Range expansion of the Bluetongue vector, *Culicoides imicola*, in continental France likely due to rare wind-transport events

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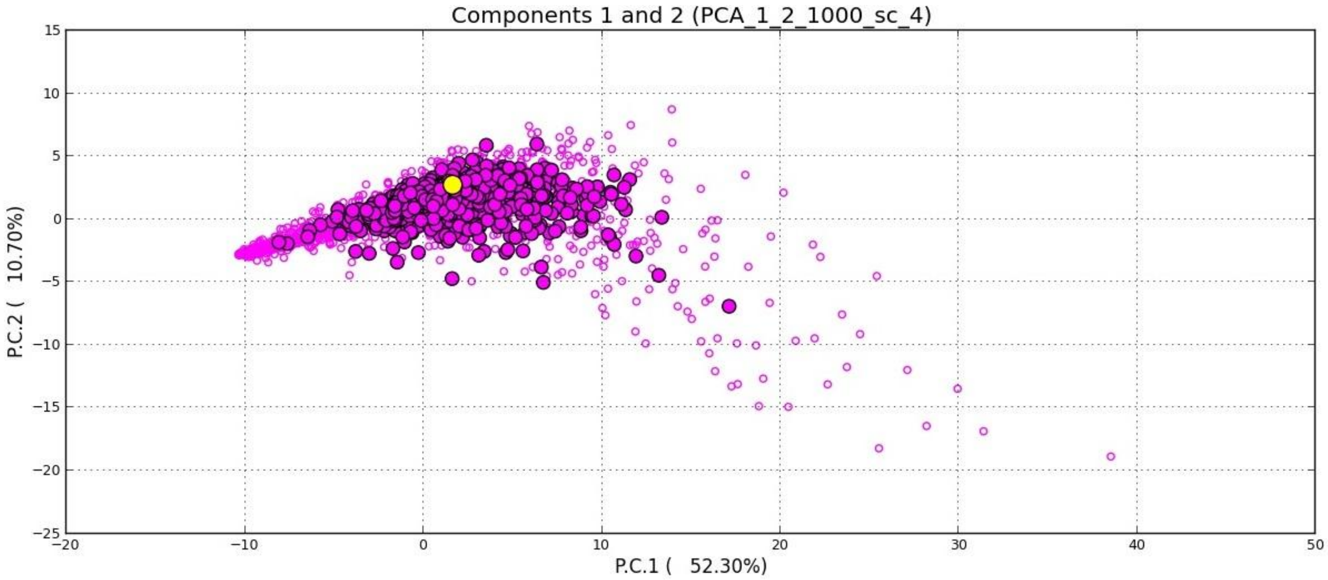
Supplementary Information



Supplementary Figure 1 | Principal component analysis (PCA) in the space of summary statistics performed under the most probable scenario (Scenario 4). The observations are the simulated datasets and the variables are the summary statistics. Small pink circles correspond to simulated statistics with parameters drawn from the prior distributions, the large pink dots correspond to the simulated datasets drawn from the posteriors distributions and the yellow dot represents the real *C. imicola* data set.

Supplementary Video clip 1 | NAME simulations of individual trajectories. Full 36-hour back-trajectories for all particles together are presented for each day during the full observation period. Maps were generated using ArcGIS software v10.2.2 (ESRI, Redlands, CA).

Supplementary Table 1 | Details of the entomological surveys realized in Spain and in France from 2008 and 2012.

