INFLUENCE OF HE-NE AND DIODE LASER ON DATE PALM CALLUS
Khater, M.S.
National Institute of Laser Enhanced Science (NILES) Cairo University, Egypt.

ABSTRACT

This study was evaluating the effect of diode laser with power 160 mw and He-Ne laser with power 7 mw on date palm callus. Diode laser was irradiate Date Palm Callus for 4,8,12 and 16 minutes and 5, 10, 15 and 20 minutes for He-Ne laser respectively. Generally 10 min irradiation in He-Ne and 8 min in diode laser significantly increased the content of total soluble sugars and total indol compared with control. In addition 20 min irradiation in He-Ne and 16 min in diode laser increased the content of total phenols and total soluble amino acids compared with control. DNA analysis and protein Electrophoreses were study the similarity of genetic at different duration of Laser irradiation for date palm callus.

DNA analysis showed a higher similarity between 10 min and control treatment (93%) while Lower similarity (85%) between 15 min and control treatment was recorded.

INTRODUCTION

Palms belong to family (Areaceae) (Corner, 1996) which contain several members, such as oil palm (Elasis guineensis Tacq) , date palm (Phoenix dactylifera L.) and cocount palm (Cocos nucifera L.) are widely cultivated for their fruit crop products. Date palm is one of the most important crops in the Arab world.

Date palm have large percentage of sucrose up to 80% and considerable amounts of fiber, fat, protein, minerals and vitamins.

In addition, date palms have been proven to be one of the most salt tolerant fruit crops therefore, have the potential to help combat desertification processes (Bauchireb and Clark, 1997).

(Salyaev et al., 2001) showed that laser irradiation stimulated the morphogenetic processes in plant tissues. (salyaev et al., 2003) found that five min callus irradiation by the He-Ne laser with wavelength 632.8 nm and intensity 10 mW can motivate lipid peroxidation processes in tissues of the plant. (Chen Rumin el al., 1992) showed that He-Ne laser with power 20 mw radiation enhanced the growth, differentiation of carnation callus and increase the callus fresh weight 170% compared with control treatment and laser argon with power 1000 mw can significantly promote bud differentiation for carnation.

(Wang et al., 2001) found that laser He-Ne with power 0.37mw for 1hour increased the growth of the callus and biomass of hairy roots. On the other hand (Wang Mansi et al., 1994) discover that magnetic field and laser He-Ne enhanced the biosynthesis and growth of ONOSMA PANICULATUM callus.

The objective of this work was to study Influence of He-Ne and Diode Laser with power 7, 160 mw respectively on date palm callus.

MATERIALS AND METHODS

This work was carried out at the National Institute of Laser Enhanced Science - Cairo University and Central Laboratory for Research and Development of Date Palm - Agricultural Research Center, Ministry of Agriculture during 2011-2012, to study the effect of He-Ne and Diode Laser on date palm callus. Preparation of culture media used for culturing according to (Murashige and Skoog, 1962)

1- Laser devices

Two types of laser were used as follows:

Diode laser with power 160 mw at 4,8,12 and 16 min
He-Ne laser with power 7 mw at 5,10,15 and 20 min

The spot diameter of laser diode was 2 mm, thus it is suitable for direct irradiation to Date Palm Callus without any divergence.

2-Chemical analysis:

Total soluble sugars, Total soluble amino acids, Total soluble indols and Total soluble phenols were measured according to (Dubois et al., 1956; Rosen, 1957; Larsen et al., 1962; A.O.A.C, 1990) Respectively.

3- Molecular studies:

1-DNA Fingerprints were analyzed according to (Porebski et al., 1977).
2-Protein analysis electrophoresis (SDS-PAGE) were analyzed according to (Laemmli, 1970)

4- Statistical analyses:

R2 and LSD descriptive measures of quantitative data using ANOVA (analysis of variance) analysis test for independent samples. p-Values <0.05 were considered significant for R2 and not significant for LSD (Table 1,2).

