Research Article

Age and growth assessment of *Chondrostoma regium* (Heckel, 1843) (Teleostei: Cyprinidae) inhabiting the Zayandeh River (Iran) using different structures

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Abstract: This study investigated the age and growth of *Chondrostoma regium* in upstream of the Zayandeh River by using different calcified structures (i.e. scale, otolith and vertebra). Also, the potential variability of age and growth estimates derived from different structures and their competency for further applications in population and life history studies of this species were statistically evaluated and compared. Based on the aging performed by using different calcified structures, *C. regium* specimens composed of 4 age groups of 2 to 5 years. Considering all aging structures (i.e., scale, otolith and vertebra) the major proportion of the sample was those within the age groups of 3 and 4. The von Bertalanffy’s growth models of *C. regium* was fitted separately for the data derived from each of the aging structures. Among all the fitted models, the scale-based model showed the smallest measures of the information criteria with AIC=296.88 and SBC=300.86. Based on all the statistical indices for model comparison, the von Bertalanffy’s growth model using the direct scale-based observations showed the most reliability and the best performance for growth parameters estimation of *C. regium* in this study. Hence, for *C. regium* inhabiting upstream of the Zayandeh River the most statistically sound von Bertalanffy’s growth equation could be written as $L_t=303 \left(1-e^{-0.11 (t-0.34)}\right)$.

Given the importance of growth studies in effective management and conservation of native fish populations as well as considering the highly changing and under pressure environment of the Zayandeh River, our study attempted to provide updated information on growth characteristics of *C. regium* in this aquatic ecosystem. The results of this study may help to improve our understanding of its population dynamics and may provide help in designing better conservational plans for this native species.

Keywords: Aging, Bertalanffy models, Cypriniformes, Otoliths, Scales, Vertebra, Growth, Iran.

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Introduction

Identification of fish age structure is considered as a major prerequisite and baseline for studying their population dynamics, population assessment and effective management as well as fish stock conservation. Aging procedure in fish has been routinely based on distinguishing and counting visible yearly growth rings appeared on certain calcified/ossified structures of the body including scales, otoliths, vertebra, opercular bones and fin rays (Blackwell et al. 2016). Evaluation and comparison of age structures achieved from aging by using
different calcified structures has been conventionally considered as an efficient approach to discern the most appropriate structure to be used for aging of a given fish species (Khan & Khan 2009). The most appropriate and reliable ageing structure might differ amongst fish species, therefore, the assessment and comparison of the potential precision and accuracy of these structures (to be used for aging) could be crucial (Ma et al. 2010).

In the present study, as a preliminary attempt, the competency of various calcified structures (including scale, vertebra and otolith) for age determination and growth estimation in *Chondrostoma regium* (Heckel, 1843) inhabiting the Zayandeh River is statistically evaluated and compared. Zayandeh is the only permanent riverine system in central arid and semi-arid parts of Iran which has a major role in provision of crucial services (e.g. human consumption water, agronomic and animal food products, industrial factories, etc.) for the population residing alongside this river and also it supports aquatic living resources. During the last decades, this unique aquatic environment as well as its biotic components have suffered from numerous ecological threats including industrial contamination (Varnosfaderany et al. 2009; Bateni et al. 2013; Gilannejad et al. 2016), overharvesting of water, environmental destruction, contemporary impacts of climate changes, declining of precipitation and droughts. *Chondrostoma regium* (locally named “Nazak”) is one the inhabitant fish in this ecosystem with an omnivorous feeding regime (Mahboobi-Soofiani et al. 2014). The population of this species exhibits a relatively wide-ranging dispersion along Zayandeh riverine ecosystem, but still there seems to be a lack of sufficient available information on various aspects of its biology and ecology (Mahboobi-Soofiani et al. 2014) which appears to be the key prerequisite for its population assessment as well as its stock conservation in the river. Also, more recent studies by Keyvani et al. (2016) and Kiani et al. (2018) provided more detailed information on age and growth of *C. regium* in the Beheshtabad River (in Chaharmahal & Bakhtiar Province of Iran) and Bibi-sayyedian river (in Isfahan Province of Iran), respectively. The main objectives of our study were (1) investigation of age and growth of *C. regium* in upstream of the Zayandeh River by using different calcified structures (2) statistical evaluation of variability of age and growth estimates derived from different structures and comparison of their competency for further applications in population and life history studies of this species.

