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Gene Expression Profile of some Antioxidant-Related Genes in Five Species of the Family *Asteraceae* in Egypt

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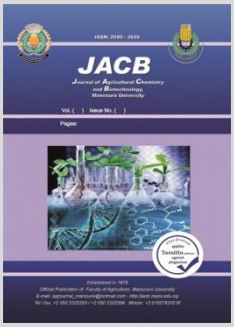

ABSTRACT

The *Asteraceae* family is one of the largest flowering plant families and is known for its antioxidant, anti-inflammatory, and antimicrobial activity. Their pharmacological effects can be related to their range of phytochemical compounds, including polyphenols, phenolic acids, flavonoids, acetylenes, and triterpenes. In this study, Sunflower *(Helianthus annuus*), Gazania (*Gazania rigens*), Gaillardia (*Gaillardia pulchella*), Zinnia (*Zinnia elegans*), and Chrysanthemum (*Chrysanthemum morifollium*) were evaluated for their antioxidant activity using the DPPH free radical scavenging assay. Total phenol content (TPC) and the gene expression profiles of some antioxidant-related genes, including ascorbate peroxidase 3 *APX3*, catalase *CATA1*, and Phenylalanine ammonia lyase *PAL*, were also analyzed. Results revealed that Sunflower and Chrysanthemums plants had the highest phenolic contents of about (3.26±0.39 and 2.99±0.22 mg GAE/g), respectively. The expression of *PAL* gene was about 4-fold and 2-fold higher in Chrysanthemums and Zinnia s' flowers, respectively, in comparison to the *sunflower*s. *APX3* gene expression was upregulated in *Chrysanthemum*, *Gazania*, and *Gaillardia* 's leaves in comparison to the Sunflowers'. Our results give an insight into the antioxidant potential of some uncommonly used plants of the family *Asteraceae.*

Keywords: *Asteraceae,* Antioxidant activity, Gene expression.

INTRODUCTION

The *Asteraceae* (Compositae) family is one of the largest flowering plant families, with over 1100 genera and 25000 species. (Zareh, 2005). Species from this family are frequently highlighted because of their anti-inflammatory, analgesic, antioxidant, and antipyretic properties (Odom *et al*., 2006). Many *Asteraceae* species have been shown to have pharmacological properties and to contain essential phytochemicals such as polyphenols, flavonoids, and diterpenoids. (Koc *et al*., 2015).

Egypt's lands, particularly the desert, are wealthy in various medicinal plants (Boulos, 1995). Phytochemicals found inside these plants, for example, flavonoids, alkaloids, terpenoids, and phenolics, have many medical benefits and get into a lot of food industries (Ramawat, 2008).

Sunflower *(Helianthus annuus)* is a type of oilseed crop native to North America. It is grown all throughout the world, and the majority of its products are used to make animal feed. (Yegorov *et al*., 2019). The production of Sunflower seeds is crucial to producing edible oils, as the seeds have around 40–45% oil. The oil extracted from it is characterized by the quality of its chemical and natural properties. However, the use of Sunflower meals in the human diet is restricted due to the presence of anti-nutrients (saponins, protease inhibitors, and arginase inhibitors), insoluble, and a minuscule amount of solvent residue in the meal after extraction, the use of sunflower meals in the human diet is restricted. (Grasso *et al*., 2019).

Gazania has been used in traditional medicine to treat toothaches and miscarriages, and it was frequently combined with aloe in purgative medicines. Only a few research have evaluated the biological benefits of gazania, including its anti-inflammatory, antioxidant, and hepatoprotective properties for *G. nivea* and *G. rigens*, respectively. (Hammoda, 2009). The various secondary metabolites found in the *Asteraceae* family and *Gaillardia* species are characterized by sesquiterpene lactones, which can be used as chemotaxonomic markers. Gaillardia sp. extracts have antiparasitic, antitumoral, and cytotoxic properties. (Raal *et al*., 2011; Tong Yao *et al*., 2013). Several species of the Zinnia genus are being investigated for their possible biological effects, such as their insecticidal, antifungal, antioxidant, hepatoprotective, antibacterial, antiviral, and antimalarial properties. (Gomaa *et al*., 2018). The State Ministry of Health of China has officially acknowledged *Chrysanthemum morifolium* flowers as traditional medicine and nutritious food that can be used to make tea or food. (Yuan *et al*., 2020). Many substances are recognized as biologically active components, including terpenoids (mostly represented by essential oils), hydroxycinnamic acid derivatives (primarily described by chlorogenic acid), and flavonoids. (Liu *et al*., 2010).

