

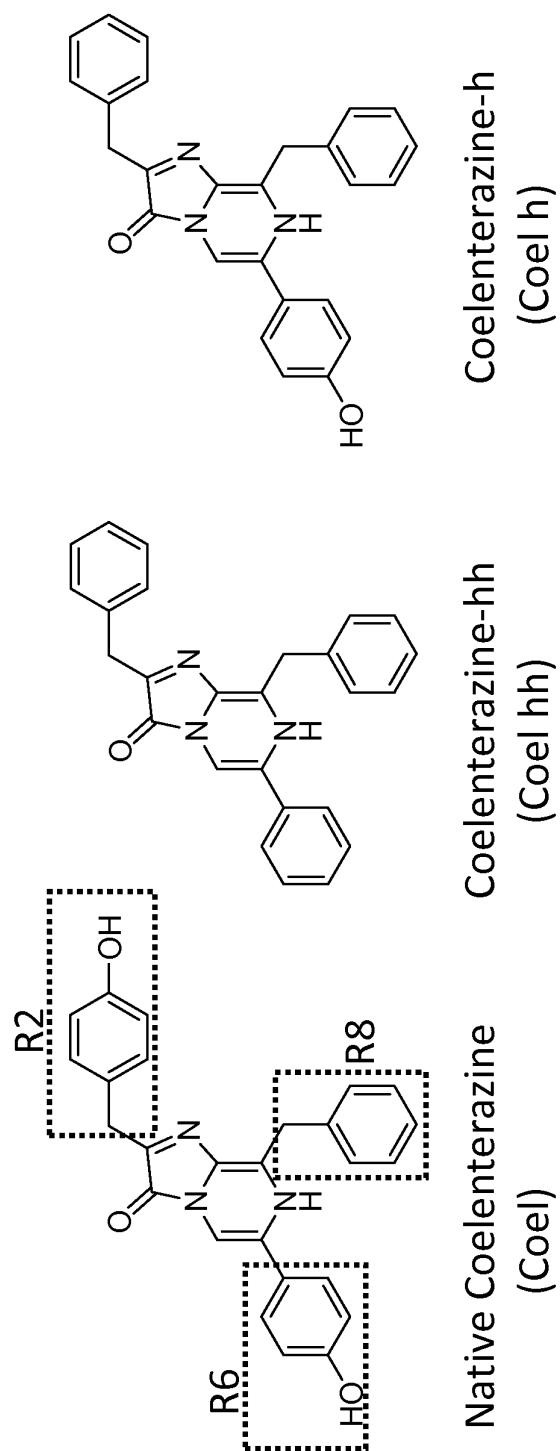


FIG. 1



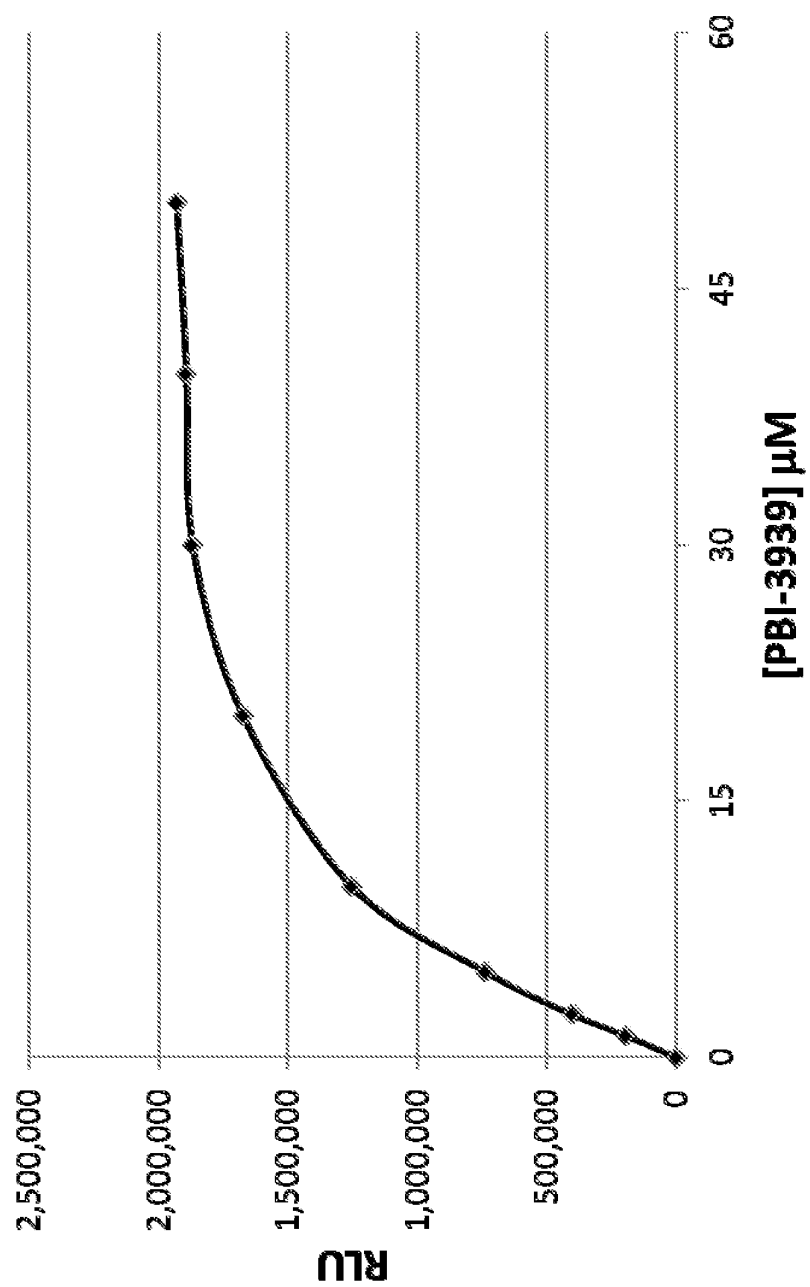


FIG. 3

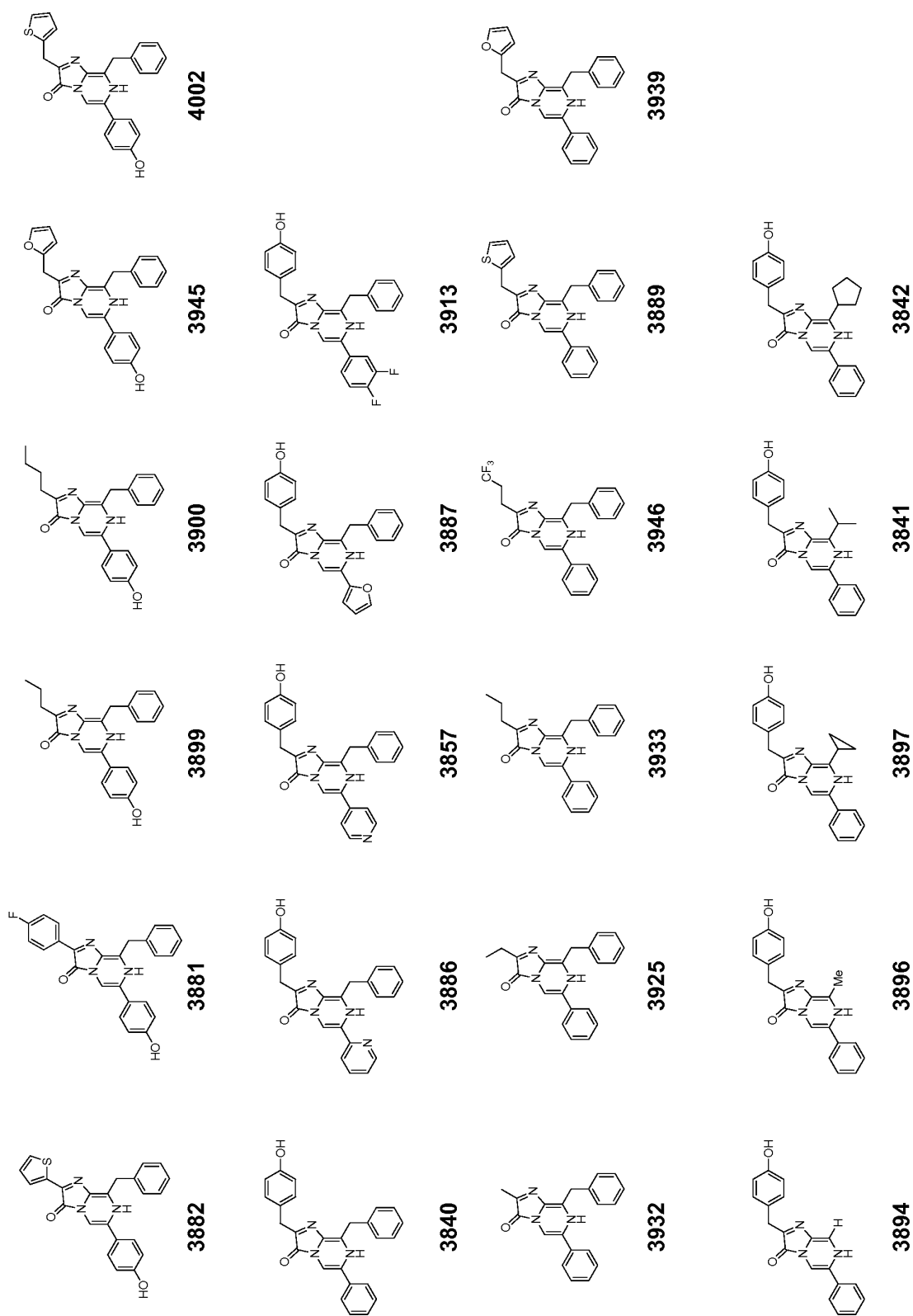


FIG. 4

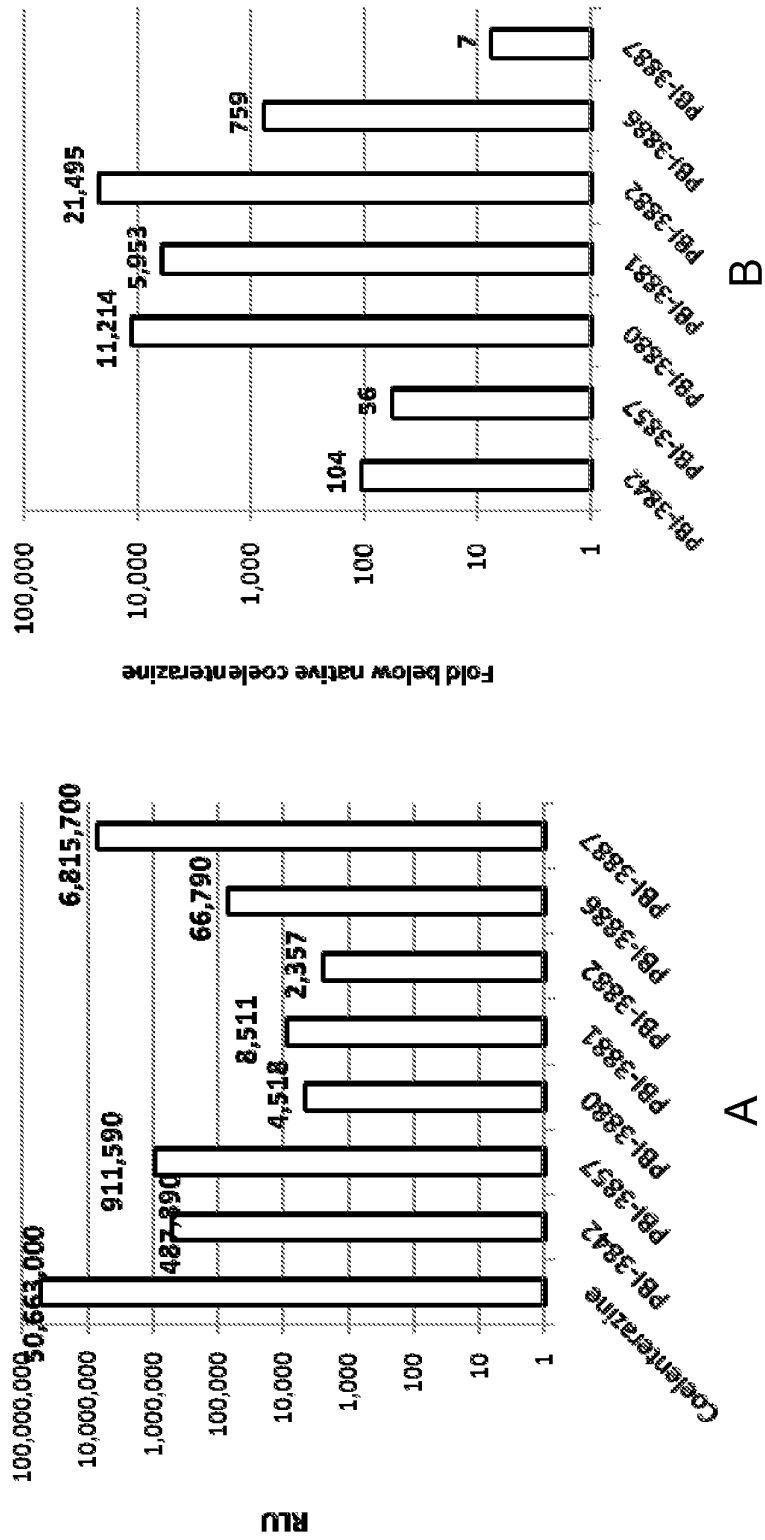


FIG. 5

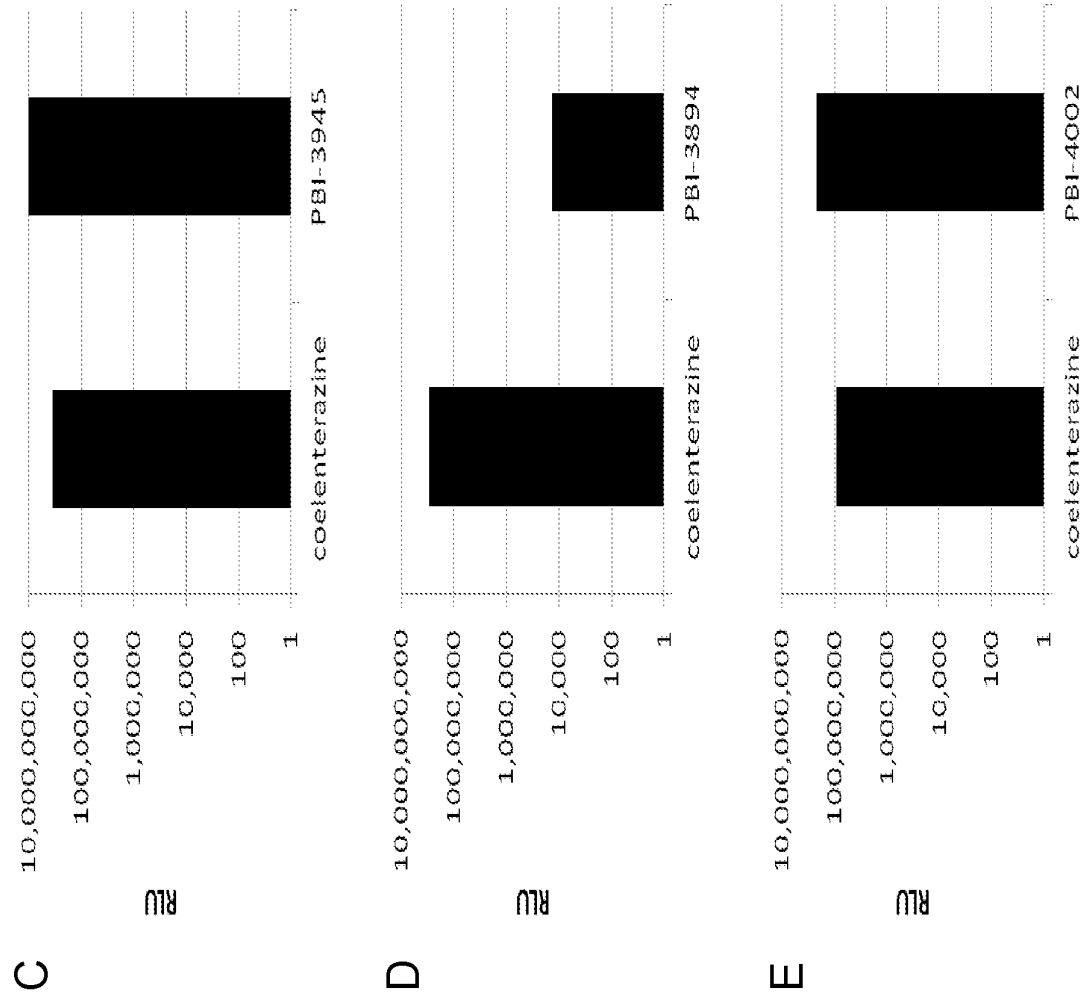


FIG. 5

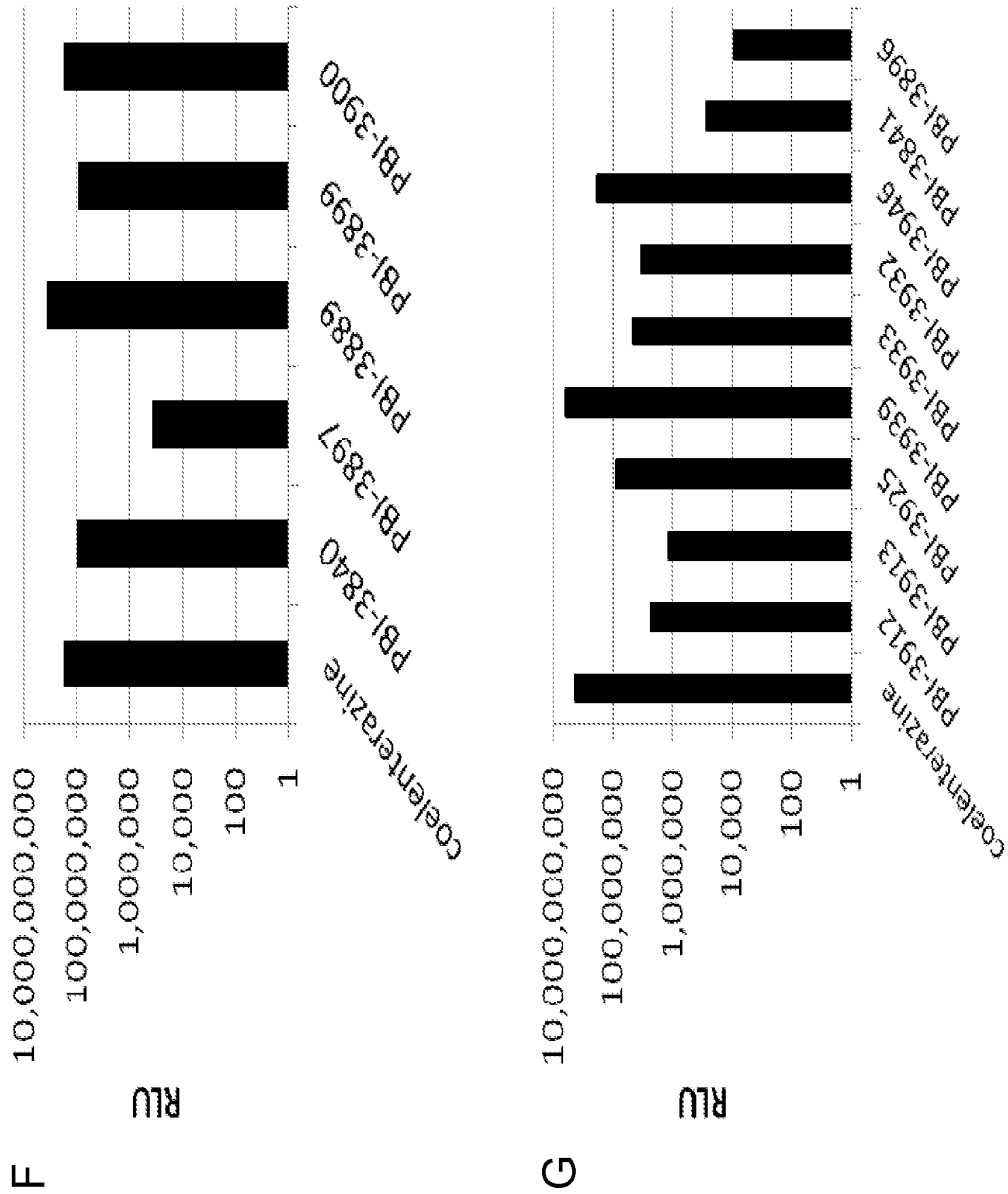


FIG. 5

| POSITION       | Clone(AA sequence)             | Colentel <sup>h</sup> | h <sub>h</sub> | 2-me        | PBI-3840     | PBI-3897     | PBI-3889     | PBI-3899     | PBI-3900    |
|----------------|--------------------------------|-----------------------|----------------|-------------|--------------|--------------|--------------|--------------|-------------|
| 15             | 03F2 (G15R)                    | 0.004                 | 0.059          | 0.024       | 0.004        | 0.008        | 0.003        | 0.003        | 0.011       |
| 18             | 40H11 (Q18L)                   | 0.38                  | 0.26           | 0.51        | 0.20         | 0.28         | 0.53         | 0.17         | 0.43        |
| 20             | 16C5 (Q20R)                    | <b>5.66</b>           | <b>1.96</b>    | <b>1.88</b> | <b>9.02</b>  | <b>4.72</b>  | <b>6.50</b>  | <b>4.92</b>  | <b>2.41</b> |
| 22             | 14C1 (L22I)                    | 0.17                  | 0.63           | 0.47        | 0.41         | 0.14         | 0.08         | 0.43         | 0.66        |
| 27             | 11E8 (L27V)                    | 0.34                  | <b>1.74</b>    | <b>1.88</b> | 0.55         | 0.35         | 0.03         | 0.88         | <b>2.22</b> |
| 54             | 18F8 (F54S)                    | 0.21                  | 0.40           | <b>1.78</b> | 0.13         | 0.53         | 0.55         | 0.18         | 0.69        |
| 54             | 29H7 (F54I)                    | <b>11.30</b>          | <b>1.71</b>    | <b>1.48</b> | <b>24.59</b> | <b>18.11</b> | <b>18.09</b> | <b>10.57</b> | <b>2.99</b> |
| 58             | 30E4 (V58I)                    | 0.27                  | 0.89           | 1.00        | 0.96         | 0.43         | 0.21         | 0.65         | 1.02        |
| 72             | 04G7 (L72Q)                    | <b>4.46</b>           | 1.25           | 1.15        | <b>9.36</b>  | <b>5.65</b>  | <b>8.84</b>  | <b>4.04</b>  | <b>1.53</b> |
| 75             | 28B2 (M75K)                    | <b>4.16</b>           | <b>1.57</b>    | <b>2.30</b> | <b>5.43</b>  | <b>4.92</b>  | <b>5.67</b>  | <b>4.03</b>  | <b>2.11</b> |
| 77             | 18C4 (F77T)                    | <b>1.62</b>           | 0.55           | 1.22        | <b>7.82</b>  | <b>2.06</b>  | <b>2.28</b>  | <b>1.33</b>  | 0.86        |
| 77             | 42E4 (F77C)                    | 0.13                  | 0.46           | 0.99        | <b>3.43</b>  | 0.20         | 0.02         | 0.56         | 0.58        |
| 77             | 50D12 (F77T)                   | <b>1.48</b>           | 0.52           | 1.15        | <b>7.75</b>  | <b>1.87</b>  | <b>2.20</b>  | <b>2.46</b>  | 0.78        |
| 89             | 35D11 (K89E)                   | <b>3.02</b>           | 0.59           | 1.26        | <b>5.27</b>  | <b>3.39</b>  | <b>5.19</b>  | <b>1.41</b>  | <b>2.32</b> |
| 90             | 10F2 (I90T)                    | <b>2.20</b>           | 0.48           | 0.66        | 0.00         | <b>3.90</b>  | <b>5.41</b>  | <b>1.36</b>  | 0.55        |
| 92             | 04A12 (L92H)                   | 0.89                  | 0.39           | 0.61        | <b>7.31</b>  | <b>3.72</b>  | <b>14.25</b> | 0.53         | 0.45        |
| 109            | 43F9 (Y109F)                   | 0.48                  | 0.33           | 0.97        | 0.36         | <b>1.49</b>  | <b>24.83</b> | 0.94         | 0.56        |
| 127            | 30D6 (V127A)                   | 0.23                  | 0.50           | 0.54        | 0.26         | 0.81         | 1.00         | 0.57         | 0.42        |
| 139            | 27B8 (D139G)                   | 0.01                  | 0.03           | 0.14        | 0.00         | 0.03         | 0.00         | 0.16         | 0.02        |
| 164            | 25A1 (C164S)                   | <b>2.47</b>           | <b>1.79</b>    | <b>1.53</b> | <b>2.74</b>  | <b>3.37</b>  | <b>5.10</b>  | <b>1.63</b>  | <b>2.87</b> |
| 14-89          | 25F2 (A14V,K89E)               | <b>3.05</b>           | 0.81           | 1.22        | <b>5.43</b>  | <b>3.82</b>  | <b>6.21</b>  | <b>1.58</b>  | <b>2.32</b> |
| 27-33          | 47D8(L27M,K33N)                | 0.59                  | <b>1.71</b>    | <b>2.05</b> | 0.85         | 0.95         | 0.27         | <b>2.12</b>  | <b>1.85</b> |
| 49-72          | 26F1 (E49K,L72Q)               | <b>6.19</b>           | <b>1.48</b>    | <b>1.35</b> | <b>10.72</b> | <b>6.90</b>  | <b>9.99</b>  | <b>1.32</b>  | <b>4.76</b> |
| 54-66          | 48G3 (F54I,S66T)               | <b>7.33</b>           | 1.06           | 0.91        | <b>16.39</b> | <b>11.27</b> | <b>8.28</b>  | <b>1.42</b>  | <b>6.63</b> |
| 54-67          | 35B8 (F54S,G67S)               | 0.33                  | 0.75           | <b>2.49</b> | 0.19         | 0.78         | 0.87         | <b>2.25</b>  | 0.27        |
| 55-117         | 05H8 (D55G,I117F)              | 0.24                  | 0.53           | 0.62        | 0.00         | 0.25         | 0.22         | 0.65         | 0.21        |
| 56-93          | 46A11(I56V,H93R)               | 0.35                  | 0.61           | <b>1.53</b> | 0.22         | 0.42         | 0.18         | <b>1.50</b>  | 0.36        |
| 68-75          | 04E2 (F68S,M75K)               | <b>3.43</b>           | 1.20           | <b>1.53</b> | <b>4.37</b>  | <b>3.91</b>  | <b>4.12</b>  | <b>1.62</b>  | <b>3.24</b> |
| 77-126         | 27B9 (F77T,T126R)              | 0.87                  | 0.15           | 0.69        | <b>4.87</b>  | 0.80         | 1.00         | 0.79         | 0.32        |
| 77-39          | 33E2 (F77T,T39I)               | 0.95                  | 0.34           | 0.69        | <b>6.30</b>  | <b>1.32</b>  | <b>1.64</b>  | <b>0.74</b>  | <b>1.47</b> |
| 89-148         | 50E7 (K89E,S148T)              | <b>1.81</b>           | 0.48           | 0.85        | <b>3.74</b>  | <b>2.23</b>  | <b>3.20</b>  | <b>0.94</b>  | <b>1.59</b> |
| 90-77          | 02D10 (I90V,F77T)              | <b>2.05</b>           | 0.70           | 1.17        | <b>17.39</b> | <b>6.32</b>  | <b>10.88</b> | 1.05         | <b>2.37</b> |
| 106-164        | 17E6 (M106K,C164S)             | <b>4.14</b>           | <b>2.46</b>    | <b>2.52</b> | <b>3.77</b>  | <b>5.06</b>  | <b>7.72</b>  | <b>2.71</b>  | <b>3.80</b> |
| 23-58-169      | 47G12 (E23K,V58L,A169V)        | 0.06                  | 0.44           | 0.49        | <b>1.49</b>  | 0.10         | 0.01         | 0.68         | 0.58        |
| 33-76-89       | 17C5 (K33N,I76N,K89E)          | 1.27                  | 0.68           | <b>2.02</b> | <b>1.33</b>  | <b>2.22</b>  | 1.12         | <b>2.24</b>  | 1.07        |
| 68-90-145      | 04D10 (F68S,I90V,P145L)        | <b>4.17</b>           | <b>1.40</b>    | <b>1.65</b> | 0.00         | <b>6.09</b>  | <b>5.96</b>  | <b>1.72</b>  | <b>3.82</b> |
| 59-109-113-136 | 41H10 (I59T,Y109F,P113T,L136M) | 0.06                  | 0.06           | 0.26        | 0.04         | 0.46         | <b>8.09</b>  | 0.25         | 0.06        |

FIG. 6A

| Position       | Clone(AA sequence)             | Colenterazine | H     | h,h   | v     | PBI-3912 | PBI-3913 | PBI-3925 | PBI-3897 | PBI-3899 |
|----------------|--------------------------------|---------------|-------|-------|-------|----------|----------|----------|----------|----------|
| 15             | 03F2 (G15R)                    | 0.003         | 0.040 | 0.018 | 0.022 | 0.005    | 0.019    | 0.005    | 0.002    | 0.003    |
| 18             | 40H11 (Q18L)                   | 0.20          | 0.18  | 0.30  | 0.11  | 0.13     | 0.14     | 0.06     | 3.24     | 0.09     |
| 20             | 16C5 (Q20R)                    | 4.08          | 1.30  | 1.14  | 5.25  | 5.27     | 5.34     | 4.83     | 3.33     | 3.32     |
| 22             | 14C1 (L22I)                    | 0.13          | 0.56  | 0.40  | 0.13  | 0.10     | 0.11     | 0.21     | 0.06     | 0.39     |
| 27             | 11E8 (L27V)                    | 0.16          | 1.01  | 0.95  | 0.22  | 0.07     | 0.08     | 0.26     | 0.02     | 0.45     |
| 54             | 18F8 (F54S)                    | 0.16          | 0.37  | 1.44  | 0.55  | 0.17     | 0.22     | 0.26     | 0.38     | 0.14     |
| 54             | 29H7 (F54I)                    | 10.34         | 1.45  | 1.06  | 8.28  | 18.52    | 15.79    | 18.92    | 11.15    | 9.62     |
| 58             | 30E4 (V58I)                    | 0.17          | 0.63  | 0.69  | 0.20  | 0.19     | 0.21     | 0.83     | 0.13     | 0.43     |
| 72             | 04G7 (L72Q)                    | 2.96          | 1.02  | 1.03  | 0.69  | 5.81     | 5.81     | 5.65     | 7.42     | 3.09     |
| 75             | 28B2 (M75K)                    | 2.93          | 1.35  | 1.53  | 1.06  | 3.98     | 3.89     | 3.86     | 4.73     | 2.93     |
| 77             | 18C4 (F77T)                    | 1.01          | 0.53  | 0.85  | 1.31  | 0.78     | 0.74     | 7.08     | 1.69     | 2.30     |
| 77             | 42E4 (F77C)                    | 0.10          | 0.34  | 0.70  | 0.61  | 0.03     | 0.04     | 0.88     | 0.01     | 0.41     |
| 77             | 50D12 (F77T)                   | 0.88          | 0.46  | 0.72  | 1.21  | 0.62     | 0.60     | 5.94     | 1.41     | 1.92     |
| 89             | 35D11 (K89E)                   | 2.17          | 0.53  | 0.89  | 0.76  | 3.64     | 3.98     | 3.51     | 3.89     | 1.77     |
| 90             | 10F2 (I90T)                    | 1.86          | 0.39  | 0.49  | 1.71  | 10.44    | 11.10    | 3.76     | 3.61     | 1.20     |
| 92             | 04A12 (L92H)                   | 0.65          | 0.08  | 0.27  | 27.76 | 4.48     | 2.79     | 5.01     | 6.72     | 0.60     |
| 109            | 43F9 (V109F)                   | 0.36          | 0.20  | 0.76  | 0.84  | 0.73     | 0.60     | 1.04     | 18.04    | 0.31     |
| 127            | 30D6 (V127A)                   | 0.16          | 0.31  | 0.30  | 0.16  | 0.32     | 0.45     | 0.50     | 0.43     | 0.15     |
| 139            | 27B8 (D139G)                   | 0.00          | 0.02  | 0.08  | 0.10  | 0.02     | 0.04     | 0.01     | 0.00     | 0.00     |
| 164            | 25A1 (C164S)                   | 3.10          | 1.71  | 1.47  | 3.91  | 3.04     | 3.29     | 3.43     | 4.33     | 3.14     |
| 14-89          | 25F2 (A14V,K89E)               | 3.31          | 0.75  | 1.16  | 1.34  | 5.26     | 5.90     | 5.28     | 4.68     | 2.46     |
| 27-33          | 47D8(L27M,K33N)                | 0.52          | 1.41  | 1.46  | 1.55  | 0.41     | 0.41     | 1.21     | 0.17     | 1.05     |
| 49-72          | 26F1 (E49K,L72Q)               | 3.61          | 1.09  | 0.76  | 2.24  | 6.83     | 6.59     | 6.42     | 6.33     | 3.40     |
| 54-66          | 48G3 (F54I,S66T)               | 10.15         | 1.40  | 1.01  | 7.67  | 17.73    | 14.90    | 18.29    | 10.14    | 9.44     |
| 54-67          | 35B8 (F54S,G67S)               | 0.23          | 0.46  | 1.73  | 1.28  | 0.23     | 0.29     | 0.32     | 0.42     | 0.18     |
| 55-117         | 05H8 (D55G,I117F)              | 0.18          | 0.38  | 0.46  | 0.44  | 0.12     | 0.14     | 0.10     | 0.14     | 0.16     |
| 56-93          | 46A11(I56V,H93R)               | 0.24          | 0.39  | 0.96  | 0.88  | 0.10     | 0.11     | 0.21     | 0.11     | 0.24     |
| 68-75          | 04E2 (F68S,M75K)               | 2.39          | 1.01  | 1.04  | 0.74  | 3.01     | 3.04     | 3.61     | 3.58     | 2.47     |
| 77-126         | 27B9 (F77T,I126R)              | 0.34          | 0.15  | 0.37  | 0.51  | 0.28     | 0.24     | 2.16     | 0.58     | 0.74     |
| 77-39          | 33E2 (F77T,I39I)               | 0.50          | 0.24  | 0.35  | 0.67  | 0.42     | 0.38     | 3.27     | 0.96     | 0.96     |
| 89-148         | 50E7 (K89E,S148T)              | 2.97          | 0.66  | 1.15  | 1.50  | 4.57     | 5.01     | 4.36     | 3.91     | 2.28     |
| 90-77          | 02D10 (I90V,F77T)              | 1.44          | 0.63  | 0.80  | 3.79  | 2.80     | 2.47     | 24.43    | 7.82     | 2.14     |
| 106-164        | 17E6 (M106K,C164S)             | 2.71          | 1.50  | 1.52  | 4.80  | 2.79     | 2.92     | 2.93     | 3.47     | 2.48     |
| 23-58-169      | 47G12 (E23K,V58L,A169V)        | 0.05          | 0.41  | 0.39  | 0.31  | 0.03     | 0.03     | 1.24     | 0.01     | 0.48     |
| 33-76-89       | 17C5 (K33N,I76N,K89E)          | 1.53          | 0.63  | 1.74  | 2.16  | 0.86     | 0.94     | 1.38     | 0.91     | 1.03     |
| 68-90-145      | 04D10 (F68S,I90V,P145L)        | 2.63          | 0.88  | 0.95  | 2.74  | 4.40     | 4.42     | 5.96     | 4.82     | 2.48     |
| 59-109-113-136 | 41H10 (I59T,Y109F,P113T,L136M) | 0.08          | 0.07  | 0.29  | 0.20  | 0.23     | 0.22     | 0.41     | 7.52     | 0.07     |

FIG. 6B

| POSITION       | Clone(AA sequence)                | coel         | h           | h,h         | 2 me         | PBI-3889    | PBI-3939    | PBI-3933    | PBI-3932     | PBI-3946    | PBI-3897     | PBI-3841     | PB-3896      | PBI-3925     |
|----------------|-----------------------------------|--------------|-------------|-------------|--------------|-------------|-------------|-------------|--------------|-------------|--------------|--------------|--------------|--------------|
| 15             | 03F2 (G15R)                       | 0.003        | 0.041       | 0.021       | 0.005        | 0.025       | 0.019       | 0.003       | 0.009        | 0.003       | 0.002        | 0.003        | 0.013        | 0.004        |
| 18             | 40H11 (Q18L)                      | 0.31         | 0.34        | 0.48        | 0.19         | 0.43        | 0.39        | 0.09        | 0.08         | 0.09        | <b>3.86</b>  | <b>4.08</b>  | <b>61.25</b> | 0.09         |
| 20             | 16C5 (Q20R)                       | <b>2.84</b>  | 1.04        | 0.92        | <b>3.91</b>  | 0.86        | 1.15        | <b>2.79</b> | <b>4.70</b>  | <b>2.18</b> | <b>2.70</b>  | <b>3.59</b>  | <b>3.78</b>  | <b>3.81</b>  |
| 22             | 14C1 (L22I)                       | 0.12         | 0.52        | 0.39        | 0.30         | 0.33        | 0.29        | 0.20        | 0.15         | 0.20        | 0.06         | 0.09         | 0.12         | 0.20         |
| 27             | 11E8 (L27V)                       | 0.25         | <b>1.64</b> | <b>1.48</b> | 0.40         | <b>1.53</b> | <b>1.44</b> | 0.47        | 0.52         | 0.61        | 0.03         | 0.04         | 0.09         | 0.40         |
| 54             | 18F8 (F54S)                       | 0.2          | 0.6         | <b>1.6</b>  | 0.1          | <b>1.4</b>  | 1.3         | 0.4         | 0.3          | 0.5         | 0.4          | 0.4          | 0.5          | 0.3          |
| 54             | 29H7 (F54I)                       | <b>6.77</b>  | 1.18        | 0.78        | out of range |             |             |             |              |             |              |              |              |              |
| 58             | 30E4 (V58I)                       | 0.17         | 0.66        | 0.72        | 0.62         | 0.80        | 0.91        | 0.63        | 0.90         | 0.48        | 0.12         | 0.14         | 0.27         | 0.83         |
| 72             | 04G7 (L72Q)                       | <b>4.07</b>  | 1.27        | 0.93        | <b>7.23</b>  | 1.08        | 1.27        | <b>5.03</b> | <b>7.66</b>  | <b>3.14</b> | <b>6.40</b>  | <b>7.30</b>  | <b>6.20</b>  | <b>6.85</b>  |
| 75             | 28B2 (M75K)                       | <b>3.25</b>  | <b>1.44</b> | <b>1.68</b> | <b>3.97</b>  | <b>1.86</b> | <b>2.01</b> | <b>3.77</b> | <b>4.33</b>  | <b>3.33</b> | <b>3.89</b>  | <b>3.85</b>  | <b>3.41</b>  | <b>4.15</b>  |
| 77             | 18C4 (F77T)                       | 0.86         | 0.48        | 0.74        | <b>4.98</b>  | 0.96        | 0.81        | <b>3.37</b> | <b>4.84</b>  | <b>1.81</b> | <b>1.32</b>  | 0.56         | 0.91         | <b>5.04</b>  |
| 77             | 42E4 (F77C)                       | 0.07         | 0.30        | 0.65        | <b>1.89</b>  | 0.77        | 0.46        | 0.50        | <b>2.18</b>  | 0.34        | 0.02         | 0.02         | 0.08         | 0.63         |
| 77             | 50D12 (F77T)                      | 0.87         | 0.50        | 0.76        | <b>4.89</b>  | 0.99        | 0.85        | <b>3.45</b> | <b>4.92</b>  | <b>1.86</b> | 1.30         | 0.55         | 0.88         | <b>5.14</b>  |
| 89             | 35D11 (K89E)                      | <b>2.17</b>  | 0.60        | 0.88        | <b>3.39</b>  | 0.97        | 1.02        | <b>2.28</b> | <b>3.65</b>  | <b>2.09</b> | <b>3.17</b>  | <b>2.95</b>  | <b>2.56</b>  | <b>3.01</b>  |
| 90             | 10F2 (I90T)                       | <b>1.68</b>  | 0.37        | 0.51        | <b>2.60</b>  | 0.72        | 0.62        | <b>2.08</b> | <b>4.64</b>  | <b>1.35</b> | <b>2.99</b>  | <b>1.93</b>  | <b>4.51</b>  | <b>3.57</b>  |
| 92             | 04A12 (L92H)                      | 0.59         | 0.03        | 0.24        | <b>5.44</b>  | 0.23        | 0.11        | <b>1.86</b> | <b>3.25</b>  | 0.88        | <b>8.81</b>  | <b>23.30</b> | <b>22.13</b> | <b>3.53</b>  |
| 109            | 43F9 (Y109F)                      | 0.25         | 0.20        | 0.65        | 0.11         | 0.73        | 0.64        | 0.64        | 0.71         | 0.44        | <b>13.07</b> | <b>4.80</b>  | 1.20         | 0.85         |
| 127            | 30D6 (V127A)                      | 0.22         | 0.40        | 0.46        | 0.15         | 0.48        | 0.52        | 0.65        | 0.75         | 0.62        | 0.50         | 0.55         | 0.53         | 0.66         |
| 139            | 27B8 (D139G)                      | 0.00         | 0.02        | 0.11        | 0.01         | 0.10        | 0.09        | 0.02        | 0.01         | 0.02        | 0.01         | 0.01         | 0.04         | 0.01         |
| 164            | 25A1 (C164S)                      | <b>2.10</b>  | <b>1.30</b> | 1.12        | <b>1.71</b>  | 1.28        | <b>1.30</b> | <b>2.30</b> | <b>2.26</b>  | <b>2.11</b> | <b>3.14</b>  | <b>5.76</b>  | <b>1.96</b>  | <b>2.66</b>  |
| 14-89          | 25F2 (A14V,K89E)                  | <b>1.93</b>  | 0.53        | 0.73        | <b>2.80</b>  | 0.98        | 1.00        | <b>2.15</b> | <b>3.78</b>  | <b>1.88</b> | <b>3.07</b>  | <b>2.32</b>  | <b>2.94</b>  | <b>3.01</b>  |
| 27-33          | 47D8(L27M,K33N)                   | 0.38         | 1.26        | <b>1.37</b> | 0.46         | <b>1.73</b> | <b>1.79</b> | 1.27        | 0.92         | <b>1.46</b> | 0.16         | 0.21         | 0.25         | 1.02         |
| 49-72          | 26F1 (E49K,L72Q)                  | <b>4.26</b>  | 1.22        | 0.91        | <b>7.26</b>  | 1.10        | 1.26        | <b>5.21</b> | <b>7.70</b>  | <b>3.23</b> | <b>6.66</b>  | <b>7.28</b>  | <b>6.41</b>  | <b>6.91</b>  |
| 54-66          | 48G3 (F54I, S66T)                 | <b>10.93</b> | <b>1.88</b> | 1.21        | out of range |             |             |             |              |             |              |              |              |              |
| 54-67          | 35B8 (F54S,G67S)                  | 0.19         | 0.59        | <b>1.64</b> | 0.10         | <b>1.44</b> | 1.30        | 0.47        | 0.30         | 0.58        | 0.43         | 0.51         | 0.47         | 0.32         |
| 55-117         | 05H8 (D55G,I117F)                 | 0.17         | 0.37        | 0.49        | 0.08         | 0.51        | 0.54        | 0.15        | 0.09         | 0.19        | 0.12         | 0.10         | 0.13         | 0.10         |
| 56-93          | 46A11(I56V,H93R)                  | 0.21         | 0.45        | 0.99        | 0.12         | 0.97        | 0.80        | 0.26        | 0.13         | 0.14        | 0.09         | 0.06         | 0.15         | 0.18         |
| 68-75          | 04E2 (F68S,M75K)                  | <b>2.80</b>  | 1.14        | 1.27        | <b>4.47</b>  | <b>1.50</b> | <b>1.62</b> | <b>3.71</b> | <b>4.35</b>  | <b>3.04</b> | <b>3.91</b>  | <b>3.46</b>  | <b>2.89</b>  | <b>4.20</b>  |
| 77-126         | 27B9 (F77T,T126R)                 | 0.55         | 0.26        | 0.54        | <b>3.68</b>  | 0.67        | 0.59        | <b>1.91</b> | <b>2.34</b>  | 1.06        | 0.67         | 0.32         | 0.49         | <b>2.58</b>  |
| 77-39          | 33E2 (F77T,T39I)                  | 0.70         | 0.35        | 0.51        | <b>4.35</b>  | 0.67        | 0.53        | <b>2.39</b> | <b>4.01</b>  | 1.20        | 1.12         | 0.50         | 0.67         | <b>3.74</b>  |
| 89-148         | 50 E7 (K89E, S148T)               | <b>2.62</b>  | 0.76        | 1.04        | <b>3.34</b>  | 1.21        | 1.24        | <b>2.76</b> | <b>4.62</b>  | <b>2.49</b> | <b>2.89</b>  | <b>2.61</b>  | <b>2.58</b>  | <b>3.71</b>  |
| 90-77          | 02D10 (I90V,F77T)                 | <b>1.55</b>  | 0.60        | 0.82        | <b>13.12</b> | 1.08        | 0.84        | <b>6.71</b> | <b>30.62</b> | <b>1.77</b> | <b>8.13</b>  | <b>4.09</b>  | <b>12.85</b> | <b>20.94</b> |
| 106-164        | 17E6 (M106K,C164S)                | <b>1.65</b>  | 1.06        | 1.11        | <b>1.32</b>  | 1.27        | 1.29        | <b>1.98</b> | <b>1.77</b>  | <b>1.82</b> | <b>2.58</b>  | <b>4.79</b>  | <b>1.60</b>  | <b>2.16</b>  |
| 23-58-169      | 47G12 (E23K,V58L,A169V)           | 0.05         | 0.42        | 0.46        | 1.16         | 0.62        | 0.79        | 0.84        | <b>1.54</b>  | 0.30        | 0.02         | 0.03         | 0.12         | <b>1.32</b>  |
| 33-76-89       | 17C5 (K33N,I76N,K89E)             | 1.11         | 0.59        | <b>1.48</b> | 0.93         | <b>1.89</b> | <b>1.79</b> | 1.30        | 1.13         | <b>1.56</b> | 0.74         | 0.32         | 0.65         | 0.99         |
| 68-90-145      | 04D10 (F68S,I90V,P145L)           | <b>3.91</b>  | <b>1.33</b> | <b>1.45</b> | <b>5.59</b>  | <b>1.74</b> | <b>1.99</b> | <b>6.12</b> | <b>8.68</b>  | <b>4.09</b> | <b>3.86</b>  | <b>3.64</b>  | <b>6.08</b>  | <b>8.66</b>  |
| 59-109-113-136 | 41H10 (I59T, Y109F, P113T, L136M) | 0.06         | 0.07        | 0.24        | 0.03         | 0.26        | 0.20        | 0.25        | 0.30         | 0.16        | <b>5.44</b>  | <b>2.21</b>  | 0.43         | 0.36         |

FIG. 6C

| POSITION       | Clone(AA sequence)                | coel. | h, h  | PBI-3945 |
|----------------|-----------------------------------|-------|-------|----------|
| 15             | 03F2 (G15R)                       | 0.002 | 0.010 | 0.024    |
| 18             | 40H11 (Q18L)                      | 0.38  | 0.53  | 0.39     |
| 20             | 16C5 (Q20R)                       | 2.58  | 0.82  | 0.80     |
| 22             | 14C1 (L22I)                       | 0.12  | 0.33  | 0.44     |
| 27             | 11E8 (L27V)                       | 0.21  | 1.20  | 1.30     |
| 54             | 18F8 (F54S)                       | 0.59  | 0.25  | 0.03     |
| 54             | 29H7 (F54I)                       | 6.20  | 0.65  | 1.29     |
| 58             | 30E4 (V58I)                       | 0.18  | 0.69  | 0.77     |
| 72             | 04G7 (L72Q)                       | 4.19  | 1.01  | 1.49     |
| 75             | 28B2 (M75K)                       | 4.11  | 2.21  | 2.06     |
| 77             | 18C4 (F77T)                       | 0.93  | 0.74  | 0.33     |
| 77             | 42E4 (F77C)                       | 0.08  | 0.75  | 0.28     |
| 77             | 50D12 (F77T)                      | 0.81  | 0.64  | 0.29     |
| 89             | 35D11 (K89E)                      | 2.18  | 0.88  | 0.61     |
| 90             | 10F2 (I90T)                       | 1.45  | 0.38  | 0.32     |
| 92             | 04A12 (L92H)                      | 0.62  | 0.27  | 0.03     |
| 109            | 43F9 (Y109F)                      | 0.22  | 0.54  | 0.07     |
| 127            | 30D6 (V127A)                      | 0.11  | 0.22  | 0.19     |
| 139            | 27B8 (D139G)                      | 0.00  | 0.08  | 0.01     |
| 164            | 25A1 (C164S)                      | 0.56  | 0.61  | 0.63     |
| 14-89          | 25F2 (A14V, K89E)                 | 1.91  | 0.66  | 0.49     |
| 27-33          | 47D8 (L27M, K33N)                 | 0.40  | 1.24  | 1.12     |
| 49-72          | 26F1 (E49K, L72Q)                 | 1.17  | 1.08  | 1.14     |
| 54-66          | 48G3 (F54I, S66T)                 | 6.17  | 0.66  | 1.31     |
| 54-67          | 35B8 (F54S, G67S)                 | 0.13  | 0.99  | 0.24     |
| 55-117         | 05H8 (D55G, I117F)                | 0.11  | 0.30  | 0.30     |
| 56-93          | 46A11 (I56V, H93R)                | 0.15  | 0.64  | 0.26     |
| 68-75          | 04E2 (F68S, M75K)                 | 4.11  | 1.78  | 1.94     |
| 77-126         | 27B9 (F77T, T126R)                | 0.49  | 0.47  | 0.13     |
| 77-39          | 33E2 (F77T, T39I)                 | 0.58  | 0.44  | 0.20     |
| 89-148         | 50E7 (K89E, S148T)                | 2.06  | 0.85  | 0.59     |
| 90-77          | 02D10 (I90V, F77T)                | 1.85  | 1.01  | 0.56     |
| 106-164        | 17E6 (M106K, C164S)               | 2.16  | 1.30  | 1.35     |
| 23-58-169      | 47G12 (E23K, V58L, A169V)         | 0.04  | 0.37  | 0.47     |
| 33-76-89       | 17C5 (K33N, I76N, K89E)           | 1.31  | 1.75  | 0.60     |
| 68-90-145      | 04D10 (F68S, I90V, P145L)         | 3.01  | 1.11  | 1.07     |
| 59-109-113-136 | 41H10 (I59T, Y109F, P113T, L136M) | 0.05  | 0.20  | 0.02     |

