

## The sludge and manure effects on the growth of citrus trees

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*Abstract:* The objective of this study was to evaluate the agricultural use of organic solid wastes. Hence two types of wastes were selected to be applied in a citrus orchard: the sewage sludge (the most common on the urban world) and the manure (the most common on the agricultural fields). The sewage sludge is a residue originated from the wastewater treatment – the solid phase. Its application as an organic fertilizer may represent an alternative to the pollution consequences in nature. The effect of sludge and manure application on the growth of orange trees (*Citrus sinensis* [L.] Osb.), established on a Fluvisol, was evaluated in relation to the biometric parameters – number of leaves per tree, diameter of the trunk, leaf area, specific leaf area and leaf chlorophyll (estimated by using SPAD values). In general, the citrus plants response to the application of sewage sludge and of manure was positive, compared to the control trees. The application of the manure and especially the sewage sludge (once this fertilizer/soil amendment is available in larger amounts) may be a profitable alternative application to the use of mineral fertilizers and to other organic soil amendments. Moreover, as the other possible destinations of sewage sludge (landfill or incineration) are associated to potential environmental problems, its agricultural use when correctly applied, represents a better alternative. The application of this solid wastes, as a clean and safe technique to preserve the environment contamination, can also be successfully used as an organic amendment of citrus orchards.

*Key-Words:* citrus leaves, manure, plant development, sewage sludge, soil organic amendments

## 1 Introduction

In the last five decades and especially in the last years, the improving of fertilizers use in crops has been a trend on agricultural studies especially the organic amendment management [1, 2].

The economic aspects related to the application of treated sewage sludge to the soil, are principally originated by better crop productions and by the decreasing of chemical fertilizers and amendments that it represents [3, 4].

Social and environmental requirements to give an adequate destination to sewage sludge have stimulated the use of this material as soil fertilizer or amendment. This alternative allows the utilization of minerals and organic matter present in sewage sludge, which can be beneficial for the soil structure and fertility, and, consequently, for the growth of plant species.

In the Algarve, sewage sludge, due to their reduced toxicity (trace elements), represents an advantage for agriculture use [5] and has been occasionally used as fertilizer, in citrus and in carob trees (*Ceratonia siliqua*). The application of sludge in agriculture has some potentialities in the sea shore and mountain areas, where the soils are characterized by low pH and low organic matter content. Thus, the application of these residues, as soil amendment, may represent an advantage for the crop productions [6].

With the World population's growth, the demand of fertilizers was expected to rise. World fertilizer production was expected to outstrip demand over the 2008-2013 period and should support higher levels of food and biofuel production. The FAO report estimated that world fertilizer supply (nitrogen, phosphate and potash nutrient) should increase by some 34 million tonnes representing an annual growth rate of 3 percent between 2007/08 and 2011/12, comfortably sufficient to cover demand growth of 1.9 percent annually [7].

Total production is expected to grow from 206.5 million tonnes in 2007/08 to 241 million tonnes in 2011/12. Fertilizer demand will

increase from 197 million tonnes today to 216 million tonnes in 2011/12.

According to the same authors, world nitrogen supply is forecast to rise by 23.1 million tonnes by 2011/12; world phosphate fertilizer supply will increase by 6.3 million tonnes and potash supply by 4.9 million tonnes.

The population increase and the industrial development produce enormous amount of organic residues which, nowadays, generate great environmental problems. On the other hand, inorganic fertilizers production consumes energy and its resources are limited. A potential alternative for reduce the inorganic fertilizers use consists in a better use of the organic residues. By using sewage sludge, improving crop yield may be achieved with less chemical hazardous pollution as it was demonstrated, when compared to application of inorganic fertilizers [8]. Currently, new organic wastes appear at a significantly extent. Their deposition in landfills and incineration must be avoided.

A brief classification of organic residues which may be used in agricultural soils is shown in Table 1 [9].

