Table S1. List of surface sediment samples collected in the Mediterranean Sea, locality, date, geographical coordinates and depth.

Sample	Locality	Date (dd/mm/yyyy)	Latitude (DD.ddddd)	Longitude (DD.ddddd)	Depth
MS15_09	Bari	07/11/2015	41.30400	16.92400	115
MS15_55	Gargano Sud	10/11/2015	41.51400	16.32300	35
MS15_56	Gargano Sud	11/11/2015	41.55400	16.23000	22
MS15_120	Leuca	16/11/2015	39.54720	18.40894	600
MS15_121	Leuca	16/11/2015	39.57741	18.40133	515
MS15_137	Gallipoli Sud	17/11/2015	39.78800	17.97600	94
MS15_145	Crotone	18/11/2015	39.58355	16.87080	65
MS15_147	Crotone	18/11/2015	39.48957	17.10846	155
MS15_151	Crotone	18/11/2015	39.34500	17.19500	227
MS15_152	Crotone	18/11/2015	39.25500	17.15800	188
MS15_159	Crotone	18/11/2015	38.99725	17.25582	321
MS15_161	Crotone	19/11/2015	38.90300	17.20400	225
MS15_174	Crotone	19/11/2015	38.87500	17.14000	180
MS15_175	Crotone	19/11/2015	38.76444	16.89083	564
MS15_181	Roccella Ionica	20/11/2015	38.34500	16.52400	85
MS15_196	Augusta	21/11/2015	37.24600	15.28200	81
MS15_185	Riace	20/11/2015	38.34600	16.50300	51
MS16_I_155	Orosei	28/07/2016	40.26010	9.90530	446
MS16_I_157	Orosei	28/07/2016	40.20990	9.67640	276
MS16_I_166	Nora Canyon	30/07/2016	38.80270	8.92530	123
MS16_I_168	Nora Canyon	30/07/2016	38.65490	9.04010	495
MS16_II_28	Gela	04/09/2016	36.91869	14.27464	70
MS16_II_31	Gela	04/09/2016	36.83518	14.20068	415
MS16_II_34	Gela	04/09/2016	36.75515	14.12661	766
MS16_II_44	Augusta	04/09/2016	37.22468	15.29330	128
MS16_II_76	Porto Cesareo	09/09/2016	40.25050	17.46871	189
MS16_II_77a	Porto Cesareo	09/09/2016	40.15356	17.40916	930
MS16_II_77b	Porto Cesareo	09/09/2016	40.15356	17.40916	930
MS16_II_104a	S. Maria Leuca	11/09/2016	39.80009	18.64332	145
MS16_II_104b	S. Maria Leuca	11/09/2016	39.80009	18.64332	145