### Materials and Methods

In March and April 2017, specimens of *C. regium* (total of 63 fish) were captured using a riverine seine in upper parts of the Zayandeh River (Fig. 1). Standard lengths and weights of *C. regium* samples were recorded at the 0.1mm and 0.01g accuracy, respectively, and initially the length-weight relationship (LWR) was assessed using the conventional equation \( W = aL^b \). Aging structures including scales (taken from above the lateral line), otoliths and vertebra were removed from each individual specimen, afterwards they were cleaned, rinsed (in case of vertebra with using boiling water) and dried to be used in further age determination. Age readings for each calcified structure were achieved independently by counting annual growth increment rings using an optical microscope. Only the specimens with the readable and assignable ages in all structures (i.e. scale, otolith and vertebra) were considered for further growth analysis (overall of 54 fish). For the back-calculation procedure, we measured and recorded the total scales radius and the distances from the center to each of the growth rings by using an ocular micrometer. Back-calculation procedure was performed according to the well-known Fraser-Lee formula (as quoted in Francis 1990):

\[
L_i = C + (L_c - C)(S_i / S_c)
\]

Where, \( L_i \) is the length at the time when growth ring \( i \) was appeared, \( L_c \) is the length of fish at the time of sampling, \( S_i \) is the observed distances between the center to the growth ring \( i \), \( S_c \) is the recorded total scale radius and \( C \) is the intercept parameter derived
The von Bertalanffy’s growth analysis model was used to determine growth parameters for each of the aging structures, separately. The classical von Bertalanffy’s growth model was fitted to the observed length-at-age data for each aging structure:

\[ L_t = L_\infty(1 - e^{-kt}) \]

In the von Bertalanffy’s growth model, parameters of \( L_t \), \( L_\infty \), \( k \), \( t \) and \( t_0 \) are the length at age (time), the asymptotic length, body growth rate coefficient, age and the hypothetical average fish age at zero length (also known as the initial condition parameter), respectively. The von Bertalanffy’s growth parameters of \( C. \) regium (i.e. \( k \), \( L_\infty \) and \( t_0 \)) were estimated by the application of NLIN procedure of SAS software along with the iterative Levenberg–Marquardt algorithm. For evaluation and better comparison of the precision and competency of different growth models fitted based on the scale, otolith and vertebra aging data, not only the conventional quality-of-fit measure (R^2) but also Akaike’s Information Criterion (AIC) and Schwarz/Bayes Criterion (SBC) were also computed and considered in our study (Schwarz 1978; Akaike 2011). AIC and SBC can help to have a better and more precise comparison especially when comparing several different regression models (finally the best and most parsimonious model will be chosen based on the minimum AIC and SBC).

**Results**

Based on the aging performed by using different calcified structures, \( C. \) regium specimens in this study composed of 4 age groups of 2 to 5 years. Considering all aging structures (i.e., scale, otolith and vertebra) the major proportion of the sample was those within age groups of 3 and 4. Descriptive statistics of the observed ages (based on counting the annual growth rings on different aging structures) and standard lengths of \( C. \) regium specimens are given in Table 1 (as fish grew older, standard lengths showed a clear increasing trend in all aging structures). The variability of \( C. \) regium standard lengths for each age class based on the scale, otolith and vertebra aging observations were also illustrated in Figure 2. The observed standard length and weight relationship of \( C. \) regium samples collected for this study was described as \( W = 0.009L^{3.21} \) (Fig. 3).
Table 1. Descriptive statistics of the observed ages (based on counting the annual growth rings on scales, vertebra and otolith) and standard lengths of *Chondrostoma regium* in the Zayandeh River.

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Scale</th>
<th>Vertebral</th>
<th>Otolith</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>Mean±SD (mm)</td>
<td>n</td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>71.9±14.0</td>
<td>10</td>
</tr>
<tr>
<td>3</td>
<td>24</td>
<td>92.7±15.4</td>
<td>18</td>
</tr>
<tr>
<td>4</td>
<td>16</td>
<td>118.1±17.7</td>
<td>18</td>
</tr>
<tr>
<td>5</td>
<td>10</td>
<td>134.4±12.2</td>
<td>8</td>
</tr>
</tbody>
</table>

**Fig. 2.** Boxplots of *Chondrostoma regium* standard lengths showing its age variability using different aging structures, including scales (a), otolith (b) and vertebra (c).

**Fig. 3.** The observed standard length and weight relationship of *Chondrostoma regium* samples in the Zayandeh River.

Prior to the back-calculation procedure, the potential relationship between the scales radius and the standard length and weight were assessed. These linear relationships for *C. regium* are presented in Figure 4. The results of linear regression revealed statistically significant positive relationships between the variability of the scales radius and the variabilities of the standard length and weight ($R^2=0.70$, $P<0.05$ and $R^2=0.32$, $P<0.05$ for the length and weight, respectively) of *C. regium*. However, the observed relationship between the scales radius and the standard length was more than twice as strong as that of between the scales radius and the weight (Fig. 4).

