

Fig. 1A

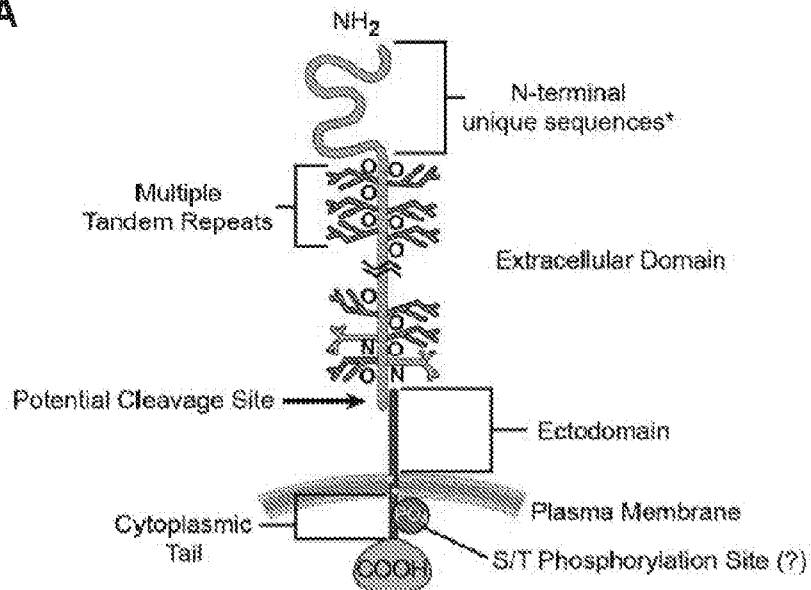


Fig. 1B

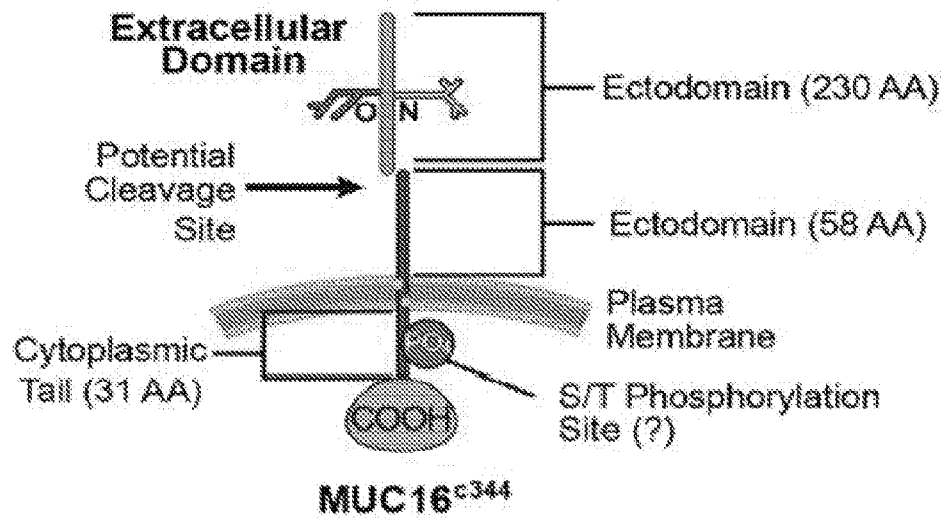
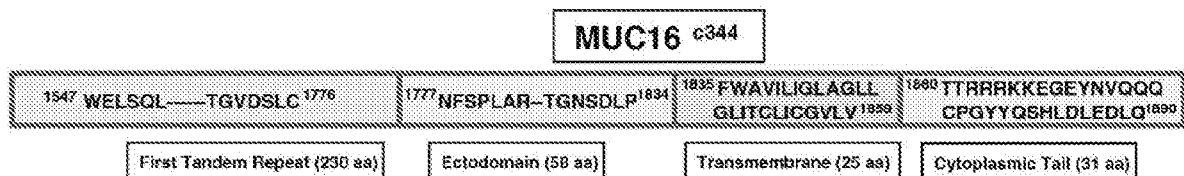


Fig. 1C

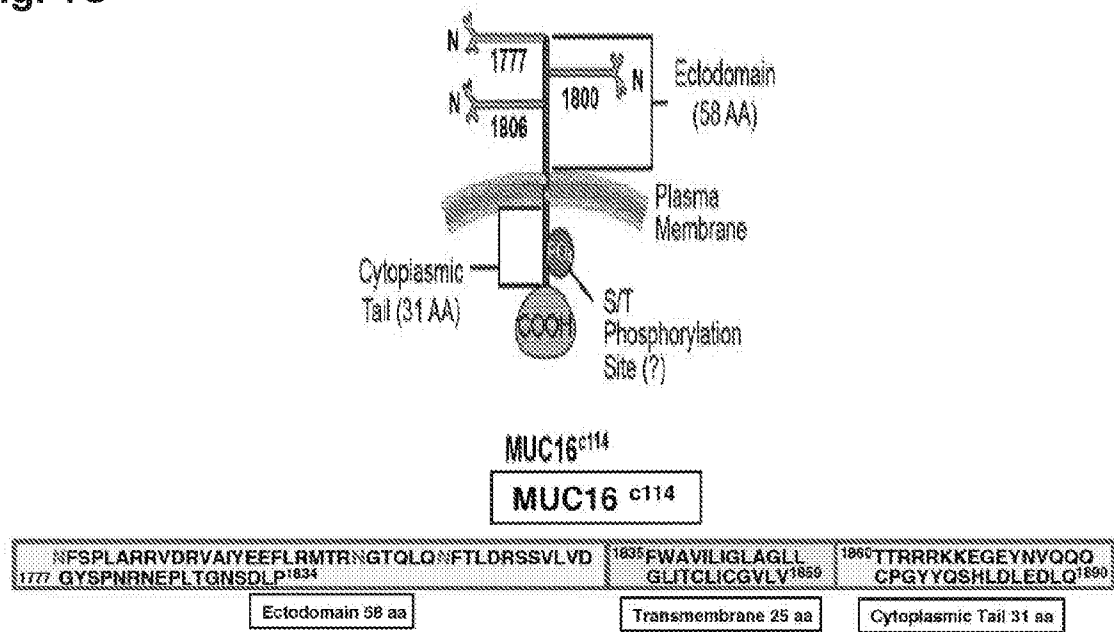


Fig. 1D

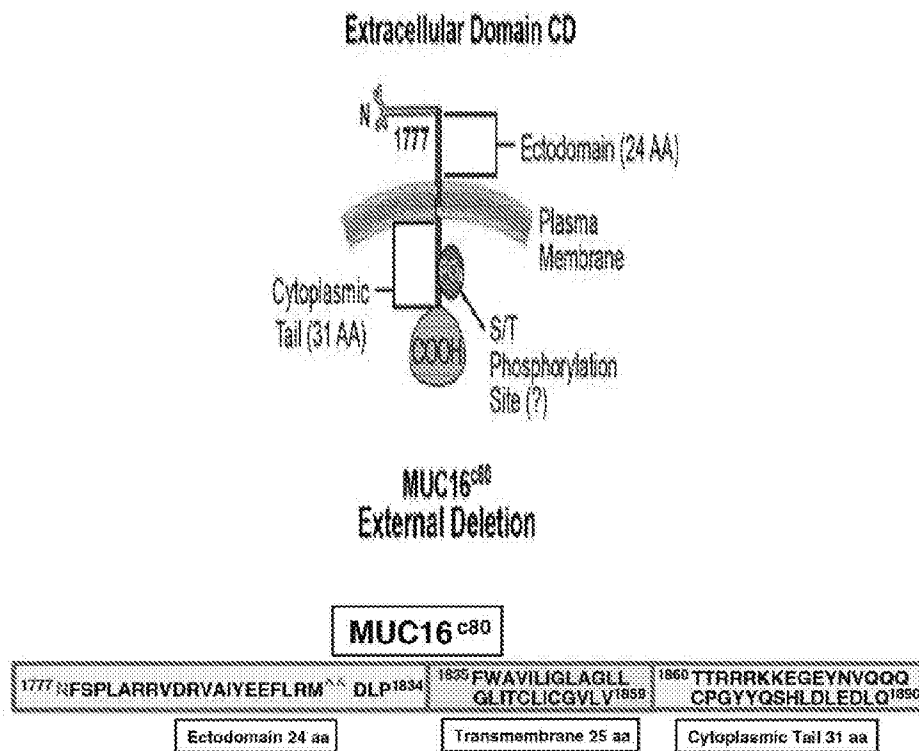
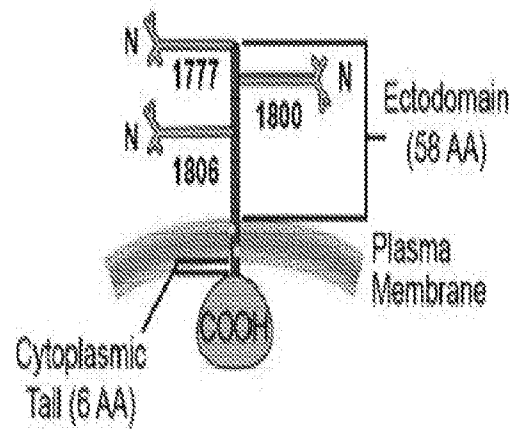


Fig. 1E



**MUC16<sup>c86</sup>**  
Internal Deletion

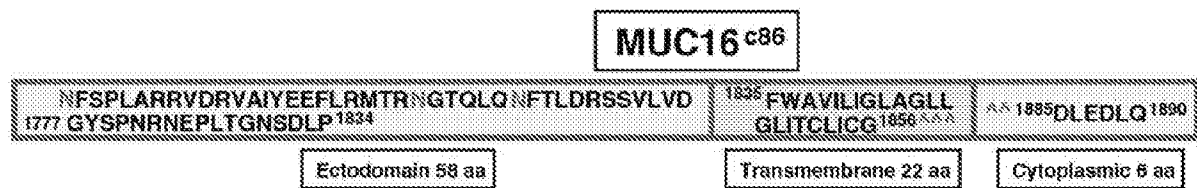


Fig. 1F

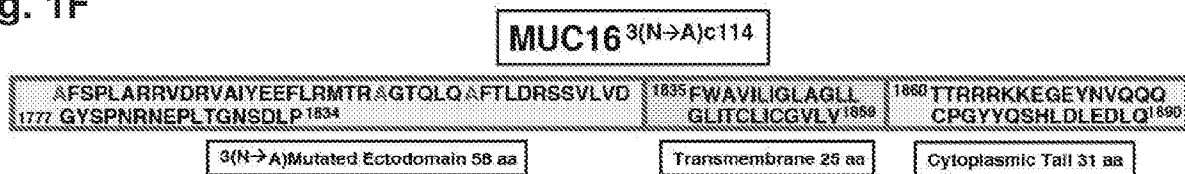


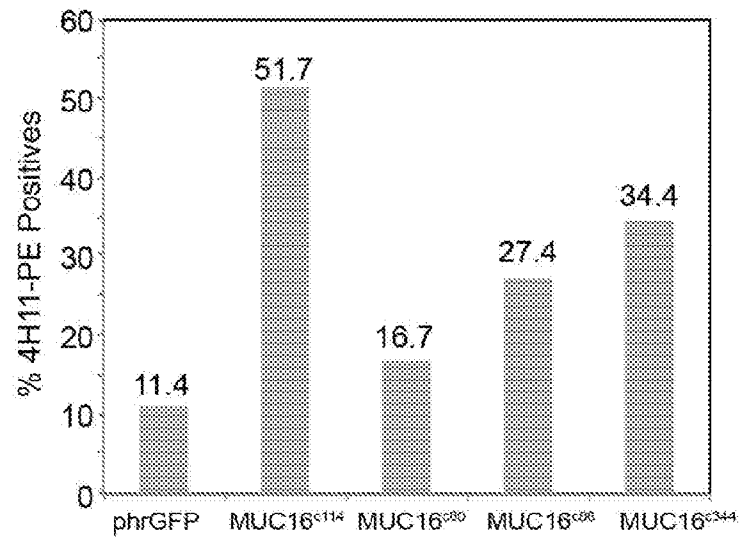
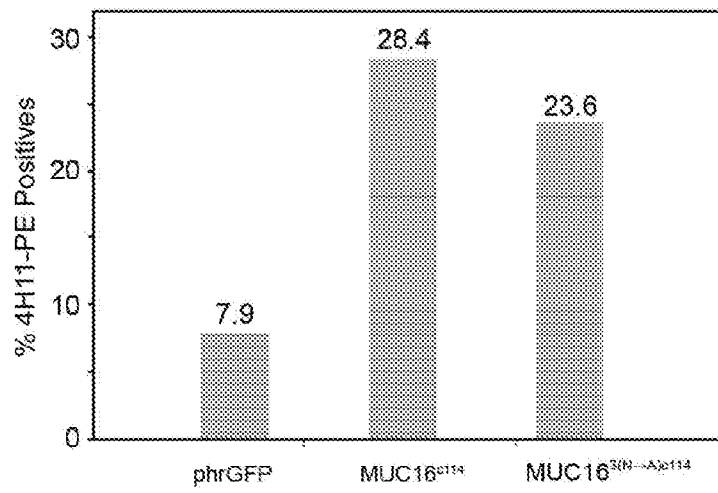
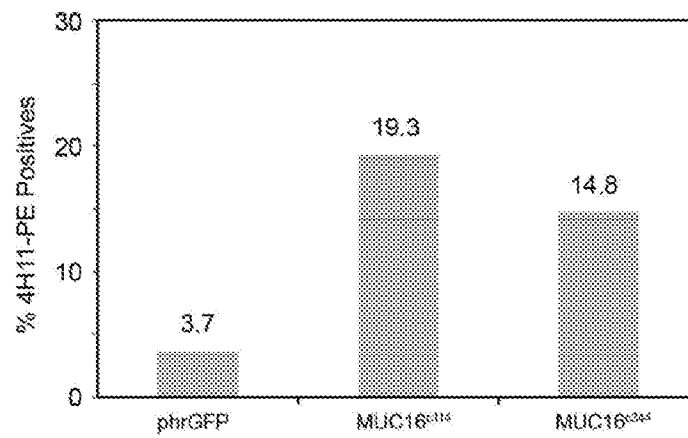
**Fig. 1G****Fig. 1H****Fig. 1I**

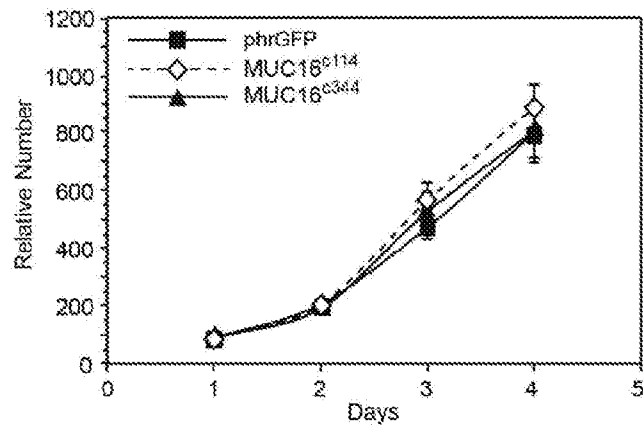
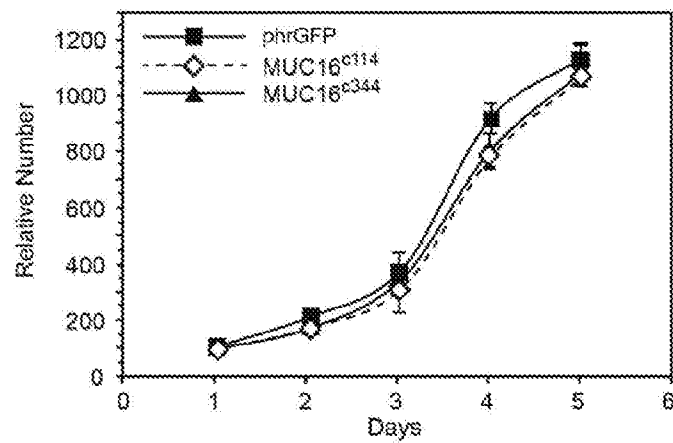
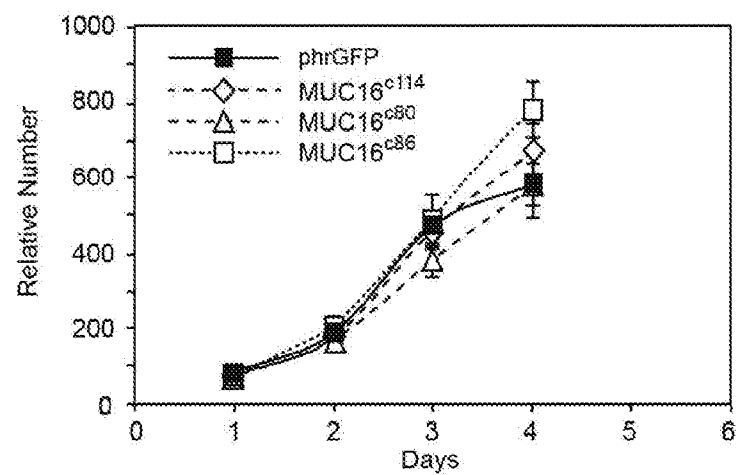
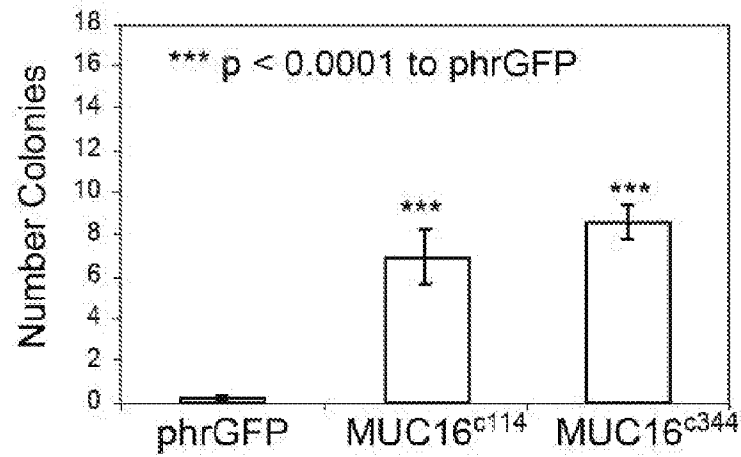
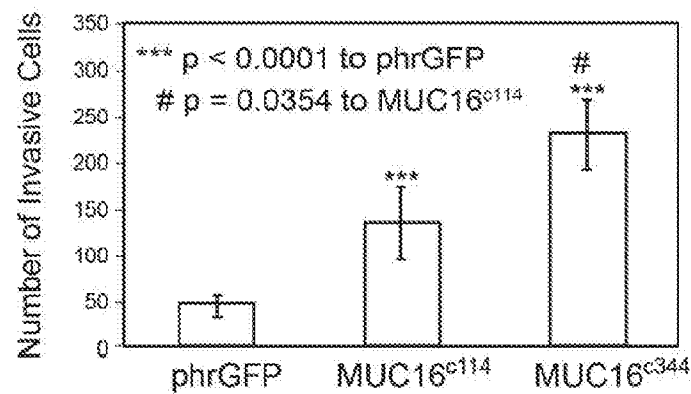
**Fig. 2A** 3T3 Transfectant *in vitro* Growth**Fig. 2B** A2780 Transfectant *in vitro* Growth**Fig. 2C** 3T3 Transfectant *in vitro* Growth

**Fig. 3A****Fig. 3B 3T3 Transfectant Matrigel Invasion****Fig. 3C**

| Symbol | 3T3 phr vs MUC16 <sup>c114</sup> |             | 3T3 phr vs MUC16 <sup>c344</sup> |             |
|--------|----------------------------------|-------------|----------------------------------|-------------|
|        | p value                          | Fold Change | p value                          | Fold Change |
| CXCL12 | 0.0008                           | 10.74       | 0.0071                           | 3.48        |
| NF2    | 0.0075                           | -1.31       | 0.0317                           | -1.28       |
| FN1    | 0.0166                           | -3.06       | 0.0377                           | -2.20       |
| CTBP1  | 0.0283                           | -1.76       | 0.2043                           | -1.36       |
| SMAD4  | 0.0324                           | -2.42       | 0.1759                           | -1.34       |
| CDH11  | 0.0790                           | 2.73        | 0.0057                           | 9.25        |
| MMP2   | 0.1634                           | 1.43        | 0.0445                           | 1.86        |
| MMP9   | 0.7675                           | 1.03        | 0.0315                           | 1.12        |

Fig. 3D

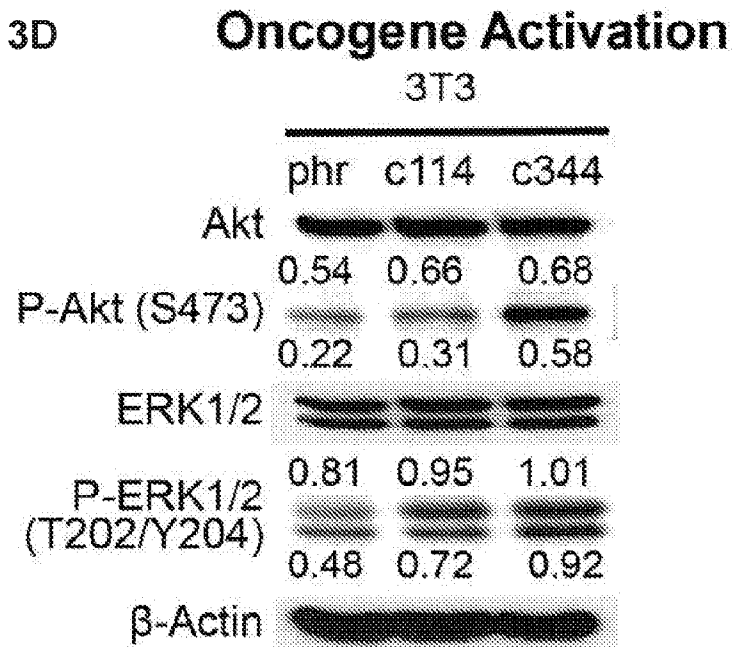


Fig. 3E

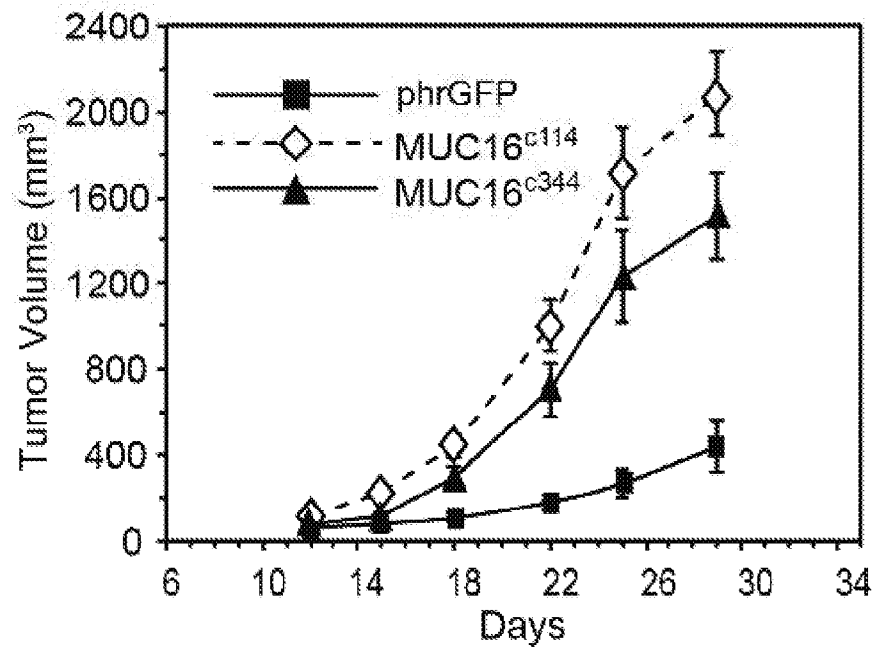


**Fig. 4A** A2780 Transfectant Matrigel Invasion

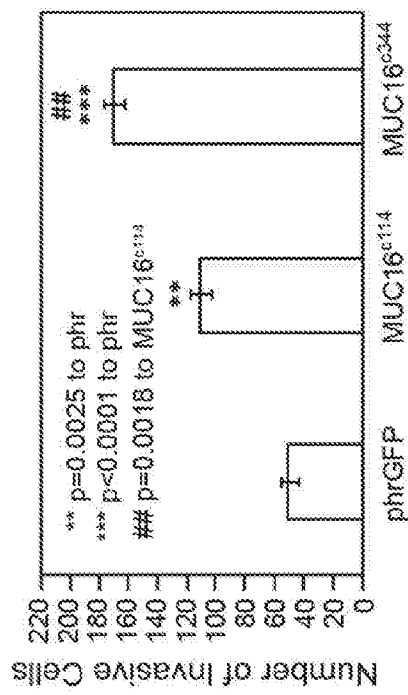
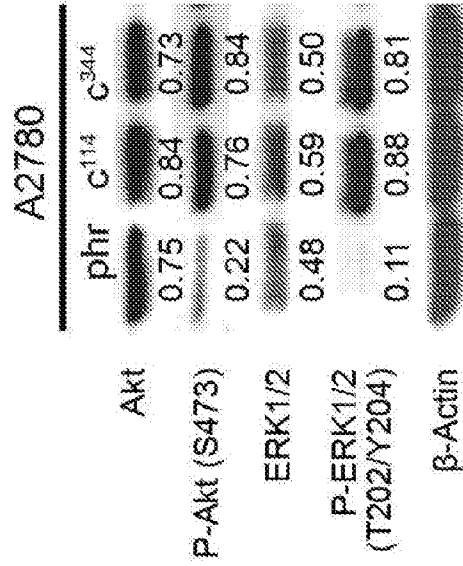


**Fig. 4B** Oncogene Activation



**A2780 Transfectant Xenograft Growth**

**Fig. 4C**

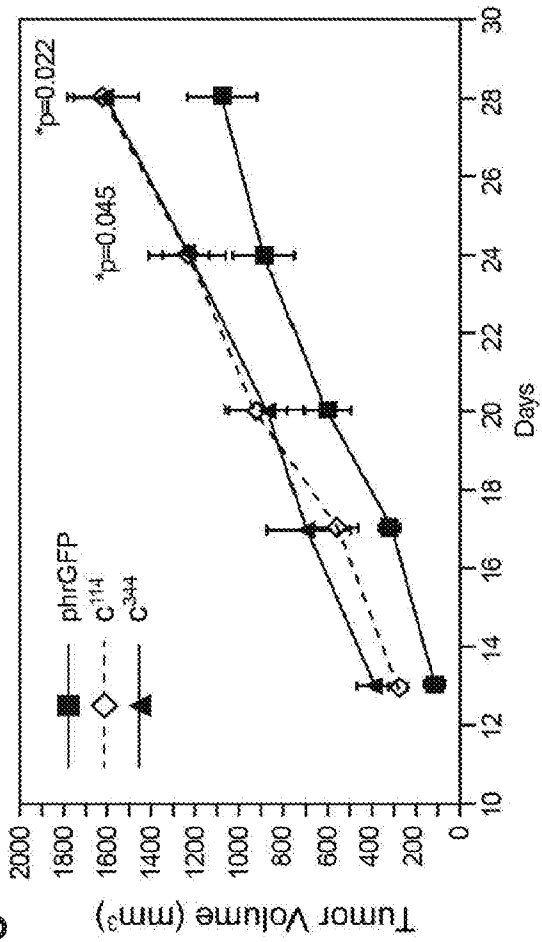


Fig. 5A

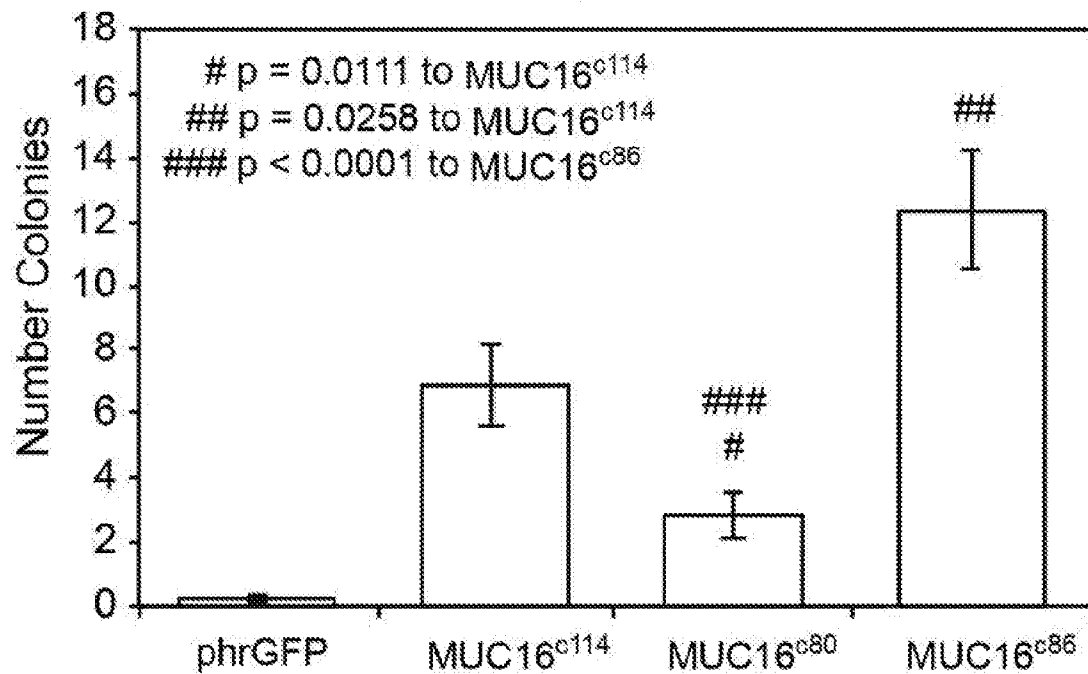
**3T3 Soft Agar Growth**

Fig. 5B

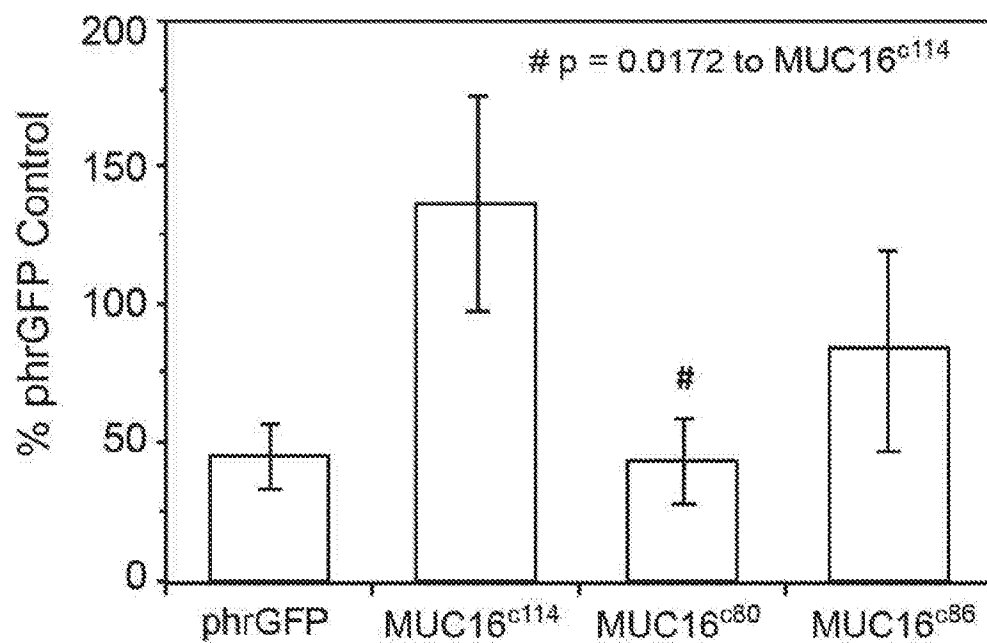
**3T3 Transfectant Matrigel Invasion**

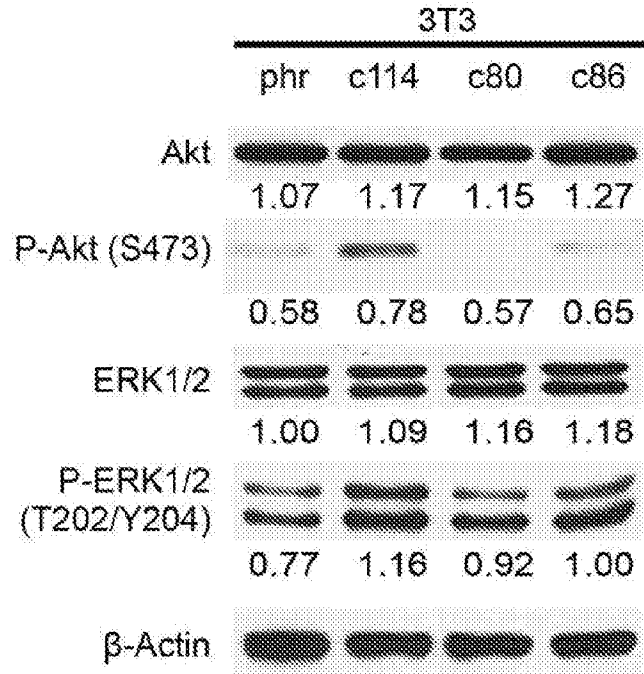
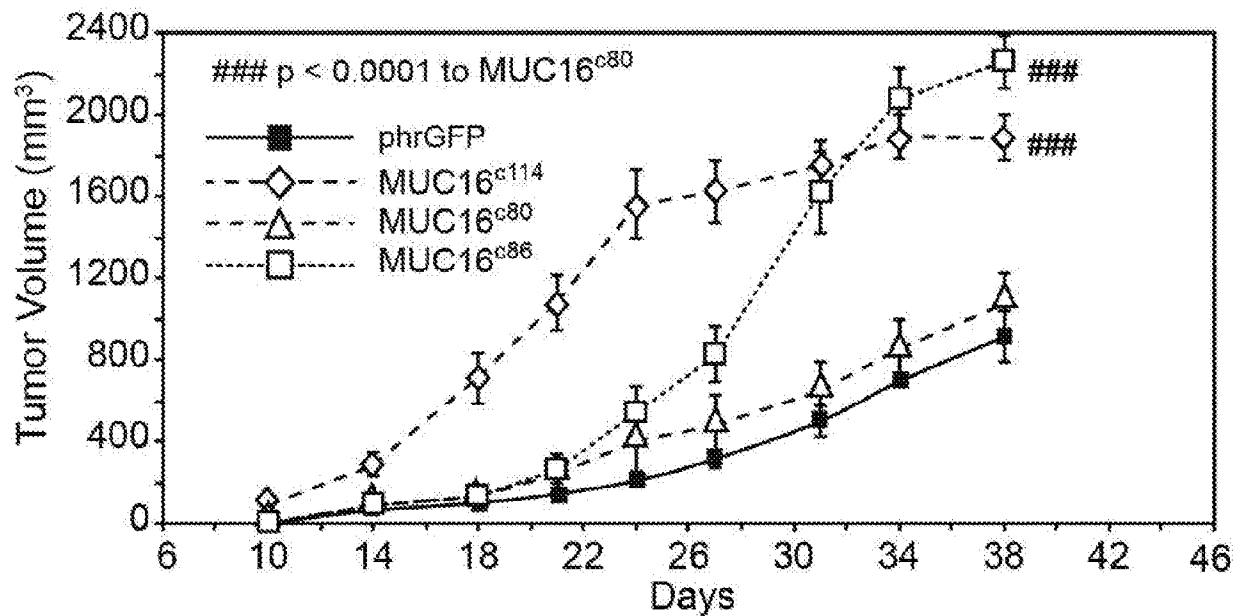
**Fig. 5C****Oncogene Activation****Fig. 5D****3T3 Transfectant Xenograft Growth**

