

## MapReduce

MRPrimer is based on the MapReduce framework, whose dataflow is shown in Figure S1. One round of MapReduce consists of two user-defined functions, *Map* and *Reduce*. Here, *Reduce* is optional, and can therefore be omitted. The input data for a round is distributed over the disks of a cluster of computers. The partial data in each computer is processed by Map functions, shuffled via network, processed by Reduce functions, and then stored on the disks; the resultant data is then fed to the Map functions of the next MapReduce round. The input/output format of the data, i.e., the signatures of the Map and Reduce functions, are formally defined as follows.

$$\text{Map: } \langle k1, v1 \rangle \rightarrow \text{list}(\langle k2, v2 \rangle)$$
$$\text{Reduce: } \langle k2, \text{list}(v2) \rangle \rightarrow \langle k3, v3 \rangle$$

The Map function takes a pair of *key* and *value*,  $\langle k1, v1 \rangle$ , as input (e.g.,  $k1$  is a sequence ID, and  $v1$  is the sequence itself), and then returns a list of pairs,  $\langle k2, v2 \rangle$ , in a different domain as output. The Reduce function takes a pair of key and list of values, i.e.,  $\langle k2, \text{list}(v2) \rangle$ , as input, and then returns a pair of key and value, i.e.,  $\langle k3, v3 \rangle$ , in a different domain as output. Here, we note that  $v2$  in the output of Map becomes  $\text{list}(v2)$  in the input of Reduce because the shuffle process gathers all  $v2$ s having the same  $k2$ , which are scattered over computers, into a single list of  $v2$ s on a single computer. MRPrimer relies extensively on this feature of MapReduce to efficiently perform large-scale and complicated computation.

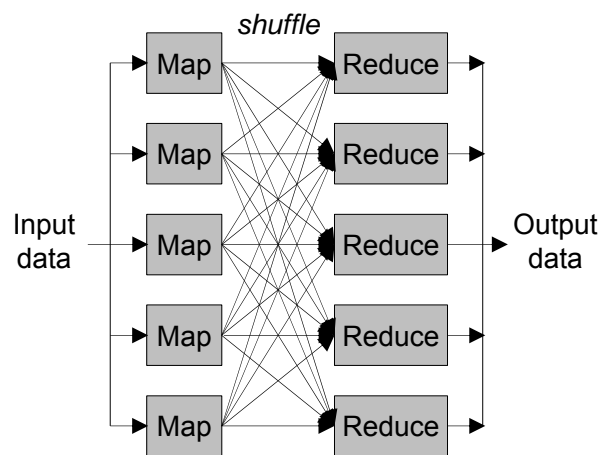
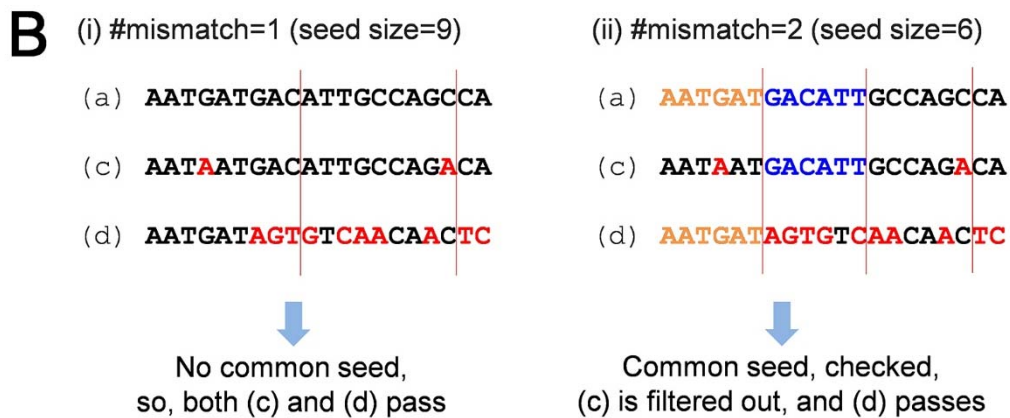
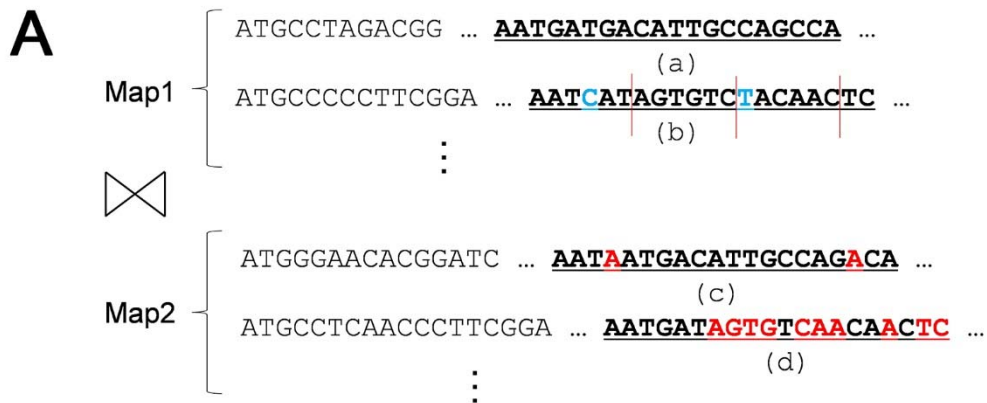


Figure S1. Dataflow of the MapReduce framework.



**Figure S2. An example of the 5' cross-hybridization filtering step.** There is a 20-mer primer (a) in Map1, and three candidate primers of the same length, (b), (c), and (d), in Map2. We assume that the user-defined value for the maximum mismatches at the 5' end is 4. Then, primer (b) is filtered out because it has the same 16-mer subsequence (in blue) as primer (a) in an off-target sequence (only two mismatches in red). Primers (c) and (d) pass because they have no such identical subsequence. However, since it is similar to primer (a), primer (c) might cross-hybridize to the off-target sequence to which primer (a) belongs.



**Figure S3. An example of the general cross-hybridization filtering step.** (A) There are two primers, (a) and (b), in Map1, and two candidate primers, (c) and (d), in Map2. Primer (c) is similar to primer (a), with only two mismatches (in red). Primer (d) is quite different from primer (a), although it is similar to primer (b), with only two mismatches (in cyan). (B) In the first execution of Steps 4 and 5 with #mismatch = 1, the seed size is nine. Primers (c) and (d) have no common seed with primer (a), and so they pass. In the second execution of Steps 4 and 5 with #mismatch = 2, the seed size is 6. Primer (c) has a common seed (in blue) with primer (a), and so both the prefix and suffix of the seed are checked. Due to the presence of only two mismatches (in red), primer (c) is filtered out. Primer (d) also has a common seed (in orange) with primer (a), and so it is checked. It can pass Step 4 due to the large number of mismatches (in red), but cannot pass Step 5 because it is identified as similar to primer (b).

**Table S1. Summary of the filtering constraints used in PrimerBank.**

|                    | <b>Parameter</b>                 | <b>Value</b>       |
|--------------------|----------------------------------|--------------------|
| <b>Each primer</b> | primer length                    | 19–23 bp           |
|                    | melting temperature (TM)         | 60–63 °C           |
|                    | GC content                       | 35–65%             |
|                    | self-complementarity             | <5-mer             |
|                    | 3' self-complementarity          | <4-mer             |
|                    | contiguous residue               | <6-mer             |
|                    | Gibbs free energy ( $\Delta G$ ) | $\geq -9$ kcal/mol |
| <b>Primer pair</b> | length difference                | $\leq 3$ -mer      |
|                    | TM difference                    | $\leq 5$ °C        |
|                    | product size                     | 100–250 bp         |
|                    | pair-complementarity             | <9-mer             |
|                    | 3' pair-complementarity          | <4-mer             |