MS16_II_108a	S. Maria Leuca	11/09/2016	39.82152	18.52397	125
MS16_II_108b	S. Maria Leuca	11/09/2016	39.82152	18.52397	125
MS16_II_119a	S. Cataldo	12/09/2016	40.50142	18.60157	270
MS16_II_119b	S. Cataldo	12/09/2016	40.50142	18.60157	270
MS16_II_121a	S. Cataldo	12/09/2016	40.43669	18.41888	104
MS16_II_121b	S. Cataldo	12/09/2016	40.43669	18.41888	104
MS16_III_26	Bari	09/12/2016	41.33600	17.19300	608
MS16_III_34	Bari	09/12/2016	41.11800	17.05900	82
MS16_III_41	Barletta	09/12/2016	41.42500	16.37500	39
MS16_III_45	Barletta	10/12/2016	41.50600	16.66800	99
MS16_III_49	Vieste (DART)	10/12/2016	42.24300	15.92300	119
MS16_III_65	Vieste (DART)	10/12/2016	42.02200	15.86900	45
MS16_III_69	Pescara	11/12/2016	42.55100	14.45200	91
MS16_III_72	Pescara	11/12/2016	42.51900	14.40000	75
MS16_III_75	Pescara	11/12/2016	42.47600	14.33400	26
MS16_III_79	S. Benedetto Tronto	11/12/2016	42.80600	14.07500	22
MS16_III_85	S. Benedetto Tronto	11/12/2016	42.88800	14.18600	73
MS16_III_90	Civitanova Marche	11/12/2016	43.29100	14.03800	55
MS16_III_96	Civitanova Marche	11/12/2016	43.20800	13.89000	17
MS16_III_102	Senigallia	12/12/2016	43.78600	13.31800	20
MS16_III_110	Senigallia	12/12/2016	43.85800	13.42200	49
MS16_III_113	Ancona	12/12/2016	43.72100	13.77100	65
MS16_III_122	Ancona	12/12/2016	43.61900	13.65000	1
MS16_III_126	Rimini	14/12/2016	44.22400	12.68900	22
MS16_III_129	Rimini Boa E1	14/12/2016	44.14000	12.57000	10
MS16_III_147	MEDA S1	15/12/2016	44.73500	12.45400	22
MS16_III_155	Delta Po	16/12/2016	44.64000	12.60000	31
MS16_III_157	Chioggia	16/12/2016	44.98500	12.62100	30
MS16_III_165	Chioggia	16/12/2016	45.00100	12.80700	33
MS16_III_184	Trieste	16/12/2016	45.65700	13.65900	23
MS17_I_83_C	Golfo di Napoli	12/07/2017	40.73183	14.23217	187
MS17_I_84_C R1	Golfo di Napoli	12/07/2017	40.77417	14.16750	127
MS17_I_88_C R2	Golfo di Napoli	12/07/2017	40.77133	14.17750	180
MS17_I_90_C R1	Golfo di Napoli	12/07/2017	40.75163	14.15683	120

MS17_I_94_C R2	Golfo di Napoli	12/07/2017	40.75733	14.11500	95
MS17_I_107_C R2	Isole Pontine	14/07/2017	40.91117	12.88283	65
MS17_I_111_C	Anzio	14/07/2017	41.38317	12.68417	41
MS17_I_114_C R2	Elba Nord	15/07/2017	43.01733	10.34417	110
MS17_I_118_C R2	Elba Nord	15/07/2017	43.02167	10.47183	58
MS17_I_120_C	Livorno	15/07/2017	43.46717	10.24817	66
MS17_I_125_C R2	Gorgona	16/07/2017	43.40917	9.91250	111
MS17_I_131_C	Capraia	16/07/2017	43.00000	9.79050	110
MS17_I_142_C	Anzio	18/07/2017	41.37867	12.42800	139
MS17_II_008_C R2	Egadi	22/07/2017	37.96283	12.14567	171
MS17_II_025_C R1	Porto Empedocle	24/07/2017	37.12983	13.34650	440
MS17_II_032_C R1	Gela	24/07/2017	36.75850	14.13350	781
MS17_II_035_C R1	Gela	24/07/2017	36.83633	14.20133	418
MS17_II_039_C R2	Gela	24/07/2017	36.92000	14.28000	70
MS17_II_041_C R1	Augusta	25/07/2017	37.22983	15.30000	130
MS17_II_066_C R1	Soverato	26/07/2017	38.68000	16.61983	692
MS17_II_089_C R3	Amendolara	28/07/2017	39.93067	16.70850	99
MS17_II_096_C R2	Amendolara	29/07/2017	39.93183	16.71083	81
MS17_II_108_C R1	Campomarino	30/07/2017	40.13283	17.41517	1018
MS17_II_143_C R1	Bari	02/08/2017	41.25650	17.14750	151
MS17_II_145_C R1	Bari	02/08/2017	41.18833	17.10900	125
MS17_II_147_C R1	Bari	03/08/2017	41.11833	17.06467	84
MS17_II_167_C R2	Brindisi	04/08/2017	40.76633	17.93650	106
MS17_II_169_C R1	Brindisi	04/08/2017	40.76217	17.94350	106
MS17_II_172_C R1	Brindisi	04/08/2017	40.76500	17.93883	106
MS17_II_176_C R2	Brindisi	04/08/2017	40.76367	17.94117	106
MS17_III_33	Lesina	09/08/2017	42.03683	15.31517	68
MS17_III_76	Chioggia	11/08/2017	44.99183	12.69167	32
MS17_III_81	Venezia	11/08/2017	45.29100	12.57217	24
MS17_III_94	Trieste	12/08/2017	45.62450	13.47083	21

Table S2. List of microalgal species used to test the new primer specificity.

