# Supplement

**Supplement method:**

*The conduction of Fisher exact test.* The intracellular concentrations were calculated by adding the uptake results in 3 OATP isoforms (OATP1B1, 1B3 and 2B1) together with weighting. The intracellular concentrations were calculated as the equation presented below:

Total intracellular concentration =

X/10*OATP1B1 concnetraion+Y/10*OATP1B3 concentration+Z/10* OATP2B1 concentration

Concentrations are intracellular concentration of phenolic glucuronide in 3 cell lines generated from cell uptake experiment. X, Y, Z are integers and are weightings to individual isoform, which follows the description below:

X+Y+Z=10, 0≤X≤10, 0≤Y≤10, 0≤Z≤10.

To conduct Fisher exact test, all the compounds were classified into 4 groups by their recycle ratio (high and low) and intracellular concentration (high and low). Recycle ratio higher than 50% is classified into high group and recycle ratio lower than 50% is classified into low group. Total intracellular concentration higher than average concentration of all the compounds is classified into high group and lower than average concentration of all the compounds is classified into low group. The number of substrates that belongs to the classification classified groups (e.g., high recycle and high uptake, high

recycle and low uptake) was counted. A 2*2 table was generated based on the classification of all the tested glucuronides. Then Fisher exact test was applied to test the statistically significant correlation of recycle ratio and intracellular concentration. $p < 0.05$ was considered of significant correlation. Different X, Y, Z weighting combinations were tested and individual p values were calculated by Fisher exact test.
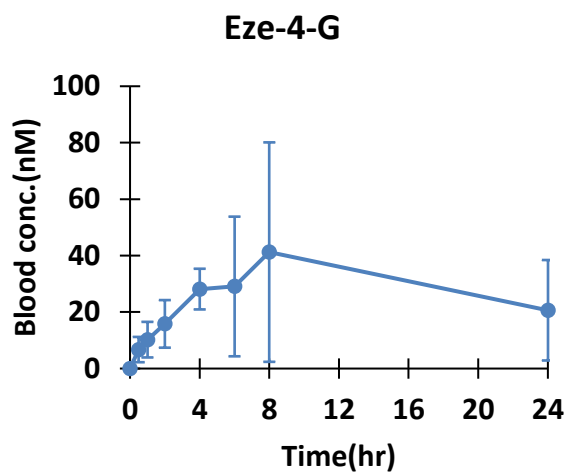
## Supplement Figures

A



Won-7-G

B



Bai-7-G

**C**



Eze-4-G

**D**



Ace-G
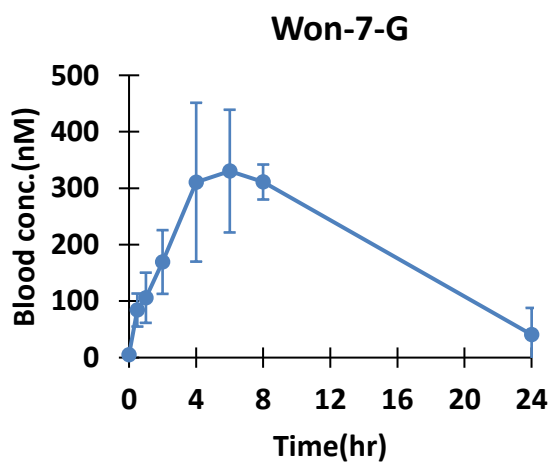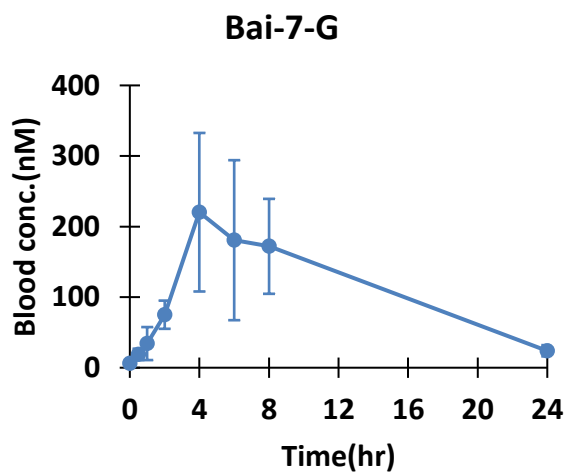
Fig. S1 The blood concentraiton-time curves of single PK study of wogonin/baicalein and ezetemibe/acetaminophene. The blood concentration of wogonin-7-glucuronide(A), baicalein-7-glucuronide(B), ezetemibe-4'-glucuronide(C) and acetaminophene-glucuronide(D) were summarized in Fig.S1.