Table 1. Brief classification of treated residues used in the agricultural soils

Organic wastes/residues		References
Urban and Industrial	Urban sludge Industrial sludge	[10, 11, 12]
Mills	Mill manure	[13, 14]
Livestock	Manure	[15, 16, 17]
Agro-industry	Carob pulp Sugar cane pulp Wine pulp Beer pulp Olive pulp	[9, 18]

The effects of manure and treated sewage sludge application were studied in a citrus orchard. The aim of this work was to show the effects of these wastes on the plant growth.

## 2 Material and methods

### 2.1 Experimental procedure

An experimental plot was established in an orange (*Citrus sinensis* [L.] Osb.) orchard, in the Algarve, southern Portugal, on the same conditions, as related in [19].

A 20 ha plantation (6 m x 2 m tree compass) took place in May and twelve trees were chosen to conduct the experiments. A drip irrigation system was used – Ram Netafim self-compensating drippers, incorporated in the pipes, at a distance of 0.75 cm with a constant 2.3 L h<sup>-1</sup> discharge.

The irrigation periods occurred six months per year, between May and October.

The amount of water was 4.6 L per tree per day (700 m<sup>3</sup>ha<sup>-1</sup>year<sup>-1</sup>).

Two different types of organic soil amendments were tested: sewage sludge (S) and bovine manure (M), which were compared to the control treatment (C), where any fertilizer application was done.

The experimental procedure was a randomized block design with 3 treatments x 4 replications, in a total of 12 young orange trees.

### 2.2 Soil

The soil of the experimental site was classified as Fluvisol [20], defined as an alluvial soil, till 1 m depth, characterized by a “Terric” superficial layer (FLtr), originated from calcareous materials.

The soil was characterised for texture, pH(H<sub>2</sub>O), organic matter (OM), electric conductivity (EC, dSm<sup>-1</sup>), N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O, Fe, Mn, Zn, total and active lime (CaCO<sub>3</sub>), C, cation exchangeable capacity (CEC) and the ratio of C to N (C/N) was also calculated. These results are presented in Table 2.

Table 2 – Soil physical and chemical parameters before the plantation of the orange trees.

Soil Parameters	Values
Texture class	Silt loam
Sand (%)	28.1
Silt (%)	45.5
Clay (%)	26.3
pH (H <sub>2</sub> O)	8.0
OM (%)	1.8
EC (dS m <sup>-1</sup> )	0.9
N (%)	0.1
P <sub>2</sub> O <sub>5</sub> (g kg <sup>-1</sup> )	29.1
K <sub>2</sub> O (g kg <sup>-1</sup> )	617
Fe (g kg <sup>-1</sup> )	250
Mn (g kg <sup>-1</sup> )	128
Zn (g kg <sup>-1</sup> )	4.0
Total lime (%)	21.1
Active lime (%)	10.2
Organic Carbon (%)	1.0
C/N	12.3
Ca (cmol <sub>c</sub> kg <sup>-1</sup> )	7.5
Mg (cmol <sub>c</sub> kg <sup>-1</sup> )	4.4
Na (cmol <sub>c</sub> kg <sup>-1</sup> )	1.7
K (cmol <sub>c</sub> kg <sup>-1</sup> )	0.7
Exchangeable H (cmol <sub>c</sub> kg <sup>-1</sup> )	0,0
CEC (cmol <sub>c</sub> kg <sup>-1</sup> )	14.7

### 2.3 Climate

The climate of the Algarve can be considered as Mediterranean and in particular the south shore, according to Köppen, is classified as *Csa*, with semi-arid characteristics, identified by mild rainy winters and warm-dry summers. Snow and frost are not expected. The annual average temperature, in Faro, ranges between 12°C in January and 23°C in August, a difference of only 11°C [21]. The annual average value for potential evapotranspiration (ETp; Penman

Monteith) was 1280 mm, and for precipitation around 450 mm.

ETp and precipitation have an opposite pattern, during the year. So, during the hottest semester, when the photosynthesis is more efficient and the water is more essential for plant growth, this resource is less available; irrigation is the only solution to balance this situation. The average of the shortage of water, defined by the difference between the values of evapotranspiration and precipitation rates, verified along the driest months, reaches 940 mm per year.