Antioxidants from our diet are essential for endogenous antioxidants in protection against oxidative stress. A nutrient-antioxidant deficit is one of the reasons for various chronic and degenerative illnesses. Each nutrient has a unique purpose in terms of its composition and antioxidant capacity (Donaldson, 2004; Willcox *et al.*, 2004). Plants are

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regarded as the primary source of antioxidant compounds, which are mostly phenolic. *Asteraceae* is the largest family of blooming plants and the second-largest plant family overall in the kingdom of plants. Hence, research into *Asteraceae* plants is necessary, especially for those that are regarded as weeds. In fact, weeds have seen the most intensive control in agricultural fields, yet their potential for bioprospecting may not have been fully investigated. (Kasote *et al*., 2015; Zhang and Tsao, 2016).

Gene expression is one of the most important process in studying the response in plants by which the information encoded in a gene is turned into a function. Caverzan et al., (2016a) and Scandalios, (1994) found that enzymatic antioxidants include a large and versatile set of enzymes superoxide dismutase [SOD], ascorbate peroxidase [APX], and catalase [CAT] which are present in all subcellular compartments of the plant cell also, PAL, C4H, 4CL, and CHS are important enzymes in this pathway and facilitating the biosynthesis of flavonoids. An improved understanding of these enzymes is vital for identifying targets for biotechnological manipulation of product accumulation (Voloudakis *et al*., 2006). Likewise, ascorbate Peroxidase (APX), peroxidase (POD), and catalase (CAT) are found in Chrysanthemum (Chrysanthemum morifolium Ramat.) and had a big role in antioxidant activity (Chakrabarty *et al*., 2007a)

Despite the spread of these plants in Egypt and their high levels of medicinal substances, antioxidants, and nutrients, comprehensive research is needed to confirm their effectiveness and explore potential applications. Thus, the primary objective of this work is to identify novel and natural antioxidants in five species of family *Asteraceae* in Egypt, Sunflower, Gazania, Gaillardia, Zinnia, and Chrysanthemum. The antioxidant activity in all selected plants was verified by analyzing the amount of total phenolic content (TPC) using gallic acid as a standard and measuring the scavenging antioxidant activity using the DPPH assay. Quantitative real-time PCR was also carried out to assess the relative gene expression of certain genes associated to antioxidants: Ascorbate peroxidase 3 *APX3*, catalase *CATA1*, and phenylalanine ammonia lyase *PAL*.

MATERIALS AND METHODS

Plant material

The fresh flowers of Sunflower (*Heilanthus annuus*), Gazania (*Gazania rigens*), Gaillardia (*Gaillardia Pulchella*), Zinnia (*Zinnia elegans*) and Chrysanthemum (*Chrysanthemum morifollium*) (figure 1) were collected from a private garden and directly frozen at -20 °C until usage.

According to the system of A.Engler, each of Helianthus, Gazania, Gaillardia, Zinnia and Chrysanthemum are in the same subfamily Asteroideae. Both of Helianthus and Zinnia are in tribe Heliantheae. The three other genera are in different three tribes (Table 1).

Figure 1. The five studied species from *Asteraceae* family (a) Sunflower, (b) Gazania, (c) Gaillardia, (d) Zinnia, (e) Chrysanthemum.

Table 1. Pedigree of Studied species.