**Table S2. Primers for non-OR genes used in the biological experiments to validate MRPrimer.**

| Gene    | Sequence |                         | Size (bp) | Gene   | Sequence |                         | Size (bp) |
|---------|----------|-------------------------|-----------|--------|----------|-------------------------|-----------|
| Adcy6   | F        | GACCAACTGCGTAAGGACCAT   | 178       | Gnai2  | F        | CTTATGACTTGGTGCTGGCTGA  | 183       |
|         | R        | TCAGGGTGGAGTATGGGAACA   |           |        | R        | ACTCAGGGAAACAGATGGTCAG  |           |
| Adcy9   | F        | AATGAAACAAGGGGACGAGGAG  | 119       | Gnai3  | F        | GCAGATGATGCCCGACAGTTA   | 136       |
|         | R        | TAAAGGGGCGGAATGCTATCG   |           |        | R        | ATCCCTGGACCTGCTAAAGC    |           |
| Anxa2   | F        | GAGACGGTGATTTTGGGCCTAT  | 127       | Gnal   | F        | TGGACGATGAAGGAGTGAAG    | 124       |
|         | R        | GGTTGGTTCGTGAGCAGATGAT  |           |        | R        | GGTCTGTGGGTGTGTAGTCAA   |           |
| Anxa4   | F        | AGAGAAGAGATGGGGGACAGA   | 168       | Gnao1  | F        | TCACCCTTGACCATCTGCTTTC  | 107       |
|         | R        | GCCAACAGGGCATCTTCAAAG   |           |        | R        | TTGTTGGGTGAGCGGTTTTTG   |           |
| Cacna1c | F        | GCAGCGTAAGGATGAGTGAAGA  | 170       | Gnaq   | F        | GGTTGATGTGGAGAAGGTGTCT  | 183       |
|         | R        | TAGAGAGGCAGAGCGAAGGAA   |           |        | R        | TGTGTAGGCAGATAGGAAGGGT  |           |
| Cacna1i | F        | AAACGTGCTCCTGCTCTGTTT   | 159       | Gnas   | F        | TACGATCAGGACGACTACGAGAC | 175       |
|         | R        | TCGTCTCTTCTGGTTGGTAGT   |           |        | R        | GAGTGAGTGACTGGTTGAAGGT  |           |
| Cacnb1  | F        | AGATGACCGACAACAGGAACC   | 100       | Gnat2  | F        | TGGACGTCATCAGGAAGTTGT   | 186       |
|         | R        | CAGCCCTCCAGCTCATTCTTAT  |           |        | R        | CGATGATGCCTGTTGTCTTGAC  |           |
| Clcn3   | F        | GAGCATCTCGAGCAACTAAAGC  | 162       | Gnaz   | F        | CAGAGAGCAAGGGTGAGATTACA | 139       |
|         | R        | TTCTGTCTCTCTCTGTCCTCA   |           |        | R        | AGGTCGTTCAAGGTAGTAGGCT  |           |
| Clcn4-2 | F        | TGGAGTCTTTGGGGGTTTATGG  | 114       | Ifnar2 | F        | ACTGGCCCCCTATGAGAGAAGAA | 191       |
|         | R        | ACCGCAATAACCTCCAACACT   |           |        | R        | TCGTCTAGGAGGATGGTGTCTT  |           |
| Clcn6   | F        | ATCCTTGGGGAGACACAGGAA   | 152       | Insig1 | F        | GCTGTATTGCCGTGTTTCGTT   | 111       |
|         | R        | CACTTCAACGCCTCGTATCTT   |           |        | R        | TCCACCACAAACCCAAAGAGA   |           |
| Clcn7   | F        | CGACACAGCGTCTAATCACAAC  | 136       | Kcnb1  | F        | GGAGAAAAATGGAGAGGGCGT   | 180       |
|         | R        | GGACCTCTCCACAAACACCTT   |           |        | R        | TTCAAGTGCTGCGGACTAGAC   |           |
| Fpr1    | F        | GGTTCATCATTGGGTTTCAGCAC | 124       | Kcnc3  | F        | CAGAAGACAAGAGCCCAATCAC  | 128       |
|         | R        | ACAAAGGAGAGAACCCGCAAA   |           |        | R        | GCGGGACTTCTCGTAACCTTT   |           |
| Fpr2    | F        | TCCCTGCCTTATAGTCTTGAGAG | 108       | Kcnc4  | F        | GGAGGTAGAAACAGAGCCCAT   | 185       |
|         | R        | TGGGGCCTTTAACTCAATGTCT  |           |        | R        | ACAAACCACTCAATCCACCTC   |           |
| Fpr3    | F        | CATTCTCACTTTGCCCTTTTCC  | 100       | Kcnd1  | F        | ACTGCAGCCCTGGTTTTCTAC   | 128       |
|         | R        | AACAGAGTTGCCCCAGGATAC   |           |        | R        | TCACCACACGACTGCTCTTTG   |           |
| Gcg     | F        | CACCAGCGACTACAGCAAATAC  | 169       | Kcnh2  | F        | ACCTGCTTACTGCCCTCTACT   | 133       |
|         | R        | CTGGCCCTCCAAGTAAGAACT   |           |        | R        | GACTTTCAGGACGGGCATAT    |           |
| Gna11   | F        | TCCGCACAATCATCACCTACC   | 136       | Kcnh6  | F        | TACAGCAAATGCCCCAAGTC    | 105       |
|         | R        | CTCTGTGGCCCATCAAACCTCA  |           |        | R        | GTCTGTTTCATCTGGGCTTGA   |           |
| Gna12   | F        | CTAGAAAGGCCACCAAGGGAAT  | 152       | Kcnj11 | F        | TCGTGTCCAAGAAAGGCAACT   | 110       |
|         | R        | CGAGGACACCATGAACAGGATA  |           |        | R        | AGTGTGTGGCCATTTGAGGTC   |           |
| Gnai1   | F        | CGGAAGAGGAGTGAAGCAGTA   | 152       | Kcnj9  | F        | TCGTCTCACCTCTCGTCATCA   | 178       |
|         | R        | CCCAGCAAGCACGAAAAGTT    |           |        | R        | CACAACACTTCATCCACCAGGTA |           |

