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Pigments in the petals of Rangoon creeper (*Combretum indicum*) at different stages of flower maturity

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Abstract: *Combretum indicum is a dichogamous and self-incompatible plant. The flowers have a white corolla at anthesis. The petals change colour gradually with the progression of anthocyanin accumulation and turn pale pink, then intermediate pink and lastly deep pink. Different pollinators visit the flowers at the different colour stages. In the present investigation the pH of the petal extract, the percent pink cells in the upper epidermis of the petals, and the photosynthetic pigments chlorophyll a, chlorophyll b and carotenoids in the petals have been studied at the four identified flower colour stages. The pH of the petal extract did not change with the change in flower colour. The number of pink epidermal cells in the upper surface of petals increased from zero to 93% as the petals passed from the white to deep pink stage with the accumulation of anthocyanins, the principal anthocyanin being cyanidin 3-O-glucoside. The photosynthetic pigments chlorophyll a, chlorophyll b and carotenoids were present at all the four flower colour stages. The absorbance of chlorophyll a and chlorophyll b was the highest in the deep pink stage, and that of carotenoids was highest at the white stage. Chlorophyll b showed higher absorbance than chlorophyll a at all the four stages. We suggest that the antioxidant anthocyanin along with chlorophyll b and carotenoids protect the petals from oxidative damage, thus ensuring that the flowers do not wither for three days.*

Key Words: *Combretum indicum, pH, anthocyanin, photosynthetic pigments, antioxidant.*

1. INTRODUCTION:

Combretum indicum (L.) DeFilipps (syn. *Quisqualis indica* L.; common names Rangoon creeper or Burma creeper), is a woody climber with drooping spikes bearing fragrant tubular flowers and is often grown as an ornamental plant. The floral rewards to pollinators are nectar and pollen. A study by Eisikowitch & Rotem [1] in Israel showed that the flowering period for each flower is three days, and seeds do not set. The buds open at dusk; the flowers are white and horizontally oriented, and visited only by hawkmoths during the initial hours from blooming and at night. On the following day the flowers turn pink and later red, become pendulous, and are visited by solitary bees, honeybees, flies, and sunbirds but not hawkmoths. The work of Willmer *et al.* [2] was largely in agreement with that of Eisikowitch & Rotem [1]; and they observed that the diameter of the corolla of pink and red flowers was slightly wider than that of white flowers.

Based on controlled pollinations in Southwest China, Yan *et al.* [3] showed that the species is dichogamous and self-incompatible; and nectar production and scent emission are related to flower colour change. The white flowers emitted scent more than 3-fold that of pink or red flowers. Nectar production was continuous at the white stage; the production declined and then eventually stopped when the flowers had turned red [3]. Cross-pollination was effectively carried out by bees, moths and butterflies; bees collected pollen whereas moths and butterflies collected nectar. In the first hour after anthesis bees visited the flowers; thereafter, only hawkmoths visited the flowers but less frequently till the following morning. Bees and butterflies visited the pink flowers. Very few butterflies visited the red flowers. Fruit set was best when white flowers were pollinated. The study suggested that diurnal pollination compensated where nocturnal pollination was unsuccessful. The combination of signals, namely patterns of nectar secretion and scent emission, and floral colour change, which attracts different pollinators promotes reproductive fitness in *Q. indica* [3] (syn. *C. indicum*). The flowers change colour from white to pink and finally red because of anthocyanin synthesis. Cyanidin 3-O-glucoside

is the major anthocyanin, which accumulates gradually in the vacuoles of the epidermal and subepidermal cells of the petals [4].

Changes in colours of turgid flowers is common in angiosperms, and is observed in at least 456 species covering 268 genera and 78 diverse families [3]. Floral colour changes, which are visual cues to insect pollinators, occur during the ontogeny of flowers [5], indicating functional convergence [6]. The colour change can be in the whole flower or localized to a certain region; and the pigments responsible for colour changes are anthocyanins, carotenoids and betalains, and the most common pigment synthesised is anthocyanin, which has been demonstrated in 68 families [6]. Specific light-reflecting structures on the perianth surface can also produce colour [7]. It is the colour contrast of the flowers compared to the vegetative parts that guides pollinators to the flowers [7].