**Fig. 6A** MUC16<sup>c114</sup> Construct

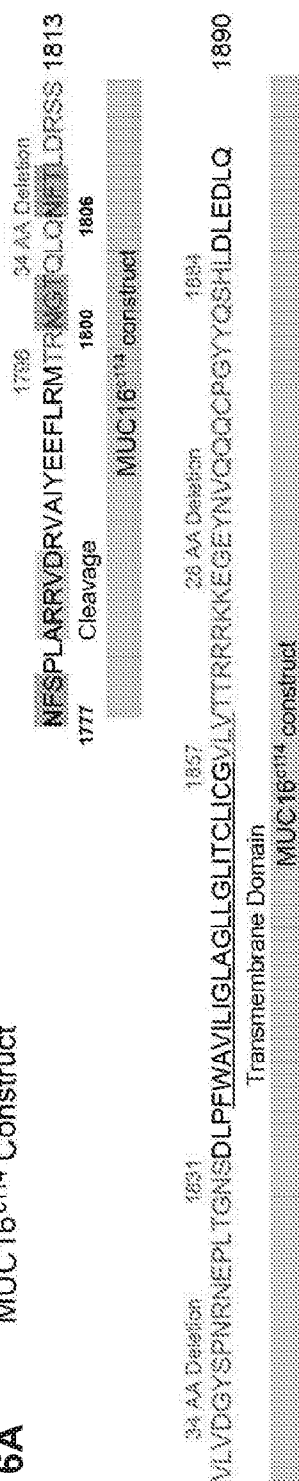


**Fig. 6B** MUC16<sup>c344</sup> Construct

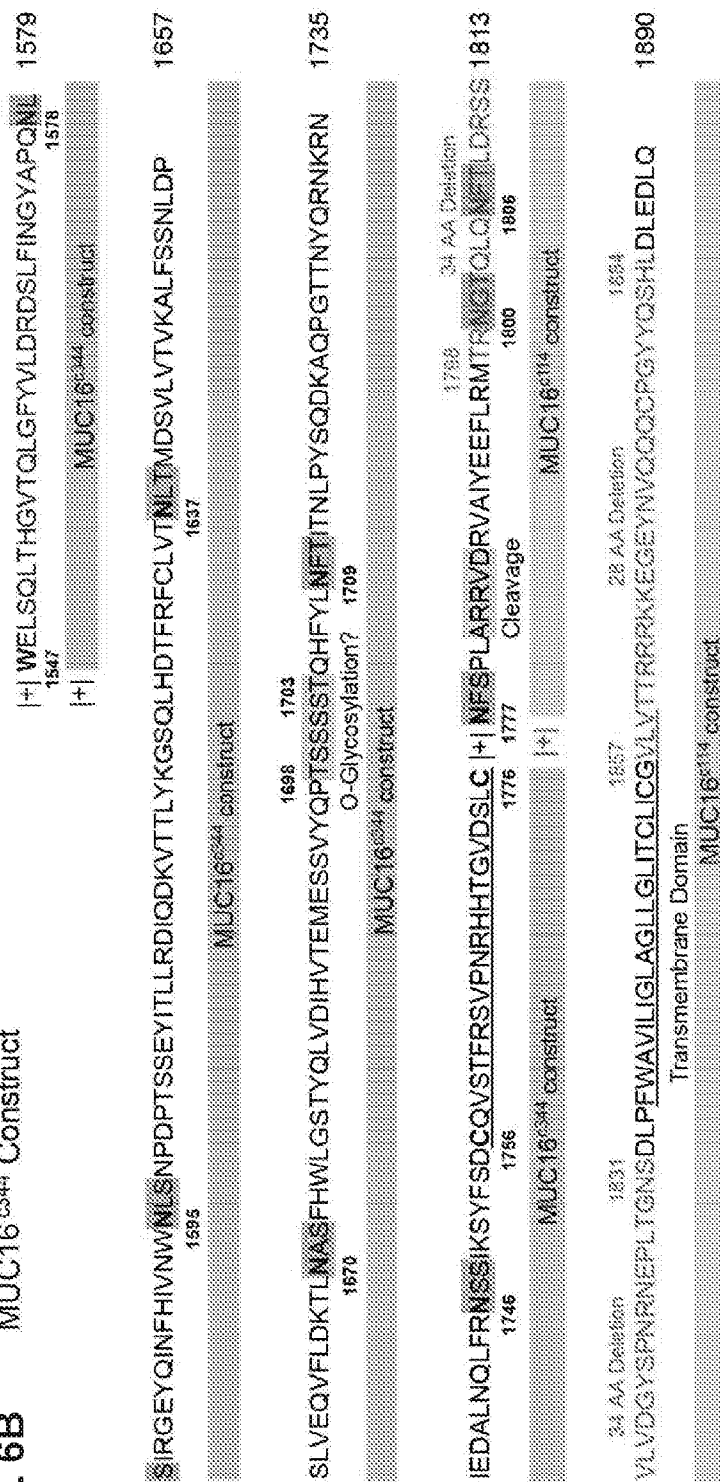


Fig. 7A

## 3T3 Transfectant Matrigel Invasion

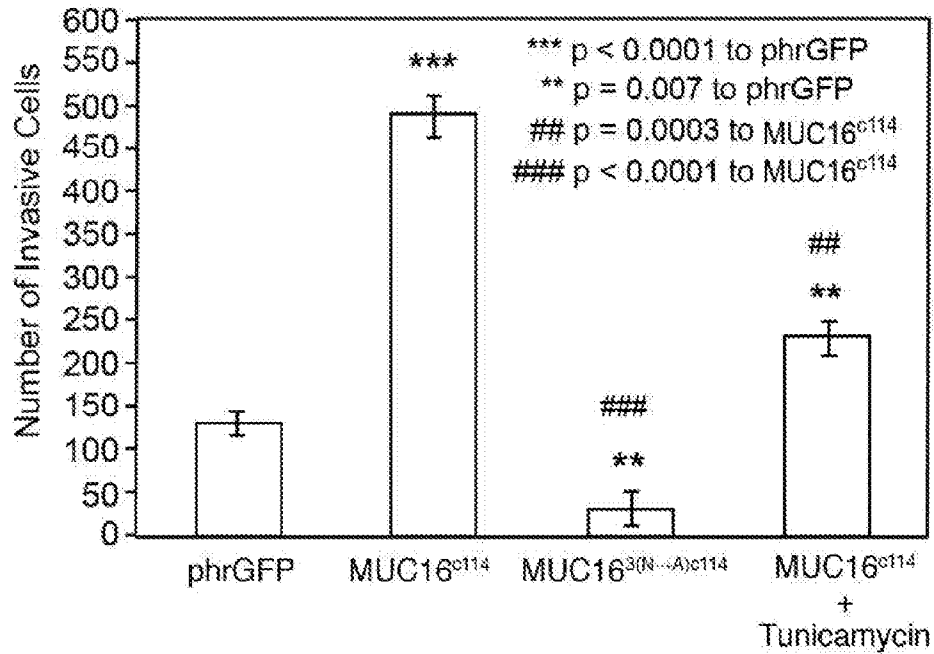


Fig. 7B

## 3T3 Transfectant Matrigel Invasion

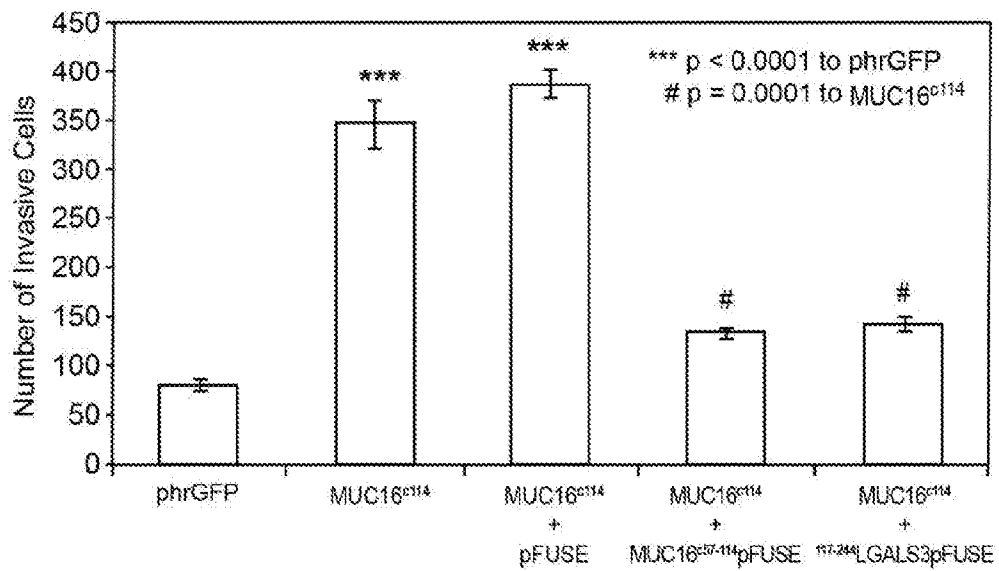


Fig. 7C

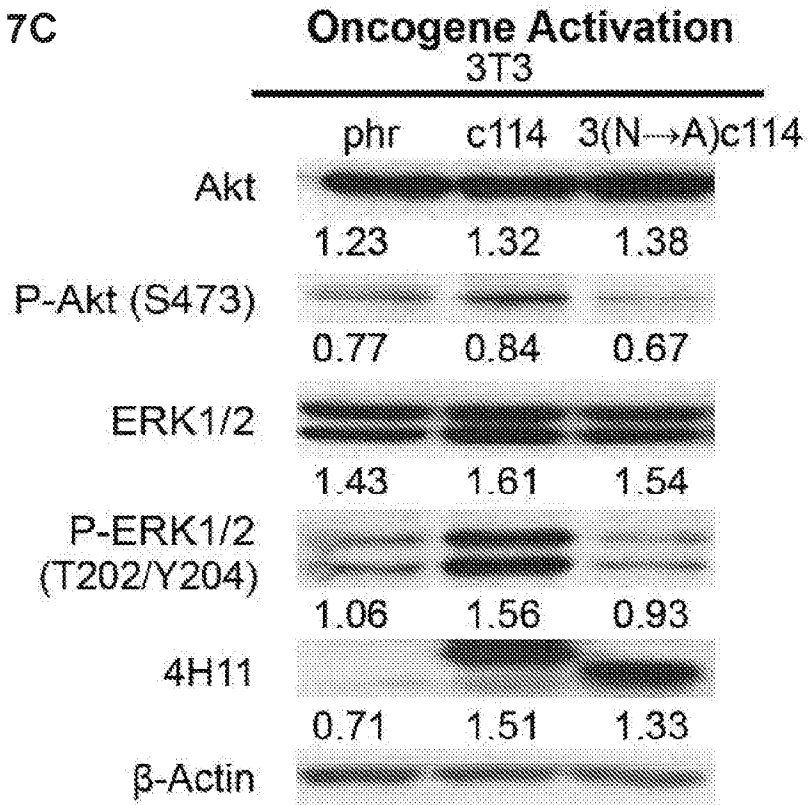


Fig. 7D

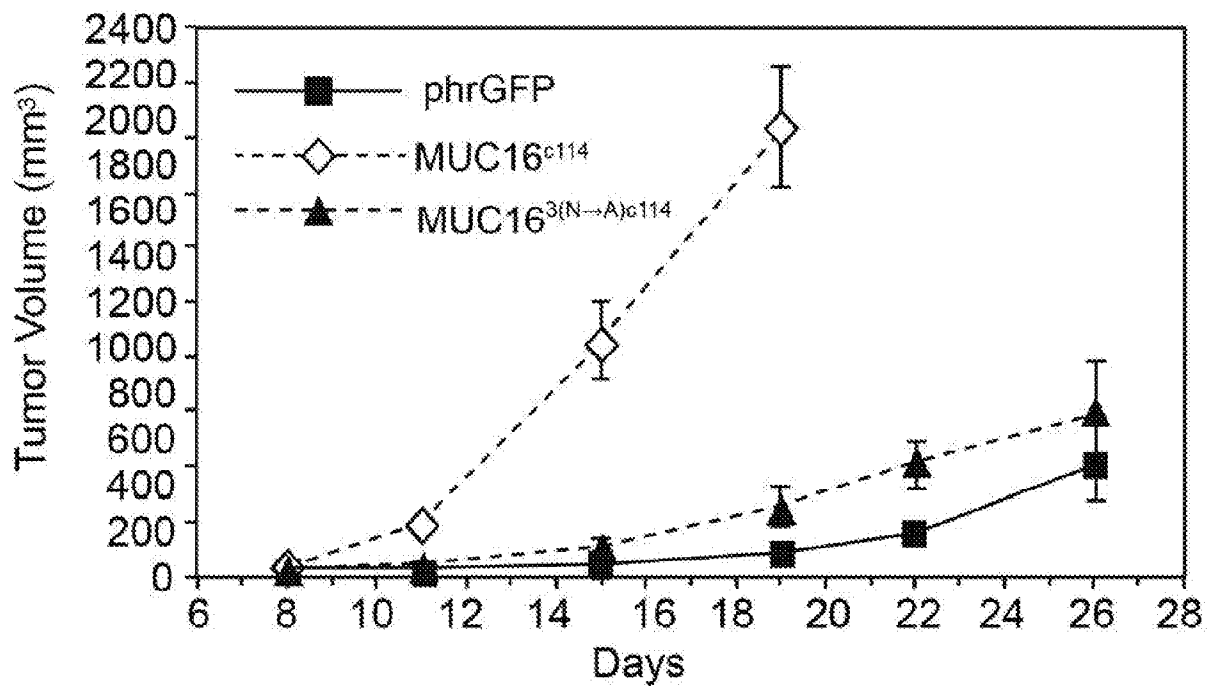
**3T3 Transfectant Xenograft Growth**

Fig. 7E

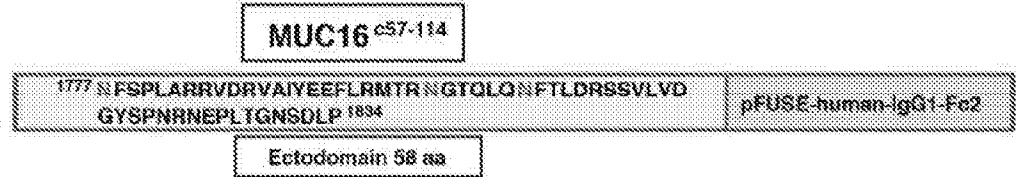
Ectodomain MUC16<sup>c57-114</sup>-construct (58 aa)-pFUSE-human-IgG1-Fc2

Fig. 7F

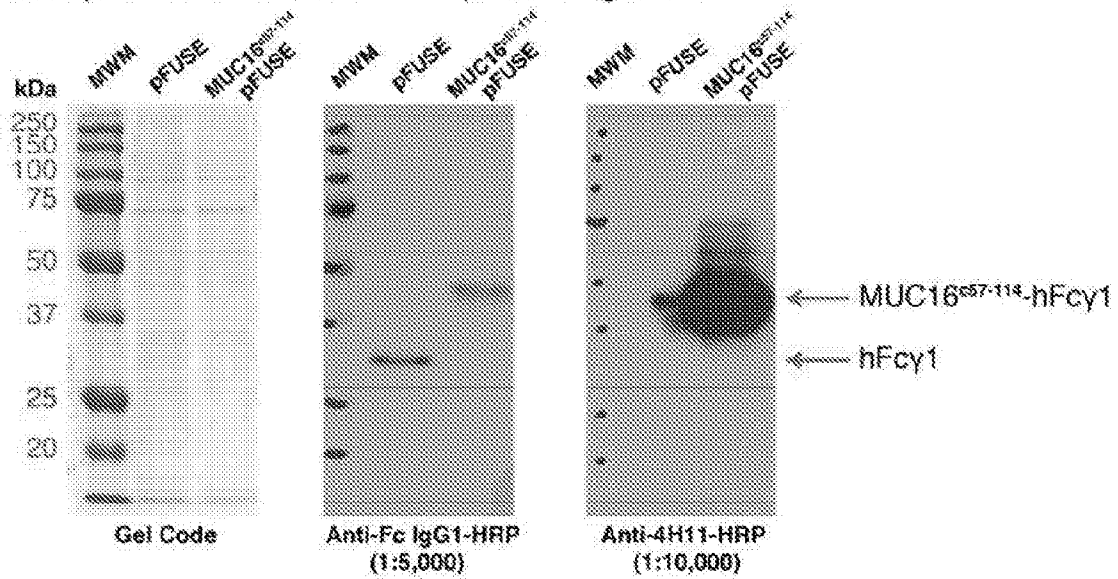
293 Expression of MUC16<sup>c57-114</sup>-pFUSE-hIgG1-Fc2

Fig. 7G

117-244 LGALS3 Sugar binding protein (128 aa)-pFUSE-human-IgG1-Fc2

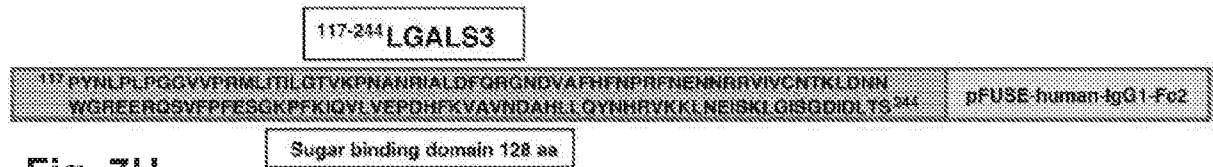
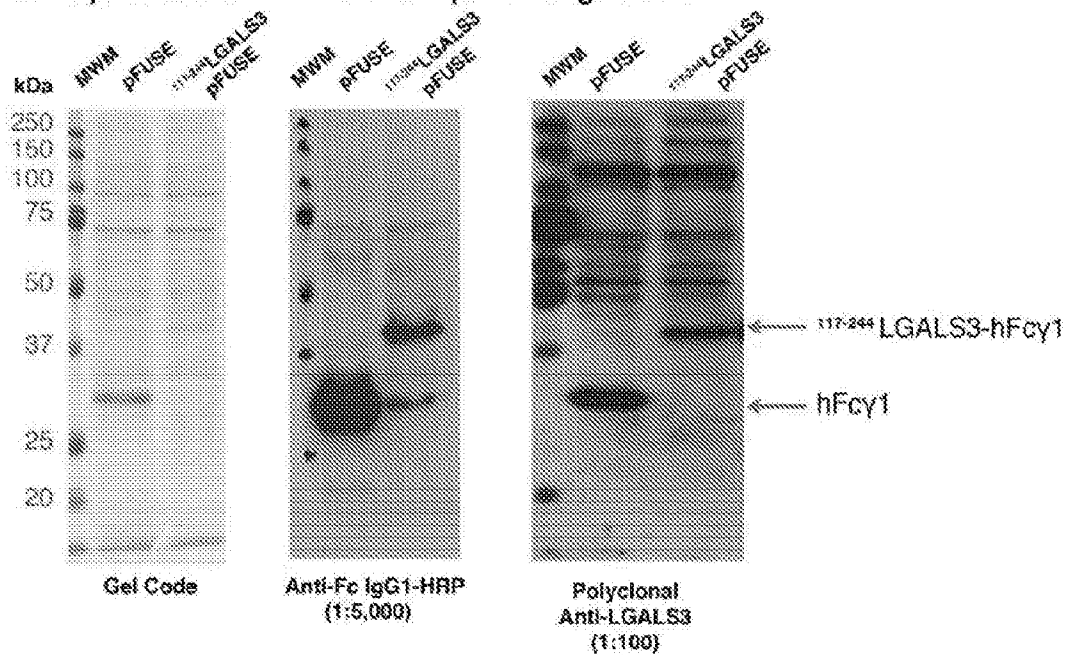


Fig. 7H

293 Expression of 117-244 LGALS3-pFUSE-hIgG1-Fc2



Ectodomain MUC16<sup>c57-c114</sup> construct (58 aa) pFUSE-hlgG1-Fc2

NFSPLARRVDRVAIYEEFLRMTRNGTQLQNFTLDRSSVLVDGYSPNRNEPLTGNSDLP - pFUSE-hlgG1-Fc2

<sup>117-244</sup>LGALS3 Sugar binding protein (128 aa)

PYNLPLPGGVVPRMLITLGTVPNANRIALDFQRCNDVAFHFNPRFNENNRVIVCNTKLDNNWGRE

ERQSVFPFESCKPFKIQVLVEPDHFKVAVNDAHLLQYNHRVKKLNEISKLGISGDIDLTS - pFUSE-hlgG1-Fc2

**Fig. 8**



Fig. 9B

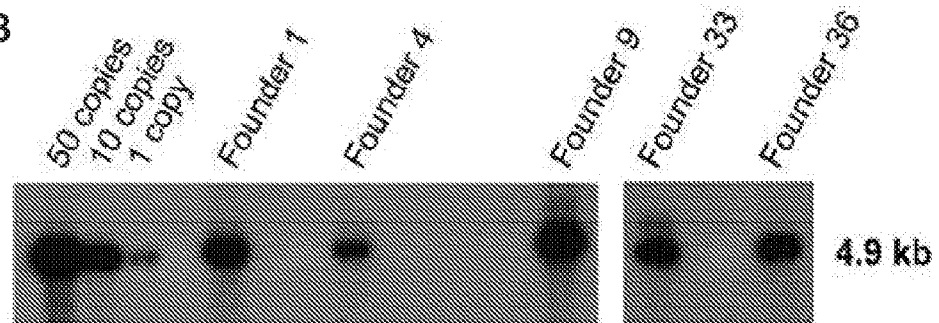


Fig. 9C

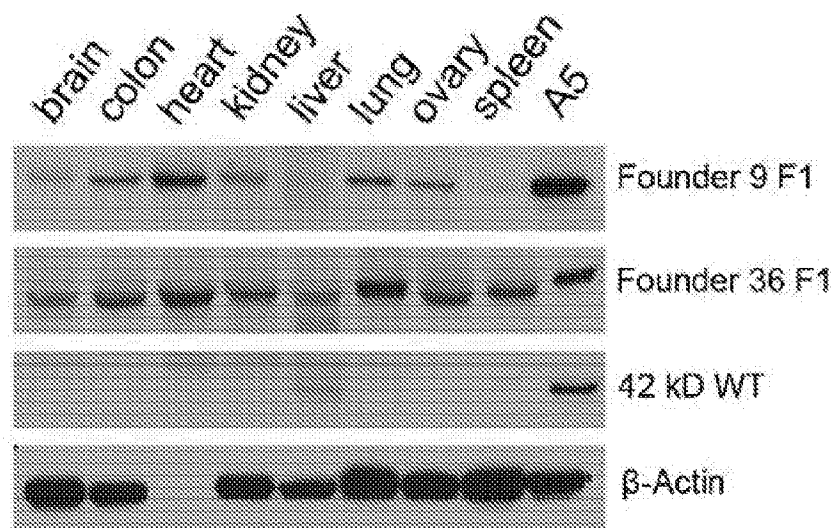


Fig. 9D

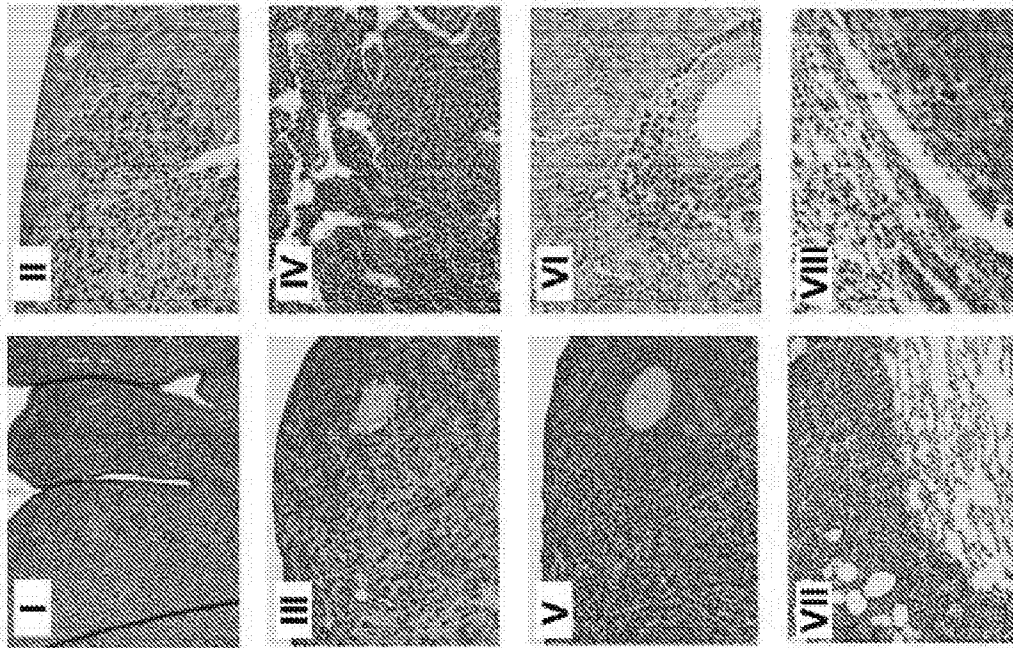
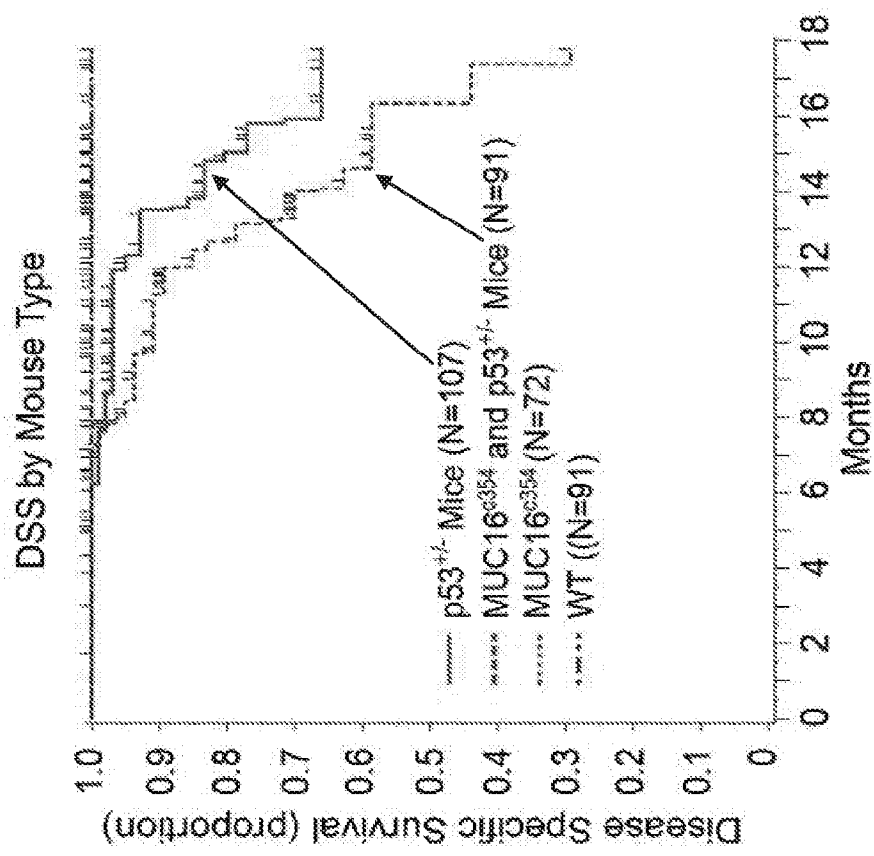


