



Determination of Most Reliable Bony Structure For Ageing of *Carassius carassius* (Linnaeus, 1758) Inhabiting In Duhok River (Iraq)

Jihad Saleem Mohammed¹ and Serap Saler^{1*}

¹Fisheries Faculty, Firat University, Elazığ, Turkey.

Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

In this study, it was aimed to make a comparative age determination in the population of *Carassius carassius* (Linnaeus, 1758) living in Duhok River. For this purpose, 71 fish samples (57 male and 14 female) were obtained. The bony structures vertebrae, scales, otoliths and operculum used in the age determination. The age groups determined for each bony structure. The number and percentage distributions of individuals in these groups were expressed with tables. The comparison of age matching between bony structures was expressed by figures. The most reliable bony structures were found as vertebrae, otolith, scales and operculum respectively. The highest consistency was observed in the vertebrae (70.42%). The ages changed between I and IX age in *Carassius carassius* in this study.

Keywords: Age determination; *carassius carassius*; bony structures; Duhok river.

1. INTRODUCTION

Age information is one of the most important variables to be determined in fisheries biology

studies. In order to understand the vital characteristics of fish species and populations (longevity, age of participation in stock, sexual maturity age, reproductive periods, migration,

*Corresponding author: Email: serapsaler@gmail.com;

mortality rates, etc.), data on age and growth are needed. Therefore, age determination should be done in a healthy way [1]. Studies on age growth have offered avenues for fisheries management problem resolution. Das [2] is in agreement that with fish age information, it is possible to derive first fish age maturity, growth estimation, population dynamics comprehension, and harvesting time optimization. Fish age knowledge is a valuable asset in the study of the fish population defining features and it may be a necessary to undertaking detailed research in the ecology and life history strategies. Knowledge about age is the foundation of all mortality, productivity, and growth rate calculations; thereby making it a crucial biological variable. Biological history calculations need age knowledge.

Carassius carassius is a freshwater cyprinid that is widely distributed in Eurasia: Spain across Europe, and north-central Asia to Northern China [3]. It has been successfully introduced into fresh waters throughout the World. It occurs in shallow ponds, lakes rich in vegetation and slow moving rivers. It burrows in mud in the dry season or winter. Since it is a fast growing and hardy fish that can withstand adverse environmental conditions, is widely recognized to be resistant to some ecological conditions and is known for its morphological plasticity [4]. Although there are some studies on age determination of *Carassius* species, no research on comparative age determination of *Carassius carassius* has been found.

In Eğirdir and Bafra Fish Lakes, vertebra was found to be the reliable bony structure for Bafra

Fish Lake population and otolith for Eğirdir Lake of the Crucian Carp (*Carassius carassius* L., 1758). In this study age determination has been made by using scales and the age distribution of these fish were found to be between III-VII years [5].

Şası [6] determined growth and condition of invasive species Prussian carp, *carassius gibelio* (Bloch, 1782) in a dam lakes from Büyük Menderes Basin and found the age compositions of specimens between III-VI years.

In Iraq some researches about *Carassius* species have been made as population status of gold fish *Carassius auratus* [7,8].

No previous research has been found for determination of most reliable bony structure for ageing of *Carassius carassius* in Duhok River. On the other hand, no study has been conducted on the Duhok River population of the species. In this study, age data obtained from five different bony formations were evaluated in order to determine the most appropriate bony structure for age determination of *Carassius carassius* in Duhok River and the harmony of bony structures with each other was compared.

2. MATERIALS AND METHODS

2.1 Description of the Sampling Sites

In total, 71 *Carassius carassius* samples were captured from Duhok River, 36°45'38.88" N, 42°48'11.12" E (in Northern Part of Iraq) Fig. 1 between November 2017 and March 2018.