RESULTS AND DISCUSSION

The data shown in Tables 1&2 showed the effect of He-Ne and diode Laser on date palm callus. Total soluble amino acids contents in callus treated with He-Ne for 20 min are 0.225 which is twice of control 0.109 (Table 1). While the irradiation of diode Laser are increasing from 0.109 for untreated to reach its maximum 0.589 for 16 min (Table 2).This result goes in line with the finding of (Zeinab et al., 2013) who showed that red laser irradiation at wavelength 650 nm at the different treatment periods on palm callus cultures. Increased values of total amino acids after irradiation compared with the untreated callus.

On the other hand total soluble sugars (TSS) (Table 1) show that the different treatment time of irradiation with He-Ne TSS content progressively with increasing exposure, giving its maximum 1.34 at 20 min. In addition diode Laser increased TSS in the same
manner, so the highest content was recorded at 16 min 0.591, while 0.400 in control (Table 2). The present result is in congeniality with the finding of (Zeinab et al., 2013) who found that red laser irradiation at wavelength 650 nm increased the total soluble sugars on palm callus cultures at the different time compared with the untreated callus.

Total phenols and Total Indols as result of applied treatments are shown in (Table 1&2). The data indicates a significant for R2 increase in total phenols and Total Indols content in irradiated palm callus as compared to untreated treatment. After irradiation total phenols content reached its highest value 0.588 at 20 min duration comparing with control treatment 0.105. on the other hand Total Indol are for untreated treatment 0.205 and reached its highest 0.629 at 20 min in red laser irradiation (Table 1).

While total phenols content for unirradiated sample was 0.105 and reached its maximum 0.229 at treatments 16 min for total phenols and was 0.205 and reached its maximum 0.480 at 16 min diode Laser irradiation for Total Indols (Table 2).

Table (1) Effect of He-Ne Laser duration treatments on the contents of Total soluble amino acids, Total soluble sugars, Total phenols and Total Indol in palm callus .

<table>
<thead>
<tr>
<th>Treatments (min)</th>
<th>Total soluble amino acids (mg/g-1 Dw)</th>
<th>Total soluble sugars (mg/g-1 Dw)</th>
<th>Total phenols (mg/g-1 Dw)</th>
<th>Total Indol (mg/g-1 Dw)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.109</td>
<td>0.400</td>
<td>0.105</td>
<td>0.205</td>
</tr>
<tr>
<td>5</td>
<td>0.121</td>
<td>0.652</td>
<td>0.145</td>
<td>0.343</td>
</tr>
<tr>
<td>10</td>
<td>0.172</td>
<td>0.977</td>
<td>0.269</td>
<td>0.455</td>
</tr>
<tr>
<td>15</td>
<td>0.202</td>
<td>1.233</td>
<td>0.409</td>
<td>0.553</td>
</tr>
<tr>
<td>20</td>
<td>0.225</td>
<td>1.348</td>
<td>0.588</td>
<td>0.629</td>
</tr>
<tr>
<td>R2</td>
<td>0.97</td>
<td>0.98</td>
<td>0.96</td>
<td>0.98</td>
</tr>
<tr>
<td>LSD</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

NS = not significant

Table (2) Effect of diode Laser duration treatments on the contents of Total soluble amino acids, Total soluble sugars, Total phenols and Total Indol in palm callus .

<table>
<thead>
<tr>
<th>Treatments (min)</th>
<th>Total soluble amino acids (mg/g-1 Dw)</th>
<th>Total soluble sugars (mg/g-1 Dw)</th>
<th>Total phenols (mg/g-1 Dw)</th>
<th>Total Indol (mg/g-1 Dw)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.109</td>
<td>0.400</td>
<td>0.105</td>
<td>0.205</td>
</tr>
<tr>
<td>4</td>
<td>0.180</td>
<td>0.461</td>
<td>0.111</td>
<td>0.274</td>
</tr>
<tr>
<td>8</td>
<td>0.359</td>
<td>0.528</td>
<td>0.145</td>
<td>0.369</td>
</tr>
<tr>
<td>12</td>
<td>0.413</td>
<td>0.584</td>
<td>0.198</td>
<td>0.449</td>
</tr>
<tr>
<td>16</td>
<td>0.589</td>
<td>0.591</td>
<td>0.229</td>
<td>0.480</td>
</tr>
<tr>
<td>R2</td>
<td>0.97</td>
<td>0.95</td>
<td>0.95</td>
<td>0.98</td>
</tr>
<tr>
<td>LSD</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

NS = not significant

2.Molecular studies in palm callus cultures
A. Protein analysis:
Protein Electrophoresis analysis was obtain to study the similarity of the genetic at different time of Laser irradiation for cultures of date palm callus. 42.5 % Monomorphic and 7 total band had been found (Figure 1). Molecular weight 20 KDa was not showed at 10 min, on the other hand Molecular weight 13 KDa had not been appear at 10 and 15 min. Molecular weight 29 KDa was not showed in the callus treated for 5,10 and 15 min (Figure 1).

