



Identification of Some Citrus Genotypes using Leaf Shape Analysis Based on Elliptical Fourier Descriptors

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ABSTRACT: Computer-based identification of plant species has been the subject to numerous investigations in the last few decades. Image analysis of plants organs have been performed using different features such as shape, color, texture and by employing various mathematical models. In this work, identification of eight citrus genotypes has been carried out through image analysis of their leaves. Elliptical Fourier Descriptors (EFDs) have been used to model the outline of the leaves and subsequent statistical analyses have been performed on these EFDs. The results from the discriminant analysis indicate success rates of greater than 95% in correct identification of different genotypes, which is comparable with the results in the existing literature. Furthermore, principal component and hierarchical clustering analyses have been proven to be efficient methods for classification of these genotypes. The results show that the image-processing method is effective in identification of the studied genotypes.

Key words: Image analysis, Elliptical Fourier Descriptors, Leaf Image, Shape Software, Chain code

INTRODUCTION

Image processing technology has found widespread application in agriculture. These include likes of the product recognition system used in the supermarket (Bolle *et al.*, 1996); automated process of picking the ripe tomatoes, Arefi *et al.* (2011); using thermal imaging as an approach to convert the pattern of invisible radiation from an object into visible images to facilitate the feature extraction and analysis (Vadivambaland Jayas. 2011); automatic detection of the symptoms of diseases (Dubey *et al.*, 2015). Furthermore, images are among the important data used for species identification. Classification of plants from their apparent structure is not an easy task for a layman and most of the methods used in plant taxonomy, such as molecular biological techniques are expensive and time-consuming and require expertise in botany (Du *et al.*, 2006). Moreover, although analytical methods like molecular markers are used for classification, this type of classification is not feasible due to wide variations in genetic (Spiegel-Roy and Goldschmidt, 1996).

Different species of plants contain specific shape features, which could be utilized for the identification process. Specifically, leaves seem to be the most common organs used for this purpose. Identification of plants through image analysis of their leaves is more advantageous compared to molecular biological approaches as it does not require the expertise of a botanist to perform the task (Jamil *et al.*, 2015).

Petry and Kuhbauch in 1989 carried out one of the earliest automatic leaf identification based on the shape parameters to classify weed species. Work of Arora *et al.* (2012), is based on leaves shape to identify different species. Sparse representation of leaf tooth features has been proposed as an identification method (Jin *et al.* 2015). Features other than shape such as color and texture could also contribute to recognition of various plant species from each other such as the image analysis of the cross-sections of leaf midrib using fractal descriptors (da Silva *et al.*, 2015). Alternative combinations of shape, color and texture were evaluated to obtain accuracy in the plant identification (Jamil *et al.*, 2015, Bama *et al.*, 2011, Kadir *et al.*, 2011). Images taken from plants have been subjected to different modeling approaches to acquire the desirable data such as leave outline, its color, etc. An automatic plant identification system was developed by applying the histogram of oriented gradient to recognize plants based on the leaf information (Pham *et al.*, 2013). Using Flavia data set, performance of this method was compared with Hu Descriptor, which showed better accuracy. Douglas Peucker approximation algorithm was adopted to obtain the shape of leaves (Du *et al.*, 2006). Then a modified dynamic programming algorithm was proposed for matching and recognizing the plant leaf. Probabilistic neural network was introduced to implement a leaf recognition algorithm based on a predefined number of morphological features (Wu *et al.*, 2007).

Quantitative trait loci mapping of the wheat seeds was performed by application of phenotypic data and Elliptical Fourier's descriptors (Williams, 2013). EFDs, developed by Kuhl and Giardina (1982), are used to describe closed contours. Since then, they have found a variety of applications in the analysis of biological forms and organs.

In the present work, EFDs have been employed to classify different genotypes of citrus family based on the shape of their leaves. Identification of citrus species is an urgent step for qualification, germoplasm conservation programs and breeding (Milori *et al.*, 2013). Shape of leaves exhibits wide variations in

genus citrus and could be considered as a key taxonomic feature to classify the citrus species (Iwata *et al.*, 2002). Implementation of EFDs and statistical analysis on the results have been performed using SHAPE and SPSS software respectively.

