Armor Loss Across Threespine Stickleback Populations

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Marine vs. Freshwater Stickleback Fish

Marine:

- Found in salt water environments
- ~30-40 lateral armor plates
- Defined dorsal and pelvic spines

Freshwater:

- Found in lakes, rivers, and streams
- ~0-12 lateral armor plates
- Little to no dorsal or pelvic spines

Parallel Evolution

- Different freshwater populations across the world show similar features
- Same gene in all populations responsible for these features
- Gene responsible identified as Pitx1

Evolutionary Advantages

- Marine Stickleback armor and spines
  - Armor prevents predators with softer teeth from eating them

- Freshwater lack of armor
  - Allows easier movement and faster swimming
  - Certain insects cannot latch on to spines to try and capture the sticklebacks

Crosses Used

- Ancestral Marine fish (ANC)
  - Robust, bony armory and well defined pelvic structures with complete lateral plates
- Two independently derived freshwater population
  - Population from Mud Lake, AK (DER1)
    - Lack lateral plates but have complete pelvis structure
  - Population from Boot Lake, AK (DER2)
    - Small subset of lateral plates but very reduced, or no pelvic structure

Overview of the Study

- Obtained fish from Anchorage, Alaska
- Established stocks using both marine ancestral fish and freshwater fish
- Incubated the embryos from the cross
- Developed embryo cartilage and bone structures were stained and compared

Summary of Data

- Bone and cartilage development in the post embryonic stage
  - Many bones and cartilage form after hatching
- Presence of feature vs. time feature developed
  - Logistic regression was used to quantify when certain areas developed bone/cartilage
  - Compared the timing of these cartilage formations and ossifications between each population
  - Compared timing of development of different fin rays and the three spines

Methods

- Utilized lab rearing to minimize environmental impacts on development
- Cartilage and bone initiate between 15 and 30 dpf, sampling emphasized here
- Alizarin staining used to detect pelvic structures
- Logistic Regression used to model a binary function.

Fig. 1a

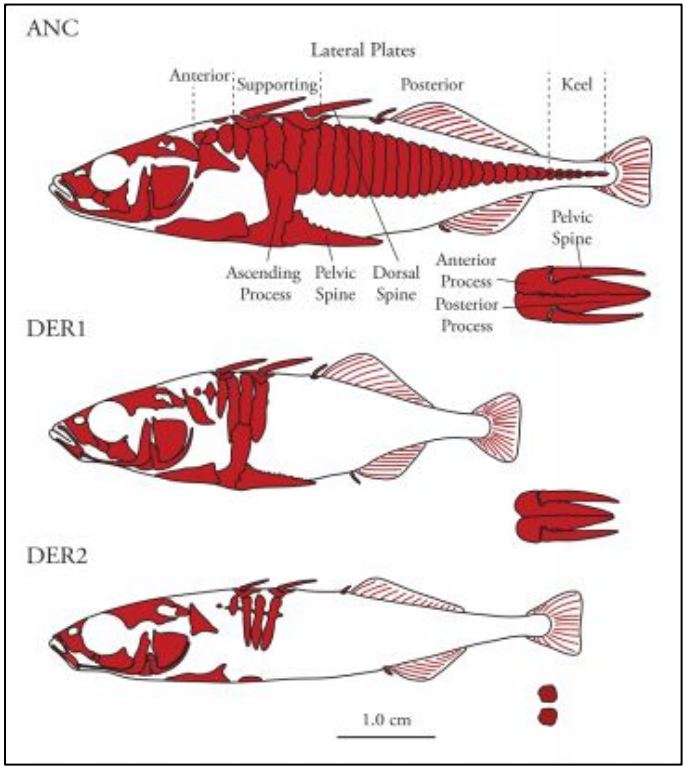

ANC
Lateral Plates
Anterior
Supporting
Posterior
Keel
Pelvic Spine
Ascending Process
Pelvic Spine
Dorsal Spine
Anterior Process
Posterior Process
DER1
DER2
1.0 cm