FIG. 6D

| Clone                                                | 18 | 20 | 54 | 59 | 72 | 77 | 89 | 92 | 109 | 113 | 127 | 136 | 164 |
|------------------------------------------------------|----|----|----|----|----|----|----|----|-----|-----|-----|-----|-----|
| QC#1 (Q20R, L92H, F77C)                              |    | X  |    |    |    | X  |    | X  |     |     |     |     |     |
| QC#3 (F77C)                                          |    |    |    |    |    | X  |    |    |     |     |     |     |     |
| QC#5 (L92H)                                          |    |    |    |    |    |    |    | X  |     |     |     |     |     |
| QC#6 (F54I, F77C)                                    |    |    | X  |    |    | X  |    |    |     |     |     |     |     |
| QC#7 (Q20R, F54I, F77C)                              |    | X  | X  |    |    | X  |    |    |     |     |     |     |     |
| QC#8 (Q20R, F54I)                                    |    | X  | X  |    |    |    |    |    |     |     |     |     |     |
| QC#9 (F54I, F77C, L92H)                              |    |    | X  |    |    | X  |    | X  |     |     |     |     |     |
| QC#10 (Q20R, F54I, F77C, L92H)                       |    | X  | X  |    |    | X  |    | X  |     |     |     |     |     |
| QC#13 (Q20R, Y109F)                                  |    | X  |    |    |    |    |    |    | X   |     |     |     |     |
| QC#14 (L92H, Y109F)                                  |    |    |    |    |    |    |    | X  | X   |     |     |     |     |
| QC#15 (Y109F)                                        |    |    |    |    |    |    |    |    | X   |     |     |     |     |
| QC#16 (F54I, L92H, Y109F)                            |    |    | X  |    |    |    |    | X  | X   |     |     |     |     |
| QC#20 (F54I, Y109F)                                  |    |    | X  |    |    |    |    |    | X   |     |     |     |     |
| QC#22 (Q18L, L92H, Y109F)                            | X  |    |    |    |    |    |    | X  | X   |     |     |     |     |
| QC#27 (Q18L, F54I, L92H, Y109F)                      | X  |    | X  |    |    |    |    | X  | X   |     |     |     |     |
| QC#32 (Q18L, I59T, L92H, Y109F, P113T, V127A, K136M) | X  |    |    | X  |    |    |    | X  | X   | X   | X   | X   |     |
| QC#36 (Q20R, F54I, L72Q, K89E, C164S)                |    | X  | X  |    | X  |    | X  |    |     |     |     |     | X   |
| QC#37 (F54I, L72Q, K89E, C164S)                      |    |    | X  |    | X  |    | X  |    |     |     |     |     | X   |
| QC#38 (F54I, C164S)                                  |    |    | X  |    |    |    |    |    |     |     |     |     | X   |
| QC #1 C2 (Q18L, Y109F)                               | X  |    |    |    |    |    |    |    | X   |     |     |     |     |
| QC #1 B4 (Q18L, F54I)                                | X  |    | X  |    |    |    |    |    |     |     |     |     |     |
| QC #1 C3 (Q18L, F54I, Y109F)                         | X  |    | X  |    |    |    |    |    | X   |     |     |     |     |
| QC#1 A7 (F54I, K89E, C164S)                          |    |    | X  |    |    |    |    |    |     |     |     |     | X   |
| QC#1 A11 (Q20R, F54I, L72Q, K89E)                    |    | X  | X  |    | X  |    | X  |    |     |     |     |     |     |
| QC#1 B9 Q20R, F54I, L72Q, C164S)                     |    | X  | X  |    | X  |    |    |    |     |     |     |     | X   |
| QC#1 D11 (F54I, L72Q, K89E, C164S)                   |    |    | X  |    | X  |    | X  |    |     |     |     |     | X   |

FIG. 7

| Sample                                                  | coelenterazine | h     | h,h   | PBI-3840 | PBI-3925 | PBI-3912 |
|---------------------------------------------------------|----------------|-------|-------|----------|----------|----------|
| QC#1 (Q20R, L92H, F77C)                                 | 0.17           | 0.03  | 0.19  | 0.74     | 1.03     | 0.56     |
| QC#3 (F77C)                                             | 0.09           | 0.36  | 0.89  | 0.16     | 1.06     | 0.04     |
| QC#5 (L92H)                                             | 0.73           | 0.04  | 0.38  | 4.49     | 6.92     | 5.64     |
| QC#6 (F54I, F77C)                                       | 0.85           | 0.59  | 0.91  | 1.89     | 3.80     | 0.41     |
| QC#7 (Q20R, F54I, F77C)                                 | 0.72           | 0.22  | 0.20  | 1.85     | 2.83     | 1.50     |
| QC#8 (Q20R, F54I)                                       | 6.48           | 0.89  | 0.34  | 11.73    | 21.51    | 35.04    |
| QC#9 (F54I, F77C, L92H)                                 | 0.39           | 0.07  | 0.16  | 0.81     | 1.58     | 0.64     |
| QC#10 (Q20R, F54I, F77C, L92H)                          | 0.27           | 0.05  | 0.07  | 0.75     | 2.24     | 1.53     |
| QC#13 (Q20R, Y109F)                                     | 0.21           | 0.07  | 0.25  | 1.08     | 1.74     | 4.18     |
| QC#14 (L92H, Y109F)                                     | 0.06           | 0.00  | 0.08  | 0.78     | 1.84     | 2.63     |
| QC#15 (Y109F)                                           | 0.21           | 0.15  | 0.54  | 0.95     | 0.68     | 0.46     |
| QC#16 (F54I, L92H, Y109F)                               | 0.35           | 0.02  | 0.05  | 1.01     | 3.73     | 8.75     |
| QC#20 (F54I, Y109F)                                     | 4.49           | 0.82  | 0.51  | 9.41     | 12.43    | 10.39    |
| QC#22 (Q18L, L92H, Y109F)                               | 0.05           | 0.01  | 0.05  | 0.15     | 0.06     | 0.14     |
| QC#27 (Q18L, F54I, L92H, Y109F)                         | 0.08           | 0.01  | 0.10  | 0.68     | 0.59     | 1.06     |
| QC#32 (Q18L, I59T, L92H, Y109F,<br>P113T, V127A, K136M) | 0.003          | 0.001 | 0.007 | 0.022    | 0.015    | 0.020    |
| QC#36 (Q20R, F54I, L72Q, K89E, C164S)                   | 0.99           | 0.14  | 0.06  | 1.34     | 1.86     | 16.93    |
| QC#37 (F54I, L72Q, K89E, C164S)                         | 2.01           | 0.30  | 0.14  | 3.16     | 4.63     | 23.45    |
| QC#38 (F54I, C164S)                                     | 3.39           | 0.65  | 0.48  | 5.25     | 4.56     | 9.48     |
| C1A4E                                                   | 1.00           | 1.00  | 1.00  | 1.00     | 1.00     | 1.00     |

FIG. 8A

| Sample                                               | Coel. | h     | h,h   | 2-me  | PBI-3889 | PBI-3939 | PBI-3933 | PBI-3932 | PBI-3946 | PBI-3841 | PBI-3896 | PBI-3925 |
|------------------------------------------------------|-------|-------|-------|-------|----------|----------|----------|----------|----------|----------|----------|----------|
| QC#1 (Q20R, L92H, F77C)                              | 0.13  | 0.02  | 0.11  | 1.62  | 0.15     | 0.12     | 0.45     | 3.87     | 0.18     | 0.25     | 0.17     | 0.58     |
| QC#3 (F77C)                                          | 0.07  | 0.25  | 0.55  | 1.14  | 0.63     | 0.38     | 0.41     | 1.79     | 0.28     | 0.04     | 0.04     | 0.55     |
| QC#5 (L92H)                                          | 0.51  | 0.03  | 0.24  | 4.99  | 0.23     | 0.10     | 1.68     | 2.88     | 0.80     | 25.89    | 23.26    | 3.28     |
| QC#6 (F54I, F77C)                                    | 0.69  | 0.48  | 0.67  | 22.92 | 0.31     | 0.07     | 0.51     | 35.10    | 0.44     | 0.08     | 0.15     | 2.55     |
| QC#7 (Q20R, F54I, F77C)                              | 0.54  | 0.18  | 0.15  | 22.57 | 0.09     | 0.03     | 0.39     | 32.08    | 0.21     | 0.39     | 0.41     | 2.24     |
| QC#8 (Q20R, F54I)                                    | 5.32  | 0.79  | 0.28  | 28.12 | 0.49     | 0.11     | 2.80     | 40.88    | 1.43     | 26.40    | 43.55    | 17.18    |
| QC#9 (F54I, F77C, L92H)                              | 0.26  | 0.05  | 0.10  | 3.69  | 0.13     | 0.09     | 0.55     | 5.63     | 0.22     | 0.21     | 0.23     | 1.03     |
| QC#10 (Q20R, F54I, F77C, L92H)                       | 0.20  | 0.03  | 0.05  | 5.91  | 0.07     | 0.05     | 0.45     | 11.18    | 0.19     | 0.54     | 0.45     | 1.58     |
| QC#13 (Q20R, Y109F)                                  | 0.26  | 0.10  | 0.31  | 1.05  | 0.37     | 0.29     | 0.66     | 3.37     | 0.25     | 19.06    | 74.46    | 1.82     |
| QC#14 (L92H, Y109F)                                  | 0.10  | 0.003 | 0.11  | 1.18  | 0.11     | 0.06     | 0.52     | 2.24     | 0.07     | 13.47    | 65.50    | 1.64     |
| QC#15 (Y109F)                                        | 0.29  | 0.22  | 0.74  | 0.38  | 0.83     | 0.69     | 0.68     | 0.74     | 0.47     | 6.43     | 1.61     | 0.89     |
| QC#16 (F54I, L92H, Y109F)                            | 0.44  | 0.02  | 0.06  | 6.00  | 0.11     | 0.06     | 0.46     | 9.48     | 0.03     | 5.53     | 269.14   | 3.50     |
| QC#20 (F54I, Y109F)                                  | 4.22  | 0.90  | 0.50  | 5.86  | 1.19     | 0.61     | 3.68     | 14.10    | 1.82     | 44.89    | 15.18    | 12.45    |
| QC#22 (Q18L, L92H, Y109F)                            | 0.06  | 0.01  | 0.06  | 0.06  | 0.06     | 0.05     | 0.05     | 0.03     | 0.01     | 14.10    | 242.95   | 0.04     |
| QC#27 (Q18L, F54I, L92H, Y109F)                      | 0.13  | 0.02  | 0.12  | 0.33  | 0.15     | 0.12     | 0.30     | 0.24     | 0.10     | 0.60     | 561.32   | 0.40     |
| QC#32 (Q18L, I59T, L92H, Y109F, P113T, V127A, K136M) | 0.004 | 0.001 | 0.011 | 0.01  | 0.01     | 0.01     | 0.01     | 0.01     | 0.00     | 16.29    | 51.72    | 0.02     |
| QC#36 (Q20R, F54I, L72Q, K89E, C164S)                | 0.78  | 0.14  | 0.07  | 23.49 | 0.02     | 0.002    | 0.09     | 28.30    | 0.06     | 64.90    | 49.30    | 2.04     |
| QC#37 (F54I, L72Q, K89E, C164S)                      | 1.87  | 0.32  | 0.17  | 26.86 | 0.08     | 0.01     | 0.32     | 33.42    | 0.22     | 71.56    | 37.60    | 5.30     |
| QC#38 (F54I, C164S)                                  | 4.51  | 0.92  | 0.67  | 11.22 | 0.49     | 0.06     | 1.15     | 12.46    | 1.13     | 22.07    | 11.03    | 6.46     |
| CI44E                                                | 1.00  | 1.00  | 1.00  | 1.00  | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     |

FIG. 8B

| Sample                                              | Coel   | h <sub>h</sub> | PBI-3939 | PBI-3945 | PBI-3840 | PBI-3932 | PBI-3925 | PBI-9894 | PBI-3896 |
|-----------------------------------------------------|--------|----------------|----------|----------|----------|----------|----------|----------|----------|
| QC C2(Q18, Y109F)                                   | 0.01   | 0.06           | 0.04     | 0.04     | 0.03     | 0.01     | 0.01     | 19.87    | 24.80    |
| QC B4(Q18, F54)                                     | 5.71   | 1.84           | 1.23     | 1.19     | 6.04     | 8.23     | 7.02     | 67.35    | 719.58   |
| QC C3(Q18, F54, Y109F)                              | 0.18   | 0.52           | 0.65     | 0.22     | 0.49     | 0.13     | 0.46     | 407.08   | 454.48   |
| QC E6(Q18, I59T, Y109F, P113T, V127A, K136M)        | 0.0004 | 0.01           | 0.01     | 0.002    | 0.01     | 0.01     | 0.00     | 3.88     | 9.03     |
| QC A7(F54, K89E, C164S)                             | 2.63   | 0.21           | 0.01     | 0.28     | 4.05     | 17.21    | 4.86     | 11.82    | 23.95    |
| QC A11(Q20R, F54, L72Q, K89E)                       | 3.76   | 0.13           | 0.02     | 0.31     | 3.21     | 94.34    | 15.76    | 46.29    | 127.88   |
| QC B9(Q20R, F54, L72Q, C164S)                       | 1.67   | 0.10           | 0.005    | 0.10     | 1.89     | 25.28    | 3.37     | 13.58    | 42.27    |
| QC D11(F54, L72Q, K89E, C164S)                      | 1.95   | 0.13           | 0.01     | 0.16     | 2.45     | 27.69    | 4.84     | 13.38    | 33.69    |
| QC #6(F54, F77C)                                    | 0.67   | 0.76           | 0.06     | 0.36     | 1.24     | 39.37    | 3.14     | 0.52     | 1.32     |
| QC #7(Q20R, F54, F77C)                              | 0.51   | 0.15           | 0.03     | 0.07     | 1.01     | 32.09    | 2.26     | 1.67     | 1.30     |
| QC #9(F54, F77C, L92H)                              | 0.22   | 0.10           | 0.09     | 0.06     | 0.33     | 4.97     | 1.01     | 0.26     | 0.32     |
| QC #8(Q20R, F54)                                    | 6.37   | 0.35           | 0.14     | 0.93     | 8.42     | 46.87    | 21.22    | 13.40    | 33.57    |
| QC #13(Q20R, Y109F)                                 | 0.18   | 0.26           | 0.25     | 0.02     | 0.79     | 2.54     | 1.58     | 6.85     | 49.23    |
| QC #14(L92H, Y109F)                                 | 0.04   | 0.07           | 0.04     | 0.001    | 0.29     | 1.35     | 1.43     | 4.53     | 37.03    |
| QC #16(F54, L92H, Y109F)                            | 0.30   | 0.05           | 0.05     | 0.01     | 0.46     | 7.07     | 3.42     | 19.64    | 157.70   |
| QC #20(F54, Y109F)                                  | 4.25   | 0.56           | 0.62     | 1.04     | 6.93     | 13.08    | 11.84    | 8.73     | 10.47    |
| QC #22(Q18, L92H, Y109F)                            | 0.04   | 0.05           | 0.04     | 0.01     | 0.09     | 0.03     | 0.05     | 38.41    | 138.45   |
| QC #27(Q18, F54, L92H, Y109F)                       | 0.07   | 0.08           | 0.10     | 0.01     | 0.28     | 0.17     | 0.42     | 392.98   | 283.85   |
| QC #32(Q18, I59T, L92H, Y109F, P113T, V127A, K136M) | 0.003  | 0.01           | 0.01     | 0.0002   | 0.02     | 0.01     | 0.02     | 11.79    | 33.46    |
| QC #36(Q20R, F54, L72Q, K89E, C164S)                | 0.92   | 0.06           | 0.002    | 0.05     | 1.07     | 24.56    | 2.12     | 10.40    | 35.04    |

FIG. 9

| Clone                                               | 18 | 20 | 54 | 71 | 77 | 90 | 92 | 109 | 127 |
|-----------------------------------------------------|----|----|----|----|----|----|----|-----|-----|
| QC#2 1 (Q20R,F77C,I90V,I92H,V127A)                  |    | X  |    |    | X  | X  | X  |     | X   |
| QC#2 2 (Q20R,G71D,F77C,I90V,I92H,V127A)             |    | X  |    | X  | X  | X  | X  |     | X   |
| QC#2 3 (Q20R,G71D,F77C,I92H,V127A)                  |    | X  |    | X  | X  |    | X  |     | X   |
| QC#2 6 (F54I,G71D,F77C,I92H,V127A)                  |    |    | X  | X  | X  |    | X  |     | X   |
| QC#2 7 (F54I,G71D,F77C,I90V,I92H)                   |    |    | X  | X  | X  | X  | X  |     |     |
| QC#2 8 (F54I,G71D,F77C,I90V,I92H,V127A)             |    |    | X  | X  | X  | X  | X  |     | X   |
| QC#2 11 (Q20R,F54I,G71D,F77C,I90V,I92H,V127A)       |    | X  | X  | X  | X  | X  | X  |     | X   |
| QC#2 16 (G71D,I90V,I92H,Y109F,I127A)                |    |    |    | X  |    | X  | X  | X   | X   |
| QC#2 18 (G71D,I92H,Y109F,I127A)                     |    |    |    | X  |    |    | X  | X   | X   |
| QC#2 21 (Q20R,F54I,G71D,F77C,I90V,I92H,V127A)       |    | X  | X  | X  | X  | X  | X  |     | X   |
| QC#2 26 (Q18L,G71D,F77Y,I90V,I92H,Y109F,V127A)      | X  |    |    | X  | X  | X  | X  | X   | X   |
| QC#2 31 (Q18L,F54I,G71D,F77Y,I90V,I92H,Y109F,V127A) | X  |    | X  | X  | X  | X  | X  | X   | X   |
| QC#2 41 (G71D,F77C,I90V,I92H,V127A)                 |    |    |    | X  | X  | X  | X  |     | X   |
| QC#2 44 (G71D,F77C,I90V,V127A)                      |    |    |    | X  | X  | X  |    |     | X   |
| QC#2 A3 (Q20R,G71D,F77C,I92H)                       |    | X  |    | X  | X  |    | X  |     |     |
| QC#2 C6 (F54I,G71D,F77C,I92H)                       |    |    | X  | X  | X  |    | X  |     |     |
| QC#2 A7 (Q20R,F54I,G71D,F77C,I92H,V127A)            |    | X  | X  | X  | X  |    | X  |     | X   |
| QC#2 C10 (I90V,I92H,Y109F,V127A)                    |    |    |    |    |    | X  | X  | X   | X   |
| QC#2 E1 (F54I,I90V,I92H,Y109F,V127A)                |    |    | X  |    |    | X  | X  | X   | X   |
| QC#2 E2 (F54I,G71D,I90V,I92H,Y109F)                 |    |    | X  | X  |    | X  | X  | X   |     |
| QC#2 E4 (Q18L,G71D,F77Y,I92H,Y109F,V127A)           | X  |    |    | X  | X  |    | X  | X   | X   |
| QC#2 F4 (Q18L,F77Y,I90V,I92H,Y109F,V127A)           | X  |    |    |    | X  | X  | X  | X   | X   |
| QC#2 E7 (Q18L,F54I,F77Y,I90V,I92H,Y109F,V127A)      | X  |    | X  |    | X  | X  | X  | X   | X   |
| QC#2 E8 (Q18L,F54I,I90V,I92H,Y109F,V127A)           | X  |    | X  |    |    | X  | X  | X   | X   |
| QC#2 F7 (Q18L,F54I,G71D,F77Y,I92H,Y109F,V127A)      | X  |    | X  | X  | X  |    | X  | X   | X   |
| QC#2 H11 (G71D,F77C)                                |    |    |    | X  | X  |    |    |     |     |
| IE1 (I90V/I92H)                                     |    |    |    |    |    | X  | X  |     |     |
| IE2 (I90V,Y109F)                                    |    |    |    |    |    | X  | X  | X   |     |
| IE3 (I90V/I92H/Y109F)                               |    |    |    |    |    | X  | X  | X   | X   |

FIG. 10

| Sample                                              | Coel   | h,h    | PBI-3939 | PBI-3945 | PBI-3840 | PBI-3932 | PBI-3925 | PBI-3894 | PBI-3896 |
|-----------------------------------------------------|--------|--------|----------|----------|----------|----------|----------|----------|----------|
| QC#2 1 (Q20R,F77C,I90V,I92H,V127A)                  | 0.04   | 0.04   | 0.05     | 0.004    | 0.17     | 4.04     | 0.62     | 0.19     | 0.13     |
| QC#2 2 (Q20R,G71D,F77C,I90V,I92H,V127A)             | 0.07   | 0.05   | 0.07     | 0.01     | 0.24     | 6.19     | 0.98     | 0.26     | 0.20     |
| QC#2 3 (Q20R,G71D,F77C,I92H,V127A)                  | 0.09   | 0.07   | 0.09     | 0.01     | 0.30     | 5.02     | 0.88     | 0.34     | 0.21     |
| QC#2 6 (F54I,G71D,F77C,I92H,V127A)                  | 0.11   | 0.07   | 0.08     | 0.03     | 0.19     | 2.43     | 0.51     | 0.15     | 0.11     |
| QC#2 7 (F54I,G71D,F77C,I90V,I92H)                   | 0.40   | 0.11   | 0.12     | 0.09     | 0.39     | 13.65    | 2.15     | 0.38     | 0.41     |
| QC#2 8 (F54I,G71D,F77C,I90V,I92H,V127A)             | 0.11   | 0.06   | 0.08     | 0.03     | 0.16     | 4.12     | 0.68     | 0.15     | 0.18     |
| QC#2 11 (Q20R,F54I,G71D,F77C,I90V,I92H,V127A)       | 0.10   | 0.03   | 0.05     | 0.02     | 0.17     | 6.66     | 1.11     | 0.40     | 0.24     |
| QC#2 16 (G71D,I90V,I92H,Y109F,I127A,)               | 0.11   | 0.06   | 0.04     | 0.001    | 0.29     | 1.49     | 1.51     | 7.72     | 55.11    |
| QC#2 18 (G71D,I92H,Y109F,I127A)                     | 0.08   | 0.04   | 0.03     | 0.0005   | 0.28     | 1.33     | 1.29     | 7.30     | 48.97    |
| QC#2 21(Q20R,F54I,G71D,F77C,I90V,I92H,V127A)        | 0.27   | 0.02   | 0.02     | 0.004    | 0.31     | 7.57     | 2.49     | 30.02    | 160.54   |
| QC#2 26 (Q18L,G71D,F77Y,I90V,I92H,Y109F,V127A)      | 0.00   | 0.01   | 0.01     | 0.001    | 0.00     | 0.01     | 0.02     | 1.38     | 7.86     |
| QC#2 31 (Q18L,F54I,G71D,F77Y,I90V,I92H,Y109F,V127A) | 0.01   | 0.03   | 0.03     | 0.004    | 0.01     | 0.05     | 0.06     | 8.04     | 77.32    |
| QC#2 41 (G71D,F77C,I90V,I92H,V127A)                 | 0.05   | 0.07   | 0.08     | 0.01     | 0.12     | 2.50     | 0.37     | 0.10     | 0.07     |
| QC#2 44 (G71D,F77C,I90V,V127A)                      | 0.17   | 0.15   | 0.15     | 0.03     | 0.15     | 2.92     | 0.41     | 0.18     | 0.10     |
| QC#2 A3 (Q20R,G71D,F77C,I92H)                       | 0.23   | 0.20   | 0.22     | 0.03     | 0.52     | 6.75     | 1.11     | 0.39     | 0.24     |
| QC#2 C6 (F54I,G71D,F77C,I92H)                       | 0.44   | 0.18   | 0.18     | 0.13     | 0.60     | 12.27    | 2.43     | 0.50     | 0.30     |
| QC#2 A7 (Q20R,F54I,G71D,F77C,I92H,V127A)            | 0.13   | 0.05   | 0.06     | 0.02     | 0.28     | 6.93     | 1.25     | 0.32     | 0.22     |
| QC#2 C10 (I90V,I92H,Y109F,V127A)                    | 0.06   | 0.04   | 0.02     | 0.0004   | 0.22     | 1.08     | 1.05     | 4.33     | 34.52    |
| QC#2 E1 (F54I,I90V,I92H,Y109F,V127A)                | 0.18   | 0.04   | 0.03     | 0.004    | 0.25     | 2.74     | 1.66     | 6.96     | 49.09    |
| QC#1 E2 (F54I,G71D,I90V,I92H,Y109F)                 | 0.39   | 0.06   | 0.04     | 0.01     | 0.24     | 9.71     | 3.63     | 25.77    | 185.63   |
| QC#2 E4 (Q18L,G71D,F77Y,I92H,Y109F,V127A)           | 0.0005 | 0.009  | 0.008    | 0.0005   | 0.001    | 0.009    | 0.004    | 0.58     | 3.48     |
| QC#2 F4(Q18L,F77Y,I90V,I92H,Y109F,V127A)            | 0.0002 | 0.0057 | 0.0062   | 0.0003   | 0.0013   | 0.0069   | 0.0048   | 0.56     | 4.18     |
| QC#2 E7 (Q18L,F54I,F77Y,I90V,I92H,Y109F,V127A)      | 0.003  | 0.013  | 0.015    | 0.002    | 0.006    | 0.020    | 0.024    | 3.17     | 36.43    |
| QC#2 E8 (Q18L,F54I,I90V,I92H,Y109F,V127A)           | 0.06   | 0.04   | 0.05     | 0.002    | 0.13     | 0.05     | 0.11     | 145.44   | 331.26   |
| QC#2 F7 (Q18L,F54I,G71D,F77Y,I92H,Y109F,V127A)      | 0.01   | 0.03   | 0.03     | 0.004    | 0.01     | 0.03     | 0.04     | 3.22     | 30.50    |
| QC#2 H11 (G71D,F77C)                                | 0.07   | 0.69   | 0.61     | 0.24     | 0.13     | 3.46     | 1.02     | 0.16     | 0.10     |
| CIA4E                                               | 1.00   | 1.00   | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     | 1.00     |

FIG. 11

| Sample                                         | Coel.  | ih    | PBI-3939 | PBI-3945 | PBI-3889 | PBI-3840 | PBI-3932 | PBI-3925 | PBI-3894 | PBI-3896 | PBI-3897 |
|------------------------------------------------|--------|-------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| C1A4E                                          | 1      | 1     | 1        | 1        | 1        | 1        | 1        | 1        | 1        | 1        | 1        |
| I90V                                           | 1.49   | 0.77  | 1.21     | 0.51     | 1.12     | 2.86     | 22.13    | 14.69    | 8.48     | 2.30     | 6.43     |
| Y109F                                          | 0.13   | 0.42  | 0.35     | 0.15     | 0.42     | 0.58     | 0.37     | 0.41     | 1.05     | 0.35     | 12.70    |
| LE2 (I90V,Y109F)                               | 0.31   | 0.29  | 0.26     | 0.06     | 0.33     | 1.80     | 10.20    | 6.09     | 5.15     | 3.04     | 60.24    |
| QC #27(Q18L/L92H/Y109F/F54I)                   | 0.15   | 0.06  | 0.08     | 0.01     | 0.06     | 0.23     | 0.12     | 0.20     | 196.86   | 140.68   | 13.83    |
| QCH2 E7 (Q18L,F54I,F77Y,I90V,L92H,Y109F,V127A) | 0.002  | 0.02  | 0.02     | 0.0022   | 0.0076   | 0.0054   | 0.02     | 0.02     | 1.83     | 6.57     | 31.17    |
| QCH2 F4(Q18L,F77Y,I90V,L92H,Y109F,V127A)       | 0.0008 | 0.005 | 0.0046   | 0.0004   | 0.0021   | 0.0021   | 0.01     | 0.05     | 0.24     | 0.80     | 7.04     |
| QCH1 A11 (Q20R, F54I, L72Q, K89E)              | 3.50   | 0.12  | 0.02     | 0.16     | 0.08     | 3.51     | 82.84    | 11.19    | 59.12    | 24.00    | 42.51    |

FIG. 12

| Positions in common with C1A4E library |
|----------------------------------------|
| 18                                     |
| 22                                     |
| 68                                     |
| 72                                     |
| 75                                     |
| 90                                     |
| 92                                     |
| 106                                    |
| 109                                    |

| Sample     | POSITION     | Sequence             | COE/ENTERAZINE | H/H  | PBI-3897/PBI-3896 | PBI-3894 |
|------------|--------------|----------------------|----------------|------|-------------------|----------|
| 29H3       | 18           | L18I                 | 1.17           | 0.32 | 0.94              | 2.37     |
| 25A11      | 21           | V21L                 | 0.06           | 0.11 | 0.53              | 0.55     |
| 4F3        | 67           | G67S                 | 1.75           | 0.63 | 1.37              | 1.24     |
| 24B12      | 68           | F68Y                 | 1.88           | 0.88 | 1.52              | 1.62     |
| 29C4       | 72           | L72Q                 | 1.69           | 0.77 | 1.37              | 2.33     |
| 3H11       | 75           | M75K                 | 2.33           | 1.09 | 1.90              | 2.19     |
| 3F10       | 76           | I76F                 | 0.50           | 0.81 | 1.27              | 0.17     |
| 27E 4      | 90           | I90T                 | 0.24           | 0.17 | 0.76              | 0.10     |
| 1B6        | 92           | H92R                 | 0.10           | 0.46 | 0.98              | 0.06     |
| 47H12      | 22-74        | L22F-E74K            | 0.03           | 0.12 | 0.29              | 0.02     |
| 11F10      | 158-1        | F1I,V158I            | 2.49           | 2.53 | 2.37              | 1.67     |
| 48D5       | 11-109       | R11Q,F109Y           | 6.24           | 2.74 | 0.39              | 1.04     |
| 18A8       | 18-98        | L18I, V98F           | 0.64           | 0.19 | 0.54              | 1.25     |
| 43G9       | 21-106       | V21M,M106I           | 0.02           | 0.04 | 0.05              | 0.23     |
| 33A5       | 31-102       | F31I,V102M           | 0.73           | 0.68 | 0.67              | 1.06     |
| 11G9       | 32-92        | Q32H,H92R            | 0.10           | 0.47 | 1.02              | 0.06     |
| 9D7        | 45-76        | V45E,I76V            | 1.66           | 0.71 | 1.33              | 1.72     |
| 33H2       | 92-159       | H92R,T159S           | 0.05           | 0.37 | 0.58              | 0.04     |
| 1G5        | 92-170       | H92R,G170R           | 0.58           | N/A  | 1.50              | 0.16     |
| 19C10      | 18-49-86     | L18Q,E49D,H86R       | 3.29           | 0.15 | 0.52              | 0.05     |
| 44H1       | 21-47-51     | V21M, S47P, G51E     | 0.01           | 0.02 | 0.02              | 0.10     |
| 14C2       | 46-106-168   | L46Q, M106I, L168F   | 1.77           | 0.97 | 1.52              | 1.51     |
| 24D7       | 48-99-76     | G48R,I99V,I76F       | 0.38           | 0.63 | 1.04              | 0.10     |
| 18E 8      | 55-92-96     | D55E,H92Q,T96A       | 1.40           | 3.78 | 2.41              | 0.39     |
| 7D9        | 74-92-99     | E74I, H92R,I99T      | 0.14           | 0.36 | 0.77              | 0.04     |
| 8C2        | 21-68-69-142 | V21M,F68L,Q69H,L142V | 0.01           | 0.02 | 0.02              | 0.11     |
| QC #27 HT7 |              |                      | 1.00           | 1.00 | 1.00              | 1.00     |

FIG. 13

| Sequence                                        | Clone     | 21 | 68 | 72 | 75 | 76 | 90 | 92 | 158 |
|-------------------------------------------------|-----------|----|----|----|----|----|----|----|-----|
| F68Y, L72Q, M75K, V158I                         | QC27 #1   |    | X  | X  | X  |    |    |    | X   |
| I90T, H92R                                      | QC27 #2   |    |    |    |    |    | X  | X  |     |
| F68Y, L72Q, M75K, I90T, H92R, V158I             | QC27 #3   |    | X  | X  | X  |    | X  | X  | X   |
| V21L, I76F, I90T, H92R                          | QC27 #4   | X  |    |    |    | X  | X  | X  |     |
| V21L, I76F                                      | QC27 #6A  | X  |    |    |    | X  |    |    |     |
| V21L, F68Y, L72Q, M75K, I90T                    | QC27 #7A  | X  | X  | X  | X  |    | X  |    |     |
| V21L, I76F, H92R                                | QC27 #8   | X  |    |    |    | X  |    | X  |     |
| V21L, F68Y, L72Q, M75K, H92R, V158I             | QC27 #9A  | X  | X  | X  | X  |    |    | X  | X   |
| V21L, F68Y, L72Q, M75K, I76F, I90T, H92R, V158I | QC27 #10  | X  | X  | X  | X  | X  | X  | X  | X   |
| F68Y, L72Q, M75K, H92R                          | QC27 #12A |    | X  | X  | X  |    |    | X  |     |
| V21L, H92R                                      | QC27 #13  | X  |    |    |    |    |    | X  |     |
| F68Y, L72Q, M75K, H92R, V158I                   | QC27 #14A |    | X  | X  | X  |    |    | X  | X   |
| V21L, F68Y, L72Q, M75K, I90T, H92R, V158I       | QC27 #5   | X  | X  | X  | X  |    | X  | X  | X   |
| V21L, I90T, I76F                                | QC27 #6   | X  |    |    |    | X  | X  |    |     |
| V21L, F68Y, L72Q, M75K, I76F, V158I             | QC27 #9B  | X  | X  | X  | X  | X  |    |    | X   |

FIG. 14

| Sample     | AA change                                       | Coelenterazine | h,h  | PBI-3897 | PBI-3841 | PBI-3896 | PBI-3894 |
|------------|-------------------------------------------------|----------------|------|----------|----------|----------|----------|
| QC27 #1    | F68Y, L72Q, M75K, V158I                         | 2.59           | 3.95 | 2.90     | 2.90     | 5.19     | 6.72     |
| QC27 #2    | I90T, H92R                                      | 0.0189         | 0.17 | 0.19     | 0.19     | 0.02     | 0.00     |
| QC27 #3    | F68Y, L72Q, M75K, V158I, I90T, H92R             | 0.1212         | 0.77 | 0.88     | 0.92     | 0.10     | 0.02     |
| QC27 #4    | V21L, I76F, I90T, H92R                          | 0.0006         | 0.00 | 0.06     | 0.08     | 0.04     | 0.00     |
| QC27 #6A   | V21L, I76F                                      | 0.0060         | 0.07 | 0.26     | 0.20     | 0.12     | 0.03     |
| QC27 #7A   | V21L, F68Y, L72Q, M75K, I90T                    | 0.0370         | 0.40 | 1.76     | 1.54     | 1.09     | 0.16     |
| QC27 #8    | V21L, I76F, H92R                                | 0.0730         | 0.21 | 0.50     | 0.59     | 0.28     | 0.11     |
| QC27 #9A   | V21L, F68Y, L72Q, M75K, H92R, V158I             | 0.0041         | 0.08 | 0.81     | 1.35     | 0.32     | 0.03     |
| QC27 #10   | V21L, F68Y, L72Q, M75K, I76F, I90T, H92R, V158I | 0.0049         | 0.01 | 0.33     | 0.48     | 0.20     | 0.02     |
| QC27 #12A  | F68Y, L72Q, M75K, H92R                          | 0.2767         | 2.64 | 2.82     | 3.17     | 0.31     | 0.09     |
| QC27 #13   | V21L, H92R                                      | 0.0007         | 0.01 | 0.12     | 0.20     | 0.05     | 0.00     |
| QC27 #14A  | F68Y, L72Q, M75K, H92R, V158I                   | 0.2283         | 2.38 | 2.51     | 2.71     | 0.26     | 0.07     |
| QC27 #5    | V21L, F68Y, L72Q, M75K, I90T, H92R, V158I       | 0.0013         | 0.02 | 0.15     | 0.25     | 0.07     | 0.01     |
| QC27 #6    | V21L, I90T, I76F                                | 0.0014         | 0.00 | 0.09     | 0.05     | 0.03     | 0.00     |
| QC27 #9B   | V21L, F68Y, L72Q, M75K, I76F, V158I             | 0.0530         | 0.43 | 1.14     | 1.19     | 0.93     | 0.32     |
| QC27 ctrl. |                                                 | 1              | 1    | 1        | 1        | 1        | 1        |

FIG. 15

| Sample    | Position   | AA change        | Coelenterazine | H           | HH           | PBI-3841    | PBI-3897    |
|-----------|------------|------------------|----------------|-------------|--------------|-------------|-------------|
| 30D12     | 22         | L22F             | 0.08           | 0.01        | 0.08         | 0.28        | 0.26        |
| 26D4      | 36         | V36M             | <b>1.70</b>    | <b>1.40</b> | <b>2.19</b>  | 1.21        | 1.08        |
| 40B10     | 76         | I76F             | <b>3.66</b>    | 0.54        | 0.57         | <b>1.65</b> | <b>1.97</b> |
| 5C3       | 128        | T128A            | 0.19           | 0.26        | 0.45         | 0.43        | 0.47        |
| 22C2      | 128-154    | T128S,T154S      | 0.24           | 0.33        | 0.52         | 0.43        | 0.46        |
| 22G9      | 135-152    | N135Y,R152H      | 0.09           | 0.13        | 0.16         | 0.20        | 0.24        |
| 39H11     | 18-145     | L18I,P145Q       | <b>42.10</b>   | <b>3.20</b> | <b>18.30</b> | <b>2.51</b> | <b>2.37</b> |
| 36C11     | 18-99      | L18V,I99T        | <b>125.68</b>  | <b>2.64</b> | <b>24.99</b> | <b>2.15</b> | <b>2.17</b> |
| 3E 2      | 67-159     | G67D,T159I       | 0.39           | 0.51        | 0.63         | 0.77        | 0.80        |
| 48C5      | 46-51-138  | L46Q,G51R,I138S  | 0.43           | 0.49        | 0.96         | 0.87        | 0.95        |
| 3C10      | 51-112-113 | G51V,R112C,P113H | 0.24           | 0.31        | 0.72         | 0.93        | 0.67        |
| QC279a HT |            |                  | 1              | 1           | 1            | 1           | 1           |

FIG. 16

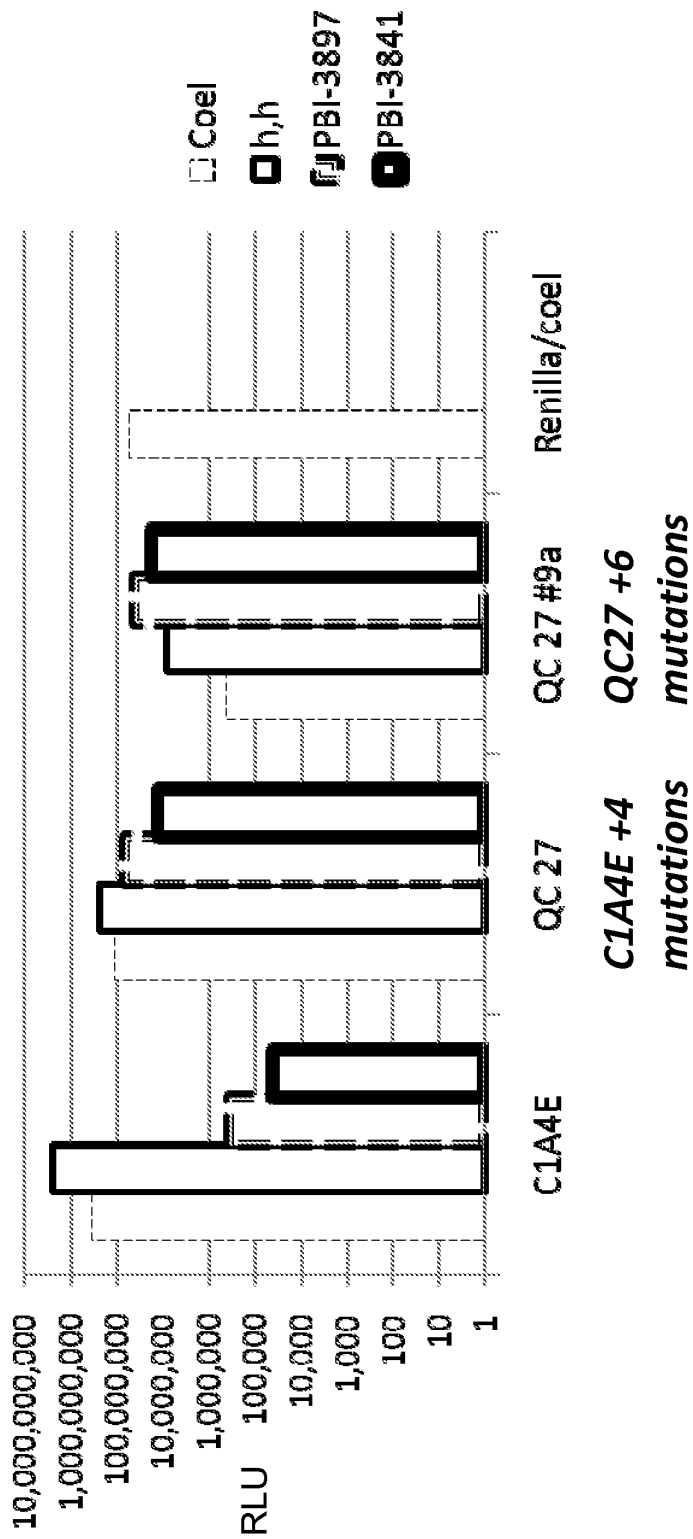


FIG. 17

| Sample   | Position  | AA change       | Coelenterazine | h <sub>h</sub> | PBI-3939    | PBI-3945 | PBI-3889 | PBI-4002 |
|----------|-----------|-----------------|----------------|----------------|-------------|----------|----------|----------|
| IW 29H12 | 19        | D19N            | 0.002          | 0.03           | 0.04        | 0.15     | 0.03     | 0.11     |
| IW 47E11 | 20        | Q20P            | 0.054          | 1.05           | <b>1.36</b> | 1.22     | 1.22     | 1.12     |
| IW 36E11 | 38        | V38I            | 0.169          | 0.96           | 1.26        | 1.18     | 1.23     | 1.09     |
| IW 21D1  | 44        | I44F            | 0.128          | 0.67           | 0.65        | 0.39     | 0.73     | 0.38     |
| IW 5B8   | 76        | I76N            | 0.208          | 0.32           | 0.39        | 0.50     | 0.36     | 0.44     |
| IW 50G8  | 77        | Y77N            | 0.141          | 0.86           | 0.23        | 0.61     | 0.62     | 0.85     |
| IW 27E6  | 95        | G95S            | 0.106          | 0.73           | 0.70        | 0.61     | 0.78     | 0.62     |
| IW 2G6   | 95        | G95D            | 0.004          | 0.07           | 0.07        | 0.33     | 0.06     | 0.30     |
| IW 27B7  | 110       | F110I           | 0.022          | 0.13           | 0.18        | 0.44     | 0.17     | 0.37     |
| IW 26H7  | 149       | L149I           | 0.266          | 0.94           | 0.82        | 0.90     | 0.89     | 0.94     |
| IW 21A4  | 152       | R152S           | 0.242          | 0.78           | 0.83        | 0.63     | 0.99     | 0.60     |
| IW 5C2   | 38-43     | V38F,K43N       | 0.008          | 0.45           | 0.51        | 0.56     | 0.46     | 0.51     |
| IW 43D3  | 49-32     | E49G,Q32P       | 0.170          | 0.93           | 1.23        | 1.15     | 1.20     | 1.07     |
| IW 20E12 | 60-119    | I60V,V119M      | 0.289          | 0.38           | 0.44        | 0.62     | 0.43     | 0.57     |
| IW 26B6  | 18-69-124 | Q18H,Q69H,K124M | 0.006          | 0.29           | 0.39        | 0.29     | 0.40     | 0.26     |
| IW 23F3  | 33-43-94  | K33N,K43N,Y94F  | 0.004          | 0.24           | 0.25        | 0.34     | 0.21     | 0.34     |
| IW+HT7   |           |                 | 1.00           | 1.00           | 1.00        | 1.00     | 1.00     | 1.00     |

FIG. 18

| Sample | position | AA             | Coel. | H           | H,H         | PBI-3939    | PBI-3945    | PBI-3889    | PBI-4002    |
|--------|----------|----------------|-------|-------------|-------------|-------------|-------------|-------------|-------------|
| 34B9   | 18       | Q18L           | 0.22  | 0.52        | 0.74        | 0.92        | 0.38        | 1.12        | 0.48        |
| 8E 3   | 27       | L27V           | 0.19  | 1.06        | 1.12        | <b>1.35</b> | 1.17        | <b>1.33</b> | 1.11        |
| 33C4   | 28       | S28Y           | 0.07  | 0.83        | 0.80        | 0.78        | 0.88        | 0.83        | 0.80        |
| 21C5   | 33       | K33N           | 0.96  | <b>1.41</b> | <b>1.68</b> | <b>1.68</b> | <b>1.36</b> | <b>1.74</b> | <b>1.34</b> |
| 25F10  | 36       | V36E           | 0.14  | 0.39        | 0.46        | 0.40        | 0.36        | 0.60        | 0.41        |
| 25H7   | 40       | P40T           | 0.003 | 0.13        | 0.27        | 0.47        | 0.28        | 0.46        | 0.23        |
| 28A8   | 135      | N135D          | 0.04  | 0.50        | 0.69        | 0.73        | 0.51        | 0.86        | 0.47        |
| 44G12  | 155      | I155T          | 0.08  | 0.51        | 0.85        | 0.63        | 0.48        | 0.87        | 0.48        |
| 46A6   | 32-6     | F6Y,Q32L       | 0.12  | 0.61        | 0.76        | 0.75        | 0.57        | 0.80        | 0.56        |
| 26D5   | 18-51-86 | Q18L,G51R,H86L | 0.47  | 0.95        | <b>1.39</b> | <b>1.58</b> | 0.62        | <b>1.95</b> | 0.83        |
| 3E 12  | 6-42-50  | F6Y,Q42H,N50K  | 0.12  | 0.60        | 0.84        | 1.03        | 0.61        | 1.04        | 0.61        |

FIG. 19

| Sequence                            | Sample | 19 | 20 | 27 | 32 | 38 | 43 | 49 | 58 | 77 | 95 | 110 | 149 |
|-------------------------------------|--------|----|----|----|----|----|----|----|----|----|----|-----|-----|
| Q20P,Q32P,E49G,I149I                | C1.1   |    | X  |    | X  |    |    | X  |    |    |    |     | X   |
| Q20P,Q32P,E49G,F77N,G95D,F110,I149I | C4.13  |    | X  |    | X  |    |    | X  |    | X  | X  | X   | X   |
| Q20P,I27N,V38I,I149I                | C5.16  |    | X  | X  |    | X  |    |    |    |    |    |     | X   |
| I27N,V38I,I149I                     | C5.19  |    |    | X  |    | X  |    |    |    |    |    |     | X   |
| D19N,Q20P,V38I,I149I                | C7.26  | X  | X  |    |    | X  |    |    |    |    |    |     | X   |
| V38I,G95D,F110,I149I                | C8.28  |    |    |    |    | X  |    |    |    |    | X  | X   | X   |
| D19N,Q20P,V38I,G95D,F110,I149I      | C8.29  | X  | X  |    |    | X  |    |    |    |    | X  | X   | X   |
| I27N,V38F,K43N,I149I                | C11.40 |    |    | X  |    | X  | X  |    |    |    |    |     | X   |
| I27N,V38F,K43N,V58I,I149I           | C11.42 |    |    | X  |    | X  | X  |    | X  |    |    |     | X   |
| I27N,V38F,K43N,V58I,F110,I149I      | C12.46 |    |    | X  |    | X  | X  |    | X  |    |    | X   | X   |
| Q20P,I27N,I149I                     | C13.50 |    | X  | X  |    |    |    |    |    |    |    |     | X   |
| Q20P,I27N,V38I,F110I                | C14.51 |    | X  | X  |    | X  |    |    |    |    |    | X   |     |
| I27N,V38I,F110,I149I                | C14.52 |    |    | X  |    | X  |    |    |    |    |    | X   | X   |

FIG. 20

| Sample  | Sequence                             | Codenterazine | H        | H/H      | PBI-3939 | PBI-3945 | PBI-4002 | PBI-3932 | PBI-3840 |
|---------|--------------------------------------|---------------|----------|----------|----------|----------|----------|----------|----------|
| C1.1    | Q20P,Q32P,E49G,L149I                 | 0.00006       | 0.00026  | 0.00033  | 0.00069  | 0.00050  | 0.00051  | 0.00014  | 0.00008  |
| C4.13   | Q20P,Q32P,E49G,F77Y,G95D,F110I,L149I | 0.00006       | 0.00008  | 0.00009  | 0.00008  | 0.00005  | 0.00006  | 0.00013  | 0.00010  |
| C5.16   | Q20P,L27V,V38I,L149I                 | 0.0002        | 0.0013   | 0.0003   | 0.0011   | 0.0034   | 0.0036   | 0.0001   | 0.0001   |
| C5.19   | L27V,V38I,L149I                      | 0.0010        | 0.2747   | 0.0663   | 0.1595   | 0.5082   | 0.5129   | 0.0015   | 0.0003   |
| C7.26   | D19N,Q20P,V38I,L149I                 | 0.0001        | 0.0017   | 0.0011   | 0.0020   | 0.0032   | 0.0037   | 0.0001   | 0.0000   |
| C8.29   | D19N,Q20P,V38I,G95D,F110I,L149I      | 0.000016      | 0.000025 | 0.000036 | 0.000016 | 0.000014 | 0.000031 | 0.000040 | 0.000025 |
| C11.40  | L27V,V38F,K43N,L149I                 | 0.00010       | 0.01172  | 0.00714  | 0.01853  | 0.04124  | 0.02485  | 0.00036  | 0.00006  |
| C11.42  | L27V,V38F,K43N,V58I,L149I            | 0.0001        | 0.0016   | 0.0011   | 0.0044   | 0.0093   | 0.0040   | 0.0001   | 0.0001   |
| C12.46  | L27V,V38F,K43N,V58I,F110I,L149I      | 0.00002       | 0.00004  | 0.00005  | 0.00001  | 0.00008  | 0.00006  | 0.00006  | 0.00004  |
| C13.50  | Q20P,L27V,L149I                      | 0.0001        | 0.0124   | 0.0036   | 0.0027   | 0.0109   | 0.0100   | 0.0001   | 0.0000   |
| C14.51  | Q20P,L27V,V38I,F110I                 | 0.00003       | 0.00010  | 0.00004  | 0.00001  | 0.00008  | 0.00011  | 0.00003  | 0.00004  |
| C14.52  | L27V,V38I,F110I,L149I                | 0.00007       | 0.00041  | 0.00008  | 0.00006  | 0.00034  | 0.00046  | 0.00012  | 0.00008  |
| IVY-H17 |                                      | 1             | 1        | 1        | 1        | 1        | 1        | 1        | 1        |