MATERIALS AND METHODS

In this work, 500 leaves from eight genotypes of Citrus family have been selected. The leaves have been collected from Ramsar-Mazandaran Province of Iran with the scientific names of their genotypes shown in Table 1.

Table 1: Scientific and common names of citrus genotypes.

Scientific Name	Common Name
<i>Citrus limon</i> cv. Eurica	Lemon
<i>Citrus reticulata</i> cv. Page	Page mandarin
<i>Fortunella Margarita</i>	kumquat
<i>Citrus unshiu</i>	Satsuma mandarin
<i>Citrus sinensis</i> cv. Navel	Thompson orange
<i>Citrus reticulata</i> cv. Yunesi	Yunesi mandarin
<i>Citrus aurantium</i>	Sour orange
<i>Citrus sinensis</i>	Orange (wild type)

The leaves were scanned with Canon 550 MP and 300dpi accuracy, and the obtained images were filtered to remove the ones with poor quality, leaving 430 scanned samples as input for the image analysis. Images obtained thus far were fed into SHAPE software where they were processed as described in the following sections. SHAPE is a free computer program developed by Iwata and Ukai (2002) and is used for quantitative analysis of the biological images.

A. Generation of Chain Coder

Based on the work of Kuhl FP and Giardina CR, 1982, in this step, outline of each image is turned into a chain

of numerical codes, which define the direction of the successive points on the perimeter of the leaf with respect to each other. Fig. 1 presents an example of a chain code.

The chain code generates an approximation of a continuous contour through a chain of linear fits. The code of contour is then the chain *V* of length *k* as below.

$$V = s_1 s_2 s_3 \dots s_k \dots (1)$$

Eqn. 1 describes the chain *V* of length *k*, in which *s_i* is an integer between 0 and 7 oriented in the direction (/4)*s_i*

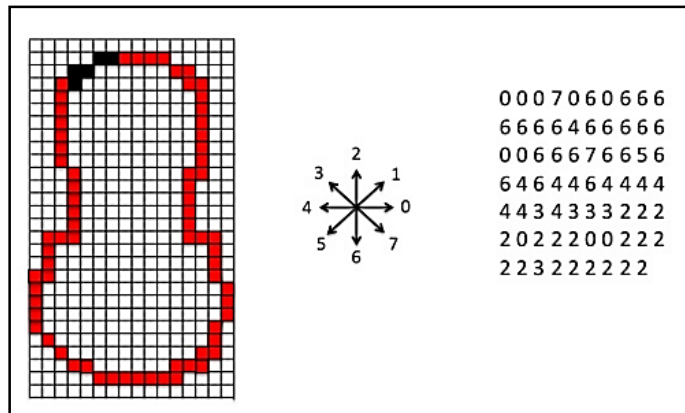


Fig. 1. Example of a chain code.

Given that the time needed to traverse s_i is t_i , the time T required to traverse the entire contour is calculated according to Eqn. 2.

$$T = \sum_{i=1}^k \Delta t_i \dots (2)$$

$T=t_k$ is known as the basic period. Projections of the chain on x and y axes, as link s_i is traversed are:

$$x_i = \text{sgn}(5 - s_i) \text{sgn}(2 - s_i), \dots (3)$$

$$y_i = \text{sgn}(4 - s_i) \text{sgn}(s_i), \dots (4)$$

Where

$$\text{Sgn}(Z) \begin{cases} 1 & Z > 0 \\ 0 & Z = 0 \\ -1 & Z < 0 \end{cases} \dots (5)$$

Choosing the origin as an optional starting point for the chain code sums of the projections on x and y of the contour areas follows:

$$x_p = \sum_{i=1}^p \Delta x_i \dots (6)$$

$$y_p = \sum_{i=1}^p \Delta y_i \dots (7)$$

The chain code determined in this way is an identifier for the entire outline of the leaf and is used in the next section of the software to progress the image analysis process.