Fig.S2 The elimination-half life of 4 glucuronides were correlated with their corresponding recycle ratios.
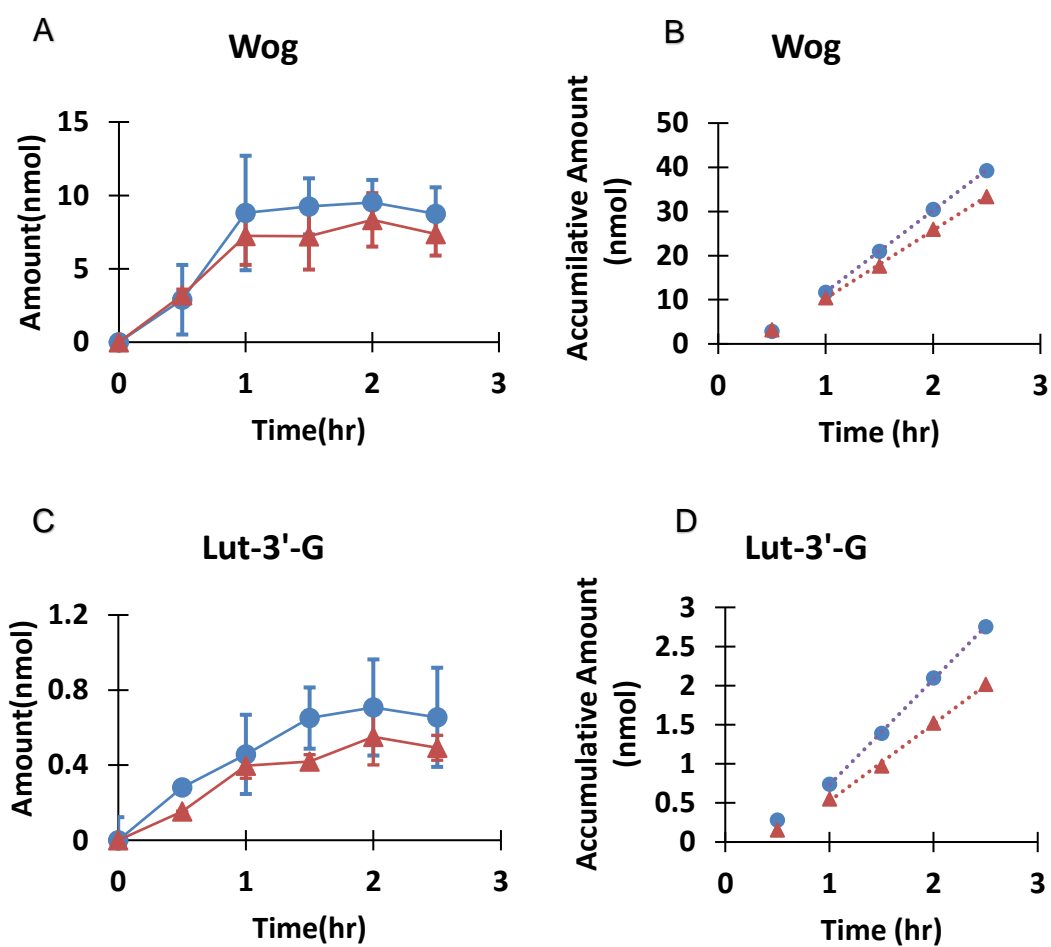
Fig. S3 The bile secretion amount (A) and accumulative bile secretionmount(B) of wogonin-7-glucuronide, the bile secretion amount (C) and accumulative bile secretionmount(D) of luteolin-3'-glucuronide were summarized in Fig.S3.
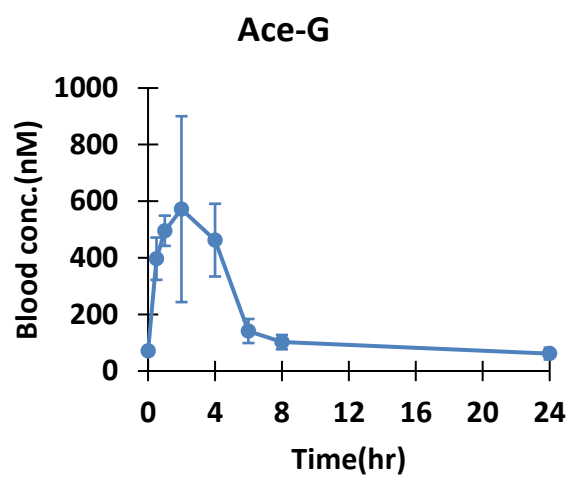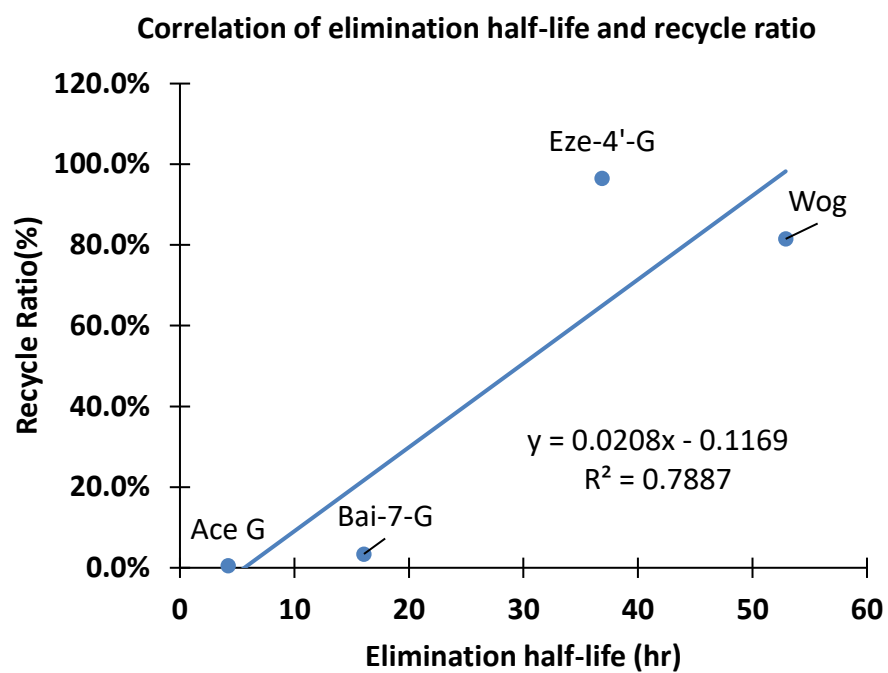
A



