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FORWARD

Dear Colleagues,

Today, I write you about how our journal is moving to the new volume as we are now in 2016, eleven years without stop, despite the challenges we faced, and despite all constraints that our beloved Arab countries have while they are looking for more development achievements. What I want to say, is that the only weapon, as well as the tool to proceed to the gate of development is science and how we can use and adopt all the ways that make our cultures, our thoughts and our talents and research efforts to be converted into practices to improve life for us and for the coming generations and let the other parts of the world listen to us very appreciately.

By this year, IJST had been awarded a new scientific impact factor, that is (the Global Impact Factor- GIF) of a value scored 0.81. In addition, IJST had awarded an increase of the value scored for SJIF to be 4.487.

By the beginning of the current year, a new Editorial Board Member has joined IJST, and it is our pleasure to welcome Prof. Taha Al- Samarrai from University of Samarra and wishing him the best times while in our IJST journey.

For all what we achieved, I would like to present my deepest thanking and great recognitions for all people and institutes who faithfully gave IJST their concerns, their cares, and their patiences to keep it as one of the leading journals in Arab and international worlds.

Thanks a lot for Prof. Jamal Abbas and Dr. Abdullah Al- Shebani from University of Kufa, Dr. Atheer Al- Douari, Prof. Hazim Al- Daraji from University of Baghdad, Prof. Waleed Al- Murrani for his endless support from Plymouth University, Prof. Abdulbari Abbas Al- Faris from University of Basrah, and finally to the one who stands always behind this great effort and performs her best with no disperence, non stopping, and with full of faith, loyalty and creative footprints at IJST, the Editorial Board Secretary of IJST. With you all, IJST is now here, and will continue as long as we breath, as we believe on our goal, and as we have the power from God to be with you.

IJST was a fruitful effort issued by the International Centre for Advancement of Sciences and Technology – ICAST, which tries to take part in both globalization and revolution in information and communication technologies, because S&T development becoming not only the key elements of economic growth and industrial competitiveness, but also essential for improving the social development, the quality of life and global environment. ICAST took then a decision to establish a scientific alliance with TSTC (Tharwa for scientific Training & Consultations) and this alliance comes to support the efforts towards publishing IJST.

Today, we announce a new issue of our journal, that is the third issue from the eleven volume of IJST, September, 2016.

Finally, I hope that all significant figures of sciences whom joined the editorial board, the researchers, and the readers of our journal will keep IJST between their eyes and contribute in continuing its journey, with their remarks, valuable recommendations and their researching outcomes.

Thanks a lot for all who support IJST.

Editor-in-Chief

IJST

Abdul Jabbar Al- Shammar
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مصدر... المدينة المستدامة

تحليل علمي لأهم تطبيقات الاستدامة في المدينة ومحاولة توظيفها لتحسين الواقع البيئي لمدينة بغداد

مالة حسين مرسي
ENGLISH SECTION
Salt tolerance of Jordanian tomato landraces at seedling stage

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ABSTRACT

Tomato (Lycopersicon esculentum) is one of the major vegetable crops grown in Jordan. Many Jordanian tomato landraces are still grown in small farms due to quality and special consumer demands. In this study, thirty nine (39) tomato landraces were exposed to three salinity levels (without salinity, 4 and 6 dS m⁻¹) at seedling stage using NaCl. The results showed that increasing salinity to 6 dS m⁻¹ caused death to 64% of the seedlings of all tomato accessions. Fourteen seedlings accessions were alive and highly varied in their influenced to salinity for fresh and dry shoot and root weights, shoot and root growth rates. Accession 24 (Jo970) showed the highest tolerance to salinity at seedling stage, while the accession 37 (Jo957) was the most sensitive to salinity.

Keywords: Tomato (Lycopersicon esculentum), landraces, salinity
INTRODUCTION

Tomato (Lycopersicon esculentum Mill.) is an annual herbaceous plant belongs to the Solanaceae family. It is one of the most important vegetable crops in many countries including Jordan. The average annual area planted with tomato during 2001-2005 in Jordan was 9.1 thousand hectares with an average production 426.9 thousand tons (1). Most of tomato cultivars grown in Jordan are hybrids with high water and fertilizer requirement and low tolerance to environmental stresses. However, tomato landraces which is less sensitive to environmental stresses and grown mainly under rain fed condition are still grown in small farms due to, quality and special demand of some consumers. These landraces are valuable sources of genetic characteristic, which is of plant breeders interest to include in breeding programs for crop improvement. Since 1983, seed samples of tomato landraces were collected from local farmers throughout the country and conserved in the gene bank of the National Center of the Agricultural Research and Extension (NCARE). High variations in vegetative and reproductive traits of these landraces were recorded (2). These variations were expected due to the fact that these landraces were collected from different regions and different farmers. Tomato in Jordan is transplanted in nurseries and grown in open field or under greenhouses (3). The shortage of good quality water in local resources is becoming an important issue. For this reason, the use of available saline water become important and should receive immediate consideration.

Tomato acts as a model crop for the use of saline and poor quality water because of the wealth of knowledge available on physiology and genetics of this species (4). It has been catalogued as moderately sensitive to salinity at all stages of plant development, and as a result, tomato yield is reduced under salt stress (5). Guerrier (6) pointed out that tomato species have significant difference in salt tolerance. Foolad and Lin (7) showed that wild tomato species are more salt tolerant than cultivated tomato. Generally, salt tolerance is increased with plant age (8). Commercial tomato cultivars are most vulnerable to salinity at early seedling growth stages (9). Al-karak (10) found that increasing level of salinity had negative effect on tomato growth. Plants grown under saline conditions are subjected to different types of stresses including water stress caused by osmoticum, mineral toxicity, and disturbance in the mineral nutrition of the plant (11-13). Cuartero and Fernandez-Munoz (4) reported that plants showed adaptive responses to salinity. While Franco et al. (14) reported that plants could survive under salty conditions expressing their pre-existing genetic information for tolerance. Early application of salt can induce salt adaptation in some tomato cultivars (15). Tomato landraces response to different level of salt is expected.

The present study aimed to examine the effect of three levels of NaCl salt stress at tomato landraces seedling stage.

MATERIALS AND METHODS

Thirty-nine (39) tomato landraces from Jordan were used in this study (table 1). Seeds of these landraces were provided by the National Centre for Agricultural Research and Extension (NCARE) Amman, Jordan. The experiment was conducted at the glasshouse of Jordan University of Science and Technology (JUST). Twenty-two seeds from each tomato landraces were sown directly in polystyrene trays (209 cells) filled with peatmoss and perlite (2:1). One seed were sown in each cell and each trays received 1 ml of liquid fertilizer (20:20:20) with 1L of salinity treatment (control, 4 dSm⁻¹, 6 dSm⁻¹) immediately after sowing then covered with plastic sheet. Four days after sowing, the trays were placed on suspended support. The experiment was carried out under greenhouse condition using completely randomized design with three replicates. Plants were harvested after they had been grown for 45 days. For each treatment, 5 plants were chosen randomly from each replicate and separated into shoots and roots to determine fresh and dry weights. Shoots and roots were oven dried at 65°C for 72 hrs.

Table (1): Region of collection and fruit shape of tomato landraces used in the study

<table>
<thead>
<tr>
<th>Code</th>
<th>Accession No.</th>
<th>Region of Location</th>
<th>Fruit shape</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>111A</td>
<td>Kharja</td>
<td>Round</td>
</tr>
<tr>
<td>2</td>
<td>111B</td>
<td>Kharja</td>
<td>Round</td>
</tr>
<tr>
<td>3</td>
<td>960</td>
<td>Shatanah</td>
<td>Slightly flattened</td>
</tr>
<tr>
<td>4</td>
<td>951</td>
<td>Al al</td>
<td>Round</td>
</tr>
<tr>
<td>5</td>
<td>952</td>
<td>Al al</td>
<td>Round</td>
</tr>
<tr>
<td>6</td>
<td>956</td>
<td>Hebras</td>
<td>Flattened</td>
</tr>
<tr>
<td>7</td>
<td>972</td>
<td>Wadi Musa</td>
<td>Flattened</td>
</tr>
<tr>
<td>8</td>
<td>973</td>
<td>Rhaba</td>
<td>Flattened</td>
</tr>
<tr>
<td>9</td>
<td>967A</td>
<td>Rhaba</td>
<td>Flattened</td>
</tr>
<tr>
<td>10</td>
<td>967B</td>
<td>Rhaba</td>
<td>Flattened</td>
</tr>
<tr>
<td>11</td>
<td>971A</td>
<td>Rhaba</td>
<td>Flattened</td>
</tr>
<tr>
<td>12</td>
<td>971B</td>
<td>Rhaba</td>
<td>Flattened</td>
</tr>
<tr>
<td>13</td>
<td>971</td>
<td>Rhaba</td>
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</tr>
<tr>
<td>14</td>
<td>961</td>
<td>Ain Jamah</td>
<td>U shape</td>
</tr>
<tr>
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</tr>
<tr>
<td>16</td>
<td>988</td>
<td>Ain Al-Baida</td>
<td>Flattened</td>
</tr>
<tr>
<td>17</td>
<td>989</td>
<td>Ain Al-Baida</td>
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</tr>
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<td>958</td>
<td>Sakh</td>
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</tr>
<tr>
<td>20</td>
<td>974A</td>
<td>Rhaba</td>
<td>High round</td>
</tr>
<tr>
<td>21</td>
<td>974B</td>
<td>Rhaba</td>
<td>Slightly flattened</td>
</tr>
<tr>
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</tr>
<tr>
<td>23</td>
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<td>24</td>
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</tr>
<tr>
<td>26</td>
<td>991</td>
<td>Afra</td>
<td>Flattened</td>
</tr>
<tr>
<td>27</td>
<td>991A</td>
<td>Ain Al-Baida</td>
<td>Round</td>
</tr>
<tr>
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<td>991B</td>
<td>Ain Al-Baida</td>
<td>Round</td>
</tr>
<tr>
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</tr>
<tr>
<td>30</td>
<td>959</td>
<td>Anjara</td>
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<tr>
<td>31</td>
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<td>Rhaba</td>
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</tr>
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<td>32</td>
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<td>Round</td>
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<tr>
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<td>985</td>
<td>Ain Al-Baida</td>
<td>Flattened</td>
</tr>
<tr>
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<td>986</td>
<td>Ain Al-Baida</td>
<td>Flattened</td>
</tr>
<tr>
<td>36</td>
<td>987</td>
<td>Ain Al-Baida</td>
<td>Flattened</td>
</tr>
<tr>
<td>37</td>
<td>957</td>
<td>Hebras</td>
<td>Round</td>
</tr>
<tr>
<td>38</td>
<td>955</td>
<td>Qasim</td>
<td>Slightly flattened</td>
</tr>
<tr>
<td>39</td>
<td>980A</td>
<td>Afra</td>
<td>Round</td>
</tr>
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</table>
Relative reduction of shoot or root = [shoot (root) fresh weight at control - shoot (root) fresh weight at salinity treatment / shoot (root) fresh weight at control]*100

Growth rate of shoot (root) = shoot (root) fresh weight / 45 days

Relative reduction of shoot (root) growth rate = [shoot (root) growth rate at control - shoot (root) growth rate at salinity treatment / shoot (root) growth rate at control]*100

In this experiment, at salinity level 4 dS m⁻¹ only 34 landraces were survival and 14 landraces were survival at salinity level 6 dS m⁻¹. Relative reduction of each level was determined compared with control. Data were statistically analyzed using the Genstat program six edition. Means were separated according to LSD (0.05).

RESULTS

Results showed that shoot and root growth rates of the tested tomato landraces were decreased significantly with increasing salinity when compared to the control (tables 2 and 3). At 4 dS m⁻¹ salinity level, the lower relative reduction in shoot fresh weight is 22% for accession 21(Jo 974A) whereas the highest relative reduction was 69% for accession 1(Jo111A). At salinity level 6 dS m⁻¹, the lowest relative reduction in shoot fresh weight was 51% for accession 24(Jo 970), while the highest relative reduction was 76% for accession 37(Jo 957).

The lowest relative reduction in root fresh weight at 4 dS m⁻¹ salinity was 38% for accession 36 (Jo 987) while the highest relative reduction was 90% for accession 25(Jo 969). At 6 dS m⁻¹ salinity level, the lowest relative reduction in root fresh weight was 87% for accession 24(Jo 970) where as the highest relative reduction was 97% for accession 33(Jo 963). At salinity level 4 dS m⁻¹, the lowest relative reduction in shoot dry weight was 19% for accession 21(Jo 974A), whereas the highest relative reduction was 68% for accession 33(Jo 963). At 6 dS m⁻¹ salinity level, the lowest relative reduction in shoot dry weight was 50% for accession 39(Jo 980A), while the highest relative reduction was 76% for accession 37(Jo 957).

The lowest relative reduction in root dry weight at 4 dS m⁻¹ was 28% for accession 23(Jo 978), while the highest relative reduction was 83% for accession 29(Jo 964). At 6 dS m⁻¹ salinity level, the lowest relative reduction in root dry weight was 56% for accession 24(Jo 970), where as the highest relative reduction was 80% for accession 36(Jo 987).

In this experiment the lowest relative reduction in shoot growth rate at 4 dS m⁻¹ salinity level was 24% in accession 19(Jo 958), while the highest relative reduction was 83% for accession 29(Jo 964). At 6 dS m⁻¹ salinity level, the lowest relative reduction in shoot growth rate was 51% for accession 24(Jo 970), where as the higher relative reduction was 76% for accession 37(Jo 957). The lowest relative reduction in root growth rate at 4 dS m⁻¹ salinity level was 24% for accession 21(Jo 974A), while the higher relative reduction was 84% for accession 28(Jo 991B). At 6 dS m⁻¹ salinity level, the lowest relative reduction in root growth rate was 44% in accession 23(Jo 978), while the highest relative reduction was 83% for accession 33(Jo 963).

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**DISCUSSION**

Salinity is currently one of most severe a biotic factor limiting agricultural production. Plants are subjected through their life cycle to different stresses; some of these plants can tolerate these stresses in different ways depending on plant species and the type of stress. Salinity affects some major processes such as growth (16). In this study, tomato landraces exhibited significant difference among each other at each salinity level during seedling growth, this possibly due to high genetic variation among them. Shoot and root growth decreased with increasing salinity and it was vary among tomato landraces. These results are in agreement with the finding of other researchers (17,18), who attributed this reduction in shoot and root growth under salinity to lower osmotic...
potential in the medium which decrease water content in the root and in the shoot, or to ion toxicity (19). Dumbroff and Cooper (20) reported that, in seedling stage of development, the younger the salinised seedling, the less the shoot growth.

Reduction in root growth is a common response to increasing salinity (17,18). Different reasons are possible for the reduction in root growth under salinity: cell growth restriction because of low water potential in the external medium, interference of the saline ions with the plants nutrition or the toxicity of accumulated ions leading to cell death (4). Cuartero and Fernandez-Munoz (4) stated that tomato root growth was less affected by salinity than shoot growth.

Bolarin et al. (8) indicated that the reduction in shoot dry weight is starting below 6 dS m⁻¹ in tomato plant. Reduction in shoot dry weight not attributed to the reduction in the number of leaves, this occurs only at EC above 6 dS m⁻¹ (21) but attributed to reduction in leaf area (22).

Root dry weight was decreased among landraces, different reason is possible: cell growth restriction, because of low water potential of external medium; interference of saline ions with plant nutrition or toxicity of accumulated ions. Similar outcome was obtained by Compos et al. (23), dry weight of root reduced by water salinity (1, 2, 3, 4, 5 dS m⁻³) in tomato plant.

The reduction in shoot and root dry weight due to increase salinity might be a result of a combination of osmotic and specific ion effects of Cl and Na (24). In general, salinity inhibited more shoot dry weight than root dry weight which reveals the ability of the plants in maintaining a higher root surface for water uptake, in response to the reduction of the osmotic potential of the soil solution (23).

Growth rate among landraces was reduced in response to salinity and this might be due to the reduction in photosynthetic rate as reported by (4). Growth rate of L. esculentum cultivar (VF 36) were reduced under salinity compared to salt tolerant species of L. cheesmanii (25).

REFERENCES

Effect of Citrullus Colocynthis water and alcoholic extracts on the viability of Old World Screw worm (OWS) chrysomya bezzaina fly

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ABSTRACT

The present study was carried out to investigate the effects of aqueous and alcoholic extracts of Citrullus Colocynthis on the larvae of OWS fly. Results showed that the cold-water extracts gave a high mortality (89.444%) more than hot water extracts mortality (86.111%). The results of alcoholic extract of Citrullus Colocynthis showed 100% mortality in larvae in each instars. Many concentrations of extracts were prepared (0.1,0.5,1.0) and yielded improvement of chemicals plant compounds (alkaloids, phenols, tannins, resins and oils). This study is conceded as a first investigation in Iraq to treat myiasis wounds infested by larvae of old world screw worm by natural plants products instead of chemical insecticides.

Keywords: OWS fly, Natural products, water and alcoholic extracts, Citrullus colocynthis

الملخص باللغة العربية

ستف هذا الدراسة إلى اختبار تأثير المستخلص المائي والكحولي لنبات الكوكز وافترشة عالمية لاثراء الأطوار البرقية لل-fly إمساك العالم القديم كرايسمونا بزيكا. أوضحنا النتائج أن المستخلص المائي البارد أعطى أفضل نسبة هلاك للأطوار البرقية بلغت معدلها (89.444%)، بينما بلغت معدل نسبة الهلاك للعطور البرقية لمستخلص الكوكز (86.111%). كما أوضحنا نتائج استخدام المستخلص الكحولي لنبات الكوكز في كل الأطوار البرقية بلغت 100% وكافة التراكيب المحضره (0.5, 1.0) مغاملا. وتم الكشف عن المواد الكيميائية الداخلة في تركيب الكوكز، ووجد أنه يحتوي على (الكُلِدات، الفينولات، الصابونين، النترات، الزئبق والزئبق). وتعتبر هذه الدراسة الأولى من نوعها في العراق التي تتم في إيجاد طريقة فعالة لمعالجة الجروح الخائبة بالتنويد. ببرفيه النباتية للعالم القديم باستخدام النباتات النباتية بدلا من المواد الحشرية.
**INTRODUCTION**

The old world screw fly (OWS) *Chrysomya bezziana* Villeneuve (1914) is a member of the insect family Calliphoridae and is a blood obligate parasite of worm-blood animals in the tropics and sub-tropics (1). *Chrysomya bezziana* larvae causes cutaneous myiasis (strikes) on host, which results in loss of condition; maiming, infertility and death of the host (2-5). There were 120,789 cases of OWS in Iraq recorded in animals and 22 cases in human until 2007 (6). *Chrysomya bezziana* larvae causes myiasis, which can be defined as infections of live tissues of human and animals by larvae of OWS *Chrysomya bezziana*, caliphoridae, diptera, causes damage or death (1,2,4). In Basra province, there are many cases of OWS *Chrysomya bezziana* reported in dogs, camel, buffalo, cattle, sheep and horses (1,2,7-9). Plant water extract was used in subterranean termites, as insecticide instead of chemical method (10). In addition, the extraction of *citrullus colocynthis* is used to control the larvae of sarcophagi haemorrhoidalis (11). The extracts of *Ricinus* and *Peganum harmala* are used on instars of mosquito culex pipiens molestus (13). The effects of some plant extractions on the mortality of the larval mosquitoes *culex pipiens molestus* was studied by (13). Classical chemical insecticides had harmful effects both on environment and human health. This requires use of natural plant products as substitutes, which give a good result to treat sucker plant insects (14). Natural products are also used as antibacterial agents (15). Secondary compound extractions showed good effect on activity of *Muscadomastica* fly (16).

**MATERIALS AND METHODS**

**Preparation of aqueous extracts**

*Citrullus Colocynthis* fruits were collected from Al-Rumela city, west of Basra province, Iraq, then washed carefully to remove any suspicious particles from sand or others, then dried under sun light. Philips machine was used for crushing and grinding dry plant (5). Thirty (30) gm of dried powder of plants were put in a 1000ml flask containing 500 ml cold water and normal saline mixed together by move magnetic for 15 sec. Then the sample was left for 24 hrs to be mixed carefully. The mix was filtered and transmitted to a sterile tube, then suspended to centrifuge with 300 circle / min. The supernate was collected and moved to oven at (45°C) for dryness and then kept in refrigerator until use. One mg of crude (dry plant) was solved in 100 ml distilled water containing 500 ml cold water and normal saline mixed together by move magnetic for 15 sec. Then the sample was left for 24 hrs to be mixed carefully. The mix was filtered and transmitted to a sterile tube, then suspended to centrifuge with 300 circle / min. The supernate was collected and moved to oven at (45°C) for dryness and then kept in refrigerator until use. One mg of crude (dry plant) was solved in 100 ml distilled water to have concentration 1% as stock, and 0.1%, 0.5% were prepared from this stock. Hot water method is only different from the above method by adding hot water instead of cold water. Larvae of *chrysomya bezziana* OWS were prepared in the laboratory from infested lamb as a culture. Forty eight (48) Petri dishes where prepared for each and concentrated in triple dishes, with three dishes as control for each instar and each concentration. Ten (10) larvae were put in each dish and the extractions were added to each one with different concentrations and different instar with control use too. Fresh sheep liver (50 gm) were used as a supplement food for larvae of OWS to growth. All dishes were incubated in the cold incubator at 25°C.

**Preparation of alcoholic extract**

Thirty (30) gm of dry powder of *Citruluscolocynthis* fruits were kept in a thimble and fixed in Soxhelet extracts apparatus. A glass containing of 300 ml ethyl Alcohol was heated for 24 hrs at 50°C. Alcoholic extracts were transmitted to the rotary evaporator to get a pure crude which then was dried and put into black serial glass and then kept in the refrigerator. One (1) mg of crude was diluted by adding 99 ml of distal water to have 1% and then diluted to 0.1,0.5 (17).

**Statistical analysis**

Data were based on completed randomized design by using variance with coefficient limits of P< 0.05.

**RESULTS AND DISCUSSION**

Results showed that the mean value of killed larvae of *Citrulus Colocynthis* in cold water extraction (89.444) was prerogative than the mean value of killed larvae of *Citrulus Colocynthis* in hot water extract (86.111) (tables 1 and 2). Three concentrations were used in the study (0.1, 0.5, 1.0) mg/ml of distilled water and the percentage of mean killed larvae showed (80%, 96.666%, 95.666%) respectively for cold water extraction, compared with the mean of same concentrations and same extractions of plant in hot water extraction (71.666%, 91.666%, 95.000%) respectively. Results also showed that alcoholic extracts showed mortality percentages for all instars larvae in all concentrations (table 3). There were a significant degree under p=< 0.05 statistically. The first instar larva for all concentrations and for cold and hot water extractions showed % mortality more susceptible to alcohol, and this result was in agreement with those concluded by (18,19). Chemical compound extracts were detected (table 4).
Table (1): Effects of Secondary compounds of *Citrullus colocynthis* in cold water extract in larvae instars of *(OWS)* *Chrysomya bezziana*

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<th>% conc. Mg/ml/ml</th>
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R.L.S.D To con.=34.65, R. L. S. D TO inst.= 21.43, P=0.05, p To conc. = 0.001, p To inst. = 0.02 Cons. * inst.=5.72

Table (2): Effects of secondary compounds of *Citrullus colocynthis* hot water extract in larvae instars of *(OWS)* *Chrysomya bezziana*

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<td>91.666</td>
<td>75</td>
<td>0</td>
</tr>
<tr>
<td>95</td>
<td>95</td>
<td>80</td>
</tr>
<tr>
<td>95.000</td>
<td>75</td>
<td>0</td>
</tr>
<tr>
<td>85</td>
<td>85</td>
<td>100</td>
</tr>
<tr>
<td>86.111</td>
<td>78.33</td>
<td>0.00</td>
</tr>
</tbody>
</table>

R. L. S. D TO con.= 36.53, R.L.S.D To inst. = 28.84, P=0.05 P To con. = 0.001, P To inst.=0.006 , Cons. * inst.=5.65

Table (3): Effects of secondary compounds in alcoholic extract of *Citrullus colocynthis* in larvae instars of *(OWS)* *Chrysomya bezziana*

<table>
<thead>
<tr>
<th>% Mean mortality rate conc.</th>
<th>% Mortality rate in instar</th>
<th>% conc. Mg/ml/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>3rd inst.</td>
<td>control</td>
<td>2nd inst.</td>
</tr>
<tr>
<td>100.000</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>100.000</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>100.000</td>
<td>100</td>
<td>0</td>
</tr>
</tbody>
</table>

R.L.S.D. To con.=0.063, R.L.S.D. To inst. =27.755, F < 0.05 P To con.=0.001, P To inst. = 0.001, Cons. * inst.=0.00

Table (4): Detected chemical compounds of *Citrullus colocynthis*

<table>
<thead>
<tr>
<th>Plant</th>
<th>Alkaloid reagents</th>
<th>Phenols reagents</th>
<th>Saponins reagents</th>
<th>Tannins reagents</th>
<th>Resins reagents</th>
<th>Oil reagents</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Citrullus colocynthis</em></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

No toxicity was observed on experimental rates for all plant extracts, and thus this result was in agreement with (20) who showed that the cold extract of *Citrullus colocynthis* at 16% conc. gave better result of mortality rate than hot water extract on Ch. bezziana larvae instar. Cold water extracts of *Chrysanthemum cinerarinfolium* demonstrated more effect in mortality on adults of *Tribolium castaneum* than hot water extracts (21). Persisting of enzymes in cold water plants extracts for long time without analysis produced new materials or may cause changing in activity of natural product compounds to be more active (22). The first and
second instar larvae are more sensitive to toxic substances of plants extracts than third instar, by which it can change the toxic compounds in extract to intoxick by special enzymes (M.F.O.), which the first and second instars can not yield because they lack these enzymes (23). In previous study, it was proved that the first and second instars were more effective to plants extracts due to their thin layer of cuticles (24), in addition to that the high mortality in alcoholic extract than water extract were due to alcohol extract all active component in the plants, that’s Intone whit present results. Many investigators demonstrated that the alcoholic extracts were hard effective on larvae instars of Chrysomya bezziana, Trogoderma granarium, Aphis fabae and Aphis gossypii (20,25,26).

REFERENCES

Effects of copper nanoparticles on reproductive organs of male albino rats

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ABSTRACT

Copper nanoparticles (Cu-NPs) are widely used in various industrial and commercial applications and little is known about their potential toxicity on reproductive system. In the present study, the effect of Cu-NPs on the weight of some reproductive system organs and the sperm characteristics of male albino rats were examined. Rats were administrated with 0.5 ml of 20 and 40 mg/kg/BW Cu-NPs intraperitoneally for 3, 6 and 9 days. The results showed statistically significant decrease in the final body weight and increase in the relative sex organs (testis, epididymis, seminal vesicles and prostate gland) with both Cu-NPs treatments compared to control group. A significant decrease was found in the percentage of sperm viability and increase in the percentage of sperm abnormalities and sperm concentration were observed with both Cu-NPs compared to the control with concentration and time effects. Thus, the results of this study revealed for the first time that intraperitoneal injection of Cu-NPs has a negative influence on the effectiveness and activity of male reproductive system in albino rats.

Keywords: Copper nanoparticles, Organs weight, Sperm characteristic, Rats, Testes, Epididymes

الملخص باللغة العربية

 потهدت الجزئات النانوية (Cu-NPs) بشهارة واسعة من خلال استخداماتها في العديد من الصناعات، كما أن لها تطبيقات تجارية كثيرة، ولكن لا تزال المعلومات حول تأثيرها على الجهاز التناسلي غير كافية.

تعد هذه الدراسة إلى الكشف عن تأثير جسيمات النحاس النانوية (Cu-NPs) على أوزان بعض الأعضاء التناسلية ومواصفات النطف في ذكور الجرذان البيضاء.

تم حقن الجرذان داخل البريتيون بمقدار 0.05 مل من تركيزين مختلفين من جسيمات النحاس النانوية (Cu-NPs) 20-40 ملغ / كيلوغرام وزن الجسم لمدة 3-9 أيام. وقد أظهرت النتائج وجود فروقات إحصائية معروفة تتمثل بالانخفاض في الوزن النسبي للأعضاء التناسلية (الخصيت، البربخ، الحويصلات المنوية، وعدة البروستاتا) لكل الجرعتين من جسيمات النحاس النانوية مقارنةً بمجموعة السيطرة. من ناحية أخرى، خفضت النسبة المنوية لحوية النطف إحصائياً مع زيادة ملحوظة في النسبة المنوية لتشوهات النطف وتقلباتها لكل Cu-NPs الجرعة من مجموعة السيطرة، وكان التغير في هذه النسب متلازماً بالتركيز ومدة التعريض. وقد خصصت الدراسة للمرة الأولى إلى أن حقن الجسيمات النانوية داخل البريتيون في الجرذ له تأثير كبير على نشاط وفعالية الجهاز التناسلي الذكري للجرذ الأبيض.
INTRODUCTION

Nanoparticles are coming into our daily life in excessive quantity. The unique characteristics of nanosubstances, such as size, large surface area, ultrahigh reactivity, and shape effects allow them to produce many specific effects regarding their bulky states (1). Recently, a new form of Cu metal has been engineered containing of nanoscale copper particles (Cu-NPs), which are considered as type of metal-containing nanoparticles (2). Cu-NPs are used in lubricants, polymers/plastics, textiles, metallic coating, antimicrobial, home appliances, wear resistance and inks, etc (3,4). Therefore, nano copper are likely to distribute in the environment and enter human body via different paths such as effluent, disposal, and consumer products (3) and cause toxicity via pulmonary, oral, nasal, skin or other routes of exposure. Then, nanoparticles can penetrate through cell membrane via blood-brain barrier and blood-testis barrier (5,6) to reach and impact all organs of the body (7). In *in vitro* study, Cu-NPs and CuO-NPs were found to be able to generate oxidative stress (8,9). In fish, Ostaszewska et al.(10) found that 0.15 mg L⁻¹ of Cu-NPs caused pathological changes in epidermis (irregular structure and pyknotic nuclei), gills (lifting epithelium, fusion of lamellae and epithelial necrosis), and liver (dilatation of sinusoid space, pyknotic nuclei and overfilled blood vessels) of *Siberian sturgeon* larvae after 21 days of exposure. Additionally, Al-Bairuty et al.(11) found that 20 and 100 µg/g of Cu-NPs caused pathologies in gills, liver, kidney, gut, brain and muscle of rainbow trout after 10 days of exposure. In *in vivo* study, oral administration of Cu-NPs in mice caused injuries in the kidney, liver and spleen (3). However, there are still scarce and controversially evident of effects of Cu-NPs on spermatogenesis parameters and other reproductive indices such as organs weight and pathologies. Little is known about the effects of other types of nanoparticles on reproductive organs and sperm characteristic in animals. For example oral administration of 20 µg/kg/day Ag-NPs (AgNPs) caused disorganization of the germinal epithelium with loss of some spermatogenic cells types (spermatocytes and spermatids) and degeneration, necrosis as well as atrophy of the seminiferous tubules after 90 days of exposure (12). Kim et al. (13) found that oral administration of AgNPs in male albino rats caused decline in body weight, accumulation of NPs and enlargement of testis as well as some hepatocyte toxicity after 90 days of exposure. Oral administration of AgNPs in male albino mice caused decrease in the weights of testis and tunica albuginea, epididymis head and tail, and decline in the percentage of sperm vitality and sperm concentrations as well as an increase in the percentage of sperm abnormalities were observed in both testis and epididymis after 5, 10, 15 days of exposure (14). Ema et al. (15) had reviewed that nanoparticles are able to penetrate into reproductive tissue through biological barriers and may damage various cells such as reduce sperm numbers, viability and change cell functions, as well as inhibit the embryo development. However, there is a gap information about the effect of intraperitoneal injection of Cu-NPs on the male reproductive system indices. Therefore, the aim of this study was to investigate the effect of intraperitoneal injection of different doses of Cu-NPs (20 and 40 mg/kg/BW) on body weight, the weight of some organs of male reproductive system, and sperm characteristic in testes and epididymes (head, tail) of albino rats.

MATERIALS AND METHODS

Characterization of copper nanoparticles (Cu-NPs)

The powder of Copper nanoparticles were purchased from Xuzhou Hongwu Nanometer material CO.,LTD. with 20-30 nm size and purity 99.99% (manufacture Hongwunanometer).

Animals and treatments

Male albino rats (197- 200 gms weight and 10-12 weeks old) were obtained from National Center for Control and Pharmaceutical Research /Baghdad/ Iraq and housed in animal house of biological department/ College of Education for Pure Sciences/ Ibn Al-Haitham. Animals were fed with standard commercial pelleted diet and tap water and maintained under a 12:12 h light: dark cycle and room temperature 22–24°C. Rats were kept in polycarbonate cages with woodchip bedding and acclimatized to the environment for 2 weeks prior to experimental use. Animals were randomly divided into 3 groups (n =54). The animals were assigned to each experimental group, with 18 animals per group and 6 per day.

**Group 1** - Control group of animals were intraperitoneal injected with 0.5 ml of distilled water once a day for 3, 6, and 9 days (each day have 6 animals).

**Group 2** -Experimental group of animals were intraperitoneal injected with 0.5 ml of 20 mg/kg/BW Cu-NPs once a day for 3, 6, and 9 days (each day have 6 animals).

**Group 3** - Experimental group of animals were intraperitoneal injected with 0.5 ml of 40 mg/kg/BW Cu-NPs once a day for 3, 6, and 9 days (each day have 6 animals).

Preparation of copper nanoparticles

The low (20mg/kg/BW) and high (40 mg/kg/BW) concentrations of Cu-NPs suspensions were prepared by weighting 0.8 and 1.6 g of Cu-NPs powder and diluted with 100 ml of distilled water, then stirred with magnetic stirrer overnight and next dispersed by ultrasonic vibration for 30 min. Before injection, the low and high concentrations of Cu-NPs were vortexed in order to avoid the aggregation of the particles.

Animal weighting, sample collection and preparation

Before and after the end of 3, 6, and 9 days of the exposure duration, six rats were weighted by using a Mettler pc Balance in order to record Initial and final body weights. Then, rats were anaesthetized (using diethyl ether) and sacrificed for samples collection from each groups.
Organs samples

Testis, epididymis (head and tail), seminal vesicle and prostate gland from each rats were collected and weighed by using a sensitive balance. The coefficient-relative organ weights were calculated as organ wet weight, (gm/body weight of animal *100%).

Evaluation of sperm viability, morphology and concentration

The spermatozoa were collected from the testis and epididymis (head and tail) and examined for live and dead sperms, sperm cell concentration and sperm abnormalities according to (16).

Statistical analysis

All data values were giving as mean ± standard error (S.E.). The resulting data of the current study was analyzed using the SPSS software, version 21. Data were tested for treatment, time, and treatment x time interaction effects by using general linear model/multivariate followed by least squares difference post hoc test. When a statistically significant effect was showed by this model, a one way ANOVA was used to assess for simple effects. The differences were considered significant at level p ≤0.05.

RESULTS

Effect of Cu-NPs on body weight and coefficient-relative reproductive organs weight

The results exhibited significantly decrease (P ≤ 0.05) in the average body weights of animals after 3, 6 and 9 days of injected with 20 and 40 mg/kg Cu-NPs compared to the same animals groups before injected (table 1). Some concentration and time effects were observed in the average body weight after treated with Cu-NPs in this study where there were more significantly decrease in the average body weight of animal injected with 40 mg/kg Cu-NPs for 9 days compared with 20 mg/kg Cu-NPs for 3 and 6 days (table 1). The average body weight of control animals showed a statistically significant increased (p ≤ 0.05) after injected with distill water for different periods (6, 9) compared to the same control animal before injected (table 1).

