IN SILICO PRACTICAL – MORPHOLOGICAL VARIATION IN DIFFERENT POPULATIONS OF STICKLEBACK

Three-spine stickleback (*Gasterosteus aculeatus*) were collected from 3 different locations, from an estuary in Kent, a pond in the Botanic Gardens in Dundee, and a freshwater stream in the Outer Hebrides in Scotland. You are provided with images of 5-6 fish collected from each location which were fixed and stained with Alizarin Red/Alcian Blue and then cleared to reveal the skeleton of the fish, with bone shown in red and cartilage blue. There is a side view of each fish and also a view of the underside of the fish to show the pelvic region. For each set of images, you are provided with a scale bar.

Figure 14.20 (below) provides a diagram indicating features of the bony anatomy of a fully spined stickleback.



Blue = 3 dorsal spines, red= pelvic girdle and right pelvic spine, yellow= lateral armor plates on the right side of the body

KENT- SPECIMEN 1





KENT – SPECIMEN 3





KENT – SPECIMEN 5





HEBRIDEAN - SPECIMEN 1





HEBRIDEAN - SPECIMEN 3





HEBRIDEAN - SPECIMEN 5





BOTANICAL GARDENS - SPECIMEN 1





BOTANICAL GARDENS – SPECIMEN 3





BOTANICAL GARDENS – SPECIMEN 5



ENVIRONMENTAL CALCIUM CONCENTRATIONS IN DIFFERENT STICKLEBACK HABITATS

Location (Type of Habitat)	Sample	рН	[Ca2+]		Giles (1983) [Ca2+]
			mg/ml	mM	mM / pH
River Rife, Kent, England, UK	1		116.40	2.91	
(river, estuarine)	2		116.00	2.90	
	Mean		116.20	2.91	
Botanic Gardens, Dundee, Scotland, UK	pond 1 (a)	8.11	64.00	1.60	
(natural spring-fed pond)	pond 1 (b)	8.09	66.20	1.66	
	pond 2 (a)	8.33	78.70	1.97	
	pond 2 (b)	8.36	72.20	1.81	
	Mean	8.22	70.28	1.76	
North Uist, Outer Hebrides, Scotland, UK	Loch Scabhagh	6.17	1.90	0.05	0.035
(Inland Lochs, Lochans)	Loch Na Garbh a Bhainn Ard	6.05	2.20	0.06	
	Loch Fada	6.25	2.10	0.05	0.056 / 5.4
	Stream (Loch Fada to Loch Struth Mhoir)		2.00	0.05	
	Mean	6.16	2.05	0.05	

Giles, N. (1983) J. Zool., Lond. 199, 535-544.

EXERCISE

- 1) Characterize the phenotype of the fish from the three different locations by carrying out the following analysis on each fish.
 - a) Measure its total length.
 - b) Measure the depth of its body.
 - c) Score each fish for the number of dorsal spines (colored blue in the diagram).
 - d) Score each fish for the extent of the pelvic structures present by scoring for presence or absence of pelvic spines and/or measuring their length (colored red in the diagram). You may find this easier using the ventral view of the pelvic region.
 - e) Score each fish for the number of bony lateral plates present (colored yellow in the diagram).
- 2) Draw up tables showing the results of your analysis. What are the main qualitative differences in the skeleton of the fish from the different locations? For example, are the dorsal spines present? Do the fish from all three locations have lateral armor plates and pelvic spines?
- 3) You may also wish to make quantitative comparisons. How do the lengths of the pelvic spines vary in fish from the different locations? Why might it be useful to compare the ratios of the length of the pelvic spines and another measurement (e.g. body length) rather than simply using the raw data? Is it possible to carry out any statistical tests on the data?
- 4) Are there any differences between right and left pelvic spines?

5) You are also provided with measurements of the calcium concentration in the water from the three different habitats. Use these data to evaluate the proposition that calcium deficiency may have been an important environmental factor in the evolution of skeletal reduction.

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Tickle, C., Cole, N.J.: Morphological diversity: taking the spine out of three-spine stickleback. *Curr Biol.* 2004, 14:R422-424.