Supplementary information

Investigation of the preparation, characterization, and whitening activity of glabridin and oxymatrine co-amorphous

Peiyi Liang[#a], Yi Hu[#a], Zhuxian Wang[a], Yinglin Guo[a], Hongkai Chen[a], CuiPing Jiang[a], Quanfu Zeng[a], Chunyan Shen[a], Yufan Wu[a], Li Liu[a], Yankui Yi[a], Hongxia Zhu[b], Zhijun Yang*[c], Qiang Liu*[a]*

[a] *School of Traditional Chinese Medicine, Southern Medical University, Guangzhou 510515, PR China*

[b] *Integrated Hospital of Traditional Chinese Medicine, Southern Medical University, Guangzhou 510315, PR China*

[c] *School of Chinese Medicine, Hong Kong Baptist University, Hong Kong 999077, PR China*

[#]*Peiyi Liang and Yi Hu contributed equally to this work.*

[*]**Corresponding author:** School of Traditional Chinese Medicine, Southern Medical University, Guangzhou, 510515, China. Tel.: +86-20-6164-8264, (Qiang Liu) *Email-address*: liuqiang@smu.edu.cn

[*]**Corresponding author:** School of Chinese Medicine, Hong Kong Baptist University, Hong Kong 999077, China. Tel.: +852-3411-2961, (Zhijun Yang) *Email- address*: yzhijun@hkbu.edu.hk

[*]**Corresponding author:** Integrated Hospital of Traditional Chinese Medicine, Southern Medical University, Guangzhou, 510315, China. Tel.: +86-20-6278-9408, (Hongxia Zhu) *Email-address*: gzzhx2012@163.com

Table S1 The pH change of Gla, physical mixture of Gla-OMT and co-amorphous Gla-OMT before and after dissolution.

buffer solution	Simples	Initial pH	pH after dissolution
1.0	Gla	1.01	1.03
	Physical mixture of Gla-OMT		1.02
	Co-amorphous Gla-OMT		1.04
4.0	Gla	4.02	4.03
	Physical mixture of Gla-OMT		4.07
	Co-amorphous Gla-OMT		4.09
6.8	Gla	6.82	6.83
	Physical mixture of Gla-OMT		6.87
	Co-amorphous Gla-OMT		6.86
Water	Gla	5.34	5.84
	Physical mixture of Gla-OMT		6.87
	Co-amorphous Gla-OMT		6.88

Table S2 Molecular docking scores of tyrosinase with Gly and OMT.

Compound	**Protein (RCSB Protein Data Bank)**	**Libdock score**
Gla	Tyrosinase（PDB:6EI4）	124.00
OMT	Tyrosinase（PDB:6EI4）	100.37

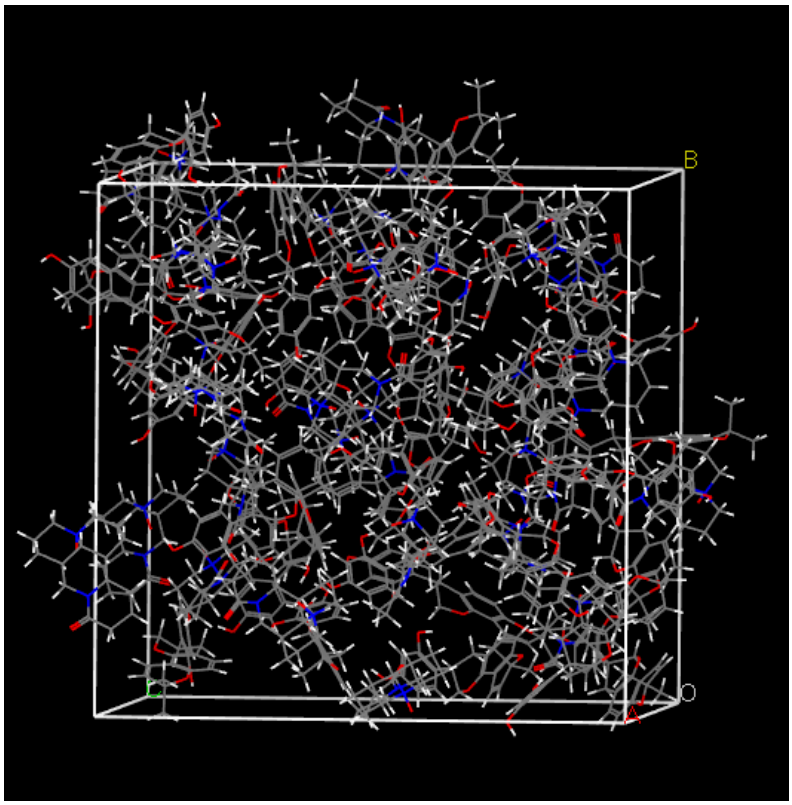

Figure S1 AC model of co-amorphous Gla-OMT.

