

|            |                                                          |        | APC binding<br>(nm) | TPP-4885<br>Fab<br>binding<br>(nm) |
|------------|----------------------------------------------------------|--------|---------------------|------------------------------------|
| Nanobodies | Nanobodies<br>Not<br>Blocking<br>TPP-4885<br>Fab Binding | Buffer | 0.06                | 0.03                               |
|            |                                                          | LP2    | 0.29                | 0.15                               |
|            |                                                          | LP3    | 0.29                | 0.16                               |
|            |                                                          | LP7    | 0.34                | 0.08                               |
|            |                                                          | LP9    | 0.33                | 0.18                               |
|            |                                                          | LP10   | 0.29                | 0.15                               |
|            |                                                          | LP11   | 0.30                | 0.16                               |
|            |                                                          | LP12   | 0.31                | 0.18                               |
|            |                                                          | LP18   | 0.29                | 0.16                               |
|            | Nanobodies<br>Blocking<br>TPP-4885<br>Fab Binding        | LP1    | 0.32                | -0.03                              |
|            |                                                          | LP4    | 0.33                | -0.03                              |
|            |                                                          | LP5    | 0.33                | -0.03                              |
|            |                                                          | LP6    | 0.31                | -0.03                              |
|            |                                                          | LP8    | 0.32                | -0.02                              |
|            |                                                          | LP13   | 0.32                | -0.09                              |
|            |                                                          | LP14   | 0.31                | -0.02                              |
|            |                                                          | LP15   | 0.31                | -0.05                              |
|            |                                                          | LP16   | 0.29                | -0.03                              |
|            |                                                          | LP17   | 0.30                | -0.00                              |
|            |                                                          | LP19   | 0.31                | -0.02                              |
|            |                                                          | LP20   | 0.30                | -0.01                              |
|            |                                                          | LP21   | 0.31                | -0.03                              |

Cluster  
1

FIG. 1A

|        |      | Analyte |      |      |      |      |      |      |
|--------|------|---------|------|------|------|------|------|------|
|        |      | LP2     | LP3  | LP10 | LP7  | LP9  | LP12 | LP18 |
| Ligand | LP2  | 0       | 0    | 0    | 0.05 | 0.04 | 0.04 | 0.06 |
|        | LP3  | 0       | 0    | 0    | 0.05 | 0.05 | 0.06 | 0.06 |
|        | LP10 | 0       | 0    | 0    | 0.05 | 0.06 | 0.05 | 0.06 |
|        | LP7  | 0       | 0    | 0.02 | 0    | 0    | 0    | 0    |
|        | LP9  | 0.04    | 0.04 | 0.04 | 0    | 0    | 0    | 0    |
|        | LP12 | 0.05    | 0.05 | 0.05 | 0    | 0    | 0    | 0    |
|        | LP18 | 0.04    | 0.03 | 0.04 | 0    | 0    | 0    | 0.01 |

FIG. 1B

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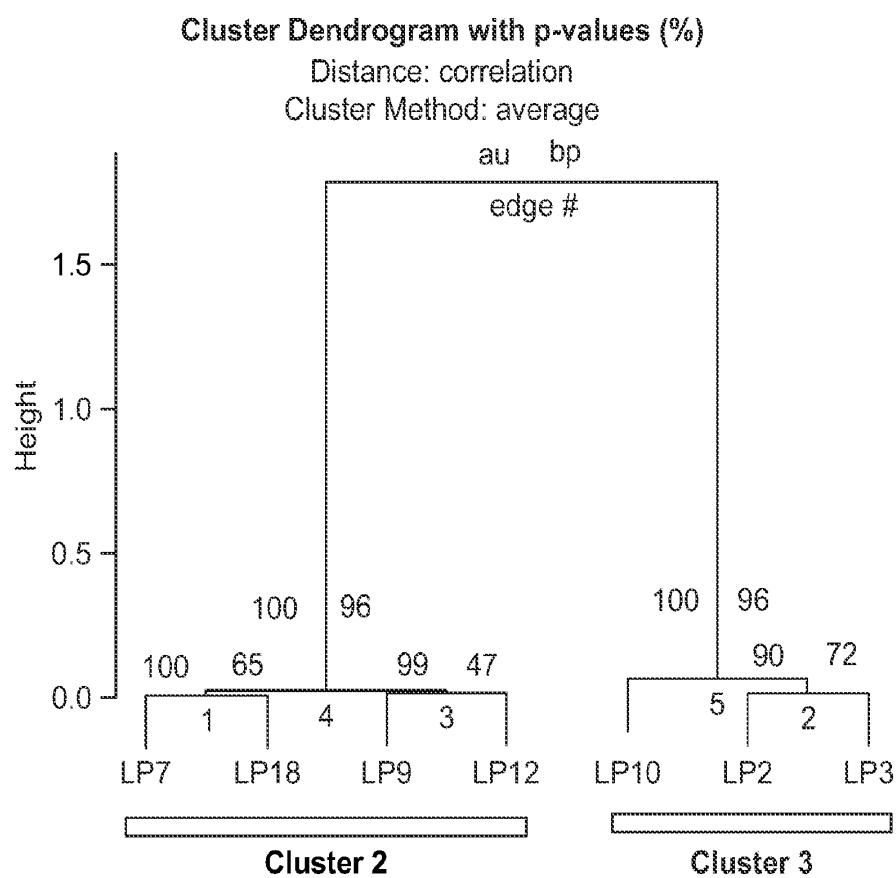


FIG. 1C

|           | # of Nanobodies(%) | Nanobodies                                                    |
|-----------|--------------------|---------------------------------------------------------------|
| Cluster 1 | 13(62%)            | LP1 LP4 LP5 LP6LP8 LP13 LP14<br>LP15 LP16 LP17 LP19 LP20 LP21 |
| Cluster 2 | 5(24%)             | LP7 LP9 LP11 LP12 LP18                                        |
| Cluster 3 | 3(14%)             | LP2 LP3 LP10                                                  |

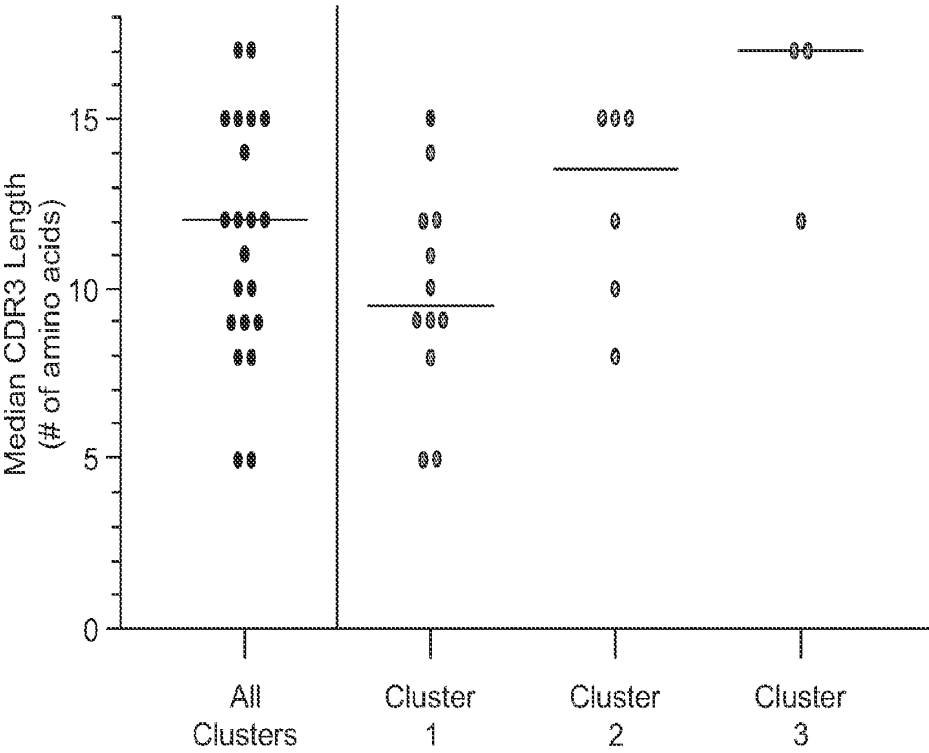


FIG. 1D

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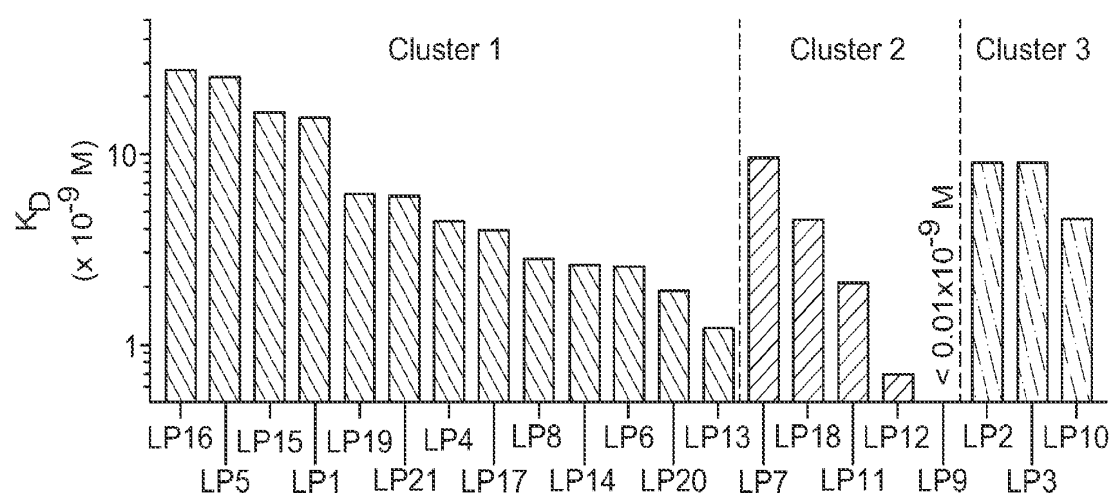


FIG. 1E

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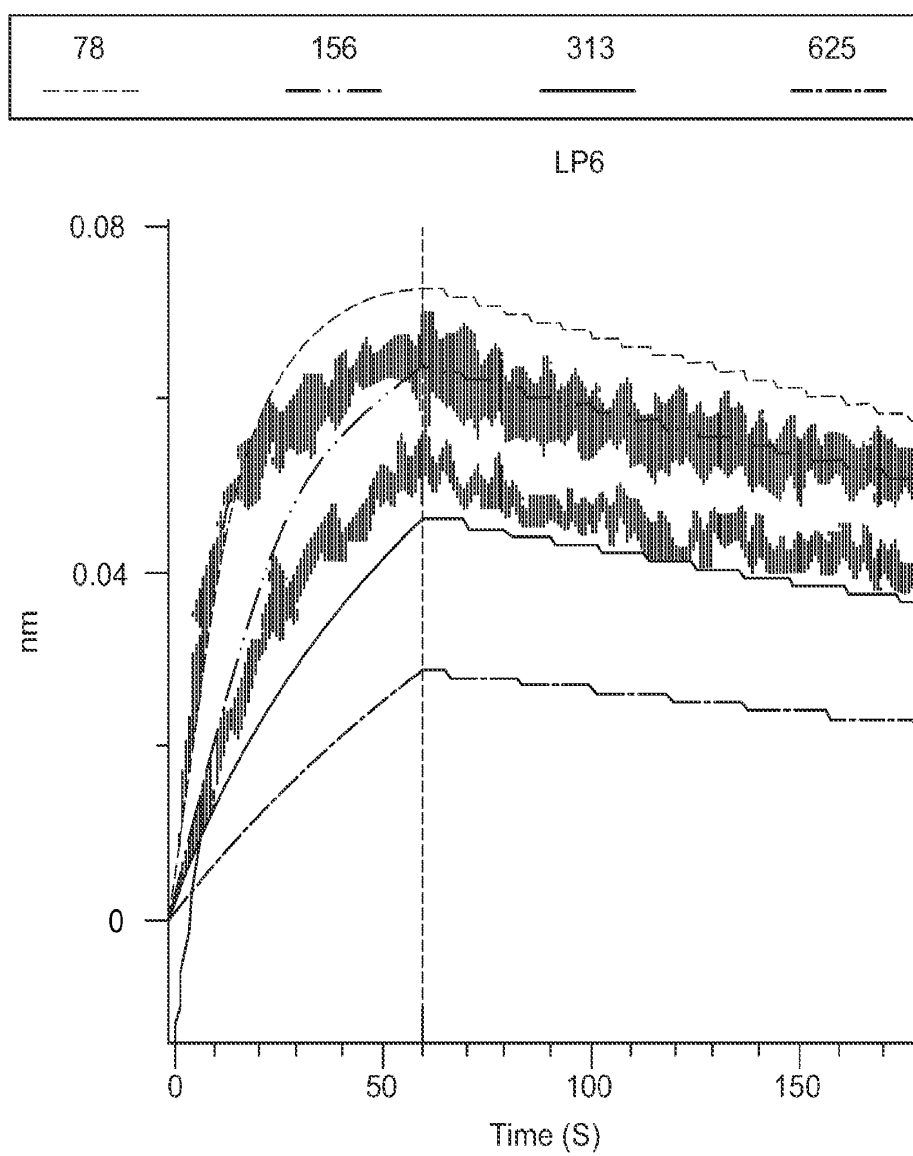


FIG. 1F

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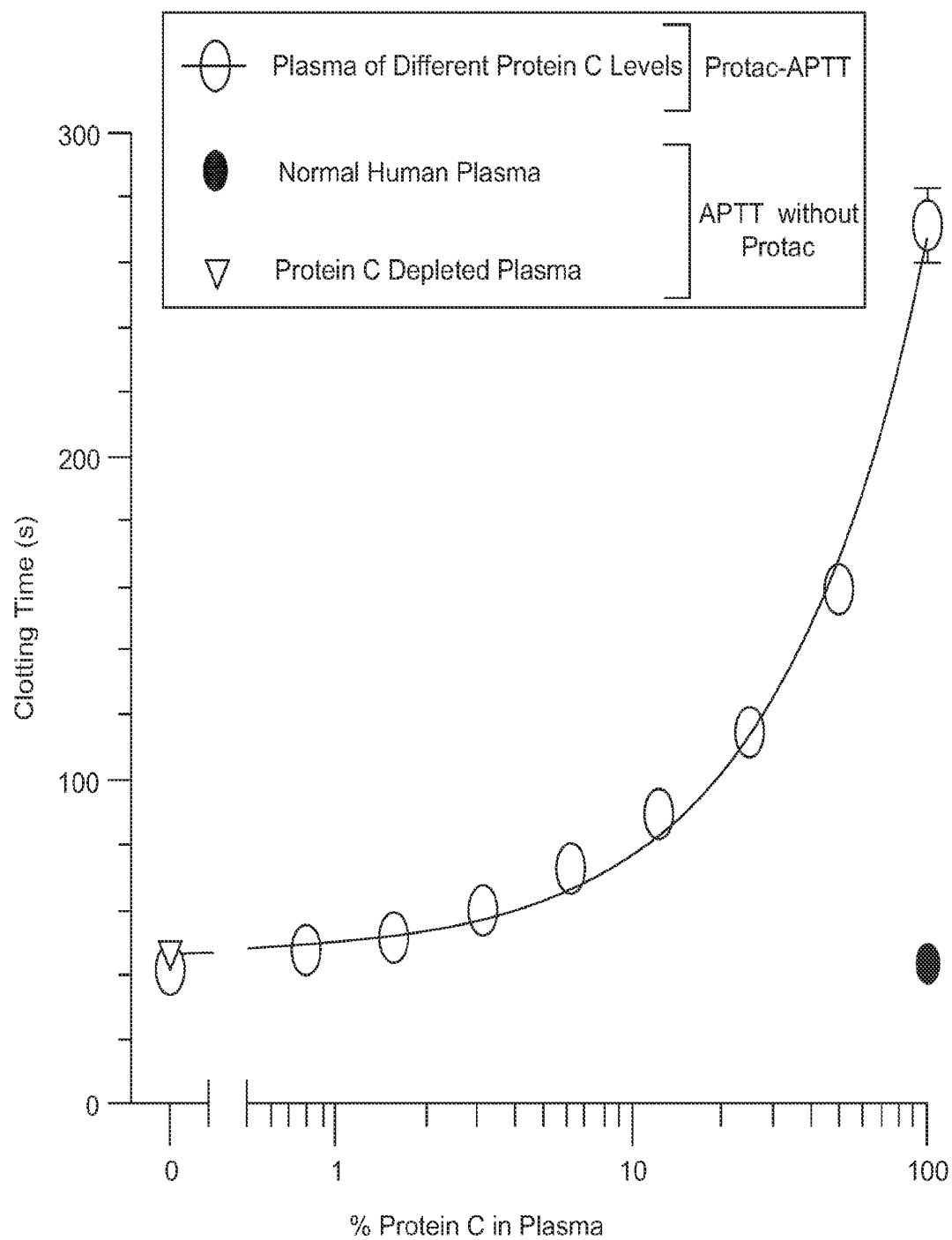


FIG. 2A

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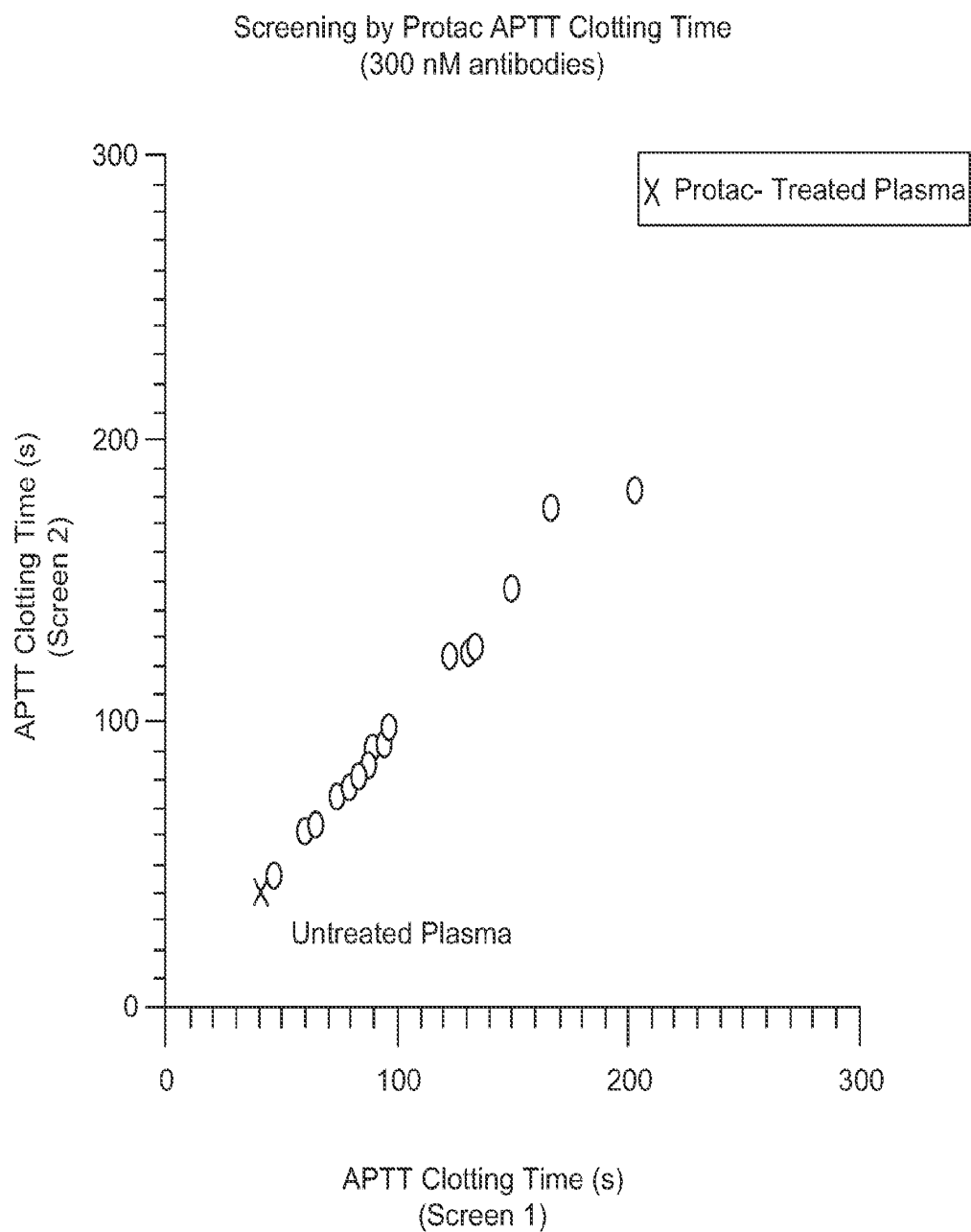