Family: Compositae (Asteraceae)					
Subfamily:	Asteroideae	Asteroideae	Asteroideae	Asteroideae	Asteroideae
Tribe:	Heliantheae	Arctotideae	Heliantheae	Heliantheae	Anthemideae
Subtribe:	Helianthinae	Gorteriinae	Melampodiinae	Helianthinae	———
Genus:	Helianthus	Gazania	Gaillardia	Zinnia	Chrysanthemum

(Note: Chrysanthemum doesn't have a designated subtribe)

Samples preparation

Air parts for the five species were dried at 50°C overnight, then ground to fine powder (Akar *et al*., 2017). The plant samples were ground in a laboratory homogenizer with 2.5 mm particle size and prepared for further analysis, the homogenized samples put separately in distilled water using a sonicator (2.2L digital ultrasonic cleaner bath, Shenzhen Derui Ultrasonic Equiment Ltd, Shenzhen – China) with

40kHz for 30min and then extracted using a magnetic stirrer equipped (Guangzhou Ikeme Technology Co., Ltd, Shanghai, China) with a heater set at 95 ºC for 4 hours. The extracts obtained were then filtered with 4 filter papers, according to (Khalaf Ashok *et al.*, 2008) with minor changes. To prepare the standard solutions, one gram of Gallic acid was dissolved in 100 ml of methanol to get a 1% solution of Gallic acid (10 mg/ml), which was termed a "standard 1 solution." Similarly, 1 g of Quercetin was dissolved in 100 ml of methanol separately to get a 1% solution of Quercetin (10 mg/ml), which was termed "standard 2 solutions." (Ullah Shirazi *et al*., 2014).

Determination of Total Polyphenol Content (TPC)

The amount of TPC in the studied plants' extracts was determined with the Folin-Ciocalteu's reagent (FCR) (Sigma-Aldrich, Merck Ltd., Cairo, Egypt) according to the method previously published by Slinkard and Singleton (1977) use gallic acid as the benchmark. The FCR is a mixture of phosphomolybdate and phosphotungstate, also known as Folin's phenol reagent, Folin-Denis reagent, and gallic acid equivalence method (GAE). (Sigma-Aldrich, Merck Ltd., Cairo, Egypt) used for the colorimetric in vitro assay of phenolic and polyphenolic antioxidants (Bärlocher *et al*., 2006).

The extracted solution was transferred into a test tube, and the final volume was adjusted to 4 ml by the addition of distilled water. Afterwards, 0.25 ml of Folin-Ciocalteu Reactive (FCR) (Fluka) was added to this mixture, and after 3 minutes, 0.75 ml of Na_2CO_3 (20%) was added. Subsequently, the mixture was shaken on a shaker for 2 hours at room temperature, and then absorbance was measured at 760 nm. Gallic acid (Sigma-Aldrich; Merck Ltd., Cairo, Egypt) was used as a standard phenolic compound. Then, the phenolic compound content was determined as Gallic acid equivalent using the calibration curve.

Scavenging antioxidant activity (DPPH) assay

The free radical scavenging activities of the extracts were measured using the method of (Lu and Yeap Foo, 2000) with some modifications. 70% methanol was prepared, 1 gm of each sample was measured (plus 10 ml of 70% methanol), 1 millimoles of DPPH (2, 2-diphenyl-1-picrylhydrazyl) was prepared, and 4 ml of the DPPH solution plus 0.1 ml of the extracted plant sample was measured, along with a blank sample (plus 3.9 ml of the DPPH solution plus 0.1 ml of distilled water). All measurements were done, and the absorbance was read at 517 nm on a UV/Vis spectrophotometer (ChromTech CT-2200), Chrom Tech, Inc., MN, USA.

RNA extraction and c-DNA synthesis

Total RNA was isolated from plant leaves and flowers and promptly frozen and preserved for use in gene expression analyses at -80 "C. The total RNA was extracted using total RNA Mint Extraction Kit (applied biotechnology company) following the manufacturer's instructions. c-DNA synthesis was prepared according to (Aseel *et al.*, 2019). 3µl of total RNA (500ng) sample were mixed with 0.5µl oligo dT, 2µl dnTPs, 0.5µl Reverse transcriptase, 2 µl buffer of reverse transcriptase and 12µl distilled water, the mixture was incubated first at 37 °C for 2 hours, then at 65 °C for 20 min and finally cooling at 4 °C for 10 min.

