Using Infrared Absorption Spectroscopy In Studying The Effect of Application Times of Ammonium Sulphate on Some Constituents of Manzanillo Olive Leaves At Full Bloom Stage

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ABSTRACT

This study was carried out during two successive seasons 2009 and 2010 on ten-years-old Manzanillo olive trees, planted at 5 X 5 m and grown in sandy soil in a private orchard located at Cairo–Alexandria desert road (about 50 Km from Cairo), Egypt. Trees were of normal growth, uniformed in vigour and received the same horticultural practices. This experiment aimed to study the effect of nitrogen fertilization dates on nitrogen, photosynthetic pigments and starch contents of Manzanillo olive leaves using FT-IR spectroscopy at full bloom stage. Also minimize the quantity of annually added amounts by 20% and the best add-on dates during the growing season. In both seasons the experiment included five nitrogen fertilization treatments with ammonium sulphate (20.6% N) as soil applications. The highest value of absorbance ratio A1658cm⁻¹/A2921cm⁻¹ (Nitrogen content) absorbance ratio A1155cm⁻¹/A2921 cm⁻¹ (pyrophrine ring the basic structure of chlorophyll molecule) and absorbance ratio A1049cm⁻¹/A2921 cm⁻¹ (starch) were obtained as a result of ammonium sulphate application at the rate of 1kg/tree in each of February, May, June and August (3.6, 2.1 and 3.08 respectively) meanwhile the lowest value was obtained in control trees (3.02, 1.61 and 2.76 respectively).

Key words: full bloom stage – Manzanillo - infrared absorption Spectroscopy.

Introduction

Infrared (IR) absorption spectroscopy is certainly one of the most important analytical techniques. One of the great advantages of infrared absorption spectroscopy is that study any sample in any state. Infrared absorption spectroscopy in identifying the molecular structure of materials organic and inorganic in the three forms solid, liquid and gas as well as to identify mutations that occur as a result of various treatments. Infrared absorption spectroscopy is a technique based on the vibrations of the atoms of a molecule. In fact, it demonstrated recently that the IR technique can be effectively used to determine the water contents of leaves (Ning et al., 1996 and Chieu and Victor, 2004) and soils (Xu et al., 1998), to determine the nitrogen concentration in tissues (Young et al., 1997) and the soluble solids content in processing tomatoes (Peris et al., 1998), as well as to measure acidity, soluble solids, and firmness of Jonagold apples (Lammertyn et al., 1998). McGlone and Kawano (1998) using IR spectroscopy to determine dry-matter and soluble solids content of kiwifruit, oil content in groundnuts (Misra et al., 2000), free fatty acids content in sunflower (Moschner and Biskeupe- Korell, 2006) and measured the quality of intact olive cvs. (Ayvalik and Gemlik) including firmness, oil content and colour (Ismail et al., 2009). An infrared spectrum commonly obtained by passing infrared radiation through a sample and determining what fraction of the incident radiation is absorbed at a particular energy. The energy at which any peak in an absorption spectrum appears corresponds to the frequency of a vibration of a part of a sample molecule (Stuart, 2004). The main aim of this investigation is to study the possibility of using infrared absorption spectroscopy in studying the effect of application times of ammonium sulphate on some constituents of Manzanillo olive leaves at full bloom stage. Also minimize the quantity of annually added amounts by 20% and the best add-on dates during the growing season. In both seasons the experiment included five nitrogen fertilization treatments with ammonium sulphate (20.6% N) as soil applications.

Material and Methods

This study was carried out during two successive seasons 2009 and 2010 on ten-years-old Manzanillo olive trees, planted at 5 X 5 m and grown in sandy soil in a private orchard located at Cairo–Alexandria desert road (about 50 Km from Cairo), Egypt. Trees were of normal growth, uniformed in vigour and received the same horticultural practices. This experiment aimed to study the effect of nitrogen fertilization dates on nitrogen, photosynthetic pigments and starch contents of Manzanillo olive leaves using FT-IR spectroscopy at full bloom stage.

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Also minimize the quantity of added annually amounts by 20% and the best add-on dates during full bloom stage. In both seasons the experiment included five nitrogen fertilization treatments with ammonium sulphate (20.6%N) as soil applications and the applied treatments involved the following:

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Date of application</th>
<th>Amount of ammonium sulphate (Kg/tree)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>January</td>
<td>1</td>
</tr>
<tr>
<td>March</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>April</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>May</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>June</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>July</td>
<td>-</td>
<td>1</td>
</tr>
<tr>
<td>August</td>
<td>-</td>
<td>1</td>
</tr>
</tbody>
</table>

Twenty leaves from the middle portion at one year old shoot each treated tree were taken in full bloom in May. The samples were dried in an oven at 50°C for 2-3 h then grounded by mortar and passed through 125-mesh sieves. Two mg of the sample was mixed with 198 mg of pure KBr (potassium bromide) to give 1% concentration. The mixture was carried out for suitable time in an agate mortar. The mixture was pressed in special mold under hydraulic press (at 40 KN) to form a transparent disk, diameter 13 mm approximately. IR spectra leaves were determined by infrared spectrophotometer.