Species	Strain	Class	Medium
Alexandrium andersoni	VGO664	Dinophyceae	f/2 - Si
Alexandrium mediterraneum	CNR-AT4	Dinophyceae	f/2 - Si
Alexandrium minutum	CBA57	Dinophyceae	f/2 - Si
Alexandrium pacificum	CNR-SRA4	Dinophyceae	f/2 - Si
Coolia monotis	CBA1	Dinophyceae	f/4 - Si
Gonyaulax spinifera	CBA5	Dinophyceae	f/4 - Si
Gymnodinium impudicum	GY6V	Dinophyceae	L1 - Si
Heterocapsa sp.	CBAB/D5	Dinophyceae	f/2 - Si
Heterocapsa triquetra	RCC4813	Dinophyceae	L1 - Si
Lingulodinium polyedrum	LPA0510	Dinophyceae	f/10 - Si
Ostreopsis cf. *ovata*	CBA1291	Dinophyceae	f/4 - Si
Prorocentrum lima	CBA3	Dinophyceae	f/4 - Si
Protoceratium reticulatum	PRA0311	Dinophyceae	f/4 - Si
Scrippsiella trochoidea	CBA3	Dinophyceae	f/2 - Si
Fibrocapsa japonica	CBA1	Raphidophyceae	f/2 - Si
Heterosigma akashiwo	HA2V	Raphidophyceae	L1
Chaetoceros socialis	CBA22	Bacillariophyceae	f/2
Cylindroteca sp.	CBA7	Bacillariophyceae	f/2
Coscinodiscus sp.	CBA1	Bacillariophyceae	f/2
Ditylum brightwellii	CBA2	Bacillariophyceae	f/2
Guinardia flaccida	CBA1	Bacillariophyceae	f/2
Pseudo-nitzschia cf. *arenysensis*	CBA163	Bacillariophyceae	f/2
Pseudo-nitzschia calliantha	CBA189	Bacillariophyceae	f/2
Pseudo-nitzschia pungens	CBA180	Bacillariophyceae	f/2
Skeletonema marinoi	CBA4	Bacillariophyceae	f/2
Thalassionema sp.	CBA10	Bacillariophyceae	f/2
Thalassiosira sp.	CBA3	Bacillariophyceae	f/2

Table S3. List of species-specific primer sequences of various diatom and dinoflagellate taxa targeting different rDNA regions, qPCR reagent concentrations, amplicon melting temperature (Tm) and size.

