

## **Older adults lack SARS CoV-2 cross-reactive T lymphocytes directed to human coronaviruses OC43 and NL63**

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**Supplementary Table S1: Demographic characteristics of the study cohort****Young adults****Older adults**

| Donor Nr. | Age (years) | sex | HLA Haplotype           | Donor Nr. | Age (years) | sex | HLA Haplotype          |
|-----------|-------------|-----|-------------------------|-----------|-------------|-----|------------------------|
| 1952587   | 20          | F   | A*02,*03; B*65, B*51    | 1952570   | 65          | M   | A*01,A*31; B*8,B*40    |
| 1952664   | 19          | M   | A*01, *24; B* 08 B*61   | 1952560   | 61          | M   | A*02; B*15, B*18       |
| 1952787   | 22          | M   | A*03; B*07              | 1952564   | 64          | M   | A*01, A*02; B*08, B*44 |
| 1952804   | 22          | M   | A*02, A*03; B*07, B*08  | 1952597   | 69          | F   | A*02; B *44, B*51      |
| 1952818   | 27          | M   | A*01,A*03; B*08,B*35    | 1952637   | 66          | F   | A*02, A*31; B*27, B*60 |
| 1952836   | 21          | M   | A*02, A*03; B*13,B*65   | 1952718   | 61          | M   | A*02, A*68; B*14, B*40 |
| 1952849   | 22          | M   | A*02,A*24; B*07,B*60    | 1952706   | 61          | F   | A*01, A*02; B*15,B*18  |
| 1952890   | 24          | F   | A*02, A*24; B* 35,B*60  | 1952731   | 62          | M   | A*01, A*68; B*40, B*51 |
| 1953028   | 24          | F   | A*01, A*02; B* 08, B*44 | 1953101   | 63          | M   | A*03; B*18, B*35       |
| 1953035   | 21          | M   | A*02,A*03; B*44, B*51   | 1953093   | 63          | M   | A*02, A*32; B*07, B*44 |
| 1953034   | 23          | F   | A*30,A*31; B*13, B*35   | 1953116   | 64          | M   | A*01, A*29; B*27, B*44 |
| 1953105   | 20          | M   | A*01, A*25; B* 08, B*18 | 1953115   | 69          | M   | A*02, A*25; B*18,B*44  |
| 1953084   | 23          | M   | A*01, A*24; B*07, B*61  | 1953122   | 62          | M   | A*03, A*28; B*07, B*49 |
| 1953138   | 23          | F   | A*03, A*24; B*07,B*08   | 1953262   | 68          | M   | A*02, A*32; B*35, B*40 |
| 1953152   | 22          | F   | A*02, A*29; B*37, B*44  | 1953270   | 64          | M   | A*02; B*13, B*51       |
| 1953273   | 21          | M   | A*024, A*68; B*07,B*35  | 1953416   | 65          | M   | A*02, A*24; B*44, B*13 |
| 1953288   | 21          | F   | A*01, A*02; B*08, B*44  | 1953403   | 66          | M   | A*02, A*03; B*07, B*27 |
| 1953922   | 20          | M   | A*01, A*31; B*62, B*35  | 1954928   | 70          | M   | A*02, A*11; B*8, B*37  |
| 1954225   | 22          | F   | A*024; B*38,B*44        | 1955149   | 65          | M   | A*24, A*68; B*61, B*44 |
| 1954843   | 23          | M   | A*02, A*31; B*07,B*62   | 1956269   | 62          | M   | A*02, A*26; B*37, B*38 |
| 1954909   | 21          | F   | A*02, A*03; B*07, B*51  | 1956347   | 68          | M   | A*24, A*31; B*35, B*60 |
| 1954945   | 23          | F   | A*03, A*24; B*65, B*18  |           |             |     |                        |
| 1956339   | 23          | F   | A*2, A*33; B*65, B*35   |           |             |     |                        |

**Supplementary Table S2: IFN- $\gamma$  ELISpot**

| Donor Nr. | RSV | NL63 | OC43 | CD3   |  | Donor Nr. | RSV | NL63 | OC43 | CD3   |
|-----------|-----|------|------|-------|--|-----------|-----|------|------|-------|
| 1952587   | 38  | 351  | 156  | 1163  |  | 1952570   | 77  | 59   | 7    | 4242  |
| 1952664   | 68  | 213  | 112  | 493   |  | 1952560   | 66  | 2    | 0    | 11928 |
| 1952787   | 91  | 420  | 255  | 1213  |  | 1952564   | 12  | 5    | 0    | 7493  |
| 1952804   | 40  | 540  | 209  | 1838  |  | 1952597   | 28  | 96   | 63   | 3921  |
| 1952818   | 25  | 9    | 10   | 1718  |  | 1952637   | 66  | 226  | 46   | 1656  |
| 1952836   | 187 | 614  | 474  | 3162  |  | 1952718   | 18  | 4    | 1    | 5530  |
| 1952849   | 52  | 26   | 0    | 2714  |  | 1952706   | 39  | 171  | 1    | 2225  |
| 1952890   | 72  | 233  | 15   | 2596  |  | 1952731   | 156 | 169  | 12   | 5596  |
| 1953028   | 76  | 260  | 14   | 1696  |  | 1953101   | 46  | 22   | 6    | 6330  |
| 1953035   | 36  | 43   | 19   | 399   |  | 1953093   | 11  | 11   | 1    | 5640  |
| 1953034   | 352 | 242  | 19   | 3387  |  | 1953116   | 80  | 23   | 0    | 2696  |
| 1953105   | 19  | 50   | 10   | 5592  |  | 1953115   | 9   | 2    | 1    | 3997  |
| 1953084   | 79  | 64   | 1    | 5021  |  | 1953122   | 61  | 34   | 14   | 13785 |
| 1953138   | 447 | 48   | 39   | 8288  |  | 1953262   | 154 | 83   | 2    | 420   |
| 1953152   | 199 | 261  | 33   | 2580  |  | 1953270   | 141 | 53   | 41   | 1447  |
| 1953273   | 146 | 672  | 510  | 8000  |  | 1953416   | 5   | 5    | 4    | 1233  |
| 1953288   | 244 | 312  | 288  | 7895  |  | 1953403   | 632 | 25   | 5    | 1292  |
| 1953922   | 262 | 111  | 8    | 5550  |  | 1954928   | 20  | 5    | 0    | 1764  |
| 1954225   | 196 | 116  | 51   | 2455  |  | 1955149   | 353 | 221  | 70   | 12244 |
| 1954843   | 231 | 93   | 32   | 3760  |  | 1956269   | 41  | 24   | 40   | 11494 |
| 1954909   | 33  | 3    | 11   | 10899 |  | 1956347   | 92  | 7    | 11   | 2591  |
| 1954945   | 161 | 1    | 0    | 29147 |  |           |     |      |      |       |
| 1956339   | 66  | 23   | 24   | 2466  |  |           |     |      |      |       |

