

APPENDIX C. Genetic and geographic distance and their relationships for 16 populations of *Desmognathus fuscus* sampled in the Washington D.C.–Baltimore metropolitan region.

TABLE C1. Pairwise F_{st} values (below diagonal) and associated P values (above diagonal) measured among *Desmognathus fuscus* populations sampled in the Washington D.C.–Baltimore metropolitan region. Significance was based on 1000 permutations.

Pop	BYN1	BYN2	WIN1	WIN2	LIT1	LIT2	GUN1	GUN2
BYN1	0	***	***	***	***	***	***	***
BYN2	0.135	0	***	***	***	***	***	***
WIN1	0.157	0.152	0	***	***	***	***	***
WIN2	0.144	0.135	0.156	0	***	***	***	***
LIT1	0.093	0.207	0.209	0.161	0	***	***	***
LIT2	0.212	0.263	0.274	0.185	0.097	0	***	***
GUN1	0.147	0.191	0.187	0.129	0.043	0.132	0	***
GUN2	0.113	0.189	0.216	0.112	0.052	0.098	0.047	0
JON1	0.145	0.175	0.162	0.106	0.090	0.155	0.070	0.076
JON2	0.112	0.198	0.107	0.121	0.079	0.156	0.071	0.071
GWY1	0.095	0.166	0.183	0.129	0.040	0.140	0.027	0.056
GWY2	0.097	0.134	0.137	0.068	0.061	0.140	0.029	0.032
PAT1	0.175	0.194	0.154	0.159	0.159	0.267	0.099	0.127
PAT2	0.155	0.200	0.145	0.157	0.176	0.256	0.155	0.153
SEN1	0.311	0.357	0.344	0.300	0.212	0.405	0.161	0.272
SEN2	0.164	0.133	0.167	0.146	0.110	0.252	0.062	0.136

Table C1. Continued.

Pop	JON1	JON2	GWY1	GWY2	PAT1	PAT2	SEN1	SEN2
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BYN1	***	***	***	***	***	***	***	***
BYN2	***	***	***	***	***	***	***	***
WIN1	***	***	***	***	***	***	***	***
WIN2	***	***	***	***	***	***	***	***
LIT1	***	***	***	***	***	***	***	***
LIT2	***	***	***	***	***	***	***	***
GUN1	***	***	***	**	***	***	***	***
GUN2	***	***	***	***	***	***	***	***
JON1	0	***	***	***	***	***	***	***
JON2	0.116	0	***	***	***	***	***	***
GWY1	0.088	0.092	0	***	***	***	***	***
GWY2	0.046	0.043	0.035	0	***	***	***	***
PAT1	0.087	0.137	0.108	0.094	0	***	***	***
PAT2	0.091	0.156	0.129	0.104	0.043	0	***	***
SEN1	0.231	0.237	0.179	0.197	0.188	0.287	0	***
SEN2	0.113	0.128	0.075	0.071	0.079	0.137	0.121	0

Notes: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

TABLE C2. Pairwise in-network (below diagonal) and out-of-network distances (above diagonal) measured among sampling localities in the Washington D.C.–Baltimore metropolitan region. All distances are expressed in kilometers (km). Bolded values indicate the distances between stream pairings established in the hierarchical study design.

Pop	BYN1	BYN2	WIN1	WIN2	LIT1	LIT2	GUN1	GUN2
BYN1	0	8.18	0.86	9.34	16.33	16.48	17.94	17.52
BYN2	12.88	0	8.68	2.29	22.84	20.74	24.11	21.98
WIN1	56.93	46.68	0	9.68	15.49	15.65	17.08	16.68
WIN2	42.92	32.67	16.83	0	22.70	19.90	23.81	21.17
LIT1	104.26	94.01	107.32	93.30	0	7.09	1.96	6.67
LIT2	98.73	88.48	101.79	87.78	12.32	0	6.48	1.28
GUN1	115.43	105.19	118.50	104.48	89.61	84.09	0	5.75
GUN2	111.21	100.96	114.27	100.25	85.38	79.86	19.70	0
JON1	136.23	125.98	139.30	125.28	144.95	139.42	156.12	151.90
JON2	132.61	122.37	135.68	121.66	141.33	135.80	152.51	148.28
GWY1	141.29	131.04	144.35	130.34	150.00	144.48	161.18	156.95
GWY2	131.39	121.14	134.45	120.43	140.10	134.57	151.28	147.05
PAT1	341.08	330.83	344.14	330.13	349.79	344.27	360.97	356.74
PAT2	334.43	324.19	337.50	323.48	343.15	337.62	354.33	350.10
SEN1	468.92	458.67	471.99	457.97	477.64	472.11	488.81	484.59
SEN2	468.53	458.28	471.60	457.58	477.25	471.72	488.42	484.20

Table C2. Continued.