Flowers change colour either as the flowers age or when flowers have been pollinated. In *Fuchsia excorticata*, *Weigela middendorffiana* and *Combretum indicum* the colour change occurs as the flowers age. In *F. excorticata* flowers the colour change is from green to red; fresh flowers are green, produce nectar and are visited by bellbirds [8]. Red flowers do not produce nectar and are avoided by bellbirds. At least three days are needed for the pollen tubes to reach the ovary and another 1.5 days for the style to abscise; that is the reason the red flowers are retained on the plant [8]. In *W. middendorffiana*, a self-incompatible species pollinated by bumble bees, the corolla is yellow and with age the inner part of the lower corolla changes from yellow to red [9]. The bumble bees get much more nectar and pollen from yellow-phase flowers than from red-phase flowers, and hence prefer yellow-phase flowers. In *W. middendorffiana* the colour change usually occurs simultaneously in all the flowers of an inflorescence; therefore, retention of red-phase low-reward flowers allows the bumble bees to move through quickly. A similar situation of retention of flowers till natural senescence is observed in *C. indicum* to increase floral colour display to pollinators [4]. Pollination-induced floral colour change is reported in *Lantana camara,* and *Tibouchina pulchra* and *T. sellowiana*. The petals of flowers of *L. camara* are yellow at anthesis, and on pollination gradually change colour to orange, scarlet, and finally magenta; the corolla abscission occurs at the magenta stage [10]. Pollination triggers the synthesis of the anthocyanin delphinidin monoglucoside which starts masking the already present carotenoids [10]. Similarly, in the self-compatible species *T. pulchra* and *T. sellowiana* which are pollinated by bees, the flowers last for three days and the deposition of pollen on the stigma induces the flower colour change from white to pink [11].

In *Combretum indicum* four sequential stages of flower maturity based on the petal colour, namely white, pale pink, intermediate pink and deep pink, were identified [12]. The objective of the present work was to determine the pH of the petal extract, calculate the percentage of pink cells in the upper epidermis of petals using light microscopy, and determine the absorbance of the photosynthetic pigments chlorophyll a, chlorophyll b and carotenoids by spectrophotometry in the petals at the four identified stages.

2. MATERIALS AND METHODS:

The inflorescences of *Combretum indicum* were collected from the plants growing in Sri Aurobindo Park, Sadik Nagar, Delhi, before 09:00 a.m. during June-July 2024. The inflorescences were brought to the laboratory in a polythene bag. The cut ends of the inflorescences were kept dipped in a beaker containing water. The flowers were segregated into the four stages of maturity identified, namely white, pale pink, intermediate pink, and deep pink [12]. The petals from the freshly collected flowers were used in the experiments. All experiments were conducted thrice.

2.1 Determination of pH:

An extract of the petals of white flowers was prepared by homogenizing 2.0 g of petals in 30 mL of distilled water using a mortar and pestle. The homogenate was filtered through muslin cloth and the filtrate was used for determining the pH. The pH paper method was used [13]. A broad-range pH paper strip was carefully picked using a clean and dry pair of forceps, and a drop of the petal extract was applied on it using a glass rod. The colour that developed was matched immediately with the reference colour chart in the pH paper booklet and the pH was noted. Based upon the broad range of the pH determined, the suitable narrow range pH paper was selected. The narrow range pH was determined following the same procedure as for the broad range pH. Identical procedure was followed using the extracts of petals of flowers at the pale pink, intermediate pink and deep pink stages.

2.2 Preparation of peel mounts of the upper epidermis of petals:

A flower with white corolla was selected and using a pair of forceps, the upper epidermis of a petal was peeled from the throat (inner central portion) of the corolla. The peel was placed with the upper surface facing away from the microslide and then trimmed to a proper shape. A drop of glycerine was added to plasmolyse the epidermal cells in order to facilitate observation, and a coverslip was carefully lowered on the peel. The preparation was examined under a binocular compound microscope, and the number of pink cells (cells containing anthocyanin) and hyaline cells (cells lacking

anthocyanin) was recorded in three non-overlapping microscopic fields selected randomly. Representative photographs were clicked using the camera of a smartphone. The percentage of pink cells was calculated. Identical procedure was followed for studying the upper epidermis of the petals of flowers at the pale pink, intermediate pink and deep pink stages. A total of 185-576 epidermal cells were scored from the three microscopic fields in each replicate. The average percentage of pink cells obtained from the three replicates was used for discussing the results.