|        |   |                         |     |         |   |                         |     |
|--------|---|-------------------------|-----|---------|---|-------------------------|-----|
| Kcnk1  | F | TTGTCACCGTTTCCTGCTTCT   | 157 | Syt7    | F | GAAAGCCATCAACGACCTAGAC  | 120 |
|        | R | AACTTCTGGTTGTAGCCTTCCC  |     |         | R | TAAGGGGCGTAGGGTGAAATG   |     |
| Kcnk12 | F | CTACTTCTGCTTCGTACCTTCA  | 159 | Syt13   | F | AGAACCTCCACTCCAACCAATC  | 110 |
|        | R | ATGGAGATGACGTTGAAGAGCG  |     |         | R | GGCCCGTTTTGTCTGTTTCTT   |     |
| Kcnk16 | F | GTCATTCTCATCTCCCACCCA   | 117 | Taar1   | F | GCGGCTGTTCTCCCTTCTTTA   | 188 |
|        | R | AACAACATAGTCCCGAAGCC    |     |         | R | GCTTTGTGGTGCTTGGCTTTT   |     |
| Kcnn1  | F | TTAACCGCGTCACCTTCAACA   | 157 | Taar2   | F | GAGGCTTACGCTGATGGAATTG  | 147 |
|        | R | CTGGTCACCTTCTGCTTATCGT  |     |         | R | GCCGTAAATCCCCACCATCAT   |     |
| Pde3b  | F | CCGTCGTTGCCTTGATTTCC    | 164 | Taar3   | F | GCGAACACAAAAGGAGCAGTAG  | 191 |
|        | R | CTTGGGTCAATCAGCAGGTCT   |     |         | R | TACCCGAGCCATACCAGAAGAT  |     |
| Pde6d  | F | GTGGTTCTTCGAGTTTGCTT    | 119 | Taar4   | F | GGCCTCTCAGAAAGCAAAATG   | 104 |
|        | R | TGATGACATTGCCGTTAGGA    |     |         | R | AGGGTAGCCAACACAACACAA   |     |
| Pde10a | F | GGACAGAGACAAGCGAGATGAA  | 160 | Taar5   | F | TGTCAAGCGGGAAAGAAAAGC   | 126 |
|        | R | GCGAATTACCTTCTCCCACTGA  |     |         | R | AGGGGTGGGGTGATGAAGTTA   |     |
| Plcb3  | F | TTAATCGGCGGCACATCACT    | 133 | Taar6   | F | CAGAGTGGCGAGAAGAGAAAGAA | 137 |
|        | R | AGCTTGGGTTCTCTTCTCTA    |     |         | R | AAATGTAGGCAGGGGTGATGAAG |     |
| Plcb4  | F | AGTGAAGGCAAGGAAGGACAAG  | 154 | Taar9   | F | CCTCCTCTGTTTTGCGTCTCT   | 119 |
|        | R | CGCTGCAGACACACAATATCC   |     |         | R | GCACAGTCCAGAAACCGATACA  |     |
| Plcg1  | F | CGACAGCACCAAGCAAAAGAC   | 116 | Vmn1r1  | F | CTGCTCTCTGGGTTGTTAGT    | 170 |
|        | R | CAAAGCGCAGAAAGGCAAACCT  |     |         | R | GGGAAATGCTGGTGTGTGAA    |     |
| Pld3   | F | TCCTTCTACTGGACCCTCACAA  | 124 | Vmn1r26 | F | TTTATTCTCCGGTCTGTGCCA   | 100 |
|        | R | CAGCGATGCGAACCTTTACAC   |     |         | R | TAGTGGACCTGTATGGTGGAGAT |     |
| Reep1  | F | TACAAGGCTGTGAAGTCCAAGG  | 157 | Vmn1r54 | F | ACTACATCGTGCTCTCTGGCA   | 180 |
|        | R | GCCAGGCTACAAACGCTATTT   |     |         | R | GAGGGAAAAGCTGGTGATATGG  |     |
| Reep2  | F | ACCCTGTACCCAGCCTATTCTT  | 157 | Vmn1r65 | F | GAGGACAACAGAAGAAGTGGCT  | 142 |
|        | R | GCTCAAAGTAGAAGGGGAACCA  |     |         | R | TTGGATGATCTGAATGGGCCTC  |     |
| Reep3  | F | GTGGTGCTGGTGTGTTGAATG   | 121 | Vmn1r66 | F | ACATCCACAGCTCTCAGGTTTC  | 164 |
|        | R | CAGTGATAGAGGGCAAAGACGAT |     |         | R | GACCATCACCAGACACCAACT   |     |
| Reep5  | F | GTGGCTTTGTATCTGGTGTTCG  | 102 | Vmn1r67 | F | GGTGTGTACCTTCTGGCATT    | 174 |
|        | R | GGGACTCTCGATGGCTTTCAT   |     |         | R | CTGAGTCTGGGCACAAAAGTAC  |     |
| Rtp1   | F | TACCCTCTTTCCCCACGTTCT   | 180 | Vmn1r70 | F | AGAGTTTGCAAGGGATTTTCC   | 142 |
|        | R | ACACATTGTGCTTGAGGTTGGG  |     |         | R | AGCACAGGACCAGAGAAGAAC   |     |
| Rtp2   | F | CGAGCAGTGTTACGATGAGGAT  | 174 | Vmn1r71 | F | CCAACACATCCGTAGCACTCA   | 109 |
|        | R | TTCTTGAGGCATCGGTATAGG   |     |         | R | GGTGAGAGAACAAAAGGCCAGA  |     |
| Rtp4   | F | TTCTCCCCATCAAAGAGCTG    | 170 | Vmn1r72 | F | TAACCCAAGGGCTGATGCT     | 178 |
|        | R | GGGCAAATGCAGCAATAGACA   |     |         | R | TGAAAACCATGAGCAGTAGGC   |     |
| Scn1b  | F | ACGTCTACCGTCTCCTTCTT    | 171 | Vmn1r73 | F | ACTAAGAGTATCAGGTCCCAGGT | 101 |
|        | R | CCATCTCTGCCACAAGCCATAT  |     |         | R | ACAATGCAGCTCCCACATTTT   |     |
| Sstr3  | F | GCGAACAGCCTTCATCATCTAC  | 100 | Vmn1r77 | F | ATTGGCCCCTTCTGCTTAGTCT  | 160 |
|        | R | CGACCGCACCTTTACCACAAT   |     |         | R | AGAGTACAGCTCGCACATGATC  |     |

|          |   |                         |     |          |   |                         |     |
|----------|---|-------------------------|-----|----------|---|-------------------------|-----|
| Vmn1r78  | F | TCTACTCTGCTTCTCTGATGGCT | 160 | Vmn1r212 | F | GGAGTTCTGACTGGATTTTGGC  | 135 |
|          | R | GATAGTTTTGGTGGCTTGGTCC  |     |          | R | AACCTTCTTGGTGGGATCTGA   |     |
| Vmn1r80  | F | GTTACGGCCTACTCCAATACC   | 139 | Vmn1r224 | F | ACATTGGCTCCAGAACATCTCC  | 141 |
|          | R | GAAAGCAGGGTAGAAACAGGTT  |     |          | R | CCAGCCACCAACCAGGATTATA  |     |
| Vmn1r82  | F | AACTCTGGCCAACCTCTTGTC   | 181 | Vmn1r228 | F | GGGACATGGCAGTAGGAATAGT  | 170 |
|          | R | GATTATGGCCGCTTGGAAACA   |     |          | R | GGAGAGAAGGATCAAGGCGTTG  |     |
| Vmn1r84  | F | GGTCTGTGTTTGAGCATCATGG  | 178 | Vmn1r230 | F | GCCAGGAATTTGGGAACAGGAA  | 124 |
|          | R | TGGAGTAGGAGAGGACAAAGGT  |     |          | R | CTGTGGGCTTTCGTTTGTGTT   |     |
| Vmn1r87  | F | CTCATCAGAAGAAGCCCGTAGA  | 175 | Vmn1r231 | F | TCGCATGAATCAAGAGCCACT   | 127 |
|          | R | GAAAGGCCCTAGTAACACTGT   |     |          | R | TAATCATCCACCAGCCAGCAC   |     |
| Vmn1r88  | F | CCTACGTTTGCTCCTGGCTTA   | 145 | Vmn1r232 | F | GGAACATGGCAATAGGAGTAGGA | 183 |
|          | R | CACGGCCAACGAGAGTCATAT   |     |          | R | GGGGGAAACCTTTGGAGATAATG |     |
| Vmn1r89  | F | CTTCTCCTCACTCACGATCTCT  | 110 | Vmn1r233 | F | CTGGTCTCTGGCAAATGTAGCT  | 139 |
|          | R | CTACTGTGGAGATGCTGGGAT   |     |          | R | GGCTAGAGGCTTTGGGGAAAT   |     |
| Vmn1r179 | F | ACCAATCGACACTACAGAGGC   | 156 | Vmn1r234 | F | GTGCATCAGCTCTCCCTATACT  | 199 |
|          | R | ACTCCAATGCCTCACAAATGC   |     |          | R | GCACACAGCACCAGGGATAAT   |     |
| Vmn1r194 | F | CTGTTCGTGATCTCGTCTTCCA  | 138 | Vmn1r235 | F | GGCTTCTGCTCTGTTTGTCTTG  | 192 |
|          | R | CTCTGATCTCTGGGCTGAAAGT  |     |          | R | GGACTCCGTGGATGATTGTGA   |     |
| Vmn1r195 | F | GGCATTGCAGGCTGTAAAAT    | 181 | Vmn1r236 | F | CTGTACTATGTAAAGGAGTGCCC | 139 |
|          | R | TACAAAGGAGGAGGAGAGAGGG  |     |          | R | CTAAGAAATGAGGTGCTGCCA   |     |
| Vmn1r196 | F | CACAGTGGTCCAAGCAGTTATC  | 156 | Vmn1r237 | F | TCCTGGGCAACTCCTTCTTAGT  | 160 |
|          | R | GCTGTGTCTCTGATGGAAAGGA  |     |          | R | TTCAACCCAAAGGCAGACACA   |     |
| Vmn1r211 | F | GCTGTAAAGTTGCTGTCTACCTG | 165 |          |   |                         |     |
|          | R | GGCAGAGGAGTGAGGAAAGAAT  |     |          |   |                         |     |

