

ABSTRACT NADPH-P450 oxidoreductase (POR) is a membrane bound flavoprotein that donates electrons to a wide spectrum of heme-containing proteins, among which are several steroidogenic and many xenobiotics-metabolizing enzymes. Given the important role of POR protein in drug metabolism and pharmacogenomics, there is a particular need to understand the contributions of POR genetic variants to these processes. Mutations in *POR* gene cause a disorder called POR deficiency, which manifests with a wide phenotypic spectrum ranging from disordered steroidogenesis to skeletal malformation, namely, Antley-Bixler syndrome (ABS). The aim of the present work was to investigate the *POR* gene in patients suspected to have POR deficiency syndrome from Czech Republic and to perform genotyping in Czech and Jewish control populations. We analyzed 644 alleles in unrelated individuals from the general Czech population and 1128 alleles in Jewish population, where 330 alleles were of Ashkenazi and 798 of Sephardic Jews. We have also studied the impact of selected new genetic variants on POR activity and identified fourteen amino acid variations, two of which we have studied in detail to establish their influence on POR activity. Using the available human POR three-dimensional structure, we then modelled the newly identified variants to describe these defects at the molecular level. Through this study, notably, we have systematically performed analysis of the *POR* gene and are providing POR deficiency diagnostics - the only laboratory in Czech Republic to provide this service. In conclusion, we have, for the first time, defined *POR* allele frequencies in the Czech and Jewish populations that have 14 novel amino acid variations of which two variations studied in detail impinge upon POR activity.