Supporting Information

Metabolism of gartanin in liver microsomes and its modulating effects on cytochrome P450s

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Parent drug (M0)

The retention time (11.20 min), $[M-H]^-$ and MS/MS spectra of the molecular ion at *m/z* 395.0658 (M0) were the same as those of the gartanin standard. Therefore, M0 was confirmed as the unchanged parent drug. Gartanin gave the characteristic fragment ions at *m/z* 351.0123, 339.0141, and 296.9763. The cleavage of gartanin at C-22 and C-17 produced a daughter ion at *m/z* 351, which created further fragment ions at *m/z* 339.0141. However, the structure of the fragment ion at *m/z* 296.9763 was not inferred.

M1 and M2

The molecular formulas of M1 (t_R = 9.42 min) and M2 (t_R = 10.21 min) were determined as $C_{23}H_{24}O_7$ with the $[M-H]^-$ at *m/z* 411.0722 and 411.0719, respectively. Their *m/z* was 16 Da higher than that of gartanin, indicating that they were the hydroxylated metabolite of the parent drug. The hydroxyl group was added to the parent molecule at C-18(19) or C-22(23). Based on the MS/MS spectra of M1 and M2, two side chains underwent a series of cleavage at C-22, C-21, and C-17 and generated the product ion at *m/z* 381.0663, 351.0247, and 337.0116. The m/z 351 fragment was the same as gartanin, due to cleavage of the isoprenyl group, indicating the new

hydroxyl should be substituted on a terminal isoprenyl methyl group [1-3].

M3

Metabolite M3 ($C_{23}H_{24}O_8$) with $[M-H]^-$ at *m/z* 427.0648 was 32 Da more than M0 and was inferred as two hydroxylated metabolites of the parent compound gartanin. Two hydroxyl groups were bound to C-23(24) and C-18(19) according to the M3 information of fragment ions. Two side chains formed a series of cleavages and produced the daughter ions at *m/z* 397.0592, 379.0516, and 351.0247.

M4

Metabolite M4 had a $[M-H]^-$ ion at *m/z* 429.0787 that was 34 Da higher than M0 and was inferred as the double hydroxylation and reduction product of M0 according to the MS/MS spectra. M4 had the same fragment ion at *m/z* 296 with the parent drug, suggesting that two hydroxyl groups were added into the side chain of the parent molecule.