**Table S3. Primers for OR genes used in the biological experiments for MRPrimer.**

| Gene    | Sequence |                        | Size (bp) | Gene    | Sequence |                         | Size (bp) |
|---------|----------|------------------------|-----------|---------|----------|-------------------------|-----------|
| Olfr6   | F        | TCTCACGAGTTGCCTTCTGTG  | 156       | Olfr345 | F        | CACCCCTCTGCACTATTACAAAA | 174       |
|         | R        | CAGTGGCTGAGAGTGGGAATAT |           |         | R        | CAACCAGTCAGAGAGGTCACA   |           |
| Olfr11  | F        | TTCTGGTGTCCATTGGATCC   | 104       | Olfr350 | F        | CTCTTATGGTCACATTGTGGCC  | 181       |
|         | R        | TTGTGGGACTGTGCATGTGAT  |           |         | R        | CACAGAAGCAACTATGCCCTTG  |           |
| Olfr16  | F        | TTCTGGGATTCTCCAGCTTTG  | 168       | Olfr354 | F        | CAGCCATCCCCCATTCTACT    | 114       |
|         | R        | TGAGCTTGCCAACATGCTCA   |           |         | R        | GAACAGCTAGAAACACCAAGCC  |           |
| Olfr17  | F        | CAGCACTGCCATCCTCACATA  | 149       | Olfr356 | F        | TACGCTGCTGATTAGCTAGC    | 158       |
|         | R        | TCAATGCAGCCTTCACCTCAT  |           |         | R        | CTTTGTGGAGGTTGCAGCAAA   |           |
| Olfr38  | F        | CACAAGGCTGGTCATCACATC  | 127       | Olfr368 | F        | GAAATGTGGGCATGGTGCTAC   | 106       |
|         | R        | AGCAAGGGTTTCACAGGCTAT  |           |         | R        | GCAGAGGAATAGAAGGCATCCA  |           |
| Olfr48  | F        | TGTGGGCTTGACTCAGAACATG | 112       | Olfr417 | F        | ATAGACAAGGACAGCCGCATT   | 172       |
|         | R        | GCTGCTGATGGTGACCATGATA |           |         | R        | GTCCACATGCAACATTGGTCAT  |           |
| Olfr50  | F        | GTTCTGCTAGTGATGGTGTCT  | 200       | Olfr429 | F        | AGCCTTGGCAGTAGTCTACTCT  | 106       |
|         | R        | GTGATGACCACCACTGCTAAAG |           |         | R        | TCTCATAAGCTGCCTCTTCAGG  |           |
| Olfr51  | F        | ACCTGCATGCTCTCTTGATA   | 160       | Olfr432 | F        | AGGTATTGTGAGAGCTGTGCTG  | 118       |
|         | R        | ACCAATCCACCAATGGTGAAG  |           |         | R        | GACAATGAAGGCAGAGCCAAAG  |           |
| Olfr65  | F        | ATTGCTTCTACAGATGAGCGGG | 143       | Olfr450 | F        | ATCAGGCTTCATACCCCGATGT  | 121       |
|         | R        | GTGATGTGGACGACATGAGGAA |           |         | R        | GGATGAGCTTTTGGGCTGAAAC  |           |
| Olfr71  | F        | CAGCAACAGAACTGCAGTCTC  | 102       | Olfr452 | F        | CACAAGGCTGGTCATCACATC   | 140       |
|         | R        | ACCAGGTACATGACTGAGCAC  |           |         | R        | CCAATCGGACCACAGCTAGAAT  |           |
| Olfr74  | F        | TGCTAGCCCTTTCTCCTCTGA  | 147       | Olfr552 | F        | GTCAGCAATGTTGTTGGCCAT   | 105       |
|         | R        | CACTGGCTGAACGCATCTTAAG |           |         | R        | AGGCCAATCTTGATGATGAGGG  |           |
| Olfr140 | F        | CATCCTGCATTCAACAGCTCAG | 108       | Olfr553 | F        | CAGTACTTACCCATGCAGTGGT  | 103       |
|         | R        | GCCAGCTCCAGTAAAGGATACA |           |         | R        | GGCAGCCTTAGGCCGAATTAGA  |           |
| Olfr248 | F        | CCATCTGCAATGCCCTCAAATA | 186       | Olfr556 | F        | CTCCATCATTGTGGGCTCAGAT  | 141       |
|         | R        | GCCTCAGAACTGCCCTCATAT  |           |         | R        | AGAGCCATGACCCCAACATGA   |           |
| Olfr273 | F        | TGTTTCATGGCATCCGCATCA  | 136       | Olfr560 | F        | GTCATGGAATCCTCAGTGCTGT  | 151       |
|         | R        | AGCTTGAGGACAGCTAAGACCT |           |         | R        | TCGGTGTCAACATTACAGTCCC  |           |
| Olfr275 | F        | GTGGAAACTGGACCTGTGATTC | 113       | Olfr575 | F        | GGCATTGGATCCATAAAGAGC   | 112       |
|         | R        | ACCATGGAAGTGTCTCCACAG  |           |         | R        | AGCGGTGCATGGATGCTAAA    |           |
| Olfr283 | F        | TAAGCTCAGCCTCTGGGAGAA  | 118       | Olfr577 | F        | TTTCATGCTCAAACGGTTCCC   | 151       |
|         | R        | GTCGATCCGGAATTTGGCATG  |           |         | R        | GTCCACTCCCCTGTAGAAACA   |           |
| Olfr305 | F        | TTCATGGCTCTCCTGGATCAG  | 153       | Olfr592 | F        | CCGCTTACCCTTTTGTCAAGC   | 110       |
|         | R        | TAATCCAGTGCCCCAGGAAAC  |           |         | R        | AAAGGCCATACACAGCGTTGA   |           |
| Olfr307 | F        | CCCTTGACCTACAGCTCCAAA  | 145       | Olfr599 | F        | AGTACTCGCAGCTACTGACCT   | 155       |
|         | R        | GCACAAGCCAGAACGGAAATAG |           |         | R        | GCAGAACGCCAGATTCCATTC   |           |



|          |   |                          |     |          |   |                         |     |
|----------|---|--------------------------|-----|----------|---|-------------------------|-----|
| Olfr606  | F | GGGTAACAGGGAAGCTGTTCCAT  | 124 | Olfr1043 | F | GCCAAATGCGCTGGTGAATTT   | 181 |
|          | R | ACCATAGCAGGCAAGGACATG    |     |          | R | CCGCGGCATGAGAATGACATA   |     |
| Olfr622  | F | AGGTCTAGTTGGCCTGATGAGA   | 179 | Olfr1052 | F | GCCAAGCTCCCAGTATTCTGT   | 136 |
|          | R | GGGCCACTGAAATTCATATGC    |     |          | R | CTCTATGGCTCTCTTTGCAGCT  |     |
| Olfr631  | F | CTCATTGCGCTGTCTATGGA     | 137 | Olfr1055 | F | TTGAGACTGGTCAAGTCGCATC  | 107 |
|          | R | AGTCCCATCATGGGCACAAAG    |     |          | R | TCTATTCAAGGCCTGCTTACC   |     |
| Olfr648  | F | GCTGGCAATGACTGATCTCATC   | 100 | Olfr1089 | F | ATAGGCATCACTAATCGGCCTG  | 125 |
|          | R | GAGGCAGCCTGAAATGTGATT    |     |          | R | TGCAGTCGAGGGTCCACTATT   |     |
| Olfr661  | F | CTGGGCATTCTTGCTATTGTGG   | 145 | Olfr1090 | F | ACCTCAGTCCAGTCATTCTGATG | 135 |
|          | R | CCATGGCAACAAAGCAATGGA    |     |          | R | CTCCAAGTCTATGTAGGGCAAA  |     |
| Olfr745  | F | CGGATCCAAGAACAGCTCCAT    | 163 | Olfr1093 | F | TGGAGTTGTGCATGGTGCTAT   | 167 |
|          | R | CTGATCCCATCTCACAGCACAA   |     |          | R | CAATCAAGCCACCAAGTACAAG  |     |
| Olfr768  | F | GAGCGTAATTGGTAACCTGACC   | 150 | Olfr1106 | F | CACTACTTACCACAGAGGGCTT  | 137 |
|          | R | GACGCCATTGTGTAGAGGAATC   |     |          | R | TCCTAAGCACGATCCCAACAC   |     |
| Olfr788  | F | CATGAACAGGAAACTCTGCACC   | 107 | Olfr1111 | F | TTGCAGATGCTGAGTGCCTTA   | 147 |
|          | R | AAGCACAGTAATCCTGCTGGAG   |     |          | R | AGGTCACGCTTCCGCTAAAA    |     |
| Olfr812  | F | ACCAGTCAGGAGAGTTAGAGTTC  | 128 | Olfr1112 | F | CCTAGCTTGTGGGGACACTTT   | 135 |
|          | R | GAGGATGATCGTTAAGTTCCCC   |     |          | R | GCAGATGGAAGCTTCAGAATGC  |     |
| Olfr827  | F | AGCCTTTATACTAGCAGCCATGG  | 100 | Olfr1124 | F | TGCTGCATTCTTATCCTGGGAG  | 152 |
|          | R | CAACTGGACACAGACACTCCTT   |     |          | R | CCACTTATCCAGGAGCCAATCA  |     |
| Olfr855  | F | GACTGACAAGAAACATGGAGCC   | 109 | Olfr1128 | F | GGGGTCTTTGGAGATACAGAATG | 138 |
|          | R | AGGAAGAGGCTGAAGATGAGAC   |     |          | R | TGCATAGGCAACAGCCACAAG   |     |
| Olfr885  | F | GGTGCCTTGTCCACATAGTT     | 168 | Olfr1133 | F | CCTGTGCATTGAGATTGTCCTTC | 145 |
|          | R | GGGCACTGTTATATTGACACCA   |     |          | R | CACCCTACCACACAGCCAATTA  |     |
| Olfr933  | F | GACTTTGCTCACTGCTAGTGT    | 167 | Olfr1136 | F | GTTGGCTGTTTTCTCCAACCTCC | 136 |
|          | R | CAGCTTTATGTCAGAGCAGGAG   |     |          | R | CTCTACTAGACATGTCCACAGCA |     |
| Olfr988  | F | CATGGGCTTCCTAAATGCTTCTG  | 184 | Olfr1152 | F | ATGGACCATCAGCTTCACATCC  | 116 |
|          | R | TACAGTGCTCACTAGGTTGAACC  |     |          | R | CTCTTGGCAAGCAGGTCCAATA  |     |
| Olfr992  | F | TCACAGGATGCTTGGCTCAAT    | 159 | Olfr1157 | F | CCCTGGCATGTATGGTCCAAT   | 168 |
|          | R | CAAGCAGCAGCAAAATGCAGAG   |     |          | R | GGTAGCATCCAGACACCAGTAT  |     |
| Olfr1009 | F | TGATCGGACCCTATGCTGTTG    | 191 | Olfr1161 | F | CTCAAGATGCAATCATCCAGGGG | 144 |
|          | R | AATCAATACAGCGCCTGCCA     |     |          | R | CACTTTGAATGTGAGCTGGGAG  |     |
| Olfr1010 | F | GCGGTGTTACAGCAAGTACTCT   | 107 | Olfr1162 | F | GCAAGTCGTTAAAGTGGCCTC   | 134 |
|          | R | TGGAGTCGTGAATCAGCACAG    |     |          | R | ACTGAAGAGGGATCTTTGTGCTC |     |
| Olfr1014 | F | CTGGATCTTGGATTGTCCACAG   | 129 | Olfr1164 | F | GGGCTGGGATATTTCTCAAC    | 149 |
|          | R | CTCAGTATACCCAAGTCCAGCA   |     |          | R | CAGGCATGCCATTTGACTGAA   |     |
| Olfr1022 | F | CAACACTGCAGTGATGGATTC    | 146 | Olfr1176 | F | TCCCCACTAAAGGAGCACTACA  | 120 |
|          | R | GGGAATTGGTCCTGATCAGCA    |     |          | R | GTGAGCTCTTTGACTTGAGCAC  |     |
| Olfr1030 | F | CATATGCTGCGTGTGTTAGTCCAG | 193 | Olfr1179 | F | CTTGCTGTGCTGGTGCTTTTA   | 185 |
|          | R | AGGCCACTAAGGAAGCCATAGA   |     |          | R | CAGTCCCATCATGCCTGAATTG  |     |