Relative gene expression analysis

Quantitative real-time PCR qRT-PCR was performed using gene-specific primers for *APX3, CATA1*, and *PAL*, and the *Actin-2* as the housekeeping gene (Table 2). Following reaction mixture 10 µl of SYBR Green was mixed with 1 µl of forward primer, 1 µl of reverse primer, 2 µl of c-DNA and 6 µl of distilled water. The qRT-PCR program was optimized for all primers as follows; 95 °C for 5 min then start 45 cycles of 95°C for 10 s, 60°C for 20 s, and 72°C for 20s. After real-time PCR, melting curves were carried out to show the amplification of each target individual gene product.

Table 2. Primers used in this study.

Primers	Sequence (5′–3′)	accession number	Tm	Reference
Actin F	5'-GCTAACAGGGAAAAGATGACTC-3'	AF28264.1	56°C to 60°C	(Tatiana Ș *et al*., 2015)
Actin R	5'-ACTGGCATAAAGAGAAAGCACG-3'	AF28264.1	56°C to 60°C	(Tatiana Ș *et al*., 2015)
APX3* F	5'-CCCAAATGCTACCAAAGGTG-3'	BU032190.1	57°C to 60°C	(Tatiana Ș *et al*., 2015)
APX3* R	5'-ATGTGCTCTTCCAAGGGTGT-3'	BU032190.1	57°C to 60°C	(Tatiana Ș *et al*., 2015)
CATA1 F	5′-CTTCCCGCTTGAATGTGAAG-3′	L28740	56°C to 60°C	(Azpilicueta *et al*., 2008)
CATA1 R	5′-CCGATTACATAAACCCATCATCG-3′	L28740	56°C to 60°C	(Azpilicueta *et al*., 2008)
PAL F	5'-CGGATTCTTCGAGTTAAG-3'	Y12461	57°C to 60°C	(Göpfert *et al*., 2006)
PAL R	5'-CTTACGGTTGACTTCATGTTC-3'	Y12461	57°C to 60°C	(Göpfert *et al*., 2006)

Data analysis

qRT-PCR was used to determine the mRNA expression levels for the five samples using Thermo Scientific's Maxima SYBR Green/ROX PCR Master Mix in accordance with the manufacturer's instructions. Three replicates of each sample's analysis were carried out in three separate runs. According to Livak and Schmittgen (2001), the $\Delta\Delta C_T$ value was calculated to normalize the target gene expression $\Delta\Delta C_T = \Delta C_{T(test)} - \Delta C_T$(calibrator). ΔC_T(calibrator) displays the difference between the C_T of the target gene and the C_T of reference gene for sunflower as a control specie, while the ΔC_T (test) = C_T target gene - C_T reference gene for each other studied species. The following formula was used to estimate fold changes (FC) in gene expression between the experiment and the control: $FC=2^{-\Delta\Delta CT}$. Differences between means were tested for significance at 0.05 using an unpaired, two-tailed *student test*

For TPC and antioxidant activity, the data were examined using One-way ANOVA with the IBM SPSS program 25, Armonk, New York, the United States. Duncan's test was used to examine the mean differences at (a) confidence level of 99.5% (p 0.05). For the three copies, the acquired data were reported as mean standard deviation.

RESULTS AND DISCUSSION

Results

Determination of Total Phenolic Content (TPC)

Plant samples were tested for the determination of total phenols. The mean values were determined after signing

the standard curve of absorbance for standard Gallic acid. The total phenolic content varied significantly in the five studied plants; the total phenolic content of the studied plants' leaves ranged from 1.18 to 2.98 mg GAE/g respectively.

Sunflower and Chrysanthemum had the highest phenolic content, 2.91±0.66 and 2.99±0.15, respectively (Figure 2); among the samples, the lowest total phenolic content was observed in Gazania and Gaillardia, 1.18±0.07 and 1.59±0.35, respectively. Same result as (Francesco Gai *et al.*, 2020).

