Marine forests of the Mediterranean-Atlantic *Cystoseira tamariscifolia* complex show a southern Iberian genetic hotspot and no reproductive isolation in parapatry

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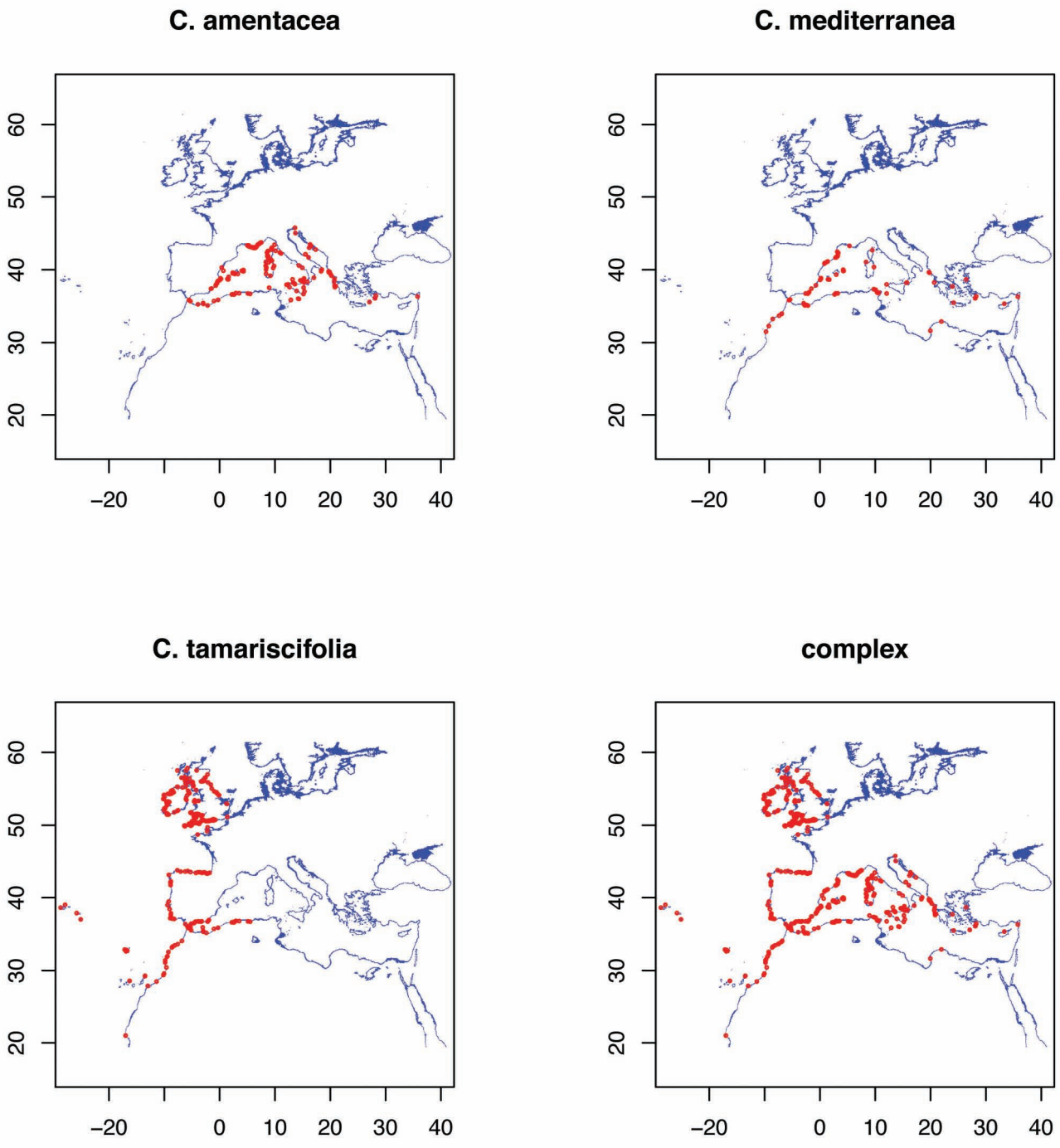
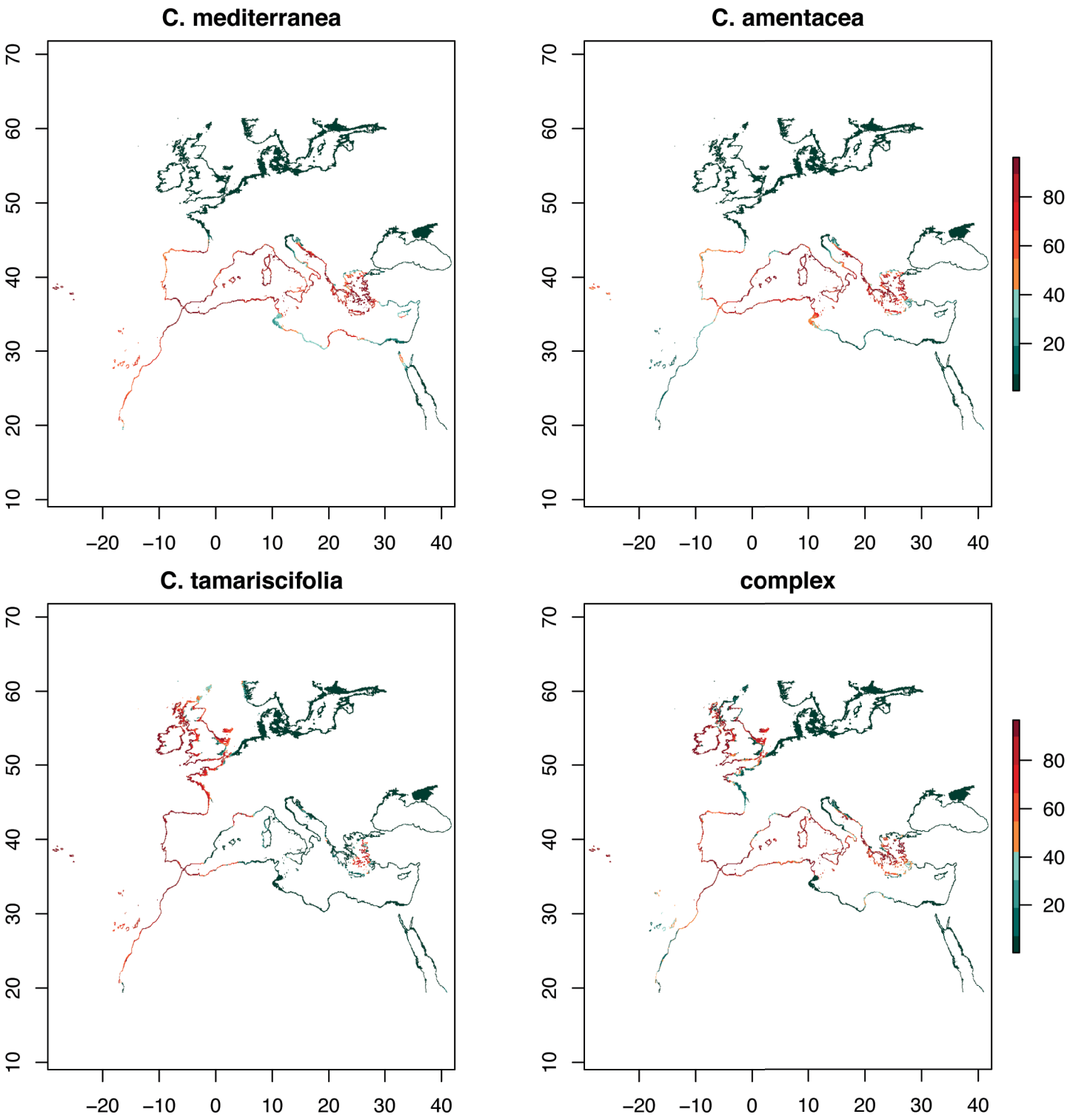
Supplementary material:

