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RESEARCH PAPER

Application of Image Processing for Gastropod Identification: Analysis of Shell Morphology

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Abstract

Gastropods, a diverse group of invertebrates with single shells, are often identified through their unique shell shapes and patterns. Computer-assisted taxonomic assessments from shell morphology can save time and eliminate human error in taxonomic studies on Gastropods. Recently computer-based applications have focused on species identification based on visual records of individuals, reducing errors and biases due to human interpretation. This study evaluated the applicability of data library created from images of 10 individuals of six different Gastropod species taken from four different angles using a stereo microscope for species identification. These images were processed into digital data with SqueezeNet and Inception v3 algorithms and analyzed using cosine distance and hierarchical clustering techniques. Analysis carried out with the library data showed that images of the same species in our library dataset were clustered together. When the analyses were repeated using visual records of the species being aimed for identification, it was observed that 90% of the newly considered individuals were classified under their respective species clades. This suggests that taxonomic identification is an applicable methodology that can be applicable quickly from images taken in field or laboratory conditions, potentially serving as a preliminary evaluation in advanced species identification studies.

Introduction

The class Gastropoda belongs to the Molluscs, which are prehistoric animals and are of great ecological importance. Gastropods consist of a single solid shell that surrounds their body. The morphologies of gastropod shells exhibit significant taxonomic variability, with distinct shapes and patterns indicative of their respective class, order, family, genus, and species. Researchers frequently utilize these shell morphologies, which display a wide spectrum of sizes, chromatic variations, and forms, including helical, conical, and planar configurations. (Onpans et al., 2018; Sultana et al., 2021). Although the basic characters for morphological species determination have been defined, there are difficulties in identification of Gastropods due to the wide variety of morphological characters. Moreover, gastropods exhibit ontogenetic variability in morphological characters, coupled with a pronounced capacity for phenotypic plasticity. This phenomenon is further complicated by the presence of both synonymous and homonymous species within the taxonomic framework (Altaf et al., 2017). Therefore, the identification of gastropod species requires experienced qualified experts. Although molecular species identification methods are frequently used to eliminate this complexity, they are expensive, time-consuming and require part of the organism (Altaf et al., 2017; Borges et al., 2016; Sultana et al., 2021). Furthermore, the use of computer-assisted automated systems for morphological species determination will allow non-experts in this field to do species identification fast and affordable way (Onpans et al., 2018; Sultana et al., 2021).

Computer-assisted automated systems are a powerful method to overcome the problems that during morphological species identification and can significantly help in species identification by facilitating

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the reliable identification of any specimen in a population (Godfray et al., 2004; Kalafi et al., 2018; Soberón & Peterson, 2004). Among these methods, especially image-based analyses are widely used in the field of biodiversity (Francoy et al., 2008; Leow et al., 2015; Wen et al., 2009; Yousef Kalafi et al., 2016). Images processing has application in several scientific areas, including the identification of numerous species, such as spiders (Lameed, 2012), wild animals (wolf, fox, brown bear, deer and wild boar) (Matuska et al., 2014; Shalika & Seneviratne, 2016), plants (Dyrmann et al., 2016), fish (Andayani et al., 2019; Li & Hong, 2014), birds (Marini et al., 2013), microalgae (Chong et al., 2023, 2023), and disease detection (Iqbal et al., 2018). Image processing is a part of computer-assisted vision systems designed by advanced and high-end machines that can fulfill the function of the human eye. This method enables to obtain many important information about the object of interest by enriching, filtering and segmenting the pixels in digital images. Thus, by processing the details that the human eye cannot see, it transforms visual data into numerical data and allows them to be analyzed statistically (Gurau et al., 2013; Kini et al., 2023). This method, which is based on the detection of morphological characters through images, plays an important role in species classification studies (Kalafi et al., 2018).

Nowadays, collecting high quality images of gastropod shells and sharing them digitally makes it possible to make identification through images (Onpans et al., 2018). Image processing technique helps in taxonomy based on shell morphology in Gastropods in areas such as sharpening images and detecting details. This technique when applied to species identification studies from shell morphology stands out with the advantages of eliminating problems due to human error, reducing dependence on experts and obtaining accurate results in a short time (Gurau et al., 2013; Onpans et al., 2018).

In this study, it was aimed to (*i*) create a library for the identification of Gastropod species based on visual records of 60 individuals of 6 species (*Neverita josephinia* (Risso, 1826), *Steromphala varia* (Linnaeus, 1758), *Ergalatax junionae* (Houart, 2008), *Conomurex persicus* (Swainson, 1821), *Columbella rustica* (Linnaeus, 1758) *and Cerithium lividulum* (Risso, 1826)) taken from 4 different angles and (*ii*) use the image processing method to identify species based on the created libraries.

Materials and Methods

Capturing Images

The shell structures of 60 individuals of 6 different species (*N. josephinia* [NJ], *S. varia* [SV], *E. junionae* [EJ], *C. persicus* [CP], *C. rustica* [CR] and *C. lividulum* [CL]) were photographed from 4 different angles (dorsal, ventral, apex and siphon) using an Olympus SZX-16 (Olympus, Japan) stereo microscope at 1X magnification under constant light. A reference library was created by grouping each of the 240 images obtained in folders belonging to their species and shooting angle. Furthermore, two specimens (three for *S. varia*) of each species that were not part of the library were captured using same photography techniques and organized based on their shooting angles in a separate folder named sample dataset. These images in the sample dataset folder were used in subsequent analyses for species identification by image processing. The species used in the creation of the library were named with the abbreviation of the species names, while the specimens to be used in the species identification analysis were included in the analysis by placing an X next to the species names (Table 1).

In order to remove distortion caused by the background of the images, the backgrounds were removed using code created with Python programming language (Sanner, 1999). For this purpose, rembg (Das, 2023) and pillow (Clark, 2024) libraries were used. The images with removed backgrounds were saved in "png" format and the subsequent analyses were performed with use of these images.

