Phenotypization of cytochrome CYP2D6 activity with dextromethorphan can predict tamoxifen activation in breast cancer patients

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Background. Tamoxifen (TAM) activity is mainly due to its active metabolite endoxifen (END) which is prevalently formed by the hepatic cytochrome CYP2D6; CYP2D6 metabolic activity can be estimated by a phenotyping test, using destromethorphan (DMT) as a drug probe, and by genotyping CYP2D6 polymorphisms responsible for the enzyme function. Our study aimed at predicting steady-state END plasma levels by means of CYP2D6 pheno/genotypization, in order to identify a priori patients who are more likely to respond to TAM anti-estrogenic treatment.

Methods. One hundred and twenty early breast cancer patients (pts) treated with TAM (20 mg/die) were the study population. Plasma concentrations of END were determined by HPLC 1 month (END-1m) and 4 months after starting TAM treatment (END-ss), i.e. when pts were considered to have stable TAM and metabolites plasma levels.
A phenotyping test was performed before starting therapy: pts were given DMT 15 mg per os and urine was collected for the following 8 hours. DMT and its metabolite dextrorphan (DOR) concentrations were measured in urine (by HPLC) and the log transformed urinary metabolic ratio (log MR = [DMT]/[DOR]) was calculated.
DNA was extracted from peripheral blood leukocytes and the CYP2D6 variant alleles with normal (*1, *2), null (*3, *4, *5 and *6) and reduced (*9, *10 and *41) activity were detected, through DHPLC and RFLP assays. Patients were classified in three different functional groups as Extensive (EM), Intermediate (IM) and Poor (PM) Metabolizers, according to the CYP2D6 genotype. Data were analyzed by means of ANOVA or regression analysis, as required.

Results. ENDss plasma levels varied between 2.4 and 39.2 ng/ml (median: 8.7 ng/ml); their frequency distribution appeared to identify three groups with different TAM activation capability. DMT/DOR logMR varied between -3.1 and +1.2 (median: -1.6) and showed a three-modal distribution as well, with -0.52 and -1.5 as cut off values. CYP2D6 alleles with normal, null and reduced activity were found in 51.0, 26.8 and 22.2% of cases, respectively. Fourteen pts had two null alleles and 58 pts had at least one null or reduced activity allele. Patients with homozygous (hom) or heterozygous (het) defective mutations had significantly lower ENDss plasma levels compared to pts with wild type (wt) genotype (hom: 3.2 ±1.1 ng/mL; het: 8.7±5.2 ng/mL; wt: 14.4±8.1 ng/mL; ANOVA, $p<0.0001$). END-ss was significantly correlated with DMT/DOR logMR ($r^2 = 0.49$; $p<0.0001$) and with END-1m ($r^2 = 0.76$; $p<0.0001$). Multiple regression analysis carried out to identify which variables could predict END-ss showed that END-1m and logMR (but not CYP2D6 genotype) were independently associated with END-ss, according to the equation:
END-ss = 0.055 - 1.53 ´ logMR + 1.11 ´ END-1m ($r^2 = 0.77$; $p<0.0001$).

Conclusions. Our finding suggest that END-ss can be predicted by determining individually the CYP2D6 phenotype before starting TAM and by measuring END plasma concentration after 1 month of treatment.

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