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Time/day</th>
<th>Initial body weight before treatment (gm)</th>
<th>Final body weight after treatment (gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>3 days</td>
<td>200.67 ± 2.91</td>
<td>206.5 ± 3.34 a</td>
</tr>
<tr>
<td>20 mg/kg Cu-NPs</td>
<td>3 days</td>
<td>202.67 ± 3.99</td>
<td>195.83 ± 4.02 a</td>
</tr>
<tr>
<td>40 mg/kg Cu-NPs</td>
<td>3 days</td>
<td>197.00 ± 3.36</td>
<td>180.10 ± 3.80 *b</td>
</tr>
<tr>
<td>Control</td>
<td>6 days</td>
<td>200.66 ± 1.81</td>
<td>214.00 ± 2.47 *a</td>
</tr>
<tr>
<td>20 mg/kg Cu-NPs</td>
<td>6 days</td>
<td>197.00 ± 2.69</td>
<td>181.83 ± 2.81 *b</td>
</tr>
<tr>
<td>40 mg/kg Cu-NPs</td>
<td>6 days</td>
<td>204.66 ± 3.44</td>
<td>167.66 ± 4.36 *c</td>
</tr>
<tr>
<td>Control</td>
<td>9 days</td>
<td>199.83 ± 2.66</td>
<td>222.00 ± 1.94 *a</td>
</tr>
<tr>
<td>20 mg/kg Cu-NPs</td>
<td>9 days</td>
<td>198.17 ± 1.68</td>
<td>167.17 ± 1.83 *b</td>
</tr>
<tr>
<td>40 mg/kg Cu-NPs</td>
<td>9 days</td>
<td>197.66 ± 0.66</td>
<td>146.17 ± 2.21 *c</td>
</tr>
</tbody>
</table>

Data are mean ± SEM (n = 6 rats/treatment). * Significantly different between initial body weights and final body weights (ANOVA, P ≤ 0.05). Values with different letters within column were significantly different (ANOVA, p ≤0.05). ● Significantly different between day 3 and 9 at the same treatment (ANOVA, p ≤0.05). ▲ Significantly different between day 6 and 9 at the same treatment (ANOVA, p ≤0.05) ■ Significantly different between day 3 and 6 at the same treatment (ANOVA, p ≤0.05)

The coefficient reproductive organs weights (Testis, epididymis head, epididymis tail, seminal vesicle and prostate glands) were statistically significant increased (p ≤0.05) with both 20 and 40 mg/kg of Cu-NPs at different time points compared to the control groups (table 2). More increasing in the coefficient reproductive organs weights were observed over a long time of exposure. The coefficient weight of tunica albuginea was statistically significant increased (p ≤ 0.05) with high concentration of Cu-NPs at day 9 compared to the control group (table 2). A concentration effect was observed in the coefficient weight of prostate gland and a time effect detected in the coefficient weights of tunica albuginea, epididymis tail and prostate gland (table 2).
perm abnormalities in testis and epididymis (head, tail) showed a time effect (table 4). Table 5 reveals significant increase (p ≤ 0.05) in the percentage of sperm abnormalities in the testis and epididymis head and tail with both Cu-NPs concentrations over longtime of exposure. The elevation of sperm abnormalities in the testis and epididymis (head, tail) showed concentration and time effects with more increasing with high Cu concentration at day 9 when compared to low Cu concentration at day 3 and 6 (table 5).

The sperm abnormalities that observed with both Cu-NPs concentrations at day 3, 6 and 9 involved deformed head, detached head, coiled head, curled tail, curved tail and loss of head spine when compared to normal sperm structure that consist of head with spine, middle piece, and tail (figure 1).

Table (3): The percentage of sperms viability (sperms life) in rats injected with 20 and 40 mg/kg of Cu-NPs for 3, 6 and 9 days

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Time/Days</th>
<th>Control</th>
<th>20 mg/kg Cu-NPs</th>
<th>40 mg/kg Cu-NPs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Testes</td>
<td>3</td>
<td>0.024 ± 0.002 a</td>
<td>0.024 ± 0.001 a</td>
<td>0.024 ± 0.000 a</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.025 ± 0.001 a</td>
<td>0.025 ± 0.000 a</td>
<td>0.027 ± 0.001 a</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.024 ± 0.002 a</td>
<td>0.029 ± 0.002 a</td>
<td>0.035 ± 0.001 b</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>0.555 ± 0.025 a</td>
<td>0.576 ± 0.021 a</td>
<td>0.578 ± 0.021 a</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.517 ± 0.009 a</td>
<td>0.578 ± 0.022 a</td>
<td>0.662 ± 0.032 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.486 ± 0.057 a</td>
<td>0.671 ± 0.036 b</td>
<td>0.675 ± 0.022 b</td>
</tr>
<tr>
<td>Testis tissues</td>
<td>3</td>
<td>0.109 ± 0.005 a</td>
<td>0.111 ± 0.005 a</td>
<td>0.106 ± 0.005 a</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.098 ± 0.004 a</td>
<td>0.114 ± 0.006 b</td>
<td>0.127 ± 0.003 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.093 ± 0.008 a</td>
<td>0.128 ± 0.008 b</td>
<td>0.135 ± 0.003 b</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>0.078 ± 0.002 a</td>
<td>0.086 ± 0.005 ab</td>
<td>0.105 ± 0.009 b</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.081 ± 0.005 a</td>
<td>0.108 ± 0.003 b</td>
<td>0.125 ± 0.009 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.069 ± 0.004 a</td>
<td>0.124 ± 0.007 b</td>
<td>0.118 ± 0.009 b</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>0.271 ± 0.021 a</td>
<td>0.565 ± 0.010 b</td>
<td>0.567 ± 0.033 b</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.224 ± 0.008 a</td>
<td>0.417 ± 0.041 b</td>
<td>0.626 ± 0.108 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.276 ± 0.012 a</td>
<td>0.592 ± 0.056 b</td>
<td>0.638 ± 0.124 b</td>
</tr>
<tr>
<td>Epididymes</td>
<td>3</td>
<td>0.172 ± 0.020 a</td>
<td>0.337 ± 0.057 b</td>
<td>0.313 ± 0.043 b</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.226 ± 0.006 a</td>
<td>0.410 ± 0.048 b</td>
<td>0.356 ± 0.045 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>0.142 ± 0.010 a</td>
<td>0.386 ± 0.027 b</td>
<td>0.263 ± 0.030 c</td>
</tr>
</tbody>
</table>

Table (2): The coefficient -relative organs weight in albino rats injected with 20 and 40 mg/kg of Cu-NPs for 3, 6 and 9 days

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Time/ Days</th>
<th>Control</th>
<th>20 mg/kg Cu-NPs</th>
<th>40 mg/kg Cu-NPs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Testis tissues</td>
<td>3</td>
<td>56.50 ± 0.21 a</td>
<td>65.00 ± 0.31 b</td>
<td>66.17 ± 0.47 b</td>
</tr>
<tr>
<td>Testic tissues</td>
<td>6</td>
<td>57.11 ± 0.33 a</td>
<td>63.83 ± 0.31 b</td>
<td>63.83 ± 0.31 b</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>58.50 ± 0.21 a</td>
<td>60.83 ± 0.31 b</td>
<td>60.83 ± 0.31 b</td>
</tr>
<tr>
<td>Epididymes</td>
<td>3</td>
<td>58.50 ± 0.21 a</td>
<td>61.33 ± 0.31 b</td>
<td>61.33 ± 0.31 b</td>
</tr>
<tr>
<td>Epididymis head</td>
<td>6</td>
<td>59.17 ± 0.31 a</td>
<td>64.17 ± 0.48 b</td>
<td>64.17 ± 0.48 b</td>
</tr>
<tr>
<td>Epididymis tail</td>
<td>9</td>
<td>59.17 ± 0.31 a</td>
<td>59.17 ± 0.31 b</td>
<td>59.17 ± 0.31 b</td>
</tr>
</tbody>
</table>

Table (3): The percentage of sperms viability (sperms life) in rats injected with 20 and 40 mg/kg/BW of Cu-NPs for 3, 6 and 9 days

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Time/Days</th>
<th>Control</th>
<th>20 mg/kg Cu-NPs</th>
<th>40 mg/kg Cu-NPs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Testis</td>
<td>3</td>
<td>96.67 ± 0.21 a</td>
<td>66.17 ± 0.47 b</td>
<td>64.33 ± 0.88 c</td>
</tr>
<tr>
<td>Testic tissues</td>
<td>6</td>
<td>96.50 ± 0.43 a</td>
<td>63.83 ± 0.31 b</td>
<td>61.67 ± 0.33 c</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>97.33 ± 0.33 a</td>
<td>60.83 ± 0.31 b</td>
<td>60.33 ± 0.06 c</td>
</tr>
<tr>
<td>Epididymes head</td>
<td>3</td>
<td>97.83 ± 0.31 a</td>
<td>64.17 ± 0.48 b</td>
<td>60.33 ± 0.06 c</td>
</tr>
<tr>
<td>Epididymis tail</td>
<td>6</td>
<td>97.67 ± 0.21 a</td>
<td>61.33 ± 0.31 b</td>
<td>58.17 ± 0.31 c</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>98.17 ± 0.31 a</td>
<td>59.17 ± 0.31 b</td>
<td>53.67 ± 0.49 c</td>
</tr>
</tbody>
</table>

Effect of Cu-NPs on sperm parameters in testes and epididymis

The effect of daily injected to 20 and 40 mg/kg Cu-NPs for 3, 6 and 9 days on sperm cells life, sperm concentration and sperm abnormalities in testis and epididymes (head and tail) of albino rats were shown in tables (3-5). The percentage of sperm life in the testis and epididymes (head and tail) showed significant decrease (p ≤ 0.05) compared to control groups of each treatment and time point with more decreasing with high concentration of Cu-NPs and longtime of exposure (table 3). The sperm concentrations in the testis and epididymes (head and tail) were exhibited significant increased (p ≤ 0.05) with both Cu-NPs concentration compared to the control (table 4). A concentration effects of Cu-NPs were observed in the testis and epididymes tail with less increase with 40 mg/kg Cu-NPs when compared to 20 mg/kg Cu-NPs at the same time point (table 4). The sperm concentration in the testis and epididymes (head, tail) showed a time effect (table 4). Table 5 reveals significant increase (p ≤ 0.05) in the percentage of sperm abnormalities in the testis and epididymes head and tail with both Cu-NPs concentrations over longtime of exposure. The elevation of sperm abnormalities in the testis and epididymes (head, tail) showed concentration and time effects with more increasing with high Cu concentration at day 9 when compared to low Cu concentration at day 3 and 6 (table 5).The sperm abnormalities that observed with both Cu-NPs concentrations at day 3, 6 and 9 involved deformed head, detached head, coiled head, curled tail, curved tail and loss of head spine when compared to normal sperm structure that consist of head with spine, middle piece, and tail (figure 1).
Table (4): The sperms concentration in rats injected with 20 and 40 mg/kg/BW of Cu-NPs for 3, 6 and 9 days

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Time/Days</th>
<th>Control</th>
<th>20 mg/kg Cu-NPs</th>
<th>40 mg/kg Cu-NPs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Sperms concentrations *10^3</td>
<td>Sperms concentrations *10^3</td>
</tr>
<tr>
<td>Testes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>51979.2 ± 408.8 a</td>
<td>71666.7 ± 2000.4 b</td>
<td>68125.0 ± 2060.3 b</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>54270.8 ± 987.1 a</td>
<td>54270.8 ± 1254.3 a</td>
<td>57500.0 ± 2212.7 a</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>51354.2 ± 298.3 a</td>
<td>66562.5 ± 576.2 b</td>
<td>54270.8 ± 746.8 c</td>
<td></td>
</tr>
<tr>
<td>Epididymis Head</td>
<td>3</td>
<td>26541.7 ± 150.2 a</td>
<td>45958.3 ± 318.9 b</td>
<td>42666.7 ± 333.3 c</td>
</tr>
<tr>
<td>6</td>
<td>31208.3 ± 1309.3 a</td>
<td>32291.7 ± 1918.6 a</td>
<td>31958.3 ± 3159.9 a</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>26708.3 ± 135.7 a</td>
<td>32041.7 ± 1109.5 b</td>
<td>29916.7 ± 1394.4 b</td>
<td></td>
</tr>
<tr>
<td>Tail</td>
<td>3</td>
<td>30750.0 ± 129.1 a</td>
<td>68291.7 ± 881.3 b</td>
<td>65833.3 ± 1247.8 b</td>
</tr>
<tr>
<td>6</td>
<td>36000.0 ± 766.5 a</td>
<td>36125.0 ± 1938.4 a</td>
<td>50375.0 ± 1311.4 b</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>34125.0 ± 201.6 a</td>
<td>40541.7 ± 439.8 b</td>
<td>30416.7 ± 1457.3 c</td>
<td></td>
</tr>
</tbody>
</table>

Data are mean ± SEM (n = 6 rats/treatment). Values with different letters within row were significantly different (ANOVA, p ≤ 0.05). ● Significantly different between day 3 and 9 at the same treatment (ANOVA, p ≤ 0.05). ▲ Significantly different between day 6 and 9 at the same treatment (ANOVA, p ≤ 0.05). ■ Significantly different between day 3 and 6 at the same treatment (ANOVA, p ≤ 0.05).

Table (5): The percentage of sperms abnormalities in rats injected with 20 and 40 mg/kg/BW of Cu-NPs for 3, 6 and 9 days

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Time/Days</th>
<th>Control</th>
<th>20 mg/kg Cu-NPs</th>
<th>40 mg/kg Cu-NPs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Percentage of sperms abnormalities</td>
<td>Percentage of sperms abnormalities</td>
</tr>
<tr>
<td>Testes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>7.83 ± 0.31 a</td>
<td>46.63 ± 0.60 b</td>
<td>57.33 ± 0.49 c</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>6.73 ± 0.40 a</td>
<td>73.63 ± 0.61 c</td>
<td>95.33 ± 0.33 c</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>8.33 ± 0.33 a</td>
<td>88.33 ± 0.33 b</td>
<td>55.33 ± 0.49 c</td>
<td></td>
</tr>
<tr>
<td>Epididymis Head</td>
<td>3</td>
<td>6.50 ± 0.22 a</td>
<td>42.17 ± 0.60 b</td>
<td>54.50 ± 0.42 c</td>
</tr>
<tr>
<td>6</td>
<td>6.83 ± 0.31 a</td>
<td>63.67 ± 0.67 b</td>
<td>73.17 ± 0.79 c</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>5.67 ± 0.33 a</td>
<td>61.17 ± 0.48 b</td>
<td>70.83 ± 0.47 c</td>
<td></td>
</tr>
<tr>
<td>Tail</td>
<td>3</td>
<td>5.50 ± 0.22 a</td>
<td>40.50 ± 0.43 b</td>
<td>51.17 ± 0.60 c</td>
</tr>
<tr>
<td>6</td>
<td>5.67 ± 0.33 a</td>
<td>61.17 ± 0.48 b</td>
<td>70.83 ± 0.47 c</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>5.83 ± 0.17 a</td>
<td>82.50 ± 0.50 b</td>
<td>87.17 ± 0.31 c</td>
<td></td>
</tr>
</tbody>
</table>

Data are mean ± SEM (n = 6 rats/treatment). Values with different letters within row were significantly different (ANOVA, p ≤ 0.05). ● Significantly different between day 3 and 9 at the same treatment (ANOVA, p ≤ 0.05). ▲ Significantly different between day 6 and 9 at the same treatment (ANOVA, p ≤ 0.05). ■ Significantly different between day 3 and 6 at the same treatment (ANOVA, p ≤ 0.05).

Figure (1): (A) (cu) normal sperm shape and (B) types of sperm abnormalities in rats intraperitoneal injection with 20 and 40 mg/kg Cu-NPs for 9 days. Normal sperm (n) consist of head with spine, middle piece, and tail. The sperm abnormalities involved deformed head (df); detached head (dt); loss of head spine (ls); coiled head (ch); coiled tail (co) and curved tail (cu).
DISCUSSION

Body and reproductive organs weights

Decline in the average body weight reported in this study are consistent with Zhang et al. (17), who found that the intraperitoneal injection of gold nanoparticles after 14 days in mice induced a decline in the body weight. The authors suggested that this way of exposure may cause some toxicity. Decreasing the average body weight observed in this study may attributed to the disturbance in different metabolic activity that resulted from intraperitoneal injection. Other types of nanoparticles (25 mg/kg TiO$_2$ NP) also caused decrease in the rat body weight when intraperitoneal injection (18). Another ways of exposure to nanoparticles also caused decrease in the body weight (13, 17). For example, Kim et al. (13) found that oral exposure to 30, 125, and 500 mg/kg of Ag-NPs for 13 weeks in male rats produced a significant decrease (P < 0.05) in the bodyweight after 4 weeks of exposure. A study by Zhang et al (17) found that oral administration of 1100 µg/kg gold nanoparticles over 28 days caused a decrease in the body weight and the authors suggested that oral exposure to NPs could produce some effects on the digestive system. The body weight of male albino rats fed 1% and 2% TiO2 NP showed decline after 65 days of exposure (19).

The results of this work revealed that intraperitoneal injection of 20 and 40 mg/kg Cu-NPs resulted in significant increase in testis, epididymis head and tail, seminal vesicle and prostate glands relative weights compared to control group at all-time point (3, 6 and 9 days). The elevation of relative sex organs weights observed in this study may be related to histopathological changes in sex organs (20) or to the level of serum testosterone that play a major role in the maintenance of structural integrity and functional efficacy of the sex accessory glands (21). Kong et al. (20) illustrated that administration of different doses Ni NPs caused increase in the ratio of epididymis weight over body weight and testes pathologies. A study by Bakare et al. (22) also found that intraperitoneal administration of 0.5 ml TiO$_2$ NPs in mice caused pathological changes in the testicular tissue such as congestion of the inter-stitiumoedema, vacuolation and necrosis. Garcia et. al. (23) found that intravenously injected of 1 mg/kg Ag NPs in mice caused elevation serum and intratesticular testosterone concentration after 15 days of injection.

Sperm parameters

The present study revealed that intraperitoneal injection of 20 and 40 mg/kg Cu-NPs at different time points caused a decrease in the percentage of sperm life in the testis and epididymes (head and tail) with concentration and time effect. This reduction is occur due to the effects of nanoparticles on the germ cells, which lead to disturbance of spermatogenic process. Gromadzka-Ostrowska et al. (24) exhibited that nanoparticles have the ability to penetrate the blood-testis barrier and cause toxic effects on male germ cells and reduced sperm quality. A study by Xu et al. (25) illustrated mitochondrial damaging and decline the levels of ATP resulting oxidative stress in the testis, DNA damaging and decrease in the quantity and quality of epididymal sperm in mice injected with 20 mg/kg of silica nanoparticles for 15 and 35 days. An increase in the percentage of sperm abnormalities and sperm concentration that observed in the current study may be caused due to the effect of Cu-NPs on the sertoli cells function or oxidative stress. The results of this study is in consistence with the suggestion of Kruszewski et al. (26) who reported that Ag-NPs could react with cellular DNA and stimulated inflammation, oxidative damage and cellular dysfunction that created genetic mutation and sperm cells with abnormal morphology. In testes, the sperm cells are almost immature, and they are getting mature through passing the epididymides tail (27). Talebi et al. (28) found that oral administration of 50 and 300 mg/kg zing nanoparticles for 35 consecutive days caused the presence of vacuoles in the cytoplasm of sertoli cells and affect the function of these cells which led to increase the percentage of sperm abnormalities. Exposure to silver nanoparticles can cause the inflammation and oxidative damage as well as increase the rates of sperm abnormalities and genetic mutations (29). Smith et al. (30) found that at 4–8 days post-injection of anatase titanium dioxide nanoparticles in mice induce structural and functional sperm deficiency associated with infertility, and DNA damage via oxidative stress. The oral administration with 200 mg/kg of Ag NPs in mice for 5, 10, and 15 days caused decrease in the percentage of sperm viability and sperm concentration as well as an increase in the percentage of sperm abnormalities that observed in the testis and epididymis (14). In rats, the oral exposure to 100 mg/kg of TiO$_2$ NPs for 8 weeks also caused decline in the percentage of sperm motility, viability and sperm concentration as well as an increase in the percentage of sperm abnormalities (31).

Generally, the intraperitoneal injection of Cu-NPs could affect immature sperm cells in the testes and mature one that stocked in the epididymis. The reduction in the number of sperm viability, and the elevation of sperm concentration and abnormalities in the testis and epididymis in this study, depending on concentration and time of exposure. However, Gromadzka-Ostrowska et al. (24) found that a dose-dependent (5 and 10 mg/kg body mass) and time-dependent (24 hrs, 7 and 28 days) decline the sperm count in the rat epididymis, and elevation in number of dead sperm after intravenously administrated with Ag-NPs.

CONCLUSION

The present obtained results demonstrated that copper nanoparticles can be considered as a reproductive toxicant by diminish the number of sperm parameter indices in addition to decline the body weight, which led to a negative effects on the activity of male reproductive system. The effects of nanocopper on reproductive indices are concentration and time-dependent. Further research is needed to clarify exposure by measuring the hormone levels and pathological changes.
RECOMMENDATION

The researchers recommend to study the effect of Cu-NPs on female reproductive system and embryonic development in addition to investigate the effect of these materials on immuno-histo system.

REFERENCES

13. Kim YS.; Song MY.; Park JD.; Song KS.; Ryu HR.; Chung YH.; Chang HK.; Lee JH.; Oh KH.; Kelman BJ.; Hwang IK. and Yu IJ. (2010). Sub chronic oral toxicity of silver nanoparticles. Part Fibre Toxicol. 7:20–23.
Effect of combination of 635 nm Red laser and infrared 810 nm light on wound healing infected by *Acinetobacter Baumannii*

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**ABSTRACT**

Infection is the most important factor for delayed wound healing. Many therapeutic techniques are used for enhancing infected wound healing including the use of two different laser sources. Each wavelength has different biological effect on healing process. An elliptical full thickness skin wound was created aseptically on a back of 45 adult’s female BALB/C mice. The wounds were infected with *Acinetobacter Baumannii*, and the mice were randomly divided into two groups, first group (non-irradiated, controls), 15 animals (live in each subgroup) divided according to days of irradiation. Infected (irradiated groups) randomly divided into two principle groups, first set exposed for 5 minutes, the second set exposed to 15 minutes. Animals killed on day 3, 5 and 10 after contamination. Laser phototherapy was carried out with a diode [810nm, power=57 mw, continuous wave laser, power density 11.4 mw/cm²], [635 nm, power = 48.5 mw, continuous wave laser, power density 9.4 mw/cm²], the output of each laser fitted with a beam expander to irradiate a circular area of diameter (2.5) cm (area = 5 cm²). Phototherapy started directly after surgery and repeated for 3, 5 and 10 days. Histological analysis showed that control subjects had lower granulation tissue compared to irradiated subjects. Irradiated subjects also had a lower amount of inflammation compared to controls. Irradiated subjects also showed a more intense re-epithelialization. The granulation tissues were mostly mature in these subjects at the end of the experimental time especially when both wavelengths were used. The present study showed that cold laser therapy has a certain effect on healing of infected wound, especially with association of 810nm+635 nm.

**Keywords:** infected wound, combination of two wavelengths of laser, *Acinetobacter Baumannii*

**الملخص باللغة العربية**

يشكل استعمال منظومة ليزرية مؤلفة من طولين موجيين مختلفين إحدى الطرق الفعالة في علاج الجروح الملوثة لما لها من تأثير كبير في تحسين عملية الالتمام. في هذه الدراسة، تم إجراء التجربة على 45 فأرًا تم إحداث جرح في منطقة ظهر أدمها وتلوثه بالبكتيريا (Acinetobacter Baumannii) الأحمر 635 نانومتر + الأشعة تحت الحمراء 810 نانومتر لفترات زمنية مختلفة (5، 15 دقيقة) لمدة (3، 5، 10 أيام) وتم مقارنتهما بمجموعة السيطرة غير المعروضة للليزر. تم استخدام تقنيات التصوير والتعليم المكاني لالتمام الجرح في استجابة النتائج. حيث ثبت أن الأطوال الموجية المستعملة كان لها تأثير فعال في تحسين االتمام الجروح الملوثة، خاصة باستخدام المنظومة (الأحمر+الأشعة تحت الحمراء).
INTRODUCTION

The skin is the largest body organ and essentially composed of epidermis and dermis. It has five vital functions: protection, sensation, thermoregulation, excretory function, and metabolism (1). Wound healing is a complex biological process, which results in the restoration of tissue integrity. Physiologically, it can be broken down into four distinct phases: homeostasis, inflammation, proliferation and tissue remodeling (2).

Skin flora originate from varying parts of body, but generally do not cause disease. Despite they are not pathogenic in their region, various microorganisms may promote pathogenic ones as a result of discrepancy of the local circumstances and body immunity, and a possibility of complication in particular surgical procedure arises (3). Wound infection is the principle familiar complication in healing wound and preceding both vascular and cellular reaction in tissue. The healing of infected tissue take place after the clarification of infection process and eliminates necrotic tissue (3,4).

Previous studies approved the benefits of cold laser therapy (CLT) at some wavelengths in improving tissue repair by accelerating cell division, increasing phagocytic function of leucocytes, increasing granulation tissue formation and collagen deposition (5). Cold laser therapy stays a questionable for non-healing wounds. Wound healing, infection is essential point. Patients with infected wounds have a risk of developing bacteremia, which have lethal consequences and healthcare cost.

There are sporadic literatures on the effect of CLT on healing of wounds that colonized by pathogenic bacteria, such as S.aureus, A.Baumannii and Pseudomonas aeruginosa (6). Acinetobacter Baumannii is a gram-negative bacilli that had been emerged in recent years as leading cause of nosocomial infections associated with elevated morbidity and mortality, A. Baumannii treatment became a problem in recent years due to its increasing antibiotic resistance strains, so many studies were conducted to discover new modalities of treatment (7).

A combination of two wavelengths with different absorption manners may accelerate wound healing. In spite of various studies and researches that referred to the use of cold lasers in the treatment of wounds, few of these researches examined the effect of laser on the infected wound, as well as few of them applied the usage of combination of two wave lengths at the same time (5). Thus, the present study aimed to evaluate the benefit of combination of two wavelengths of cold laser by using beam combiner (an optical system) in treating wounds infected with A. Baumannii mice.

MATERIALS AND METHODS

Forty-five (45) females of BALB/c mice weighing (18-32) gram were enrolled in the study. Animals were kept in individual plastic cage in hygiene conditions with wood chip bedding and maintained at 22°C in day/night light cycle and fed with standard pelleted laboratory diet and had water libidum.

Irradiation procedure

All equipments were calibrated prior to the study to make sure they delivered an accurate dose during the study protocol. The method of irradiation was standardized before experiment. Low energy continuous wave portable 635nm and 810 nm from (Laser Scientific Ltd, UK) was used in all experiments of irradiation. The output power was measured using a laser power meter (SOLO PE Genetc -EoInc, Canada). Cold laser therapy was started immediately after surgery and repeated at 3,5,10 days. This protocol was chosen because the conventional clinical approach to laser therapy for wounds, were 3-5 exposures per week in 24-hours interval (6, 8-10).

Beam combiner

Components of beam combiner used in this study: beam splitter (is a common optical component that partially transmitted and partially reflects an incident light beam, usually in equal properties and its designed for well defined dividing ratio (r:t) reflectance=transmission for visible light source of light (laser No.1, 635nm, laser No.2, 810nm). Two beam expanders, plane mirror at 45°C (figure 1).

Figure (1): Beam Combiner System

Treatment parameters for beam combiner

Area of spot size at wound surface, (area=5cm²), Irradiance for 810 nm is equal to 11.4mW/cm², Irradiance for 635 nm = 9.4 mW/cm². Dose equals to 3.4J/cm² for 810 nm at 5 minutes. Dose equals to 3J/cm² for 635 nm at 5 minutes. Total doses for 810 nm and 635 nm for 5 minutes equal to 6.4J/cm². Dose equals to 10.26 J/cm² for 810 nm at 15 minutes, and dose equals to 9 J/cm² for 635 nm for 15 minutes is equal to 19.26 J/cm².
Wound model

On day zero, the day of wounding and inoculation, mice were anaesthetized with injection of ketamine at 130mg/kg and xylazine at 10mg/kg was given via injection for pain management. Hair was clipped from the cervical to mid-lumber dorsum. The operative site was prepared aseptically with alcohol 70% and an elliptic full thickness skin wound was created aseptically with scalpel in all mice on the shaved back of the animal skin defect overlying the thoracic spinal column and adjacent musculature (8, 9, 11, 12). Each wound was measured approximately (1.4-2.0 cm). The wound was left uncovered during whole period of experiments.

Percentage of wound closure

At 3, 5, and 10 days after wounding, the area of wounds of all mice were recorded. The wound area of all mice was measured at regular intervals with a caliper. The wound area for all ellipses was calculated as follows (12):

\[ \text{Area} = \frac{L/W}{2} \times \pi \times (cm)^2 \]

Where L and W are the length and width respectively (1-14).

Percentage of wound closure was calculated using the following Formula (13):

\[ \text{Percentage of wound closure} = \left( \frac{\text{Area of 1 day} - \text{Area of x days}}{\text{Area of 1 day}} \right) \times 100\% \]

Bacterial strain and inoculation preparation

Swab samples were taken from wound areas of patients whose wounds were infected with Acinetobacter Baumannii and were suspected (using sterile disposable cotton swabs in transport media). These samples were collected from patients hospitalized at Al-Yarmook teaching hospital in the Baghdad. One isolate of Acinetobacter Baumannii was selected according to the resistance test to several antibiotics. A. Baumannii was identified by using microscopic, cultural characteristics, biochemical test and API system (14,15).

Immediately after the creation of wound, a bacterial suspension containing 10s cells in 50 μl sterile normal saline was inoculated on the surface of each wound with a pipette tip and then was smeared on to the wound surface with a sterile disposable cotton swab (14).

Study design

Prior to surgery, an inoculation of the wound with bacterial suspension was done. Animals were divided into two principle groups, irradiated and infected group (30 animals) and control non-irradiated group (15 animals). Wounds were irradiated for 3, 5 and 10 days beginning on day one immediately post inoculation. This regimen was chosen because the common clinical approach to laser therapy for wounds is three and five exposures approximately (1.4-2.0 cm). The wound was left uncovered during whole period of experiments.

Histopathological evaluation

At 3, 5, and 10 days after wounding, five mice were selected from each group randomly and killed by ether inhalation. The tissue specimens were stained with hematoxylin and eosin and examined with a semi-quantitative method (16). Histopathological parameters: polymorphnuclear leucocytes (PMNL), re-epithelialization, fibroblasts, angiogenesis, granulation tissue formation and collagen fibers deposition were evaluated. The sections were examined by histopathologist's experts and assessed on a scale of 0-3 (6, 10).

Sections were graded for wound healing according to seven parameters related to acute inflammatory response and repair: polymorphnuclear leucocytes, granulation tissue, fibroblasts, collagen deposition, and evidence of epithelialization. Each feature was semi quantitatively evaluated (from 0=absent or no evidence, to 3= prominent =marked) based on well-defined and reproducible histological feature as described by (17).

Statistical analysis

Statistical analysis and reporting of obtained data were carried out by using the computerized database structure; statistical package for social science (SPSS V.20, computer software was used for this purpose). Frequency distribution was done for the study variables. Data were reported and presented as mean ±SD and or (95% confidence interval) for the normally distributed variables. The bootstrapping was done for small groups to the 1000 sample size and the statistical significance of difference between mean of a normally distributed continuous parametric variables of two groups was assessed using the independent samples students t-test; and the Analysis of variance (ANOVA) were used to compare continuous parametric variables between more than two groups. Statistical tests were
approved by assuming a null hypothesis of no difference between mean of variable, a P value ≤0.05 and ≤0.005 was considered statistically significant. Histopathological parameters were compared via Chi-square test.

**RESULTS**

Wounds infected with *Acinetobacter Baumannii* revealed inflammation sequences after 48 hrs.

**Percentage of wound closure**

No significant differences were shown for wounds closure between contaminated groups exposed to 6.4J/cm², (27.497±11.058), and controls (20.011±7.037) at day 3 after treatment.

While significant difference was observed for wound closure between contaminated group exposed to (19.26 J/cm²) (52.887±10.941) and controls (20.011±7.037) (P=0.025) (table 2).

Significant differences were observed for wound closure between contaminated groups exposed to (6.4J/cm²) (47.191±8.805), and controls (34.751±5.852) at day 5 after treatment, while the group exposed to (19.26/cm²) showed high significant difference for wound closure between contaminated group (62.537±6.706) and controls (34.751±5.852) (P=0.001) (table 2).

At day 10 after treatment, contaminated groups exposed to (19.26 J/cm²) showed very high significant difference for wound closure between contaminated groups exposed to (6.4 J/cm²) (58.435±10.789) and controls (48.256±18.0) (p=0.0085) (table 2).

**Histological observations**

Healing process characterized by incomplete epithelization and presence of collagen and granulation tissue with absent of necrosis were observed at irradiated infected groups on day 3, after the exposure for 15 minutes (figure 2).

Progressive healing was seen in the infected wound exposed to 15 minutes on day 5 with complete epithelialization, granulation and enhance collagen deposition compared to no healing seen in control group (no epithelialization, no granulation tissue formation and no collagen deposition) (figure 3).

A complete healing with complete re-epithelialization mature granulation formation and collagen fibers were seen in the infected wound exposed to 15 minutes on day 10 compared to incomplete healing of control group (figure 4).

<table>
<thead>
<tr>
<th>Day of examination (n= 10)</th>
<th>Duration of irradiation (min)</th>
<th>Contaminated wound (m±SD)</th>
<th>Control (m±SD)</th>
<th>Calculated t-test(P-value)</th>
<th>ANOVA* F value (P-value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 3 (n= 10)</td>
<td>5 (n= 5)</td>
<td>27.497±11.058</td>
<td>20.011±7.037</td>
<td>1.28 (0.24)</td>
<td>15.28 (0.005)***</td>
</tr>
<tr>
<td></td>
<td>15 (n= 5)</td>
<td>52.887±10.941</td>
<td>47.191±8.805</td>
<td>-1.41 (0.20)</td>
<td>6.712</td>
</tr>
<tr>
<td>Day 5 (n= 10)</td>
<td>5 (n= 5)</td>
<td>58.435±10.789</td>
<td>48.256±18.000</td>
<td>3.47 (0.0085)**</td>
<td>22.17</td>
</tr>
<tr>
<td></td>
<td>15 (n= 5)</td>
<td>79.064±2.865</td>
<td>6.58 (0.0002)**</td>
<td>(0.0001)*****</td>
<td></td>
</tr>
</tbody>
</table>

*Bootstraping was done for the independent samples up to the sample size 1000.
**=t-test; statistically significant at level of significance of 0.05, 0.005.
***= one way ANOVA; statistically significant at level of significance of 0.05, 0.005
**DISCUSSION**

Cold laser therapy (CLT) is a type of phototherapy, which concerns the application of CLT light in the red or near infrared wavelengths to treat different diseases and injuries. The light is regularly monochromatic and coherent and produces by lasers and used to enhance healing, regeneration. Increase the rate quality and tensile strength of healing tissue (18,19).

The present study aimed to investigate the effects of using two wavelengths CLT on healing processes of wound infected with *Acinetobacter Baumannii* Bacteria. Two fundamental considerations were made from the outset of study: a cutaneous wound model was selected based on findings of a previous studies, where an open wound did not negatively affect animal health during more than 15 days protocol (20). Many positive effects of CLT have been recorded in repair of cutaneous wounds.

Both experimental and human researches has shown enhanced collagen synthesis, these being a property to increased ATP (Adenosine triphosphate) production and enhanced cell metabolism and fibroblast reproduction or spreading positive significant wound healing seen in the irradiated infected groups expressed by decrease inflammation and increase granulation tissue formation, collagen deposition compared to non-irradiated control group, this may be due to the effect of combination of two wavelengths laser (red + infrared) which leads to increase ATP (Adenosine triphosphate)
production and cell metabolism and fibroblast proliferation. For infected wounds in rats estimated by Histopathological and immune histochemical, Ferreira et al. noticed a huge amount of macrophages in the lesions treated with laser 632.8 nm. This explained that laser therapy augment the phagocytic capacity of macrophages, assisting cleaning of the wound Ferreira study (21).

This result is in agreement of same finding noticed by (22,23).

A complete distinguishing of the effect of light on healing is remaining far away, and the mechanism of stimulation of specific cells such as fibroblast, remain unknown. For this reason of knowledge demonstrates why it is so hard to explain effective protocols for the treatment of various wound types of in compromised subjects. Another important parameter in this study was the irradiance (energy density). Our results showed that on day 5, the infected group showed a positive effect of complete healing of the wound at (19.26 J/cm²) expressed by complete re-epithelialization, granulation tissue formation, increase collagen deposition and decreased inflammation which goes with the wound closure percentage and this finding suggest that the beneficial effect may be due to the direct effect of laser on host tissue (10). Using a combination of (Red 635nm+IR 810nm) cold laser, we achieved a best result in wound healing with dose 19.26 J/cm² at 15 minutes for 5 and 10 days where complete healing of wound was seen compared to incomplete healing shown by control. Using optical system in this study is to interfere two wavelengths (red and 810nm) to reduce illuminated time. It was noticed that if an animal’s model exposed to laser No 1, for 5 minutes and then to laser No.2 for another 5 minutes, the summation of time exposure will be 10 minutes. While using (optical system) (beam combiner it reduce the time of exposure. (Two types of laser at the same time) i.e. 5 minutes only.

The results showed that both doses have the same noticeable effect on wound healing but the effect is more clear at (19.26J/cm²) and this in accordance with the result of Castano study (24), who noticed that when irradiance is constant, the biological effect of laser needs enough time and this effect depends on the total dose (absorbed photon) more than on intensity of laser (irradiance) and this incident called (Importance of irradiated time). The principle advantage of beam combiner is to use certain illuminated time, because illuminated or irradiation time is the most important factor affecting photo biological response (24,25).