Obtaining Quantitative Data from Images

Image embedding algorithms SqueezeNet (Iandola et al., 2016) and Inception V3 (Xia et al., 2017) were used to extract quantitative data from images. The SqueezeNet algorithm is a small and fast image identification model based on ImageNet (Deng et al., 2009). The Inception v3 algorithm is also based on ImageNet and includes Google support. In our study, both algorithms were used in the process of obtaining digital data from the visual records to be analyzed. Image embedding analysis provided a total of 2047 numerical data for each image which were then used for the subsequent analysis.

Table 1. Species used in the study, sampling location, sampling date and their nomenclature used in the analysis

Specimen Name	Library Dataset	Sample Dataset	Location (Antalya, Türkiye)	Date (D/M/Y)
Neverita josephinia	NJ	NJ-X	Kundu	16.01.2015
Steromphala varia	SV	SV-X	Beldibi	12.12.2004
Ergalatax junionae	EJ	EJ-X	Beldibi	10.02.2005
Conomurex persicus	CP	CP-X	Beldibi	12.12.2004
Columbella rustica	CR	CR-X	Beldibi	12.12.2004
Cerithium lividulum	CL	CL-X	Phaselis	29.04.2005

To handle multiclassification issues in digital data acquired from images, the multinomial logistic regression classifier (Softmax Regression) was employed to derive logistic regression (Wright, 1995). The logistic regression model was cross-validated, and the significance of the model was assessed.

To evaluate the performance of the classification model used, Test&Score analysis was performed with the results of logistic regression analysis. To determine the misclassifications and low accuracy confusion matrix analysis was used. Images located in wrong species clusters within the created confusion matrix were removed from the analysis, so as not to cause a false assessment.

Hierarchical ClusIter AnaIlysis

Statistical distance calculation and hierarchical clustering analysis were applied to classify the images. The distance calculation was performed with the Cosine algorithm, which uses the cosine of the angle between two vectors of an inner product of data in the spatial plane (Lahitani et al., 2016). The hierarchical clustering analysis was repeated for both library creation and species identification. First, the samples evaluated for library creation were subjected to hierarchical clustering analysis to determine how the species were separated from each other. Then, the sample dataset prepared for species identification was included in the optimized libraries and it was determined under which species these samples were clustered in the library. Finally, all trees obtained as from the clustering analysis were visualized and colored.

Results

Within the scope of the study, images of the shell structures of 60 individuals of 6 different species, taken from 4 different angles, were classified by image processing (Figure 1). After removing the backgrounds of the images, analyses were performed to obtain quantitative data from the images. Finally, species classification was performed as a result of hierarchical clustering analysis.