Won

B

CC

Eze

C

**Ace**



D

**Bai**
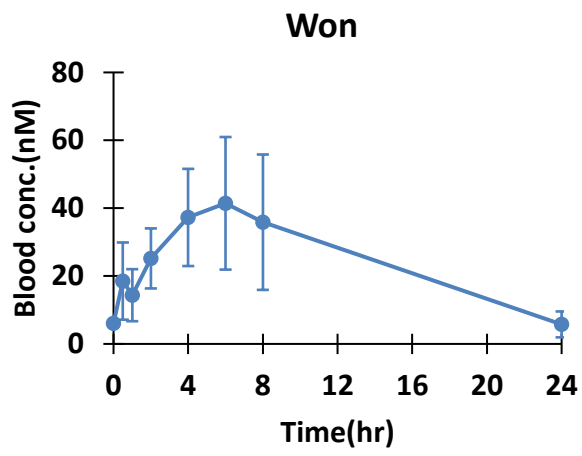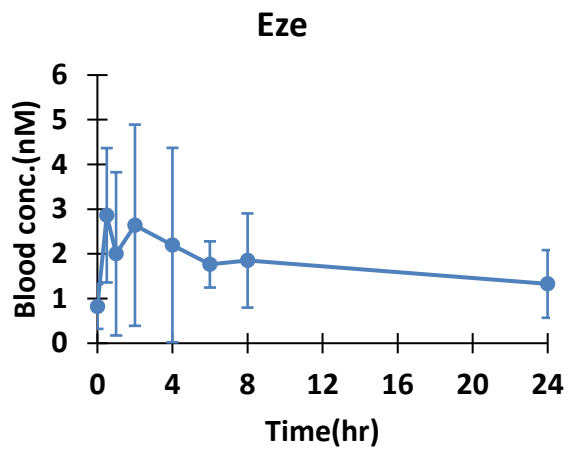


Fig. S4 The blood concentraiton-time curves of single PK study of wogonin/baicalein and ezetemibe/acetaminophene. The blood concentration of wogonin (A), baicalein (B), ezetemibe (C) and acetaminophene (D) were summarized in Fig.S4