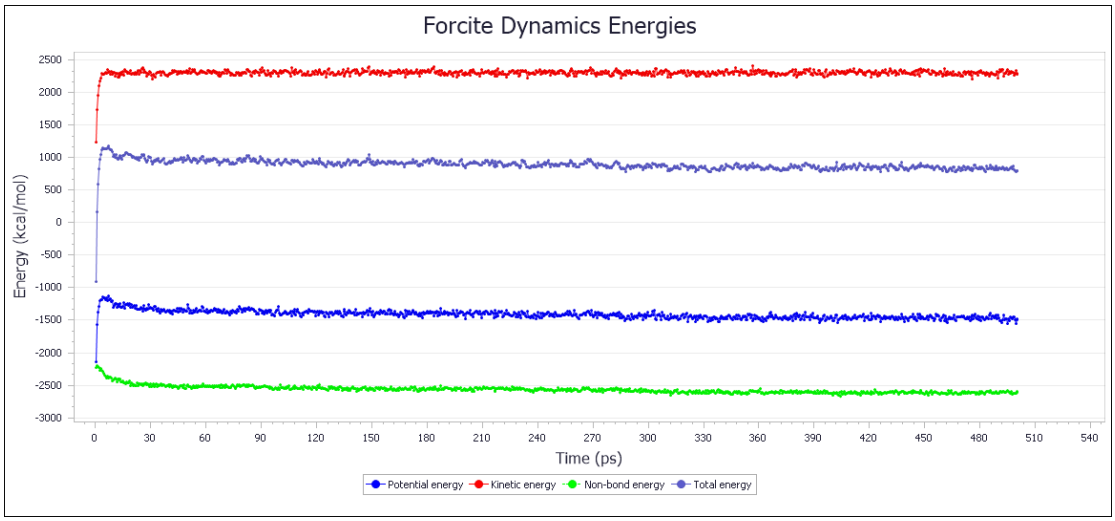

Figure S2 Density as a function of time during the optimization of NPT kinetics for the Gla-OMT coamorphous model.

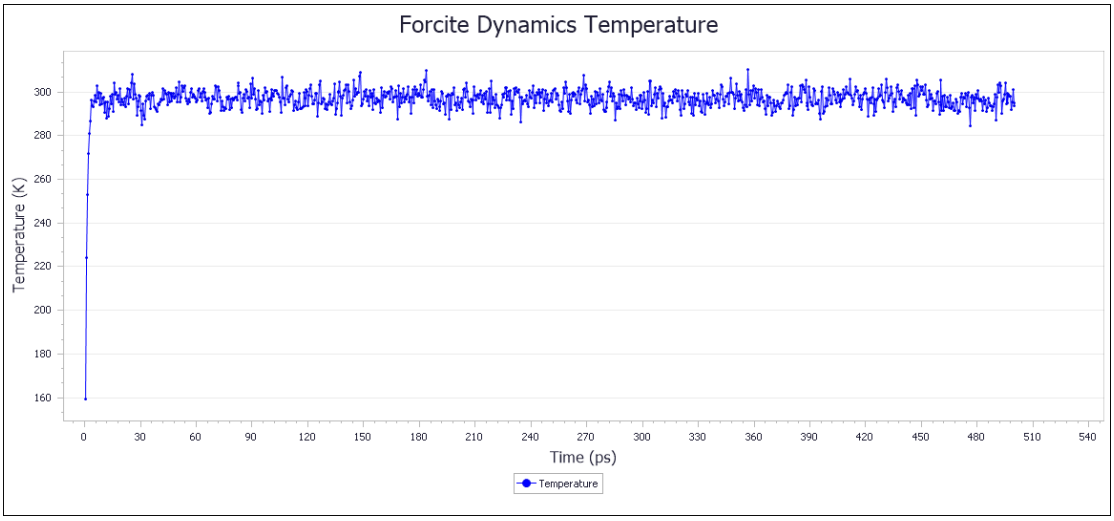

Figure S3 Energy as a function of time during the optimization of NPT kinetics for the Gla-OMT coamorphous model.

