Dried Blood Spot Analysis for Chiral Compounds: Determination of Warfarin Enantiomers in Human Blood Using HPLC and Tandem Mass Spectrometer

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Objective: Study the application of dried blood spot (DBS) technology on the chiral drugs using warfarin as a model. The effect of DBS on the chiral chromatography, inter-conversion of warfarin enantiomers in DBS in sample preparation, storage condition and time would be studied. Method: Warfarin enantiomers were spiked into fresh human blood, then the blood was spotted onto DBS cards. A disc was cut and the analytes were extracted with methanol. A bioanalytical method for the determination of R- and S-warfarins in human blood has been developed with a linear range of 5-5000 ng/mL. In the method, R-and S-warfarins were baseline separated and determined using a chiral HPLC-MS/MS system. Results: The result indicated that the extract of DBS did not interfere with the chiral chromatography in the method. There was no enantiomer inter-conversion was observed in the sample preparation procedures. The DBS card has been put in room temperature for 1 month, and no stability issue or inter-conversion of enantiomers was observed. Implications: The DBS technology did not cause any stability issue or inter-conversion of warfarin. It has a great potential to apply to the discovery or development programs for chiral drugs.