There has been many projects suggested that the photons absorbed by cytochrome C oxidase, which leads to dissociation of the inhibitory molecule, nitricoxide, from oxygen binding sites within the enzyme, thus elevating the enzyme activity in cellular respiration. The primary cellular effect of CLT is to elevate the activity of enzyme; it makes sense to suppose that this increased activity should take for a sufficiently enough time to have real effect on cellular metabolism.

Wavelengths are an important and essential parameter of CLT because it interacts with certain molecules (26,27). It is accepted that red light affect directly on mitochondria, infrared light interact with cell membrane and each type of light after specific therapeutic actions. This indicates that the use of associated wavelengths may further improve the outcome treatment (26, 2, 29).

REFERENCES

The effect of silver nanoparticles prepared with *Lawsonia inermis* extract on some multiple antibiotics resistant bacteria

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**ABSTRACT**

The present study was conducted to investigate the effect of silver nanoparticles that were prepared by using the *Lawsonia inermis* extract against two multiple antibiotics resistant Gram negative pathogenic bacteria (*E. coli* and *Proteus mirabilis*) and one gram positive pathogenic bacteria (*Staphylococcus aureus*).

Results showed that the observed color of the mixed solution of extract and 1 mM silver nitrate changed to dark brown. The solution is considered to be an evidence of the nanoparticles formation, where the UV-VIS spectrophotometer showed the absorption peak 400 nm. However, the study of XRD diffraction showed peaks (111, 200, 220) and the antibacterial activity of the particles prepared showed the antibacterial activity against tested species.

**Keywords**: silver nanoparticles, *Lawsonia inermis*, bacteria

**الملخص باللغة العربية**

هـذـا الـدـرـاـسـة تـعـدـد تأثير جسيمات القـلـة النـائوـية الحـبـرة باستـخدام مـستـخلـص نـبات الـذنـب، وـبـكـتـيـريـا الـمـرـضـة الـعـالـية تـمـثـل~(E. coli) وـأـنـوـاع البكتـيرـيـا المـرـضـة البـكـريـة (Proteus mirabilis) وأنـواع البكتـيرـيـا البكـريـة (Staphylococcus aureus) المـتـضـدادـة للـثـقـلات الحـيويـة (1Mm).

أظهرت النتائج تغيير نـزل مـحياة القـلـة بـتركيز (1Mm) ونـبـت الـذنـب إـلـى الـثـقـلات الحـيويـة، حيثـ قـلـة النـائوـية حـبـرة UV-VIS وـأـظهـر طـيف الأـشـعـة بـمـعالـة العـائـة (400 nm). وأظهـر طـيف الأـشـعـة تحت الحـمراء (220), وأظهـر طـيف الأـشـعـة تحت الحـمراء (220) وأظهـر طـيف الأـشـعـة تحت الحـمراء (220). من ناحية أخرى، تبين أن هذه الجسيمات تأثيرها تبليطًا على أنواع البكتيريا المرضية ذات المقاومة المطلوبة للمضادات الحيوية المستخدمة.
INTRODUCTION

The resistance of bacteria to antibiotics is a clinical problem, which relies on many medicinal plants as an alternative to antibiotics (1-3). Henna plant is a small tree and scientifically called Lawsonia inermis. It can be grown in many tropical and nontropical areas. It is used mainly and commonly in cosmetic industry.

Henna Plant contains special components in its leaves such as several biologically active substances that act as analgesic as well as it can be used to reduce body temperature, improve the color and health of the hair. In addition, it has been studied in cancer cases and to inhibit the growth of bacteria, parasites and viruses (3,4).

Recently, nanoparticles field has got a lot of attention of scientists in the nanotechnology science to reveal high activities of materials (1). The nanoparticles of noble metals such as gold, silver, zinc and platinum has been used in many products, which are in contact with the human body like as detergents, cosmetics and toothpaste. In addition to the studies of pharmaceutical and medical uses for an antibacterial, anticancer and antipyretic (5-9).

The preparation of nanoparticles process depends on the biological methods, which are more significant than the physical and chemical processes because of the cost, low exhausting of energy and heat and non-toxic, which is considered as environmentally friendly (10-12).

Many studies had pointed out the use of plant extracts as a reductant in preparation of silver nanoparticles, such as the use of Aloe vera plant, which showed inhibitory effect on the E.coli and Staphylococcus aureus (13), where the silver nanoparticles was prepared by using the pomegranate peel extract as a reducing agent, which had inhibitory effect on E.coli and Pseudomonas aeruginosa and Klebsiella pneumoniae (14).

In addition, other plants such as Artemisia pallens (15), Murrayako enigii and Zea mays (16) and bananas in addition to lemon, black pepper and Myrtus communis had been used as reducing agents for silver nanoparticle preparation (17-19).

In addition, other plants such as Artemisia pallens (15), Murrayako enigii and Zea mays (16) and bananas in addition to lemon, black pepper and Myrtus communis had been used as reducing agents for silver nanoparticle preparation (17-19).

Because of bacterial resistance to antibiotics as well as the evolution of multiple antibiotics resistance, many plants have been adopted as alternative sources to antibiotics. Therefore, this study aimed to use the henna powder as a reducing agent in the preparation of silver nanoparticles and to study its influence on a number of human pathogenic bacteria.

MATERIALS AND METHODS

Samples

Clinical samples were collected from different hospitals in Kirkuk city. The samples were planted in bacteriological culture media and then the grown isolates were identified depending on the reliable diagnosis sources (20-22). The diagnosis was confirmed by using API20E system and API staph system.

Plant extract preparation

Henna powder was obtained from local markets. A weight of 2 g of powder was dissolved in 50 ml of sterile distilled water. The solution was left for three hrs. on a magnetic motor, then left for one hr. before the filtration with Whatman No.1. filter paper (1).

Antibiotic sensitivity

To study the sensitivity of the bacteria to a number of antibiotics, disc diffusion method was adopted, where the results were compared with standard diameters of inhibition (23, 24).

Preparation of silver nanoparticles

10 ml of Henna extract were added to 90 ml of 1mM silver nitrate solution. To obtain reduction process, the solution was left at room temperature until observing the color change. The process was carried out in the dark to avoid photo oxidation (25).

Silver nanoparticles characterization

According to (25), UV-VIS- Spectrophotometer device (JASCO) was used to detect the absorption spectrum of the particles. The size of particles were determined by using the x-ray diffraction ((XRD) device type (Shimadzu). In addition, the scanning electron (TSCCAN - VEGA) was used.

Antibacterial activity

Well- Diffusion method in Mueller –Hinton agar was followed to study the inhibitory effects of the particles on the growth of the number of humans pathogenic bacteria. The bacteria was characterized as a multiple-antibiotics resistance. The prepared media was poured in the dishes to be harden. The bacterial inoculum was prepared and compared to standard MacFarland tube (1 * 10⁸) cfu; Muller Hinton dishes were inoculated using cotton swabs. Using the cork borer, a number of wells were developed in each plate (6 mm) diameter. The holes were individually fuelled with 60 ul of Henna extract, solution of silver nitrate 1mM and silver nanoparticles. Finally, the dishes were incubated at 37 ºC for 24 hrs. (26).
RESULTS AND DISCUSSION

Three species of gram positive and gram negative bacteria were selected (three isolates of each species), and characterized according to their resistance to the largest numbers of antibiotics among bacterial isolates obtained. The species were found to be resistant to five beta-lactam antibiotics in addition to other types of antibiotics as shown in table (1).

<table>
<thead>
<tr>
<th>Isolates</th>
<th>AMC</th>
<th>AX</th>
<th>AM</th>
<th>IP</th>
<th>CAZ</th>
<th>CRO</th>
<th>CTX</th>
<th>GM</th>
<th>AK</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>E. coli</em></td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
</tr>
<tr>
<td><em>Staph.aureus</em></td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
</tr>
<tr>
<td><em>P.mirabilis</em></td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>S</td>
</tr>
</tbody>
</table>

Table (1): Antibiotics resistant of tested bacteria

The reason of beta-lactam antibiotics resistance may be due to production of beta-lactamase enzymes especially for resistance to B-lactam antibiotics or because of other resistance mechanisms. The mechanisms are either naturally depend on the genotype or acquired (27).

Figure (1) shows the first step in the preparation of silver nanoparticles as well as the process of changing the color of a mixture of Henna extract solution and 1mM silver nitrate to dark brown, where the obtained results were same as in (28-30).

![Figure (1): Synthesis of silver nanoparticles](image)

The formation of silver nanoparticles was confirmed by studying the UV/VIS absorption spectrum as a second step, which was 400 nm as shown in figure (2) that are consistent with (31).

![Figure (2): UV-visible absorption spectra obtained for silver nanoparticles synthesized by *Lawsonia inermis*](image)

Figure (3) reflected the third step of confirmation of silver nanoparticles formation. The study of X-ray diffraction showed the presence of three peaks of the diffraction rays (111, 200 and 220) which were considered to (34.3, 41.5 and 63.25)0 respectively. The result was closed to Joint Committee on Powder diffraction Standards (JCPDS). The particle sizes were (5 nm to 111 and 8 nm to 200 and 7nm 220 nm) peaks using sherrer equation (32).

\[
D = \frac{0.9\lambda}{\beta \cos \theta}
\]

Where,
\(\lambda\) = wave of X-ray (0.1541 nm)
\(\beta\) = (full width at half maximum)
\(\theta\) = Diffraction angle
\(D\) = particles diameter

![Figure (3): the XRD pattern of the silver nanoparticles formed by *Lawsonia inermis*](image)

The average particle size was 6.6nm, where is closed to (33), who indicated that the particle sizes were (5.3, 8.2 and 5.1) nm to the peakes (111, 200 and 220) respectively. The results of the present study also were in agreement with (34), where the particle size was 7.40 nm. Figure (4) showed the accumulation of silver nanoparticles using a scanning electron microscope in different magnification.

Through the study of the effect of silver nanoparticles on some pathogenic bacterial isolates, the inhibition of the growth reflects the effectiveness of silver nanoparticles on selected pathogenic bacteria which are characterized as multiple - antibiotic resistance as shown in table (2). According to the results, the higher inhibition zone is 32 mm for *Staph. aureus* using absolute concentrations of silver nanoparticles while in concentration 35%, the lowest inhibition zone is 8 mm for the isolates (No. 2 and 3). The highest zone of inhibition of *P.miribilis* growth is 34 mm for the isolates (No. 2 and 3) in absolute concentration while the lowest zone was 5 mm for the same isolates in concentration 35%. The largest diameter of inhibition was 35 mm for isolate (No. 2) of *E.coli* in the absolute concentration while lowest zone for the isolates (No. 2 and 3) was 8 mm in the concentration 35%.
Figure (4): Scanning electron micrograph of silver nanoparticles

The selected isolates were characterized as a multiple-antibiotic resistance. In the other hand, there was little inhibition against three isolates in concentration 25%. However, more future studies needed to improve production or combination with antibiotics to promote the antibiotics activity. The effectiveness of the prepared particles were tested where it was more than the effect of Henna extract alone and silver nitrate 1mM. Additionally, the results showed that the effect of the particles prepared with henna extract was more than the effect of particles prepared using a *Clitoria ternatea* extract on same types of bacteria as the inhibition zone which was not exceed more than 17 mm (35).

Table (2): Antibacterial activity of silver nanoparticles resistant of tested bacteria

<table>
<thead>
<tr>
<th>Concentration of silver nanoparticles %</th>
<th>Staph. aureus</th>
<th>Proteus mirabilis</th>
<th>E.coli</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Zone of inhibition for isolate No.1(mm)</td>
<td>Zone of inhibition for isolate No.2(mm)</td>
<td>Zone of inhibition for isolate No.3(mm)</td>
</tr>
<tr>
<td>100</td>
<td>30</td>
<td>32</td>
<td>30</td>
</tr>
<tr>
<td>50</td>
<td>21</td>
<td>18</td>
<td>17</td>
</tr>
<tr>
<td>35</td>
<td>9</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>25</td>
<td>0</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Henna extract</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>AgNO₃</td>
<td>15</td>
<td>15</td>
<td>15</td>
</tr>
<tr>
<td>Concentration of silver nanoparticles %</td>
<td>Zone of inhibition for isolate No.1(mm)</td>
<td>Zone of inhibition for isolate No.2(mm)</td>
<td>Zone of inhibition for isolate No.3(mm)</td>
</tr>
<tr>
<td>100</td>
<td>32</td>
<td>34</td>
<td>34</td>
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<tr>
<td>50</td>
<td>20</td>
<td>19</td>
<td>17</td>
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<tr>
<td>35</td>
<td>7</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>25</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Henna extract</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>AgNO₃</td>
<td>15</td>
<td>15</td>
<td>15</td>
</tr>
<tr>
<td>Concentration of silver nanoparticles %</td>
<td>Zone of inhibition for isolate No.1(mm)</td>
<td>Zone of inhibition for isolate No.2(mm)</td>
<td>Zone of inhibition for isolate No.3(mm)</td>
</tr>
<tr>
<td>100</td>
<td>34</td>
<td>35</td>
<td>34</td>
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<td>50</td>
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<td>23</td>
<td>24</td>
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<tr>
<td>25</td>
<td>1</td>
<td>1</td>
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<td>Henna extract</td>
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<td>20</td>
</tr>
<tr>
<td>AgNO₃</td>
<td>16</td>
<td>16</td>
<td>16</td>
</tr>
</tbody>
</table>
CONCLUSION
When *Lawsonia inermis* extracts were mixed with [1mM] AgNO₃ solution, the color of the reaction medium had changed to dark brown, which indicated the formation of the nanoparticles formation, where the UV-VIS spectrophotometer showed the absorption at peak 400 nm and XRD showed peaks at (11, 20, 220). However, the study of antibacterial activity of the particles formed appeared to have the inhibitory activity against multiple – antibiotics resistant bacteria (*E. coli, Staphylococcus aureus, proteus mirabilis*).

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REFERENCES
25. Kumar KS. and Kathireswari P. (2016). Biological synthesis of Silver nanoparticles (Ag-


Evaluation of her/2-neu gene status using FISH/CISH techniques in Iraqi breast carcinoma patients

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ABSTRACT

The present study aimed to examine the concordance between FISH/CISH techniques for assessment of amplification of her2neu gene in Iraqi breast carcinoma patients. Seventy four (74) Iraqi breast cancer patients were involved at the study from the Histopathology Department at the Central Public Health Laboratory in Baghdad, Iraq. Amplification of HER2/neu was detected in (33.8%) by fluorescence in situ hybridization and (13.51%) showed high amplification by chromogenic in situ hybridization and (32.43%) showed low amplification. The results of chromogenic in situ hybridization were significantly correlated with the results of two-color fluorescence in situ hybridization with the same tumors. In addition, the study involved the correlation between HER2/neu level, progesterone (PR) and estrogen levels (ER).

Keywords: her2neu gene, FISH/CISH techniques, progesterone (PR), estrogen (ER).

الملخص باللغة العربية

سعت هذه الدراسة إلى اختبار العلاقة بين طريقة التهجين الورمي وطريقة التهجين الموضعي النووي لقياس التعبير الجيني للجين her2neu لتحديد مدى تأثيره على النتائج المثبتة بفموس في سرطان الثدي لدى النساء من النساء العراقيات المصابات بسرطان الثدي، حيث تم جمع أربع وسبعين (74) عنة من النساء في مستشفى البحرين في مدينة بغداد، العراق. أظهرت النتائج أن التصاعف الجيني الموضعي الموسمي كان بنسبة 33.8%، فيما كانت النسبة بطريقة التهجين الورمي الموسمي النموذجي 32.43% للتصاعف المنخفض، و51.31% للتصاعف العالي. كما انتشرت النتائج توافقيًا قويًا بين الطريقتين. وقد سعت الدراسة أيضاً إلى اختبار علاقة هرموني البروجسترون والإستروجين بالجين her 2 neu
INTRODUCTION

Breast cancer is considered one of the most common types of cancer worldwide. The incidence rate of this type of cancer was elevated in the last few years in Iraq. In 2011, The Iraqi Cancer Registry reported that the breast cancer was the most frequent type of cancer among women and accounted for approximately one-third of the registered female cancer (1). Her-2/neu is a proto-oncogene located on chromosome 17q21 and encodes an 185-kD trans membrane glycoprotein. Her-2/neu belongs to the human epidermal growth factor receptor family that plays an important role in the regulation of cell growth, differentiation and survival. Her-2/neu gene was found to be over expressed, amplified or both in different human cancers, including breast cancer. Amplification of this gene was recorded to be occurred in 10-34% of breast carcinomas (2). The studied hormonal receptors in breast cancer were estrogen receptor (ER) and progesterone receptors (PR). If the breast cancers are classified by positive immunohistochemistry (IHC) expression of ER and PR, then, they have different pathological, and molecular characteristics (3). It could be explained that risk factors are closely associated with breast tumors ER+ and PR+, while etiology of breast cancer ER− expression and PR− should be independent of hormone exposure (4).

Fluorescence in situ hybridization (FISH)

It is a significant technique to study chromosomal abnormality in tumor cells. The technique reached high level of detection sensitivity (individual genes can be detected), and high multiplicity (i.e. in the same nucleus several probes can be applied) (5). This delicate process consists of hybridizing a DNA probe to its complementary sequence on chromosomal preparations. Probes commonly are labeled by two ways directly, by incorporation of fluorescent nucleotides, or indirectly, by incorporate reporter molecules to the probe, which subsequently is detected by fluorescent antibodies. Finally, probes and the targets DNA sequences are observed in situ by fluorescent microscope (3,6).

Chromogenic in situ hybridization (CISH)

Chromogenic in situ hybridization (CISH) technique was introduced in last few years in which the DNA probe was detected by an immune peroxidase reaction (7). This method is similar to FISH, but it does not require the fluorescense microscopy for final examination. The bright field microscope is being used for evaluation of slides instead of fluorescent microscope slides, which provides more morphological features that can be observed and archived in contrast to fish slide, which cannot be archived. Although two dual color protocol for CISH were found, this technique was basically done using one color. The most significant advantage of the two colors is gene amplification distinguished from chromosomal aneuploidy by a reference probe, which is more accurate, faster and easier process (8).

MATERIALS AND METHODS

Patients

Seventy-four (74) Iraqi breast cancer patients who had lumpectomy with lymphnode clearance or total mastectomy were involved at the study from the archive of the center health laboratory, histopathology department, Bagdad, Iraq. Most of these cases included invasive ductal and invasive lobular carcinoma type.

Scoring of fluorescent in situ hybridization signal

Fluorescent in situ hybridization was performed with a florescent method for molecular marker (Her2/neuDNA) using kretch digestion kits and probe (POSEIDON-KREATECH, Netherland). The Her2/neu (17q12) specific delegate DNA probe was optimized to detect copy numbers of the Her2/neu gene region at region 17q12 within the cell. To facilitate her-2/neu identification, chromosome 17 Satellite (SE) was included as internal control. The probe was direct-labeled with platinum bright 495 (green region). The Her2/neu (17q12) specific DNA probe was designed as dual-color assay to chromosome 17 and amplification involving the Her2/neu gene region at (17q12), which shall show several red signals, while the chromosome 17 (internal control) centromere region shall provide 2 green signals. Otherwise, two red signals (R) and two green (G) signals will consider normal chromosomes 17(2 R2G).

Quantification of her-2/neu signals fluorescent in situ hybridization was detected under fluorescence microscopy at power 40x and 100x, and different filters (FITC, TRITC TAXSAS RED) for the counting of positive cells were carried out at oil immersion (100x.).

Ratio of her-2/CEP17 shall be calculated and if the ratio is equal or more than two, it was considered amplified, and there is no amplification if the ratio less than two was obtained for tumor cells between 20 to 60 cells, based on kits used (POSEIDON-KREATECH, Netherland).

Scoring of chromogenic in situ hybridization

CISH method is depends on a simple bonding reaction of a specific DNA probe to an enzymatic indicator for create chromogenic reaction. The presence of nucleic acid sequence in cells can be detected with in situ hybridization using labeled DNA probes. The zyto Dot SPEC her-2/neu probe, consist of digoxigenin labeled polynucleotide which target sequence of the her -2/neu, and the
duplex formation visualized using primary anti-digoxigenin antibody, which was detected by secondary enzyme-conjugated antibody. The enzymatic reaction of DAB leads to the formation of strong permanent brown signals that can be visualized by light microscope.

The slides were stained by Zyto dot spec her-2/neu probe and kits and evaluated using an light microscope equipped with 40x dry objectives scanned for best area represented tumor.

Scoring of the signal: normal gene copy number was defined as 2 dots shaped signals per nucleus. And Low-level amplification was consider up to 5 dots signals per nucleus in 50% of counting cancer cells, or when a small gene copy cluster was noticed. High level of Amplification of HER-2 was defined when a large gene copy cluster in 50% of carcinoma cells or 5 signalor more of separate gene copies were seen. Images were captured using a Pixera PVC100C digital camera (Pixera Corp., Los Gatos, CA).zyto Dot SPEC her-2/neu probe and kits.

The scoring protocol for ER, PR

Staining by immunohistochemistry was done as described by (9) using the commercially used ER,PR monoclonal antibodies provided by Dako company with detection kit.

RESULTS AND DISCUSSION

Distribution of sample study according to FISH results

Ratio of her-2/neu gene (red signal) to chromosome CEP17 (green signal) ratio higher than 2.2 was considered as amplification, otherwise a ratio less than 2.2 was reported as non-amplification (and a ratio between 1.8 and 2.2 was taken as an equivocal result and counting additional 20 tumor nuclei were counted for the equivocal cases (10) (table 1 and figure 1). In the present study, seventy four (74) cases were examined for her-2/neu by FISH and (58.1%) showed no amplification, while 33.8 % (25 cases) were amplified for her-2/neu gene. Many studies had confirmed the suggestion that protein over expression without gene amplification could be as result from an increase of chromosome 17 copy number, which lead to increase the number of copies of her-2/neu per nucleus in the absence of “true” amplification of her-2/neu gene (11, 12). ASCO/CAP guidelines stated that at least 8 per cent of equivocal cases on FISH exhibit polysomy of the chromosome 17 (13).

Table 1: Distribution of sample study according to FISH results

<table>
<thead>
<tr>
<th>FISH result</th>
<th>Number</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amplified</td>
<td>25</td>
<td>33.8</td>
</tr>
<tr>
<td>Non-amplified</td>
<td>43</td>
<td>58.1</td>
</tr>
<tr>
<td>Equivocal</td>
<td>6</td>
<td>8.1</td>
</tr>
<tr>
<td>Total</td>
<td>74</td>
<td>100%</td>
</tr>
</tbody>
</table>

Chi-square (χ²) --- 12.574 ** (P<0.01).

Figure 1: AFISH for HER-2/neu gene signals (Orange) amplification (more than 2 copies of gene per cell (x100).b:Polysomy of chromosome 17 (more than 2 copies per cell. (A): HER-2/neu gene signals (Orange) amplification (more than 2 copies of gene per cell. (B): polysomy of chromosome 17 (more than 2 copies per cell

Distribution of sample study according to her-2/neu CISH

Significant difference were recorded in which 13.51% of cases had high amplification, while 32.43% had low amplification and the largest percentage 54.06% were non amplified as in table (2), figure (2). A previous study conducted by (14), the amplification was reported in all cases tested, 39 of tumors were determined as non-amplified, which were similar to the results of the present study, while 9 cases had low amplification and 27 cases had high amplification.
Table (2): Distribution of sample study according to her-2/neu CISH results

<table>
<thead>
<tr>
<th>CISH result</th>
<th>Number</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amplified High.</td>
<td>10</td>
<td>13.51</td>
</tr>
<tr>
<td>Amplified Low.</td>
<td>24</td>
<td>32.43</td>
</tr>
<tr>
<td>Non-amplified</td>
<td>40</td>
<td>54.06</td>
</tr>
<tr>
<td>Total</td>
<td>74</td>
<td>100%</td>
</tr>
</tbody>
</table>

Chi-square ($\chi^2$) --- 9.857 **  

** (P<0.01)

Figure (2): A: Low amplified 3-6 her-2/neu gene copies B: High amplified her-2/neu more than 6 gene copies of her-2/neu by CISH (X100)

Correlation of her-2/neu amplification by FISH with estrogen receptor and progesterone receptors

A significant correlation was found in the results of the present study between hormonal receptor and her-2/neu status (table 3). A significant inverse association was noted between hormonal receptor (ER status) and her-2/neu gene amplification in the current study, and these results were in agreement with those obtained by (15). The reason for this inverse relation between hormone receptor ER and her-2/neu amplification is suggested to be due to complex interactive processes, signaling between ER receptor and other growth factor signaling pathways in breast cancer cells (16). The results showed that 64.3 % of cases of ER positive showed non amplification of her-2/neu, while 26.2 of ER positive cases had her-2/neu amplification. The results obtained from studies (10, 17) showed that 18 % of breast cancer were both ER over expression and her-2/neu amplification, also there were significant but not inverse correlation of her 2/neu with PR receptor. In the present study, 25 cases had her-2/neu amplification 46.1% and were positive for PR and 27.1% were negative. This came in concordance with (18). Several studies had suggested that ER-positive breast cancers with her-2/neu amplification could be less responsive to tamoxifen (19).

Table (3): Association of estrogen receptor and progesterone receptor by IHC with Her-2/neu amplification by FISH

<table>
<thead>
<tr>
<th>Total</th>
<th>Amplified</th>
<th>%</th>
<th>Non-amplified</th>
<th>%</th>
<th>Equivocal</th>
<th>%</th>
<th>Chi-Square</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estrog. receptor Positive</td>
<td>42</td>
<td>11</td>
<td>26.2</td>
<td>27</td>
<td>64.3</td>
<td>4</td>
<td>9.5</td>
</tr>
<tr>
<td>Neg.</td>
<td>32</td>
<td>14</td>
<td>43.75</td>
<td>16</td>
<td>50.0</td>
<td>2</td>
<td>6.25</td>
</tr>
<tr>
<td>Progest. receptor Positive</td>
<td>26</td>
<td>12</td>
<td>46.1</td>
<td>12</td>
<td>46.2</td>
<td>2</td>
<td>7.7</td>
</tr>
<tr>
<td>Neg.</td>
<td>48</td>
<td>13</td>
<td>27.1</td>
<td>51</td>
<td>64.6</td>
<td>4</td>
<td>8.3</td>
</tr>
</tbody>
</table>

** (P<0.01).

Correlation of her-2/neu amplification by CISH with ER,PR

There were negative relations between her-2/neu amplification and ER in our findings. Out of 32 cases were negative for ER (34.38%) and showed low amplification, while (21.87%) had high amplification and (43.75%) of ER negative cases were found non amplified as in the table (4). 48 cases of PR negative 25.00% showed low amplification and 10.42% had high amplification and 64.85% showed no amplification, where these results were in agreement with (20), who showed a significant negative association with both ER and PR positivity.

Comparison between FISH and CISH for her-2/neu amplification

There were significant correlations between FISH and CISH in which out of the 25 FISH amplified cases, 24 cases showed amplification and one showed no amplification by CISH, so the concordance was 96% between the two methods. Out of the 43 FISH non-amplified cases, 38 were non-amplified and 5 were low amplified <5 by CISH. Out of the 6 cases at the range (1.8-2.2) equivocal FISH cases, 3 showed low level of amplification with CISH and the 2 other cases had high amplifications (table 5). Our findings showed complete agreement with (21) who reported that out
of 18 cases showed amplification by FISH, 17 cases showed amplification by CISH and only one case was non amplified. In addition, out of 23 FISH non-amplified her-2/neu cases, 21 were non-amplified and two were amplified by CISH, which were very similar to results obtained by many studies reporting that a concordance between the two methods were in the range of 83%–100% and not necessary 100% (22-24).

Table (4): Correlation of her-2/neu amplification by CISH with ER, PR

<table>
<thead>
<tr>
<th></th>
<th>Total</th>
<th>Low amplified N=24</th>
<th>%</th>
<th>High amplified N=10</th>
<th>%</th>
<th>Non amplified N=40</th>
<th>%</th>
<th>Chi-square</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estrogen receptor</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>42</td>
<td>13</td>
<td>31.00</td>
<td>3</td>
<td>7.1</td>
<td>26</td>
<td>61.9</td>
<td>10.94 **</td>
</tr>
<tr>
<td>Negative</td>
<td>32</td>
<td>11</td>
<td>34.38</td>
<td>7</td>
<td>21.87</td>
<td>14</td>
<td>43.75</td>
<td>8.63 **</td>
</tr>
<tr>
<td>Progesterone receptor</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>26</td>
<td>12</td>
<td>46.2</td>
<td>5</td>
<td>19.2</td>
<td>9</td>
<td>34.6</td>
<td>8.59 **</td>
</tr>
<tr>
<td>Negative</td>
<td>48</td>
<td>12</td>
<td>25.00</td>
<td>5</td>
<td>10.42</td>
<td>31</td>
<td>64.58</td>
<td>10.64 **</td>
</tr>
</tbody>
</table>

** (P<0.01).

A high concordance between the FISH an CISH was reported by (21), and agreed with our results as well as with those obtained by (25,26) that showed high concordance between the two methods. A recent study had considered CISH as a reasonable alternative for FISH (7). This minor difference in the result of the two methods could be due to the difference in the scoring system used for FISH, which is reported as a ratio of her-2/neu signal to the chromosome 17 signal, while CISH used in the study was single-colored assay, and only her-2/neu average number of signals was reported. One case in our study to amplification by CISH was found non-amplified by FISH, in addition this case had polysomy. Also 4 cases which are equivocal by FISH were found amplified by CISH and had polysomy of chromosome 17. The polysomy 17 was found in these cases. The reason of this erroneous of CISH results reported by (21) were in agreement with our results. This could be the major problem of single color CISH that cannot detect polysomy cases as concluded by (27), who also reported high concordance between the two methods and that 8 tumors were amplified by CISH but not by FISH. In the present study, 10 tumors were amplified by CISH but not with FISH and 5 cases were equivocal and 5 cases were non amplified by FISH. The differences between the two methods FISH and CISH is related to a lack of CISH chromosome 17 interpretation with one color CISH method.

CONCLUSION

There was a significant correlation between FISH and CISH for assessment of her-2/neu in breast cancer patients. The advantage of two-color FISH is the ability to distinguish chromosomal amplification from aneuploidy using a probe for chromosome 17 centromere, her-2/neu which is very useful for equivocal cases. This property is not found with one color CISH used in the present study.

Acknowledgements

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REFERENCES

4. Althuis MD.; Fergenbaum JH.; Garcia-Closas M. et. al. (2004). Etiology of hormone receptor--defined
Prevalence of scabies in Anbar province / west of Iraq

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ABSTRACT

The frequency and prevalence of scabies *Sarcoptis scabiei* with some possible risk factors were studied in Anbar province, Iraq between May 2010 to February 2011. It was found that out of 401 patients with skin diseases there were 200 scabies infestation cases (49.87%). Scabies frequency in women (65%) was highly significant (p<0.001) compared to men. Higher scabies cases in rural (61%) then in urban residents. As age groups increased from 13 years to >60 years scabies incidents decreased from 31.5% to 5.5% respectively. It was also found that the number of scabies patients declined as the education level progresses from illiteracy (46.2%) to college level (1%). Other possible risk factors studied were inconsistent as factors affecting scabies infestation in this study.

Keywords: scabies, pruritus, Anbar province
INTRODUCTION

Scabies is a skin disease caused by the mite Sarcopit scabei and is endemic in resource-poor urban and rural communities throughout the world. (1). Scabies is spread mainly through direct skin to skin contact with an infected person or by contaminated beddings, clothing and towels. (2). The parasites largely target poor people of undeveloped regions with poor hygienic levels (3). The Department of Health of South Australia (2008) pointed out that scabies infestation is not an indication of poor hygiene and social class (4). Approximately 300 million people are infected with Scabies worldwide (5).

Characteristics and symptoms of the disease are lesions at the mite innovation sites with chronic scratching and redness of the skin appear 4-6 weeks after infection and itching, more so at night. The itching is due to allergic reaction. These symptoms are as a result of burrow sites by the female mite to lay eggs and hatch and become adult mites within of days. The mite is tiny (0.2-0.4mm long) (6). Burrow sites of infestation are found on soft and thin skin and folds surfaces like axilla, elbows, finger webs, genitalia, buttocks, waists and knees. Symptoms of scabies may persist up to 12 months following treatments, and patients who completes treatment may still be contagious (7).

The most effective and safest treatment is believed to be permethrin 5% Scabicide cream preparation. The cream is applied on whole body and left for 8-14 hours (8). Kala (2011) reported that many herbal plant healers have been traditionally used as antiscabies and other skin diseases in India (9). Crusted Norwegian scabies is a rare manifestation of scabies characterized by crusted lesions uncontrolled perforation of mites in the skin. The occurrence of the disease in human is related to poor immune system and virus infected patients (10).

In Iraq, limited relied community based data on the prevalence of scabies is available. During the last few decades, scabies infection in Iraq was scare. After the occupation of Iraq by the US, British and their allies in 2003, scabies patients were significantly increased, may due to tens of thousands of prisoners, declining hygiene standards ...etc.

Therefore, this study was devoted to survey the prevalence of scabies in the largest province in Iraq (Anbar) during 2007-2011 in a Dermatology Private Clinic in Ramadi, the capital of the province. The study also aimed to assess possible risk factors may be related to the disease including socioeconomic, education, age, sex, and prisoners' contacts.

MATERIALS AND METHODS

The study was conducted in Ramadi city, the largest province west of Iraq. The city is about 600,000 inhabitants, located 120 Kilometers west of Baghdad, the capital of Iraq. All patients and control (with no scabies) who presented to the Private Dermatology Clinic in Ramadi during a period of nine months (19 May 2010 to 15 February 2011) were included in the study. Generally, the patients visited the dermatology clinic from different towns in Anbar province. A presumptive diagnosis of scabies was based on symptomatic complaints of pruritus and physical examination of the sites involved. The entire body of each patient was examined. Cases of scabies were diagnosed by to the senior dermatologist in the private clinic. After that, Patients were sent to the researcher in another room where patients were asked using suitable questionnaire. Scabies was diagnosed grossly, microscopically and clinically for the presence of burrows or erythematous popular vesicular, postural or lesions associated with different degrees of itching as absent, weak, moderate or severe and family history if other family member with similar symptoms. A Consent was taken from each patient with the approval of the dean of the College of Medicine, University of Anbar for the ethics of the study.

Statistical analysis

Frequency procedures of (2004) were utilized to calculate frequency and percentages. Chi-Square test was utilized to test if differences are present among different levels of two factors tested for all different factors in the study. Logistic procedure of SAS (2004) was utilized to calculate risk factors by computing odds ratio and 95% confidence interval by multivariate logistic regression analysis.

RESULTS AND DISCUSSION

Out of a total sample of (401) patients of different skin diseases presented to the Dermatology Clinic, there were 200 scabies infestation positive and 201 negative. Figure (1) represents the distribution of scabies patients with relation to variable age groups. Out of the total patients, 49.87% were infected with scabies. The highest scabies incidents were in the age group (13-29) years old. This age group is considered most active and interacted with the surrounding environment. As the age groups increase from 13-50 scabies patients were less reaching a minimum of 5.5% in simple Chi-Square analysis. The frequency of scabies patients with female (65%) was highly significant compared to male (35%) (table 1).
Results obtained from a study conducted in Brazil (11) found no consistence pattern of age distribution in scabies patients, which comprised 8.8% of population in slum urban population in Brazil. This result may suggest that women may be exposed to the might infestation more than men. This result also showed that higher incidents of scabies patients are found in the rural residents (61.06%) more than in Urban (table 2). This may be due to less health education in rural societies than in urban. Figure (2) shows the relationship between scabies patient infestation and the level of education. It was found that as the education level progresses from illiteracy (46%), primary, secondary, high school and institution and colleges (1%), the scabies patients would decrease. Education seems to give persons the ability of understanding health and frequency of diseases with methods of control and treatments more than uneducated people.

From a total skin disease patients of 259 studied with relation to their income levels of thousand Iraqi Dinar/month there were 58 (22.3%) scabies patients. Scabies infestation frequency and percentage with relation to the income levels of their families were found to be variable (figure 3). It is interesting to note that the highest incidence of infestation in this study was in the highest income group 18 out 58 scabies patients, which represents (31%). Similar results were found with the two highest income levels of individual (percapita) (figure 4) and the lowest was in the lowest income group.
The frequency of percentages of scabies patients with relations to the number of family number are presented in figure (5). It was found that the highest incidents of scabies patients is in the highest family number group of 10 or more. It represented 23(22.3%) out of a total interviewed patients of 103 scabies patients, which is 25.8% out of a total 314 skin disease patients. The lowest scabies patients were found to be in the lowest family number of 2, which was only about 2%.