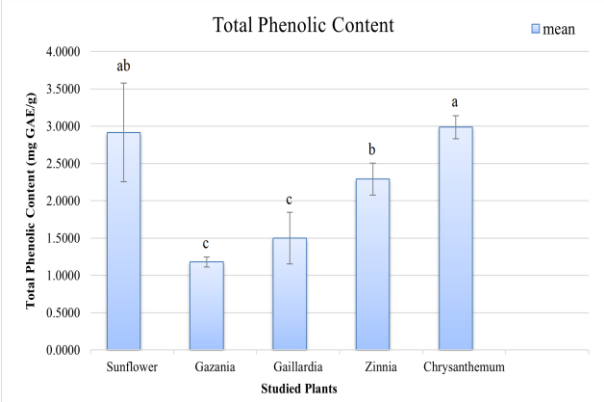

Figure 2. Measurement of total phenolic content (TPC) and each bar represent the mean ± and SD of three replica, Duncan's letters refer to a = the highest mean, b = lower and c = the lowest, when there the same letter to different samples that's mean there is no significant differences between them.

The antioxidant activity by DPPH

The extracts of the five examined plant species Sunflower, Gazania, Gaillardia, Zinnia, and Chrysanthemum showed no statistically significant differences in antioxidant activity among the five extracts when in vitro testing for antioxidant activity using the DPPH method. (Figure 3).

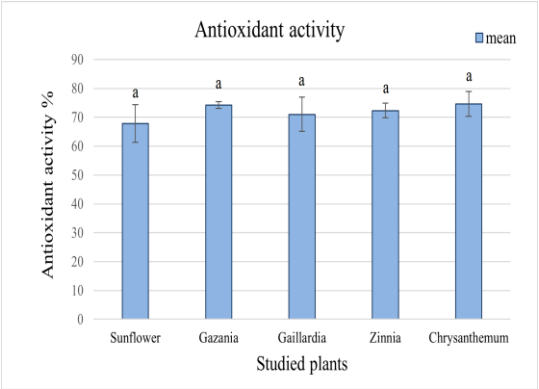

Figure 3. Measurement of antioxidant activity and each bar represents the mean ± and SD of three replicates. Duncan's letters refer to a = the highest mean, when there the same letter to different samples that's mean there is no significant differences between them.

qRT-PCR for the antioxidant-related genes

In order to evaluate the antioxidant potential of various species of the family *Asteraceae*, gene expression analysis was conducted in this study. RNA was extracted from the leaves and flowers of the studied plants, transcribed to c-DNA, and then amplified using antioxidant gene-specific primers. Because sunflower has a high antioxidant potential, this study compared antioxidant activity in Zinnia, Gaillardia, Gazania, and Chrysanthemum plants to Sunflower. In the leaves (figure 4),