2.3 Extraction of pigments:

Extracts were prepared using the petals of freshly collected flowers at the white, pale pink, intermediate pink and deep pink stages by homogenizing 2.0 g of petals in 10 mL of acetone in a mortar using a pestle. The homogenate was filtered through muslin cloth, and the final volume of the filtrate was made up to 10 mL using acetone. The acetone extract, containing both anthocyanin and photosynthetic pigments, was transferred to a screw-cap vial and 10 mL of petroleum ether was added and the mixture was shaken. The colour of the two fractions formed was noted, and the upper petroleum ether fraction was drawn using a fine-nozzle dropper and transferred to a boiling tube. Methanolic KOH (30%, 10 mL) was added to the petroleum ether fraction and shaken for 10 minutes, allowed to stand, and then 10 mL of distilled water was added. This step resulted in two fractions; the lower fraction consisting of 30% methanolic KOH mixed with distilled water containing chlorophyll a and chlorophyll b, and the upper petroleum ether fraction containing carotenoids. Both fractions were separated and stored in labelled boiling tubes with the mouths of the tubes covered with aluminium foil. The absorbance of the two fractions was recorded using a PC-based double-beam spectrophotometer (Systronics 2206). The absorbance of the methanolic KOH + distilled water fraction was measured at 650 nm for chlorophyll a and at 490 nm for chlorophyll b using 15% methanolic KOH as the blank. For carotenoids, the absorbance of the petroleum ether fraction was measured at 490 nm using petroleum ether as the blank [14-16]. The average absorbance from the three replicates was used to interpret the results.

3. RESULTS:

3.1 The four sequential stages of floral maturity:

The corolla of the flowers of *C. indicum* begins to turn pink gradually from white. In the first two flowers in Figure 1 A, anthocyanin synthesis has begun in the throat region of the tubular flowers. The corolla of the third flower has turned deep pink completely. The lower surface view of the corolla of the flowers in Figure 1 A shows that the anthocyanin synthesis starts later in the lower surface, is not uniform, and is far lesser than the upper surface (Figure 1 B). The corolla changes from white to deep pink through the pale pink and intermediate pink stages; the intensity of pinkness increases as the flower matures from the pale pink to the deep pink stage.

Figure 1: The progression of anthocyanin synthesis in the petals of flowers of *Combretum indicum.* A. The upper surface view of the petals of three flowers. B. The lower surface view of the petals of the three flowers in A.

3.2 pH of the petal extracts:

The pH of the petal extract was 4.0 to 6.0 in the broad range and 5.0 in the narrow range at all the four stages of flower maturity, namely white, pale pink, intermediate pink and deep pink.

3.3 Peel mounts of the upper epidermis of petals:

The epidermal cells of the petals were polygonal and lacked intercellular spaces, and stomata were observed very rarely. Occasionally, trichomes were present in the peel mounts (Figure 2 B). As the petals transitioned from white to pale pink, intermediate pink, and finally deep pink, the number of pink cells gradually increased from zero to 93%, while the hyaline cells correspondingly decreased (Table 1, Figure 2 A, B). This showed that anthocyanin biosynthesis in the hyaline epidermal cells was progressive and as the gradual accumulation of anthocyanin occurred the pinkness of the corolla intensified.

Table 1: Anthocyanin-containing cells in the upper epidermis of the petals of *Combretum indicum* at the four stages of flower maturity

Replicate No.*	Percent anthocyanin-containing/ pink cells**			
	Petal colour			
	White	Pale pink	Intermediate pink	Deep pink
1	0	32.8	54.8	94.1
2	0	39.9	63.1	95.3
3	0	42.4	71.5	89.7
Average	0	38.4	63.1	93.0

* In each replicate the data on number of pink cells and number of hyaline cells were collected from three non-overlapping microscopic fields chosen at random.
**A total of 185-576 epidermal cells were scored from the three microscopic fields in each replicate.