Fig. 1b-d

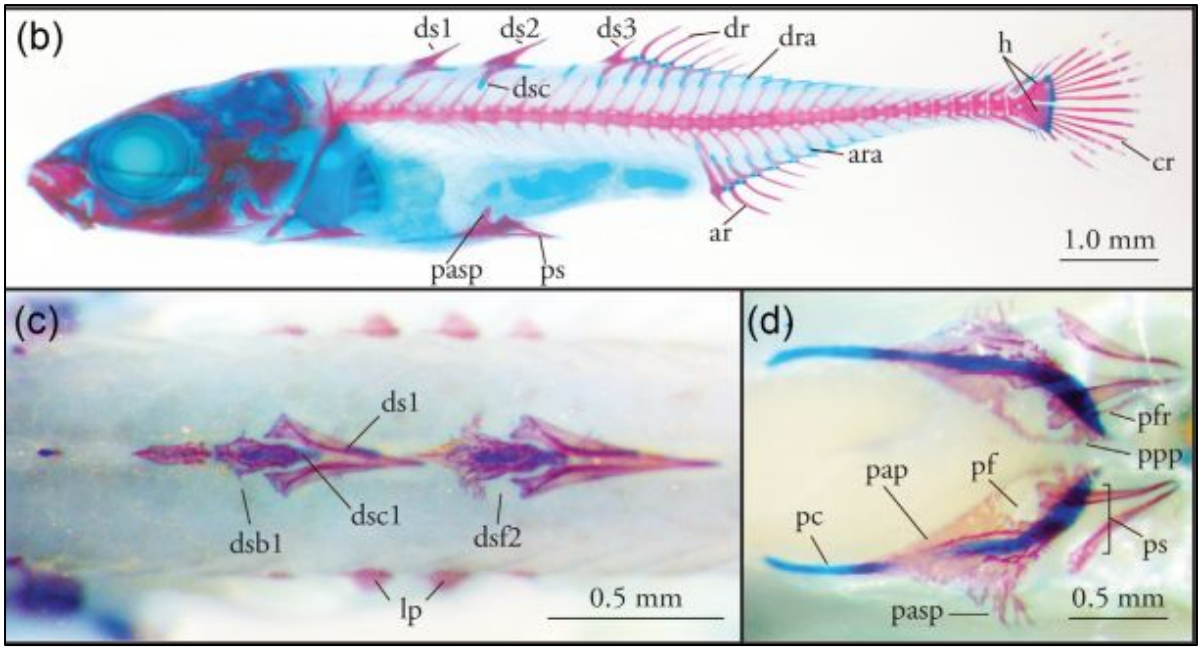

(b)
ds1
ds2
ds3
dr
dra
h
dsc
ara
cr
ar
pasp
ps
1.0 mm
(c)
ds1
dsc1
dsb1
dsf2
lp
0.5 mm
(d)
pfr
ppp
pap
pf
pc
ps
pasp
0.5 mm