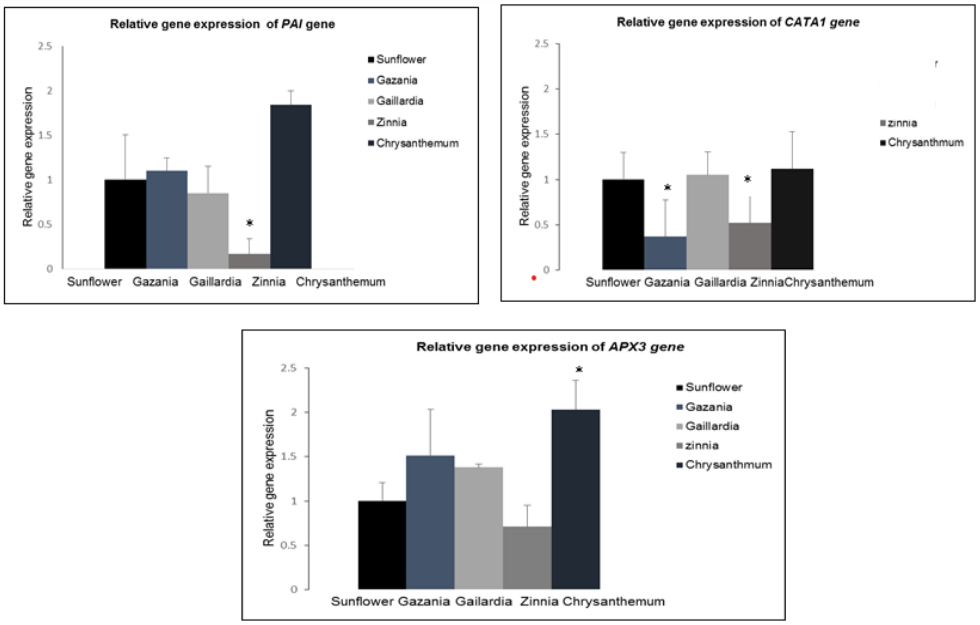

Figure 4. Quantitative real-time PCR of *PAL, CATA1and APX3* genes in the leaves of 5 different species of *Asteraceae* family. For each sample, RNA was extracted, transcribed to C-DNA and normalized to the house keeping gene B-Actin 2. Error bar represents + SD. Differences between means were tested for significance at 0.05 using an unpaired, two-tailed *student test.*

The PAL gene catalyses the first step of the phenylpropanoid pathway, which is one of the best understood secondary metabolism pathways in higher plants, leading to the synthesis of various physiologically important

metabolites, including flavonoids, lignin, coumarins, stilbenes, etc. (Dixon and Paiva, 1995; Liu et al., 2006). The expression rate of the PAL gene in leaves was significantly down-regulated in Zinnia by 0.17 fold. On the other hand, the same gene was slightly but not significantly up-regulated in Chrysanthemum and Gazania by 1.84 and 1.1 fold but in Gaillardia the PAL gene wasn't significantly down-regulated compared to Sunflower by 0.85 fold. In addition, the CATA1 gene is the gene responsible for the synthesis of the enzyme catalase and is considered a home-containing protein concerning cell protection from the toxic effects of ROS (Balogun et al., 2016). CATA1 was also analyzed in the studied plants, and its expression varied among different species. In Gazania and Zinnia, the gene expression of CATA1 was significantly down-regulated by 0.37 and 0.52 fold, respectively, while no significant difference was observed in the other species compared to Sunflower. Moreover, the APX3 gene, which plays an important role in the anti-oxidation metabolism in plant cells (Narendra et al., 2006), exhibits a significant up-regulation in Chrysanthemum by 2.03 fold, but no significant difference was observed in Gazania, Gaillardia, or Zinnia.

In the flowery parts (figure 5), *PAL* was significantly up-regulated in Chrysanthemum and Zinnia by 4.65 and 2.26 folds, respectively, but exhibits an insignificant up-regulation in Gaillardia. On the contrary, the *PAL* gene was significantly downregulated in Gazania by 0.41 fold. Results also revealed differences in gene expression of the *CATA1* gene among the studied species. *The CATA1* gene showed significant down-regulation in Gaillardia and Chrysanthemum by 0.4 fold for each. While in Gazania and Zinnia, it was downregulated by 0.95 and 0.59 folds, respectively, compared to Sunflower. The *APX3* gene expression was also analysed in the flowers of the sample plants. The *APX3 gene* was significantly up-regulated in Gazania by 1.8 fold, as well as significantly down-regulated in Gaillardia by 0.13 fold. In Chrysanthemum, the gene expression was slightly but not significantly up-regulated compared to Sunflower.