FIG. 2B

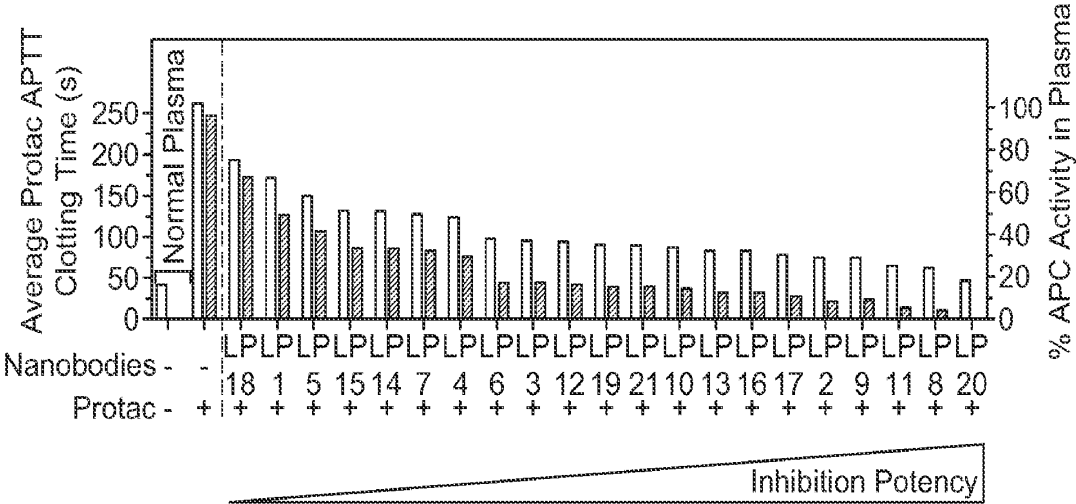


FIG. 2C

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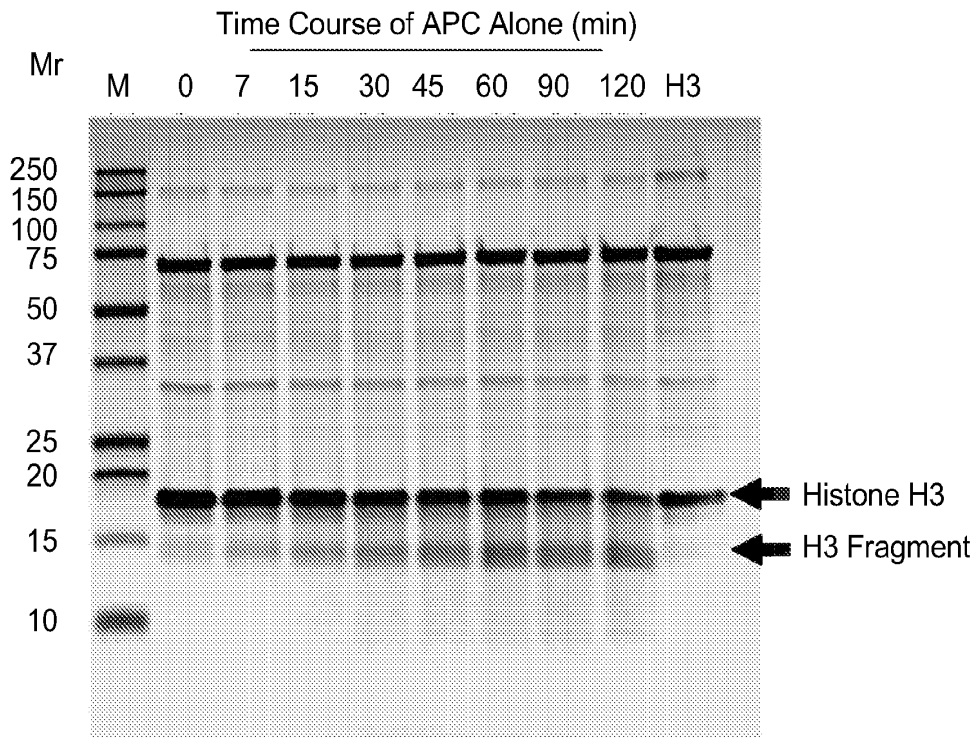


FIG. 3A

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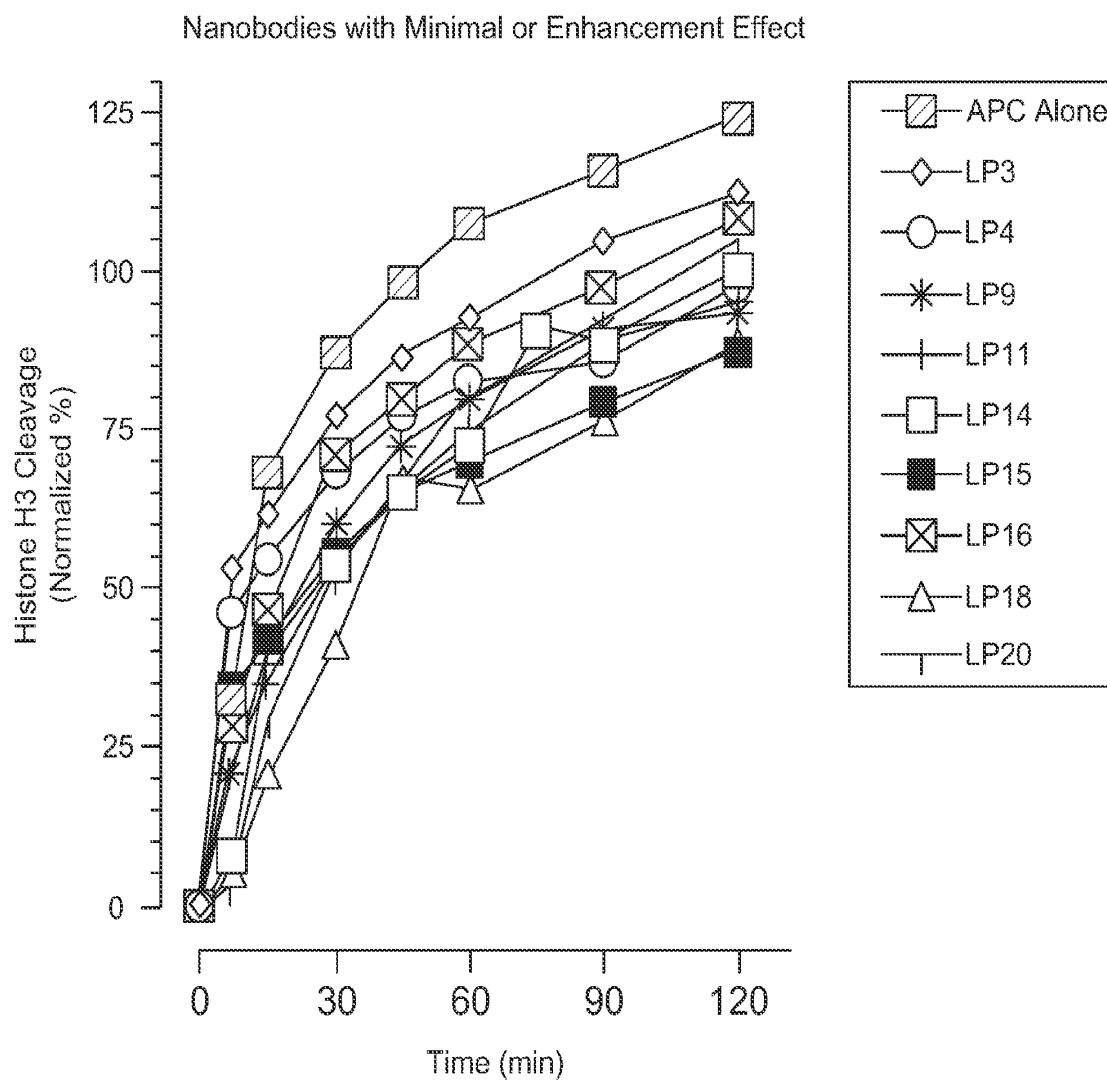


FIG. 3B

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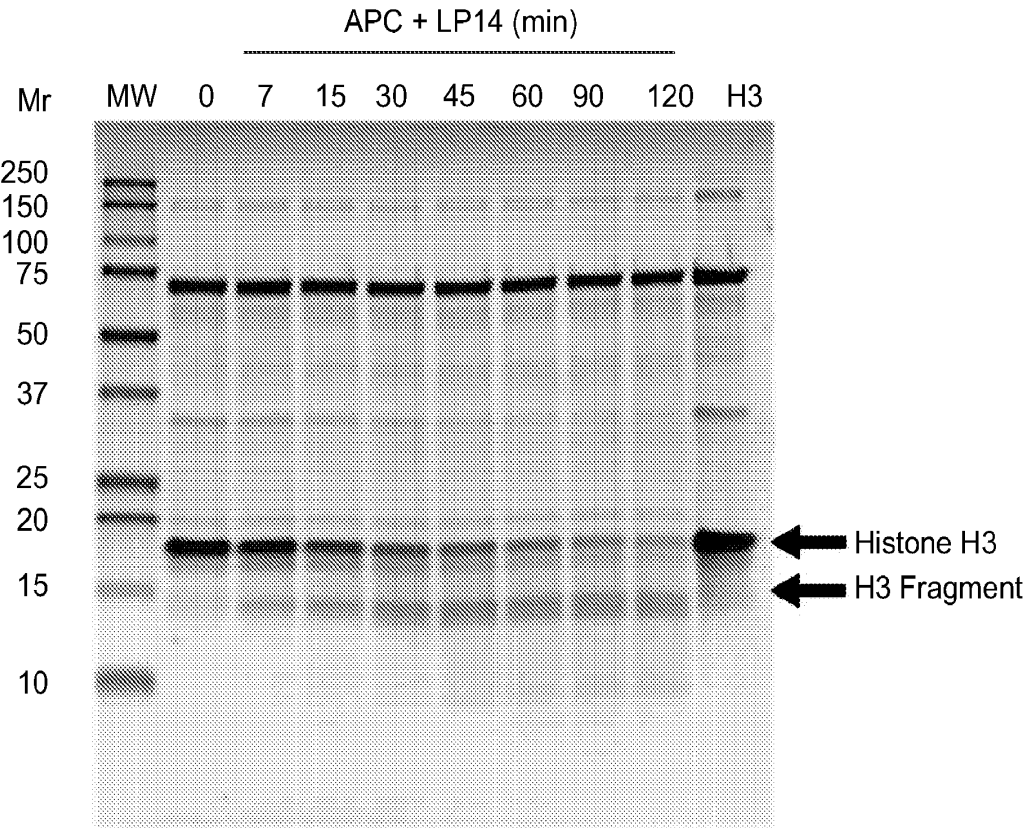


FIG. 3C

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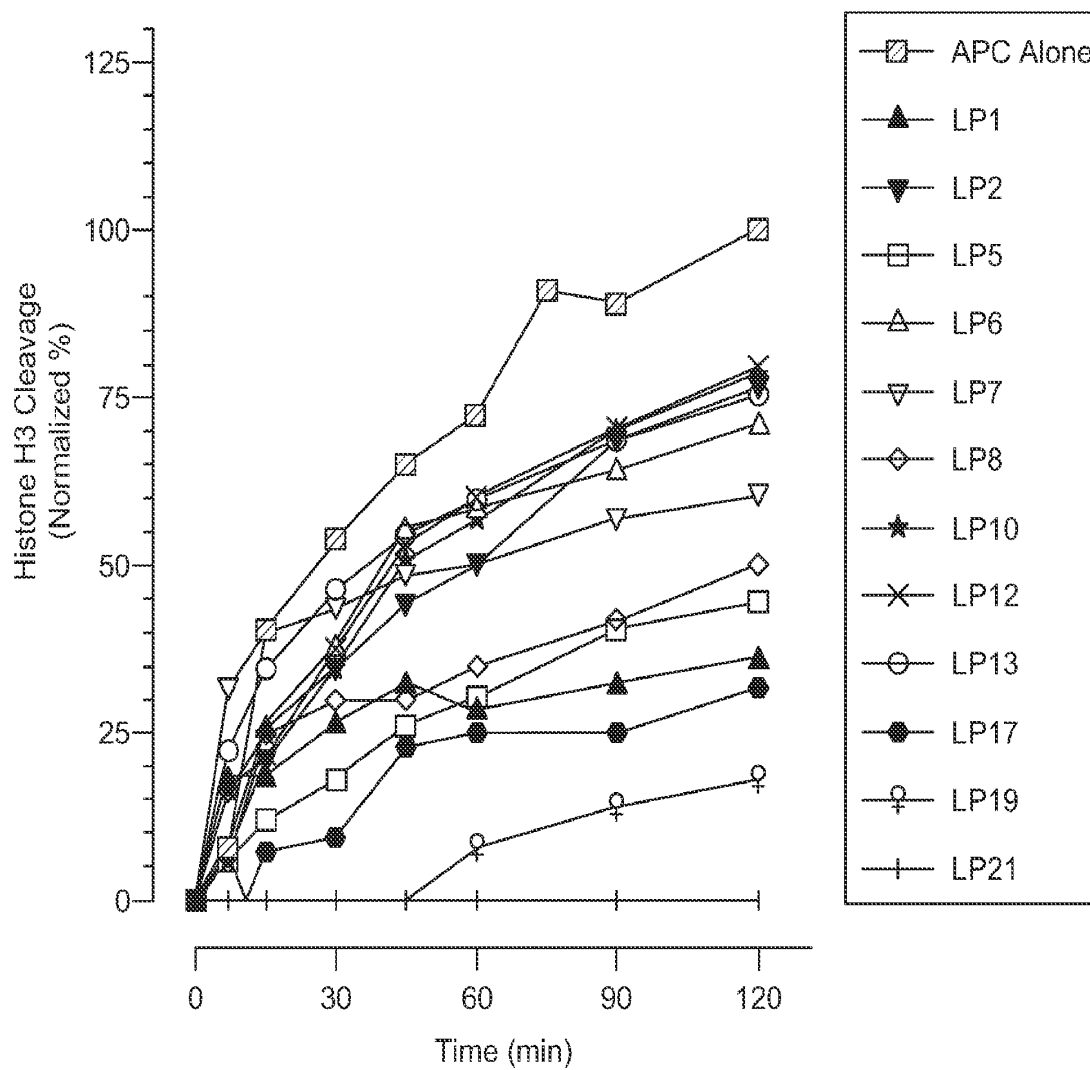


FIG. 3D

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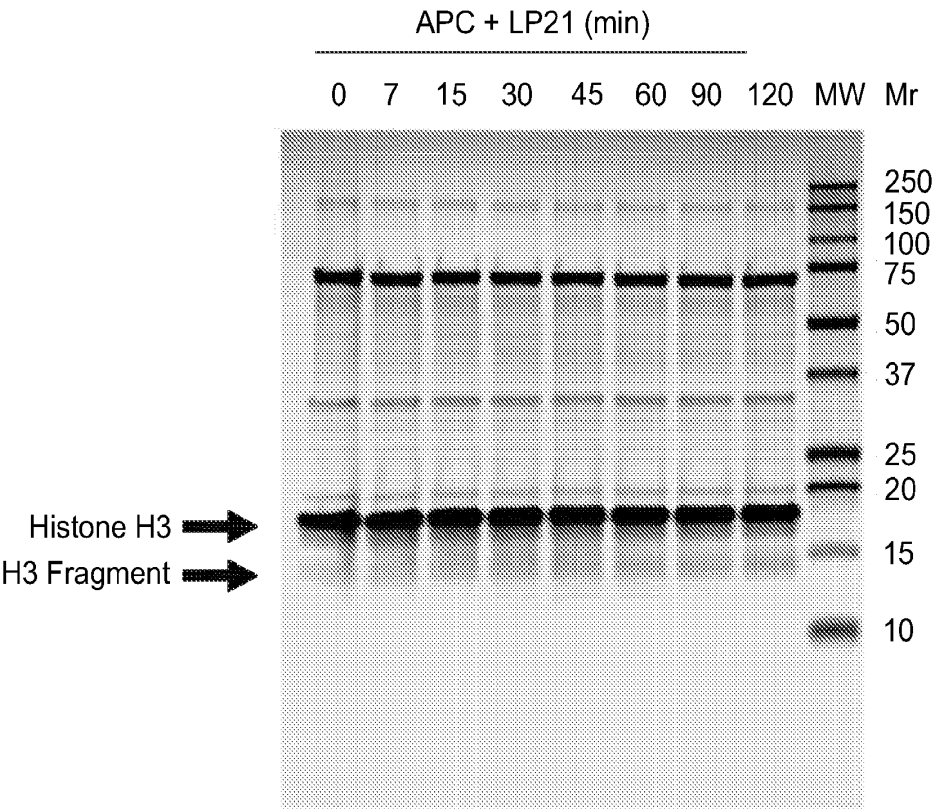