Taxon	Primer name	Forward primer sequence (5′–3′) Reverse primer sequence (5′–3′)	Primer concentration [nM]	$MgCl_2$ [mM]	Amplicon Tm (°C)	Amplicon size (bp)	Primer locations	Reference
Chaetoceros socialis	CsocF CsocR	5′-GGAGCGTCTGAGTATGGTCGT-3′ 5′-GCCATCCAGACGCAAAAGTGT-3′	300	3.5	83.98	100	ITS2 (5′ → 3′) ITS2 (3′ ← 5′)	This study
Ditylum brightwellii	DbrighF DbrighR	5′-ACCGCGTGTCGCTTATGTAT-3′ 5′-ACCAAACGACTTTCGGGGTT-3′	300	2.5	83.46	103	ITS1 (5′ → 3′) ITS1 (3′ ← 5′)	This study
Skeletonema spp.	SkelsppF SkelsppR	5′-CGATACACTGGTAGCGAGCC-3′ 5′-TTCAGTTCGGTAATGGGCGG-3′	200	1.5	86.52	118	ITS1 (5′ → 3′) ITS1 (3′ ← 5′)	This study
Thalassiosira spp.	ThalsppF ThalsppR	5′-AGCGGGAAGAGCTCACCAT-3′ 5′-AAGAGACTTGGCCCGGGAAC-3′	200	2.5	84.35	99	LSU (5′ → 3′) LSU (3′ ← 5′)	This study
Pseudo-nitzschia spp.	Pseudo 5′ Pseudo 3′	5′-CGATACGTAATGCGAATTGCAA-3′ 5′-GTGGGATCCRCAGACACTCAGA-3′	300	1.5	82.10	111	5.8S (5′ → 3′) 5.8S (3′ ← 5′)	Penna et al. (2007)
Bacillariophyceae	1209f Diatom18SR1	5′-CAGGTCTGTGATGCCCTT-3′ 5′-CAATGCAGWTTGATGAWCTG-3′	300	3.5	84.95	189	SSU (5′ → 3′) SSU (3′ ← 5′)	Giovannoni et al. (1988) Godhe et al. (2008)
Alexandrium minutum	ITS1m 5.8S 3′	5′-CATGCTGCTGTGTTGATGACC-3′ 5′-GCAMACCTTCAAGMATATCCC-3′	200	2.5	82.0	212	ITS1 (5′ → 3′) 5.8S (3′ ← 5′)	Galluzzi et al. (2005)
A. pacificum	ITS1c 5.8S-3′	5′-AGCATGATTTGTTTTTCAAGC-3′ 5′-GCAMACCTTCAAGMATATCCC-3′	200	2.5	81.90	226	ITS1 (5′ → 3′) 5.8S (3′ ← 5′)	Penna et al. (2007)
A. tamarense/A. mediterraneum	5.8S 5’ ITS2t	5′- TGTTACTTGTACCTTTGGGA-3′ 5′-ACAACACCCAGGTTCAAT-3′	300	2.5	81.34	134	5.8S (5′ → 3′) ITS2 (3′ ← 5′)	Penna et al. (2007)
Gonyaulax spinifera	GspinF_for	5′-GAAACTCCTTCTGTGGATGC-3′	200	3.5	82.90	154	LSU (5′ → 3′)	Perini et al. (2019)

	GspinF_for	5′-TCACAGTTCCCTCATGGTACT-3′					LSU (3′ ← 5′)	
Gymnodinium spp.	GymnosppF	5′-CAGCGACGGATGTCTCGGTT-3′	200	2.5	84.20	102	5.8S (5′ → 3′)	This study
	GymnosppR	5′-TGCGTTCAAGTTTCTGTCGGT-3′					5.8S (3′ ← 5′)	
Heterocapsa triquetra	HtriF	5′-ATCCTCCTTGCGAGGGTTGG-3′	200	2.5	86.27	73	ITS2 (5′ → 3′)	This study
	HtriR	5′-AATGCGTCAGGGGTGGGAAA-3′					ITS2 (3′ ← 5′)	
Lingulodinium polyedrum	Lpoly RT-for	5′-AACTCGTTGGCGAGCATTTT-3′	400	2.5	81.90	58	ITS2 (5′ → 3′)	Perini et al. (2019)
	Lpoly RT-rev	5′-CGCTAGCAAAGCACTCGCTTA-3′					ITS2 (3′ ← 5′)	
Protoceratium reticulatum	Pret RT-for	5′-GGTGCAGTGAAATGTATTAGGCATT-3′	200	2.5	78.90	83	5.8S (5′ → 3′)	Perini et al. (2018)
	Pret RT-rev	5′-TCCCAAAAACATAGAATACGTTCAAT-3′					5.8S (3′ ← 5′)	
S. trochoidea-species complex	StcompF	5′-GGGTGTGCTTGTGCGTCAAA-3′	200	1.5	83.39	81	ITS2 (5′ → 3′)	This study
	StcompR	5′-AGCAAGTTGTGCGCCAAGAG-3′					ITS2 (3′ ← 5′)	
Heterosigma akashiwo	HaF	5′-CCGACGGGCGTGGTAGC-3′	200	2.5	83.47	73	ITS2 (5′ → 3′)	Yuan et al. (2015)
	HaR	5′-TCCTCTGTCAGAACAACCGAAGT-3′					ITS2 (3′ ← 5′)	
Dinophyceae	EUK528f	5′-CCGCGGTAATTCCAGCTC-3′	300	3.5	84.07	110	SSU (5′ → 3′)	Elwood et al. (1985)
	Dino18SR1	5′-GAGCCAGATRCDCACCCA-3′					SSU (3′ ← 5′)	Lin et al. (2006)