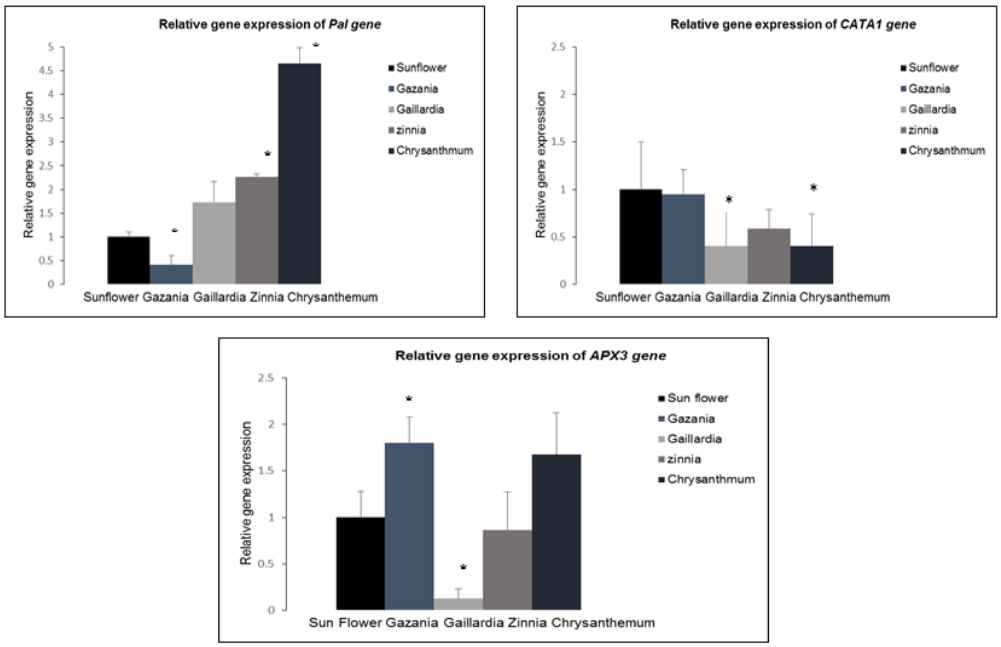

Figure 5. Quantitative real-time PCR of *PAL, CATA1and APX3* genes in the flowers of 5 different species of *Asteraceae* family. For each sample, RNA was extracted, transcribed to C-DNA and normalized to the house keeping gene *B-Actin 2*. Error bar represents + SD. Differences between means were tested for significance at 0.05 using an unpaired, two-tailed *student test.*

Discussion

Catalase is an essential antioxidant enzyme, helps to break down hydrogen peroxide and preserve cellular redox equilibrium. Catalase has been linked to the development of several prevalent diseases, including diabetes, Alzheimer's disease, Parkinson's disease, and others, according to numerous investigations from diverse laboratories. (Nandi *et al.*, 2019). In this context, the catalase's expression rate varied sharply between the leaves and the flowery parts.

In Gazania, the floral parts had higher *CATA1* expression. In the leaves of the same plant *CATA1* expression was the lowest compared to Sunflower. *CATA1* levels expression in the leaves of Gaillardia and Chrysanthemum were higher when compared to Sunflower. On the other hand *CATA1* expression in Gaillardia and Chrysanthemum floral parts were lower compared to Sunflower. Balogun and Ashafa (2016) reported that Gazania is a high source of Catalase. Chakrabarty *et al*., (2007) discovered that Catalase expression is up regulated in Chrysanthemum and found equivalent results.

PAL has a crucial role in secondary phenylpropanoid metabolism and is one of the most extensively studied genes with respect to plant responses to biotic and abiotic stress (Kim and Hwang, 2014). Hence, *PAL* expression rates showed some similarities and some differences between the leaves and the flowery parts. Interestingly, *PAL* expression was elevated in Chrysanthemum floral parts. In the same plant leaves *PAL* expression showed the highest level compared to Sunflower. In addition (Yang *et al*., 2017) reported that Chrysanthemum cultivars are promising sources of natural antioxidants. The expression of *PAL* in Gazania leaves was higher compared to Sunflower. In the same plant floral parts,

the expression of *PAL* gene was the lowest in comparing to Sunflower. In Zinnia and Gaillardia, *PAL* expression of the floral parts was higher. Otherwise *PAL* gene expression in the leaves of the pervious plant showed lower levels compared to Sunflower. A quite similar results were suggested by Moharram *et al*., (2017) and Tugbaeva *et al*., (2022) proving that Gaillardia and Zinnia have a high percentage of antioxidants in aerial parts.

APX3 gene plays an important part in the ascorbate-glutathione cycle by utilizing the reducing capacity of ascorbate to convert H_2O_2 into water and create monodehydroascorbate, (MDHA) (Asada, 1992). However, there were differences in the relative gene expression of *APX3* between the examined leaves and flowers. The relative expression of the *APX3* gene in Zinnia was down regulated in the leaves. Likewise in the flowers of Zinnia the APX3 expression was lower compared to Sunflower. In Gaillardia, the *APX3* gene was high levels in leaves. In floral parts of the same plant *APX3* gene expression showed the lowest level comparing to the Sunflower. In line with the outcomes attained by Chakrabarty *et al*., (2007), who stated that Chrysanthemum leaves had a significant increase in *APX* activity.

