

Table S1. Physicochemical and anti-HBV activity of PAC5 derivatives

| HepG2.2.2.15 cells                     |          |                              |          |        |                                  |
|----------------------------------------|----------|------------------------------|----------|--------|----------------------------------|
| Compounds                              |          | Physicochemical <sup>a</sup> |          |        | IC <sub>50</sub> (μM)            |
|                                        |          | Mw                           | ClogP    | tPSA   | HBsAg                            |
| PA                                     | XM-1     | 746.71                       | -1.5694  | 312.05 | 14.96±0.5                        |
| 1a                                     | XM-2     | 438.43                       | 0.5356   | 162.98 | NA <sup>f</sup>                  |
| 1b                                     | XM-3     | 318.32                       | -2.213   | 136.68 | NA                               |
| 1c                                     | HY-9     | 776.47                       | 6.518    | 140.98 | NA                               |
| 1d                                     | HY-2     | 508.56                       | 3.086    | 107.98 | NA                               |
| 1e                                     | HY-3     | 388.46                       | 0.753    | 81.68  | NA                               |
| 2a                                     |          | 452.46                       | 0.98759  | 151.98 | NA                               |
| 2b                                     | HY-31    | 480.51                       | 2.0456   | 151.98 | 106.89±28.3                      |
| 2c                                     | HY-32    | 494.54                       | 2.5746   | 151.98 | 0.03±0.01                        |
| 2d                                     | HY-34    | 508.56                       | 3.1036   | 151.98 | 0.1±0.01                         |
| 2e                                     | HY-22    | 558.57                       | 2.25     | 140.98 | 139.8±3.7                        |
| 2f                                     | HY-36    | 586.63                       | 3.008    | 140.98 | 141±3.6                          |
| 2g                                     | HY-23    | 614.68                       | 4.066    | 140.98 | NA                               |
| 3a                                     | HY-25    | 465.5                        | 0.508599 | 154.78 | 155.52±1.1                       |
| 3b (PAC3)                              | HY-26    | 479.53                       | 1.0376   | 154.78 | 170.74±0.5                       |
| 3c                                     | HY-30    | 493.55                       | 1.5666   | 154.78 | 44.46±3.1                        |
| 3d (PAC5)                              | HY-16    | 507.58                       | 2.0956   | 154.78 | 0.81±0.9                         |
| 3e                                     | HY-4     | 521.61                       | 2.6246   | 154.78 | 15.97±21.77                      |
| 3f                                     | HY-15    | 535.63                       | 3.1536   | 154.78 | 8.42±0.9                         |
| 3g                                     | HY-7     | 549.66                       | 3.6826   | 154.78 | 16.91±9.05                       |
| 3h                                     | HY-5     | 523.58                       | 1.0616   | 164.01 | 31.47±7.07                       |
| 3i                                     | HY-17    | 541.6                        | 2.0766   | 154.78 | 13.91±0.9                        |
| 3j                                     | HY-10    | 601.65                       | 1.7346   | 173.24 | NA                               |
| 4a                                     | HY-1     | 565.62                       | 1.0546   | 181.08 | NA                               |
| 4b                                     | HY-14    | 593.67                       | 1.8926   | 181.08 | 49.21±6.6                        |
| 4c                                     | HY-13    | 607.7                        | 2.6416   | 181.08 | 70.13±14.7                       |
| 4d                                     | HY-39    | 621.72                       | 2.9506   | 181.08 | 3.83±0.5                         |
| 4e                                     | HY-40    | 635.75                       | 3.3496   | 181.08 | 70.42±3.8                        |
| 4f                                     | HY-27    | 593.67                       | 2.1126   | 181.08 | 169.79±1                         |
| 4g                                     | HY-24    | 621.72                       | 3.1706   | 181.08 | NA                               |
| 4h                                     | HY-33    | 593.67                       | 2.1126   | 181.08 | 23.11±2.5                        |
| 4i                                     | HY-21    | 607.7                        | 2.6416   | 181.08 | 133.52±2.6                       |
| 4j                                     | HY-28    | 621.72                       | 2.9506   | 181.08 | 134.58±0.6                       |
| 4k                                     | HY-29    | 635.75                       | 3.6996   | 181.08 | NA                               |
| 4l                                     | HY-37    | 635.75                       | 3.4796   | 181.08 | 95.37±10.6                       |
| 4m                                     | HY-38    | 649.78                       | 3.8786   | 181.08 | 81.88±3.7                        |
| 4n                                     | HY-12    | 678.78                       | 1.8508   | 210.18 | 118.508±18.4                     |
| 4o                                     | HY-18    | 664.75                       | 1.3218   | 210.18 | NA                               |
| 4p                                     | HY-19    | 740.85                       | 2.8698   | 210.18 | NA                               |
| 3AT                                    |          |                              |          |        | inhibition (%) 44.6±2.1 (100 μM) |
| Probe of PAC5                          | LJ1-26-1 |                              |          |        | 0.55±0.1                         |
| NC                                     | LJ1-66-1 |                              |          |        | 164.6±3.47                       |
| BC                                     | LJ1-76-1 |                              |          |        | 101.33±16.8                      |
| HepG2 cells (transfected PBSK-rtM204I) |          |                              |          |        |                                  |
| 3d                                     | HY-16    | 507.58                       | 2.0956   | 154.78 | 2.11±0.01                        |