Table S4. List of the environmental variables used in the RDA and MRT. All variables were used to train the MRT, while only those with short names followed by an asterisk (VIF<10) were used in the RDA.

No.	Name of the variable (mean value if not specified)	Source	Short name
1	Sampling depth	Field data from sampling	Depth*
2	Bottom salinity	Boyer et al. (2005)	Botsal*
3	Bottom temperature	Boyer et al. (2005)	Bottemp*
4	Calcite concentration	Feldman & McClain (2010) Tyberghein et al. (2012)	Calcite
5	Chlorophyll *a* concentration (mean)	Feldman & McClain (2010) Tyberghein et al. (2012)	Chl
6	Chlorophyll *a* concentration (annual range)	Feldman & McClain (2010) Tyberghein et al. (2012)	Chl range
7	Euphotic depth	Feldman & McClain (2010)	Zeumean
8	Human impact to marine ecosystems	Halpern et al. (2008)	Impact*
9	Nitrate concentration	Feldman & McClain (2010) Tyberghein et al. (2012)	NO_3
10	Nutrient input (fertilizers)	Halpern et al. (2008)	Nutrients
11	Ocean acidification	Halpern et al. (2008)	Acid*
12	Phosphate concentration	Boyer et al. (2009) Tyberghein et al. (2012)	PO_4*
13	Pollutants (inorganic)	Halpern et al. (2008)	Inorg
14	Pollutants (organic)	Halpern et al. (2008)	Orgpol
15	Salinity	Boyer et al. (2009) Tyberghein et al. (2012)	Sal
16	Sea surface temperature (mean)	Feldman & McClain (2010) Tyberghein et al. (2012)	Sst
17	Sea surface temperature (annual range)	Feldman & McClain (2010) Tyberghein et al. (2012)	Sstrange
18	Shipping intensity	Halpern et al. (2008)	Shipping*
19	Silicate concentration	Boyer et al. (2009) Tyberghein et al. (2012)	Silic
20	Seafloor slope	GIS based on bathymetry	Slope*
21	Otter trawling	VMS based estimation (see Russo et al. 2014)	Fishing*

Table S5. Mean standard curves, mean Ct cell^{-1}, rDNA copy number cell^{-1} and efficiency obtained from diatom and dinoflagellate cultured strains by qPCR assay.

Taxon	Mean standard curves	Mean Ct cell^{-1} ± SD[a]	rDNA copy number cell^{-1} ± SD[b]	Efficiency (%)
Chaetoceros socialis	y = -3.4696x + 38.441	34.27 ± 0.29	15 ± 2	94
Ditylum brightwellii	y = -3.4302x + 34.205	25.48 ± 0.13	396 ± 140	96
Skeletonema spp.	y = -3.4549x + 34.536	29.92 ± 0.10	25 ± 5	95
Thalassiosira spp.	y = -3.549x + 34.422	25.35 ± 0.17	344 ± 38	91
A. tamarense/A. mediterraneum	y = -3.3218x + 36.286	22.51 ± 0.03	14676 ± 1662	100
Gymnodinium spp.	y = -3.377x + 35.00	26.41 ± 0.14	412 ± 58	98
Heterocapsa triquetra	y = -3.5951x +35.62	25.71 ± 0.13	570 ± 53	90
S. trochoidea species complex	y = -3.5916x + 38.039	24.33 ± 0.13	5625 ± 1375	90
Heterosigma akashiwo[c]	y= -3.406x + 38.253	n.d.	367 ± 12	97