Fig. 9E



Transgenic (Tg) / WT

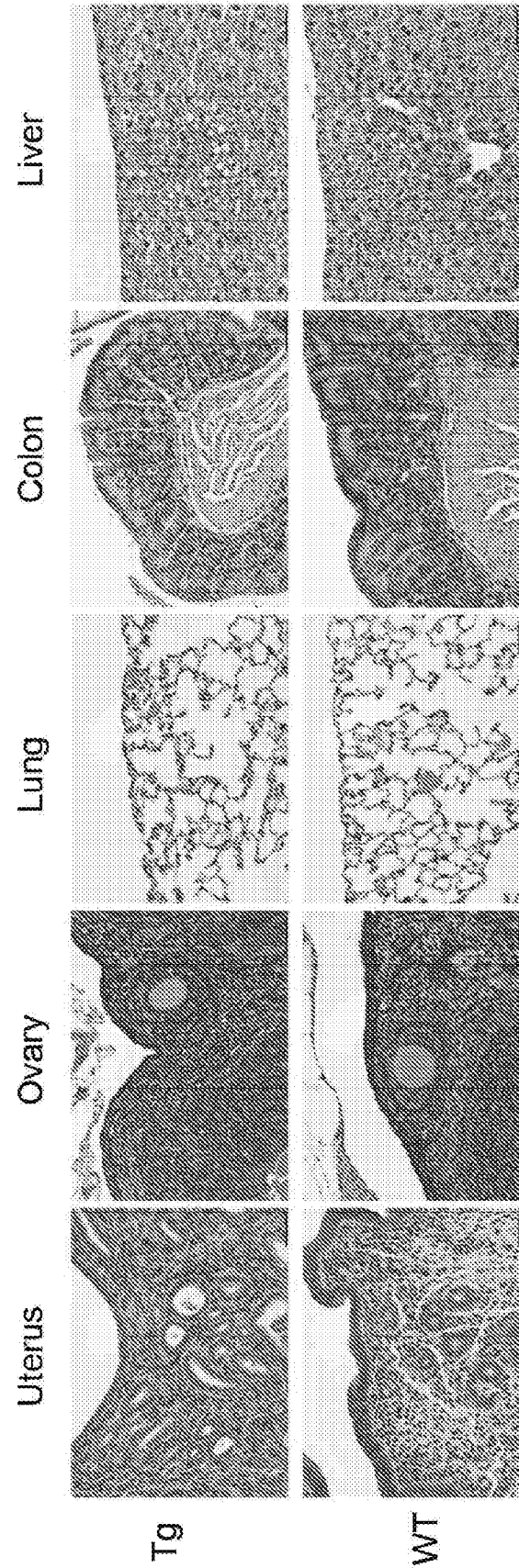


Fig. 10

Fig. 11A MUC16 Transcript Number

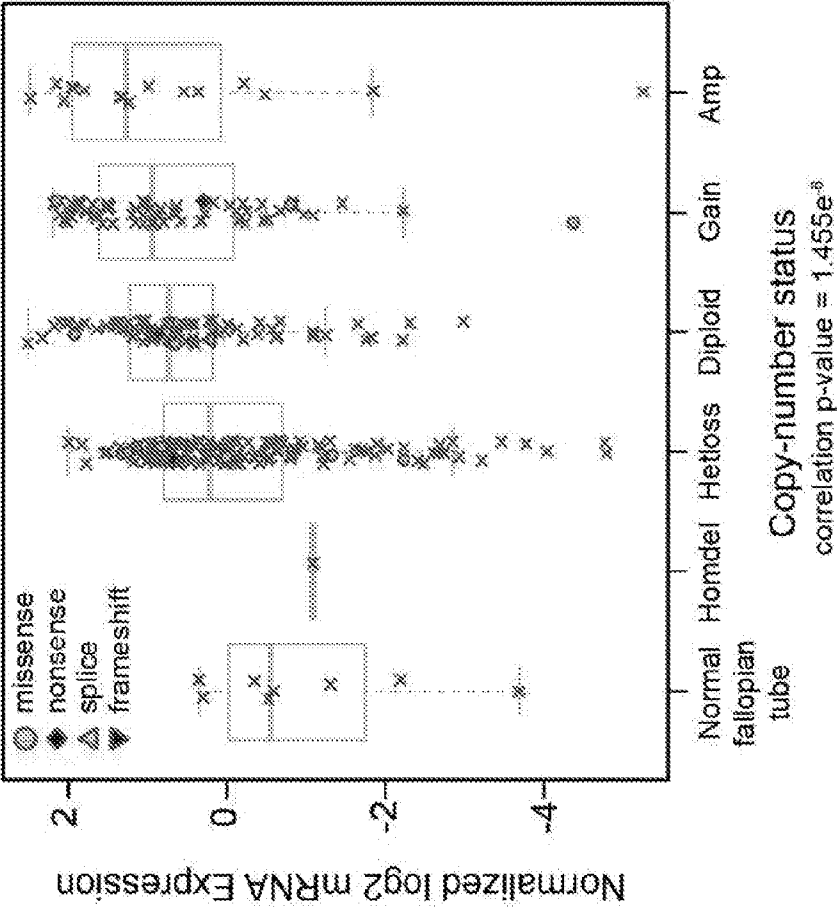


Fig. 11B

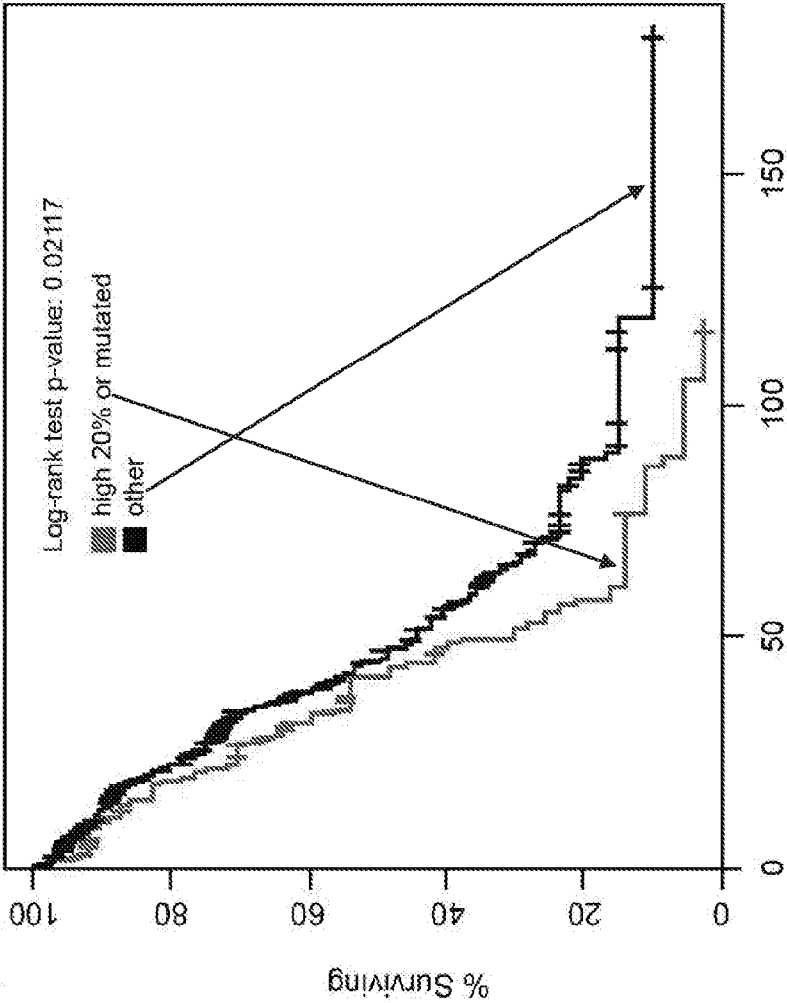
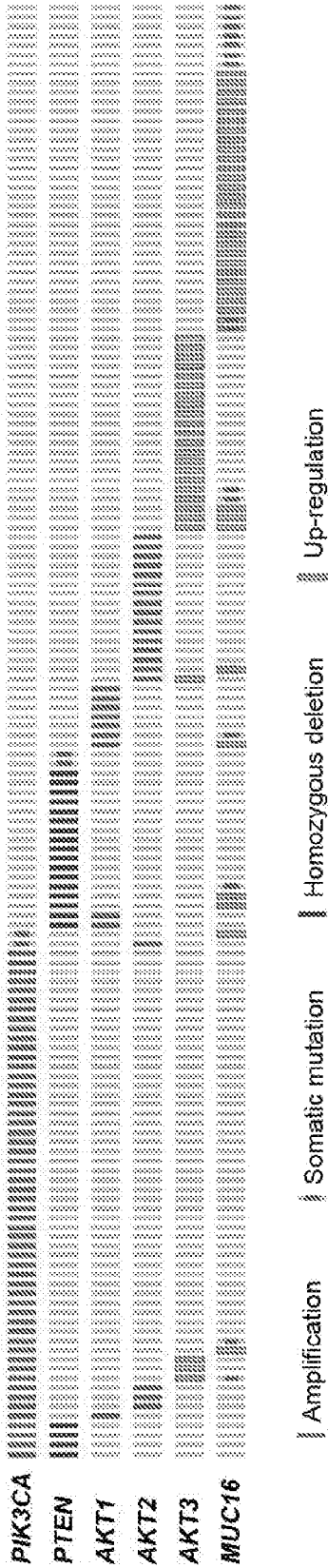


Fig. 11C



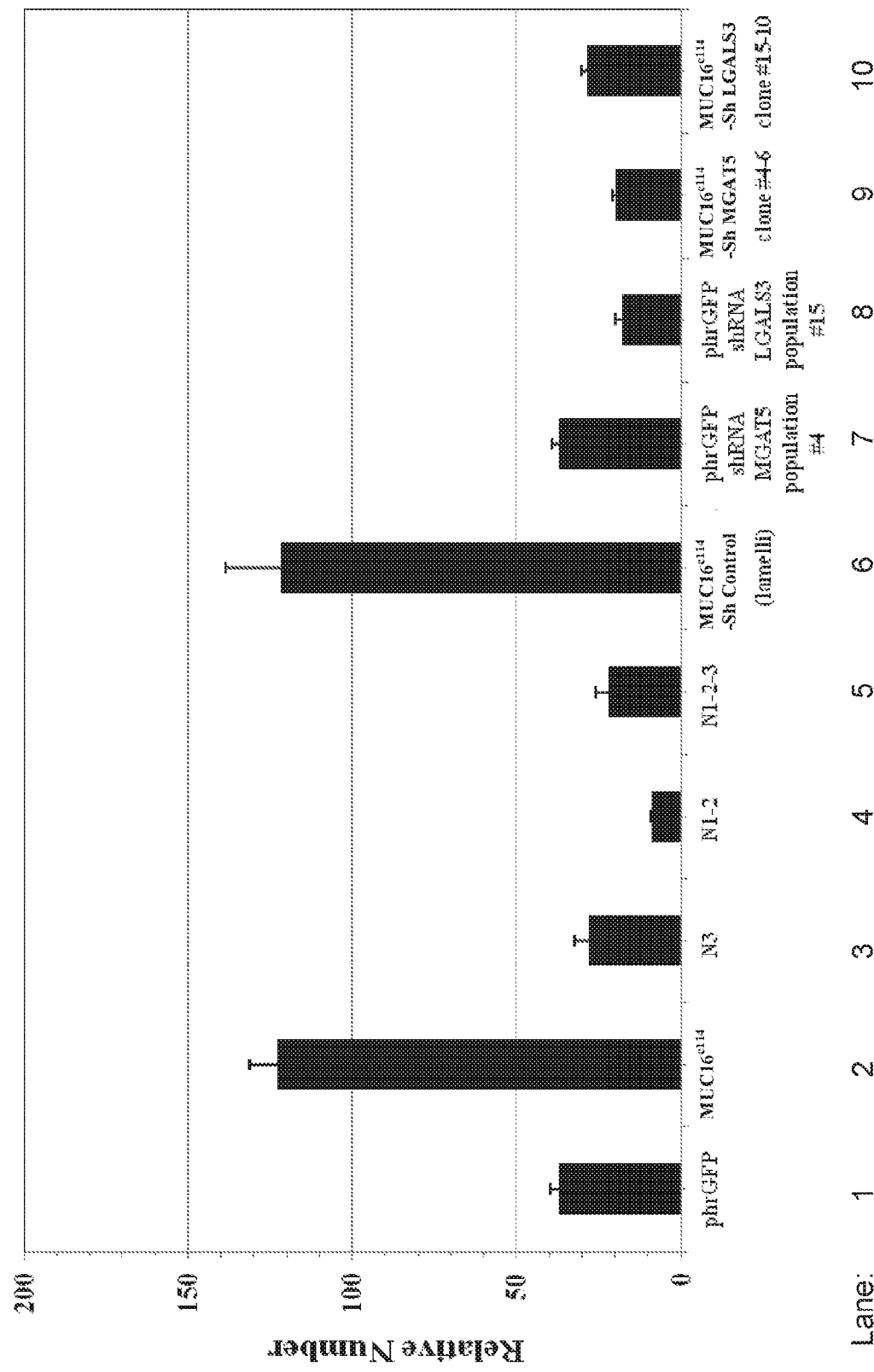


Fig. 12A

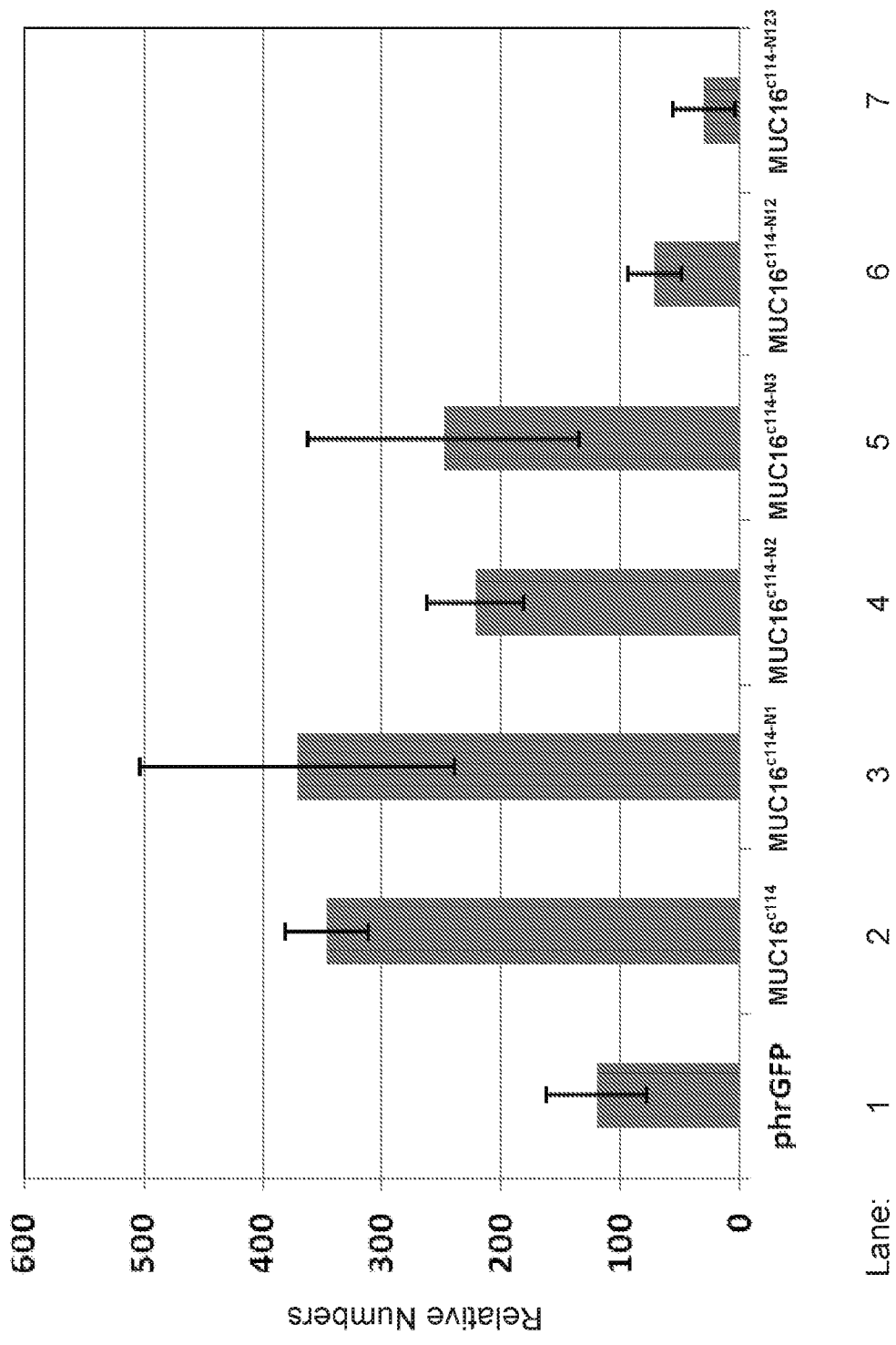


Fig. 12B

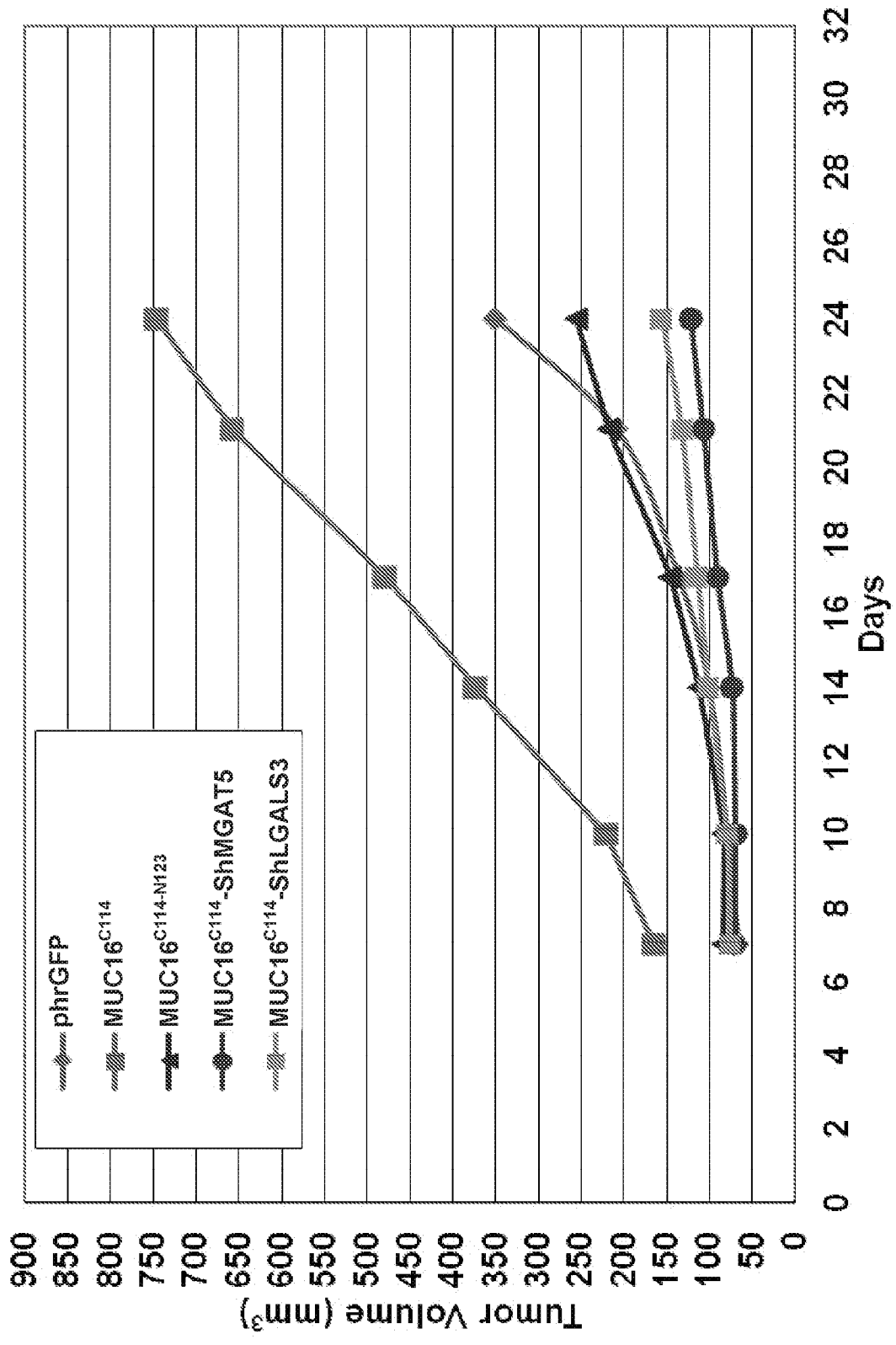


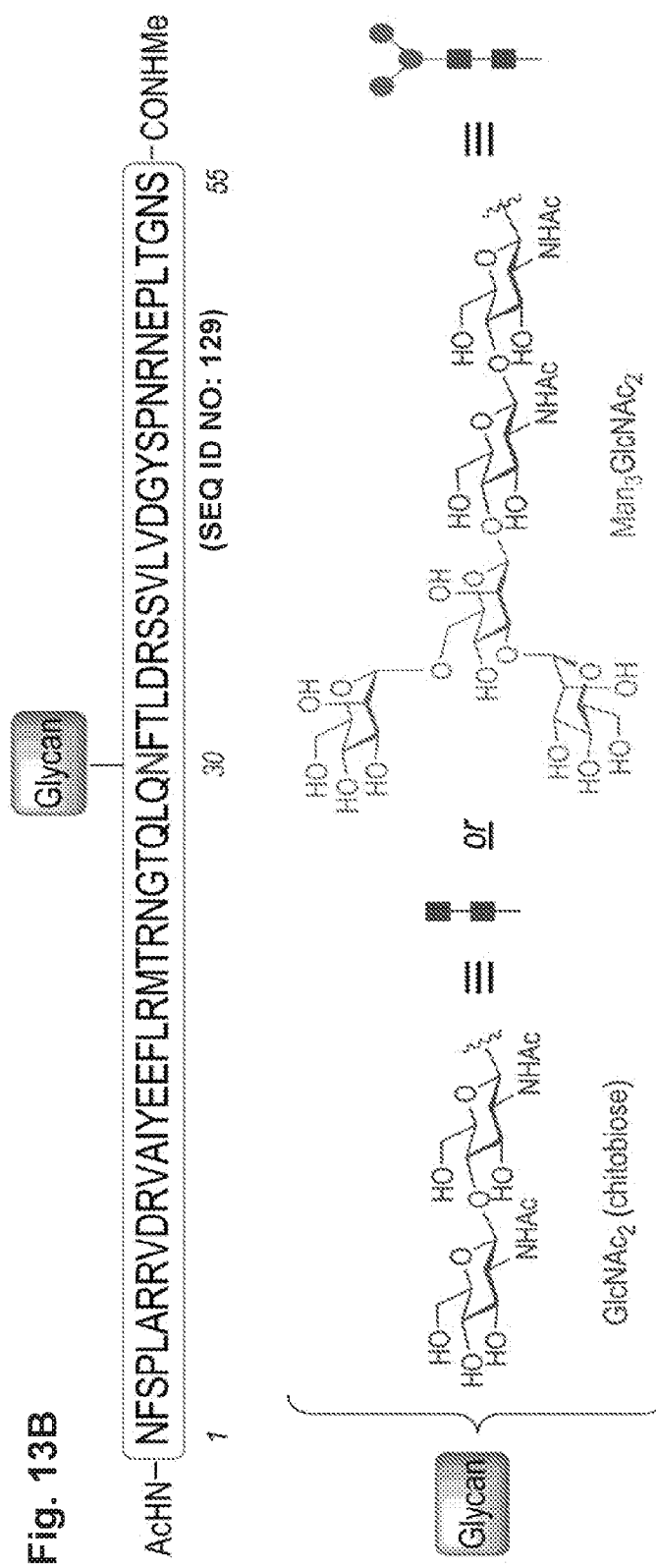
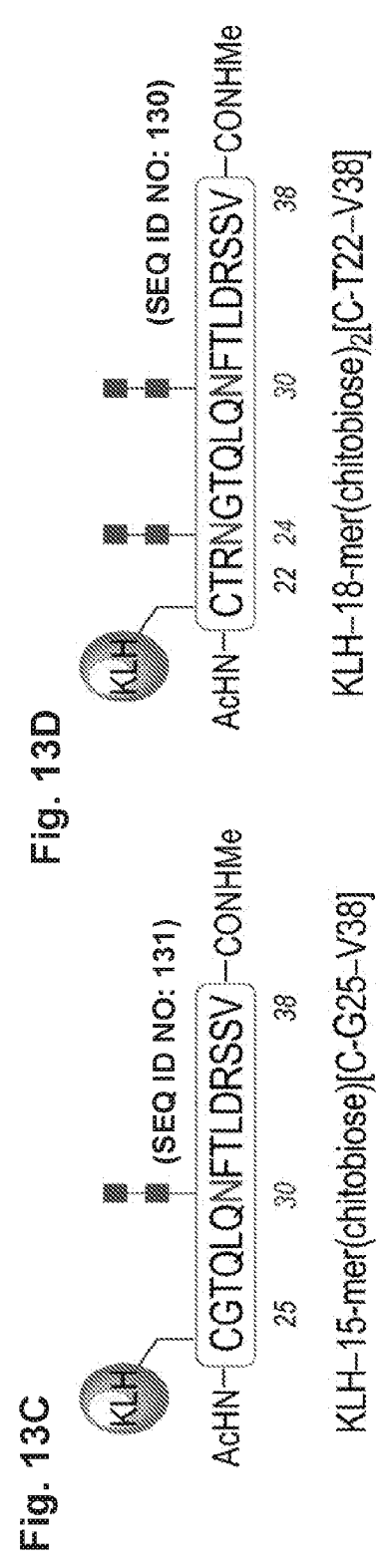
Fig. 12C

| Observed<br>m/z | Charge<br>state | Structural assignment                                                                                    | Mass<br>[M+Na] <sup>+</sup> | Intensity/Charge<br>state | Glycan<br>% |
|-----------------|-----------------|----------------------------------------------------------------------------------------------------------|-----------------------------|---------------------------|-------------|
| 1142            | 1               | M2N2F<br>               | 1142                        | 1.02E+03                  | 32.47%      |
| 1170            | 1               | M3N2<br>                | 1171                        | 3.56E+02                  | 11.33%      |
| 1346            | 1               | M3N2F<br>               | 1345                        | 4.18E+02                  | 13.30%      |
| 1376            | 1               | M4N2<br>                | 1376                        | 1.91E+02                  | 6.08%       |
| 1580            | 1               | M5N2<br>                | 1580                        | 3.78E+02                  | 12.03%      |
| 1784            | 1               | M6N2<br>               | 1784                        | 8.68E+01                  | 2.76%       |
| 1988            | 1               | M7N2<br>              | 1988                        | 3.70E+01                  | 1.18%       |
| 1134            | 2               | Gal2N2M3N2F<br>       | 2244                        | 1.91E+02                  | 6.06%       |
| 1228            | 2               | NeuAc1Gal2N2M3N2<br>  | 2431                        | 1.50E+02                  | 4.76%       |
| 1316            | 2               | NeuAc1Gal2N2M3N2F<br> | 2605                        | 1.65E+02                  | 5.25%       |
| 1408            | 2               | NeuAc2Gal2N2M3N2<br>  | 2792                        | 7.70E+01                  | 2.45%       |
| 1496            | 2               | NeuAc1Gal2N2M3N2F<br> | 2966                        | 7.30E+01                  | 2.32%       |
|                 |                 |                                                                                                          | Total                       | 3.14E+03                  | 100.00%     |

**Figure Legend**

|                                                                                     |                                                                                     |                                                                                     |
|-------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------|
|  |  |  |
|  |  |                                                                                     |