FIG. 21

| Sequence                                     |  | clone  | 6 | 18 | 27 | 28 | 33 | 34 | 36 | 40 | 50 | 51 | 135 | 155 |
|----------------------------------------------|--|--------|---|----|----|----|----|----|----|----|----|----|-----|-----|
| F6Y,Q18L,K33N,N135D                          |  | C1.1   | X | X  |    |    | X  |    |    |    |    |    | X   |     |
| F6Y,Q18L,K33N,N135D,I155T                    |  | C1.2   | X | X  |    |    | X  |    |    |    |    |    | X   | X   |
| F6Y,K33N,N135D,I155T                         |  | C1.3   | X |    |    |    | X  |    |    |    |    |    | X   | X   |
| K33N,P40T,N135D,I155T                        |  | C2.6   |   |    |    |    | X  |    | X  |    |    |    | X   | X   |
| F6Y,Q18L,K33N,P40T,N135D,I155T               |  | C2.7   | X | X  |    |    | X  |    | X  |    |    |    | X   | X   |
| F6Y,K33N,P40T,N135D,I155T                    |  | C2.8   | X |    |    |    | X  |    | X  |    |    |    | X   | X   |
| F6Y,Q18L,K33N,V36E,P40T,N50S,G51RN135D,I155T |  | C3.13  | X | X  |    |    | X  |    | X  | X  |    |    | X   | X   |
| F6Y,Q18L,N135D,I155T                         |  | C4.16  | X | X  |    |    |    |    |    |    |    |    | X   | X   |
| Q18L,N135D,I155T                             |  | C4.17  |   | X  |    |    |    |    |    |    |    |    | X   | X   |
| F6Y,Q18L,L27V,S28Y,N135D,I155T               |  | C4.18  | X | X  | X  | X  |    |    |    |    |    |    | X   | X   |
| F6Y,Q18L,L27V,S28Y,P40T                      |  | C5.23  | X | X  | X  | X  |    |    |    | X  |    |    |     |     |
| Q18L,L27V,S28Y,K33N,V36E,N135D,I155T         |  | C6.26  |   | X  | X  | X  | X  |    | X  |    |    |    | X   | X   |
| F6Y,Q18L,L27V,S28Y,K33N,V36E,N135D,I155T     |  | C6.28  | X | X  | X  | X  | X  |    | X  |    |    |    | X   | X   |
| F6Y,Q18L,P40T,N135D,I155T                    |  | C7.31  | X | X  |    |    |    |    |    | X  |    |    | X   | X   |
| F6Y,Q18L,L27V,S28Y,K33N,P40T,N135D,I155T     |  | C7.33  | X | X  | X  | X  | X  |    |    | X  |    |    | X   | X   |
| Q18L,P40T,N135D,I155T                        |  | C7.34  |   | X  |    |    |    |    |    | X  |    |    | X   | X   |
| F6Y,Q18L,K33M,L34M,N135D,I155T               |  | C8.36  | X | X  |    |    | X  | X  |    |    |    |    | X   | X   |
| Q18L,K33M,L34M,N135D,I155T                   |  | C8.38  |   | X  |    |    | X  | X  |    |    |    |    | X   | X   |
| F6Y,Q18L,K33M,L34M,I55T                      |  | C8.40  | X | X  |    |    | X  | X  |    |    |    |    |     | X   |
| F6Y,Q18L,K33M,L34M,P40T,N135D                |  | C9.44  | X | X  |    |    | X  | X  |    | X  |    |    | X   |     |
| F6Y,Q18L,L27V,S28Y,N50S,G51R,N135D           |  | C10.46 | X | X  | X  | X  |    |    |    |    | X  | X  | X   |     |
| F6Y,Q18L,N50S,G51R,N135D,I155T               |  | C10.48 | X | X  |    |    |    |    |    |    | X  | X  | X   | X   |
| Q18L,N50S,G51R,N135D,I155T                   |  | C10.49 |   | X  |    |    |    |    |    |    | X  | X  | X   | X   |
| Q18L,L27V,S28Y,N50S,G51R,N135D,I155T         |  | C10.50 |   | X  | X  | X  |    |    |    |    | X  | X  | X   | X   |
| F6Y,Q18L,L27V,S28Y,N50S,G51R,N135D,I155T     |  | C11.54 | X | X  | X  | X  |    |    |    |    | X  | X  | X   | X   |

FIG. 22

| Sample | Sequence                                      | Codentertazine | H         | HH        | PBI-3939  | PBI-3945  | PBI-3889  | PBI-4002  | PBI-3932  | PBI-3840  |
|--------|-----------------------------------------------|----------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| C1.1   | F6Y,Q18L,K33N,N135D                           | 0.0055128      | 0.130     | 0.463     | 0.444     | 0.127     | 0.290     | 0.159     | 0.0083435 | 0.017     |
| C1.2   | F6Y,Q18L,K33N,N135D,I155T                     | 0.0000068      | 0.0015688 | 0.0043713 | 0.0066058 | 0.0013479 | 0.0027420 | 0.0009303 | 0.0000844 | 0.0000782 |
| C1.3   | F6Y,K33N,N135D,I155T                          | 0.0000257      | 0.0088297 | 0.036     | 0.048     | 0.0083438 | 0.023     | 0.0057396 | 0.0002600 | 0.0004430 |
| C2.6   | K33N,P40T,N135D,I155T                         | 0.0000009      | 0.0000288 | 0.0001711 | 0.0017877 | 0.0001365 | 0.0002859 | 0.0000858 | 0.0000035 | 0.0000025 |
| C2.7   | F6Y,Q18L,K33N,P40T,N135D,I155T                | 0.0000008      | 0.0000091 | 0.0000318 | 0.0000611 | 0.0002445 | 0.0003744 | 0.0002560 | 0.0000013 | 0.0000020 |
| C2.8   | F6Y,K33N,P40T,N135D,I155T                     | 0.0000004      | 0.0000107 | 0.0000641 | 0.0007157 | 0.0000848 | 0.0001958 | 0.0000447 | 0.0000019 | 0.0000011 |
| C3.13  | F6Y,Q18L,K33N,V36E,P40T,N50S,G51R,N135D,I155T | 0.0000003      | 0.0000006 | 0.0000015 | 0.0000037 | 0.0000010 | 0.0000031 | 0.0000018 | 0.0000064 | 0.0000017 |
| C4.16  | F6Y,Q18L,N135D,I155T                          | 0.0000299      | 0.0046508 | 0.012     | 0.017     | 0.0052284 | 0.010     | 0.0037895 | 0.0000946 | 0.0002504 |
| C4.17  | Q18L,N135D,I155T                              | 0.0000222      | 0.0032164 | 0.0081703 | 0.011     | 0.0034948 | 0.0066376 | 0.0026060 | 0.0000717 | 0.0001691 |
| C4.18  | F6Y,Q18L,I27V,S28Y,N135D,I155T                | 0.0000008      | 0.0000325 | 0.0001094 | 0.0003355 | 0.0001157 | 0.0001447 | 0.0000548 | 0.0000060 | 0.0000025 |
| C5.23  | F6Y,Q18L,I27V,S28Y,P40T                       | 0.0000579      | 0.0002759 | 0.0000464 | 0.0004978 | 0.0017611 | 0.0002812 | 0.0013044 | 0.0000144 | 0.0000132 |
| C6.26  | Q18L,I27V,S28Y,K33N,V36E,N135D,I155T          | 0.0000055      | 0.0000276 | 0.0001950 | 0.0004882 | 0.0000838 | 0.0003453 | 0.0000663 | 0.0003772 | 0.0000586 |
| C6.28  | F6Y,Q18L,I27V,S28Y,K33N,V36E,N135D,I155T      | 0.0000016      | 0.0000067 | 0.0000451 | 0.0001166 | 0.0000200 | 0.0000805 | 0.0000158 | 0.0000852 | 0.0000157 |
| C7.31  | F6Y,Q18L,P40T,N135D,I155T                     | 0.0000003      | 0.0000017 | 0.0000024 | 0.0000174 | 0.0000084 | 0.0000113 | 0.0000067 | 0.0000018 | 0.0000005 |
| C7.33  | F6Y,Q18L,I27V,S28Y,K33N,P40T,N135D,I155T      | 0.0000004      | 0.0000007 | 0.0000013 | 0.0000017 | 0.0000008 | 0.0000011 | 0.0000006 | 0.0000016 | 0.0000009 |
| C7.34  | Q18L,P40T,N135D,I155T                         | 0.0000007      | 0.0000021 | 0.0000024 | 0.0000253 | 0.0000131 | 0.0000159 | 0.0000101 | 0.0000048 | 0.0000009 |
| C8.36  | F6Y,Q18L,K33M,L34M,N135D,I155T                | 0.0000024      | 0.0000249 | 0.0000898 | 0.0001137 | 0.0000298 | 0.0000823 | 0.0000269 | 0.0000163 | 0.0000195 |
| C8.38  | Q18L,K33M,L34M,N135D,I155T                    | 0.0000047      | 0.0000956 | 0.0003420 | 0.0004501 | 0.0001128 | 0.0003207 | 0.0001027 | 0.0000476 | 0.0000710 |
| C8.40  | F6Y,Q18L,K33M,L34M,I155T                      | 0.0002632      | 0.010     | 0.015     | 0.018     | 0.011     | 0.015     | 0.0097703 | 0.0008447 | 0.0018540 |
| C9.44  | F6Y,Q18L,K33M,L34M,P40T,N135D                 | 0.0000041      | 0.0000139 | 0.0000153 | 0.0000379 | 0.0000423 | 0.0000474 | 0.0000534 | 0.0000284 | 0.0000486 |
| C10.46 | F6Y,Q18L,I27V,S28Y,N50S,G51R,N135D            | 0.0002613      | 0.016     | 0.015     | 0.030     | 0.026     | 0.014     | 0.014     | 0.0004421 | 0.0002678 |
| C10.48 | F6Y,Q18L,N50S,G51R,N135D,I155T                | 0.0000064      | 0.0005384 | 0.0011439 | 0.0016621 | 0.0006785 | 0.0010704 | 0.0004835 | 0.0000176 | 0.0000290 |
| C10.49 | Q18L,N50S,G51R,N135D,I155T                    | 0.0000184      | 0.0023853 | 0.0052253 | 0.0072753 | 0.0028335 | 0.0045995 | 0.0021987 | 0.0000853 | 0.0001334 |
| C10.50 | Q18L,I27V,S28Y,N50S,G51R,N135D,I155T          | 0.0000095      | 0.0000547 | 0.0001427 | 0.0004411 | 0.0002044 | 0.0001929 | 0.0000965 | 0.0000697 | 0.0000099 |
| C11.54 | F6Y,Q18L,I27V,S28Y,N50S,G51R,N135D,I155T      | 0.0000009      | 0.0000157 | 0.0000467 | 0.0001215 | 0.0000541 | 0.0000547 | 0.0000266 | 0.0000065 | 0.0000023 |

FIG. 23

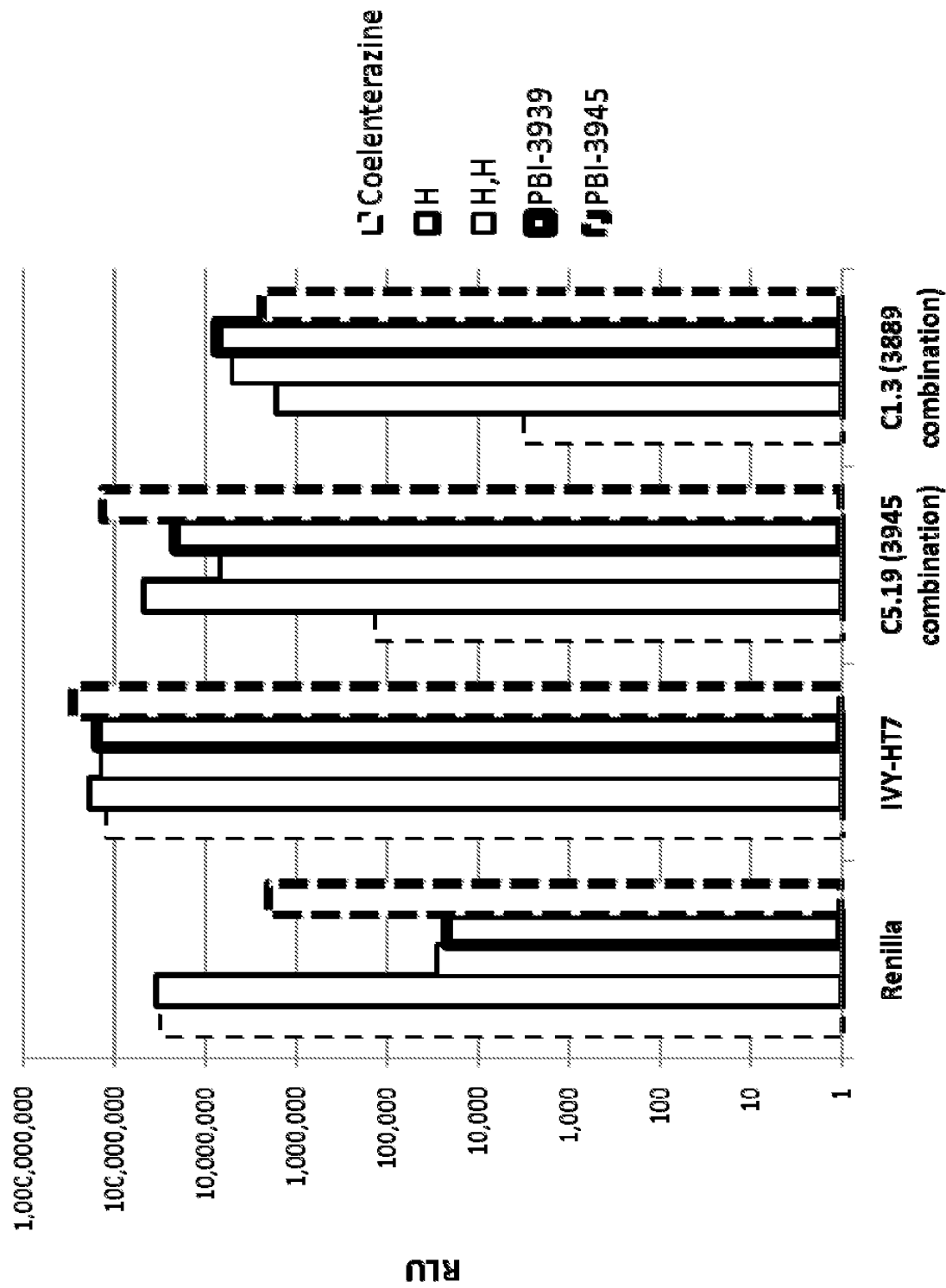


FIG. 24

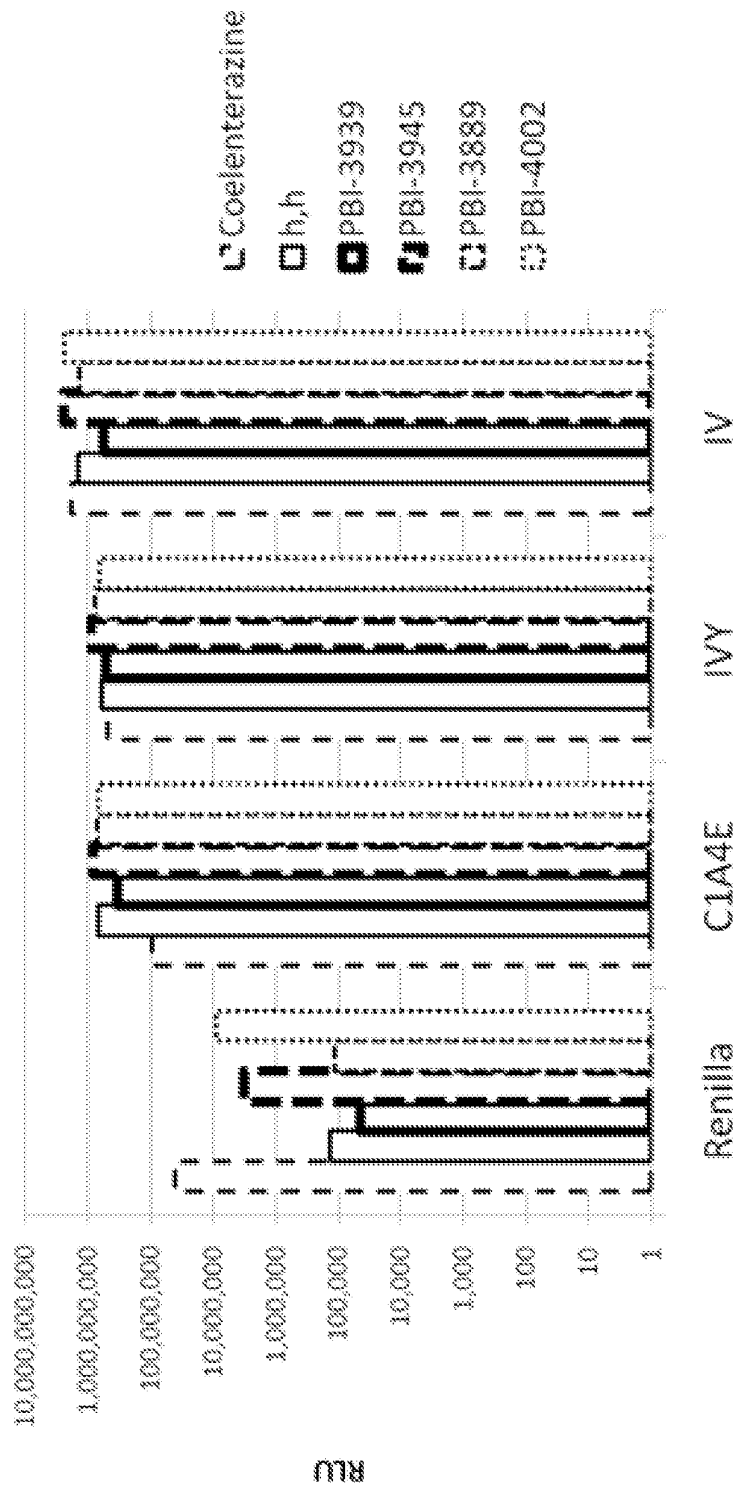


FIG. 25

| Sequence                      | done  | 1 | 4 | 18 | 27 | 38 | 38 | 68 | 70 | 72 | 75 | 102 |
|-------------------------------|-------|---|---|----|----|----|----|----|----|----|----|-----|
| Q18L                          | 1D6   |   |   | X  |    |    |    |    |    |    |    |     |
| F1J,L27A,V88                  | 8A3   | X |   |    | X  |    | X  |    |    |    |    |     |
| F1J,L27V                      | 8A7   | X |   |    | X  |    |    |    |    |    |    |     |
| F1J,V88                       | 8E2   | X |   |    |    |    | X  |    |    |    |    |     |
| L27V                          | 8F2   |   |   |    | X  |    |    |    |    |    |    |     |
| M75K,L72Q,F68Y,Q18L           | 9B8   |   |   | X  |    |    |    | X  |    | X  | X  |     |
| M75K,L72Q,F68Y,L27A,Q18L,V88  | 9F6   |   |   | X  | X  |    | X  | X  |    | X  | X  |     |
| F1J,M75K,L72Q,F68Y,L27A,V88   | 10A7  | X |   |    | X  |    | X  | X  |    | X  | X  |     |
| F1J,M75K,L72Q,M70V,Q18L       | 10G10 | X |   | X  |    |    |    |    | X  | X  | X  |     |
| K33N                          | 14A2  |   |   |    |    |    | X  |    |    |    |    |     |
| F1J,M75K,L72Q,F68Y,K33N,M102E | 14C4  | X |   |    |    |    | X  |    |    | X  | X  | X   |
| F4K,M75K,L72Q,F68Y,K33N       | 15C1  |   | X |    |    |    | X  |    |    | X  | X  |     |

FIG. 26

| Sample | Sequence                      | Coel. | H    | HH   | PBI-3939 | PBI-3945 | PBI-3889 | PBI-4002 |
|--------|-------------------------------|-------|------|------|----------|----------|----------|----------|
| IV     |                               | 1     | 1    | 1    | 1        | 1        | 1        | 1        |
| 1D6    | Q18L                          | 0.77  | 0.92 | 2.16 | 4.08     | 1.04     | 2.27     | 1.32     |
| 8A3    | F1I,L27V,V38I                 | 0.01  | 0.81 | 0.87 | 0.43     | 0.90     | 0.48     | 0.77     |
| 8A7    | F1I,L27V                      | 0.43  | 1.55 | 1.95 | 1.26     | 1.50     | 1.50     | 1.50     |
| 8E 2   | F1I,V38I                      | 0.13  | 0.53 | 0.61 | 0.75     | 0.60     | 0.64     | 0.51     |
| 8F2    | L27V                          | 0.40  | 1.54 | 1.87 | 1.06     | 1.53     | 1.37     | 1.49     |
| 9B8    | M75K,L72Q,F68Y,Q18L           | 1.40  | 1.69 | 2.52 | 4.37     | 1.44     | 2.35     | 1.72     |
| 9F6    | M75K,L72Q,F68Y,L27V,Q18L,V38I | 0.09  | 2.05 | 1.67 | 1.11     | 1.98     | 1.04     | 1.62     |
| 10A7   | F1I,M75K,L72Q,F68Y,L27V,V38I  | 0.08  | 1.40 | 1.17 | 0.82     | 1.74     | 0.89     | 1.23     |
| 10G10  | F1I,M75K,L72Q,M70V,Q18L       | 1.03  | 1.51 | 2.30 | 4.25     | 1.35     | 2.33     | 1.60     |
| 14A2   | K33N                          | 1.12  | 1.62 | 1.93 | 1.80     | 1.80     | 2.23     | 1.66     |
| 14C4   | F1I,M75K,L72Q,F68Y,K33N,V102E | 1.27  | 1.33 | 1.08 | 1.36     | 1.53     | 0.84     | 1.11     |
| 15C1   | E4K,M75K,L72Q,F68Y,K33N       | 2.63  | 2.61 | 2.10 | 2.60     | 2.94     | 1.67     | 2.20     |

FIG. 27

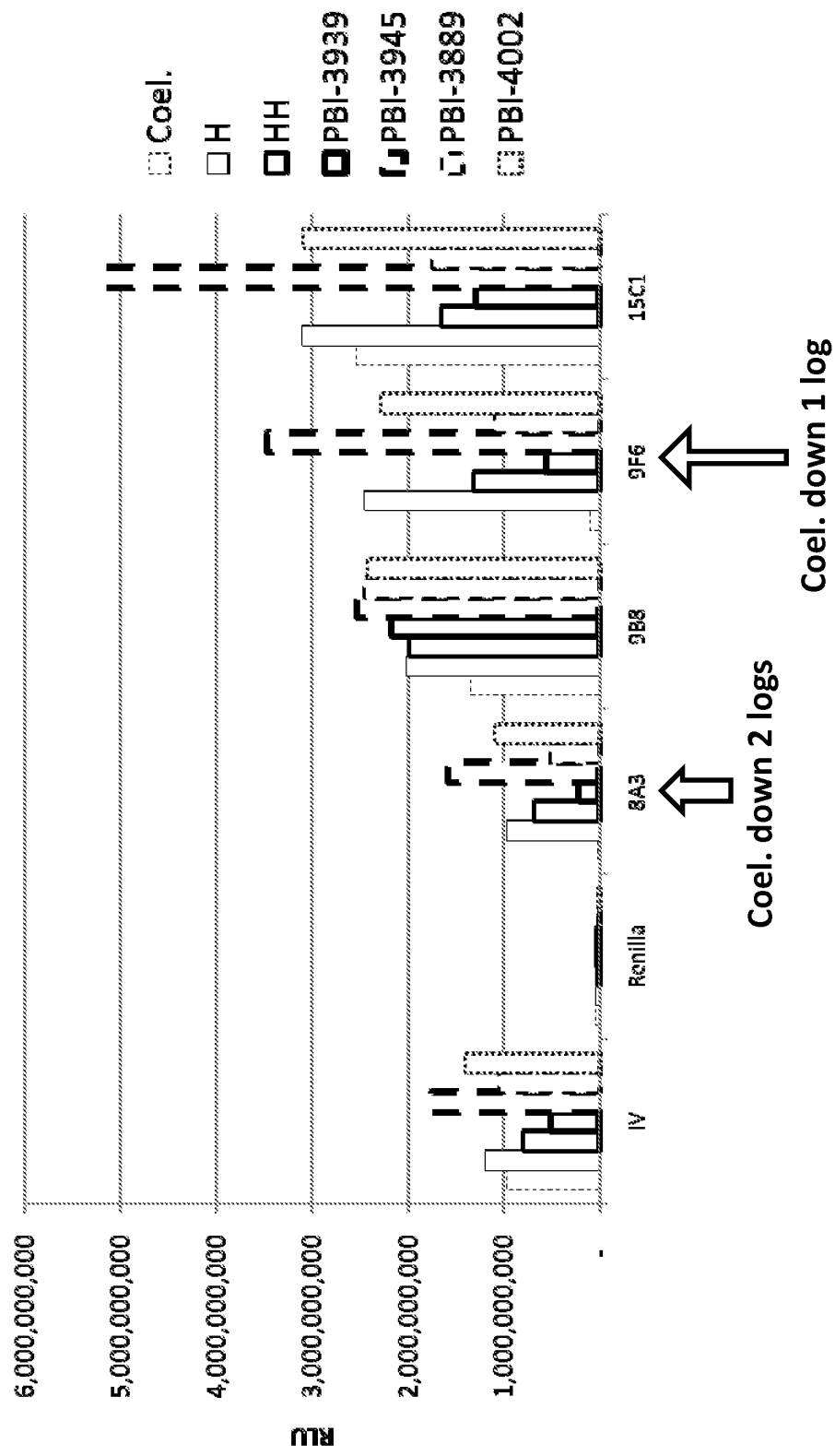


FIG. 28

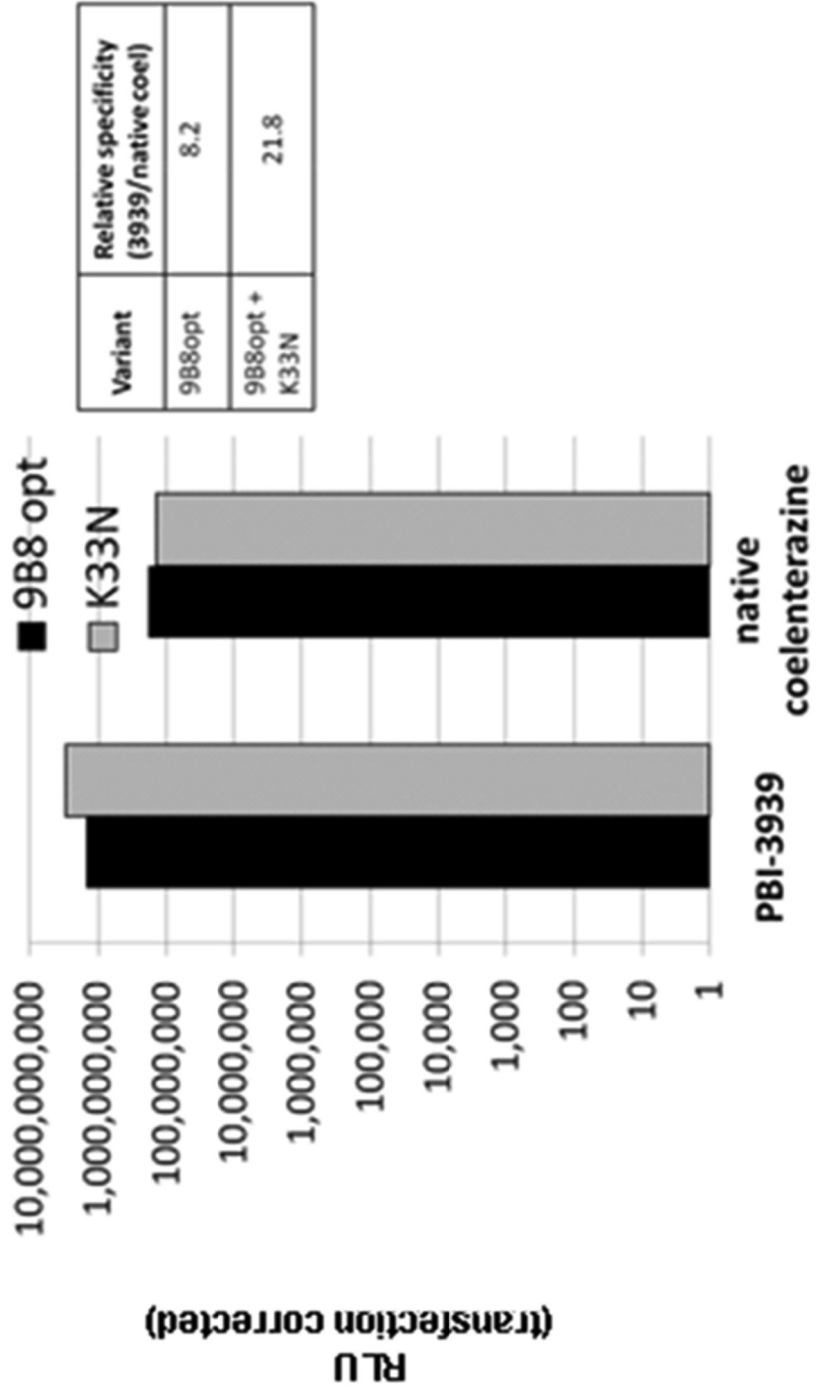


FIG. 29

|   |                                            |                                         |
|---|--------------------------------------------|-----------------------------------------|
| A | Native coelenterazine                      |                                         |
|   | Amino acid                                 | Relative activity                       |
|   | Ala                                        | 1.0                                     |
|   | Phe                                        | 3.2                                     |
|   | Lys                                        | 19.2                                    |
| C | PBI-3939                                   |                                         |
|   | Amino acid                                 | Relative activity                       |
|   | Ser                                        | 1.8                                     |
|   | Glu                                        | 4.6                                     |
|   | Ile                                        | 10.0                                    |
| B | Coelenterazine-h                           |                                         |
|   | Amino acid                                 | Relative activity                       |
|   | Ser                                        | 1.1                                     |
|   | Ile                                        | 1.4                                     |
|   | Lys                                        | 11.6                                    |
| D | Relative activity (in context of wt OgLuc) |                                         |
|   | Amino acid substitution                    | Relative activity (in context of N166R) |
|   | L27V                                       | 62.2                                    |
|   | A33N                                       | 0.6                                     |
|   | K43R                                       | 1.1                                     |
|   | Relative activity (in context of wt OgLuc) |                                         |
|   | Amino acid substitution                    | Relative activity (in context of N166R) |
|   | M75K                                       | 65.7                                    |
|   | T39T                                       | 118.6                                   |
|   | L72Q                                       | 2.0                                     |
|   | Relative activity (in context of wt OgLuc) |                                         |
|   | Amino acid substitution                    | Relative activity (in context of N166R) |
|   | F68D                                       | 3.0                                     |
|   |                                            | 5.4                                     |

FIG. 30

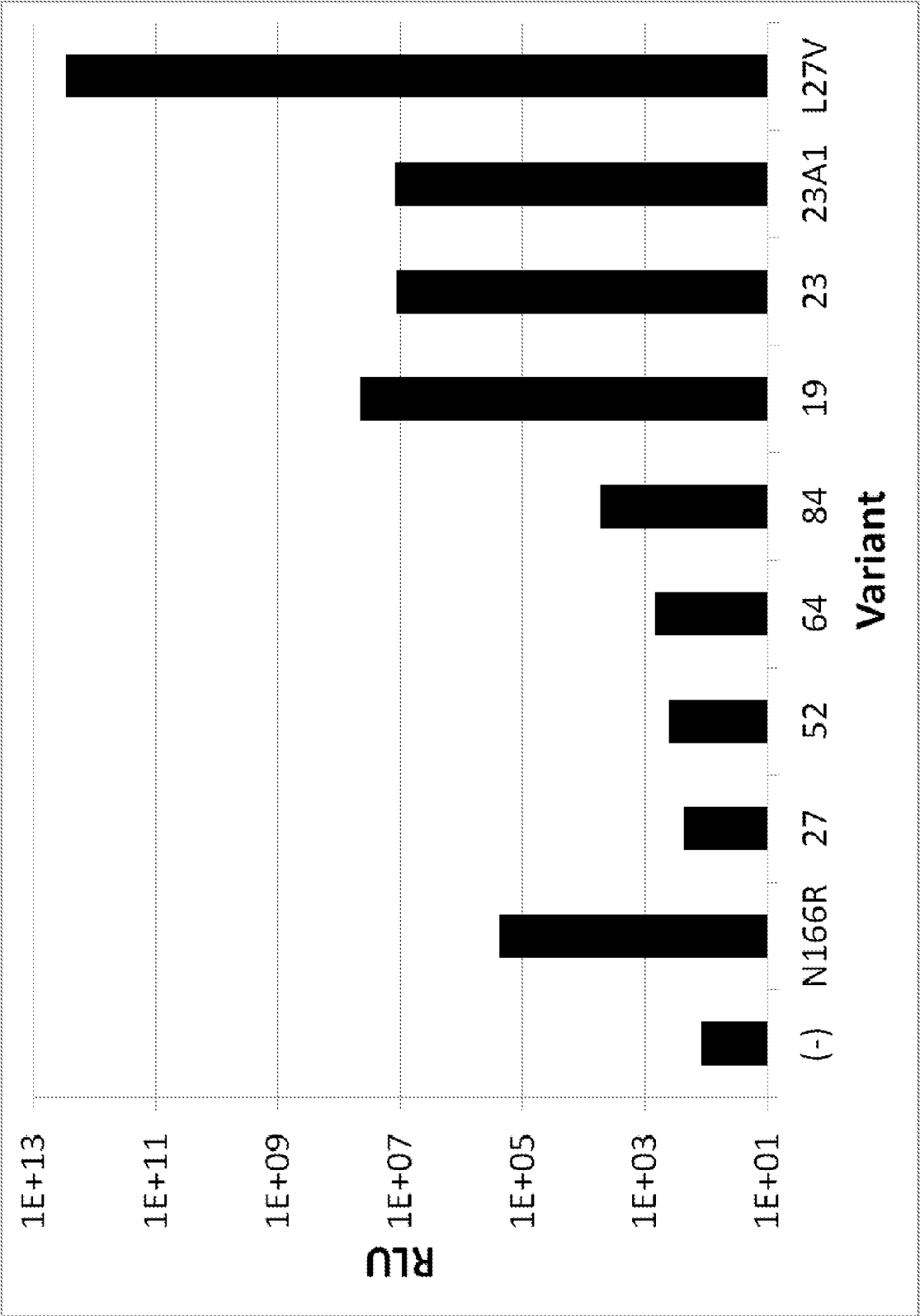


FIG. 31

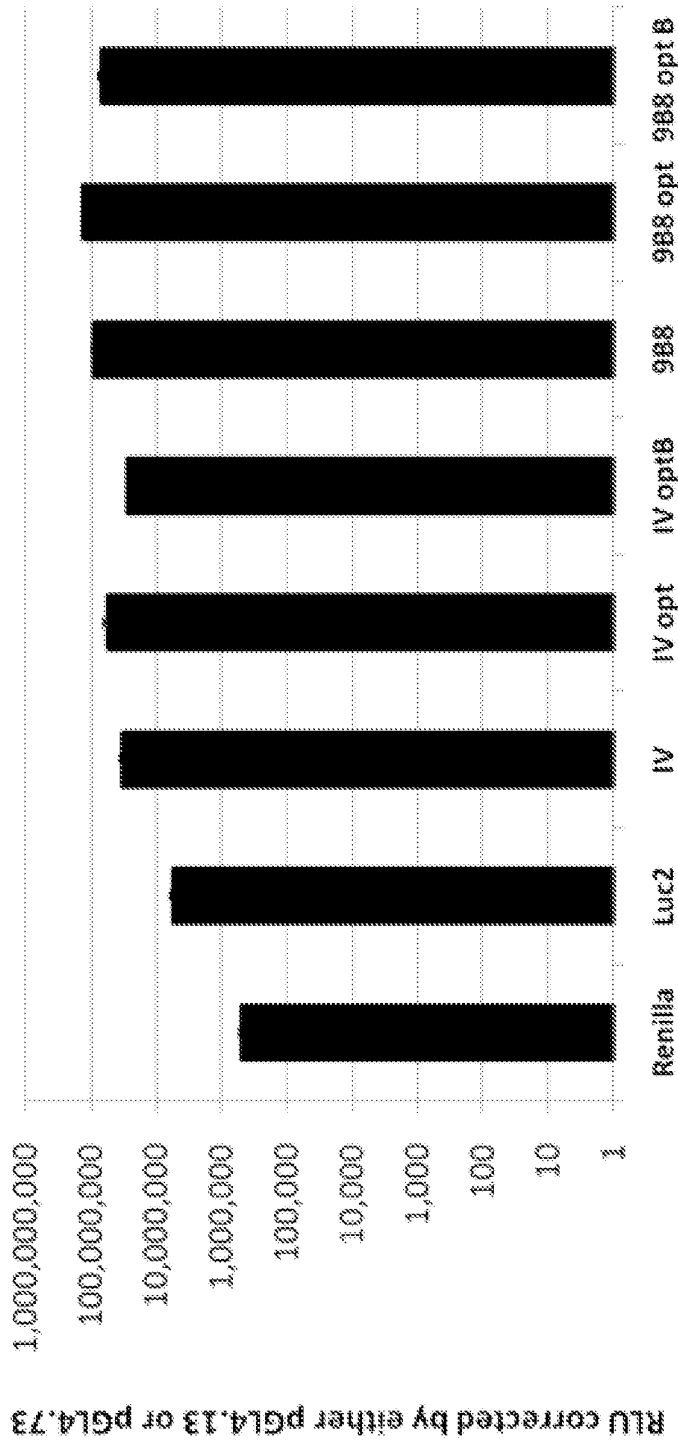


FIG. 32

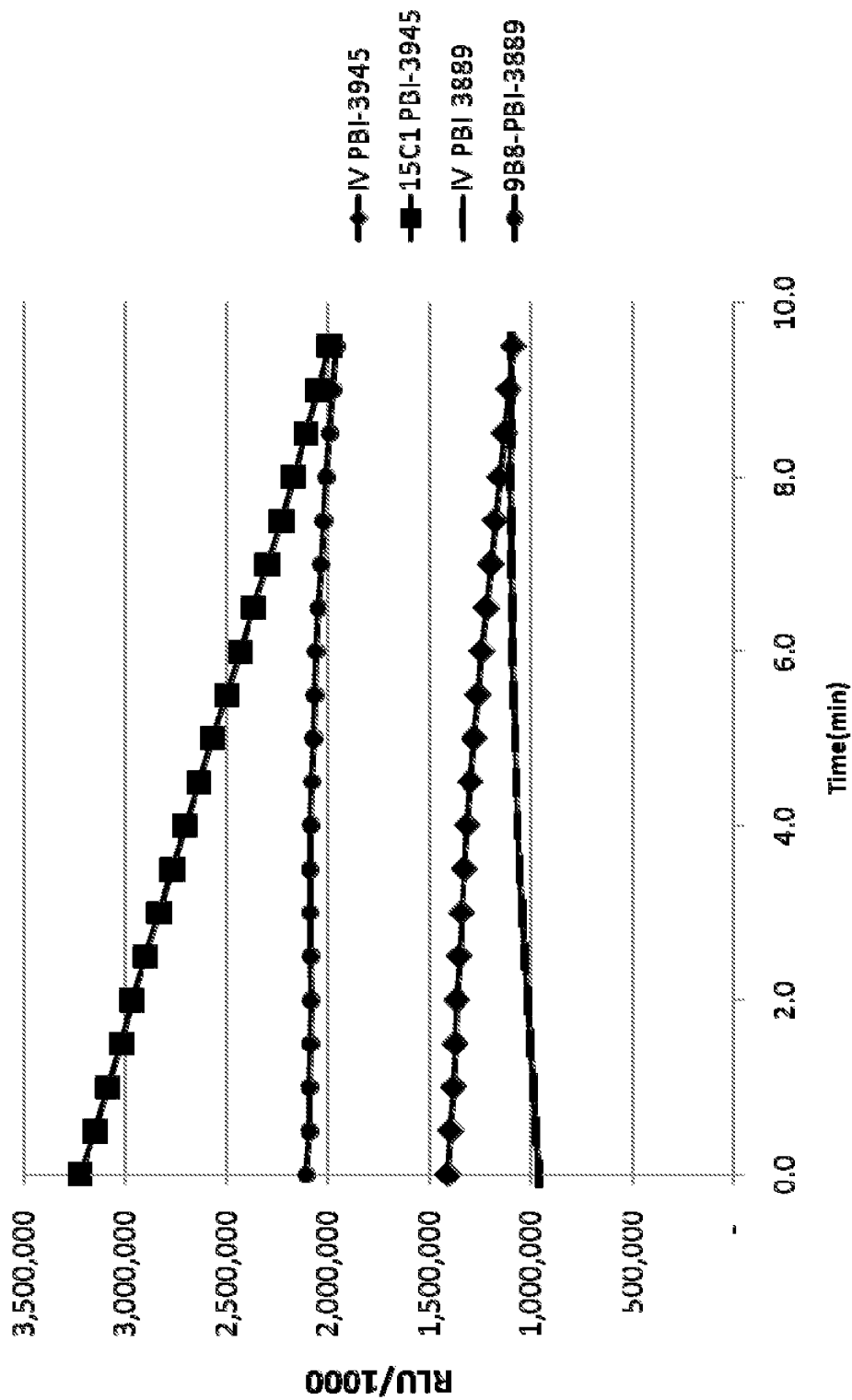


FIG. 33

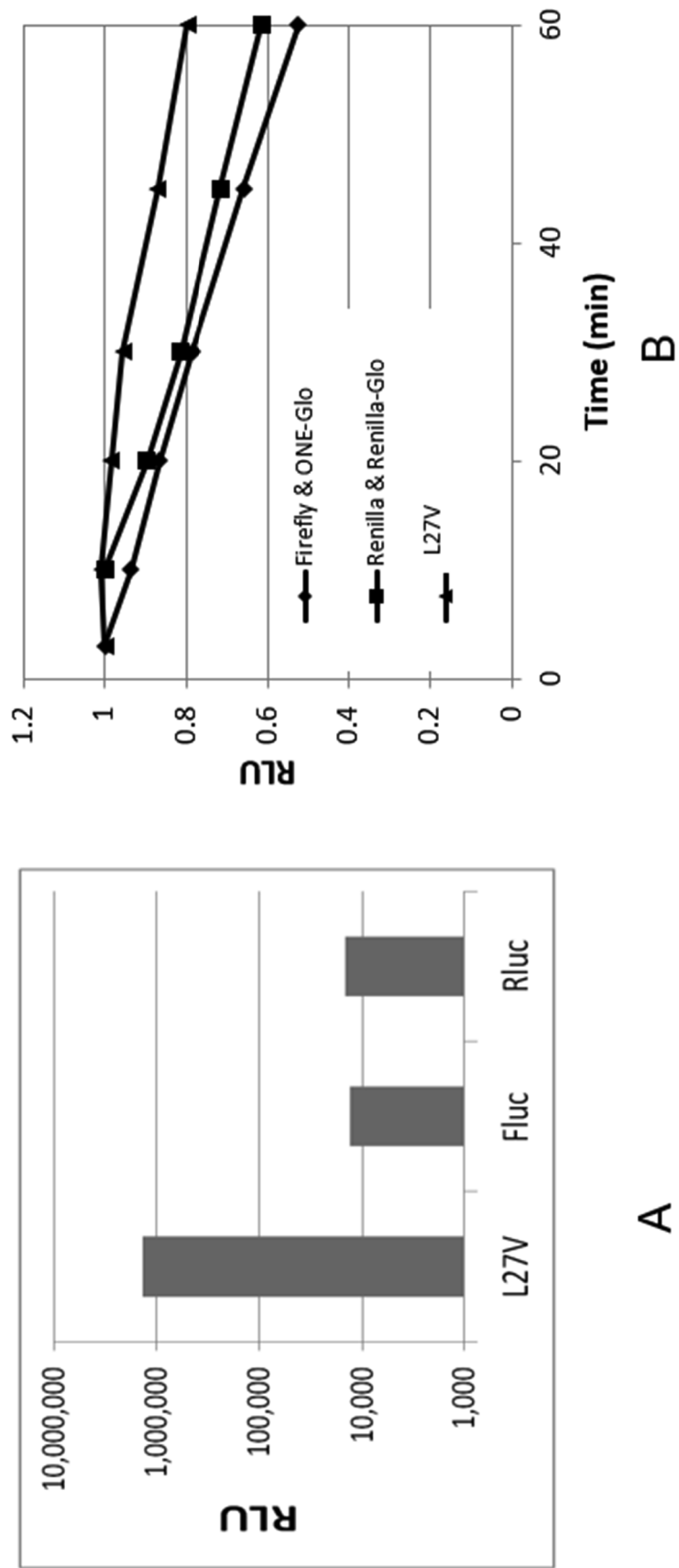


FIG. 34

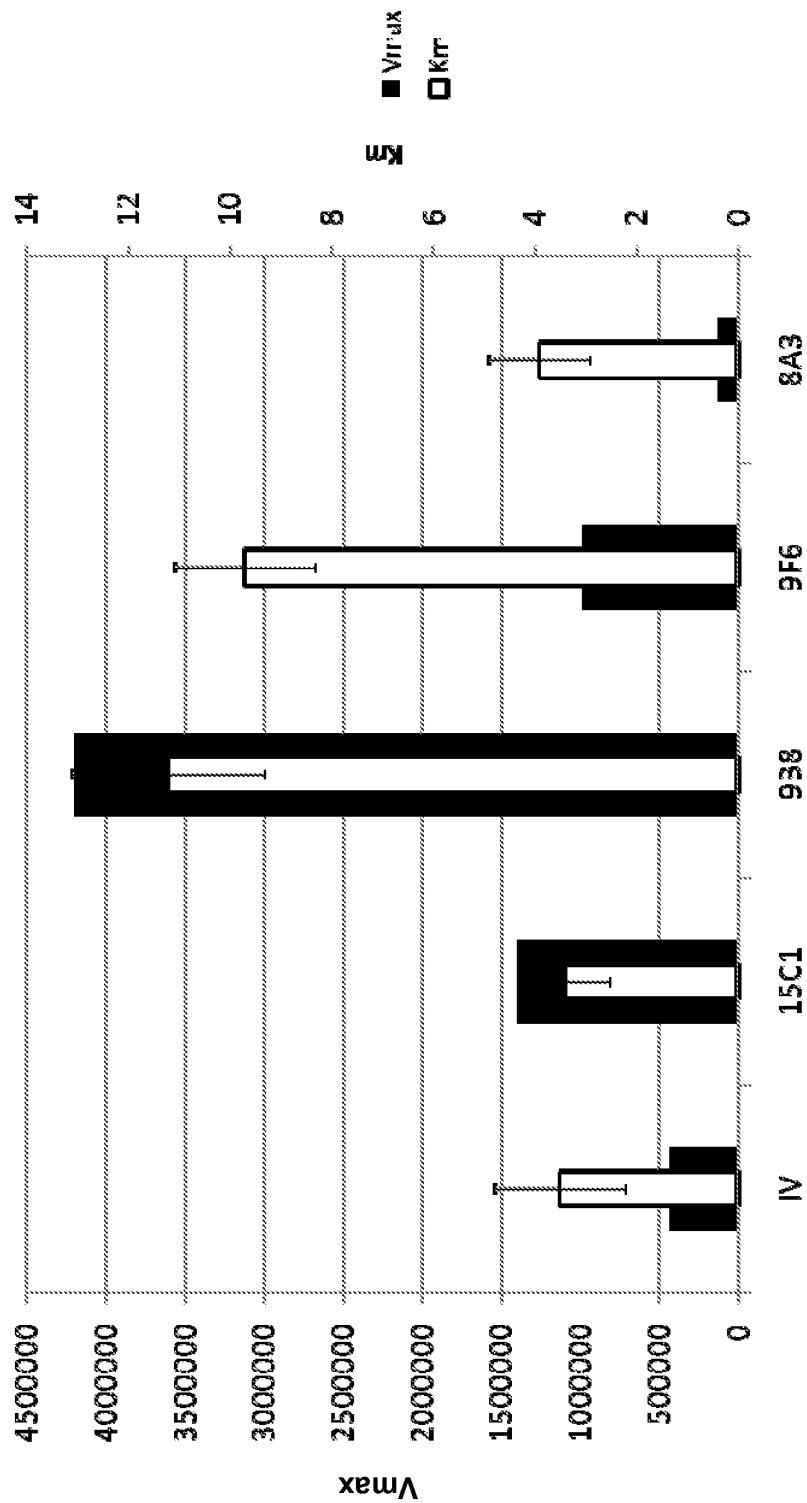
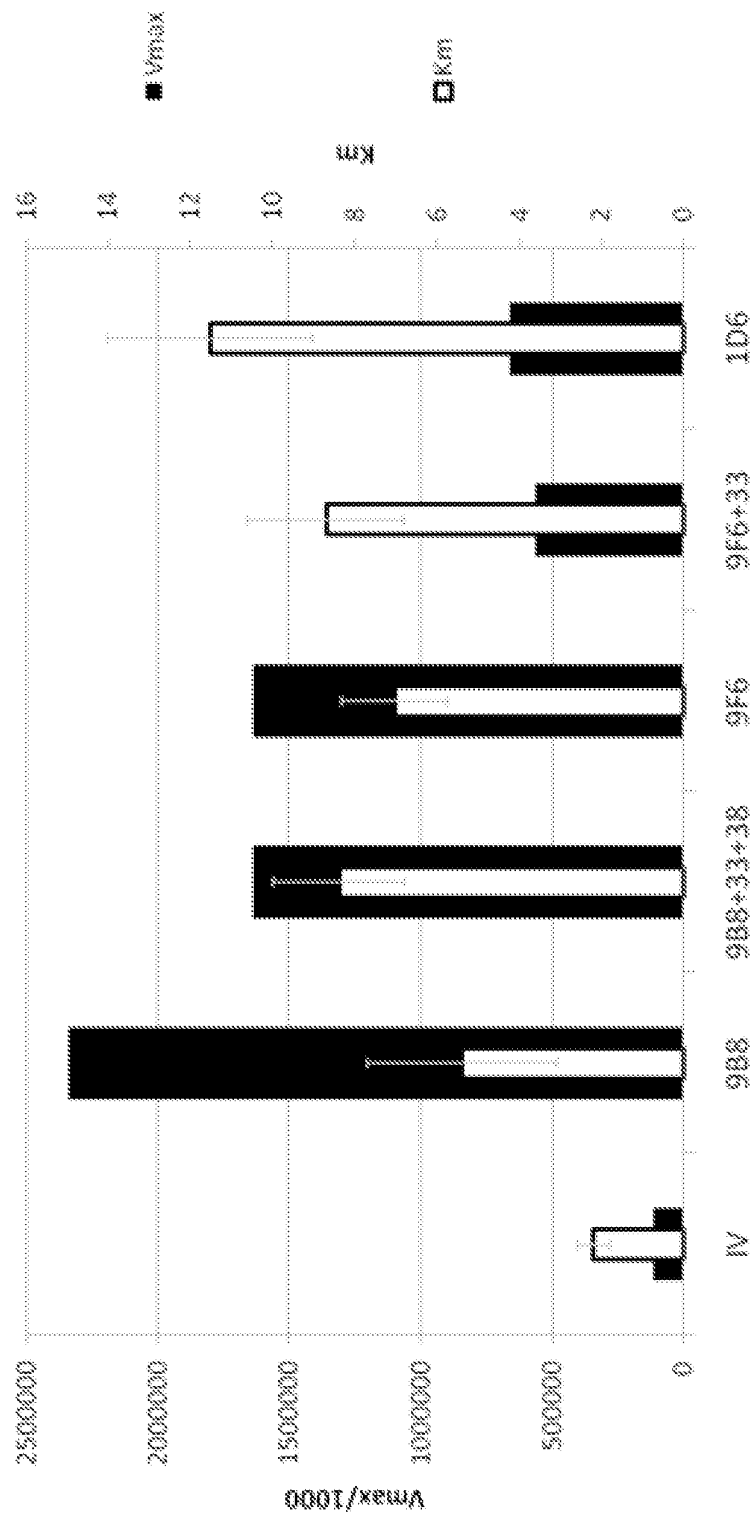