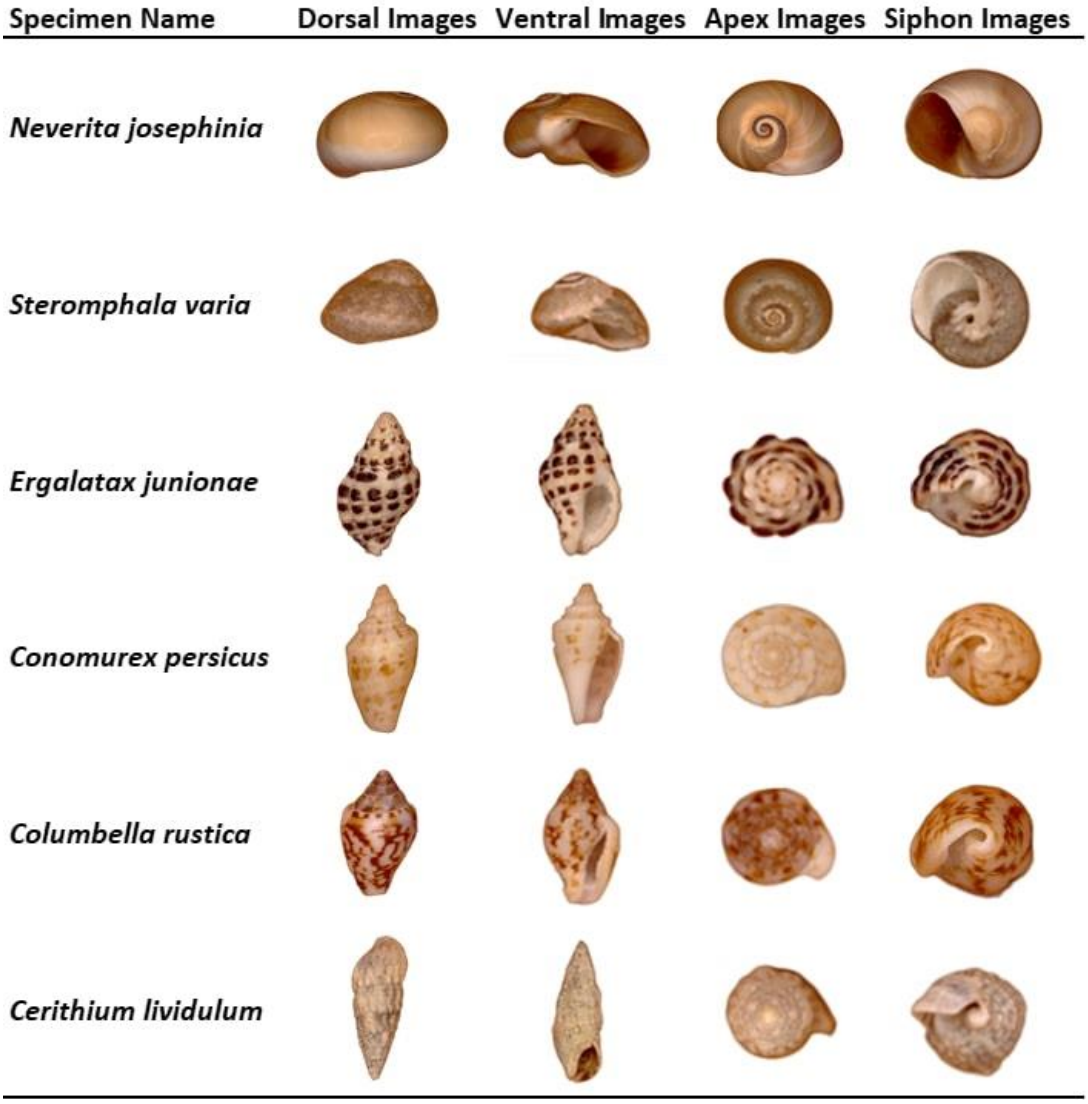

Figure 1. Images of the species used in this study taken from 4 different angles

As a result of the clustering analysis performed to create a library based on the dorsal angle images, it was seen that all individuals belonging to the species clusters were clearly separated from each other. Confusion matrix analysis (Table 2) shows that all individuals were perfectly distributed (100%). There were no deviations observed when clustering the images of each individual under their own species. However, NJ and SV species, which have similar shell morphology compared to the other 4 species, were clustered in a separate cluster from the other species (Figure 2).

After the creation of the dorsal library, the sample dataset was included in the analysis to evaluate the possibility of species identification. Despite the overall success of species identification, it was noted that the CL-X1 individual was grouped together with an individual from the CP species under the CL species category. Similarly, the CL-X2 individual was clustered under the CP species category (Figure 3).

The clustering analysis conducted to create a library on the ventral angle images revealed distinct separation among all species (98%). The confusion matrix analysis revealed that only one deviation was observed when clustering the images (Table 3). One of the CR individuals is classified under CP species. However, unlike the dorsal library, NJ and SV species

Table 2. Confusion matrix analysis of dorsal library

		Predicted Class						
		CL	CR	CP	NJ	EJ	SV	Total
Actual Class	CL	10	0	0	0	0	0	10
	CR	0	10	0	0	0	0	10
	CP	0	0	10	0	0	0	10
	NJ	0	0	0	10	0	0	10
	EJ	0	0	0	0	10	0	10
	SV	0	0	0	0	0	10	10
	Total	10	10	10	10	10	10	60
Accuracy		100%	100%	100%	100%	100%	100%	100%
Error		0%	0%	0%	0%	0%	0%	0%

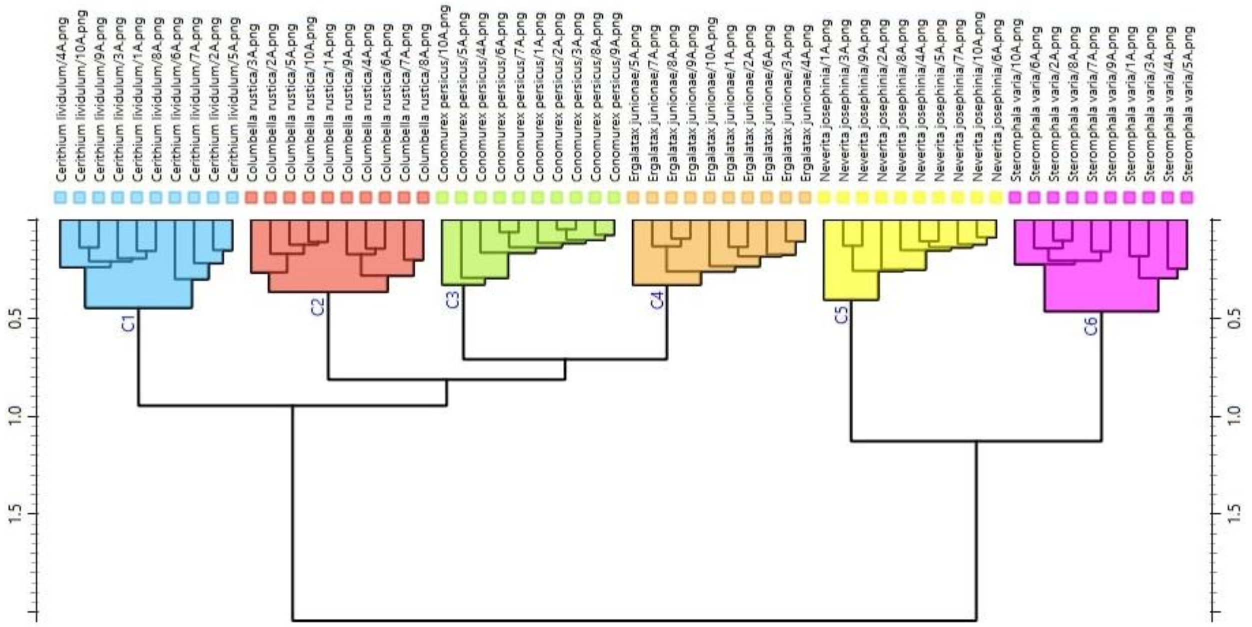

Figure 2. Library tree obtained from hierarchical clustering analysis of dorsal images of species.

Table 3. Confusion matrix analysis of ventral library

		Predicted Class						
		CL	CR	CP	NJ	EJ	SV	Total
Actual Class	CL	10	0	0	0	0	0	10
	CR	0	9	1	0	0	0	10
	CP	0	0	10	0	0	0	10
	NJ	0	0	0	10	0	0	10
	EJ	0	0	0	0	10	0	10
	SV	0	0	0	0	0	10	10
	Total	10	9	11	10	10	10	60
Accuracy		100%	90%	100%	100%	100%	100%	98%
Error		0%	10%	0%	0%	0%	0%	2%

with similar shell morphology were placed in a separate cluster from the other species (Figure 4).

With the addition of the sample dataset to evaluate the possibility of species identification to the ventral library, it was observed that species identification was generally successful. However, the EJ-X2 individual caused the EJ-X1 individual to be clustered under the CP species, while one of the EJ individuals used in the creation of the library was placed under the CL species (Figure 5).