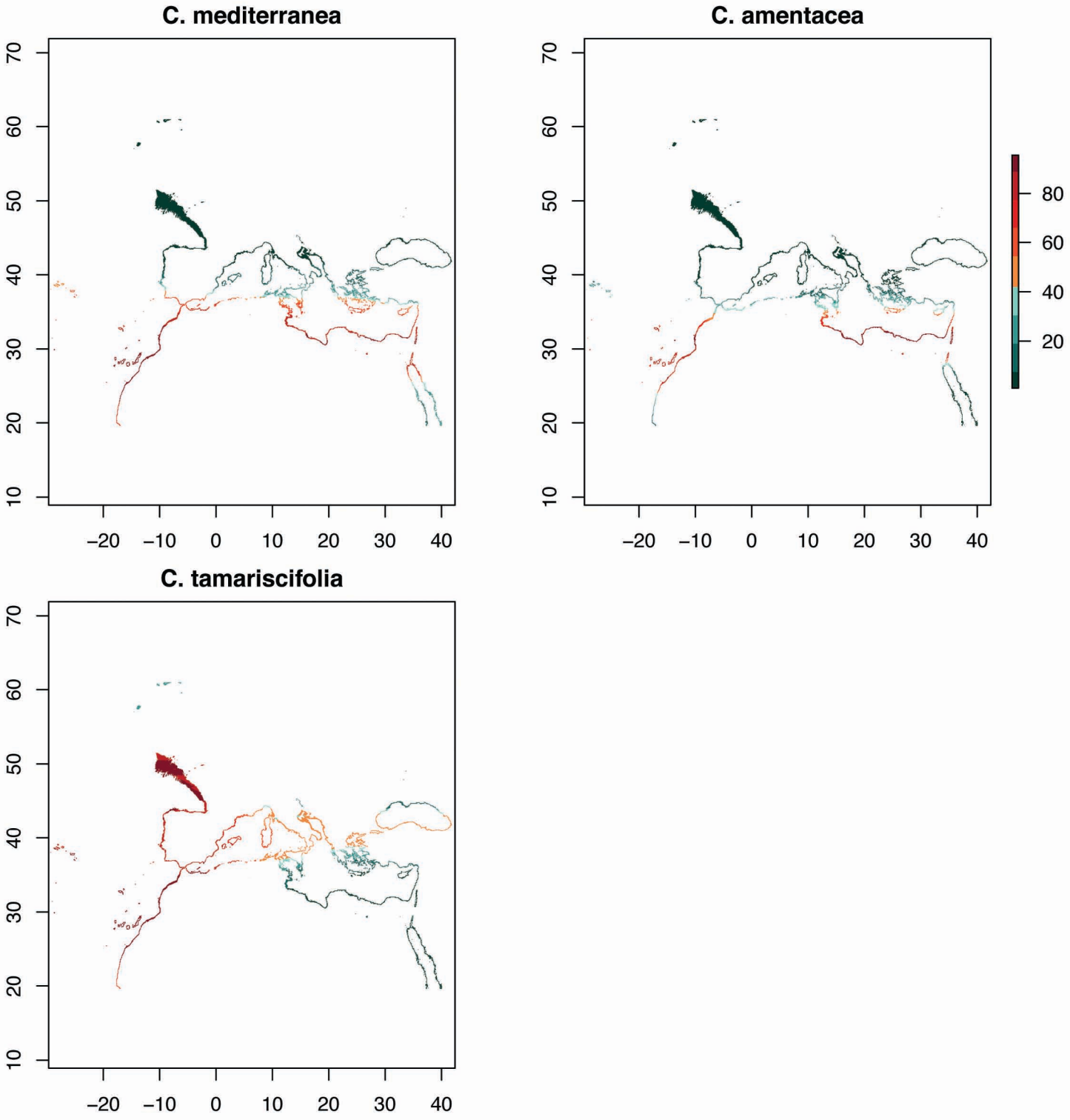
Appendix S1 Supplementary data and results regarding SDMs and list of specimens deposited in the herbarium of the University of Algarve.
Appendix S2 Supplementary data and results regarding the parapatric populations of *C. amentacea* and *C. tamariscifolia* from "El Playazo".
Appendix S3 Supplementary results regarding genetic structure, diversity and differentiation.
Appendix S4 Supplementary results regarding genetic isolation by distance.



Supplementary S1.1. Georreferenced records used to build Spatial Distributuion Models of *Cystoseira amentacea, C. mediterranea, C. tamariscifolia* and *C. ericaefolia* complex.



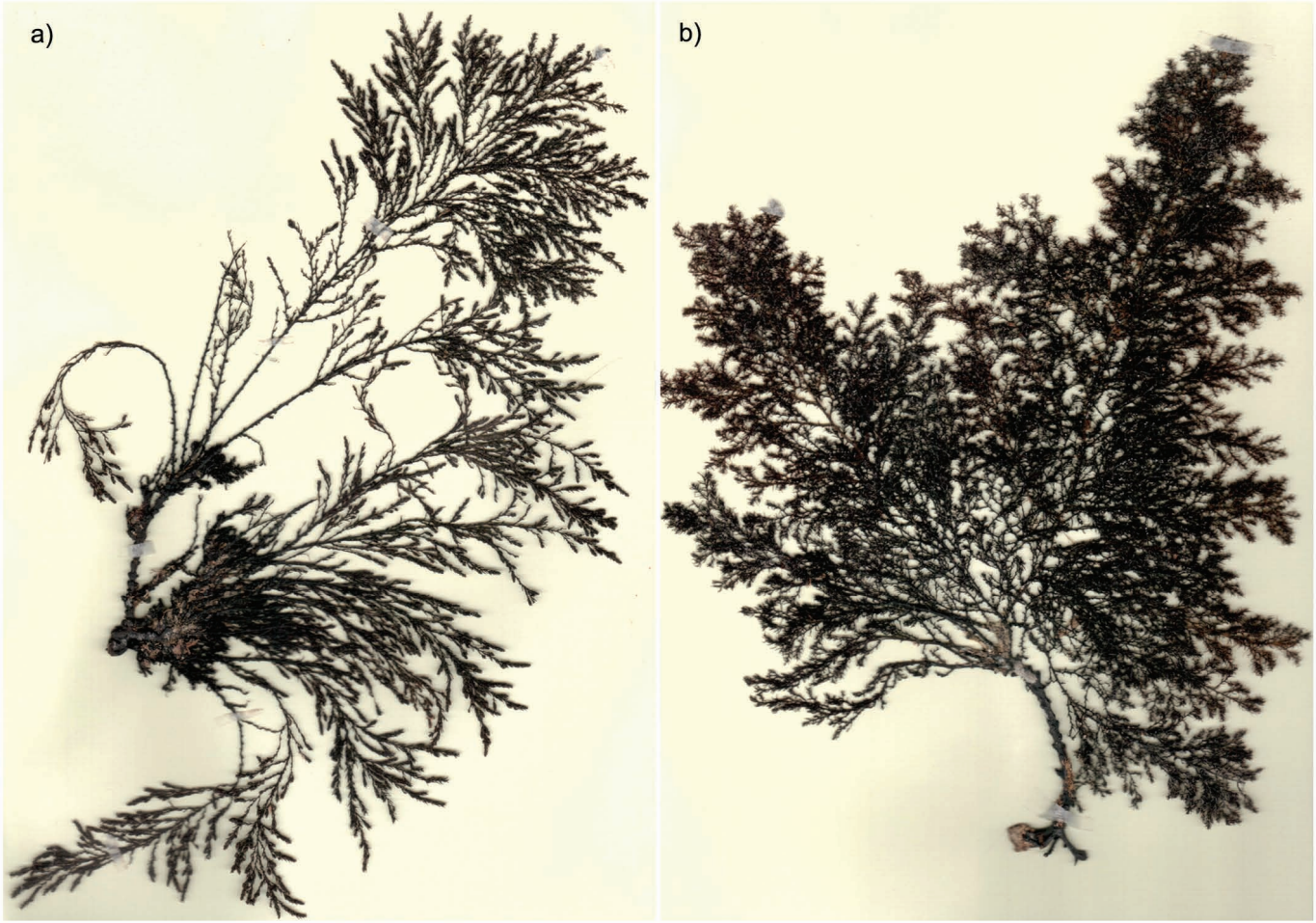
Supplementary S1.2. Assemble map of distribution of *Cystoseira amentacea, C. mediterranea, C. tamariscifolia* and *C. ericaefolia* complex for the present time using six presence-absence algorithms.