Fig. 2

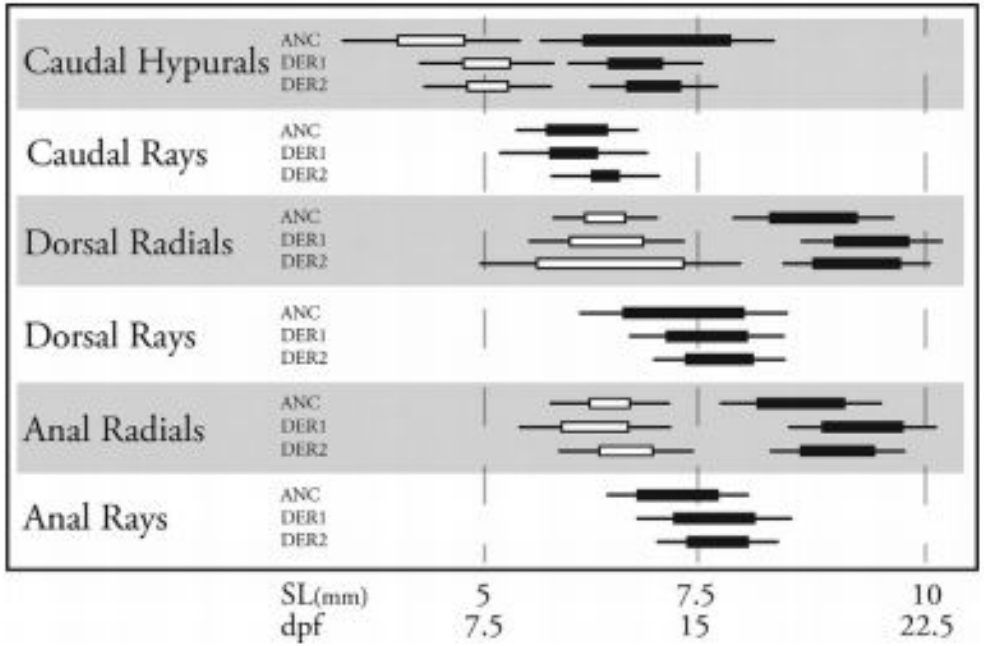

Caudal Hypurals
ANC
DER1
DER2
Caudal Rays
ANC
DER1
DER2
Dorsal Radials
ANC
DER1
DER2
Dorsal Rays
ANC
DER1
DER2
Anal Radials
ANC
DER1
DER2
Anal Rays
ANC
DER1
DER2
SL(mm)
dpf
5
7.5
7.5
15
10
22.5

