

Complete Genotype with Project Merge Tool

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Introduction

Obtaining a complete genetic profile for wildlife and plant research or medical research is often complicated by overlapping marker ranges and/or incompatible chemistry; making it necessary to amplify the same samples multiple times using different sets of markers or locus specific primers. Traditionally, researchers export the genotyping results from several multiplexes into a spread sheet and manually combine allele calls for each individual.

GeneMarker 1.90 addresses this problem with a new tool: Merge Projects. Researchers can conveniently combine two or more GeneMarker projects (each project using a panel containing a unique set of markers or loci) into a single, comprehensive report. This report represents a single view of multiplexes (a super panel) of markers/loci from the individual projects, providing a complete genotype for each sample. This merged report may be saved in a spread sheet as a genome-wide genotype, or imported into other special applications, such as, Clustering Analysis or Relationship Testing and Kinship Analysis, improving the robustness of these analyses by including information from a greater number of markers.

Procedure – Merge Projects

1. Import data files (fsa, abi, ab1, scf) or open a saved GeneMarker Project file
2. Select the Run icon to launch the Run Wizard – using a panel with uniquely named markers
3. Repeat with data files amplified with a second multiplex of uniquely named markers
4. Save each project for a set of individual files
5. Select Tools → Merge Project → Open Merge Project → Add project files → OK
6. Select File Name Group icon → Match Files by group identifier or character position → Match → OK
7. Click Report Settings Icon to activate the Allele Report Settings dialog
8. Save the merged project report as a .txt file.

	MAF65	MAF64	BMC1222	BN848	OARCP0026	Rt9	BH1225	MAF209	BM4505	OxwAE16	TGLA0387	FCB266	TGLA1222														
1 BBS-054 PA	128	116	120	112	291	285	234	232	136	130	134	132	252	246	114	110	110	277	263	98	80	137	131	89	101	134	131
2 SPBBRS-001	118	116	116	116	291	287	236	236	138	136	132	128	**	**	110	106	263	263	94	80	141	135	91	89	133	133	
3 SPBBRS-002	118	116	116	116	287	285	244	236	144	144	136	132	252	248	106	106	268	268	94	82	141	131	95	101	134	134	
4 SPBBRS-003	118	118	118	118	291	285	244	236	146	146	128	128	246	242	118	106	263	262	86	82	141	133	95	91	134	133	
5 SPBBRS-004	Disabled																										
6 SPBBRS-005	126	118	122	116	287	285	236	236	138	138	128	122	252	246	114	110	271	271	86	80	141	135	97	89	134	134	
7 SPBBRS-006	116	116	122	120	291	285	244	236	146	126	134	128	260	260	118	106	271	262	86	82	137	131	95	101	135	133	
8 SPBBRS-007	118	116	122	110	285	285	232	228	146	146	128	128	260	248	118	118	262	262	82	82	141	137	95	95	134	133	
9 SPBBRS-008	118	116	122	118	285	285	244	236	126	126	134	128	260	248	118	118	263	262	86	82	141	137	95	95	135	133	
10 SPBBRS-009	118	116	118	110	285	285	236	232	148	144	128	128	248	248	114	106	271	262	**	**	137	131	95	101	133	133	
11 SPBBRS-010	116	116	122	118	287	285	244	234	146	136	136	132	252	250	118	114	277	275	98	80							
12 SPBBRS-011	116	116	120	110	291	285	236	228	146	126	128	128	248	246	118	106	268	263	86	82							
13 SPBBRS-012	124	116	122	116	291	285	236	234	148	138	136	128	252	252	106	106	277	277	80	80							
14 SPBBRS-013	116	116	122	118	291	291	244	244	146	146	128	128	248	246	118	118	275	258	82	80							
15 SPBBRS-014	126	124	122	122	291	287	238	232	144	144	132	132	252	246	120	106	263	263	92	80							
16 SPBBRS-015	128	128	118	118	287	287	234	232	136	130	138	120	244	244	114	114	263	263	92	86							
17 SPBBRS-016	126	118	122	112	291	285	236	236	144	138	128	128	252	248	114	114	263	258	82	80							
18 SPBBRS-017	118	118	122	122	291	285	244	236	126	126	128	128	260	242	106	106	275	262	98	80							
19 SPBBRS-018	118	116	120	116	287	285	244	240	146	146	138	128	260	242	118	118	268	258	86	86							
20 SPBBRS-019	118	118	122	118	291	285	236	228	138	126	132	128	260	260	118	106	263	258	82	82							
21 SPBBRS-020	124	118	118	110	285	285	244	244	126	126	128	128	260	248	118	106	262	262	86	82							
22 SPBBRS-021	118	118	118	116	287	285	236	228	146	146	106	106	277	258	86	82											
23 SPBBRS-022	118	116	120	118	291	285	244	238	146	146	138	134	260	260	118	118	262	258	86	80							
24 SPBBRS-023	118	118	120	110	291	287	244	234	144	126	128	128	248	248	120	118	262	258	80	80							
25 SPBBRS-024	**	**	122	118	**	**	244	228	146	146	132	128	252	248	120	118	276	268	86	80							
26 SPBBRS-025	126	124	122	122	291	287	244	236	146	126	128	128	250	248	118	106	258	258	98	98							
27 SPBBRS-026	126	118	122	122	291	285	236	236	144	138	128	128	248	246	118	106	271	258	98	80							
28 SPBBRS-027	126	118	116	110	291	285	236	234	146	138	132	128	252	250	114	106	277	263	98	80							
29 SPBBRS-028	124	116	116	116	291	291	236	236	144	138	132	128	260	260	106	106	263	263	94	94							
30 SPBBRS-029	126	124	116	116	291	291	244	234	138	138	132	128	260	250	110	106	263	262	80	80							