**Supplement Tables:**

Table S1. Biliary glucuronide secretion rate and recycle ratio of 16 different glucuronides

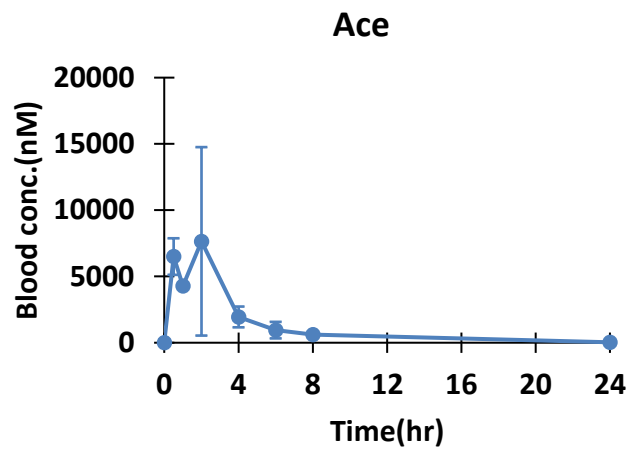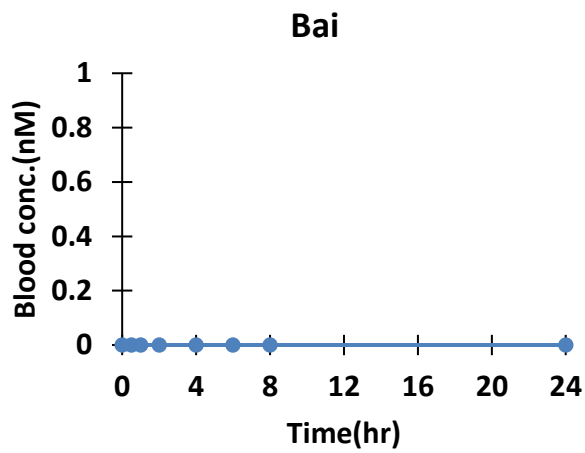| Infused compound (10uM) | Secretion rate (nmol/hr) | Recycle ratio (%) |
|---|---|---|
| Ezetimibe-4'-G | 19.31±1.85 | 96.5±9.2 |
| Wogonin-7-G(wogonoside) | 16.3±4.28 | 81.5±21.4 |
| Genestein-7-G | 9.78[c] | 59.3[c] |
| Raloxifene-4'-G | 11.48±2.32 | 57.4±11.6 |
| Apigenin-7-G | 10.64±4.49 | 53.2±22.4 |
| Raloxifene-6-G | 10.53±1.51 | 52.6±7.5 |
| Chrysin-7-G | 9.86[c] | 49.3[c] |
| Icaritin-7-G | 7.46±0.72 | 37.3±3.6 |
| Biochanin A-G | 3.28[c] | 27.8[c] |
| Icaritin-3-G | 5.35±1.12 | 26.8±5.6 |
| Scuttelarin | 4.34±1.18 | 21.7±5.9 |
| Luteolin-3'-G | 1.33±0.34 | 6.7±1.7 |
| Baicalin | 0.69±0.29 | 3.4±1.5 |
| Luteolin-7-glycoside | 0.67±0.56 | 3.4±2.8 |
| Quercetin-3-G | 0.43±0.57 | 2.2±2.8 |
| Ace-G | 0.10±0.06 | 0.5±0.3 |

a The glucuronide secretion rate at the steady-state was calculated by the linear regression of accumulative amount secreted in bile vs. time curve

b.

$$RR \text{ (Recycle ratio) } \% = \frac{\text{Steady-state biliary glucuronide excretion rate}}{\text{Portal vein infusion rate}}$$

c. Data from previous study [55].