The study also examined the effects of sharing towels and blankets, having showers daily, the presence of animals in the house, and the presence of neglected persons on the scabies infestation spreading within the family members, which were found to be variable and not consistent (table 3). Data collected from patients revealed that these factors are not considered valid for scabies mite infestation within the family members surveyed.

![Figure (5): Frequency and percentages of Scabies patients and healthy individuals in the study with number of family members](image)

Table (3): Frequency and percentages of scabies patients and healthy individuals in the study related to variable factors

<table>
<thead>
<tr>
<th>Scabies</th>
<th>Factors</th>
<th>Sharing towel and blanket</th>
<th>Washing daily</th>
<th>Presence of Animals</th>
<th>Presence of neglected person</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Negative</td>
<td>Frequency 52</td>
<td>149</td>
<td>7</td>
<td>194</td>
<td>110</td>
</tr>
<tr>
<td></td>
<td>Percent 25.87</td>
<td>74.13</td>
<td>3.48</td>
<td>96.52</td>
<td>54.73</td>
</tr>
<tr>
<td>Positive</td>
<td>Frequency 123</td>
<td>64</td>
<td>10</td>
<td>190</td>
<td>89</td>
</tr>
<tr>
<td></td>
<td>Percent 65.78</td>
<td>34.22</td>
<td>5</td>
<td>95</td>
<td>87.25</td>
</tr>
<tr>
<td>Total</td>
<td>Frequency 175</td>
<td>213</td>
<td>17</td>
<td>384</td>
<td>199</td>
</tr>
<tr>
<td></td>
<td>Percent 45.10</td>
<td>54.90</td>
<td>4.24</td>
<td>95.76</td>
<td>65.68</td>
</tr>
</tbody>
</table>

Chi-square test between each factor with incidence of scabies at p<0.01

Table (4) shows the results of logistic regression analysis and the trend of significance of scabies patients. It was found that age groups and education levels are highly significant (P< 0.001) factors affecting scabies patient infestation in this study.

Table (4): Logistic regression analysis results

<table>
<thead>
<tr>
<th>Factors</th>
<th>DF</th>
<th>Chi-Square</th>
<th>Pr&gt;Chi Square</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>5</td>
<td>7.4934</td>
<td>0.1865</td>
</tr>
<tr>
<td>Gender</td>
<td>1</td>
<td>18.5751</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>Residence</td>
<td>1</td>
<td>0.0963</td>
<td>0.7563</td>
</tr>
<tr>
<td>Education levels</td>
<td>5</td>
<td>20.5668</td>
<td>0.001</td>
</tr>
<tr>
<td>Number of family members</td>
<td>8</td>
<td>10.2934</td>
<td>0.245</td>
</tr>
<tr>
<td>Sharing towel blanket</td>
<td>1</td>
<td>3.2998</td>
<td>0.0693</td>
</tr>
<tr>
<td>Washing daily</td>
<td>1</td>
<td>0.0018</td>
<td>0.9659</td>
</tr>
<tr>
<td>Presence of Animals</td>
<td>1</td>
<td>6.2694</td>
<td>0.0123</td>
</tr>
<tr>
<td>Presence of neglected person</td>
<td>1</td>
<td>3.9776</td>
<td>0.0461</td>
</tr>
</tbody>
</table>
The presence of animals and the presence of neglected persons are significant (P= 0.0123 and P= 0.0461 respectively). Other factors (Gender, number of family sharing towels and blankets and showering) were non-significant. Table (5) represents the odds ratio and multiple logistic regression and probability test of mean values of different factors affecting scabies infestation. It was found that gender is highly significant (p<0.001) and the presence of animals and neglected persons in the house were significant ( P= 0.0123 and P=0.10401) respectively.

Table (5): Odds ratio, 95% Wald Confidence Limits, and probability for X² test of multiple logistic regression

<table>
<thead>
<tr>
<th>Factors</th>
<th>Level</th>
<th>Level</th>
<th>Odds Ratio</th>
<th>95% Wald Confidence Limits</th>
<th>Pr&gt;Chi Square</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>1</td>
<td>6</td>
<td>95.687</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Age</td>
<td>2</td>
<td>6</td>
<td>&gt;999.999</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Age</td>
<td>3</td>
<td>6</td>
<td>&gt;999.999</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Age</td>
<td>4</td>
<td>6</td>
<td>&gt;999.999</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Age</td>
<td>5</td>
<td>6</td>
<td>&gt;999.999</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Gender</td>
<td>F</td>
<td>M</td>
<td>310.264</td>
<td>22834</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Residence</td>
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<td>0.757</td>
<td>0.131</td>
<td>4.379</td>
</tr>
<tr>
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<td>&gt;999.999</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Education levels</td>
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<td>5</td>
<td>&gt;999.999</td>
<td>48.703</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Education levels</td>
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<td>526.678</td>
<td>20.229</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Education levels</td>
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<td>5</td>
<td>&gt;999.999</td>
<td>42.392</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Education levels</td>
<td>4</td>
<td>5</td>
<td>120.183</td>
<td>1.556</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Number of family members</td>
<td>2</td>
<td>10</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Number of family members</td>
<td>3</td>
<td>10</td>
<td>0.07</td>
<td>0.002</td>
<td>2.557</td>
</tr>
<tr>
<td>Number of family members</td>
<td>4</td>
<td>10</td>
<td>0.007</td>
<td>&lt;0.001</td>
<td>0.259</td>
</tr>
<tr>
<td>Number of family members</td>
<td>5</td>
<td>10</td>
<td>0.01</td>
<td>&lt;0.001</td>
<td>0.3</td>
</tr>
<tr>
<td>Number of family members</td>
<td>6</td>
<td>10</td>
<td>0.041</td>
<td>0.002</td>
<td>0.866</td>
</tr>
<tr>
<td>Number of family members</td>
<td>7</td>
<td>10</td>
<td>0.039</td>
<td>0.001</td>
<td>1.316</td>
</tr>
<tr>
<td>Number of family members</td>
<td>8</td>
<td>10</td>
<td>0.534</td>
<td>0.023</td>
<td>12.664</td>
</tr>
<tr>
<td>Number of family members</td>
<td>9</td>
<td>10</td>
<td>0.106</td>
<td>0.002</td>
<td>4.981</td>
</tr>
<tr>
<td>Sharing towel and blanket</td>
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<td>1</td>
<td>4.844</td>
<td>0.883</td>
<td>26.575</td>
</tr>
<tr>
<td>Washing daily</td>
<td>0</td>
<td>1</td>
<td>0.013</td>
<td>&lt;0.001</td>
<td>&gt;999.999</td>
</tr>
<tr>
<td>Presence of Animals</td>
<td>0</td>
<td>1</td>
<td>23.512</td>
<td>1.986</td>
<td>278.414</td>
</tr>
<tr>
<td>Presence of neglected person</td>
<td>0</td>
<td>1</td>
<td>32.343</td>
<td>1.062</td>
<td>985.126</td>
</tr>
</tbody>
</table>

Watkins (2010) reported that scabies infestation can be spread rapidly in schools by direct contact between pupils (12). Therefore, school health agents should be able to recognize scabies symptoms and offer appropriate advice to families and school employees to control further spread in the school and the community. Raza et. al. (13) identified risk factors for scabies among male soldiers in Pakistan including itching in family, dormitory males, infrequent bathing, infrequent changing clothes, low education and sharing beds. They pointed out that overcrowding, large family size and sharing of towels and hospitalization were not risk factors. Andersen (2004) added that education assessment and treatment of the disease are more important measures to control the disease (14). Scabies lesions generally present as small erythematous papules.
Infections and allergic reaction can result from scratching.
Watkins (2012) revealed that scabies female deposits its eggs under the skin and dies after about a hatched month. The eggs are hated after two days and the male dies after mating (15). Venna et. al. (2001) described the infestation and life cycle of scabies and lice, risk factors and medical and non-medical treatments (16). Olley (2011) stated that scabies is associated with close proximity like care homes and the spread of infection rather difficult to control (17). Risk assessment for contact tracing of scabies infections are divided into 3 risk levels, high, which includes workers with intimate direct contacts, medium, which includes workers with intermittent contact and, low for those who have no intimate contact (18). A study conducted by Tjioe and Rissers (19) discussed life cycle, clinical preventative, epidemiology, diagnosis, treatment and control. A delay in diagnosis may induce rapid spread of scabies disease in mentally retarded and elderly patients. Immediate diagnosis and treatment are therefore necessary. Normal scabies (Scabies vulgaris) and crusted (scabies crustos, novegica) can be healed by effective treatments or topical application of permethrin 5% and 2 oral doses of ivermectin 200 ug/kg (one week apart). Currie and McCarthy (2010) discussed the therapeutics recommendation of permethrin and ivermectin for scabies and the clinical problem and the mechanism of their benefit and potential adverse effects (20). Sharma and Singal (2011) believed that permethrin had a rapid onset of action (21).

REFERENCES
6. Department of Public Health, Division of Communicable Disease Control, State of California (2010). Scabies, MS 7307, Sacramento, CA
Study of paronychia as risk factor of diabetic patients in Ramadi province

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ABSTRACT

Paronychia is an acute and chronic inflammation (bacterial and fungal) of skin surrounds a toenail or fingernail due to exposure to moisture or harsh chemical for long periods. The aim of the present study was to investigate the paronychia in diabetic patients visiting Al-Ramadi General Teaching Hospital. A total of (135) patients with nail infection were categorized into two groups according to disease status: 89 patients (acute paronychia) and 46 patients (chronic paronychia). Out of 135 patients, 59 were diabetic women and men and the other 63 patients were not suffered from diabetes, but they were antibiotics abuse, while 13 patients; nor diabetic neither antibiotics abuse. Gram positive bacteria Staphylococcus aureus and Streptococcus pyogenes showed high incidence of infection in acute cases, while candida albicans showed high incidence of infection in chronic cases. Paronychia is more common in adult women than men. Acute and chronic paronychia infections are more occurred among diabetic patient and antibiotic users.

Keywords: Paronychia, Diabetes, Staphylococcus aureus, Streptococcus pyogenes, candida albicans
INTRODUCTION

Paronychia is either acute paronychia (an infection of the folds of tissue surrounding the nail of a finger or, less commonly, a toe, lasting less than six weeks) or chronic paronychia (an infection of the folds of tissue surrounding the nail of a finger or, less commonly, a toe, lasting more than six weeks) (1,2). Paronychia is either acute paronychia (an infection of the folds of tissue surrounding the nail of a finger or, less commonly, a toe, lasting less than six weeks) or chronic paronychia (an infection of the folds of tissue surrounding the nail of a finger or, less commonly, a toe, lasting more than six weeks) (1,2). The infection generally starts in the paronychium at the side of the nail, with local redness, swelling, and pain (3). The risk of paronychia increases in patients suffered from diabetes (4), due to decline of immune response (5). Bacteria such as Staphylococcus aureus (S.aureus) and Streptococcus pyogens were responsible for acute causes (6,7). Acute paronychia is usually precipitated by direct or indirect trauma to the cuticle or nail fold, and may be from relatively minor events, such as dishwashing, an injury from a splinter or thorn, nail biting, biting or picking at a hangnail, finger sucking, an ingrown nail, or manicure procedures. In chronic paronychia, the cuticle separates from the nail plate, leaving the region between the proximal nail fold and the nail plate vulnerable to infection (4). Paronychia might be associated with diabetes, drug-induced immunosuppression (5), or systemic diseases such as pemphigus (6). Health services were affected by wars, widespread violence and internal displaced movements (7,8). Several articles stressed on difficulties in glycemic control or misuse of antibiotic (9,10). The western region of Iraq suffered from all these circumstances. Thus, this study was carried out to throw a light on one of paronychia risk factors, diabetics in patients attending the Al-Ramadi General Teaching Hospital.

MATERIALS AND METHODS

Patients

A total sample of (135) patients with nail infection were included in this study. They were recruited for the period 2nd May to 30th October 2014. Out of 135 patients, 59 were diabetic women and men and 63 non-diabetic patients, but they were antibiotics abuse, while 13 another patient; nor diabetic neither antibiotics abuse.

Specimens collections

Pus or exudate and affected nails were swabbed with sterile swabs for affected areas. Swabs were kept in sterile test tubes for bacteriological study.

Bacterial and fungal isolations and characterization were done by using standard methods (11).

Blood glucose level determination

To confirm the diabetic status of patients, blood glucose levels were determined (12).

Case history

The forms were used to categorize the patients if they are diabetic or used antibiotics not prescribed by physician.

Statistical analysis

Student test was used to explain results obtained.

RESULTS

Table (1) shows the types of microorganisms isolated from patients with paronychia attending Al-Ramadi General Teaching Hospital. Gram positive cocci (Staphylococcus aureus and Streptococcus pyogenes) were isolated from acute cases only, on other hand Candida albicans was isolated from patients with chronic paronychia exclusively. Gram negative rods (Pseudomonas aeruginosa, Proteus vulgaris, Escherichia coli and Klebsiella spp.) were isolated from both acute and chronic paronychia. Fifty-nine (48%) of the studied patients were diabetic, as judged by the case history and confirmed by fasting blood glucose level (data not shown). Adult women showed the majority percentage (42) (71.2%). Antibiotics abuse also revealed another risk factor for paronychia, 63 (51.6%) of positive cases for paronychia used antibiotics without need or prescribed by physician.

Female represented superior than male and accounted 39(61.9%) against 24(38.1%) (table 2).

Statistical analysis revealed highly significant difference between women diabetic and antibiotic users more than in men (p< 0.01).

Table (1): Isolation of microorganisms from chronic and acute paronychia

<table>
<thead>
<tr>
<th>Isolates</th>
<th>No. of isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Acute infection</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>22</td>
</tr>
<tr>
<td>Streptococcus pyogenes</td>
<td>18</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>13</td>
</tr>
<tr>
<td>Proteus vulgaris</td>
<td>10</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>8</td>
</tr>
<tr>
<td>Klebsiella spp.</td>
<td>6</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>0</td>
</tr>
</tbody>
</table>
DISCUSSION

The present study was conducted in an area suffering from war and displacement and relocation of people and showed that the female more susceptible to paronychia. That not surprised, because other researcher shows the same finding previously (13,14). The wet conditions during the housewife work might explain this finding, wet conditions (house-wife work) is causing loss of nail cuticle following separation of proximal and lateral nail folds which creates a dead space allowing the entrance of water (15,16), detergent and bacteria or candida under the nail folds followed by inflammation leading to main feature of chronic paronychia. Minor traumas and manicuring predispose to acute paronychia (17), which might contribute to female predominance in this study. A male predominance was observed in elderly patients (data not shown in tables). This finding might be attributed to the reduction of wet conditions in housewife working and manicuring process among females in their elderly age. In the line of other studies (18), acute paronychia was a predominant one. The main factor associated with the development of acute paronychia is direct or indirect trauma to the cuticle or nail fold. This enables pathogens to inoculate the nail, resulting in infection. The most common cause of acute paronychia is direct or indirect trauma to the cuticle or nail fold. Such trauma may be relatively minor, resulting from ordinary events, such as dishwashing, an injury from a splinter or thorn, nail biting, finger sucking, an ingrown nail, pushing back the cuticles, artificial nail application, or other nail manipulation (19). The finding that gram positive cocci and gram negative rods were isolated from patients with paronychia is consistent with that from studies of Iraq (17). However, it is inconsistent with that from developed countries e.g. USA (19, 20), which reported gram positive cocci only. This difference might be attributed to the difference in environmental sanitation between Iraq (developing country) and developed country. Contact with animals, no sewage disposal, and shortage of preventive services …etc, or predisposing factors e.g. biting or sucking finger as cause of trauma. The study revealed that 40% of chronic paronychia cases showed candida albicans. The observed high rate of candida albicans might be attributed to the high prevalence of diabetes mellitus in Iraq. Last Iraqi national survey documented a high prevalence of hyperglycemia (20). It was mentioned that diabetic patients easily get fungal infection (21).

The observed rate of candida albicans (40%) is similar to that reported in India (41%) (22). Rate of candida albicans in chronic paronychia (40%) is much lower than that reported in Iran (84%) (23). The difference might be attributed to differences in life style. Difference in sample might be contributed to this variation, also. It was a growing trend of fingernail onychomycoses in Iraq in the last decades (24).

### Table (2): The predisposing factors of paronychia

<table>
<thead>
<tr>
<th>Predisposing factors</th>
<th>Women</th>
<th></th>
<th>Men</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Diabetic patients</td>
<td>42</td>
<td>71.2</td>
<td>17</td>
<td>28.8</td>
</tr>
<tr>
<td>Antibiotic uses</td>
<td>39</td>
<td>61.9</td>
<td>24</td>
<td>38.1</td>
</tr>
<tr>
<td>Total</td>
<td>81</td>
<td>66.4</td>
<td>41</td>
<td>33.6</td>
</tr>
</tbody>
</table>

### REFERENCES

Antibiotics susceptibility pattern to *Salmonella enteric serotype Typhi* in Iraqi hospital, 2013

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**ABSTRACT**

Enteric fever is countered as a public health problems. The treatment with fluoroquinolone, 3rd generation Cephalosporine and Azithromycin, are now increasingly resistant. The preset study aimed to investigate the antibiotic susceptibility pattern for *S. Typhi* which has been isolated from different Iraqi hospitals in year 2013. One hundred (100) strains of *S. Typhi* were tested from different Iraqi hospitals to (12) different types of antibiotics according to EUCAST-2013. The percentages of resistant to Ampicillin, Chloramphenicol and Trimethoprim- Sulfamethoxazol were (74-77%), while for Ciprofloxacin, Cefotaxime and azithromycin 77%, 14% and 17% respectively. All strains showed full susceptibility (100%) to meropenem, tigecyclin and tetracycline. The strains of *Salmonella typhi* which were studied in CPHL, 2013 had shown increasingly resistant to all antibiotics which were used for treatment (1st and 2nd line treatment due to acquire of resistance genes). This study concludes, that is an increasing resistant trend for *Salmonella typhi* to Ciprofloxacin and 3rd cephalosporin. In some reports carabapenem (Imipenem and Meropenem) are potential third lines for multi-drug resistant isolate.

**Keywords:** EUCAST: European Committee on Antimicrobial Susceptibility testing, CPHL: Central Public Health Lab – Iraq – Baghdad, CLSI: Clinical and Laboratory Standard institute
**INTRODUCTION**

Enteric fever is countered as a major public health problem especially in developing countries. Chloramphenicol has been the treatment of choice for typhoid fever for (40) years, but widespread emergence of multi drug resistance of S. Typhi, which resistant to 1st line of drug (resistance to Ampicillin, Chloramphenicol and Trimethoprim-Sulfamethoxazole) had necessitated the search for other therapeutic options (1).

The 2nd lines of treatment were fluoro-quinolone and 3rd generation Cephalosporins such as Ceftriaxone, Cefixime and Cefotaxime are now increasingly being used the treatment typhoid fever. With the developing of resistance to fluoro-quinolone and 3rd generation cephalosporin, are now increasingly being used in the treatment of typhoid fevers such as India, Pakistan, Middle East and Africa (2).

Salmonella resistance to cephalosporine is largely due to production of enzymes by bacteria called Extended-spectrum β-lactamase [ESBL]. Sal. typhi was found to produce a wide variety of ESBL types including TEM, SHV and CTX-M enzymes. The CTX-M-15 resistance gene was found in Kuwait and United Arab Emirates, and had the problem for these salmonella strains to pose for treatment (1).

**MATERIALS AND METHODS**

One hundred (100) isolates of Salmonella typhi were submitted to Central Public Health Lab (CPHL) in Bagdad from different Iraqi hospitals. The sources of isolated were from blood, stool and other body sites. The identification of Salmonella typhi was carried and confirmed by using (API 20E) system Biomerieux, and antisera for salmonella poly and mono (Bio-Rad).

The strains were classified as Susceptible , intermediate or resistant according to ( EUCAST ) version 3.0 April 2013- with standard strain were done with Escherichia coli ATCC 25922.

The panel of antibiotics tested included Ampicillin - 10µg ; Ampicillin - Clavulanic 20+10 µg ; Chloramphenicol 30µg ; Trimethoprim-Sulfamethoxazole[1.25-23.75 ]µg; Ciprofloxacin 5µg ; Naídixic acid 30µg ; Cefotaxim 30 µg ; meropenem 10 µg ; Imipenem 10µg ; Tigecycline 15µg ; Azithromycin 15µg and Tetracyline 30µg . At present, no clinical Azithromycin breakpoint has been defined for Enterobactericeae; included Salmonella by either (CLSI) or EUCAST (3). The MIC (minimal inhibitory concentration) of azithromycin concentration reported in the literature is in the range of 4-32 mg /ml, and most strains the MIC are 4 to 8 mg/ml. We consider strains was susceptible when zone diameter of inhibition by disk containing 15mg more ≥ 21mm ,and equal to MIC=16mg/l.

**RESULTS**

Typhoid fever is a systemic infection, and the most source isolates were from blood and stool as shown in table (1).

<table>
<thead>
<tr>
<th>No.</th>
<th>Site of infection</th>
<th>No. of isolates</th>
<th>% of isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Blood</td>
<td>84</td>
<td>84%</td>
</tr>
<tr>
<td>2</td>
<td>Stool</td>
<td>10</td>
<td>10%</td>
</tr>
<tr>
<td>3</td>
<td>Urine</td>
<td>4</td>
<td>4%</td>
</tr>
<tr>
<td>4</td>
<td>bone marrow</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td>5</td>
<td>Pus ( wound )</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>100</td>
<td>100%</td>
</tr>
</tbody>
</table>

Multi – drug resistant typhoid fever (MDR) is defined as typhoid fever caused by Salmonella typhi strains which are resistant to all three first – line recommended drugs for treatment. Ie Chloramphenicol, Ampicillin, Trimethoprim-sulfamethoxazol and ciprofloxacin (4).

According to the results obtained, the percentages of resistance to first line antibiotic were found to be 74 – 77% as shown in the table (2).

According to results in table (2), the resistance to second – line antibiotics like the fluoroquinolone like ciprofloxacin; third- generation cephalosporine like cefotaxime and azithromycin, are often now used for treatment of uncompleted typhoid fever; were found to be 12-77%.

The isolates of Sal. Typhi reported susceptible to fluoroquinolone i.e Ciprofloxacin must be sensitive to both Naídixic acid and ciprofloxacin antibiotics. The CLSI reported special zone for Breakpoints of Ciprofloxacin for Sal. Typhi .

The resistance to called potential third line of drugs Imipenem, meropenem and tigecyclin were 0-2%. Also Tetracyline antibiotic is found rolled back as filled susceptible for Salmonella typhi isolated in CPHL-2013.
**DISCUSSION**

Determining the antibiotic sensitivity pattern of *Salmonella typhi* isolates is necessary since it will guide the physicians in making the right choice of drug when treating the patients with typhoid fever. The sensitivity of *Sal. Typhi* had continuously changed, initially chloramphenicol was used. In 1989 outbreak was caused by strain of *Sal. Typhi*, resistant to Chloramphenicol, ampicillin and Trimethoprim-sulfamethaxol i.e. (1st line of treatment MDR *Sal. Typhi* ) and were reported in many developing countries, especially Pakistan and India (5).

Before 1970, a few sporadic isolations of Chloramphenicol resistant were reported in Aden, Chile and Kuwait (5).

The MDR *Sal. typhi* strains were also reported from all parts of the world, in Quetta Pakistan for instance 69%, where in Vietnam 89% isolates between 1998 and 2002 (6).

In 1995, 28% of all the isolates of *Sal. typhi* from humans in the USA were resistant to first line of drug.

From the results obtained at the present study, the resistant for Ampicillin, Chloramphenicol and Trimethoprim-sulfamethaxol were 76%,74% and 77% respectively in Iraq.

The second line antibiotics used for treatment *Sal. Typhi* were fluoroquinolone (Ciprofloxacin, gatifloxacin,levofoxacin,Ofloxocacin, perfoxacin) third-generation Cephalosporin (Ceftriaxone, Cefotaxime, Cefixime and Cepodoxime proxetil and azithromycine are used for treatment MDR *Sal. Typhi* , thus, there are increasing resistant percentages trend of *Sal. typhi* to 2nd line of treatment i.e fluoroquinolone, 3rd generation cephalosporin, which range from (12-77%) of isolates.

In *Salmonella*, resistance to 3rd cephalosporine is largely due to production of enzymes by bacteria called "Extended-Spectrum-β-lactamase –ESBLs" which can hydrolyze Cephalosporine. A type of ESPLs called (CTX-M), which displays a level of resistance to Cefotaxime and Ceftrixone was found in strains *Sal. Typhi* from Kuwait and United Arab Emirates (1).

Resistance to 3rd Cephalosporine also was reported in UK and USA, especially among patients who travelled from developing world-east.

Also, nosocomial infection caused by ESBLs producing *Sal. Typhi* was reported from Latin America, France, Senegal, Africa, Asia and Europe (7).

The emergence of ESBLs in MDR *Sal.Typhi* represents a new challenge and has become a matter of concern, especially in developing countries. Fluoroquinalone used in treatment are Ciprofloxacin, Gatifloxacin, Levofloxacin and ofloxacin.

The resistant *Sal. Typhi* to fluoroquinalone was reported in India and UK in 1992 (2, 5, 8).

For strain susceptible to ciprofloxalin, (CLSI) reported a special zone of breakdowns (CLSI-2013) for *Salmonella typhi* and sensitive to Nalidixic acid disc.

Currently, Azithromycin is recommended for the treatment of both Shigellosis and Salmonellosis by the World Health Organization and the American Academy of Pediatrics (3) and its increasingly used for the management of uncomplicated enteric fever. The MIC of Azithromycin, which was reported in the literature are in the range 4-32 µg/ml and most strains of the MIC are 4 to 8 µg/ml (1).

At present, no clinical Azithromycin break points were defined for Enterobacteriacae including Salmonella by either CLSI or EUCAST (3). Azithromycin was given once daily in a dose of 1000 mg on the first day and 500 mg a day for (6) or more days.

At the moment, the emergence of resistant strains to three major secondary drugs like Ciprofloxacin, cefotaxime and Azithromycin is posing a major problem. Tigecycline, glycycline, tetracycline analogue and lack cross-resistance with other compounds and are very potent inhibiting strains of *salmonella typhi*.

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**Table (2): Numbers and percentages of susceptible and resistant for *Salmonella typhi* tested in CPHL - 2013**

<table>
<thead>
<tr>
<th>No.</th>
<th>Antibiotics</th>
<th>Number of susceptible</th>
<th>Number of resistant</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ampicillin</td>
<td>24 (24%)</td>
<td>76 (76%)</td>
</tr>
<tr>
<td>2</td>
<td>Amoxicillin-Clavulanic acid</td>
<td>25 (25%)</td>
<td>75 (75%)</td>
</tr>
<tr>
<td>3</td>
<td>Chloramphenicol</td>
<td>26 (26%)</td>
<td>74 (74%)</td>
</tr>
<tr>
<td>4</td>
<td>Trimethoprim-sulfamethaxol</td>
<td>23 (23%)</td>
<td>77 (77%)</td>
</tr>
<tr>
<td>5</td>
<td>Nalidixic acid</td>
<td>23 (23%)</td>
<td>77 (77%)</td>
</tr>
<tr>
<td>6</td>
<td>Ciprofloxacin</td>
<td></td>
<td>77 (77%)</td>
</tr>
<tr>
<td>7</td>
<td>Cefotaxime</td>
<td>88 (88%)</td>
<td>12 (12%)</td>
</tr>
<tr>
<td>8</td>
<td>Azithromycin</td>
<td>83 (83%)</td>
<td>17 (17%)</td>
</tr>
<tr>
<td>9</td>
<td>Imipenem</td>
<td>98 (98%)</td>
<td>2 (2%)</td>
</tr>
<tr>
<td>10</td>
<td>Meropenem</td>
<td>100 (100%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>11</td>
<td>Tigecycline</td>
<td>100 (100%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>12</td>
<td>Tetracycline</td>
<td>100 (100%)</td>
<td>0 (0%)</td>
</tr>
</tbody>
</table>
Tetracycline is an old antibiotic and used for treatment of Typhoid before 1980. however, the strain of Sal.Typhi is found now susceptible to both Tetracycline and Tigycyclin.

If we closely look at the results, the only meropenem showed 100% susceptible and Imipenem showed 98% susceptible both are penems classes of antibiotic with broad spectrum activity and are stable to hydrolysis by extended – spectrum β-lactamaes producing strain (ESBLs) these antibiotics are alternative to drug – resistant strain of Sal. Typhi isolates.

CONCLUSION

This study concludes that there is an increasing resistance for Salmonella typhi to 2nd line drugs like Ciprofloxacin and 3rd generation Cephalosporin. The Carbapenems (Imipenem and Meropenem) are potential third line for multi-drug resistant Salmonella typhi.

REFERENCES

Epidemiological study of pulmonary and extrapulmonary tuberculosis from Iraqi patients

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ABSTRACT

The study included 2328 samples of patients who attended to Specialized Chest and Respiratory Disease Center/ National Reference Laboratory for Tuberculosis (NRL) in Baghdad in the period from (February 2015 to February 2016). These samples included (1806) of pulmonary TB patients and (522) Extra-pulmonary samples. Methods included direct examination by Zeihl-Nelsen stain, then cultures were examined traditionally and been followed by phenotypic-based identification methods.

Out of a total sample 2328 of different specimens, 351(15.08%) were positive by direct examination (smear microscopy) and 433(18.6%) specimens were able to grow on solid media, and it was found that cultures had detected 82 negative specimens by smear microscopy. Results showed that males were more affected significantly 1356 (58.25%) than females 972 (41.75%), and the higher percentage of positive culture was in Baghdad (12.07%) than other governorates (6.53%).

The most sites of specimens samples were sputum 1462 (62.8%) with high significant difference than other samples 866 (37.2 %) and the total pulmonary TB was 1806 (77.58%) than total extra pulmonary TB 522 (22.42%). The ages of patients were ranged from <1 year to >60 years. Age range of 30-40 years of age group was observed in (0-9) years with percentage (5.36%). More incidence was a previously treated Pulmonary Tuberculosis patients (old case) (25.44%) with highly significant than new patients (new cases) (10.19%), but the higher percentage of positive culture was in multidrug resistant (MDR) (32.51%).

Keywords: tuberculosis, epidemiology, MDR

الملخص باللغة العربية

تضمنت الدراسة 2328 عينة جمعت من المرضى المراجعين للمختبر المركزي للإصابات والنازعات المسببة في بغداد للعاصمة العراقية، على مدى فترة من شهر شباط 2015 إلى شهر شباط 2016. شملت العينات (1806) مريضاً بمرض التهاب الرئة (M. Tuberculosis) و(522) مريضاً بمرض نزل التهاب الرئة، واستخدمت طرق الفحص المجسم المسح الحدي أوZEIHIL-NELSEN، لم تتغير على الألواح الرطبة الخاصة بطبقات الأشجار المجسمة المتفجرة. نتائج الدراسة تشير أن 351 (15.08%) عينة كانت إيجابية للإصابة بالتهاب الرئة (M. Tuberculosis) و (433) (18.6%) عينة كانت قابلة للنمو. النتائج تظهر أن الذكور أكثر تأثراً بواتجاع 1356 (58.25%) من الإناث 972 (41.75%). وتظهر أن معدل الإصابة في بغداد (12.07%) أعلى من معدل الإصابة في其他 (6.53%).

العمر المثير للاهتمام في المعاينة كان من 30 إلى 40 سنة، في حين تم عزل (22.42%) من العينات في فئة من ست إلى عمر 60 سنة. النتائج تظهر أن ارتفاع نسبة الإصابة في مرضى النزل التهاب الرئة (25.44%) أعلى من معدل الإصابة في المرضى الجدد (10.19%). ولكن من النسبة الأعلى للحالات الإيجابية كانت في المرضى المثلث المتوفر (MDR) (32.51%).

Keywords: تهاب الرئة، الوبائي، MDR
INTRODUCTION

Tuberculosis (TB) stands as a major global health problem, ranking as the second highest cause of death from an infectious disease globally, after the human immunodeficiency virus (HIV). Iraq had been identified as middle-TB burden country in the world and ranks 8th of 22 EMR countries according to estimated incidence of all forms of TB. Iraq contributed 3% of total cases in the EMR and considered among the eight high TB burdened countries in the region (1). According to a recent report of the Ministry of health (MOH) in Iraq, the incidence rate 43/100,000 with nearly 8268 new and relapse cases reported for 2014 (2). The case detection rate is 54% for all forms of TB cases, mainly due to the poor security situation in some governorates, which report fewer cases as many cases are displaced or lost to follow up (1). The conventional techniques for TB diagnosing worldwide, can be done by symptoms, chest radiography, and sputum smear microscopy. Light microscopy with Ziehl Neelsen (ZN) stain is easier, cheaper and used in the diagnosis of TB in many resources poor setting, while culture is considered the gold-standard for diagnosis, the sensitivity of culture is far higher than microscopy (3,4).

The aim of this study was to determine epidemiology of M. tuberculosis isolates in Baghdad, as well as in other Iraqi governorates, to assess their transmission dynamics.

MATERIALS AND METHODS

Sample collection

The study was implemented at specialized center for chest and respiratory diseases in National Reference Laboratory for Tuberculosis (NRL) in Baghdad. During the period from (1/2/2015...1/2/2016). The samples were selected from 2328 TB patients that attended to the center which were, including pulmonary and extra pulmonary TB diseases. Samples were collected from two gender their ages were between (few months-95 year). Samples were collected from new cases (new patients), previously treated Pulmonary Tuberculosis patients (retreated patients). Baseline demographic data (including: name, age, gender, residence address, evidence of previous anti-TB treatment was compiled from patient’s medical records. sputum was collected from patient according to (National Tuberculosis Program in Iraq) three samples were collected from each patient. First one was taken from patient when he just reached the center; second sample collected at early morning before breakfast; and third one was collected at any other time on the day. The samples were divided into two group: pulmonary TB (1806) samples and (522) samples extra – pulmonary TB. The pulmonary TB patients (sputum Bronchial wash and Bronchoalveolar lavage) On the other hand, the extra – pulmonary TB samples were taken included (urine, pleural fluid, fine needle aspiration, gastric fluid, ascitic fluid, CSF etc.).

Processing of samples

The specimens were digested and decontaminated by using Sodium hydroxide procedure (Petroff decontamination procedure). An equal volumes of sputum were mixed with 4% sodium hydroxide (previously sterilized by autoclaving) in sterile centrifuge tube. The mixture was allowed to stand at room temperature for 15 minutes with occasional gentle shaking, and the mixture centrifuged at 3000 rpm for 15 minutes. The resultant supernatant was discarded and the sediment neutralized by distilled water or by drop with a 2 mol / 1N HCl solution containing 2% of phenol red combined with shaking until the color changes from red to yellow. And the sediment used for preparing the concentrated smear method and inoculating culture media (5).

Smears stained with Acid Fast Bacilli staining

Two smears were prepared from all decontaminated samples and stained with Ziehl Neelsen (ZN). All smears were examined with 100x oil immersion objectives using light microscopy.

Mycobacterial culture

Lowenstein Jensen (LJ) medium was prepared by method described by (5) and Löwenstein-Jensen (LJ) medium with sodium pyruvate was prepared in the same manner but with 12 g of sodium pyruvate instead of the glycerol. Decontaminated specimens was cultured on (LJ)media, (2-4) drops of the centrifuged sediment distributed over the surface of 3 slopes of (LJ) medium by plastic Pasteur pipettes used for each slope, and an additional one slope containing sodium pyruvate. All cultures were incubated at 37°C for 8 weeks, cultures were examined 48 hrs. after incubation; thereafter, cultures were examined weekly. The growth was observed or discarded as a negative after 8 weeks. All The positive growth of M. tuberculosis thus obtained was identified by their rate of growth, colonial morphology, Ziehl Neelsen staining and biochemical tests. There were several tests done for identification and typing of TB bacilli was prepared such as: Growth on pyruvate (stone brink) media, Growth on P.N.B (P-nitro benzoic acid) media, growth on T.C.H (thiophene-2- carboxyl acid Hydrazide) media, Niacin test, Nitrate reduction test and catalase test (5).

RESULTS

All samples (2328) were examined for acid fast stain. 351(15.08 %) samples were positive for acid fast stain but after culturing on LJ medium
433 (18.6%) samples showed cultural, microscopic features and biochemical characterization to be identified as *M. tuberculosis*. (table 1). It was found that cultures detected 82 negative specimens by smear microscopy. This result means that the cultivation rate of *M. tuberculosis* from clinical specimens was higher than direct examination, while negative of specimens were (81.4%, 84.92%) respectively with significant different, and the male was more affected significantly 271 (62.6%) than female162 (37.4%) from 433 positive specimens in culture.