FIG. 35



Position 33 is K33N  
Position 38 is V38I

FIG. 36

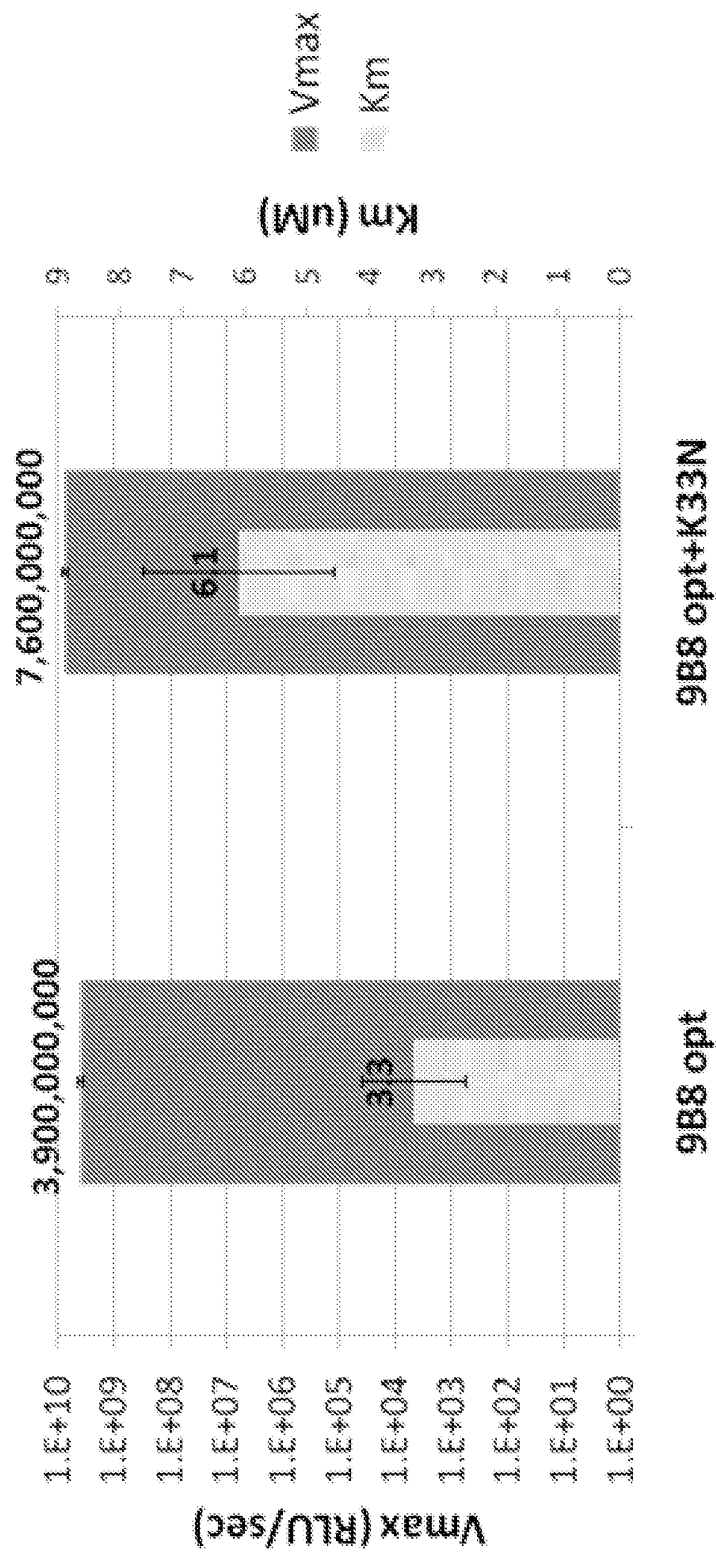


FIG. 37

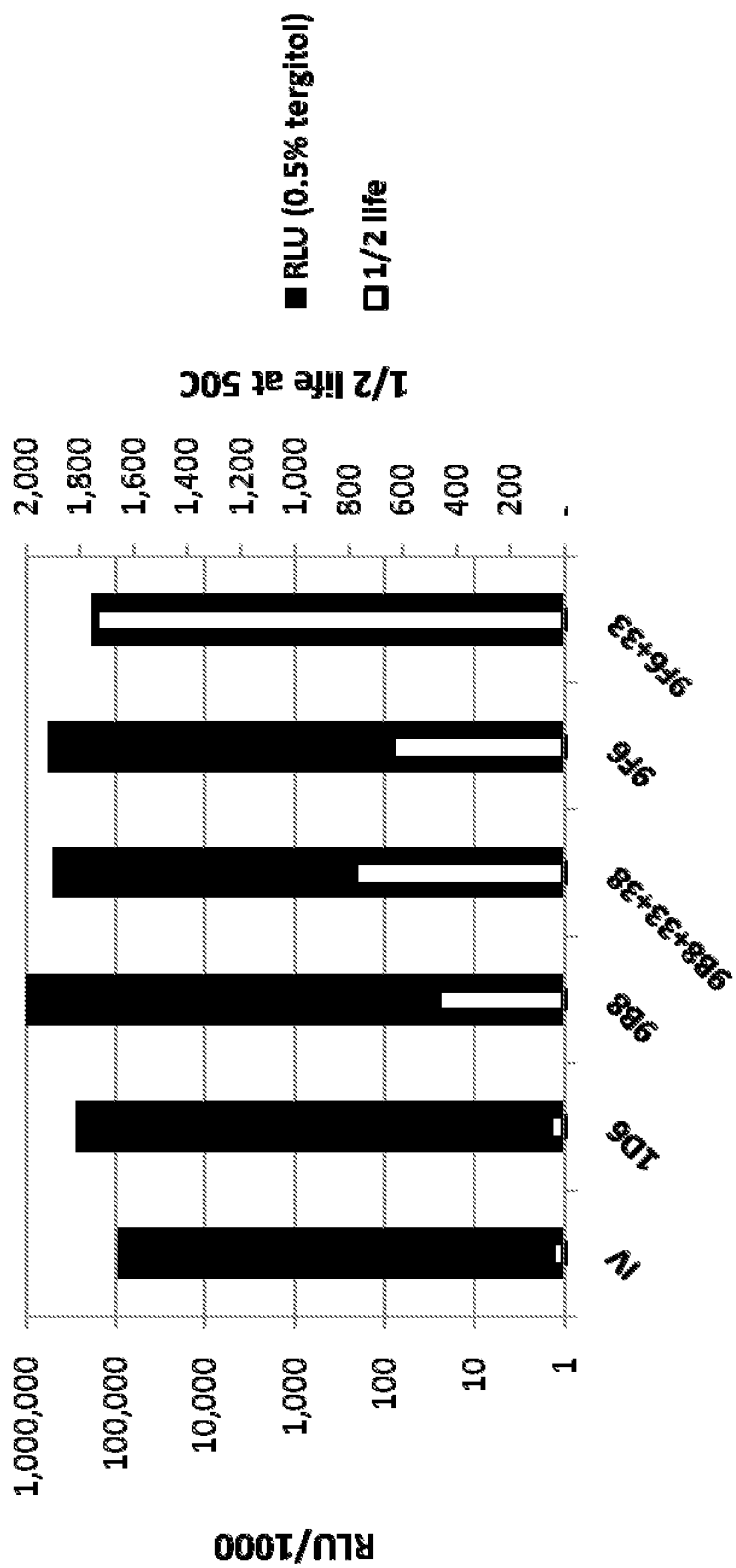


FIG. 38

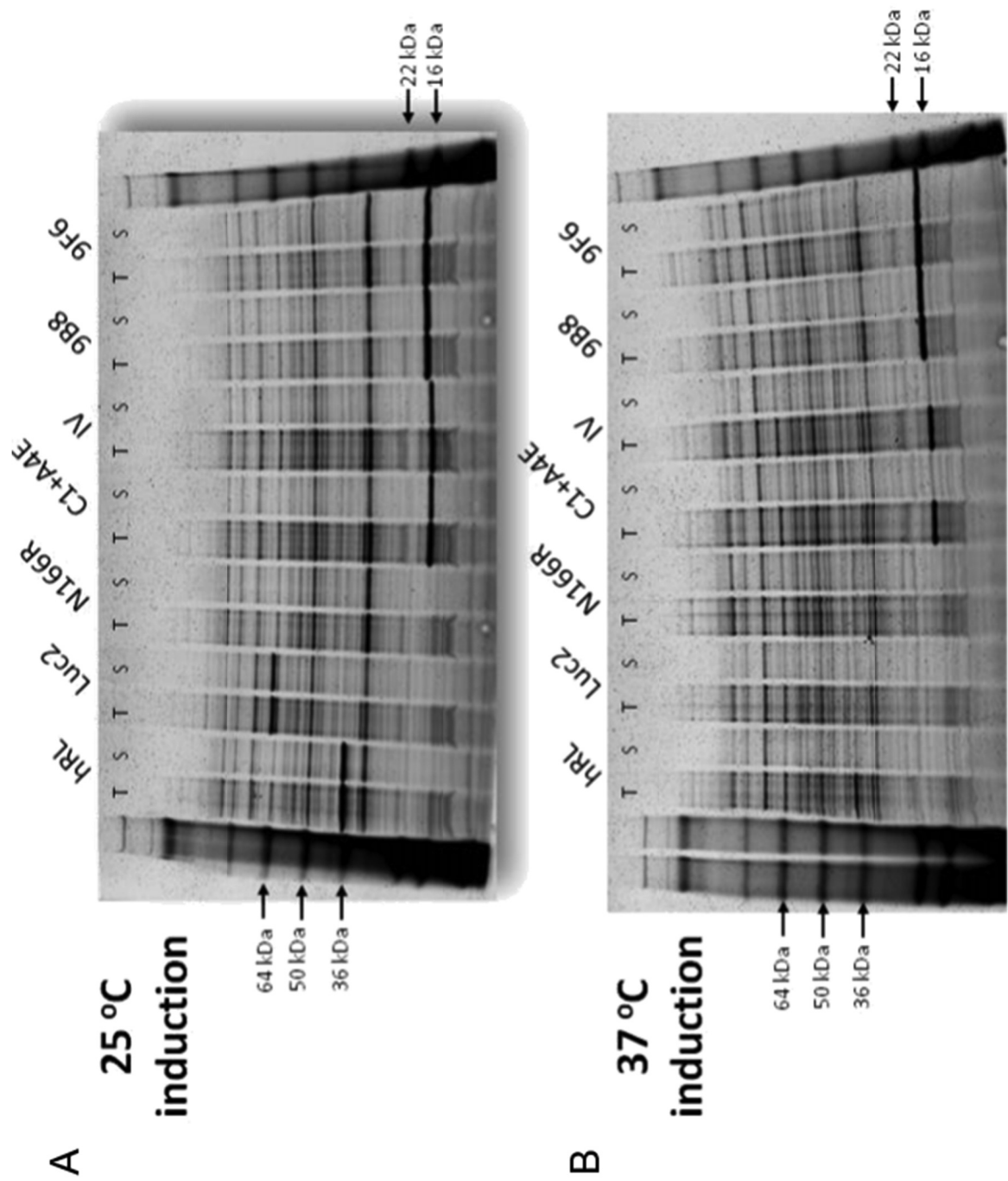
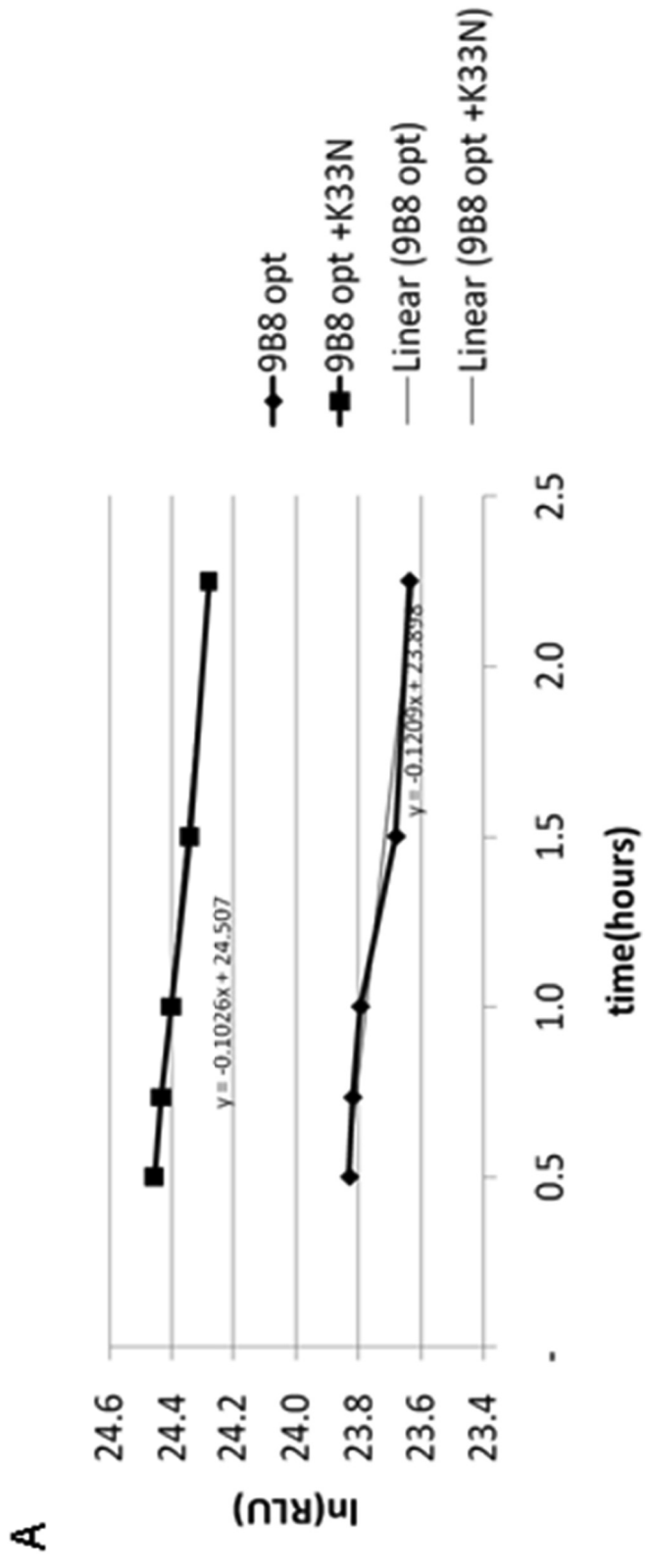


FIG. 39



**B**

| Sample        | 1/2 life (hours) 60C |
|---------------|----------------------|
| 9B8 opt       | 5.7                  |
| 9B8 opt +K33N | 6.8                  |

FIG. 40

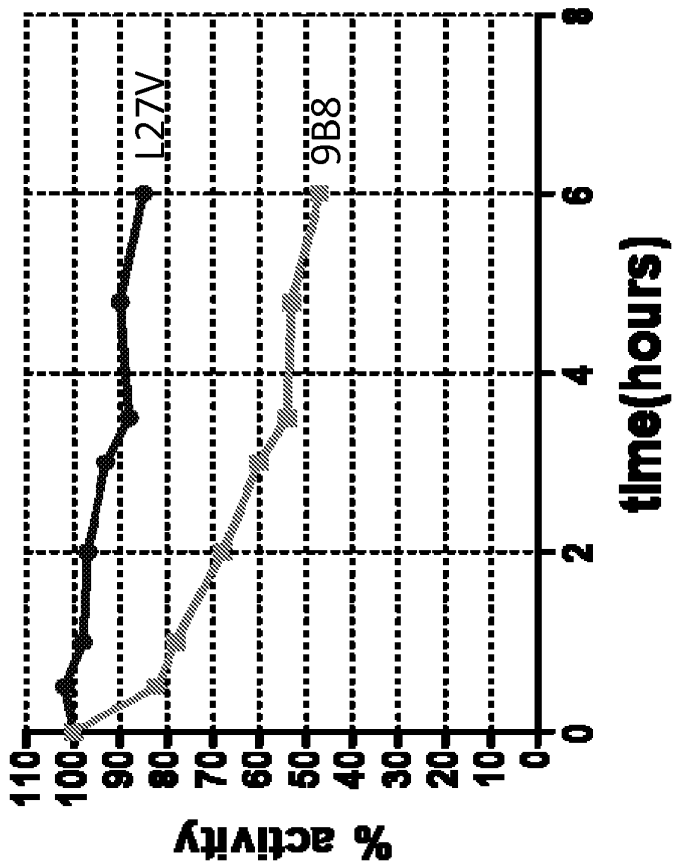


FIG. 41

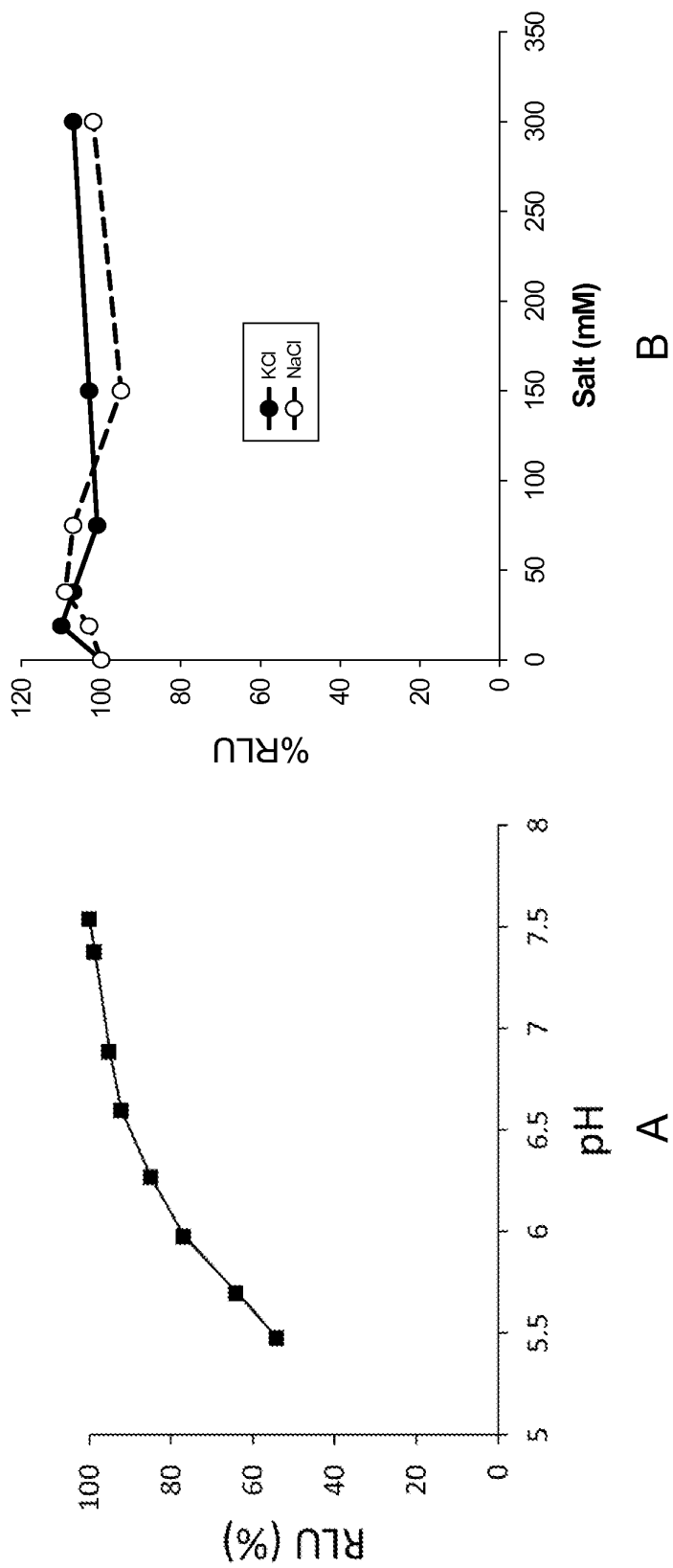


FIG. 42

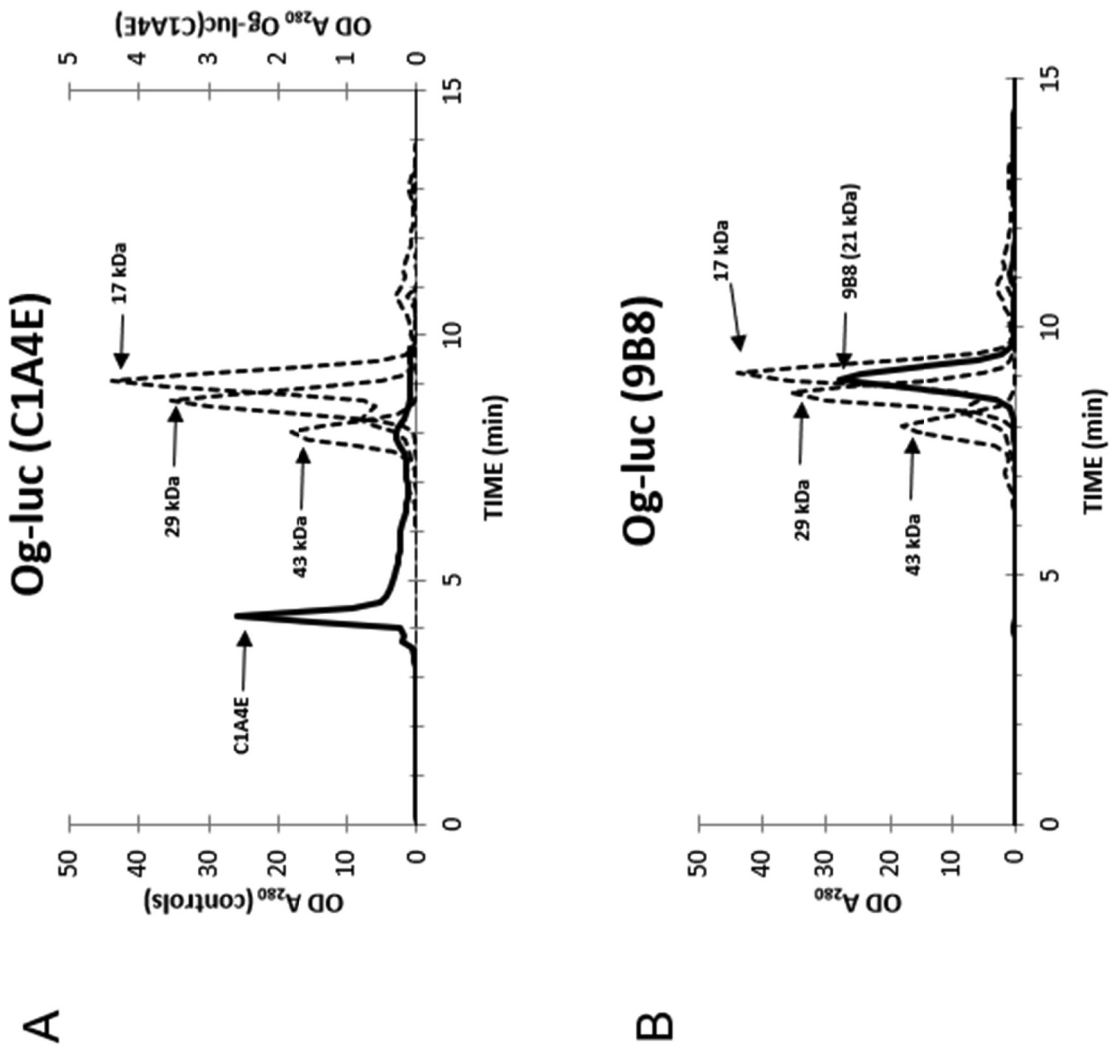


FIG. 43

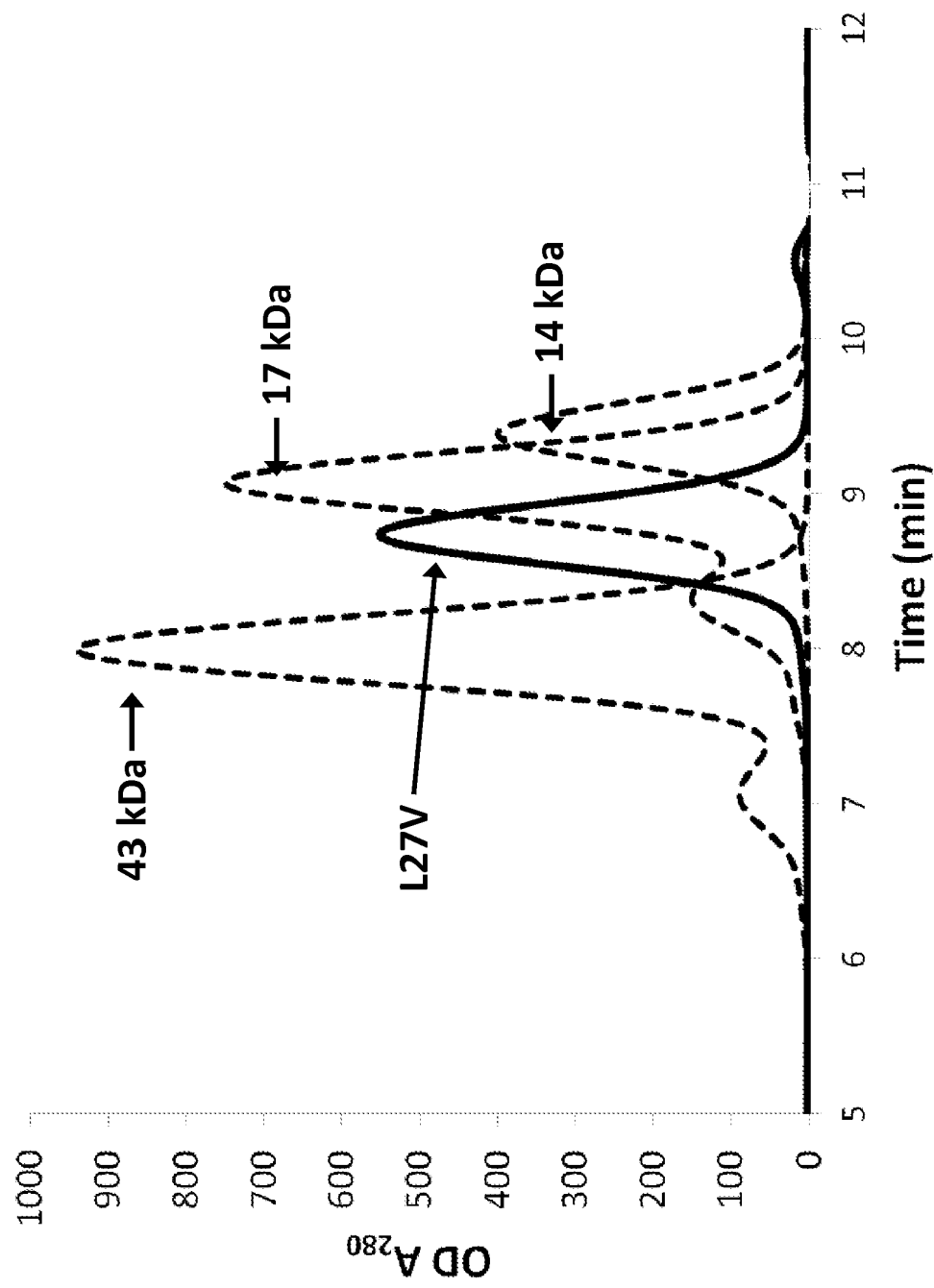


FIG. 44

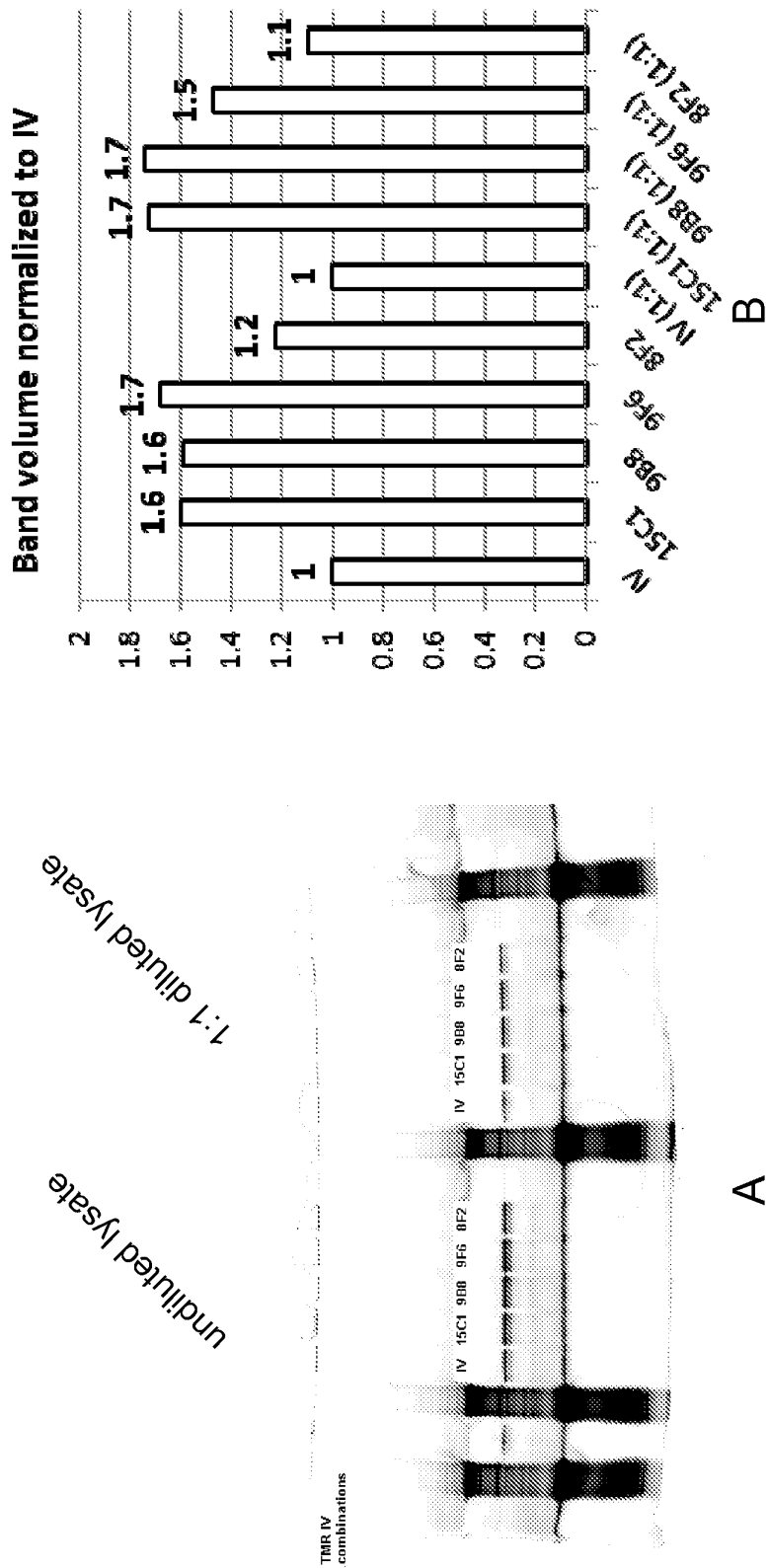


FIG. 45

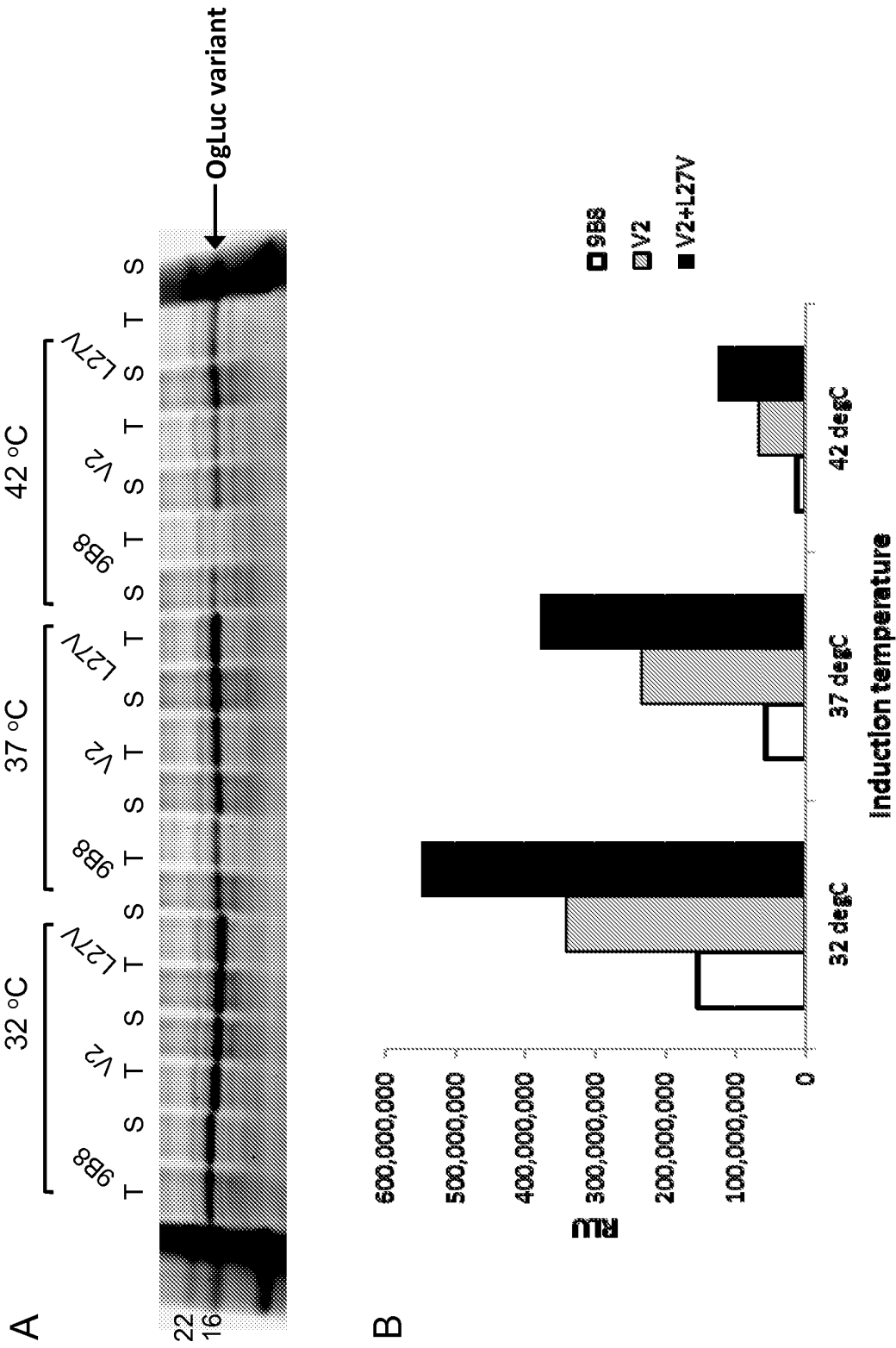


FIG. 46

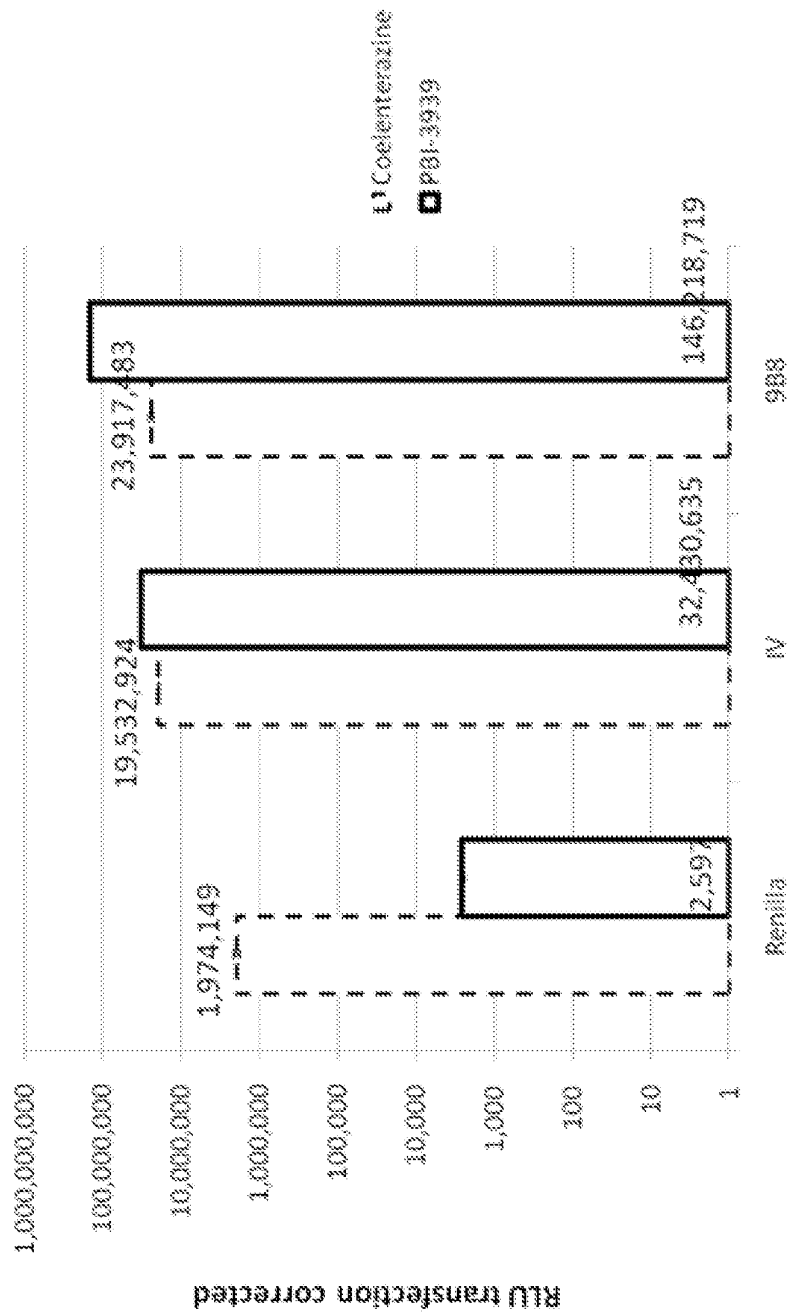


FIG. 47

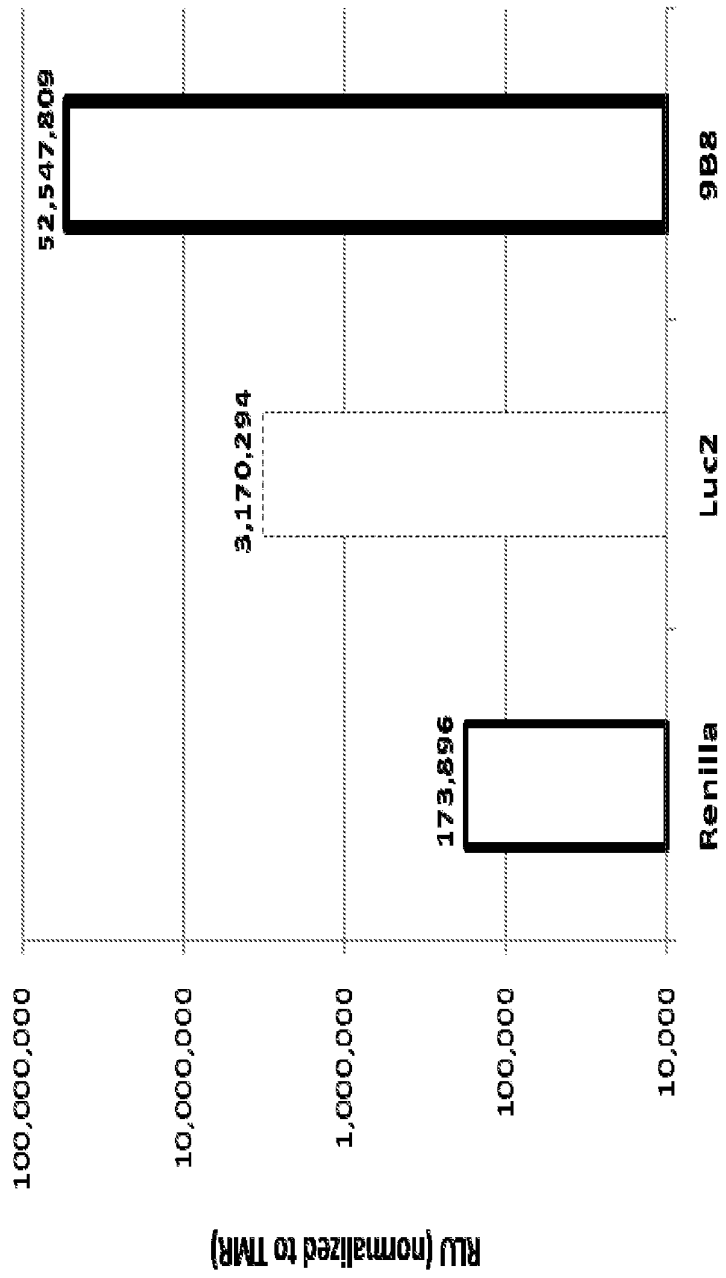


FIG. 48

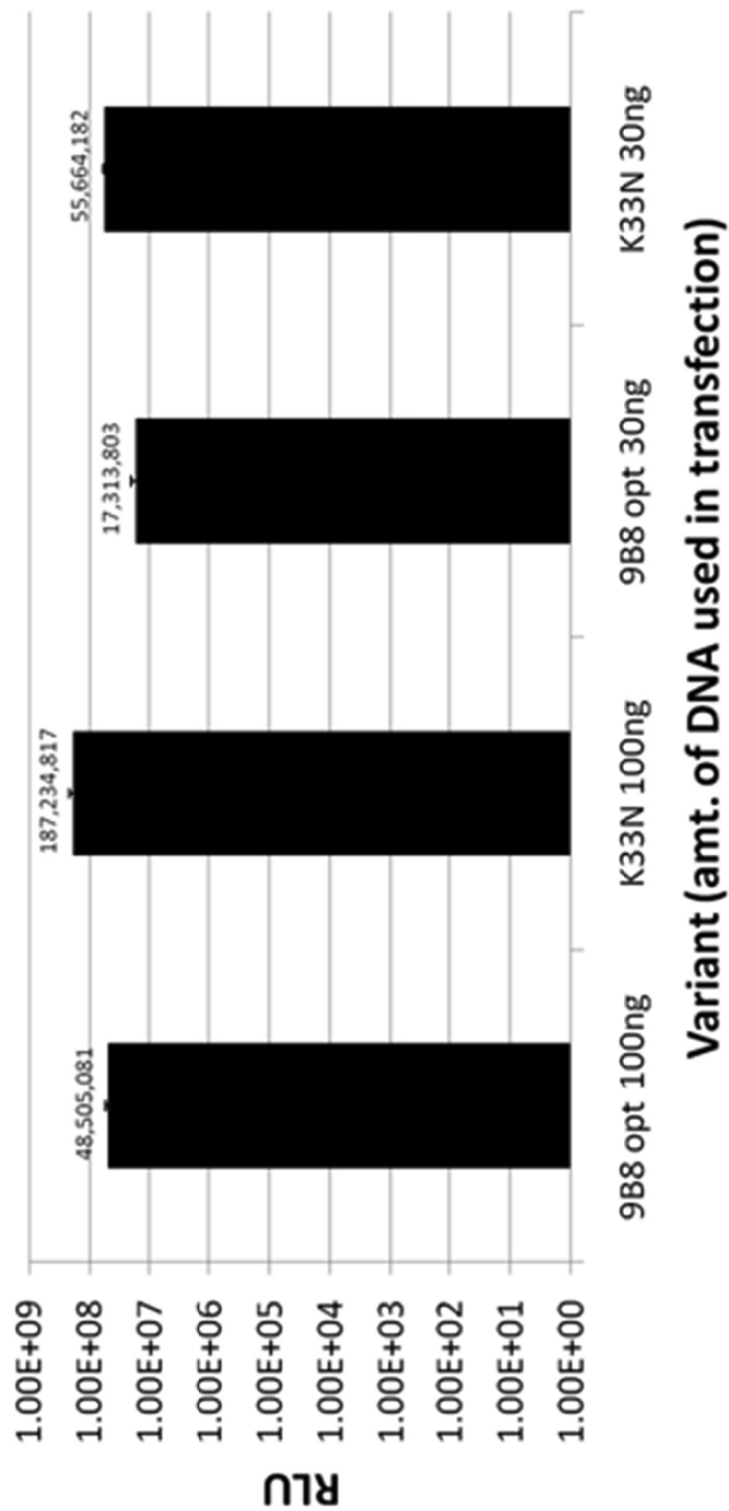


FIG. 49

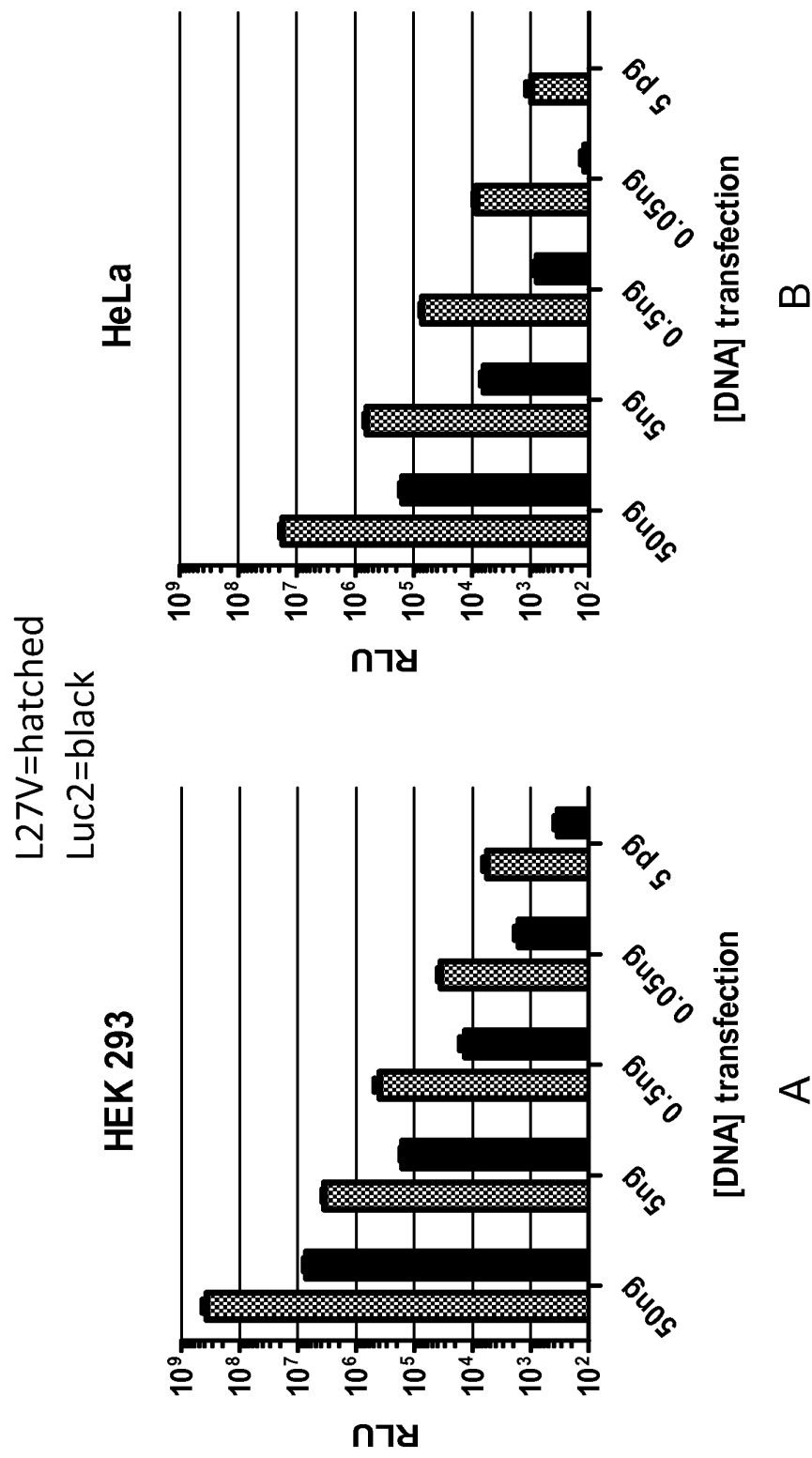


FIG. 50

L27V=hatched,  
Luc2=black

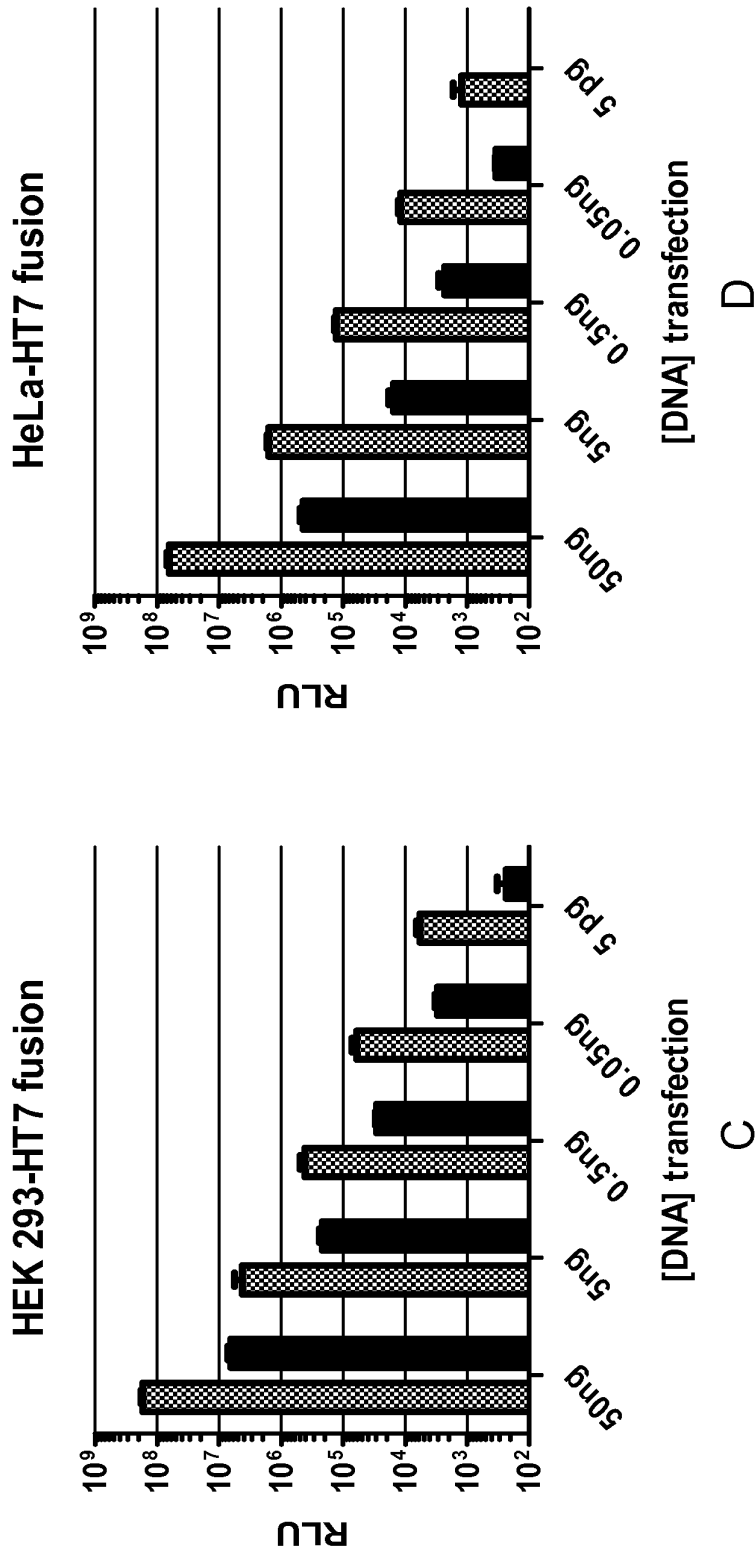
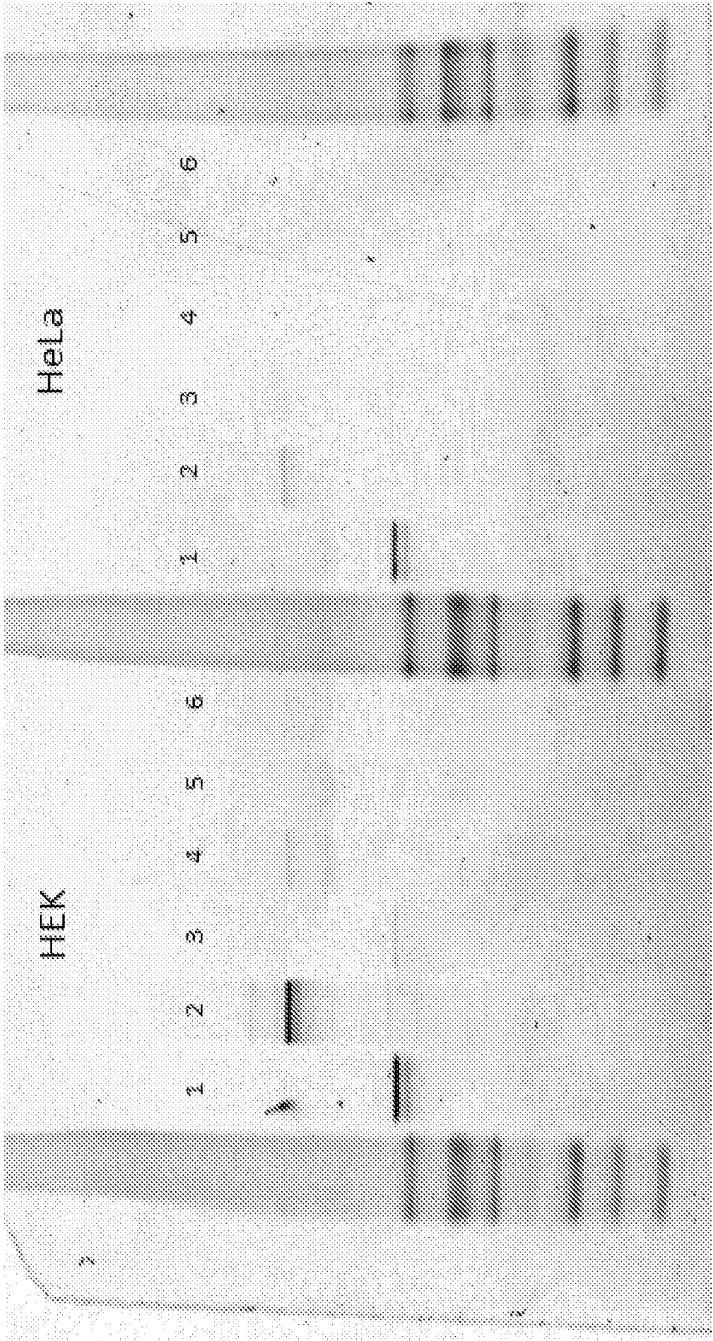


