



Measuring amphibian gene flow across species, scale and sampling scheme in a managed forest

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Introduction

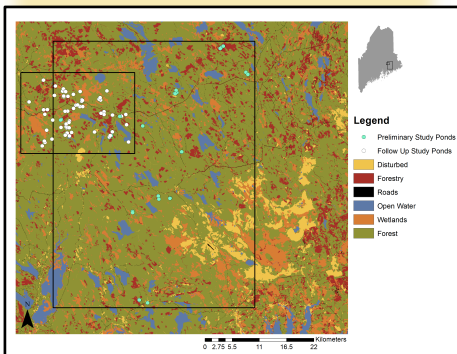
In the face of anthropogenic landscape change, understanding the factors that affect dispersal and gene flow is essential for conservation management. Landscape genetics provides a mechanism to examine how landscape features affect gene flow in natural populations.

Simulation studies suggest the strongest relationship between gene flow and landscape pattern occurs when the scale of sampling and analysis match the scale of the phenomena being studied¹. When using empirical data, however, it can be a challenge to synchronize these scales and may require analysis at multiple scales^{2,3}.

The purpose of this study was to:

Utilize a landscape genetics approach to investigate how the relationship between genetic distance and landscape pattern is influenced by two different

- sampling schemes
- spatial scales
- thematic resolutions
- focal species



This map shows the ponds sampled for both the preliminary and follow up studies; it also indicates the spatial extents of each study area.

Literature cited

1. Anderson, C., B. K. Epperson, M. J. Fortin, R. Holderegger, P. M. A. James, M. S. Rosenberg, K. T. Scribner, and S. Spear. 2010. Considering spatial and temporal scale in landscape-genetics studies of gene flow. *Molecular Ecology* 19:3565-3575.
2. Murphy, M. A., J. S. Evans, and A. Storfer. 2010. Quantifying *Bufo boreas* connectivity in Yellowstone National Park with landscape genetics. *Ecology* 91:252-261.

Methods

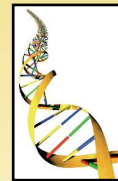
Sampling

- We compared data from a preliminary study conducted at a larger spatial extent with clustered sampling to a follow up study with a smaller spatial extent and continuous sampling (see Figures below).
- At each pond we collected ~ 20 embryonic samples from egg masses of the spotted salamander (*Ambystoma maculatum*) and the wood frog (*Lithobates sylvatica*).

	Preliminary Study	Follow Up Study	
Scale	Large	Fine	
Extent	40 x 52 km	15 x 10 km	
Sampling	Clustered	Continuous	
Thematic Resolution	Multivariate (Expert Opinion) Binary		
Species	Spotted salamander Wood frog		
			Spotted salamanders
	<ul style="list-style-type: none"> • 7 pond clusters • 23 ponds • 6 microsatellite loci • 411 samples 	<ul style="list-style-type: none"> • 53 ponds • 8 microsatellite loci • 1225 samples 	
	<ul style="list-style-type: none"> • 7 pond clusters • 20 ponds • 6 microsatellite loci • 287 samples 	<ul style="list-style-type: none"> • 34 ponds • 7 microsatellite loci • 780 samples 	Wood frogs

Genetic analysis

- DNA was extracted and amplified in multiplexed PCR reactions using microsatellite markers.
- We used the program STRUCTURE⁴ to estimate the number of subpopulations (K).
- To estimate the genetic distance between ponds we used the metrics F_{ST} and chord distance (D_c).



Landscape Analysis

- To characterize the landscape, we obtained a landcover map from the Maine Office of GIS and merged this with wetland, stream, and road layers.
- We modeled the landscape using a multivariate approach where resistance values were derived from expert opinion⁵.
- We also modeled the landscape using a binary approach where we isolated each variable of interest including open water, streams, wetlands, forestry, roads, slope, and disturbed land (agriculture, development and bare ground) and assigned high resistance to that variable and low resistance to the background.
- We conducted a least-cost path analysis and correlated the path length with genetic distance (F_{ST} and D_c) using partial Mantel tests to control for geographic distance.