B. Elliptical Fourier Descriptors

The chain codes obtained in the previous step are analyzed by Elliptical Fourier Descriptor method, Kuhl FP and Giardina CR, 1982. This simplifies the long chain of codes into a predefined number of coefficients. Given that x from Eqn. 6 is expanded by Fourier's series, one will have:

$$x(t) = A_0 + \sum_{n=1}^{\infty} a_n \cos \frac{2n\pi t}{T} + b_n \sin \frac{2n\pi t}{T} \dots (8)$$

Where the coefficients a_n and b_n are determined by:

$$a_n = \frac{T}{2n^2\pi^2} \sum_{p=1}^k \frac{\Delta x_p}{\Delta t_p} \left[\cos \frac{2n\pi t_p}{T} - \cos \frac{2n\pi t_{p-1}}{T} \right] \dots (9)$$

$$b_n = \frac{T}{2n^2\pi^2} \sum_{p=1}^k \frac{\Delta x_p}{\Delta t_p} \left[\sin \frac{2n\pi t_p}{T} - \sin \frac{2n\pi t_{p-1}}{T} \right] \dots (10)$$

In a similar way the projection y could be expanded to obtain coefficients c_n and d_n . Using a greater number of harmonies n more precise presentation of x and y could be obtained through above transformation. In the current work, the value of k has been set to 20, meaning that 20 harmonies have been used to generate the outline of a leaf. Considering that each harmony contains 4 coefficients i.e. a_n, b_n, c_n and d_n , in total 80 parameters have been determined for each leaf as the outcome of this section.

C. Randomness Checks

To ensure that the selected leaves form a random representative of their group, distribution of their EFDs has been examined prior to further analyses. It was observed that EFDs followed a normal distribution which indicated random nature of the chosen leaves. Fig. 2 presents the distribution of the a_2 coefficient related to the second harmony between the EFDs of Lemon's genotype. As it is seen from this figure, the probability of occurrence resembles the normal distribution function.

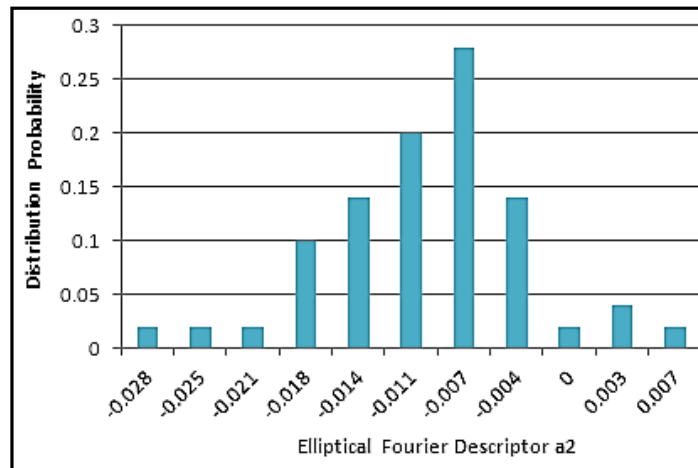


Fig. 2. Distribution of a sample EFD for leaves of lemon.

D. Statistical Analysis

The EFDs generated have been converted into an Excel file and then imported into SPSS software for further analyses. SPSS is a software package developed by IBM and is used for statistical analysis. Current work includes the following statistical analyses based on the results obtained from SPSS.

1. Canonical Discriminant Analysis (CDA)
2. Principal Component Analysis (PCA)
3. Hierarchical Cluster Analysis (HCA)

In SPSS, EFDs have been subjected to principal component analysis (PCA) so that the dimension of data, being 80, is reduced to a manageable size which could simplify the subsequent statistical interpretation.

To carry out PCA, covariance matrix of the EFDs was calculated as follows.

