

# EVALUATION OF VENTRICULAR STIFFNESS OF FROGS AND SNAKES

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## Introduction

The vertebrate hearts have changed in structure and properties to adapt to different environments. Since the transition from aquatic to terrestrial environments was one of the most dramatic habitat changes during vertebrate evolution, terrestrialization may have prompted changes in the structure and properties of the heart. Previously, we showed that the ventricle of terrestrial toads was stiffer than that of aquatic frogs [1]. This result suggests that ventricles may have become stiff owing to terrestrialization. However, it is possible that the ventricles of the animals examined were not representative of their respective habitats because we examined only one type in each habitat. Here, to test further the hypothesis that terrestrialization caused ventricular stiffening, we measured ventricular stiffness in different animal species from those used in the previous study [1]. We compared the ventricular stiffness of frogs (aquatic) with that of snakes (terrestrial and arboreal) as a representative of reptiles that are less dependent on water than amphibians.

## Methods

Three species of adult frogs and snakes with different habitats were used in the experiment: *X. borealis* (aquatic frog), *E. quadrivirgata* (terrestrial snake), and *E. climacophora* (arboreal snake).

To analyze the passive mechanical properties of the ventricles, we obtained the relationship between the ventricular pressure  $P$  and the ventricular volume  $V$  by introducing cardioplegic solution into diastolic-arrested ventricle. The ventricular volume was normalized to the ventricular weight in each animal to obtain a pressure  $P$ -normalized volume  $nV$  relationship. The  $P$ - $nV$  relationship was fitted with

$$P = a \cdot e^{b \cdot nV} + c \quad (1)$$

to determine the stiffness constants  $a$ ,  $b$ , and  $c$  [2, 3]. Constant  $b$  was used as the stiffness measure of the ventricle following a previous study [4].

## Results

*X. borealis* (aquatic) had a smaller ventricle than other two species. The ventricles of *E. quadrivirgata* (terrestrial) and *E. climacophora* (arboreal) were more elongated than those of *X. borealis* (aquatic), indicating that the snake heart has a shape corresponding to their elongated bodies.

Figure 1 shows the stiffness constant  $b$  of ventricles. The stiffness constants  $b$  of *E. quadrivirgata* (terrestrial) and *E. climacophora* (arboreal) were significantly higher than that of *X. borealis* (aquatic). The stiffness constant

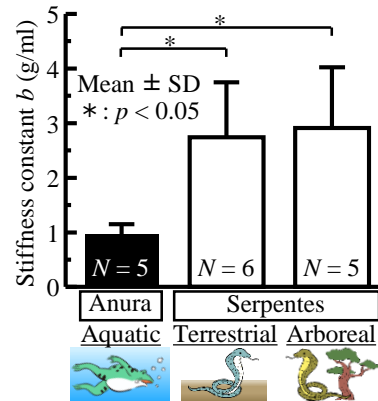


Figure 1: Stiffness constant  $b$  of ventricles of *X. borealis* (aquatic), *E. quadrivirgata* (terrestrial), and *E. climacophora* (arboreal).

$b$  of *E. quadrivirgata* (terrestrial) was equivalent to that of *E. climacophora* (arboreal).

## Discussion

In this study, we investigated the passive mechanical property of the ventricles of frogs and snakes in different habitats. The results suggested that the ventricles of terrestrial and arboreal species were stiffer than those of aquatic species. Compared to the previous studies [1], the ventricles of terrestrial and arboreal snakes were stiffer than those of aquatic frogs. Furthermore, the ventricular stiffness of terrestrial and arboreal snakes was comparable to that of terrestrial toads, even though their body shape differed significantly from terrestrial toads. After the terrestrialization of amphibians and the separation of reptiles from amphibians, amphibians and reptiles underwent their own unique evolution, for example, habitat diversification and loss of limbs. Our findings suggest that ventricular stiffness has been preserved even after amphibians and reptiles evolved independently. The combined results support the hypothesis that the transition from aquatic to terrestrial environments is key in evolutionary ventricular stiffening.

## References

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