Table 2 shows the back-calculated mean standard length-at-age data for *C. regium* samples in the study area. The standard lengths derived for each age class from the back-calculation procedure indicated a
The von Bertalanffy's growth models of *C. regium* was fitted separately for the data derived from each of the aging structures (i.e. scale, otolith and vertebra) as well as for the derived data from the back-calculation procedure. Table 3 shows the estimated growth parameters (i.e. $k$, $L_\infty$ and $t_0$) obtained from the von Bertalanffy’s growth models using different input data. Performance of all the fitted growth models were statistically compared using $R^2$, AIC and SBC measures (Table 3). The model using otolith as the aging structure showed the lower estimates of $L_\infty$ and the higher estimates of $k$ than other models. However, the lowest measure of $t_0$ was estimated in the growth model with the data derived from vertebra-based aging. Statistical comparison among different growth models showed the better quality-of-fit in case of scales and the back-calculated data ($R^2=0.61$). Among all the fitted models based on different aging structures, the scale-based model showed the smallest measures of the information criteria with AIC=296.88 and SBC=300.86. Based on all the statistical indices for model comparison (i.e., $R^2$, AIC and SBC), the von Bertalanffy’s growth model using the direct scale-based observations (as the input data) showed the most reliability and the best performance for growth parameters estimation in this study. Hence, for *C. regium* inhabiting upstream of the Zayandeh River, strong conformity and robust agreement with the directly observed standard lengths ($R^2=0.94$, $P<0.05$ as graphically represented in Figure 5).

Table 2. The back-calculated mean standard length-at-age (mm) of *Chondrostoma regium* in upstream of the Zayandeh River.

<table>
<thead>
<tr>
<th>Age (year)</th>
<th>$L_1$</th>
<th>$L_2$</th>
<th>$L_3$</th>
<th>$L_4$</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>55.9</td>
<td>64.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>59.5</td>
<td>71.5</td>
<td>85.4</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>63.4</td>
<td>75.2</td>
<td>92.0</td>
<td>107.8</td>
</tr>
<tr>
<td>5</td>
<td>63.4</td>
<td>75.9</td>
<td>92.0</td>
<td>107.8</td>
</tr>
</tbody>
</table>

Table 3. Statistical comparison of the estimated growth parameters using different aging structures for *Chondrostoma regium* in the Zayandeh River.

<table>
<thead>
<tr>
<th></th>
<th>Scale</th>
<th>Vertebra</th>
<th>Otolith</th>
<th>Back-calculation</th>
</tr>
</thead>
<tbody>
<tr>
<td>$L_\infty$</td>
<td>303.3</td>
<td>283.5</td>
<td>273.4</td>
<td>306.2</td>
</tr>
<tr>
<td>$k$</td>
<td>0.11</td>
<td>0.09</td>
<td>0.13</td>
<td>0.10</td>
</tr>
<tr>
<td>$t_0$</td>
<td>- 0.34</td>
<td>- 1.53</td>
<td>- 0.41</td>
<td>- 0.40</td>
</tr>
<tr>
<td>$R^2$</td>
<td>0.61</td>
<td>0.50</td>
<td>0.53</td>
<td>0.61</td>
</tr>
<tr>
<td>AIC</td>
<td>296.88</td>
<td>310.39</td>
<td>307.38</td>
<td>318.99</td>
</tr>
<tr>
<td>SBC</td>
<td>300.86</td>
<td>314.37</td>
<td>311.36</td>
<td>323.18</td>
</tr>
</tbody>
</table>

Fig. 4. The observed relationships between the scales radius and the standard length (a) and weight (b) of *Chondrostoma regium* in the Zayandeh River.
The most statistically sound von Bertalanffy’s growth equation could be written as $L_t = 303 (1 - e^{-0.11(t - (-0.34))})$.

**Discussion**

To our knowledge, this is the first study to assess the competency of different aging structures for growth analysis in *C. regium* in the study area. The observed variability of age estimates using scales, otolith and vertebra revealed that the measures of *C. regium* growth parameters can be influenced by the applied aging structure. Therefore, further research to validate the precision of age determination in *C. regium* are essentially required for acquiring a better perception of its population dynamics in the highly under pressure ecosystem of the Zayandeh River. Based on the statistical indices, our study suggested that implementing the otolith- and vertebra-derived age estimates in growth model of *C. regium* leads to a poorer model performance comparing to the application of scale-based age estimates; thus, among various aging structures, scales could be considered to be the most appropriate calcified structure in age estimation studies of *C. regium* inhabiting the Zayandeh ecosystem. For many cyprinids, historically age has been determined using scales and in several species such as *Hypophthalmichthys molitrix, Labeo rohita, Channa marulius, Cyprinus carpio* and *Prochilodus nigricans* scales were found to be the most suitable aging structure (Johal et al. 2001; Khan & Khan 2009).