FIG. 50



- 1. L27V 50ng
- 2. Luc2 50ng
- 3. L27V 5 ng
- 4. Luc2 5 ng
- 5. L27V 0.5ng
- 6. Luc2 0.5ng

FIG. 50

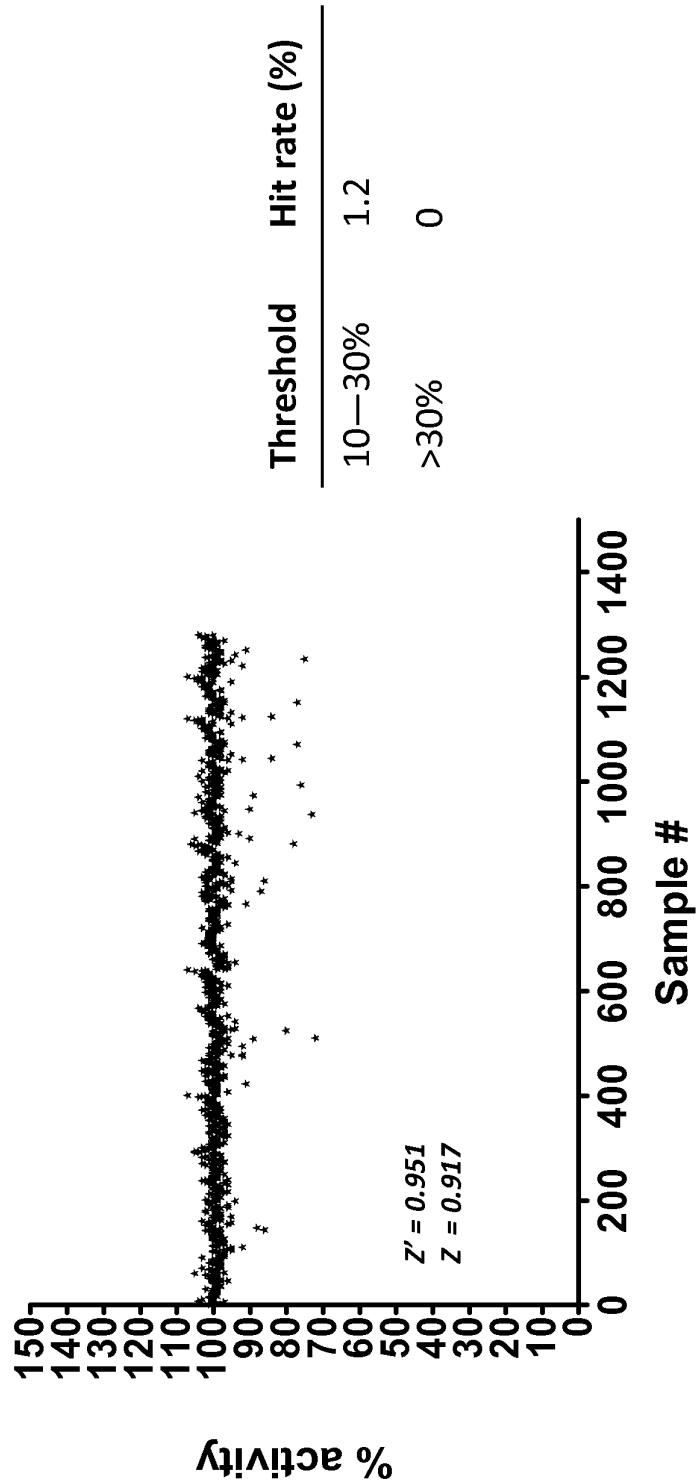
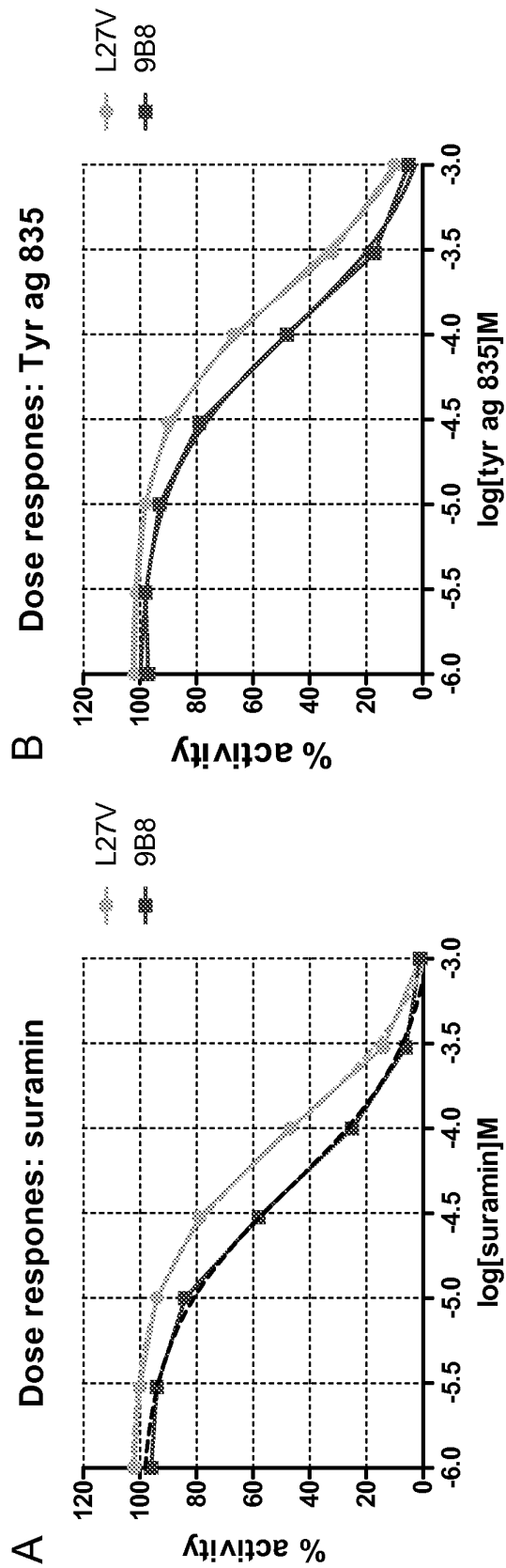
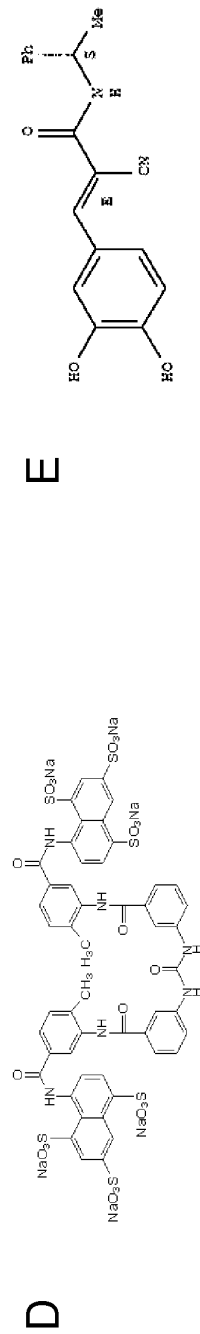


FIG. 51



**C**

| Sample     | 9B8 IC50 (uM) | L27V IC50(uM) |
|------------|---------------|---------------|
| Suramin    | 44.4          | 100.4         |
| Tyr ag 835 | 105           | 198           |



Tyr ag 835

Suramin

FIG. 52

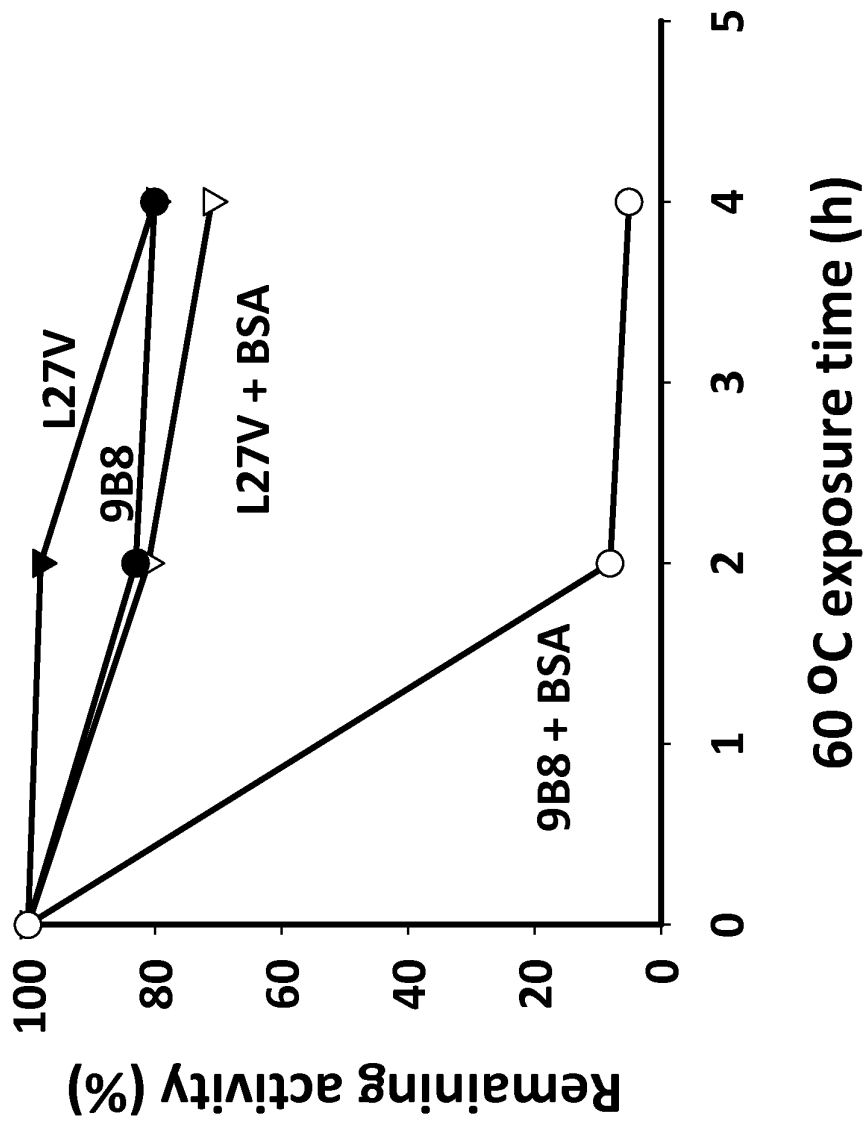


FIG. 53

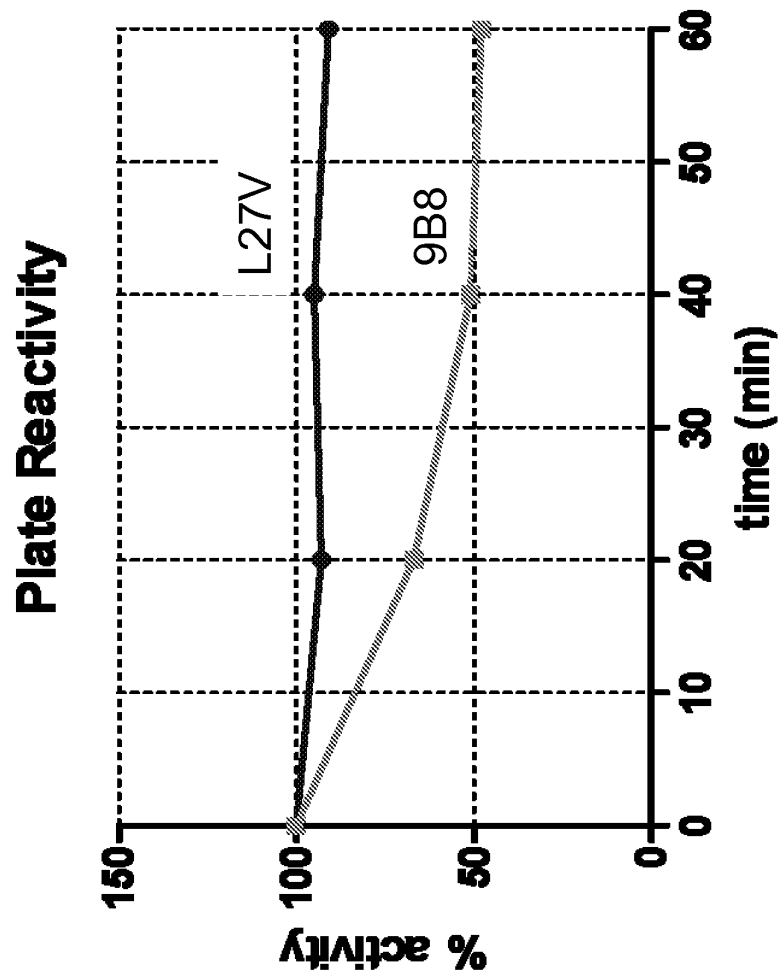


FIG. 54

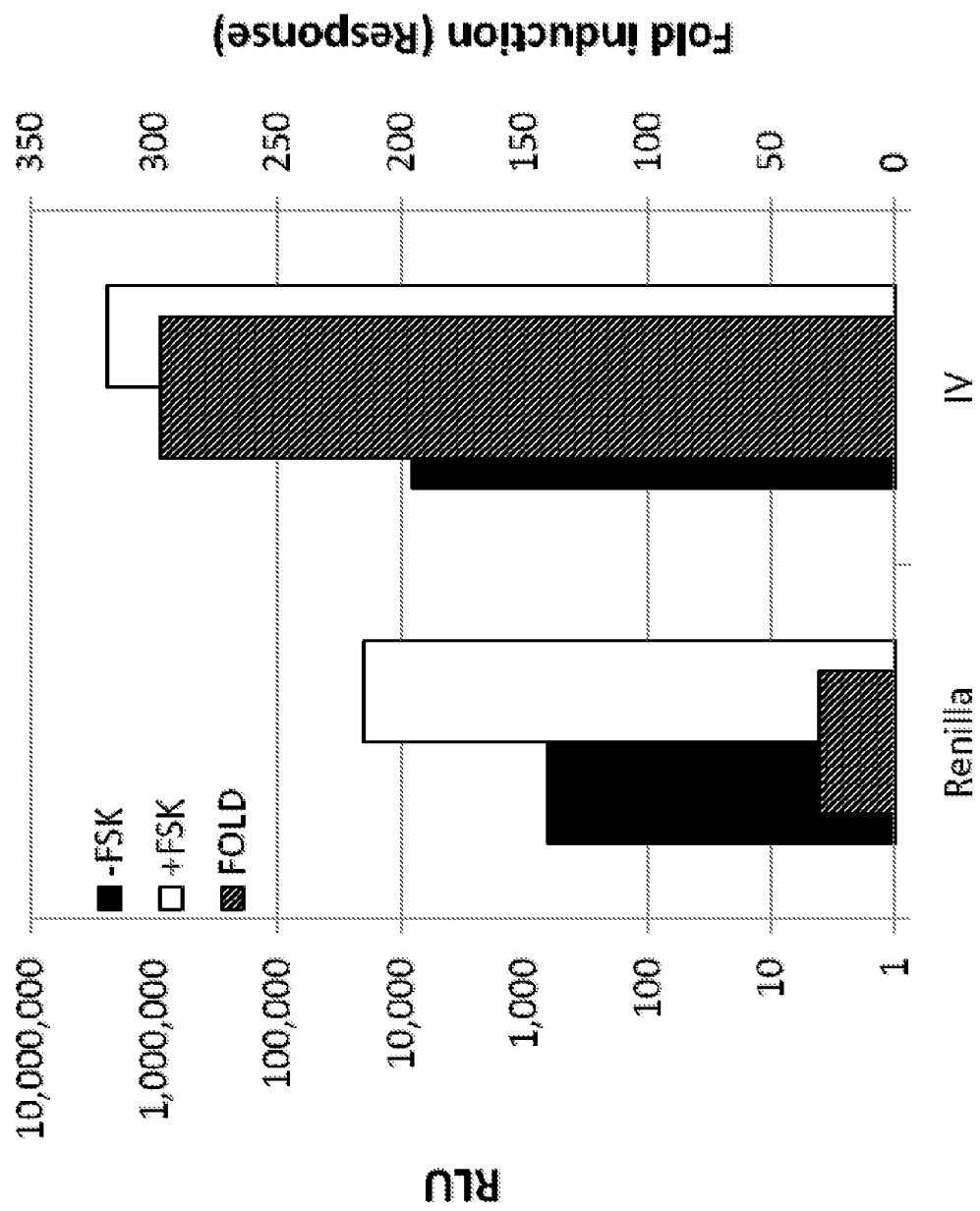


FIG. 55

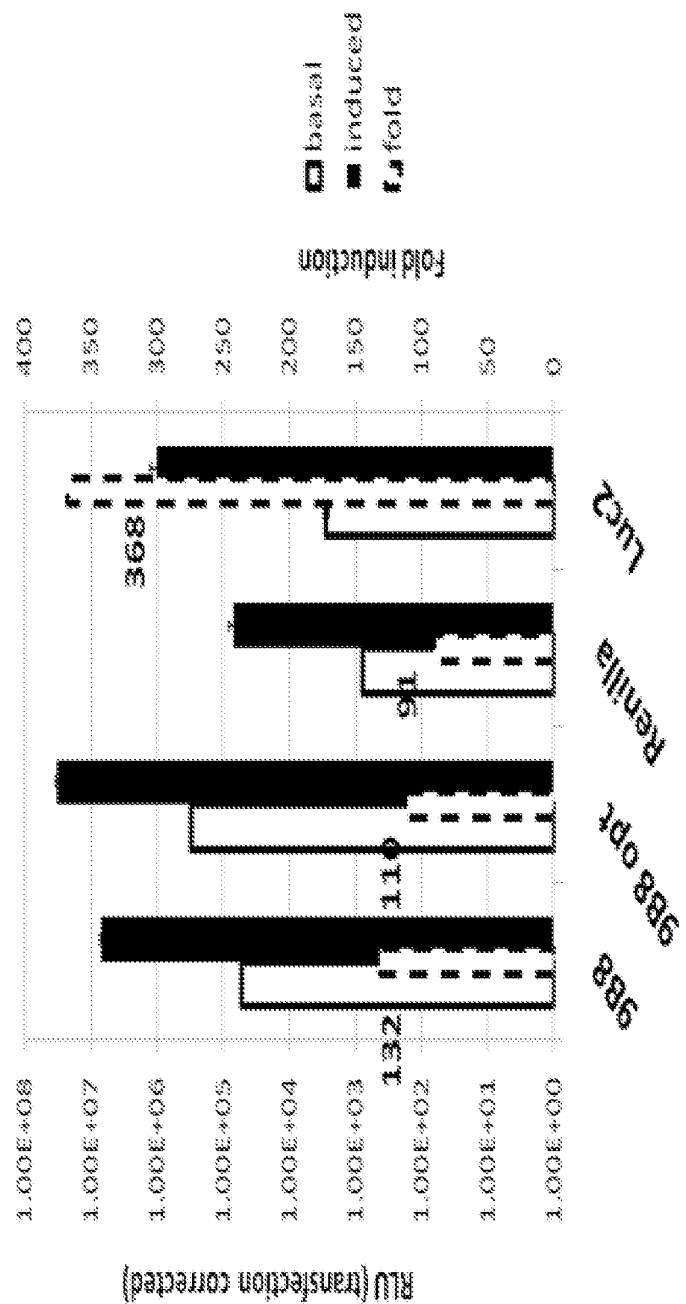


FIG. 56

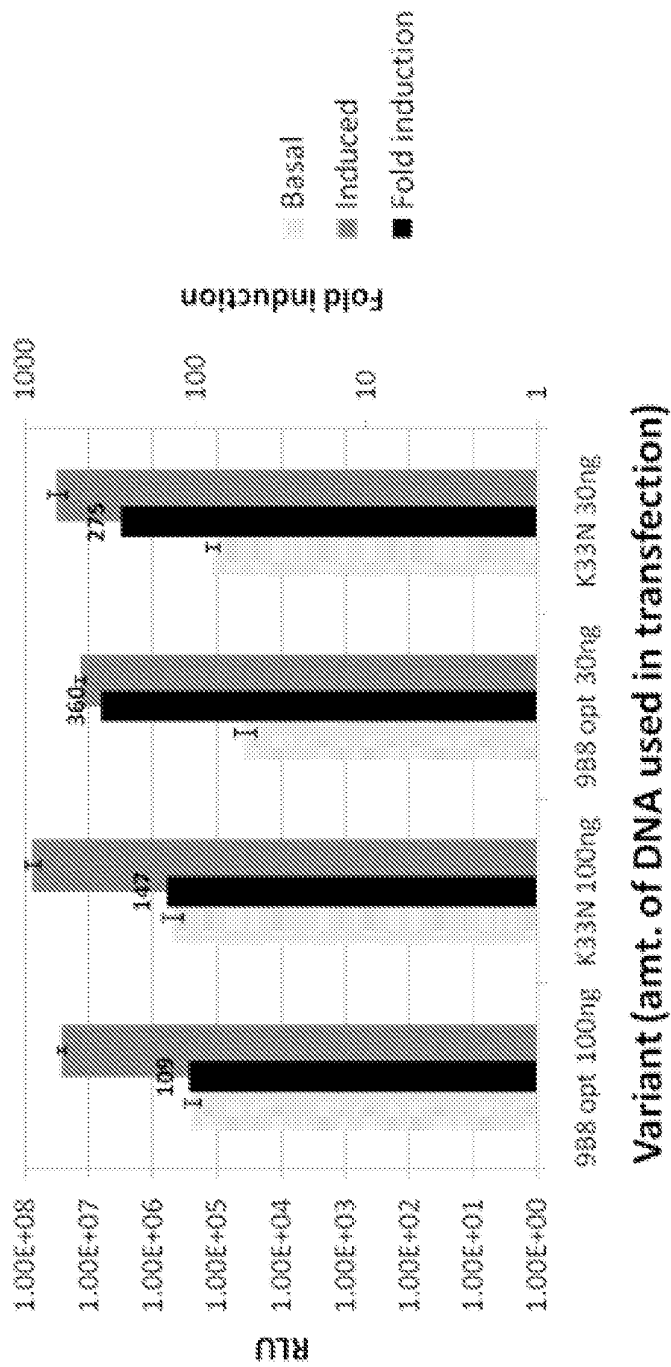


FIG. 57

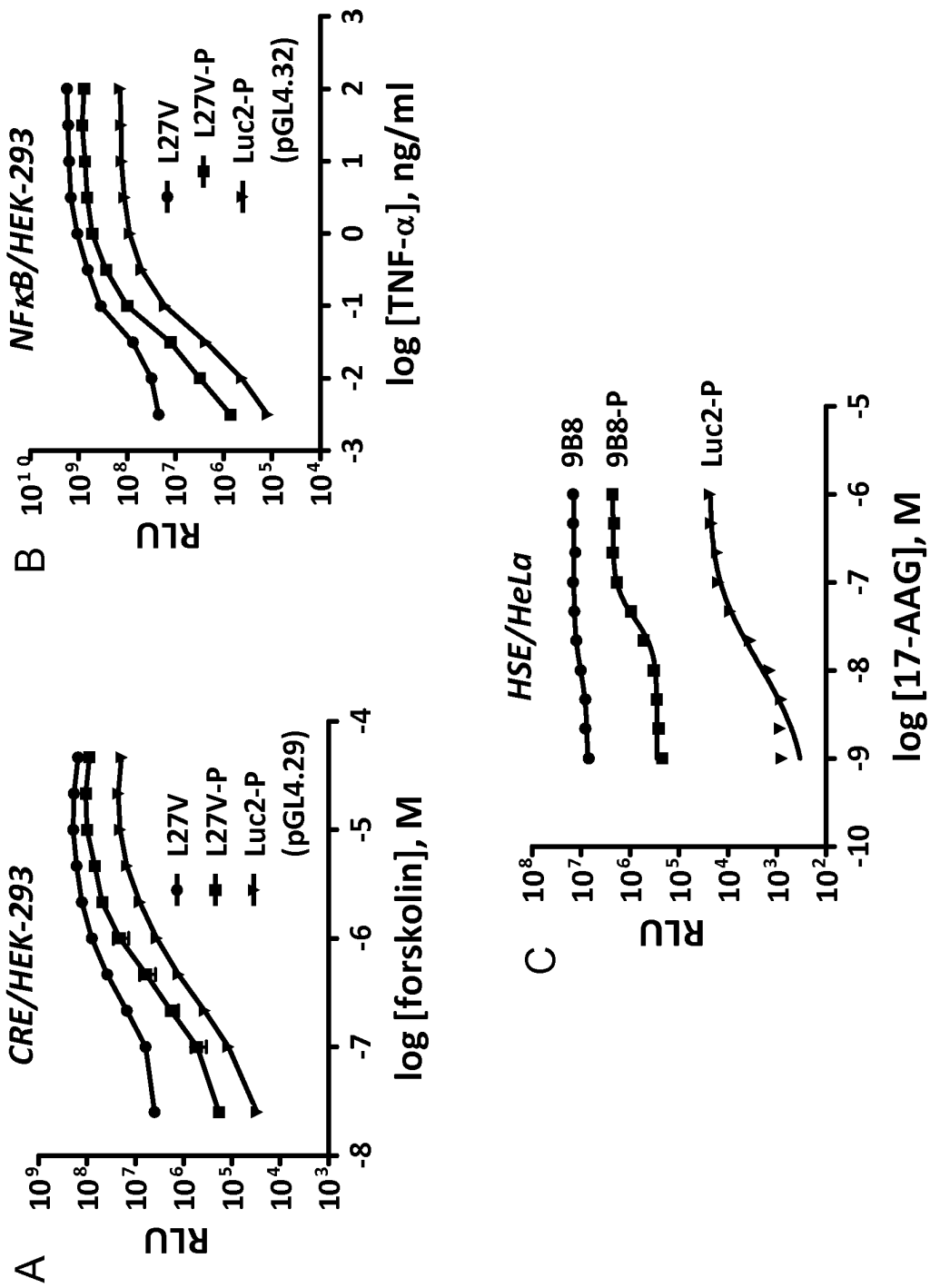
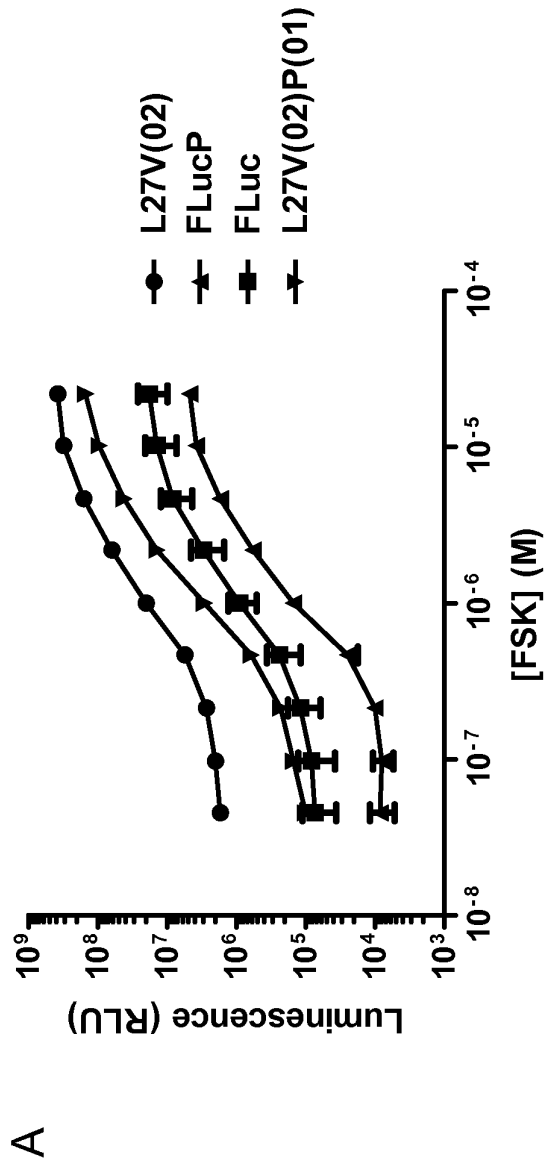


FIG. 58



**B**

|                  | FLucP | FLuc | L27V02P | L27V02 |
|------------------|-------|------|---------|--------|
| Avg. RLU / luc2P | 1.00  | 7.5  | 21.6    | 168.5  |
| S/B              | 781   | 268  | 1768    | 224    |
| EC50 (μM)        | 6.3   | 5.9  | 10.3    | 5.9    |

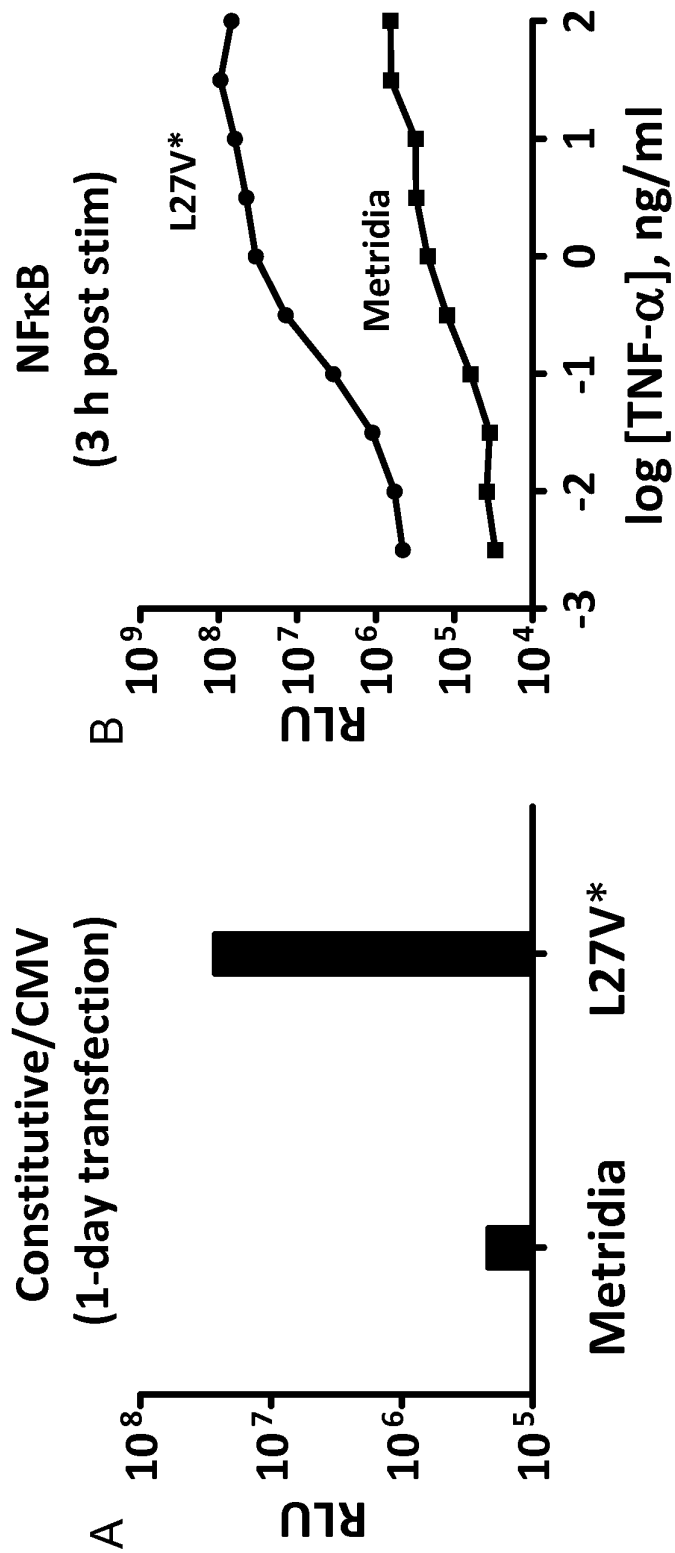
FIG. 59

C

| Exp. system | RLUs<br>OLuc/FLuc | RLUs<br>OLucP/FLucP | S/B,<br>OLuc | S/B,<br>OLucP | S/B,<br>FLuc | S/B,<br>FLucP |
|-------------|-------------------|---------------------|--------------|---------------|--------------|---------------|
| 293/CRE     | 22                | 22                  | 224          | 1768          | 268          | 781           |
| 293/NFkB    | 75                | 8                   | 22           | 514           | 233          | 347           |
| Hela/NFkB   | 14                | 2                   | 40           | 47            | 54           | 44            |
| HepG2/NFkB  | 267               | 5                   | 7            | 11            | 11           | 19            |
| Jurkat/NFkB | 16                | 6                   | 18           | 15            | 6            | 11            |
| Jurkat/CRE  | 19                | 4                   | 14           | 123           | 28           | 40            |
| ME180/CRE   | 56                | 12                  | 2            | 6             | 4            | 5             |
| ME180/NFkB  | 149               | 21                  | 7            | 19            | 7            | 25            |
| HCT116/NFkB | 22                | 16                  | 10           | 605           | 4            | 47            |
| U2OS/NFkB   | 114               | 16                  | 15           | 18            | 11           | 20            |

