Table S1. Long-read PCR parameters for each primer pair.

Primer pair code*	**1**	**3**	**4**	**6**	**7**	**8**	**9**	**10**	**12**	**13**	**15**	**16**
Number of PCR cycles	35	35	35	30	35	30	35	35	35	30	30	35
T_a (°C) (annealing)	53	53	55	60	55	60	53	53	48	60	60	48
Elongation step (min)	8	7.5	9.5	10	11	10	11	11	11	11	11	10

* Primer pair as designated in Uribe-Convers et al. [1].

Table S2. Flow cytometry and ploidy determination results for *Dactylis glomerata.*

ID	**Country**	**Accession no.**	**DNA ratio with int. standard ***	**Ploidy**
D-000	Cultivar	cv. Sparta	1.32	4x
D-000b	Cultivar	cv. Sparta 2	1.29	4x
D-001	Poland	P6	1.34	4x
D-002	Poland	P34	1.34	4x
D-003	Poland	P96	1.33	4x
D-004	Poland	P112	1.34	4x
D-005	Poland	P156	1.30	4x
D-006	Poland	P182	1.31	4x
D-007	Poland	P216	1.31	4x
D-008	Poland	P248	1.38	4x
D-009	Poland	P282	1.32	4x
D-010	Germany	G6	1.32	4x
D-011	Germany	G10	1.34	4x
D-012	Germany	G52	1.33	4x
D-013	Germany	G66	1.36	4x
D-014	Germany	G68	1.38	4x
D-015	Germany	G100	1.36	4x
D-016	Germany	G102	1.37	4x
D-017	Germany	G130	1.37	4x
D-018	Germany	G132	1.39	4x
D-019	Germany	G152	1.30	4x
D-020	Germany	G184	1.39	4x
D-021	Germany	G220	1.36	4x
D-022	Germany	G272	1.31	4x
D-023	Germany	G276	1.36	4x
D-024	Denmark	D2	1.42	4x
D-025	Denmark	D6	1.38	4x
D-026	Denmark	D62	1.40	4x
D-027	Denmark	D68	1.36	4x
D-028	Denmark	D92	1.39	4x
D-029	Denmark	D96	1.40	4x
D-030	Denmark	D128	1.43	4x
D-031	Denmark	D132	1.43	4x
D-032	Denmark	D156	1.43	4x
D-033	Denmark	D160	1.37	4x
D-034	Denmark	D182	1.41	4x
D-035	Denmark	D184	1.47	4x

D-036	Denmark	D236	1.32	4x
D-037	Denmark	D240	1.40	4x
D-038	Denmark	D246	1.38	4x
D-039	Denmark	D272	1.36	4x
D-040	Denmark	D274	1.45	4x
D-041	Sweden	S6	1.38	4x
D-042	Sweden	S10	1.43	4x
D-043	Sweden	S42	1.35	4x
D-044	Sweden	S54	1.40	4x
D-045	Sweden	S82	1.38	4x
D-046	Sweden	S88	1.42	4x
D-047	Sweden	S114	1.47	4x
D-048	Sweden	S118	1.38	4x
D-049	Sweden	S132	1.31	4x
D-050	Sweden	S140	1.34	4x
D-051	Sweden	S160	1.38	4x
D-052	Sweden	S164	1.42	4x
D-053	Sweden	S180	1.39	4x
D-054	Sweden	S184	1.39	4x
D-055	Sweden	S211	1.40	4x
D-056	Sweden	S216	1.37	4x
D-057	Sweden	S241	1.38	4x
D-058	Sweden	S242	1.37	4x
D-059	Sweden	S253	1.41	4x
D-060	Sweden	S254	1.38	4x
D-061	UK	UK62	1.42	4x
D-062	UK	UK30	1.28	4x
D-063	UK	UK59	1.32	4x
D-064	UK	UK106	1.31	4x
D-065	UK	UK146	1.31	4x
D-066	UK	UK172	1.29	4x
D-067	Ireland	IE4	1.34	4x
D-068	Ireland	IE48	1.25	4x
D-069	Ireland	IE196	1.30	4x
D-070	Ireland	IE226	1.32	4x
D-071	Ireland	IE244	1.30	4x
D-072	Ireland	IE272	1.28	4x