In the library created with the images taken from the apex angle, it was determined that the species classification was done successfully (93%). However, unlike the dorsal and ventral libraries, individuals belonging to CP, CR and CL species were observed to be scattered in this library. To be able to progress with the identification phase of our analysis 3 individuals of each species that caused confusion were identified by confusion matrix analysis and removed from the hierarchical clustering analysis (Table 4). Thus, a correct library was created, and identification analyses were continued over this library (Figure 6). Also with the apex library, it was observed that species identification was performed successfully. All individuals clustered within their own species, whereas SV-X3 clustered within the CR species (Figure 7).

Data gathered form siphon angle revealed that the individuals of the CR species were widely dispersed across the obtained tree, leading to confusion in the library (Table 5). It was also observed that 3 individuals belonging to CP species settled under different species in the same way. For this reason, the entire CR species and the 3 individuals of the CP species were removed from the analysis and the library was created in a correct

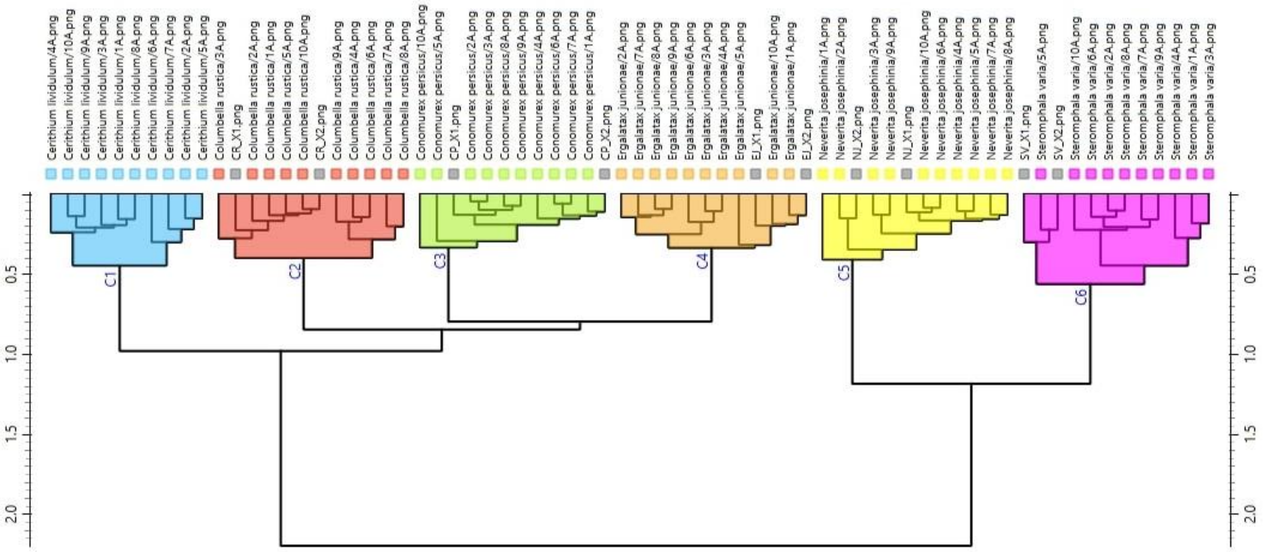

Figure 3. Tree obtained from identification analysis of dorsal images of the species.

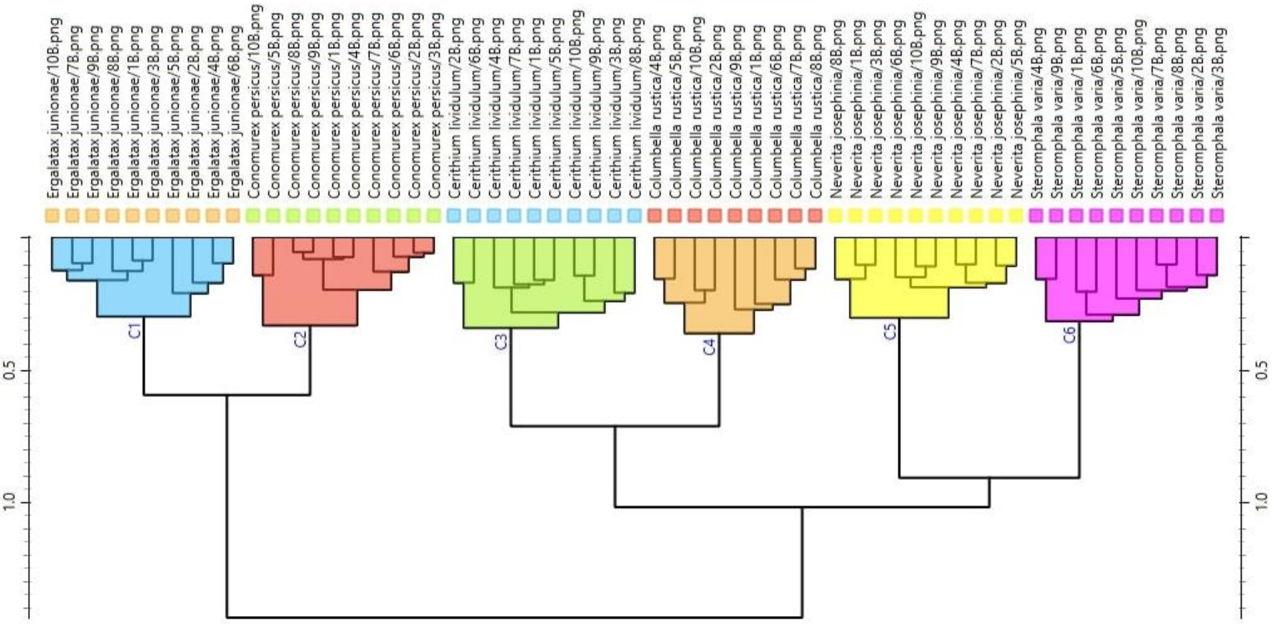

Figure 4. Library tree obtained from hierarchical clustering analysis of ventral images of species.