**Fig. 13A**



| ID     |      | 15 vs 15s Peptides                              |      |      |                      |                    |                                                                      |      |      |                      |                    | 18 vs 18s Peptides                              |          |      |      |                      |                                                                      |          |      |      |                      | Study<br>ck<br>EL<br>IS<br>Av<br>by |                    |
|--------|------|-------------------------------------------------|------|------|----------------------|--------------------|----------------------------------------------------------------------|------|------|----------------------|--------------------|-------------------------------------------------|----------|------|------|----------------------|----------------------------------------------------------------------|----------|------|------|----------------------|-------------------------------------|--------------------|
|        |      | ELISA on 12/24/13<br>Sups from time of freezing |      |      |                      |                    | ELISA during fusion screen<br>(ELISA performed on<br>Different Days) |      |      |                      |                    | ELISA on 12/24/13<br>Sups from time of freezing |          |      |      |                      | ELISA during fusion screen<br>(ELISA performed on<br>Different Days) |          |      |      |                      |                                     |                    |
|        |      | Dilution                                        | 15   | 15S  | 15S /<br>Ratio<br>TB | Fold<br>over<br>TB | Dilution                                                             | 15   | 15S  | 15S /<br>Ratio<br>TB | Fold<br>over<br>TB | Agree                                           | Dilution | 18   | 18S  | 18S /<br>Ratio<br>TB | Fold<br>over<br>TB                                                   | Dilution | 18   | 18S  | 18S /<br>Ratio<br>TB |                                     | Fold<br>over<br>TB |
| 4H11   | neat | 0.02                                            | 0.02 | 0.02 | 0.02                 | neat               | 0.02                                                                 | 0.02 | 0.02 | 0.02                 | neat               | 0.02                                            | 0.02     | 0.02 | 0.02 | 0.02                 | 0.02                                                                 | 0.02     | 0.02 | 0.02 | 0.02                 | 0.02                                | ELISA              |
| 9B11   | neat | 0.02                                            | 0.02 | 0.02 | 0.02                 | neat               | 0.02                                                                 | 0.02 | 0.02 | 0.02                 | neat               | 0.02                                            | 0.02     | 0.02 | 0.02 | 0.02                 | 0.02                                                                 | 0.02     | 0.02 | 0.02 | 0.02                 | 0.02                                | Agree              |
| TB     | 1:10 | 0.72                                            | 0.29 | 0.40 | 0.40                 | 1:1000             | 0.81                                                                 | 0.77 | 0.95 | 0.95                 | 1:1000             | 0.81                                            | 0.77     | 0.95 | 0.95 | 0.95                 | 0.95                                                                 | 0.95     | 0.95 | 0.95 | 0.95                 | 0.95                                | Agree              |
| H01B05 | 400  | 0.34                                            | 0.47 | 1.39 | 1.46                 | 400                | 0.55                                                                 | 0.52 | 0.95 | 1.12                 | No                 | 400                                             | 0.47     | 0.60 | 1.28 | 1.34                 | 400                                                                  | 0.43     | 0.78 | 1.81 | 1.29                 | Yes                                 |                    |
| H02E01 | 400  | 0.42                                            | 0.37 | 0.88 | 2.10                 | 400                | 0.31                                                                 | 0.22 | 0.71 | 0.83                 | No                 | 400                                             | 0.37     | 0.47 | 1.27 | 1.34                 | 400                                                                  | 0.28     | 0.51 | 1.82 | 1.39                 | Yes                                 |                    |
| H04G02 | 80   | 0.38                                            | 0.05 | 0.13 | 0.33                 | 40                 | 0.87                                                                 | 0.51 | 0.59 | 0.94                 | Yes                | 80                                              | 0.34     | 0.33 | 0.97 | 1.02                 | 80                                                                   | 0.37     | 0.61 | 1.65 | 1.24                 | No                                  |                    |
| H07B12 | 400  | 0.44                                            | 0.25 | 0.57 | 1.42                 | 200                | 0.77                                                                 | 0.80 | 1.04 | 1.22                 | ?                  | 400                                             | 0.33     | 0.51 | 1.55 | 1.63                 | 400                                                                  | 0.53     | 0.92 | 1.74 | 1.24                 | ?                                   |                    |
| H09A06 | 100  | 0.96                                            | 0.31 | 0.32 | 0.81                 | 100                | 0.61                                                                 | 0.40 | 0.66 | 1.18                 | ?                  | 100                                             | 1.00     | 0.92 | 0.92 | 0.97                 | 400                                                                  | 0.25     | 0.37 | 1.48 | 1.56                 | No                                  |                    |
| H09G09 | 400  | 0.51                                            | 0.27 | 0.53 | 1.32                 | 200                | 0.47                                                                 | 0.31 | 0.66 | 1.77                 | ?                  | 400                                             | 0.50     | 0.62 | 1.24 | 1.31                 | 400                                                                  | 0.24     | 0.52 | 2.17 | 1.37                 | Yes                                 |                    |
| H09H07 | 200  | 0.71                                            | 0.21 | 0.30 | 0.74                 | 200                | 0.51                                                                 | 0.30 | 0.59 | 1.06                 | ?                  | 200                                             | 0.62     | 0.69 | 1.11 | 1.17                 | 200                                                                  | 0.40     | 0.51 | 1.28 | 1.34                 | ?                                   |                    |
| H10C06 | 400  | 0.41                                            | 0.26 | 0.63 | 1.59                 | 100                | 0.62                                                                 | 0.49 | 0.79 | 1.42                 | Yes                | 400                                             | 0.38     | 0.56 | 1.47 | 1.55                 | 400                                                                  | 0.25     | 0.37 | 1.48 | 1.56                 | Yes                                 |                    |
| H12H08 | 200  | 0.68                                            | 0.26 | 0.38 | 0.96                 | 200                | 0.37                                                                 | 0.18 | 0.49 | 1.31                 | No                 | 200                                             | 0.68     | 0.76 | 1.12 | 1.18                 | 400                                                                  | 0.17     | 0.36 | 2.12 | 1.34                 | ?                                   |                    |
| H13A07 | 400  | 0.26                                            | 0.14 | 0.54 | 1.35                 | 200                | 0.51                                                                 | 0.53 | 1.04 | 1.22                 | Yes                | 400                                             | 0.23     | 0.34 | 1.48 | 1.56                 | 400                                                                  | 0.23     | 0.53 | 2.40 | 1.64                 | Yes                                 |                    |
| H15A08 | 400  | 0.59                                            | 0.25 | 0.42 | 1.06                 | 400                | 0.33                                                                 | 0.19 | 0.58 | 1.55                 | No                 | 400                                             | 0.60     | 0.67 | 1.12 | 1.18                 | 400                                                                  | 0.30     | 0.59 | 1.97 | 1.24                 | ?                                   |                    |
| H16C05 | 400  | 0.62                                            | 0.23 | 0.37 | 0.93                 | 100                | 0.58                                                                 | 0.45 | 0.78 | 1.40                 | No                 | 400                                             | 0.63     | 0.68 | 1.08 | 1.14                 | 400                                                                  | 0.24     | 0.36 | 1.50 | 1.58                 | No                                  |                    |
| H18C06 | 400  | 0.44                                            | 0.14 | 0.32 | 0.80                 | 200                | 0.70                                                                 | 0.29 | 0.41 | 1.11                 | ?                  | 400                                             | 0.45     | 0.55 | 1.22 | 1.29                 | 400                                                                  | 0.39     | 0.64 | 1.64 | 1.17                 | ?                                   |                    |
| H19C11 | 400  | 0.30                                            | 0.10 | 0.33 | 0.83                 | 400                | 0.16                                                                 | 0.19 | 1.19 | 1.39                 | No                 | 400                                             | 0.20     | 0.36 | 1.80 | 1.89                 | 400                                                                  | 0.38     | 0.47 | 1.24 | 0.88                 | No                                  |                    |
| H21C03 | 200  | 0.73                                            | 0.36 | 0.49 | 1.23                 | 200                | 0.40                                                                 | 0.33 | 0.83 | 1.49                 | Yes                | 200                                             | 0.7      | 0.78 | 1.11 | 1.17                 | 200                                                                  | 0.34     | 0.48 | 1.41 | 1.49                 | ?                                   |                    |
| H21G12 | 50   | 0.99                                            | 0.32 | 0.32 | 0.81                 | 50                 | 0.62                                                                 | 0.30 | 0.48 | 0.87                 | Yes                | 50                                              | 0.92     | 0.88 | 0.96 | 1.01                 | 50                                                                   | 0.31     | 0.46 | 1.48 | 1.56                 | No                                  |                    |
| H05H10 | 50   | 0.30                                            | 0.32 | 1.07 | 2.67                 | 50                 | 0.16                                                                 | 0.25 | 1.56 | 4.19                 | Yes                | 50                                              | 0.28     | 0.33 | 1.18 | 1.24                 | 50                                                                   | 0.13     | 0.26 | 2.00 | 1.26                 | Yes                                 |                    |
| H16H08 | 20   | 0.12                                            | 0.16 | 1.33 | 1.13                 | 20                 | 0.11                                                                 | 0.26 | 2.16 | 3.81                 | Yes                | 20                                              | 0.06     | 0.14 | 2.13 | 2.46                 | 20                                                                   | 0.13     | 0.33 | 2.54 | 1.97                 | Yes                                 |                    |
| H21F08 | 20   | 0.20                                            | 0.28 | 1.40 | 1.50                 | 40                 | 0.14                                                                 | 0.34 | 2.43 | 3.91                 | Yes                | 20                                              | 0.09     | 0.25 | 2.78 | 2.92                 | 20                                                                   | 0.17     | 0.43 | 2.53 | 1.91                 | Yes                                 |                    |