FIG. 3E

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| Nanobody  | % Histone H3 Cleavage<br>Relative to APC Alone |
|-----------|------------------------------------------------|
| LP1       | 35                                             |
| LP2       | 81                                             |
| LP3       | 116                                            |
| LP4       | 102                                            |
| LP5       | 46                                             |
| LP6       | 63                                             |
| LP7       | 77                                             |
| LP8       | 53                                             |
| LP9       | 98                                             |
| LP10      | 84                                             |
| LP11      | 102                                            |
| LP12      | 88                                             |
| LP13      | 81                                             |
| LP14      | 133                                            |
| LP15      | 91                                             |
| LP16      | 116                                            |
| LP17      | 32                                             |
| LP18      | 95                                             |
| LP19      | 18                                             |
| LP20      | 112                                            |
| LP21      | 0                                              |
| APC alone | 100                                            |

FIG. 3F

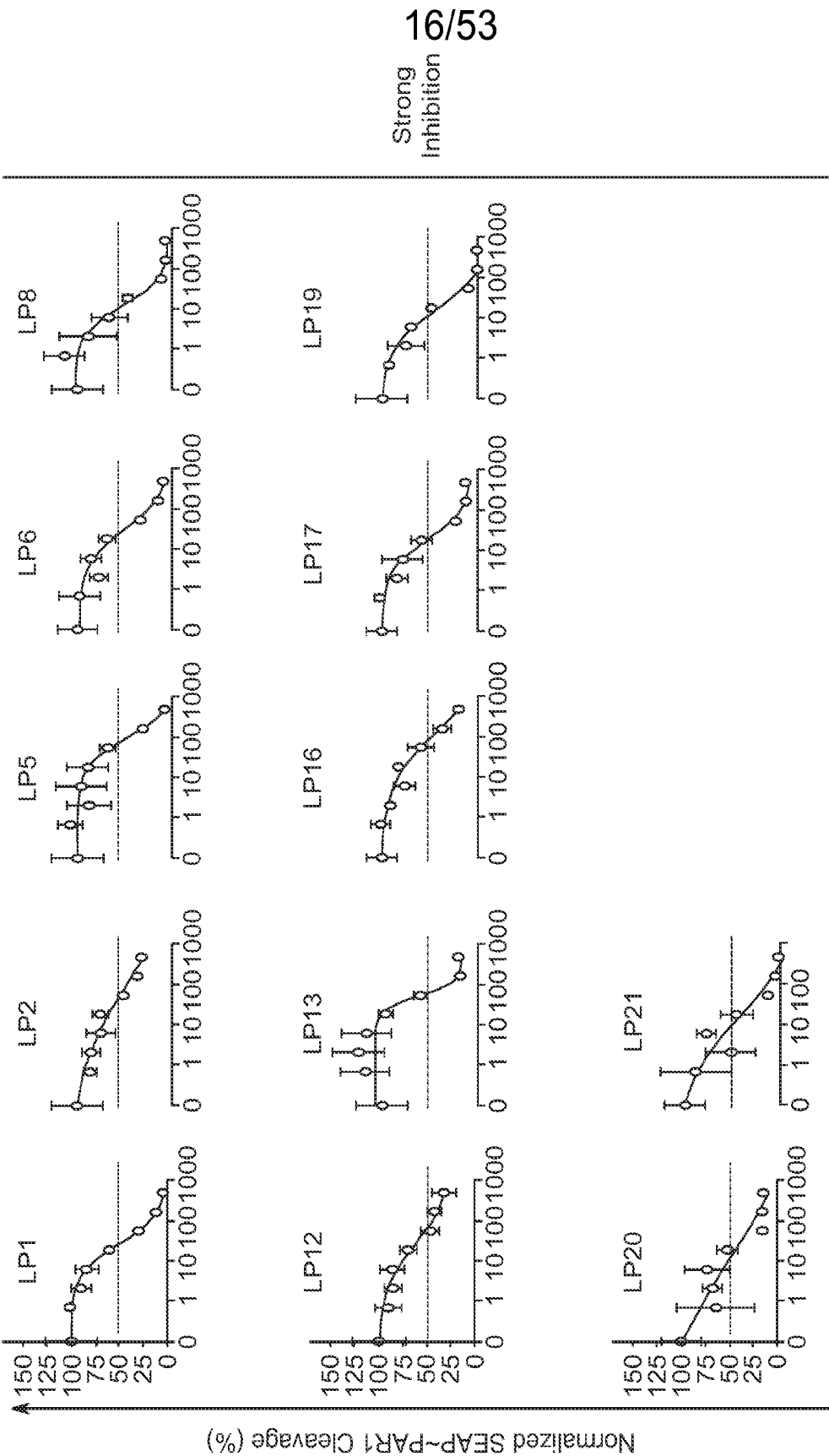


FIG. 4A

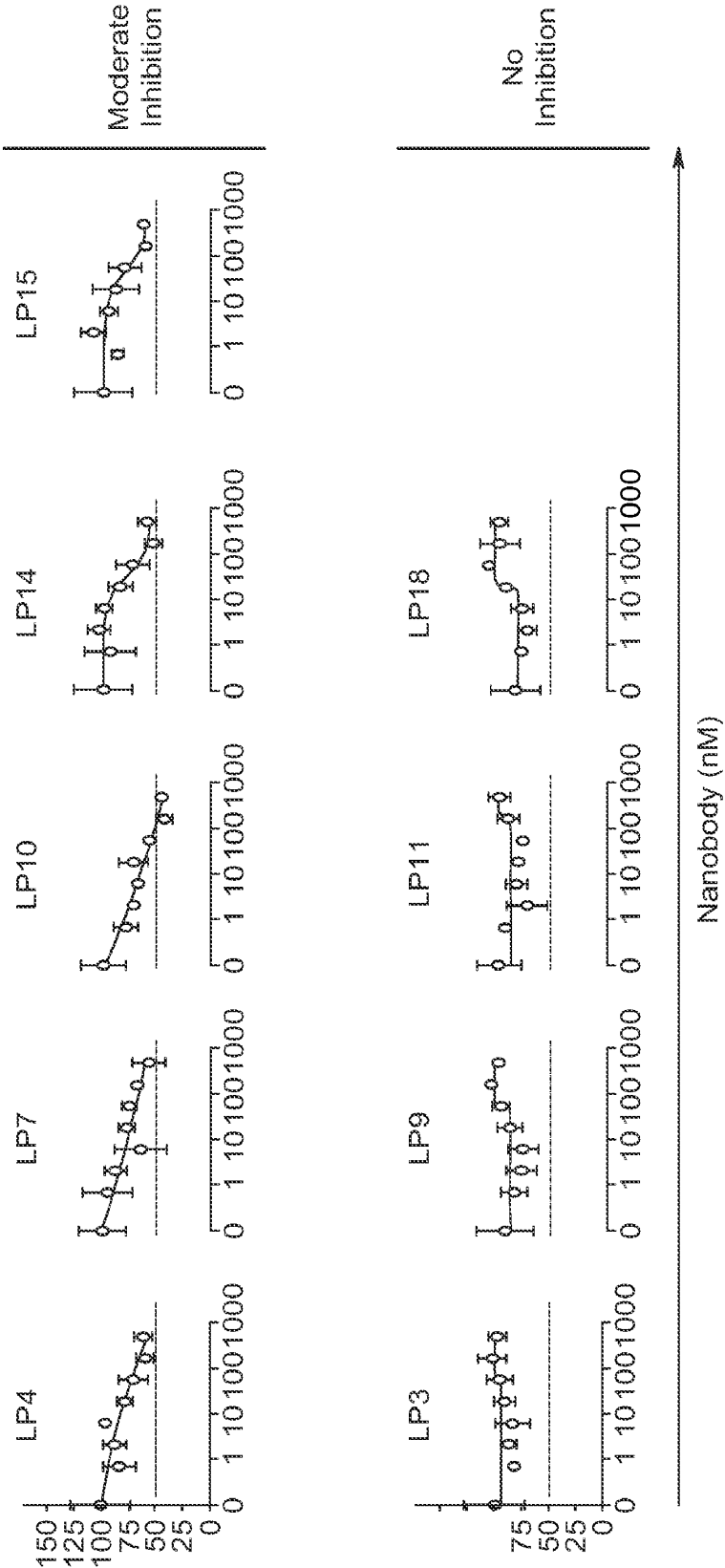


FIG. 4A (Continued)

| Cell Line | PAR1 Sequence |   |         |   |                  | Cleavage Site |
|-----------|---------------|---|---------|---|------------------|---------------|
| WT PAR1   | ...L38 D P    | R | S F L L | R | N P N D K Y52... | R41 and R46   |
| R41Q PAR1 | ...L38 D P    | Q | S F L L | R | N P N D K Y52... | R46           |
| R46Q PAR1 | ...L38 D P    | R | S F L L | O | N P N D K Y52... | R41           |

FIG. 4B

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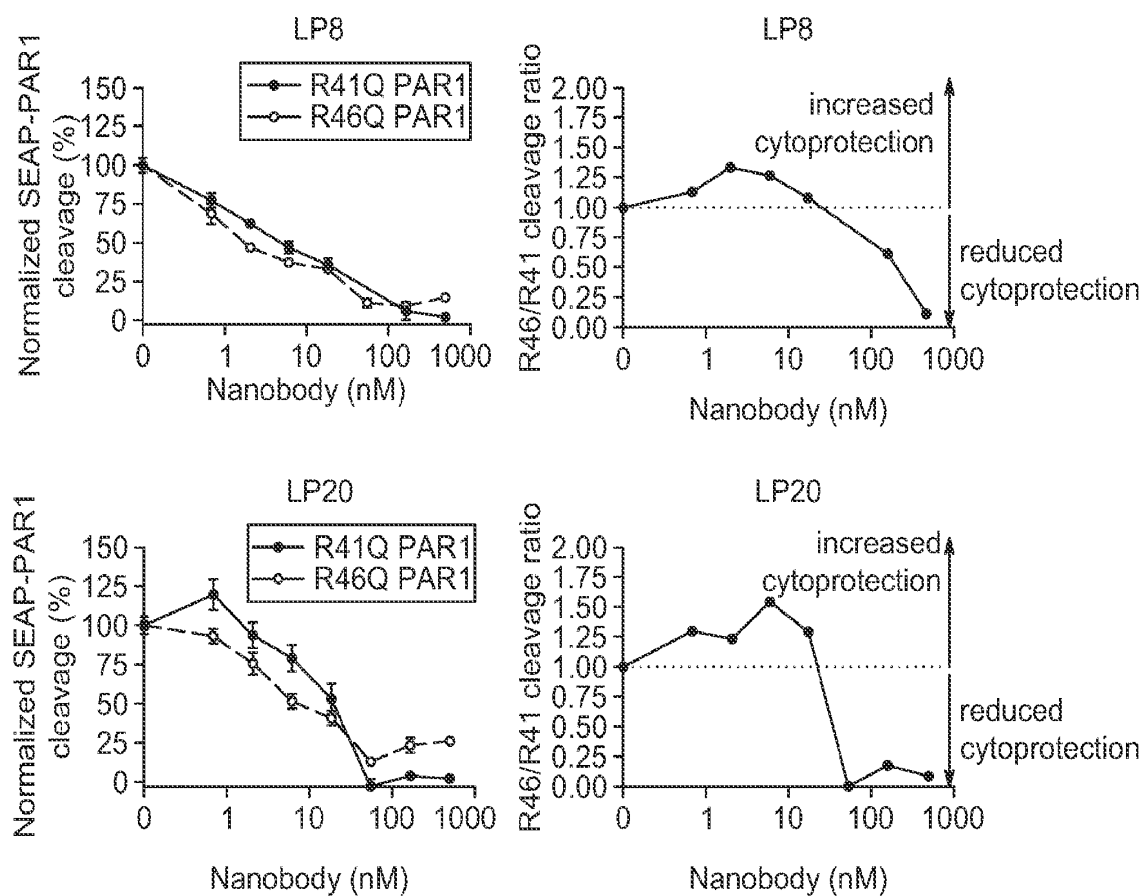


FIG. 4C

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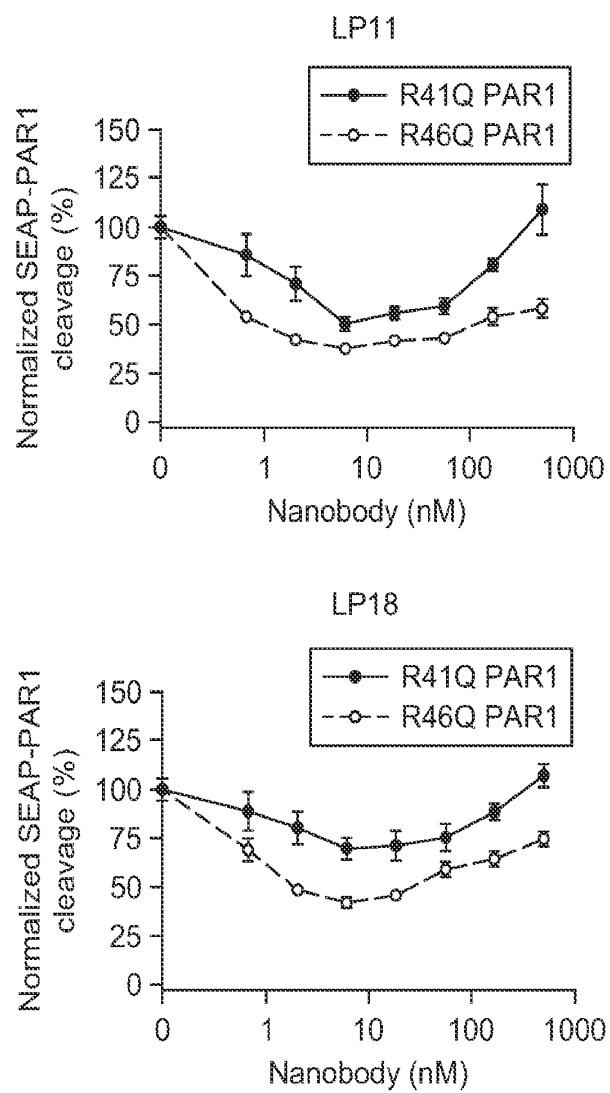


FIG. 4D

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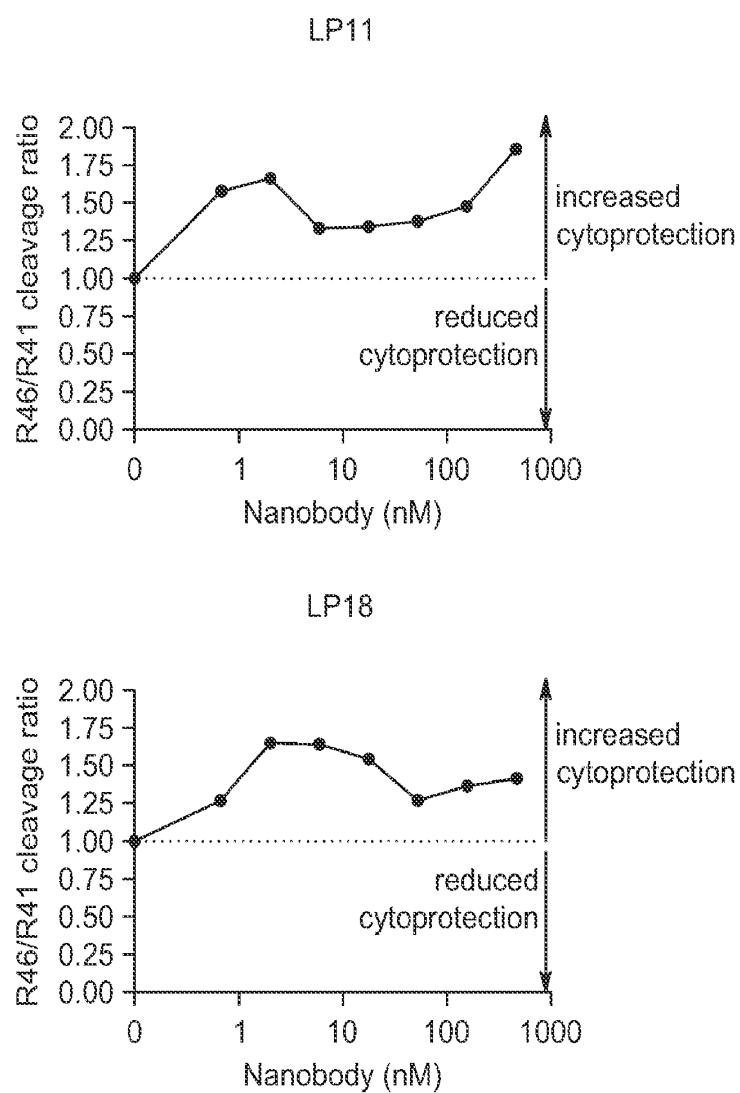


FIG. 4D (Continued)

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|          | R46 Cleavage (%)       |          |          |          |
|----------|------------------------|----------|----------|----------|
| Ab (nM)  | LP8                    | LP11     | LP18     | LP20     |
| 500      | 1.903854               | 108.5549 | 106.6904 | 2.146834 |
| 166.6667 | 5.77                   | 80.08773 | 88.3821  | 3.773875 |
| 55.55556 |                        | 59.08649 | 75.16057 | -2.18392 |
| 18.51852 | 35.56682               | 55.64895 | 70.9784  | 52.99545 |
| 6.17284  | 46.6056                | 50.27576 | 69.52457 | 79.02337 |
| 2.057613 | 62.21896               | 70.50425 | 79.99503 | 93.66718 |
| 0.685871 | 77.28031               | 85.14592 | 88.54834 | 119.9055 |
| 0        | 100                    | 100      | 100      | 100      |
|          |                        |          |          |          |
|          | R41 Cleavage (%)       |          |          |          |
| Ab (nM)  | LP8                    | LP11     | LP18     | LP20     |
| 500      | 14.66883               | 57.89803 | 74.49582 | 25.97716 |
| 166.6667 | 9.487648               | 53.6736  | 64.04361 | 23.38926 |
| 55.55556 | 10.83467               | 42.52482 | 58.82161 | 12.85918 |
| 18.51852 | 33.00104               | 41.29221 | 45.48716 | 40.86204 |
| 6.17284  | 37.11524               | 37.47665 | 41.90987 | 51.33857 |
| 2.057613 | 46.68803               | 42.00603 | 48.26064 | 75.67375 |
| 0.685871 | 68.41719               | 53.50785 | 68.98976 | 93.11083 |
| 0        | 100                    | 100      | 100      | 100      |
|          |                        |          |          |          |
|          | R46/R41 Cleavage Ratio |          |          |          |
| Ab (nM)  | LP8                    | LP11     | LP18     | LP20     |
| 500      | 0.13                   | 1.87     | 1.43     | 0.08     |
| 166.6667 | 0.61                   | 1.49     | 1.38     | 0.16     |
| 55.55556 | -                      | 1.39     | 1.28     | 0        |
| 18.51852 | 1.08                   | 1.35     | 1.56     | 1.3      |
| 6.17284  | 1.26                   | 1.34     | 1.66     | 1.54     |
| 2.057613 | 1.33                   | 1.68     | 1.66     | 1.24     |
| 0.685871 | 1.13                   | 1.59     | 1.28     | 1.29     |
| 0        | 1                      | 1        | 1        | 1        |