### 2.3 Amendments

The treated sewage sludge was collected from the urban wastewater treatment of Vilamoura (Algarve). The manure came from a beef cattle stable near the experimental site. These amendments were applied to the soil at a depth between 0.5 and 1.0 m, during the plantation of the trees. The physical and chemical parameters of the soil amendments are shown in Table 3.

Table 3 – Physical and chemical parameters of the soil amendments.

Parameters	Sludge (S)	Manure (M)
pH (H <sub>2</sub> O)	6.6	7.4
OM (%)	55.7	72.4
EC (dSm <sup>-1</sup> )	14.5	21.2
Water content (%)	12.3	7.5
N (g kg <sup>-1</sup> )	31.7	23.8
P (g kg <sup>-1</sup> )	14.5	9.70
K (g kg <sup>-1</sup> )	10.9	31.0
Ca (g kg <sup>-1</sup> )	52.5	28.1
Mg (g kg <sup>-1</sup> )	1.60	3.20
C/N	17.6	30.4

The amounts of sludge and manure applied per tree (g) and respective nutrient contents (N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O and MgO) are presented in Table 4.

The sludge and manure quantities applied under each tree were 9.5 kg and 12.6 kg. These amounts represent the needs of the plant, in relation to N, during the first 3 years of growth.

Sewage sludge and manure were applied and incorporated to the soil at one meter depth before the citrus tree plantation.

### 2.4 Plant analysis

Several plant biometric determinations were done on the end of the 1<sup>st</sup> and 2<sup>nd</sup> experimental year, in November, as follows: number of leaves per tree (NLT); trunk perimeter (TP); leaf area (LA); specific leaf area (SLA), specific leaf disk area (SLDA) and degree of leaf chlorosis (SPAD).

Table 4 – Amounts and nutrient contents of sludge (S) and manure (M) applied per tree (g).

	Sludge (S)	Manure (M)
Amount	9500	12600
N	300	300
P <sub>2</sub> O <sub>5</sub>	314	280
K <sub>2</sub> O	124	470
Mg	15	40

The number of leaves per tree (NLT) is a biologic parameter used to define the vegetative development of the trees.

The trunk perimeter (TP, cm) was measured at a 20 cm height. This is a no destructive biologic parameter and is used to define the trees vigour and it is easy to determine and very explicit in terms of results [22].

Leaf area (LA, cm<sup>2</sup>) is considered as a biometric parameter which defines the vegetative development, in function of time and of the experimental treatments. The LA was measured using a leaf area measurer “AT Delta-T” (Devises Lda., Burwell, Cambridge, UK).

The specific leaf area (SLA, cm<sup>2</sup>g<sup>-1</sup>) is expressed as the ratio of leaf area (LA) to leaf dry weight [23]; this parameter is useful to evaluate the vegetative development, as a function of time and of the experimental

treatments; in each treatment, 5 leaves per tree were collected, oven dried at 60°C, until constant weight and the respective dry weight was determined.

The specific leaf disk area (SLDA, cm<sup>2</sup>) was determined in leaf discs of 50.3 mm<sup>2</sup>, in different leaves, then oven dried at 60°C and weighted, in a precision scale. This parameter is also useful to evaluate the vegetative development.

The degree of leaf chlorosis [24] was evaluated using a portable SPAD-502 meter (Minolta Co., Osaka, Japan). This apparatus measures the absorbances of the leaf at two wavelengths in the red range (~ 650 nm), which corresponds to the pick of absorption by chlorophyll and in the infrared range (~ 940 nm), used as reference. Using these two transmittances, the SPAD-meter calculates a numerical value (SPAD units), which is proportional to the amount of chlorophyll in the citrus.

The number of samples was 59 for each treatment and parameter, except to the number of leaves per tree (NLT), which was reduced to 8.

## 2.5 Statistical analysis

The effects of treatments were evaluated using analysis of variance (ANOVA) and the means compared using the Duncan Multiple Range Test (DMRT) at the 95% significance level, using the SPSS 11.0 [25].

## 3. Results

### 3.1 Sludge and manure effects on the 1st year

The ANOVA was done between the means of the parametric values of the plant development to the treatments.