Table (1): Distribution of samples according to sex

<table>
<thead>
<tr>
<th>Governorate</th>
<th>Total</th>
<th>+Ve</th>
<th>-Ve</th>
<th>+Ve</th>
<th>-Ve</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baghdad</td>
<td>1596</td>
<td>23</td>
<td>1353</td>
<td>1375</td>
<td>1085</td>
</tr>
<tr>
<td>Al-Basra</td>
<td>23</td>
<td>0.99</td>
<td>127</td>
<td>117</td>
<td>1085</td>
</tr>
<tr>
<td>Ninawa</td>
<td>6</td>
<td>0.26</td>
<td>14</td>
<td>5</td>
<td>1085</td>
</tr>
<tr>
<td>Kerbala</td>
<td>51</td>
<td>2.28</td>
<td>14</td>
<td>39</td>
<td>1085</td>
</tr>
<tr>
<td>Wasit</td>
<td>95</td>
<td>4.02</td>
<td>18</td>
<td>77</td>
<td>1085</td>
</tr>
<tr>
<td>DhiQar</td>
<td>103</td>
<td>4.42</td>
<td>22</td>
<td>81</td>
<td>1085</td>
</tr>
<tr>
<td>Diyala</td>
<td>108</td>
<td>4.64</td>
<td>19</td>
<td>89</td>
<td>1085</td>
</tr>
<tr>
<td>Salah Al-</td>
<td>54</td>
<td>2.32</td>
<td>6</td>
<td>48</td>
<td>1085</td>
</tr>
<tr>
<td>Din</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Babylon</td>
<td>58</td>
<td>2.62</td>
<td>16</td>
<td>72</td>
<td>1085</td>
</tr>
<tr>
<td>Al-Qadisiya</td>
<td>33</td>
<td>1.42</td>
<td>6</td>
<td>27</td>
<td>1085</td>
</tr>
<tr>
<td>Muthanna</td>
<td>21</td>
<td>0.9%</td>
<td>6</td>
<td>15</td>
<td>1085</td>
</tr>
<tr>
<td>Al-Anbar</td>
<td>8</td>
<td>0.34</td>
<td>2</td>
<td>6</td>
<td>1085</td>
</tr>
<tr>
<td>Maysan</td>
<td>20</td>
<td>0.9%</td>
<td>6</td>
<td>24</td>
<td>1085</td>
</tr>
<tr>
<td>An-Najaf</td>
<td>35</td>
<td>1.5%</td>
<td>12</td>
<td>31</td>
<td>1085</td>
</tr>
<tr>
<td>Kirkuk</td>
<td>7</td>
<td>0.3%</td>
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<td>6</td>
<td>1085</td>
</tr>
<tr>
<td>Sulaymaniyah</td>
<td>3</td>
<td>0.26</td>
<td>1</td>
<td>5</td>
<td>1085</td>
</tr>
<tr>
<td>Dobuk</td>
<td>5</td>
<td>0.26</td>
<td>0</td>
<td>5</td>
<td>1085</td>
</tr>
<tr>
<td>Arbil</td>
<td>29</td>
<td>1.25</td>
<td>5</td>
<td>24</td>
<td>1085</td>
</tr>
<tr>
<td>Total</td>
<td>2328</td>
<td>433</td>
<td>1895</td>
<td>1895</td>
<td></td>
</tr>
</tbody>
</table>

Chi-square χ² = 9.39*, P-value= 0.0245

*M. tuberculosis* cells appeared as a single cells or aggregated .The colonies of *M. tuberculosis* on LJ media were circular, rough, appears as brown, granular colonies. Table (2) summarizes the biochemical characterization results.

Table (2): Biochemical characterizations of *M. tuberculosis*

<table>
<thead>
<tr>
<th>Niacin Production</th>
<th>Nitrate reduction</th>
<th>Catalase test</th>
<th>TCH</th>
<th>PNB</th>
<th>Pyruvate medium</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>+</td>
<td>-</td>
<td>resistant</td>
<td>Sensitive</td>
<td></td>
</tr>
</tbody>
</table>

Results shown in table (3) revealed that the higher percentage of positive culture was in Baghdad than other governorate. Samples from Baghdad were 1596 out of 2328 (68.56%). Only 281(12.07%) specimens were positive by cultivation on LJ media, other governorates represented 732  out of 2866 (31.44%) with positive specimens 152 (6.53 %). Results shown in table (4) reflected that sputum were in 1462 (62.8%), while the other samples 866 (37.2 %) and the total pulmonary TB were 1806 (77.58%) of total extra pulmonary TB 522 (22.42%).

In table (5), the higher appearance of *M. tuberculosis* (28.47 %) was in the age group (30-39) years with highly significant difference between age groups (P < 0.001) . Finally, More incidence was found among retreated type of patients (25.44%) with highly significant with new type (10.19%), but the higher percentage of positive culture was in MDR (32.51%) as shown in table (6).

Table (3): Distribution of samples according to Governorates

<table>
<thead>
<tr>
<th>Governorate</th>
<th>Total</th>
<th>+Ve</th>
<th>-Ve</th>
<th>+Ve</th>
<th>-Ve</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baghdad</td>
<td>1596</td>
<td>1375</td>
<td>1085</td>
<td>1281</td>
<td>1315</td>
</tr>
<tr>
<td>Al-Basra</td>
<td>23</td>
<td>127</td>
<td>1085</td>
<td>1375</td>
<td>1315</td>
</tr>
<tr>
<td>Ninawa</td>
<td>6</td>
<td>14</td>
<td>1085</td>
<td>1375</td>
<td>1361</td>
</tr>
<tr>
<td>Kerbala</td>
<td>51</td>
<td>14</td>
<td>1085</td>
<td>1375</td>
<td>1361</td>
</tr>
<tr>
<td>Wasit</td>
<td>95</td>
<td>18</td>
<td>1085</td>
<td>1375</td>
<td>1357</td>
</tr>
<tr>
<td>DhiQar</td>
<td>103</td>
<td>22</td>
<td>1085</td>
<td>1375</td>
<td>1353</td>
</tr>
<tr>
<td>Diyala</td>
<td>108</td>
<td>19</td>
<td>1085</td>
<td>1375</td>
<td>1356</td>
</tr>
<tr>
<td>Salah Al-</td>
<td>43</td>
<td>6</td>
<td>1085</td>
<td>1375</td>
<td>1369</td>
</tr>
<tr>
<td>Din</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Babylon</td>
<td>58</td>
<td>16</td>
<td>1085</td>
<td>1375</td>
<td>1359</td>
</tr>
<tr>
<td>Al-Qadisiya</td>
<td>33</td>
<td>6</td>
<td>1085</td>
<td>1375</td>
<td>1369</td>
</tr>
<tr>
<td>Muthanna</td>
<td>21</td>
<td>6</td>
<td>1085</td>
<td>1375</td>
<td>1369</td>
</tr>
<tr>
<td>Al-Anbar</td>
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</tr>
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</tr>
<tr>
<td>An-Najaf</td>
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<td>12</td>
<td>1085</td>
<td>1375</td>
<td>1363</td>
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<tr>
<td>Kirkuk</td>
<td>17</td>
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<td>1085</td>
<td>1375</td>
<td>1374</td>
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<tr>
<td>Sulaymaniyah</td>
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</tr>
<tr>
<td>Dobuk</td>
<td>5</td>
<td>0</td>
<td>1085</td>
<td>1375</td>
<td>1374</td>
</tr>
<tr>
<td>Arbil</td>
<td>28</td>
<td>5</td>
<td>1085</td>
<td>1375</td>
<td>1367</td>
</tr>
<tr>
<td>Total</td>
<td>2328</td>
<td>433</td>
<td>1895</td>
<td>1895</td>
<td>1895</td>
</tr>
</tbody>
</table>

Chi-square χ² = 30.590*  p < 0.22

Table (4): Distribution of samples according to Site of infection

<table>
<thead>
<tr>
<th>Site of infection</th>
<th>Total</th>
<th>+Ve</th>
<th>-Ve</th>
<th>+Ve</th>
<th>-Ve</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sputum</td>
<td>1462</td>
<td>1373</td>
<td>1083</td>
<td>1083</td>
<td>2086</td>
</tr>
<tr>
<td>Bronchial Wash</td>
<td>286</td>
<td>189</td>
<td>1083</td>
<td>1083</td>
<td>271</td>
</tr>
<tr>
<td>Bronchoalveolar</td>
<td>58</td>
<td>35</td>
<td>1083</td>
<td>1083</td>
<td>83</td>
</tr>
<tr>
<td>Peritoneal fluid</td>
<td>287</td>
<td>15</td>
<td>1083</td>
<td>1083</td>
<td>232</td>
</tr>
<tr>
<td>Gastric fluid</td>
<td>35</td>
<td>3</td>
<td>1083</td>
<td>1083</td>
<td>32</td>
</tr>
<tr>
<td>CSF</td>
<td>17</td>
<td>0</td>
<td>1083</td>
<td>1083</td>
<td>17</td>
</tr>
<tr>
<td>Articular fluid</td>
<td>64</td>
<td>2</td>
<td>1083</td>
<td>1083</td>
<td>62</td>
</tr>
<tr>
<td>Abscess</td>
<td>19</td>
<td>1</td>
<td>1083</td>
<td>1083</td>
<td>18</td>
</tr>
<tr>
<td>Urine</td>
<td>59</td>
<td>1</td>
<td>1083</td>
<td>1083</td>
<td>58</td>
</tr>
<tr>
<td>Pericardial fluid</td>
<td>4</td>
<td>0</td>
<td>1083</td>
<td>1083</td>
<td>4</td>
</tr>
<tr>
<td>Aspiration fluid</td>
<td>16</td>
<td>1</td>
<td>1083</td>
<td>1083</td>
<td>15</td>
</tr>
<tr>
<td>Peritoneal fluid</td>
<td>6</td>
<td>1</td>
<td>1083</td>
<td>1083</td>
<td>5</td>
</tr>
<tr>
<td>Total EPTB</td>
<td>522</td>
<td>489</td>
<td>1083</td>
<td>1083</td>
<td>1895</td>
</tr>
<tr>
<td>Total</td>
<td>2482</td>
<td>379</td>
<td>1083</td>
<td>1083</td>
<td>1895</td>
</tr>
</tbody>
</table>

Chi-square χ² = 227.368**, P-value= 0.00
Table (5): Distribution of samples according to age groups

<table>
<thead>
<tr>
<th>Age group (years)</th>
<th>Total</th>
<th>Result</th>
<th>Percentage of positive culture (%) from total positive result</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 - 9</td>
<td>112</td>
<td>+Ve 6</td>
<td>5.36</td>
</tr>
<tr>
<td>10- 19</td>
<td>132</td>
<td>+Ve 28</td>
<td>21.45</td>
</tr>
<tr>
<td>20- 29</td>
<td>352</td>
<td>+Ve 71</td>
<td>20.17</td>
</tr>
<tr>
<td>30- 39</td>
<td>418</td>
<td>+Ve 119</td>
<td>28.47</td>
</tr>
<tr>
<td>40- 49</td>
<td>390</td>
<td>+Ve 85</td>
<td>21.79</td>
</tr>
<tr>
<td>50- 59</td>
<td>377</td>
<td>+Ve 67</td>
<td>17.77</td>
</tr>
<tr>
<td>&gt; 60</td>
<td>527</td>
<td>+Ve 57</td>
<td>10.82</td>
</tr>
<tr>
<td>Total</td>
<td>2328</td>
<td>+Ve 433</td>
<td>18.6</td>
</tr>
</tbody>
</table>

Chi-square $\chi^2=64.543**$, $P$-value = 0.00

Table (6): Distribution of samples according to type of patients

<table>
<thead>
<tr>
<th>Type of patients</th>
<th>Total</th>
<th>Result</th>
<th>Percentage of positive culture (%) from type of patient</th>
</tr>
</thead>
<tbody>
<tr>
<td>Retreated</td>
<td>397</td>
<td>+Ve 101</td>
<td>25.44</td>
</tr>
<tr>
<td>New</td>
<td>1325</td>
<td>+Ve 135</td>
<td>10.19</td>
</tr>
<tr>
<td>MDR</td>
<td>606</td>
<td>+Ve 197</td>
<td>32.51</td>
</tr>
<tr>
<td>Total</td>
<td>2328</td>
<td>+Ve 433</td>
<td>18.6</td>
</tr>
</tbody>
</table>

Chi-square $\chi^2=151.614**$, $P$-value = 0.00

**DISCUSSION**

The results showed that cultivation rate (18.6 %) of *M. tuberculosis complex* from clinical specimens was higher than direct examination (15.08%), because culture methods can detect cases with low mycobacterial loads. Culture techniques have been estimated to detect as many as 10 –100 of viable organism per ml of specimen in comparison with 5000-10,000 bacilli/ml that are required for detection by smear as mentioned by (6). The smear microscopy plays an important role in the early diagnosis of mycobacterial infections which is rapid, inexpensive and very useful method to identify highly contagious patients, its usefulness depends largely on the quality of the sputum specimen and the performance quality of the laboratory (7). However, sensitivity of sputum Ziehl–Neelsen (Z-N) staining does not exceed 60% to 70% as compared with sputum culture. cultivation *M. tuberculosis* consider as a gold standard for TB diagnosis, it can performance on a variety of specimens' pulmonary and extra pulmonary specimens. It is much more sensitive than microscopy (8).

Regarding the extra-pulmonary TB (EPTB) specimens, (9) had mentioned that the pulmonary tuberculosis infected lungs mainly, while the extra-pulmonary tuberculosis can present as pleural effusions, tuberculous lymphadenitis, tuberculous meningitis, abdominal tuberculosis and tuberculosis of bones and joints. Although Initial diagnosis is dependent on the smear microscopy for acid fast bacilli (AFB) by Ziehl Neelsen (ZN) staining and culture , there are various reports regarding the sensitivity of Z-N smear for extra-pulmonary specimen ranging from as low as 0% to as high as75% as reported by (10), these findings were clear in this study, as a total of 33 extra-pulmonary specimens which were diagnosed with conventional staining and solid culture. The majority of TB cases were more from Baghdad city with percentage (68.56%) than other governorates at percentage (31.44%) as shown in Table 3 , so the high percentage of TB disease were from Baghdad may be related to the high density of population associated with crowded conditions, and because of the availability of more diagnostic methods facilities than other governorates that enable to record more TB cases among suspected TB patients , with difficulties for patients to reach Baghdad. Differences in tuberculosis notification rates between men and women may reflects the nature of work that, the males work in various fields, non sanitary, and crowded area especially in cases of poor ones. Thus, males are more exposed to infection.This result agreed with Shaker and Saleh who found that males were more than females in TB cases (63.2% males and 36.8%females) (11) , and in 2014 according to WHO , there were an estimated 5.4 million incident cases of TB among men, 3.2 million among women and 1.0 million among children (1).

The most site of specimens are shown in Table 4 was the pulmonary TB (PTB) that higher percentage of positive culture than total extra pulmonary TB (EPTB). In this study the lower reduction in EPTB
compared with incidence of PTB, such results are expected and attributed to that TB is a lung disease and sputum represented the best specimen for it. Other study showed high percentage compared with this result such as the result of Al-Otaibi and Hazmi in Saudi Arabia, who reported percentage of EPTB was 57.5% and PTB was 42.5% (12), but another study found the percentage of EPTB and PTB in Turkey were 25.9% and 74.1% respectively (13).

The current result of infection age group was shown in table 5 revealed that low percentage (7.39%) of tuberculosis cases among age less than 19 years from 433 positive culture may be attributed to BCG vaccine given at young ages while the high percentage of infection above 19 year returned to many factors such as smoking and other immune depression factors like diseases, this was in accordance with study in Baghdad by Al-Rubayai which found the highest percentage of tuberculosis were at the age group above 15 years and the mean age was 35.8 years (14), and in Iraq in 2014, among 8268 cases 585 (7%) cases aged under 15 years (1) and this study was in accordance with study in Basra which found the higher appearance of M. tuberculosis (24.2%) was in the age group 30-39 years (15).

In table (6), the higher percentage of positive culture was in MDR (32.51%), the major factors behind the high prevalence rate of MDR-TB include poor access to laboratory diagnosis and effective treatment. Therefore, the undiagnosed cases continue to spread the MDR-TB. An empiric treatment of patients infected with TB or MDR-TB strain can also result in the spread of MDR-TB. The approach of empiric treatment without the drug susceptibility testing (DST) in many developing countries is believed to aggravate the problem of MDR-TB in patients already infected with strains resistant to one or more drugs (1). Therefore, a rapid and reliable diagnostic tool with simple experimental protocol can significantly help in decreasing the prevalence rate of MDR-TB strain.

CONCLUSION

Results obtained by this study suggest that cultivation rate is more sensitive than direct method. The gender and age may be risk factors for disease, males with high risk than female with MDR-TB infection.

REFERENCES

Isolation of *E. coli* O157:H7 from bovine milk sample in Baghdad province

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**ABSTRACT**

The present study aimed to isolate *E. coli* and *E. coli* O157:H7 from bovine milk samples. One hundred samples of bovine milk were collected from three areas of Baghdad province. All milk samples were subjected to culture on specific media (Sorbitol MacConkey agar plus cefixime potassium tellurite (SMA-CT)) and Chrom agar then biochemical and serological tests using latex kit were done. The results demonstrate that 47% (47 out of 100 milk samples) and 22% (22 out of 100 milk samples) were positive to *E. coli* and *E. coli* O157:H7 respectively from these three areas in Baghdad. In west Rathwania 23 and 16 out of 50 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. In east Rathwania 17 and 5 out of 34 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. In Al-Huria city 7 and 1 out of 16 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. The results of sensitivity test showed that the *E. coli* O157:H7 were resistant 100% to the antibiotic, Doxycycline 30 (Do 30), Trimethoprim 5 (TMP5), Ceftriaxone 30 (CRO30).

**Keywords:** *E. coli* O157:H7, milk samples
INTRODUCTION

Milk is considered a serious threat foodstuff and plays a significant medium for bacterial growth (1). It can be the major resource of food borne pathogens, and there have been many outbreaks due to food-poisoning related with direct consumption of untreated milk. The existence of pathogenic microbes in milk increased because of fecal contamination during the milking. Contaminated milking parlor, equipment and floors can facilitate the spread of these pathogens to the udders. Good hygiene, including the removal of fecal material from udders and ensuring a clean environment, is therefore important (2).

Bovine and other ruminants carry E. coli O157:H7 without clinical symptoms and these animals shed this pathogen with feces for long time lead to contamination of food (3). Kedhier (2006) found that 90 % of fecal samples of healthy cattle were positive to E. coli O157:H7 (4). Ruminants, especially cattle are the major reservoir of Shiga Toxin E. coli (STEC), and human infectivity is often related to eating of contaminated beef or closed contact with the animals (5).

STEC strains are extremely significant harmful to human with low infectious dose, through using of contaminated water or food leading to food-borne disease. Raw milk is used by wide range of people especially who work at farms and consider that the milk is safe and have useful health benefits which are lost by pasteurization.

The existence of food borne microorganisms in milk may attributed to close contact with contaminated sources in the dairy cattle farm and shedding of the pathogens from the udder of an infected animal (6). E. coli O157:H7 cause severe abdominal pain, hemorrhagic colitis, diarrhea, thrombocytopenic, hemolytic–uremic syndrome, and purpura .The pathogenic factors of entero-hemorrhagic E. coli such as Shiga toxins, the chromosomal LEE locus that carries factors (eaeA, tir) involved in the attaching and effacing process, and a big plasmid carry the hemolysin genes (7).

Infections can also be acquired by close contact with animals and by human-to-human spread (8). Studies show that there is a considerable increase in the resistance of E. coli O157:H7 to antibiotics (9). The effectiveness of antibiotics therapy for STEC infections is unresolved because antibiotics may destroy bacteria cell walls, in this manner releasing Shiga toxins (10). However, many studies propose that some antibiotics, if treated at the beginning of the course of infection, may prevent disease development to hemorrhagic uremic syndrome (HUS) (11). Because STEC infections are not aggressively (12) they are not recommended for treating STEC O157:H7 infections.

MATERIALS AND METHODS

Collection of fresh milk samples

The samples of milk were collected from three areas in Baghdad province. Ten (10) ml of fresh milk samples of 100 cows were collected for the microbiological analysis. Milk was stripped by hand directly into sterile test tubes after cleaning the udder with warm disinfectant solution and transporting as soon as possible to the laboratory in a cooled container (13).

Bacterial isolation

Each sample was cultured on MacConky, Eosin Methylen blue, Sorbitol MacConkey agar plus cefixime potassium tellurite (SMA-CT) and Chrome agar. E. coli O157:H7 was incubated at 37°C for 24 hrs. aerobically. Gram stain was performed by taking a single colony from a growth. Confirmation by biochemical tests (IMVIC, Urea test) was done to confirm the diagnosis of isolated bacteria (14).

Serological test

Latex Agglutination Test was done for isolated E. coli O157:H7 by latex agglutination test kit, (Oxoid).

RESULTS AND DISCUSSION

Culturing on MacConkey and eosin methylene blue

Results of culturing samples exhibit different morphological characteristic colonies of E. coli. The colonies appear pink on MacConkey agar and blue metallic sheen appear on eosin methylene blue. The gram stain was done on isolated bacteria and it appears as gram negative bacteria (figure 1).

Figure (1): Colonies of E. coli on EMB showed metallic sheen dye
Culturing on specific media for *E. coli* O157:H7 - Sorbitol MacConky agar plus cefsixime potassium tellurite (SMA-CT)

The suspected colonies of *E. coli* O157:H7 show tiny, circular and colorless with smoky center on SMA-CT (figure 2). *E. coli* O157:H7 cannot ferment Sorbitol that is differs from most other strains of *E. coli* in being incapable to ferment Sorbitol. The isolation of *E. coli* O157:H7 on SMA-CT was similar to what mentioned by (15), who revealed that Sorbitol MacConkey agar was an alternative of conventional MacConkey agar used in the isolation of *E. coli* O157:H7.

**Figure (2): Colorless colonies with smoky center of **

*E.coli O157:H7* **on SMA-CT**

Culturing on Chrome agar *E. coli* O157: H7

*E. coli* O157:H7 appeared on chrome agar as mauve color colonies while another strains of *E.coli* appear blue color colonies when cultured on the same agar and incubated for 24 hrs. at 37 °C (figure 3). This result confirms that the chrome media is very essential to diagnose *E. coli* O157:H7. These results agreed with (16), who revealed that *E. coli* O157:H7 appear mauve color due to utilize one of chromogenic substrates. The growth of mauve color colonies on Chrome agar is considered as presumptive identification for *E. coli* O157:H7. Non *E. coli* O157:H7 bacteria may use other chromogenic substrates resulting in blue to blue green colored colonies whereas none of the chromogenic substrates are utilized, colonies may appear as a natural color (17). Studies conducted all over the world had shown that chrome agar is an effective supplemental medium for the diagnosis of probable STEC strains and indicated that these technique have higher specifically, exclusivity and performance in compare with other technique (18). Philips et. al. (2005) showed that the improved diagnostic concert and competence of Chrome agar assist to diagnose *E. coli* O157:H7 cases and outbreaks (19).

Tavakoli et. al. (2008) stated that chromogenic media have more benefit and can be a suitable choice for conventional and routine diagnostic procedure (20).

The present study showed that 47% and 22% of fresh milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. From 100 milk samples 47 samples were positive to *E. coli* and 22 were positive to *E. coli* O157:H7 from three areas in Baghdad province. In west Rathwania, 23 and 16 out of 50 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. In East Rathwania, 17 and 5 out of 34 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively. In Al-Huria city 7 and 1 out of 16 milk samples were positive to *E. coli* and *E. coli* O157:H7 respectively (table 1). These results were analyzed by use Chi-square. Chi square value was 4.88 and P=0.087 for positive isolates of *E. coli* and Chi-square value was 0.0870, P=0.04 for positive isolates to *E. coli* O157:H7. Results revealed that the differences among areas were significant (P=0.04).

Presence of *E. coli* and *E. coli* O157:H7 in milk is considered as an indicator to contaminated milk by these bacteria either by unhealthy conditions in milk production or unclean of equipments and milk handlers or fecal contaminate of udder and milk. The result variations in microbiological value of fresh milk in different studies attributed to many factors such as: geographic area, farm size, different sample size and the type of methods of diagnosis (21). Although safe milk is produced from healthy cows and milking process is performed under clean circumstances but bovine feces were spotted about floor of farm and attached to the milking and feeding equipments leading to contamination of feeding equipments and infected feed. The results obtained here are in agreement with those obtained by (22), who isolated *E.coli* from fresh milk samples at 63% in Sudan and they mentioned the way of milk contamination with *E. coli*. Soomro et. al. (2002) isolated *E. coli* from milk samples with percentage 75 % in Pakistan (23), while the results of *E. coli* O157:H7 isolation percentage were in agreement with that found by (24) who found that *E. coli* O157:H7 are presented in fresh milk samples of cattle in Baghdad province with percentage 72%.

Table (1) shows the positive isolates of *E.coli* and *E.coli*157: H7

<table>
<thead>
<tr>
<th>Areas from Baghdad Province</th>
<th>No. of samples</th>
<th>No. of positive samples of <em>E. coli</em></th>
<th>No. of positive samples of <em>E.coli</em> O157:H7</th>
</tr>
</thead>
<tbody>
<tr>
<td>West Rathwania</td>
<td>50</td>
<td>23</td>
<td>16</td>
</tr>
<tr>
<td>East Rathwania</td>
<td>34</td>
<td>17</td>
<td>5</td>
</tr>
<tr>
<td>Al- Huria city</td>
<td>16</td>
<td>7</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>47(47.4%)</td>
<td>22(22.4%)</td>
</tr>
<tr>
<td>Chi-square value</td>
<td>0.211</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>0.89</td>
<td>0.04</td>
<td></td>
</tr>
</tbody>
</table>

Differences among areas were significant (P=0.04)
It was found that 86 out of 150 (57.34%) of fresh milk samples were positive to *E. coli* O157:H7 (figures 3 and 4) as results obtained by (25), (12) isolated *E. coli* O157:H7 from fresh milk samples with 8% percentage, while (26) isolated *E. coli* O157:H7 in 6 cattle fresh milk samples out of 40 samples (15%) from Baghdad Province. Lye *et al.* (2013) found *E. coli* O157:H7 in raw cow milk with percentage 8.75% (27). Dairy cattle are important reservoirs of STEC. This microorganism contaminates milk during close contact with dairy cattle and its environment (28). Oksuz *et al.* (29) explained that the most outbreaks due to *E. coli* O157:H7 were related with fodder cattle origin especially those contaminated with feces of cattle. It was confirmed by (30) that *E. coli* O157:H7 are shedded from healthy cattle feces. The results of sensitivity test to antibiotics showed that all isolates of *E. coli* O157:H7 were resistance 100% to Doxycycline 30(Do30), Trimethoprim 5 (TMP5) and Ceftriaxone 30(CRO30).

**Figure (3): The mauve colonies of *E. coli* O157: H7 on chrome media**

**Figure (4): Latex agglutination test 4.5 positive agglutination to *E. coli* O157:H7**

Recognition of *E. coli* O157:H7 in fresh milk samples revealed that raw milk is a serious public health concern of less educated people and farm families. They still consume milk without using boiling or pasteurization. Effective and continuous training accompanied with emphasize on the safety and health issues related to raw milk hazards is really needed. To protect public health, more stringent regulations and strategies are in demand (27).

**REFERENCES**

13. Vendramin T.; Kich DM.; Molina RD.; de Souza CFV.; Salvadori RU.; Pozzobon A. and Bustamante-filho IC. (2014). Molecular screening


Assessment of natural radioactivity level and hazard index in cultivation medium (Peat moss and perlite)

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ABSTRACT

The background level of radiation in the natural environment surrounds us at all times. Levels of natural occurring radioactivity in Cultivation medium such as peat moss and perlite that widely used within gardeners and horticultural uses to achieve better growth for plants have been investigated using HPGe detector through gamma-ray spectrometry. The activity concentration of radionuclides in the $^{226}$Ra, $^{232}$Th-series and $^{40}$K has been determined in seven samples of peat moss and one of perlite. The activity concentration of $^{226}$Ra, $^{232}$Th and $^{40}$K for Peat moss and perlite samples were ranged from 6.2±2.3 Bq/kg to 54.3±28.4 Bq/g from 2.1±0.3 Bq/kg to 29.2±6.4 Bq/kg and from 99.3±11.6 Bq/kg to 551.2±38.1 respectively. The study also examined some radiation hazard indices: The Radium Equivalent ($Ra_{eq}$), Absorbed Dose Rates (D), Annual Effective Dose Rate (AEDR), External Hazard Index (Hex) Internal Hazard Index (Hin) and total excess lifetime cancer risk (ELCR). All the health hazard indices are well below samples were less than the global limit.

Keywords: Radiological Hazards; Peat moss; Perlite; Cultivation medium; HPGe Detector
**INTRODUCTION**

Peat moss (sphagnum peat moss) is partially fossilized plant matter that is formed in poorly oxygenated waters of marshes, bogs such as mosses. This is used as a soil conditioner, which increases the soil's capacity to hold water and nutrients (1). Perlite is a volcanic glass that is heated to 1,600 degrees F. (871 C), where upon it pops much like popcorn and expands to 13 times its former size used in soil mixes to improve aeration and modify the soil substructure, keeping it loose well-draining and defying compaction. The major radionuclide of concern are potassium, uranium, thorium and their decay products, some of which like radium and radon are intensely radioactive but occur in low concentrations. Most of these sources have been decreasing due to radioactive decay since the formation of the earth, because there is no significant amount currently transported to the earth. Thorium and uranium primarily undergo alpha and beta decay. They aren't easily detectable. However, many of their daughter products are strong gamma emitters (2). Volcanic rocks (and their plutonic equivalents) commonly are enriched in radioactive isotopes especially potassium (3). Many gropes studied the radioactivity of Bog sphagnum, volcanic rocks soil and fertilizers (4-6).

The aim in this work was to determine the concentrations of radionuclide of \(^{226}\text{Ra}\), \(^{232}\text{Th}\) and \(^{40}\text{K}\) in some kinds of peat moss and perlite that were collected from Iraqi markets.

**MATERIALS AND METHODS**

**Sampling and samples preparation**

Seven (7) samples of peat moss and one of perlite were collected of from the Iraqi markets. The samples were dried in oven about 200°C for 6 hrs., then crushed and sieved through 200 µm mesh to be homogenize sample. The samples weighted and carefully sealed in closed container. They were kept for about four weeks to reach secular equilibrium between thorium and radium contents of the samples and their daughter radionuclides (7). The average sample weight was 500 g.

**Experimental set up**

Gamma ray spectrometry analysis of the samples for natural radioactivity was carried out by HpGe detector with the resolution (FWHM) of 1332KeV to \(^{60}\text{Co}\) is 2.2KeV, and relative efficiency 20%. The detector was controlled by computer with Gain 2000 a software program. The system also contained the usual electronic components of preamplifier, amplifier and power supply in one unit with the Digital Spectrum Analyzer 2000 (DSA-2000) (CANBERRA).

The detector was situated in a well shield the measuring station against all background radioactivity in the room. Measurement takes 3600 sec and subtraction of the background was measured for the same counting time. The gamma spectra were collected. The activities of \(^{226}\text{Ra}\) and \(^{232}\text{Th}\) in each sample were determined by measuring the characteristic γ-peaks of their daughters. The energy regions selected for the corresponding radionuclides (609KeV,1120KeV,1764KeV) for \(^{214}\text{Bi}\) to measured \(^{226}\text{Ra}\) and (911KeV,969 KeV) for \(^{208}\text{Ac}\) to measured \(^{232}\text{Th}\) but the activity concentration of \(^{40}\text{K}\) was measured directly on gamma ray of 1461 KeV (8).

**RESULTS AND DISCUSSION**

**Activity concentration**

The activity concentration of \(^{226}\text{Ra}\), \(^{232}\text{Th}\) and \(^{40}\text{K}\) were calculated by using the following equation (9).

\[
A (Bq) = \frac{\text{CPS}}{(\text{DPS. I. eff})} \pm \frac{\text{CPS error}}{(\text{DPS. I.eff})} ...(1)
\]

Where CPS is the net count rate per second (figure 1), I is the intensity and eff is the efficiency of the detector. The \(^{226}\text{Ra}\) activity concentration ranges from 6.2±2.3 Bq/Kg to 54.3±28.4 Bq/g. \(^{232}\text{Th}\) activity concentration ranges from 2.1±0.3 Bq/Kg to 29.2±6.4 and \(^{40}\text{K}\) activity concentration ranges from Bq/Kg, as reported in table (1). Figure (2) describes the activity concentration of \(^{226}\text{Ra}\), \(^{232}\text{Th}\) and \(^{40}\text{K}\) (Bq·Kg\(^{-1}\)) in peat moss and perlite cultivation medium used in Iraq.
Table (1): The activity concentration of $^{226}$Ra, $^{232}$Th and $^{40}$K in peat moss and perlite cultivation medium used in Iraq

<table>
<thead>
<tr>
<th>No.</th>
<th>Samples</th>
<th>$^{226}$Ra Bq/kg</th>
<th>$^{232}$Th Bq/kg</th>
<th>$^{40}$K Bq/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Perlite Saudi</td>
<td>35.2±13.4</td>
<td>29.2±6.4</td>
<td>551.2±38.1</td>
</tr>
<tr>
<td>2</td>
<td>Peat moss Alfayafy</td>
<td>37.7±17.6</td>
<td>6.1±1.8</td>
<td>106.8±16.5</td>
</tr>
<tr>
<td>3</td>
<td>Peat moss Culvita</td>
<td>39.7±25.3</td>
<td>4.3±1.8</td>
<td>114.3±16.5</td>
</tr>
<tr>
<td>4</td>
<td>Peat moss Alasala</td>
<td>14.1±2.8</td>
<td>4±0.4</td>
<td>188.5±14.1</td>
</tr>
<tr>
<td>5</td>
<td>Peat moss Green fresh</td>
<td>17.5±3.4</td>
<td>11.2±0.9</td>
<td>153±13.2</td>
</tr>
<tr>
<td>6</td>
<td>Peat moss Gulf</td>
<td>14.3±3.4</td>
<td>6.4±1</td>
<td>99.3±11.6</td>
</tr>
<tr>
<td>7</td>
<td>Peat moss Chift Chiler</td>
<td>54.3±28.4</td>
<td>13.3±3.9</td>
<td>181.8±20.7</td>
</tr>
<tr>
<td>8</td>
<td>Peat moss Pot grond</td>
<td>6.2±2.3</td>
<td>2.1±0.3</td>
<td>109±10.8</td>
</tr>
</tbody>
</table>

Figure (2): Activity concentrations of $^{232}$Th, $^{226}$Ra and $^{40}$K (Bq/Kg) in peat moss and perlite used in Iraq

Radium equivalent activity

Assessment of radiological hazards was made by calculating the radium equivalent activities, external and internal hazard indices. The radium equivalent activity ($Ra_{eq}$) is a weighted sum of activities of the $^{226}$Ra, $^{232}$Th and $^{40}$K based on the assumption that 370 Bq/kg of $^{226}$Ra, 259 Bq/kg of $^{232}$Th and 4810 Bq/kg of $^{40}$K produce the same gamma-ray dose rates as given by the following equation (10):

$$Ra_{eq} = A_{Ra} + 1.43A_{Th} + 0.077A_{K} \ldots (2)$$

The results obtained for the radium equivalent activity index $Ra_{eq}$ of all samples of peat moss and perlite were varied from 17.59 to 119.4 Bq/kg as listed in table (2). It is observed that the values of radium equivalent index of all samples are not exceed 370 Bq/kg the maximum permissible limit. Therefore, the results of this study will contribute to the national data regarding natural radioactivity levels (11).

External hazard ($H_{ex}$)

The external hazard index is defined to be obtained from $Ra_{eq}$ expression through the supposition that its maximum allowed value (equal to unity) corresponds to the upper limit of $Ra_{eq}$ (370Bq/kg). $H_{ex}$ which is given by this equation (12).

$$H_{ex} = \frac{A_{K}}{370} + \frac{A_{Th}}{259} + \frac{A_{Ra}}{4810} \ldots (3)$$
Internal hazard ($H_{in}$)

The internal hazard index, which is given by the expression:

$$H_{in} = \frac{A_{Ra}}{185} + \frac{A_{Th}}{225} + \frac{A_{K}}{1910} \ldots \ (4)$$

The external and internal hazard indices of samples were varied from 0.047 for pot grond peat moss to 0.31 mGy/y, from 0.06 for pot grond peat moss to 0.415 mGy/y for perlite Saudi, respectively as listed in table (2). It is noticed that external and internal hazards indices are lower than unity for all samples (13).