Fig. 3a-d

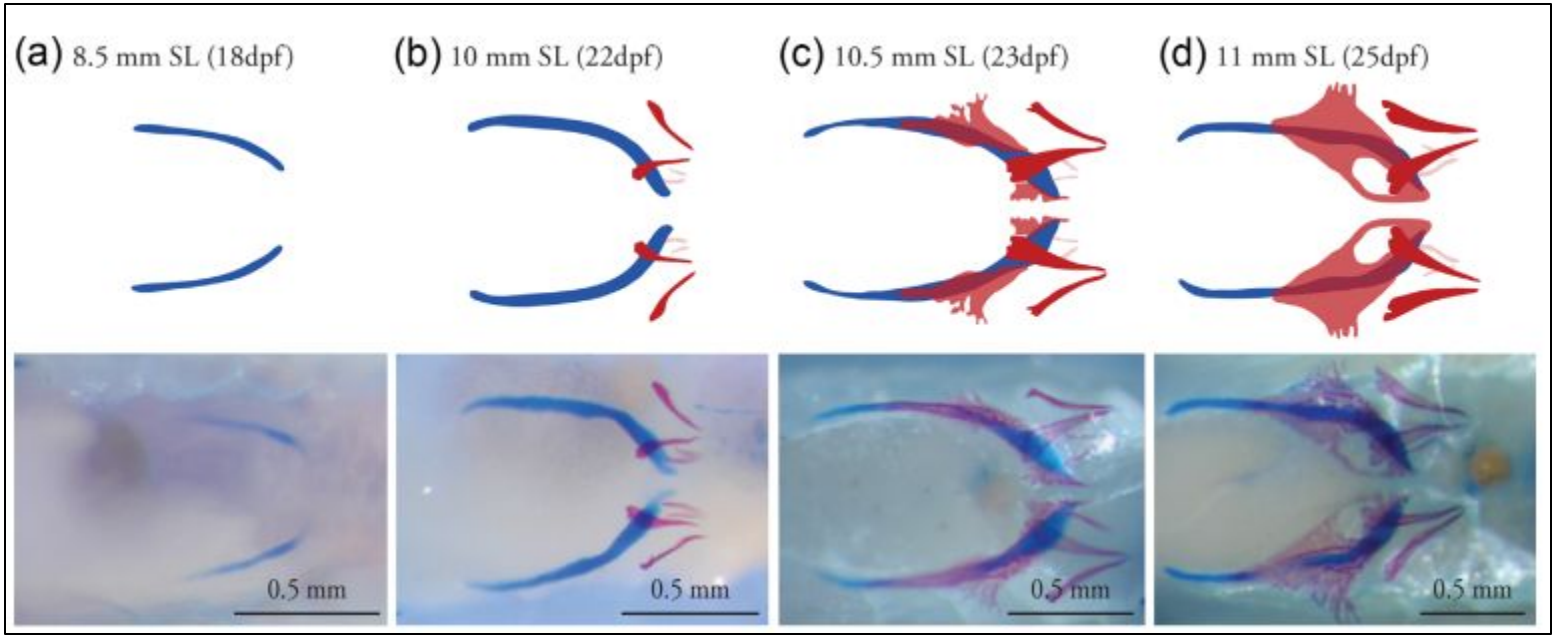

(a) 8.5 mm SL (18dpf)
(b) 10 mm SL (22dpf)
(c) 10.5 mm SL (23dpf)
(d) 11 mm SL (25dpf)
0.5 mm
0.5 mm
0.5 mm
0.5 mm