|          |   |                         |     |          |   |                         |     |
|----------|---|-------------------------|-----|----------|---|-------------------------|-----|
| Olfr1180 | F | ACATGCACTCAGCTCATGGAC   | 104 | Olfr1278 | F | ATTCCAGGGTTGTGTCTTCCAG  | 170 |
|          | R | TCAGTCAGGAGTTTGGGAGTCA  |     |          | R | CCCAAGCACCAGATAGAAGCATA |     |
| Olfr1184 | F | CATGCTACAGGTCTTTGCCATG  | 144 | Olfr1279 | F | GAACGATGTGTGGGTTTCCTC   | 131 |
|          | R | ATCAGGATATGGCACCTGCTTC  |     |          | R | TCTCCACTCCTCCCATAATGTG  |     |
| Olfr1208 | F | GGGTGATGTCCATTCTGACCTT  | 195 | Olfr1289 | F | TAACCCTTGTAGCCATGGCCTT  | 116 |
|          | R | TGTCGTCCCAACAGAATCAC    |     |          | R | AATTGCCCATGCAGTAGCCA    |     |
| Olfr1230 | F | AGGCCTGTGGTCACTTATCCT   | 129 | Olfr1301 | F | GACCATTGTGTTGGTGCAGTA   | 183 |
|          | R | CCACAGTTTCTCAGAGCATGT   |     |          | R | CACAACCTGCCAAAAGCTGACT  |     |
| Olfr1234 | F | GCAAGGCCTACATCCACATTTT  | 164 | Olfr1305 | F | CCACGGATGTGCCTGCTAATA   | 158 |
|          | R | AGCCCACTTACGATGGAGCTA   |     |          | R | GTGCATGCAAGCTTGACAAGT   |     |
| Olfr1239 | F | GTTGTGGTCAGCCCAAGTTTAG  | 171 | Olfr1311 | F | ATGGGGAATCCAGTTGCTTCTC  | 110 |
|          | R | GTGTCCTATGAAAAGCTGGCTC  |     |          | R | AATGCAGGTTGGAGTCAGCAG   |     |
| Olfr1240 | F | AGTCATCGTTGTTTGCTACCTC  | 101 | Olfr1340 | F | GACCTGGATTTTGCAGCTATGG  | 101 |
|          | R | AAGTGTCAGTACATGCTAAGCCC |     |          | R | CCAGGGCTATGCATAGGGTTT   |     |
| Olfr1258 | F | GTTGAACTTCCTCCTTCTGTTGG | 174 | Olfr1389 | F | AAGGCTGGATGTCAGATTGCA   | 178 |
|          | R | GACAAAGTGGCTGAAGGTCTCA  |     |          | R | GACCAAGGCAAGGAAAATGAGG  |     |
| Olfr1260 | F | ATGTGGCTTCCTGGTTTCCAC   | 133 | Olfr1404 | F | ATGGCTGCACTGCAATCATCT   | 125 |
|          | R | GAGCGGAAAAGGTCACAGAT    |     |          | R | CCGCAGAGTGATACCAAAGGA   |     |
| Olfr1262 | F | GCACCATATCCTTCAATGGCTG  | 183 | Olfr1406 | F | GCCATATTCAGGCTGCCATTT   | 141 |
|          | R | CGATTCCAGCTGCTACCACTAA  |     |          | R | GGCTGTGATGACCATAAGGCTA  |     |

