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Quantification of biopigments from *Nerium oleander* L. and assessing their antioxidant potential

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Abstract
The present investigation was to quantify the biopigments from *Nerium oleander* L. and also to assess the antioxidant potential. Two different Nerium cultivars Red and Pink type were selected for the experiment. The collected flowers were dried under shade for the removal of moisture content and also to prevent the destruction of anthocyanins, as they are sensitive to light. The dried flowers were grinded to obtain fine powder. Different solvents were used for the extraction of anthocyanin and for the estimation of their antioxidant potential from the flower sample. The solvents used were Acetone, Ethanol, Methanol and Distilled Water in different concentrations. It provides measurements in the UV and VIS range. It has a spectral range of 220nm to 830nm. Based on the conducted experiments, it is visible that the anthocyanin content extracted using the solvent Ethanol has higher anthocyanin when compared with other solvents. The anthocyanin content is high in 100% ethanol (3.54 mg/k 3 g eq) and 100% methanol extraction in red flowers (3.40 mg/k 3 g eq): The antioxidant potential is high in 100% ethanol (2.97 %) and 50% ethanol (2.53 %) extraction in red flowers.

Keywords: Nerium, solvent, anthocyanin, antioxidant

Introduction
Nerium oleander is an evergreen shrub in the family Apocynaceae. It is the only species currently classified in the genus Nerium. It is commonly known as oleander. The *oleander* is most prevalent, and its alluring flowers make it a particular hazard for accidental ingestion. The plant contains a number of related cardiac glycosides similar in activity to digitalis. The main glycosides are oleandrin, neriine, Cardenolides, gentiobiosyl, oleandrin and odoroside are also present. In addition, a variety of other pharmacologically active compounds, including folinerin, rosagenin, rutin and oleandomycin have been identified in the plant. The total level of antioxidant activity was higher in Crude *Nerium oleander* leaves extract (72.8%), as compared to flower (68%) and Superoxide radical whereas scavenging activity was higher in Crude flower extract (66%) as compared to leaves 25%. Anti-aging skin creams are abundant in today's cosmetic market place. Future product development includes nerium AD Eye Cream Spot Cream, Skin Repair Cream, Blemish Cream and lots of such products. *Nerium oleander* aqueous extract is used as a novel anti-HIV therapeutic.

Materials and Methods
An experiment was conducted at Faculty of Horticulture, Tamil Nadu Agricultural University, Coimbatore during 2018-2019. The main aim of this experiment was to standardize the solvent for extraction of biopigments, to assess the anthocyanin extract of different nerium collections and to assess their antioxidant potential.

To standardize the solvent suitable for extraction of anthocyanin from nerium flowers
The experiment was carried out in Completely Randomized block Design with 6 treatments each with three replications. Solvents such as Methanol (50 and 100% concentration), Ethanol (50 and 100 % concentration), Acetone and distilled water of 100% concentration were used. 2.5 g of the powdered flower sample (Red and Pink) was taken in a conical flask and were added with the respective solvents of different concentrations.
The samples were then foiled and placed in the mechanical shaker for 12 hours for complete diffusion of the pigments into the solvent. They were then filtered by using Whattman Filter No.16 and the extracts were collected in a beaker and foiled.

To assess the anthocyanin extract of different nerium collections

The experiment was carried out in Completely Randomized block Design (CRD) with the cultivars of Red and Pink nerium flowers, to assess the cultivar possessing higher anthocyanin content. After the extraction, the sample is prepared for anthocyanin estimation. Monomeric anthocyanin content was measured by spectrophotometric pH differential diagnosis protocol.

The extracts were mixed thoroughly with 0.025M KCl as pH 1 buffer in 1:3 ratio of extract to buffer. The absorbance measured at 520 and 700 nm against distilled water as blank. The extracts were then combined similarly with sodium acetate buffer pH 4.5.The absorbance of these solutions were measured at the same wavelengths. The anthocyanin content was calculated, expressed as cyanidin-3-gluciside equivalent in mg/l.

$$\text{Total monomeric anthocyanin (mg/k}^3\text{ g eq)} = \frac{\text{AxMWxDFx10}^3}{\varepsilon \text{xl}}$$

Where,

- A (absorbance) = (A520nm-A700nm) pH1.0 - (A520nm-A700nm) pH4.5
- MW (molecular weight) = 449.2g/mol for cyanidin-3-glucoside.
- DF= Dilution factor
- ε= Molar extinction coefficient of cyd-3-glu(26900 L/mol cm)
- l= Path length in cm
- 10^3= Factor for conversion from g to mg

The statistical analysis of data was carried out as suggested by Padma S. Vankar *et al*. (2010) [1].

To assess the antioxidant potential of nerium flowers through DPPH assay

The experiment was carried out in Completely Randomized block Design with the extracts from 6 treatments and 3 replications to assess the antioxidant potential using DPPH assay. The antioxidant activity of different extracts of nerium flowers were measured using the DPPH (2, 2-diphenyl-1-picryl hydrazyl hydrate) radical scavenging assay.

3.0ml reaction mixture which comprises of 1.0ml of DPPH in 0.3mM methanol, 1.0ml of the extract and 1.0ml of methanol. The blank is the respective solvent used for the extraction of pigments.

The reaction mixture is incubated for 10 mins in dark and then the absorbance is measured at 517nm and the radical scavenging activity was obtained by,

$$\text{Radical scavenging activity \%} = \frac{\text{(OD Control-OD sample)}}{\text{OD control}} \text{ X } 100$$

The statistical analysis of data was carried out as suggested by Mariam Mohadjerani (2012).