Fig. 3e-g

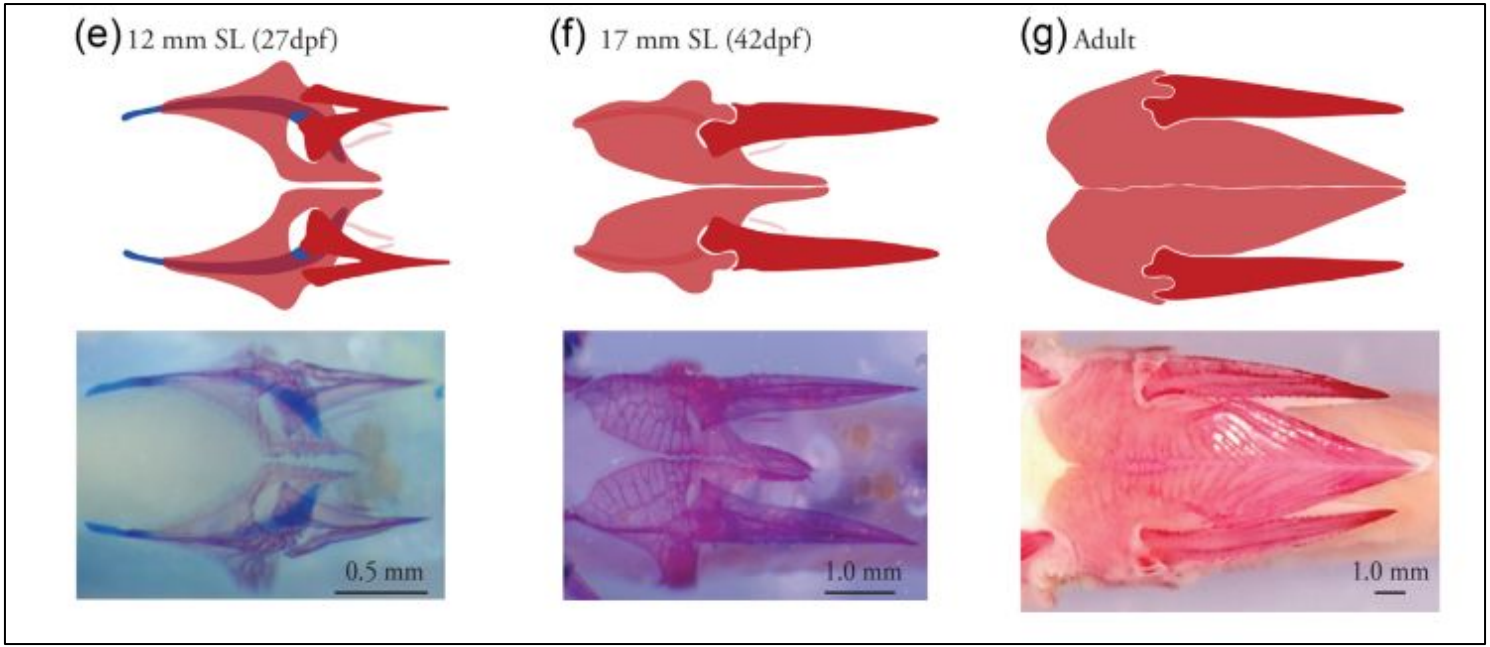


Fig. 4

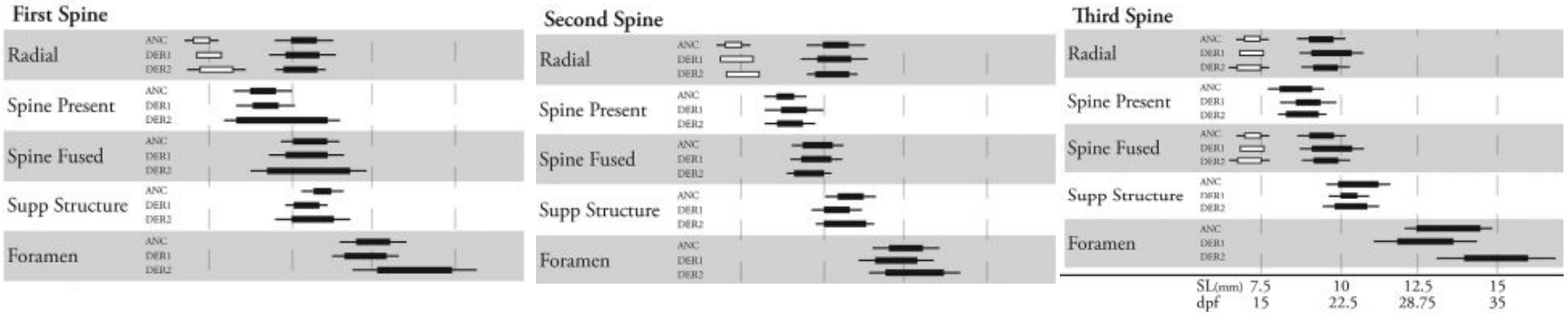
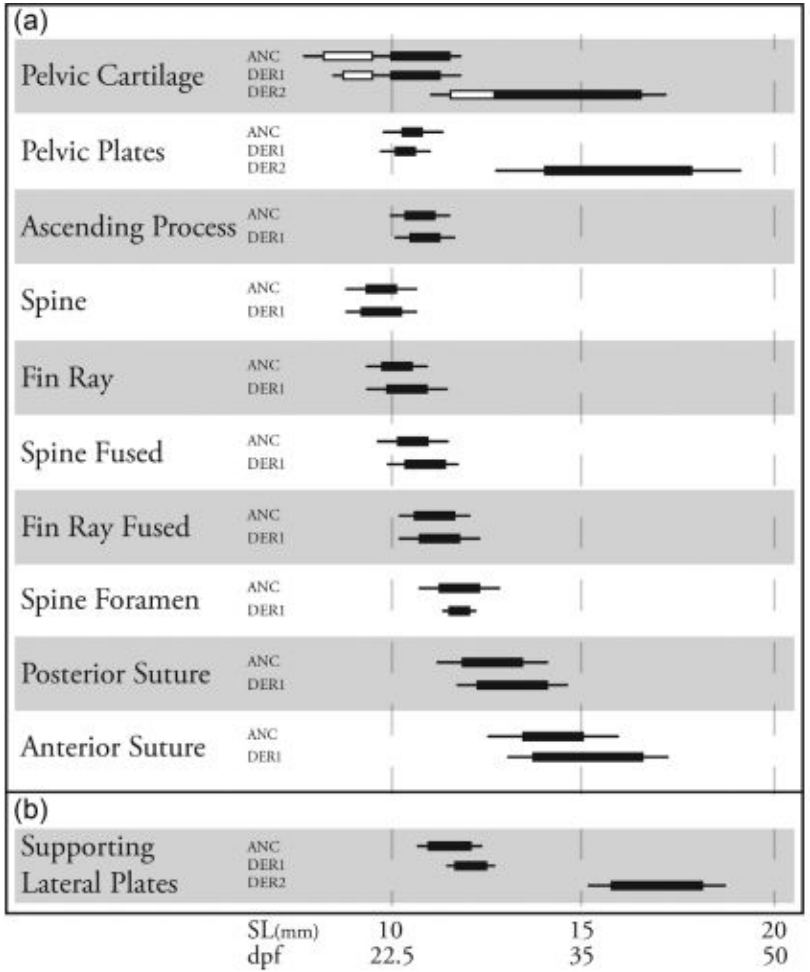
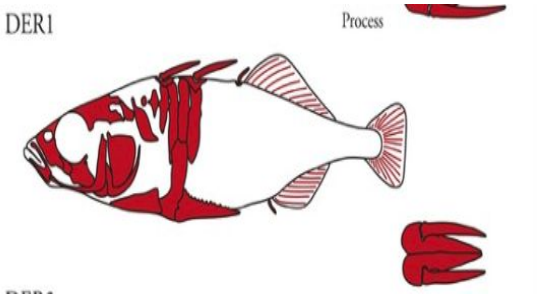

