that the high morphological uniformity found in the genus *Branchiostoma* exists also at a karyological level, if the older doubtful results are not considered.

In particular the genus *Branchiostoma* seems to be characterized by chromosomes which are all autosomes, regularly graded in size, mainly telocentric, with only 2 metacentric pairs; the chromosomes numbers are 19 (n) and 38 ($2n$) respectively and the content of DNA is very low, in comparison with that of vertebrates[9]. Spermatogenesis is chiasmatic.

A comparison of chromosome numbers, morphology and content of chromosomal DNA – allowing that the results obtained within the genus *Branchiostoma* can be extrapolated to include other amphioxi – indicates a closer relationship between amphioxi and some ascidians from the order Enterogona, rather than to pelagic tunicates[10] and a considerable separation of lower chordates from the vertebrates[7,9,11].

Riassunto. In entrambe le popolazioni di *Branchiostoma lanceolatum* del Golfo di Napoli e delle coste del Devonshire, Inghilterra, é stato trovato il numero aploide 19. Si sono individuati sia cromosomi metacentrici che telocentrici ma non cromosomi sessuali differenziati. È stata notata una certa somiglianza tra i cromosomi dei cefalocordati e di alcuni ascidiacei.

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[9] N. B. Atkin and S. Ohno, Chromosoma *23*, 10 (1967).

[10] D. Colombera, in press.

[11] K. M. Taylor, Chromosoma *21*, 181 (1967).

[12] This research was supported by grant No. 72.01034/04 115.0542 from the Institute of Marine Biology, C.N.R., Venice, to the Institute of Comparative Anatomy, University of Padova, Italy.

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Critical Thermal Maximum: Ecotypic Variation Between Montane and Piedmont Chorus Frogs (*Pseudacris triseriata*, Hylidae)

The critical thermal maximum (CTM) of ectothermic vertebrates provides a convenient index to adaptations to the thermal environment that have occurred at the subcellular level of organization[1]. Whereas most prior attempts to correlate CTM of amphibians with variations in the thermal environment have relied upon interspecific comparisons[2–7], few investigators have reported ecotypic variation of this character within a single species[6–9]. We here report differences in CTM of montane and piedmont chorus frogs (Hylidae: *Pseudacris triseriata*) that correspond with important differences in ambient temperatures of the respective environments[10].

Male chorus frogs were captured from breeding congregations in Larimer County in northcentral Colorado in May and June, 1973. Three populations were sampled both in the piedmont (elevation 1542–1631 m) and in the Front Range of the Rocky Mountains (elevation 2763–3040 m). Five animals from each population were acclimated in darkness at constant temperatures of 5 °C and 20 °C for 4–12 days prior to determining CTM by the method of Dunlap[5]. Experiments were performed between 05.00 and 11.00 Mountain Daylight Time in order to minimize the effect of circadian variations in CTM[11–13]. All frogs were held at 5 °C until beginning acclimation; and the interval between capture and

Table I. Critical thermal maxima of montane and piedmont chorus frogs following acclimation at 5 °C or 20 °C

Elevation		Acclimation temperature 5 °C	20 °C
Piedmont	Population 1	37.0 ± 0.051	38.7 ± 0.037
	Population 2	37.0 ± 0.037	38.6 ± 0.040
	Population 3	37.0 ± 0.066	38.6 ± 0.049
	$\overline{\overline{X}}$	37.0 °C	38.6 °C
Montane	Population 1	35.4 ± 0.073	36.9 ± 0.060
	Population 2	35.5 ± 0.051	38.1 ± 0.037
	Population 3	35.3 ± 0.037	38.1 ± 0.058
	$\overline{\overline{X}}$	35.4 °C	37.7 °C

Each mean (± the standard error) is based upon measurements of 5 different animals.

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[1] G. N. Somero, Am. Nat. *103*, 517 (1969).

[2] B. H. Brattstrom, Comp. Biochem. Physiol. *24*, 93 (1968).

[3] J. S. Frost and E. W. Martin, Copeia *1971*, 750 (1971).

[4] D. L. Claussen, Comp. Biochem. Physiol. *44A*, 137 (1973).

[5] D. G. Dunlap, Physiol. Zool. *41*, 432 (1968).

[6] H. Heatwole, N. Mercado and E. Ortiz, Physiol. Zool. *38*, 1 (1965).

[7] V. H. Hutchison, Physiol. Zool. *34*, 92 (1961).

[8] P. V. Cupp, jr. and E. D. Brodie, jr., Am. Zool. *12*, 689 (1972).

[9] J. R. Spotila, Ecol. Monogr. *42*, 95 (1972).

[10] The average air temperature for June–August is about 12 °C for the montane localities and 19 °C for the piedmont habitats. Colorado State Planning Commission, Water Conservation Board, and State Engineer, *Water Resources Survey, Climatological Data of Colorado* (Denver 1938).

[11] J. J. Mahoney and V. H. Hutchison, Oecologia *2*, 143 (1969).