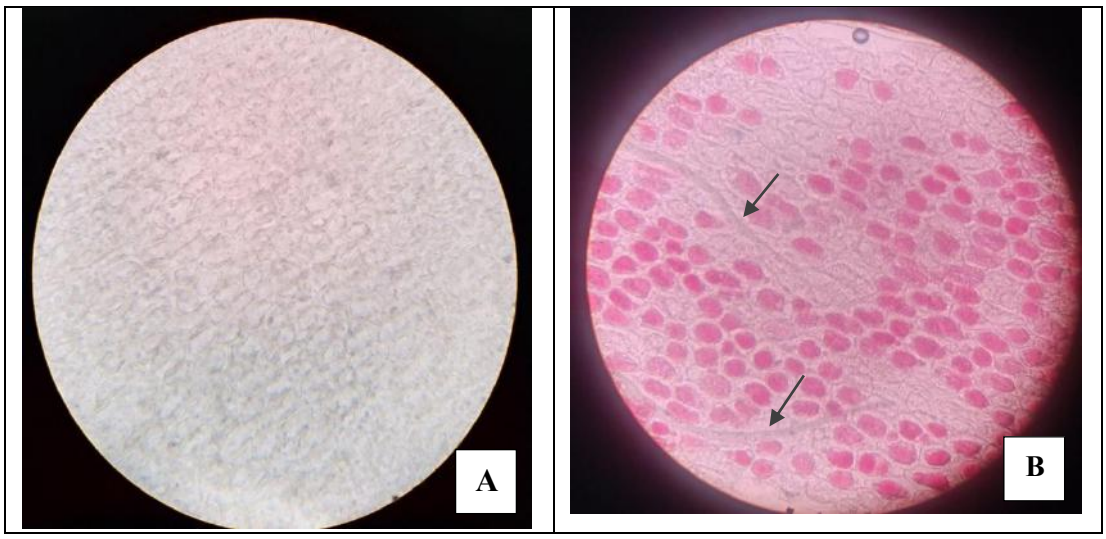


Figure 2: Peel mounts of the upper epidermis of the petals of *Combretum indicum* flowers mounted in glycerine. A. Flower at the white stage; B. Flower at the intermediate pink stage. Trichomes (arrows) are clear. A, B: 32x.

3.4 Pigments in the extracts:

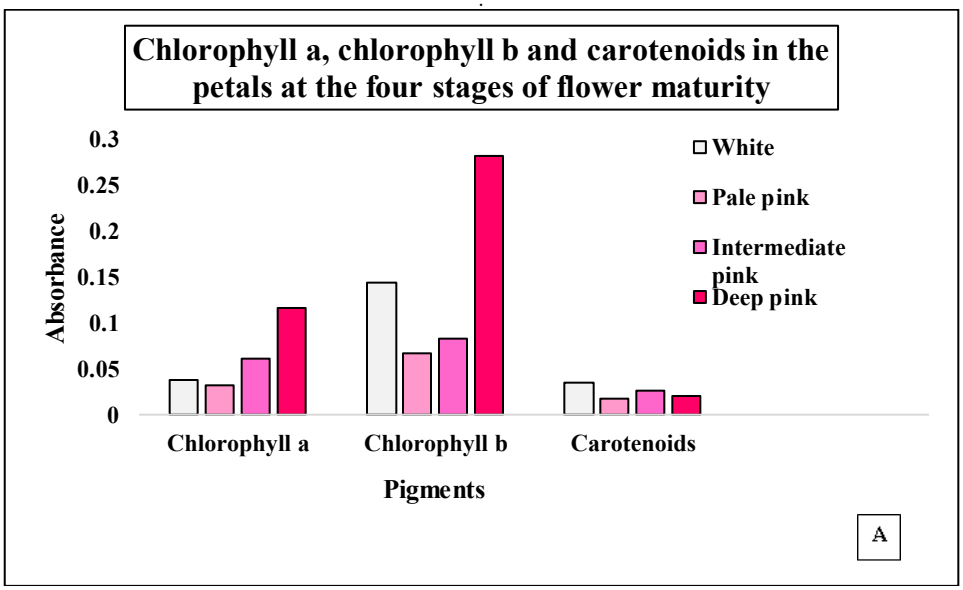
When petroleum ether was added to the acetone extracts of the petals, the lower acetone fractions were pale lemon yellow, peach coloured, pink, and brownish red at the white, pale pink, intermediate pink and deep pink stages, respectively; and the upper petroleum ether fraction was visually colourless and transparent at all the four stages. On adding methanolic KOH to the petroleum ether fraction, the lower methanolic KOH (+ distilled water) fraction showed variation in colour up to the intermediate pink stage, and was pale yellow-green, pale orange or colourless but turbid; and pale greenish yellow or orange at the deep pink stage. However, the upper petroleum ether fraction in this step was also visually colourless and transparent at all the four flower colour stages. The addition of methanolic KOH followed by distilled water allowed the separation of chlorophyll a and chlorophyll b from carotenoids. The carotenoids were relatively more hydrophobic than the chlorophylls and, therefore, remained in petroleum ether. Spectrophotometric analysis of the methanolic KOH + distilled water, and petroleum ether fractions showed that chlorophyll a, chlorophyll b and carotenoids were present at all the four stages of the corolla (Table 2). The absorbance of chlorophyll a and