Jasco FT/IR-430 Fourier Transform Infrared Spectrometer was used for recording the IR spectra. Spectra were recorded in a spectral range of 400-4000cm⁻¹ and the obtained spectrum was automatically recorded on a computer.

For quantitative measurements, the absorbance ratios (amount of infrared absorbance by the various bands in the molecule) were calculated using software Jasco by the Beer-Lambert’s law. Where there is a direct correlation between the absorbance and the number of molecules absorbent material to radiation or concentration. Then absorbance was measured using the base line method as the vertical distance from the maximum absorption to the base line connecting the two wings of the band (Fig. 1). Absorbency C-H, N-H, C-N-C and C-O were measured to determine absorbance ratio to track the amount of change in chlorophyll molecule, protein and starch as a result of application with ammonium sulphate during the growing season.

Fig. 1: The base line method.

Results and Discussion

The result of FT-IR spectrum leaves at full bloom in May. Careful examinations of Fig.(1) shows that values of all absorbance ratios were increased with increasing the N-fertilizer levels (Table 1).
Fig. 2: FT-IR spectra of leaves at the full bloom in May.

T1. Trees were fertilized on Jan.(1k/tree), Mar.(1k/tree), May (1k/tree) and Aug.(1k/tree).
T2. Trees were fertilized on Jan.(0.5k/tree), Mar.(1.5k/tree), June(1.5k/tree) and Aug. (0.5k/tree).
T3. Trees were fertilized on Jan. (1k/tree), Mar. (1.5k/tree), May (1k/tree) and Aug. (0.5k/tree).
T4. Trees were fertilized on Feb. (1k/tree), May (1k/tree), June (1k/tree) and Aug. (1k/tree).
T5. Control (recommended dose according to the ministry of Agriculture).

Table 1: Effect of fertilization with ammonium sulphate on absorbance ratios A1658cm\(^{-1}\)/A2921cm\(^{-1}\), A1155cm\(^{-1}\)/A2921cm\(^{-1}\) and A1049cm\(^{-1}\)/A2921 cm\(^{-1}\) [May].

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Absorbance ratio A1658 cm(^{-1})/A2921 cm(^{-1}) (Nitrogen content)</th>
<th>Absorbance ratio A1155 cm(^{-1})/A2921 cm(^{-1}) (chlorophyll)</th>
<th>Absorbance ratio A1049 cm(^{-1})/A2921 cm(^{-1}) (starch)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.14</td>
<td>1.78</td>
<td>2.86</td>
</tr>
<tr>
<td>2</td>
<td>3.38</td>
<td>1.97</td>
<td>2.99</td>
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<tr>
<td>3</td>
<td>3.5</td>
<td>2.02</td>
<td>3.06</td>
</tr>
<tr>
<td>4</td>
<td>3.6</td>
<td>2.1</td>
<td>3.08</td>
</tr>
<tr>
<td>Control</td>
<td>3.02</td>
<td>1.61</td>
<td>2.76</td>
</tr>
</tbody>
</table>

a. Absorbance ratio A1658 cm\(^{-1}\)/A2921 cm\(^{-1}\) (Nitrogen content):

The results illustrated in Table (1) & Fig. (3) show that the highest value of absorbance ratio A1658cm\(^{-1}\)/A2921cm\(^{-1}\)(Nitrogen content) was obtained with application ammonium sulphate at rate of 1kg/tree in each of February, May, June and August since it reached (3.6), meanwhile the lowest value was obtained in control trees (3.02).

Fig. 3: Effect of fertilization with ammonium sulphate on absorbance ratios A1658 cm\(^{-1}\)/A2921 cm\(^{-1}\), A1155cm\(^{-1}\)/A2921cm\(^{-1}\) and A1049cm\(^{-1}\)/A2921 cm\(^{-1}\) [May].
b. Absorbance ratio $A_{1155 \text{ cm}^{-1}}/A_{2921 \text{ cm}^{-1}}$ (Chlorophyll):

Regarding absorbance ratio $A_{1155 \text{ cm}^{-1}}/A_{2921 \text{ cm}^{-1}}$ (pyrophrine ring the basic structure of chlorophyll molecule), results illustrated in Table (1) & Fig.(3) show that the highest value was recorded with application of ammonium sulphate at rate of 1kg/tree in each of February, May, June and August (2.1), meanwhile the lowest value was obtained in control trees (1.61).

c. Absorbance ratio $A_{1049 \text{ cm}^{-1}}/A_{2921 \text{ cm}^{-1}}$ (starch):

Concerning absorbance ratio $A_{1049 \text{ cm}^{-1}}/A_{2921 \text{ cm}^{-1}}$ (starch), results illustrated in Table (1) & Fig.(3) show that the highest value was recorded with application ammonium sulphate at rate of 1kg/tree in each of February, May, June and August (3.08), meanwhile the lowest value was obtained in control trees (2.76).

From the illustrated data it can be concluded that fertilization with ammonium sulphate at rate 1 kg / tree in each of February, May, June and August resulted in the highest values of nitrogen, chlorophyll and starch. Also this treatment reduced the added amount of ammonium sulphate with 20 % .

References