After the first experimental year, significant differences were observed among the mean values of trunk perimeter (TP); leaf area (LA); specific leaf area (SLA), specific leaf disk area

(SLDA) and degree of leaf chlorosis (SPAD). The differences observed among the mean values of NLT (number of leaves per tree) were not significant.

The statistical Duncan test was used to the TP values and the mean values were associated into 2 homogeneous groups (a) and (b). It was observed higher values in the S and M treatments than in control (C).

Fig. 1 shows the mean values, standard deviation and the Duncan test of the perimeter of the trunk values (TP, cm) among treatments. A positive relation was observed between the use of the S and M amendments and the perimeter of the trunks values (TP).

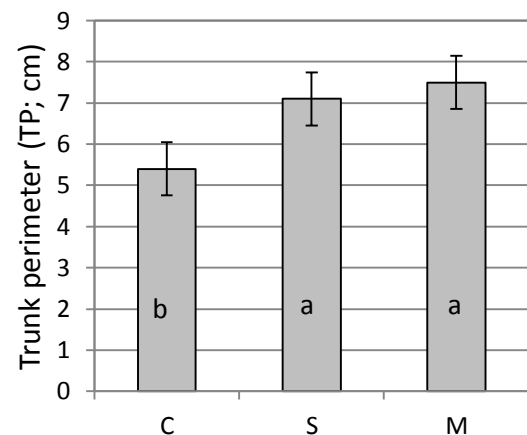


Fig. 1 – 1<sup>st</sup> year - Trunk Perimeter (TP, cm) values for the treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at  $P < 0.05$  (Duncan Test).

Fig. 2 shows the mean values, standard deviation and the Duncan test of the leaf area (AF cm<sup>2</sup>) among treatments. It was shown that the values of manure (M) treatment were significantly higher than those of control (C). Thus, it was observed a positive relation between the manure application and the plant development.

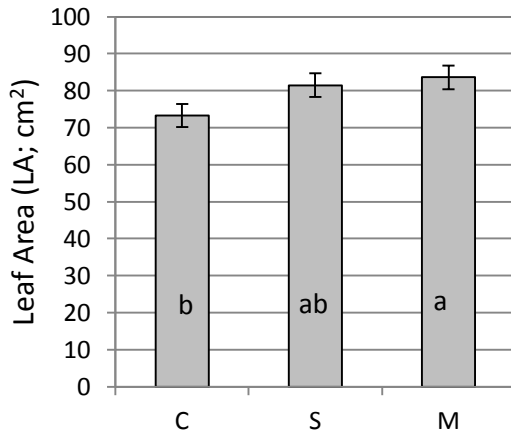


Fig. 2 – 1<sup>st</sup> year - Leaf area (LA, cm<sup>2</sup>) values for the different treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at P<0.05 (Duncan Test).

Fig. 3 shows the mean values, standard deviation and the Duncan test of the specific leaf area (SLDA, cm<sup>2</sup>g<sup>-1</sup>), among treatments. The SLDA was significantly lower in plants treated with sludge (S) and manure (M), compared to control (C). This shows that the leaves of plants of control were thinner than those where manure and treated sewage sludge were applied.

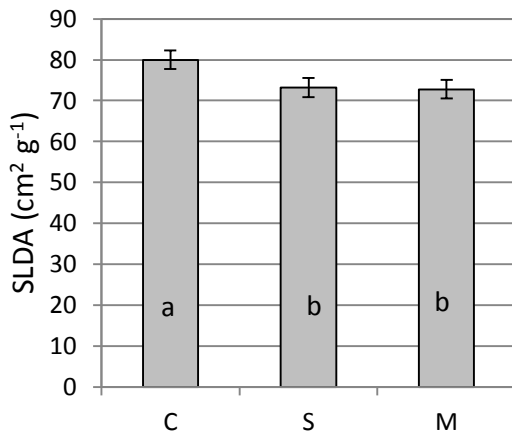


Fig. 3 – 1<sup>st</sup> – year – Specific leaf disk area (SLDA, cm<sup>2</sup>g<sup>-1</sup>) values for the different treatments: C – control, S – sewage sludge, M –

manure. Columns with the same letter were not significantly different at P<0.05 (Duncan Test).