chlorophyll b was the highest at the deep pink stage, and that of carotenoids was the highest at the white stage (Table 2, Figure 3 A, B).

Table 2: Absorbance of chlorophyll a, chlorophyll b, and carotenoids in the petals of *Combretum indicum* at the four stages of flower maturity

Pigment	Wavelength (nm)	Absorbance*			
		Petal colour			
		White	Pale pink	Intermediate pink	Deep pink
Chlorophyll a	650	0.038	0.032	0.061	0.116
Chlorophyll b	490	0.143	0.067	0.082	0.282
Carotenoids	490	0.035	0.018	0.026	0.020

*Average of three replicates.



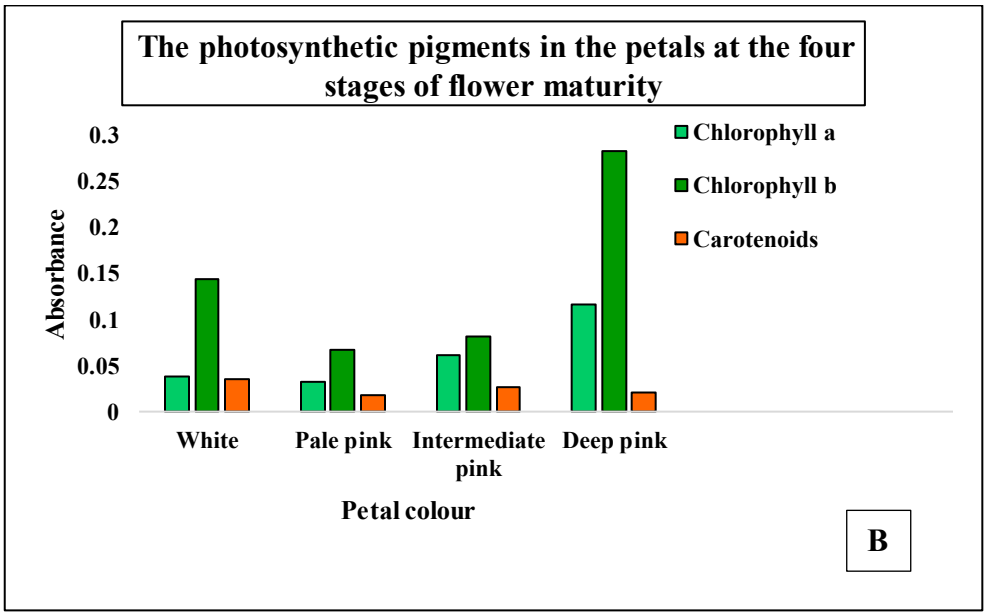


Figure 3: The photosynthetic pigments in the petals of *Combretum indicum* at the four stages of flower maturity. In A, the absorbance of chlorophyll a, chlorophyll b and carotenoids have been shown individually at the four stages of flower maturity; in B, the absorbance of chlorophyll a, chlorophyll b and carotenoids have been shown at each stage of flower maturity.

4. DISCUSSION:

Our result that the pH of the extract of the petals remains the same at the four stages of flower maturity is in agreement with the work of Yan *et al.* [17] who reported that the differences in petal pH were insignificant and could not explain the colour alterations in *Quisqualis indica* (syn. *C. indicum*). Pollination and ethylene did not induce petal colour change, and the rate of petal colour change was not affected at temperatures of 20-30ºC; however, no anthesis occurred at a constant temperature of 15ºC or 35ºC [17].