FIG. 4E

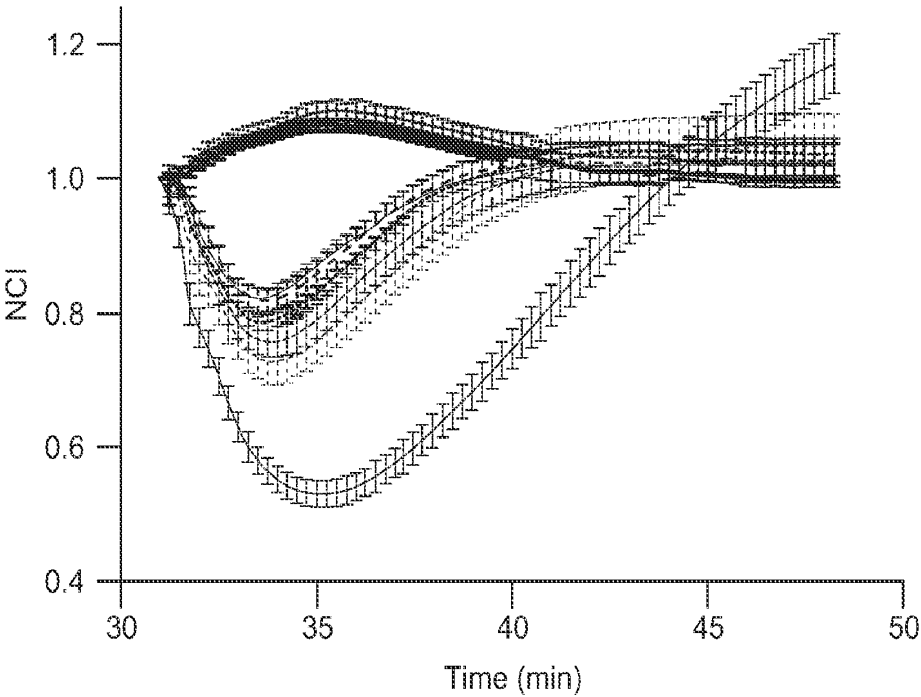
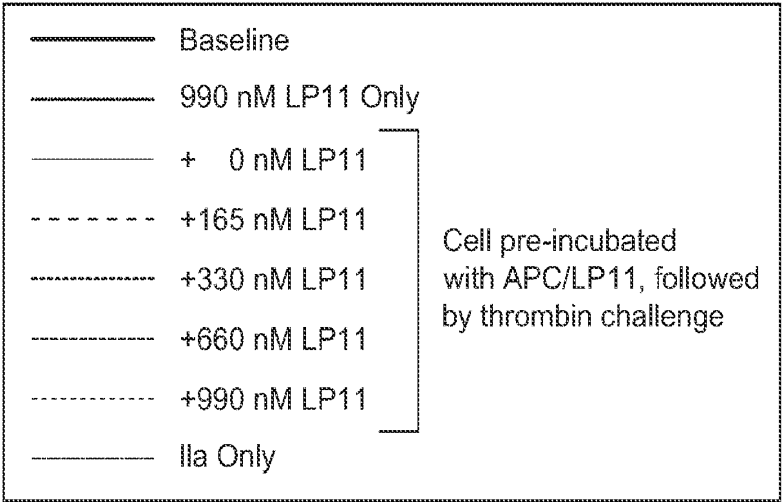


FIG. 5A

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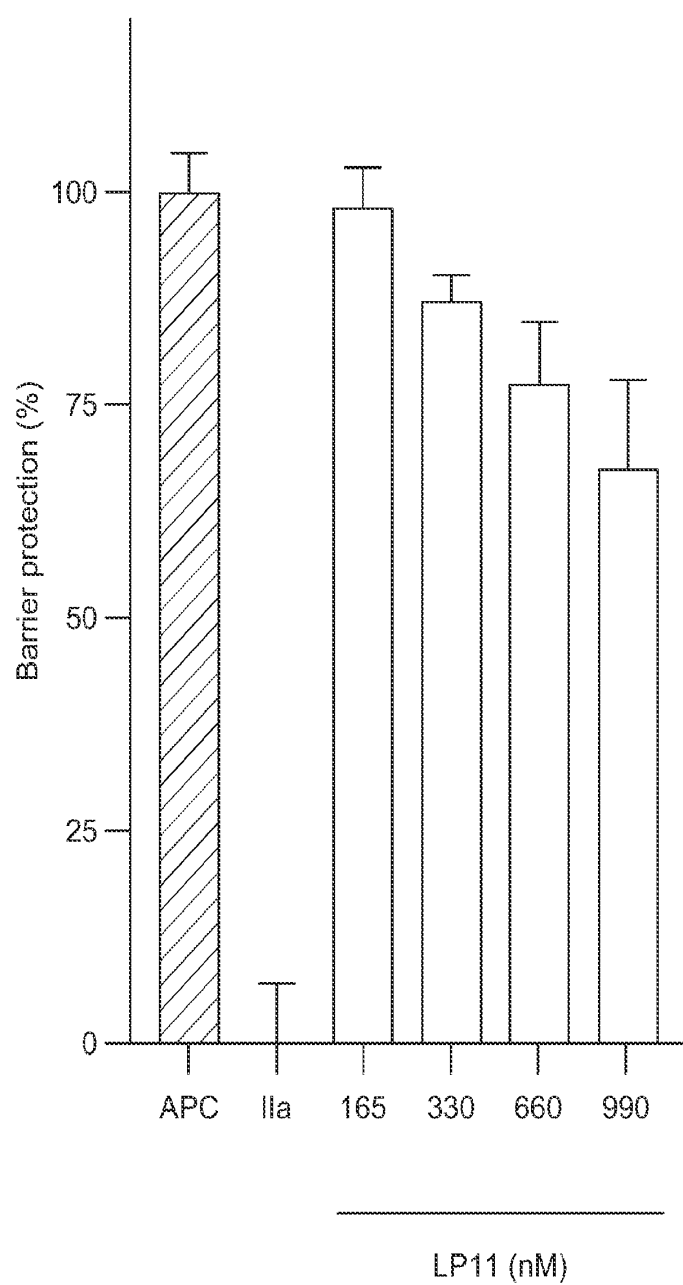


FIG. 5B

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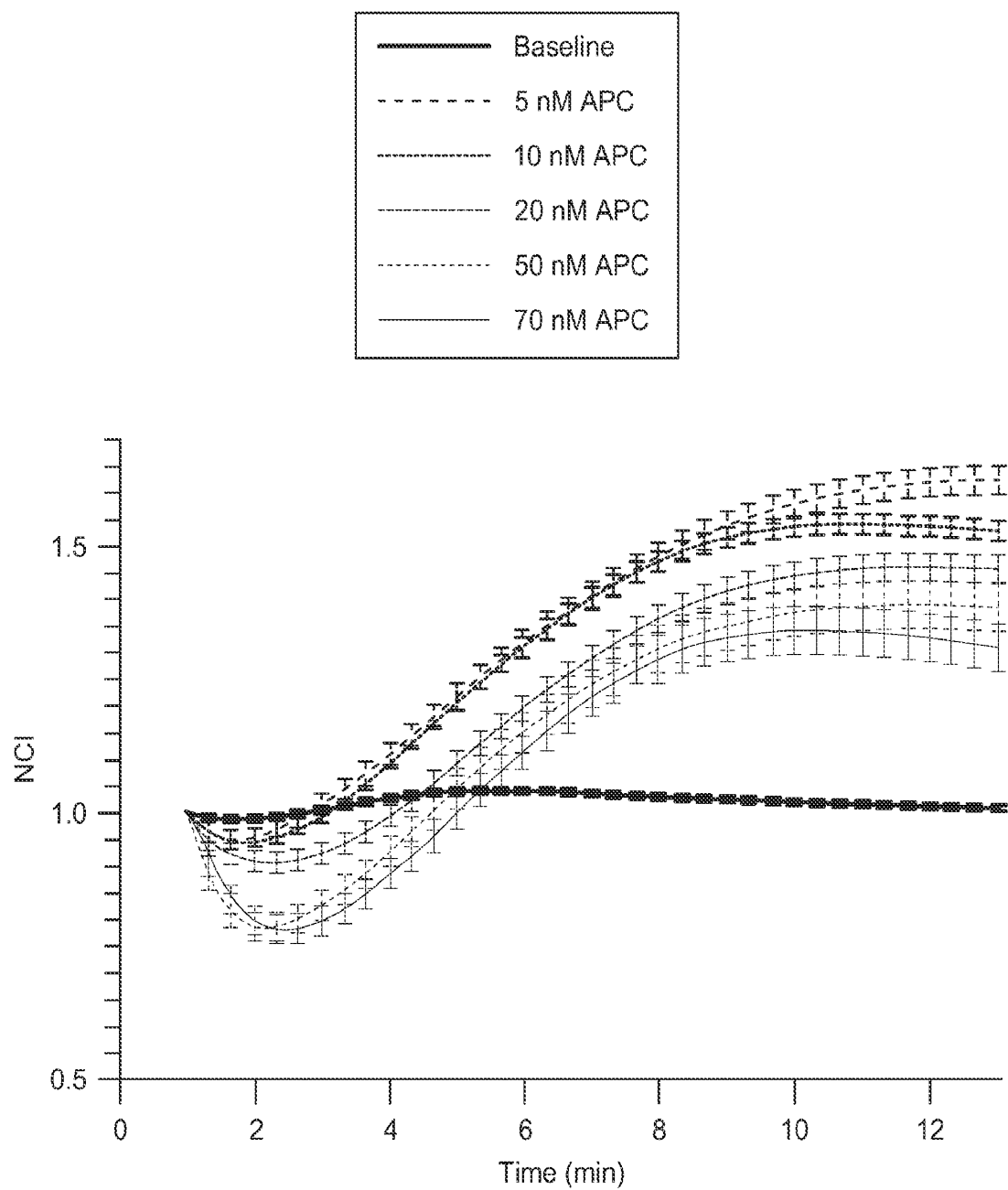


FIG. 5C

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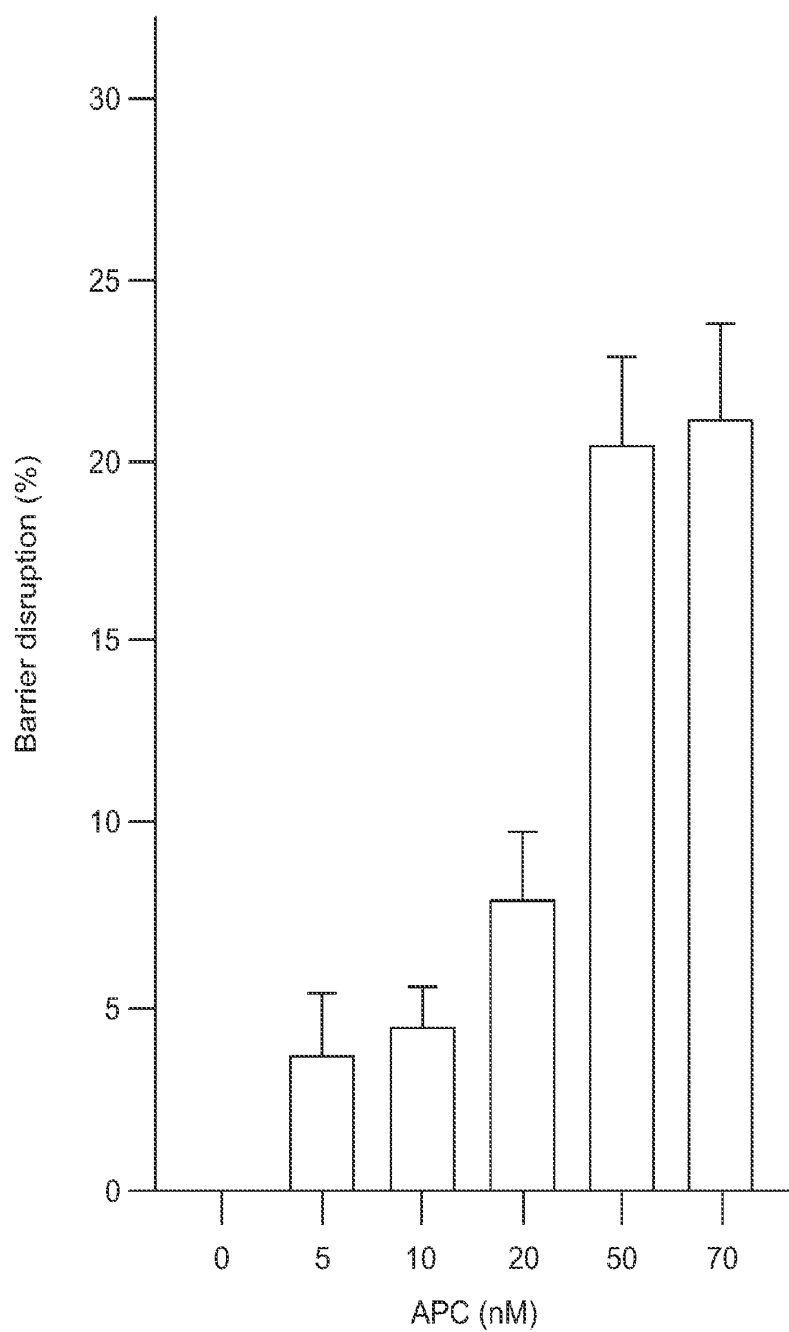


FIG. 5D

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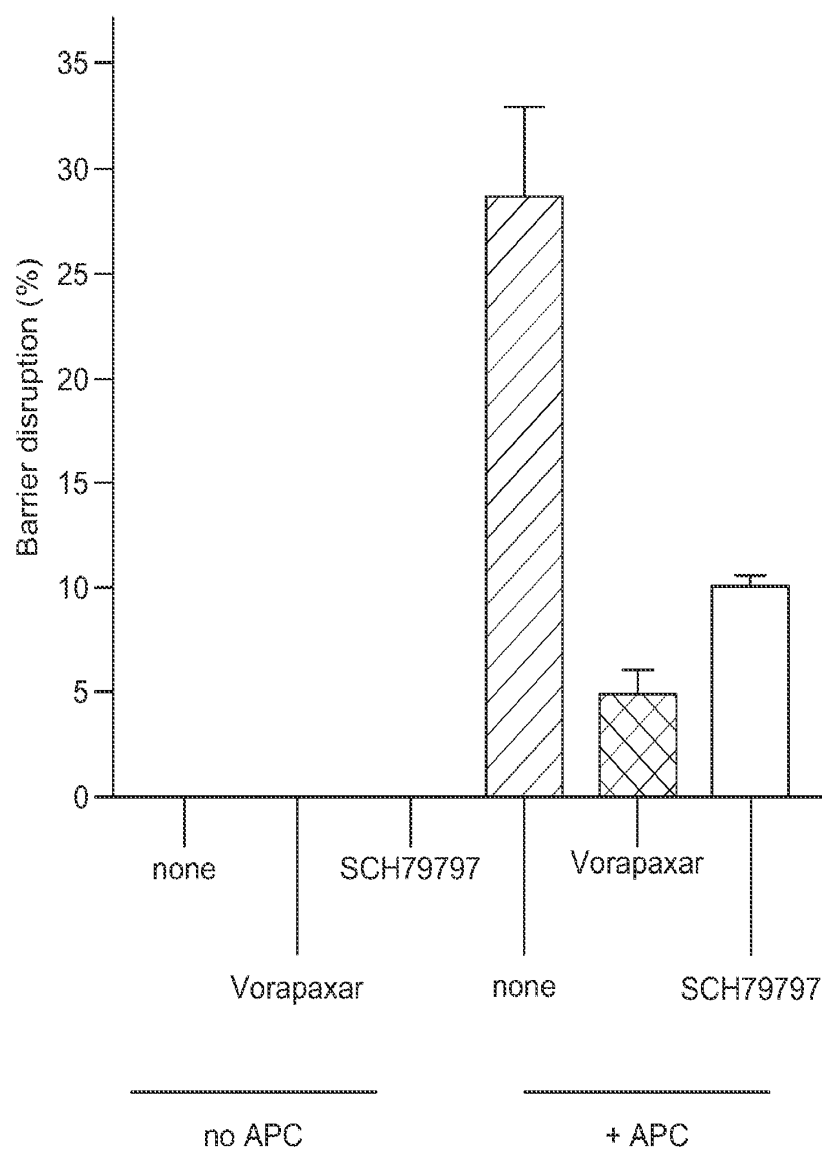


FIG. 5E

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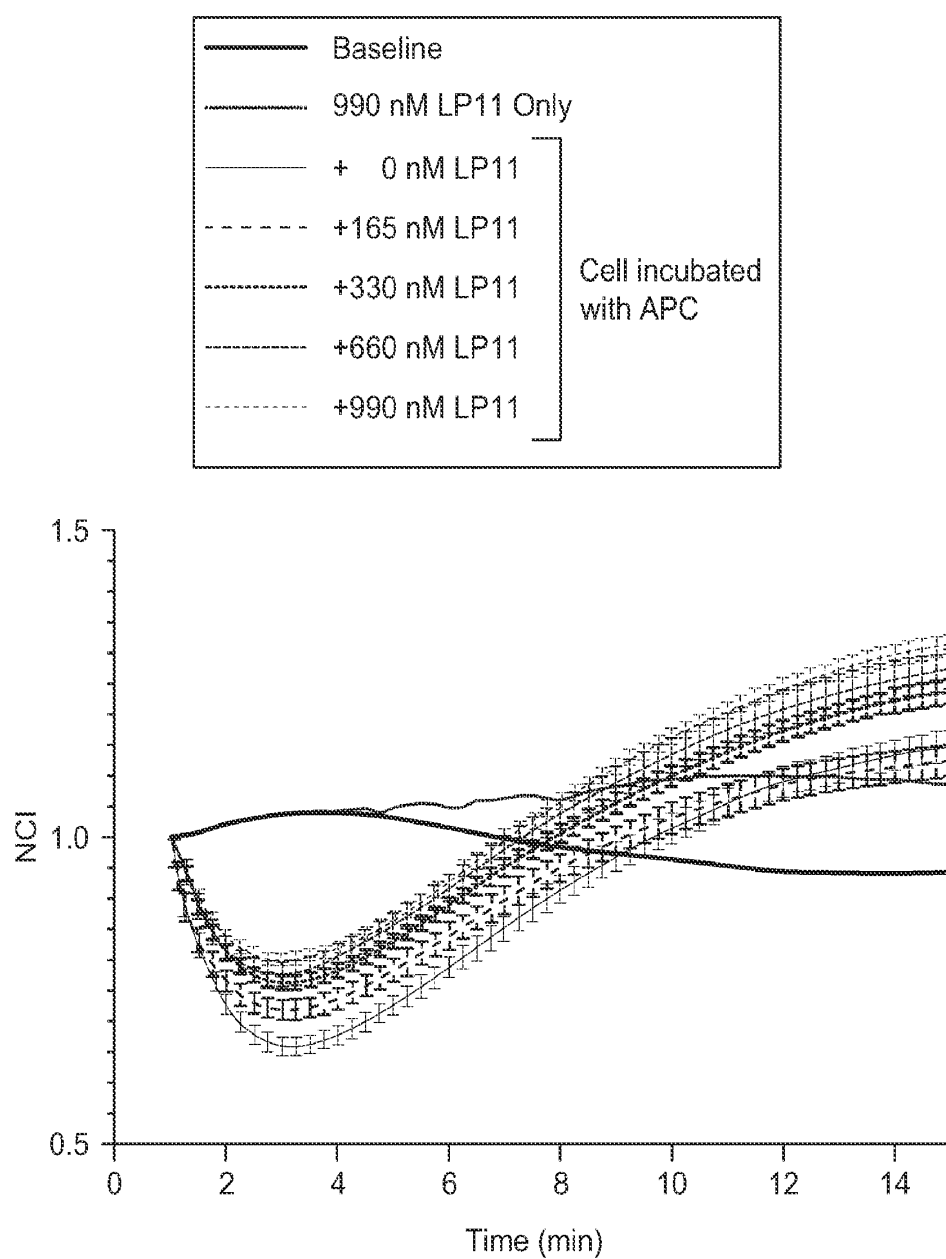