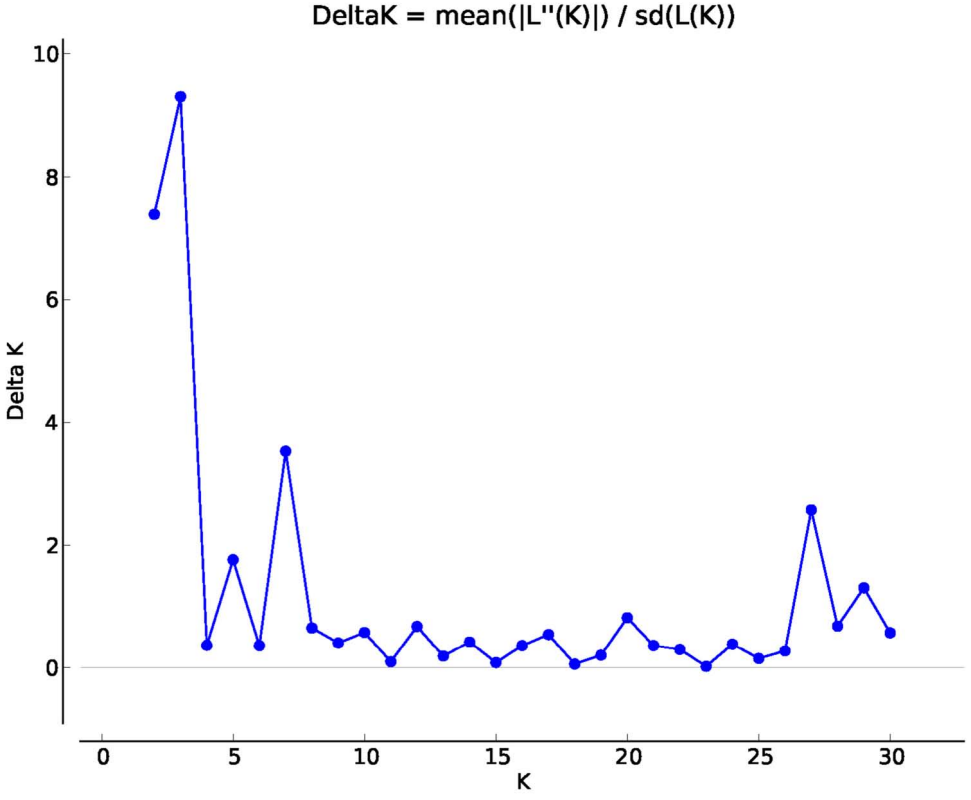
Supplementary S1.3. Ensemble map of distribution of *Cystoseira amentacea, C. mediterranea, C. tamariscifolia* and *C. ericaefolia* complex for the Last Clacial Maximum using six presence-absence algorithms.

Species	Region	Local	Code	Data	nº ALGU	Latitude	Longitude	Details
Cystoseira tamariscifolia	Cornwall	Polzeath	Po	30/6/11	A999	50.5858	4.882	2 samples in silica
Cystoseira tamariscifolia	Bretanha	Roscoff	Ro	28/7/10	A996	48.7294	4.0108	2 samples in silica
Cystoseira tamariscifolia	Cantabria	La Noja	Nj	11/4/11	A993	43.4962	3.5247	2 samples in silica
Cystoseira tamariscifolia	Asturias	Porcia	Pr	15/4/11	A989	43.5676	6.8754	2 samples in silica
Cystoseira tamariscifolia	Viana do Castelo	Praia Norte, Viana do Castelo	VC	28/4/10	A988	41.6993	8.8567	2 samples in silica
Cystoseira tamariscifolia	Alentejo	Odeceixe	Od	18/6/10	A992	37.4388	8.804	2 samples in silica
Cystoseira tamariscifolia	Algarve	Praia Manuel Lourenço, Albufeira	Ab	17/8/10	A995	37.0761	8.2769	2 samples in silica
Cystoseira tamariscifolia	Cádiz	El Chato, Cadiz	Ca	30/9/12	A994	36.477676	-6.264495	2 samples in silica
Cystoseira tamariscifolia	Cádiz	El Mirlo (Tarifa)	Ta	14/1/09	A972	36.0596	-5.7198	Herbarium sheet
Cystoseira tamariscifolia	Cádiz	Tarifa	Ta	14/1/09	A998	36.059609	-5.719784	2 samples in silica
Cystoseira tamariscifolia	Málaga	Calaburras	Cb	9/6/12	A973	36.5061	-4.6397	Herbarium sheet
Cystoseira tamariscifolia	Málaga	Calaburras	Cb	26/1/09	A987	36.5061	-4.6397	2 samples in silica
Cystoseira tamariscifolia	Granada	La Herradura	He	21/6/08	A974	36.7361	-3.7577	Herbarium sheet
Cystoseira tamariscifolia	Granada	La Herradura	He	22/6/12	A986	36.736105	-3.757676	2 samples in silica
Cystoseira amentacea	Almeria	Guardias Viejas	GV	23/6/08	A965	36.6950	-2.8496	Herbarium sheet
Cystoseira tamariscifolia	Almeria	El Playazo (Cabo de Gata)	Ps	4/8/12	A975	36.8593	-2.0032	Herbarium sheet
Cystoseira tamariscifolia	Almeria	El Playazo (Cabo de Gata)	Ps	3/8/08	A990	36.8593	-2.0032	2 samples in silica
Cystoseira amentacea	Almeria	El Playazo (Cabo de Gata)	Pi	4/8/12	A966	36.8593	-2.0032	Herbarium sheet
Cystoseira amentacea	Almeria	El Playazo (Cabo de Gata)	Pi	3/8/08	A976	36.8593	-2.0032	2 samples in silica
Cystoseira amentacea	Alicante	Santa Pola	SP	27/1/13	A977	38.196482	-0.514723	2 samples in silica
Cystoseira mediterranea	Tarragona	Punta de La Mora	PM	20/11/12	A982	41.126227	1.344316	2 samples in silica
Cystoseira mediterranea	Girona	Cala Frances, Blanes	Bl	20/12/12	A983	41.681354	2.814728	2 samples in silica
Cystoseira mediterranea	Girona	Cap de Creus	CC	30/7/10	A985	42.317225	3.31634	1 sample in silica
Cystoseira mediterranea	Baleares	Porto Colom, Mallorca	Ml	2010	A984	39.416826	3.275612	2 samples in silica
Cystoseira amentacea	Sicily	Altavilla (north Sicily), Sant'Elia	SE	2013	A980	38.046146	13.559728	5 samples in silica
Cystoseira amentacea	Sicily	Mazara del Vallo (south Sicily)	Close SC	2013	A981	37.651799	12.577311	2 samples in silica
Cystoseira amentacea	Calabria	Crotone (Calabria)	Cr	2013	A979	39.093961	17.112076	2 samples in silica
Cystoseira amentacea	Puglia	Otranto (Puglia)	Ot	2013	A978	40.0322	18.4534	3 samples in silica

Supplementary S1.4. List of specimens deposited in the herbarium of the University of Algarve.