The gradual increase in the percentage of pink cells in the upper epidermis of the petals from zero at the white stage to 93% at the deep pink stage shows that the number of epidermal cells that have started to synthesize and accumulate anthocyanin increased with the age of the flower. Anthocyanin accumulates in the vacuoles of the epidermal and subepidermal tissues of plants [4, 18]. The white flowers are horizontal [1] and it is the upper epidermis which receives direct incident light. Light has been shown to be the major factor in inducing the sequential petal colour change from white to pink to red because the petals do not change colour in dark [17]. Light microscopic studies using petal sections have shown a differential accumulation of anthocyanin on the petal adaxial and abaxial surfaces: the adaxial surface which is mainly exposed to light accumulates more anthocyanin than the abaxial surface [4]. This proved that probably light was an important factor influencing the accumulation of anthocyanin in the vacuoles of cells in the peripheral epidermal and subepidermal layers of the petals [4]. The pink and red flowers are pendulous [1]; and in such an orientation the upper surface of the petals will not receive direct incident light. Cyanidin 3-O-glucoside was the major anthocyanin, and the anthocyanin composition was the same during the different stages of ontogeny in coloured flowers although the individual anthocyanin concentrations changed [4].

In apple, grape, lychee, pear and tomato fruits induction of anthocyanin accumulation is light-dependent whereas in the tubers of some varieties of potato, turnip and sweet potato the regulation of anthocyanin biosynthesis is light-independent [19]. Bagged apple (*Malus domestica*) fruits in dark continued to change colour and the pericarp attained the normal colour in darkness following light induction, contrary to previous studies which had shown that anthocyanins do not accumulate in apple peel in darkness [20]. The amount of cyanidin 3-O-galactoside in the apple fruit peel was 84.71% of the total anthocyanin after seven days of dark treatment [20].

The progressive deepening of colour of the acetone fraction of petals from pale lemon yellow in white flowers to brownish red in deep pink flowers of *C. indicum* correlates with the initiation and progression of biosynthesis and accumulation of anthocyanin in the petals. The visually colourless and transparent petroleum ether fraction at all the four stages was indicative of the fact that photosynthetic pigments, although present, were very less in concentration. However, when methanolic KOH was added followed by distilled water, the chlorophylls got saponified and separated from the carotenoids. The result suggests that all the photosynthetic pigments play a role in absorbing light. The higher absorbance shown by chlorophyll b as compared to chlorophyll a at each of the four stages shows that chlorophyll b provides photoprotection to the petals. Carotenoids, although present at low concentrations at all the four stages, along with chlorophyll b absorb the short wavelength radiations. Our work is in agreement with the report of Afify & Hassan [21] wherein the flowers of *Quisqualis indica* (syn. *C. indicum*) were shown to contain chlorophyll a, chlorophyll b, xanthophylls, carotenoids, anthocyanin, phenolics and flavonoids. It is known that anthocyanins are antioxidants, and all parts of *Quisqualis indica* (syn. *C. indicum*) have been shown to have good antioxidant activity; the flowers are rich in phenolics and the flavonoid quercetin, which supports the plant's use in traditional medicine [22]. In the drought-tolerant *Bougainvillea glabra* and *Delonix regia*, the chlorophyll a, chlorophyll b and carotenoid contents were more in senescing flowers compared to fresh flowers [16]. The presence of the antioxidant vacuolar pigment betacyanin only in senescing flowers of *B. glabra*, and the presence of more anthocyanins in senescing flowers of *D. regia* than in the fresh flowers [16] corroborates the current work and the importance of antioxidants to the flowers.

The function of the antioxidant enzymes superoxide dismutase, catalase, ascorbate peroxidase, glutathione reductase, glutathione peroxidase, and dehydroascorbate reductase is to decrease oxidative stress and improve anthocyanin accumulation [18]. We have reported the presence of catalase and oxidase activity at all the four stages of flower maturity in *C. indicum*; however, catalase activity was maximum at the white stage and oxidase activity was maximum at the intermediate pink stage [12]. It is likely that the enzymatic antioxidant catalase, oxidase, and the accumulated nonenzymatic antioxidant anthocyanin protect the petals from oxidative stress [12]. Our earlier study showed that catalase activity was present in fresh flowers of *Bougainvillea glabra* but in senescing flowers the activity decreased; and catalase activity was absent in fresh flowers of *Delonix regia* but low activity was present in senescing flowers [16]. During senescence reactive oxygen species (ROS) levels increase, and concomitantly the ROS scavenging antioxidant enzymes such as superoxide dismutase, catalase and ascorbate peroxidase increase; however, at the last phase of senescence the activity of ROS scavenging enzymes decreases [23]. Both antioxidant pigments and antioxidant enzymes help in detoxifying the ROS which form as an organ senesces.