Figure 1: Three different multiplexes were used to amplify 30 DNA samples. Each multiplex contained primers for 4 or 5 independently assorting loci. The merge project tool in GeneMarker provided a single genotype for each individual with 14 markers. Allele drop out is indicated by **. The report style pictured here is a Marker Table suitable for further analysis in Relationship Testing or Kinship Analysis. Saving as a Peak Table would provide a spread sheet with marker and allele name, fragment size (MW), peak height, height ratio, peak area, area ratio. The Bin Table format would be selected for further analysis using clustering algorithms for phylogeny.

Results and Discussion

Figure 1 illustrates a three-fold increase in the number of loci in one report made possible by merging projects. The .txt tab delimited format of the saved report provides the ability to save any number of loci and samples into one merged report.

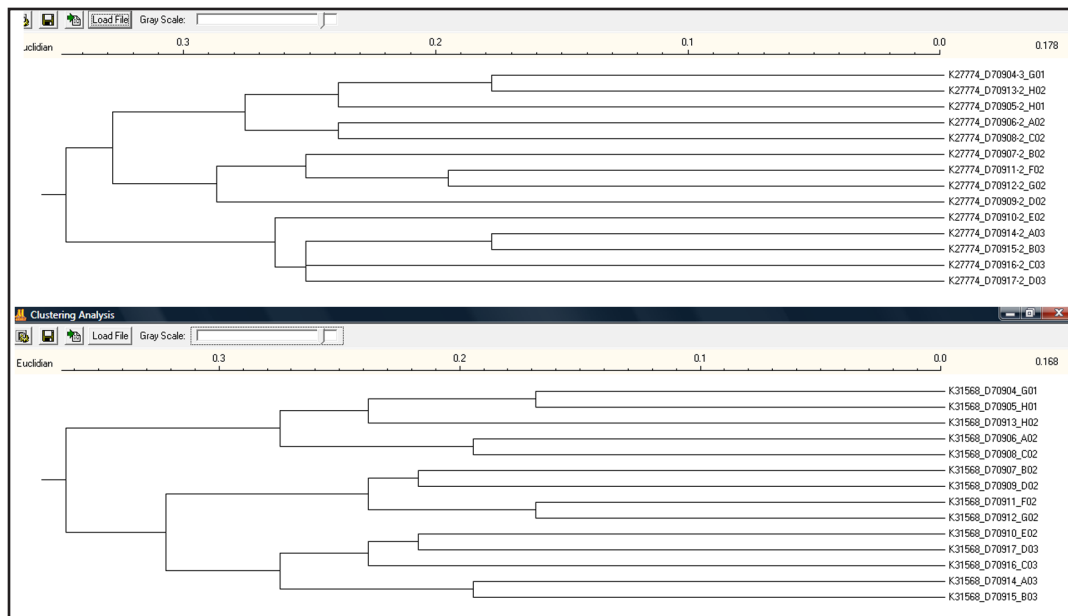


Figure 2. Expanded discriminatory power is possible by including 9 loci from two projects (upper dendrogram) in contrast to the lower dendrogram that is based on the 4 loci of one multiplex.

Overlapping marker ranges or incompatible chemistries are common challenges in microsatellite analysis. GeneMarker's Merge Project tool enables researchers to combine genotype information on the same samples analyzed with different multiplexes. The result is a master report that provides genotype information from multiple kits or multiplexes with the flexibility to be saved in a variety of formats: Marker Table, Peak Table or Bin Report.

Examples of clinical research applications with Merge Project include pathogen and genetic disorder research. Often several PCR kits are used to screen individuals for a variety of pathogens that cause similar symptoms, such as influenza strains or drug resistant tuberculosis strains. Results from these multiple kits are presented in one report table, providing more complete information for each patient in a single report. Multiple kits are also required for complete genetic profiles for genetic disorders, such as breast cancer or Duchenne muscular dystrophy (DMD). Merged projects provides complete genotypes, allowing combination of kit results for one disorder, or combining results from kits for different disorders on one report for each individual.

Ecology and agriculture applications include compilation of a genome-wide report for each individual in one table. The table can be saved as a spread sheet for import into other statistics packages, or used with GeneMarker applications; providing greater discriminatory power for kinship analysis and relationship testing, and expanded rigor for clustering algorithms used in phylogeny, lineage and evolution studies.

GeneMarker has been designed to provide genetic researchers with a biologist friendly genotyping tool; with unique pattern recognition and sizing technology providing >99% accuracy, easy linked navigation, management control and tracking, exportable LIMS reports, bulk printing capabilities, instrument compatibility with ABI®, MegaBACET™ and Beckman Coulter™. Unique post-genotyping applications in GeneMarker of interest to ecology and population genetics research include: cluster analysis and phylogeny, relationship testing and kinship analysis, microsatellite instability, and TILLING®.

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