		2008					2009					2010					2011					2012				
				Maximum *C. imicola* collection					Maximum *C. imicola* collection					Maximum *C. imicola* collection					Maximum *C. imicola* collection					Maximum *C. imicola* collection		
code	Location	Nb night collection during the year	Total nb of *Culicoides* collected	Collection date (DD /MM/YY)	Nb of *C. imicola*	Nb of other *Culicoides* collected	Nb night collection (DD /MM/YY)	Total nb of *Culicoides* collected	Collection date (DD /MM/YY)	Nb of *C. imicola*	Nb of other *Culicoides* collected	Nb night collection during the year	Total nb of *Culicoides* collected	Collection date (DD /MM/YY)	Nb of *C. imicola*	Nb of other *Culicoides* collected	Nb night collection during the year	Total nb of *Culicoides* collected	Collection date (DD /MM/YY)	Nb of *C. imicola*	Nb of other *Culicoides* collected	Nb night collection during the year	Total nb of *Culicoides* collected	Collection date (DD /MM/YY)	Nb of *C. imicola*	Nb of other *Culicoides* collected
Monitoring sites																										
Spain																										
23	Aramunt	52	590	-	0	-	52	375	02/07/2009	2	5	52	7,594	-	0	-	52	6,801	25/08/2011	1	38	52	3,188	-	0	-
26	Caldes	52	30,905	07/08/2008	129	699	52	9,801	10/09/2009	1,464	37	52	6,786	16/09/2010	11,627	358	52	22,817	01/09/2011	6,344	194	52	1,705	11/10/2012	739	0
31	Garcia	52	297	-	0	-	52	163	03/09/2009	1	0	52	184	20/05/2010	1	18	52	843	-	0	-	52	236	-	0	-
29	Piera	52	1,303	11/09/2008	1	28	52	2,001	08/10/2009	4	340	52	50,026	09/09/2010	7	5,207	52	8,551	03/11/2011	1	214	26	729	-	0	-
25	Susqueda	52	24,702	-	0	-	52	17,904	16/07/2009	296	4,654	52	40,123	01/07/2010	283	4,250	52	26,218	26/05/2011	279	4,103	26	3,604	24/05/2012	2	420
24	Avia	52	30	-	0	-	52	729	-	0	-															
32	Montbrio	52	10	-	0	-	52	64	-	0	-	19	3	29/04/2010	1	2										
27	Alguaire	52	29	-	0	-	52	110	-	0	-															
33	Amposta	52	76	-	0	-	52	667	31/12/2009	1	16															
30	Bonastre	52	44	21/08/2008	4	4	52	63	15/10/2009	24	12															
28	Sant Iscle	52	25,763	-	0	-	52	2,391	15/10/2009	3	54															

22	Vilanova Muga	52	6,486	27/03/2008	1	13	52	7,887	-	0	-															
France																										
9	**St Jean Pla de Corts**	20	144	**07/10/2008**	**3**	20																				
2	Vebre						25	8,972	-	0	-	26	23,788		0		25	2,305		0		27	1,155		0	
1	Clermont						23	740	-	0	-	26	446		0		26	5,306		0		27	3,069		0	
5	Argeles Sur Mer						23	3,080	-	0	-															
4	Argeles Sur Mer											25	92		0		24	81		0		21	62		0	
3	Toreilles	21	229	-	0	-																				
Sentinel sites																										
France																										
6	Vives						1	7	19/10/2009	0	7															
10	Ceret						1	16	19/10/2009	0	16															
18	**Reynes**						1	1	19/10/2009	0	1	1	408	11/10/2010	0	408	1	105	18/10/2011	0	105	1	466	**25/09/2012**	**8**	466
15	**Ceret**											1	186	11/10/2010	0	186	1	194	18/10/2011	0	194	1	2	**26/09/2012**	**3**	2
17	Amélie Les Bains Palalda						1	1	19/10/2009	0	1															
7	Taillet						1	1	19/10/2009	0	1															
21	**Maureillas** Las Illas											1	0	11/10/2010	0	0	1	109	18/10/2011	0	109	1	10	**25/09/2012**	**11**	10
20	Maureillas Las Illas											1	6	11/10/2010	0	6	1	0	18/10/2011	0	0	1	20	25/09/2012	0	20
14	**Albere**											1	6	11/10/2010	1	6	1	3	18/10/2011	0	3	1	1	**26/09/2012**	**2**	1

19	**Cerbere**			1	0	21/10/2009	0	0	1	5	14/10/2010	0	5	1	3	19/10/2011	0	3	1	9	**24/09/2012**	**3**	9
16	Banyuls			1	0	19/10/2009	0	0	1	2	14/10/2010	0	2	1	2	19/10/2011	0	2	1	0	24/09/2012	0	0
13	Montesquieu Des Alberes			1	0	20/10/2009	0	0	1	9	13/10/2010	0	9	1	0	18/10/2011	0	0	1	0	26/09/2012	0	0
11	Laroque Des Alberes								1	1	13/10/2010	0	1	1	37	17/10/2011	0	37	1	7	24/09/2012	0	7
12	**Collioure**								1	1	14/10/2010	0	1	1	14	19/10/2011	0	14	1	103	**25/09/2012**	**1**	103
8	Sorede								1	41	13/10/2010	0	41	1	1	19/10/2011	0	1	1	53	24/09/2012	0	53

For *C. imicola* collections, we reported only the data for the maximum abundance collection date. Nb = number. Locations and collection dates in bold are positive sites for *C. imicola* in the Pyrénées-Orientales department.

Supplementary Table 2 | COI and Cytb concatenated genetic diversity and haplotype composition within sample site.