FIG. 5F

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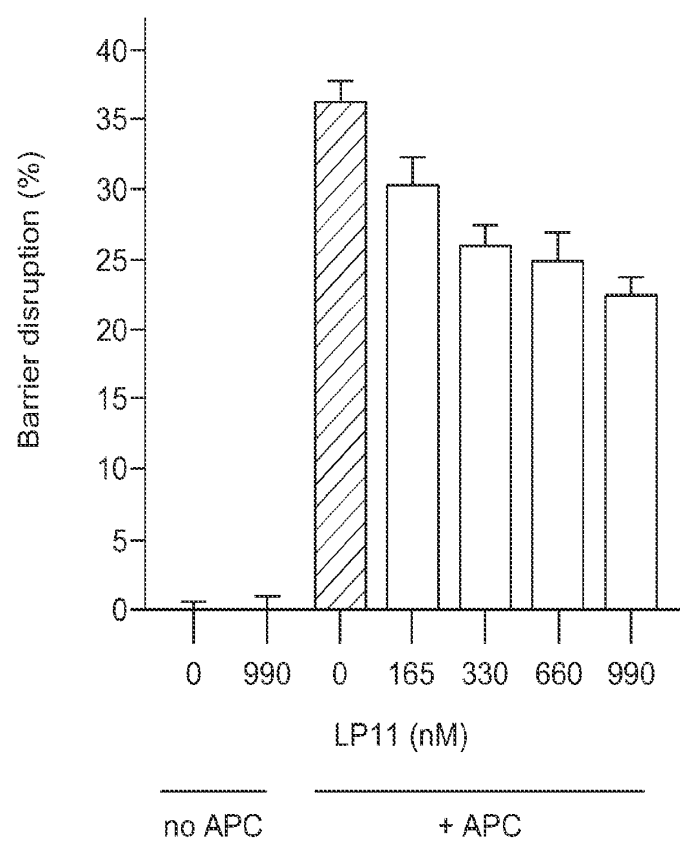


FIG. 5G

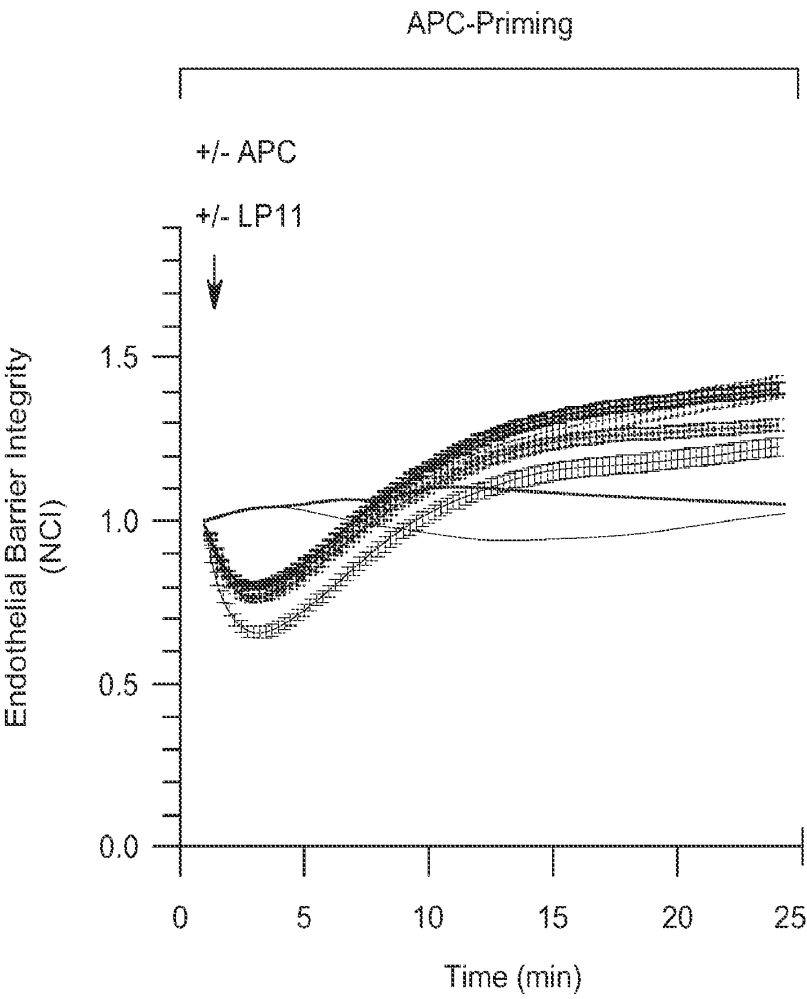
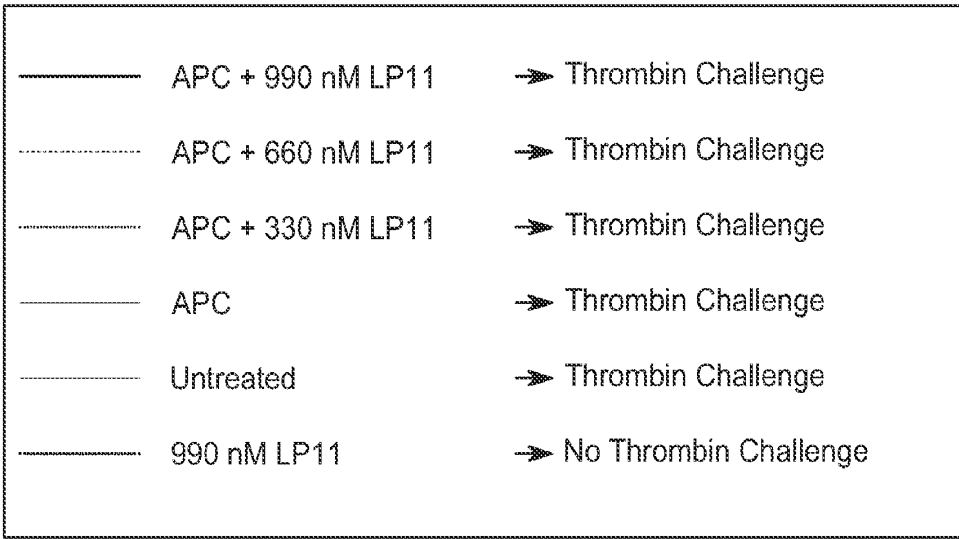
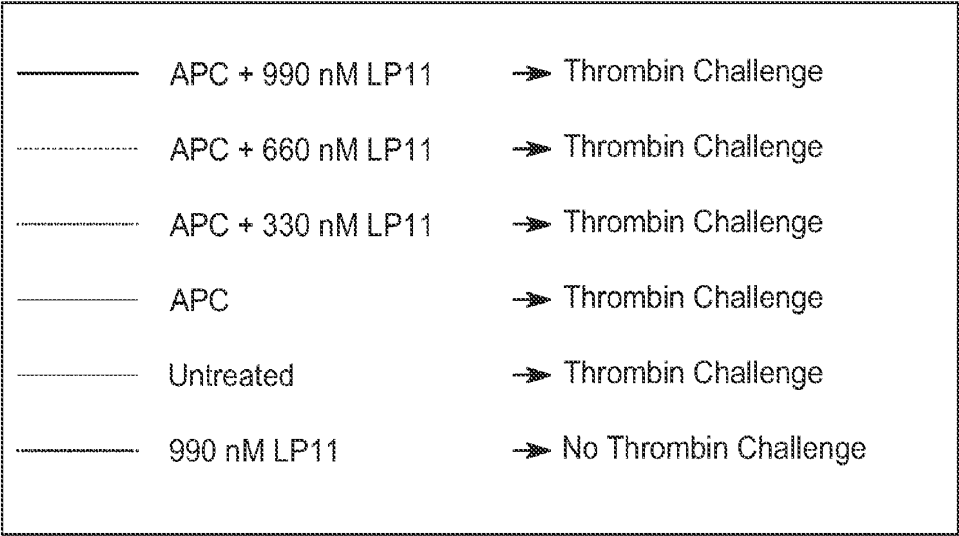


FIG. 5H



Thrombin-Mediated  
Endothelial Disruption

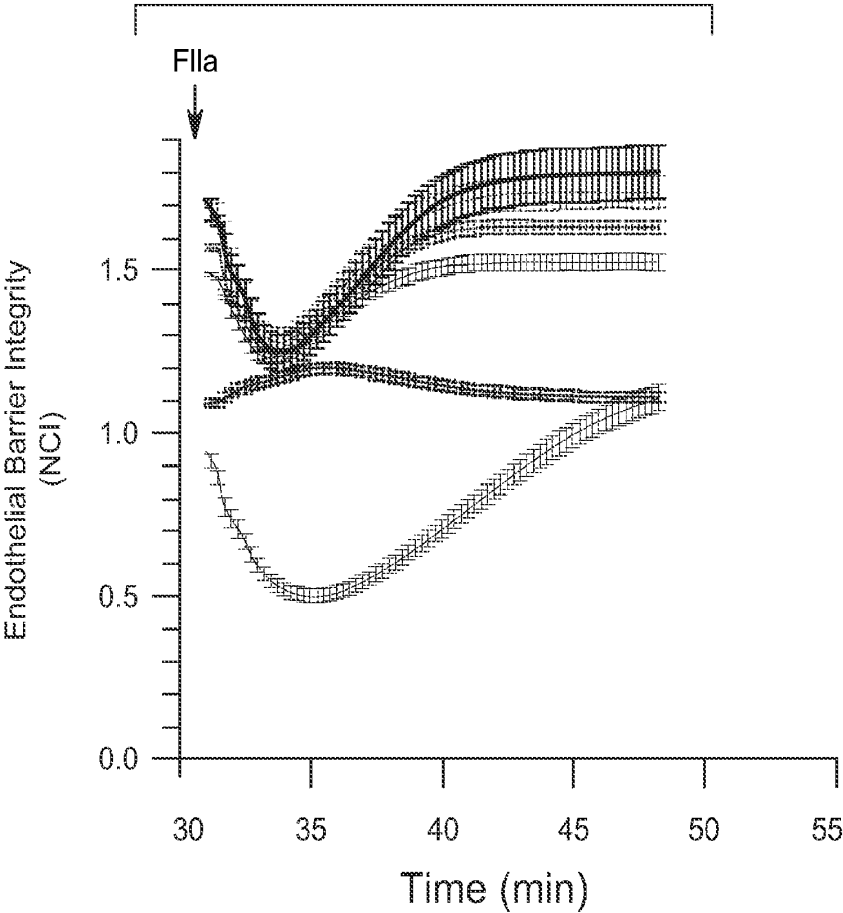


FIG. 5H (Continued)

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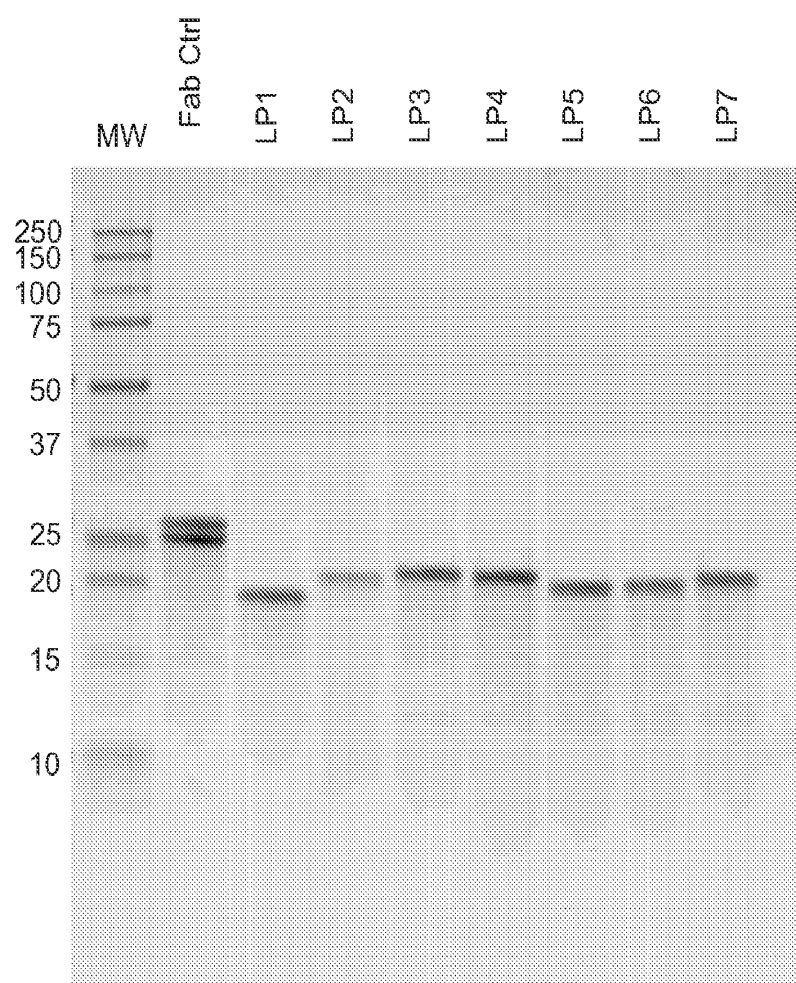


FIG. 6A

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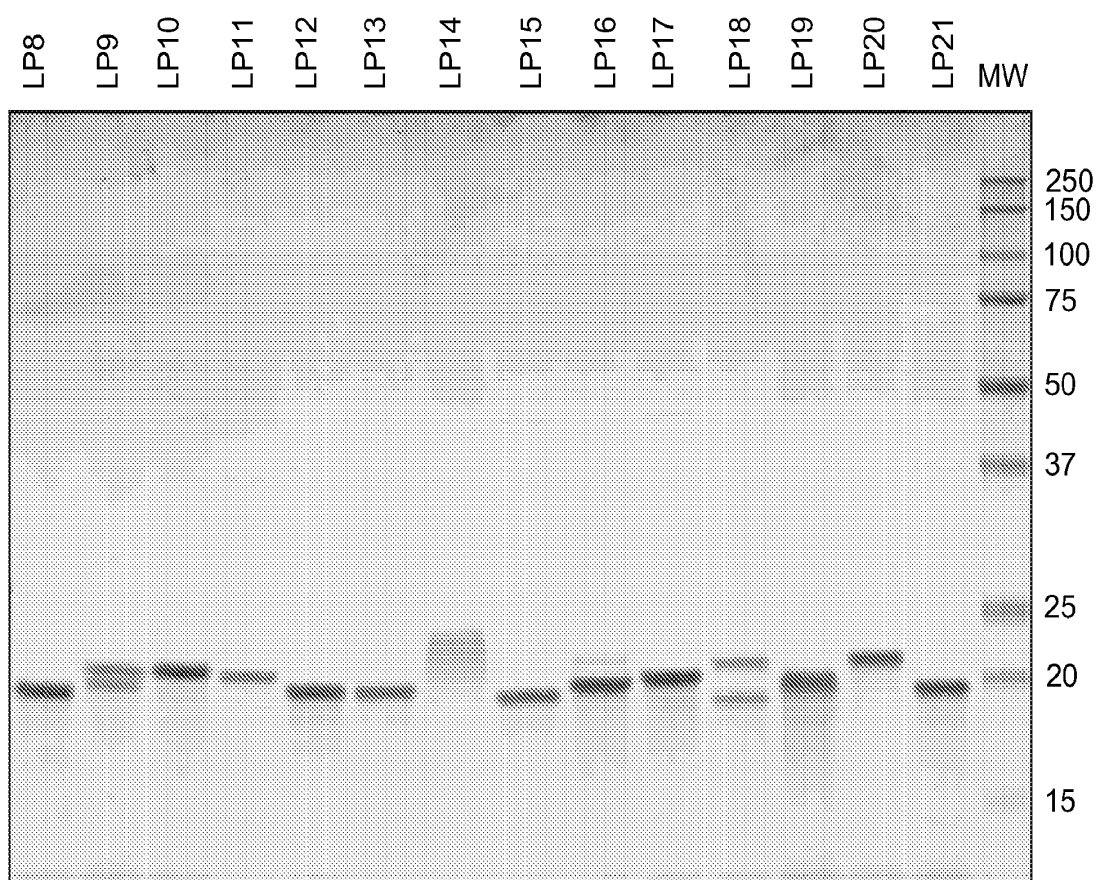