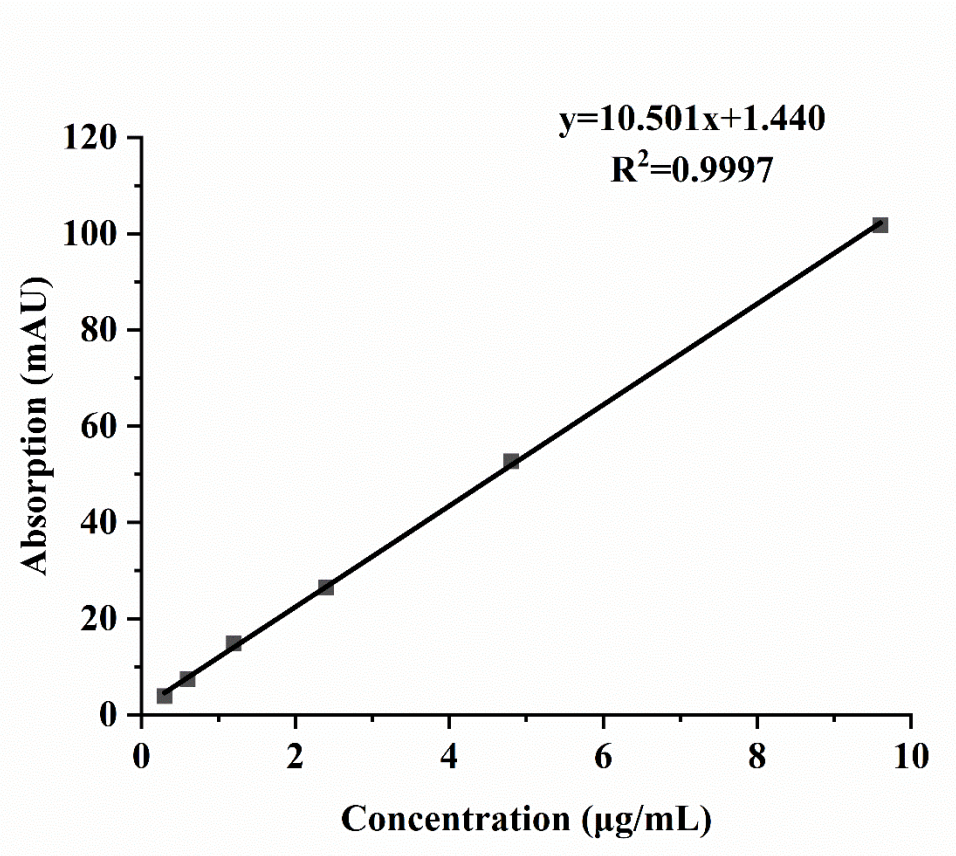

Figure S4 Glabridin standard curve.

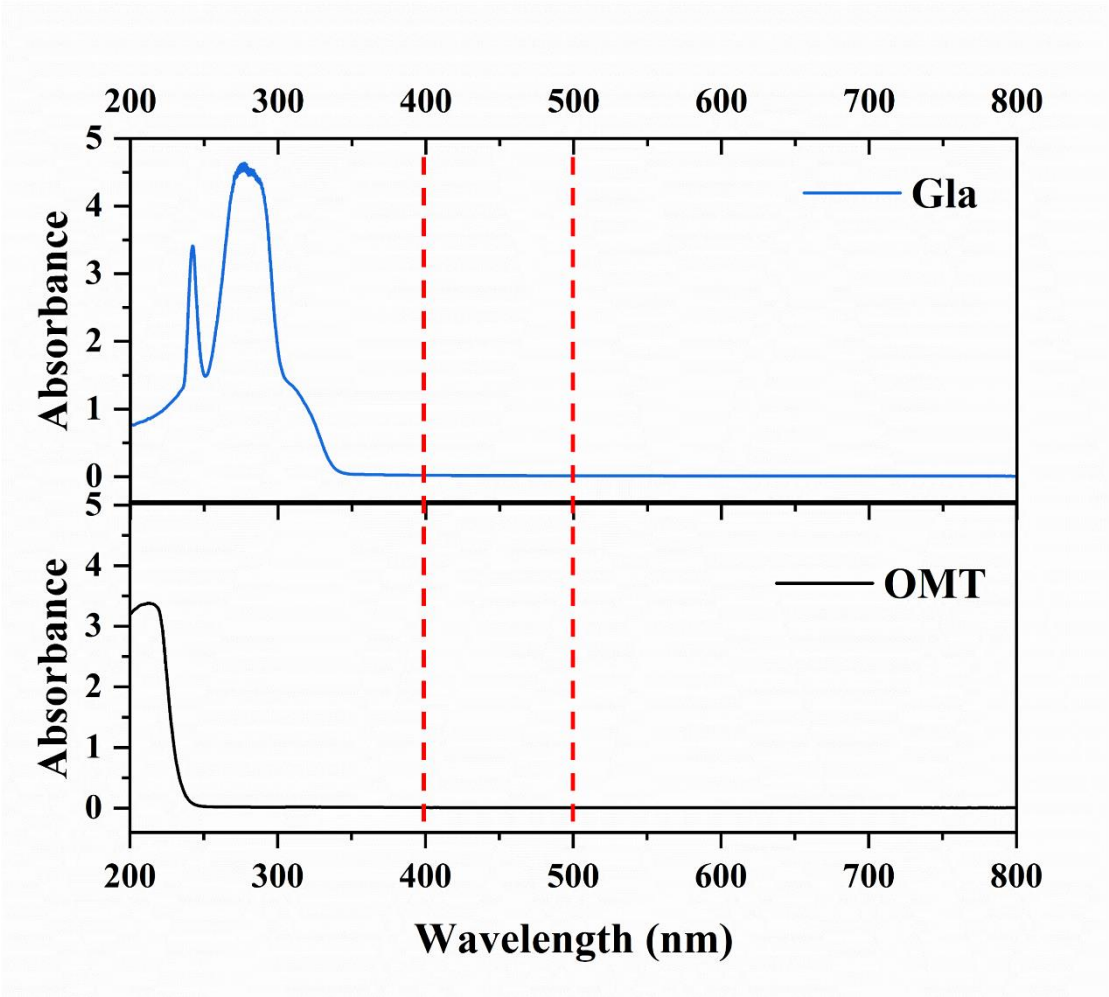

Figure S5 The UV-Vis absorption spectra of Gla and OMT in the range of 200-800 nm.