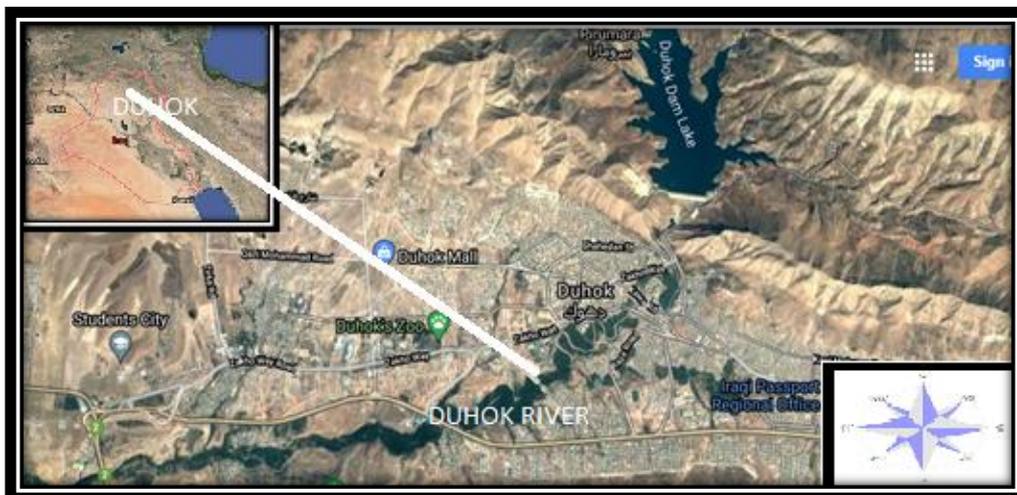


Fig. 1. Sampling stations in Duhok River, Northern Iraq

The samples were caught using gill nets of multiple sizes (36, 40, 44, 50, 60, 70, 80, 90 mm). The mesh sizes of the trammel nets were 100, 120, 140 and 160 mm.

2.2 Biological Features

Body of the species laterally compressed and thick, round abdomen; standard length 2.1-2.8 times body height and 3.1-4.1 times head length; length of head larger than body height; small and short head; short snout; terminal mouth and arch shaped; lower jaw slanting upwards slightly; thick lip; no palpus; 37-54 gill rakes on the first outer gill arch, gill rake long; one row of pharyngeal teeth on each side, laterally compressed, formula 4-4; 27-30 scales in lateral line. Tip of pectoral fin can reach the base of ventral fin; dorsal fin ray: 3, 15-19; pectoral fin ray: 1, 16-17; ventral fin ray: 1, 8; anal fin ray: 3, 5; body colour: silvery grey, deeper colour in dorsal portion, lighter colour in abdomen; colour of fins: greyish [9].

2.3 Taking of Bony Structures

Scales, vertebrae, operculum, otoliths (lapillus and lagenar asteriscus) were taken from each fish for age determination. Preparation of bony structures for age determination was made according to Chugunova [10].

The scales were taken from the area between the lateral line and the dorsal fin on the left side of the fish. The scales of each fish were placed in petri dishes and warm water was added to them, the scales were separated from each other with the help of two forceps, and the scales were kept for 10-12 hours. At the end of the waiting period, the pigment layer on the flakes was cleaned with the help of a brush. and kept in a 3% NaOH solution until they were cleaned and turned into a preparation by passing through 96% alcohol. 4-10th vertebrae were removed from each sample and kept in hot water and removed from meat, skin, etc. For detailed cleaning, fine-tipped forceps were used and the vertebrae after the cleaning process were completed were left to dry. The otoliths were taken from the right and left sides of the head of the samples and cleaned in alcohol. The opercules extracted from both parts of the head were treated with hot water and cleared of residue. The vertebra, otolith and opercules were dried in an oven at 103 ° C and made ready for examination.

All bony structures were subjected to preliminary examination. Age readings were repeated 3

times at 10x magnification by the same reader, at different times, under a stereo-binocular microscope.

As well as whole the structure after process of washing, cleaning and drying has been preserved in a referenced packet and then recorded on this packet about date, number and name of the structure until reading under microscope.

In order to determine the most appropriate bony structure in age readings, mean age, percent agreement (PA), Average Percent Error (MAPE), and Coefficient of Variation (CV) were calculated using the relevant literature [11-14].

3. RESULTS AND DISCUSSION

In this study, a total of 71 *Carassius carassius* samples from Duhok River were determined. Age determinations were made from 4 bony structures of these obtained fish. The dorsal fin bone was thin and fractured and was not used for age determination.

Distribution of bony structures ages according to age groups, average ages in bony structures, Precision of age estimates obtained from readings of three readers have been obtained from the data and given in Table 1.

In operculum the highest age group was determined with 16 samples as 22.53% in V age group, in otolith in 17 samples with 23.94% in II age group, in scale 14 samples with 19.73% in VI age group and in vertebrae in 18 samples with 23.58% II age group. Minimum, maximum and average values of total length and weight, Percent Agreement (N %) of the bony structures, average ages in bony structures of *C. carassius* in Duhok River have been determined and given in Tables 2, 3 and 4.