[12] C. R. Johnson, Comp. Biochem. Physiol. *41A*, 727 (1972).

[13] D. G. Dunlap, Copeia *1969*, 852 (1969).

Table II. Analysis of variance of critical thermal maxima of chorus frogs

Source	SS	df	MS	F	*P*
Acclimation temperature	56.454	1	56.454	100.4	< 0.001
Elevation	24.067	1	24.067	42.8	< 0.001
Interaction (temperature × elevation)	1.803	1	1.803	3.2	> 0.1
Among populations (pooled)	4.499	8	0.562	43.2	< 0.001
Error	0.632	48	0.013		

Since there was significant variation in CTM among the populations of an experimental group, the Mean Squares/Among Populations was used as the denominator in calculating F-ratios for higher levels of variation[14]. SS, sums of squares; df, degrees of freedom; MS, mean squares; F, F-ratio.

determination of CTM was approximately 10 days for montane frogs and 30 days for piedmont animals.

Critical thermal maxima were subjected to 2-way analysis of variance, the fixed factors being elevation and acclimation temperature. One level of nesting (populations) was incorporated into the experimental design to permit us to distinguish altitudinal effects from interpopulation effects[14].

The CTM of chorus frogs acclimated at 20 °C was about 2 °C higher than that of frogs held at 5 °C (Tables I and II), which is the expected pattern of response[5,13,15]. More important, however, the CTM of piedmont frogs averaged 1.2 °C higher than that of montane animals (Tables I and II). Additionally, the effect of thermal acclimation on CTM was similar in frogs from montane and piedmont populations (Table II).

The CTM of amphibians has been reported either to be positively correlated with body weight[16] or to be independent of body weight[5-7]. Assuming that the former situation obtains in chorus frogs, it is possible that the CTM of the larger montane frogs[17,18] was displaced upward relative to that of the smaller piedmont animals. However, since the observed CTM of the montane frogs still is lower than that of piedmont frogs (Table I), the difference in mean values seems not to be attributable to the influence of body weight.

Additionally, the CTM of amphibians has been reported either to decrease in starved animals[16] or to be independent of nutritional state[7]. Since piedmont frogs were held without food for a longer interval before study than montane frogs, it is possible that the CTM of piedmont animals was depressed accordingly. However, since the recorded CTM of piedmont frogs still is higher than that of montane frogs, the mean difference in CTM seems not to be attributable to different nutritional states of the experimental animals.

We conclude, therefore, that the different CTMs of montane and piedmont chorus frogs represent ecotypic variation within this species. The observed difference in CTM between montane and piedmont chorus frogs is precisely what one would predict as an outcome of evolutionary compensation for temperature[1]. The montane frogs, which occupy generally cooler habitats[10], presumably have acquired rate limiting enzymes in intermediary metabolism that function optimally at lower mean temperatures than do equivalent enzymes in piedmont frogs. Consequently, negative modulation of enzyme function by increasing temperature leads to metabolic inactivation, and death, at lower body temperatures in montane frogs than in piedmont animals[1].

Zusammenfassung. Das kritische thermale Maximum von Fröschen (Hylidae: *Pseudacris triseriata*) aus Bergpopulationen liegt niedriger als jenes von Tieflandpopulationen, wobei es sich offenbar um das Ergebnis einer physiologischen Evolution handelt: Adaptation an kühleres Höhenklima.

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8 October 1973.

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[14] R. R. SOKAL and F. J. ROHLF, *Biometry* (Freeman, San Francisco 1969).

[15] N. HOLZMAN and J. J. McMANUS, Comp. Biochem. Physiol. *45A*, 833 (1973).

[16] R. V. SEIBEL, Herpetologica *26*, 208 (1970).

[17] D. PETTUS and A. W. SPENCER, SWest. Nat. *9*, 20 (1964).

[18] G. C. PACKARD, Physiol. Zool. *44*, 90 (1971).

[19] We thank D. L. KILGORE, JR., and M. J. PACKARD for critical reviews of drafts of the manuscript, and T. J. BOARDMAN for assistance with statistical analysis of the data.

Nicotinic Acid Suppressors in *Aspergillus nidulans*

Suppressor genes have been studied both in higher organisms and micro-organisms; and mainly in the latter, the study has led to important contributions to the understanding of gene action. In fungi, several instances have been reported of differences in dominance of suppressors between diploids and heterokaryons[1-3]. In respect to the nicotinic acid requirement caused by the *nic8* mutant of *A. nidulans*, several authors reported the presence of suppressors of such gene. WARR and ROPER[4], for instance, looking for resistant mutants to various inhibitors, found that *p*-fluorophenylalanine (*p*FA) resistant genes also suppress the nicotinic acid requirement caused by *nic8*.

We have isolated and studied suppressors of the *nic8* gene of *A. nidulans* in relation to several aspects, such as map location, resistance to *p*FA, growth rate and dominance or recessiveness of the isolated suppressors in heterokaryons and diploids. For that the minimal medium