[a]mean Ct values measured in 2 independent experiments ± standard deviation (SD)
[b]mean rDNA gene copy number calculated in triplicate ± standard deviation (SD)
[c]from Yuan et al. (2015)
n.d., not determined

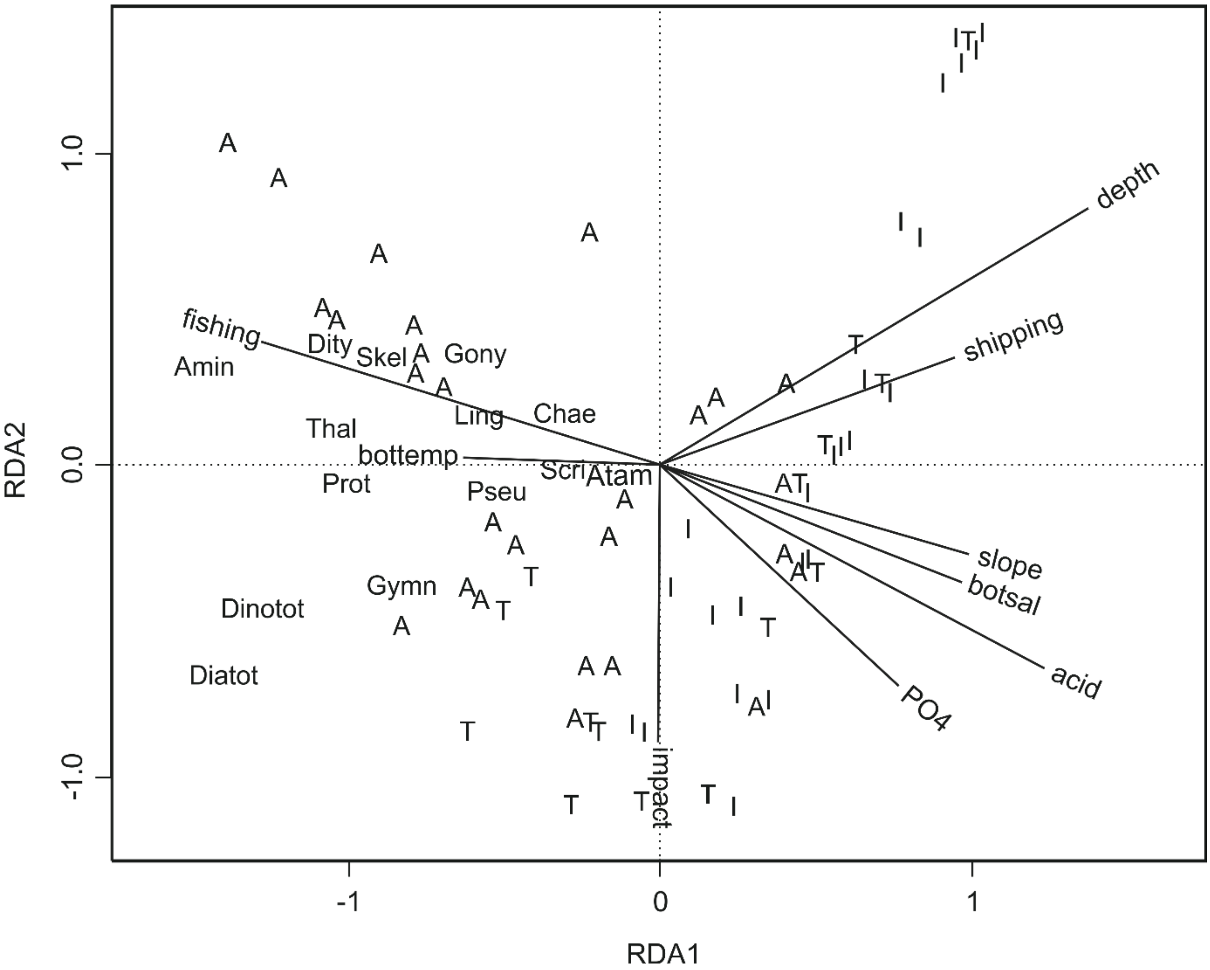

Figure S1. Redundancy Analysis triplot: samples are labelled according to the basin where they were collected (A = Adriatic Sea, I = Ionian Sea and T = Tyrrhenian Sea), while short names are shown for taxa and for environmental variables (solid lines). Full names can be found in Table 1. All the resting stages point towards the positive end of the first canonical axis (RDA1), thus showing that samples that are found in that semi-plane are the most abundant. The latter include almost all the samples from the Adriatic Sea. Short names for the resting stage taxa can be found in Table 1 next to the full names, while