In *C. carassius* the average total length was determined as 37.42 cm. Its minimum and maximum values were changed between 22.20-70.00 cm. The highest weight of fish sample was recorded as 6870 g, minimum value as 176 g (Table 2).

When the age adjustment of *C. carassius* bony structures was examined, it was found that the highest adaptation of the reader was in the vertebrae with 70.42% and then in the otoliths with 66.19 %. The lowest adjustment was recorded in operculum with 43.66% (Table 3).

Table 1. Distribution of bony structures ages according to age groups in *C. carassius* in Duhok

	Age groups																		
	I		II		III		IV		V		VI		VII		VIII		IX		N
Bony structures	N	N%	N	N%	N	N%	N	N%	N	N%	N	N%	N	N%	N	N%	N	N%	
Operculum	3	4.21	11	15.50	9	12.67	11	15.50	16	22.53	15	21.12	4	5.64	2	2.82	0	0	71
Otolith	2	2.81	17	23.94	10	14.08	13	18.31	14	19.72	12	16.91	1	1.41	2	2.82	0	0	71
Scale	3	4.21	12	16.90	9	12.67	11	15.50	15	21.12	14	19.73	4	5.64	2	2.82	1	1.41	71
Vertebrae	5	7.04	18	25.35	12	16.90	11	15.50	13	18.31	9	12.65	1	1.41	1	1.41	1	1.41	71

Table 2. Minimum, maximum and average values of total length and weight of *C. carassius* in Duhok River

	Total length(cm)	Fork length (cm)	Standard length (cm)	Weight (g)
Average	37.42	33.91	31.53	1141.33
Minimum	22.20	26.00	24.00	176
Maximum	70.00	65.00	58.60	6870
N	71	71	71	71

Table 3. Percent agreement (N%) of the bony structures of *C. carassius* in Duhok River

Compared bony structures	Compliance groups					
	3/3	N	2/3	N	1/3	Total
Operculum	43.66	31	56.34	40	-	100
Otolith	66.19	47	33.81	24	-	100
Scale	50.70	36	49.30	35	-	100
Vertebra	70.42	50	29.58	21	-	100

Table 4. Average ages in bony structures of *C. carassius*

Bony Structures	N	AG	SE
Vertebra	71	4.12	0.16
Otolith	71	3.32	0.10
Scale	71	4.23	0.16
Operculum	71	4.15	0.15

N: Number of fish, AG: Average age SE: Standard error

Table 5. Precision of age estimates obtained from readings of three readers

Bony Structures	N	PA	(APE) (±SE)	CV(±SE)
Vertebra	71	70.42	11.92 (±0.86)	24.49(±1.01)
Otolith	71	66.19	12.05(±0.49)	25.62(±1.04)
Scale	71	50.70	16.26(±0.59)	34.48(±1.25)
Operculum	71	43.66	17.38(±0.51)	44.70(±1.71)

N: Sample size, PA: Percent agreement, APE: Average percent error, CV: Coefficient of variation, SE: Standard error

The highest percent agreement was found in the vertebra (70.42) followed by otolith, scale and operculum respectively (Table 5). In addition, APE (Average Percent Error) and CV (Coefficient of Variation) were also calculated for the four bony structures. In the vertebra, these two data were calculated lower than in other bony structures. When the vertebrae are considered in terms of percent agreement, APE and CV, it has been found that PA is higher than other structures, APE and CV is lower. This shows that the vertebrae are more effective in reading age compared to other bony structures.

The relationship of each bony structure with the other bony structure have been observed and given in graphics (Fig. 2).

Fig. 2 shows that for *C. carassius*, the age relationship between the otolith and the

vertebrae was; without age difference is 50, the number of samples with 1 age difference is 19, and the number of samples with 2 age difference is 2.

Variations in the age estimates in diverse aging structures were examined. In this study, variations were observed in the age estimates from different aging structures. For example, an analysis of the vertebrae indicated rings when compared with other structures. A number of researchers have raised doubt about the accuracy of using scales to estimate the age of fish [15]. This is because of reabsorption and deposition of false annuli as a result of stress. Also, the clarity of the annuli can be affected when the fish gets older. In *C. carassius* scales are big. But a problem occurs during the age reading because of false and half ring age rings on the scale. In this study due to the large size of

the scales in *C. carassius*, the samples which were not observed as a whole in the field of view of the microscope were determined with the help of a magnifying glass. In addition, deterioration was observed in the center. However, a large number of false rings were found.