First Spine
Radial
Spine Present
Spine Fused
Supp Structure
Foramen
ANC
DER1
DER2
Second Spine
Radial
Spine Present
Spine Fused
Supp Structure
Foramen
ANC
DER1
DER2
Third Spine
Radial
Spine Present
Spine Fused
Supp Structure
Foramen
ANC
DER1
DER2
SL(mm) 7.5 10 12.5 15
dpf 15 22.5 28.75 35

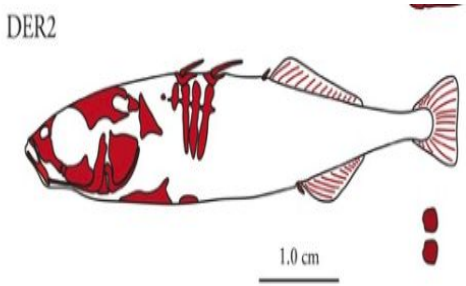
Fig. 5

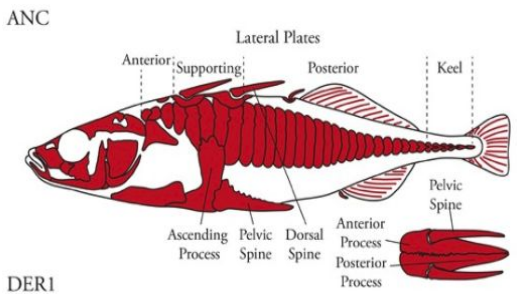

(a)
Pelvic Cartilage
ANC
DER1
DER2
Pelvic Plates
ANC
DER1
DER2
Ascending Process
ANC
DER1
Spine
ANC
DER1
Fin Ray
ANC
DER1
Spine Fused
ANC
DER1
Fin Ray Fused
ANC
DER1
Spine Foramen
ANC
DER1
Posterior Suture
ANC
DER1
Anterior Suture
ANC
DER1
(b)
Supporting Lateral Plates
ANC
DER1
DER2
SL(mm) 10 15 20
dpf 22.5 35 50

Conclusion

- Overall post-hatching growth rate is nearly the same among the three compared populations.
- Able to quantify the variation in the development of the derived skeletal traits that differ among the three studied populations. (DER1, DER2, ANC)







Quotes from Paper

- Divergent adult morphologies emerge through altered developmental programs for specific traits in an otherwise highly congruent developmental context
- The population with the most divergent morphology also is the most delayed and sports the broadest variation among individuals in ontogenetic timing of defensive traits
- DER1 and DER2 arrive at a roughly parallel complement of adult lateral plates, but differ markedly in the schedule of their development implicating additional loci beyond (*eda)* that could affect initiation timing and subtler morphological differences

Conclusions

- The fish still harbor developmental competence for pelvic apparatus
- The altered adult phenotypes result from modular developmental changes specific to the divergent traits
- Changes in initiation time and in sequence of events appear to have accompanied the evolution of specific pelvic structure morphologies

Conclusions

- Delayed pelvic cartilage and plate formation, show that the bony outgrowths are outgrowths of the periostia of the underlying cartilages.

- Pitx1 expression is strongest where the pelvic spines will emerge

- Possible that in DER2 stickleback, there is reduced expression levels of pitx1, permitting occasional expression of reduced pelvic traits in some individuals

Threespine Stickleback

- Long history of scientific interest
- Loss of the armor is resulting from relaxed selection on this developmental fate
- Proves effectiveness of “micro-evo-devo” research program
- Observations of post-embryonic skeletal development
- Fish species are excellent models for studying developmental differences among populations