Body Size and Age
Assessment among Advertising Male Chorus Frogs

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Calling males in breeding choruses of Pseudacris maculata and P. triseriata are often flanked by one or two smaller, non-calling males (J. Platz, pers. obs.). It is often assumed that larger individuals are older because amphibians have indeterminate growth. If the body size-age correlation is true, then the inference from our original observations above is that satellite males should be younger than calling males. Skeletochronology as a technique for determining age has in the last decade or so been successfully applied to a limited number of anuran taxa: Hemelaar and Van Gelder (1980) and more recently Höglund and Säterberg (1989) for Bufo bufo; Lykens and Forester (1987) for P. crucifer; Leclair and Castanet (1987) for Rana pipiens; and Gibbons and McCarthy (1984) for R. temporaria. The present study was undertaken to determine if skeletochronology techniques could be applied to male chorus frogs, and if so, to determine whether body size is correlated with age well enough to allow aging of males based on snout-vent length (SVL), thus avoiding the need to kill individuals. This work is part of a larger ongoing project involving a similar assessment of the body size-age relationship between calling males and attending satellites.

All specimens were calling males collected in April and May of 1980. Locales included two species (populations 1–4 represent the Boreal chorus frog, Pseudacris maculata, and 5–6 the Western chorus frog, P. triseriata) as follows: South Dakota, (1) Marsh Lake, Hamlin County, (2) Springfield, Bon Homme Co.; Nebraska, (3) Dodge Park, Omaha, Douglas Co., (4) Cozad, Dawson Co., (5) Ogallala, Keith Co.; Kansas, (6) Arkansas City, Cowley Co. Individuals were preserved in 10% formalin and transferred to 40% isopropyl alcohol and stored until sectioned. SVLs were obtained to the nearest 0.01 mm with dial calipers.

Bone sectioning and staining procedures followed Hemelaar and Van Gelder (1980) with the exception of their use of 70% ethanol for temporary storage of bone samples. Femurs were removed from the frogs and decalcified in 5% nitric acid for 3 h and left in water overnight. The diaphysis was cut from each femur and sectioned on a Spencer model 880 freeze microtome. A series of mid-diaphyseal sections 15 to 20 microns thick were mounted on glass slides treated with an albumen-glycerin preparation, containing equal proportions of egg white and glycerin with thymol added. All preparations were then stained in Cabisco’s Delafeld hematoxylin for 15 min and washed in tap water for an equivalent period. Stained slides were examined and photographed at 125× under a compound light microscope.

Stained sections (Fig. 1) showed a series of narrow, concentric, hematoxylinophilic rings separated by wider zones of paler background material. The narrower, dark-staining rings are interpreted to be the result of climatically-imposed winter periods of arrested growth or “resting lines” (RL). Intervening, broader, pale zones are interpreted to represent active bone deposition during one growing season. Because this is an annual phenomenon, a wide zone and subsequent RL are interpreted to represent one year. On occasion (Fig. 1) a pair of very closely spaced dark concentric rings occurred. According to Klevezal and Kleinenberg (1967), double lines are caused by interruptions in hibernation and therefore we regarded these as equivalent to one season. Because all frogs were collected in April and May, the outer bone perimeter was counted as an RL for bone growth, which is consistent with observations by Smirina (1972) indicating that renewed bone growth starts only after the reproductive season is over. Limited attempts to section the penultimate phalanx of the second digit of the hind foot as an alternative method failed to produce good sections using the present freeze technique.

FIG. 1. Representative stained cross section of the femur of a two year old male P. triseriata (no. 8650). Arrows indicate lines of arrested growth; MC = medullary cavity. Note that RL 1 is a double line. Scale bar represents 20 μm.
The femurs of 27 individuals from the six localities were sectioned. Fig. 2A represents seven individuals from Marsh Lake, South Dakota. Three were determined to be two years old, and the remaining were scored as three years old. The largest individual (SVL 24.4 mm) was judged to be two years old, while three smaller males (SVL 23.6, 23.5, 23.2 mm) were determined to be three years old. Two additional males (SVL 22.8 and 22.3 mm) were two years old, and the smallest animal (SVL 22.2 mm) was three years old.

Five individuals from Cozad, Nebraska (Fig. 2B) reveal a similar trend. The largest two animals (one in excess of 27 mm SVL) were determined to be two years old, while the three year old was 23.4 mm SVL, followed by a two-year old only 1 mm smaller. Fig. 2C represents the largest sample (N = 10), from Arkansas City, Kansas, which contained almost equal numbers of animals two and three years of age. Again the largest specimen was a two-year old while four of the two-year olds have SVLs similar to the three largest three-year olds. Two of the three-year olds are among the smallest members of the population with the exception of one two-year old (SVL 25.1 mm).