C. indicum flowers have been used for dyeing fabric. An aqueous extract of flowers has been used traditionally to dye silk textiles using aluminium, ferrous sulphate and potassium dichromate [24]. Ariffin *et al.* [25] showed that natural colourants extracted from the petals could be used to dye silk fabric to different shades of brown with colour fastness and a uniform colour could be obtained with the technique of infra-red dyeing. Using a microwave, the dye extraction time in water from flowers dried in shade has been minimized to 120 seconds [26]. Considering that *C. indicum* is a vigorous, hardy, quick-growing climber which can easily be propagated vegetatively [27], it is recommended that the potential of this under-utilized versatile plant could be tapped on a commercial scale for use as colourants in the textile industry, and in medicine after clinical studies.

4. CONCLUSION:

The epidermal cells in the upper surface of petals which lacked anthocyanin at the white flower stage gradually accumulated anthocyanin, and 93% of the cells contained anthocyanin at the deep pink stage. The pH of the petal extract remained 5.0 at all the four stages of flower maturity. Chlorophyll a, chlorophyll b and carotenoids were present at all stages. Chlorophyll a and chlorophyll b showed the highest absorbance at the deep pink stage whereas carotenoids showed the highest absorbance at the white stage. The absorbance of chlorophyll b was higher than chlorophyll a at all the four stages. The combination of the antioxidant anthocyanin, and the photosynthetic pigments chlorophyll b and carotenoids, along with the antioxidant enzyme catalase, and oxidase prevent the petals from oxidative damage helping in the retention of the flower until pollination.

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REFERENCES:

1. Eisikowitch, D., & Rotem, R. (1987). Flower orientation and colour change in *Quisqualis indica* and their possible role in pollinator partitioning. *Botanical Gazette, 148*(2), 175-179. https://doi.org/10.1086/337645
2. Willmer, P., Stanley, D. A., Steijven, K., Matthews, I. M., & Nuttman, C. V. (2009). Bidirectional flower colour and shape changes allow a second opportunity for pollination. *Current Biology, 19*(11), 919-923. https://doi.org/10.1016/j.cub.2009.03.070
3. Yan, J., Wang, G., Sui, Y., Wang, M., & Zhang, L. (2016). Pollinator responses to floral colour change, nectar, and scent promote reproductive fitness in *Quisqualis indica* (Combretaceae). *Scientific Reports, 6*, Article 24408. https://doi.org/10.1038/srep24408
4. Ghissing, U., Goswami, A., & Mitra, A. (2023). Temporal accumulation of pigments during colour transformation from white to red in *Combretum indicum* (L.) DeFilipps (syn. *Quisqualis indica* L.) flowers. *Natural Product Research, 37*(3), 529–533. https://doi.org/10.1080/14786419.2021.1984467
5. Weiss, M. R., & Lamont, B. B. (1997). Floral color change and insect pollination: A dynamic relationship. *Israel Journal of Plant Sciences, 45*(2-3), 185-199. https://doi.org/10.1080/07929978.1997.10676683
6. Weiss, M. R. (1995). Floral color change: A widespread functional convergence. *American Journal of Botany, 82*(2), 167-185. https://doi.org/10.1002/j.1537-2197.1995.tb11486.x
7. Nadot, S., & Carrive, L. (2020). The colourful life of flowers. *Botany Letters, 168*(11), 120-130. https://doi.org/10.1080/23818107.2020.1839789
8. Delph, L. F., & Lively, C. M. (1989). The evolution of floral colour change: Pollinator attraction versus physiological constraints in *Fuchsia excorticata*. *Evolution, 43*(6), 1252-1262. https://doi.org/10.1111/j.1558-5646.1989.tb02572.x
9. Ida, T. Y., & Kudo, G. (2003). Floral colour changes in *Weigela middendorffiana* (Caprifoliaceae): Reduction of geitonogamous pollination by bumble bees. *American Journal of Botany, 90*(12), 1751-1781-1757. https://doi.org/10.3732/ajb.90.12.1751
10. Mohan Ram, H. Y., & Mathur, G. (1984). Flower colour changes in *Lantana camara*. *Journal of Experimental Botany, 35*(11), 1656-1662. https://doi.org/10.1093/jxb/35.11.1656
11. Pereira, A. C., da Silva, J. B., Goldenberg, R., Melo, G. A. R., & Varassin, I. G. (2011). Flower colour change accelerated by bee pollination in *Tibouchina* (Melastomataceae). *Flora, 206*, 491-497. https://doi.org/10.1016/j.flora.2011.01.004

