**Additional File 1: Primers for each marker used in the study.** All markers except Sssp3016 (Gene bank accession number: AY372820) are listed in [46]**.** Fluorescent dye, working primer concentration, multiplex group and genotyping groups (panel) of each primer pair are indicated in the table 1.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Locus | Label | Panel2 | Multiplex number | Primer (µM) |
| CA060177 | pet | 1 | MP1 | 0.36 |
| Ssa 407 | ned | 1 | MP1 | 0.36 |
| CA064557 | ned | 1 | MP1 | 0.27 |
| Ssa197 | vic | 1 | MP1 | 0.10 |
| Ssa 171 | fam | 1 | MP1 | 0.35 |
| Ssa 412 | vic | 1 | MP1 | 0.27 |
| CA040282 | pet | 1 | MP2 | 0.10 |
| SSsp2216 | fam | 1 | MP2 | 0.20 |
| CA039983 | pet | 1 | MP2 | 0.10 |
| SSsp2215 | fam | 1 | MP2 | 0.10 |
| CA047220 | vic | 1 | MP2 | 0.30 |
| SSsp2210 | ned | 1 | MP2 | 0.30 |
| Ssa85 | vic | 1 | MP2 | 0.20 |
| CA062844 | pet | 1 | MP2 | 0.10 |
| Ssleer 15.1 | fam | 2 | MP3 | 0.36 |
| SSsp1605 | vic | 2 | MP3 | 0.04 |
| Ssa124 | vic | 2 | MP3 | 0.20 |
| Ssa202 | ned | 2 | MP3 | 0.40 |
| Ssf43 | ned | 2 | MP3 | 0.10 |
| Ssosl 438 | fam | 2 | MP3 | 0.25 |
| Sssp3016 | pet | 2 | MP3 | 0.30 |
| Ssd30 | vic | 2 | MP4 | 0.40 |
| Ssosl25 | ned | 2 | MP4 | 0.30 |
| Sleen82 | fam | 2 | MP4 | 0.20 |
| Ssa98 | ned | 2 | MP4 | 0.20 |
| Sleei53 | fam | 2 | MP4 | 0.15 |
| Sssp2201 | fam | 2 | MP4 | 0.40 |
| EST405 | ned | 2 | MP4 | 0.33 |
| Ssosl311 | vic | 2 | MP4 | 0.40 |

1PCR reactions were performed at 8 µl consisted 1 µl of genomic DNA (variable concentration), 0.04 to 0.40 µM of each primer, and 1X QIAGEN multiplex PCR master mix (Qiagen Inc. Valencia, CA, USA). The PCR program was as follows: 15 minutes at 95 ˚C for initial denaturing followed by 36 cycles of denaturing at 94 ˚C for 30 seconds, annealing at 60/59 ˚C for 1.5 minutes and elongating at 72 ˚C for 1 minute with final elongation at 72 ˚C for 5 minutes. Annealing temperature was 59 for MP1 and 60 for the remaining multiplex reactions.

2The multiplex PCRs were pooled for fragment analysis as follows: for panel 1, 1.6 µl of MP1 and 1.7 µl of MP2 were pooled to 100 µl of H2O and for panel 2, 1.5µl of MP3 and 1.5 µl of MP4 were pooled to 100 µl of H2O.