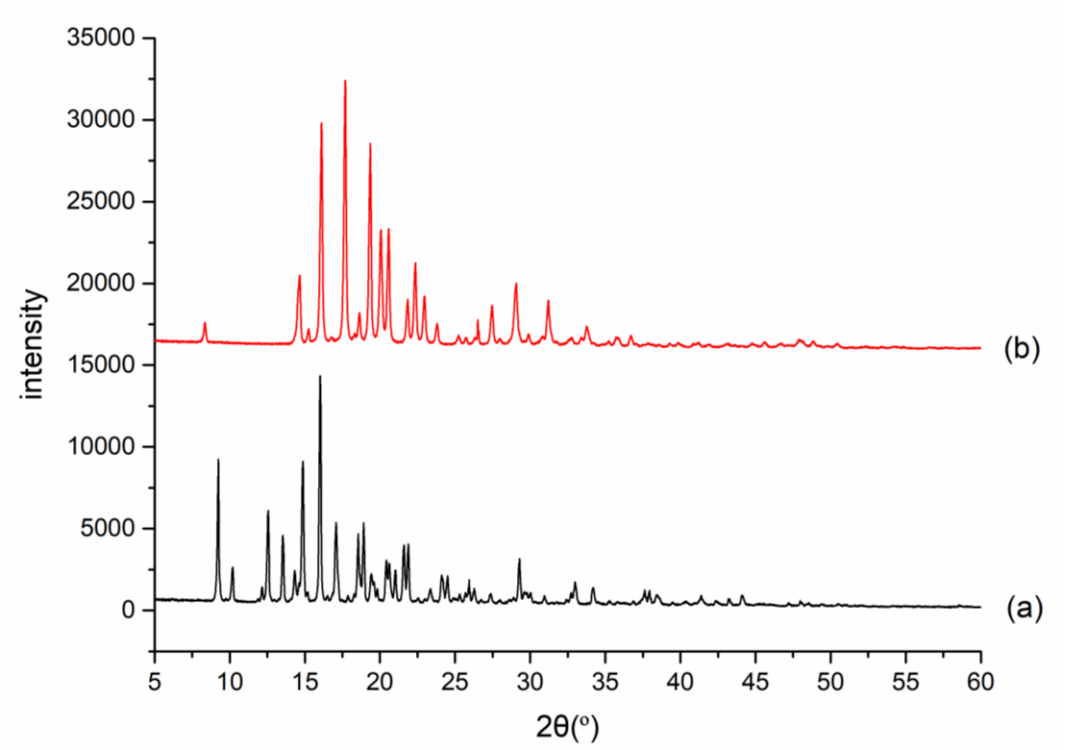

Figure S6 PXRD patterns of OMT evaporation products (a) and Gla evaporation products (b).