Supplementary Table S3: Peptides libraries from HCoV-NL63 HCoV-OC43 (Spike protein)

| HCoV-NL63-Pool 1 (S1) |                   | HCoV-NL63-Pool 2 (S2) |                  |
|-----------------------|-------------------|-----------------------|------------------|
| 1                     | MKILFLILPLASGFF   | 41                    | GSTLDVQSRVLRQLR  |
| 2                     | LLVPLASGFFVNSA    | 42                    | GVSRLOPLRLTCLWPR |
| 3                     | ASGFTVPLASGFFVNSA | 43                    | GVSRLOPLRLTCLWPR |
| 4                     | CVNMAVNSAIVQD     | 44                    | GVSRLOPLRLTCLWPR |
| 5                     | CVNMAVNSAIVQD     | 45                    | GVSRLOPLRLTCLWPR |
| 6                     | CVNMAVNSAIVQD     | 46                    | GVSRLOPLRLTCLWPR |
| 7                     | CVNMAVNSAIVQD     | 47                    | GVSRLOPLRLTCLWPR |
| 8                     | CVNMAVNSAIVQD     | 48                    | GVSRLOPLRLTCLWPR |
| 9                     | CVNMAVNSAIVQD     | 49                    | GVSRLOPLRLTCLWPR |
| 10                    | CVNMAVNSAIVQD     | 50                    | GVSRLOPLRLTCLWPR |
| 11                    | CVNMAVNSAIVQD     | 51                    | GVSRLOPLRLTCLWPR |
| 12                    | CVNMAVNSAIVQD     | 52                    | GVSRLOPLRLTCLWPR |
| 13                    | CVNMAVNSAIVQD     | 53                    | GVSRLOPLRLTCLWPR |
| 14                    | CVNMAVNSAIVQD     | 54                    | GVSRLOPLRLTCLWPR |
| 15                    | CVNMAVNSAIVQD     | 55                    | GVSRLOPLRLTCLWPR |
| 16                    | CVNMAVNSAIVQD     | 56                    | GVSRLOPLRLTCLWPR |
| 17                    | CVNMAVNSAIVQD     | 57                    | GVSRLOPLRLTCLWPR |
| 18                    | CVNMAVNSAIVQD     | 58                    | GVSRLOPLRLTCLWPR |
| 19                    | CVNMAVNSAIVQD     | 59                    | GVSRLOPLRLTCLWPR |
| 20                    | CVNMAVNSAIVQD     | 60                    | GVSRLOPLRLTCLWPR |
| 21                    | CVNMAVNSAIVQD     | 61                    | GVSRLOPLRLTCLWPR |
| 22                    | CVNMAVNSAIVQD     | 62                    | GVSRLOPLRLTCLWPR |
| 23                    | CVNMAVNSAIVQD     | 63                    | GVSRLOPLRLTCLWPR |
| 24                    | CVNMAVNSAIVQD     | 64                    | GVSRLOPLRLTCLWPR |
| 25                    | CVNMAVNSAIVQD     | 65                    | GVSRLOPLRLTCLWPR |
| 26                    | CVNMAVNSAIVQD     | 66                    | GVSRLOPLRLTCLWPR |
| 27                    | CVNMAVNSAIVQD     | 67                    | GVSRLOPLRLTCLWPR |
| 28                    | CVNMAVNSAIVQD     | 68                    | GVSRLOPLRLTCLWPR |
| 29                    | CVNMAVNSAIVQD     | 69                    | GVSRLOPLRLTCLWPR |
| 30                    | CVNMAVNSAIVQD     | 70                    | GVSRLOPLRLTCLWPR |
| 31                    | CVNMAVNSAIVQD     | 71                    | GVSRLOPLRLTCLWPR |
| 32                    | CVNMAVNSAIVQD     | 72                    | GVSRLOPLRLTCLWPR |
| 33                    | CVNMAVNSAIVQD     | 73                    | GVSRLOPLRLTCLWPR |
| 34                    | CVNMAVNSAIVQD     | 74                    | GVSRLOPLRLTCLWPR |
| 35                    | CVNMAVNSAIVQD     | 75                    | GVSRLOPLRLTCLWPR |
| 36                    | CVNMAVNSAIVQD     | 76                    | GVSRLOPLRLTCLWPR |
| 37                    | CVNMAVNSAIVQD     | 77                    | GVSRLOPLRLTCLWPR |
| 38                    | CVNMAVNSAIVQD     | 78                    | GVSRLOPLRLTCLWPR |
| 39                    | CVNMAVNSAIVQD     | 79                    | GVSRLOPLRLTCLWPR |
| 40                    | CVNMAVNSAIVQD     | 80                    | GVSRLOPLRLTCLWPR |