FIG. 6B

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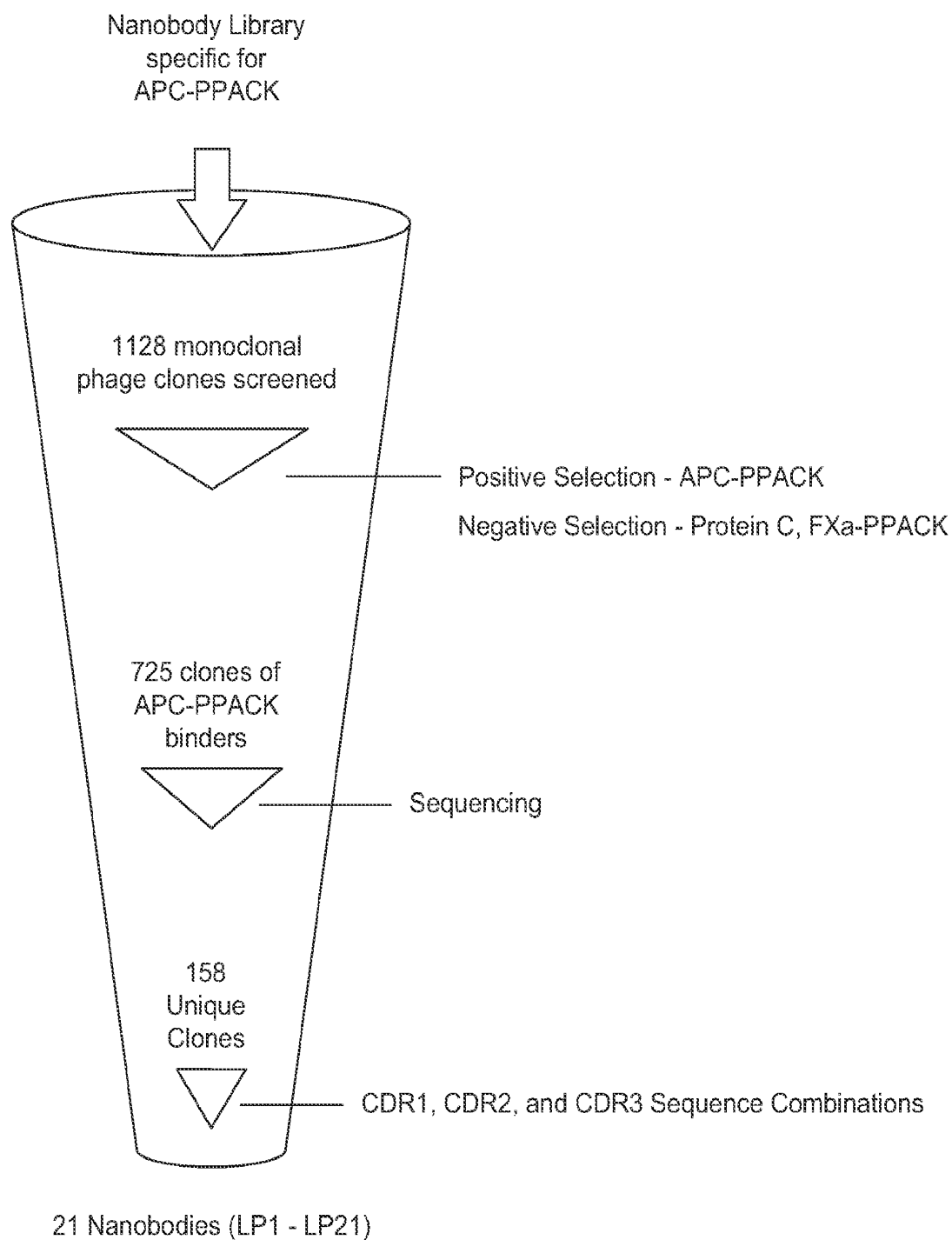


FIG. 7

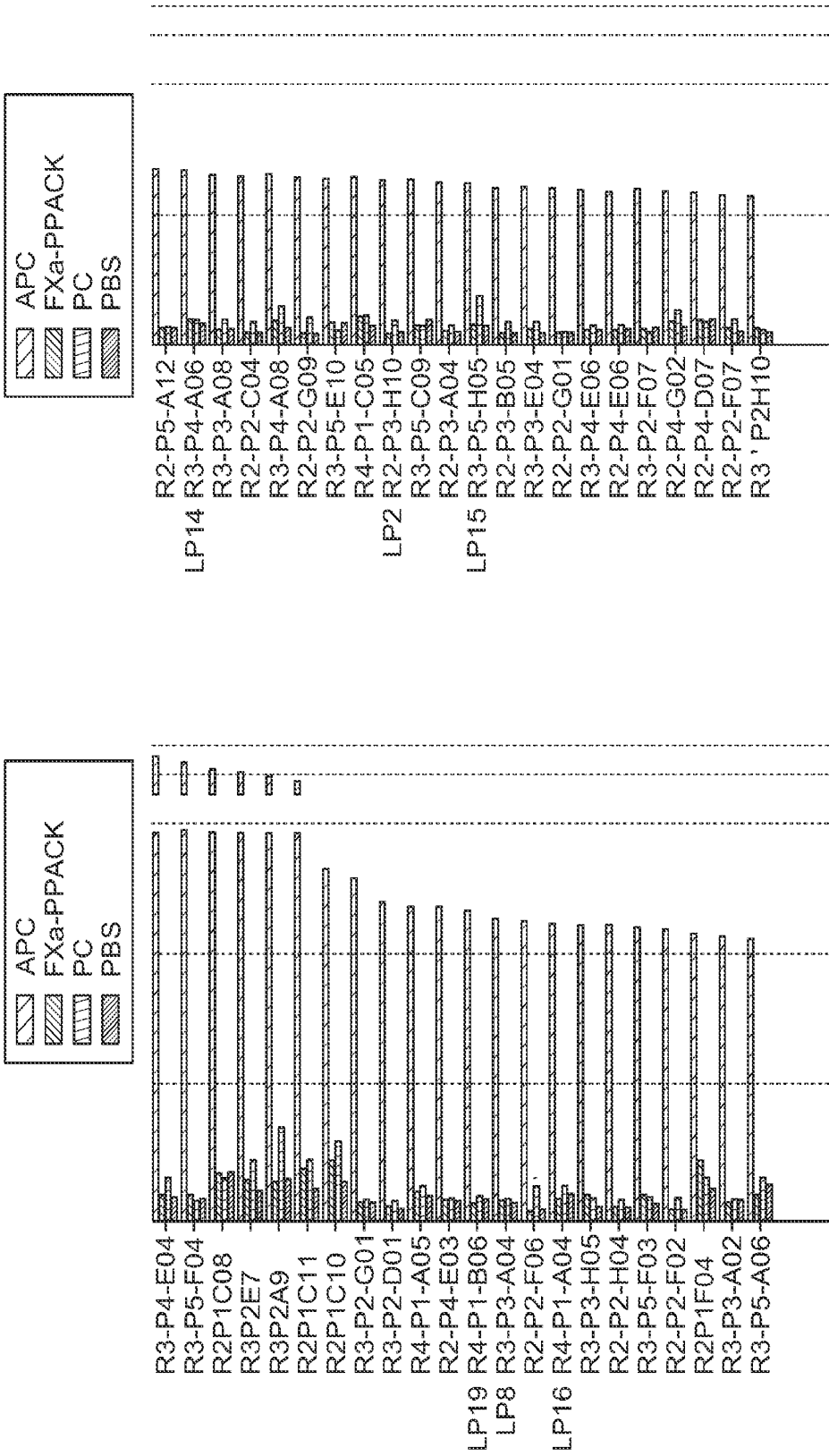


FIG. 8

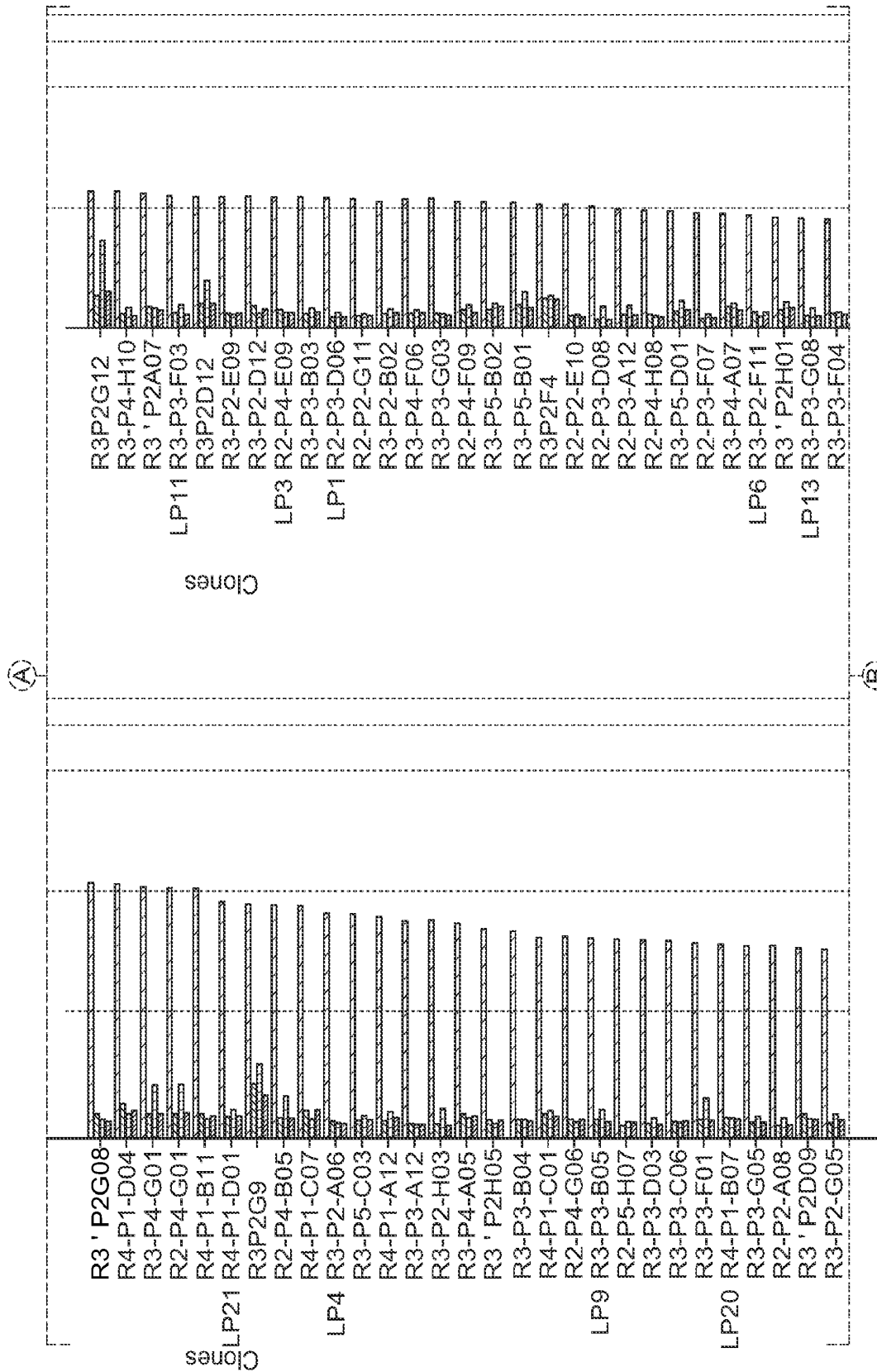


FIG. 8 (Continued)

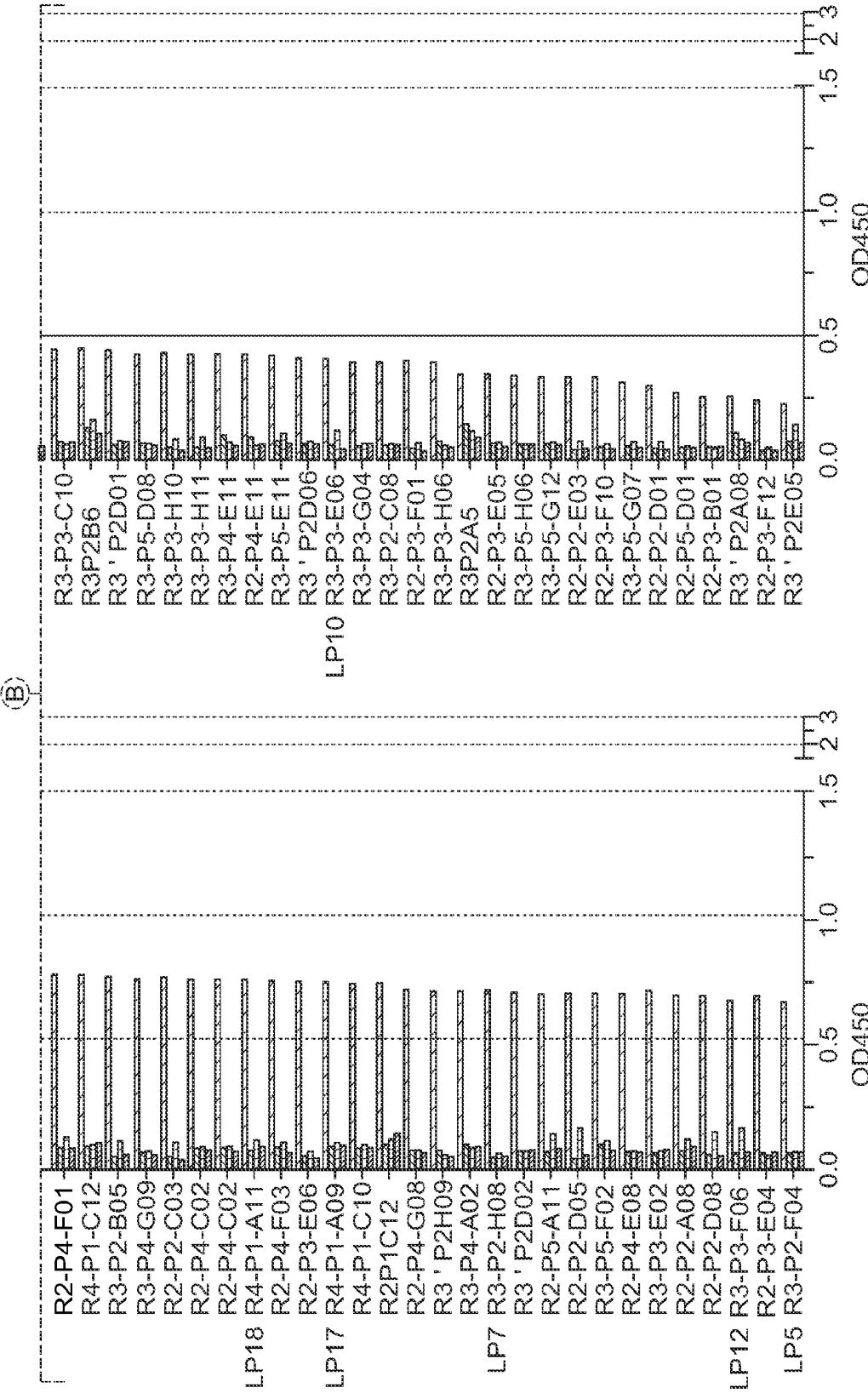


FIG. 8 (Continued)

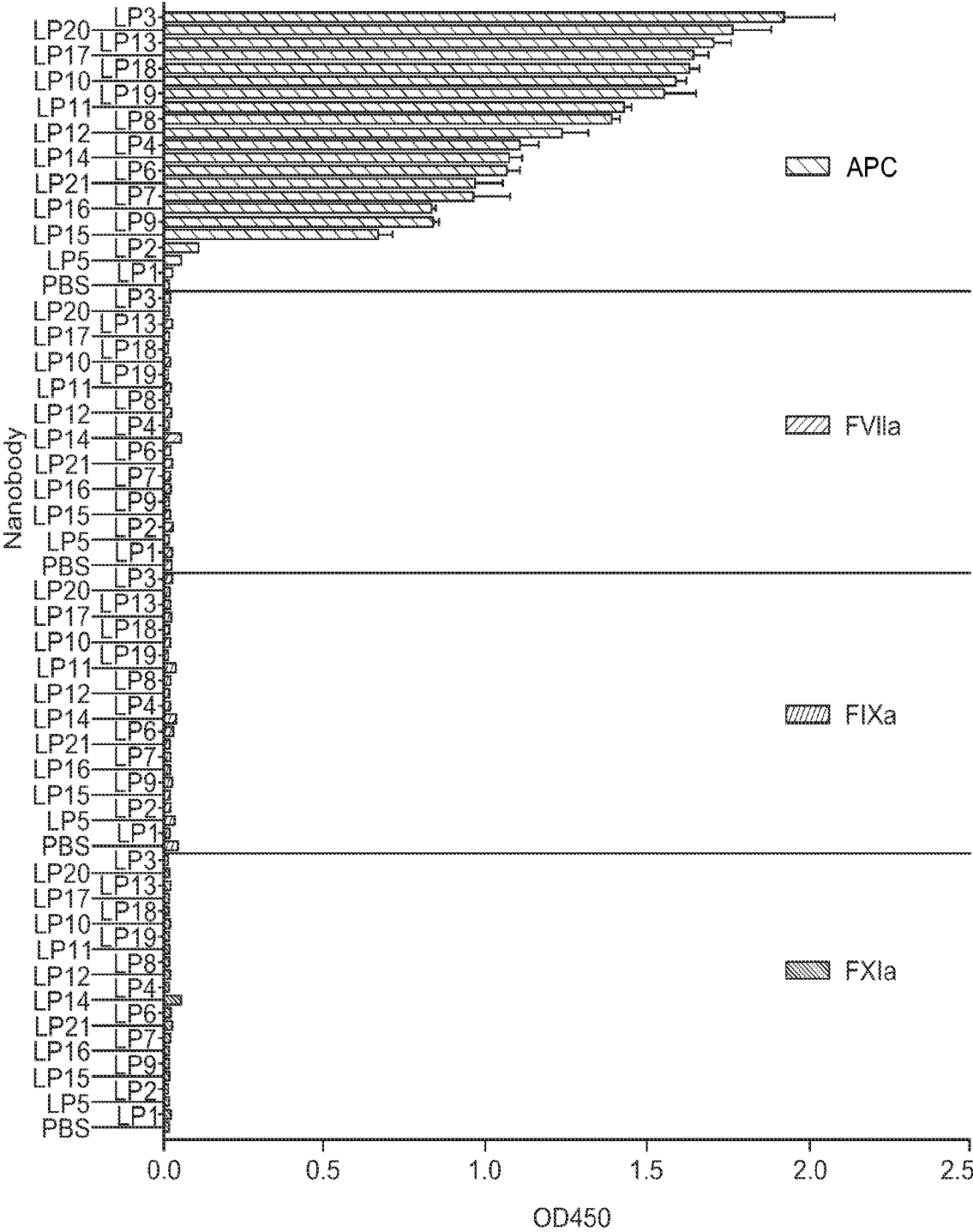


FIG. 9

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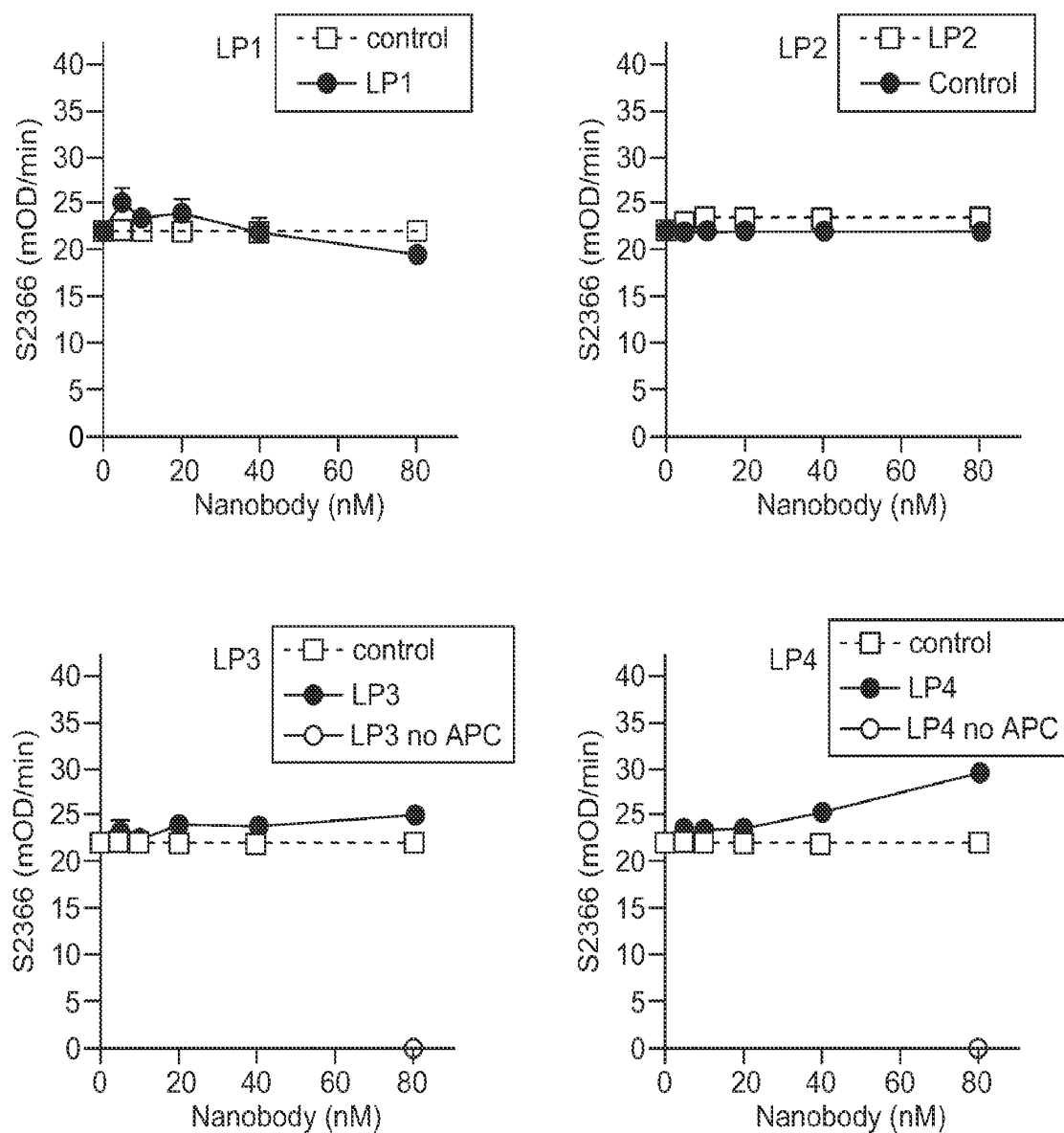