**Table S4. Primers used in the comparative biological analysis.**

|     |        | MRPrimer |                         |           | PrimerBank |                         |           |
|-----|--------|----------|-------------------------|-----------|------------|-------------------------|-----------|
| No. | Gene   | Sequence |                         | Size (bp) | Sequence   |                         | Size (bp) |
| 1   | Olf5   | F        | GCTAGGTGGGCTATTGGTTTCT  | 186       | F          | TTGTGGGCACAGAGTGCATT    | 133       |
|     |        | R        | AAGCAACCCCAAAGGATGACA   |           | R          | CCTAGCCATGAAATCATAGCCAA |           |
| 2   | Olf39  | F        | CATGCAGTGCCTCACTCAAGT   | 166       | F          | CAGATGATCCTAAATTGCAGCCT | 113       |
|     |        | R        | CCAGCACATTAGCACAAAGGAAG |           | R          | GAGATGGGAATCAGAAGTACAG  |           |
| 3   | Olf49  | F        | ATTCTGGGCTTTCTCTGAC     | 185       | F          | CTTCCCAAGATGCTAACCAAC   | 172       |
|     |        | R        | GTCCTGTGATGATGTTGGTTAGC |           | R          | GGTGGCATAACGCAAAGGG     |           |
| 4   | Olf78  | F        | CATGCCACCTTCTGCTTATTG   | 138       | F          | ATGAGTTCCTGCAACTTCACC   | 111       |
|     |        | R        | GCTCCGCTCTGTTCTACTAT    |           | R          | TGCTACAGCATAATGGAAGC    |           |
| 5   | Olf97  | F        | TTGGGAATCAGTGCCTGCTC    | 163       | F          | AGCCTTGGCTACATGCAGTTC   | 158       |
|     |        | R        | GGTAGGTAGCAAAGACTGTGCT  |           | R          | TGAAGGGGTTTAGAGTGGGAG   |           |
| 6   | Olf118 | F        | TGCTCCCCACTCCATTACTCA   | 233       | F          | CACTTGCTTGTGGCGATACAT   | 153       |
|     |        | R        | ACTACGGCCACAAAGATTGCA   |           | R          | CTTTATGGCGACCTCAGGTG    |           |
| 7   | Olf122 | F        | GCTGGTAATGCCCTCACGT     | 109       | F          | AGGAGAACAGCTTGTCTGTCA   | 60        |
|     |        | R        | CCTCAAATAGGTGGCAGACGT   |           | R          | CTCCAGGGACCTCAGAGAACT   |           |
| 8   | Olf130 | F        | GTCGCACATGCTGGTAGTAGT   | 223       | F          | TGACACTGGTAGGCAACACAG   | 176       |
|     |        | R        | AAGAATTTCTTCCAGTGTGT    |           | R          | TGTGGCAGTAATTGTCTTGGC   |           |
| 9   | Olf190 | F        | CTGGCTTTTGTGGATGCTTCC   | 162       | F          | CTGTGACATCGTTCCATTGCT   | 263       |
|     |        | R        | TGCCATAGCTCCCAACAAGAA   |           | R          | CTTGGGACACGGGGAAAATATAC |           |
| 10  | Olf218 | F        | GAGCATGGCTATTGTCCAGGTTA | 168       | F          | CTGCAATCCTTAAGGTATTCCGG | 102       |
|     |        | R        | AACACAAGGACACACACGCT    |           | R          | ACCTGGACAATAGCCATGCTC   |           |
| 11  | Olf222 | F        | GGTCATCCTGACAGTGAATT    | 114       | F          | CACCCCAAAGGACTTTATCCTC  | 103       |
|     |        | R        | GCATCATTCCCGTAGTATCAG   |           | R          | CCCAGCATAGCCAGAATGT     |           |
| 12  | Olf235 | F        | GGCTGTAACCTGGAACCTTTCC  | 127       | F          | GTGTTTTGGAATAGCAAATGCCT | 180       |
|     |        | R        | GGGGGCTGTGGAAGTATATAG   |           | R          | GAGCTGGATCGCAAATAGACAA  |           |
| 13  | Olf267 | F        | CTGACTGCTCTGCTGGAACTA   | 168       | F          | ACCCGAGGTTAGAGATTGTTCT  | 111       |
|     |        | R        | TGGAATGGGCAGAAGGAGAATAC |           | R          | TTTGAAGGCGTGAATCCAGGA   |           |
| 14  | Olf340 | F        | CATCAGTCGCATCTCCAAAATG  | 250       | F          | TTGGGACTCCCCATTGAG      | 195       |
|     |        | R        | AGGGAGTTGGCAGTAGATAAAGT |           | R          | TGGAGATGCGACTGATGAGAA   |           |
| 15  | Olf348 | F        | GTAACACTGTCCACCACTTCTTC | 161       | F          | ACCCCTCCACTATACAAGAATC  | 89        |
|     |        | R        | AGTGGCTCCAATGCGTCCATA   |           | R          | AAGGGCACCAGCAAATGATAA   |           |
| 16  | Olf355 | F        | TCCTTTCCCACTTCCATTAC    | 198       | F          | GATGTGCCCTACTGGTGACC    | 336       |
|     |        | R        | AGCAATGCATAGGAAGGGAGTC  |           | R          | AAAAGAGGGTTACCACAGTGAAG |           |
| 17  | Olf362 | F        | TGCCCCACACTTCTCTTTT     | 136       | F          | TGTGCCCGAATGCTTAC       | 122       |
|     |        | R        | GAGAGGCCACGCAAGAGATAAT  |           | R          | TTCCCAATATGGTGGTCAGATA  |           |

|    |         |   |                         |     |   |                         |     |
|----|---------|---|-------------------------|-----|---|-------------------------|-----|
| 18 | Olfr401 | F | CCCACAGGCTTACAGTTCCAT   | 187 | F | TATCCCTCTATGTTGGGTCGG   | 150 |
|    |         | R | CAGGATGAGGCCACCAAAATTC  |     | R | CAGATGGCTAGGAAGCGGT     |     |
| 19 | Olfr424 | F | GGTTCCTGCATCTTTGGCTTTC  | 124 | F | GGATGGATACCCGTCTTCACA   | 127 |
|    |         | R | CCAAACGCAACACAGGTTCAA   |     | R | ATGGTCCTTTGCTTACTGATGAG |     |
| 20 | Olfr433 | F | GGGCACTGACAAACTTATTGCCT | 119 | F | CATGTCTCACTGCCAGTTTACC  | 105 |
|    |         | R | TCTTTTCACTGACCCTTTCACG  |     | R | CATGCCACCTTGTGCTCAC     |     |
| 21 | Olfr453 | F | TCTGGGCTTAGGAGGGATTGA   | 145 | F | CCTGGTGGATGTGTCTTATGC   | 135 |
|    |         | R | ACCACCGACCCAAGAAACAAT   |     | R | AACTCAATCCCTCCTAAGCCC   |     |
| 22 | Olfr460 | F | GTGGGAAACACGGTCATCATTG  | 127 | F | TCCTGGCTCTGTAAACCTACG   | 188 |
|    |         | R | GCATCACGGGCACAATAACAG   |     | R | CGGGCACAATAACAGTTGTAACC |     |
| 23 | Olfr461 | F | CCTTTTTCGACCTTCCTTCT    | 181 | F | GGCACCAAAGAACTACACCAC   | 109 |
|    |         | R | AAGAAGCCCCTGGAGCATATA   |     | R | GACGTTTGTGACGATGGACTA   |     |
| 24 | Olfr510 | F | TGGCTTCCATTGACATAGCCA   | 100 | F | ACCACACTGTAGTCACAGAGT   | 123 |
|    |         | R | ATGCCACACCCAATGTAGGAT   |     | R | TGGTGCTTAGATTCCCAGACA   |     |
| 25 | Olfr539 | F | TCTGTACCTCTTCTGTGATTCT  | 245 | F | GTGGTCCCAAGGTTATCACCC   | 156 |
|    |         | R | AAGACCGATGAACCATAACAACA |     | R | TGCAGCCATAAGACAACAAGG   |     |
| 26 | Olfr558 | F | TCCCTTTGTGTCCCTCTACCT   | 153 | F | TCAATAGCAATGAATCCAGTGCC | 114 |
|    |         | R | TTGGCATGGATGAGGTGGAAA   |     | R | GCACAGCAATAAGGTAGAGGGAA |     |
| 27 | Olfr569 | F | GCCTTGTGGCTATCACTGAC    | 109 | F | GGAATCCCAGGGTTGGAGAAT   | 87  |
|    |         | R | GGATGAGGCAGGCGTTGTATT   |     | R | GGTGATATTTCCAGTCAGTGCC  |     |
| 28 | Olfr611 | F | GAGGAGGCTCTACTTTTGTCTG  | 134 | F | TTCCCCACACTGTTGAGAATCT  | 141 |
|    |         | R | CCAAGGTAGAGAGCACCACAA   |     | R | CACATACCAATCGAAGGCCAT   |     |
| 29 | Olfr613 | F | TGGTTAGAGCGGAGCAGAATC   | 247 | F | CCTTCTGGTTAGAGCGGAGC    | 110 |
|    |         | R | AGTGGAGCACAGATAGCAACC   |     | R | CCAAGACACTAGGCATTGTTGAC |     |
| 30 | Olfr619 | F | GCTCGCATACTTTGTGCTGTG   | 215 | F | GGCTCACCTATTGTGGGAAGA   | 74  |
|    |         | R | GGATTGAGGGAGGGTGGTAGA   |     | R | ACAGGCCAACCTGGCAATG     |     |
| 31 | Olfr632 | F | CTTCTGTGCTGGGGGTGTTAT   | 128 | F | ATGAAGGTGTCTATCCACCACG  | 100 |
|    |         | R | GTCCAGAGCCATAGCAAATAGC  |     | R | GCAGTGAAATCCAATGATGAGCC |     |
| 32 | Olfr635 | F | AAACACCACCATCCTAACCCTTA | 188 | F | CCATCCTAACCCTTATCCGCA   | 117 |
|    |         | R | GGAAGAAGAAGTGGGCAAAACA  |     | R | GCATGACTGTAGGGAGTGTGG   |     |
| 33 | Olfr653 | F | CTTCCACCCTCCCACATTGT    | 112 | F | CCACCCTCCCACATTGTTTT    | 78  |
|    |         | R | ACCATTTCCAACCAGAGCAAGG  |     | R | GAGCAGAAGGGAATAGCAATCC  |     |
| 34 | Olfr677 | F | AGCTTGTGCACCCATCAAGAT   | 156 | F | CCTTGTGGGAAACATCACCAT   | 175 |
|    |         | R | GTGTTTAGCGCCTTCAATCGG   |     | R | GCCCCGAAACTGATCTCC      |     |
| 35 | Olfr693 | F | CGCCTCAGTTATCAGTCCCAAA  | 107 | F | CCATGTACCTCTTGCTTGAGC   | 67  |
|    |         | R | TACCCAGTGCCAGTTCCAAGAA  |     | R | TTGGGACTGATAACTGAGGCG   |     |
| 36 | Olfr694 | F | CAGCCCTGTACTTTTTAGCCATA | 122 | F | GAGCTGCTCTGTGCCACTATC   | 173 |
|    |         | R | AAGCAAGTCCATGAGAGAGAGC  |     | R | ATGGCCTTTGGAGTGATGACT   |     |
| 37 | Olfr740 | F | TCCTGTTCTGTTCTTCTCTTC   | 178 | F | CCCGACCTCTGAGCATGAAG    | 83  |
|    |         | R | TCAGAGGTCGGGCTTAGATACA  |     | R | TCACAGGATTAACGAGTGGAGT  |     |