Fig. 14

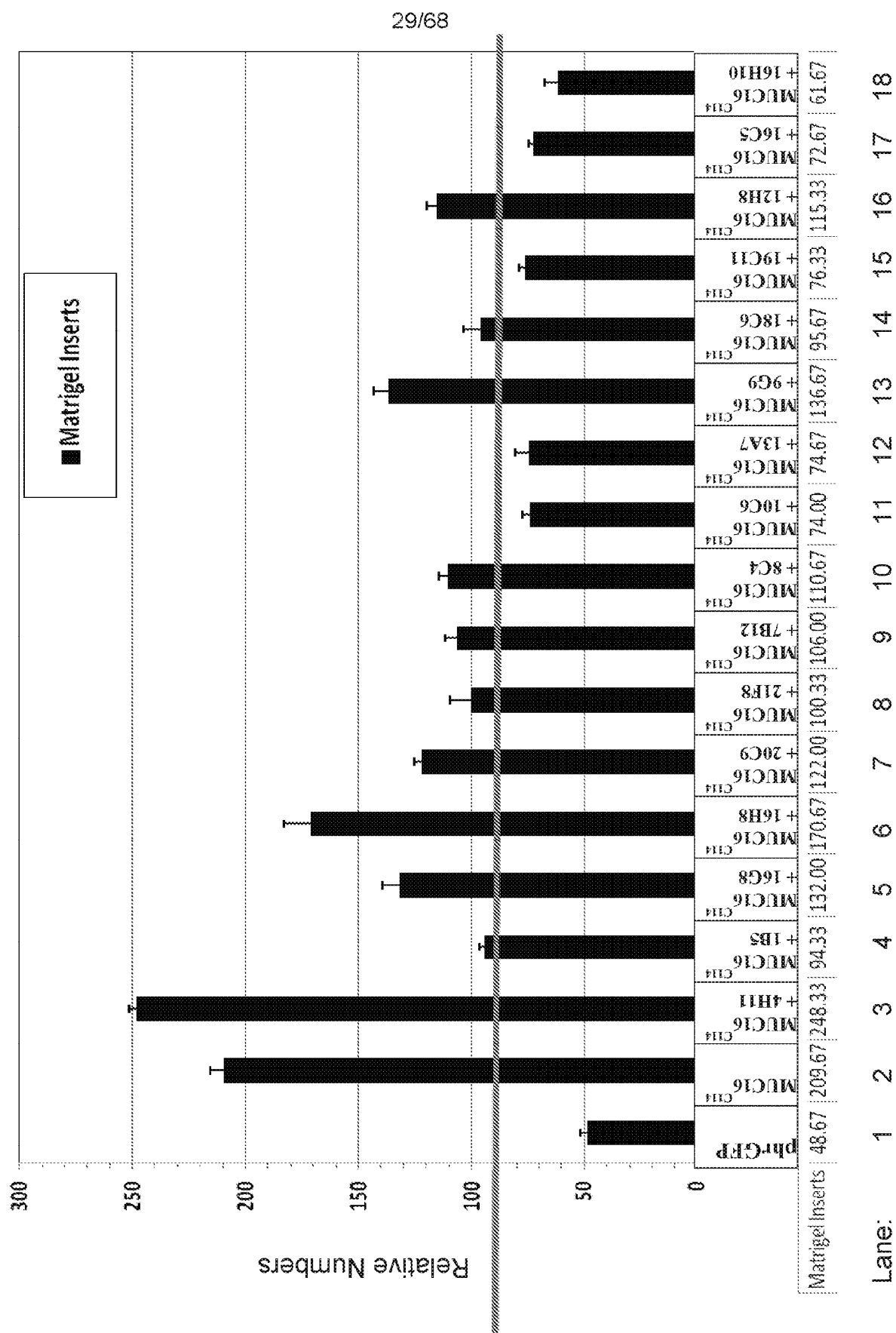


Fig. 15A

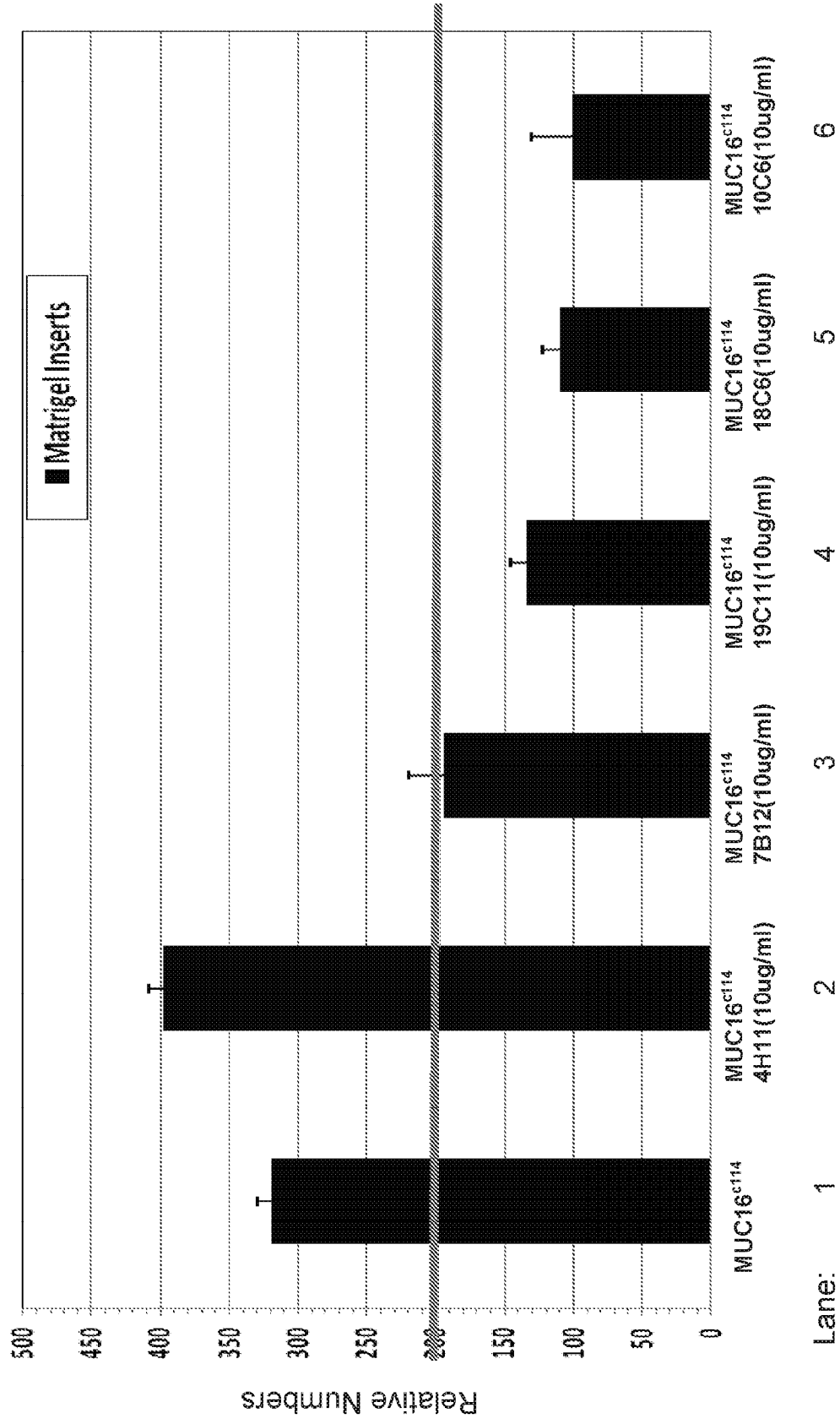


Fig. 15B

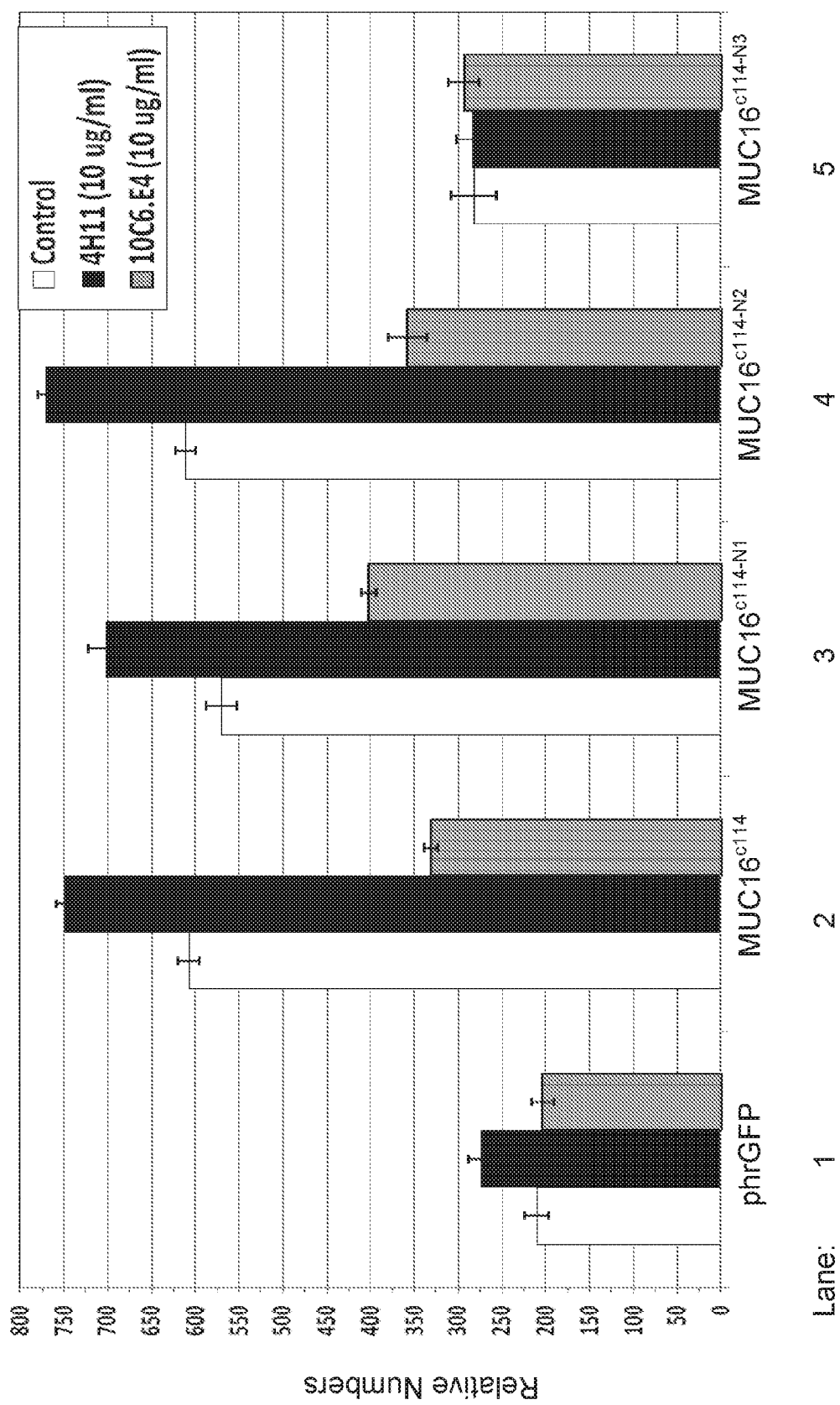


Fig. 16A

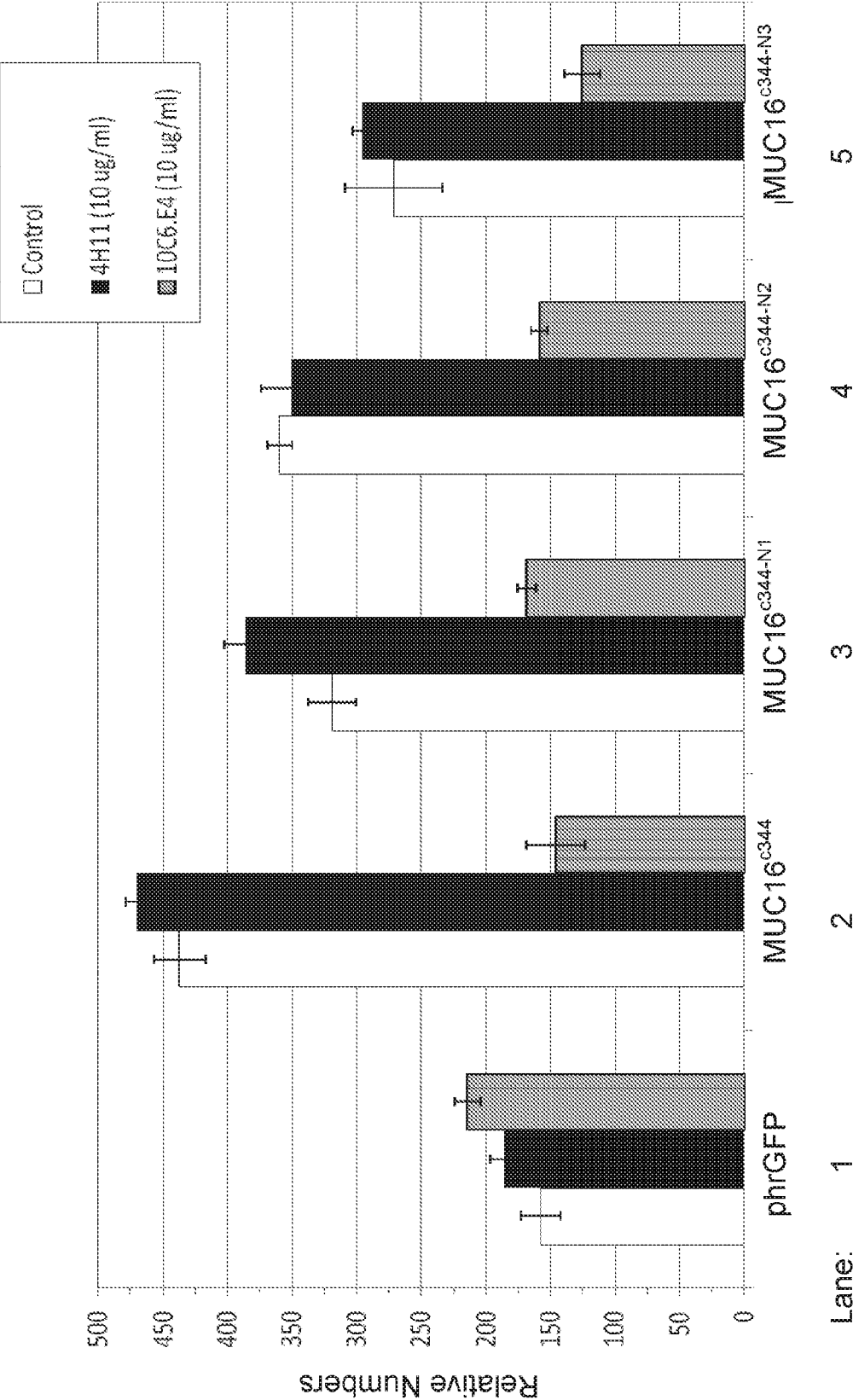


Fig. 16B

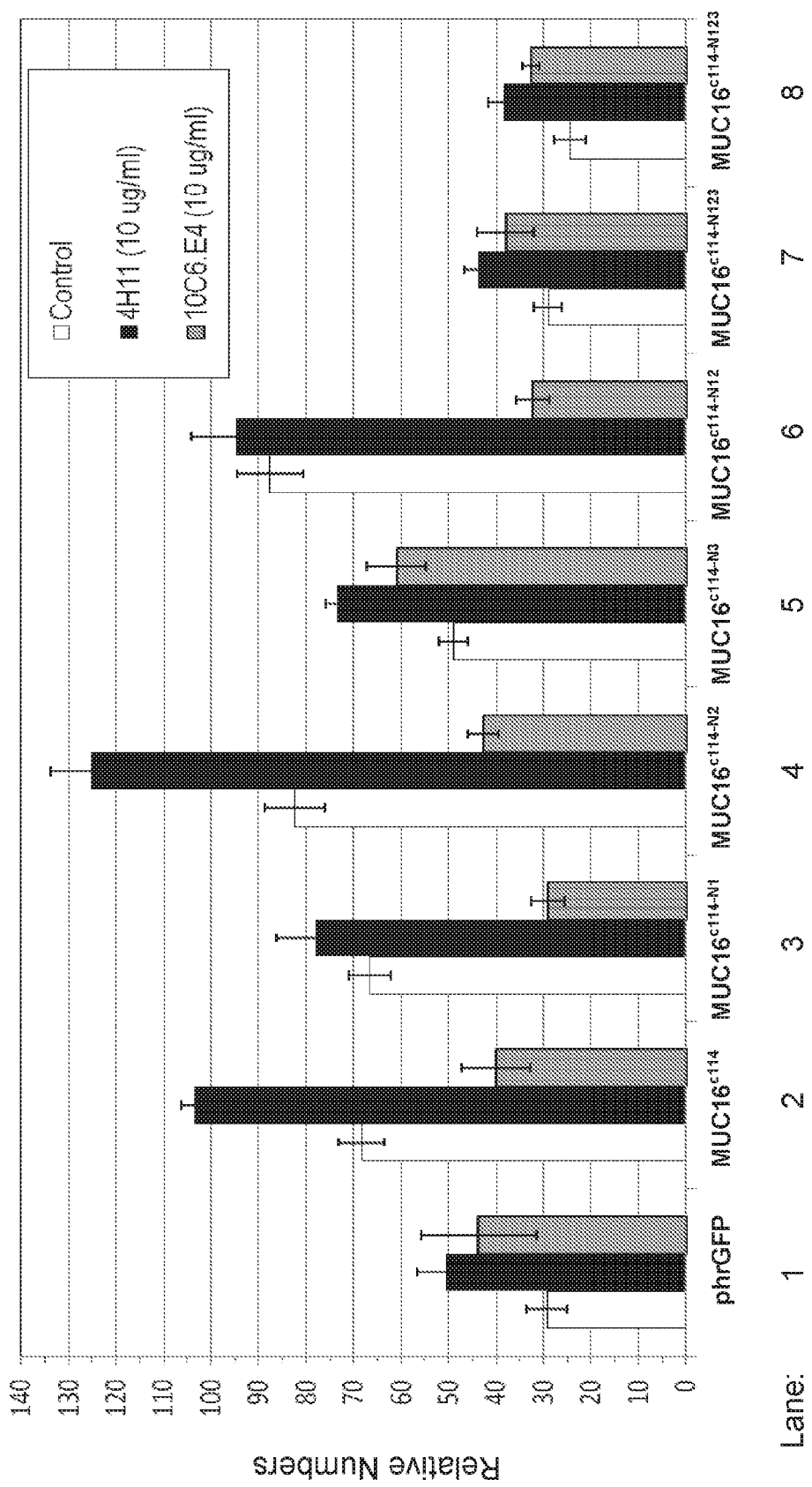


Fig. 16C

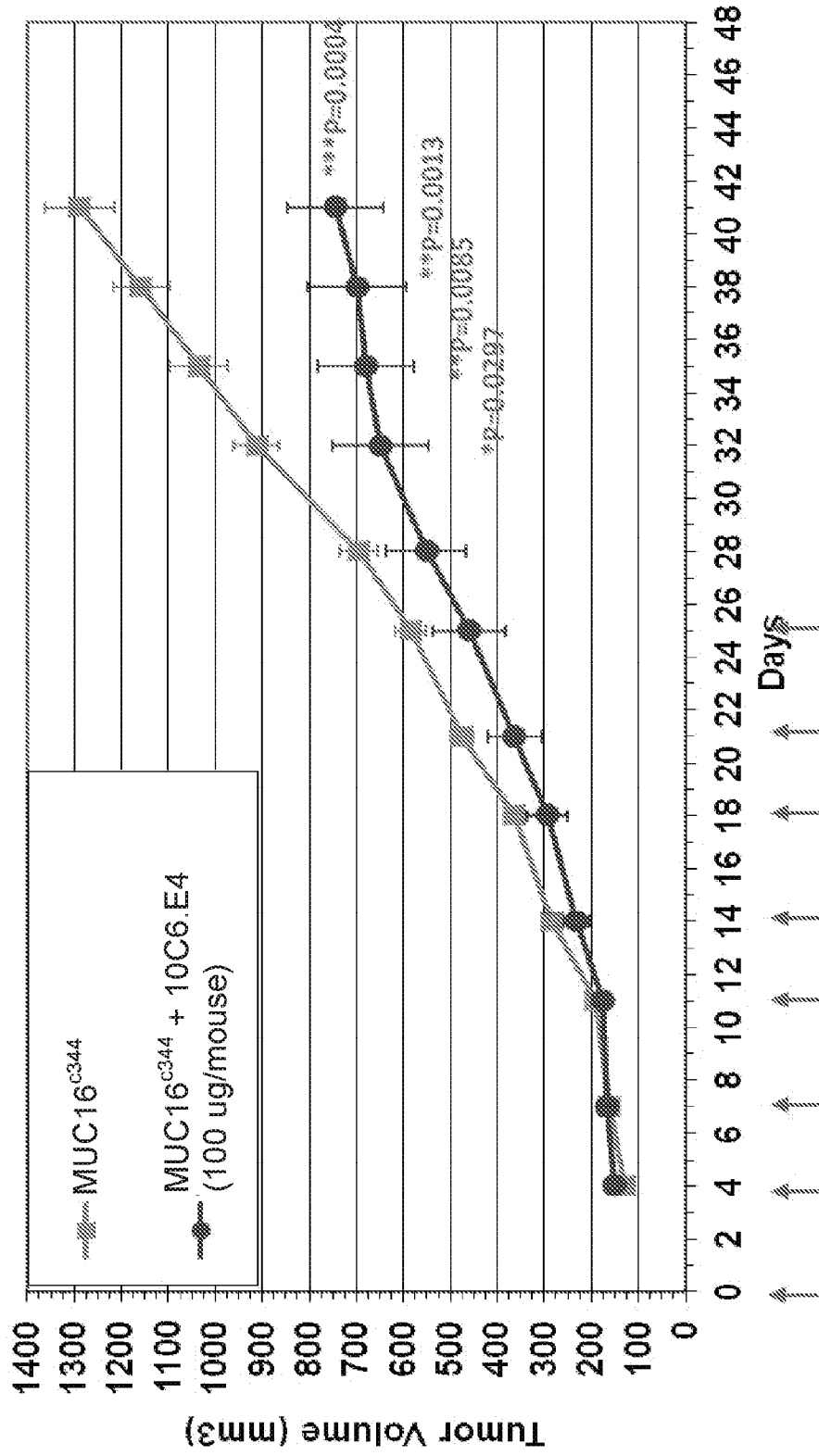


Fig. 16D

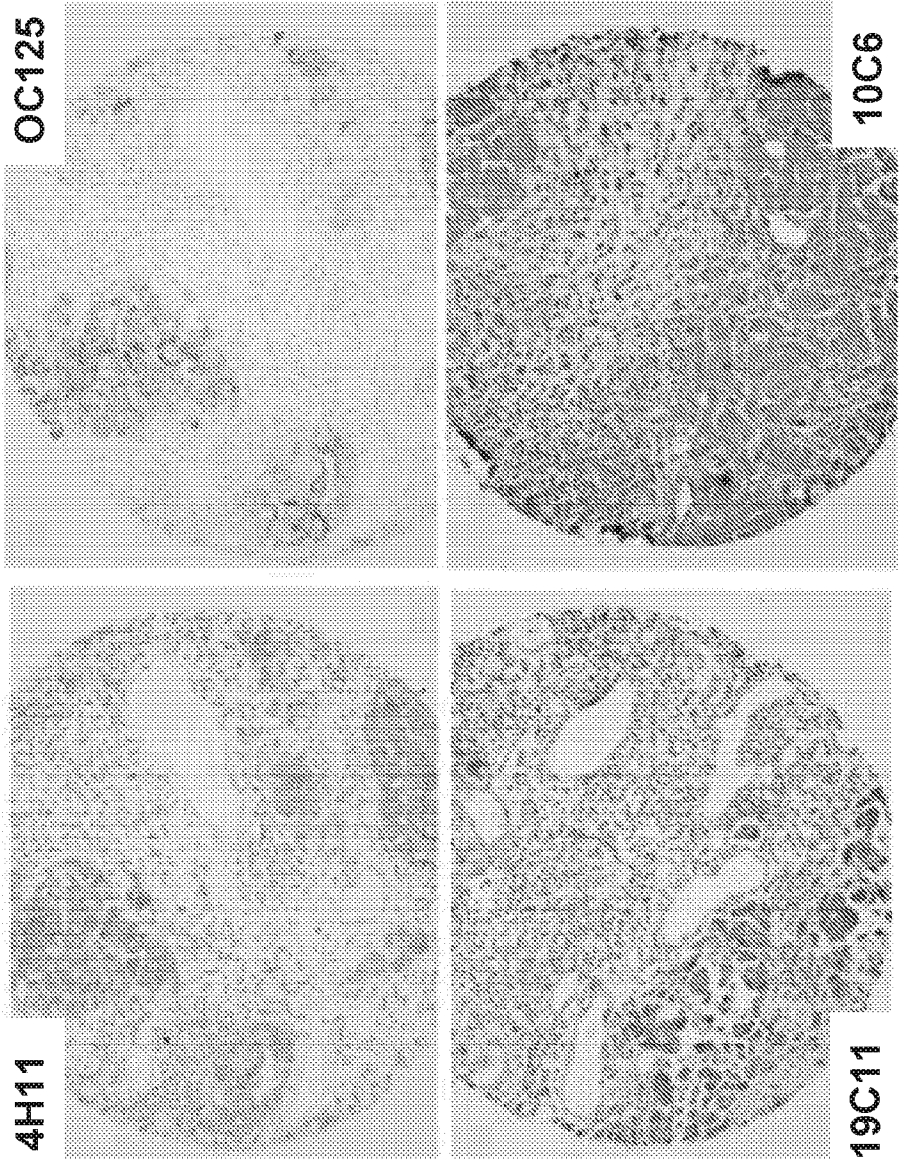


Fig. 16E

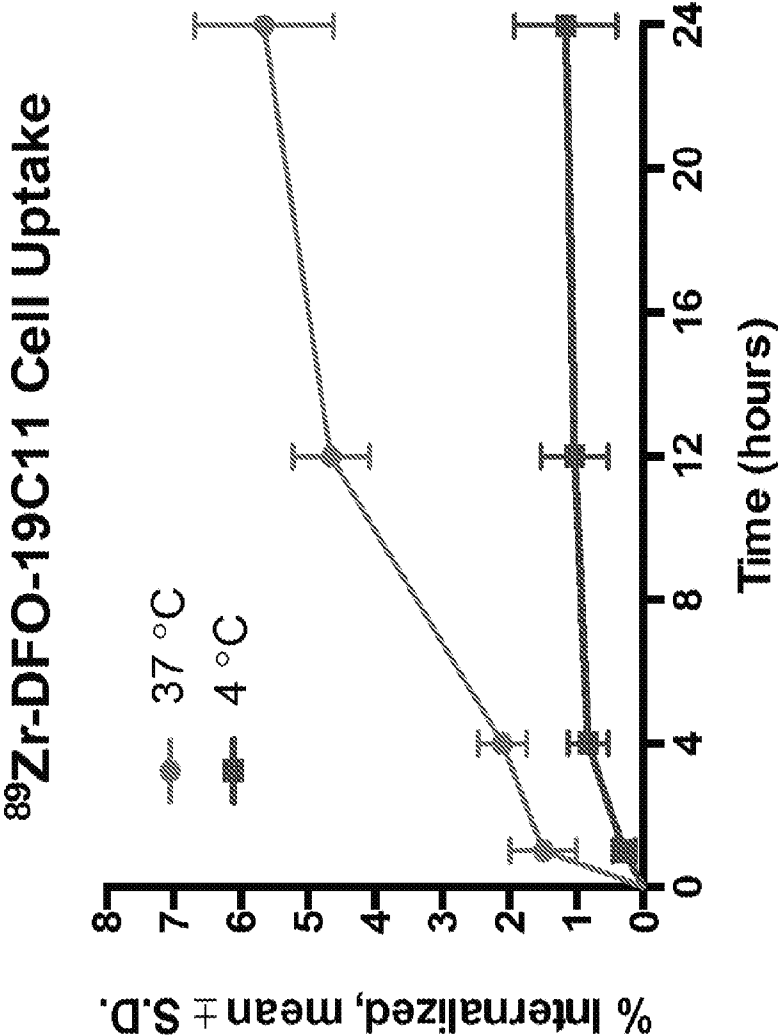


Fig. 17

Fig. 18A

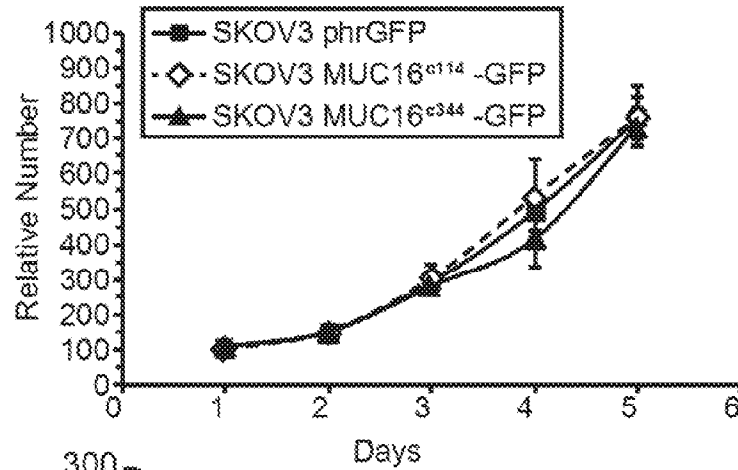


Fig. 18B

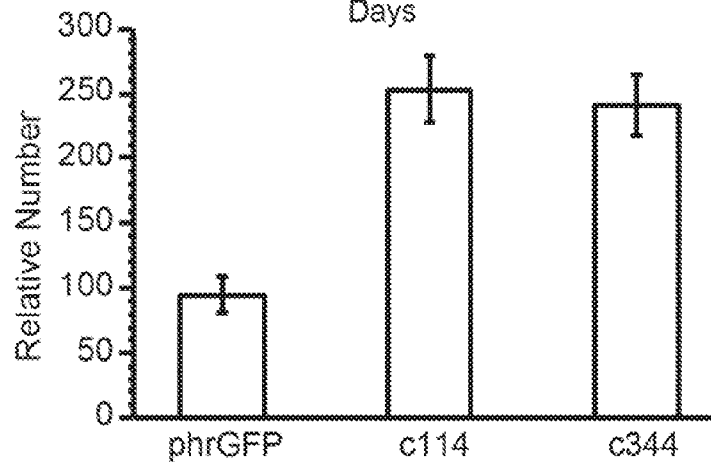
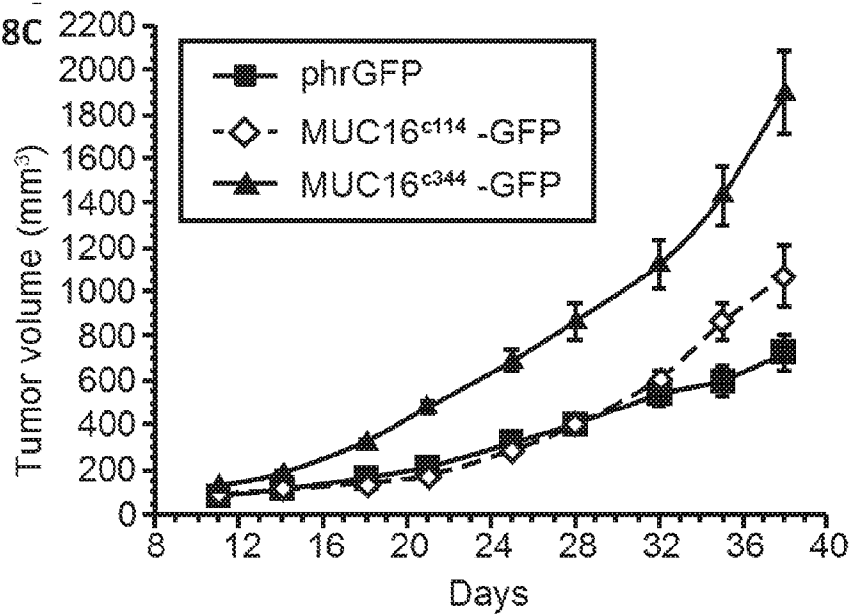


Fig. 18C



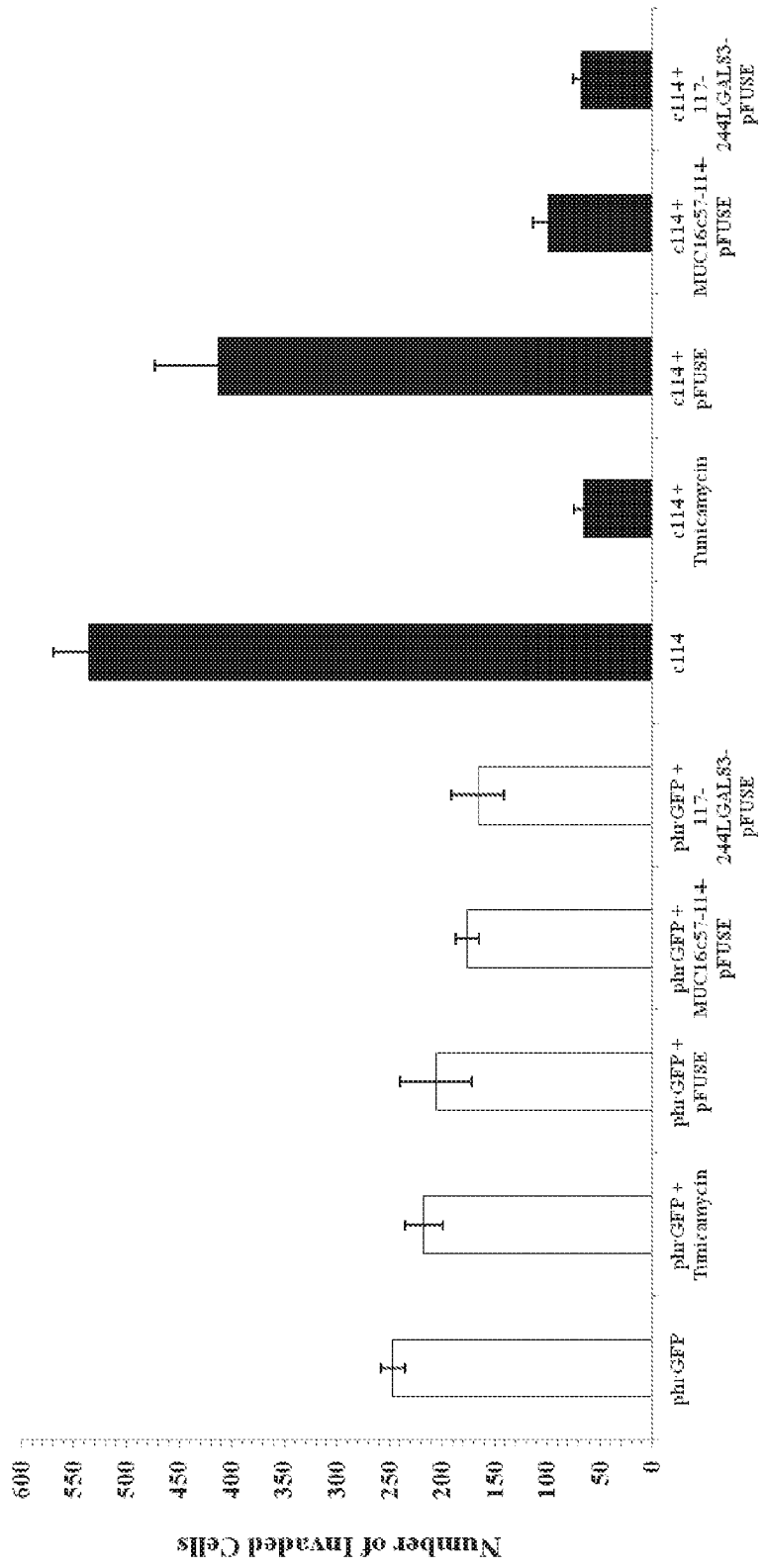


Fig. 19A

Fig. 19B

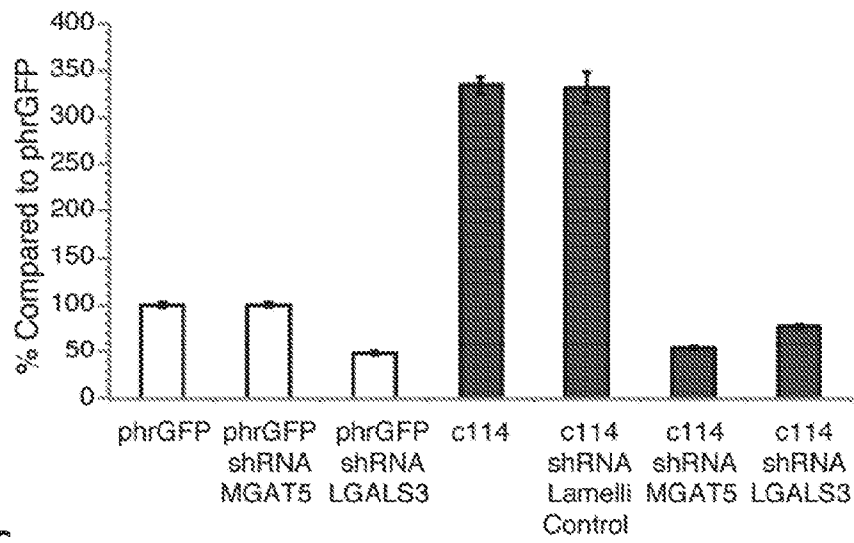


Fig. 19C

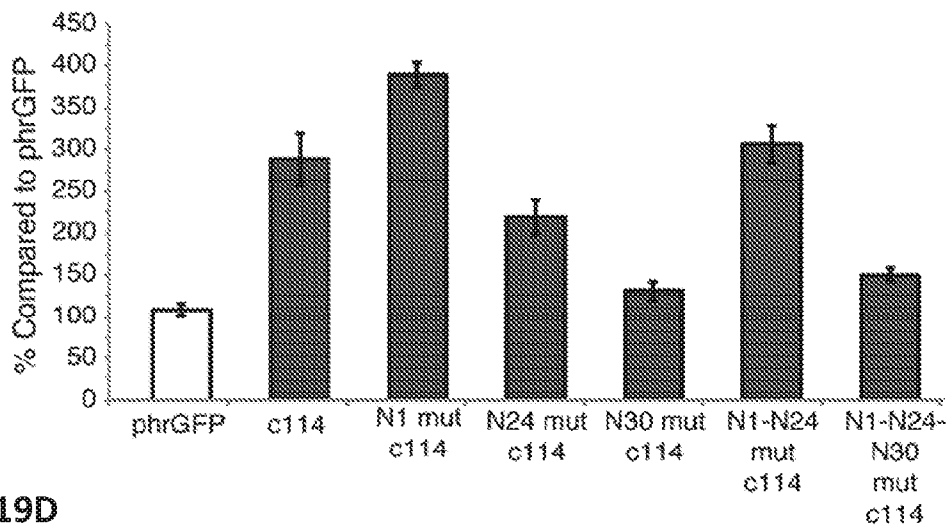


Fig. 19D

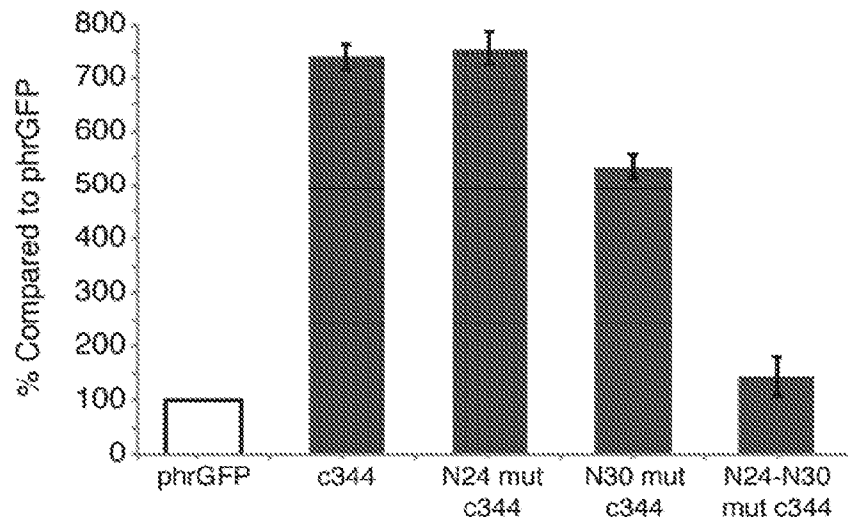


Fig. 19E

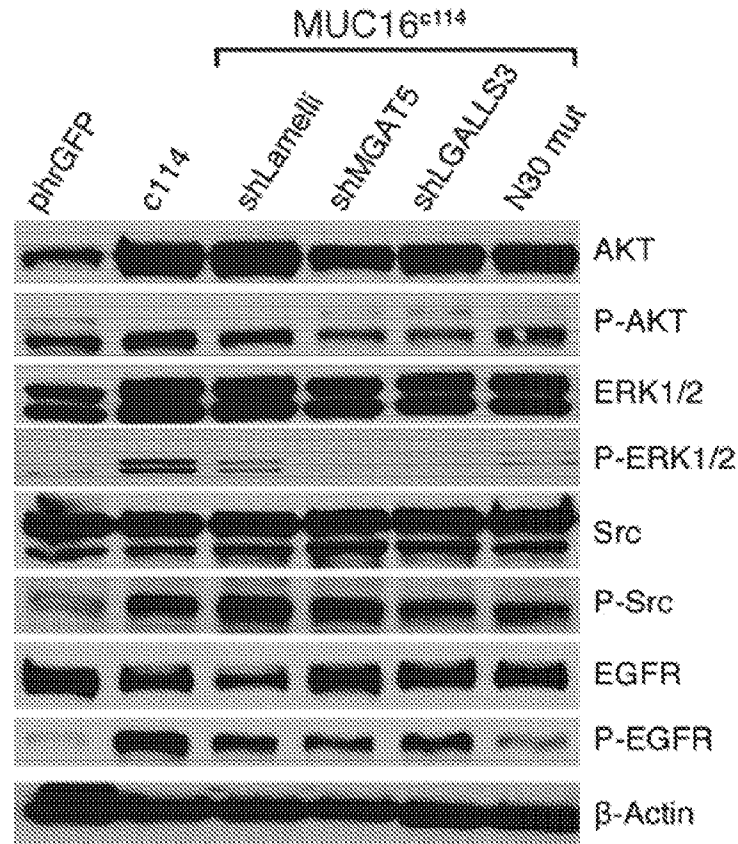


Fig. 19F

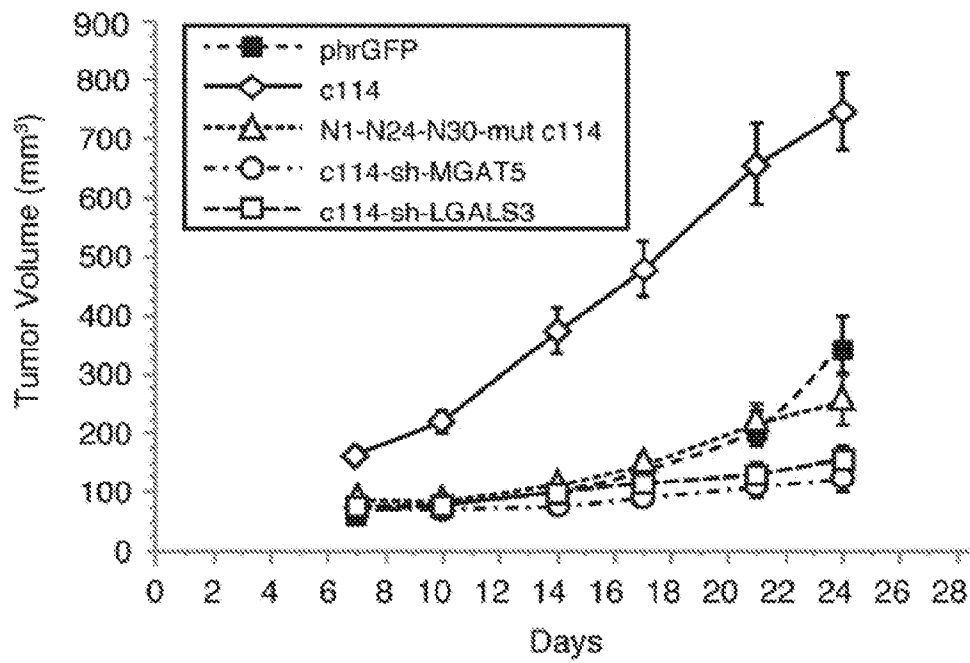


Fig. 20A

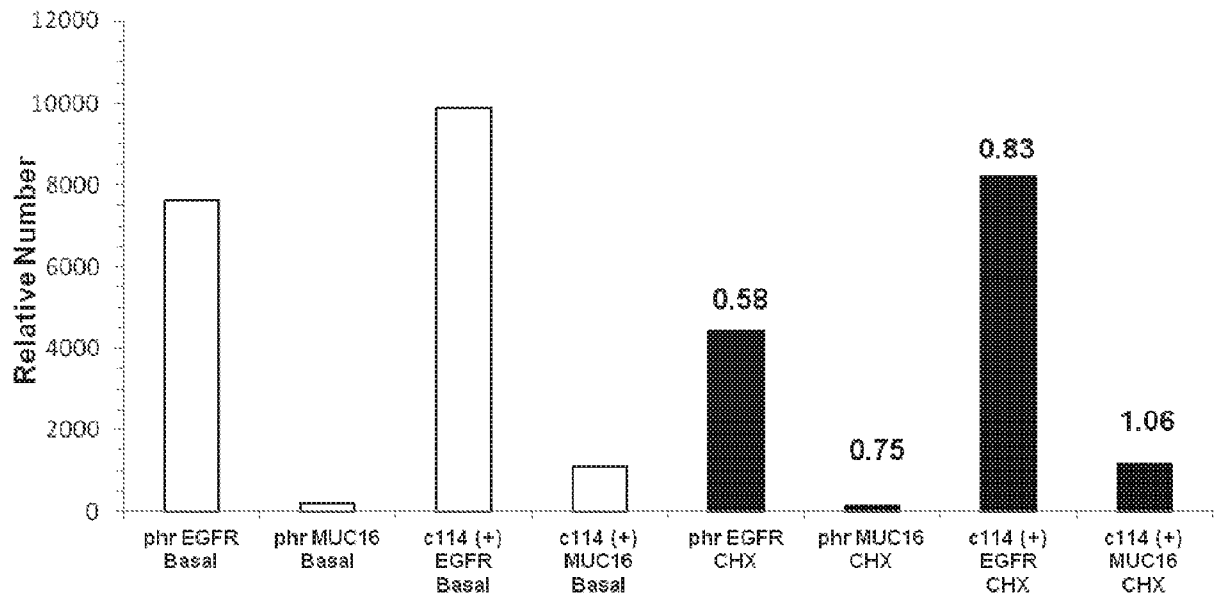
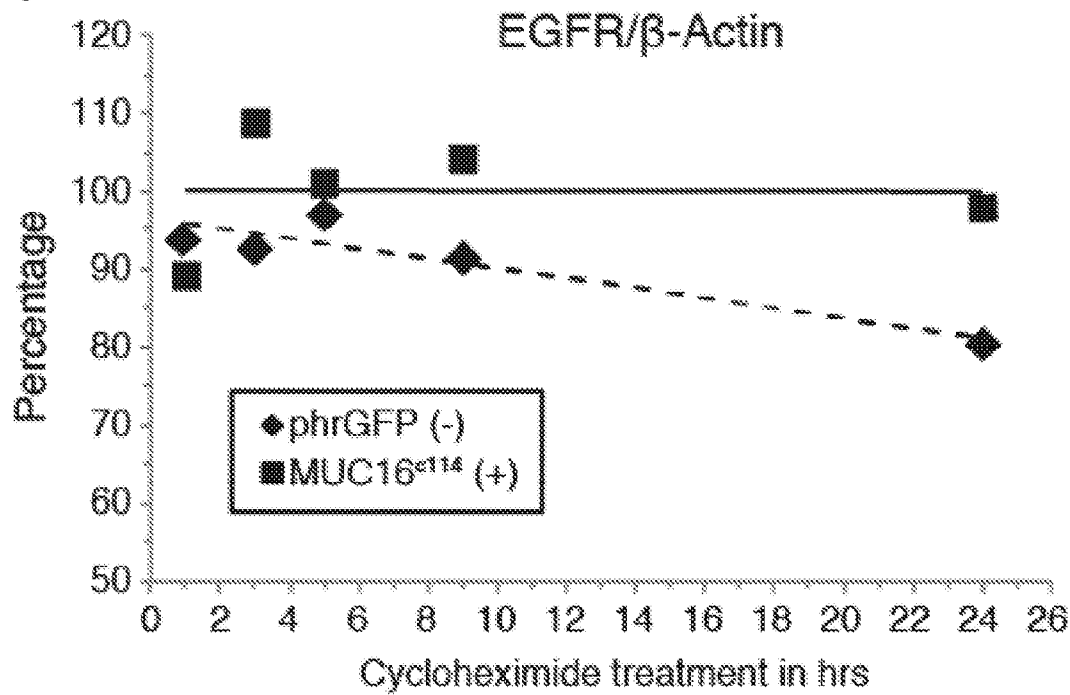


Fig. 20B



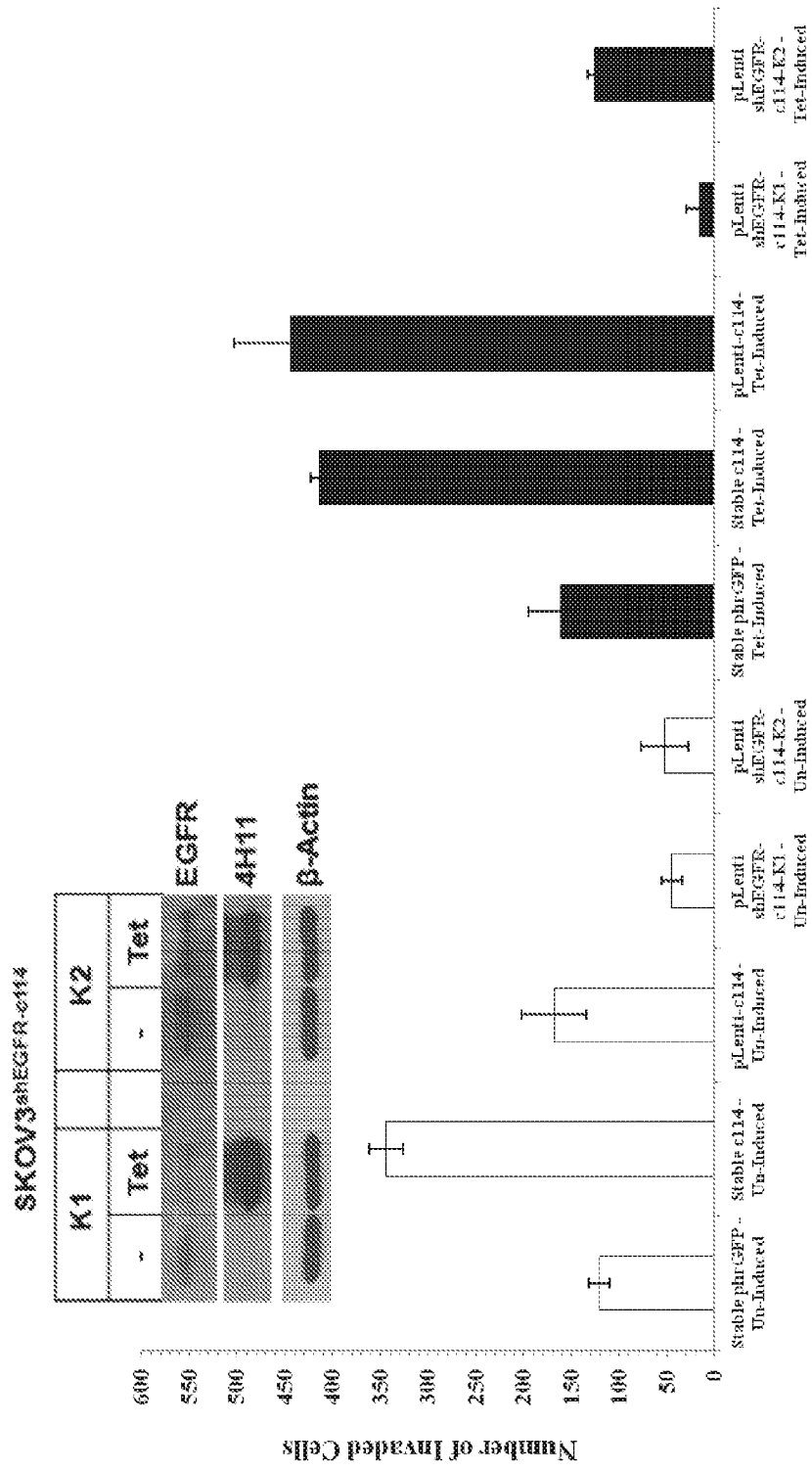


Fig. 20C

Fig. 20D

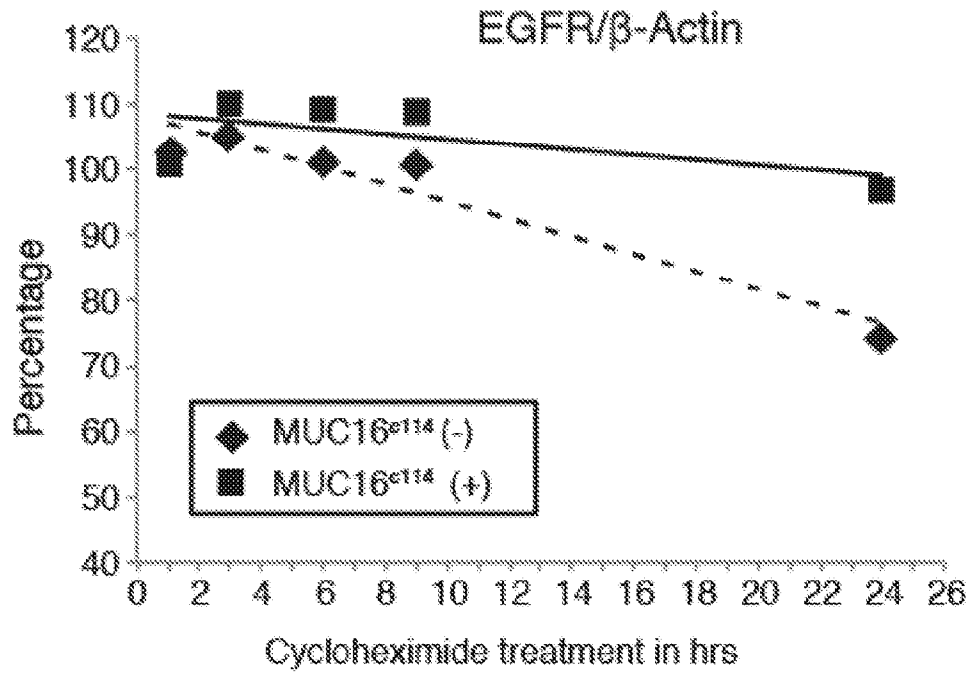


Fig. 21

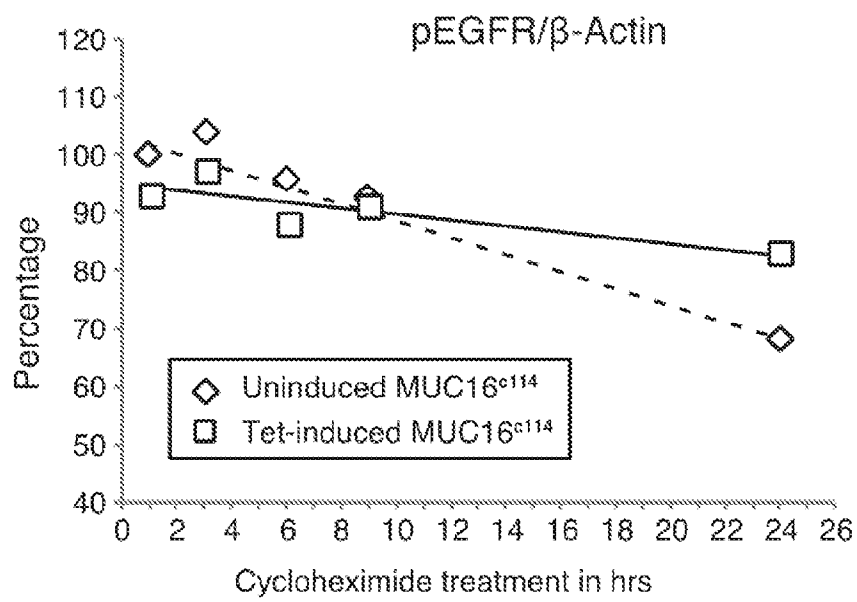


**Fig. 22A**

N-glycans listed in this table were detected through total ion mapping (M: Mannose, N: N-acetylglucosamine, F: fucose, Gal: Galactose, NeuAc: N-acetylneuraminic acid)

| Structural assignment |  | Mass<br>[M+Na] <sup>+</sup> | Intensity/Charge<br>state | Glycan<br>% |
|-----------------------|--|-----------------------------|---------------------------|-------------|
| M2N2F                 |  | 1142                        | 0.10x10 <sup>-2</sup>     | 32.47%      |
| M3N2                  |  | 1171                        | 3.56x10 <sup>-2</sup>     | 11.33%      |
| M3N2F                 |  | 1345                        | 4.18x10 <sup>-2</sup>     | 13.30%      |
| M4N2                  |  | 1376                        | 1.91x10 <sup>-2</sup>     | 6.08%       |
| M5N2                  |  | 1580                        | 3.78x10 <sup>-2</sup>     | 12.03%      |
| M6N2                  |  | 1784                        | 0.87x10 <sup>-2</sup>     | 2.76%       |
| M7N2                  |  | 1988                        | 0.37x10 <sup>-2</sup>     | 1.18%       |
| Gal2N2M3N2F           |  | 2244                        | 1.91x10 <sup>-2</sup>     | 6.06%       |
| NeuAc1Gal2N2M3N2      |  | 2431                        | 1.50x10 <sup>-2</sup>     | 4.76%       |
| NeuAc1Gal2N2M3N2F     |  | 2605                        | 1.65x10 <sup>-2</sup>     | 5.25%       |
| NeuAc2Gal2N2M3N2      |  | 2792                        | 0.77x10 <sup>-2</sup>     | 2.45%       |
| NeuAc2Gal2N2M3N2F     |  | 2966                        | 0.73x10 <sup>-2</sup>     | 2.32%       |
| Total                 |  |                             | 3.14 x10 <sup>-3</sup>    | 100.00%     |