those for the environmental variables are shown in the rightmost column in Table S4. Three resting stage taxa (Hets, Apac and Hetc) whose coordinates were very close to the origin and therefore very poorly represented, are not shown in the ordination. The first two canonical axes, both significant to the permutation test, accounted for a very large share of variance, i.e. 49.88% (RDA1 44.27% and RDA2 5.59%).

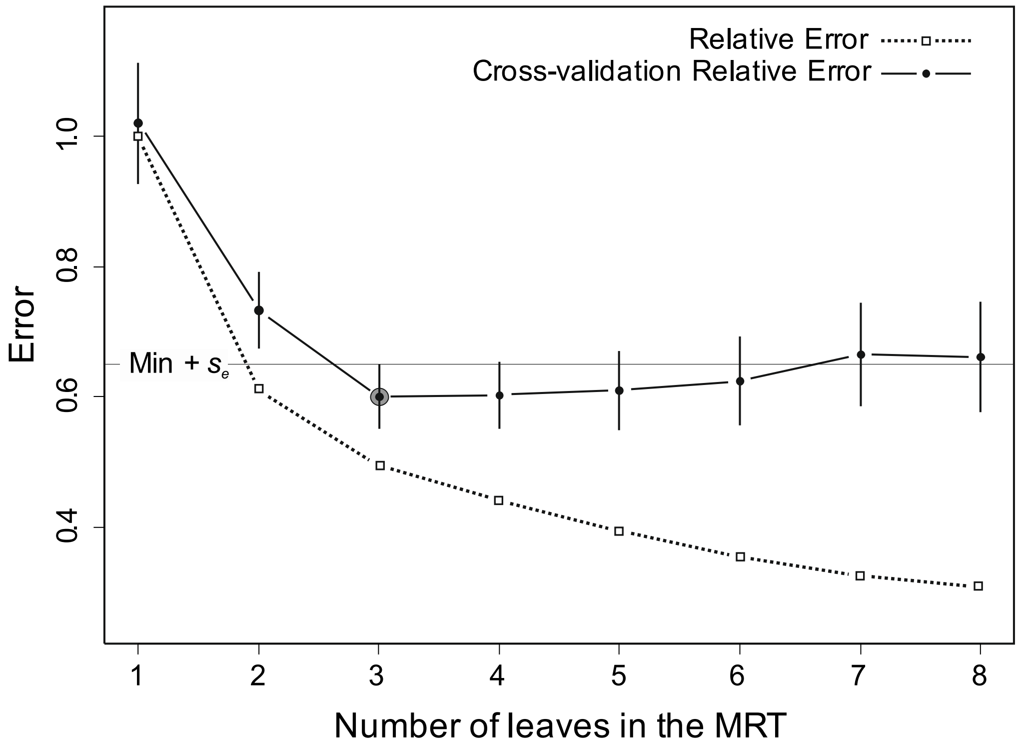

Figure S2. Relative error (dotted line) and cross-validation relative error (solid line) for the Multivariate Regression Tree (MRT). The best MRT has three leaves and corresponds both to the minimum cross-validation error and to the most parsimonious solution among those within one standard error above the optimum (i.e. below the solid horizontal line).

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