3. Galpern, P., M. Manseau and P. Wilson. 2012. Grains of connectivity: analysis at multiple spatial scales in landscape genetics. *Molecular Ecology* (online).
4. Pritchard, J. K., M. Stephens, and P. Donnelly. 2000. Inference of population structure using multilocus genotype data. *Genetics* 155:945-959.
5. Compton, B. W., K. McGarigal, S. A. Cushman, and L.R. Gamble. 2007. A resistant-kernel model of connectivity for amphibians that breed in vernal pools. *Conservation Biology* 21:788-799.

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Results

- The STRUCTURE analysis indicated both species in each study lacked sub-structure and were a single population.
- Few landscape features were significantly correlated with genetic distance in the preliminary study, but several more features became significant in the follow up study.
 - In spotted salamanders, disturbed land, open water, and streams were significant or marginally significant in both studies; additionally, in the follow up study roads and geographic distance were marginally significant.
 - In wood frogs, no landscape variables were significantly correlated with genetic distance; in the follow up study geographic distance, disturbed land, wetlands, and streams were significant or marginally significant.
- The multivariate resistance model was not correlated with genetic distance for either species.

Results from Mantel tests of least-cost paths and genetic distance

Model	Preliminary Study				Follow Up Study					
	D_c	R	P	F_{ST}	D_c	R	P	F_{ST}		
IBD	-0.13	0.86	-0.20	0.98	0.11	0.08*	0.08	0.13		
Disturbed	0.05	0.19	0.08	0.08*	0.05	0.05*	0.04	0.10*	Spotted salamanders	
Forestry	-0.10	0.67	0.05	0.32	0.05	0.24	-0.03	0.66		
Roads	0.04	0.23	0.08	0.11	0.05	0.07*	0.04	0.12		
Water	0.18	0.02*	0.12	0.06*	0.10	0.03*	0.09	0.04*		
Slope	-0.02	0.52	0.00	0.48	-0.12	0.91	-0.11	0.89		
Wetlands	0.04	0.36	-0.05	0.68	0.05	0.29	0.09	0.16		
Streams	0.05	0.22	0.08	0.09*	0.06	0.05*	0.04	0.09*		
Multivariate	0.07	0.27	0.06	0.30	0.07	0.14	0.02	0.36		
Model	R	P	R	P	Model	R	P	R	P	
IBD	-0.10	0.74	-0.05	0.60	Disturbed	0.09	0.06*	0.03	0.29	Spotted salamanders
Disturbed	-0.02	0.58	-0.04	0.66	Forestry	0.10	0.17	-0.01	0.49	
Forestry	-0.20	0.89	-0.17	0.86	Roads	0.06	0.14	0.01	0.45	
Roads	-0.02	0.57	-0.05	0.69	Water	0.07	0.12	0.02	0.34	Wood frogs
Water	-0.08	0.74	-0.11	0.82	Slope	-0.07	0.69	-0.08	0.74	
Slope	0.04	0.39	-0.04	0.55	Wetlands	0.14	0.10*	0.06	0.28	
Wetlands	-0.21	0.97	-0.21	0.96	Streams	0.08	0.08*	0.02	0.36	
Streams	-0.01	0.96	-0.05	0.67	Multivariate	0.09	0.14	0.02	0.40	
Multivariate	-0.13	0.83	-0.13	0.82						

* Indicates significance as $P < 0.05$; * indicates significance at $P < 0.10$

Conclusion

- Geographic distance alone does not explain the genetic patterns.
- Landscape resistance to dispersal is species specific; wood frogs have fewer dispersal barriers.
- Power is increased with a finer-scale sampling scheme.
- A binary thematic resolution was more powerful than a multivariate model in this study.
- More landscape features were significantly related to genetic distance using D_c which is consistent with the metric being more sensitive to recent genetic differentiation as compared to F_{ST} .

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