Fig. 4 shows the mean values, standard deviation and the Duncan test of the specific leaf area (SLA, cm<sup>2</sup>g<sup>-1</sup>), among treatments. Specific leaf area was significantly affected by the treatments. This means that the leaves of plants of manure (M) were thicker than those where organic amendments were not applied.

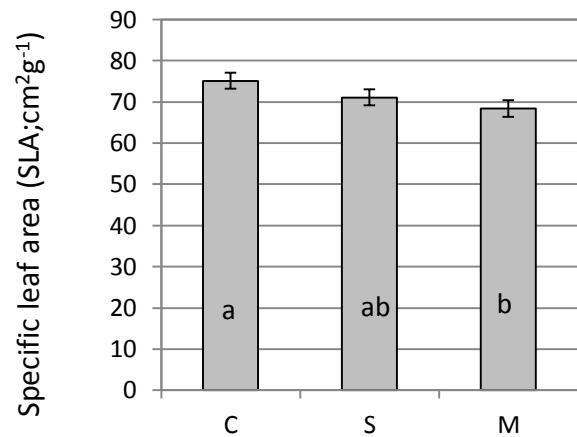


Fig. 4 – 1<sup>st</sup> year – Specific leaf area (SLA, cm<sup>2</sup>g<sup>-1</sup>) values for the different treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at P<0.05 (Duncan Test).

Fig. 5 shows the mean values, standard deviation and the Duncan test of the SPAD values among treatments. SPAD values were within the range obtained by [33] for green leaves in citrus trees, indicating an adequate nutritional level of the plants in this experiment.

The SPAD was significantly higher in plants treated with S and with M, compared to C. This shows that, in control plants, the chlorophyll level was affected probably due to nutritional imbalances, as iron chlorosis, which was frequently observed in citrus established in calcareous soils.

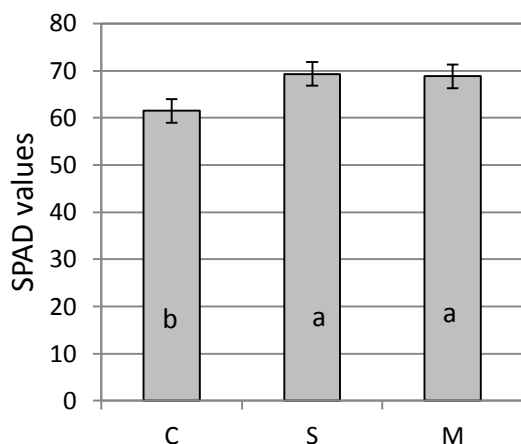


Fig. 5 – 1<sup>st</sup> year – SPAD values for the different treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at  $P < 0.05$  (Duncan Test).

### 3.2 Sludge and manure effects on the 2nd year

After the second experimental year, significant differences were observed among the mean values of trunk perimeter (TP), leaf area (LA), and specific leaf disk area (SLDA).

The number of leaves per tree (NLT), specific leaf area (SLA), and SPAD values were not significantly affected by the treatments, and are shown in Table 5.

Table 5 - Number of leaves per tree (NLT); specific leaf area (SLA,  $\text{cm}^2\text{g}^{-1}$ ) and total leaf chlorophyll (SPAD) for each treatment: sewage sludge (S), manure (M) and control (C).

	NLT	SLA	SPAD
C	396	54.0	76.4
S	510	52.1	75.6
M	447	53.1	74.9

Fig. 6 shows the variation of the perimeter of the trunk values (TP, cm) among treatments. A positive response was observed by the organic wastes application. In relation to the perimeter of

the trunks values (TP), it was observed that sludge (S) and manure (M) were both significantly higher than the control treatment (C). The young citrus plants amendment with tested wastes presented higher values of trunk perimeter (about 69%) compared to the control plants.