In this research the age of fish samples were recorded between I and IX age groups. The highest age distribution has been determined between II and VI age groups. The highest age

was as IX age in scales and vertebrae. The highest number of age group was recorded in 17 samples as II age. In Eğirdir Lake the highest age of *Carassius auratus* has been determined as V age. Average length 23.77 ± 0.181 cm, length of female 24.96 ± 0.223 cm and length of male 23.05 ± 0.238 cm were found. Average weight 363.61 ± 12.861 g, weight of female 455.99 ± 9.103 g and weight of male 308.13 ± 17.159 g were found [16].

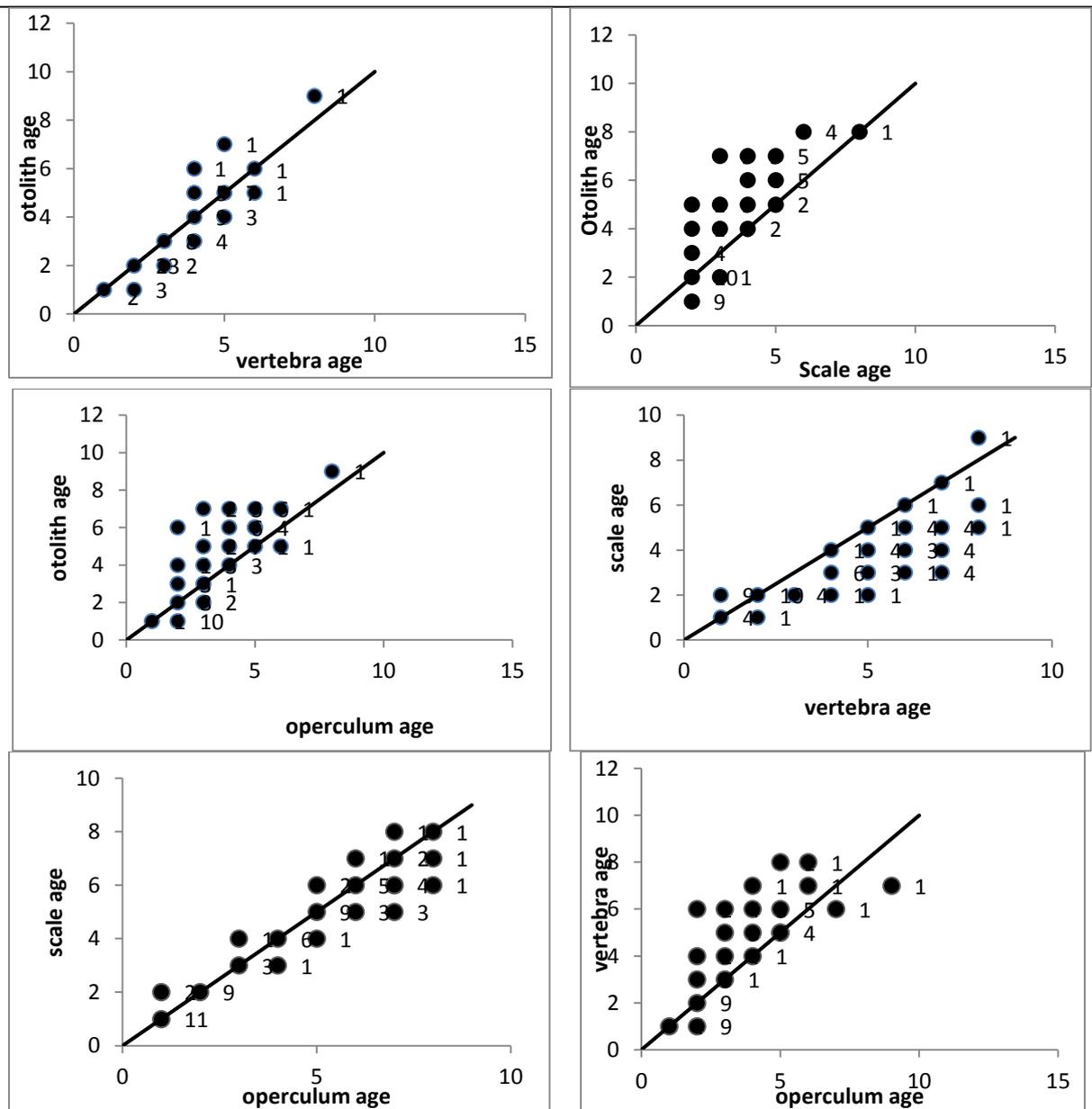


Fig. 2. Comparison of otolith and vertebra age(a), otolith and scale age (b), otolith and operculum age (c), scale and vertebra age (d), scale and operculum age (e), vertebra and operculum age (f) in population of *C. carassius* in Duhok River