Supplementary S2. Habit of putative specimens of *Cystoseira amentacea* (a) and *C. tamariscifolia* (b) from “El Playazo”



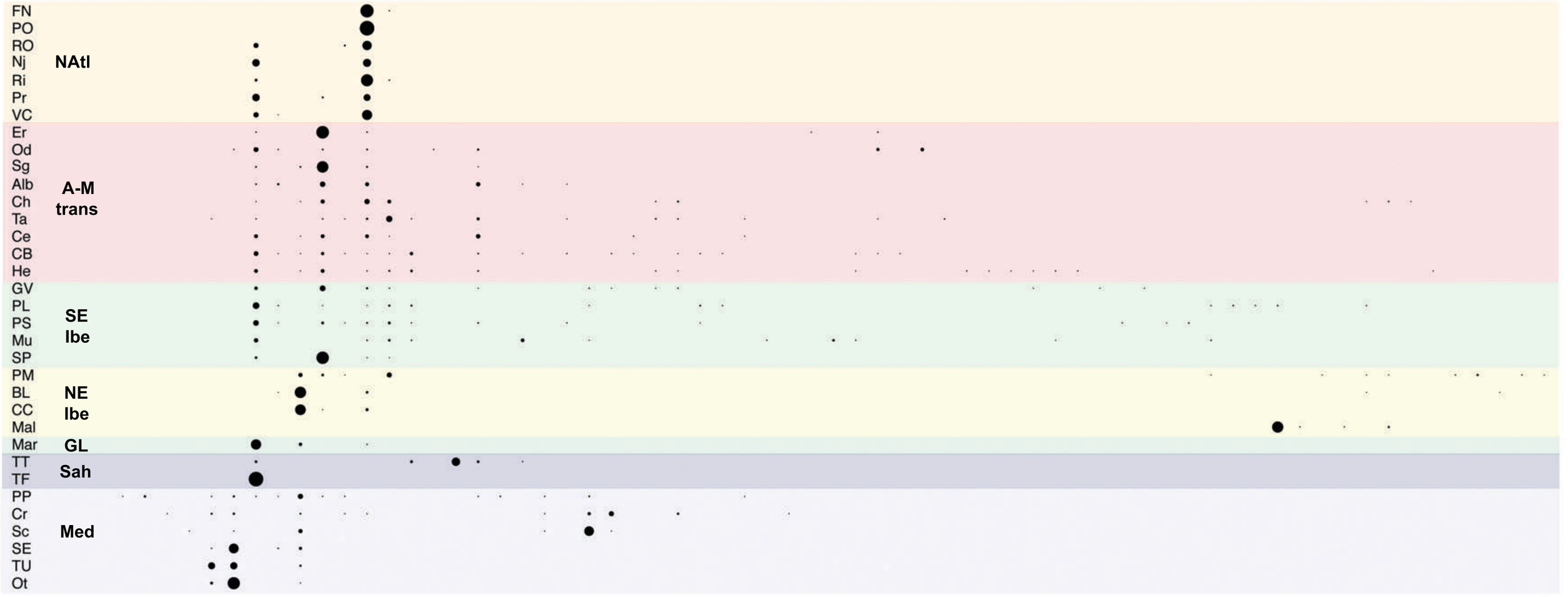
Supplementary S3.1. Plot of ΔK as a function of the number of cluster (K) across the 14 runs.