FIG. 59

HEK-293



\*IL-6 signal sequence

FIG. 60

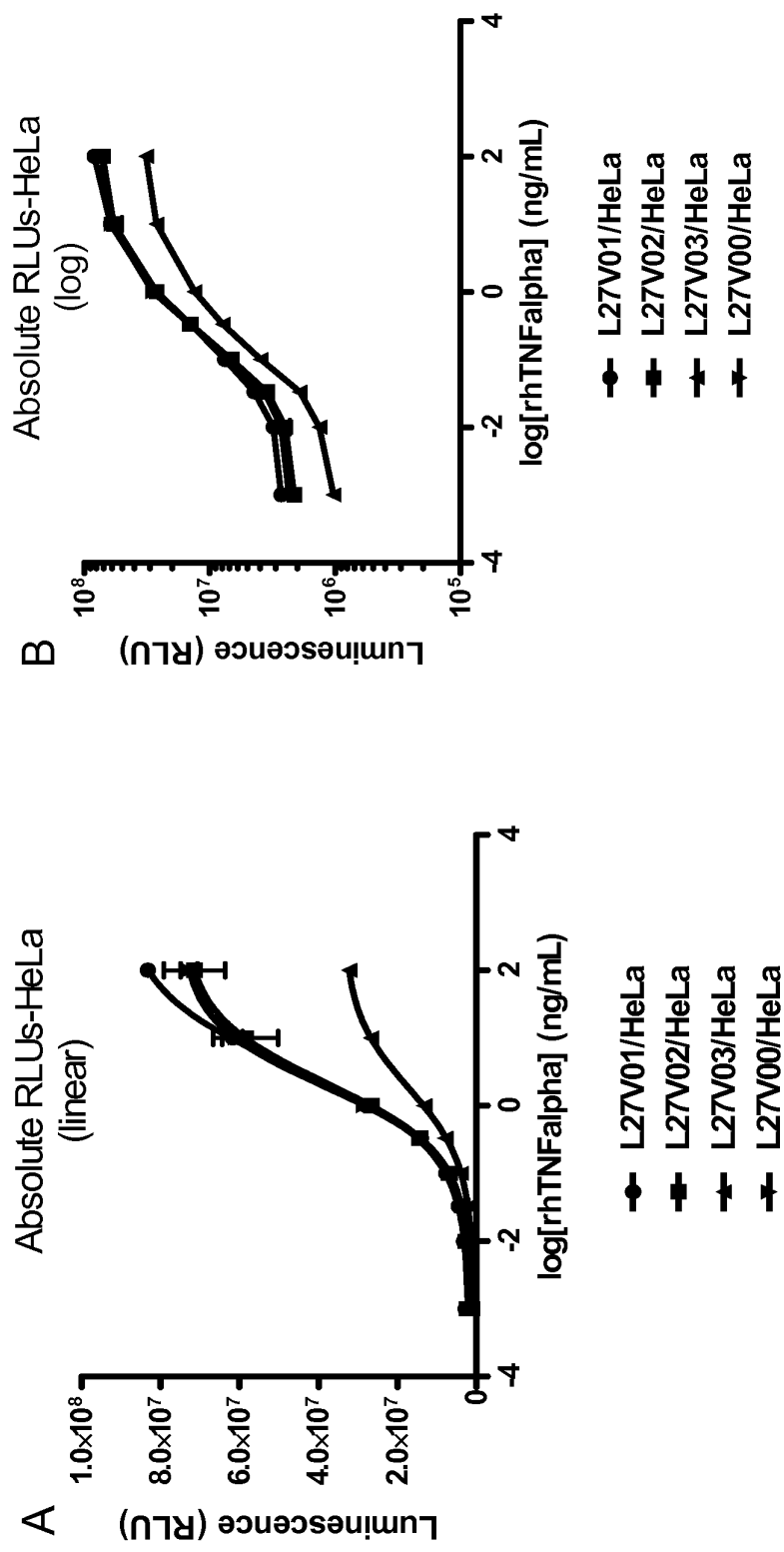


FIG. 61

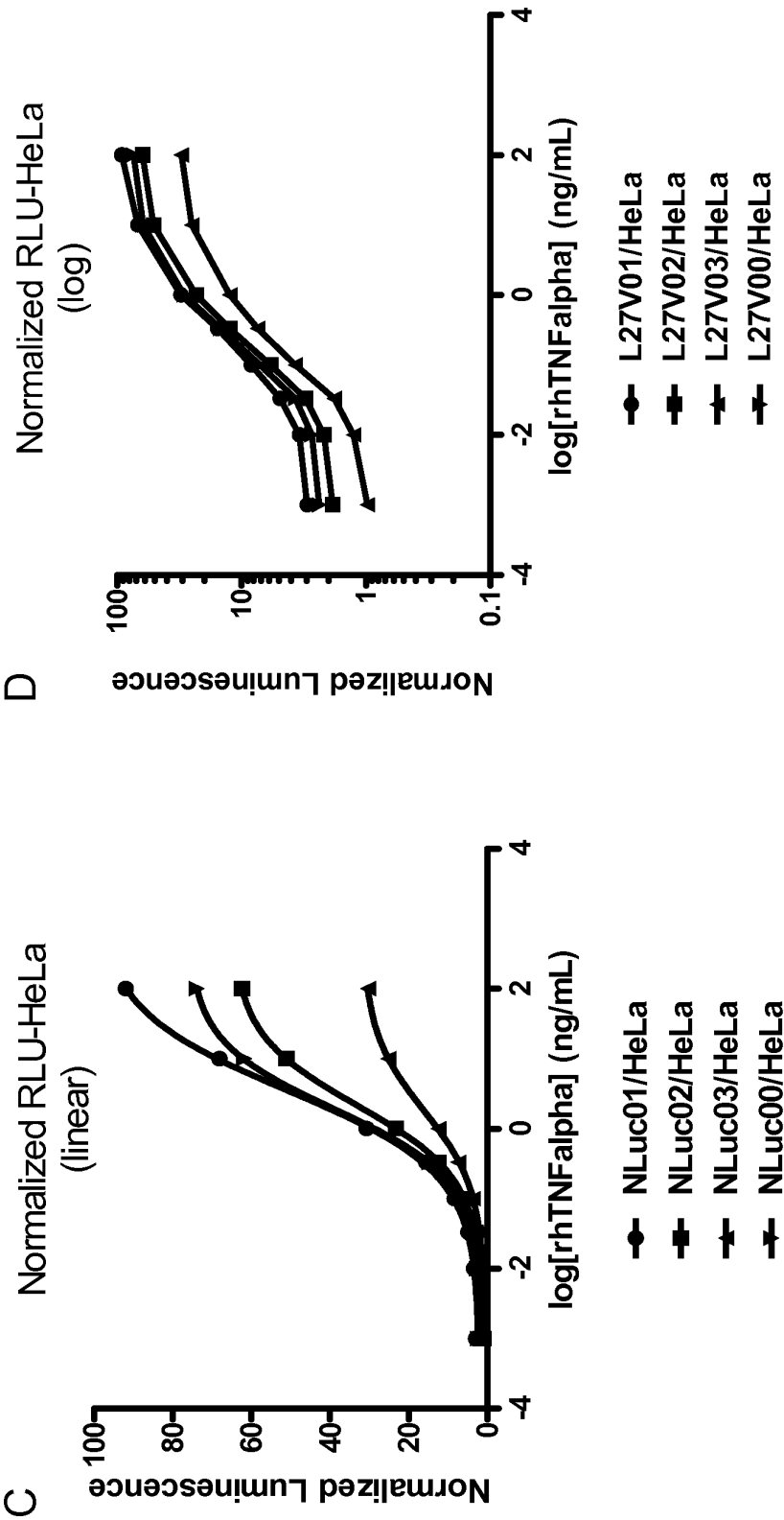


FIG. 61

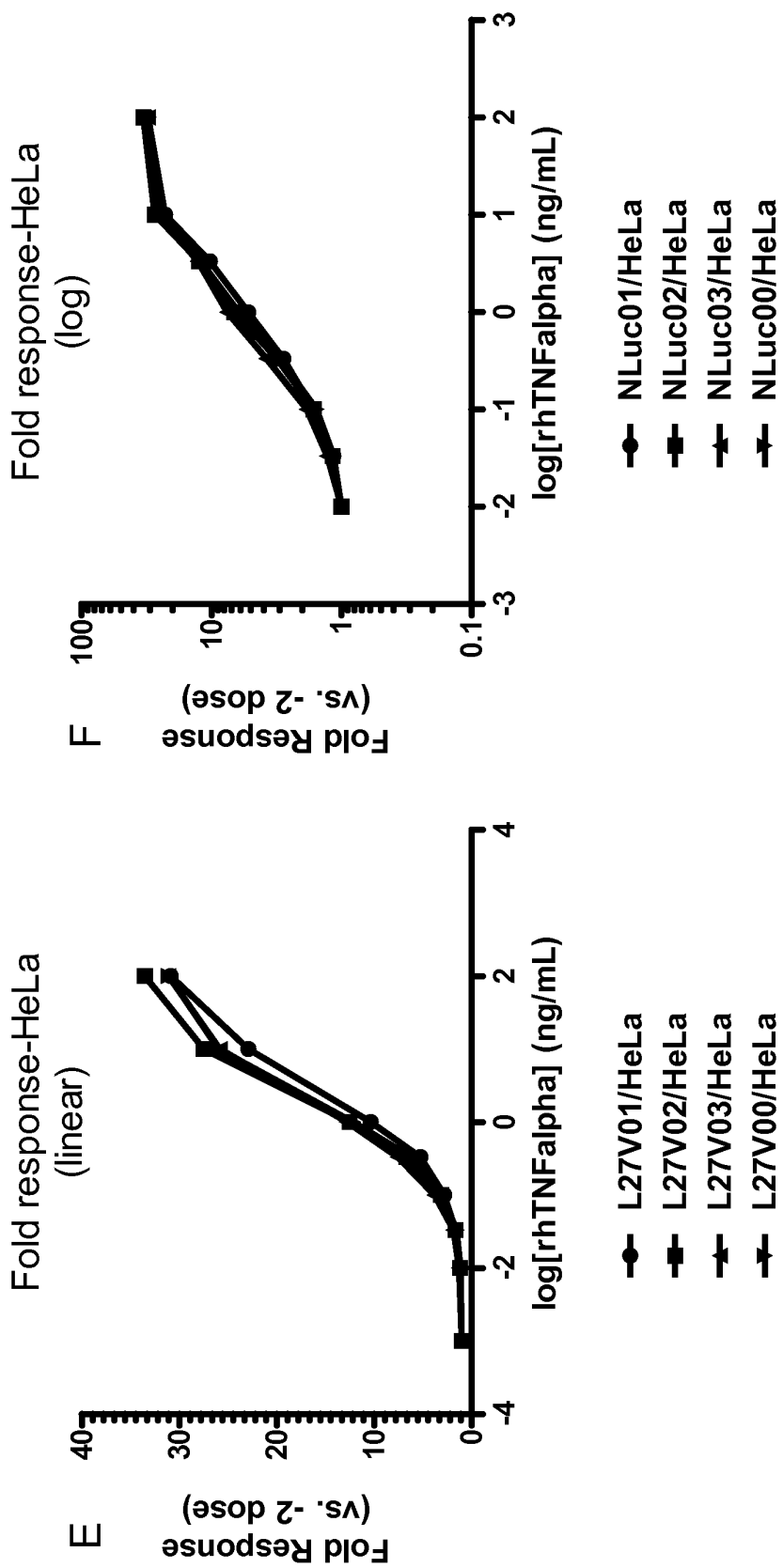


FIG. 61

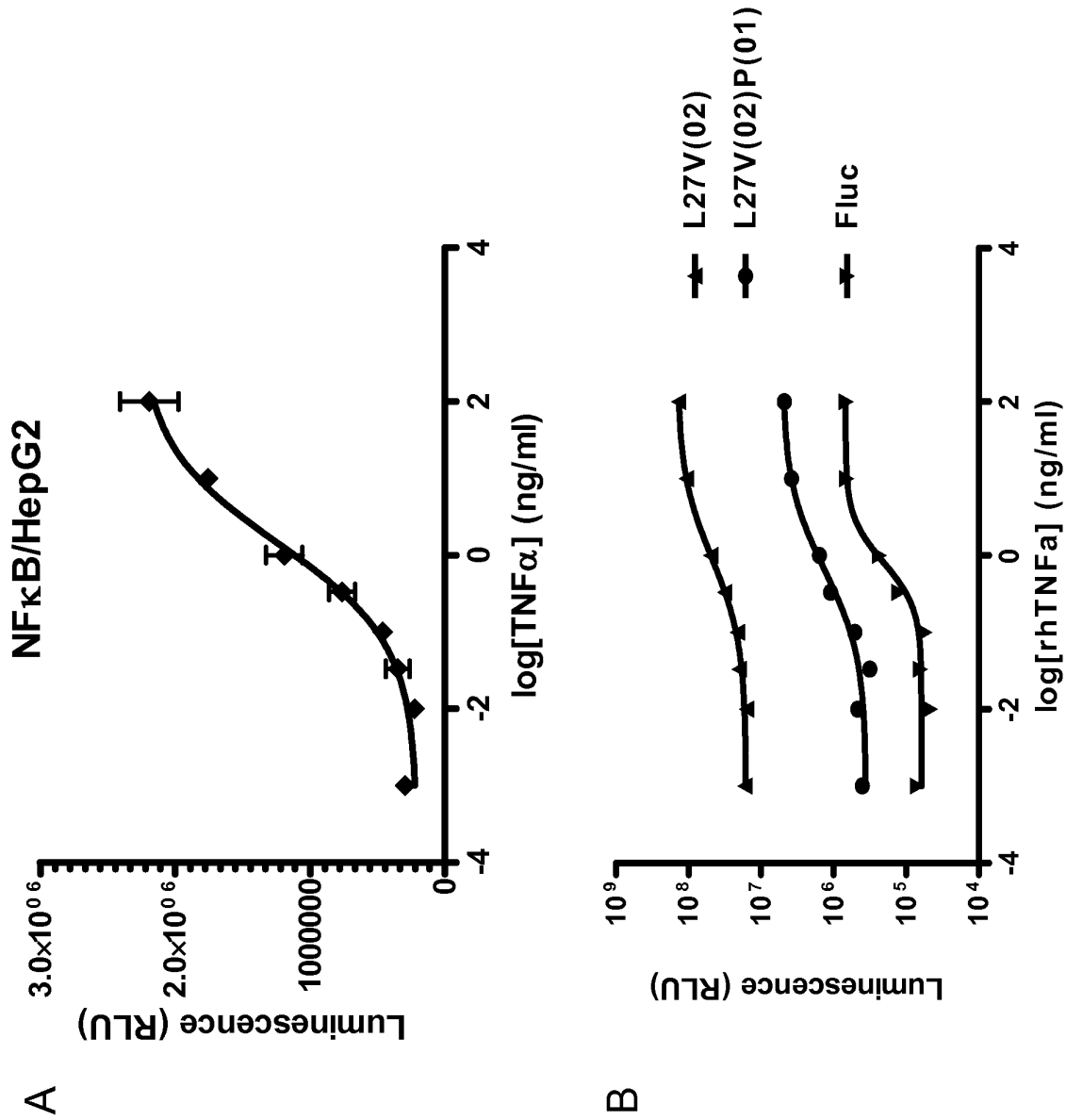


FIG. 62

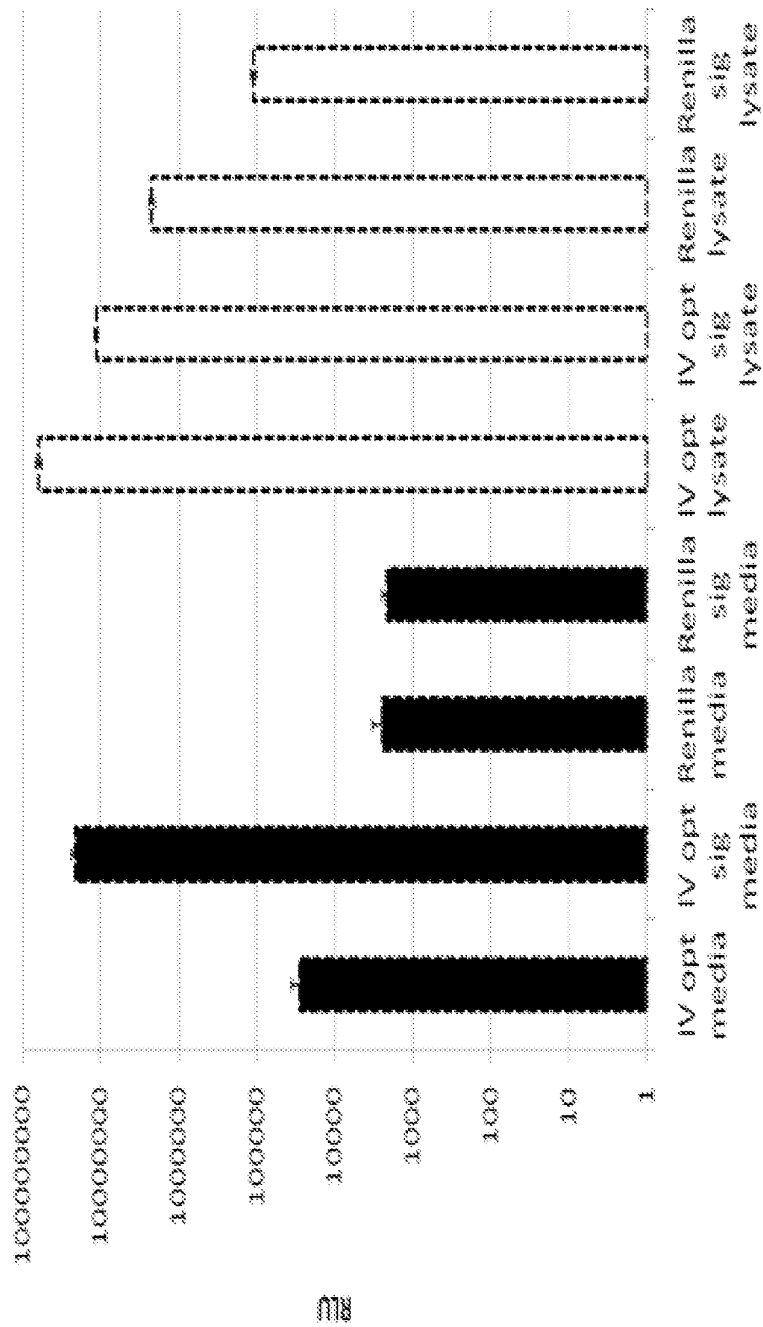


FIG. 63

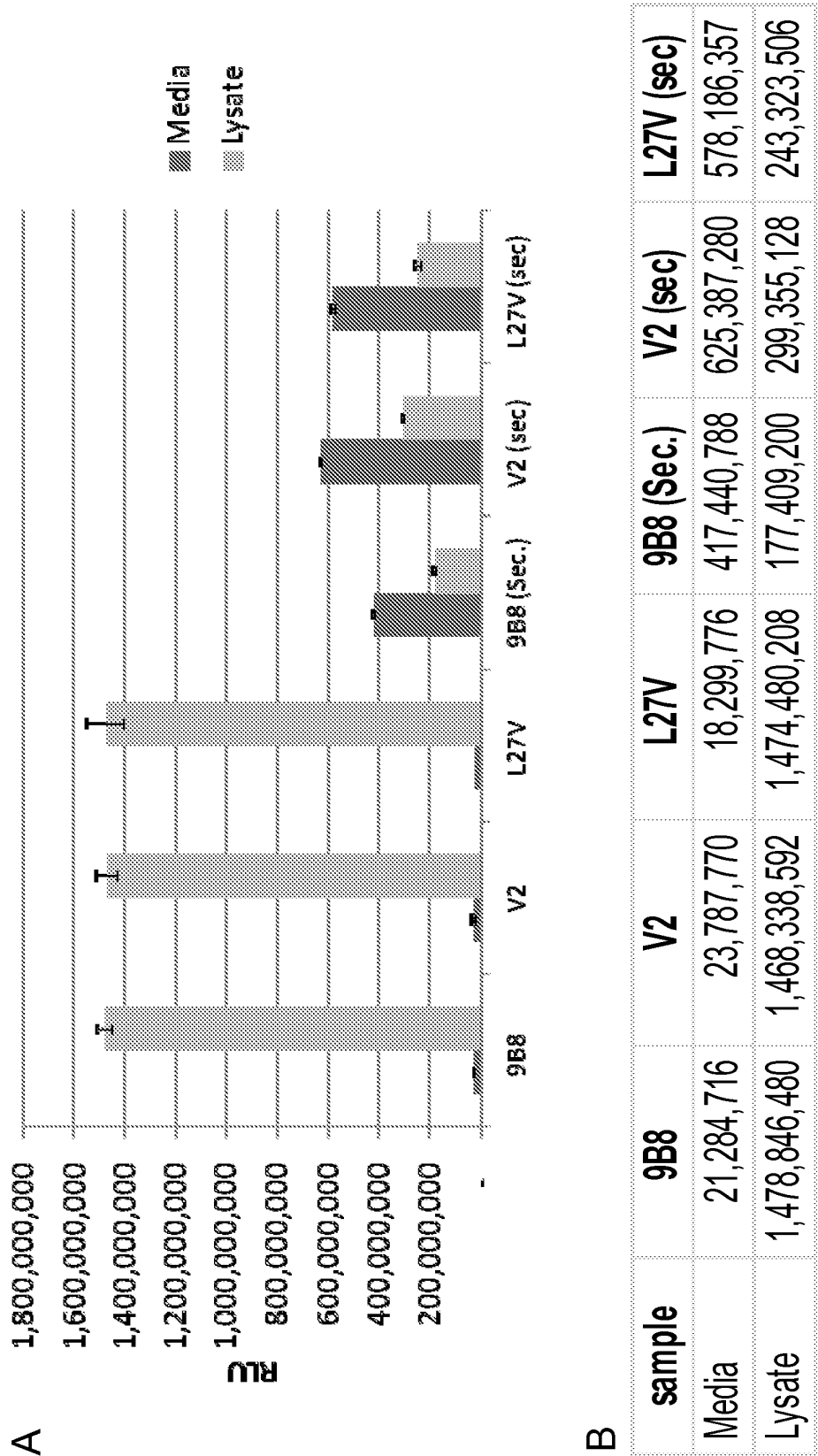


FIG. 64

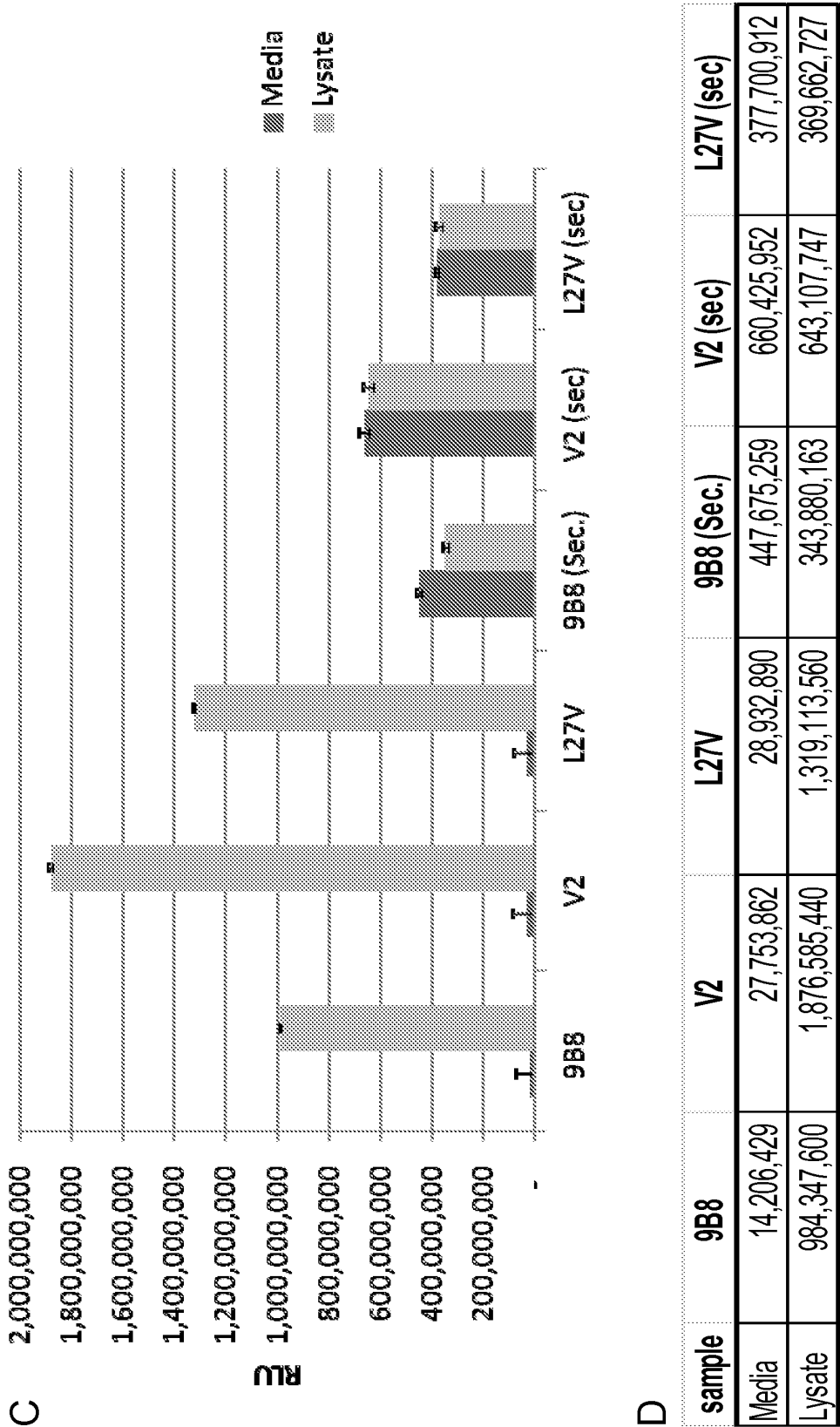


FIG. 64

| A     |  | Met-luc (Ready-To Glow™) | 9B8-3939      | V2-3939       |
|-------|--|--------------------------|---------------|---------------|
| DNA   |  |                          |               |               |
| 100ng |  | 2,502,116                | 3,307,464,875 | 3,992,245,760 |
| 10ng  |  | 297,024                  | 33,947,989    | 51,683,999    |

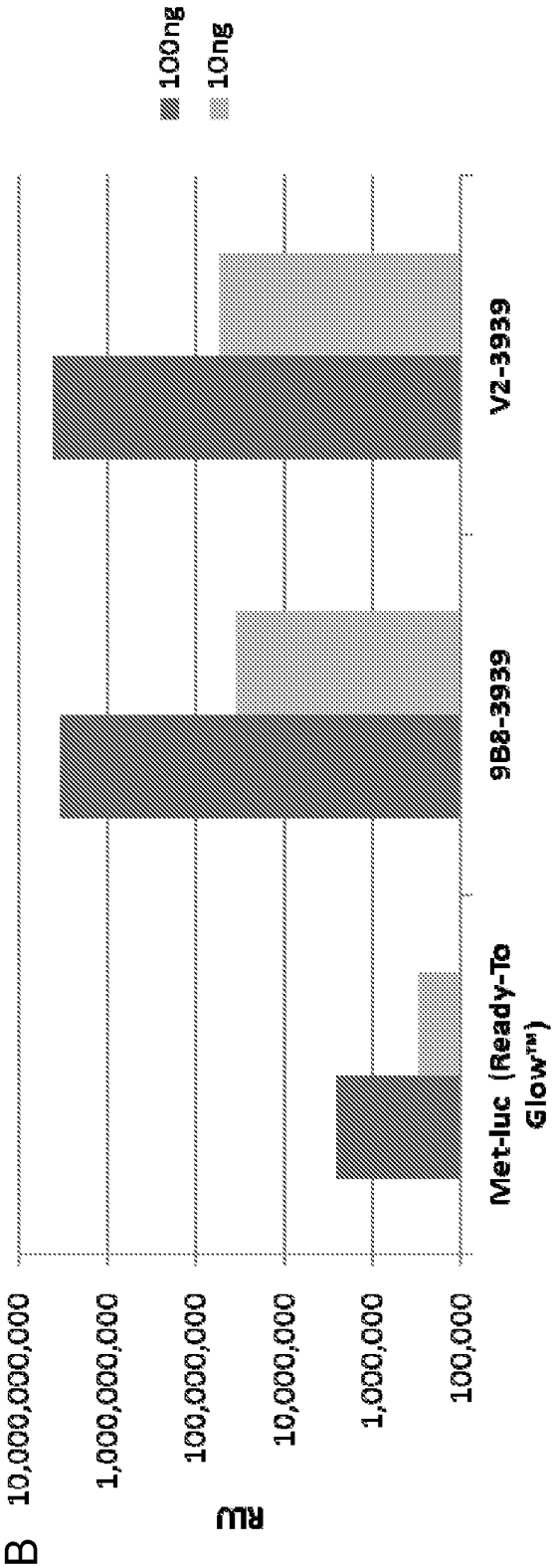
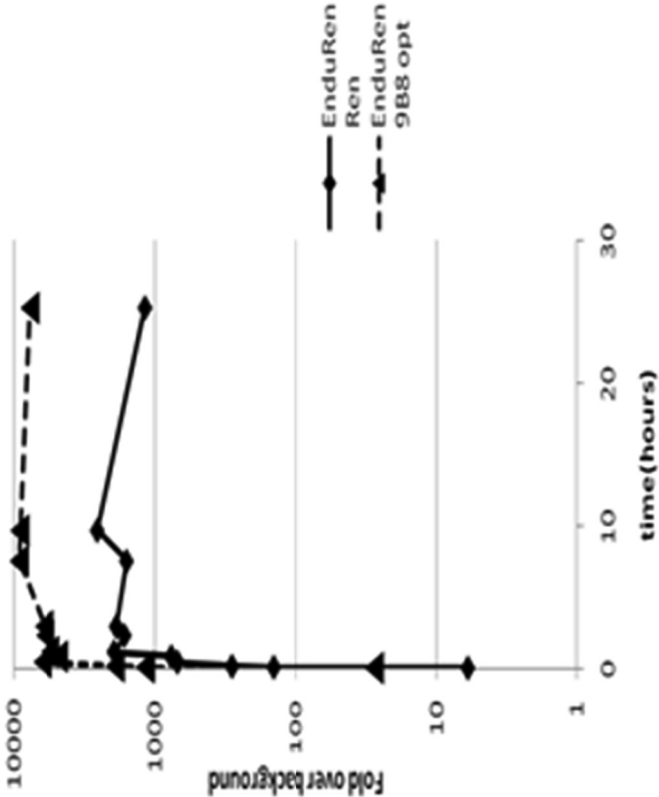


FIG. 65

A



B

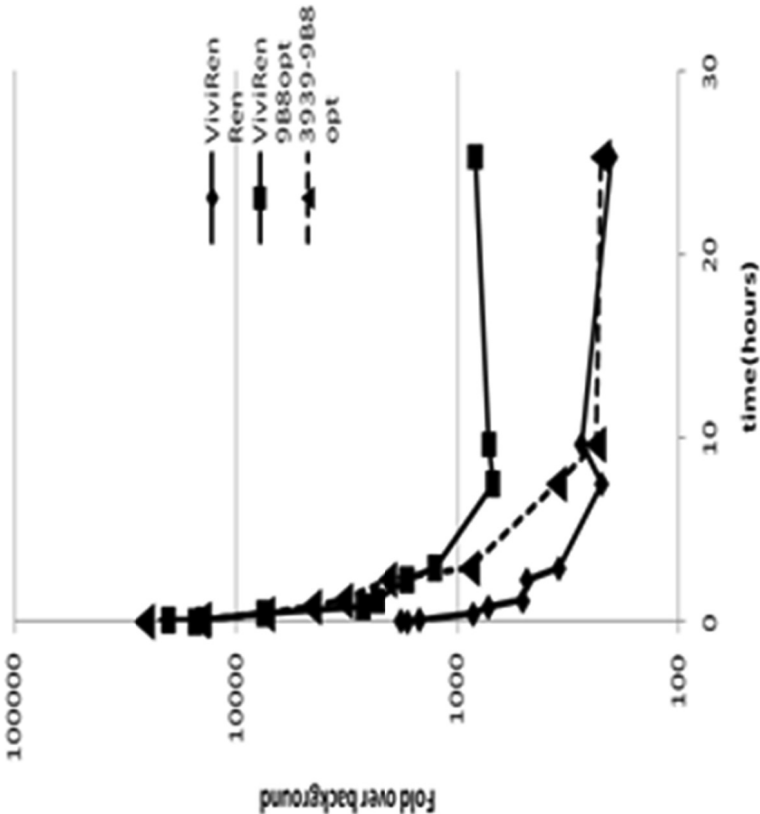
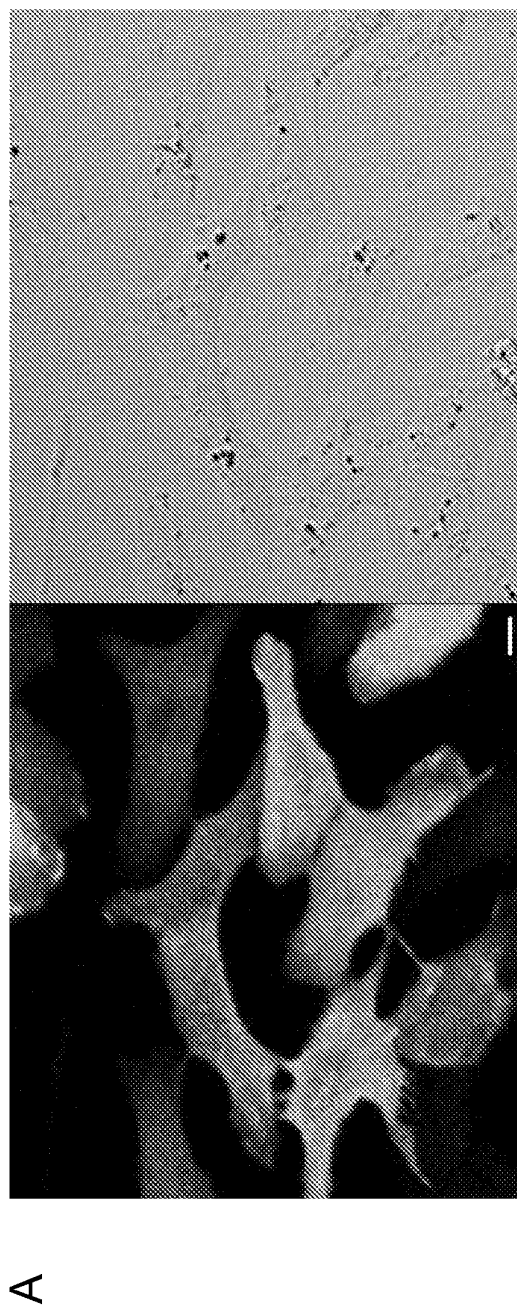
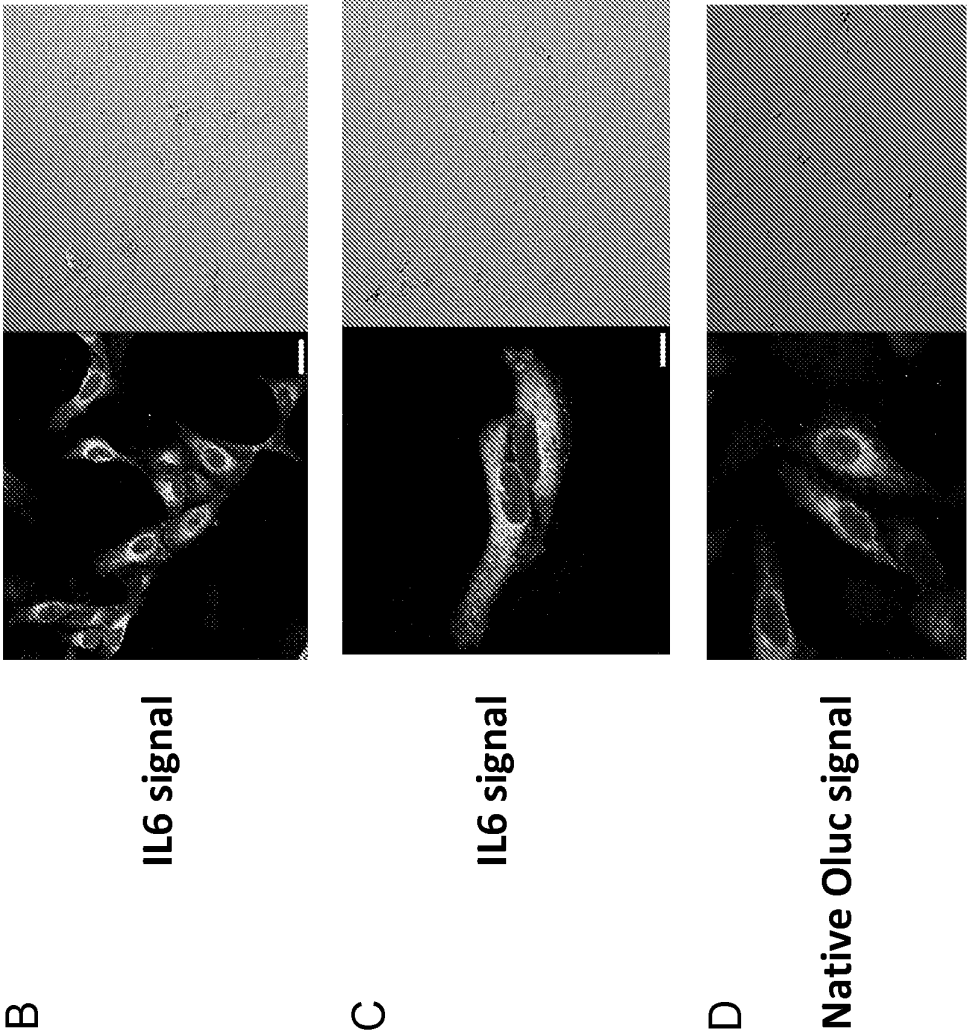