Table 4. Confusion matrix analysis of apex library

		Predicted Class						
		CL	CR	CP	NJ	EJ	SV	Total
Actual Class	CL	9	1	0	0	0	0	10
	CR	1	9	0	0	0	0	10
	CP	1	1	8	0	0	0	10
	NJ	0	0	0	10	0	0	10
	EJ	0	0	0	0	10	0	10
	SV	0	0	0	0	0	10	10
	Total	11	11	8	10	10	10	60
Accuracy		90%	90%	80%	100%	100%	100%	93%
Error		10%	10%	20%	0%	0%	0%	7%

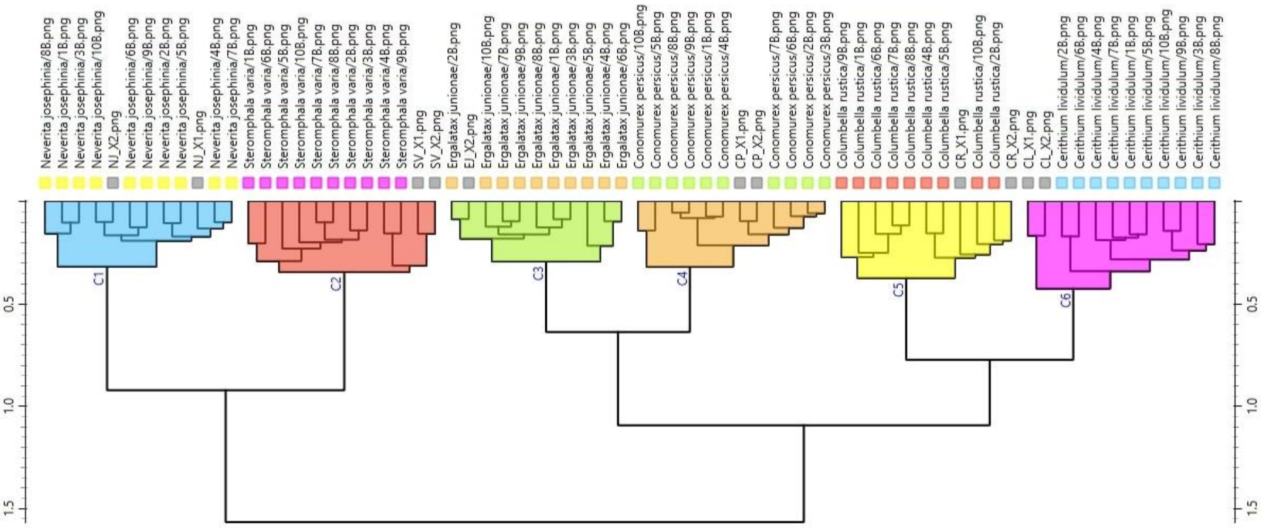

Figure 5. Tree obtained from identification analysis of ventral images of the species.

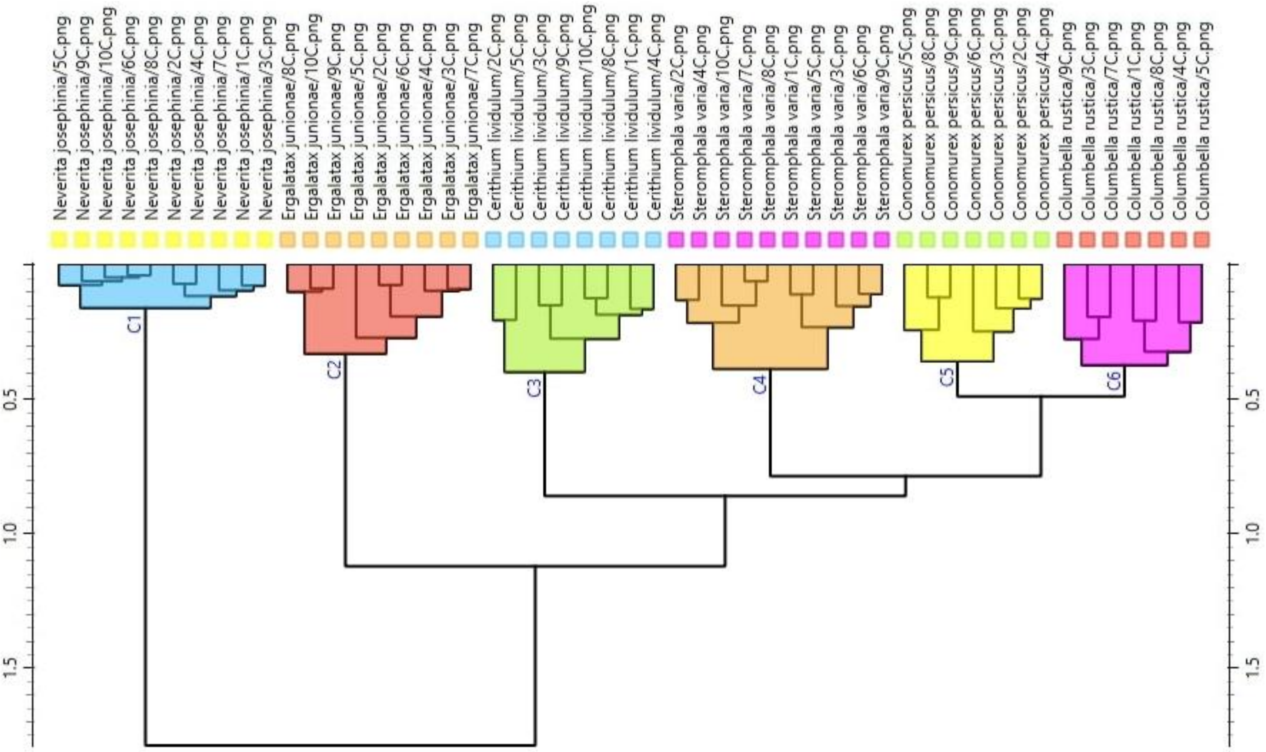

Figure 6. Library tree obtained from hierarchical clustering analysis of apex images of species.

way. Only after the removal of individuals belonging to the CR species was the library able to clearly distinguish between the species and the library has been created that can be used for further analysis. (Figure 8). As a result of confusion matrix analysis, the accuracy rate was calculated 92% for siphon library.