Site	**N**	**H**	**Hd ± SD**	***π* ± SD**	**Haplotype composition**																														
					H1	H2	H3	H4	H5	H6	H7	H8	H9	H10	H11	H12	H13	H14	H15	H16	H17	H18	H19	H20	H21	H22	H23	H24	H25	H26	H27	H28	H29	H30	H31
Algeria	17	7	0.86 ± 0.05	0.0015 ± 0.0002	5	3	1	3	1	2	2																								
Morocco	14	11	0.95 ± 0.04	0.0020 ± 0.0002		1					3	1	1						2							1	1	1	1	1	1				
Portugal	15	7	0.81 ± 0.09	0.0013 ± 0.0003						2	7		1		2																		1	1	1
Continental Spain	22	11	0.83 ± 0.07	0.0013 ± 0.0002		2					9		1	1	1	1	1	3	1	1	1														
Balearic Islands, Spain	8	3	0.67 ± 0.12	0.0007 ± 0.0001	4						3	1																							
Pyrénées-Orientales, France	17	6	0.75 ± 0.09	0.0010 ± 0.0002	2	3				1	8											2		1											
Var, France	16	3	0.54 ± 0.09	0.0056 ± 0.0001		10				5															1										
Corsica, France	16	6	0.85 ± 0.02	0.0013 ± 0.0001	1	5		3			5											1	1												
Sardinia, Italy	8	3	0.68 ± 0.12	0.0007 ± 0.0002				3		1	4																								

For each site, the number of samples (N), the number of haplotypes (H), haplotype (Hd) and nucleotide (π) diversity and their standard deviations (SD) are indicated.

Supplementary Table 3 | P-values inferred by the Wilcoxon test of bottleneck, F_{IS} values within samples and effective population sizes

Country	Sites	code	IAM	TPM	SMM	F_{IS}
Algeria	Skikda	J	0.0185*	0.5000	0.9355	0.107
	Wilaya de Jijel	I	0.0136*	0.3671	0.8984	0.050
Morocco	Khemisset	H	0.4101	0.9179	0.9951	0.030
	Sidi Yahia El Gharb	G	0.5898	0.9179	0.9970	0.140
Portugal	Castelo Branco	B	0.1250	0.5898	0.9179	0.096
	Beja	A	0.0644	0.5898	0.8984	0.122
Balearic Islands, Spain	Majorca	F	0.1015	0.5898	0.9814	0.097
Continental Spain	Girona	E	0.0644	0.5898	0.1503	0.080
	Toledo	D	0.0097**	0.4101	0.8984	0.010
	Huelva	C	0.0019**	0.0644	0.5449	0.025
Continental France	Pyrénées-Orientales	M	0.0068**	0.1503	0.3671	-0.015
	Roquebrune-sur-Argens	K	0.0019**	0.0097**	0.3261	-0.038
	Bormes-les-Mimosas	L	0.0097**	0.1503	0.4550	0.024
Corsica, France	Figari	N	0.0097**	0.2480	0.8496	0.036
	Pietracorbara	O	0.0019**	0.0644	0.8750	0.112
Sardinia, Italy	San Giovanni Suergiu	P	0.0185*	0.2481	0.7871	0.052

For the bottleneck analysis, p-values correspond to the Wilcoxon unilateral test under each mutation model (Infinite Allele Mutation: IAM, Two-phase model: TPM and Stepwise Mutation Model: SMM). Statistical significance is tested over 10000 replicates ($^{**}P < 0.01$, $^{*}P < 0.05$). P-values obtained for the Hardy-Weinberg test were adjusted for multiple comparisons at the nominal level (5%) of 0.00037.

Supplementary Table 4 | Mitochondrial pairwise F_{ST} values between *C. imicola* populations.

	Algeria	Sardinia, Italy	Corsica, France	Pyrénées-Orientales, France	Var, France	Morocco	Portugal	Balearics Island, Spain
Sardinia, Italy	0.03188							
Corsica, France	0.04440	0.00444						
Pyrénées-Orientales, France	0.04421	**0.19543**	0.04354					
Var, France	**0.15972**	**0.34694**	0.10000	0.03702				
Morocco	**0.07275**	**0.11014**	0.04071	0.00905	0.03303			
Portugal	0.06163	**0.07438**	**0.07590**	0.02891	**0.07054**	0.02240		
Balearics Island, Spain	0.02336	**0.28571**	**0.24953**	**0.19914**	**0.47986**	0.12475	**0.19556**	
Continental Spain	**0.12118**	**0.12695**	**0.07619**	**0.05231**	0.05430	0.01229	0.03447	**0.20952**

Significant values are highlighted in bold.