▼ Fucose    ■ N-acetylglucosamin    ● Mannose  
 ○ Galactose    ◆ N-acetylneuraminic acid

Fig. 22B

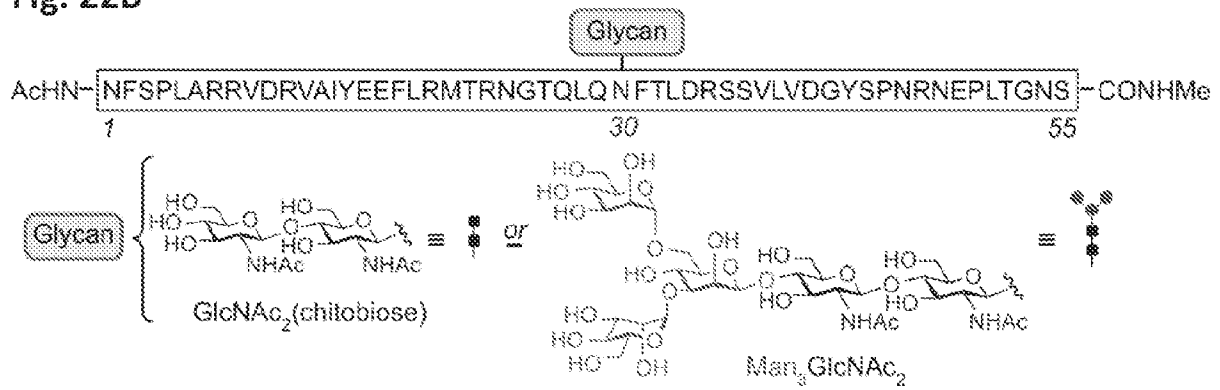


Fig. 22C

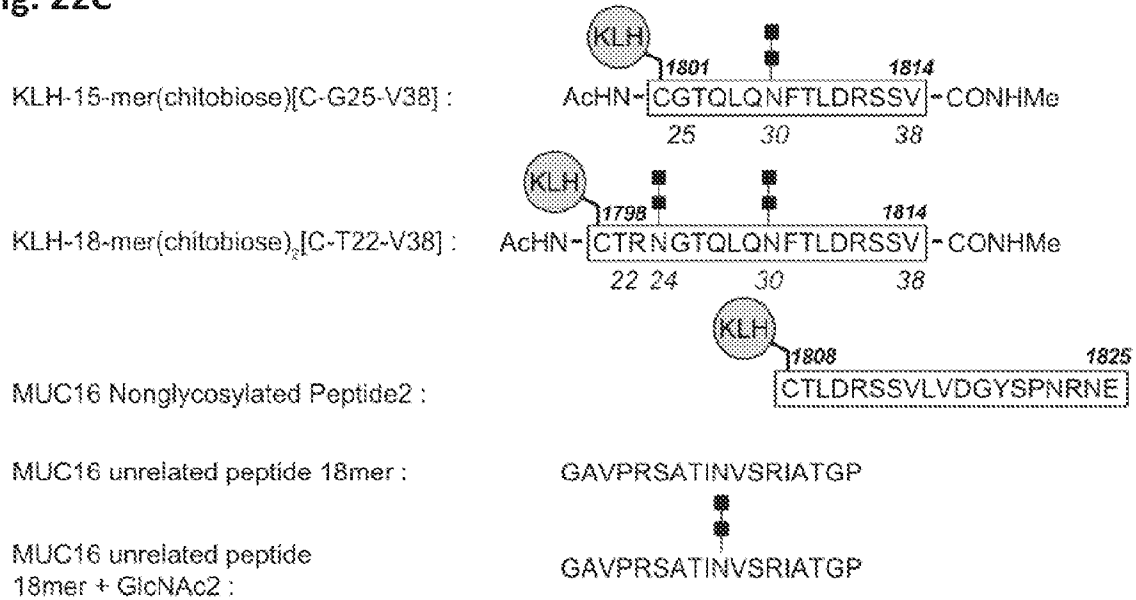


Fig. 23A

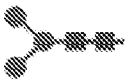
| Peptides                            | Structure                                                                           | Blank | 4H11  | Anti-glycosylated-MUC16<br>ectodomain antibodies |       |       |       |
|-------------------------------------|-------------------------------------------------------------------------------------|-------|-------|--------------------------------------------------|-------|-------|-------|
|                                     |                                                                                     |       |       | 7B12                                             | 10C6  | 18C6  | 19C11 |
| MUC16 unrelated<br>peptide 18mer    |                                                                                     | 0.013 | 0.045 | 0.04                                             | 0.03  | 0.02  | 0.03  |
| MUC16 unrelated<br>18mer + GlcNAc2  |    | 0.014 | 0.038 | 0.03                                             | 0.03  | 0.02  | 0.03  |
| MUC16 15mer                         |                                                                                     | 0.021 | 0.049 | 1.206                                            | 0.543 | 1.061 | 1.167 |
| MUC16 15mer +<br>GlcNAc2            |  | 0.019 | 0.06  | 1.323                                            | 0.573 | 1.442 | 1.514 |
| MUC16 18mer                         |                                                                                     | 0.032 | 0.076 | 1.09                                             | 1.06  | 1.01  | 0.98  |
| MUC16 18mer +<br>GlcNAc1            |  | 0.042 | ND    | 1.03                                             | 1.01  | 0.95  | 0.96  |
| MUC16 18mer +<br>GlcNAc2            |  | 0.029 | 0.063 | 1.06                                             | 1.13  | 1.06  | 1.10  |
| MUC16 18mer +<br>GlcNAc2-Trimannose |  | 0.035 | ND    | 1.05                                             | 0.99  | 0.89  | 0.99  |
| MUC16 18mer +<br>GlcNAc2-FUCOSE1    |  | 0.044 | ND    | 1.10                                             | 1.08  | 1.03  | 1.01  |
| MUC16 Nonglycosylated<br>Peptide2   |                                                                                     | 0.029 | 2.139 | 0.02                                             | 0.02  | 0.02  | 0.03  |

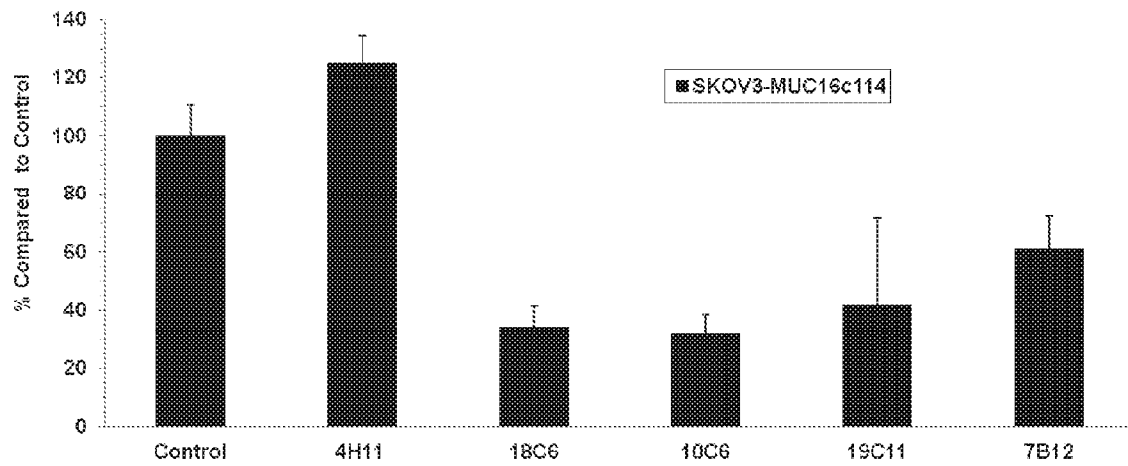
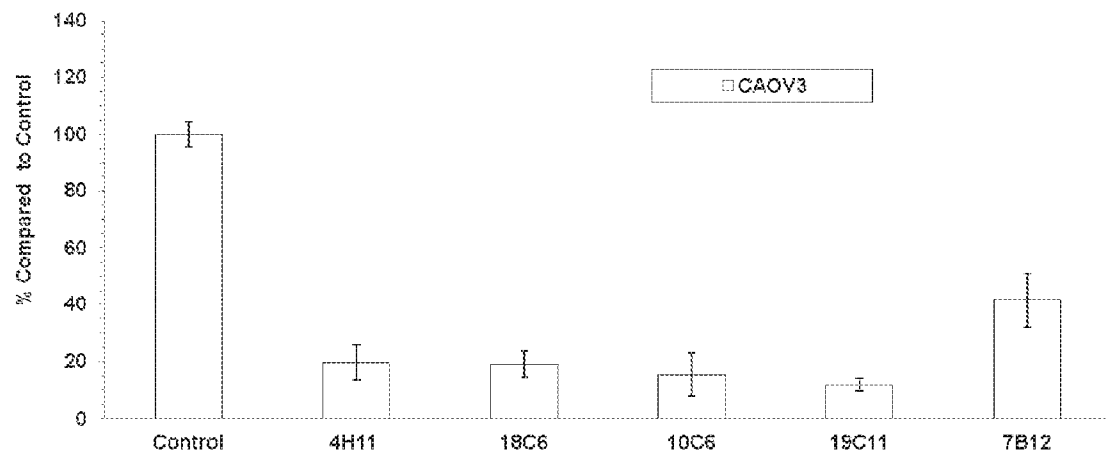
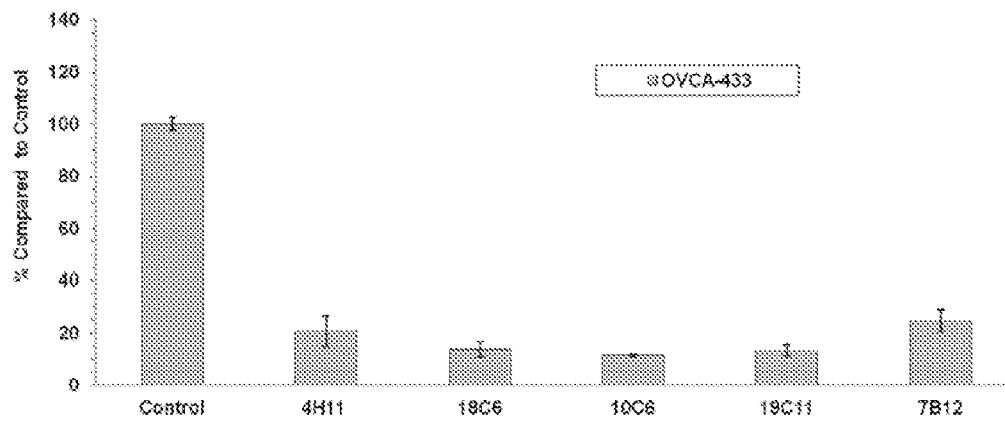
**Fig. 23B****Fig. 23C****Fig. 23D**