| HCoV-OC43 Pool 1 (S1) |                 | HCoV-OC43 Pool 2 (S2) |                  |
|-----------------------|-----------------|-----------------------|------------------|
| 1                     | MFLILSLIPLAFVIG | 41                    | FVNLGMLSHYVMIPL  |
| 2                     | ISLPLAFVIGLSDT  | 42                    | MALSHYVMIPLTNSK  |
| 3                     | FVNLGMLSHYVMIPL | 43                    | YAMPLTNSKLVNDFSK |
| 4                     | FVNLGMLSHYVMIPL | 44                    | YAMPLTNSKLVNDFSK |
| 5                     | FVNLGMLSHYVMIPL | 45                    | YAMPLTNSKLVNDFSK |
| 6                     | FVNLGMLSHYVMIPL | 46                    | YAMPLTNSKLVNDFSK |
| 7                     | FVNLGMLSHYVMIPL | 47                    | YAMPLTNSKLVNDFSK |
| 8                     | FVNLGMLSHYVMIPL | 48                    | YAMPLTNSKLVNDFSK |
| 9                     | FVNLGMLSHYVMIPL | 49                    | YAMPLTNSKLVNDFSK |
| 10                    | FVNLGMLSHYVMIPL | 50                    | YAMPLTNSKLVNDFSK |
| 11                    | FVNLGMLSHYVMIPL | 51                    | YAMPLTNSKLVNDFSK |
| 12                    | FVNLGMLSHYVMIPL | 52                    | YAMPLTNSKLVNDFSK |
| 13                    | FVNLGMLSHYVMIPL | 53                    | YAMPLTNSKLVNDFSK |
| 14                    | FVNLGMLSHYVMIPL | 54                    | YAMPLTNSKLVNDFSK |
| 15                    | FVNLGMLSHYVMIPL | 55                    | YAMPLTNSKLVNDFSK |
| 16                    | FVNLGMLSHYVMIPL | 56                    | YAMPLTNSKLVNDFSK |
| 17                    | FVNLGMLSHYVMIPL | 57                    | YAMPLTNSKLVNDFSK |
| 18                    | FVNLGMLSHYVMIPL | 58                    | YAMPLTNSKLVNDFSK |
| 19                    | FVNLGMLSHYVMIPL | 59                    | YAMPLTNSKLVNDFSK |
| 20                    | FVNLGMLSHYVMIPL | 60                    | YAMPLTNSKLVNDFSK |
| 21                    | FVNLGMLSHYVMIPL | 61                    | YAMPLTNSKLVNDFSK |
| 22                    | FVNLGMLSHYVMIPL | 62                    | YAMPLTNSKLVNDFSK |
| 23                    | FVNLGMLSHYVMIPL | 63                    | YAMPLTNSKLVNDFSK |
| 24                    | FVNLGMLSHYVMIPL | 64                    | YAMPLTNSKLVNDFSK |
| 25                    | FVNLGMLSHYVMIPL | 65                    | YAMPLTNSKLVNDFSK |
| 26                    | FVNLGMLSHYVMIPL | 66                    | YAMPLTNSKLVNDFSK |
| 27                    | FVNLGMLSHYVMIPL | 67                    | YAMPLTNSKLVNDFSK |
| 28                    | FVNLGMLSHYVMIPL | 68                    | YAMPLTNSKLVNDFSK |
| 29                    | FVNLGMLSHYVMIPL | 69                    | YAMPLTNSKLVNDFSK |
| 30                    | FVNLGMLSHYVMIPL | 70                    | YAMPLTNSKLVNDFSK |
| 31                    | FVNLGMLSHYVMIPL | 71                    | YAMPLTNSKLVNDFSK |
| 32                    | FVNLGMLSHYVMIPL | 72                    | YAMPLTNSKLVNDFSK |
| 33                    | FVNLGMLSHYVMIPL | 73                    | YAMPLTNSKLVNDFSK |
| 34                    | FVNLGMLSHYVMIPL | 74                    | YAMPLTNSKLVNDFSK |
| 35                    | FVNLGMLSHYVMIPL | 75                    | YAMPLTNSKLVNDFSK |
| 36                    | FVNLGMLSHYVMIPL | 76                    | YAMPLTNSKLVNDFSK |
| 37                    | FVNLGMLSHYVMIPL | 77                    | YAMPLTNSKLVNDFSK |
| 38                    | FVNLGMLSHYVMIPL | 78                    | YAMPLTNSKLVNDFSK |
| 39                    | FVNLGMLSHYVMIPL | 79                    | YAMPLTNSKLVNDFSK |
| 40                    | FVNLGMLSHYVMIPL | 80                    | YAMPLTNSKLVNDFSK |