	FN	PO	RO	Nj	Ri	Pr	VC	Er	Od	Sg	Alb	Ch	Ta	Ce	CB	He	GV	PL	PS	Mu	SP	PM	BL	CC	Mal	Mar	TT	TF	PP	Cr	Sc	SE	TU	Ot
FN	0.00	0.31	0.27	0.47	0.57	0.51	0.57	0.72	0.88	0.64	0.69	0.87	0.89	0.74	0.85	0.74	0.85	0.92	0.81	0.97	0.84	0.91	0.79	0.73	1.00	0.99	0.90	1.00	0.82	0.71	0.93	1.00	0.82	0.83
PO	0.75	0.00	0.06	0.25	0.19	0.16	0.26	0.47	0.81	0.65	0.49	0.51	0.83	0.65	0.69	0.66	0.71	0.75	0.76	0.88	0.94	0.69	0.69	0.65	0.78	0.98	0.89	1.00	0.81	0.76	0.93	0.93	0.88	0.88
RO	0.63	0.19	0.00	0.19	0.22	0.10	0.24	0.42	0.79	0.60	0.53	0.55	0.83	0.65	0.67	0.63	0.74	0.74	0.75	0.88	0.91	0.67	0.69	0.65	0.88	0.93	0.88	0.93	0.79	0.68	0.91	1.00	0.82	0.83
Nj	0.59	0.35	0.28	0.00	0.09	0.10	0.19	0.39	0.78	0.60	0.49	0.43	0.73	0.51	0.58	0.60	0.70	0.65	0.63	0.83	0.88	0.65	0.79	0.65	0.86	0.88	0.93	0.87	0.87	0.78	0.83	0.99	0.78	0.79
Ri	0.69	0.34	0.35	0.11	0.00	0.08	0.10	0.41	0.81	0.70	0.47	0.35	0.75	0.64	0.62	0.70	0.70	0.67	0.72	0.83	0.95	0.58	0.74	0.62	0.81	0.95	0.95	0.96	0.89	0.83	0.88	1.00	0.89	0.90
Pr	0.70	0.32	0.21	0.11	0.12	0.00	0.14	0.32	0.77	0.60	0.45	0.40	0.79	0.61	0.55	0.63	0.65	0.59	0.68	0.80	0.92	0.53	0.68	0.62	0.80	0.87	0.90	0.88	0.82	0.71	0.87	0.99	0.83	0.84
VC	0.80	0.54	0.48	0.30	0.21	0.28	0.00	0.51	0.87	0.79	0.62	0.51	0.86	0.74	0.67	0.80	0.79	0.73	0.80	0.84	0.96	0.61	0.83	0.77	0.82	0.93	0.99	0.93	0.95	0.79	0.91	0.99	0.83	0.84
Er	0.63	0.45	0.42	0.24	0.30	0.25	0.47	0.00	0.72	0.21	0.25	0.38	0.66	0.46	0.45	0.41	0.46	0.61	0.60	0.76	0.73	0.56	0.66	0.61	0.75	0.98	0.86	0.97	0.74	0.74	0.80	0.95	0.79	0.79
Od	0.74	0.63	0.61	0.40	0.48	0.47	0.64	0.32	0.00	0.62	0.67	0.80	0.61	0.57	0.72	0.61	0.67	0.79	0.77	0.59	0.95	0.93	0.87	0.83	0.97	0.92	0.80	0.78	0.76	0.81	0.85	0.79	0.88	0.87
Sg	0.52	0.46	0.43	0.27	0.37	0.33	0.52	0.09	0.24	0.00	0.27	0.59	0.51	0.33	0.46	0.30	0.41	0.65	0.50	0.78	0.67	0.76	0.70	0.62	0.79	0.97	0.81	0.95	0.62	0.66	0.77	0.82	0.75	0.76
Alb	0.50	0.36	0.38	0.23	0.27	0.26	0.43	0.11	0.26	0.10	0.00	0.41	0.39	0.32	0.31	0.23	0.32	0.44	0.42	0.63	0.76	0.62	0.57	0.48	0.53	0.97	0.75	0.90	0.72	0.83	0.80	0.85	0.94	0.93
Ch	0.62	0.42	0.43	0.23	0.24	0.26	0.43	0.18	0.32	0.21	0.16	0.00	0.46	0.46	0.50	0.47	0.63	0.64	0.71	0.85	0.92	0.50	0.83	0.76	0.67	0.98	0.93	0.99	0.80	0.84	0.89	0.88	0.93	0.93
Ta	0.56	0.48	0.48	0.28	0.35	0.35	0.50	0.22	0.21	0.14	0.12	0.15	0.00	0.30	0.45	0.35	0.54	0.55	0.47	0.54	0.83	0.68	0.75	0.64	0.64	0.98	0.86	0.94	0.70	0.80	0.67	0.77	0.86	0.85
Ce	0.49	0.40	0.39	0.19	0.29	0.27	0.44	0.14	0.17	0.09	0.09	0.13	0.06	0.00	0.33	0.18	0.43	0.51	0.41	0.68	0.82	0.78	0.80	0.70	0.76	0.91	0.84	0.89	0.66	0.74	0.76	0.76	0.79	0.79
CB	0.52	0.40	0.40	0.21	0.28	0.25	0.41	0.14	0.21	0.11	0.08	0.14	0.09	0.06	0.00	0.18	0.25	0.16	0.18	0.50	0.71	0.46	0.59	0.49	0.60	0.87	0.82	0.87	0.64	0.76	0.67	0.77	0.90	0.89
He	0.50	0.41	0.40	0.23	0.32	0.29	0.47	0.14	0.19	0.08	0.07	0.14	0.08	0.04	0.03	0.00	0.37	0.41	0.30	0.67	0.73	0.66	0.66	0.57	0.69	0.91	0.76	0.85	0.58	0.71	0.75	0.78	0.86	0.85
GV	0.56	0.45	0.45	0.26	0.33	0.30	0.48	0.16	0.21	0.11	0.09	0.18	0.12	0.08	0.05	0.08	0.00	0.26	0.28	0.57	0.70	0.58	0.65	0.57	0.67	0.89	0.77	0.80	0.63	0.82	0.74	0.77	0.94	0.93
PL	0.57	0.46	0.45	0.26	0.33	0.29	0.46	0.21	0.25	0.18	0.13	0.19	0.13	0.10	0.03	0.09	0.06	0.00	0.16	0.46	0.76	0.47	0.61	0.50	0.55	0.84	0.83	0.81	0.67	0.79	0.64	0.81	0.92	0.91
PS	0.52	0.45	0.44	0.24	0.33	0.31	0.47	0.19	0.23	0.14	0.12	0.20	0.10	0.08	0.04	0.06	0.06	0.04	0.00	0.51	0.57	0.68	0.64	0.48	0.68	0.81	0.83	0.84	0.65	0.73	0.57	0.78	0.88	0.87
Mu	0.63	0.54	0.54	0.35	0.42	0.40	0.54	0.29	0.24	0.25	0.21	0.29	0.16	0.17	0.13	0.18	0.16	0.14	0.15	0.00	0.71	0.62	0.62	0.53	0.67	0.91	0.95	0.91	0.86	0.92	0.66	0.91	0.97	0.96
SP	0.91	0.84	0.80	0.66	0.72	0.73	0.82	0.56	0.67	0.47	0.47	0.57	0.48	0.45	0.41	0.43	0.44	0.46	0.38	0.49	0.00	0.80	0.79	0.71	0.84	0.84	0.97	0.97	0.91	0.99	0.78	0.93	0.98	0.96
PM	0.67	0.53	0.52	0.35	0.38	0.36	0.51	0.27	0.39	0.29	0.25	0.23	0.23	0.23	0.15	0.21	0.20	0.17	0.22	0.26	0.57	0.00	0.49	0.47	0.59	0.98	0.94	0.99	0.70	0.89	0.79	0.91	0.92	0.91
BL	0.72	0.62	0.61	0.48	0.52	0.50	0.66	0.38	0.47	0.34	0.29	0.40	0.32	0.31	0.25	0.28	0.29	0.28	0.28	0.33	0.65	0.32	0.00	0.12	0.73	0.93	0.86	0.99	0.66	0.90	0.71	0.86	0.95	0.95
CC	0.64	0.54	0.53	0.35	0.41	0.40	0.58	0.29	0.37	0.25	0.21	0.31	0.22	0.22	0.16	0.19	0.20	0.18	0.17	0.23	0.56	0.25	0.11	0.00	0.71	0.93	0.87	1.00	0.61	0.82	0.53	0.87	0.94	0.94
Mal	0.84	0.73	0.73	0.58	0.62	0.62	0.73	0.49	0.59	0.44	0.33	0.42	0.35	0.37	0.31	0.36	0.37	0.32	0.36	0.41	0.75	0.43	0.57	0.49	0.00	0.99	0.91	0.99	0.90	0.94	0.81	0.83	0.98	0.97
Mar	0.77	0.70	0.68	0.50	0.58	0.56	0.69	0.48	0.48	0.42	0.41	0.44	0.38	0.34	0.32	0.35	0.36	0.35	0.33	0.41	0.67	0.48	0.55	0.48	0.64	0.00	0.89	0.77	0.96	0.99	0.99	0.97	0.99	1.00
TT	0.69	0.62	0.61	0.46	0.53	0.51	0.65	0.39	0.38	0.32	0.30	0.37	0.30	0.27	0.26	0.26	0.27	0.29	0.28	0.37	0.65	0.42	0.47	0.40	0.56	0.48	0.00	0.49	0.87	0.95	0.97	0.94	1.00	1.00
TF	0.81	0.75	0.73	0.58	0.65	0.64	0.73	0.57	0.54	0.50	0.47	0.53	0.46	0.43	0.41	0.43	0.43	0.43	0.43	0.50	0.76	0.57	0.64	0.58	0.70	0.58	0.42	0.00	0.98	1.00	1.00	0.94	1.00	1.00
PP	0.65	0.57	0.56	0.41	0.49	0.46	0.62	0.32	0.34	0.24	0.27	0.31	0.23	0.20	0.19	0.19	0.20	0.23	0.21	0.32	0.60	0.32	0.38	0.30	0.53	0.47	0.40	0.56	0.00	0.37	0.47	0.60	0.48	0.50
Cr	0.74	0.66	0.62	0.47	0.55	0.51	0.66	0.40	0.44	0.32	0.36	0.40	0.32	0.28	0.28	0.28	0.32	0.32	0.29	0.41	0.73	0.45	0.54	0.44	0.63	0.56	0.49	0.65	0.25	0.00	0.42	0.64	0.41	0.43
Sc	0.83	0.75	0.73	0.54	0.62	0.62	0.73	0.48	0.52	0.41	0.40	0.47	0.33	0.34	0.31	0.35	0.36	0.33	0.29	0.38	0.73	0.47	0.53	0.39	0.65	0.62	0.55	0.69	0.35	0.41	0.00	0.68	0.69	0.68
SE	0.73	0.64	0.65	0.48	0.55	0.54	0.66	0.42	0.39	0.33	0.34	0.37	0.29	0.26	0.26	0.27	0.28	0.30	0.28	0.37	0.65	0.42	0.49	0.41	0.55	0.51	0.45	0.59	0.32	0.41	0.47	0.00	0.64	0.58
TU	0.89	0.81	0.77	0.61	0.69	0.69	0.78	0.55	0.62	0.47	0.50	0.54	0.46	0.41	0.44	0.45	0.49	0.48	0.46	0.54	0.84	0.58	0.67	0.60	0.76	0.69	0.63	0.75	0.42	0.49	0.68	0.53	0.00	0.02
Ot	0.90	0.82	0.77	0.61	0.69	0.69	0.79	0.55	0.62	0.46	0.49	0.54	0.45	0.41	0.43	0.44	0.48	0.47	0.45	0.54	0.85	0.58	0.67	0.60	0.76	0.69	0.62	0.75	0.43	0.50	0.67	0.50	0.09	0.00

Supplementary S3.2. Pairwise FST (lower left) and Jost's D (upper right) values of genetic distance.