Results and Discussion

Nerium oleander L. is one of the important ornamental flowering shrubs which finds place in all gardens. This ornamental shrub is suitable for commercial cultivation in all the tropical regions. The important use of nerium flowers are as loose flower for religious purpose and for garland making in addition, they are preferred for growing as shrubs in the garden along a boundary wall or to mask some areas of lawn such as picnic spots.

Nerium oleander L. is one of the important ornamental flowering shrubs which finds place in all gardens. This ornamental shrub is suitable for commercial cultivation in all the tropical regions. The important use of nerium flowers are as loose flower for religious purpose and for garland making in addition, they are preferred for growing as shrubs in the garden along a boundary wall or to mask some areas of lawn such as picnic spots.

In recent days, nerium has great demand in landscape architecture for beautification of home gardens, industrial gardens, public gardens, road dividers in highways, railway stations, airport surroundings and historical monuments.

Table 1: Quantification of biopigments (The values are expressed in mg/k^3 g eq)

Treatment	Red				Pink			
	R1	R2	R3	Mean	R1	R2	R3	Mean
100% Ethanol	3.71	3.51	3.40	3.54	3.5	3.48	3.38	3.47
50% Ethanol	3.39	3.34	3.29	3.34	3.31	3.26	3.24	3.27
100% Methanol	3.46	3.39	3.35	3.40	2.73	2.65	2.63	2.67
50% Methanol	3.24	3.21	3.18	3.21	2.32	2.26	2.23	2.27
100% Acetone	3.24	3.21	3.12	3.19	3.18	3.14	3.10	3.14
100% Water	1.98	1.94	1.87	1.93	1.07	1.01	0.95	1.01
SEd	0.08				0.04			
CD(.05)	0.19				0.09			

Table 2: Assessment of antioxidant potential

Treatment	Red (%)				Pink (%)			
	R1	R2	R3	Mean	R1	R2	R3	Mean
100% Ethanol	3.01	2.96	2.94	2.97	2.70	2.64	2.61	2.65
50% Ethanol	2.55	2.53	2.51	2.53	2.41	2.34	2.30	2.35
100% Methanol	2.38	2.32	2.29	2.33	2.11	2.05	2.02	2.06
50% Methanol	1.44	1.39	1.34	1.39	1.27	1.21	1.18	1.22
100% Acetone	1.63	1.60	1.57	1.60	1.04	1.01	0.98	1.01
100% Water	2.72	2.42	2.24	2.46	1.62	1.56	1.53	1.57
SEd	0.08				0.04			
CD(.05)	0.18				0.08			

Apart from these nerium also has a significant role in extraction of anthocyanins, which are useful in making dyes, antimicrobial and antioxidant preparations.

Standardization of solvent

Based on the conducted experiments, it is visible that the anthocyanin content extracted using the solvent Ethanol has higher anthocyanin when compared with other solvents. Hence, ethanol solvent is regarded best for the extraction of anthocyanin from nerium flowers in both red and pink cultivars.

Though methanol has extraction potential similar to that of ethanol, it cannot be widely used because of its toxicity. This is in agreement with the results of Julia Martin *et al*. (2016).

Assessment of anthocyanin content

The anthocyanin content is high in 100% ethanol (3.54 mg/k 3 g eq) and 100% methanol extraction in red flowers (3.40 mg/k 3 g eq) followed by pink flowers in 100% ethanol (3.47 mg/k 3 g eq) and 50% ethanol (3.27 mg/k 3 g eq).

The anthocyanin content of both red and pink cultivars are approximately equivalent, but there is a visual variation in the colour of the crude extracts. Significant differences were observed among the nerium cultivars estimated for anthocyanin. The maximum anthocyanin content (3.54 mg/k 3 g eq) was recorded in red flowers extracted by 100% ethanol followed by red flower (3.40 mg/k 3 g eq) extracted by 100%

methanol, while the minimum anthocyanin is recorded in pink flower (1.93mg/k 3 g eq) extracted by 100% distilled water.

Assessment of antioxidant potential

The antioxidant potential is high in 100% ethanol (2.97 %) and 50% ethanol (2.53 %) extraction in red flowers followed by pink flowers in 100% ethanol (2.65 %) and 50% ethanol (2.35%) extraction.

Significant differences were observed among the nerium cultivars estimated for antioxidant potential estimated by DPPH assay. The maximum antioxidant potential was recorded in red flowers (2.97 %) extracted by 100% ethanol followed by pink flowers (2.65 %) extracted by 100% ethanol, while the minimum antioxidant potential is recorded in pink flowers (1.01%) extracted by 100% acetone. This is in accordance with the experiment by Saranya *et al*., 2017 [2].

References

1. Padma Vankar S, Jyoti Srivastava. Evaluation of Anthocyanin Content in Red and Blue Flowers, International Journal of Food Engineering, 2010.
2. Saranya S, Archana D, Santhy KS. Antimicrobial and Antioxidant Effects of *Nerium oleander* Flower Extracts. Int. J Curr. Microbiol. App. Sci. 2017; 6(5):1630-1637.
3. Zhong FL, Sli N, Wang J, Zhao M, Sakai J, Hasegawa T *et al*. Three new triterpenes from *Nerium oleander* and biological activity of the isolate compounds. Natural Products. 2005; 68:198-206, 9.