ABSTRACT

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Warfarin is widely used anticoagulant drug. Considering the narrow therapeutic window of warfarin, it is important to fully understand its metabolism in human body. Oxidative, reductive and conjugation reactions are involved in warfarin metabolism. However, the reductive metabolism of warfarin has not been studied in details until now. The reduced metabolite of warfarin, i.e. warfarin-alcohol, is produced by the conversion of the carbonyl group of the side chain. It is known that human liver cytosolic and microsomal fractions exhibit warfarin reductase activity but the specific enzymes catalysing the reduction of warfarin are not known yet. The aim of this study was to identify the enzyme(s) participating in reduction of warfarin and to describe enzyme kinetics. Human liver cytosolic and microsomal fractions and recombinant enzymes AKR1A1, AKR1B1, AKR1B10, AKR1C1, AKR1C2, AKR1C3, AKR1C4, CBR1 and CBR3 were incubated with warfarin at various concentrations. The produced warfarin-alcohol was quantified by UHPLC and the specific activities of enzymes and subcellular fractions were determined. The warfarin reductase activity was confirmed in cytosolic and microsomal fractions. The reduction of warfarin was higher in the liver cytosol than liver microsomes. From the enzymes tested, AKR1C3 and CBR1 were found as the main enzymes participated in the production of warfarin-alcohol. Other enzymes showed only low or no activity.