Gamma radiation hazard $I_{\gamma}$

To estimate the level of $\gamma$-radiation hazard associated with the natural radionuclides, another radiation level index suggested by OECD’s NEA (14) were evaluated by using the following equation:

$$I_{\gamma} = \frac{A_{Ra}}{150} + \frac{A_{Th}}{100} + \frac{A_{K}}{1500} \ldots \ (5)$$

$A_{Ra}$, $A_{Th}$, and $A_{K}$ are the activity concentrations (in Bq/kg) of $^{226}$Ra, $^{232}$Th and $^{40}$K respectively. The radiation level index $I_{\gamma}$ of peat moss and perlite samples are varied from 0.133 for pot grond to 0.887 for perlite Saudi, which is found to be less than unity for all samples as listed in table (2).

Table (2): Radiation Hazard parameters for peat moss and perlite samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>$^{226}$Ra, $^{232}$Th and $^{40}$K</th>
<th>$H_{ex}$</th>
<th>$H_{in}$</th>
<th>$I_{\gamma}$</th>
<th>$D$ (nGy/h)</th>
<th>AEDE (µSv/y)</th>
<th>ELCR $\times 10^3$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Perlite Saudi</td>
<td>119.4</td>
<td>0.31</td>
<td>0.415</td>
<td>0.887</td>
<td>55.6</td>
<td>68.19</td>
<td>0.238</td>
</tr>
<tr>
<td>2 Peat moss Alfayafy</td>
<td>54.6</td>
<td>0.14</td>
<td>0.24</td>
<td>0.33</td>
<td>24.19</td>
<td>29.66</td>
<td>0.103</td>
</tr>
<tr>
<td>3 Peat moss Cul vita</td>
<td>54.66</td>
<td>0.13</td>
<td>0.25</td>
<td>0.38</td>
<td>24.25</td>
<td>29.75</td>
<td>0.104</td>
</tr>
<tr>
<td>4 Peat moss Almassala</td>
<td>28.6</td>
<td>0.09</td>
<td>0.13</td>
<td>0.26</td>
<td>16.27</td>
<td>19.95</td>
<td>0.069</td>
</tr>
<tr>
<td>5 Peat moss Green fresh</td>
<td>45.29</td>
<td>0.12</td>
<td>0.16</td>
<td>0.33</td>
<td>20.59</td>
<td>25.25</td>
<td>0.088</td>
</tr>
<tr>
<td>6 Peat moss Gulf</td>
<td>31.1</td>
<td>0.08</td>
<td>0.117</td>
<td>0.22</td>
<td>14.08</td>
<td>17.26</td>
<td>0.060</td>
</tr>
<tr>
<td>7 Peat moss Chiff Chiler</td>
<td>87.32</td>
<td>0.227</td>
<td>0.37</td>
<td>0.616</td>
<td>38.76</td>
<td>47.55</td>
<td>0.166</td>
</tr>
<tr>
<td>8 Peat moss Pot grond</td>
<td>17.59</td>
<td>0.047</td>
<td>0.061</td>
<td>0.133</td>
<td>8.45</td>
<td>10.37</td>
<td>0.036</td>
</tr>
</tbody>
</table>

Estimation of absorbed and effective dose

The measured activity of $^{226}$Ra, $^{232}$Th and $^{40}$K were converted into doses by applying the factors 0.462, 0.604 and 0.0417 for radium, thorium and potassium respectively (15). These factors were used to calculate the total absorbed gamma dose rate in air at 1 m above the ground level using the following equation:

$$\text{Absorbed dose } D \text{ nGy/h } = 0.462 \cdot A_{Ra} + 0.604 \cdot A_{Th} + 0.0417 \cdot A_{K} \ldots \ (6)$$

Where, $A_{Ra}$, $A_{Th}$ and $A_{K}$ are the activities (Bq·kg$^{-1}$) of radium, thorium and potassium in the samples. To estimate annual effective doses must be taken into account of:

1) The conversion coefficient from absorbed dose in air to effective dose.
2) The indoor occupancy factor. The annual effective doses are determined as follows (16):

$$\text{AEDE in Sv/y = D nGy/h } \times 0.7 \text{ Sv/Gy} \times 0.2 \times 10^{-3} \ldots \ (7)$$

Annual estimated average effective dose equivalent received by a member is calculated using a conversion factor of 0.7 Sv·Gy$^{-1}$, which is used to convert the absorbed rate to annual effective dose with an outdoor occupancy of 20% (17).

Excess Lifetime Cancer Risk (ELCR)

The Excess Lifetime cancer risk (ELCR) was calculated using the following equation (18):

$$\text{ELCR} = \text{AEDE} \times \text{DL} \times \text{RF} \ldots \ (8)$$

Where, AEDE is the Annual Equivalent Dose Equivalent, DL is the average duration of life (estimated to 70 years), and RF is the Risk Factor (Sv$^{-1}$), i.e. fatal cancer risk per Sievert. For stochastic effects, ICRP uses RF as 0.05 for public. Lifetime cancer risk, gamma index for peat moss and perlite samples were studied below the safe limit. The $I_{\gamma}$ values for peat moss and perlite samples are below the limit of unity meaning that the radiation dose is below the permissible limit of 1mSv·y$^{-1}$ recommended by IAEA (13).
CONCLUSION

The levels of natural radioactivity in peat moss and perlite samples that used in horticultural in Iraq were determined using high resolution gamma-ray spectrometry. The results can be useful in the assessment of the radiological hazard associated with the exposures and the radiation doses due to naturally radioactive element contents in samples. We noticed that the measured samples were within the recommended safety limits and did not pose any significant source of radiation hazard inhabitants.

REFERENCES

Anti hyperglycemic effects of colostrum, virgin and multipara camel milk in alloxan-induced diabetic rats

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ABSTRACT
The present study was conducted to investigate the effect of colostrums and camel milk treatment on body weight and serum glucose of alloxan induced diabetic rats. The study was divided into three experiments according to period of treatment, first experiment was undertaken to investigate the effect of colostrums through 7 days of treatment, The second experiment was undertaken to investigate the effect of virgin and multipara camel milk through 30 days of treatment, while the third experiment was undertaken to investigate the effect of virgin and multipara camel milk through 60 days of treatment. The results of experiments revealed that the diabetic male rats in second group were suffering from significant decrease at (p ≤ 0.05) in body weight and there were significant differences in serum glucose.

Keywords: colostrums, camel milk, alloxan-induced diabetic rats
INTRODUCTION

Diabetes mellitus is a chronic systemic disease characterized by an increased blood glucose concentration. The word diabetes is derived from the Greek word “diabainein” and means “to pass through”, referring to the large volume of urine, while mellitus comes from the Latin term “mell”, which means honey and refers to the sweetness of the urine from patients with untreated diabetes. Diabetes is caused by either decreased production of insulin from the pancreatic β-cells or decreased effect of insulin on target tissues or by a combination of these two. Diabetes not only causes disturbances in carbohydrate metabolism, but also affects lipid and protein metabolism. The two major categories of diabetes are type 1 and type 2 diabetes, previously also called “insulin-dependent” (IDDM) and “non-insulin-dependent” (NIDDM), or “juvenile” and “adult-onset” diabetes, respectively. Type 1 diabetes is characterized by an autoimmune reaction that leads to a total loss of function of the insulin-secreting β-cells of the islets of Langerhans in the pancreas, resulting in absolute insulin deficiency. Type 2 diabetes is the consequence of decreased insulin sensitivity (primarily in skeletal muscles, adipose tissue, and liver) and/or decreased insulin secretion from β-cells. It is the most common form of diabetes and is increasing in epidemic proportions worldwide. Considerable overlap exists between the two conditions, and type 1 and type 2 diabetes have been proposed to be different forms of the same disease, the main difference being the absence of an immune response in patients with type 2 diabetes, leading to a slower rate of β-cell loss. On the other hand, the clear lack of evidence for similar genetic factors predisposing to type 1 and type 2 diabetes supports the notion of two separate diseases. It is predicted that about 366 million people worldwide will be diabetic by the year 2030. There are 2 types of diabetes; T1D and Type2 Diabetes (T2D). T1D is a heterogeneous disorder associated with the destruction of pancreatic beta cells, with the resultant effect of absolute insulin deficiency. Type2 diabetes on the other hand is characterized by resistance to insulin action and suboptimal insulin secretion response. Causes of diabetes ranges from autoimmune-mediated destruction of beta cells and idiopathic destruction or failure of beta cells. About 5-10% of the total cases of diabetes worldwide are due to T1D, T1D is the most common type of diabetes in children and adolescents while Type2 Diabetes (T2D) is common among young adults. Type1 Diabetes (T1D) has been increasing by 2% to 5% worldwide.

MATERIALS AND METHODS

The study included three experiments and one hundred twenty (120) male rats were used.

Experiment one: was conducted to investigate the effect of the treatment of camel colostrum for 7 days. Twenty four (24) male rats were divided equally and randomly in to four groups, as follows:

Group (1) - standard normal control group consists of (6) males rats treated orally with 2 ml of normal saline for 7 days.
Group (2) - diabetic control group consists of (6) males rats that were injected intra peritoneal (I.P.) with (150mg/kg) dissolved in 1/2 ml of alloxan for induction diabetes.
Group (3) - diabetic – insulin group consists of (6 induced diabetic rats)treated with i.p injection of (6 units /kg/day) insulin.
Group (4) - diabetic – colostrum group consists of (6 induced diabetic rats) treated orally with 2 ml/day from camels colostrum from the first age group of camel (4-6) years for 7 days.

Experiment two: was conducted to investigate the effect of the treatment with virgin and multipara milk for 30 days. Forty two (42) male rats were divided equally and randomly in to seven groups, as follows:

Group (1) - standard normal control group consists of (6) males rats treated orally with 2 ml of normal saline for 30 days.
Group (2) - diabetic control group consists of (6) males rats that were injected intra peritoneal (I.P.) with (150mg/kg) dissolved in 1/2 ml of alloxan for induction diabetes.
Group (3) - diabetic – insulin group consists of (6 induced diabetic rats)treated with i.p injection of (6 units /kg/day) insulin.
Group (4) - diabetic – virgin camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the virgin camel milk for 30 days.
Group (5) - diabetic – multipara camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the multipara camel milk for 30 days.
Group (6) - virgin camel milk group consists of (6) rats treated orally with 2 ml/day from the virgin camel milk for 30 days.
Group (7) - multipara camel milk group consists of (6) treated orally with 2 ml/day from the multipara camel milk for 30 days.

Experiment three: was conducted to investigate the effect of the treatment with virgin and multipara camel milk for 60 days. Fifty four (54) male rats were divided equally and randomly in to nine groups, as follows:

Group (1) - standard normal control group consists of (6) males rats treated orally with 2 ml of normal saline for 60 days.
Group (2) - diabetic control group consists of (6) males rats that were injected intra peritoneal (I.P.)
with (150mg/kg) dissolved in 1/2 ml of alloxan for induction diabetes. 
Group (3) - diabetic – insulin group consists of (6 induced diabetic rats)treated with i.p injection of ( 6 units /kg/day) insulin. 
Group (4) - diabetic – virgin camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the virgin camel milk for 60 days. 
Group (5) - diabetic – multipara camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the multipara camel milk for 60 days. 
Group (6) - diabetic – virgin camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the virgin camel milk for 60 days , killed after 30 days from stopping treatment . 
Group (7) - diabetic – multipara camel milk group consists of (6 induced diabetic rats) treated orally with 2 ml/day from the multipara camel milk for 60 days , killed after 30 days from stopping treatment . 
Group (8) - virgin camel milk group consists of (6 rats) treated orally with 2 ml/day from the virgin camel milk for 60 days. 
Group (9) - multipara camel milk group consists of (6 rats) treated orally with 2 ml/day from the multipara camel milk for 60 days. 

The weights of animals were measured on the first and last day of the treatment, the weight difference is calculated. 
Six male rats' of each group were sacrificed at the end of the experiment at Day 7, 30, 60 and one month after stopping of treatment in group 5 and 6. , the rats before being sacrifice were first weighed then anaesthetized by placing them in a closed jar containing cotton sucked with chloroform . Rats fasting all night before sacrificing. 
Blood samples were collected via cardiac puncture. Then the blood sample were drops directly from the heart by using 5 ml disposable syringe . The blood sample were divided in to two part. The first part about 1 ml was put in tube with EDTA for hematological study which were carried out as soon as possible. 
The other parts of blood was transferred in to a clean tube , left at room temperature for 15 minutes for clotting, centrifuged at 3000 rpm for 15 minutes, and then serum was separated and kept in a clean tube in the refrigerator at ( 2-8°C) until the time of biochemical assay. 
All blood samples were collected in the morning (8.30 -10.30 am) in order to minimize the diurnal variation of hormone levels. 

RESULTS

Effect of colostrum on serum glucose and body weight of control and experimental groups of male rats

According to the results listed in table (1) , there was significant increase in level of serum glucose in the (diabetic group) at (p < 0.05 ) compared with control and other all groups of experiment , while the results showed that there was no significant differences in the level of serum glucose between the control and diabetic group treated with insulin but they revealed a significant decrease at (p < 0.05) with diabetic group treated with colostrum, which showed a significant decrease compared with diabetic group. 
The differences in body weight between the experimental group showed significant decrease in body weight in the diabetic group compared with other groups ( p < 0.05), while there were no difference between the control and diabetic group treated with insulin but the lost showed no differences with diabetic group treated with colostrum.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum glucose (mg/dl) Mean± S.D</th>
<th>Weight (mg/dl) Mean± S.D</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control group treated with 2 ml DW a day for 7 days</td>
<td>96.50± 5.75</td>
<td>87.8± 0.84</td>
</tr>
<tr>
<td>Diabetic control group (7 days)</td>
<td>201.0± 6.03</td>
<td>3.56± 0.55</td>
</tr>
<tr>
<td>Diabetic group treated with insulin (7 days)</td>
<td>78.91± 2.01</td>
<td>6.55± 0.46</td>
</tr>
<tr>
<td>Diabetic group treated with colostrum (7 days)</td>
<td>118.16± 6.08</td>
<td>6.15± 0.91</td>
</tr>
<tr>
<td>LSD</td>
<td>17.38</td>
<td>2.23</td>
</tr>
</tbody>
</table>

** refer to significant differences at (p≤0.05).

Effect of virgin and multipara she camel on serum glucose and body weight of control and experimental groups of male rats during 30 days of treatment

Table (2) shows a significant increase at ( p < 0.05 ) in the level of serum glucose in the diabetic group compared with control and other all groups . On the other hand there were no significant differences between the control group and diabetic group treated with virgin she camel milk , standard control treated with virgin and multipara she camel milk , all of them decrease significantly at ( p < 0.05 ) compared with diabetic group treated with multipara she camel milk . 
As illustrated in table (2) , the differences in the body weight showed a significant decrease in the diabetic group compared with all other groups, whereas , the diabetic group treated with insulin and diabetic group treated with insulin and diabetic group treated with virgin she camel milk revealed no significantly differences between them . And increase significantly compared with diabetic group.
treated with multipara camel milk, but decrease significantly at (p < 0.05) compared with normal control group standard control group treated with virgin she camel milk and standard control treated with multipara she camel milk, but the last showed significant decrease compared with control group that showed no significant differences with standard control group treated with virgin camel milk. The two standard control groups, virgin and multipara had no significant differences between them.

Table (2): Effect of 30 days treatment of virgin and multipara she camel milk on glucose serum and body weight of control and experimental groups of male rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum glucose (mg/dl) Mean± S.D</th>
<th>Weight (mg/dl) Mean± S.D</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control group treated with 2 ml DW/day (30 days)</td>
<td>100.75 ± 59.33</td>
<td>11.60 ± 5.71</td>
</tr>
<tr>
<td>Diabetic group (30 days)</td>
<td>296.66 ± 20.01</td>
<td>37.32 ± 2.09</td>
</tr>
<tr>
<td>Diabetic group treated with insulin (30 days)</td>
<td>116.60 ± 35.68</td>
<td>45.56 ± 2.58</td>
</tr>
<tr>
<td>Diabetic group treated with virgin she camel milk (30 days)</td>
<td>106 ± 35.11</td>
<td>45.17 ± 4.94</td>
</tr>
<tr>
<td>Diabetic group treated with multipara she camel milk (30 days)</td>
<td>192.66 ± 25.70</td>
<td>19.40 ± 2.97</td>
</tr>
<tr>
<td>Standard control group treated with virgin she camel milk (30 days)</td>
<td>101.24 ± 56.25</td>
<td>40.67 ± 6.46</td>
</tr>
<tr>
<td>Standard control group treated with multipara she camel milk (30 days)</td>
<td>94.78 ± 52.50</td>
<td>15.53 ± 4.08</td>
</tr>
<tr>
<td>LSD</td>
<td>52.45 ± 5.68</td>
<td></td>
</tr>
</tbody>
</table>

** refer to significant differences at (p≤0.05)

Table (3): Effect of 60 days treatment of virgin and multipara she camel milk on serum glucose and body weight of control and experimental groups of male rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum glucose (mg/dl) Mean± S.D</th>
<th>Weight (mg/dl) Mean± S.D</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control group treated with 2 ml DW/day (60 days)</td>
<td>111.0 ± 66.33</td>
<td>23.40 ± 12.37</td>
</tr>
<tr>
<td>Diabetic control group (60 days)</td>
<td>271.50 ± 8.16</td>
<td>63.88 ± 2.22</td>
</tr>
<tr>
<td>Diabetic group treated with insulin (60 days)</td>
<td>110.76 ± 94.00</td>
<td>48.35 ± 15.60</td>
</tr>
<tr>
<td>Diabetic group treated with virgin she camel milk (60 days)</td>
<td>87.66 ± 97.50</td>
<td>4.92 ± 12.92</td>
</tr>
<tr>
<td>Diabetic group treated with multipara she camel milk (60 days)</td>
<td>104.83 ± 88.66</td>
<td>12.18 ± 9.37</td>
</tr>
<tr>
<td>Diabetic group after one month form stopping 60 days treatment with virgin she camel milk</td>
<td>127.75 ± 44.83</td>
<td>55.97 ± 7.13</td>
</tr>
<tr>
<td>Diabetic group after one month form stopping 60 days treatment with multipara she camel milk</td>
<td>169.05 ± 39.31</td>
<td>37.81 ± 2.93</td>
</tr>
<tr>
<td>Standard control group treated with virgin she camel milk (60 days)</td>
<td>79.00 ± 104.16</td>
<td>10.41 ± 12.81</td>
</tr>
<tr>
<td>Standard control group treated with multipara she camel milk (60 days)</td>
<td>94.66 ± 99.33</td>
<td>15.48 ± 12.80</td>
</tr>
<tr>
<td>LSD</td>
<td>48.75 ± 15.50</td>
<td></td>
</tr>
</tbody>
</table>

** refer to significant differences at (p≤0.05)

Effect of virgin and multipara she camel treatment on serum glucose and body weight during 60 days of treatment

The results in the table (3) indicated a significant increase at (p < 0.05) in the level of serum glucose in the diabetic group compared to (normal control and), other experimental group, while the 3rd, 4th, 5th, 6th, 8th and 9th groups did not revealed significant differences in comparison with control group and between them, but the 7th group showed a significant increase in the level of serum glucose in compared with normal group, 3rd, 4th, 5th, 8th and 9th groups, while did not reported any significant differences with 6th group. Body weight differences through 60 days of treatment clarified on table (3), diabetic group showed a significant decrease in body weight compare with control and other groups, the result show that 3rd, 4th, 8th and 9th groups reported not significant decrease in body weight compared to control group and between them while the 5th, 6th and 7th group showed a significant decrease compared to control one, but the 6th and 7th group indicate a significant differences compared with 5th group but there was non significant differences between them, while there is no significant differences between the 3rd, 4th and 5th groups.
**DISCUSSION**

Effect of colostrum and camel milk on serum glucose body weight in male rats

The level of glucose increase significantly in the diabetic groups of the three experiment when compared to control groups due to induction the diabetes by using alloxan in laboratory male rats (1). Thus, due to alloxan are toxic analogues preferred to accumulate in pancreatic bat a cell via the glucose transporter GLUT2. Intracellular the alloxan generates reactive oxygen species (ROS) in presence of glutathione, in a cyclic redox reaction diatomic acid, the reduction product, generates superoxide radicals, hydrogen peroxide radicals, hydrogen peroxide and hydroxyl radicals, which are ultimately responsible for the death of beta cell. Alloxan also have the ability to inhibit the beta cell glucose sensor glucokinase and that inhibits glucose-induced insulin secretion (2,3), which caused glucose to stop entering cell and this rise of glucose level in the blood (4,5). Oral treatment with colostrum and camel milk leads to a significant decrease in serum glucose when compared to diabetic groups. Colostrum hypoglycemic effect is suggested to be mediated by present of vitamin E, which are work by scavenging the reactive oxygen species and caring then a way through chemical neutralization albumin that tend to be sacrificial protein can absorb (Ros) and stop them from attacking your essential protein and colostrum also contain &- LA (Alpha-lipoic Acid) which are great modifier act on gene expression to reduce inflammation, very potent chelator for heavy metal and the last important acts as enhancer of insulin sensitivity (6,7).

The mechanism for the hypoglycemic effect of camel milk against diabetics oxidative stress in the diabetes is due to contain high levels of insulin or insulin-like protein (52 units/liter) insulin. Results obtained from studies of (8-10) found that camel milk is rich in half cystine (amino acid sequence of some camel milk protein) similar insulin family of peptides superficially (10). Thus, it is considered to have therapeutic efficacy due to lack of coagulum formation of camel milk in acidic media of stomach to pass rapidly to intestine and remains available for absorption (10). Hypoglycemic effect of CM attributed effect by it is potential to rather increase the effect of insulin or by increasing the release of insulin from the pancreatic bet-cells (14), the protective effects of camels milk also including antioxidant properties and possible chelating effects on free radicals, it is was found to contain high concentration of vitamins B2, C, A and E, and very rich in magnesium and other trace elements (11-13). These vitamins are useful in preventing toxicant- induce tissue injury (14). Additionally, camel milk is rich in zinc (15), a trace element essential for more than 300 enzymes activity and it has a relation ship with many enzymes and can a activation of anti oxidant system so prevent cell damage (16-18). Body weight decrease in diabetic rats in second group due to loss or degradation of structural protein because of lack of insulin due to diabetes and this lead to lack of entry glucose in to cell which increase protein catabolism. In addition in such circumstances as absence of insulin excess of carbohydrates did not turn to fats (19-21). Another research suggested that lack of insulin lead to sugar cannot enter inside the cell, thus increasing sugar percentage in the blood, so the body tries to get rid the excess amount of sugar by excretion in the urine, thus lead to reduced amount of water in the body and this causes body weight reduction (22). Rats treated with colostrum and CM observed a significant improvement in body weight gain the posispolity of good nutrition value. Anabolic effect of colostrum and CM via strengthening the gastrointestinal tract through increasing both the rate of secretions of the digestive juices and the motility of the gastro intestinal tract (23), or may be due to CM stimulation most aspects of carbohydrate metabolism including glucose rapid up take by the cells, increase rat absorption from gastro intestinal metabolism and even increase insulin secretion and it is effect on carbohydrate metabolism (reduce hyperglycemia) (10, 23).

Isa et al (2013) suggested there were no statistical difference in the body weight on the control, diabetic, diabetic rats treated with CM (24).

**REFERENCES**


17. Ozturk NZ.; Talamé V.; Deyholos M.; Michalowski CB.; Galbraith DW.; Galbraith NG. et. al. (2002). Monitoring large-scale changes in transcript abundance in drought- and salinity-stressed barley. Plant Molec. Biol. 48(5): 551-573.


23. Mohamed AZ.; El-Asmar AZ.; Rezq AM.; Mahfouz SM.; Wassef MA.; Fouad HH.; Ahmed HH. and Taha FM. (2013). The effect of a novel curcumin derivative on pancreatic islet regeneration in experimental type-1 diabetes in rats (long term study). Diabetol. Metab. Synd. 5:75-81.

Histological effects of repeated administration of Ivermectin alone or with the combination of vitamin C on ovaries and uterus of female rabbits

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ABSTRACT

This study was done to investigate the histological effect of repeated administration of Ivermectin alone or with combination of vitamin C in ovaries and uterus of female rabbits. Total 48 mature female rabbits were divided into 8 groups of equal number (6) as follows: group one was administered 0.9 % Nacl, which act as control. Group two, three and four were administered (0.5mg /Kg, 1mg/Kg, 2mg /Kg B.W Ivermectin) respectively. Group five was administered 50mg/Kg B.W vitamin C only. Group six, seven and eight were administered 50mg/Kg vitamin C in addition to Ivermectin (0.5mg/Kg, 1mg/Kg, 2mg/Kg) respectively. The Ivermectin was given S/C weekly, while vitamin C was given daily P.O. for 8 weeks.

The results showed the histopathological changes in ovaries in all groups which treated with intermediate and high doses of Ivermectin (1mg/Kg and 2mg/Kg) includes secondary cystic follicles, circumscribe and proliferative corpus luteum, and large corpora lutea which is suggested neoplastic as luteoma. While the group which received vitamin C showed normal structure of ovaries with normal primary and secondary follicles with corpus lutum. In uterus, the main histological changes due to ivermectin treatment includes poor activity of uterus, while the histopathological effect of Vitamin C on uterus include well develop uterine horn with papillary mucosal cell epithelium.

In conclusion, Ivermectin exhibits dose response effect and causes many deteriorate histological changes in ovaries and uterus in female rabbits in intermediate and high doses, meanwhile, vitamin C has not well ameliorative these effects positively.

Keywords: vitamin C, Ivermectin, female rabbits

الملخص باللغة العربية

هدفت هذه الدراسة إلى تحديد الآثارات النسيجية للجرع المتكررة من عقار الإيفرمكتين منفرداً أو مع فيتامين ج على مبيض ورحم إناث الأرانب. شاملت الدراسة 48 أنثى أرنبية، وتم تقسيمهم إلى 8 مجموعات، أجريت كل مجموعة على 6 آرانب. جرعت المجموعة الأولى بجرعة 0.5% من محلل الملح الفسيولوجي، واعتبرت كمجموعة سلطة. وأعطت المجموعات две، ثلاث ورابعة الإيفرمكتين بجرعة 0.5، 1، 2 ملغ/كم من وزن الجسم. وأعطيت المجموعة الخامسة بجرعة 50 ملغ/كم من وزن الجسم فيتامين ج، بينما أعطيت المجموعات السادسة، السابعة، الثامنة فيتامين ج 50 ملغ/كم من وزن الجسم بالإضافة إلى الإيفرمكتين (0.5 ملغ/كم، 1 ملغ/كم و 2 ملغ/كم من وزن الجسم على التوالي. أعطيت الإيفرمكتين تحت الجلد أسبوعياً، بينما أعطيت فيتامين ج يومياً عن طريق الفم لمدة 8 أسابيع.

أظهرت النتائج حدوث تغيرات إرئائية نسيجية بписываوية في جميع المجموعات المعالمة بالإيفرمكتين بجرعات الوسطى والعالية (1 ملغ و 2 ملغ) وبدون فيتامين ج، حيث تحدث تغيرات نسيجية تحتوي على الوفيات الشاذة، وظهور جسم أصفر متطاير وحيد، وغير واضح. وعند استخدام فيتامين ج، كما ما تبين من النتائج، حدوث تغيرات نسيجية في الرحم نتيجة استخدام الإيفرمكتين، تتمثل بقطر شاذ الرحم، بينما أظهر فيتامين ج تغيرات تطعت تطوراً جديداً في الرحم، وتحل في نتائج هذه الدراسة أن الإيفرمكتين له تأثيرات تختلف باختلاف الجرعة المستخدمة، حيث تسببت العديد من التغيرات النسيجية بالياض، وفي الرحم لإثاث الأرانب في الجرع المتوسطة والعالية، بينما لم يكن استخدام فيتامين ج لتلك التغيرات بصحة إيجابية.
INTRODUCTION

The reproductive organ of the female rabbit’s considered primitive. It consists of two ovaries and oviduct, uterus, two cervixes, vagina, and external genitalia (1). The primary organs of female reproduction are ovaries which lies within the abdominal cavity with one on each side near the kidney and responsible for egg or ova production, as well as, hormones primarily Estrogen and Progestin (2).

The uterus is formed by two independent horns split over their, and it considers the organ in which the embryonic development occur also it provides muscular force expulsion of the fetus at birth (3). Ivermectin is broad spectrum antiparasitic drug belongs to family of Avermectins which includes Abamectin, Emamectin, Eprinomectin and doramectin. It is produce by the fermentative process of Streptomyces avermilitis microorganism. Ivermectin is administered in different routes such as orally, subcutaneous, pour on formulation (4). It can be given chewable treats, oral (liquid, pastes, and drenches) and it can take as topical solution for treating mites (5).

Hays and Law (6) concluded that ivermectin was moderately absorbed after oral administration, while it is well absorbed after subcutaneous or intramuscular route. Many studies demonstrated the harmful effects of ivermectin or similar avermectin family on semen quality, characteristic and testosterone level in rabbits (7,8), in goat (9), in ram (10), rats (11), bull (12), and man (13).

Abd-Elhady and Abo-Elgar (14) showed the abamectin caused marked histopathological changes in the testes of the rats which includes degeneration and necrosis of spermatogonia cell lining the seminiferous tubules, and decrease in the number of spermatocytes. Also, Elbeticha and Daas (15) observed an increased amount of interstitial connective tissue of seminiferous tubules and congested blood vessels of the rat testes after treatment with abamectin. Little papers are concerning the histopathological effect of ivermectin on female reproductive system. So this study was aimed to investigate the adverse effect of ivermectin on ovary and uterus structures and also study the role of vitamin C when used in combination of ivermectin of female rabbits.

MATERIALS AND METHODS

The Ivermectin 1% was purchased from local market (VET Product Office, KIPRO Company, Holland) and Vitamin C 500mg (Al-Shahba Labo, Syria).

Animal housing

Total (48) mature female rabbits (Lepus curriculus), (1200-2000gm) body weight and (8-12 months) of age were brought from local market in Basra Province/ Iraq. The rabbits were housed (6 rabbits / cage) in a wire silk cages measuring (100 X 50 X 50 cm) under controlled animal house condition at temperature (25 ± 3 °C) and relative humidity (50 ± 5 %) in the animal house of Veterinary Medicine College in Basra University. The rabbits were kept under observation for one months. The animals were offered a rabbit’s diet, green leaves, alfalfa, and water.

Experimental design and histological technique

Forty eight (48) mature female rabbits were divided into eight groups (6 rabbits in each group). Each group was treated for 8 week as follow: Group one: Injected (0.9 % NaCl) which acts as a control, Group two: Injected (0.5 mg/kg B.W Ivermectin), Group three: Injected (1 mg/kg B.W Ivermectin), Group four: Injected (2 mg/kg B.W Ivermectin), Group five: administered 50mg/kg B.W Vitamin C only, Group six: Injected (0.5 mg/Kg B.W Ivermectin +50mg/kg B.W Vit.C), Group seven: Injected (1mg/kg B.W Ivermectin +50mg/Kg B.W Vit. C), and Group eight: Injected (2mg/Kg B.W Ivermectin +50mg/kg B.W Vit. C). The Ivermectin were given subcutaneously and weekly, while vitamin C were given daily and orally. At the end of experiment (8 Weeks) after the animals were sacrificed, the organ samples were taken out ovary and uterus. Then the organs were fixed in10 % formalin as preservation. The preserved organs were taken and the dehydration was done by passing specimens in ascending concentration of Ethanol, infiltrated with xylene and then was embedded in paraffin. Five-micron thick sections of paraffin-embedded tissue were cut by using microtome and mounted on glass slides, then affixed of ribbon by Mayer’s albumin on glass slide, then dehydrated at hot plate overnight and later stained with hemotoxylin-eosin stain. The sections were examined by using light microscope (16).

Statistical analysis

The results were analysed by one- way ANOVA test. All statistical calculations were carried out by the aid of the statistical SPSS V. 22 (SPSS Inc.). Least significant different test (LSD) was calculated to test the differences between means.

RESULTS

The microscopic findings of ovaries of control rabbits showed numerous primary and secondary follicles, and corpora lutea (figure 1). The histological examination of ovaries in the 2nd group showed lack of graffian follicles, presence of occasional secondary follicles (figure 2), while the 3rd group revealed cystic secondary follicles (figure 3). The 4th group showed very large corpora lutea which is suggested neoplastic as luteomal (figure 4), cystic secondary follicles (figure 5). The 5th group
showed primary and secondary follicle and corpus lutum (figure 6). Whereas in 6th group revealed large corpus lutum and secondary cystic follicle (figure 7). The 7th group showed circumscribed corpus lutum proliferated luteoma like (figure 8). Moreover, the 8th group showed secondary follicles and circumscribed corpus luteum (Luteoma like) (figure 9).
The examination of uterus of control rabbits showed well developed endometrium with papillary proliferation of mucosal epithelium of endometrium (figure 10). The 2nd and 3rd groups showed papillary projection of uterine mucosal epithelium, uterus poor activity (figures 11, 12), while the 4th group revealed uterine horn with papillary mucosal cell epithelium (figure 13). The 5th group showed well developed uterine horn with papillary proliferation of endometrial epithelium (figure 14). The 6th group revealed apparent reduction in development of endometrial epithelium (figure 15). In addition, the 7th group showed slight papillary projection of uterine mucosa (figure 16), and in the 8th group showed poor or reduce development of endometrial epithelium (figure 17).
Figure (11): Uterus of rabbit treated 0.5mg/Kg Ivermectin (H&E) X50. The pointer indicate papillary projection of uterine mucosal epithelium, but reduce number of mucus gland in lamina propria.

Figure (12): Uterus of rabbit treated 1mg/Kg Ivermectin (H&E) X50. The pointer indicate papillary projection of uterine mucus epithelium.

Figure (13): Uterus of rabbit treated 2mg/Kg Ivermectin (H&E) X50. The pointer indicate uterine horn with papillary mucosal cell epithelium.

Figure (14): Uterus of rabbit treated 50mg/Kg vitamin C (H&E) X50. The pointer indicate uterine horn well develop with papillary proliferation of endometrial epithelium.

Figure (15): Uterus of rabbit treated with 0.5mg/Kg IVM+50mg/Kg vit.C (H&E) X50. The pointer indicate apparent reduction in development of endometrial epithelium.

Figure (16): Uterus of rabbit treated with 1mg/Kg IVM+50mg/Kg vit.C (H&E) X50. The pointer indicate slight papillary projection of uterine mucosa.
In the present study, the poly cystic follicles, and circumscribe corpus lutum (luteoma like) due to administration of ivermectin may be due to hormonal disturbance and/or free radical production due to ivermectin treatment. The control of ovarian stromal cells and germ cell function is a diverse prototype and oxidative stress may be acts as one of the modulators of ovarian germ cell and stromal cell physiology (17). It well documented that ovarian follicular development is regulated primarily by the differential and coordinated action of the two primary gonadotropins FSH and LH (18).

Moreover, Vegetti and Alagna, (19) found the FSH considered the principle driving force of folliculogenesis, at least beyond the stage of the preantral follicle. Some researchers found that LH could be considered as stimulator to early stages of follicular growth in ovary (20, 21). The follicle and luteal cyst in the ovaries are so common place as almost to constitute physiological variants. Theses innocuous lesion originate in un rupture graffian follicles or in follicles that have rupture and immediately sealed (22). Polycystic ovary syndrome is associated with decrease antioxidant concentration and is thus considered an oxidative state (23). Al-Gubory, et al( 24)clarified the excessive production of reactive oxygen species may overpower the body natural antioxidant defence system which is creating an environment unsuitable for normal female physiology reaction.

The results of current study is in accordance with (25) who postulated that the uses of ivermectin in overdose caused primary and mature follicles were decrease in number, as well as, hyperplasia of ovarian follicles, atretic follicles, follicular cysts and ovarian atrophy in rats and mice. Some authors found the I/M injection of Doramectin at (0.4mg, 0.6mg/Kg) for 6 weeks to guinea pigs caused degeneration of the oocytes of the small growing follicles and damage of mature follicle in ovaries at fourth dosage, whereas after fifth and sixth weeks of the treatment the ovaries seemed with excess atretic and damage growing follicles(26). On the other states, vitamin C is very important in remodelling the basal membrane during follicular growth and the ability of follicles to uptake ascorbic acid confers an advantage in term of granulosa cell survival(27). Another investigators noticed the administration of vitamin C caused increase in graffian follicle, congestion of blood vessels of the ovaries, as well as, it caused ameliorative effect on the ovary toxicity due to of KMnO4 therapy in rats (28).