Pop	JON1	JON2	GWY1	GWY2	PAT1	PAT2	SEN1	SEN2
BYN1	36.18	36.74	36.56	38.25	75.12	72.85	79.41	75.27

BYN2	38.51	38.04	39.08	39.36	76.98	74.13	81.16	76.75
WIN1	35.39	36.00	35.76	37.52	74.34	72.09	78.63	74.50
WIN2	36.98	36.31	37.59	37.58	75.25	72.29	79.40	74.95
LIT1	24.48	26.87	24.48	28.55	62.38	61.02	66.75	63.07
LIT2	20.09	21.50	20.35	23.14	58.94	57.00	63.27	59.27
GUN1	22.64	25.17	22.62	26.86	60.44	59.10	64.81	61.14
GUN2	19.37	21.04	19.58	22.69	58.12	56.28	62.46	58.52
JON1	0	4.82	1.02	5.99	38.95	36.91	43.25	39.19
JON2	10.84	0	5.77	1.68	38.95	36.21	43.11	38.75
GWY1	77.13	73.51	0	6.83	38.60	36.71	42.92	38.93
GWY2	67.23	63.61	11.46	0	37.69	34.80	41.81	37.39
PAT1	334.70	334.70	339.76	329.86	0	6.02	4.38	3.57
PAT2	328.06	324.44	333.11	323.21	11.50	0	8.15	3.18
SEN1	462.55	458.93	467.60	457.70	499.35	492.70	0	4.98
SEN2	462.16	458.54	467.21	457.31	498.94	492.31	10.73	0

TABLE C3. Comparison of correlation coefficients calculated for each genetic distance measure with both out-of-network and in-network distances. All comparisons are significant and all genetic distance measures exhibit similar relationships with both out-of-network and in-network distances.

Genetic Distance	Geographic Distance (km)	Stream Distance (km)
F_{st}	0.439***	0.396***
D	0.419***	0.427***
D_{EST}	0.449***	0.414***
G_{st}	0.426***	0.419***
$G_{st,EST}$	0.424***	0.423***

Notes: *** $P < 0.001$.