Considering C as the covariance matrix:

$$C^{q \times q} = (c_{i,j}, c_{i,j}) = cov(Dim_i, Dim_j) \dots (11)$$

Where $q=4k$ with k being the number of harmonies used in EFDs, cov , the covariance operator and Dim_i , the coefficient vector accounting for $\vec{a}_1, \vec{b}_1, \vec{c}_1, \vec{a}_2, \vec{b}_2, \vec{c}_2, \vec{a}_k, \vec{b}_k, \vec{c}_k, \vec{d}_k$. These vectors represent the EFDs calculated for the leaves

$$EVector = (eigenvector_1, eigenvector_2 \dots eigenvector_q) \dots (12)$$

$$EValue = (eigenvalue_1, eigenvalue_2 \dots eigenvalue_q) \dots (13)$$

EVector and E Value are written in descending order with respect to eigen values starting from the greatest eigen value and its corresponding vector.

To obtain principal components, PCs, linear combinations of EFDs are created using the component of EVector. For instance, the first PC is calculated as:

$$PC_1 = eigenvector_{1,1} \times \vec{a}_1 + eigenvector_{1,2} \times \vec{b}_1 + \dots + eigenvector_{1,q} \times \vec{d}_k \dots (14)$$

Likewise q^{th} PC will be as:

$$PC_q = eigenvector_{q,1} \times \vec{a}_1 + eigenvector_{q,2} \times \vec{b}_1 + \dots + eigenvector_{q,q} \times \vec{d}_k \dots (15)$$

Besides EFDs, PCs calculated in this way are another method for presentation of the outlines of leave. Each PC includes a proportion of variation, which is defined by the eigen value for that PC divided by the sum of the eigen values.

Generally, the first n PCs are retained so that the outline of a leaf could be described by as a small number of parameters as possible. Therefore, a balance should be obtained between the following conflicting options:

1. To achieve a simple solution, n should be as small as possible.
2. To avoid loss of data, the proportion of variation described via the first n PC should be as large as possible; ideally as close to one.

The results presented within this paper are based on utilization of the first five PCs. As it is shown in the results, high proportion of the variance among the input data could be captured by these PCs without significant loss of information.

within each genotype. For example \vec{a}_1 for lemon genotype is a vector composed of 50 elements corresponding to the total number of leaves within that genotype in which each element is calculated according to Eqn. 9.

Since C is a square matrix with q dimensions, set of q eigen values and eigenvectors could be calculated for it, which is shown as below:

RESULTS AND DISCUSSION

Following sections illustrate the outcome of the analyses performed on the digital images of the leaves using SHAPE and SPSS packages.

A. Canonical Discriminant Analysis

Table 2 presents the results of the CDA of the shape of the leaves. The figures shown are an indication of the level of confidence for the correct classification of an unknown leaf. According to the results, the highest and lowest success rates in identification belong to leaves from Satsuma mandarin and local orange respectively. In other words, given that a specific leaf is Satsuma mandarin it could be correctly assigned to its group by the current image analysis process with 100% success rate. On the other hand, the possibility that a leaf from local orange genotype could be accurately identified via this process stands at 90% meaning that a 10% error may occur in classification of this genotype.

Table 2: Results of the CDA produced by SHAPE.

Genotypes (common names)	Lemon	Page mandarin	Kumquat	Satsuma mandarin	Thompson orange	Yunesi mandarin	Sour orange	Orange	Total
Lemon	98.00	2.00	0.00	0.00	0.00	0.00	0.00	0.00	100.00
Page mandarin	0.00	98.00	0.00	0.00	0.00	0.00	0.00	2.00	100.00
Kumquat	0.00	0.00	96.00	0.00	0.00	0.00	4.00	0.00	100.00
Satsuma mandarin	0.00	0.00	0.00	100	0.00	0.00	0.00	0.00	100.00
Thompson orange	0.00	0.00	0.00	0.00	92.50	7.50	0.00	0.00	100.00
Yunesi mandarin	0.00	5.7	0.00	0.00	2.9	91.40	0.00	0.00	100.00
Sour orange	0.00	1.4	1.4	0.00	0.00	0.00	97.10	0.00	100.00
Orange(wild type)	0.00	2.50	0.00	5.00	0.00	0.00	2.5	90.00	100.00

95.6% of original grouped cases correctly classified.