Supplementary Table 5 | Approximate Bayesian computation (ABC) results.

Description of tested scenarios	**Posterior probability**	**95% credibility interval**	**Type I error**	**Type II error**
Independent introductions from North Africa into Spain, Sardinia, Corsica and Pyrénées-Orientales	0.07	[0.01-0.13]	0.16	0.06
Introduction from North Africa into Spain, then colonization of Pyrénées-Orientales out from Spain	0.01	[0.00-0.06]	0.18	0.06
Introduction from North Africa into Sardinia, then colonization of Pyrénées-Orientales out from Sardinia	0.29	[0.26-0.32]	0.21	0.09
Introduction from North Africa into Sardinia, then introduction in Corsica out from Sardinia, and then colonization of Pyrénées-Orientales out from Corsica	**0.62**	**[0.60-0.64]**	**0.28**	**0.06**

Type 1 error is the probability of selecting another scenario when the chosen scenario is true. Type 2 error is the mean probability of selecting the chosen scenario when it is false. The selected (most probable) scenario is highlighted in bold (see also Fig. S7).

Supplementary Table 6 | ABC model checking of the most probable scenario (Scenario 4).

summary statistics	observed value	proportion (simulated<observed)
NAL_1_1	5.0000	0.6330
NAL_1_2	4.3333	0.5850
NAL_1_3	4.6667	0.5440
NAL_1_4	5.0000	0.5915
NAL_1_5	4.4444	0.5445
HET_1_1	0.6101	0.6745
HET_1_2	0.6113	0.7160
HET_1_3	0.5909	0.6200
HET_1_4	0.6074	0.6310
HET_1_5	0.5939	0.6340
VAR_1_1	1.3344	0.3385
VAR_1_2	1.2770	0.3340
VAR_1_3	1.3534	0.3460
VAR_1_4	1.4478	0.3500
VAR_1_5	1.3576	0.3595
MGW_1_1	0.9854	0.8330
MGW_1_2	0.9000	0.6515
MGW_1_3	1.0286	0.9160
MGW_1_4	0.9712	0.7930
MGW_1_5	0.9677	0.8430
N2P_1_1&2	5.7778	0.6655
N2P_1_1&3	5.7778	0.6080
N2P_1_1&4	5.7778	0.5920
N2P_1_1&5	5.6667	0.6085
N2P_1_2&3	5.0000	0.5110
N2P_1_2&4	5.3333	0.5390
N2P_1_2&5	4.7778	0.5125
N2P_1_3&4	5.5556	0.5470
V2P_1_1&4	1.3863	0.3315
V2P_1_1&5	1.3480	0.3255
V2P_1_2&3	1.3258	0.3310
V2P_1_2&4	1.3718	0.3355
V2P_1_2&5	1.3245	0.3420
V2P_1_3&4	1.3911	0.3290
V2P_1_3&5	1.3621	0.3420
V2P_1_4&5	1.4067	0.3375
FST_1_1&2	0.0332	0.2965
FST_1_1&3	0.0352	0.4550
FST_1_1&4	0.0261	0.4365
FST_1_1&5	0.0320	0.3400
FST_1_2&3	0.0146	0.3340
FST_1_2&4	0.0159	0.1545
FST_1_2&5	-0.0004	0.2160
FST_1_3&4	0.0042	0.0420 (*)
FST_1_3&5	0.0119	0.3580
FST_1_4&5	0.0049	0.0590
LIK_1_1&2	1.0667	0.5670
LIK_1_1&3	1.0523	0.6180
LIK_1_1&4	0.9948	0.6055
LIK_1_1&5	1.0554	0.5920
LIK_1_2&1	0.9423	0.4770
LIK_1_2&3	0.8331	0.4720
LIK_1_2&4	0.8503	0.3980
LIK_1_2&5	0.8065	0.5155
LIK_1_3&1	1.0029	0.5390
LIK_1_3&2	0.9266	0.5165
DAS_1_2&3	0.3945	0.3635
DAS_1_2&4	0.3845	0.3985
DAS_1_2&5	0.4048	0.3855
DAS_1_3&4	0.3984	0.4425
DAS_1_3&5	0.4049	0.4220
DAS_1_4&5	0.3989	0.4565
DM2_1_1&2	0.0504	0.0805
DM2_1_1&3	0.0468	0.1135
DM2_1_1&4	0.0354	0.0825
DM2_1_1&5	0.0605	0.1440
DM2_1_2&3	0.0616	0.2345
DM2_1_2&4	0.0450	0.0860
DM2_1_2&5	0.0639	0.3585
DM2_1_3&4	0.0173	0.0165 (*)
DM2_1_3&5	0.0776	0.3590
DM2_1_4&5	0.0579	0.1545