The inclusion of the sample dataset in the siphon library for species identification showed that CP-X1 and CP-X2 individuals clustered under the CK species. All other species were successfully separated from each other (Figure 9). Since the CR species was removed from the library because it caused confusion during library preparation, the individuals in the sample dataset of these species could not be evaluated in the species identification analysis.

Discussions

In this study, it was aimed to classify Gastropod species using image processing method. For this purpose, a total of 240 images of 6 species taken from 4 different angles were processed and classified by image processing method. Using the library created with these images, the species of the test specimens were identified.

In the literature, dorsal and ventral images were generally used in studies on the classification of gastropods because they reflect the whole shape of the organism. (Bouzaza, 2019; Cruz et al., 2012; Onpans et al., 2018). In this study, in the library created with dorsal (100%) and ventral (98%) images, species could be clearly separated from each other without much loss of individuals. The dorsal library exhibited the highest level of discriminatory ability. This high success rate observed in the dorsal library is consistent with the literature's findings (Bouzaza, 2019; Cruz et al., 2012; Onpans et al., 2018).

In the ventral library, SV and NJ species appear wider than the other shells when viewed dorsally. In the images taken from the ventral angle, this width distinction is not so clear. For this reason, neither SV nor NJ species, which form a different cluster by separating from other species in the dorsal library, are separated from other species in the ventral library but are not clustered together in a different cluster. Therefore, it can be said that the dorsal library shows a better discrimination than the ventral library for the species used in this study. Among the images added for species identification, EJ-X2 was clustered together with CP species. Species classification was successfully performed from the other images except EJ-X2.

Table 5. Confussion matrix analysis of siphon library

		Predicted Class						
		CL	CR	CP	NJ	EJ	SV	Total
Actual Class	CL	10	0	0	0	0	0	10
	CR	1	7	1	0	0	1	10
	CP	0	0	10	0	0	0	10
	NJ	0	0	0	10	0	0	10
	EJ	0	1	0	0	9	0	10
	SV	0	0	0	0	0	10	10
	Total	11	8	11	10	9	11	60
Accuracy		90%	70%	100%	100%	90%	100%	92%
Error		10%	30%	0%	0%	10%	0%	8%

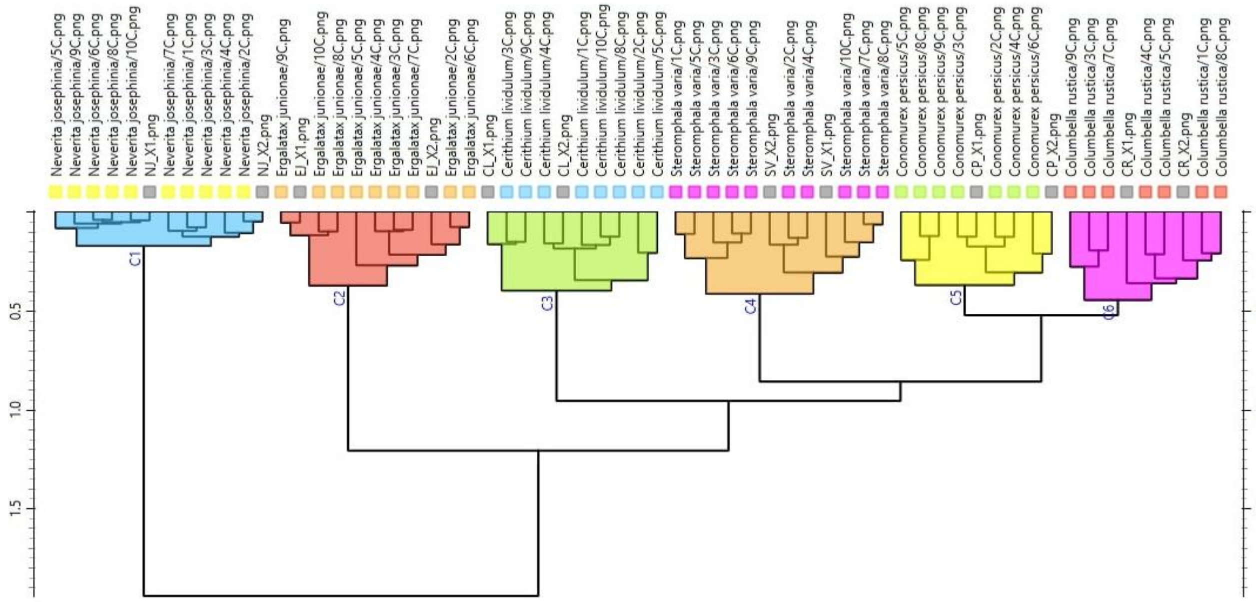

Figure 7. Tree obtained from identification analysis of apex images of the species.