![Fig(1)](attachment:Fig1.jpg)
B. DNA analysis:

DNA analysis was obtained by using five primers for the similarities between the time of Laser irradiation for palm callus. (Figure 2) showed that primer HB15 for Molecular weight of (140 and 230bp) were not showed at 5, 10 and 15 min, while Molecular weight of (400 and 450bp) were not found at 5 min, Molecular weight of (945bp) was not visible at 5 and 15 min in (Figure 3) primer HB09 Molecular weight of (530, 920 and 1050bp) were not appear for control but found at 5, 10 and 15 min.

(Figure 4) showed that primer HB08 displayed Molecular weight of (1020bp) only in control. Primer HB12 (Figure 5) displayed Molecular weight of (790 bp) in fingerprint of palm callus was detected only in the control and 5 min; while Molecular weight 760bp was not showed in 15 min. as shown in (Figure 6), revealing that B99 primer display Molecular weight (450bp) in fingerprint for 5 min and 10 min was not found, while Molecular weight (180bp) was not found in that treated with 5, 10 and 15 min and Molecular weight (225bp) had been showed at 5 min.

![Fig (2) HB08](image-url)

![Fig (3) HB09](image-url)

![Fig (4) HB15](image-url)

![Fig (5) B99](image-url)

![Fig (6) HB12](image-url)

DNA analysis was find the similarities between date palm callus and Laser duration time.

The results in (Table 3) found high similarity between untreated treatment and 10 min up to 93%, Low similarity between untreated treatment and 15 min up to 85% and middle similarity between untreated treatment and 5 min up to 91%.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Control</th>
<th>5 min</th>
<th>10 min</th>
<th>15 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>--------</td>
<td>91%</td>
<td>93%</td>
<td>85%</td>
</tr>
</tbody>
</table>

Table (3) DNA analysis similarity between different Laser duration treatments

CONCLUSION

Previous work showed that the effects of He-Ne and diode Laser with power 7, 160 mw respectively on date palm callus culture. Diode Laser was irradiate Date Palm Callus for 4, 8, 12 and 16 minutes and 5, 10, 15 and 20 minutes for He-Ne laser. Generally 10 min irradiation in He-Ne and 8 min in diode Laser were significantly increased the content of total indol and total soluble sugars compared with control. In addition 20 min irradiation in He-Ne laser and 16 min in diode Laser increased the content of total phenols and total soluble amino acids compared with control.

REFERENCES


تأثير الذايىد ليزر والهليىم نيىن ليزر على كالس نخيل البلح
محمد سليمان خاطر
المعهد القومي لعلوم النبات- جامعة القاهرة
قد أجريت هذه الدراسة لتقييم تأثير الذايىد ليزر ذو طاقة 100 ميغاوات والهليىم نيىن ليزر ذو الطاقة 7 ميغاوات على كالس نخيل البلح زراة خلال موسم 2012-2013. وقد تم تعريض كالس نخيل البلح لليزر لمدة 4، 12، 2، 0 و 10 دقيقة، و 15 و 20 دقيقة للهليىم نيىن ليزر. أظهرت النتائج أن التشريع لمدة 12 دقيقة للهليىم نيىن و 16 دقيقة للذايىد ليزر زيادة كبيرة في الأندولات والسكرات. كما أظهرت النتائج زيادة الفينولات ومجموع الأحماض الأمينية في كالس نخيل البلح. كما تم دراسة الحامض النووي لكلاس نخيل البلح وجد تشابه كبير يصل ل73% عند التعرض لمدة 10 دقيقة للهليىم نيىن، بالمقارنة بالكثرة. 