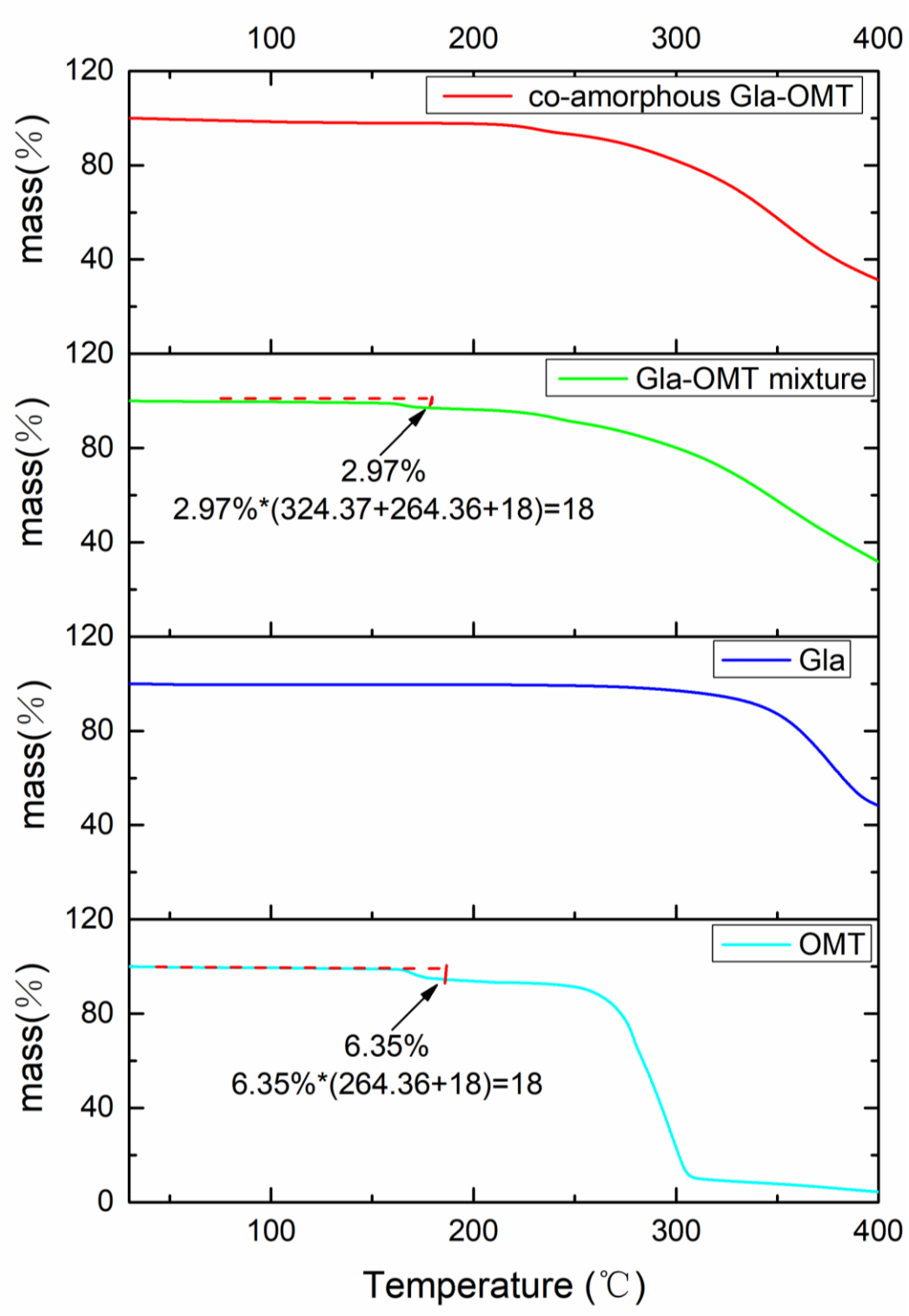

Figure S7 The TG thermograms of OMT, Gla, physical mixture of Gla-OMT, and co-amorphous Gla- OMT.

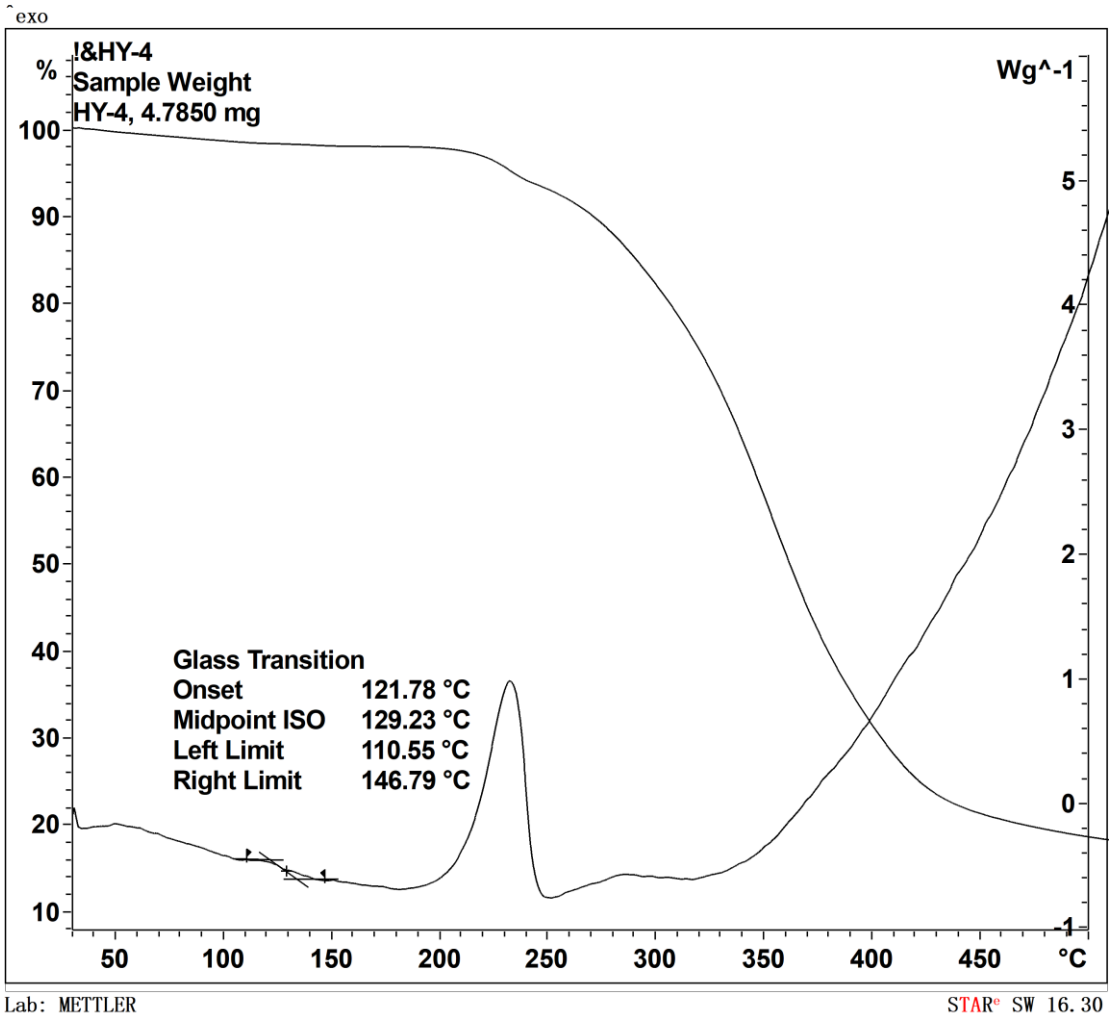

Figure S8 The TG-DSC thermograms of co-amorphous Gla- OMT.