In present study, the microscopic finding in uterus due to ivermectin therapy revealed papillary projection of uterine mucosal epithelium, reduce number of mucus gland in lamina propria, poor activity, and vacuolation in mucosal epithelial cell. This may be explained due to hormonal disruption especially estrogen which one of its function responsible for stimulation of the growth of endometrium (29). Actually, our previous work on ivermectin which showed a decrease in Estrogen level (30).

The results of this study is in tone with (26) who showed the injection of 0.4mg and 0.6mg/Kg of Doramectin I/M for 6weeks to ginea pigs caused adverse effect in uterus which appear from the 3 doses of administration which characterized by infiltration of endometrium and aggregation of mononuclear cell with congestion of perimetrial blood vessels, while from 4th doses to the end of experiment, the endometrial gland has been degenerated, atrophied and widely separated by the oedema with cellular reaction. In fact, the ivermectin therapy when given in overdoses to rats and mice caused an increase prevalence of uterine glands, uterine epithelia and hyperplasia of gland were markedly presented on diverse patterns adenoma like structure and single nodular or multiple polyp like adenoma. In present study the microscopic finding of uterus in group which administered vitamin C only has been proved well develop uterine horn with papillary proliferation of endometrial epithelium this occur may be due to protective effect of vitamin C (25).

In conclusion, Ivermectin exhibits dose response effect and causes many deleteriate histological changes in ovaries and uterus in female rabbits in intermediate and high dose, meanwhile, vitamin C has not well ameliorative these effects.

Acknowledgment:

The researcher would like to express my deep thanks to Professor Dr. Salih Khadim Majeed / Department of Pathology, Veterinary Medicine College in Basra University for his help in the histopathological reading of the slides.
REFERENCES

Molecular study of origin of Newcastle virus isolated in Iraq

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ABSTRACT

Newcastle disease (ND) caused by virulent avian paramyxovirus type 1 is responsible for devastating outbreaks in poultry flocks in Iraq. In this study, isolates obtained from ND suspected outbreaks during 2014-2016 of layers, broilers, farm and wild pigeon were characterized by intracerebral pathogenicity index (ICPI) and genetically on the basis of molecular analysis of the partial F gene sequences. The ICPI value of Iraqi pigeon isolate 1.4 and Iraqi chicken isolate 1.9-1.8. The mean death time (MDT) of NDV isolated from chicken and pigeon were 35-36, and 48 hours respectively. These are indicative of the velogenic nature of these NDV Iraqiisolates. The partial F gene sequencing of chicken/ Iraqi/2/ 2016 isolate showed identity of 98% to Newcastle virus chicken china /Jilin /YS03/ 2015, while the chicken/ Iraqi/ 2015 isolate showed identity of 87% to Newcastle virus chicken Iran /HG/ 2010. showed identity of 96% of the pigeon/ Iraqi /2/2014 to NDV pigeon / Russian /Moscow 407/04 and also showed identity of 85% to the chicken china /SD4/ 2008, while chicken/ Iraqi /4 / 2014 showed identity of 85% to the Newcastle virus chicken china SD4 / 2008. Based on the high nucleotide similarity between these Iraqi NDV isolates and the Chinese NDV strain indicates that the origin is Chinese. The similarity between viruses obtained from chicken and those obtained from wild pigeon ranged from 82-85% so further study will be useful to understand the role of wild birds in the epidemiology of NDV in Iraq.

Keywords : NDV, Chicken, Pigeon, Sequence
INTRODUCTION

Newcastle disease (ND) is highly contagious and devastating disease of poultry caused by Newcastle Disease Virus (NDV), also known as Avian paramyxovirus type -1 (APMV-1), it is classified under the genus Avulavirus of family paramyxoviridae (1). Based on genomic size and nucleotide sequences of F and L gene NDVs can be categorized as class I or class II viruses (2). Class I NDVs are occasionally isolated from wild aquatic birds and domestic poultry are mostly a virulent to chickens. Class II contains viruses that have been isolated from multiple wild birds and poultry species. Most virus within this group are virulent and cause significant economic losses to poultry industry worldwide (2).

The key contributor to ND pathogenicity is the formation of an active fusion protein upon cleavage of F protein precursor (F0), which facilitated by the presence of a number of basic amino acid residues in fusion protein cleavage site (3). NDVs that are virulent for chickens have a multiple basic amino acid sequence 112 R/K-R-Q-K/R-R 116. At C terminus of F2 protein and F (phenylalanine) at residue 117 which is the N-terminus of F1 protein, whereas the viruses of low virulence have a monobasic amino acid sequence in the same region of 112 G/E-K/R-Q-G/E 116 and L (Leucine) at residue 117 (4).

MATERIALS AND METHODS

Isolation and characterization of NDV variants

Samples were collected during NDV outbreaks in Baghdad area in poultry farms and wild pigeon. Tissue samples from dead birds were collected from suspected outbreaks around Baghdad province, samples from pigeon were also collected from suspected ND outbreaks in wild pigeons around the same area. Collected organs (trachea, lungs and brain) processed for virus isolation according to the standard virus isolation methods (5). Briefly 10% w/v organ suspension in phosphate buffer saline (PH 7.2) were homogenized, clarified by centrifugation and inoculated into 9 days old embryonated chicken eggs into amallantoic cavity. Inoculated eggs candled every 24 hours. Eggs containing dead embryo were removed from incubator and chilled at + 4°C for night. Allantoic fluid were harvested and tested by haemagglutination assay. The HA and HI assays were carried out according to the manual of world organization for animal health (5). Inactivated NDV antigen (LaSota) strain was used as positive control. Intracerebral pathogenicity index (ICPI) test was carried out in day old chicken according to (6). The mean death time (MDT) of the NDV isolates were performed according to the published protocol of (7).

Molecular characterization

Viral RNA was extracted using the viral gene – spin viral DNA/RNA extraction kit (Intron, Biotechnology, Korea). Firstly, tracheal, lungs and Caecaltissue were homogenized. Two RT-PCR assay targeting the fusion gene (traditional and nested PCR) to identify the type of NDVs strains was carried out (9,10).

The NDV primers for the traditional RT-PCR NDV are 5°-GGAGGATTTGGCAGCATTT-3° and NDVD 5° GTCAACATATACACCTCATCC-3° were used to give RT-PCR product of 318 bp (Stauboretet et al1995) (figure 1).

Two sets of primers were used for first and second round of NDV nested RT-PCR. The first round primer was used:

5°-GCAGCTCGAGGGATTGATGTGTTG-3° and
5°-TCTTTGACGAGGAGGTGTTG-3° and for second round the primers were
5°-CCCGGTGAGGACATAC-3° and
5°-TGTGAGACGAATTTGATTG-3°.

The nested RT-PCR product of 216 bp (9,10).

Sequencing

The purified RT-PCR products were sequenced using ABI Prsm 310 genetic analyzer– Jovac – Jordan (applied biosystem USA). Neoclutide sequences were exported to Bio Edit. Serotype reference sequences were obtained from the NCBI Genbank (www.NCBI.nlm.nih.gov) multiple sequence alignment were made using clustal W multiple sequence search also available from NCBI (11). Percentage of nucleotide similarity was calculated using DNA STAR software phylogenetic analysis was done using phyml software.

RESULTS

Four haemagglutinating isolates were obtained from poultry farms outbreak that reacted with monospecific antiserum specific for avian paramyxovirus, and were identified as Newcastle disease virus by HI test. Besides all the four isolates were confirmed positive for virulent NDVs. The ICPI value of the chicken / Iraq /2/2016, chicken/ Iraq 3/2015, chicken /Iraq /4/2014 and pigeon/ Iraq /12014 were 1.9, 1.8, 1.8 and 1.4 respectively while the mean death time (MDT) of the isolates were 35, 36, 36, 48 hrs. respectively (table 1).

RT-PCR product of 318 bp (figure 1) for the first round of NDV nested RT-PCR. The nested second round RT-PCR product of 216 bp was obtained (data not shown).

Phylogenetic analysis of partial F gene sequences showed percentage of nucleotide similarity between the viruses from chickens and those from pigeons ranged from 82-86%. Was calculated using DNA STAR software (figure 2).
Sequencing analysis of sample isolate compared with highly related identity by using blast software program showed identity of 96% of the pigeon/Iraqi/1/2014 to NDV pigeon /Russian/Moscow 407/04 accession number JF824018.1 and also showed identity of 85% to the chicken /Iraqi/ 4 / 2014 showed identity of 85% to the Newcastle virus chicken china SD4 / 2008 / access no. HM748947.1.

The partial F gene sequencing of chicken Iraqi to 2016 isolate showed identity of 98% to Newcastle virus chicken china /Jilin/YS03/2015 accession no. KU200238.1, while the chicken Iraqi 2015 isolate showed identity of 87% to Newcastle virus chicken Iran /HG/ 2010 accession no. JX131352.1.

Figure (1): Agarose gel electrophoresis PCR amplification of F protein gene (318bp) of NDV M ladder of 100 bp. negative control, positive control well 1 - 6 NDV chicken Iraqi isolates well 7 NDV pigeon Iraqi isolates

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Table (1): ICPI, MDT and the related virus on gene bank

Figure (2): Percentage of nucleotide similarity of the 4 most closely related Iraqi NDV isolates was calculated using DNA STAR software
DISCUSSION

ND is an endemic disease in Iraq since 1968. AG 68 L was the isolated vaccine strain. The immunity of some Iraqi isolated were studied by (12). Due to uncontrolled procedure of importing vaccines and biosafety, four new isolates were characterized indicating different origin (China, Iran, and Russia). But could be the same clad, China origin isolated MDT(45 hrs) while the similar characterized Iraqi isolated 2016 MDT was (35hrs) indicating it is higher virulent than the original one. Also, ICPI showed the same virulence (1.9), while the original isolate was (1.8). Other isolates showed the same high virulence compared with the original isolates (table 1), which in turn indicated how ND endemic became in Iraq which should be of higher mortality and long term of endemicity. Available commercial vaccine cannot protect incidence of ND due mostly to these change in virus virulence. Pigeon isolate has showed identity between Russian (96%) and china strain (85%) this could point to the responsibility of pigeon isolate in transport infection to chicken’s farms. Detection of multiple amino acids is an indication of virulence (4), this procedure could not be done due to lack of facilities, but ICPI could be a substituted. Failure to demonstrate the characteristic pattern of amino acid residues between residue 113 and 116, would require characterizations of the isolated virus by an ICPI test (13).

Vaccination with Hitchner B1 and LaSota strain belong phylogenetically to the same genotype (I and II) and are divergent from NDVs that cause the current outbreaks, ND outbreaks could occur despite intensive vaccination against NDV (14,15). This might be associated with genetic and antigenic divergence between vaccine strain and the circulating field strain (16,2).

Acknowledgement

The researcher would like to thank Miss Hazar Shaweesh for her sequencing assistance at Jovac company, Jordan

REFERENCES

قسم الدراسات والبحوث العربية

ARABIC STUDIES AND RESEARCHES SECTION
The culture of applying internet in academic and scientific research: Al- Taref University as a model

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Dept. of Sociology / Al- Taref University/ Algeria

ABSTRACT

The Study For Research On How To Use The Internet In Scientific Research And Academic Researchers The University Of Chadli Bendjedid- El Tarf / Algeria, Otherwise Big Showing The Role Who Become Lucky In Him Net Of The Internet In The Scientific Research Through What His Progress From Numerous Services For Research In Domain Their Specialization, And Sample Was Formed Studious From (112) Professor And Research Student In The High Studies Distributed On Sections And Colleges, And Gathering Of The Statements Through The Use Of The Observat Are Complete, And The Questionnaire Who Phrase Forms The Questionnaire (35) The Words On Method Was Complete Her Likert In Measure Parts Are Formed The Questionnaire (05) , And Application Method Of The Interior Consistency For Distances Of The Measure Is Complete To Measuring Extension Believed Him, And The Sixteen Releasing Already Use At That Program (Spss 16) Study Has To, Inter Alia, Results, The Most Important Of The Researchers Are The Recruitment Of The Internet In The Drafting Of Chapters And Research On The Latest Developments Of The Research, Experiments And Tests Linked To The Study And That They Do So Using Search Engines And Participation In Meetings And Seminars And Networking With Specialists, And There Are Several Obstacles Preventing Them From Using The Internet In Their Researches Ranging Between Personal Difficulties And Seminars.

The Study Of The Reached A Set Of Recommendations, Including The Need To Stimulate The Researchers To Undergo Training In The Use Of The Internet, And The Areas Of The University Of Eltarf Researchers.
Integrated (Digital Network)

In the wake of the academic social media revolution, the advent of the internet has revolutionized the way scholars communicate and collaborate. In the context of academic conferences, the internet has become a vital tool for researchers, enabling them to share their findings, network with peers, and access a wealth of information. The benefits of this digital landscape are numerous, ranging from increased access to resources to new avenues for knowledge exchange. However, as with any technological advancement, the internet also presents its challenges, including the proliferation of misinformation and the need for effective digital literacy.

To address these issues, universities and research institutions are increasingly adopting digital platforms to facilitate academic research and collaboration. These platforms offer a range of features, from social media networks to specialized research databases, enabling scholars to connect with each other and access a vast array of academic resources. However, the success of these initiatives depends on the ability of researchers to adapt to the digital landscape and effectively navigate its complexities.

In conclusion, the digital landscape has transformed the academic world, offering unprecedented opportunities for collaboration and knowledge exchange. As we continue to navigate this new territory, it is essential to cultivate a culture of digital literacy and adaptability, ensuring that the benefits of the internet are realized in full.


لا يوجد محتوى قراءة طبيعي.
الفوائد من توظيف الإنترنت في البحث العلمي الأكاديمي

إن توظيف الإنترنت في البحث العلمي الأكاديمي جدير بالاهتمام لتجاوز بعض القيود التي تؤثر على فعالية البحث، وتيسير الوصول إلى الموارد والمعلومات، وتقلص الوقت، وزيادة جدارة النتائج، وتعزيز الشفافية في العملية البحثية.

1. الوصول إلى الموارد والمعلومات: تُتيح الإنترنت الوصول إلى المكتبات الإلكترونية، المجلات العلمية،和其他 resources غير متاحة في المكتبات التقليدية.

2. تقلص الوقت: يتيح الإنترنت تجنب العقبات الفنية والاجتماعية والسياسية التي قد تمنع البحث العلمي الأكاديمي، وتسهيل التوجيه والاتصال.

3. جدارة النتائج: يسمح الإنترنت بتطبيق معايير أفضلcontest، وتعزيز الشفافية في العملية البحثية.

4. التفاعلات: يوفر الإنترنت منصة لتفاعل الباحثين وتبادل المعرفة والخبرات.

5. التنوع: يتيح الإنترنت الوصول إلى مجموعة واسعة من المواقع والمتحدثين.

6. الصفحات المختلفة: يتيح الإنترنت الوصول إلى آلات بحث متخصصة، مثل Google Scholar.

7. القدرة على حفظ ومشاركة النتائج: يتيح الإنترنت الحفاظ على النتائج، ومشاركتها مع الآخرين.

8. التفاعلية: يوفر الإنترنت منصة للتواصل الفوري، مثل المكالمات الهاتفية والمراسلات.

9. السهولة: يتيح الإنترنت الوصول إلى الموارد والمعلومات بسهولة، وتحسن جودة البحث العلمي الأكاديمي.

الاستبيان:

تم توزيع الاستبيان لمدة 35 (ساعة) موزعة على (3) شعبة، وتم توزيعه على عينة الدارسة (122) نموذج، وتم توزيع الاستبانب ذات صحة محترف في المجال، وتم توزيع الاستبانب في (25) أبريل 2015، واستمرت فترة توزيع الاستبانب 20 يومًا، وتم توزيع الاستبانب في جاهزية بكالوريوس، وکان له أثر مهم على جاهزية الباحث في العمل والإنتاجية.

الاستبيان: أسئلة الاستبيان تتناول النتائج، وتناقش الفوائد والتحديات، وتحكم في العملية البحثية، وتميز بسهولة الاستخدام، وسهولة التحليل، وسهولة التوزيع.

الاستبيان: تتناول النتائج، وتناقش الفوائد والتحديات، وتحكم في العملية البحثية، وتميز بسهولة الاستخدام، وسهولة التحليل، وسهولة التوزيع.

المراجع:


النيد والانتقائية

الإجابة عن السؤال الأول والذي نصه "ما مدى استفادة الباحثين في مراجعة الأدبيات في البحث العلمي الأكاديمي التي ينجزوها؟" حيث بوضوح الجدول رقم (2) إجابات عينة الدراسة.

من الجدول رقم (2) أن جواب توقيف الإنترنت في البحث العلمي الأكاديمي يتم ترجمته وفقًا للموطن والمصادر الحاسوبية لإجابات المجموعة على الشكل التالي:

- في الجدول الأول تأتي "سياقات اتصال وتوثيق البيانات" بمنطقة حساب (0.92) ثم "العلاقة بين البيانات في البحث، الأدبيات، الإجابة" (1.49) وال"المحترف المعايير" (1.42) والإحصاءات معينة للأعمال (1.41) "الكشف والمصدر الإلكتروني" (1.40) "الإجابة الشاملة للبحث" (1.38)، وتحل "علاقة بين إجابة "التحليل الإحصائي المعمول" أهمية خاصة لدى الباحثين في تخصيصات المجموعة وذلك لأن المعايير الإحصائية تتعامل مع

جدول رقم (1): ملاحظات صد الاتصال الدائم لجامعة المقياس وتوزيعه

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النتائج والانتقائية

الإجابة عن السؤال الأول والذي نصه "ما مدى استفادة الباحثين في مراجعة الأدبيات في البحث العلمي الأكاديمي التي ينجزوها؟" حيث بوضوح الجدول رقم (2) إجابات عينة الدراسة.

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جدول رقم (2): إجابات المجموعة عن جواب توقف الإنترنت في البحث العلمي الأكاديمي التي ينجزوها

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لا يوجد نص يمكن قراءته بشكل طبيعي من الصورة المقدمة.
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<td>0.92</td>
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</tr>
</tbody>
</table>

- التحديات من خلال التحليل البياني في البداية الساقية (4-1)، يمكن استنتاج ما يلي:
1. أن الجانب الايجابي للإنترنت في البحث العلمي الأكاديمي الذي يمكن أن يحقق على تطبيق إنترنت في البحث العلمي.
2. استخدم مزيد من الدراسات الميدانية لتوضيح الإنترنت في البحث العلمي الآلي في مجالات وتقنية أخرى مع مساعدة من استراتيجيات البحث المثالية، التدريب، وبيانات الجدري، والتدريس الالي.

المسار:
1. نحو الإسلام، صح، 1982، (276). تأثير حضوري بالطرق المختلفة في العالم الإسلامي.
5. بين عبد الله القبل، عبد الله (2004). الإنترنت المستخدم العربي، ط. دار الفكر. أ. 11.
DNA sequences analysis of 16 S r DNA gene from Pseudomonas aeruginosa bacteria isolated from wounds infection

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ABSTRACT

Ten (10) isolates of Pseudomonas aeruginosa were collected from wound clinical cases. The results revealed that the 16S rDNA gene was presented in all Pseudomonas aeruginosa isolates. The gel electrophoresis showed that the molecular weight of 16S rDNA gene was 956 bp. DNA sequences of 16S rDNA gene was done, and the results showed the presence of some gene mutations like substitution and deletion with 93% identity with the Refseq gene.
**DNA**

DNA is a molecule from the genetics of bacteria DNA Geenac biotech kit (system, UK).

**16S rDNA**

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<td>396</td>
<td>97-122 E</td>
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<tr>
<td>R</td>
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</table>

**16S rDNA**

Sequencing of 16S rDNA genes

To extract 16S rDNA from all bacteria using 16S rDNA primers, extracts are sent to NICEP, Primer R.

**BLAST**

Basic Local Alignment Search Tool (NCBI National Center for Biotechnology Information).

**Characteristics**

Aerobic, Gram-negative rods with Endospores. They are motile by polar flagella or peritrichous flagella.

**Catalase**

Positive in all strains.

**MOT**

Positive in all strains.

**Katalase**

Positive in all strains.

**Growth temperature**

At 10°C, 25°C, 37°C and 50°C.

**Growth medium**

Growth is observed on MacConkey agar, Blood agar, Citrimide agar, and Catalase Oxidase positive. 

**Motility**

Positive in all strains.

**Respiratory**

Facultative anaerobic.

**Trypticase soy agar**

Growth is observed on Trypticase soy agar.

**Cystic fibrosis**

Sensitive to all antibiotics.

**Resistant to**

None of the antibiotics tested.

**Pathology**

Infectious diseases.

**Signs**

In the elderly and immunocompromised patients.

**Clinical manifestations**

Cancer, Ulcers, Burns.

**Molecular methods**

PCR (Polymerase Chain Reaction), Sequencing.

**Therapy**

Antibiotics, Antibiotics, Antimicrobial agents.

**Prevention**

Hygiene, Personal hygiene, Water disinfection.

**References**


**Conclusion**

Pseudomonas aeruginosa is a Gram-negative, aerobic, motile, and rod-shaped bacterium. It is found in various environments, including soil, water, and the human body. It is known for its resistance to antibiotics and its ability to cause infections in immunocompromised patients. The bacteria have been associated with cystic fibrosis, burn wounds, and other infections.

**Acknowledgments**

This work was supported by the National Institutes of Health (NIH) under Grant No. 5R01AI098473-05.
Ethydium bromide: Shrink number (1): Green. 16S rDNA P. aeruginosa

pseudomonas spp. Pseudomonas aeruginosa and pseudomonas

Guanine Adenine

Sbjct 687

Thymine

Adenine

Sbjct 285

Guanine

Adenine

Sbjct 342

Guanine

Adenine

Sbjct 225

Guanine

Adenine

Sbjct 210

Guanine

Adenine

Sbjct 191

Guanine

Adenine

Sbjct 173

Guanine

Adenine

Sbjct 212

Cytosine

Sbjct 212

Cytosine

Sbjct 207

Cytosine

Sbjct 165

Cytosine

Sbjct 151

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine

Sbjct 147

Cytosine
وتم استبدال القاعدة التروجينية الجوانين Bal1 بالثانين Adenine عند الموقع 961 والأدينين Adenine بالثامن Cytosine عند موقع 974 والثانين Adenine بالثامن Guanine عند موقع 975. وتم استبدال القاعدة التروجينية جوانين Bal1 بالثامن Adenine عند موقع 976 والأدينين Adenine بالثامن Cytosine عند موقع 991 والأدينين Adenine بالثامن Cytosine عند موقع 994. وتم استبدال القاعدة التروجينية جوانين Bal1 بالثامن Adenine عند موقع 997 بالثامن Thymine وتم استبدال القاعدة التروجينية Galiane Bal1 بالثامن Thymine عند موقع 1002.

وتم إضافة إلى العديد من طفرات استبدال القواعد Galiane Thymine وتم استبدال القاعدة التروجينية Thymine بالثامن Adenine عند موقع 1005 والثامن Cytosine عند موقع 1007 و 1010، وتم استبدال القاعدة التروجينية Thymine بالثامن Cytosine عند موقع 1008 و 1010.

وتم إضافة إلى الكثير من طفرات استبدال القواعد Galiane Thymine وتم استبدال القاعدة التروجينية Galiane Thymine. وتم استبدال القاعدة التروجينية Galiane Thymine عند موقع 1005 والثامن Cytosine عند موقع 1007 و 1010، وتم استبدال القاعدة التروجينية Galiane Thymine عند موقع 1008 و 1010.

وشكل رقم (2): نتائج تحليل تسلسل الحمض النووي الجيني 16s rDNA مع الجين الأصل (ID: NC002516.2) 

**Pseudomonas aeruginosa** PAO1 (ID: NC002516.21)
Sequencing of 16s rDNA genes (Pseudomonas aeruginosa S2-SF) for comparison with the original gene Pseudomonas aeruginosa PAOI (ID: NC002516.21).
وجد أن حصول طفرات في جين 16S rDNA لـ *Helicobacter pylori* يسبب حصول مقاومة البكتيريا لـ tetracycline (13) tetracycline التي تؤثر على نوكليوتيد 30S من الريبوسومات ويمنع الربط مع amminoacyl-tRNA، مما يؤدي إلى تغيير البروتين الحيوي. وبدأت الدراسات أن سبب مقاومة البكتيريا لهذا المضاد إذا كان نتائج الإحصائيات بالطفرات الحاملة للـ *Helicobacter pylori* في جين 16S rDNA (14). وقد بذلت جهود عديدة (15) أن حصول طفرة عارمة في الجين 16S rDNA وذلك بالاستخدام بالسيتوسين Guanine و باستبدال الجين Propionibacterium Tetracycline مقاومة أيضًا في *E. coli*، بالإضافة إلى حصول طفرات في *Pseudomonas aeruginosa* لـ 16S rDNA Tetracycline.

المصادر

Ultra and histological structure of tongue in \textit{Natrix tessellata tessellata} Iraqi water snake

Abeer M. Hussain, Basma A. Jasim and Nawras A.M. Muzahim

Dept. of Biology / College of Sciences for Women/ University of Baghdad / Republic of Iraq

\textbf{ABSTRACT}

The purpose of this study was to investigate the tongue morphology and histology for Iraqi water snake (\textit{Natrix tessellata tessellata}) using light and scanning electron-microscopy (SEM) techniques. The morphological finding revealed the presence of three parts: lingual apex, lingual body and lingual root. Light microscopic shows that the tongue composed of keratinized stratified squamous epithelium based on lamina contain bundles of striated muscle its fibres including connective tissue, blood vessels and nerve. Cartilage or bone skeleton are absent. The SEM shows that the apex of the tongue is bifurcated and the dorsal surface contains medial sulcus surrounded by epithelial tissue folds.
The study investigated the histological features of the tongue epithelium in Dice snake (Natrix tessellata) using conventional histological methods. The tongue was dissected, fixed in formalin, embedded in paraffin, and sectioned at 5 µm. The sections were stained with hematoxylin and eosin (H&E) and examined under a light microscope. The tongue epithelium was found to have three layers: the stratum corneum, the stratum spinosum, and the stratum germinativum. The stratum corneum was composed of keratinized cells, the stratum spinosum consisted of cuboidal cells, and the stratum germinativum contained basal cells. The tongue epithelium was found to be highly stratified and keratinized, which is typical of species that feed on live prey. The study also observed the tongue's ability to discriminate between different tastes, and the role of the tongue in swallowing and food processing was examined. The findings suggest that the Dice snake's tongue is well-adapted for its feeding behavior, providing a functional advantage in the natural habitat.
Drastic changes in the lingual root of the human tooth. Lingual root of the human tooth (LHF:6), showing the change in the shape of the root. 

Figure 2: Magnified view of the human tooth showing the change in the shape of the root. LHF:6.

Figure 3: Magnified view of the human tooth showing the change in the shape of the root. LHF:6.

Figure 4: Magnified view of the human tooth showing the change in the shape of the root. LHF:6.

Figure 5: Magnified view of the human tooth showing the change in the shape of the root. LHF:6.
شکل رقم (7): صورة المجهر الإلكتروني الماسح توضح النهاية الأمامية المشترورة للسان في الماء

شکل رقم (8): (A) صورة المجهر الإلكتروني الماسح للسان في الماء العرقي توضحسطح القؤري لجسم الشفاه، (Longitudinal fold)، جزء مكرب للبطنات الطولية (B) (Bulges)، جزء مكرب للانتحافات (C) (Median Sulcus).

شکل رقم (9): (A) طيات طولية مقطعة، (B) نقطة، (Lingual root) توضح جذر الشفاه، وضوح العضلات القاسية في نهاية جذر الشفاه.

A survey of aquatic invertebrates in some aquatic systems in Wasit Governorate - Iraq

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ABSTRACT

The present study included a survey of aquatic invertebrates in eight sites in Wasit Governorate - Iraq, collected using a hand shovel and Eckman Dredge (15 X 15) cm² by five replicates were combined with a distance of 1-2 meters from the edge of the river, and at a depth of 50-90 cm during the month July 2015 to May 2016, were diagnosis 88 taxonomic units, 23 belonging to a group Protozoa, 4 belonging to group of Turbellaria, 5 belonging to Nematoda, 10 belonging to group of Annelida and 20 belonging to group of crustaceans. The protozoa were recorded highest percentage compared to other aggregates amounted to 42% of the total number of aquatic invertebrates. The results showed a clear difference in the temporal and spatial distribution in the preparation and the density of aquatic invertebrate species because of the difference between the study sites in the physical and chemical properties.
وصف الموقع الجغرافي لمنطقة الدراسة

من الإثباتات الحديثة في الدراسات البيئية هو إثارة أهمية أكبر للتعامل الإداري في صمغ والتحسين البيئي، فالعمل الذي يقدمه مجال دراسة البيئة، كإجابة على الحاجة إلى أن نتعامل بشكل أفضل مع التهديدات البيئية التي يتعاقب عليها العالم أكثر. يتأثر تحويت الأبحاث التي تعتمد في هذه المجال، مع تقدم نظريات التحليل البيئي هذه الأفكار في بيئة أخرى لجذب هذه الأفكار. كما أن المجامع البيئية تأتي في صمغ أكثر أهمية، وهذا لأنه ما يثير تقاسمات البيئية وتثبيت إعداد ما تكون لبيئة مثالية. وضعت هذه الأفكار في نظرًا إن نشرها يفيدنا أن الاهتمام بالبيئة يفيدنا في صمغ أكثر تحسناً.

ومع أنه من الجوانب للاقتراحات في هناك ما يمكن أن نحققه، فإنهم بيداً على أن نكون تواصلًا أكثر بين الناس. وسريعًا وسريعًا، مع النصائح العامة، وأننا نكون قادرين على أن نكون هناك نتائج أكثر تجذباً للاستفادة من صمغ هذه الأفكار. وسريعًا وسريعًا، مع النصائح العامة، وأننا نكون قادرين على أن نكون هناك نتائج أكثر تجذباً للاستفادة من صمغ هذه الأفكار.
ومقررات الماء، وتُنظم من خلال صندوق التدقيق. يتم استخدام الأجهزة المحمولة لقياس التكلفة، أخذت في المركب. يتم جمع النباتات المائية شهريا لمدة من تموز 2015، السبتمبر 2016، باستخدام مفرقة نيلية ذات أبعاد (15 X 15 سم). بينما تم جمع الراسب والحطب الخيطي، وفقاً للنماذج المثبتة والمتصلة على خرائط الدراسة، باستخدام جهاز Eckman Dredge. جرف موافق الدراسة بنزول كراء أكمان إعداد (15 x 15 سم/وأعمال). كانت تجميع على بعد 1-2 م من الضفاف، وعمق 50-90 سم، توضع عينة الراسب في مخلة دوياً تحت قطع (0.2 ملم، ثم عسل الراسب والطين، بناء على انتظار من عملية التحليل يتم عزل اللافقاريات الكبيرة Macrobiotus إلى العامة ومساحة محكمة السد واقل 48%، ويتراوح المعدل من المواقع. يتم تشير اسم الموقع، جمع عينات السد والنباتات، و Hương، و مجهزة خيطة وخصيتين. يتم استخدام جميع الإجراءات والآليات. 

جمع النماذج

والموضوع

والموضوع
والفقاريات بنسبة مئوية وصلت إلى (18 و 16) % وعلي التوالي،

ف، إن النتائج في هذه الدراسة تتفق مع منصور في إليه (10، 16) وفقاً buffering، الذي يُ떤د بعض خصائص البيئة المائية، وurence، وذلك لكون مصدر المياه فيها من الفيضانات والصرف الزراعي. 

استخدمت دراسة (مجلة رقم 2) لدرجة مرتفعة من أشهر 2015 - 2016، و 88 وحدة جمعية كانت 23 منهما تعود إلى مجموعة اللافقاريات و 4 و 4 تعود إلى مجموعة المحركات و 10 Nematodas و 5 تعود إلى الدودان الحقيقية و 20 تعود إلى Crustacea و 8 تعود إلى مجموعة العناكب و 10 تعود إلى مجموعة النواعم Mollusca و 10 تعود إلى مجموعة النباتات Insecta، تستعِد مجموعة اللافقاريات (مجلة رقم 1) في الدراسة الحالية إلى نسبة مئوية طويلة بجمعين الأخرى وصلت إلى (42 %) من المجموعات كلاً للافقاريات المائية، ونسبة المجموعات الحرارية أنواع مماثلة، وتشمل العناكب، وتمكن اللافقاريات بالإكايلية، بما في ذلك درجة الحموضة، وتيرة البحيرة، ونوعية الأكسجين، والماء. كما لا تتوفر microhabitats بشكل عام، ولكن تحت الظروف الصغرية، مثل بضعة مستويات مربعة، ضمن الأماكن أو بيئات طبية مثل: الطرق، والنظام البيئي، والحيوانات (19).

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from Sinop coasts of the Black sea. Turkish J. Mar.
Sci. 6:227-240.
10. Duquesne S. and Riddle M.(2002). Biological
monitoring of heavy –metal contamination in coastal
waters of Casey station . windmill island , east
P.: 1248.
of the United States 2nd John Willey & Sons, New
York.
13. Richards LA.(1954). Diagnosis and
improvement of saline and alkali soils. Handbook,
waters). Soil and Water Conservation Society of
analysis of phytoplankton species inhabiting the
protozoan species inhabiting the East Bank
sediment of river Tigris in Baghdad City. Iraq. J.
phytoplankton and nutrients in Al-Hammar marsh-
Iraq. Master Thesis., College of Sciences,
University of Basrah, Iraq.
of physico-chemical parameters of water on
zooplankton diversity in Nanjangud industrial area,
مناقشة رقم (1): قائمة بالنتائج النهائية للألفايرات المائية المسجلة في بعض المسطحات المائية في وسط خليط الفترة من شهر تموز / يوليو 2015 إلى شهر أيار 2016 (5 مواعيد - غير موجود)

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</tr>
<tr>
<td>63</td>
<td>Immature Cycopoda</td>
<td>+</td>
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<td></td>
<td><strong>Subclass: Harpacticoida</strong></td>
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</tr>
<tr>
<td>64</td>
<td>Nitocra sp. (Brady, 1880)</td>
<td>+</td>
<td></td>
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<td></td>
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<tr>
<td>65</td>
<td>Nauplii of Copepoda</td>
<td>+</td>
<td></td>
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<tr>
<td>66</td>
<td>Cypris sp. Zenker 1854</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>67</td>
<td>Other Ostracoda</td>
<td>+</td>
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<tr>
<td>Class</td>
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<tr>
<td>Isopoda</td>
<td>Shpaeroma annadalei annandalei Stebbing, 1911</td>
<td>-</td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td>Decapoda</td>
<td>Macrobrachium nipponese (De Haan, 1849)</td>
<td>*</td>
<td></td>
<td></td>
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<tr>
<td>Amphipoda</td>
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<td>+</td>
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<tr>
<td>Insecta</td>
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<tr>
<td>Gastropoda</td>
<td>Physa acuta Draparnaud, 1805</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>Lymania sp (Linnaeus)</td>
<td>+</td>
<td></td>
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<tr>
<td></td>
<td>Melanoides tuberculata Müller, 1774</td>
<td>-</td>
<td></td>
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<tr>
<td></td>
<td>Melanopsis costata Olivier, 1804</td>
<td>-</td>
<td></td>
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<tr>
<td></td>
<td>Melanopsis nodosa Adams, 1854</td>
<td>-</td>
<td></td>
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<tr>
<td></td>
<td>Theodoxus sp Sowerby, 1849</td>
<td>-</td>
<td></td>
<td></td>
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<tr>
<td>Bivalva</td>
<td>Pseudodontopsis euphraticus Bourguignat</td>
<td>+</td>
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<tr>
<td></td>
<td>Unio tigris Bourguignat</td>
<td>+</td>
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<tr>
<td></td>
<td>Corbicula sp Müller</td>
<td>+</td>
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</table>
Masdar: The sustainable city

A scientific interpretation of the most important sustainability applications with an attempt for adoption to improve the environmental situation in Baghdad city

Hala H. Musa

Dept. of Engineering Affairs / Headquarter of University of Baghdad / Republic of Iraq

ABSTRACT

The present study was conducted to take advantages of the experience of an environment-friendly city with a climate similar to the climate of Baghdad, that was Masdar City, which located in the United Arab of Emirates. The study also tried to make benefit of the sustainability applications that implemented in Masdar City and adopt them in Baghdad. It is an attempt to take the most important environmental solutions that are used to improve the environmental situation of Masdar city by making the emissions of CO2 and other toxic gases in the lowest levels that are accepted globally. In addition, make the dependence in electricity production on renewable energy sources and reduce the dependence on polluting fossil fuels, that was based on a lot of principles, such as, adoption of recycling principle by making benefit of the construction waste and try to be recycled to produce new environment-friendly construction materials, in addition, taking advantage of the sewage and waste to produce electricity and also for collecting the dioxide carbon to reuse it for industrial purposes and finally for the production of water that used for the purposes of cleaning and irrigation, the other principle was taking advantage of solar energy, for several purposes, such as, producing of electrical energy, filtering and sterilizing water for drinking purposes, as well as, using solar energy for heating water for domestic purposes. The research reaches to most all those principles by adopting an analyzing study for a collection of scientific applications In pursuit of finding facts and recognizing the need to melt all sciences in one pot, especially that urban planning since is a comprehensive science because it deals with all matters relating to the city and its environment, that’s only an attempt to develop solutions for the environmental problems of Baghdad, to which the problem of air pollution, the problem of the scarcity of drinking water, the problem of the deficit in the wastewater and waste treatment in addition to the problems of acute shortage of generation and producing electricity, all those problems and more resulted the bad environmental situation of Baghdad that reflected Negatively on the health of the people who live in the city and that seems clear in Statistical data. Accordingly, the research has suggested a range of solutions to improve the environmental situation of Baghdad by utilizing from the environmental applications used in Masdar City.
لا يعني على ما تقدم، فإن الاستخدام الأساسي لنيوين هو توفير
- مصدر الطاقة (100% لطاقة متجددة)
- تكون ذات خاليا أو أنها تستخدم
- استخدام الوقود الأحفوري (Free Zone)
- يتم استخدام الأنظمة ووسائل الإنتاج المجددة، وأفضل
- أن تكون فاصلة قليلا كافية (3) عن رموز النقطة.
- إن الجبهة الأدنى (60%) من المساحة، وفصائل البذور المسمى (4) بنسبة (98%)
- وتكون نسبة (3).
- متحكم في الطاقة (16).
- توفر الطاقة (18).
- rides أو استخدم الطرق الجاود (14).
- التشغيل الكهربائي واستخدام الطرق الجاود (16).
- استخدام ميادين الطاقة الجاود (16) PH.
- نتائج الميزانية مقابل الطاقة (1).
- تم استخدام نظام كهربائي (16).
- تم استخدام طاقة متجددة (98%).
- يتم استخدام الطاقة (16).
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- يتم استخدام الطاقة (16).
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- تم استخدام نظام كهربائي (16).
- تم استخدام طاقة متجددة (98%)
إضافة إلى ما ذكرنا في ما قبل، فقد كانت الكافطة الحضرية واحدة من الميقات الأساسية للمدينة الحضرية، حيث أنها كانت وجهة الأ钾 الأثري للمدينة. في حين، في أخرى، كانت هذه الكافطة واحدة من الطبقات المتشابهة، حيث أن الكافطة كانت هدف الأرباح، في حين، كان هذا النوع من الطبقات هو خيار مفيد للمواطنين في المدينة حي ثقة. 