Linear regression analysis was performed on a data set containing all individuals in Fig. 2A–C as well as additional specimens, two males from localities 2 and 5 and one from locality 3. The results revealed no significant relationship (F = 0.041, P = 0.84) between body size (SVL) and age estimates made using RLS.

Although the techniques used to section and stain femurs were effective, our results suggest that age does not correlate well with body size and that body size among chorus frogs overlapped substantially in the two- and three-year age classes. Among the three populations represented in Fig. 2A–C, the largest animal in each was a two-year old male and among the remaining 12 two-year old males, all had body lengths greater than 5 of 9 (55%) of three-year old individuals.

Results from other recent anuran studies utilizing skeletochronology reveal a mixed picture. In some species a positive correlation exists between age estimates and body size such that larger males are generally older, but in others this is not the case. In most studies there is substantial overlap in the body size of age classes, precluding size as an index of age. Both Hemelaar (1983) reporting on Bufo bufo, and Ryser (1988) for Rana temporaria, found a positive correlation between SVL and age for both males and females although substantial overlap in age classes occurs in each sex. In a sample made up largely of males, Leclair and Castanet (1987) found a positive correlation between age estimates and body size in Rana pipiens from southwestern Quebec, but again there was substantial overlap in size of age classes one and two. Höglund and Säterberg (1989) reported a positive correlation between body size and age estimates in female B. bufo, but not among males. Substantial overlap in age classes was apparent in both sexes. In a single study representing a congeneric relative of the chorus frogs, Lykens and Forester (1987) found a positive correlation between age estimates and body size for male P. crucifer.

The majority of the studies indicate a positive correlation between body size and age, suggesting that larger individuals tend to be older; however, too much overlap in body size exists to use it as a reliable index of age. In a review of amphibian and reptile papers Halliday and Verrell (1988) found weak age-body size correlations (based on several species of urodeles), and frequently nonsignificant correlations in those few anuran studies reported. In contrast, in a six-year mark and recapture study of Ascaphus truei, Daugherty and Sheldon (1982) reported reasonably distinct size classes among younger cohorts.
To fully understand the diversity of results in the current literature as well as our own study will require multigenerational skeletological studies of a variety of anurans from the same as well as disjunct populations. However, suboptimal conditions during larval development probably carry over into postmetamorphic and adult life as suggested by Hallday and Verrell (1988). Ryser (1988), for example, found a positive correlation between adult body size in R. temporaria and size at the end of the first year, supporting the suggestion that factors which influence juvenile size do carry over into adulthood. The fact that all of our specimens were collected in the same spring suggests two possibilities: (1) that individual variation in body size in relation to age is common (perhaps for genetic reasons relating to growth rate) and would be seen repeatedly if samples were taken from these same localities over a longer interval; or (2) that animals representing three-year olds in our samples were affected by environmental influences and experienced a "bad year" caused by suboptimal conditions for growth during year one but not year two. Because anurans grow throughout life, and we sampled more than one locale, and because the largest males in our samples were age two in each of three cases, the second alternative seems more likely. It should also be pointed out that two of the three populations in this paper (2 and 3) represented all of the calling males at those sites, so that although sample sizes are small from a statistical perspective, they are biologically meaningful in that they represent all that the females had to choose from.

The present study has shown that freeze-sectioned skeletological techniques can be successfully applied to chorus frog femurs but not to phalangeal sections, and that males in our breeding choruses from the Great Plains are commonly either two or three years of age and occasionally reach age four. Our results show a slight negative and non-significant correlation between age and body size among calling males, further suggesting that the assumption, often made in the past, that bigger means older is not necessarily true in chorus frogs. If older males were also usually larger, then Trivers's (1972) suggestion that females might prefer older males if age is correlated with genetic quality, would be supported. Dominant frequency is inversely correlated with body size in chorus frogs (r = -0.62, N = 158; JEP, unpubl. obs.) as is generally the case in anurans. However, our data argue against the likelihood that female chorus frogs would be able to use dominant frequency in this manner. The implications of our original observations toward an understanding of reproductive behavior in chorus frogs in the Great Plains suggests that male size rather than age may be more important in determining which males call and which operate as satellites. The fact that no one-year-old males were among those calling suggests that individuals of this age did not participate in the choruses we examined and may in fact be satellite males.

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Literature Cited


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