FIG. 10

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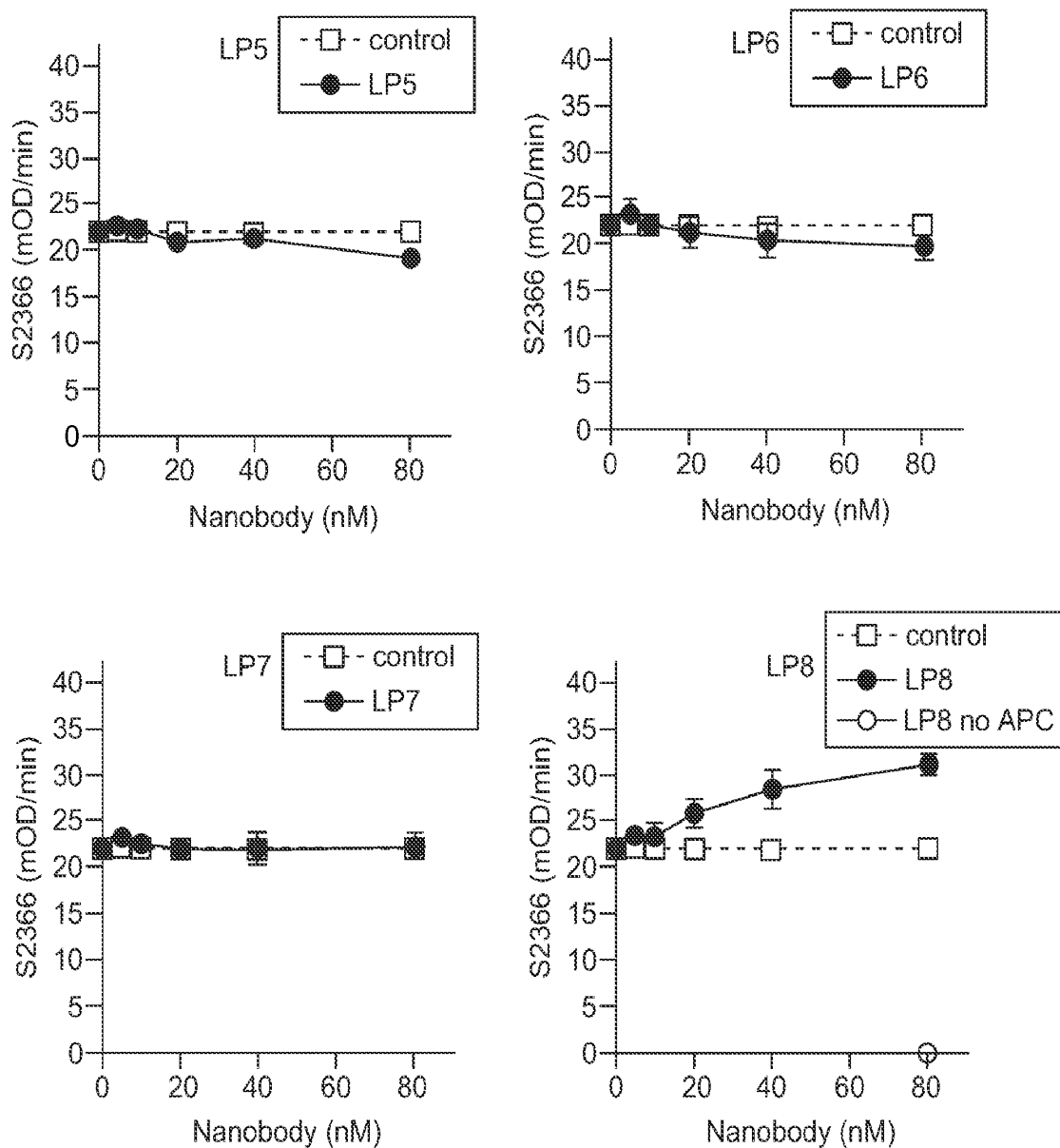


FIG. 10 (Continued)

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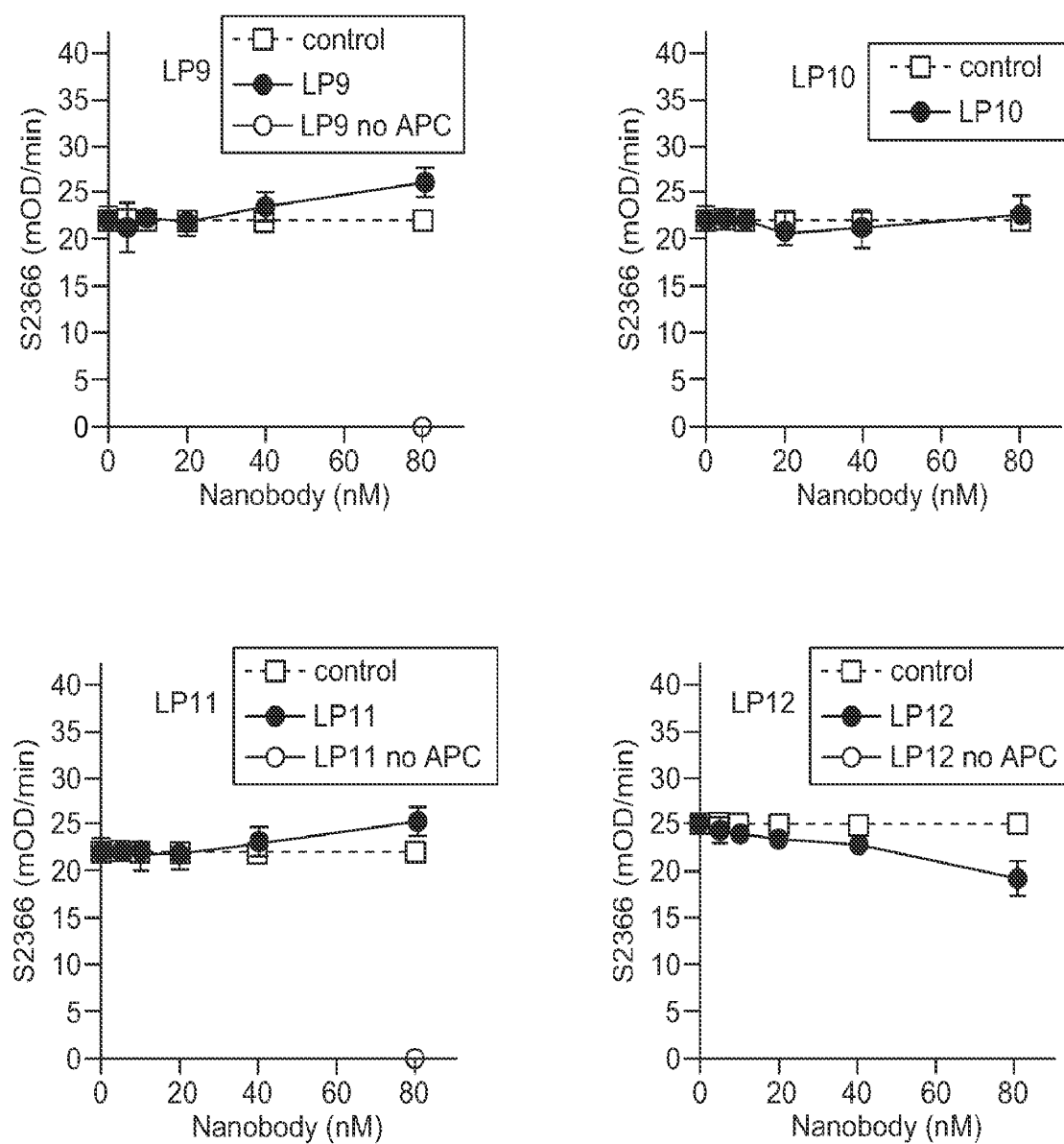


FIG. 10 (Continued)

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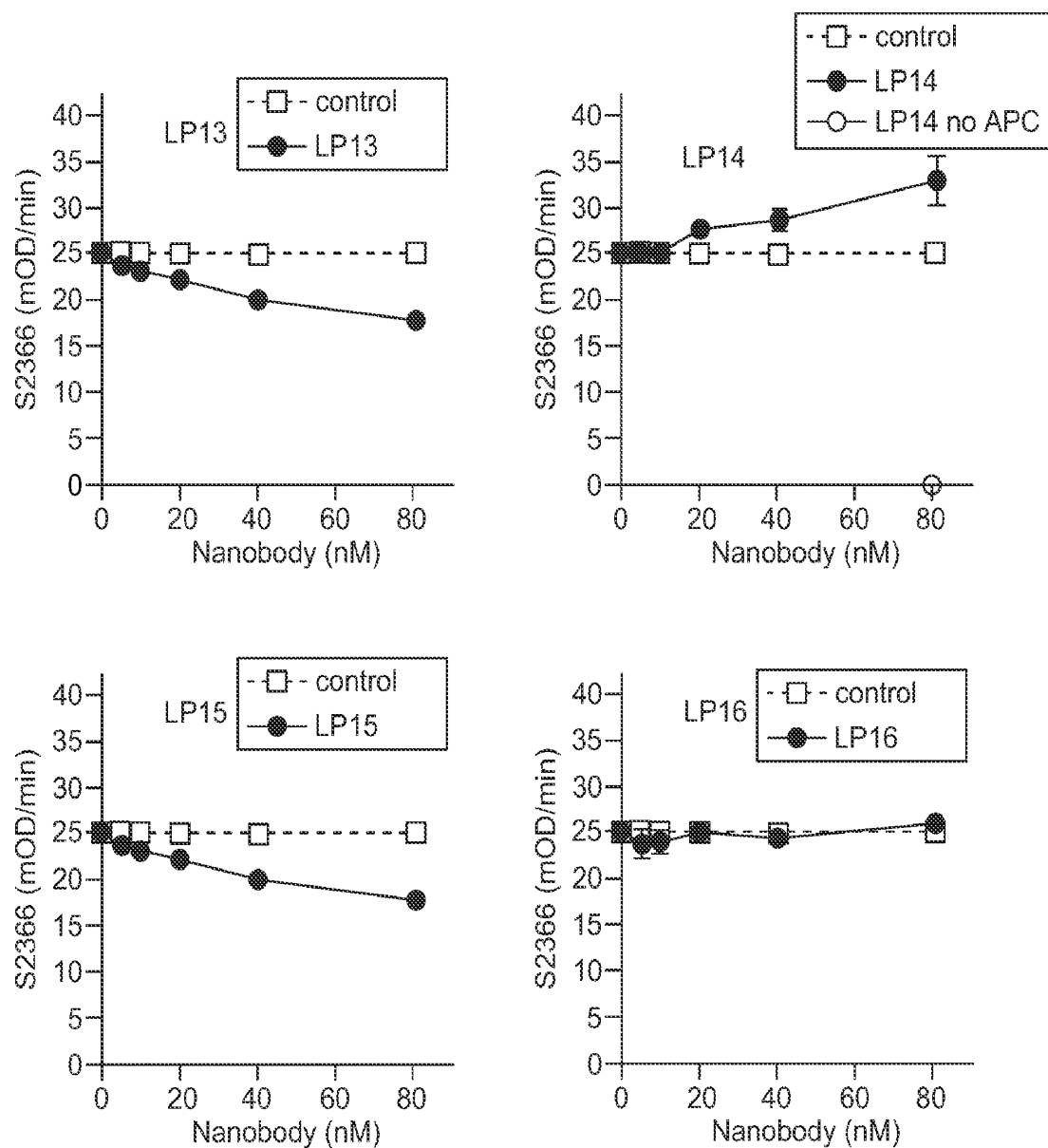


FIG. 10 (Continued)

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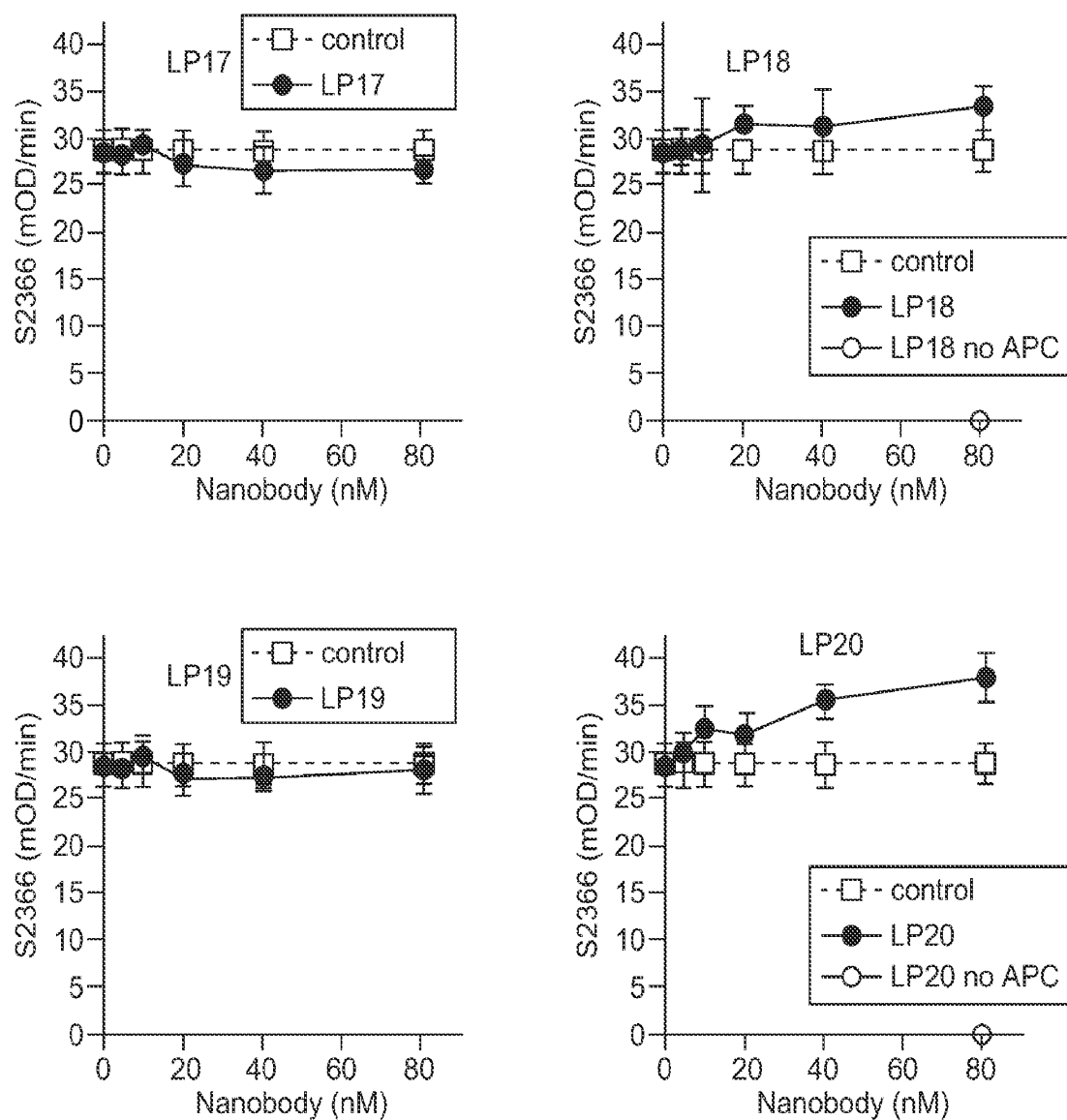


FIG. 10 (Continued)

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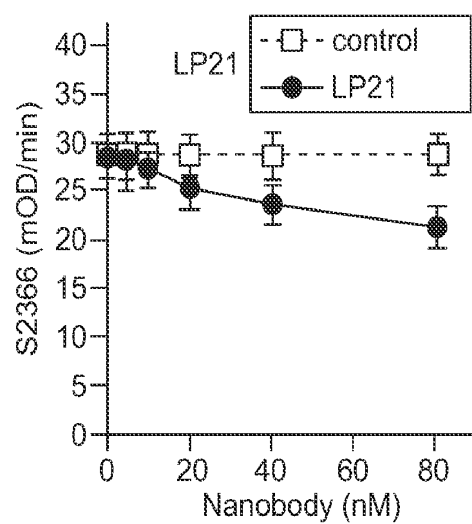


FIG. 10 (Continued)