CONCLUSION

In this study, five plants from the *Asteraceae* family (Sunflower, Gazania, Gaillardia, Zinnia, and Chrysanthemum) were studied for their total phenolic content and antioxidant activity by chemical and gene expression means. TPC analysis showed that Sunflower and Chrysanthemum had the highest phenolic contents, 3.26±0.39 and 2.99±0.22, respectively. Furthermore, using the DPPH method, aerial parts of the studied plants showed no statistically significant difference in antioxidant activity. Gene expression analysis was done to assess the antioxidant capacity of several species of the Asteraceae family. Relative expression of Catalase, Phenylalanine Ammonia-lyase, and Ascorbate Peroxidase genes were tested on extracts of the plants' flowers and leaves compared to Sunflower. This research elucidated the continuous importance of some ornamental plants available in Egypt as significant sources of antioxidants, specifically identifying certain genes responsible for these antioxidants. Likewise this study demonstrated that the leaves of Gaillardia, Chrysanthemum, and Gazania serve as rich sources of catalase. Furthermore, the *APX3* gene was found to be extractable from the leaves of these plants. The expression of the *PAL* gene was observed to be higher in the leaves of Chrysanthemum. While in the floral parts of Gazania it was lower, with contrasting results observed in Zinnia and Gaillardia. We recommended using Sunflower and Chrysanthemum as the highest antioxidant source compared with other samples. Further studies should be done on how to use the extracts of Sunflower and Chrysanthemum in medicinal properties, specific analysis about its safe to use them in nutrient diet and who should avoid using them, to make sure that the both of them could use them as we use chamomile tea that is made from the chamomile flower and is used to treat a wide range of health issues. and many other plants.

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التعبير الجيني لبعض الجينات المرتبطة بمضادات الأكسدة في خمسة أنواع من عائلة *Asteraceae* في مصر.

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الملخص

تعد العائلة النجمية Asteraceae واحدة من أكبر فصائل النباتات المزهرة وتشتهر بوجود مضادات الأكسدة ومضادات الالتهاب وأيضًا نشاطها الواضح كمضادات للميكروبات. ومن الوارد أن تكون آثارها الدوائية مرتبطة بوجود بعض المركبات الكيميائية النباتية، بما في ذلك البوليفينول والأحماض الفينولية والفلافونويد والأسيتيلين والتربتربين. في هذه الدراسة ، تم تقييم نشاط مضادات الأكسدة في كل من عباد الشمس ((Helianthus annuusوالجازانيا ((Gazania rigensو العنبر كاشمير ((Gaillardia pulchellaو الزينيا (Zinnia (elegansو الأرولا ((Chrysanthemum morifolliumوتم أيضًا تحليل إجمالي محتوى الفينول ((TPCونشاط التعبير الجيني لبعض الجينات المرتبطة بمضادات الأكسدة ، بما في ذلك APX3وCATA1و PAL . وأوضحت النتائج أن نباتات عباد الشمس والأرولا كان لهما أعلى محتوى فينولي بحوالي (3.26 ± 0.39 و 2.99 ± 0.22 مجم / GAEجم) على التوالي، وكان التعبير الجيني لل PALأعلى بمقدار 4 أضعاف و 2 ضعف في زهور الأرولا وزهور الزينيا على التوالي، وذلك عندما تم مقارنته بزهور عباد الشمس. ومن الجدير بالذكر أن التعبير الجيني لل APX3 في أوراق الأرولا والجازانيا والعنبر كاشمير كان مرتفع عند مقارنته بأوراق عباد الشمس. وكنتيجة نهائية أثبت البحث مدى ارتفاع وجود مضادات الأكسدة لبعض النباتات غير المألوفة للعائلة النجمية .Asteraceae و يوصي الباحث باستخدام أوراق كل من نبات الأرولا والجازانيا والعنبر كاشمير كأحد مصادر مضادات الأكسدة، وضرورة استكمال الأبحاث في هذا الصدد للتأكد من درجة الأمان في استخدام هذه النباتات وادراجها داخل النظام الغذائي.

الكلمات*المفتاحية: Asteraceae* ، نشاط مضادات الأكسدة ، التعبير الجيني.