Supplementary S3.3. Allele presence and frequency for the six loci of *Cystoseira* spp. populations analysed here. Sites are arranged by geographical clusters identify with STRUCTURE at K=7. For each site, a circle indicates that the corresponding allele was present; its diameter represents the frequency of that allele in the sample. These results were obtained using the MsatAllele R package (http://CRAN.R-project.org/package=MsatAllele)

Ct 2.7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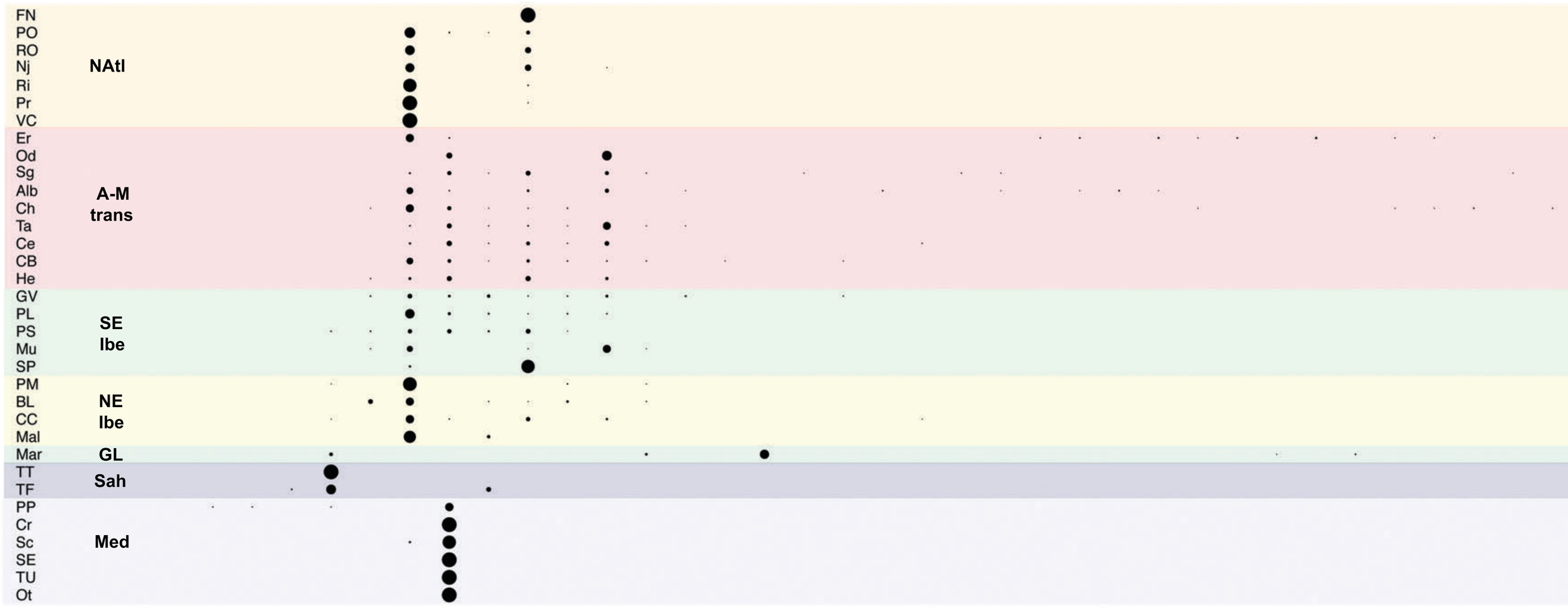
Alleles