Fig. 23E

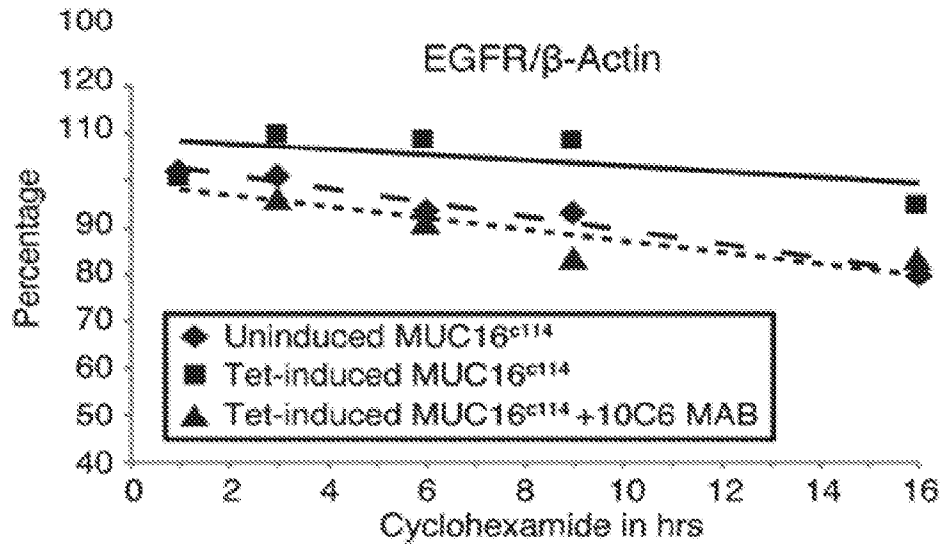


Fig. 23F

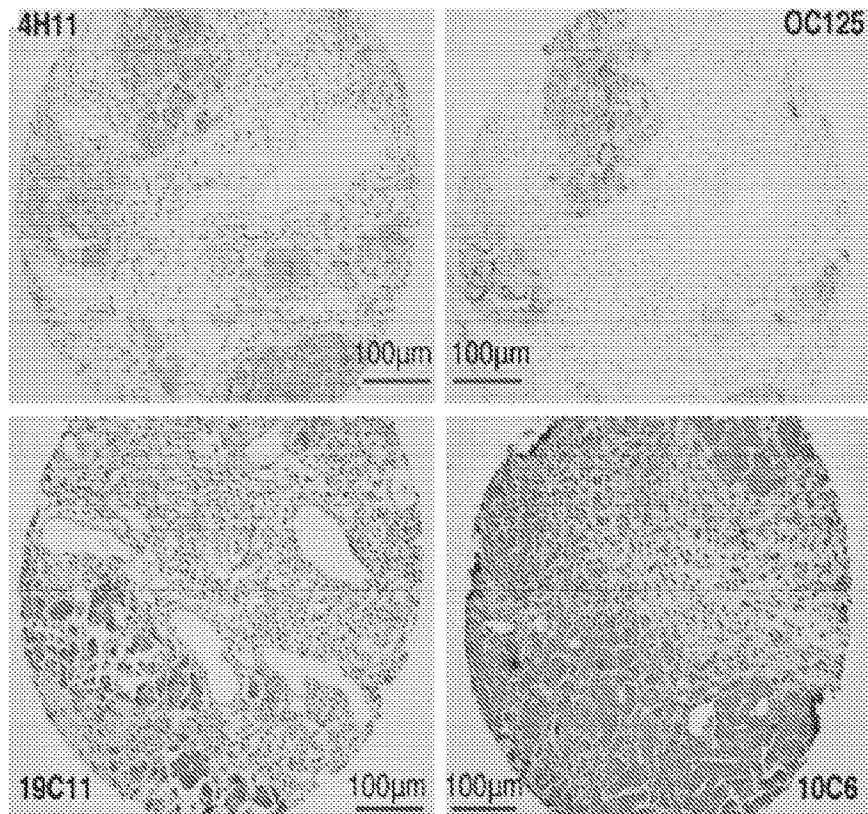


Fig. 23G

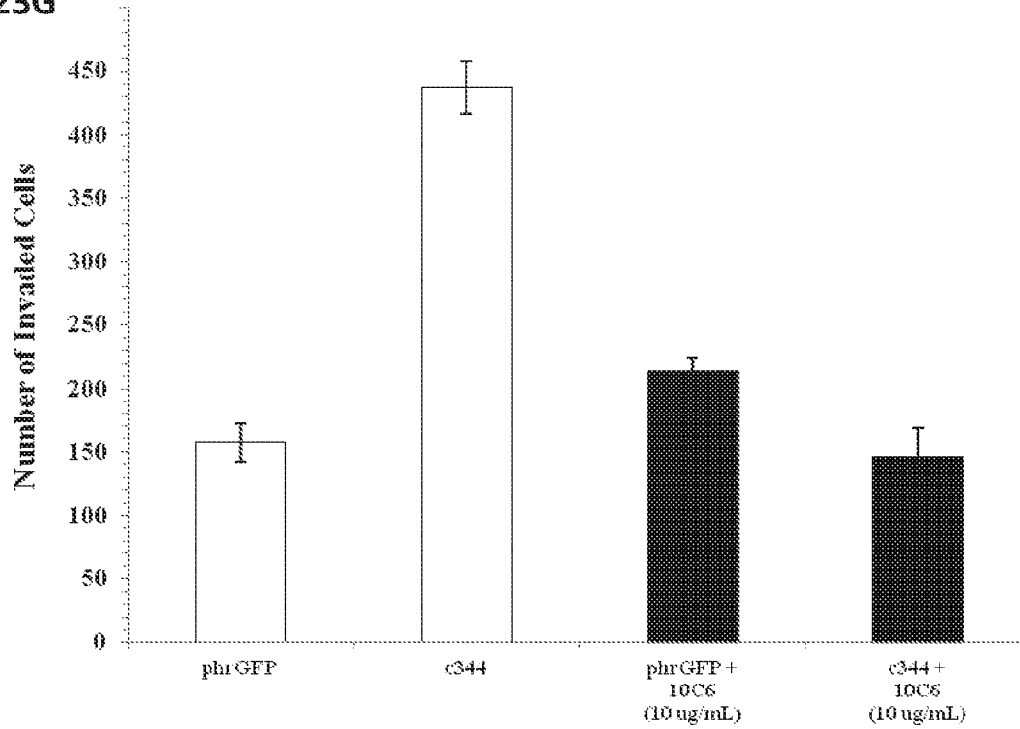


Fig. 23H

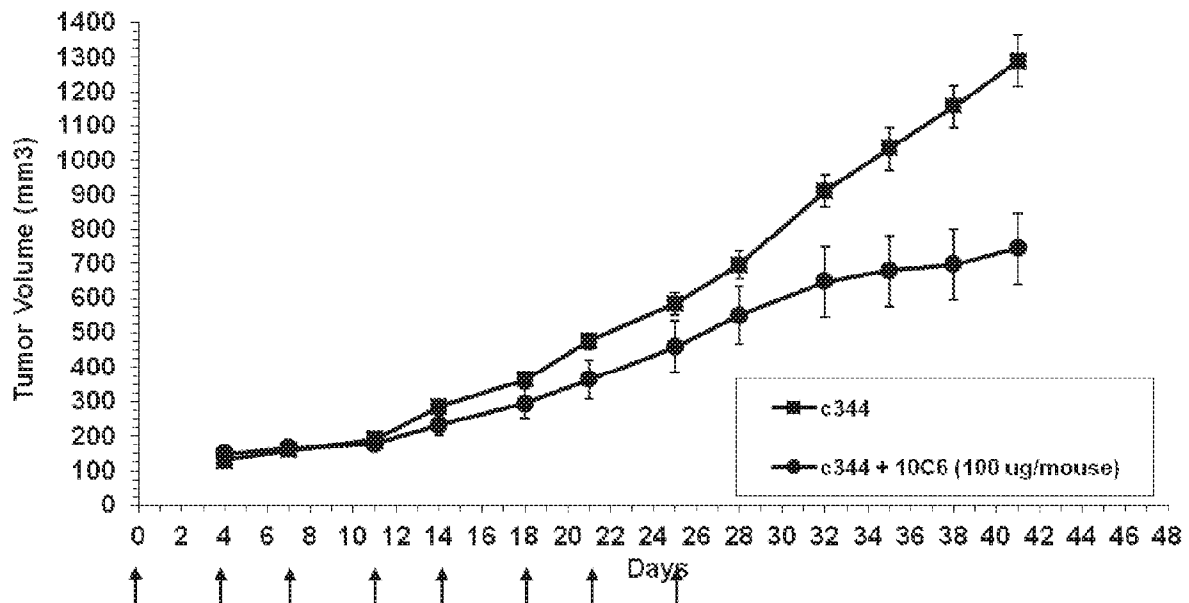


**Fig. 24A**

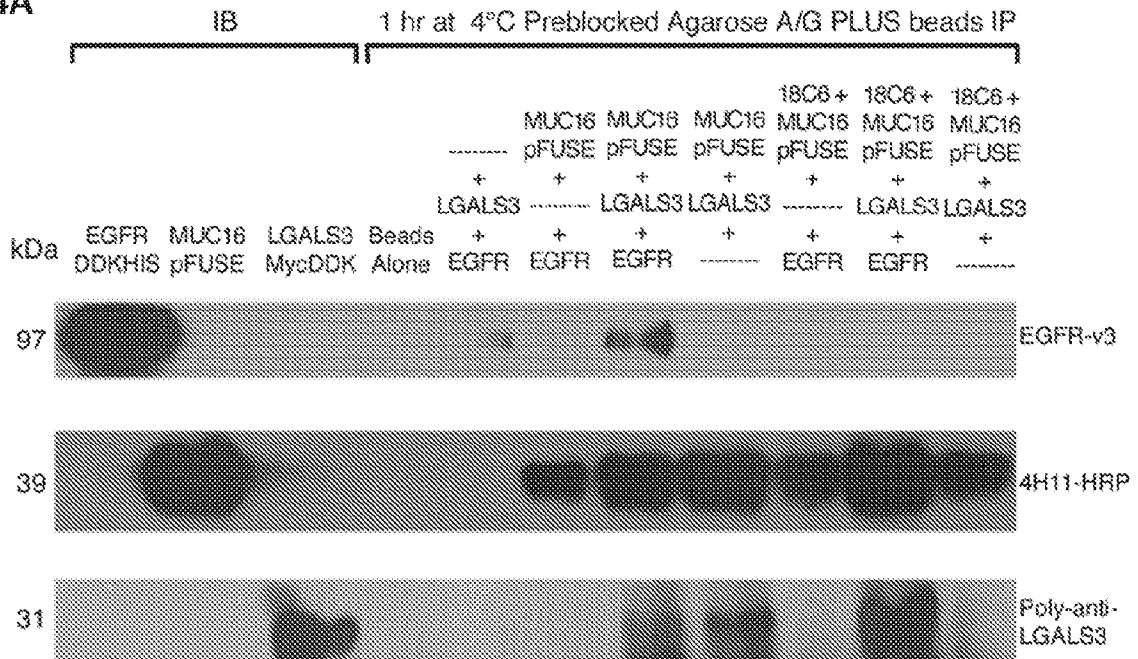


**Fig. 24B**

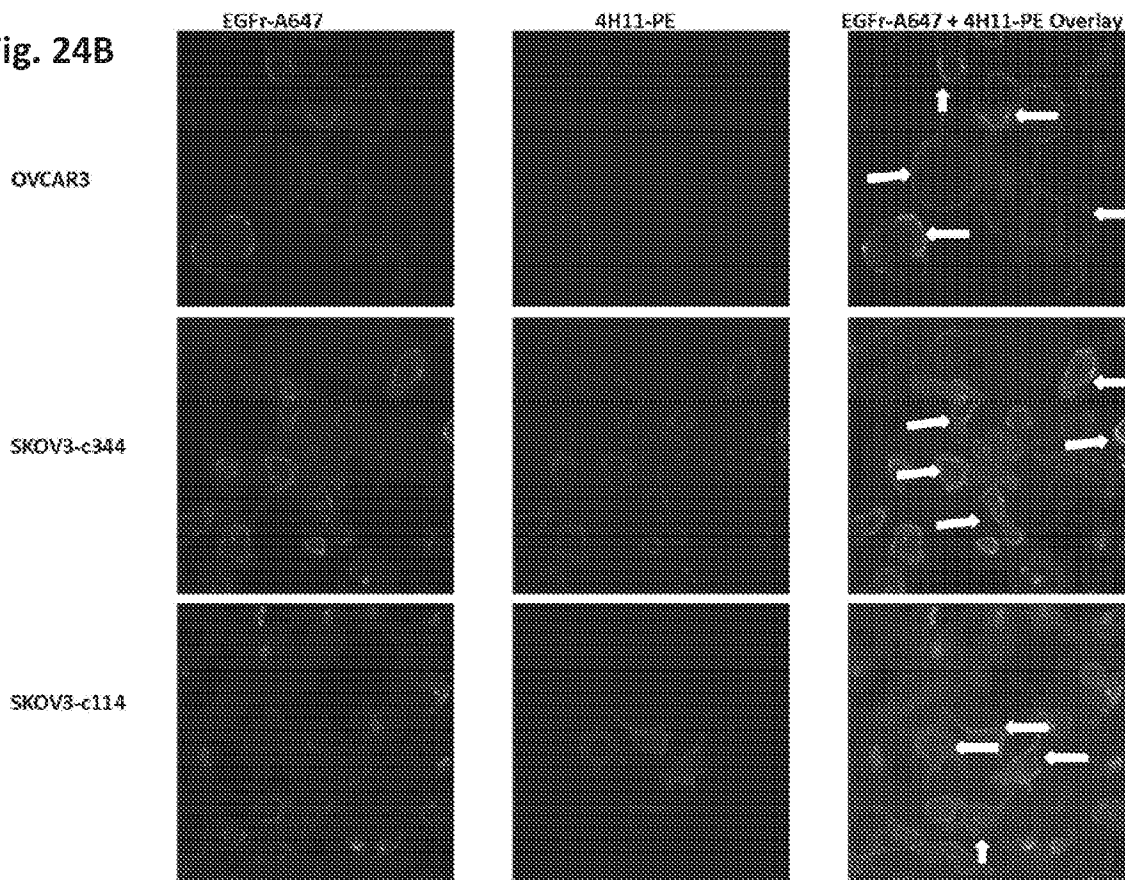


Fig. 24C

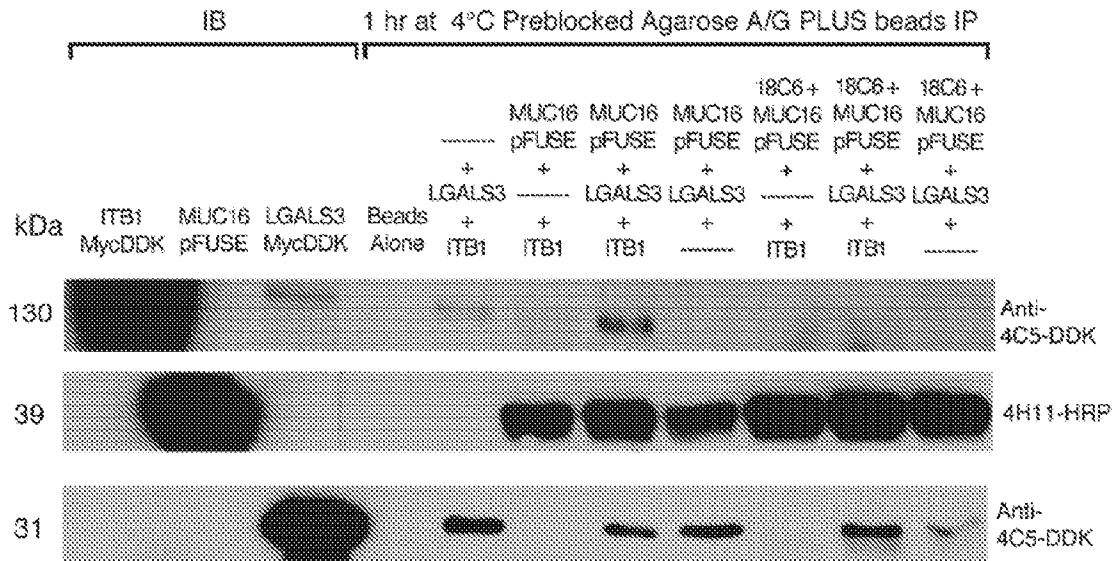


**Fig. 24D**

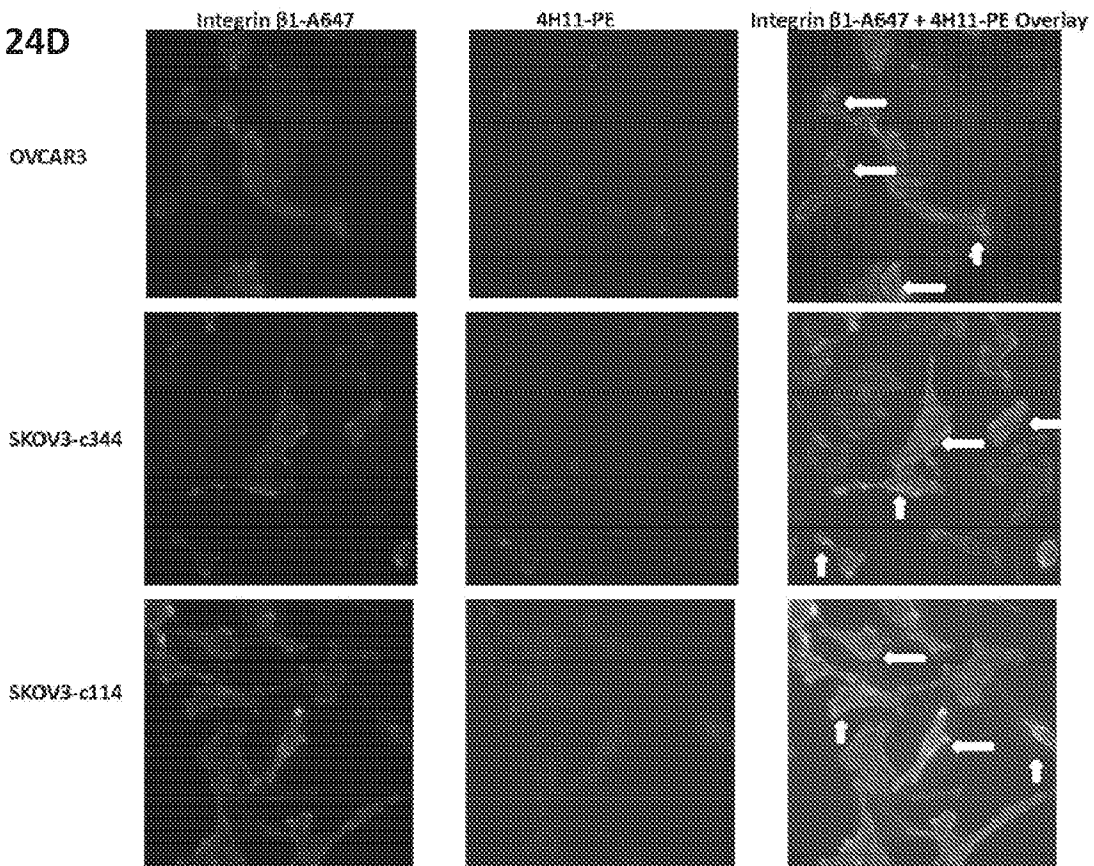


Fig. 25A

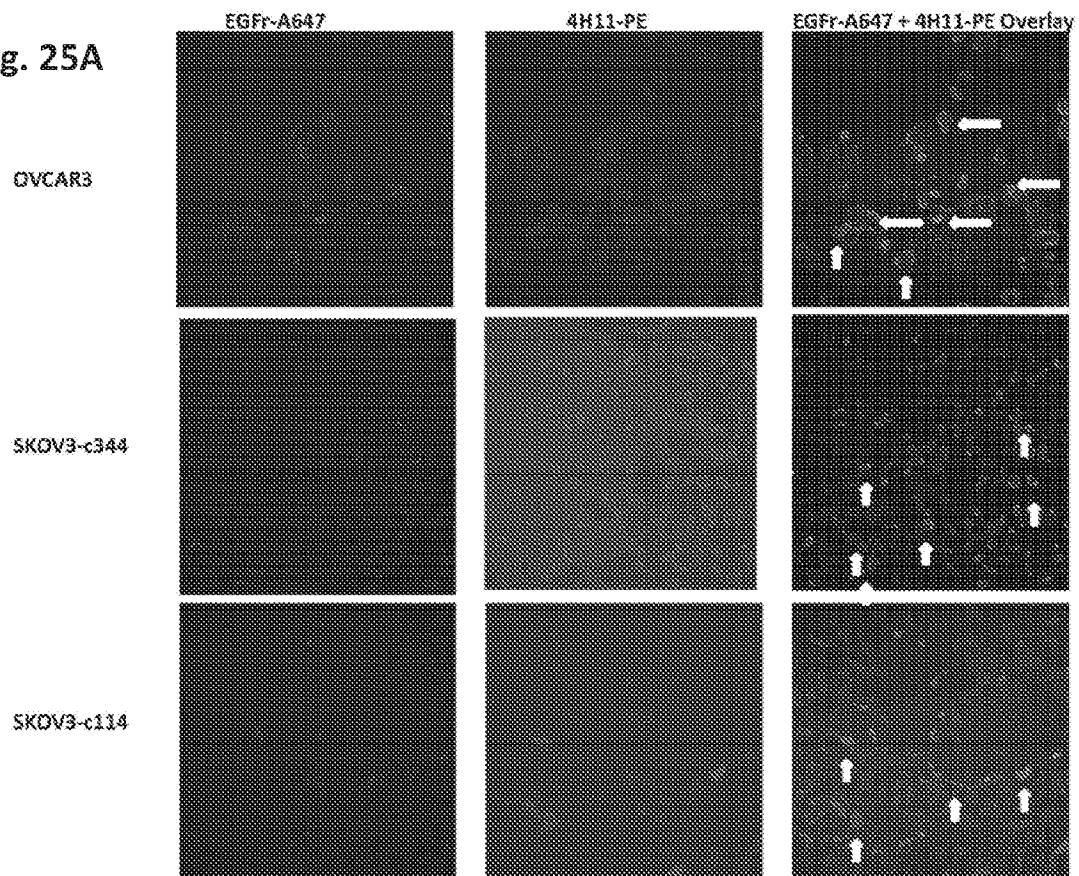


Fig. 25B

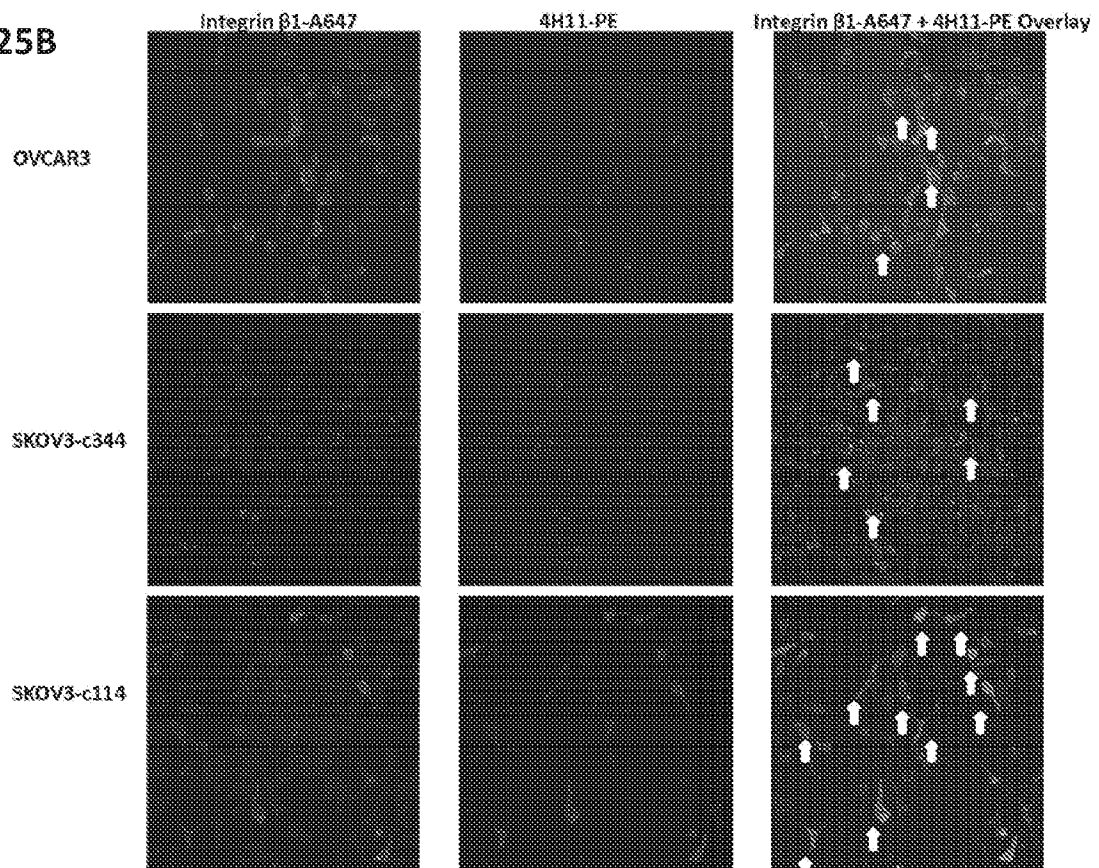


Fig. 26A

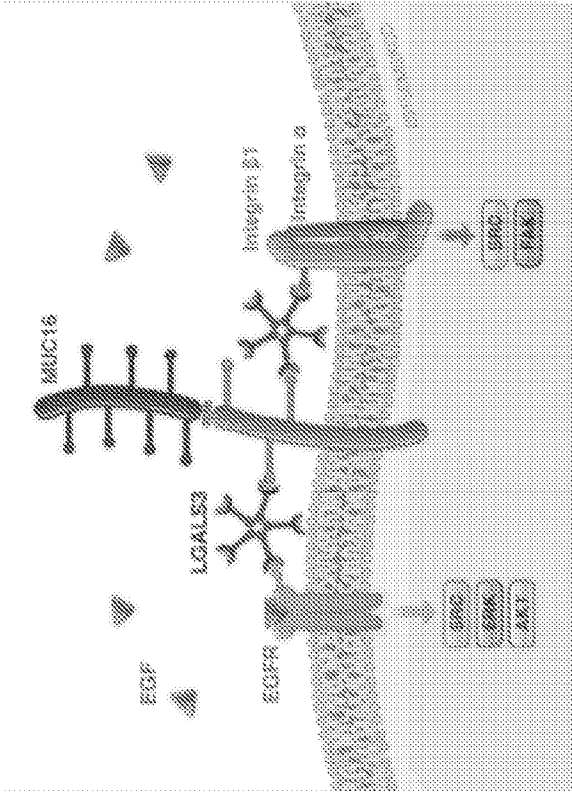
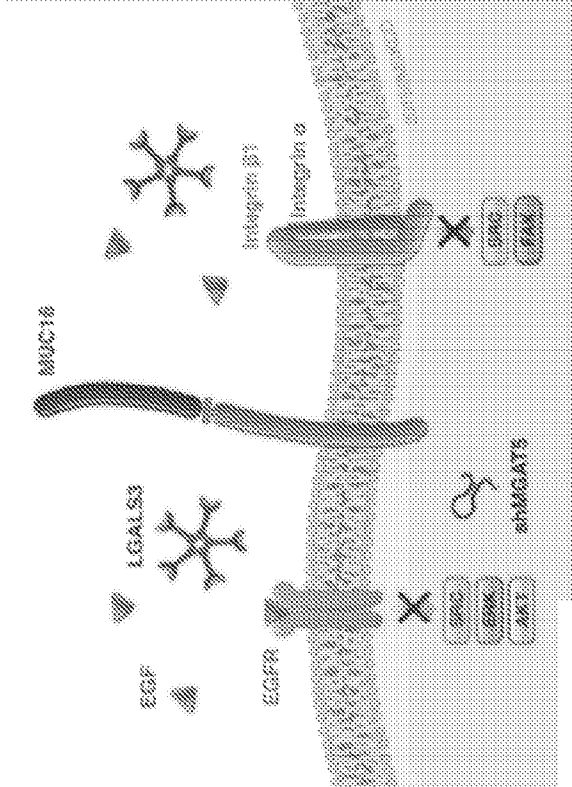


Fig. 26B



( Fig. 26C

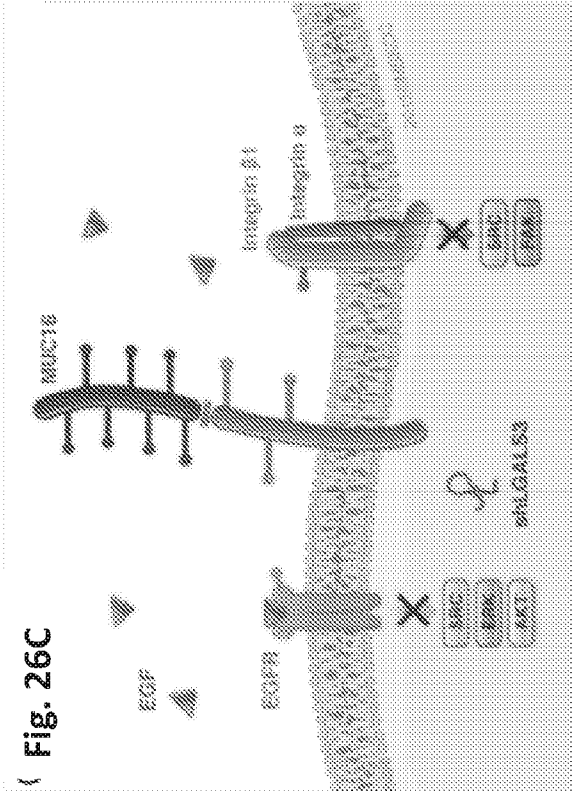
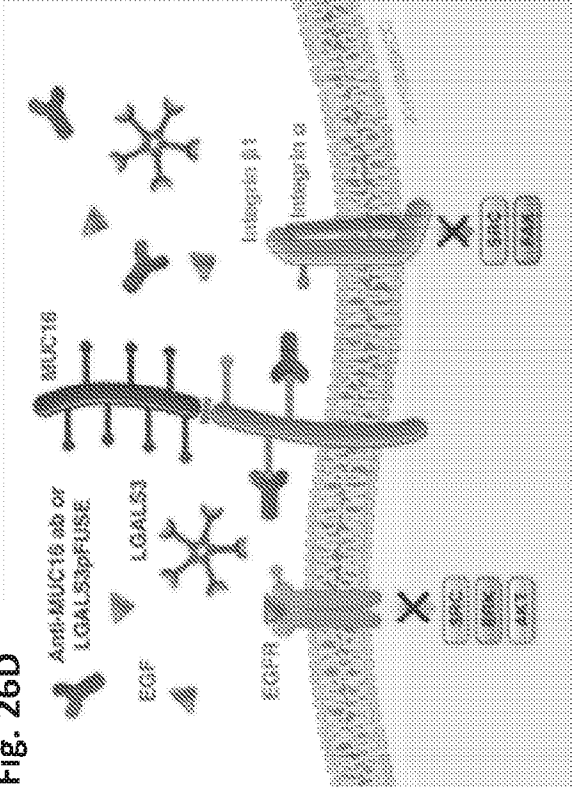