M5

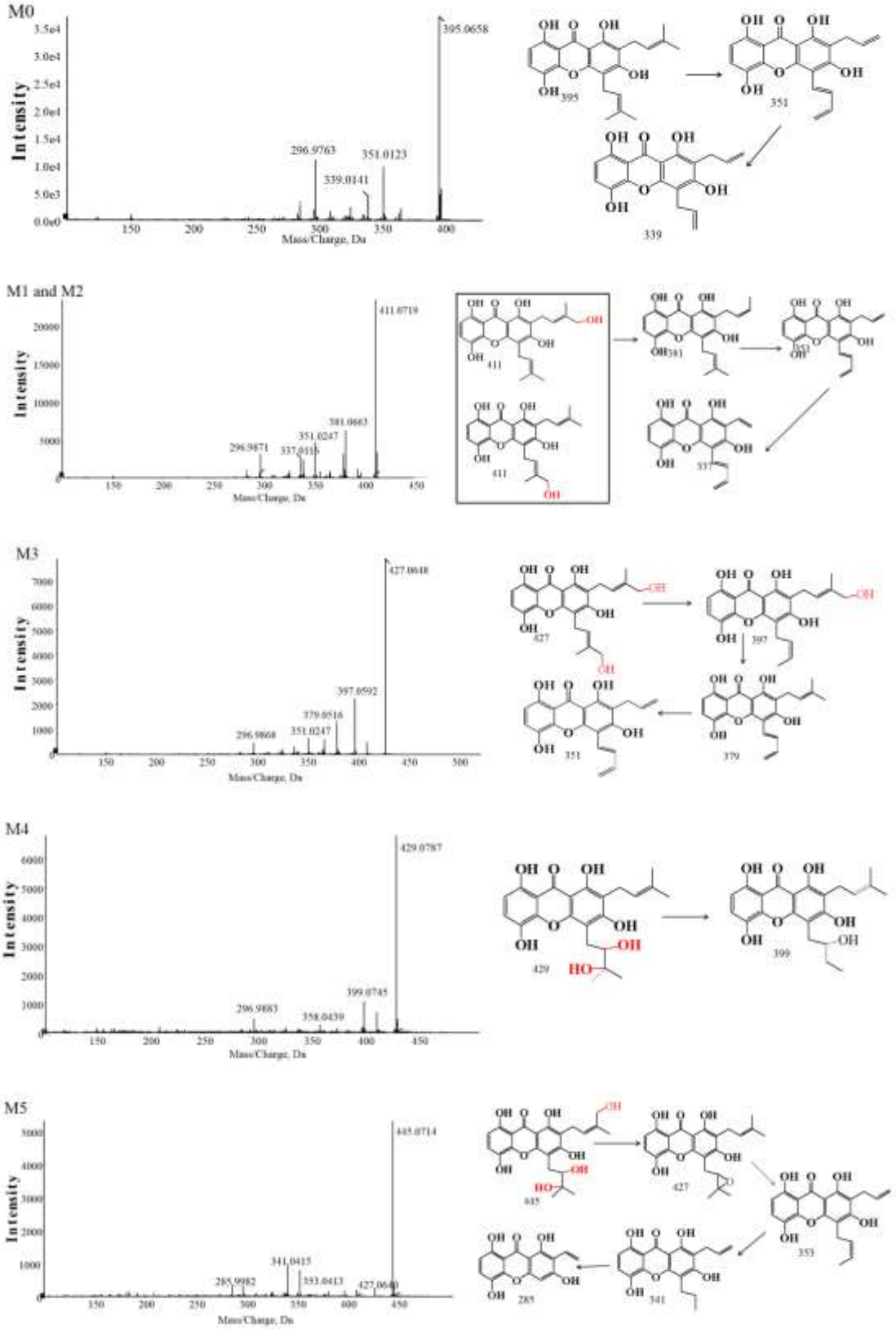
Metabolite M5 with $[M-H]^-$ at *m/z* 445.0714 was 16 Da higher than M4, suggesting that M5 was the oxidation product of M4, and the oxidation reaction occurred at C-23(24). Two side chains appeared at a series of cleavages and produced the fragment ions at *m/z* 427.0640, 353.0413, 341.0415, and 285.9982, according to the information of the MS/MS spectra.

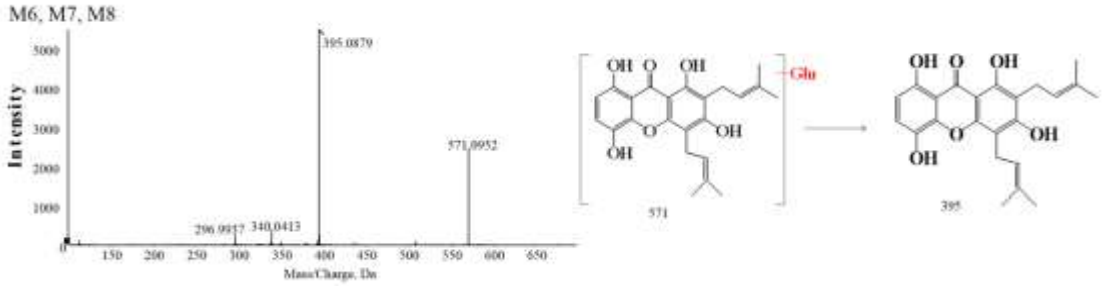
M6, M7, and M8

M6, M7, and M8 were isomers with the same $[M-H]^-$ ions at *m/z* 571. The molecular formula of M6, M7, and M8 was determined as $C_{29}H_{32}O_{12}$ and their retention times were 7.64, 9.06, and 9.76, respectively. M6, M7, and M8 had an *m/z* of 176 Da more than M0, and their main fragment ion at *m/z* 395 was originated by the loss of 176 Da (glucuronic acid residue), indicating they were glucuronidation metabolites of

gartanin.

Figure S1. Fragmentation pathway of gartanin (M_0) and its metabolites (M_1-M_8).





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