All-Norr [7] studied the population status of gold fish *Carassius auratus* in Iraq He determined the highest age as X age. Şaşı [6]. found *Carassius gibelio* age between III- VI ages in Topçam Dam Lake. Moreover in Keban Dam Lake, Duman and Şen [17] studied the comparative age determination of *C. auratus*. They stated that the age changes between I and VII. They observed the most reliable bony structures in *C. auratus* as scale and recommended otolith and vertebrae after scale as the most reliable bony structure.

The evaluation of age and growth of fish using opercular bones is well-researched in fishes drawn from temperate waters. According to Nargis [18] the use of opercular bones is more accurate compared to other methods. In fish ageing structures, scales continue to be the most popular for moronid, esocids and centrarchids. The main reason for this popularity is because it is easy to remove them and such removal has little damage on the fish [19,20]. It was concluded that the operculum was not suitable for age determination due to the abundant number of pseudo-rings and the fact that age rings were not easily distinguished. However, age rings were sometimes not seen due to thickening with advancing age. It is also necessary to pay attention to the residence time of the boiling water when making it suitable for wet determination. If left too long, the age ring will deteriorate, if left too little, it is difficult to clean and time consuming.

In this study; although similar results were obtained in the age readings of the vertebrae and otolith, the most clear age rings were observed in the vertebrae. The risk of taking fish from the ring structures; it was concluded that bony formation was the vertebrae in age determination readings because it was clear, homogeneous and showed the most confirmation at the end of three readings.

There are many studies on determining the most reliable structure for age determination in fish. Some of those; in Keban Dam Lake Altinkaya Dam Lake vertebrae was found as the most suitable bony structure for *Capoeta capoeta umbla* [21] and *Copoeta tinca* [22]. Otoliths were reported as most suitable bony structure for *Chondrostoma regium* (Heckel, 1843) and *Chalcalburnus mossulensis* (Heckel, 1843) populations of in Keban Dam Lake [23]. Gümüş [24]. studied the accumulation tracking method in the bony structures of the mirror carp, emphasized that the false rings on the vertebrae of the mirror carp were low, the ring character

was very clear and clear and it was an extremely reliable structure in both validation and age determination studies. Aydın [25] stated that *Capoeta capoeta umbla* (Heckel, 1843) showed the clearest rings in otoliths. Öztürk et al. [26]. determined the best read bony structures for the same species as the otoliths in Lake Hazar. Yüce [27] found the most clear age in otoliths in the comparative age determination from bony structures of *Mystus halepensis* (Valenciennes, 1839) in the Keban Dam Lake. Additionally In Ladik Lake it was concluded that the most reliable bony structure for age determination in *Perca fluviatilis* is the vertebra [28].

It should be noted that reliable bony formation may change in populations of the same species living in habitats with different ecological characteristics. It has been found that the reliable bony structure for age determination varies in different populations of some species and the ring deposition has proved to be annual.

Bostancı [29] reported for *Carassius auratus gibelio* vertebra as the most reliable bony structure for Bafra Fish Lake population and sagitta for Eğirdir Lake. Bostancı et al [4]. determined the highest agreement (70.3%) and lowest CV (7.0%) were with otoliths for Eğirdir Lake specimens; in Bafra Fish Lake specimens, the highest agreement and lowest CV were with vertebrae 62.9% and 8.1%, respectively. For this reason, determining the bony formation from which age data required for fish biology studies should be taken always be the first step [30].

4. CONCLUSION

In the comparative age determination of *C. carassius* in Duhok River population, the vertebra and otolith gave similar results, but it was difficult to see the rings in the center due to thickening of the otolith at very advanced ages, and the most reliable bony structures were determined as vertebra, otolith, scales and operculum respectively. In Duhok River, the use of vertebra will be extremely important in terms of the accuracy and reliability of the results to be obtained in age determination studies that will be used for future biological and population dynamics researches related to *C. carassius*.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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