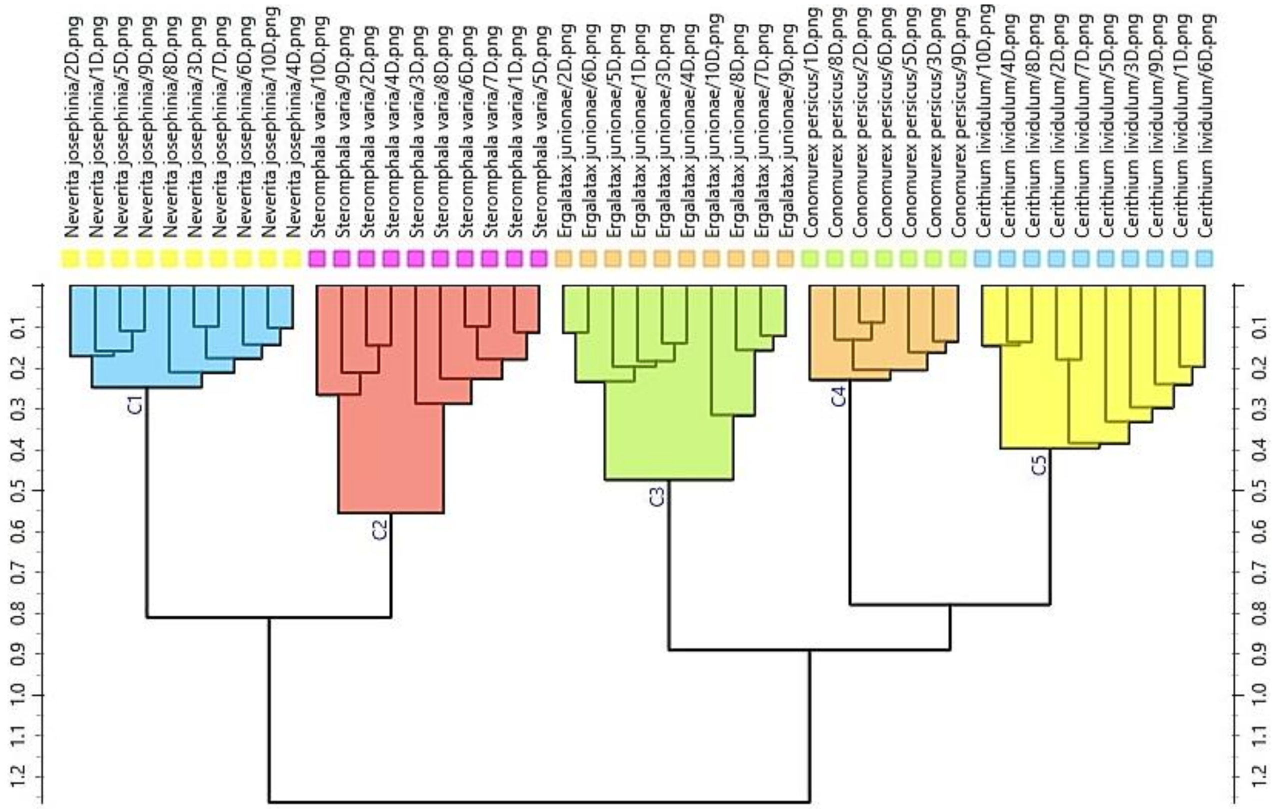

Figure 8. Library tree obtained from hierarchical clustering analysis of siphon images of species.

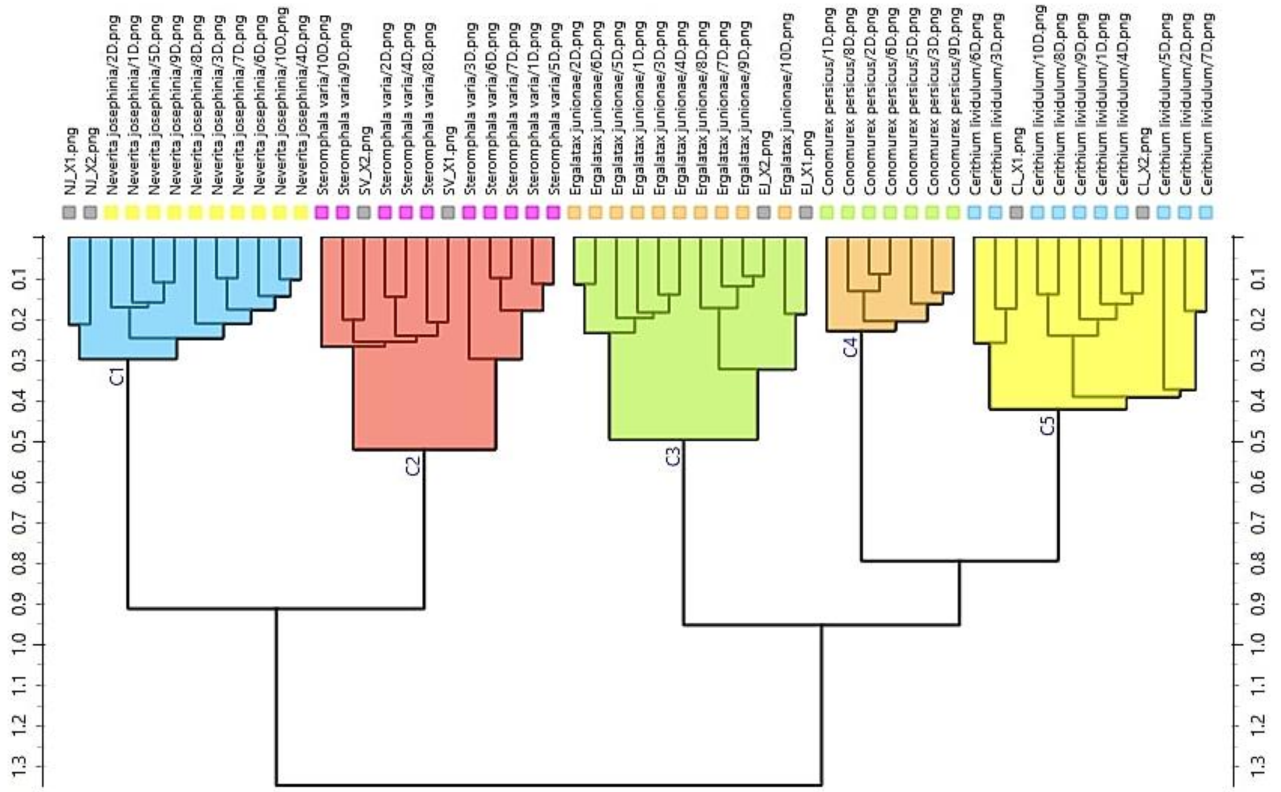

Figure 9. Tree obtained from identification analysis of siphon images of the species