| HCoV-NL63-Pool 2 (S2) |                    | HCoV-OC43 Pool 2 (S2) |                     |
|-----------------------|--------------------|-----------------------|---------------------|
| 123                   | MTYSNFGICDGLSPLV   | 163                   | LOENKQLKLVSPKNAI    |
| 124                   | GCADGSLVPRPNSS     | 164                   | IAASFNKANNIVASF     |
| 125                   | SLVPRPNSSDNGISA    | 165                   | NKANNIVASFSSVMDA    |
| 126                   | PRNSDNGISAITANL    | 166                   | IASFNFVNDVDTQTAIE   |
| 127                   | GNISAITANLSPISNV   | 167                   | SVNDVDTQTAIEAHTVT   |
| 128                   | TANLSPISNVTSQVQ    | 168                   | TQTAIEAHTVIALNK     |
| 129                   | SPISNVTSQVQVQ      | 169                   | IHTVIALNKQDVQVQ     |
| 130                   | TSQVQVQVQVQVQ      | 170                   | ALNKQDVQVQVQVQ      |
| 131                   | YQVQVQVQVQVQVQ     | 171                   | DVQVQVQVQVQVQVQ     |
| 132                   | TPVQVQVQVQVQVQ     | 172                   | GSALNHLTSLQRNFQA    |
| 133                   | YQVQVQVQVQVQVQ     | 173                   | LTSQLRHFNQVQVQVQ    |
| 134                   | NGNPRCKLLKQVTSAC   | 174                   | HFNQVQVQVQVQVQVQ    |
| 135                   | KLLKQVTSACKTIEDA   | 175                   | SNSQVQVQVQVQVQVQ    |
| 136                   | YTSACKTIEDALRLSAH  | 176                   | YDRLDSQVQVQVQVQVQ   |
| 137                   | TIEDALRLSAHLETDV   | 177                   | SDQVQVQVQVQVQVQVQ   |
| 138                   | RLSAHLETDVSSMLTF   | 178                   | DOQVQVQVQVQVQVQVQ   |
| 139                   | ELNDSMLTFDSNAFES   | 179                   | LITGRALANLAFVQVQVQ  |
| 140                   | SMLTFDSNAFESVQVQVQ | 180                   | ALNMFVQVQVQVQVQVQ   |
| 141                   | SNFVQVQVQVQVQVQ    | 181                   | VQVQVQVQVQVQVQVQ    |
| 142                   | AVNLSFVQVQVQVQVQ   | 182                   | KYTFVQVQVQVQVQVQVQ  |
| 143                   | GVNLSFVQVQVQVQVQ   | 183                   | GRRLAQVQVQVQVQVQVQ  |
| 144                   | VQVQVQVQVQVQVQVQ   | 184                   | AOQVQVQVQVQVQVQVQ   |
| 145                   | NFSRQVQVQVQVQVQ    | 185                   | ECVQVQVQVQVQVQVQVQ  |
| 146                   | AGRSQVQVQVQVQVQ    | 186                   | SNRQVQVQVQVQVQVQVQ  |
| 147                   | LEDLQVQVQVQVQVQ    | 187                   | CGNTHFVQVQVQVQVQVQ  |
| 148                   | SKVTQVQVQVQVQVQ    | 188                   | IFSVQVQVQVQVQVQVQVQ |
| 149                   | GLVQVQVQVQVQVQ     | 189                   | SAPDGLVQVQVQVQVQVQ  |
| 150                   | VDYQVQVQVQVQVQ     | 190                   | LFHLVQVQVQVQVQVQVQ  |
| 151                   | TKLSQVQVQVQVQVQ    | 191                   | LPTDYQVQVQVQVQVQVQ  |
| 152                   | ADLQVQVQVQVQVQ     | 192                   | KNKQVQVQVQVQVQVQVQ  |
| 153                   | QVQVQVQVQVQVQVQ    | 193                   | SGVQVQVQVQVQVQVQVQ  |
| 154                   | VADEQVQVQVQVQVQ    | 194                   | GRVQVQVQVQVQVQVQVQ  |
| 155                   | VADEQVQVQVQVQVQ    | 195                   | LRQVQVQVQVQVQVQVQVQ |
| 156                   | MAVQVQVQVQVQVQVQ   | 196                   | VLVQVQVQVQVQVQVQVQ  |
| 157                   | SLVQVQVQVQVQVQVQ   | 197                   | SRVQVQVQVQVQVQVQVQ  |
| 158                   | GLVQVQVQVQVQVQVQ   | 198                   | PRVQVQVQVQVQVQVQVQ  |
| 159                   | SAVQVQVQVQVQVQVQ   | 199                   | PRVQVQVQVQVQVQVQVQ  |
| 160                   | FSVQVQVQVQVQVQVQ   | 200                   | SDVQVQVQVQVQVQVQVQ  |
| 161                   | ARVQVQVQVQVQVQVQ   | 201                   | YVQVQVQVQVQVQVQVQVQ |
| 162                   | ALQVQVQVQVQVQVQ    | 202                   | FVQVQVQVQVQVQVQVQVQ |