B. Principal Component Analysis

Fig. 3 presents the shape of leaf obtained using five principal components for lemon. The images in the middle column are representative of mean value for

leaves of these genotypes while the columns on the left and right present the mean value by addition or subtraction of 2.5 times the standard deviation.

From Fig. 3, PC1 seems to affect the overall widening of the leaf along its length, while PC2 appears to identify the skewing relative to the midrib. Other PCs represent only subtle visible trends at the base or tail of the leaf and this is expected as the proportion of the variance included in the successive PCs decreases moving farther from PC1. This is further explained in Table 3. Table 3 presents the eigen values and their

cumulative variances for the first five PCs. It appears that using five PCs to identify the leaves has achieved at least 90% of the variances existed in the outlines of the leaves. As it is seen from Table 3, principal component analysis of the lemon genotype has collected the highest variance of 95.5% using the first five PCs, while in the case of Satsuma mandarin genotype 90.3% of the variance could be represented.

Table 3: Results of Principal Component Analysis of images produced by SHAPE.

Genotypes		PC1	PC2	PC3	PC4	PC5	Total Variance
Lemon	Eigen value	0.00241	0.00067	0.00042	0.00013	0.00011	0.0041
	Proportion(%)	59.20860	16.51060	10.19310	3.22670	2.59350	
	Cumulative(%)	59.20860	75.71920	85.91230	89.13890	91.73250	
Page mandarin	Eigen value	0.00108	0.00027	0.00011	0.00007	0.00005	0.0017
	Proportion(%)	63.72700	15.70770	6.37120	4.14940	2.78390	
	Cumulative(%)	63.72700	79.43470	85.80590	89.95540	92.73930	
Kumquat	Eigen value	0.00136	0.00057	0.00041	0.00009	0.00005	0.0026
	Proportion(%)	51.99140	21.62140	15.71880	3.50310	1.99200	
	Cumulative(%)	51.99140	73.61280	89.33160	92.83480	94.82670	
Satsuma mandarin	Eigen value	0.00077	0.00043	0.00027	0.00019	0.00005	0.0019
	Proportion(%)	40.66980	22.67060	14.16250	9.99780	2.88610	
	Cumulative(%)	40.66980	63.34040	77.50290	87.50070	90.38680	
Thompson orange	Eigen value	0.00194	0.00122	0.00025	0.00009	0.00007	0.0038
	Proportion(%)	51.79440	32.62110	6.69840	2.47350	1.99520	
	Cumulative(%)	51.79440	84.41560	91.11400	93.58740	95.58260	
Yunesi mandarin	Eigenvalue	0.00067	0.00052	0.00036	0.00008	0.00006	0.0018
	Proportion(%)	36.54770	28.08640	19.82900	4.24920	3.39300	
	Cumulative(%)	36.54770	64.63410	84.46310	88.71230	92.10530	
Sour orange	Eigen value	0.00120	0.00090	0.00065	0.00017	0.00005	0.0032
	Proportion(%)	37.84900	28.53300	20.47830	5.34770	1.68380	
	Cumulative(%)	37.84900	66.38200	86.86030	92.20800	93.89180	
Orange (wild type)	Eigen value	0.00122	0.00040	0.00033	0.00014	0.00008	0.0024
	Proportion(%)	51.60440	16.77130	13.99830	5.96490	3.25300	
	Cumulative(%)	51.60440	68.37570	82.37400	88.33880	91.59180	

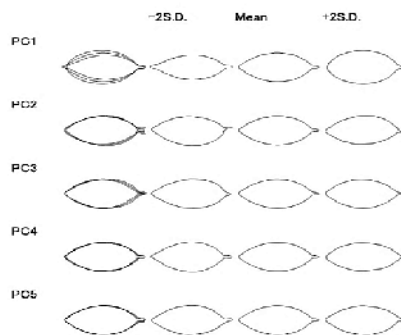


Fig. 3. Variations captured by PCs for shape of leaves in lemon genotype.