Ct 2.8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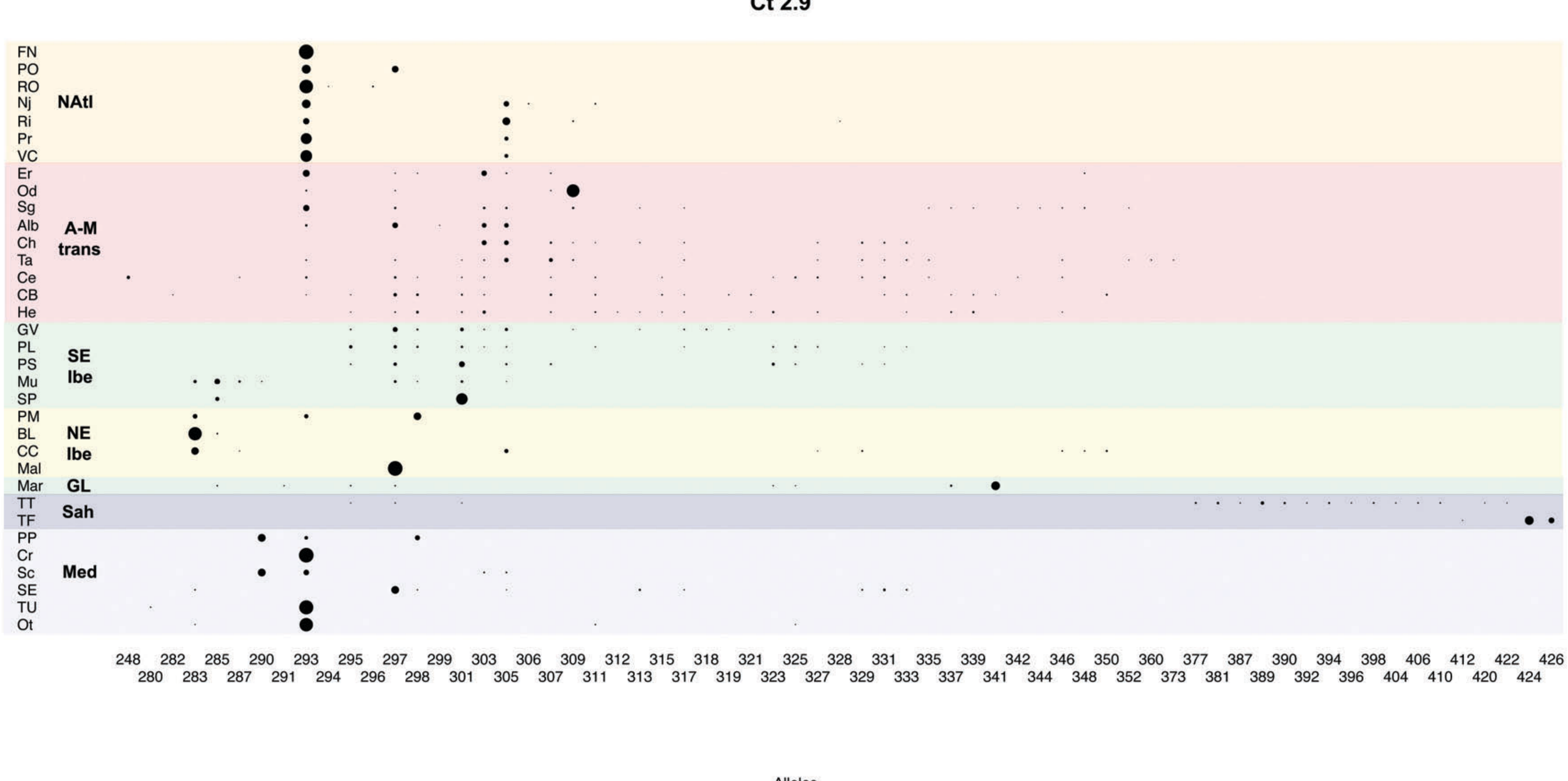
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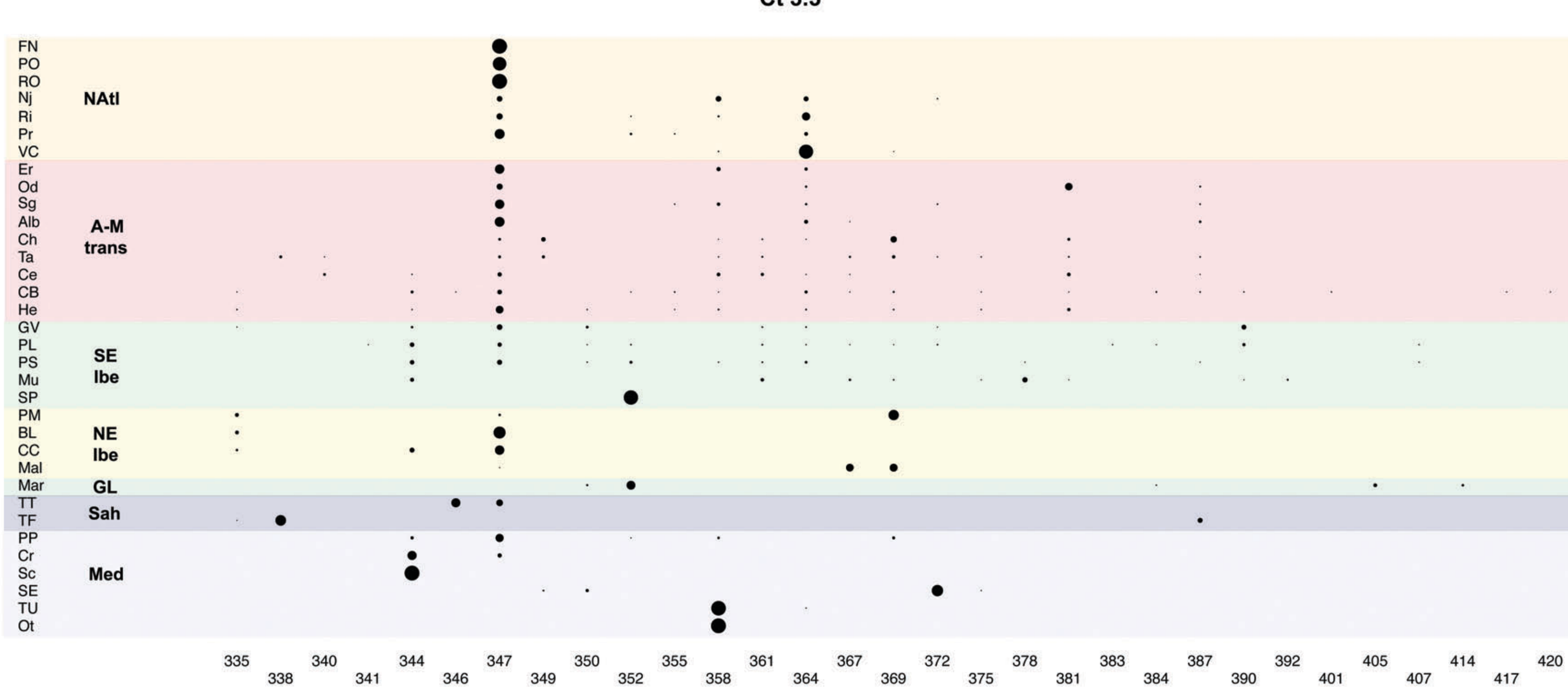
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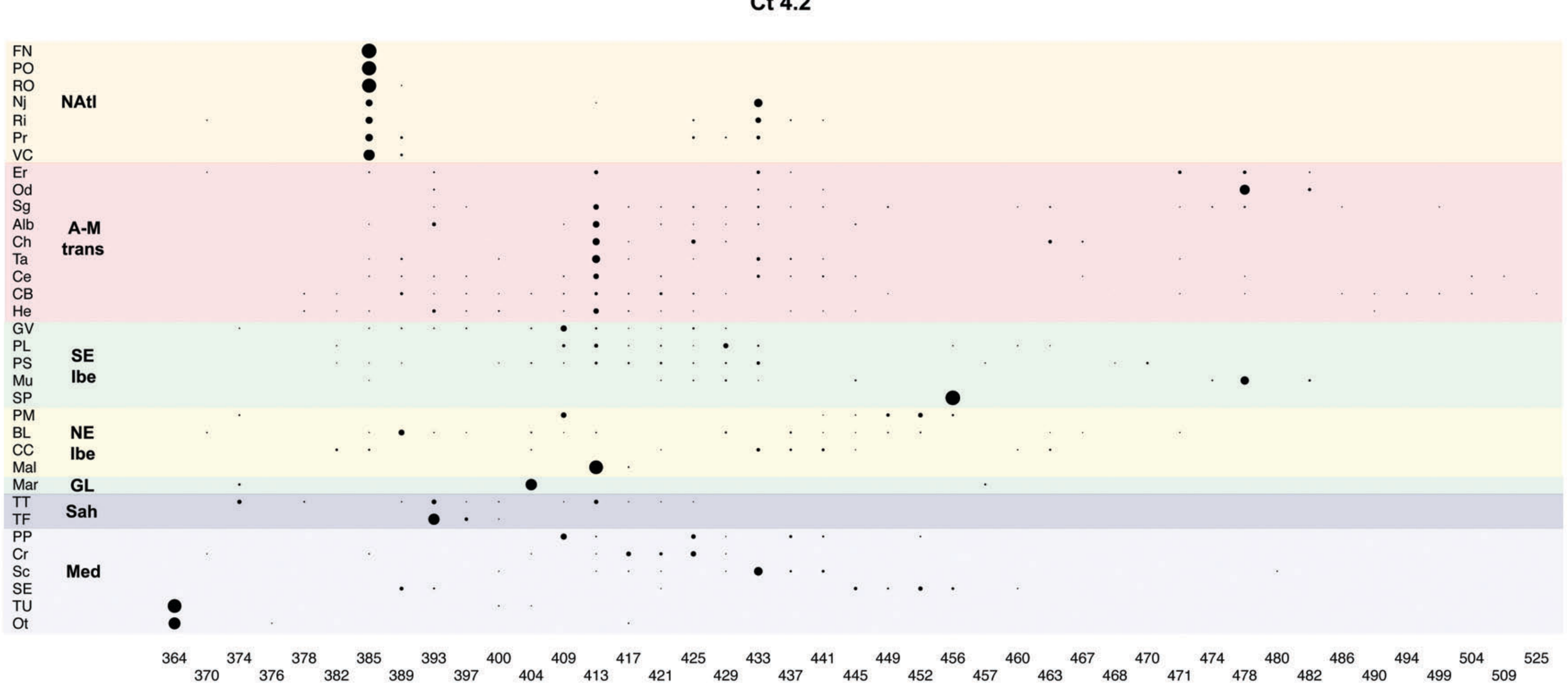
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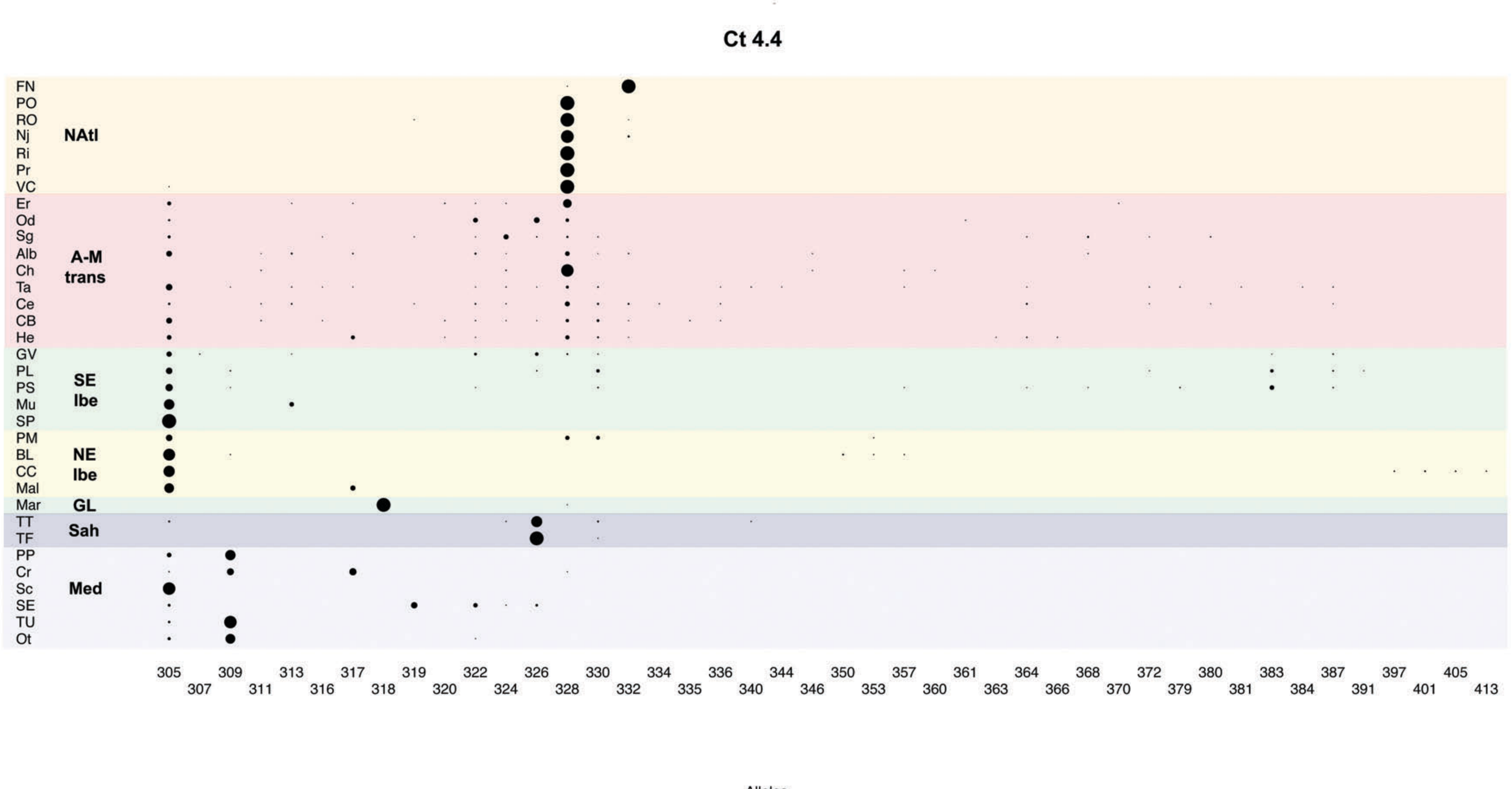
Alleles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Alleles