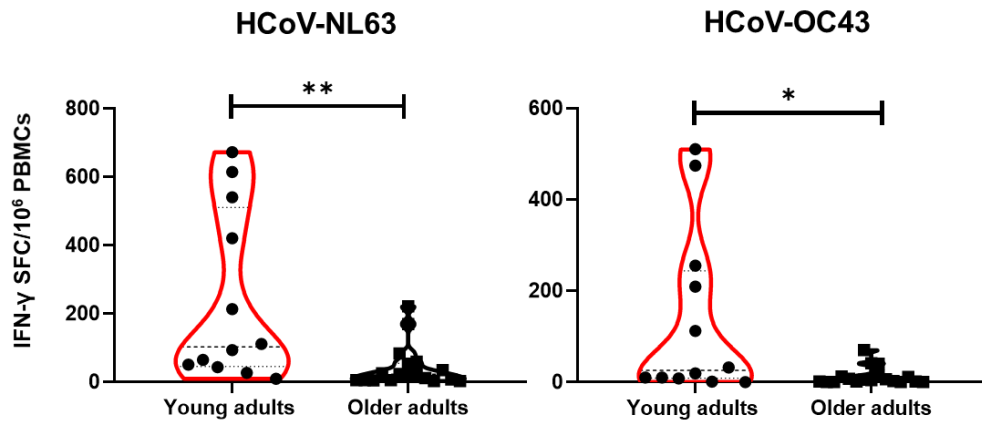
|     |                   |     |                      |
|-----|-------------------|-----|----------------------|
| 126 | TVGSGYQVQVQVQVQVQ | 206 | KAPVQVQVQVQVQVQVQ    |
| 127 | CVQVQVQVQVQVQVQVQ | 207 | LNTSPVQVQVQVQVQVQVQ  |
| 128 | NRRSQAIVTGYRFTNF  | 208 | NLPDFKEQVQVQVQVQVQ   |
| 129 | ATTGYRFTNFVQVQVQ  | 209 | EELDQVQVQVQVQVQVQVQ  |
| 130 | RFTNFVQVQVQVQVQVQ | 210 | FNQVQVQVQVQVQVQVQVQ  |
| 131 | PFTNFVQVQVQVQVQVQ | 211 | VPDLQVQVQVQVQVQVQVQ  |
| 132 | VQVQVQVQVQVQVQVQ  | 212 | LYVQVQVQVQVQVQVQVQ   |
| 133 | VQVQVQVQVQVQVQVQ  | 213 | FLDQVQVQVQVQVQVQVQ   |
| 134 | EQVQVQVQVQVQVQVQ  | 214 | VENRQVQVQVQVQVQVQVQ  |
| 135 | EFTQVQVQVQVQVQVQ  | 215 | LDQVQVQVQVQVQVQVQVQ  |
| 136 | MEQVQVQVQVQVQVQ   | 216 | VQVQVQVQVQVQVQVQVQVQ |
| 137 | TSPQVQVQVQVQVQVQ  | 217 | INLQVQVQVQVQVQVQVQVQ |
| 138 | ELQVQVQVQVQVQVQVQ | 218 | VQVQVQVQVQVQVQVQVQVQ |
| 139 | ELQVQVQVQVQVQVQVQ | 219 | VQVQVQVQVQVQVQVQVQVQ |
| 140 | AKQVQVQVQVQVQVQVQ | 220 | VQVQVQVQVQVQVQVQVQVQ |
| 141 | VQVQVQVQVQVQVQVQ  | 221 | VQVQVQVQVQVQVQVQVQVQ |
| 142 | FCQVQVQVQVQVQVQ   | 222 | VQVQVQVQVQVQVQVQVQVQ |
| 143 | ALVQVQVQVQVQVQVQ  | 223 | ICQVQVQVQVQVQVQVQVQ  |
| 144 | NELQVQVQVQVQVQVQ  | 224 | CGTQVQVQVQVQVQVQVQVQ |
| 145 | TLQVQVQVQVQVQVQVQ | 225 | KQVQVQVQVQVQVQVQVQVQ |
| 146 | NSLQVQVQVQVQVQVQ  | 226 | QVQVQVQVQVQVQVQVQVQ  |
| 147 | VQVQVQVQVQVQVQVQ  |     |                      |
| 148 | LQVQVQVQVQVQVQVQ  |     |                      |
| 149 | VQVQVQVQVQVQVQVQ  |     |                      |
| 150 | NQVQVQVQVQVQVQVQ  |     |                      |
| 151 | CGQVQVQVQVQVQVQ   |     |                      |
| 152 | QVQVQVQVQVQVQVQ   |     |                      |
| 153 | RAQVQVQVQVQVQVQVQ |     |                      |
| 154 | LFQVQVQVQVQVQVQVQ |     |                      |
| 155 | ALQVQVQVQVQVQVQVQ |     |                      |
| 156 | ACTQVQVQVQVQVQVQ  |     |                      |
| 157 | ERQVQVQVQVQVQVQVQ |     |                      |
| 158 | CVQVQVQVQVQVQVQVQ |     |                      |
| 159 | GHVQVQVQVQVQVQVQ  |     |                      |
| 160 | PKVQVQVQVQVQVQVQ  |     |                      |
| 161 | SHVQVQVQVQVQVQVQ  |     |                      |
| 162 | NOVQVQVQVQVQVQVQ  |     |                      |
| 163 | LAQVQVQVQVQVQVQVQ |     |                      |
| 164 | ASLQVQVQVQVQVQVQ  |     |                      |
| 165 | VTAAQVQVQVQVQVQVQ |     |                      |

