

Flowchart of compound extraction and purification from avocado wax using HPLC and NMR analysis

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Appendix A. of

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1. The flow chart of extracting and purifying compounds from avocado wax

Below is the flow chart of extracting and purifying compounds from avocado wax solution involves several steps, aiming to isolate fluorescent compounds using HPLC filtration. Initially, the extracted wax solution is washed with chloroform (CHCl_3), and the CHCl_3 extracts are collected. These extracts undergo SiO_2 column chromatography with a CHCl_3 -Hexane-MeOH solvent mixture, yielding four fractions: 27.5 mg, 14.4 mg, 12.1 mg, and 45.6 mg. Fraction 3 is processed again using SiO_2 column chromatography with the same solvent mixture. Fraction 2 is collected and further processed using SiO_2 column chromatography with an EtOAc-Hexane-MeOH solvent mixture, producing four fractions: 1.1 mg, 1.7 mg, 0.6 mg, and 18.1 mg. The fractions with fluorescence, specifically fractions 2 and 3 (with 2 having strong fluorescence and 3 having weak fluorescence), are then subjected to HPLC filtration with methanol (MeOH). The final products from this filtration are the filtrate containing fluorescent compounds and powder.

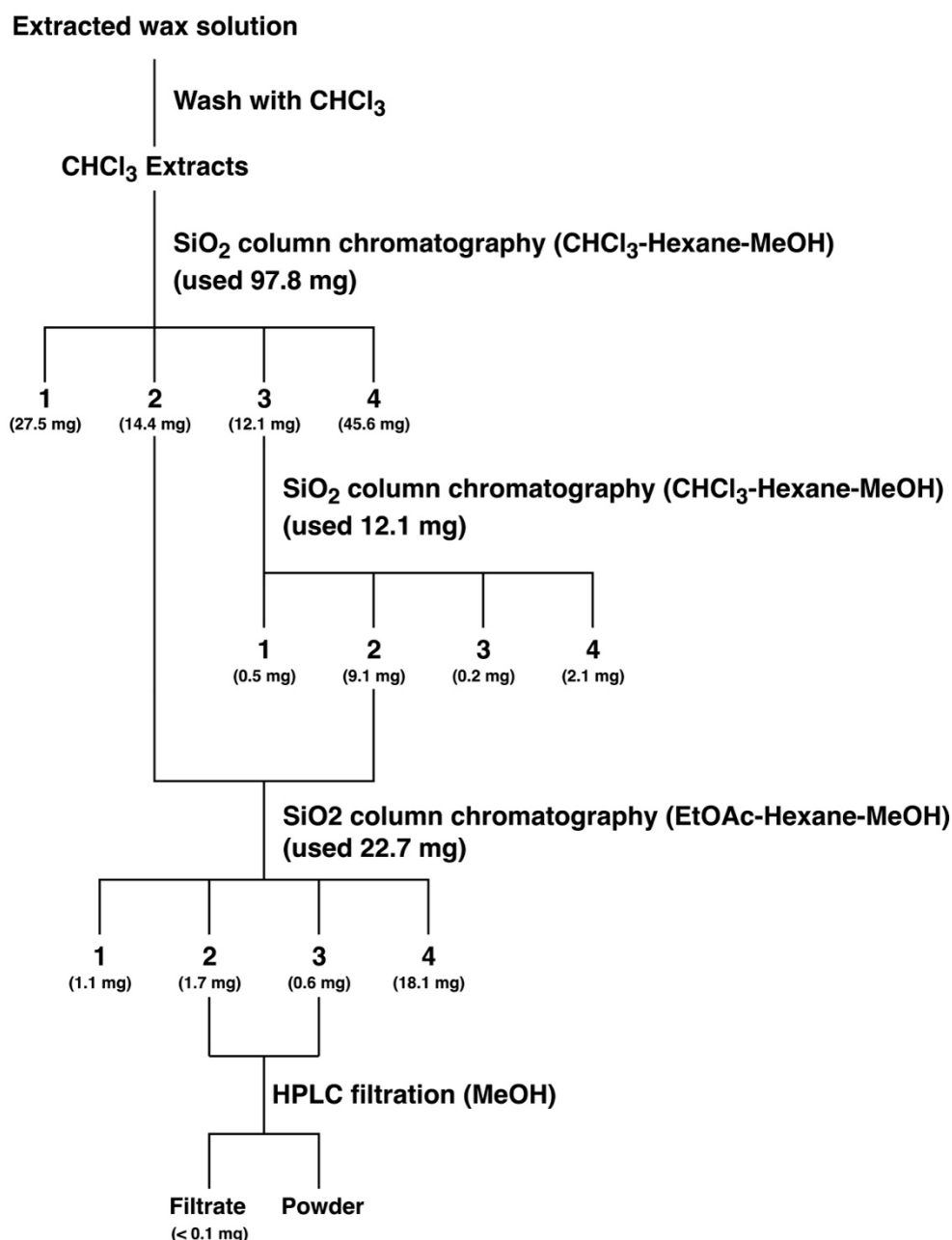


Fig. S1 The flow chart of extracting and purifying compounds from avocado wax

2. The ^1H NMR spectrum for the filtrate containing fluorescent compounds

The ^1H NMR spectrum for the filtrate containing fluorescent compounds recorded at 500 MHz in deuterated chloroform (CDCl_3). The spectrum shows a complex mixture of compounds, indicated by the multiple peaks throughout the spectrum. The presence of aromatic, vinylic, and aliphatic protons suggests a complex molecular structure with multiple functional groups. Further analysis would be needed to determine the exact structure of the compounds.

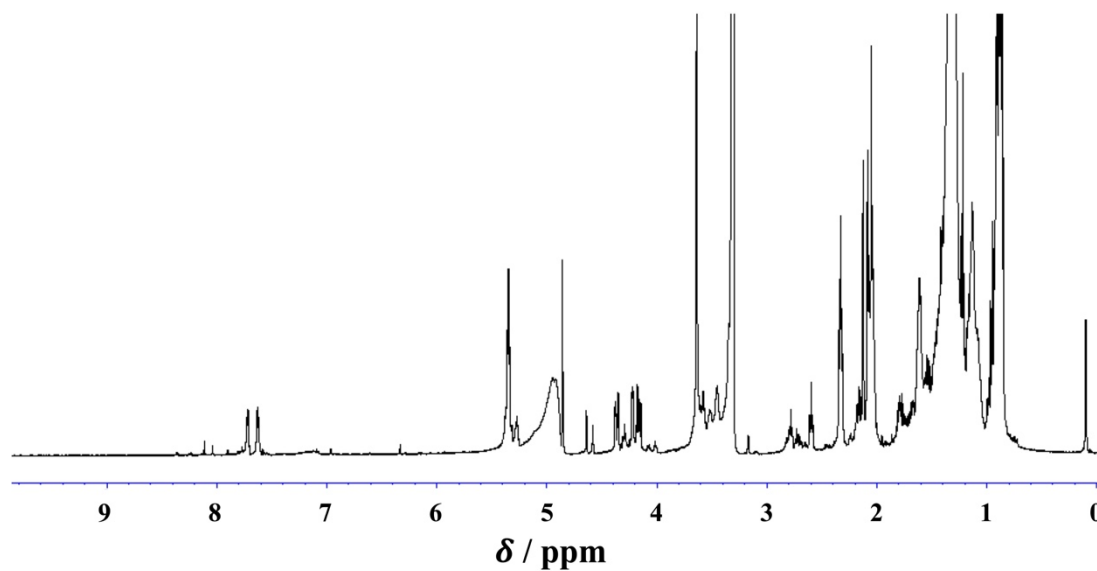


Fig. S2 The ^1H NMR spectrum for the filtrate containing fluorescent compounds