Given the importance of growth studies in effective management and conservation of native fish populations as well as considering the highly changing and under pressure environment of Zayandeh River, providing updated information on growth characteristics of *C. regium* in this aquatic ecosystem may improve our understanding of its population dynamics and help to design better conservational plans for this species. Hence, our research also attempted to contribute the basic updated key information required to better perception of the population dynamics of *C. regium* in the study area. The estimated growth parameters of *C. regium* sampled from Zayandeh River in 2007-2008 reported by Mahboobi-Soofiani et al. (2014) were 0.21 for $k$, 246-253 mm for $L_\infty$ (based on the fork length) and 0.03-0.18 for $t_0$. The $b$ exponent of the length-weight relationship in their study were found to be less than 3. Apart from the usual technical issues and the employed methods regarding the sampling procedures and the age estimation, the reasons for the observed differences between the growth characteristics of *C. regium* in their study and the current research might be associated to inter-annual environmental condition changes in the area. The estimated growth parameters of *C. regium* sampled from the Beheshtabad River (in Chaharmahal & Bakhtiari Province of Iran) during 2013-2014 reported by Keivany et al. (2018) were 0.15-0.27 for $k$, 262-319 mm for $L_\infty$ (based on the total length) and -0.48 to -2.07 for $t_0$. The $b$ exponent of the length-weight relationship in their study were found to be more than 3. Also, the measured growth parameters of *C. regium* sampled from the Bibi-sayyedan River (Tigris basin) in Isfahan Province of Iran) in 2010-2011 described by Keivany et al. (2018) were 0.20-0.28 for $k$, 189-238 mm for $L_\infty$ (based on the total length) and -0.54 to -0.58 for $t_0$. The $b$ exponent of the length-weight relationship in their study were found to be more than 3.

Alterations in local ambient conditions of...
ecosystems may influence the overall biological performance (e.g., growth) of aquatic organisms either directly (i.e., via physiological and behavioural processes) or indirectly (i.e., through intermediate or transitional ecological processes such as food web interactions). Considering the current critical condition of the Zayande River, annual surveys and monitoring of key life-history traits of *C. regium* seems to be of crucial importance and should be encouraged. This may help to have a better understating of the potential impacts of ongoing environmental changes on ecological components of this ecosystem.

**Acknowledgments**

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**References**


مقاله پژوهشی

تأثیر ارزیابی سن و رشد ماهی نازک (Chondrostoma regium (Heckel, 1843)) در رودخانه زاینده رود با استفاده از ساختارهای مختلف تعیین سن

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چکیده: در این مطالعه به ارزیابی سن و پارامترهای رشد ماهی نازک در حوضه رودخانه زاینده رود با استفاده از ساختارهای تعیین سن مختلف (فلس، اتولیت و مهره) پرداخته شده است. همچنین تغییرات سنی و پارامترهای رشد محاسبه شده حاصل از ساختارهای سنی متفاوت به صورت آماری ارزیابی و با یکدیگر مقایسه شدند. بر اساس ساختارهای تعیین سن، ماهیان نمونه‌داری شده در این تحقیق شامل 4 گروه سنی 2 تا 5 ساله بودند که بخش عمده آنها به گروه‌های سنی 4 تا 5 ساله تعلق داشتند. مدل رشد بر پایه فلس پایین‌ترین مقادیر معیارهای آماری (AIC=296.88 و SBC=300.86) را نشان داد. بر اساس تمام شاخصهای آماری مورد استفاده در این پژوهش، مدل رشد بر پایه اطلاعات ورودی از تعیین سن فلس بالاترین اطمینان و سطح عملکرد کیفی را برای استفاده در محاسبات مربوط به رشد ماهی نازک در منطقه مورد مطالعه دارا بود. بر این اساس، معمولاً رشد بر پایه فلس بالاترین اطلاعات ورودی از تعیین سن فلس بالاترین اطلاعات از مدتی به سبب اهمیت مطالعات رشد در مدیریت و حفاظت ماهیان بومی و همچنین در نظر گرفتن شرایط زیست محیطی بسیار متغیر و تحت فشار زاینده رود، پژوهش اخیر گوشیده است که اطلاعات به روزی از خصوصیات رشد ماهی نازک در این اکوسیستم بیش از افراد آورده. نتایج این مطالعه می‌تواند در درک بهتر پویایی جمعیت ماهی نازک و همچنین طراحی بهتر برنامه‌های حفاظتی این گونه در منطقه مفید باشد.

کلمات کلیدی: تعیین سن، رشد، کپورماهی شکل‌ان، مدل برپانی، اتولیت، مهره، فلس، رشد، ایران.