FIG. 66





**FIG. 67**

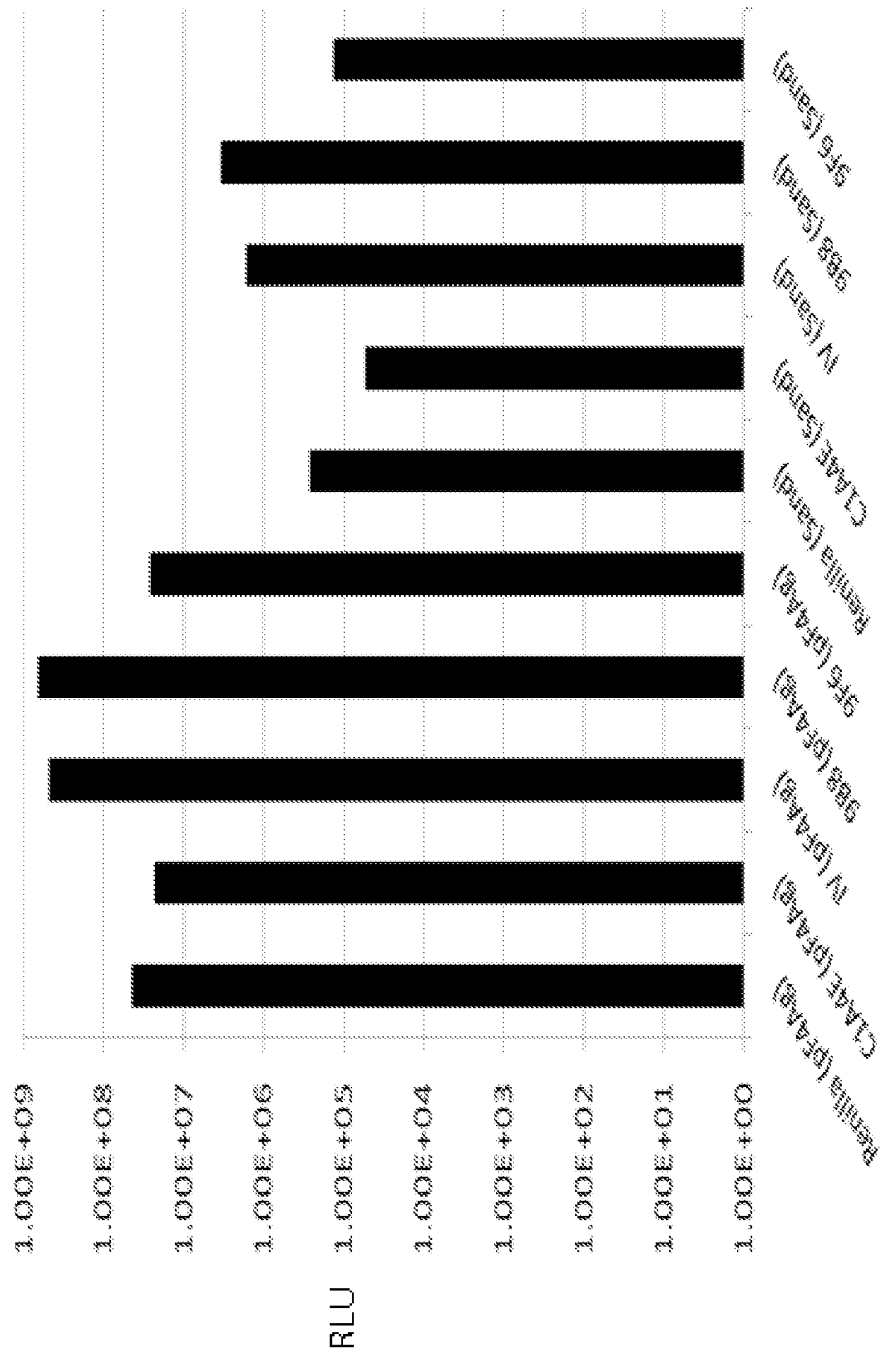


FIG. 68

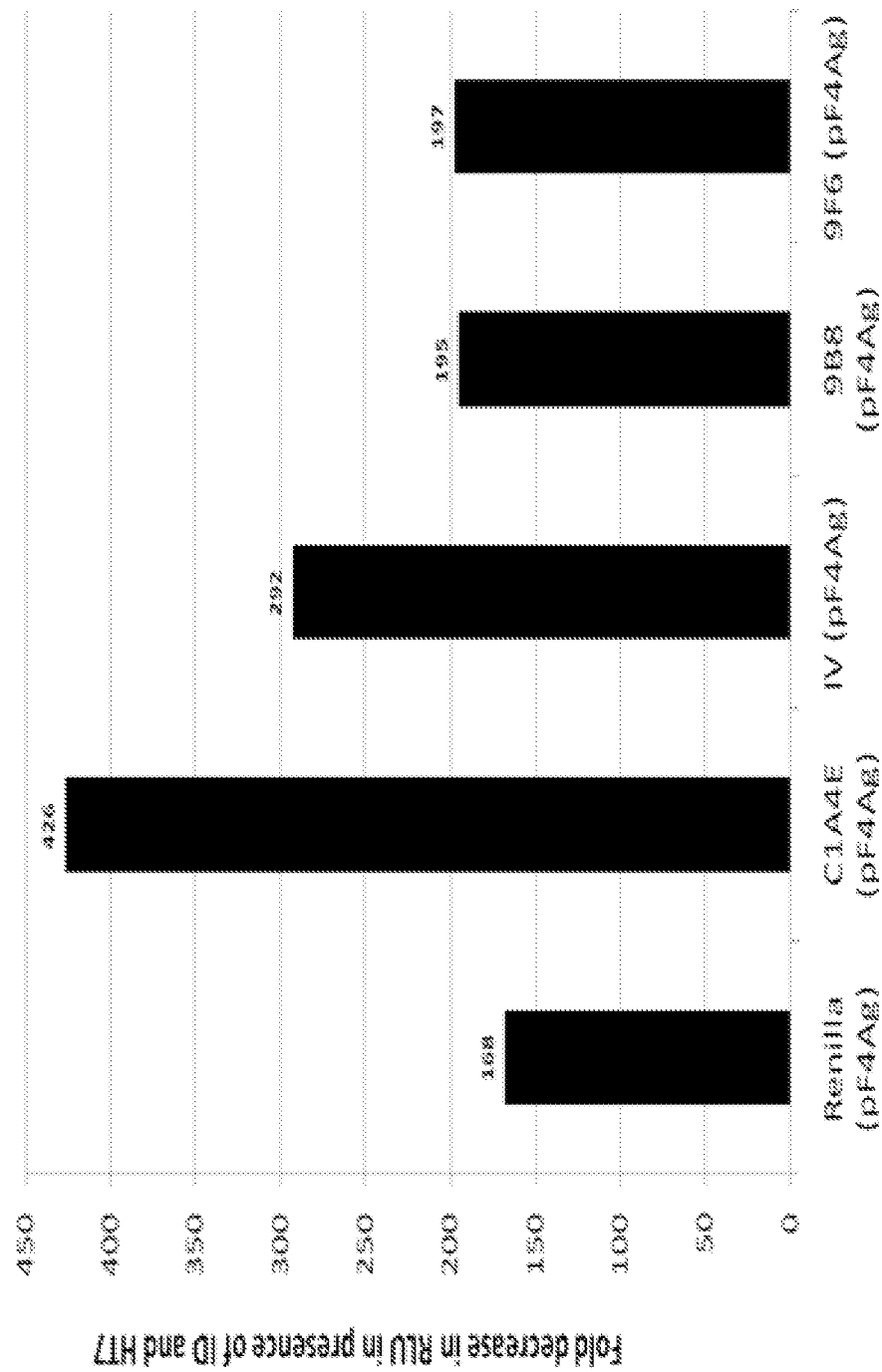


FIG. 69

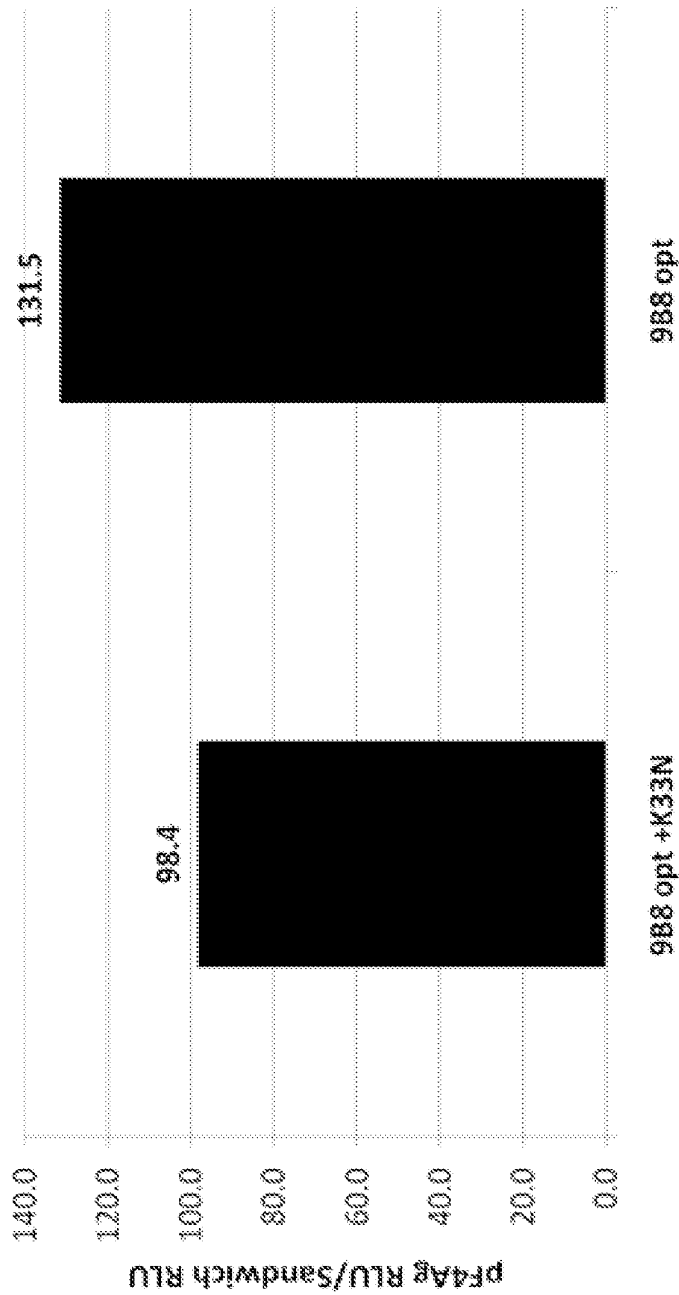


FIG. 70

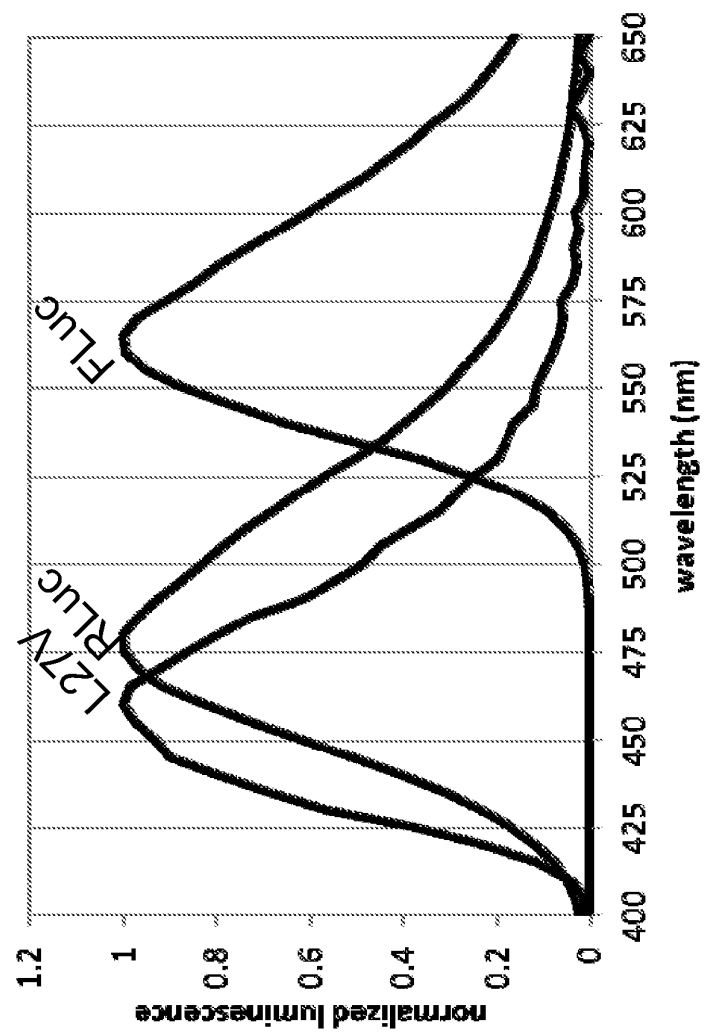


FIG. 71

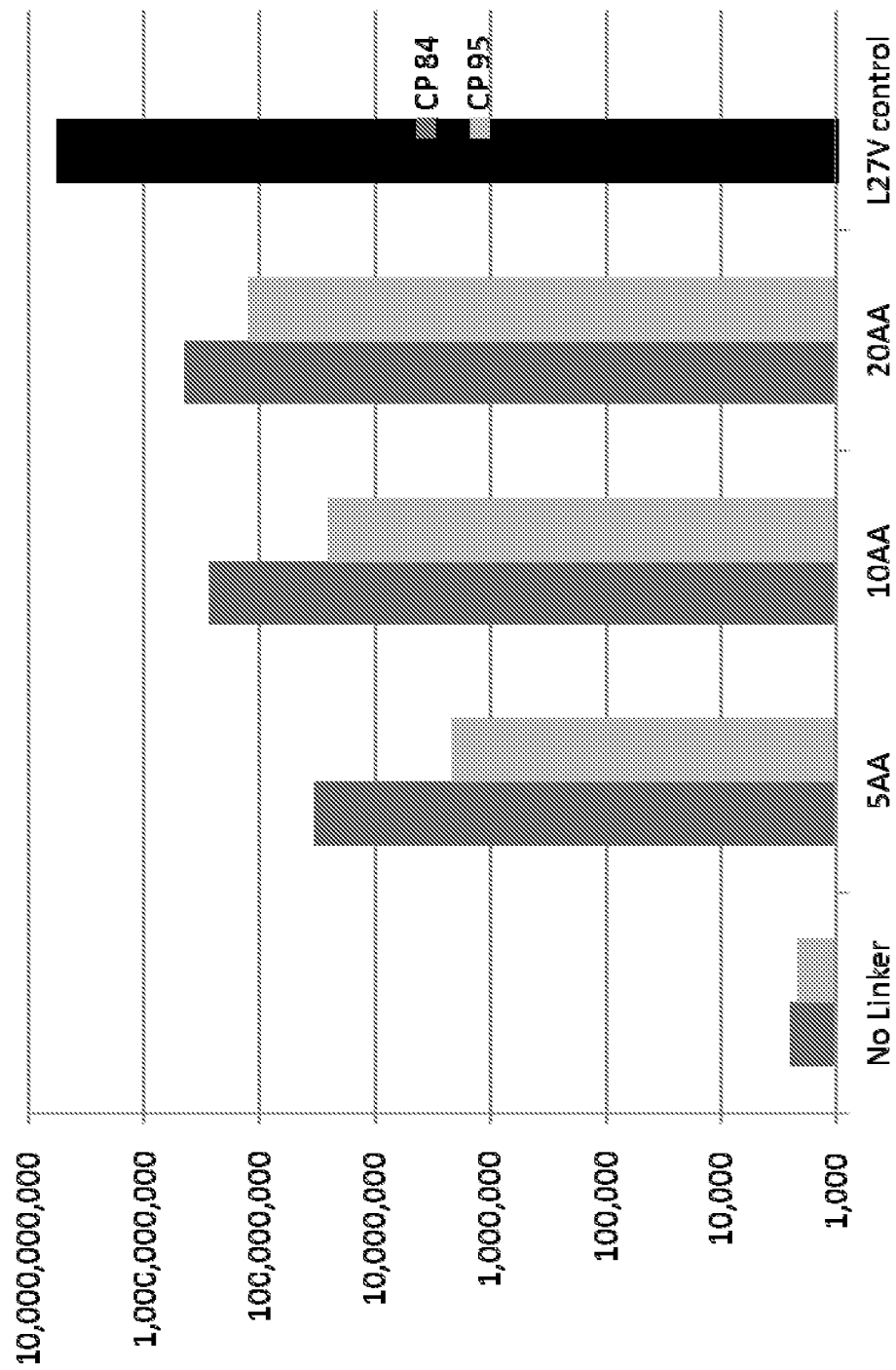


FIG. 72

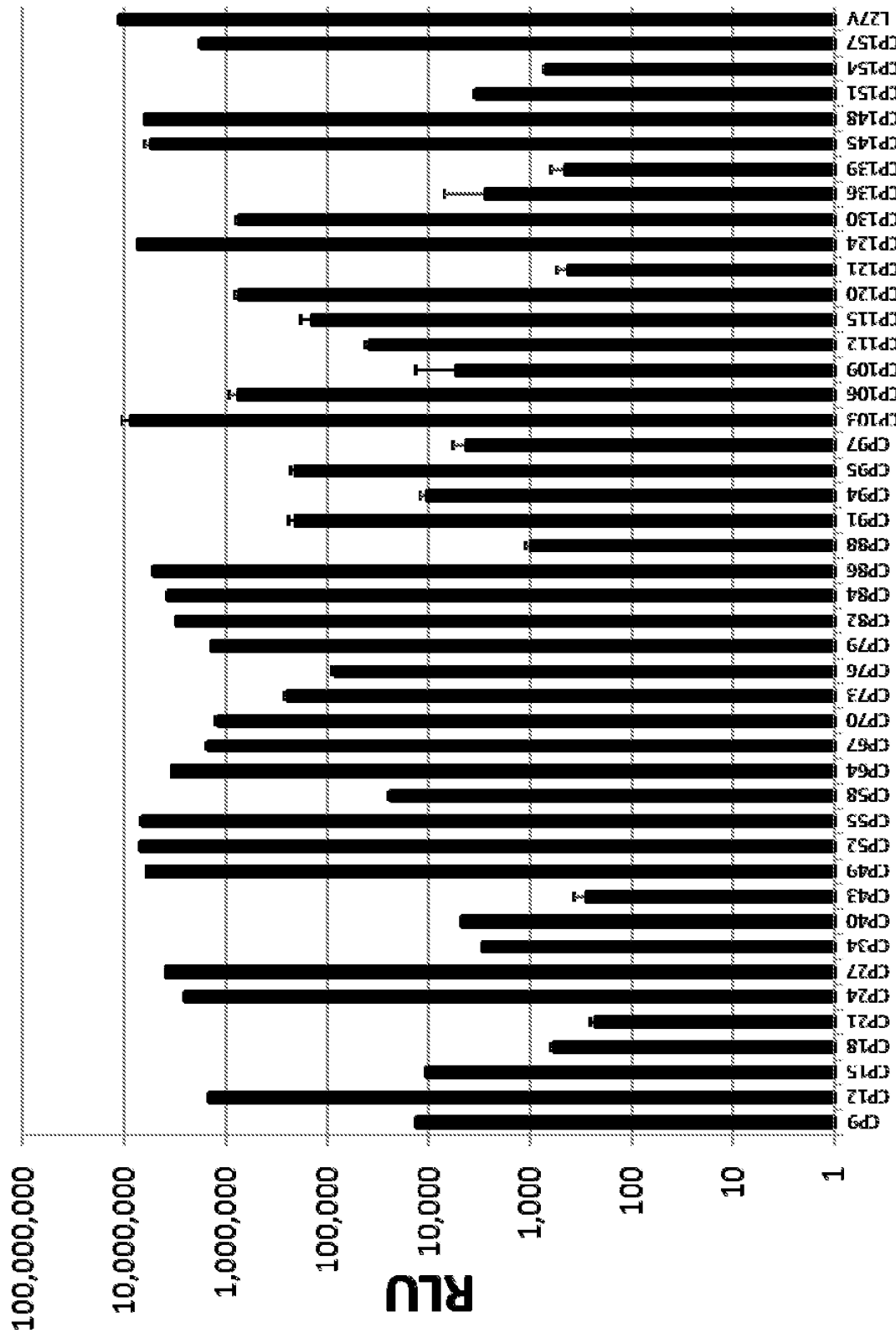


FIG. 73A



FIG. 73B

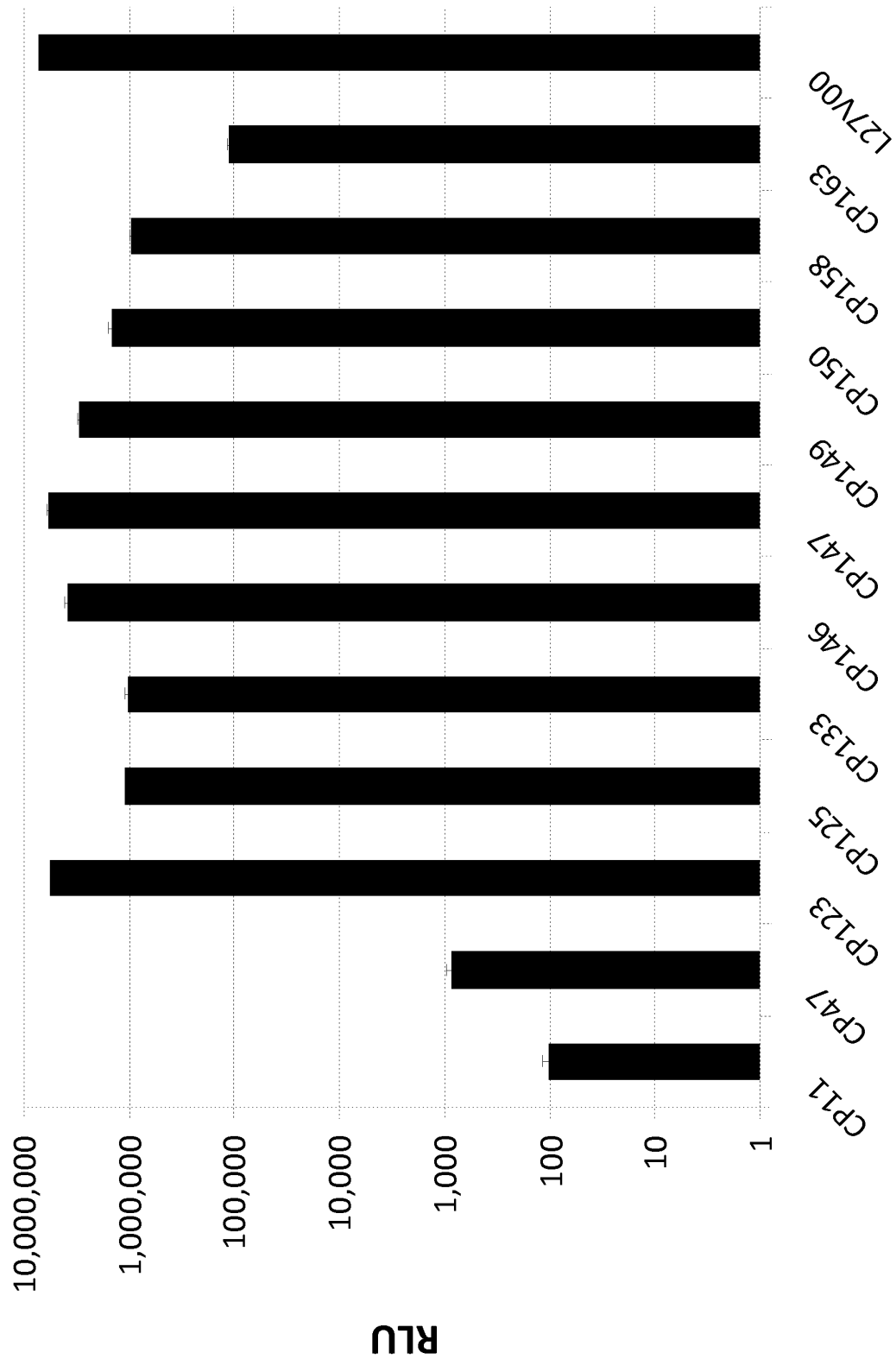


FIG. 73C

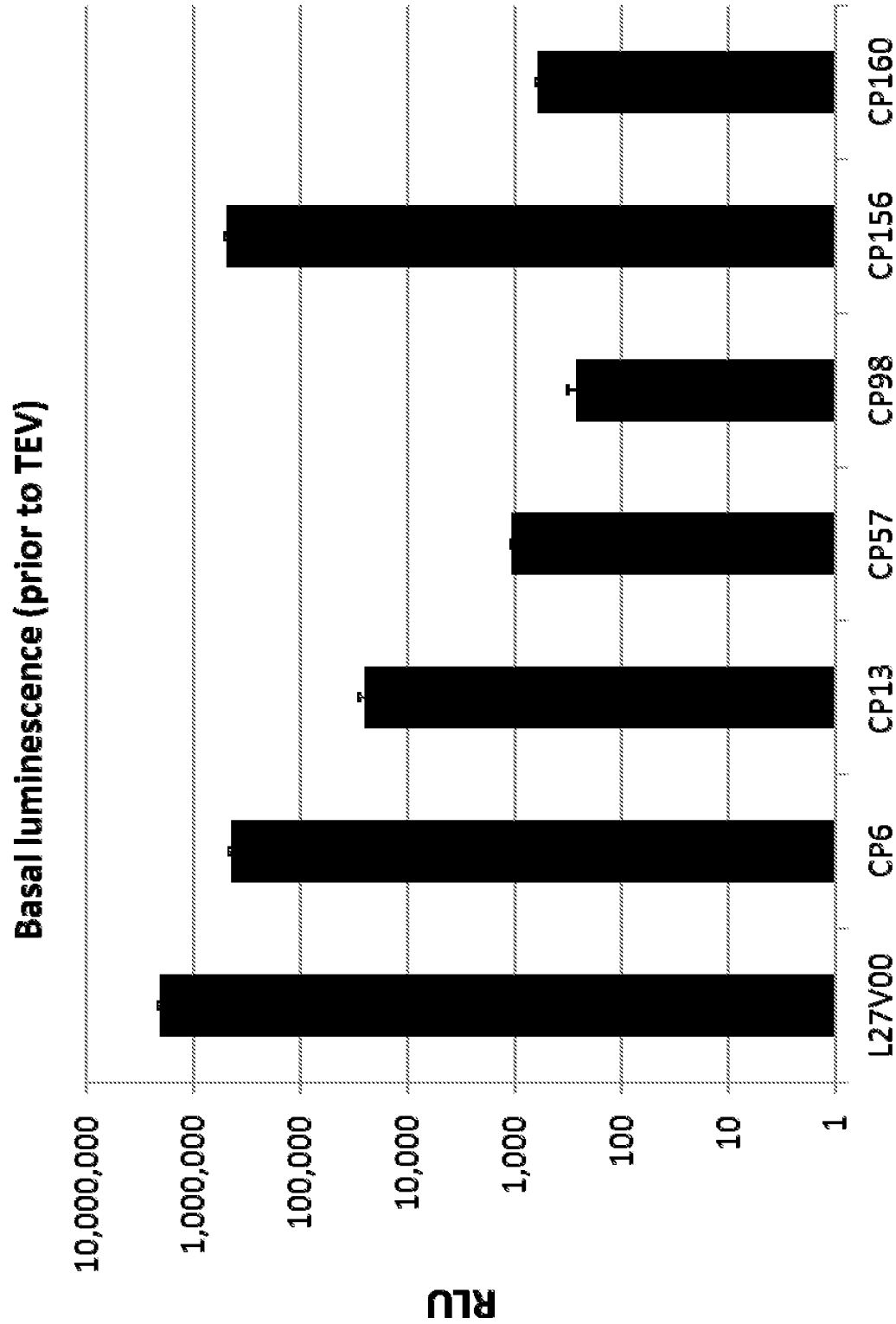


FIG. 73D

| CP site<br>("CP#") | SEQ ID NOs: | response<br>to TEV | CP site<br>("CP#") | SEQ ID NOs: | response<br>to TEV | CP site<br>("CP#") | SEQ ID NOs: | response to<br>TEV | CP site<br>("CP#") | SEQ ID NOs: | response to<br>TEV |
|--------------------|-------------|--------------------|--------------------|-------------|--------------------|--------------------|-------------|--------------------|--------------------|-------------|--------------------|
| 5                  | 114 and 115 | 1.3                | 53                 | 160 and 161 | 2100               | 95                 | 204 and 205 | 512.9              | 136                | 246 and 247 | /                  |
| 6                  | 116 and 117 | 7.0                | 54                 | 162 and 163 | 12.3               | 97                 | 206 and 207 | 9.1                | 139                | 248 and 249 | 1.9                |
| 7                  | 118 and 119 | /                  | 55                 | 164 and 165 | 7744.3             | 98                 | 447 and 448 | 5.4                | 140                | 250 and 251 | /                  |
| 9                  | 120 and 121 | 2.2                | 56                 | 166 and 167 | /                  | 100                | 208 and 209 | 7515.2             | 141                | 252 and 253 | /                  |
| 11                 | 122 and 123 | /                  | 57                 | 445 and 446 | 17.6               | 101                | 210 and 211 | 6329.9             | 142                | 254 and 255 | 5.3                |
| 12                 | 124 and 125 | 1.6                | 58                 | 168 and 169 | 116.7              | 102                | 212 and 213 | 3205.1             | 143                | 256 and 257 | 871.4              |
| 15                 | 126 and 127 | /                  | 64                 | 170 and 171 | 1.4                | 103                | 214 and 215 | 3456.8             | 144                | 258 and 259 | 1.6                |
| 18                 | 128 and 129 | 2.1                | 67                 | 172 and 173 | /                  | 104                | 216 and 217 | 3036.9             | 145                | 260 and 261 | 3.7                |
| 21                 | 130 and 131 | /                  | 70                 | 174 and 175 | /                  | 105                | 218 and 219 | 4931.6             | 146                | 262 and 263 | 6.6                |
| 24                 | 132 and 133 | 3.8                | 73                 | 176 and 177 | /                  | 106                | 220 and 221 | 5.3                | 147                | 264 and 265 | 2.0                |
| 27                 | 134 and 135 | 3.1                | 76                 | 178 and 179 | /                  | 109                | 222 and 223 | 12.3               | 148                | 266 and 267 | 3.8                |
| 34                 | 136 and 137 | 9.3                | 79                 | 180 and 181 | /                  | 112                | 224 and 225 | 82.7               | 149                | 268 and 269 | 4.4                |
| 40                 | 138 and 139 | 1.8                | 80                 | 182 and 183 | 1.3                | 13                 | 443 and 444 | 160.8              | 150                | 270 and 271 | 867.4              |
| 43                 | 140 and 141 | /                  | 81                 | 184 and 185 | 1.3                | 115                | 226 and 227 | /                  | 151                | 272 and 273 | 6.3                |
| 44                 | 142 and 143 | 12.9               | 82                 | 186 and 187 | 1.5                | 120                | 228 and 229 | 1661               | 154                | 274 and 275 | /                  |
| 45                 | 144 and 145 | 1057.7             | 83                 | 188 and 189 | 1.5                | 121                | 230 and 231 | 1.5                | 156                | 276 and 277 | 942.1              |
| 46                 | 146 and 147 | 1277.9             | 84                 | 190 and 191 | 1.2                | 123                | 232 and 233 | 323.6              | 157                | 278 and 279 | 1467.9             |
| 47                 | 148 and 149 | 1.5                | 85                 | 192 and 193 | 1.4                | 124                | 234 and 235 | 1548.2             | 158                | 280 and 281 | 1526.5             |
| 48                 | 150 and 151 | 6.7                | 86                 | 194 and 195 | 1.2                | 125                | 236 and 237 | 1141.8             | 160                | 282 and 283 | 12.4               |
| 49                 | 152 and 153 | 12.5               | 87                 | 196 and 197 | 1.5                | 129                | 238 and 239 | 40.2               | 163                | 284 and 285 | /                  |
| 50                 | 154 and 155 | 2.2                | 88                 | 198 and 199 | 1.8                | 130                | 240 and 241 | 2,043.0            | 166                | 286 and 287 | /                  |
| 51                 | 156 and 157 | 2.8                | 91                 | 200 and 201 | 324.2              | 131                | 242 and 243 | 612.5              |                    |             |                    |
| 52                 | 158 and 159 | 15.9               | 94                 | 202 and 203 | 5.5                | 133                | 244 and 245 | 1.5                |                    |             |                    |

FIG. 73E

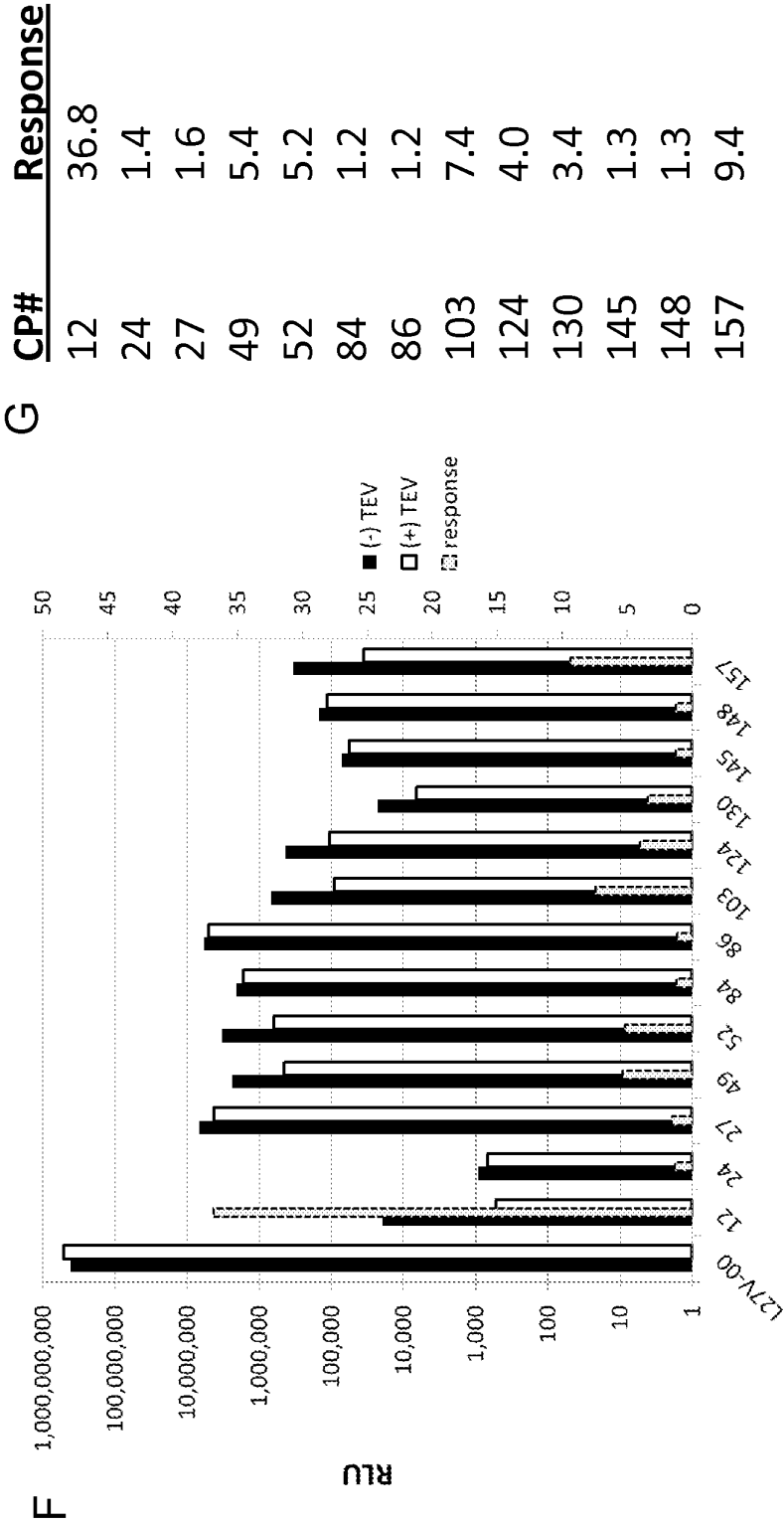


FIG. 73

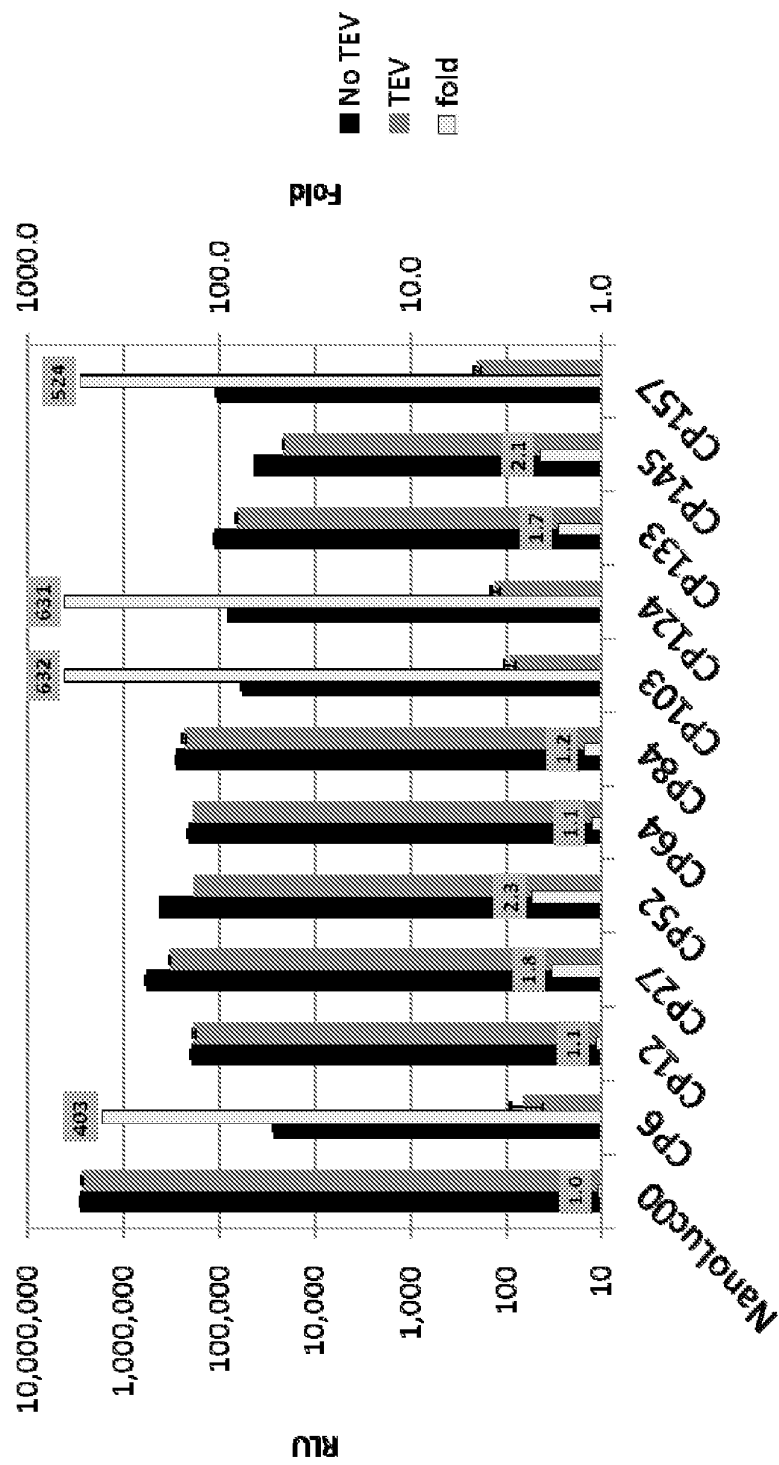


FIG. 73H

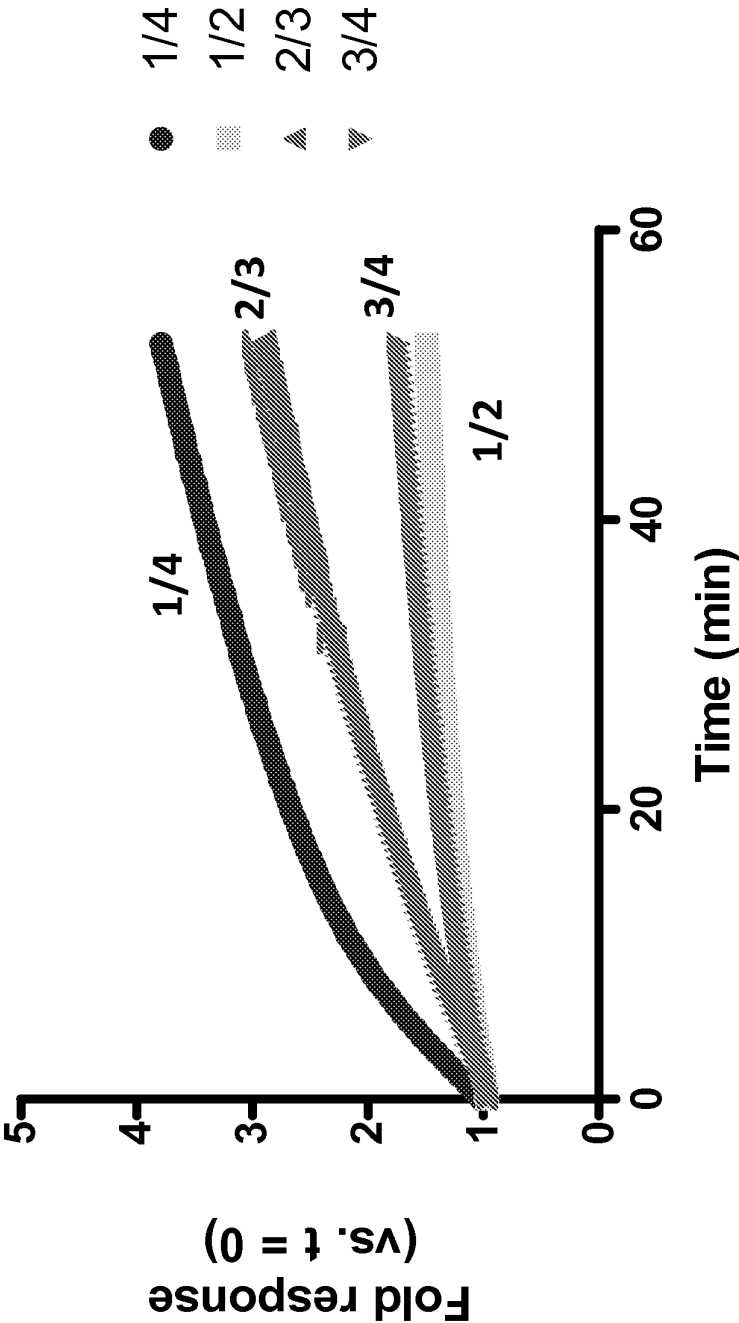


FIG. 74

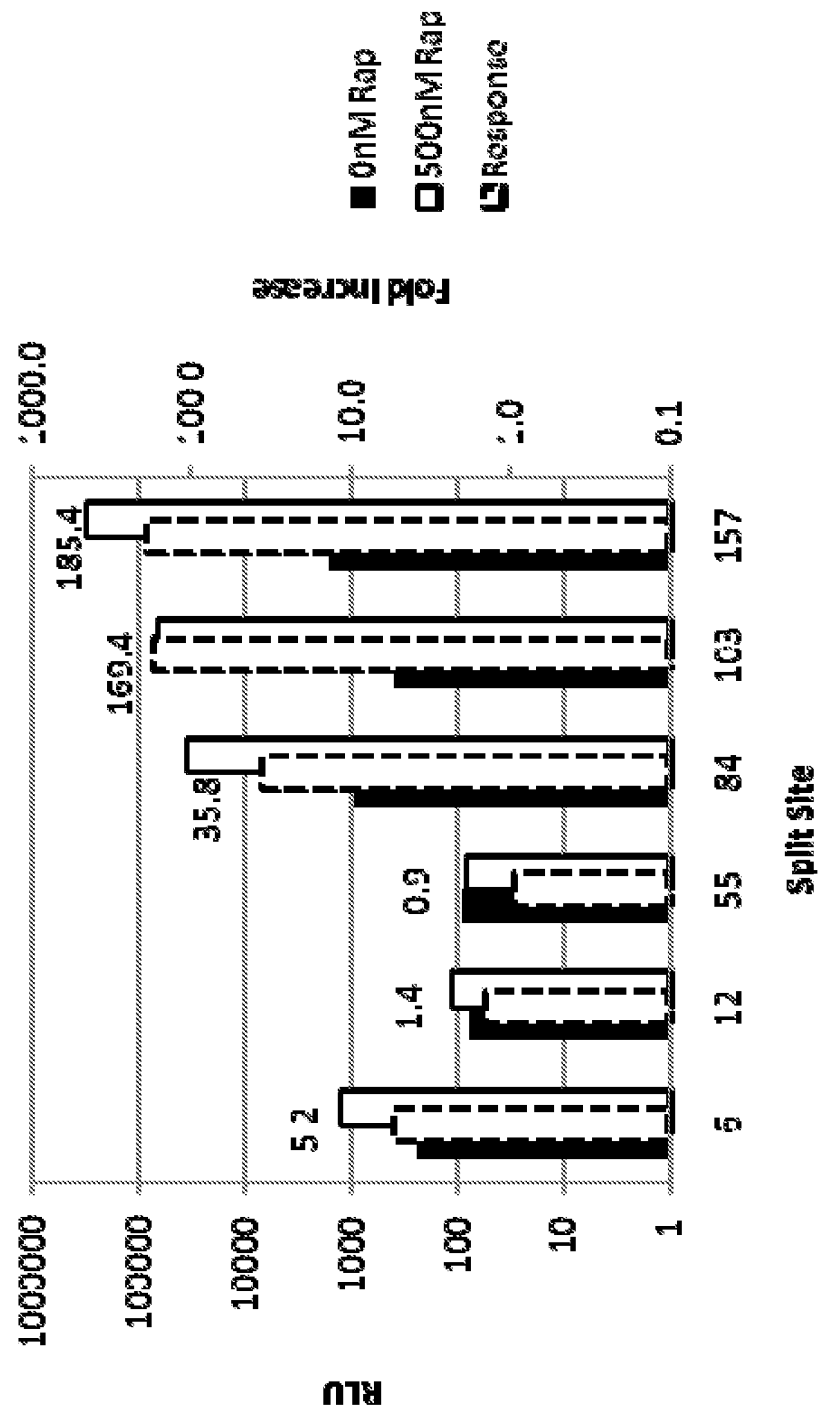


FIG. 75A

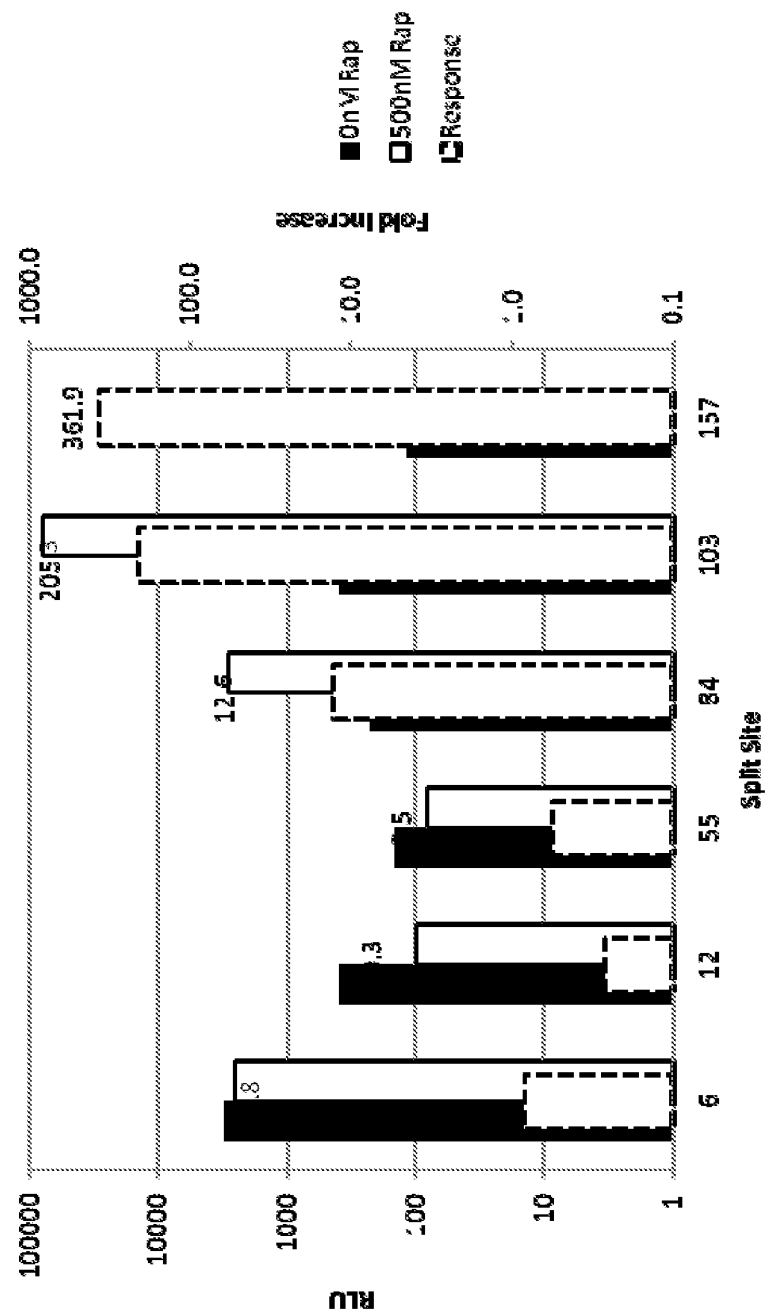


FIG. 75B

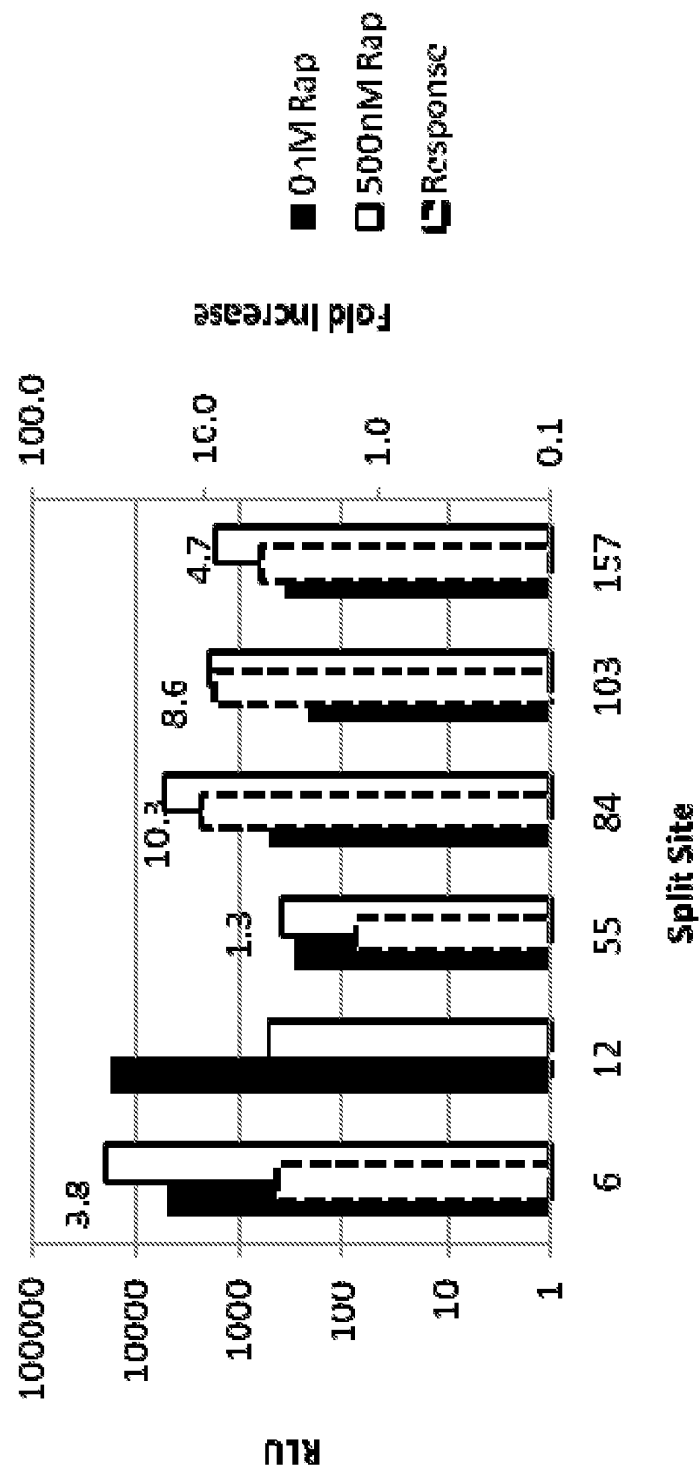


FIG. 75C

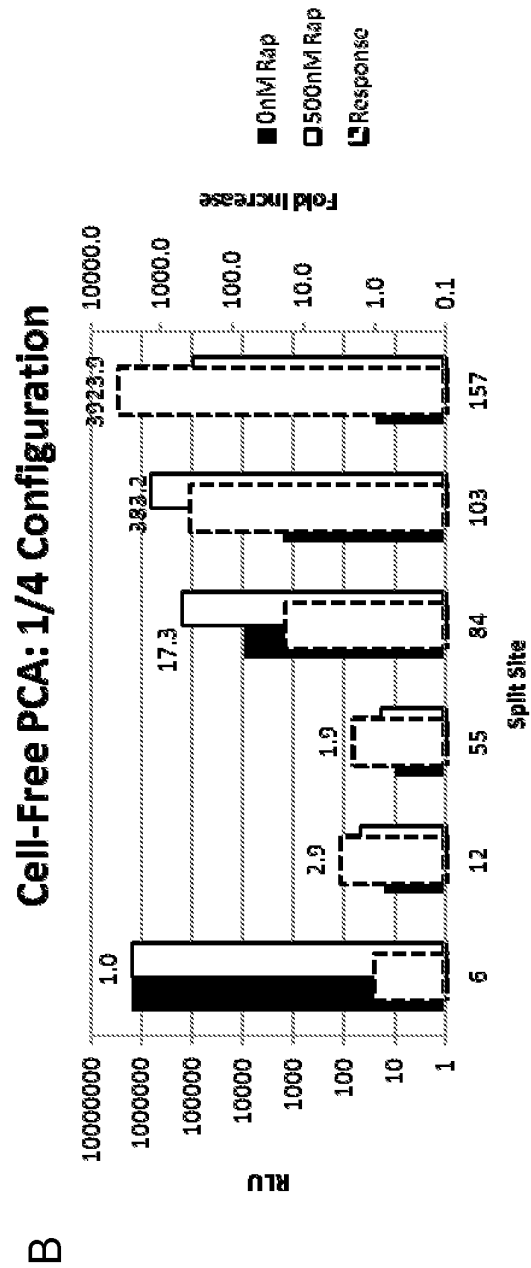
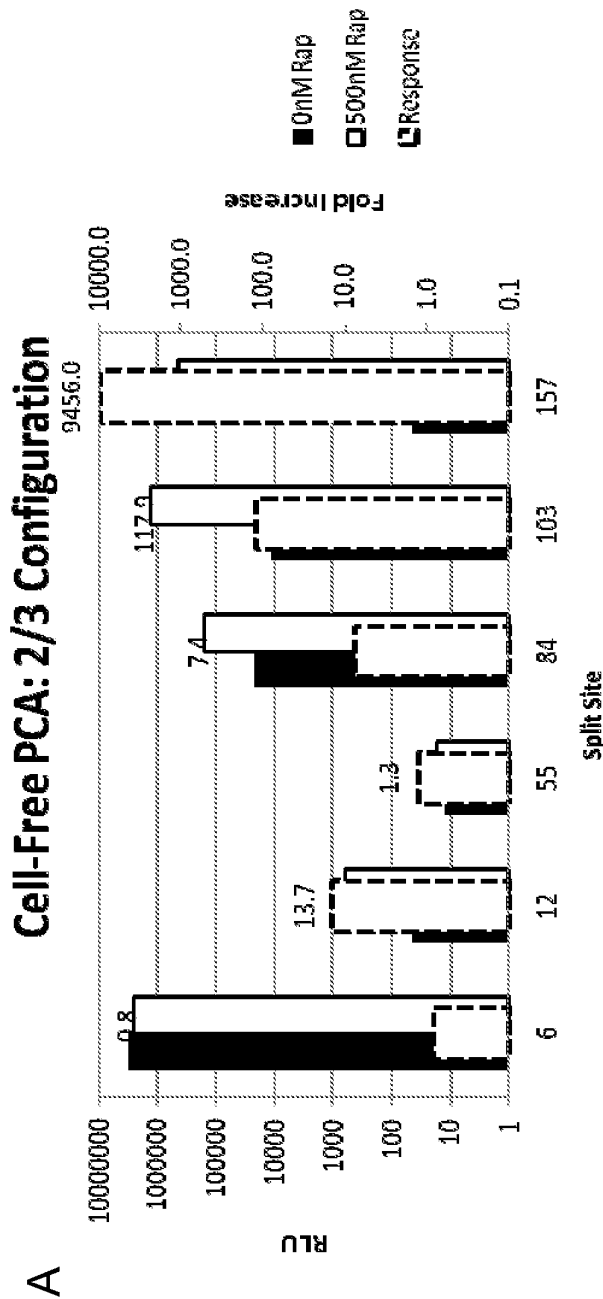


FIG. 76

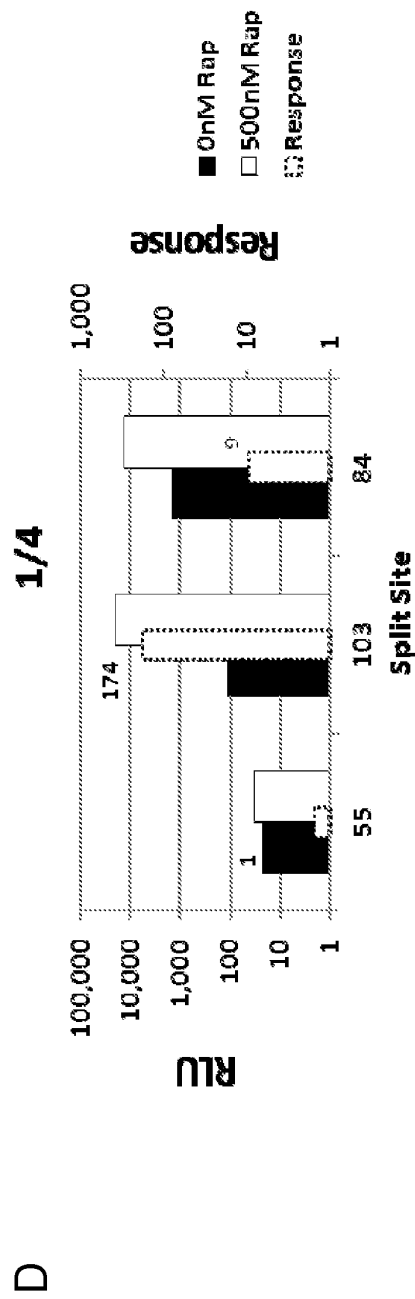
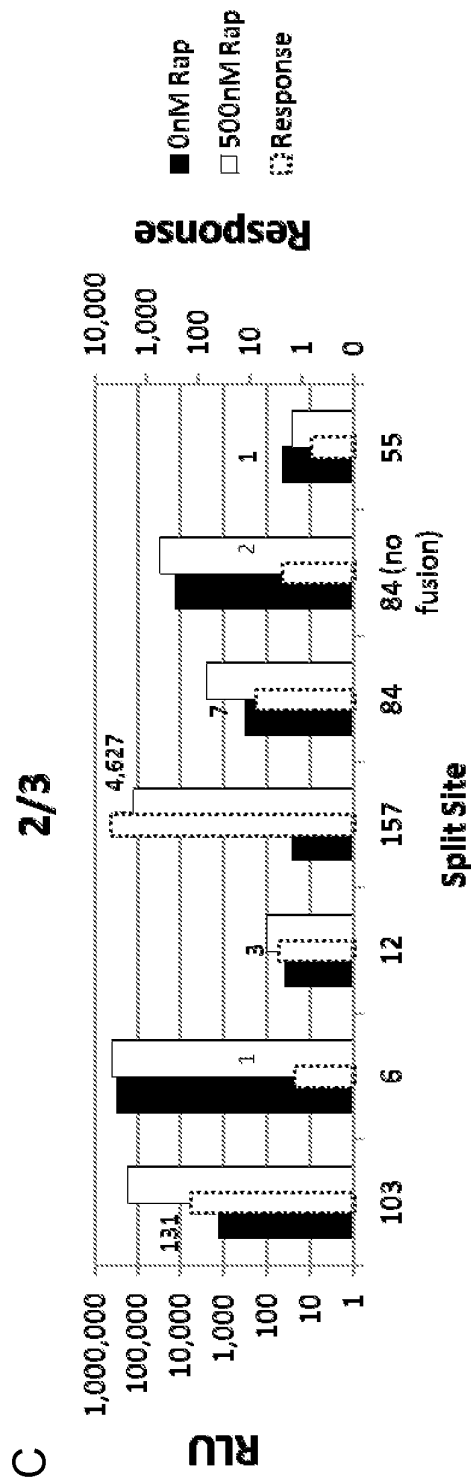