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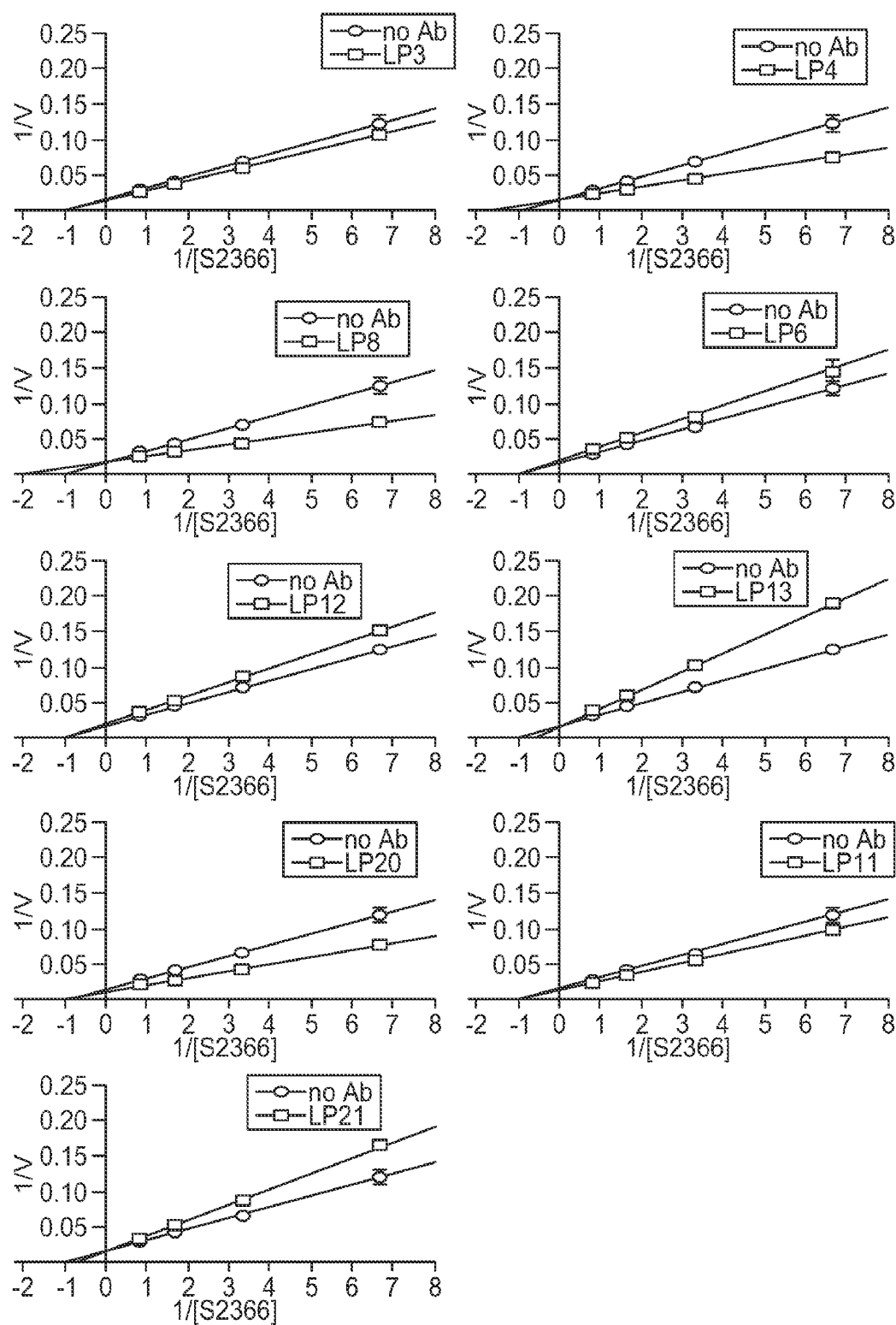


FIG. 11

SUBSTITUTE SHEET (RULE 26)

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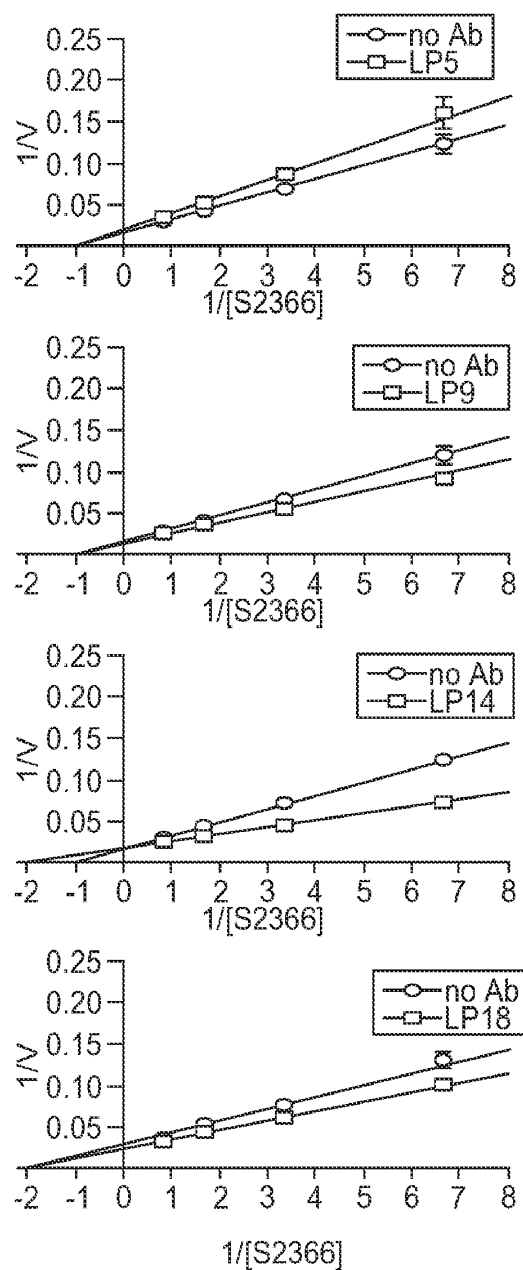


FIG. 11 (Continued)

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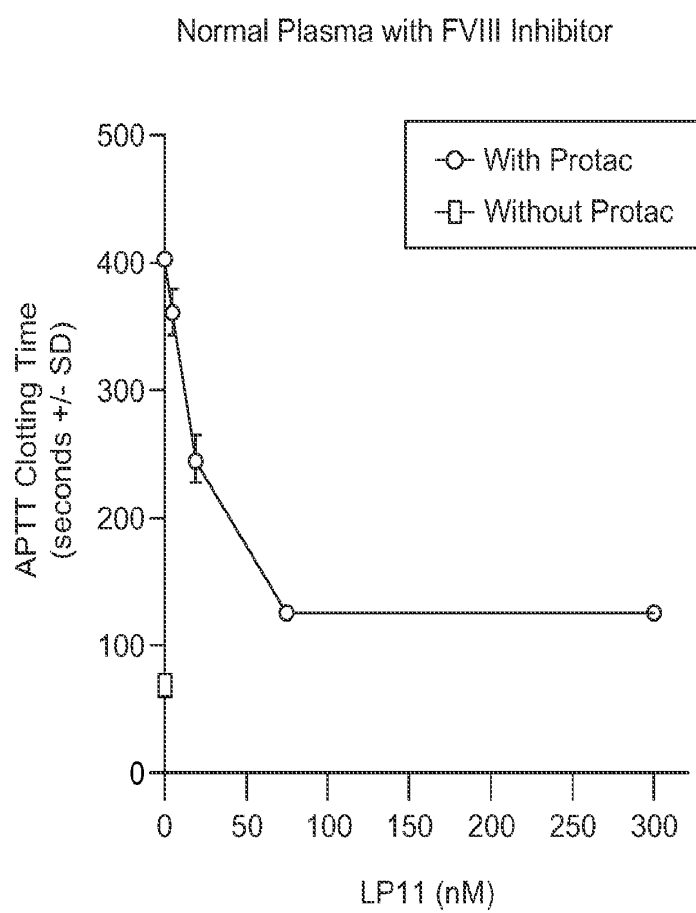


FIG. 12

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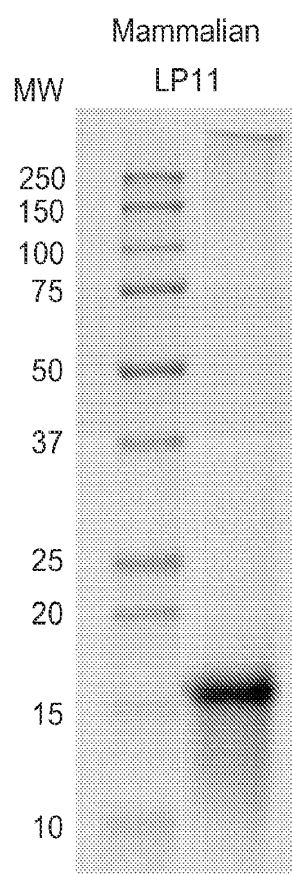


FIG. 13A

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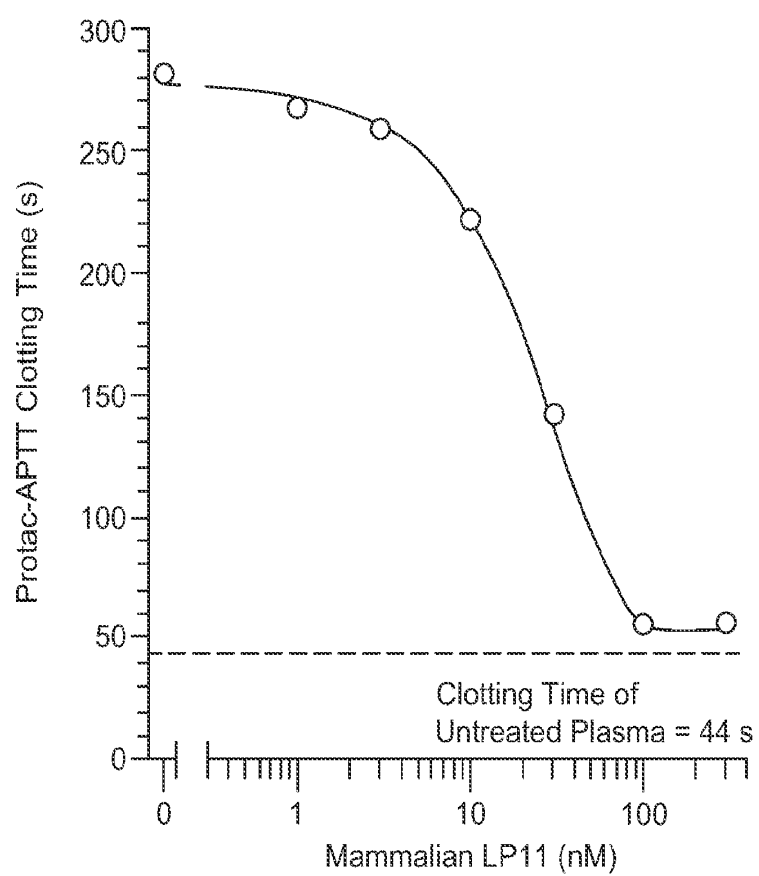


FIG. 13B

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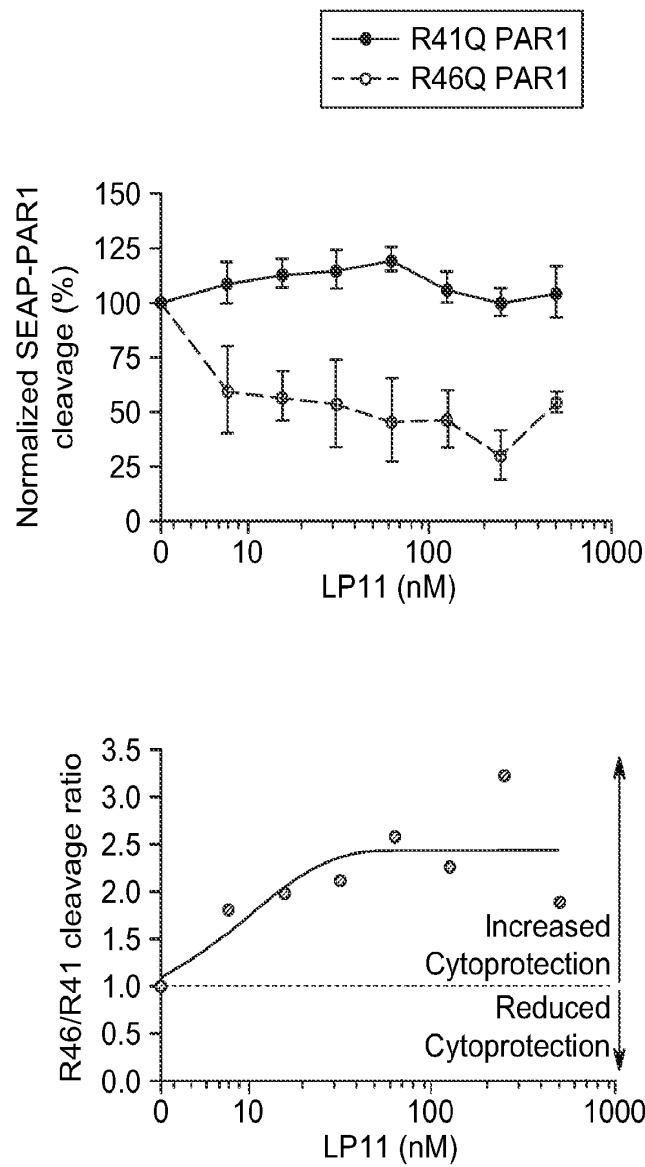


FIG. 13C

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Phage Library  
↓  
Panning Round R1

| Antigen                | Deselections                        | Washes                                                    | Input (CFU/mL)       | Output (CFU/mL)   | Ratio (Out/In)       |
|------------------------|-------------------------------------|-----------------------------------------------------------|----------------------|-------------------|----------------------|
| 100 nM Human APC-PPACK | 1xFXa-PPACK (100nM)<br>1xPC (100nM) | 6xPBST (5 quick + 1x 5 min)<br>6xPBS (5 quick + 1x 5 min) | $3.5 \times 10^{12}$ | $1.9 \times 10^6$ | $5.3 \times 10^{-7}$ |

R1

| Antigen               | Deselections                        | Washes                                                      | Input (CFU/mL)        | Output (CFU/mL)    | Ratio (Out/In)       |
|-----------------------|-------------------------------------|-------------------------------------------------------------|-----------------------|--------------------|----------------------|
| 50 nM Human APC-PPACK | 1xFXa-PPACK (100nM)<br>1xPC (100nM) | 6xPBST (5 quick + 1x 5 min)<br>6xPBS (5 quick + 1x 5 min)   | $5.5 \times 10^{11}$  | $2.8 \times 10^7$  | $5 \times 10^{-5}$   |
| 10 nM Human APC-PPACK | 1xFXa-PPACK (100nM)<br>1xPC (100nM) | 8xPBST (6 quick + 2x 5 min)<br>8xPBS (6 quick + 2x 5 min)   | $2.65 \times 10^{11}$ | $1.2 \times 10^7$  | $4.5 \times 10^{-5}$ |
| 1 nM Human APC-PPACK  | 1xFXa-PPACK (100nM)<br>1xPC (100nM) | 10xPBST (6 quick + 4x 5 min)<br>10xPBS (6 quick + 4x 5 min) | $3.15 \times 10^{12}$ | $3.85 \times 10^8$ | $1.2 \times 10^{-4}$ |

R2  
R3  
R4

| Antigen               | Deselections                        | Washes                                                    | Input (CFU/mL)    | Output (CFU/mL)   | Ratio (Out/In)       |
|-----------------------|-------------------------------------|-----------------------------------------------------------|-------------------|-------------------|----------------------|
| 50 nM Human APC-PPACK | 1xFXa-PPACK (100nM)<br>3xPC (100nM) | 6xPBST (5 quick + 1x 5 min)<br>6xPBS (5 quick + 1x 5 min) | $6.5 \times 10^9$ | $7.5 \times 10^7$ | $1.1 \times 10^{-2}$ |
| 10 nM Human APC-PPACK | 1xFXa-PPACK (100nM)<br>3xPC (100nM) | 8xPBST (6 quick + 2x 5 min)<br>8xPBS (6 quick + 2x 5 min) | $5 \times 10^8$   | $1.1 \times 10^6$ | $2.2 \times 10^{-3}$ |

R2'  
R3'

FIG. 14A

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| Panning Round | Number of Clones Screened | Binders<br>(3-Fold over Background) | Unique Clones                                         |
|---------------|---------------------------|-------------------------------------|-------------------------------------------------------|
| R2            | 470                       | 366                                 | 188                                                   |
| R3            | 470                       | 371                                 | 190                                                   |
| R4            | 94                        | 42                                  | 25                                                    |
| R3'           | 94                        | 90                                  | 59                                                    |
| Summary       | 1128                      | 725<br>(APC-PPACK positive)         | 158<br>(Sequence Unique<br>and APC-PPACK<br>positive) |

FIG. 14B

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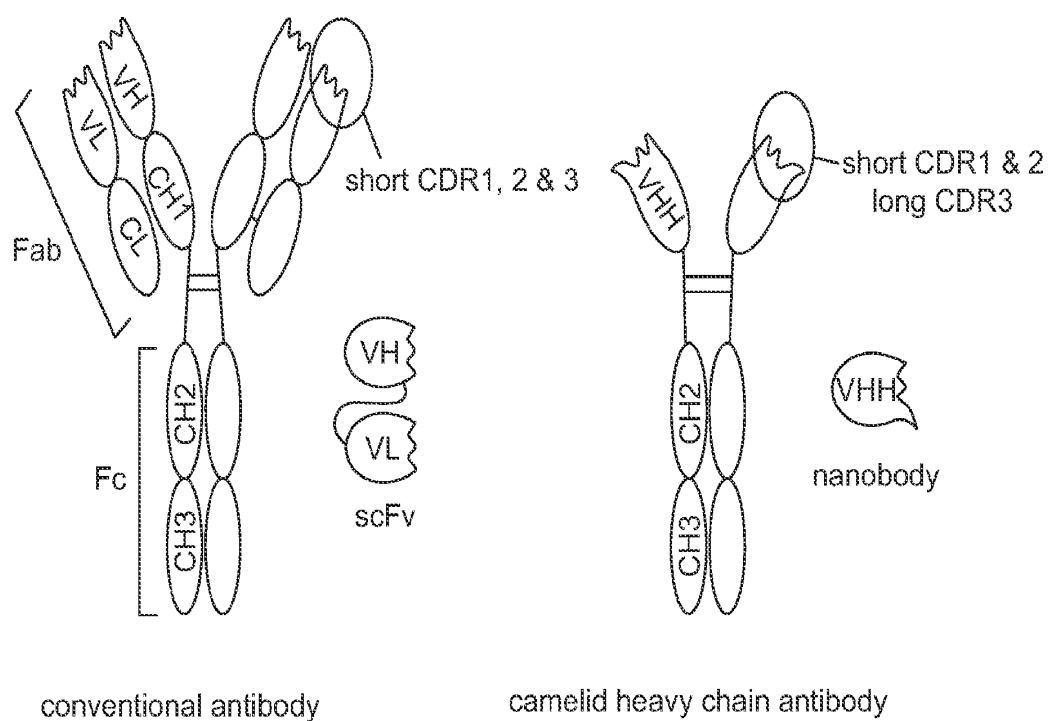


FIG. 15