C. Hierarchical Cluster Analysis

Fig. 4 portrays the dendrogram obtained through HCA of the subject leaves. Different genotypes have been joined to form the cluster tree based on the average ecological distance across each cluster. It explains the level of similarity between various genotypes and the ecological distance at which they could be clustered together. According to this, dendrogram, Thompson orange and local orange revealed the highest degree of

similarity by forming a cluster at an ecological distance of around 1. Overall, two main clusters could be identified for the eight genotypes of the leaves investigated in this paper. First cluster contains Thompson orange, local orange and lemon genotypes. While the second cluster is composed of page orange, Satsuma mandarin, yunesi mandarin and sour orange genotypes. These two clusters join at ecological distance of around 25.

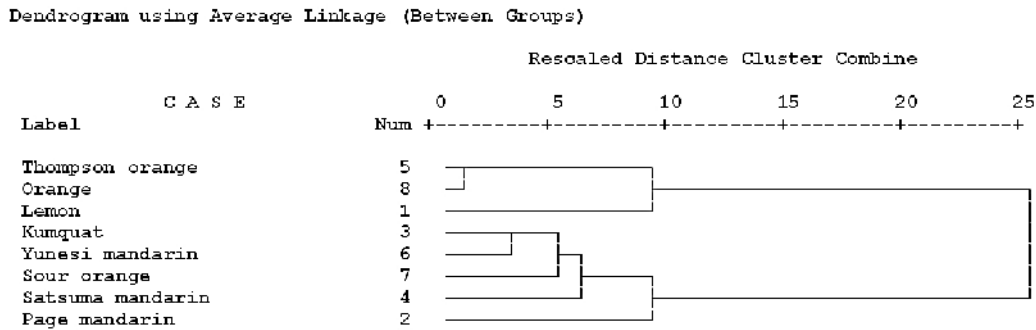


Fig. 4. Dendrogram of the Citrus Genotypes via HCA produced by SPSS.

D. Comparison of the Results

In this section, the rate of identification achieved in the results is compared with the identification rates in the existing literature. To this end, Table 4 has been prepared, which summarizes the success rates stated in a few works introduced in Section 1. According to Table 4, the rate of successful identification through the

image-processing method applied in the present study is comparable with the figures obtained by others using alternative methods. The higher success rate achieved in this study could be attributed to the utilization of a sufficient number of EFDs for accurate definition of the outlines of the leaves.

Table 4: Comparison of the Identification Rate between some similar articles.

Reference	Method	Identification Rate
Present Work	EFDs	95.6%
Jamil <i>et al.</i> (2015)	Scale Invariant Feature Transform , Color Moment; Fractal Texture Analysis	86.8%
Pham <i>et al.</i> (2013)	Histogram of oriented gradient	84.68%
da Silva <i>et al.</i> (2015)	Fractal Descriptors	87.29%
Du <i>et al.</i> (2006)	shape matching	92.3%

CONCLUSION

Digital image analysis of eight citrus genotypes has been performed to evaluate the functionality of the computer-based identification to classify these genotypes and to obtain the level of similarities between them. Outline of 430 leaves belonging to above genotypes were subjected to image processing by EFDs. Subsequently, three different statistical methods have been applied to interpret the results from various aspects. CDA revealed promising results for accurate classification of the genotypes by achieving a success rate of greater than 95%. Furthermore, principal component analysis illustrated the prominent features which could be considered in the classification process. It was shown that using the first five principal components, at least 90% of the variances among each genotype could be retained. Finally, different

genotypes of leaves were subjected to HCA which identified the overall similarity between the genotypes by clustering them into multiple groups based on the ecological distances. The obtained results confirm the capability of the image-processing techniques in efficient identification of the studied genotypes.

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