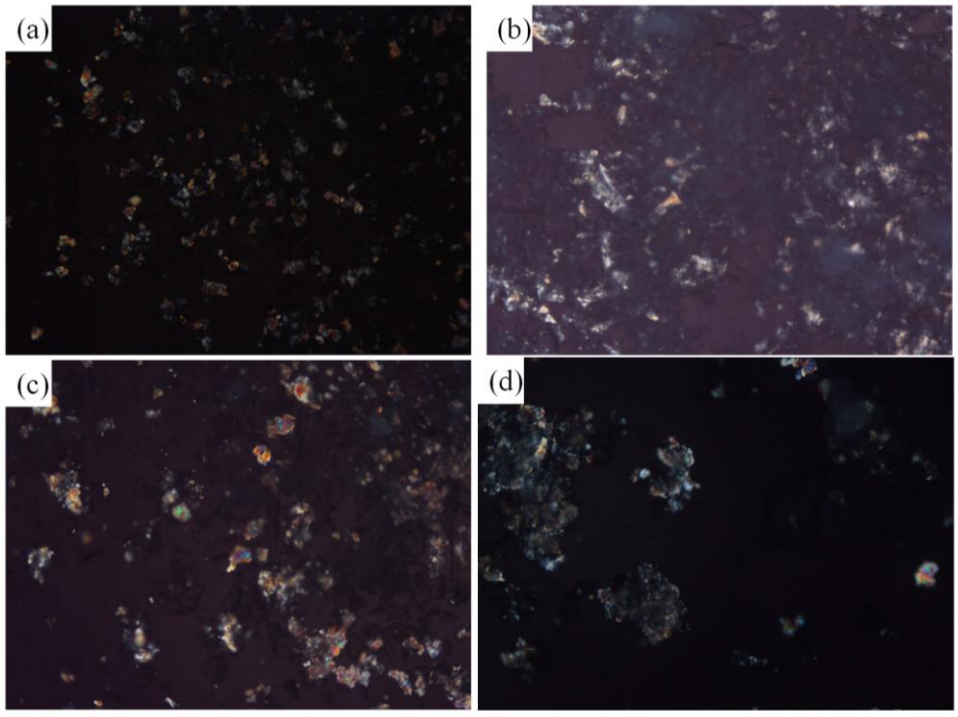

Figure S9 Polarized light microscope patterns of co-amorphous Gla-OMT after dissolving for 24h at pH 1.0 (a), 4.0 (b), 6.8 (c), and water (d).

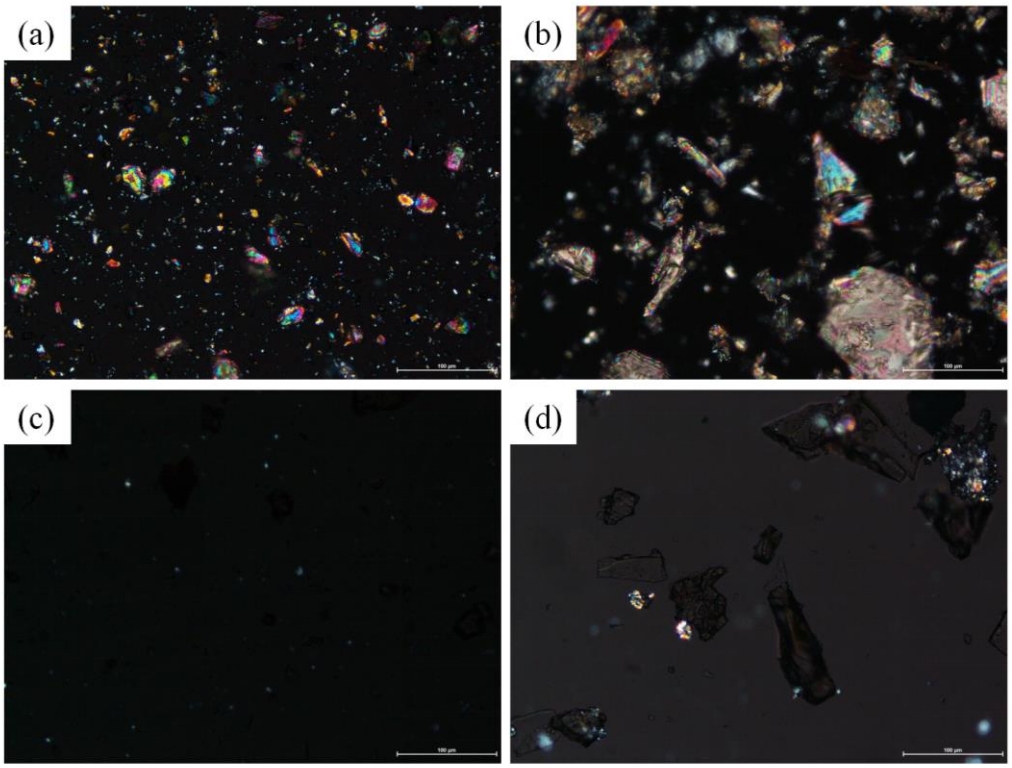

Figure S10 The polarizing microscope patterns of crystalline Gla (a), crystalline OMT (b), and co-amorphous Gla-OMT (c), as well as the polarizing microscope pattern of co-amorphous Gla-OMT after 24 hours of dissolution (d).