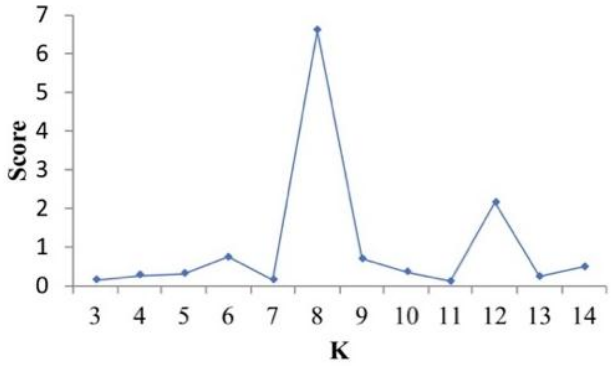

Figure S1. Optimal value of K determined by the ΔK statistic [2] using Structure Harvester [3]. Eight clusters were determined as optimal.

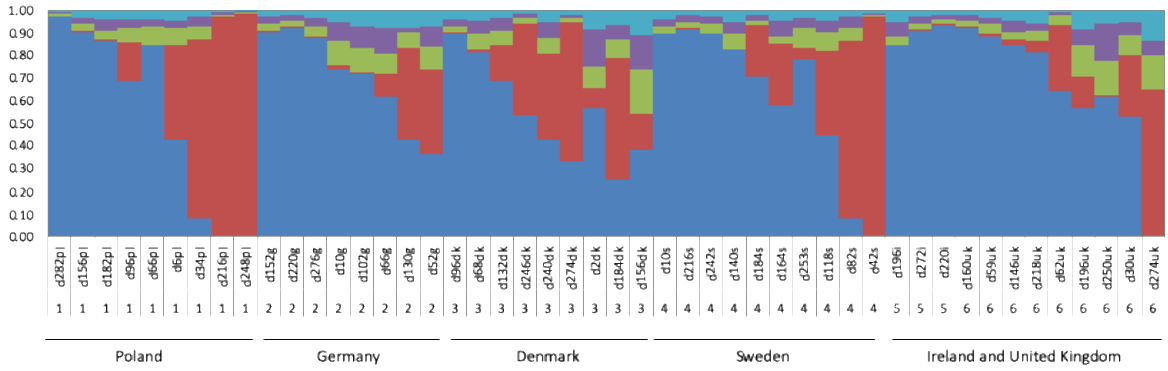

Figure S2. Structure of Dactylis glomerata genotypes estimated using Structure analysis of plastome SNPs with K=5 instead of the optimal value of K=8. The K=8 plot is presented in the main text. Each plant is shown as a vertical bar and SNP variation is partitioned into coloured segments that show the individual's estimated membership fractions in each of the K=5 clusters. K=8 was the determined as optimal by the ΔK statistic [2]. Alternative cluster numbers were compared but we have no a priori evidence to accept any of these nor do they alter our interpretation of the results based on the optimal value of K=8. We present K=5 here as an example of results with an alternative K value. K=5 was chosen a priori, instead of other alternative K values (e.g. 2, 3, 4, 6, 7), based on the altificial but rough geographical proximity of groups (national boundaries). Samples are arranged in the plot according to geographic location (1=Poland, 2=Germany, 3=Denmark, 4=Sweden, and 5=Ireland plus 6=UK combined; total K=5).

References

1. Uribe-Convers, S.; Duke, J.R.; Moore, M.J.; Tank, D.C. A long PCR–based approach for DNA enrichment prior to next-generation sequencing for systematic studies. *Appl. Plant Sci.* 2014, *2*, 1300063.
2. Evanno, G.; Regnaut, S.; Goudet, J. Detecting the number of clusters of individuals using the software structure: a simulation study. *Mol. Ecol.* 2005, *14*, 2611–2620.
3. Earl, D.A.; vonHoldt, B.M. STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conserv. Genet. Resour.* 2012, *4*, 359–361.