|                      |                 |  |  |  |                   |
|----------------------|-----------------|--|--|--|-------------------|
| <b>Probe of PAC5</b> | <b>LJ1-26-1</b> |  |  |  | <b>0.374±0.01</b> |
| <b>NC</b>            | <b>LJ1-66-1</b> |  |  |  | <b>&gt;100</b>    |
| <b>BC</b>            | <b>LJ1-76-1</b> |  |  |  | <b>&gt;100</b>    |

<sup>a</sup> In silico calculations performed using Chemdraw 14.0 software. Molecule weight (Da), ClogP, %PSA = % polar surface area

<sup>b</sup> Lamivudine (3AT) was tested as the positive control of anti-HBV;

<sup>c</sup> Compound concentration inhibiting HBsAg or HBeAg;

<sup>d</sup> Compound concentration reducing the viability of HepG2. cells culture by 50%;

<sup>e</sup> Selective index, SI = CC<sub>50</sub>/IC<sub>50</sub>

<sup>f</sup> Not active below 200 μM.

<sup>g</sup> No tested.

| 1) <sup>c</sup>         | CC <sub>50</sub> (μ M) <sup>d</sup> | SI <sup>e</sup> |
|-------------------------|-------------------------------------|-----------------|
| HBeAg                   |                                     |                 |
| 95.99±27.2              | 19.2                                | 1.5             |
| NA                      | NT <sup>g</sup>                     | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| 131.28±29.5             | NT                                  | -               |
| 83.06±6.28              | > 30                                | >1000           |
| 106.45±36.9             | > 30                                | >300            |
| NA                      | > 30                                | >0.21           |
| 51.22±9.2               | > 30                                | >0.21           |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| 175.9±9.86              | NT                                  | -               |
| 5.88±0.8                | > 900                               | >1100           |
| 65.47±10.21             | > 30                                | >1.88           |
| 33.09±0.4               | > 30                                | >3.56           |
| 24.68±0.4               | > 30                                | >1.77           |
| 277.2±4.68              | NT                                  | -               |
| 82.19±5.7               | > 30                                | >2.15           |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | > 30                                | >0.61           |
| NA                      | > 30                                | >0.43           |
| 51.99±5.1               | > 30                                |                 |
| 102.6±7.07              | NT                                  | -               |
| NA                      | > 30                                | >0.18           |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| 97.26±19.6              | NT                                  | -               |
| 65.68±5.6               | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| inhibition (%) 26.0±9.6 | NT                                  | -               |
| 0.97 ±0.01              | NT                                  | -               |
| NA                      | NT                                  | -               |
| NA                      | NT                                  | -               |
| 2.64±3.8                | NT                                  | -               |

|              |    |   |
|--------------|----|---|
| 21.645±4.413 | NT | - |
| NA           | NT | - |
| NA           | NT | - |

surface area;