**Supplementary Table S4: Peptides libraries from SARS-CoV-2 Spike protein**

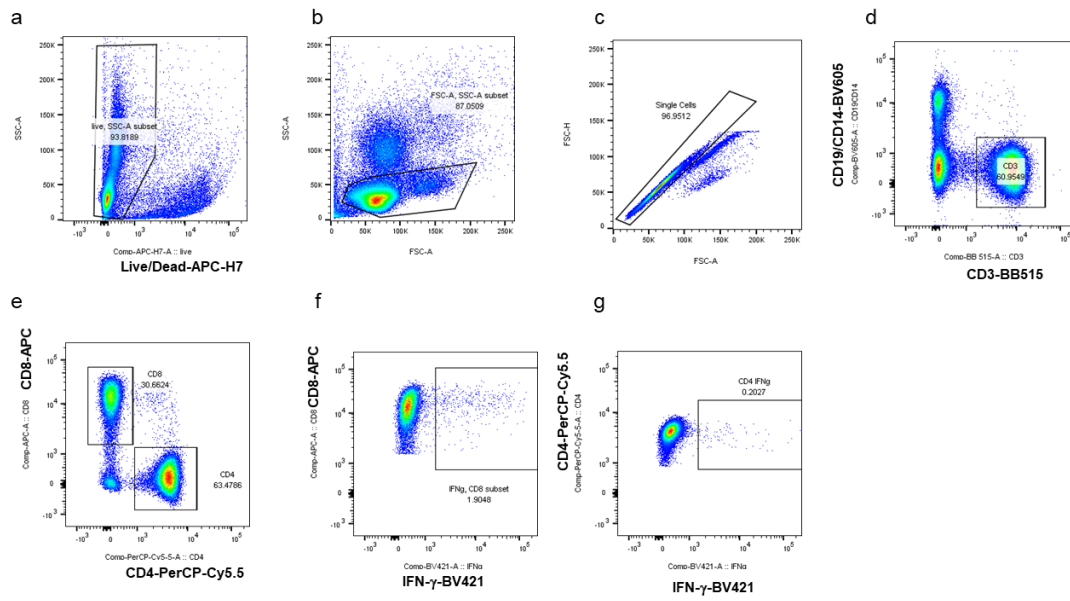
| SARS-CoV-2 Pool 1 (S1) |                  |    |                 |     |                  |     |                  |
|------------------------|------------------|----|-----------------|-----|------------------|-----|------------------|
| 1                      | MVFLVLLPLVSSOC   | 41 | SSANNCTFEYVSQPF | 81  | QPTESIVRFPNITNL  | 121 | NGVEGFNCYFPLQSY  |
| 2                      | LVLPLVSSQCVNLT   | 42 | NCTFEYVSQPLMDL  | 82  | SIVRFPNITNLCPF   | 122 | GFNCYFPLQSYGFQP  |
| 3                      | PLVSSQCVNLTTRTQ  | 43 | EYVSQPLMDLEGKQ  | 83  | FPNITNLCPFGEVFN  | 123 | YFPLQSYGFQPTNGV  |
| 4                      | SQCYNLTRTQLPPA   | 44 | QPFLMDLEGKGNFK  | 84  | TNLCPFGEVFNATRF  | 124 | QSYGFQPTNGVGYQP  |
| 5                      | NLTRTQLPPAYTNS   | 45 | MDLEGKGNFKNLE   | 85  | PFGEVFNATRFASVY  | 125 | FQPTNGVGYQPYRVV  |
| 6                      | RTQLPPAYTNSFTRG  | 46 | GKGNFKNLEFVFK   | 86  | VFNATRFASVYAWNR  | 126 | NGVGYQPYRVVLSF   |
| 7                      | PPAYTNSFTRGVYYP  | 47 | NFNKLEFVFNKIDG  | 87  | TRFASVYAWNRKRIS  | 127 | YQPYRVVLSFELLH   |
| 8                      | TNSFTRGVYYPDKVF  | 48 | LREFVFNKIDGYFKI | 88  | SVYAWNRKRISNCVA  | 128 | RVVLSFELLHAPAT   |
| 9                      | TRGVYYPDKVFRSSV  | 49 | VFNKIDGYFKIYSKH | 89  | WNRKRISNCVADYSV  | 129 | LSFELLHAPATVCGP  |
| 10                     | YYPDKVFRSSVLHST  | 50 | IDGYFKIYSKHTPIN | 90  | RISNCVADYSVLYNS  | 130 | LLHAPATVCGPKKST  |
| 11                     | KVFRSSVLHSTQDLF  | 51 | FKIYSKHTPINLVRD | 91  | CVADYSVLYNSASF   | 131 | PATVCGPKKSTNLVK  |
| 12                     | SSVLHSTQDLFLPFF  | 52 | SKHTPINLVRDLPQG | 92  | YSVLYNSASFSTFKC  | 132 | CGPKKSTNLVKNCV   |
| 13                     | HSTQDLFLPFFSNVT  | 53 | PINLVRDLPQGFSA  | 93  | YNSASFSTFKCYGVS  | 133 | KSTNLVKNCVNFNF   |
| 14                     | DLFLPFFSNVTFHFA  | 54 | VRDLPQGFSALEPLV | 94  | SFSTFKCYGVSPTKL  | 134 | LVKNCVNFNFNGLT   |
| 15                     | PFFSNVTFHFAIHVS  | 55 | PQGFSALEPLVDLPI | 95  | FKCYGVSPTKLNDLC  | 135 | KCVNFNFNGLTGTG   |
| 16                     | NVTFHFAIHVSGTNG  | 56 | SALEPLVDLPIGINI | 96  | GVSPTKLNDLCFTNV  | 136 | FNFNGLTGTGVLTES  |
| 17                     | FHAIHVSGTNGTKRF  | 57 | PLVDLPIGINTRFQ  | 97  | TKLNDLCFTNVYADS  | 137 | GLTGTGVLTESNKKF  |
| 18                     | HVSGTNGTKRFDNVP  | 58 | LPIGINTRFQTLA   | 98  | DLCFTNVYADSFVIR  | 138 | TGVLTESNKKFLPFP  |
| 19                     | TNGTKRFDNVPVLPFN | 59 | INTRFQTLALHRS   | 99  | TNVYADSFVIRGDEV  | 139 | TESNKKFLPFPQFGR  |