Supplementary S4

D Jost			$F_{ST}/(1-F_{ST})$		
	df	F-values		df	F-values
Distance	1	6.695*	Distance	1	20.650***
Region	2	41.07***	Region	2	41.445***
DistancexRegion	2	0.157	DistancexRegion	2	2.216
Residuals	38		Residuals	38	
Post hoc:			Post hoc:		
Med > A-M trans >Med			Med > Med > A-M trans		

Table S4.- Final analysis of covariance (ANCOVA) model and post hoc results for the effect of shortest sea distance (km) and Region on Jost's D and $F_{ST}/(1-F_{ST})$ genetic distances between populations from the Mediterranean, Atlantic-Mediterranean transition and North Atlantic genetic clusters (see Fig. 2 in the article). To ensure that geographic distance and genetic distances are independent variables, and to avoid any possible bias, just pairwise comparisons in a sea distance range between 150 and 700 km were considered to perform the ANCOVA.

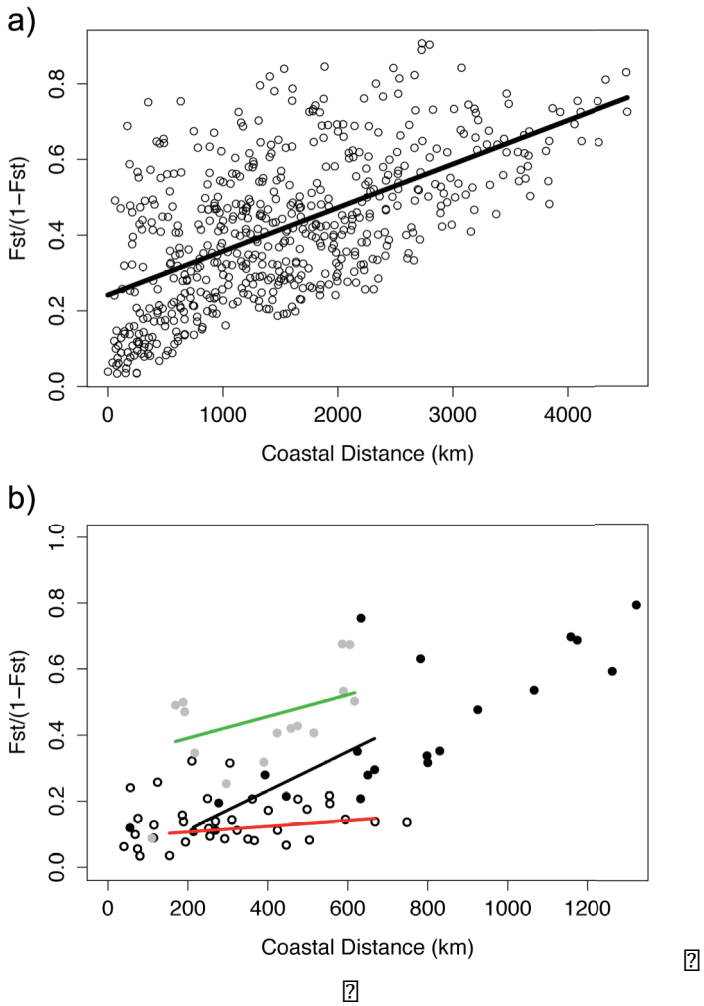

Fig S4.- Relationship between $F_{ST}/(1-F_{ST})$ genetic distance and the shortest sea distance considering: (a) all populations (mantel R = 0.593; p-value<0.001); (b) only North Atlantic (black dots and black line; mantel R = 0.829; p-value <0.001), Atlantic-Mediterranean transition (White dots and red line; mantel R = 0.141; p-value > 0.05) and Mediterranean populations (grey dots and green line; mantel R = 0.596; p-value<0.05). Jost's D results were showed in the article instead $F_{ST}/(1-F_{ST})$, because Jost's D is more appropriate to compare populations with contrasting levels of diversity.