|    |             |   |                         |     |   |                         |     |
|----|-------------|---|-------------------------|-----|---|-------------------------|-----|
| 38 | Olfr799     | F | TCCATTGCCTATGCTGCTTGTA  | 214 | F | ACAGATGACATTAGGCTGCAAA  | 227 |
|    |             | R | GGCTAATTGGTGGGAGAACGA   |     | R | GCAATGGACTTATCCCCAGATG  |     |
| 39 | Olfr828     | F | GCTCTTCATTCACTACCTCAGTC | 144 | F | GGACTGATGGTGTGCGGTT     | 444 |
|    |             | R | TTGCCCTATTGATGTGCTTCC   |     | R | CCTATTGATGTGCTTCCCAAGG  |     |
| 40 | Olfr830     | F | TGTTGGCCATCAAGTGTGACTT  | 182 | F | CCTTATGAACCCAGTTTCTGTG  | 75  |
|    |             | R | AACTACAACAAAGCAGGCCTGG  |     | R | TGCAGCAGACCATTTACAATACT |     |
| 41 | Olfr911-ps1 | F | TGGGGCTGGAAAATGGTTCTT   | 124 | F | TTGTGCCAGTGTATCATCTTT   | 120 |
|    |             | R | TTCCCCACTGCTGTTGTTGT    |     | R | CAGCAAGTATATGGGAGCTACAG |     |
| 42 | Olfr918     | F | CCACAGGTCTGCTCAATGCTA   | 154 | F | TGCTACTCTTCTGTATCCAGTCC | 251 |
|    |             | R | AAGAAAGTTGGAGGAGGGGCA   |     | R | CCAGCAAATGCCATCCCATAAG  |     |
| 43 | Olfr960     | F | AGCAACCTCTCTATCTCTGACAT | 148 | F | AGCCTGTGAAGATTCCTCTCT   | 230 |
|    |             | R | AGCACAAAACGCATCCAATACA  |     | R | GGTTGGGCTGTAGGTAGATGAC  |     |
| 44 | Olfr967     | F | GATGTCCTATCAAGTCTGCACC  | 175 | F | GCTGGATTAACAAGCACACCA   | 72  |
|    |             | R | AAAAGTAGGGGAGCAAGAAAGC  |     | R | CGTTACTGCATAGATTCCGAGG  |     |
| 45 | Olfr978     | F | TGGCCCTGGTCTTTCATCTA    | 174 | F | GGCCCTGGTCTTTCATCTAC    | 63  |
|    |             | R | TGAGGAAGCACAGACCCATAT   |     | R | GGCCACAATCCATCTACAGC    |     |
| 46 | Olfr995     | F | GTTGTCCCAATCATCAGCCTTTC | 100 | F | TATGCCACATTTGCGACCAGT   | 326 |
|    |             | R | TGACCAACACAGTGAACGTCA   |     | R | AACACAGTGAACGTGAGTTAAA  |     |
| 47 | Olfr1033    | F | ACCTTACAGCAGTTGGCATATTT | 195 | F | TCCACCCCTCATCAAGATGG    | 258 |
|    |             | R | ACTTCTGGCAATCACTTTGTCC  |     | R | GACTCCTCAGTGGGTCGTCT    |     |
| 48 | Olfr1053    | F | GTGAATGTGCTACCCAGTTGTC  | 172 | F | CATCACTGCCTGGGTTTCATCT  | 108 |
|    |             | R | CTGTAGAGGTACGGGATGCC    |     | R | GGTTGCCCATGACTGTGACT    |     |
| 49 | Olfr1079    | F | TCCTGATCATCCTTGGCTCCTAT | 128 | F | ATGTCACAAACTGTATGTTGGGT | 253 |
|    |             | R | CCCATAAAAGACAGAAACCACGG |     | R | GCAGATAGGAGCCAAGGATGA   |     |
| 50 | Olfr1145    | F | TGACTCAGACTCCGACTAAATCC | 120 | F | CTGTCTGCACTGTCTCCATC    | 391 |
|    |             | R | GGAGATTGGGAAGAGCAGAGAA  |     | R | TCTGAATTGGAATGCCACTTAGC |     |
| 51 | Olfr1204    | F | CCGCAGTTGCAGAAAATCTTG   | 201 | F | TCACACAGAATCCGCAGTTG    | 113 |
|    |             | R | ATGGAAAGTATCAGCAAGCAGTT |     | R | CAGTTGGCTGTTTGAATGGTG   |     |
| 52 | Olfr1225    | F | CCACAGCTCTGAAGGGAAATTT  | 171 | F | TGTGGCAATCTGTGATGGTG    | 265 |
|    |             | R | GAATTGAGCAATGGGGTCAACAC |     | R | TGTGGGCTTTCAGAGAGTACA   |     |
| 53 | Olfr1255    | F | CTCTTGGTCTCTTTGTTGCTGC  | 229 | F | AAAAGGAACGTGACTGAGTTCAT | 244 |
|    |             | R | GCTTTATCAATTGGCAGAGTGGT |     | R | GAAGGGAGTCCGCAATCAGC    |     |
| 54 | Olfr1284    | F | GTAAGCATCATCGTGGGAAACC  | 133 | F | GGACTCTCCAGTTCTTGGAAAAA | 78  |
|    |             | R | TGGGCACTGTGGTAGAGGAAAG  |     | R | TCCCACGATGATGCTTACATAGA |     |
| 55 | Olfr1295    | F | CTCCAGGGACTTTCCCACTCA   | 198 | F | GGGTTGTGGTTGTAACCTGC    | 104 |
|    |             | R | AGGAGTGATGTTGAGGAAAGAC  |     | R | GACAAAGCCTTAGATGCTCCAG  |     |
| 56 | Olfr1297    | F | CATTGCTTCAGGAGAGGTGGTAT | 178 | F | GGTATTGTTGGCTTAAATGGCCT | 372 |
|    |             | R | CTGCAAAAGCACTACCACGTG   |     | R | GCCTTGAAGCTCCAGTTTTC    |     |
| 57 | Olfr1303    | F | CTGCTCTGTTACTTCCCCAA    | 120 | F | GTGTCTGCGTTTGTGTTTCTG   | 106 |
|    |             | R | ACCATCTCCACTCCACCAACT   |     | R | GGATGTTCCAGCCATGCTTAAT  |     |

