## VKORC1 gene polymorphism as cardiovascular biomarker: Detection by electrochemical genosensors

*Costa, Inês F., CISA|ESS, Centro de Investigação em Saúde e Ambiente, Escola Superior de Saúde, Instituto Politécnico do Porto, Rua Dr. António Bernardino de Almeida, 400, 4200-072, Porto, Portugal*
*Sousa, António J.S.F., CISA|ESS, Centro de Investigação em Saúde e Ambiente, Escola Superior de Saúde, Instituto Politécnico do Porto, Rua Dr. António Bernardino de Almeida, 400, 4200-072, Porto, Portugal*
*Banegas, Rodrigo S., Instituto Federal Catarinense (IFC) – Campus Camboriú, Rua Joaquim Garcia s/n. Camboriú, Santa Catarina, Brazil*
*Morais, Stephanie L., REQUIMTE*
*Magalhães, Júlia, REQUIMTE*
*Domingues, Valentina, REQUIMTE*
*Delerue-Matos, Cristina, REQUIMTE*
*Ramos-Jesus, Joilson, Grupo de Estudos em Genética Humana e Médica (GEHMED), Laboratório de Genética e Biologia Molecular, Departamento de Biomedicina, Universidade do Delta do Parnaíba (UFDPar), Parnaíba, PI, Brazil*
*Ferreira-Fernandes, Hygor, Grupo de Estudos em Genética Humana e Médica (GEHMED), Laboratório de Genética e Biologia Molecular, Departamento de Biomedicina, Universidade do Delta do Parnaíba (UFDPar), Parnaíba, PI, Brazil,*
*Pinto, Giovanny R., Grupo de Estudos em Genética Humana e Médica (GEHMED), Laboratório de Genética e Biologia Molecular, Departamento de Biomedicina, Universidade do Delta do Parnaíba (UFDPar), Parnaíba, PI, Brazil*
*Santos, Marlene, CISA|ESS, Centro de Investigação em Saúde e Ambiente, Escola Superior de Saúde, Instituto Politécnico do Porto, Rua Dr. António Bernardino de Almeida, 400, 4200-072, Porto, Portugal*
*Barroso, M. Fátima, REQUIMTE*

### Abstract

Warfarin is an anticoagulant generally used to prevent cardiovascular diseases. Since of the low therapeutic index of warfarin and frequent complications of prevention or treatment, significant differences in individual doses of warfarin are needed to achieve prophylactic and therapeutic ranges.

Recent studies have been reporting that genetic variants of vitamin K epoxide reductase complex (VKORC1) influence the response to warfarin and doses [9]. So, the genetic and pharmacogenetic information of the major cardiovascular diseases plays an important role in the identification of the cardiovascular risk factors and in the diagnosis and treatment of these conditions.

This work addresses the development of a disposable electrochemical genosensor able of detecting single nucleotide polymorphism (SNP) in the VKORC1 gene. Analysing public databases, two specific 52 bp DNA probes, one with adenine (TA) and another with guanine (TG) SNP genetic variation were selected and selected and designed.

The genosensor methodology implied the immobilization of a mixed self-assembled monolayer (SAM) linear VKORC1 DNA-capture probe and mercaptohexanol (MCH) onto screen-printed gold electrodes (SPGE). To improve the genosensor´s selectivity and avoid strong secondary structures, that could hinder the hybridization efficiency, a sandwich format of the VKORC1 allele was designed using a complementary fluorescein isothiocyanate-labelled signaling DNA probe and enzymatic amplification of the electrochemical signal.

Preliminary studies indicate that differences in the electrochemical answers were obtained depending of the hybridization reaction format. In fact, higher electrochemical intensities were measured when the hybridization reaction was performed with a complementary DNA (without SNPs). These results suggested that the sensor is able to discriminate between the complementary DNA and single base mismatch targets having a great potential for the DNA polymorphism analysis