Fig. 26D



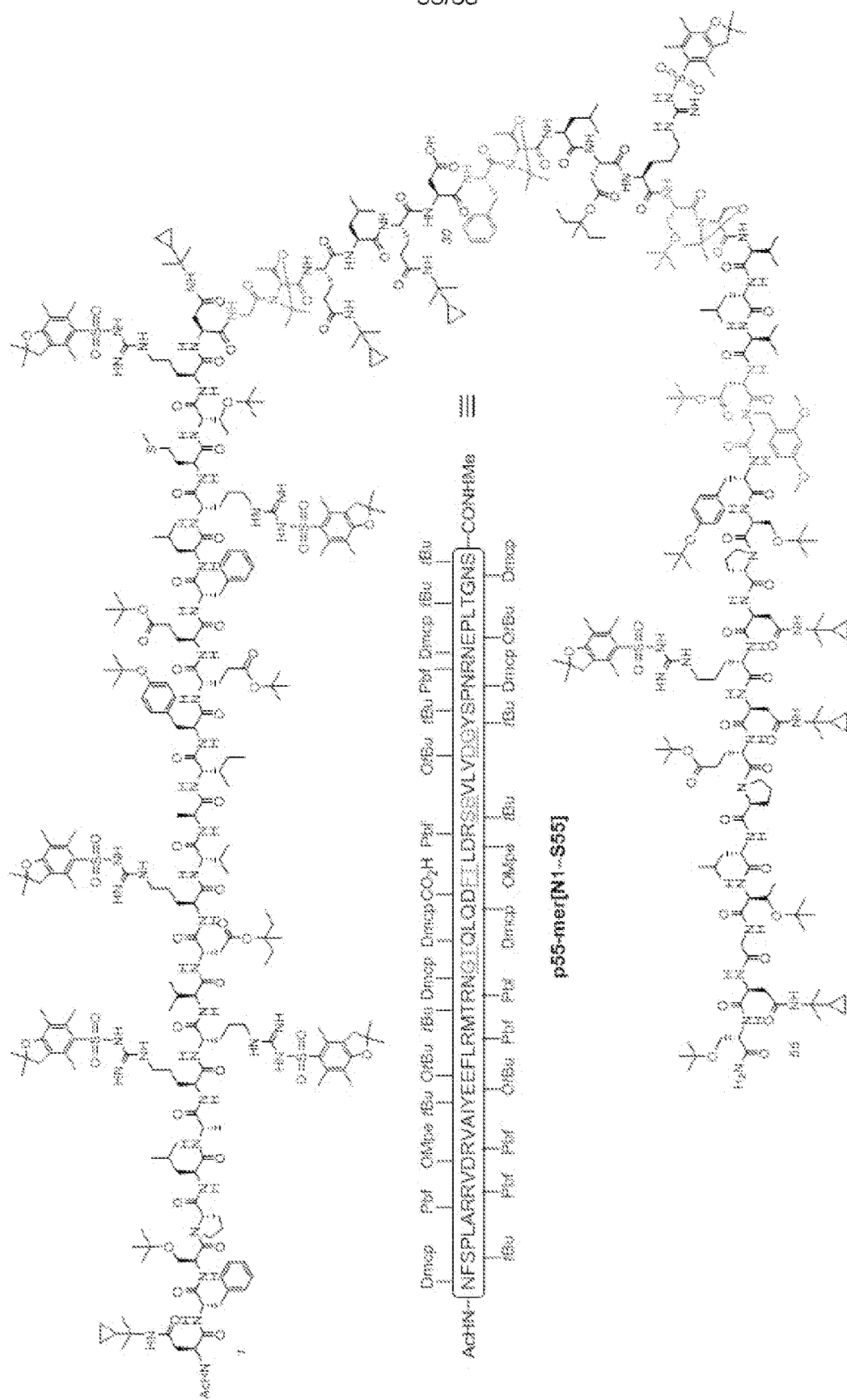


Fig. 27A

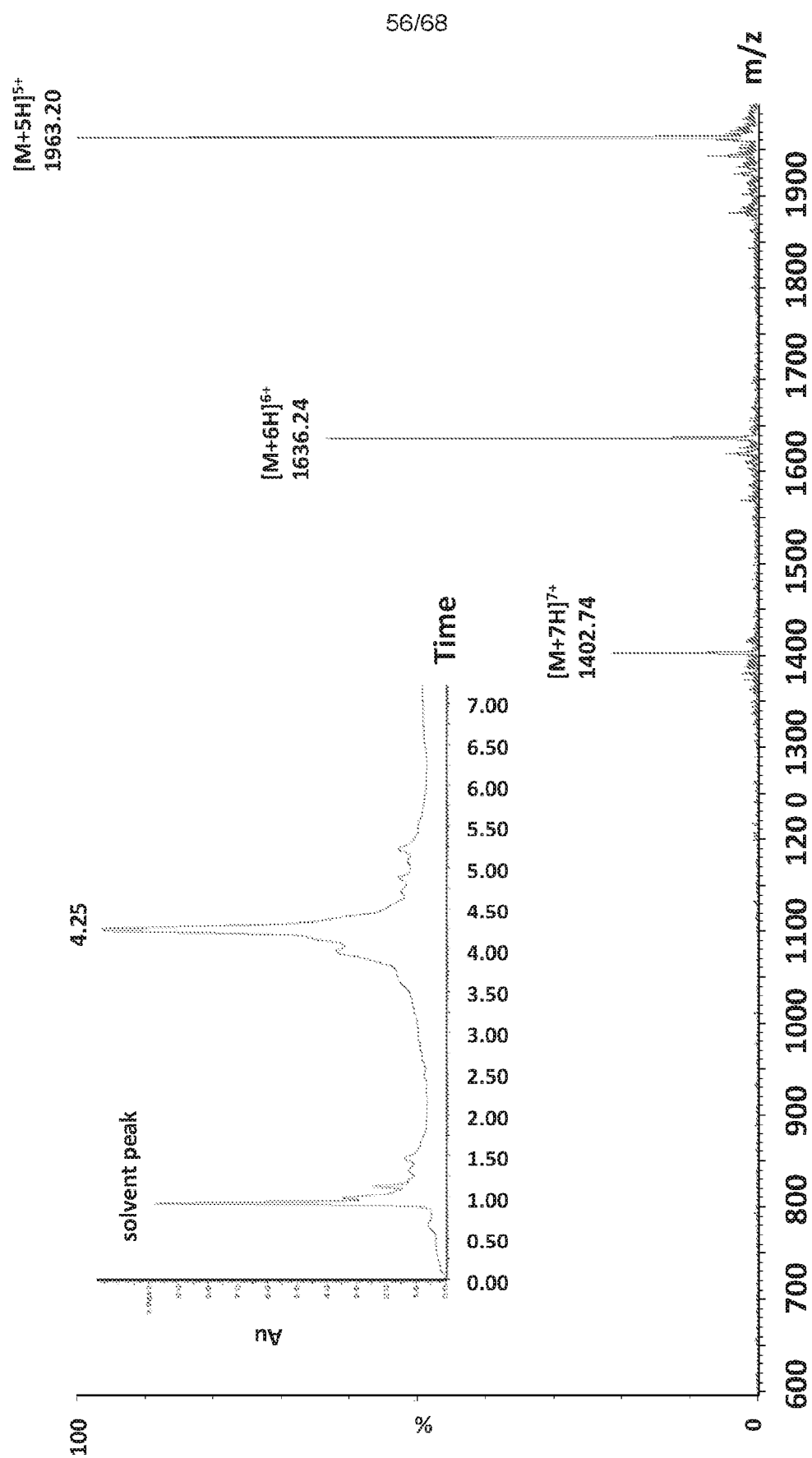


Fig. 27B

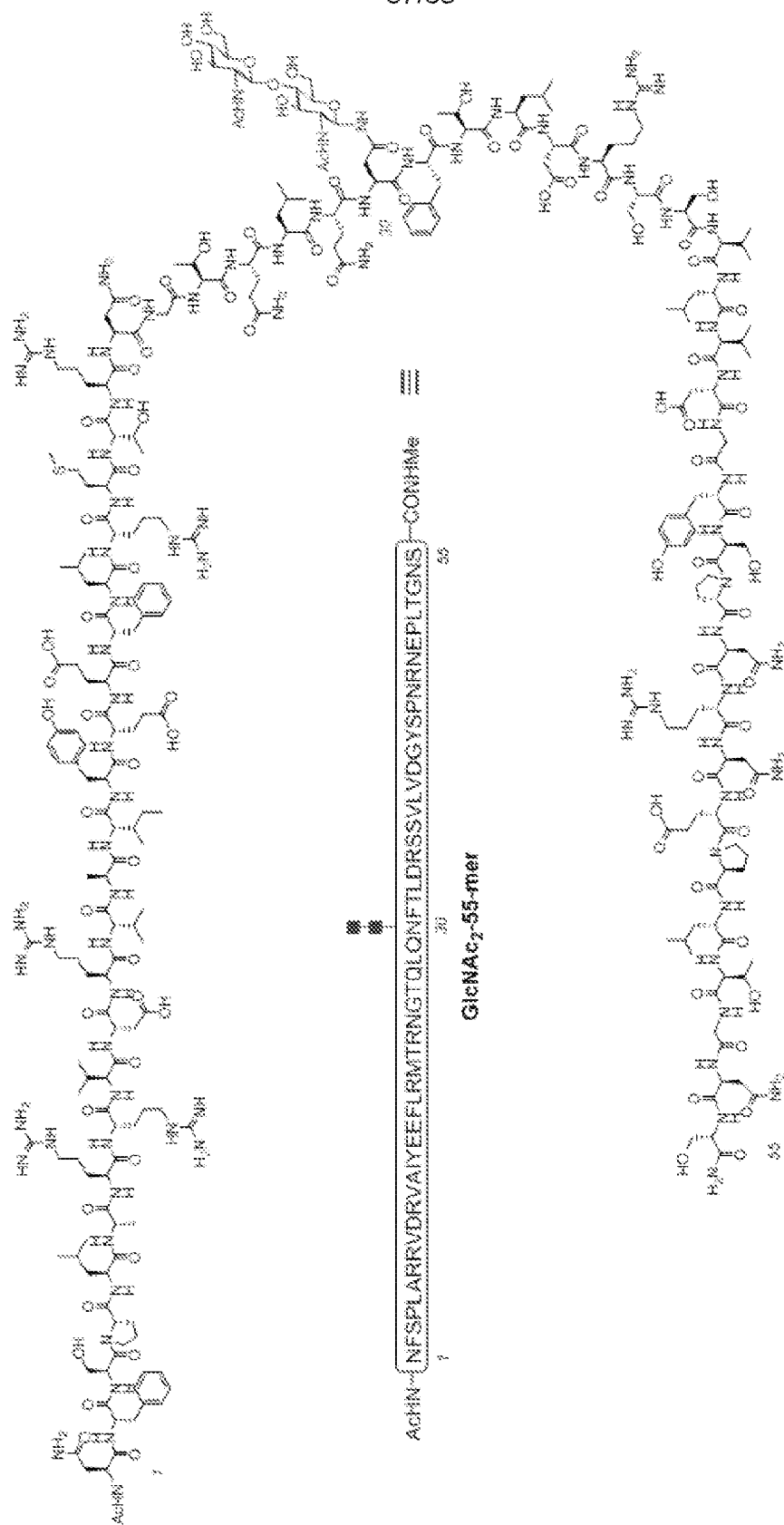


Fig. 28A

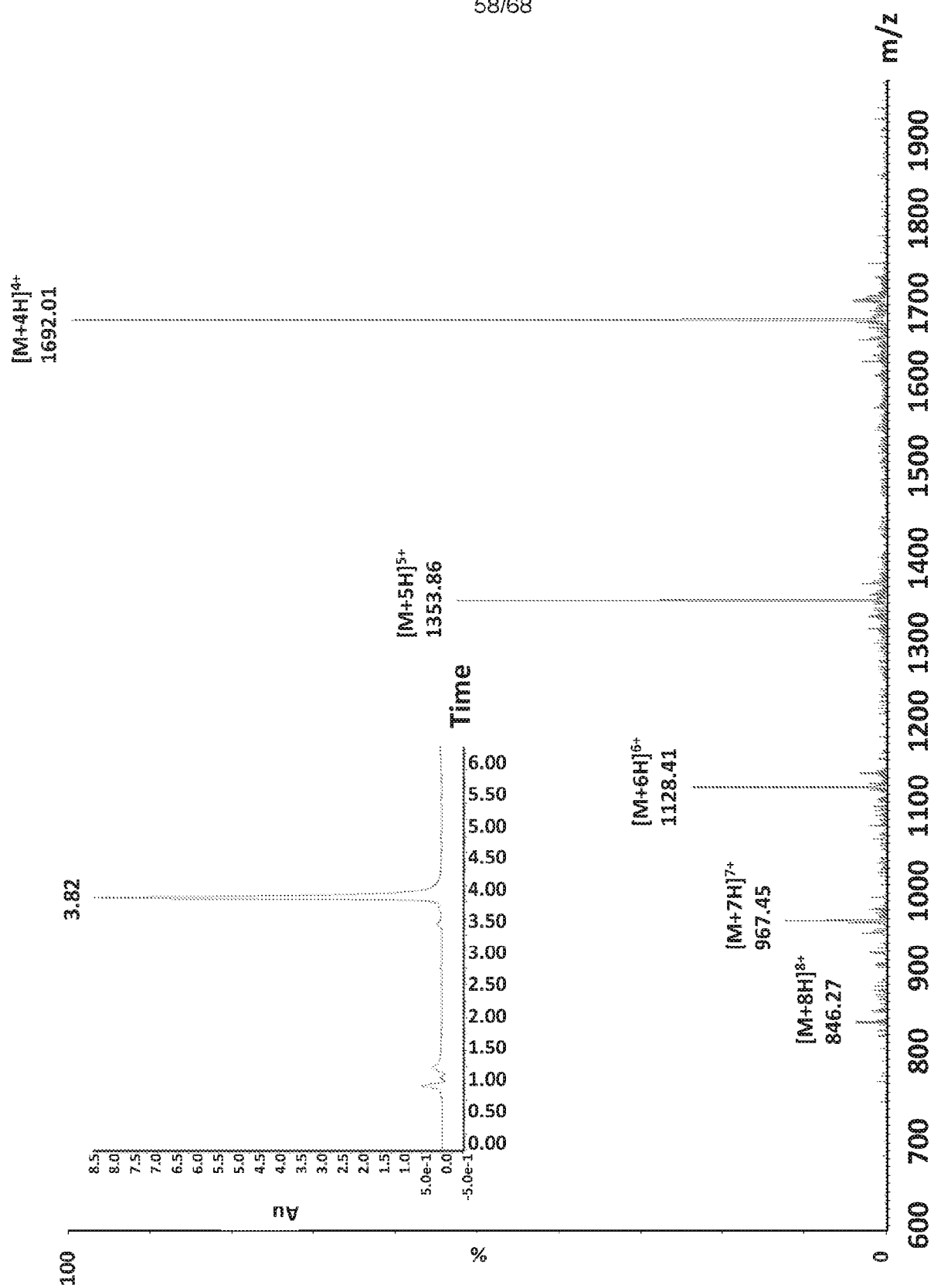


Fig. 28B

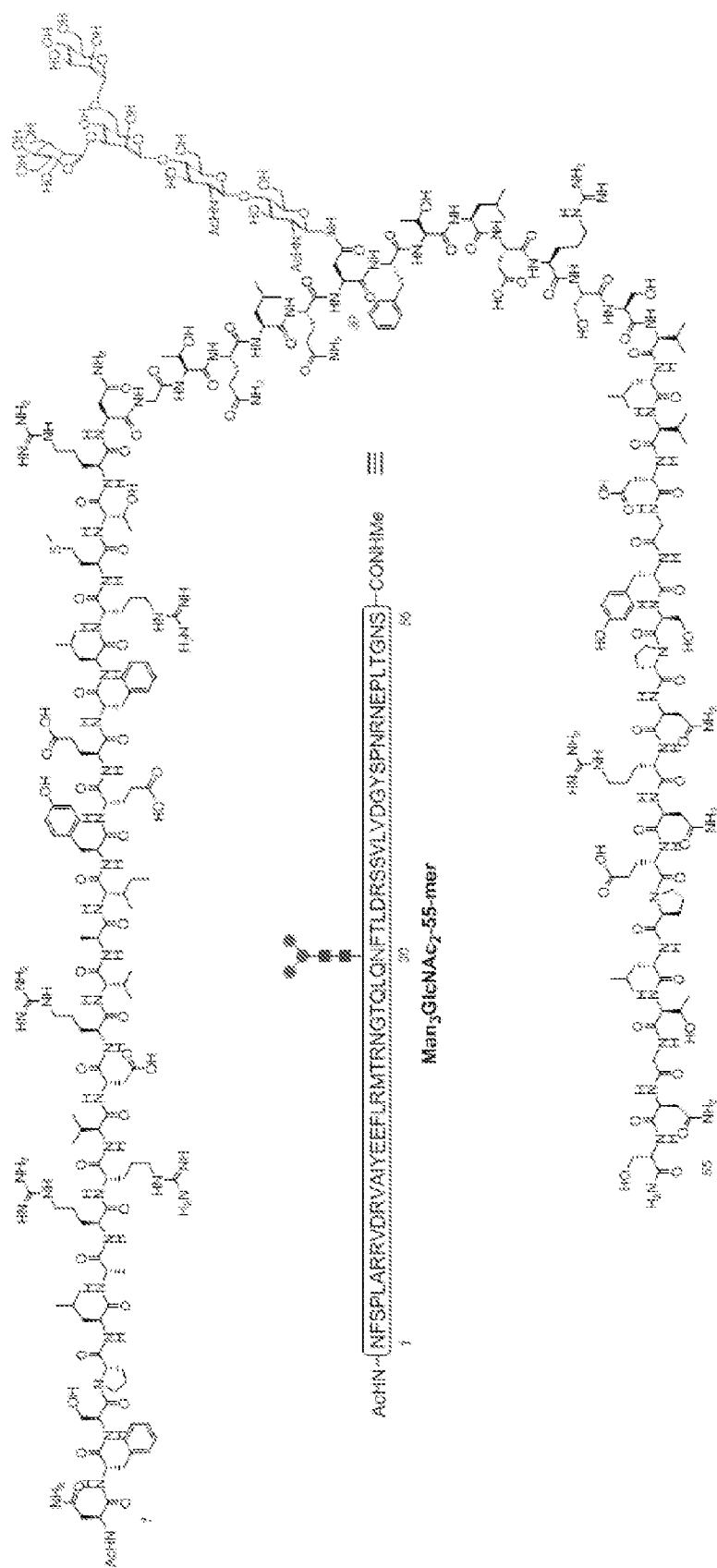


Fig. 29A

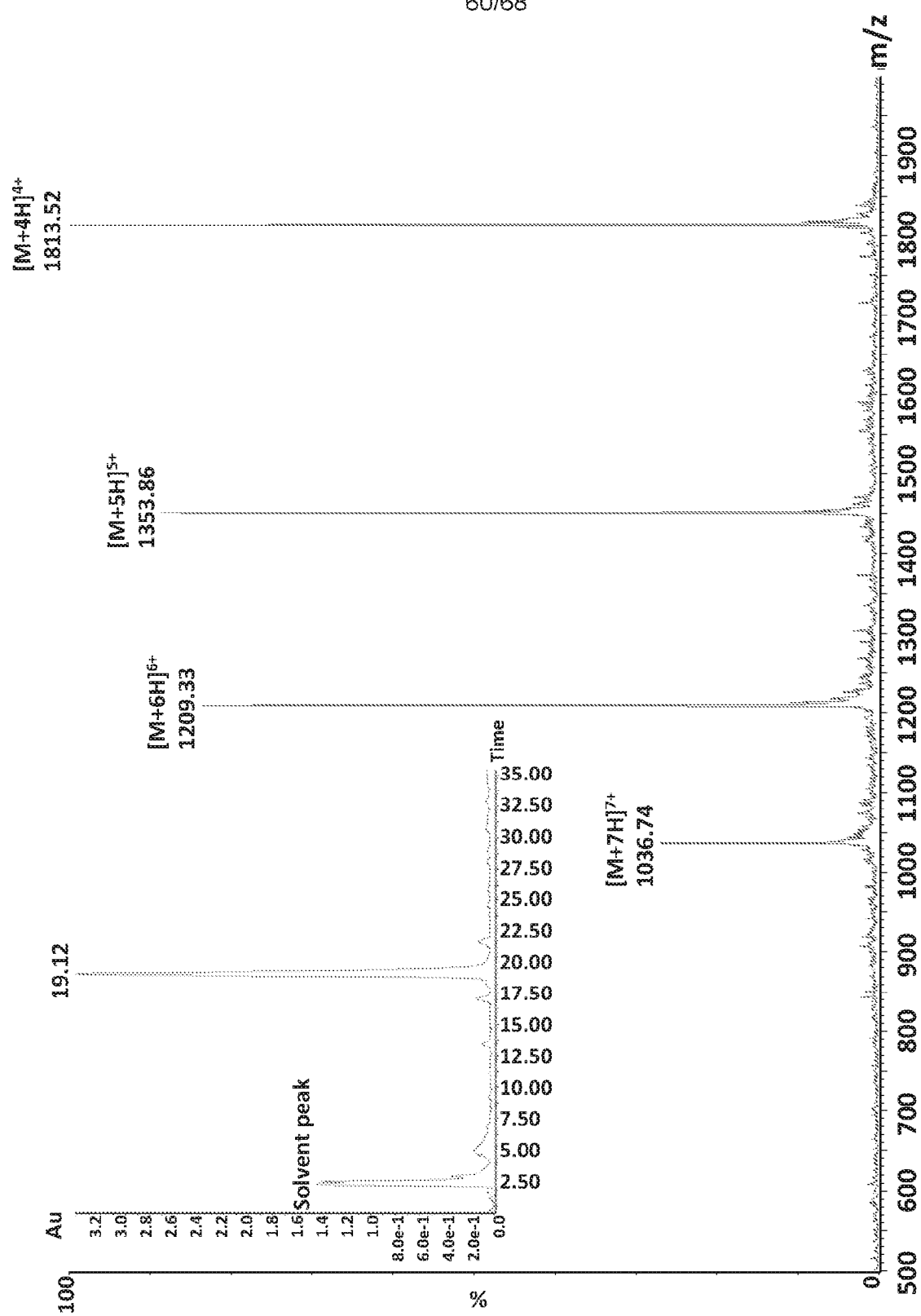


Fig. 29B

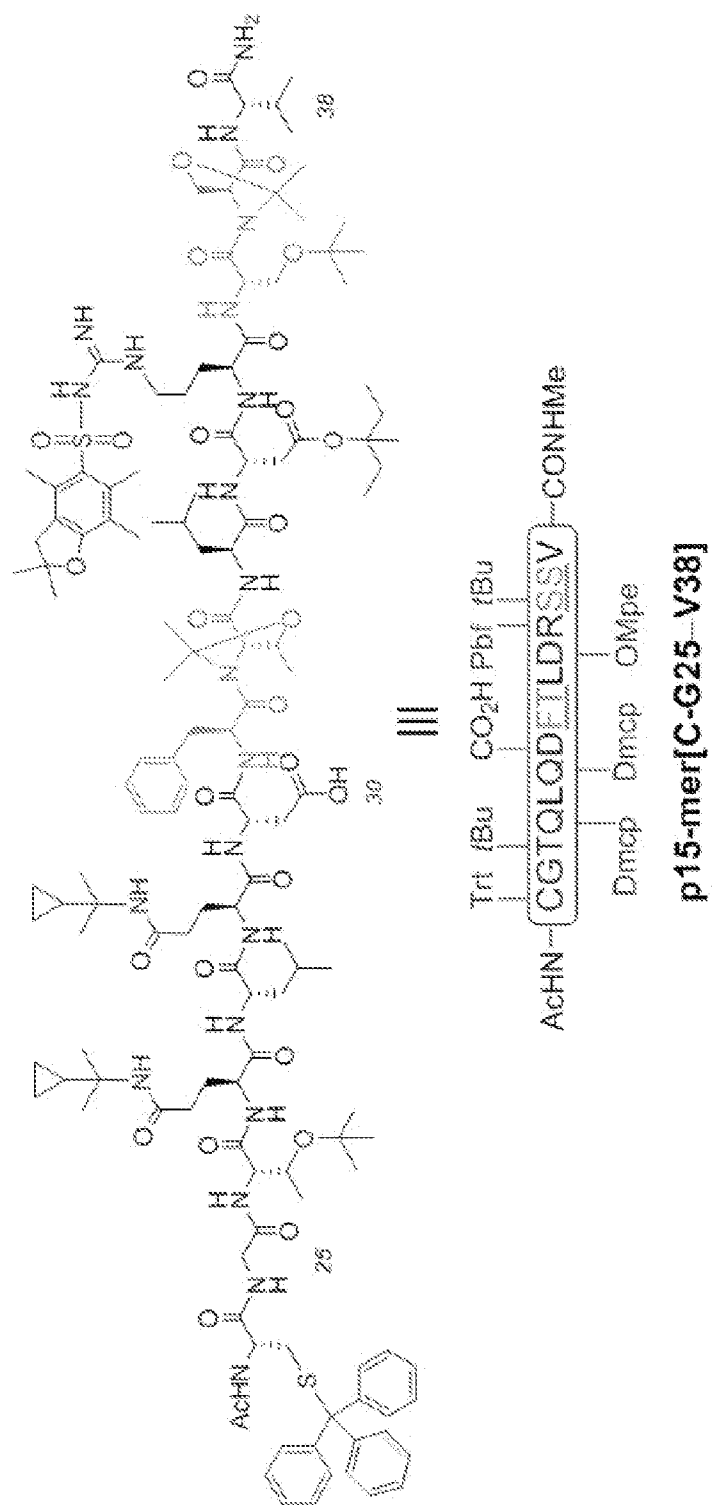


Fig. 30A

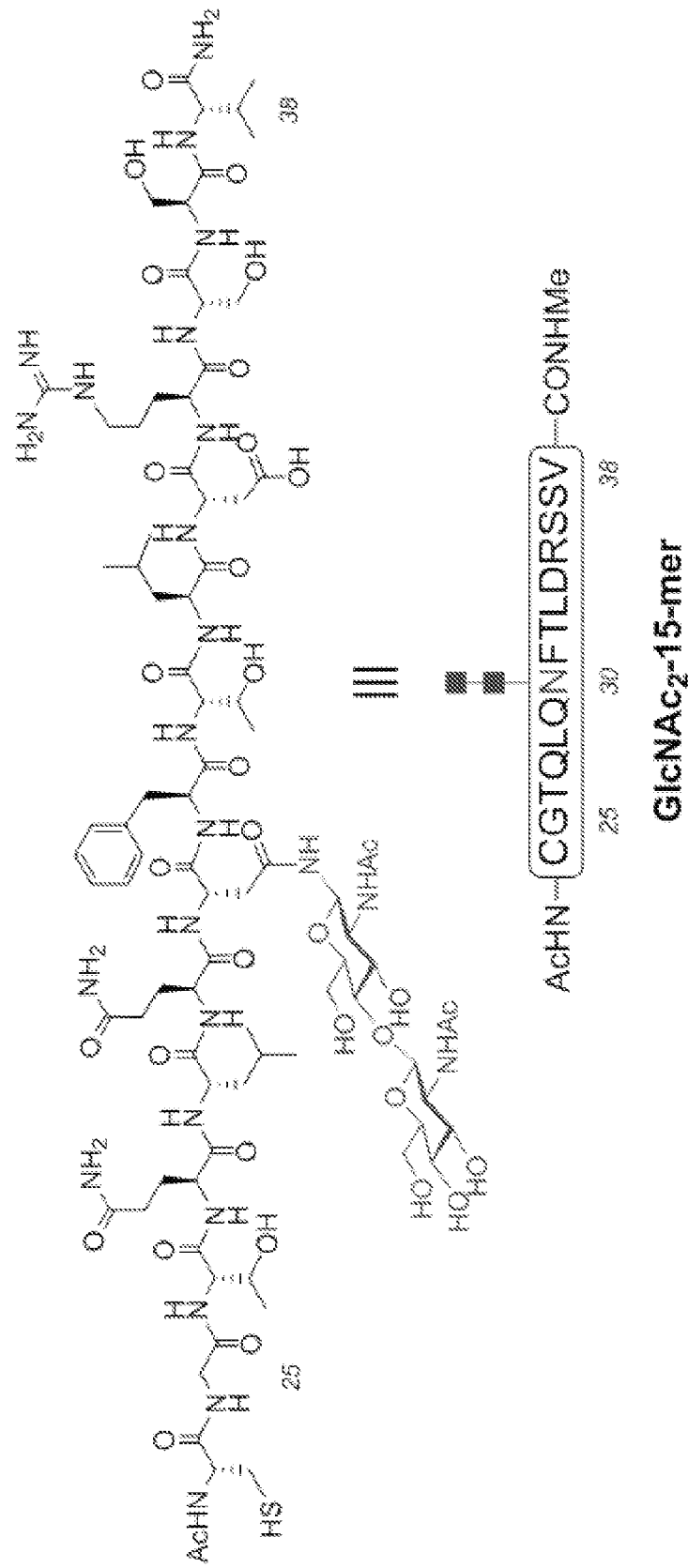


Fig. 30B

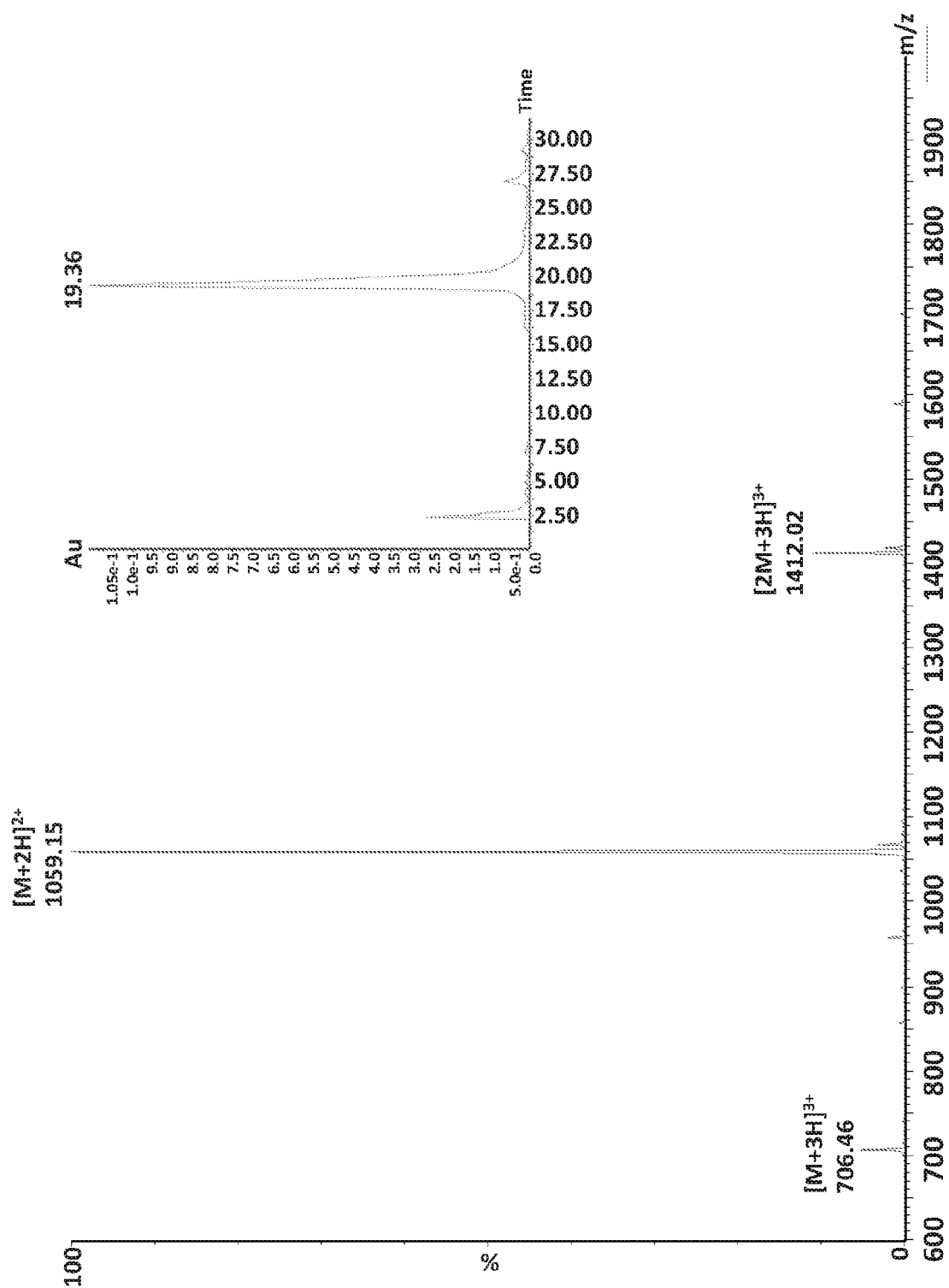


Fig. 30C

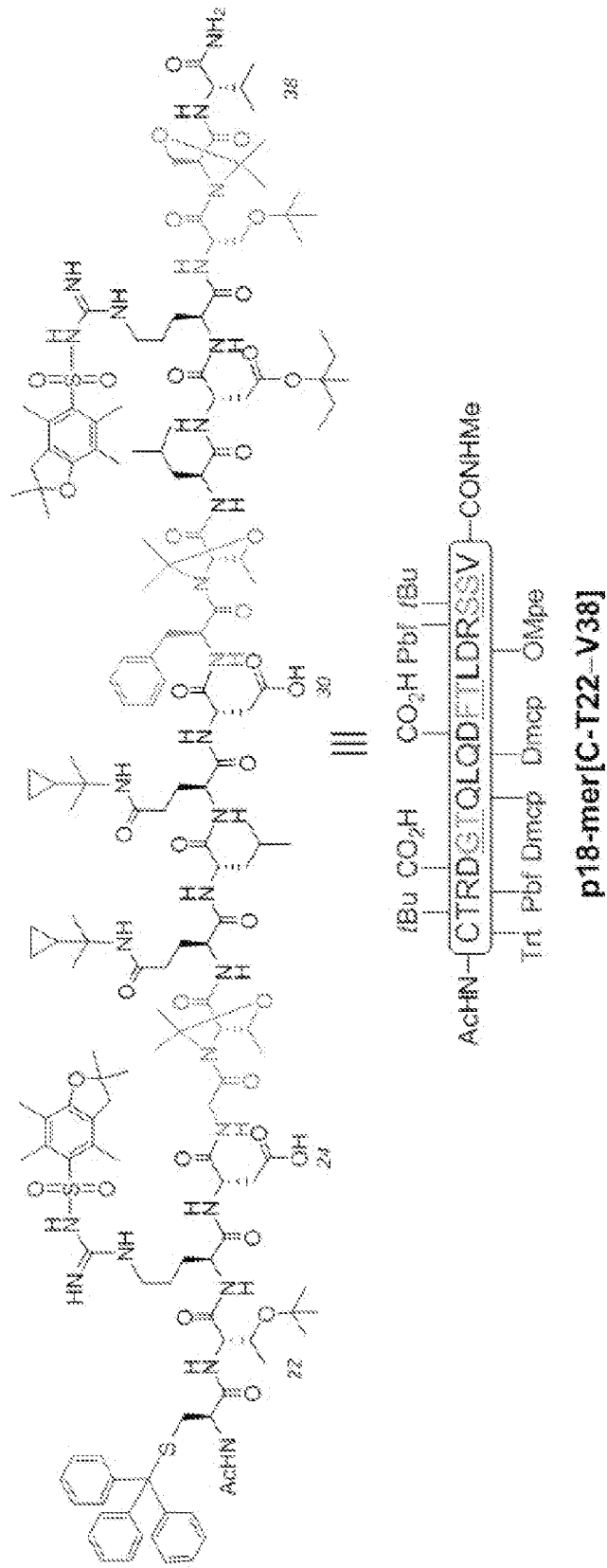


Fig. 31A

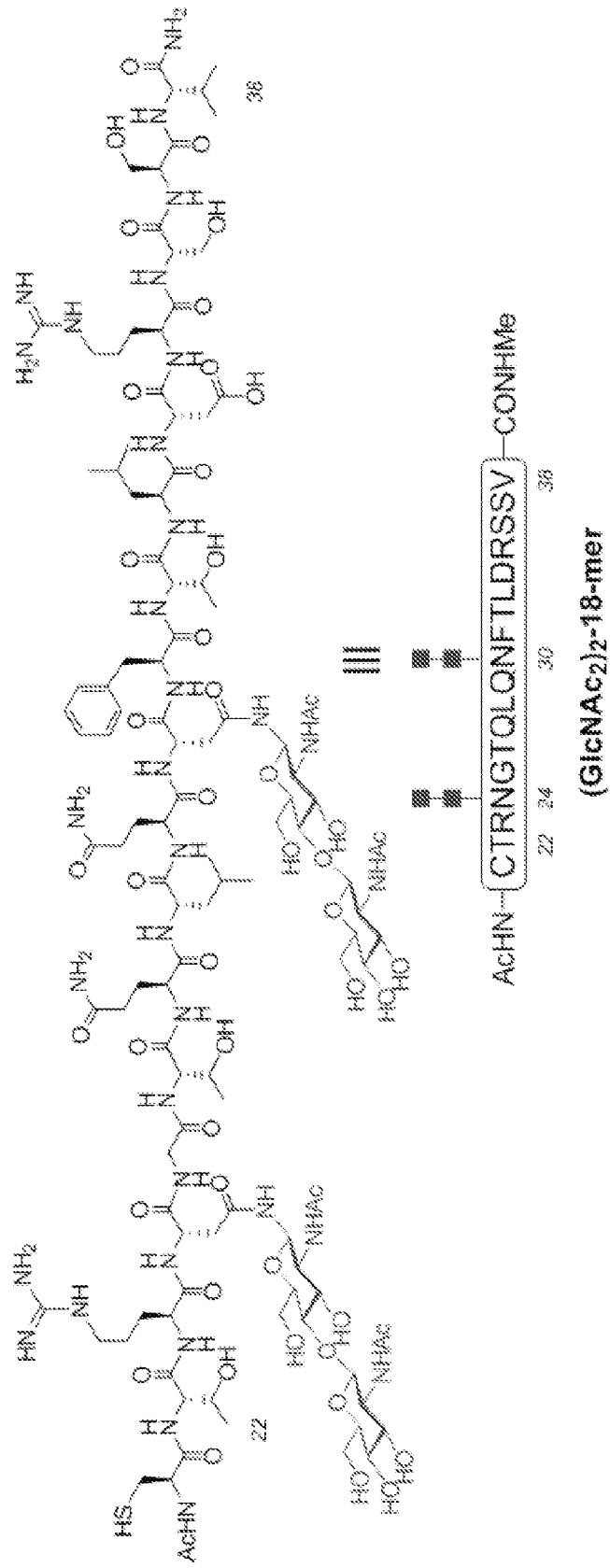


Fig. 31B

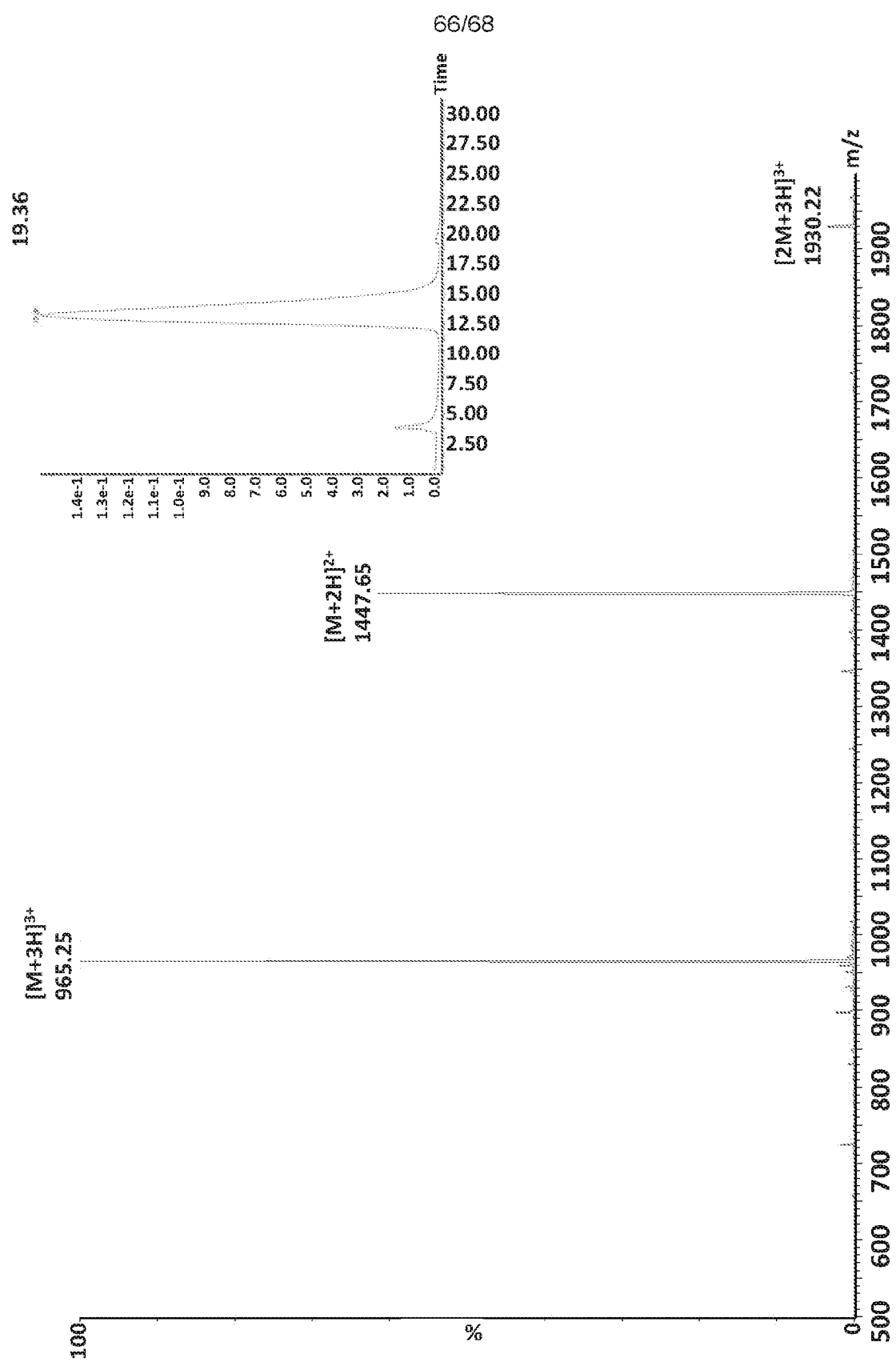


Fig. 31C

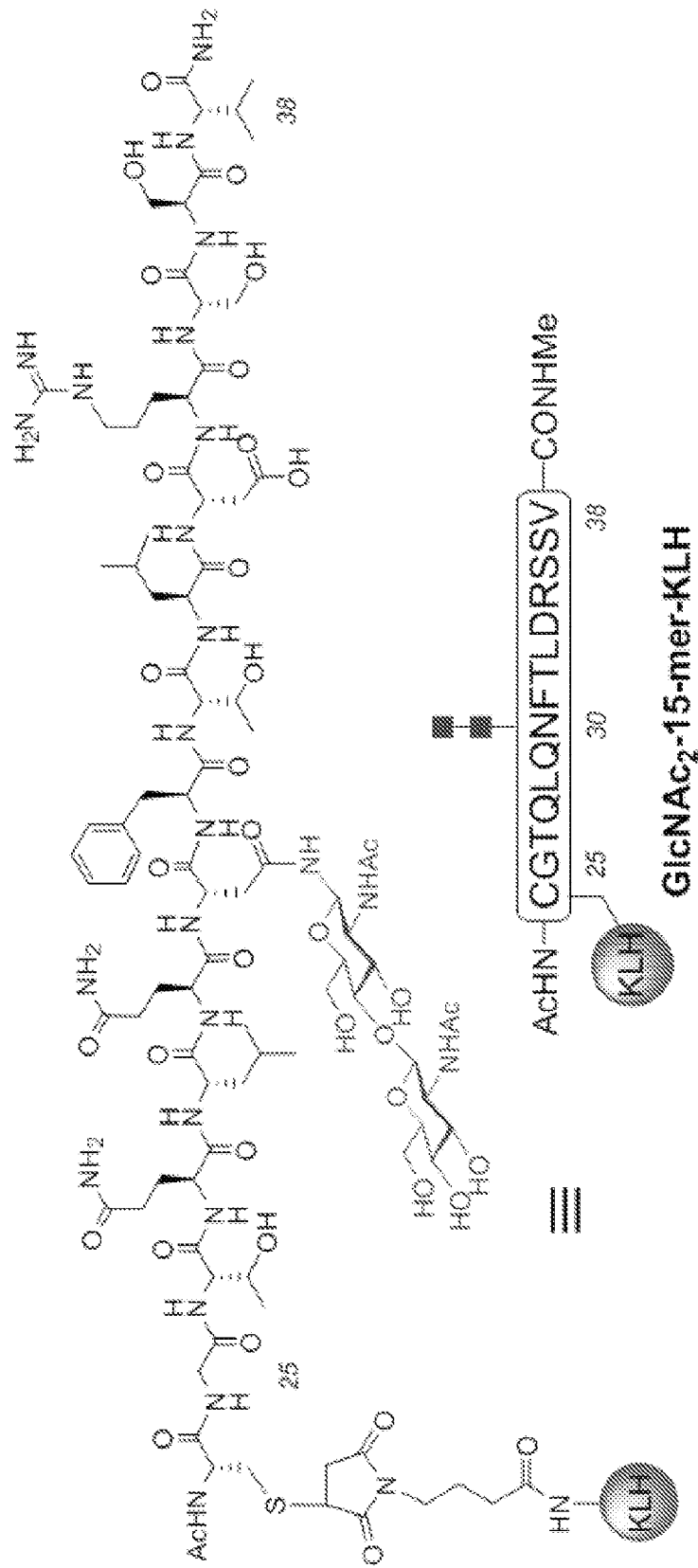


Fig. 32A

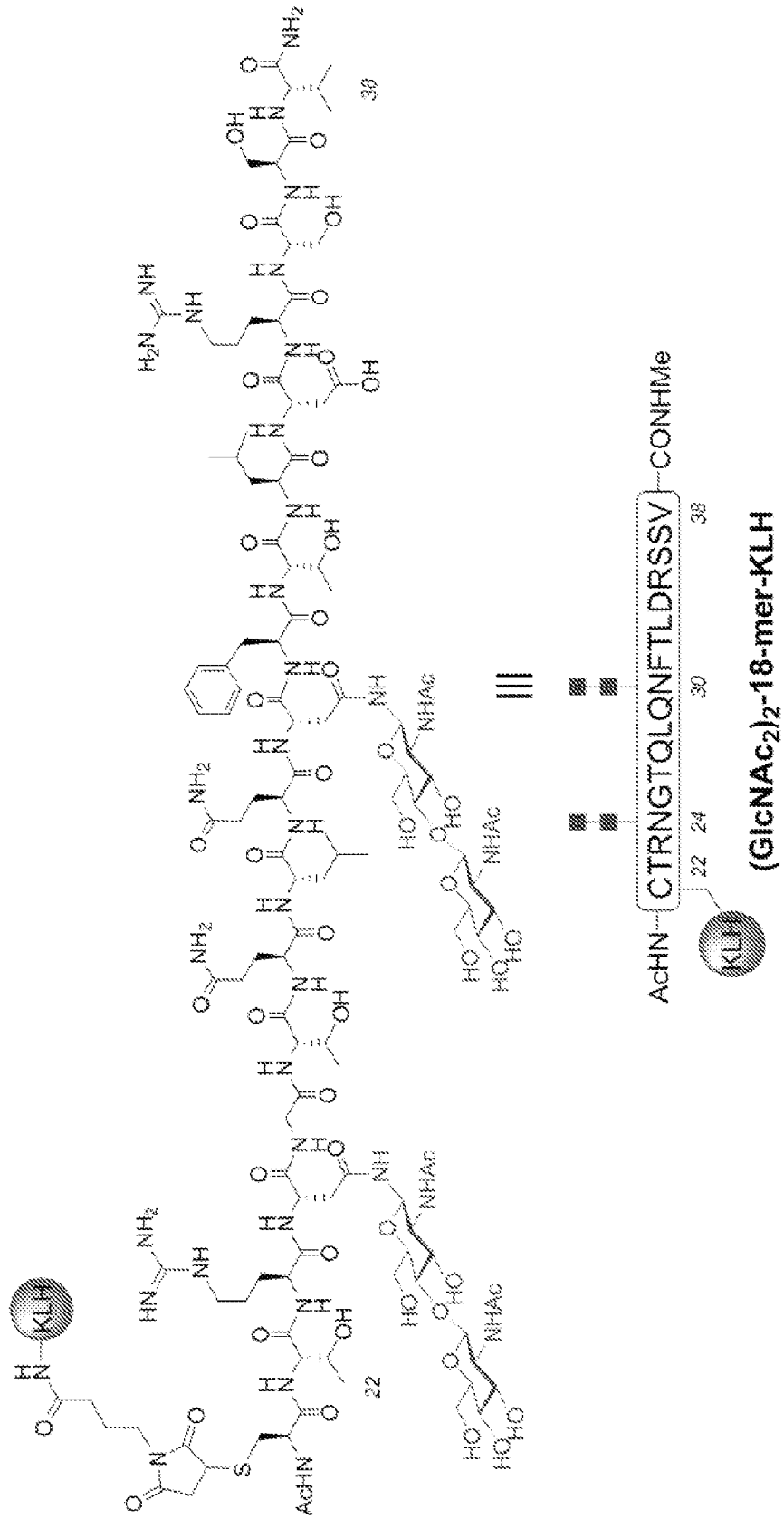


Fig. 32B