Table S2. The p values of different weighting assigned to 3 isoforms as combinations were summarized in table S1. 41 out of 66 p values are smaller than 0.05 and considered of significant correlation.

| Weighting | | | P value | Weighting | | | P value |
|---|---|---|---|---|---|---|---|
| 1B1 | 1B3 | 2B1 | | 1B1 | 1B3 | 2B1 | |
| 10 | 0 | 0 | 0.0476 | 3 | 2 | 5 | 0.0079 |
| 9 | 1 | 0 | 0.0476 | 3 | 1 | 6 | 0.0079 |
| 9 | 0 | 1 | 0.0476 | 3 | 0 | 7 | 0.0079 |
| 8 | 2 | 0 | 0.0476 | 2 | 8 | 0 | 0.1667 |
| 8 | 0 | 2 | 0.0476 | 2 | 7 | 1 | 0.1667 |
| 8 | 1 | 1 | 0.0476 | 2 | 6 | 2 | 0.1667 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 7 | 3 | 0 | 0.0476 | 2 | 5 | 3 | 0.1667 |
| 7 | 2 | 1 | 0.0476 | 2 | 4 | 4 | 0.1667 |
| 7 | 1 | 2 | 0.0476 | 2 | 3 | 5 | 0.0079 |
| 7 | 0 | 3 | 0.0476 | 2 | 2 | 6 | 0.0079 |
| 6 | 4 | 0 | 0.0476 | 2 | 1 | 7 | 0.0079 |
| 6 | 3 | 1 | 0.0476 | 2 | 0 | 8 | 0.0079 |
| 6 | 2 | 2 | 0.0476 | 1 | 9 | 0 | 0.1667 |
| 6 | 1 | 3 | 0.0476 | 1 | 8 | 1 | 0.1667 |
| 6 | 0 | 4 | 0.0079 | 1 | 7 | 2 | 0.1667 |
| 5 | 5 | 0 | 0.1667 | 1 | 6 | 3 | 0.1667 |
| 5 | 4 | 1 | 0.0476 | 1 | 5 | 4 | 0.1667 |
| 5 | 3 | 2 | 0.0476 | 1 | 4 | 5 | 0.1667 |
| 5 | 2 | 3 | 0.0476 | 1 | 3 | 6 | 0.0476 |
| 5 | 1 | 4 | 0.0079 | 1 | 2 | 7 | 0.0476 |
| 5 | 0 | 5 | 0.0079 | 1 | 1 | 8 | 0.0476 |
| 4 | 6 | 0 | 0.1667 | 1 | 0 | 9 | 0.0476 |
| 4 | 5 | 1 | 0.1667 | 0 | 10 | 0 | 0.1667 |
| 4 | 4 | 2 | 0.0476 | 0 | 9 | 1 | 0.1667 |
| 4 | 3 | 3 | 0.0476 | 0 | 8 | 2 | 0.1667 |
| 4 | 2 | 4 | 0.0476 | 0 | 7 | 3 | 0.1667 |
| 4 | 1 | 5 | 0.0079 | 0 | 6 | 4 | 0.1667 |
| 4 | 0 | 6 | 0.0079 | 0 | 5 | 5 | 0.1667 |
| 3 | 7 | 0 | 0.1667 | 0 | 4 | 6 | 0.1667 |
| 3 | 6 | 1 | 0.1667 | 0 | 3 | 7 | 0.0476 |
| 3 | 5 | 2 | 0.1667 | 0 | 2 | 8 | 0.0476 |
| 3 | 4 | 3 | 0.1667 | 0 | 1 | 9 | 0.0476 |
| 3 | 3 | 4 | 0.0476 | 0 | 0 | 10 | 0.0476 |

Table. S3 The hepatic expression level of OATP 1B1/1B3/2B1

| Transporter | OATP 1B1 | OATP 1B3 | OATP 2B1 |
|---|---|---|---|
| Expression (RPKM) | 119.3[a] | 30.2[b] | 44.0[c] |
| Relative expression (%) | 61.7 | 15.6 | 22.7 |

a https://www.ncbi.nlm.nih.gov/gene/10599

b https://www.ncbi.nlm.nih.gov/gene/28234

c https://www.ncbi.nlm.nih.gov/gene/11309

Table S4. Gender differences in bile secretion rates and recycle ratios

| Infusion compounds (10uM) | Secretion rate (nmol/hr) | | Recycle Ratio (%) | |
|---|---|---|---|---|
| | Male | Female | Male | Female |
| Won-7-G | 16.30±4.28 | 15.06±0.70 | 81.5±21.4 | 75.3±3.5 |
| Bai-7-G | 0.69±0.29 | 1.78±0.42 | 3.4±1.5 | 8.9±2.1 |
| Lut-3'-G | 1.33±0.34 | 1.67±0.32 | 6.7±1.7 | 8.3±1.6 |