| 20                     | KRFDNVPVLPFNDGVY | 60 | RFQTLALHRSYLT   | 100 | ADSFVIRGDEVIRQIA | 140 | KKFLPFPQFGRDIAD  |
| 21                     | NVPLPFDNDGVYFAST | 61 | LLALHRSYLTGDS   | 101 | VIRGDEVIRQIAPGQT | 141 | PFQFGRDIADTTDA   |
| 22                     | PFNDGVYFASTEKSN  | 62 | HRSYLTGDSSSGWT  | 102 | DEVIRQIAPGQTGKIA | 142 | FGRDIADTTDAVRDP  |
| 23                     | GVYFASTEKSNIRG   | 63 | LTPGDSSSGWTAGAA | 103 | QIAPGQTGKIADYNY  | 143 | IADTTDAVRDPQTL   |
| 24                     | ASTEKSNIRGWIFG   | 64 | DSSSGWTAGAAAYV  | 104 | GQTGKIADYNYKLPD  | 144 | TDVAVRDPQTLLEID  |
| 25                     | KSNIRGWIFGTLD    | 65 | GWTAGAAAYVGYLQ  | 105 | KIADYNYKLPDDFTG  | 145 | RDPQTLLEIDITPCS  |
| 26                     | IRGWIFGTLDLSDKTQ | 66 | GAAAYVGYLQPRTF  | 106 | YNYKLPDDFTGCVIA  | 146 | TLEIDITPCSFGV    |
| 27                     | IFGTLDLSDKTQSLI  | 67 | YVGYLQPRTFLLKY  | 107 | LPDDFTGCVIAWNSN  | 147 | LDITPCSFGVSVIT   |
| 28                     | LDLSDKTQSLIVNNA  | 68 | YLPRTFLLKYENENG | 108 | FTGCVIAWNSNLD    | 148 | PCSFVSVITPPTGN   |
| 29                     | KTQSLIVNNAIVN    | 69 | RTFLLKYENENGTID | 109 | VIAWNSNLDKSVGG   | 149 | GGVSVITPPTGNVSNQ |
| 30                     | LLIVNNAIVNVIKVC  | 70 | LKYENENGTIDAVDC | 110 | NSNLDKSVGGNYNY   | 150 | VITPPTGNVSNQAVL  |
| 31                     | NNAIVNVIKVCDFQ   | 71 | ENGTIDAVDCALDP  | 111 | LDSKVGNYNYLYRL   | 151 | GTNTSNQAVLYQDV   |
| 32                     | NVIKVCDFQFCNDP   | 72 | ITDAVDCALDPLSET | 112 | VGGNYNYLYRFRKS   | 152 | SNQAVLYQDVNCTE   |
| 33                     | KVCFQFCNDPFLGV   | 73 | VDCALDPLSETKCTL | 113 | YNYLYRFRKSNLKP   | 153 | AVLYQDVNCTEVPVA  |
| 34                     | FQFCNDPFLGVYHK   | 74 | LDPLSETKCTLKSFT | 114 | YRFRKSNLKPFERD   | 154 | QDVNCTEVPVAIHAD  |
| 35                     | NDPFLGVYHKNNKS   | 75 | SETKCTLKSFTVEKG | 115 | RKSNLKPFERDISTE  | 155 | CTEVPVAIHADQLTP  |
| 36                     | LGVYHKNNKSWMES   | 76 | CTLKSFTVEKGIYQT | 116 | LKPFERDISTEIQQA  | 156 | PVAIHADQLTPVTRV  |
| 37                     | YHKNNKSWMESEFRV  | 77 | SFTVEKGIYQTSNFR | 117 | ERDISTEIQAGSTP   | 157 | HADQLTPVTRVYVSTG |
| 38                     | NKSWMESEFRVYSSA  | 78 | EKGIYQTSNFRVQPT | 118 | STEIQAGSTPCNGV   | 158 | LTPVTRVYVSTGNSVF |
| 39                     | MESEFRVYSSANNCT  | 79 | YQTSNFRVQPTESIV | 119 | YQAGSTPCNGVEGFN  |     |                  |
| 40                     | FRVYSSANNCTFEYV  | 80 | NFRVQPTESIVRFPN | 120 | STPCNGVEGFNCYFP  |     |                  |