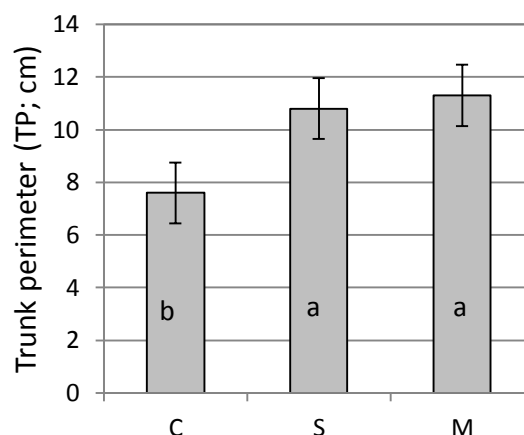


Fig. 6 – 2<sup>nd</sup> year – Trunk perimeter (TP, cm) values for the treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at  $P < 0.05$  (Duncan Test).

Fig. 7 shows the values of leaf area (LA,  $\text{cm}^2$ ) for each treatment. The leaf area increased with sludge (S) application. It was shown that the sewage sludge application improved the larger leaves, compared to the manure (M) and to the control (C).

Fig. 8 shows the specific leaf disk area values (SLDA,  $\text{cm}^2\text{g}^{-1}$ ). The SLDA was significantly lower in plants treated with sludge, compared to control (C). This means that the leaves of plants treated with sludge were thicker than those of the control (C).

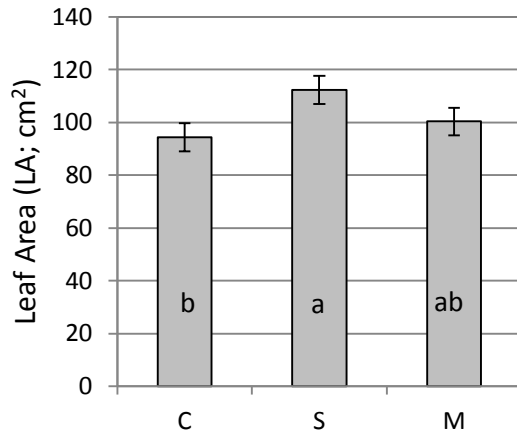


Fig. 7 – 2<sup>nd</sup> year – Leaf area (LA, cm<sup>2</sup>) values for the different treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at  $P < 0.05$  (Duncan Test).

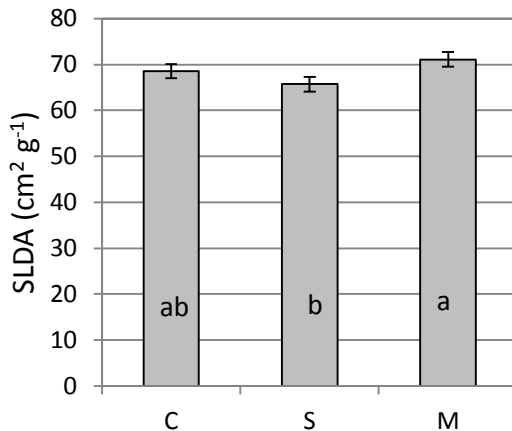


Fig. 8 – 2<sup>nd</sup> year – Specific leaf disk area (SLDA, cm<sup>2</sup>g<sup>-1</sup>) values for the different treatments: C – control, S – sewage sludge, M – manure. Columns with the same letter were not significantly different at  $P < 0.05$  (Duncan Test).

#### 4. Conclusions

This work shows that the treated sewage sludge and the manure are good soil organic amendments, once it increases the response of plants - perimeter of the trunk, leave area,

thickness of the leaves and leaf chlorophyll level (the number of leaves per tree was not significantly different). These organic wastes application represents a profitable alternative to the use of mineral fertilisers, especially the sewage sludge, since it is available in larger amounts. Moreover, as the possible destinations of urban sludge (landfill or incineration) provoke environmental problems, its agricultural use, when correctly applied, is essential to avoid these problems.

Considering the specific case of the application of sewage sludge our results were similar to those obtained by [26], where the application of sewage sludge increased the values of leaf area, total dry matter, leaf area ratio and rates of absolute and relative growth of the plants of eucalyptus (*Eucalyptus grandis*), and [27] there was an increase in leaf area of mint (*Mentha piperita* L.) treated with sewage sludge.

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