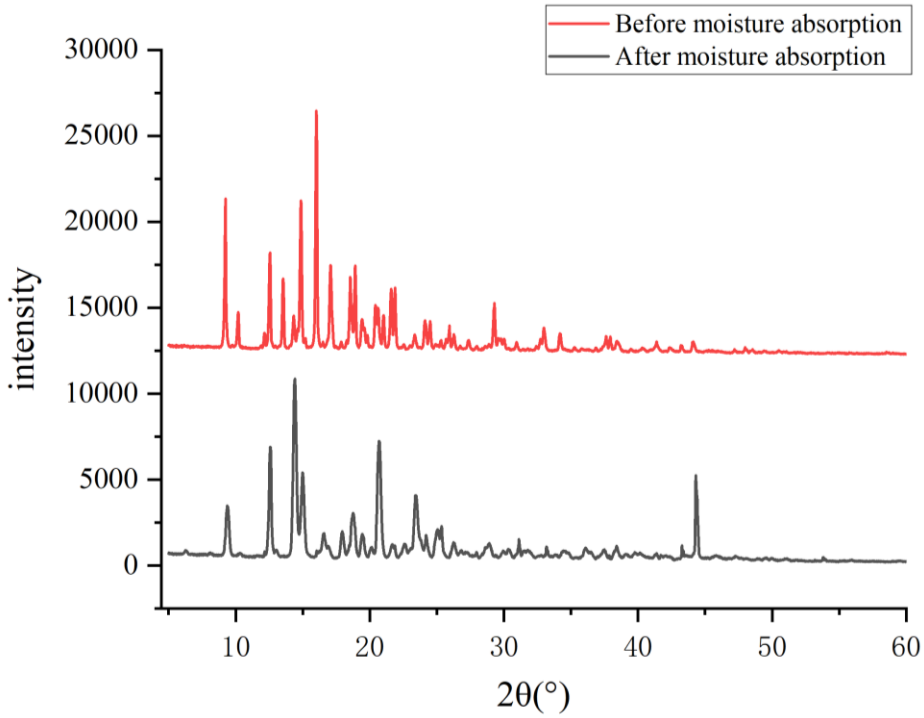

Figure S11 PXRD pattern of crystalline OMT before and after moisture absorption.

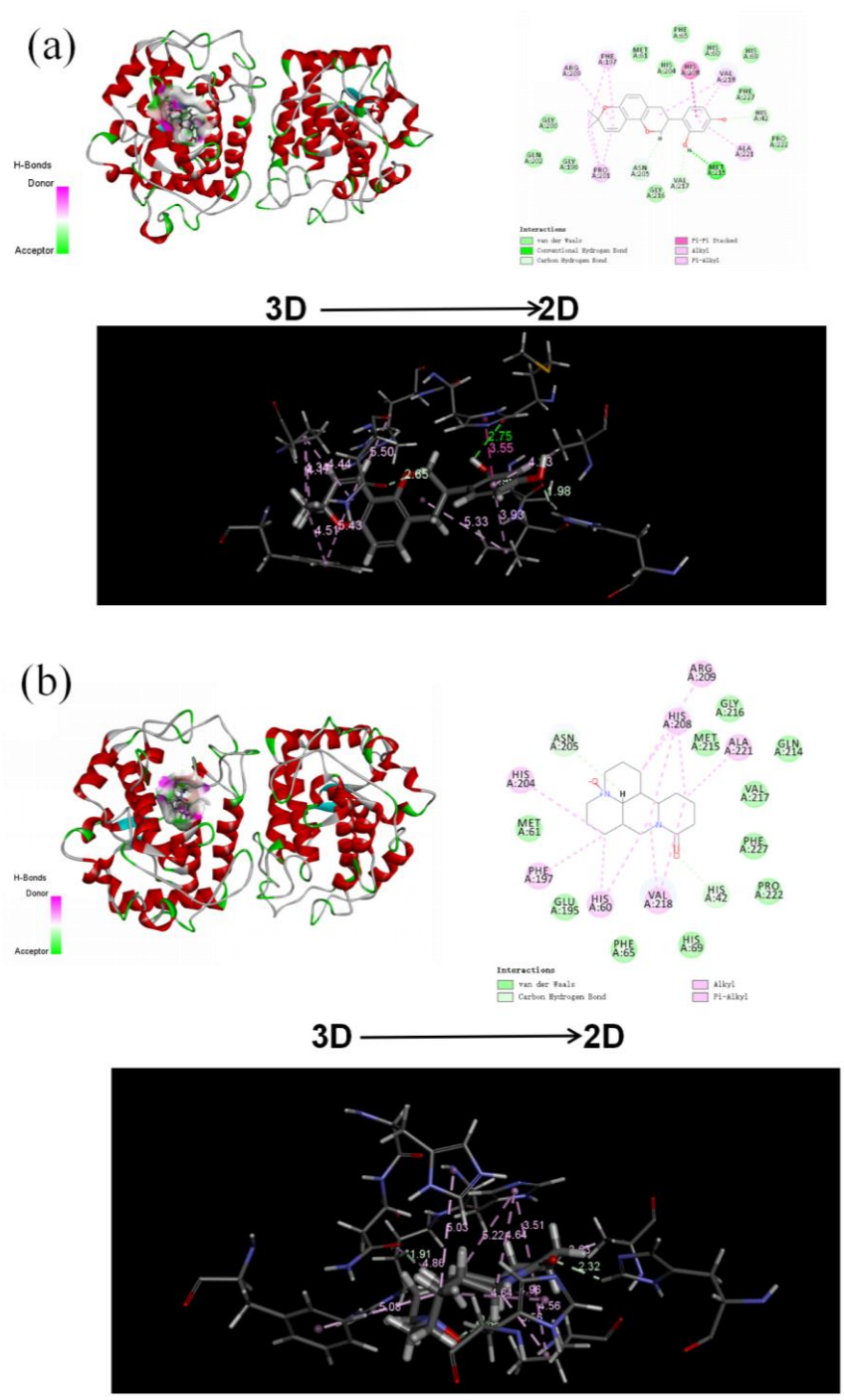

Figure S12 Molecular docking of tyrosinase (PDB: PDB:6EI4) with Gla (a) and OMT (b).