FIG. 76

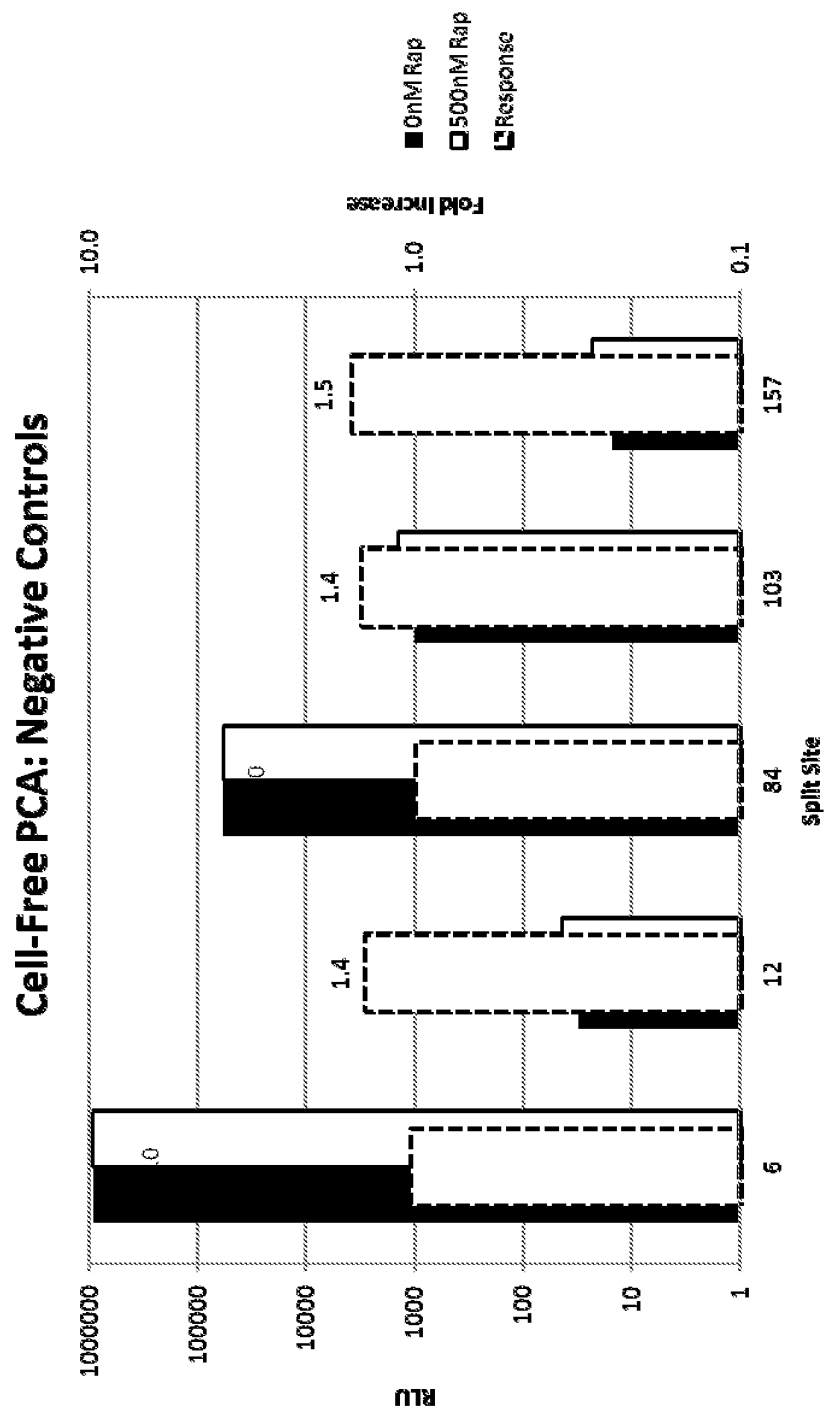


FIG. 76E

# PCA (Cell-Free, "1/4")

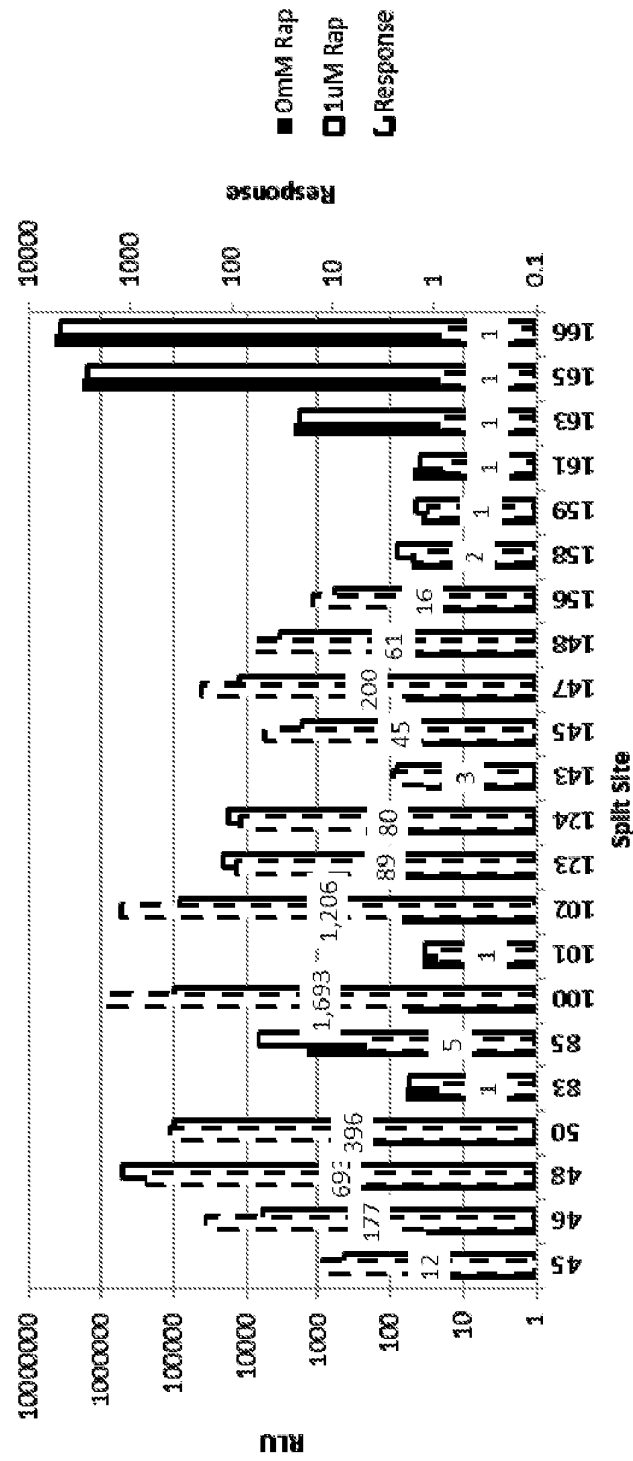


FIG. 76F

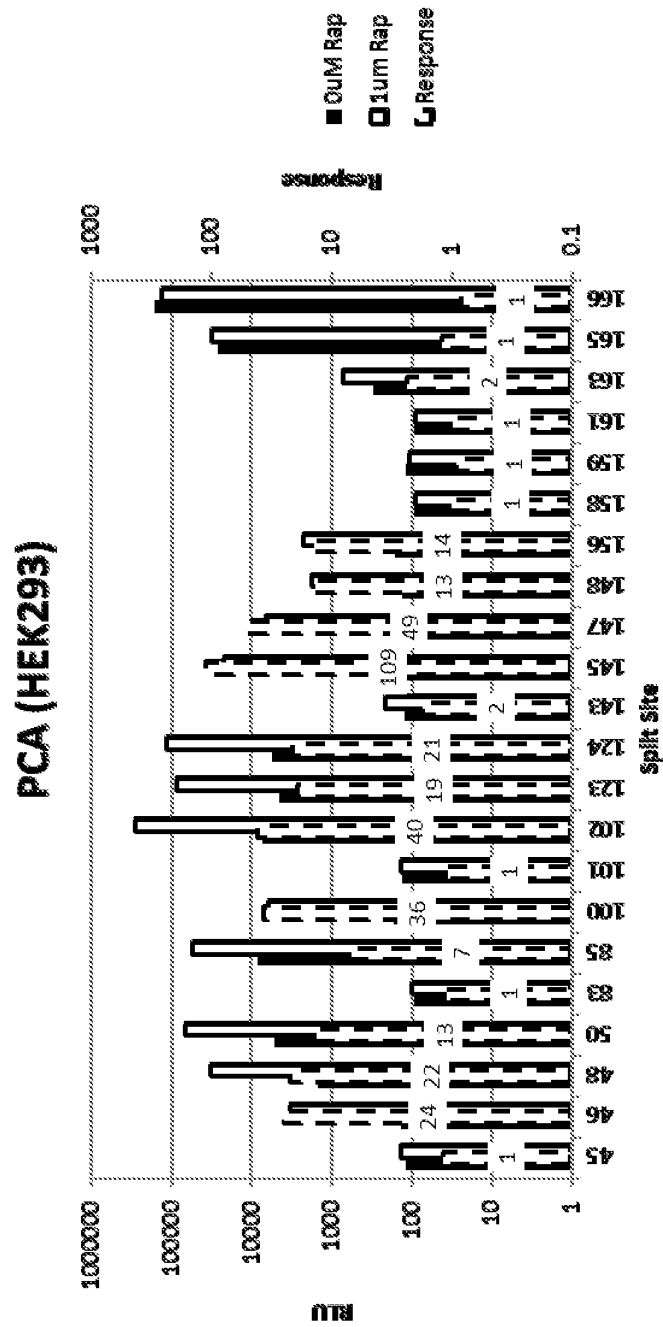


FIG. 76G

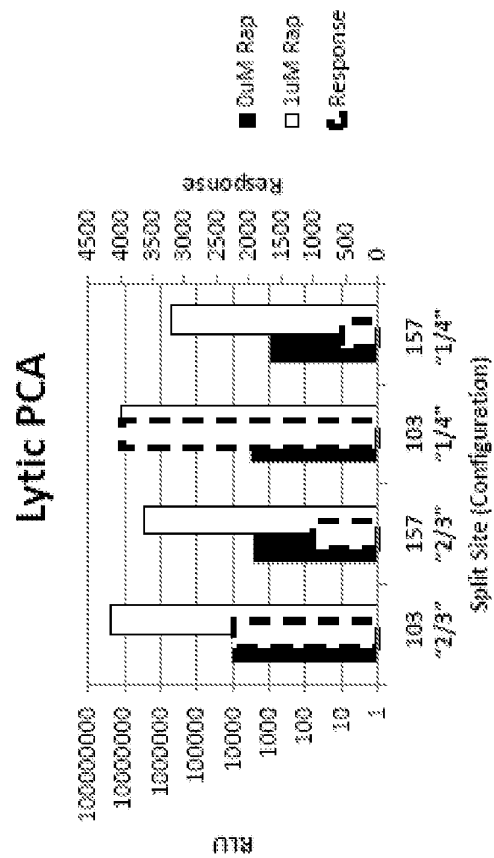


FIG. 76H

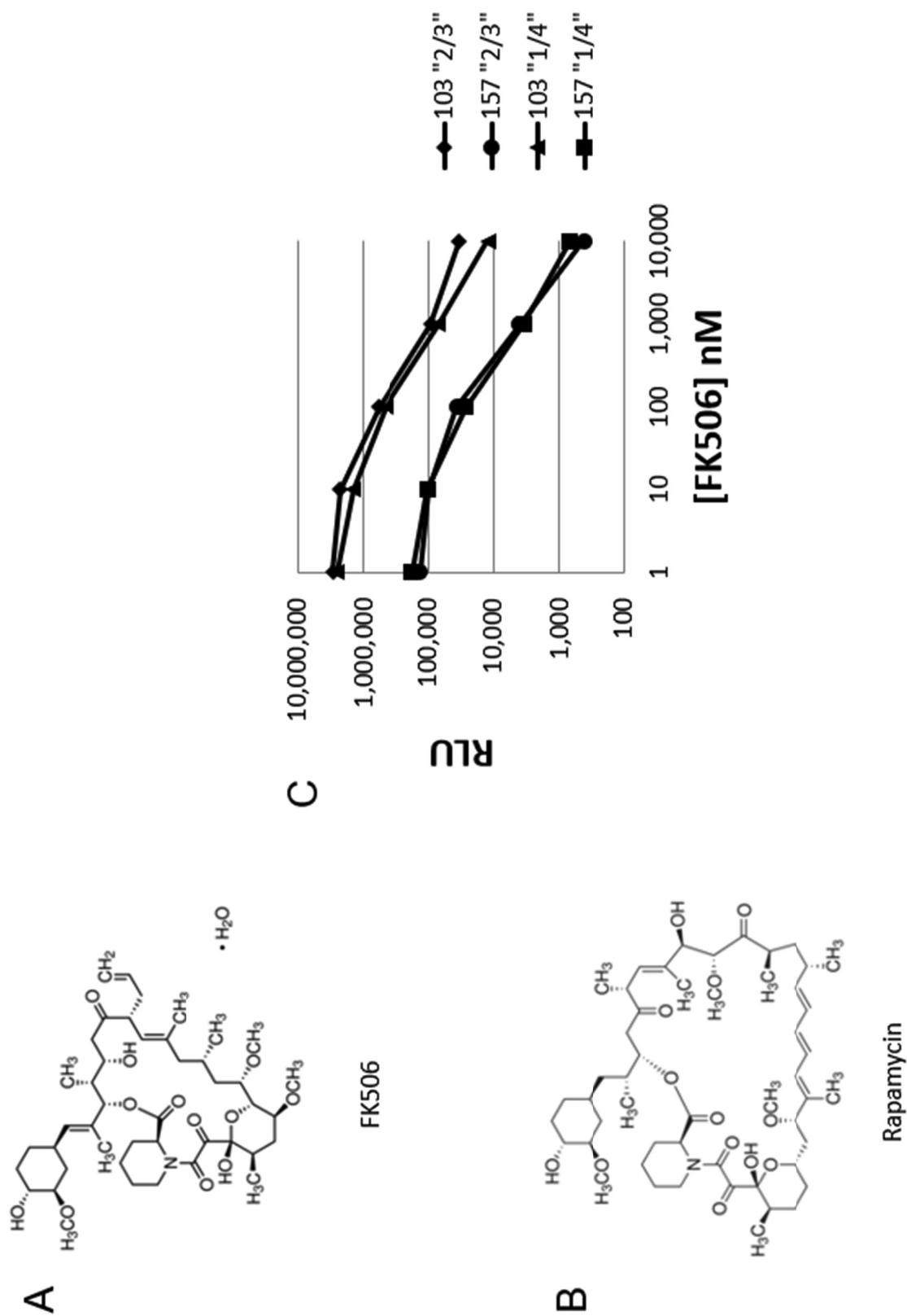


FIG. 77

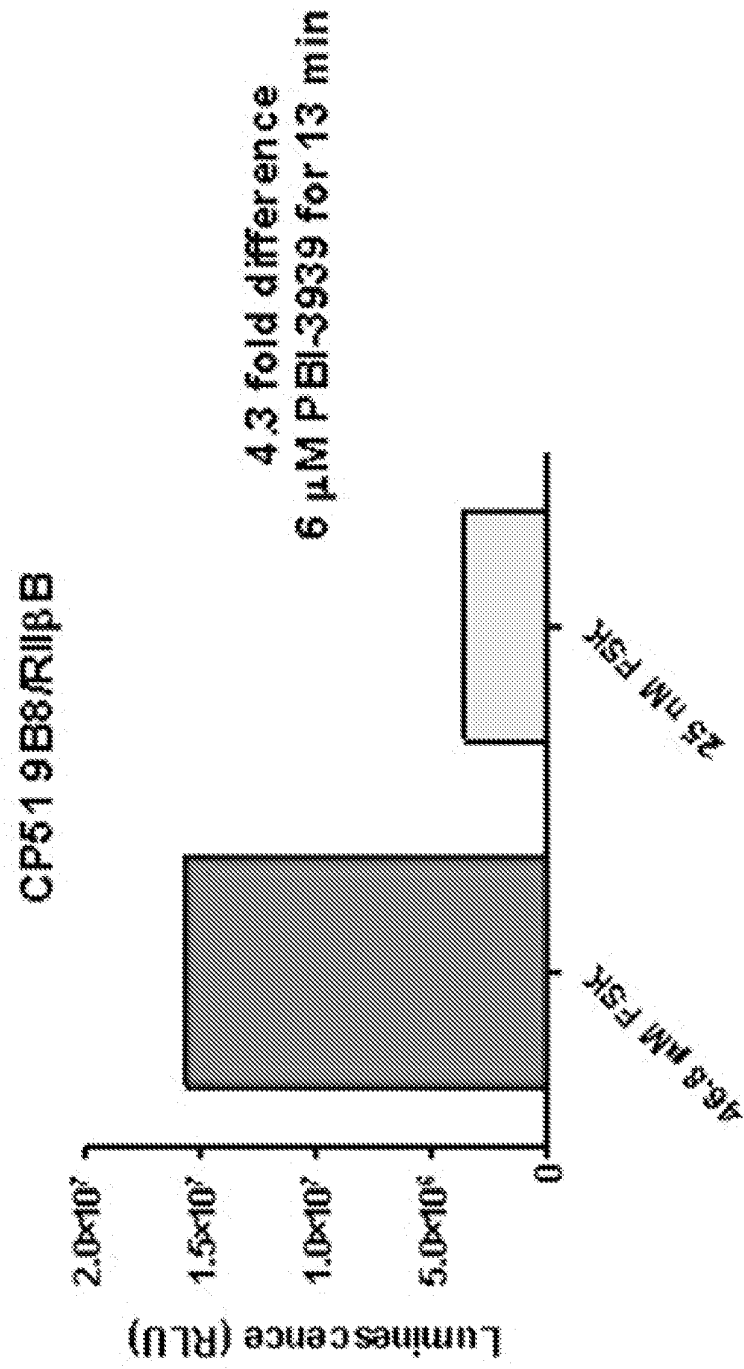


FIG. 78

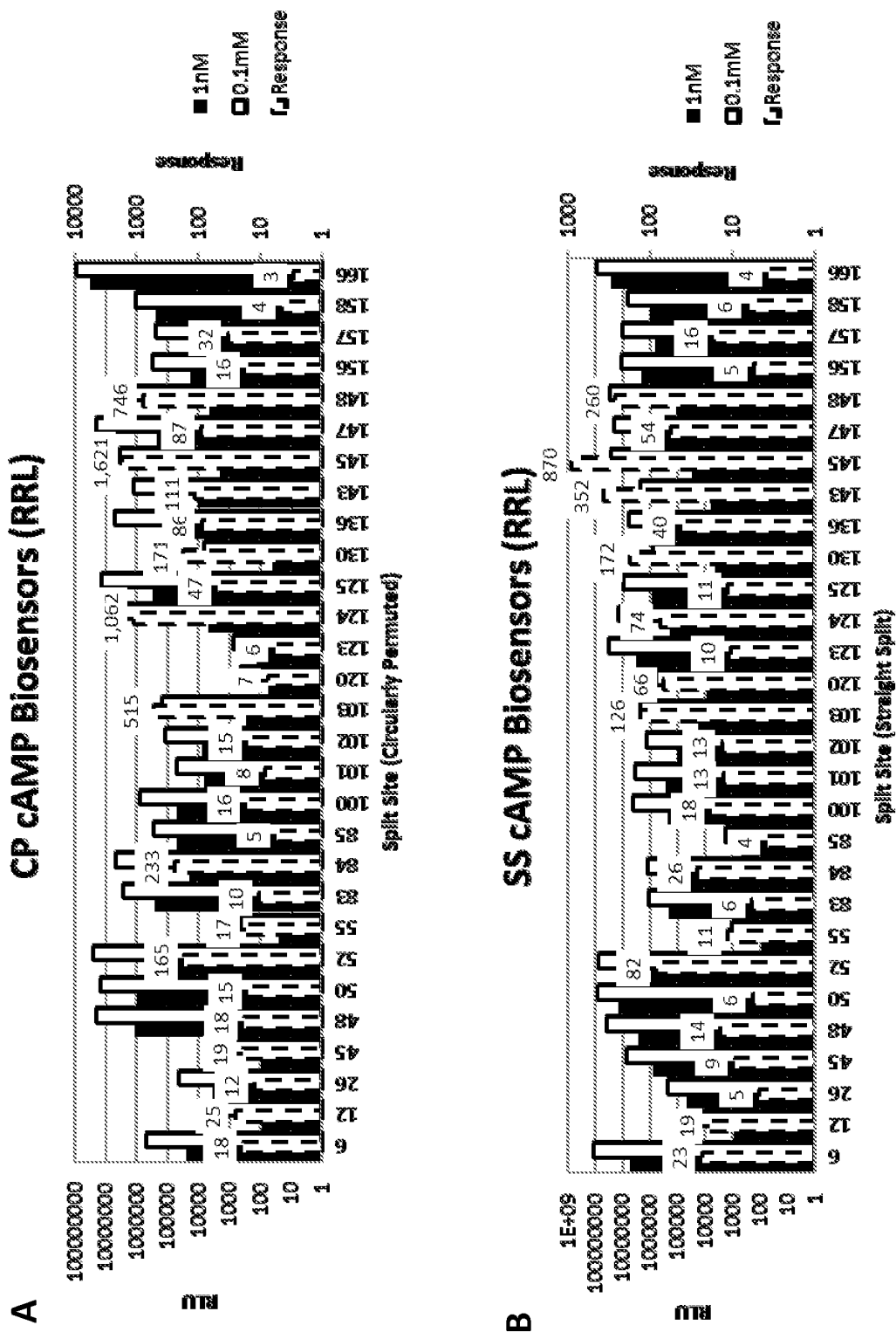


FIG. 79

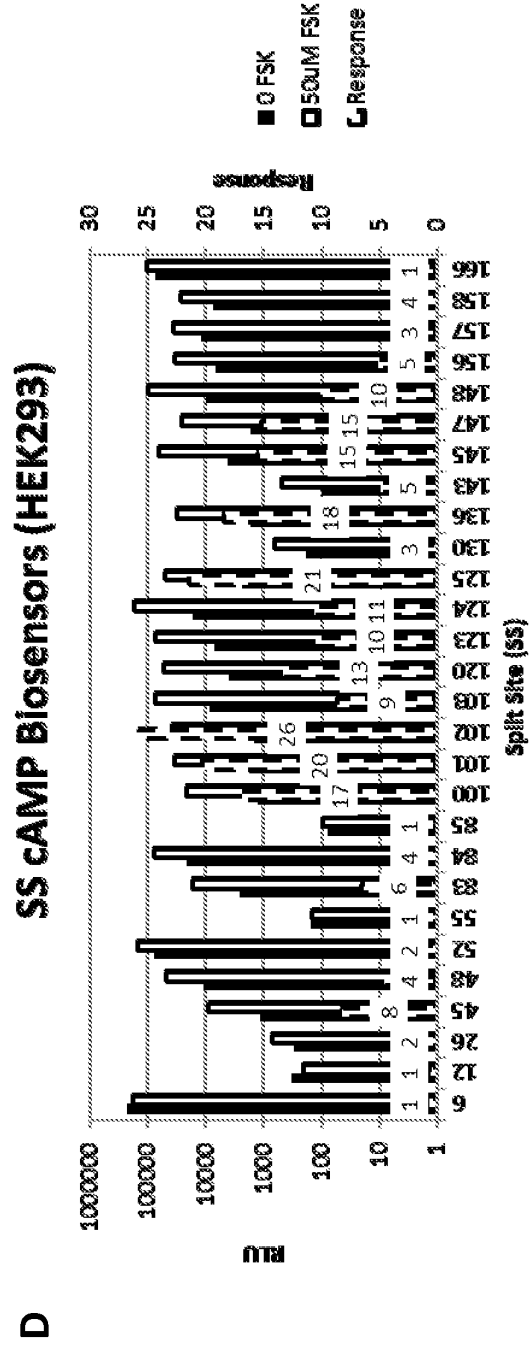
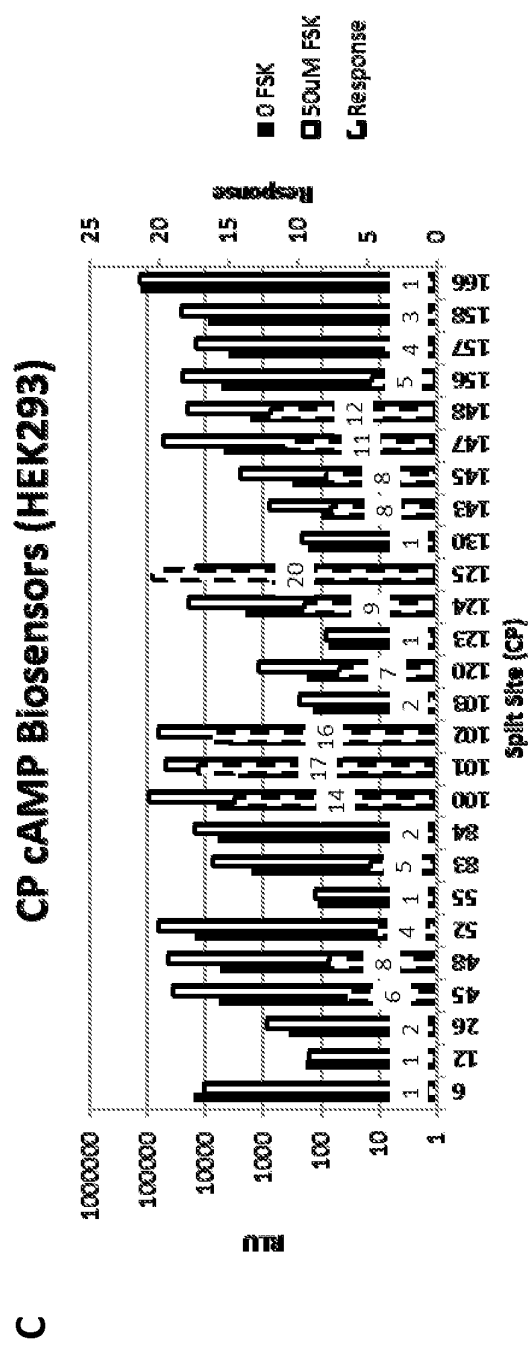


FIG. 79

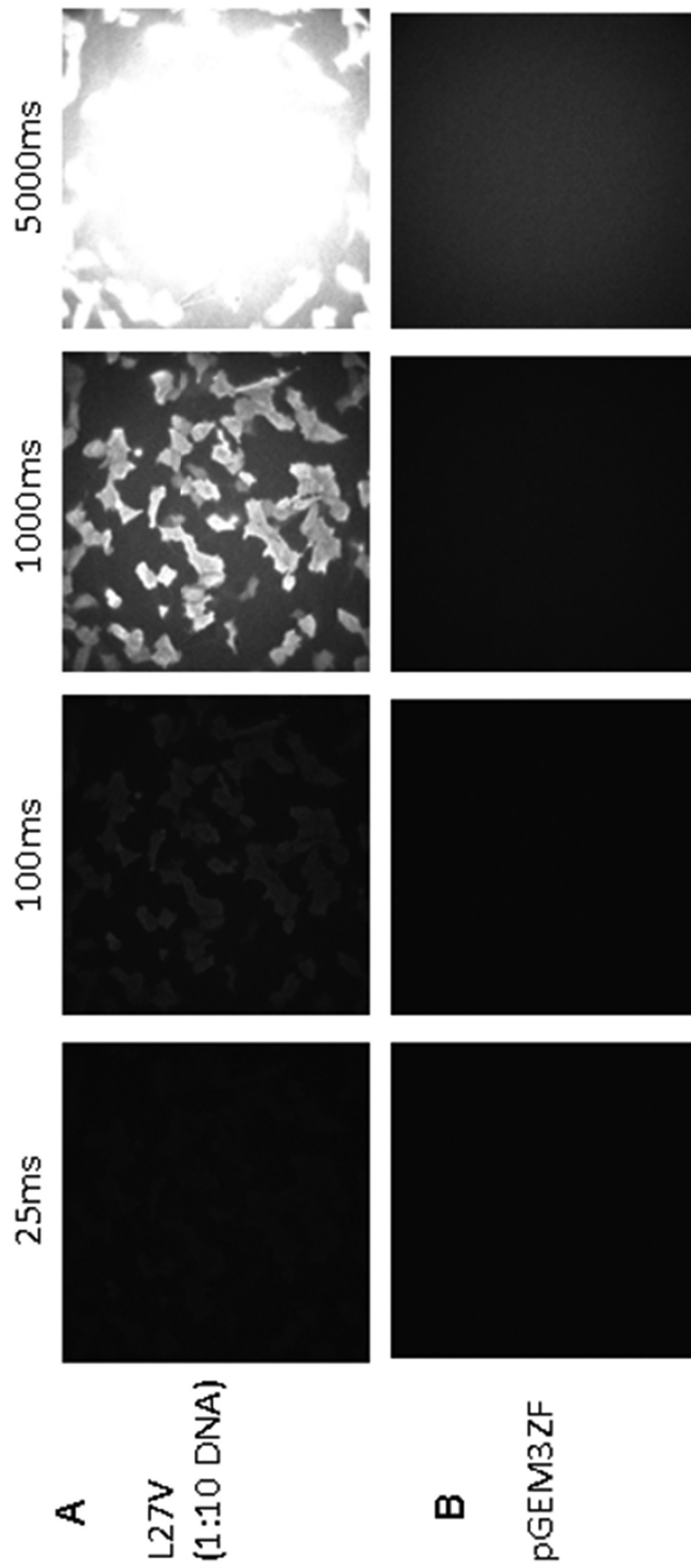


FIG. 80

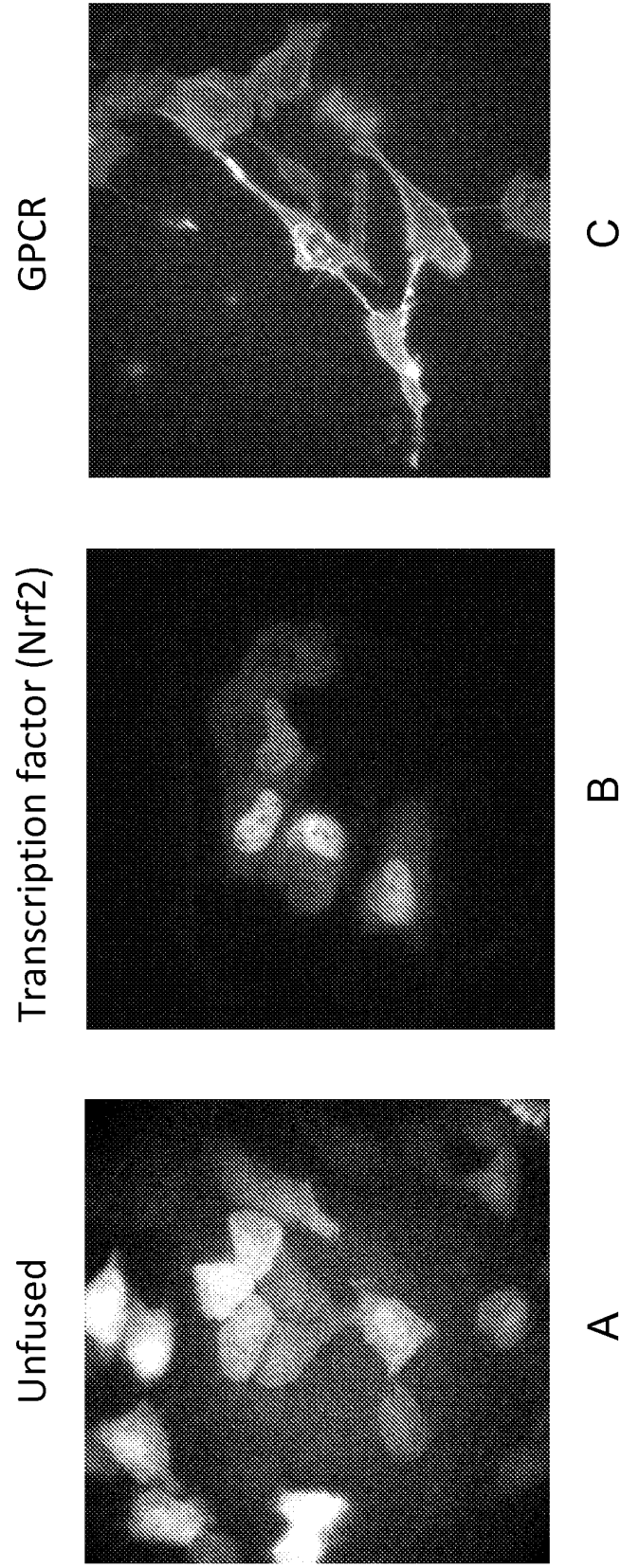


FIG. 81

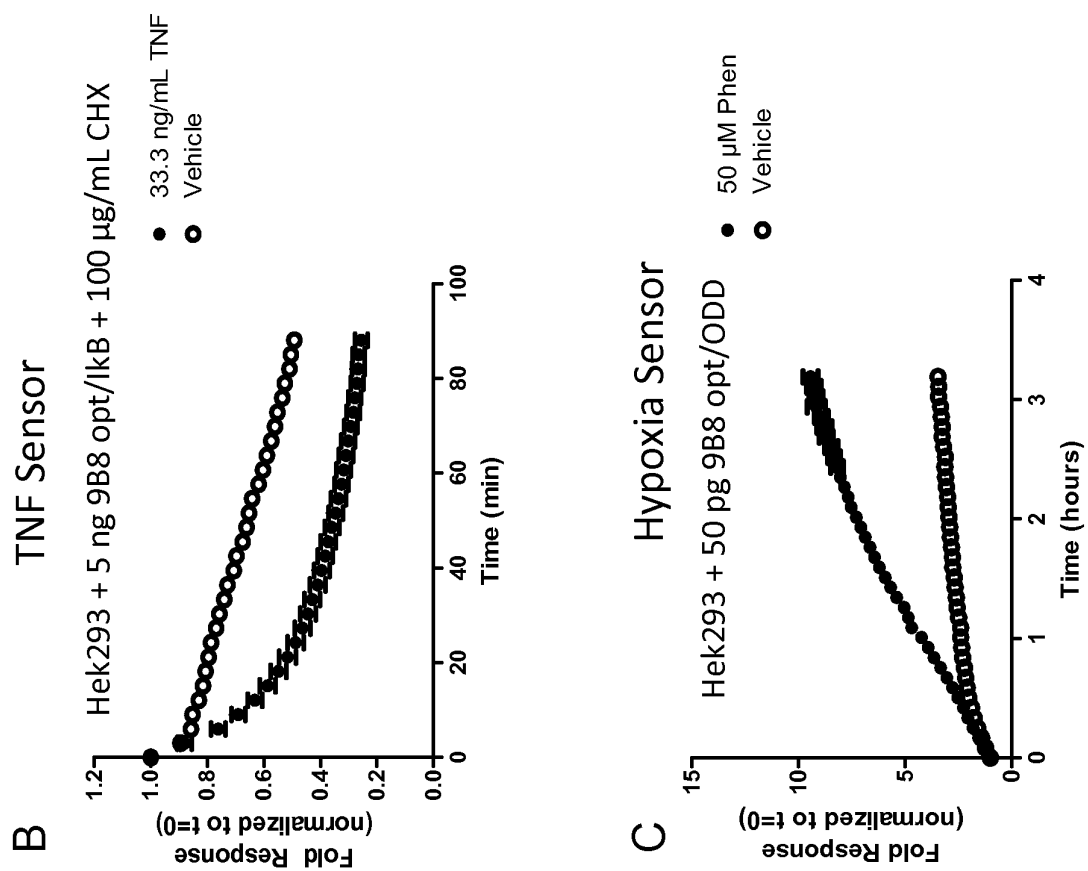


FIG. 82

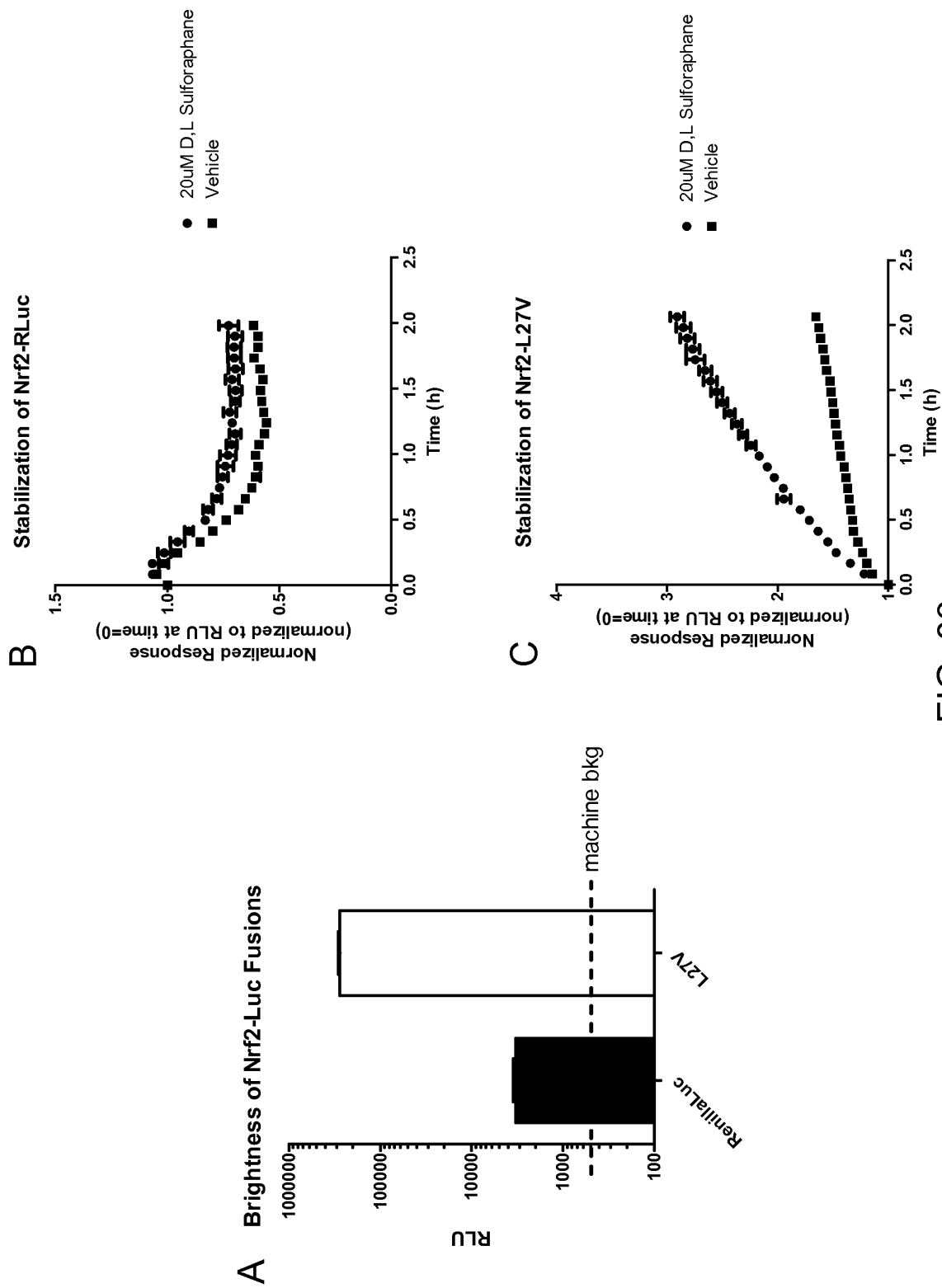


FIG. 83

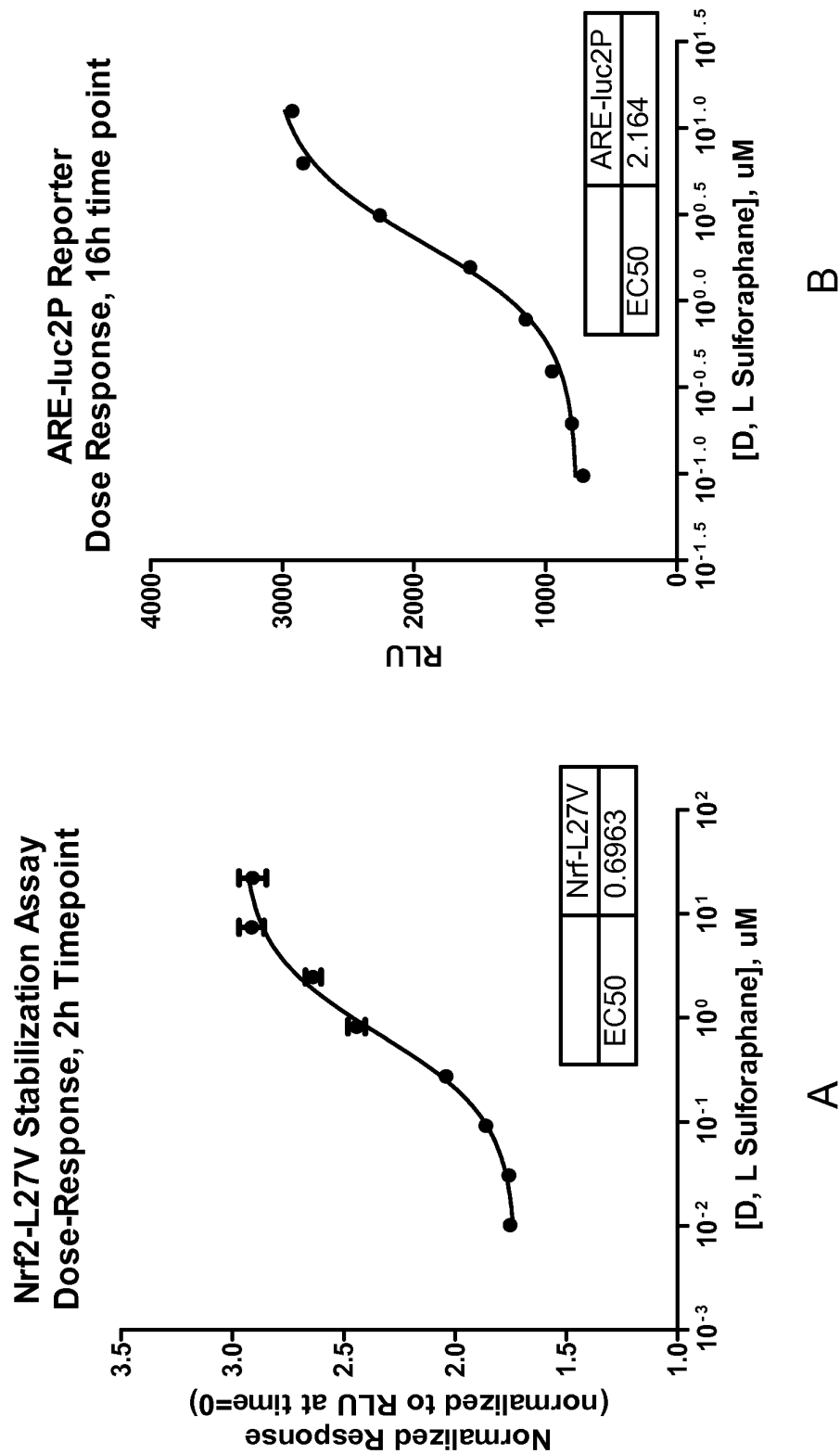
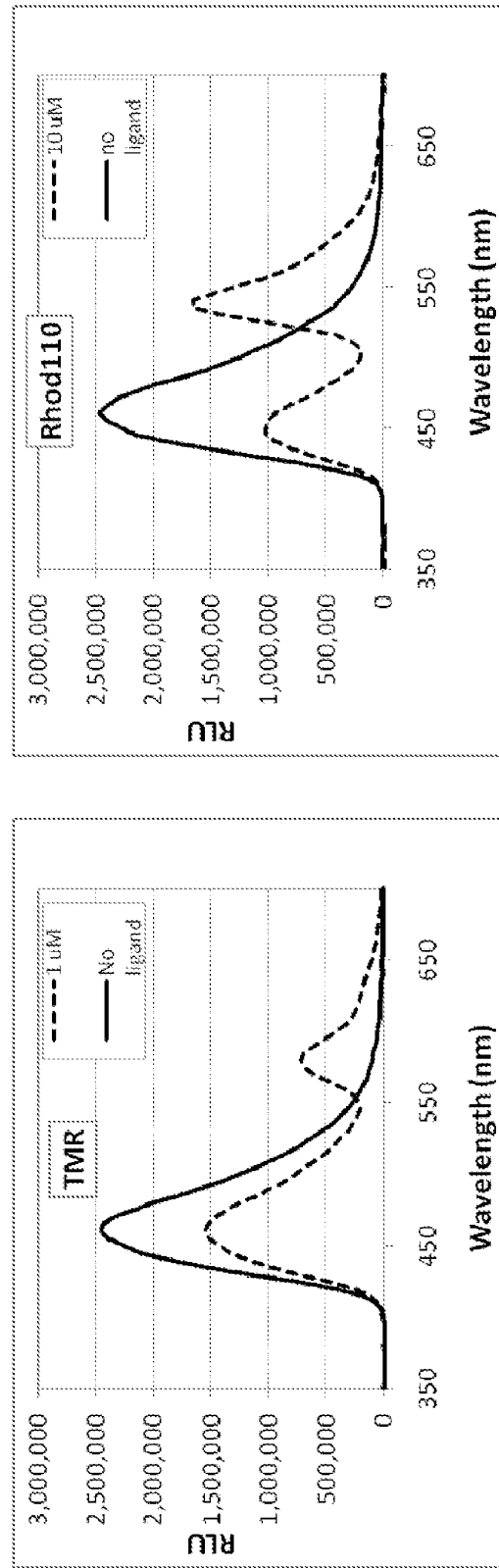


FIG. 84



A

B

FIG. 85

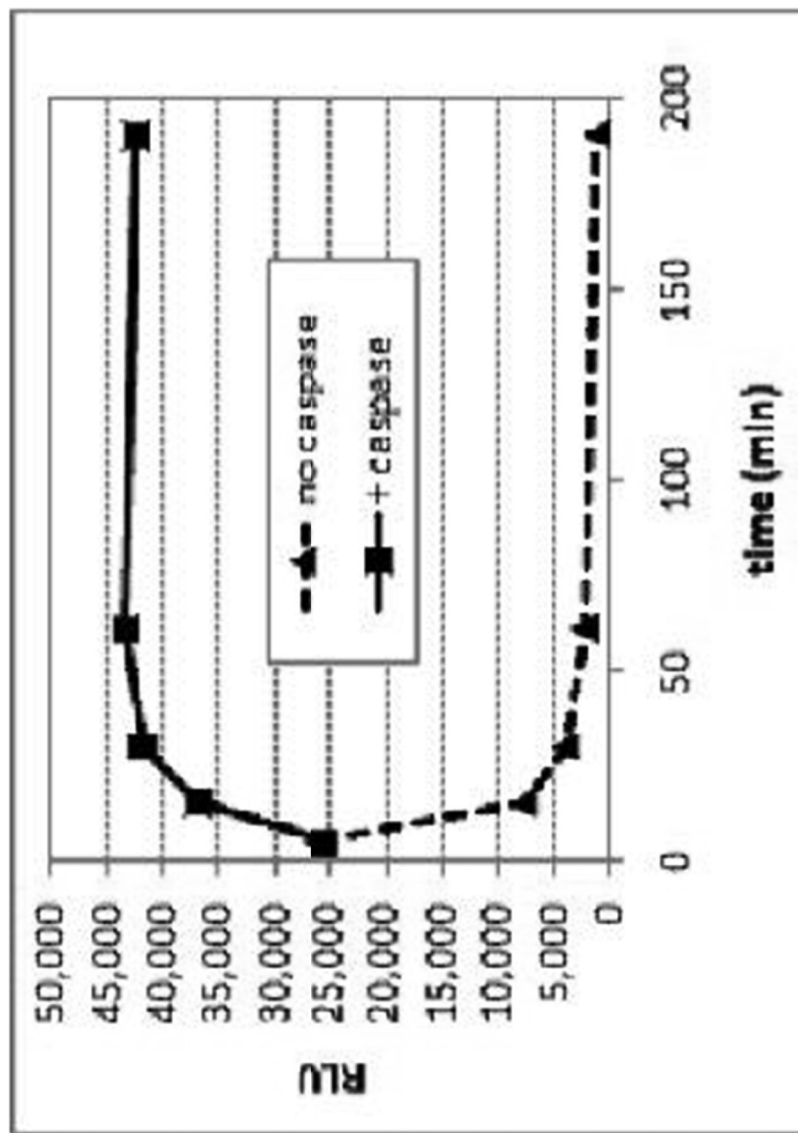


FIG. 86

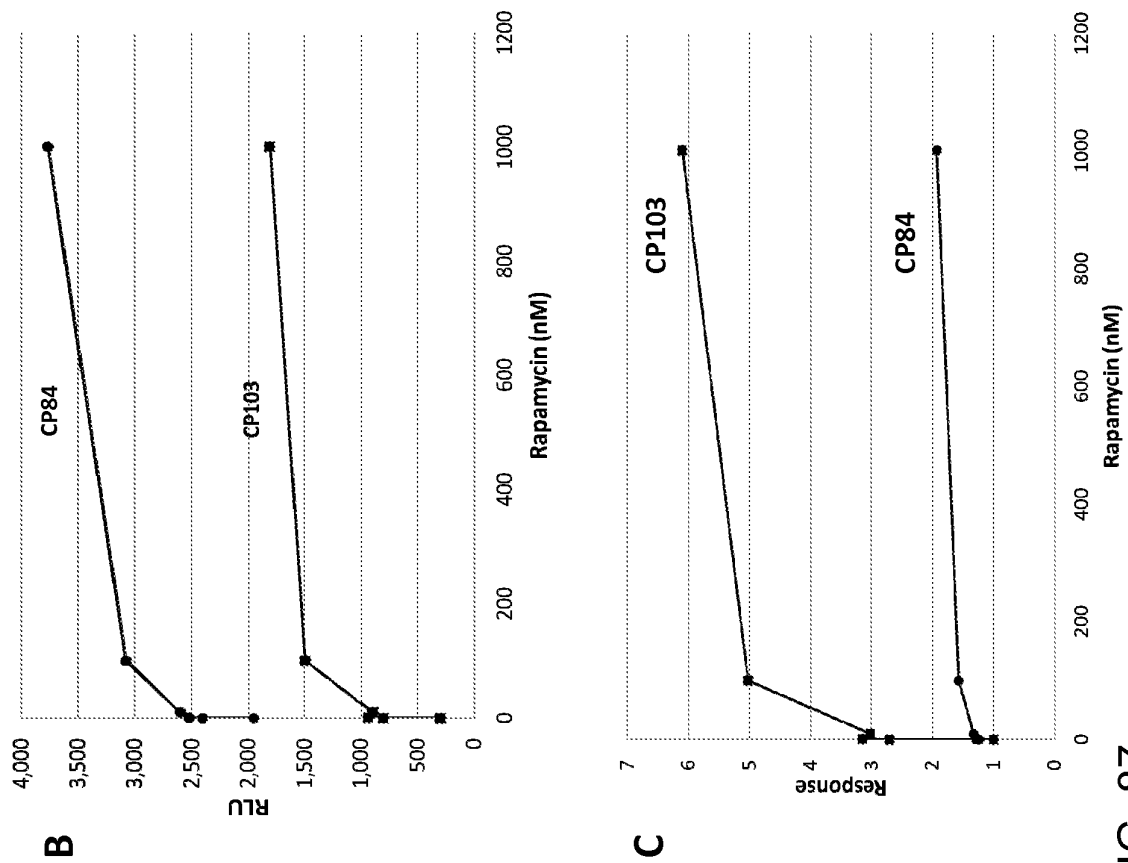


FIG. 87

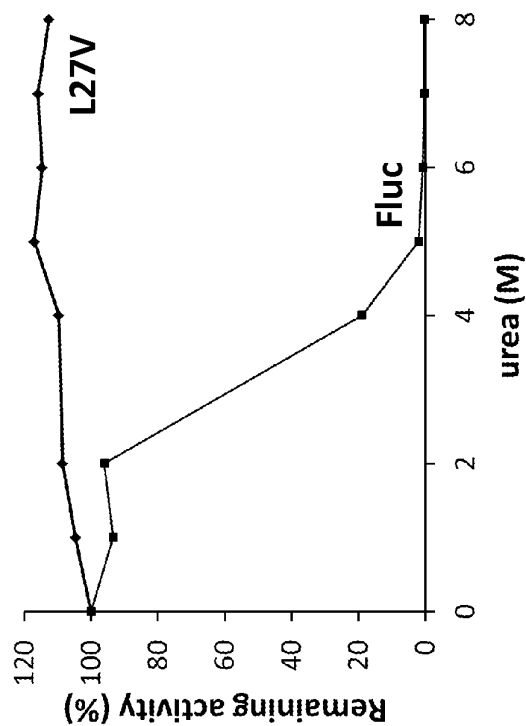


FIG. 88

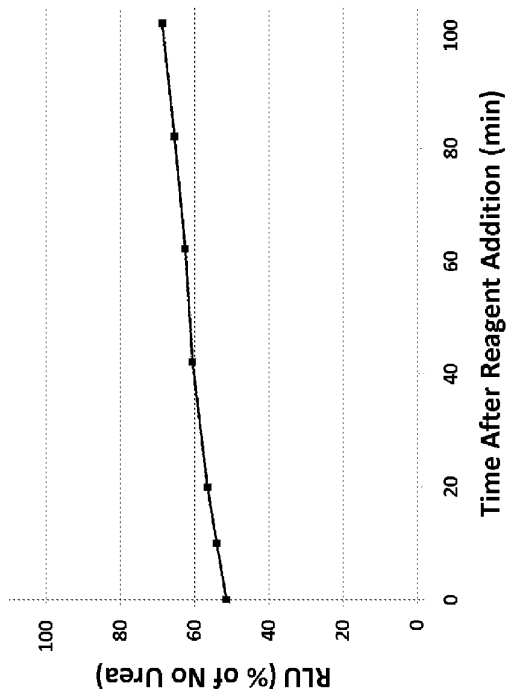
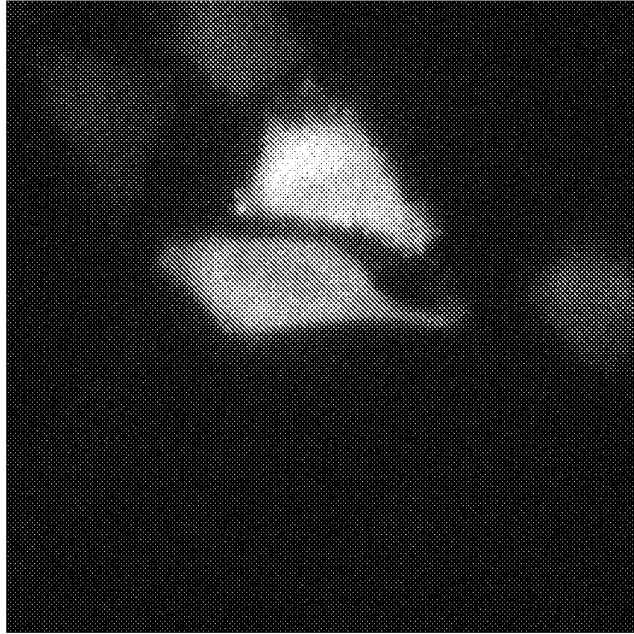


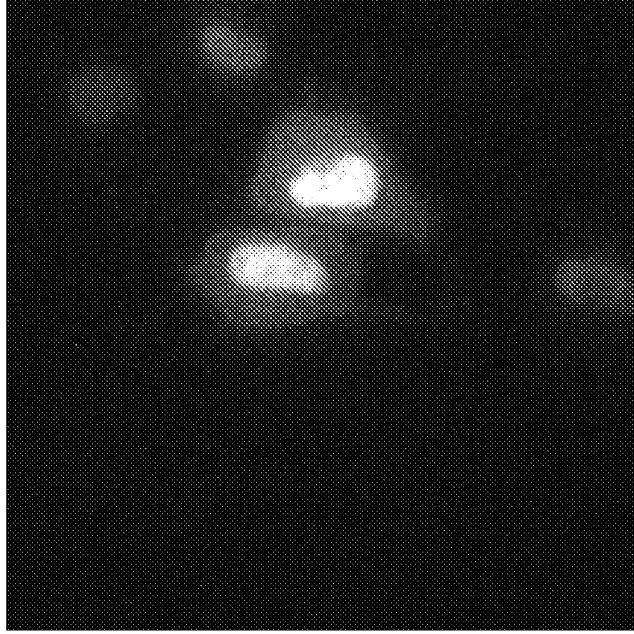
FIG. 89

**+ Dexamethasone, 0 min**



**A**

**+ Dexamethasone, 13 min**



**B**

**FIG. 90**

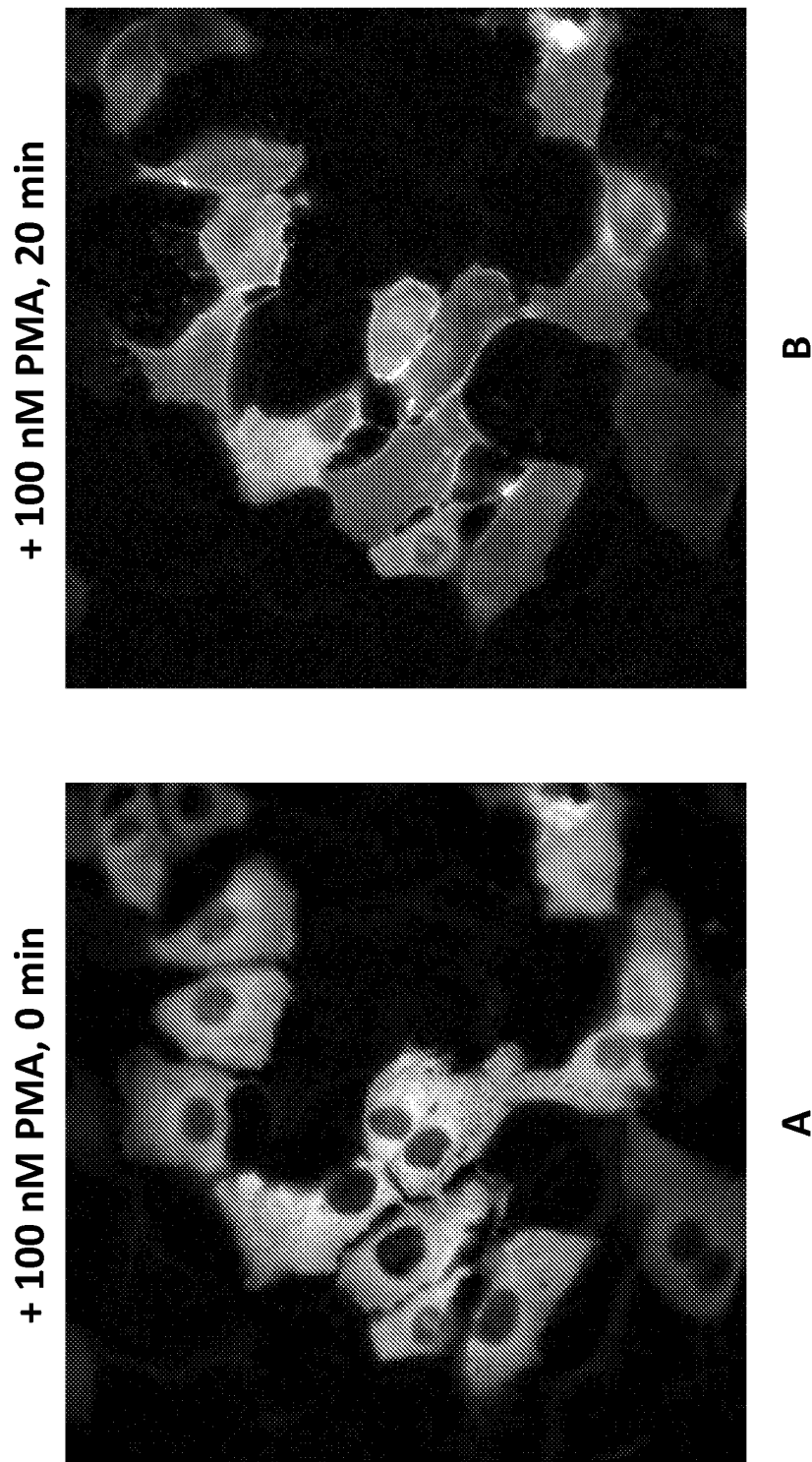


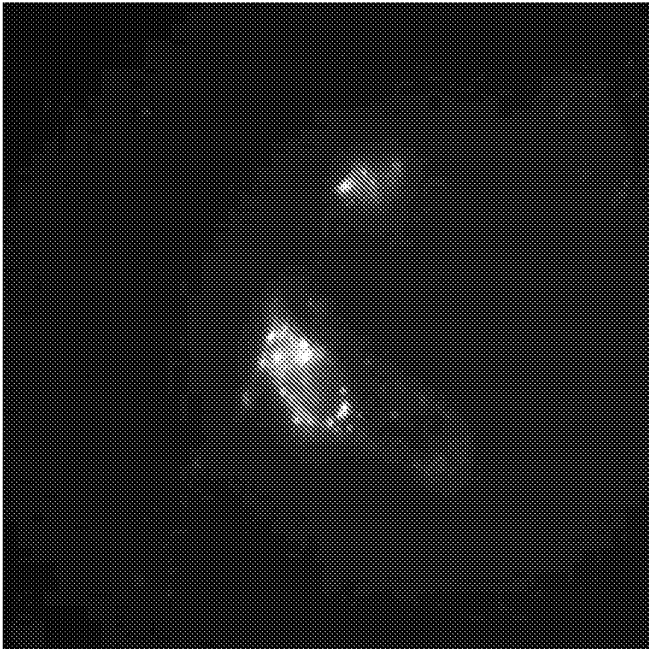
FIG. 91

untreated



A

+ 50  $\mu$ M Chloroquine, 16 h



B

FIG. 92