The apex library was obtained from fewer individuals than the dorsal and ventral libraries. While creating this library, it was observed that individuals belonging to CP, CR and CL individuals were scattered under different species. The apex images of CP, CR and EJ individuals are very similar to each other. Within these three species, where color and pattern differences are more prominent than shape, a classification was made according to coloration. This is thought to be the reason for the confusion in the hierarchical clustering analysis. In line with the results obtained in the confusion matrix analysis, the confusion was eliminated by removing 3 individuals of each species from the analysis. When the species identification analysis was performed through the apex library, it was seen that SV-X3 was placed under the CR species. There is one hole in the apex image of SV-X3 individual. This hole is caused by octopus-like creatures that are drilled predators to eat gastropods. These creatures, which are predators for gastropods, make a hole on the gastropod shell. They then secrete a poison into the shell, killing the gastropod and thus preventing it from attaching to the shell. They then consume the creature they extract from the shell as food (Gordillo & Archuby, 2012). Such holes can be perceived as different structures during image processing and can cause confusion in species classification. When a third individual of the SV species without a hole was included in the species identification analysis and the SV-X3 individual with a hole was removed from the analysis, the confusion was eliminated and all individuals were clearly separated from each other and clustered under their own species.

The siphon library showed the highest level of confusion compared to the other three libraries. As a result of the hierarchical clustering analysis, the individuals belonging to the CR species are distributed in quite different places on the tree. Siphon images of CR species are quite like CP species in terms of color and pattern. In terms of siphon structure, it is also similar to CP species. For this reason, it is thought that these two similarities prevent a clear distinction during classification. As a result of the confusion matrix analysis, it was determined that all CR individuals had similar confusion matrix results and the CR species was completely removed from the library. After the CR species was removed, all species in the library were separated from each other and the confusion was eliminated. Since the CR species was not included in the siphon library, species identification analysis could not be performed for the CR species. In the species identification analysis performed on the remaining 5 species, it was observed that CP-X1 and CP-X2 individuals were clustered under the EJ species. It was observed that CP and EJ species exhibited a more protruding structure in siphon images compared to other species. In addition, it is believed that the reason for the separation from the CL species, in which a protruding structure is also observed, is since the siphon mouth structures are more similar in CP and EJ species. As a result of the removal of CP-X1 and CP-X2 individuals from the analyses, it was observed that a successful species identification was made.

There are various studies in literature on classification by image processing. However, studies on the classification of Gastropods are quite limited. In the time series based gastropod classification study by Onpans et al. (2018) 33 individuals belonging to a total of 5 gastropod families were photographed from a single angle on a black background and classified at the family level. It was shown that centroid based distance was more effective than center point-based distance in the classification. In addition, misclassified specimens were identified using confusion matrix. The study showed that gastropods can be classified by this method. In the study by Leow et al. (2015) an automated method was tested using digital image processing and artificial neural network (ANN) methods for the classification of copepods. As a result of the study, classification was performed using ANN and species separation was realized at a rate of 93.13%. In the study by Wen et al. (2009) local feature based identification and classification of insects were performed. A total of 6 different classification methods (minimum least square linear classifier [MLSLC], K nearest neighbor classifier [KNNC], Parzen density based linear classifier [PDLC], principal component analysis expansion linear classifier [PCALC], nearest mean classifier [NMC], and support vector machine [SVM]) were tested. The strongest of these methods was NMC with 89.5% accuracy, while the weakest was KNNC with 77.4% accuracy. Andayani et al. (2019) performed classification with Probabilistic using Neural Network method over 3 species from the Scombridae family. The study was conducted with 141 photos (112 of data training and 29 of data testing) and species identification was performed with an accuracy of 89.65%. One other study carried out by Yousef Kalafi et al. (2016) focused on monogeneans, which are parasitic flatworms. Researchers classified the species using KNN method with a 90% accuracy.

As a result, in this study, it was observed that the dorsal library was able to identify species with the highest accuracy (100%), followed by the libraries created with vental (98%), apex (93%) and siphon (92%) angles, respectively. The successful classification of species in clustering analysis and obtaining high accuracy rates demonstrate that image processing techniques can successfully identify gastropod species based on shell morphology.

Conclusion

Presented study used visual records of 60 individuals of 6 different Gastropod species taken from 4 different angles were evaluated. Using image processing, the images were converted into quantitative data and species-level classification was performed. Misclassified specimens were identified using confusion matrix and the individuals causing confusion were removed. The results obtained show that this approach is an effective method for the identification and classification of gastropod species.

The main purpose of the preliminary study presented here is to develop a methodology integrated with current technological developments by gradually expanding its scope. However, in future studies, it is very important to create image libraries that will reflect academic knowledge for a more effective identification. With future studies, it is thought that a Citizen Science application platform can be developed by minimizing the costs and time for morphological species identification and expanding the area evaluated in parallel. In this way, it will be possible to create a database on the species that the end user visually records (photo and/or video) with his/her cell phone camera and to expand the library and make it universal.

Ethical Statement

Ethics committee approval is not required for this study.

Funding Information

There is no institution or organization funding this study.

Author Contribution

First Author: Conceptualization, Data Curation, Formal Analysis, Visualization, Writing -original draft; Second Author: Supervision, Methodology, Formal Analysis, Investigation, and Writing -review and editing.

Conflict of Interest

The authors declare that they have no known competing financial or non-financial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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