5. إجابة المدنية بحوار أخضر برسم حديثاً طبيعياً للمدينة مباشرة.

6. أنماط تفعيل الطرق الكافيتية الحضرية في هذه المدينة على

7. دورة مسيرة للطاقم الواحد بما في ذلك أرفف الأخبار، والمجلات،

8. وحجج التحليل أساسها، كما استُخدمت أخيراً نوع أخرى من الطاولة.

9. التكنولوجية الاستمرار في مناطق الحضرية، وفي ترiore ما، و

10. قرأت الميدان، تسببت هذه الحدود الضرر، مسارات، وحولها.

11. مبلغ مقدم للأعمال الرجاء المقدمة.

كل ذلك كان موحداً وصولاً إلى دخالت (خان الوحدة البنائية) وخارجة (خان الحدود الحضرية) كدليل ودخلنا تقيل

الأعمال، على سبيل المثال، التكتيكات الإصطناعية وما لها من أثر بسي.

http://www.2davdubai.com/}

ش提醒 (1): موقع مدينة مصدر في شبه القارة العربية المتحدة ومن إمارة أبوظبي حيث تقع هذه المدينة في قلب مصهر إمارة أبوظبي بالقرب من خط

من الحدود الثانوية للمدينة، التي يمكن من خلالها تحقيق مبادئ

المستدامية البنائية والتي تعد الأهداف الأساسية من إغاثة المدينة.

عمليات البناء، عن طريق إعادة تدوير تلك المحافل، إذ تقوم على

موقع مبنى لإعادة تدويرها حيث يعتبر مركز المعالجة، الذي يبلغ

مساحته (12) كيلومتر، يتم تحويل (96% من مخلفات البناء إلى

مواد مصنع وسائل البناء الثانوية في المدينة. كما تتبع مدينة

 مصدر نظاماً متعدد الصفائح، حيث يتم معган القماش

وارسالها إلى موقع مصنع ليغلي الصفائح عن طريق الألباب المرفعة، فيما

تستخدم لتصبح فائدة من للطاقات من الزجاج المستخدم من النفايات (13) و يتم

إشراف تحويلها في مواد نفايات (15). كل ذلك يتم بالחליף،

أيضاً، يتم إعادة النظر في النفايات المستخدمة، يتم استخدام

هذه المواد للعثور على المحور الثاني للتصيلي.

3. مصادر الطاقة المحدودة في مدينة مصدر، حيث أن من أحد

الأهداف الأساسية لإمارة مصدر، حيث توجه

إمارة مصدر، لتصبح مشاركاً وملاءّماً في عالم الطاقة الكهربائية، حيث فذ أولئك

الجهة المسماة هو إعداد للمدينة على أن يتم تحويل (22)

كفاده تتكون خلال الألوان المشكلة من (8777) و (6778) و (9778)

لتوليد الطاقة المحدودة، أبعادها محددة نسبياً (100%)

من الطاقة التي تمتلكها مدينة مصدر (1). وفقًا تماً

ثانياً: أهم الخطوة التصميمية لتطبيقات الاستدامة في البحث

1. شبكة المياه في مدينة مصدر (水源 شير) + المياه الزراعية

2. مياه الصرف الصحي: تسعى مدينة مصدر (المصدر) لتطبيق الاستدامة، وتشير استهلاك المياه، كنوبه تقع ضمن منطقة صحراء ذات

درجات حرارة عالية، ولإفقتها في المياه المائية، حيث تُحسب

المدينة على حصة معالجة تعمل بالطاقة الشمسية لتوفير حاجتها من

المياه (12). فقد تم التخطيط لدراسة استخدام شواطئ في المدينة

البحرية (13) وتسمح بالاتجاه إلى إ燹ية استدامة للمياه في المدينة

البحرية نسبياً، حيث يتم إعادة تدوير (80%) تقريباً من

المياه المستخدمة، ويتم استخدام هذه الماء في التدريب

لأغراض مختلفة (14)، إذ تستخدم المياه العاملة في تنظيف

المصادر والبيئة، والقدرة على فتح مياه الصرف الصحي

والحقلية إلى وقود محلي لتدفيع الطاقة الكهربائية

(15) ، وسيكون النتيجة الثانية كقبول الوصول إلى هذا النجف من

خلال التحليل المالي، على سبيل المثال، النتيجة المذكورة في هذا العمل.

2. معالجة وإعادة تدوير النفايات (غلابات النفايات + مخلفات

المياه)؛ تسعى مدينة مصدر إلى تقليل المخلفات النافذة عن
التخطيط المدينة بطريقة وفرت فيها أساليب خاصة لمناظر الطاقة الشمسية داخل وخارج المدينة، هذا بالإضافة إلى معايير الطاقة الحيوية وталحول الطاقة من المناظر والصحراء الصحي (CSP). أما تتميز المدينة بإنشاء كتل الطاقة الشمسية (Concentrated Solar Power)몰ع وتحولها إلى طاقة
إن أهم مصدر الطاقة الطبيعية التي تستخدم فيها المدينة هي (شكل رقم 2) (1).
- الطاقة الشمسية المحكمة (Concentrated solar power)
- الطاقة الحرارية الجيولوجية (geothermal power)
- انبايعيات المذابة ومواد الصحراء الصحي وتحولها إلى طاقة (processing waste to energy)
- المواد والتنقية من الأغراض المنزلية والأدوات الكبيرة (التدفينة والتبريد)
وخلصنا لما قدم في المحور الأول من هذه الدراسة، فقد سعت مدينة مصدر إلى التحول من المعاملات التقليدية والتصميمية المبنية إلى المعاملات الإيجابية، وكان ذلك من خلال تغيير الطاقة التي تستخدم في المدينة. وبناءً على المناورة الطبيعية وتطبيق الاستدامة. هذا ما يعكس الهدف من أجلية خلق المكرون الكربون، وتحسين استخدام المياه والاحتياجات على أساسية بالإضافة إلى تقليل التخطيط البيئي وتحسين تحليل المعالجة الطبيعية واعتدادًا بدعاية إعداد التخطيط في استخدام مراحل الاختيار ومواد الصحراء الصحي والفنين. بما يعني أن مدينة مصدر هي مدينة سريعة على جميع مستويات
(شكل رقم 4).

شاكل رقم (3): مخطط مدينة مصدر في منظمات التحالف الطاقة من المصادر المتجددة
http://web.mit.edu

شاكل رقم (2): تسمى التحالف من المصادر المتجددة في مدينة مصدر
http://www.rpd-mohser.com

نلاحظ مما قدم في المحور الثاني من هذه الدراسة، كيف تكمن في ذلك المزاحمة الشخصية للمصادر المتجددة في المدينة. وبناءً على ذلك، فقد شكلت تلك الطريقة القليفة الجوية والرياح ونظام المدينة الوسطي للحالة المدينة، أو على مستوى تخطيط المدينة كلها. إذ يتم تيثب الأتراض الخالية التماس على مطروح لأنوية وعلي
1. البلاك المركب الخزلي للحرارة

إن البلاك المركب الخزلي للحرارة مكوناً من مشتقات الوقود النووي وتكون مادة AAD ( kukhwa madaanaa) صديقة للبيئة، وهي مادة عازلة للحرارة، وكتهبي، ومستعملة في أنظمة تخزين مواد نواوية وتشمل: من خطط ( الإلكترونات، الهواء، الرمال) المغذي، المائي، الرمل، محتوى الأمور (أفاده، إداره، إداره) النتيجة فعالة ( AAD) هو خطط ( للحرارة أو الصدأ) يتم إنتاجه في محطات التصميم والترميم، وتشمل مواد التغطية المعدنية ( AAD) وتشمل مواد التغطية المعدنية ( AAD)

2. انواع الطاقة الكهربائية من مياه الصرف الصحي واتناج المياه

مجالات البحث لمرحلة الثالثة

ويأتي المصور الثاني لاستيعاب عدة من طبقات الاستدامة المعروفة المستخدمة في مدنية مصادر وصول خلأ للمهنية، في كل منها تعرض ممر على الاتجاه الاستدامة في تحسين الواقع البيئي.

الصور الثاني: تحليل تكنولوجيا الاستدامة

(شكل رقم 5). ويبعد سنديوس البلوك والكتكنت بيئاته الحركية المضادة لما يعتمد على خصائص تكييف الطاقة اللازمة للتدوير والتفاوت والتجهيز تكييف الكهرباء حوالي 40%، حيث أتم التوصيل الحراري للمنتهج 0.50 واط/م² درجة مئوية، بينما يبلغ تعامد التوصيل الحراري الأقرب 0.144 واط/م² درجة مئوية (18). أي أن هذا المنتج يعنى أن أربع اضطرابات الدروع الأحمر الفضائي. كما أنه أفضل علاج تطوير من الخلايا الدقيقة، المهدّد على تviron بيئة داخل الجسم.

وبهجة بعد ذلك تأتي علاجات للشرائح، يمكن لهذا المحور 2 في مزيد من هذه التطورات في الدورات الخائبة، وهي مستخدمة في إطلاق تفاصيل صديقة لبيئة أيضاً. }

(شكل رقم 6) مراحل عمل خلية الوقود المركبية (19).

International Journal for Sciences and Technology / ICU: 4.32 - SJIF: 4.487 - GIF: 0.81 Vol. 11, No 3, September 2016 126
ومحالات تجمع ومعالجة المياه الآمنة أثناء الأنشطة إصداراً
للالتزامات المنزلية والتعبئة البرية، والتي منها على سبيل المثال (21).
1. عزل المنازل والطوابق البليغة، التي تحدد نسبة 45-60%.
2. تطهير الهواء (40-60%).
3. الأمانة: 0.1-1%.
4. كريرتي الهيدروجين: 0-1%.

وتختلف نسبة إنتاج الغازات السامة تبعاً لما يأتي (22):
- كيوبنغيات: فقد رأت نسبة المواد العضوية في الغازات
  90% ذات العلامة الكيميائية.
- نسبة الأكسجين: تزيد تلوث البكتيريا بـ 35%.
- الهواء التي تشمل علي تحفيز الغازات وذات النتيجة 
  نقص المواد الأخرى.
- عبير المحلي والمائي: زادت نسبة المواد البكتيريا والمائي
  بـ 3% تلوث البكتيريا وذات الهواء.
- يزيد تلوث الغازات السامة: 3% تلوث البكتيريا والمائي
  زادت نسبة البكتيريا والمائي.
- عبير القالب: حيث أن الطاقم يزيدون البكتيريا والمائي
  زادت نسبة البكتيريا والمائي.
- جرعة الحرارة: حيث أن الطاقم يزيدون البكتيريا والمائي.
- عبير القالب: حيث أن القالب الحديثة تكون أكثر تأثراً
  للهواء المطهر.

ولا تعني أن الأثر السلبي للغازات السامة على صحة
ال羰ا حيث أن الغازات تكتسب مخاوف من مخاوف يمكن أن يؤدي إلى
تدفق الغازات، التأجيج الخاص، والتفاوت بين الحالة، وكون
الموازنة في الخلايا. أما الغازات هذه الغازات على مدى طويل.
تؤثر على الخلايا، وتقيداً للتنبؤ، وتفاوت البكتيريا
ش.ZERO في هذه الحالة، والإنطفاقي مما يؤدي إلى
الفرامل، فإنها تساهم في الانتقال، مما يؤدي إلى
الفرامل. في حالة الغازات، وقانون البنية، حيث تتنقل
المصادر في درجة حرارة الحبيبية، في حالات هذه
المشكلة عن طريق إعداد وحفظ مادة خاصة بكل
وحدة بناءة ورقيه إنتاج الناتج القالب في المدينة ويعتمد
منها في توليد الطاقة الكهربائية ومياه السقي المحلي (ش.(الرقم 8).

- 1.1. محطة معالجة مياه الصرف الصحي حيث يمكن تكييف غرفة
  خاصة بها في المبنى، ومائية صغيره الحجم (20).
- 1.2. ضبط قيام الغازات السامة في مكبات القالب وعربية
  الصور المعملية

يشمل هذا التطبيق الحد العلوي لمعالجة متكالفة انبعاثات الغازات
البكتيريا، حيث يمكن تكييف مكبات القالب وعربية
الصرف الصحي

ومن أهم مرايا المفاعلات الحيوية (20):
- إنتاج الكهرباء.
إن محطة معالجة مياه الصرف الصحي من نوع (FO) تعتبر تقنية من الناحية البيئية، حيث تستخدم غاز ثاني أكسيد الكربون من الجو باستخدام الطحالب لاستخلاص الماء النقي.

4. تقنية التناضح الأمامي (FO)

5. تقنية استخلاص غاز ثاني أكسيد الكربون من الجو باستخدام الطحالب

يعتبر غاز ثاني أكسيد الكربون من الغازات السلبية للبيئة، حيث يؤدي إلى ارتفاع درجة حرارة الأرض. بالإضافة إلى أنه مصدر للعناصر الحيوية للأزيمات البيئية على مختلف أشكال الحياة على الأرض وكذلك على الأطياف وغيرها.

- مادة غاز كثيرة من الصناعات.
- مؤيدة في عمليات استخلاص الغاز.
- متميزة في صناعة طاعفي الحريق.
- متميزة في خدمات النزهات.

إن محطة معالجة مياه الصرف الصحي التي تمثل بكونها محطة (FO) يمكن توصيلها في مناطق معالجة مياه الصرف الصحي المحددة، مثلاً على المحطة الأمامية، وهي لا بد من التفكير المستقبلي لتأثير الأحماض المستمرة لمثل هذه المحطات وذلك بجهد تقليل النزول البيئي من المدينة.
إن توفير الظروف الملائمة لنمو الطحالب يساعد في القيام بعملية التربة الضوئية وبالتالي على سحب غاز ثاني أكسيد الكربون من الجو. وكمثال، يمكن استخدام الطحلب كمادة أساسية في مجع لكونها رخصية ويمكن زراعتها بسهولة، ولا يزال تشمل ذلك تحقيق الاتصالات بين النباتات والحيوي. تتراوح درجة الرطوبة الحالية بين الماء 4.487 و4.287. إنها تتحمل بعض الفعالية في معالجة المخلفات الجائلة. (الشكل رقم 12)

*التحديات المروية (المصادر والملاءمة غير الضرورية، درجة الحرارة المناسبة، درجة الملوحة المناسبة، الماء (25). (الشكل رقم 13)*

6. تصفح المياه بالطاقة الشمسية أو أنظمة التدفق الدائمة

- وهي تقنية تعمل على تصفح المياه من خلال جمع الرياح الشمسية حيث يمكن الاستفادة من هذه المياه الساخنة لتعزيز الأغراض الاستعمارية كالتخليل والغسل أو الأغراض التجميلية. وتستخدم هذه الطاقة الكهربائية أو تجهيز المحاليل الزراعية. وقد تشمل استخدام نبوع من الموارد المتاحة في المنازل مثل مخزوناً كما صاغت النتائج بسبب درجات الحرارة المطلوبة للمياه وعند ذلك يمكن استخدام الأدوات المائية في دفع المياه وتوليد الطاقة الكهربائية. (الشكل رقم 15)

*الأشكال المائية (النوعية في أحد أرضية المدينة (26) (الشكل رقم 16)*

حائط-efficiency (pH) (25)

*الأشكال المائية (النوعية في أحد أرضية المدينة (26) (الشكل رقم 16)*

- 1. دالة أقل من 50 درجة مئوية للمحمات السباحة.
- 2. ساخنة بين 60-80 درجة مئوية للاستعمال المنزلي.
- 3. مخلة الحصول على البخار لغرض توليد الطاقة الكهربائية.
المحور الثالث: بغداد والاستمالة

أولاً: أعمال المؤسّسات البيئية "المتعلقة بالتطبيقات قيد البناء" للمنحة بغداد

تعد الإشارة بداية إلى أن معظم الإحصاءات المعطاة هنا لعام 2009 و 2014، و التي يعتبرها البعض نشاط من قبل الجهة المركزية للاستدامة، ولا يمكن اكتسابها بشكل فعال.

بعد عام، يشير عدد من العوامل إلى إجراءات تأسيس عام 2013، و تمثلت هذه الأدوات في العراق، و تأتي بعد مجرد 16 عاماً، أكبر مكثفة في الوطن العربي بعد العراق، و تأتي نتائج إجمالي المنشآت المحلية 36 مليون متر مكعب، و تأتي عدد من العوامل بيئية و الاجتماعية.

استخدام الطاقة الشمسية المركزية (power)

من هذا القانون، يمكن أن نقول: "الطاقة الشمسية طاقة نفعية و فعالة في enctype="application/pdf"

\[\text{International Journal for Sciences and Technology / ICV: 4.32 - SJIF: 4.487 - GIF: 0.81} \quad \text{Vol. 11, No 3, September 2016} \]


- استخدام الطاقة الشمسية المركزية (Concentrated solar)
تنخيل مدى الكثافة السكانية التي تعيدها محلياتها، وقد ترتيب على العديد من المناطق البيئية التي تستخدم عادة منهما بما يتناسب مع مصارف البحت.

شائدة لكل السكان، وأبلغت الوزارة العامة في بغداد شملت أعلى المحافظات في عدد السكان لسنة 2014، ونطاق ما تسربه 21% من مجموع سكان العراق (31) (معدل رقم 1). ونبدأ أن

جدول رقم (1): تسمى الكثافة السكانية لمدينة بغداد ونسبيها إلى العراق (31)

<table>
<thead>
<tr>
<th>المساحه</th>
<th>السكان</th>
<th>عدد السكان</th>
<th>أحسنة سن</th>
<th>الربيه من حيث الكثافة</th>
<th>الكثافة السكانية</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.175م²</td>
<td>9753873</td>
<td>21.2 %</td>
<td>1637 كم² (5 كم²)</td>
<td>2412 لكل متر²</td>
<td></td>
</tr>
</tbody>
</table>

مدينت بغداد تشمل تسمى اشتمل على 3682 مساحة سلسلة سنويون بمعدل سليم بايد 19،789 مرامور، كما هو منشأ في الترقيم السنوي للحظر المركزي للاضاءة لسنة 2009 (معدل رقم 2).

الإشعاع الشمسي لمدينة بغداد: إن العراق معرض للأشعة العلاج بناء على الكثافة المنخفضة المتخطية في بغداد ومستقلة في الكامنة (31) (معدل رقم 32).

جدول رقم (2): المعدل اليومي للإشعاع الشمسي وفترة الطبو وتبخير في بغداد (معدل رقم 2009)

4- بالإضافة إلى المقايئ غير المحسوسية من العواصف الطارئة التي ستكون اشتملة متحركة في إعدادها منذ عام 2003 (معدل رقم 21)، حيث تكمن من إعداد الفضاء المباشرة للاشعة الشمسي أدبيا في اعداد الاعضاء المعنية في إعداد المساكن المتبللة في مستويات النور لسنة 2008، وقد كان أكثر عدد الاعضاء في وضعها في محافظة بغداد، وتصميمه إلى وكذلك (21884م²) (معدل رقم 21).

ويعتبر ميسراً وشاعراً عن حجم اللعنة في مدن بغداد حيث بلغت تكلفة الدقة الأقلة في الجو ما يقارب 600 مكربة عظماء في المتر المشغوب، في حين أن الحد اليومي الوطني المقدر هو (350 مكربة عظماء / متر مشغوب) أي ما يعادل حوالي 50% بعد الحد المدبوغ. وعليه مع بعض الفروع ببعض دفع الابن يصلح بجودة الفروع، كما يعترف ببعض الجماهير في هذا المجال.

1- مدرسية: مدينت بغداد تشمل المزيد من النيافين البيئية، وثابلة مسألة في نوعية الرياح، في المدينة، في أحد أبرز هذه النشاطات، وضع تقييمات فلنصل إلى الحظر المركزي، وانشأ من الدقة والدور المركزي للاضاءة للأعمال لسنة 2009، ونبدأ أن

2- نفت أن تتماشى مع مسار الرياح لسنوات طويلة وقد اقتربت هذه الأعمال الجوية العراقية في شرائحها ذات الرقم 13 و15 د. م.

3- كانت مكماً في الغاز المحرر في الشقّات (البيئة المشغوب) في مصبات بغداد وقد بلغت (معدل رقم 2009).
شكل رقم (19): كمية الوقود المستخدم في الأفران والمراجل وغيرها حسب إحصائية عام 2014 (7)

شكل رقم (20): كمية الغازات المحروقة في الشعارات حسب عام 2014 (7)

في حدود العام 2014 أبلغ 493 كبير وب (شكل رقم 24) غير أن المرور في المدينة والمكابح السكنية على أن المياه الصافي المجهز للمواطنين في بغداد هو بعد ذلك ماء غير صالح للشرب إذ بلغت نسبة المركبة 7.13 ملليتر، كما بلغ مقدار العاملة الكلية لمواد الأطعمة 375 ملليتر، الامر الذي دعا الموارد إلى الحفاظ على المياه المعبأة لاستخدام الشرب والطهي (شكل رقم 25). وقد حددت دائرة بغداد مجموعاً من الأسباب التي أدت إلى هذا التدفق في خدمة المياه الصافي المجهز لابناء المدينة، وقد كان من مشاكل المياه الكهربائية وعدم استمراريتها الأثر الكبير على مشايع المياه في المدينة (شكل رقم 26).


1- في مناطق إمالة بغداد 23%
2- في مناطق الطراف بغداد 48%

أي أن حوالي ربع سكان بغداد لا يستهلكون من المياه الصافي بينما تقريرات الجوف تصف سكان الطراف بغداد لا يستهلكون من المياه الصافي إلا أن نسبة الفرد من المياه الصافي المستهلك كان قد ازداد قليلاً.

شكل رقم (22): مجموع الدقائق العاشرة في محطات مدنية بغداد لعام 2014 (7)
شکل رقم (23 - أ): نصيب الفرد من الماء الصافي المستهلك والم[number missing] (مليون م3) وكمية الماء المنتج من المشروعات مع المحافظة عدا القائم

شکل رقم (23 - ب): كمیتی الماء المیتزج من المشروعات والمحیطات العامة للديمایرتіا العامة للماء وامانة بغداد والمحافظة العامة للماء وناسبة المجوز في الإنتاج (مليون م3)

<table>
<thead>
<tr>
<th>المحافظة</th>
<th>النسبة المئوية من الماء المنتج من المشاريع والمحیطات العامة للديمایرتیا العامة للماء وامانة بغداد والمحافظة العامة للماء</th>
<th>النسبة المئوية من الماء المستهلك من المشاريع والمحیطات العامة للماء وامانة بغداد والمحافظة العامة للماء</th>
<th>النسبة المئوية من الماء المستهلك من المشاريع والمحیطات العامة للماء وامانة بغداد والمحافظة العامة للماء</th>
</tr>
</thead>
<tbody>
<tr>
<td>بغداد</td>
<td>70.55 %</td>
<td>47.34 %</td>
<td>8.37 %</td>
</tr>
<tr>
<td>Diyala</td>
<td>55.35 %</td>
<td>34.03 %</td>
<td>4.66 %</td>
</tr>
<tr>
<td>Al-Mansoora</td>
<td>60.55 %</td>
<td>48.76 %</td>
<td>8.11 %</td>
</tr>
<tr>
<td>Karbala</td>
<td>72.35 %</td>
<td>51.54 %</td>
<td>9.56 %</td>
</tr>
<tr>
<td>Wasit</td>
<td>67.55 %</td>
<td>45.54 %</td>
<td>7.89 %</td>
</tr>
<tr>
<td>混</td>
<td>67.55 %</td>
<td>45.54 %</td>
<td>7.89 %</td>
</tr>
</tbody>
</table>
شَكْل رقْم (24): نسب الفرد من الماء الصافي لعام 2014

شَكْل رقْم (25): الحدود الدنيا والعليا والمعدل لنتائج الفحوصات الكيمياوية والفيزيائية للماء الخام والشرب في محافظة بغداد لعام 2014
الطاقة الكهربائية: كما هو معروف فإن الكهرباء اليوم أصبحت
عدد غير قليل من المتضمنين على شبكة
الكهرباء في العراق. عرف المستخدمون بالعديد من المتضمنين على شبكة
الكهرباء بحريّة حيث وصلت إلى نقطة تصلب رقم (29) العجز الكبيرة في
الكهرباء في العراق. عدم ذلك فإن نتيجة توليد الطاقة الكهربائية في العراق
غير مبتعثة (ومضمنون بالطبع) المخاطر المخاطرة في
الاعتماد على الطاقة الكهربائية، فيما إذا كان عدد قليل من المحطات
الكهرباء والتي تزود عادة في مناطق السحوب الكثيرة أي خارج
مدينتي العراق. (شكل رقم 30). أن هذا الأمر يستدعي التفكير مدناً
في وجهاب احتمال مضايقات سريعة تلقيت من الطاقة الكهربائية
التي يشمل العجز الكبيرة في تضادي الطلب من الكهرباء، والذي تمتله
الliquidity.!
شکل رقم (28): توزیع الطاقة الكهربائية في المحافظات عدا قليم كردستان لعام 2014 (7)

شکل رقم (29): نصيب الفرد من الطاقة الكهربائية حسب المحافظة عدا قليم كردستان لعام 2014 (7)

شکل رقم (30): عدد محطات انتاج الطاقة الكهربائية حسب النوع لعام 2014 (30)
وتبلغ في الكرخ 94194 وقد كانت الأعلى بين محافظات العراق.
(7) وما لاشك في أن التلوث البيئي الذي تاعلي منه المدينة بشكل
أحد أهم أسباب التي أدت إلى ذلك كون الأطفال هم الأقل مناعة
والآكبر تأثرًا باللوتين البيئي (شكل رقم 33). 
وكم أيضًا، تشير هذه التقارير إلى أن تلوث الهواء يزيد من
خطر الإصابات التي تنتقل بها الإصابات عند الأطفال، فضلاً عن تقليل
Chance الإمكانية المقدمة للأطفال من الولادة السلمية.
الكثيره غير المحمية من مخلفات البناي والعمليات الهمه اللائي التي استخدمت هذا النوع من مواد البناي في انشاء جدرانها. وقد تم التركيز على هذا الجانب من المخلفات الصناعية كونه نسبيه صعبه بعض الانتقادات التي تم تناولها في البحث الثاني حيث من الممكن استخدامها في البناء بكميات علعبر الحدراه وخصوصاً الانتقادات عالية والتي تم تعديلها بالنظر إلى استخدام المخلفات الصناعية في المدى البعيد (شكل رقم 33). وبناءً على ذلك فإن مخلفات الصناعات الاشترائية هي الاصل على الامتناع عنوقاً من مخلفات الصناعية في القضاء عموماً (شكل رقم 35)، هذا بالإضافة إلى الكميات التي يعتبر حالي اليبنية المهمة في مدينة مصدر.

الموارد في المدينة (شكل رقم 37) والواقع عددًا 4 وحدات تقضي حدود إمارة بغداد على أن نسبة المياه المعلجة منها هي 78.7% أي أن حوالي 65% المبتكرين وحدات المعلجة لا يتم معالجتها بشكلهم المطلوب وذللك لأنها تنتج بكميات كبيرة. وتكون معالجتهم للمحلات الأربعة المخصصة للمعالجة (شفرة رقم 38)، وحسبما يمكن استنتاج أن أكثر من 15% من المياه المعلجة المطلوبة في المدينة لا يتم معالجتها تمامًا، وشكل عينياً بيئة مهينة على المدينة وسكانها.

7. قطاع المجاري: لقد تزايد الاهتمام عالمياً منذ خمسينيات القرن الماضي بالدراسات المتعلقة بموارد مياه المجاري الصحي ومعالجتها نظراً لما تحتويه من مخاطر ومواقف بيئية تشكل خطراً كبيراً على الصحة العامة. وفي هذا السياق، فإن الاتجاهات التي تتجه إلى تحسين البيئة في مدينة بغداد تحت المركزي الأول من بين محافظات القطر في كمية المياه المطلوبة المكلفة، وكذلك هو واضح في (شفرة رقم 36). وللمدينة شبكة مجاري كبيرة حيث تشير الإحصائيات أن أكثر من 95% من سكان المدينة مصرين بهذه الشبكة على أن 65% منهم فقط يرتبط شريكانهم بمحطات ووحدات المعلجة.

شكل رقم (35): التوزيع السنوي لكمية الخلافات الصناعية الضارة والخطرة والمطروحة من العامل التابعة لوزارة الصناعة والمعدن وشركات القطاع المختلط حسب القطاع لعام 2014

شكل رقم (36): كميات المياه المطلوبة من محطات المعالجة المركزية في العراق لعام 2009
شكّل رقم (37): النسبة المئوية للسكان المخدومين ببيشكات المجاري حسب المحافظة عدا الليم كردستان لعام 2014 (7)

<table>
<thead>
<tr>
<th>المحافظة</th>
<th>السكان المخدومين ببيشكات المجاري</th>
<th>السكان المجاري</th>
<th>نسبة المخدومين ببيشكات المجاري (%)</th>
</tr>
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<tbody>
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شكّل رقم (38): عدد محطات المعالجة المركزية التابعة للمديريات العامة لمجاري ودارة مدير بغداد والمجمعة للطاقات الفلكلورية في التصميمية حسب المحافظة عدا الليم كردستان لسنة 2014 (7)
8. قطاع النفايات الصناعية : تعرف النفايات الصناعية

بذلك تعزز استخدام خدماتي أو الخدمات الأخرى يقدمها

الطلبات غير المكلفة التي تنفق عن مختلف أنواع النفايات الصناعية

والنزلاقي والتجاري الصناعي الزراعي والصناعي (cej

ومنشور اداري والاقتصاد)، وحسب مصادر بغداد، فإن معدل النفايات التي

تتكفل بها سكان العاصمة بلغ 7 آلاف طن يومياً يوافق 1.6 كغم/يوم

(7) (شامل رقم 39).]

وأخدر من مراقبة مع إعداد النفايات المنطرقة في شوارع بغداد، والتي لم ترفعها كمر

ال넷ا، وبين الشكل رقم (40) السبب المنوي للنفايات التي

تتلقاها مخالفات النفايات في مدينة بغداد، وظهر في أن النفايات

المنزلية تشكل أكثر من نسبة فائدة مدينة النفايات.

ان تأثير تكسير النفايات في مناطق مدينة بغداد لم يفوّه لدى

الجميع. لكن الأرقام التي تعلناها الما، على توافرها، تأتي من

ندور الصناعي للمستهلكة عن كل فرد حسب الملاحظات عدا أقيم الكردنك لعام 2014 (7)

شَكل رقم (39): كمية النفايات الاحتياجية الموثوقة عن كل فرد حسب الملاحظات عدا أقيم الكردنك لعام 2014 (7)

شَكل رقم (40): النسب المنوية حسب مصادر النفايات الصناعية في مدينة بغداد (7)

ثانياً: الحقول البيئية المفقحة للمشاعل المطرورة في ضوء

التشريفات في البحت

1. النويية للمصادر النفاياتية للمتحدثة في الحدود العراقية، والتي تشير إلى خرط النفايات

2. استخدام خدماتي في الحدود العراقية، على سبيل تصرُّفها الصناعية للمصادر النفاياتية

3. منظورات تركز على تأثير النفايات الصناعية والمحطات الجارية النفاياتية، واقعة بأنها

withstandingها الخطر من التداعيات الصحية الناتجة عن معالجتها، ناهيك

وعن الأرقام المنوية التي تبرع أن تكون مسجية. إذا تجد ذلك

هناك 200 طن

وبعد وقوع تلك مصايف بغداد بلغ سعة تدريب تبلغ

1000 طن يومياً، وفق رؤوس مكاتب أعمال المصرف في مجال

 servicios. المشكلة الحقيقية هنا تكمن في أنه لا يوجد أي طريقة

لتنزيل النفايات أو مطرها صحياً، وما موجود حالياً عبارة

عن ميناء للنفايات تودي إلى الأحوال بالكثير من الإمساك

رغم ورش النفايات، وهذا يعني أن تؤثر البيئة سيئاً، وأن

هذا المشروع ليست سوى حكول موقعة.
استثمار الطاقة الكهربائية، خاصة في المنازل كرونة الاستدامة، يمثل كليلاً على النسب من بين الطاقة العابدة، كما ورد ذكره في نص المراجع 3. وفقاً، يكون هناك خلاف حول ترشيع الوقود والمواد البكر، حيث يستخدم استخدام الطاقة الكهربائية بصفة عامة.

4. الحرص على تنفيذ محطات إنتاج الطاقة الكهربائية من الكتلة الحيوية، مثل محطات أنابيب الطاقة من النفايات، ونتيجة لذلك، يتم استخدام الأتمات الطاقة من النفايات المكيفة، بالإضافة إلى أن الطاقة الكهربائية يمكن أن تكون مصدر شبه شهر النسيج الحضري لمدة.

5. بعد تحليل الأحماض، ناتجة عن المحطات الكهربائية، تستخدم تكنولوجيا لإنتاج الطاقة من النفايات المكيفة، وتعد هذه المحطة مساعدة للطاقة الكهربائية، ولكنها ليست مساعدة للمدينة، حيث تم استقرار ساقية بابيات تزود هذه المحطة بالطاقة الكهربائية من النفايات المكيفة، بالإضافة إلى أن هذه المحطة تجهز القاطع على إنتاج كهرباء من مواد الطاقة الكهربائية.

6. إذا تغيرت الهيكلية التشريعي، فكل الثالث يمكن أن تكون محطات إنتاج الطاقة المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايات المكيفة، وتحت إنتاج الطاقة الكهربائية من النفايا

26. موقع بوابة دبي الغازية نيلك للمشاريع الاستثمارية http://www.2daydubai.com
27. شركة إريكترو للطاقة الشمسية http://www.apricus.com
30. محطة نسم العامة في مصدر http://shamspower.ac
31. الموقع الرسمي لجهاز المركز للإحصاء http://www.cosis.gov.iq/ar/
32. تقرير إحصاءات البيئة في العراق لسنة 2009.
33. http://www.atlasObscura.com/articles/could-urban-algae-farms-clean-up-our-air

7. تقرير إحصاءات البيئة في العراق لسنة 2014.
17. Fadhil AY.; Ali AG. and Saadi D. (2013). Producing Load-Bearing Composite Blocks from AAC and Concrete. The American University of Sharjah/College of Engineering,Civil Engineering Department.

18. الموقع الرسمي لشركة النـسائط الوطنية الكويتية http://www.nicbmn.com/ar/
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