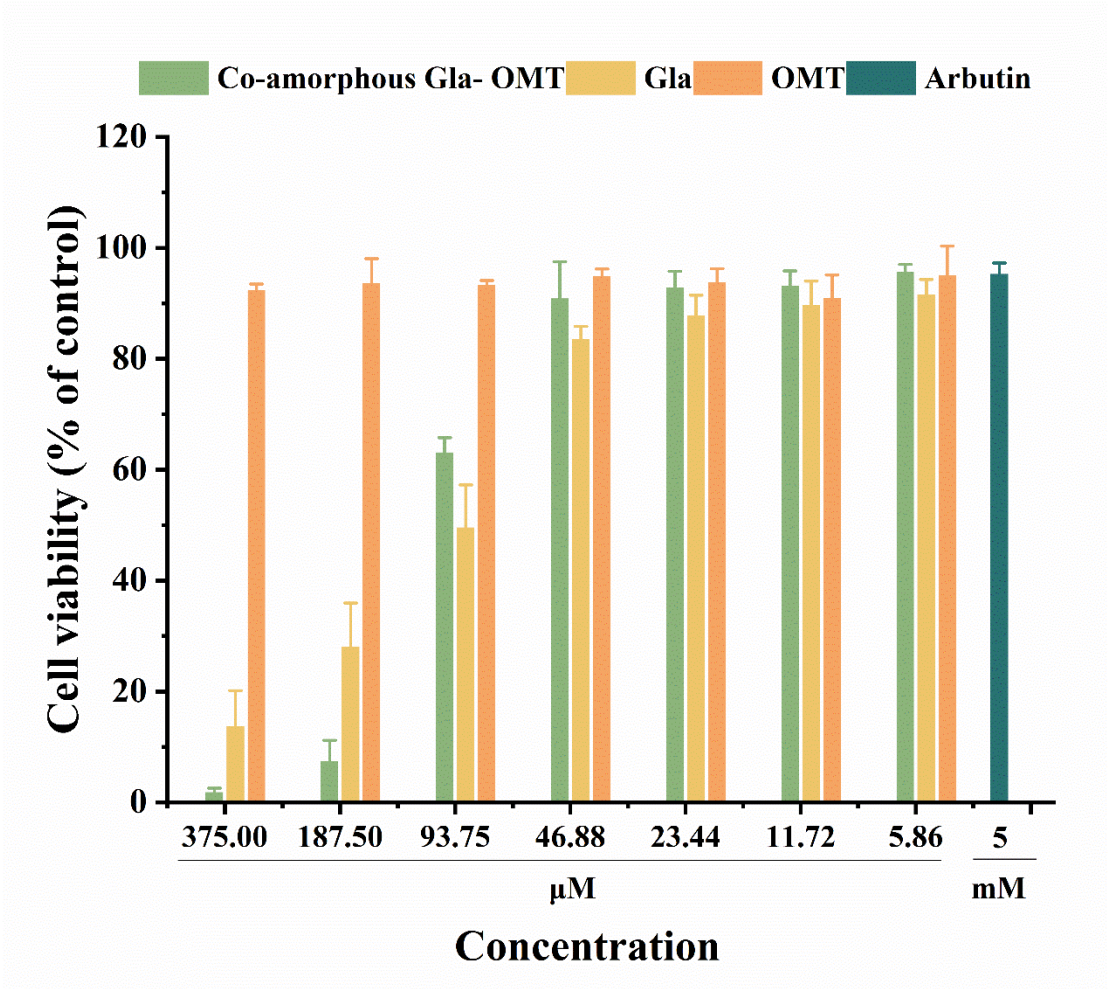

Figure S13 Effects of different drugs on cell activity

1 Polarizing Microscope

A small amount of crystalline Gla, crystalline OMT, and co-amorphous Gla-OMT samples are respectively placed on glass slides. A small amount of liquid paraffin is added to disperse them thoroughly. Morphological observations were performed using a 10×10 magnification LV100N POL polarizing microscope (Nikon, Japan). The images were collected using NIS-Elements F 4.00.00 software (Nikon, Japan).

2 Polarizing Microscope Analysis

The polarizing microscope results for crystalline Gla, crystalline OMT, and co-amorphous Gla-OMT samples are shown in **Figure S10**. Under the polarizing microscope, both crystalline Gla and crystalline OMT exhibit evident birefringence phenomena. The co-amorphous Gla-OMT form appears as irregular fragments under the polarizing microscope, lacking the birefringence phenomenon. This indicates that

co-amorphous Gla-OMT no longer possesses a crystalline structure and is in an amorphous state.