N2P_1_3&5	5.0000	0.4750	LIK_1_3&4	0.9104	0.4740
N2P_1_4&5	5.4444	0.5520	LIK_1_3&5	0.9307	0.5560
H2P_1_1&2	0.6210	0.6675	LIK_1_4&1	0.9566	0.5355
H2P_1_1&3	0.6120	0.6430	LIK_1_4&2	0.9286	0.4315
H2P_1_1&4	0.6183	0.6590	LIK_1_4&3	0.8742	0.4170
H2P_1_1&5	0.6134	0.6520	LIK_1_4&5	0.9132	0.4460
H2P_1_2&3	0.6038	0.6490	LIK_1_5&1	0.8989	0.4915
H2P_1_2&4	0.6145	0.6485	LIK_1_5&2	0.7852	0.5140
H2P_1_2&5	0.6022	0.6685	LIK_1_5&3	0.8042	0.4940
H2P_1_3&4	0.6010	0.5915	LIK_1_5&4	0.8158	0.4165
H2P_1_3&5	0.5966	0.6160	DAS_1_1&2	0.3730	0.3740
H2P_1_4&5	0.6033	0.5920	DAS_1_1&3	0.3790	0.3770
V2P_1_1&2	1.3115	0.3100	DAS_1_1&4	0.3770	0.3605
V2P_1_1&3	1.3455	0.3230	DAS_1_1&5	0.3826	0.4000

All the summary statistics (used and unused for model selection) were used. For each statistic, the observed value and probability (simulated < observed) are given. For a detail description of the summary statistics, see the DIYABC v.2.0.4 manual. Significant probabilities are represented with an asterisk.

Supplementary Table 7 | Primers characteristics.

Loci	Motif	Forward	Reverse	Allele size range	T (°C)
68	(GT)n	CTTTTCCGTTTCTTTTTATTTCTTT	GTTTCTTTCTGGTCGCGTTGGTTGCTG	99-105	60
12b	(CT)n	TTATGTGTGTATGTTAGCAAGGTCA	GTTTCTTCTTCGGATCAAAGAAATTTTGCC	129-139	50
3b	(AC)n	ATGCGGATGTTTGAAGTG	GTTTCTTTTTTGTGTCTTATTGCCC	154-175	50
31	(CAA)n	TTCTGTTCGGCTGTTGCGTT	GTTTCTTCTTTTTACGTGGTGGTCATTC	152-168	60
41b	(CT)n	GAGGAGGAGGTAGAA	GTTTCTTTCTATTAGTCAATGGTG	155-166	50
35t	(AC)n	TTTGTAAAAGCCAGTTCAACCG	GTTTCTTATCGAACGAAGGAAATAACCAC	171-194	60
88b	(AC)n	TTTGTTCGATTTGTAGTG	GTTTCTTCCTCTCTTTCATTCGC	243-256	50
16	(TG)n	TTGCCTTTGCTTGTTGAGGATG	GTTTCTTTCCTCTTTAAAATCACTGACGTG	292-299	60
88	(CAT)n	GTTGGTGCTTTGTTGTGTTGT	GTTTCTTTTTCTTTTTCTCCTTTTTGTTTCTTTC	344-348	50

Supplementary Table 8 | Prior distributions of demographic, historical and mutation parameters used for ABC inferences.

Parameter description	Parameter	Prior distribution
Stable effective population size	N	Loguniform [1000; 200000]
Time of colonization events in Pyrénées-Orientales	t_p	Uniform [40; 10000]
Time of colonization events in Sardinia, Corsica and Spain	t_s, t_c, t_e	Uniform [140; 10000]
Duration of bottleneck	D_B	Uniform [0; 50]
Founding effective population sizes	N_s	Loguniform [2; 100]
Mean mutation rate	µmicrosatellites	Loguniform [1E-6; 1E-4]
Mean parameter of the geometric distribution of length	P	Uniform [1E-1; 3E-1]
Mean mutation rate for nucleotide instability	SNI	Uniform [1E-8; 1E-4]

Effective population sizes (N) are expressed in number of diploid individuals and times of events (t) in number of generations going back to the past. Conditions among the parameters used during the simulations were ts ≥ tp, te ≥ tp, tc ≥ tp.