|    |          |   |                         |     |   |                          |     |
|----|----------|---|-------------------------|-----|---|--------------------------|-----|
| 58 | Olfr1331 | F | CCTTTGTTACCACCACCATGC   | 114 | F | TTTGGTCGTTACTCCAATCTCCC  | 240 |
|    |          | R | TTATACCCAGGCCACCGAACAT  |     | R | AGCAATGGTGTA AAAAGCACTGT |     |
| 59 | Olfr1336 | F | CCTCTTCTTA ACTTGTCTGCAC | 142 | F | GAGTTGGGCAATGTGACCAGA    | 249 |
|    |          | R | ATGGCATGCCTAGAACTGTCCT  |     | R | CCCCATGAGTAGTGTGGGC      |     |
| 60 | Olfr1344 | F | GCTTTGCACTCTCTGTACCCA   | 200 | F | GAACCCTCAATGATT CAGGAACC | 129 |
|    |          | R | AGAGGCTAGGATGAGGACCAAA  |     | R | GGGCACCATTACCCAGCAC      |     |
| 61 | Olfr1353 | F | TCTGGTTCTGGTGTCTTGGATTG | 101 | F | AACTTCTTGCTCACTATCATGGC  | 584 |
|    |          | R | AGTGTGGGATTTCTGGCTGTG   |     | R | ACGAAGAGTCTTTTTAGGGCAC   |     |
| 62 | Olfr1356 | F | CAGTCCTTCTCTTTGGGTTGC   | 145 | F | AACCTGTCCATAGCTGACATCG   | 76  |
|    |          | R | CGATGTCAGCTATGGACAGGT   |     | R | TGCTTTGTGTGCGGATATTCT    |     |
| 63 | Olfr1358 | F | GCCTTGATTGAGACCTGCATGA  | 144 | F | CCATCCCTTGC GCTACTCTG    | 112 |
|    |          | R | GCTCAGGAAGAAGATGCCAAGT  |     | R | GAAGATCATGCAGGTCTCAATCA  |     |
| 64 | Olfr1388 | F | GGCCTTCTTTTTGGTGGGATTC  | 127 | F | TCTCTCTCGACTGGACCTTCG    | 153 |
|    |          | R | GTCCAGTCGAGAGAGAGCAAT   |     | R | ACACACCTTTCATAGCTGATGG   |     |
| 65 | Olfr1392 | F | CTAACTCTCTTTGGGAACACTGC | 203 | F | CTGGCCTGCACTAGAACTCAT    | 78  |
|    |          | R | GTTATGAAGAGCTGAGACACACA |     | R | ATGGCAGTGTCCCAAAGAGA     |     |
| 66 | Olfr1408 | F | ACAGTAAGGACCAAGACCAGCT  | 125 | F | AAGATTGCCTCATCTGATGGC    | 243 |
|    |          | R | CCTACACAACACTTTCCGCAGA  |     | R | CCTACACAACACTTTCCGCAG    |     |
| 67 | Olfr1411 | F | CTCTCATATGATGGCTGTGTCCT | 102 | F | CTGCCATTGCCCTAATGC       | 210 |
|    |          | R | ACAGATACACCTTGCCCTGT    |     | R | CCAGACGGGTACAGGTTGTAG    |     |
| 68 | Olfr1420 | F | ATTAACCACTCACTCCACACCC  | 185 | F | GGGGTGCAGATGGTGATTTTT    | 177 |
|    |          | R | TCAGCTCCACCCAAGAAAACA   |     | R | CAGTGGTGCAATGGATGATGAT   |     |
| 69 | Olfr1443 | F | GACTGGAAACTTGGGGATGCT   | 121 | F | ATGGAGAACAGGACAGAGGTG    | 143 |
|    |          | R | TGGGGTAACAGCAGAGGAGTA   |     | R | AGAATCAGCACAAGCATCCCC    |     |
| 70 | Olfr1444 | F | TTTCTTCTTCGTGGGTTTGC    | 154 | F | GGTTGACAGATGACCCCAATC    | 117 |
|    |          | R | GCCACAGGTGTAAGAGCCAAT   |     | R | GGTGGGAATCCGAGAAGATGA    |     |
| 71 | Olfr1465 | F | CATGAGTGGGCTCTAAAAGGA   | 189 | F | ACATTGGGGACACCTTCAATC    | 254 |
|    |          | R | AGACACACACACACCTGAA     |     | R | TGCGGCACAAGTGGATACAG     |     |
| 72 | Olfr1494 | F | AGTCCATTCTGCTGATTTGT    | 167 | F | CCCTCTACACTACAGCCTCAT    | 147 |
|    |          | R | AGCGGGCCTCAGATATACA     |     | R | TGGTTGATTTCTGGTCATGTC    |     |
| 73 | Olfr1509 | F | CAACTGGGTGTTGGAGATTCTG  | 109 | F | CAACTGGGTGTTGGAGATTCTG   | 108 |
|    |          | R | GAGACGTGGCGTGAAGACTAT   |     | R | AGACGTGGCGTGAAGACTATG    |     |
| 74 | Olfr1511 | F | CGCCAGCAAGGTTATCGCATT   | 165 | F | TTCTGGGTGTGCTCTCCTTC     | 101 |
|    |          | R | GTGCACAACCTCCCATTCATGA  |     | R | ACCAAATGCGATAACCTTGCTG   |     |

## Evaluating the computational efficiency and scalability of MRPrimer

To demonstrate the computational efficiency and scalability of MRPrimer, we measured the elapsed time required for design of complete sets of validated primer pairs for the human and mouse CCDS databases. We conducted most of the computational experiments on a MapReduce cluster of one master node and 40 slave nodes, in which each node consisted of two Intel Xeon 8-core 2.6 GHz CPUs with 64 GB memory and a 6 TB HDD. Those nodes are connected with each other via a 1 Gbps network. All computing nodes were running on CentOS Linux version 6.4 and Apache Hadoop version 1.2.1. For the Hadoop configuration parameters, we set the number of Map per node to 4, the number of Reduce per node to 4, the Java heap memory size for Map to 8 GB, and the Java heap memory size for Reduce to 16 GB.

MRPrimer exhibited good performance in terms of computation time. Table S5 shows the elapsed times of MRPrimer at each step for human and mouse CCDS data sets. Even though MRPrimer designs all feasible and valid primer pairs, without omitting any, it finished within 1 or 2 hours. Once the results are obtained, users do not need to run the analysis again, unless the filtering constraints are changed; instead, they simply pick the appropriate primer pairs (usually the top-1 primer pair) for use in their experiments.

**Table S5. Elapsed times (sec) of MRPrimer from Step 1 to Step 7 for human and mouse CCDS data sets (Steps 4 and 5 are performed twice at #mismatch = 1 and at #mismatch = 2).**

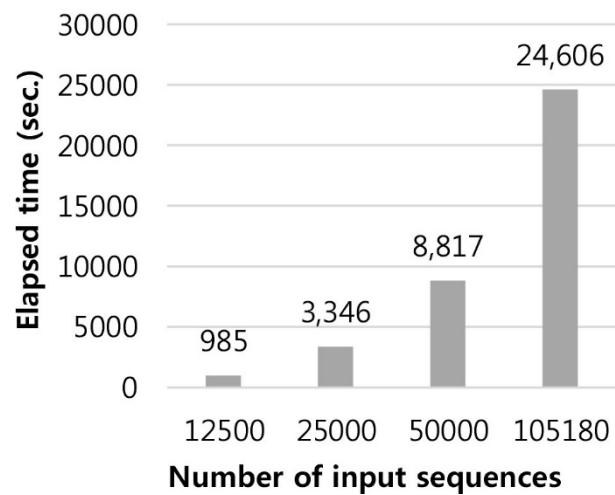
|       | Step 1 | Step 2 | Step 3 | Step 4 | Step 5 | Step 6 | Step 7 | Total |
|-------|--------|--------|--------|--------|--------|--------|--------|-------|
| human | 69     | 60     | 65     | 3648   | 56     | 586    | 29     | 4513  |
| mouse | 50     | 50     | 59     | 2590   | 55     | 224    | 30     | 3058  |

MRPrimer also exhibited a fairly scalable performance in terms of database size (i.e., the number of DNA sequences). To demonstrate this feature, we used a much larger DNA database: 105,180 DNA sequences of *Homo sapiens* from the Ensembl site (<http://asia.ensembl.org/biomart/martview/>). Figure S4 shows the elapsed times of MRPrimer as the number of sequences was varied from 12,500 to 105,180 (i.e., an entire database). Even for 105,180 sequences, MRPrimer designed all feasible and valid primer pairs within a reasonable time of less than 7 hours. Because MRPrimer is based on MapReduce, users can reduce time easily by simply adding more computers to the cluster.

Furthermore, MRPrimer was very efficient in terms of computational resources, i.e., the number of computers or the computing power of each computer. To illustrate this feature, we performed the same experiments for human and mouse CCDS data sets with a small-scale cluster of commodity PCs. The cluster consisted of one master PC and ten slave PCs, each of which had an i7-4770 4-core 3.4 GHz CPU, 16 GB memory, and 3 TB HDD. Here, we used the same number of mappers and reducers (four and four, respectively), but smaller Java heap memory sizes (4 GB for Map and 8 GB for Reduce) due to the small memory capacity of the PCs. Table S6 shows the elapsed times of MRPrimer, which still finished within 2 or 3 hours.

**Table S6. The elapsed times (sec) of MRPrimer from Step 1 to Step 7 for human and mouse CCDS data sets with a small cluster of less powerful computers (Step 4 and 5 were performed two times at #mismatch = 1 and at #mismatch = 2).**

|       | Step 1 | Step 2 | Step 3 | Step 4 | Step 5 | Step 6 | Step 7 | Total |
|-------|--------|--------|--------|--------|--------|--------|--------|-------|
| human | 303    | 106    | 165    | 7583   | 56     | 557    | 69     | 8839  |
| mouse | 259    | 91     | 110    | 3880   | 50     | 272    | 70     | 4732  |



**Figure S4. Elapsed times of MRPrimer as varying the database size.**