### Supplementary Table 2: Overview of selected individual peptides for immunological testing.

**Abbreviations:** aa: amino acid; FC: fold change; HLA: human leukocyte antigen; ID: identifier; n.a.: not applicable; n.k.: not known.

#### HLA class I:

| Sequence | Source protein | UniProt ID | aa position | Mass [Da] | HLA | HLA allele | Relative score [%] | IC₅₀ score [nM] | Evaluated in patient (UPN) | FC value (log2) | Frequency in CRC patients | Frequency in RFA patients | Involvement in cancer | Detected in patient | Detected by |
|----------|----------------|------------|--------------|-----------|-----|------------|---------------------|----------------|---------------------------|----------------|--------------------------|---------------------------|-----------------|-------------------|-----------|
| TTDCLOILAY | CTNNB1 / PLAK | P35222 / P14923 | 297-306 | 1139.55 | A*01 | A*01:01 | 87.2 | 10 | IRISS12 | 2.6 | 0/35 | 1/5 | yes | IRISS12 | MS/MS |
| DTDSAIGSFRY | DUS27 | Q5VZP5 | 888-898 | 1230.55 | A*01 | A*01:01 | 82.1 | 9 | IRISS05 / IRISS09 / IRISS12 | 4.8 | 4.8 | 2/35 | 2/5 | n.k. | IRISS05 / IRISS12 | MS/MS |
| PSENNKLTLY | VAMP7 | P51809 | 37-45 | 1964.51 | A*01 | A*01:01 | 75.0 | 22 | IRISS09 | 1.3 | 3/35 | 1/5 | n.k. | IRISS12 | MS/MS |
| VVIGNIGAL | IF6 | P09912 | 106-114 | 854.52 | A*02 | A*02:01 | 66.7 | 48 | IRISS08 | 7.0 | 0/35 | 1/5 | n.k. | IRISS12 | MS/MS |
| GLALKVQEV | TEN54 | Q812W8 | 484-493 | 955.57 | A*02 | A*02:01 | 83.3 | 2 | IRISS05 / IRISS08 | < 0 | < 0 | 9/35 | 1/5 | yes | IRISS05 / IRISS08 | MS/MS |
| KYISGPUHEL | CENPF | P49454 | 1271-1279 | 1042.54 | A*24 | A*24:01 | 77.4 | 6 | IRISS01 | 4.2 | 2/35 | 0/5 | yes | MS/MS |
| IYFEYSHAF | MSH3 | P20585 | 461-469 | 1175.53 | A*24 | A*24:02 | 71.0 | 6 | IRISS06 | 2.5 | 0/35 | 1/5 | yes | IRISS06 | MS/MS |
| VYFSKGEQL | EPHA2 | P29317 | 574-582 | 1099.56 | A*24 | A*24:02 | 77.4 | 11 | IRISS01 | 8.2 | 0/35 | 0/5 | yes | MS/MS |
| NYMPQQLTI | MACC1 | Q62N28 | 394-402 | 1035.51 | A*24 | A*24:02 | 87.7 | 6 | IRISS01 | 1.8 | 1/35 | 0/5 | yes | MS/MS |
| YYQDTPKQI | NOP16 | Q9Y3C1 | 140-148 | 1154.56 | A*24 | A*24:02 | 71.0 | 7 | IRISS01 | 2.0 | 2/35 | 0/5 | yes | MS/MS |
| VLGKKKLSL | ERO21 / ERO22 | P24390 / P33947 | 202-210 | 984.67 | B*08 | B*08:01 | 86.1 | 2 | IRISS09 | 1.6 | 3/35 | 1/5 | n.k. | IRISS12 | MS/MS |
| VPPEKDEM | KDM1A | Q8IWU6 / Q8IWU5 | 41-49 | 1049.7 | B*27 | B*27:01 | 71.1 | 25 | IRISS12 | 1.3 | 9/35 | 0/5 | yes | MS/MS |
| TQRGALFLLF | SULF1 / SULF2 | Q81WU6 / Q81WU5 | 41-49 | 1049.7 | B*27 | B*27:01 | 71.1 | 25 | IRISS12 | 1.3 | 9/35 | 0/5 | yes | MS/MS |
| AEIEADRSY | LAMC2 | Q13753 | 837-845 | 1053.48 | B*44 | B*44:02 | 73.0 | 8 | IRISS01 | 4.5 | 2/35 | 0/5 | yes | MS/MS |
| Sequence | Source protein | UniProt ID | aa position | Mass [Da] | HLA | Evaluated in patient (UPN) | FC value (log2) | Frequency in CRC patients | Frequency in RFA patients | Involvement in cancer | Detected in autologous tumor tissue | Detected by |
|----------|----------------|------------|-------------|-----------|-----|----------------------------|----------------|--------------------------|--------------------------|-----------------|---------------------------------|-------------|
| IPGYIVDDSVRVEQ | AREG | P15514 | 93-106 | 1588.81 | DR | IRISS05, IRISS06, IRISS08, IRISS09, IRISS12 | 2.7, 2.7, 2.7, 2.2, 2.2 | 3/19 | 1/4 | yes | IRISS12 (several length variants) | MS/MS |
| NPDAQYSWLGNIQQH | CEAM5 | P06731 | 442-458 | 2209.06 | DR | IRISS01 | < 0 | 0/19 | 1/4 | yes | IRISS12 (several length variants) | MS/MS |
| REGLIOWDKLLLHTHE | GPA33 | Q99795 | 52-67 | 1951.05 | DR | IRISS01, IRISS09 | 4.7, 4.7 | 0/19 | 1/4 | yes | IRISS01 (several length variants) | MS/MS |
| YELOYYEKE | EPHB2 | P29323 | 469-477 | 1263.57 | DR | IRISS12 | 7.4 | 0/19 | 14 | yes | IRISS12 (several length variants) | MS/MS |
| DPKFITSILYENNVI | EPCAM | P16422 | 177-193 | 1979.06 | DR | IRISS01, IRISS06, IRISS08, IRISS09 | 3.7, 3.9, 3.9, 3.7 | 0/19 | 1/4 | yes | IRISS01 (several length variants) | MS/MS |
| VSVYALKDTLTSRPA | FN1 | P02751 | 1789-1804 | 1619.89 | DR | IRISS05 | 3.4 | 0/19 | 0/4 | yes | | MS/MS |
| DTLTSRPAQGVVTL | FN1 | P02751 | 1797-1811 | 1343.70 | DR | IRISS09, IRISS12 | 2.8 | 0/19 | 1/4 | yes | IRISS12 | MS/MS |
| NPPSMVAAGSVVAAV | CCND1 | P24385 | 198-212 | 1254.66 | DR | IRISS01, IRISS05, IRISS08 | 0.9, 1.1, 1.2 | 1/19 | 3/4 | yes | IRISS01, IRISS05, IRISS08 | MS/MS |
| VYIFQQPASILGS (Gene fusion) | MAOM- SMAD4 | P23368 / Q13485 | 468-473; 224-232 | 1504.79 | DR | IRISS12 | < 0 | n.a. | 1/4 | yes | IRISS12 (fusion) | WTS predicted |
| TLPLPNLVRGTVQ (Mutation V104L) | mERBB3 | P21860 | 96-110 | 1676.01 | DR | IRISS06 | n.a. | n.a. | 1/4 | yes | IRISS06 mutation | WTS predicted |
| TLPLPNLVRGTVQ (Wildtype) | ERBB3 | P21860 | 96-110 | 1661.99 | DR | IRISS06 | n.a. | n.a. | n.a. | yes | | MS/MS |