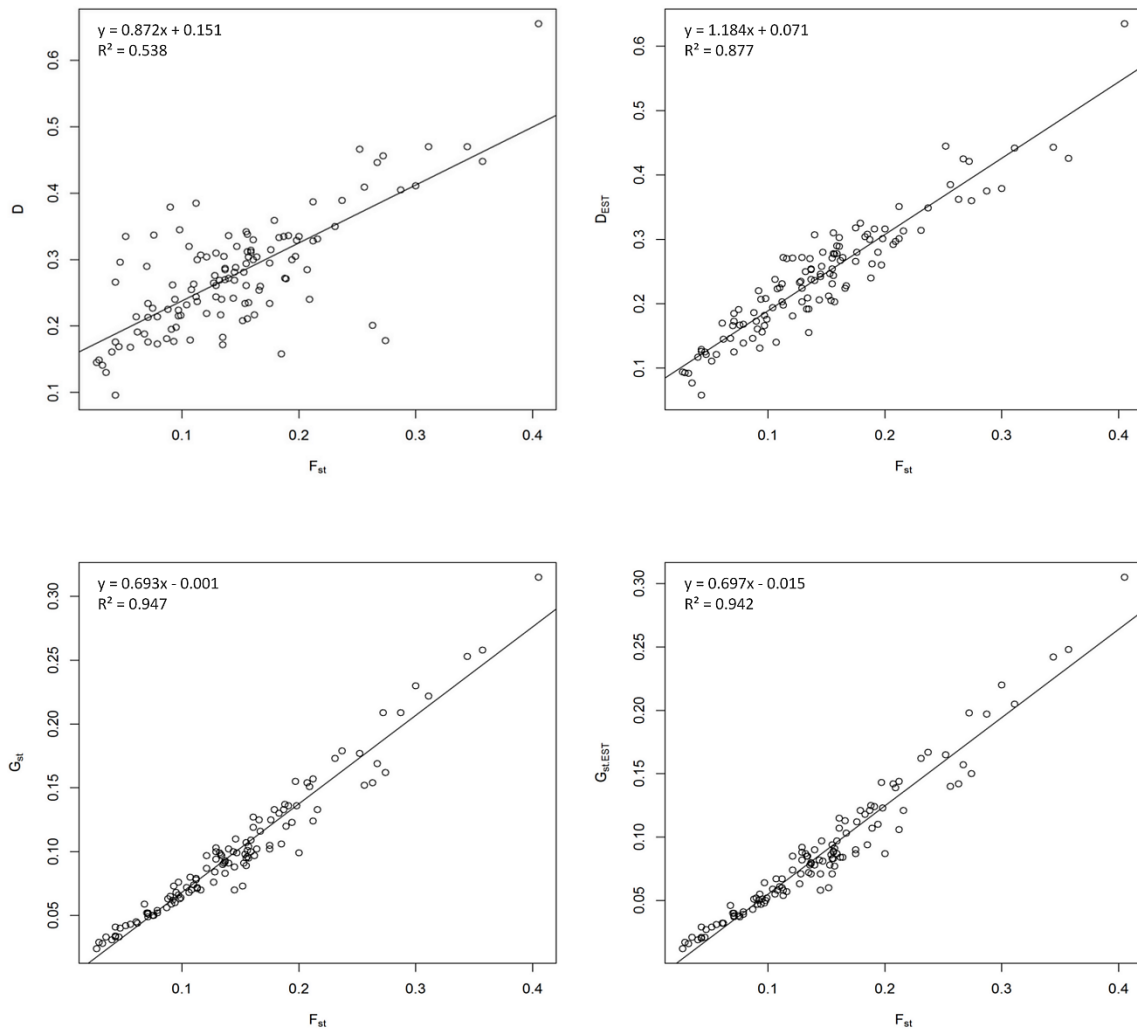


FIG. C1. Relationship between pairwise F_{st} and other genetic distance measures. F_{st} (Weir and Cockerham 1984) was compared to D and D_{est} (Jost 2008), G_{st} (Nei 1973), and $G_{st. EST}$ (Nei and Chesser 1983). Weir and Cockerham's (1984) F_{st} estimator provides an unbiased measure of relative genetic differentiation related to variance in allele frequencies among compared populations. G_{st} is another commonly used measure of relative genetic differentiation, which quantifies the difference between average within-population heterozygosity and among-

population (total) heterozygosity. $G_{st,EST}$ (Nei and Chesser 1983) is an extension of G_{st} and is calculated by using nearly unbiased estimators of within-population and among-population heterozygosity, thus providing a correction for variance in sample sizes among localities. Jost's (2008) D estimates absolute genetic differentiation by measuring the proportion of total genetic diversity, measured among paired populations, which can be attributed to the average single population. D_{EST} applies Nei and Chesser's (1983) unbiased measures of heterozygosity to the estimation of D in order to provide a correction for sample size variance.

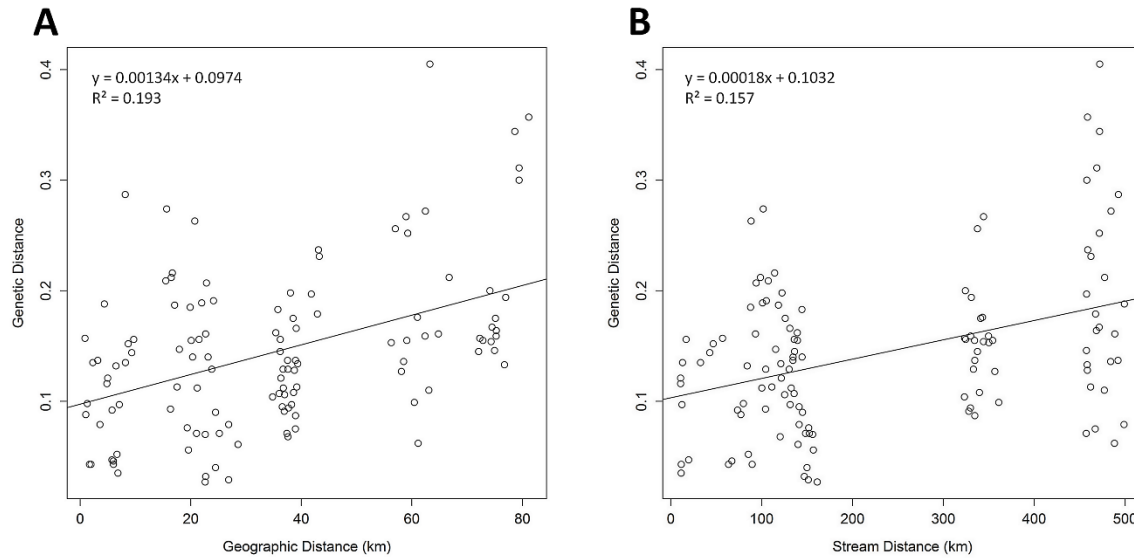


FIG. C2. Isolation-by-distance plots representing the correlation between genetic distance (F_{st}) and geographic distance (km) along out-of-network dispersal pathways (geographic distance; A), and in-network dispersal pathways (stream distance; B).

LITERATURE CITED

- Jost, L. 2008. G_{ST} and its relatives do not measure differentiation. *Molecular Ecology* 17:4015-4026.
- Nei, M. 1973. Analysis of gene diversity in subdivided populations. *Proceedings of the National Academy of Sciences* 70:3321-3323.
- Nei, M., and R. K. Chesser. 1983. Estimation of fixation indices and gene diversities. *Annals of Human Genetics* 47:253-259.
- Weir, B. S., and C. C. Cockerham. 1984. Estimating F-statistics for the analysis of population structure. *Evolution* 38:1358-1370.