| SARS-CoV-2 Pool 2 (S2) |                  |     |                 |     |                  |     |                 |
|------------------------|------------------|-----|-----------------|-----|------------------|-----|-----------------|
| 159                    | WRVYSTGSNVFQTRA  | 199 | PIKDFGGFNFSQILP | 239 | NQNAQALNTLVKQLS  | 279 | QIITDNTFVSGNCD  |
| 160                    | STGSNVFQTRAGCLI  | 200 | FGFNFQILPDPSPK  | 240 | QALNTLVKQLSNFNG  | 280 | TDNTFVSGNCDVIG  |
| 161                    | NVQTRAGCLIAEAEH  | 201 | NFSQILPDPSPKSKR | 241 | TLVKQLSNFNGAIS   | 281 | FVSGNCDVIGVGN   |
| 162                    | TRAGCLIAEAEHVNNS | 202 | ILPDPSPKSKRSFIE | 242 | QLSNFNGAISVLND   | 282 | NCDVIGVGNVTVYD  |
| 163                    | CLIAEAEHVNNSYECD | 203 | PSKPSKRSFIEDLLF | 243 | NFGAISVLNDILSR   | 283 | VIGVGNVTVYDPLP  |
| 164                    | AEHVNNSYECDIPIG  | 204 | SKRSFIEDLLFNKVT | 244 | ISSVLNDILSRDKV   | 284 | VNNTVYDPLPELDS  |
| 165                    | NNSYECDIPIGAGIC  | 205 | FIEDLLFNKVTADAG | 245 | LNILSRDKVAEAEV   | 285 | VYDPLPELDSFKEE  |
| 166                    | ECDIPIGAGICASYQ  | 206 | LLFNKVTADAGFIK  | 246 | LSRLDKVAEAEVQIDR | 286 | LOPELDSFKEEKDY  |
| 167                    | PIGAGICASYQTQTN  | 207 | KVTADAGFIKQYGD  | 247 | DKVAEAEVQIDRLITG | 287 | LDSFKEEKDYKFNH  |
| 168                    | GICASYQTQTNPRR   | 208 | ADAGFIKQYGDCLGD | 248 | AEVQIDRLITGRLOS  | 288 | KEELDYKFNHSPD   |
| 169                    | SYQTQTNPRRARSV   | 209 | FIKQYGDCLGDIAR  | 249 | IDRLITGRLOSQTY   | 289 | DKYKFNHSPDVLG   |
| 170                    | QTNPRRARSVASQS   | 210 | YGDCLGDIARLDIC  | 250 | ITGRLOSQTYVTOQ   | 290 | KNHSPDVLGDISG   |
| 171                    | PRRARSVASQSIIAY  | 211 | GDIARLDICAQKF   | 251 | LQSLQTYVTOQLIRA  | 291 | SPDVLGDISGINAS  |
| 172                    | RSVASQSIIAYTMSL  | 212 | AARDLICAQKFNGLT | 252 | QTYVTOQLIRAAEIR  | 292 | DLGDISGINASVNI  |
| 173                    | SQSIIAYTMSLGAEN  | 213 | LCAQKFNGLTVLPP  | 253 | TOQLIRAAEIRASAN  | 293 | ISGINASVNIQKEI  |
| 174                    | IAYTMSLGAENSVAY  | 214 | KQKFNGLTVLPLTDD | 254 | IRAAEIRASANLAAT  | 294 | NASVNIQKEIDRLN  |
| 175                    | MSLGAENSVAYSNN   | 215 | GLTVLPLTDEMIA   | 255 | EIRASANLAATKME   | 295 | VNIQKEIDRLNEVAK |
| 176                    | AENSVAYSNSIAIAP  | 216 | LPPLTDEMIAQYTS  | 256 | SANLAATKMECVL    | 296 | KEIDRLNEVAKNLNE |
| 177                    | VAYSNSIAIAPNFT   | 217 | LDEMIAQYTSALLA  | 257 | AATKMECVLQGSKR   | 297 | RNEVAKNLNESLID  |
| 178                    | NNSIAIAPNFTISVT  | 218 | MAQYTSALLAGTIT  | 258 | MSECVLQGSKRDFC   | 298 | VAKNLNESLIDLOEL |
| 179                    | AIPNFTISVTTEIL   | 219 | YTSALLAGTITSGWT | 259 | VLQGSKRDFCGKY    | 299 | LNESLIDLOELQYK  |
| 180                    | NFTISVTTEILPVM   | 220 | LLAGTITSGWTFGAG | 260 | SKRVDFCGKYHLMS   | 300 | LIDLOELQYKQYK   |
| 181                    | SVTEILPVMSTKTS   | 221 | TITSGWTFGAGALQ  | 261 | DFCGKYHLMSFPQS   | 301 | QELQYKQYKQYK    |
| 182                    | EILPVMSTKTSVDCT  | 222 | WTFGAGALQIPFA   | 262 | KGYHLMSFPQSAPHG  | 302 | KYEQYKQYKQYK    |
| 183                    | VSMSTKTSVDCTMYIC | 223 | GAGALQIPFAMQMA  | 263 | LMSFPQSAPHGVFL   | 303 | YKQYKQYKQYK     |
| 184                    | KTSVDCTMYICGDS   | 224 | ALQIPFAMQMAYRFN | 264 | QGSAPHGVFLHVTY   | 304 | PWYKQYKQYKQYK   |
| 185                    | DCTMYICGDSSTEC   | 225 | PFAMQMAYRFNGIV  | 265 | PHGVFLHVTYVPAQ   | 305 | WLGFIAGLIAVMT   |
| 186                    | YICGDSSTECNLLQ   | 226 | QMAYRFNGIVTQNV  | 266 | VFLHVTYVPAQEKNF  | 306 | IAGLIAVMTIMLC   |
| 187                    | DSTECNLLQYGSF    | 227 | RFNGIVTQNVLYEN  | 267 | VTVPAQEKNFITAP   | 307 | IAVMTIMLCMTS    |
| 188                    | CSNLLQYGSFCTQL   | 228 | IGVTQNVLYENQKLI | 268 | PAQEKNFITAPAI    | 308 | MTIMLCMTSCSSC   |
| 189                    | LLQYGSFCTQLNRAL  | 229 | QNVLYENQKLIANQF | 269 | KNFITAPAI        | 309 | MLCMTSCSSCKL    |
| 190                    | GSFCTQLNRALTGIA  | 230 | YENQKLIANQFNSAI | 270 | TAPAI            | 310 | MTSCSSCKLCCSG   |
| 191                    | RALNRALTGIAVEQD  | 231 | KLIANQFNSAIQKIQ | 271 | ICHGKAHFPREGV    | 311 | CSCCKLCCSGSC    |
| 192                    | RLTGIAVEQDKNTQ   | 232 | NQFNSAIQKIQDLS  | 272 | GKAHFPREGVFSNG   | 312 | KGCCSCCKCFDE    |
| 193                    | GIAVEQDKNTQEVFA  | 233 | SAIQKIQDLSSTAS  | 273 | FPREGVFSNGTHWF   | 313 | CCGCKCFDEDDSEP  |
| 194                    | EQDKNTQEVFAQVQK  | 234 | KIQDLSSTASALGK  | 274 | GVFSNGTHWFVTQR   | 314 | CCKCFDEDDSEPLK  |
| 195                    | NQEVFAQVQKQYK    | 235 | SLSSTASALGKLDV  | 275 | NGTHWFVTQRNFYE   | 315 | DEDDSEPLKQKLYH  |
| 196                    | VFAQVQKQYKTPPIK  | 236 | TASALGKLDVNVQ   | 276 | HWFTQRNFYEPQII   |     |                 |
| 197                    | VKQYKQYKTPPIKDFG | 237 | LKLDVNVQNAQAL   | 277 | TQRNFYEPQIITDN   |     |                 |
| 198                    | YKTPPIKDFGGFNFS  | 238 | QDVNVQNAQALNTLV | 278 | FYEPQIITDNFV     |     |                 |



**Supplementary Figure S1. HCoV-NL63 and HCoV-OC43-specific immunity in young and older male adults.** HCoV-specific T cells response measured as frequency of IFN- $\gamma$  spot forming cells (SFU) after stimulation of PBMCs isolated from young (n=12; red outline) and older adults (n=18; black outline) with inactivated HCoV-NL63 and HCoV-OC43. Data were background-subtracted and expressed as SFU/10<sup>6</sup> PBMCs. Dotted lines represented the median. Each dot and symbol represent a single subject. Statistical comparison between the two groups was performed using non-parametric Mann-Whitney test for unpaired samples (\*\*p<0.005; \*p<0.05).



**Supplementary Figure S2.** Gating strategy for detection of HCoV-s-specific IFN- $\gamma$ <sup>+</sup> T cells upon stimulation of PBMCs. (a) Live cells were identified followed by (b) lymphocytes selection and (c) doublets discrimination. (d) The CD3<sup>+</sup> T cells were divided into (e) CD4<sup>+</sup> and CD8<sup>+</sup> T cells subsets and the percentage of IFN- $\gamma$ <sup>+</sup> cells were calculated for each subset. Data were analyzed and images created using FlowJo software (Version 10.5.3, BD Bioscience)