12. Subramanyan, J., Beri, V., Sajid, B., Khanna, C., Samad, D., Nishika, & Mangar, R. (2025). A comparative study on certain enzymes in flowers of Rangoon creeper (*Combretum indicum*) at different stages of maturity. *International Journal of Innovative Research in Multidisciplinary Field, 11*(7), 130-136. https://doi.org/10.2015/IJIRMF/202507020
13. Kapur, P., & Govil, S. R. (2000). Environmental factors. In *Experimental plant ecology* (p. 14). CBS Publishers & Distributors.
14. Machlis, L., & Torrey, J. G. (1956). Photosynthesis. In *Plants in action: A laboratory manual of plant physiology* (p. 136). W. H. Freeman.
15. Bajracharya, D. (1999). Estimation of plant constituents. In *Experiments in plant physiology* (p. 51). Narosa Publishing House.
16. Subramanyan, J., Rajeshwari, Patel, D., & Sharma, P. (2023). Comparative study of oxidative stress biomarkers in flowers and fruits during senescence. *International Journal of Botany Studies, 8*(7), 11–17.
17. Yan, J., Wang, M., & Zhang, L. (2018). Light induces petal color change in *Quisqualis indica* (Combretaceae). *Plant Diversity, 40*(1), 28-34.
18. Sharma, H., Sharma, P., Kumar, A., Chawla, N., & Dhatt, A. S. (2024). Multifaceted regulation of anthocyanin biosynthesis in plants: A comprehensive review. *Journal of Plant Growth Regulation, 43*, 3048-3062. https://doi.org/10.1007/s00344-024-11306-x
19. Ma, Y., Ma, X., Gao, X., Wu, W., & Zhou, B. (2021). Light induced regulation pathway of anthocyanin biosynthesis in plants. *International Journal of Molecular Sciences, 22*(20), Article 11116. https://doi.org/10.3390/ijms222011116
20. Xue, X., Tian, S., Chen, R., Han, X., Wang, J., & Zhao, X. (2022). Clarifying the mechanisms of the light-induced color formation of apple peel under dark conditions through metabolomics and transcriptomic analyses. *Frontiers in Plant Science, 13*, Article 946115. https://doi.org/10.3389/fpls.2022.946115
21. Afify, A. E. M. R., & Hassan, H. M. M. (2016). Free radical scavenging activity of three different flowers—*Hibiscus rosa-sinensis*, *Quisqualis indica* and *Senna surattensis*. *Asian Pacific Journal of Tropical Biomedicine, 6*(9), 771-777.
22. Shah, A., Khan, Z., Saleem, S., & Farid, S. (2019). Antioxidant activity of an ethnobotanically important plant *Quisqualis indica* Linn. *Pakistan Journal of Pharmaceutical Sciences, 32*(1), 95-102.
23. Cavaiuolo, M., Cocetta, G., & Ferrante, A. (2013). The antioxidants changes in ornamental flowers during development and senescence. *Antioxidants, 2*(3), 132-155. https://doi.org/10.3390/antiox2030132
24. Hamdy, D. M., Othman, H. A., & Hassabo, A. G. (2021). Various natural dyes using plant palette in coloration of natural fabrics. *Journal of Textile Coloration and Polymer Science, 18*(1), 121-151.
25. Ariffin, S., Ruznan, W. S., Yazid, M. N., Suhaimi, S. A., Mazian, M. A., & Mustamam, M. M. (2021). Colour strength and washfastness of silk fabric dyed with methanolic extracted Rangoon creeper using infrared dyeing machine. *Malaysian Journal of Analytical Sciences, 25*(4), 642-648.
26. Soniya, M., & Anitha, D. (2018). Microwave extraction of dye from madhumalti flowers (*Combretum indicum*). *Journal of Research ANGRAU, 46*(3), 61-68.
27. Bose, T. K., Chowdhury, B., & Sharma, S. P. (1991). Climbers. In *Tropical garden plants in colour* (p. 205). Horticulture and Allied Publishers.