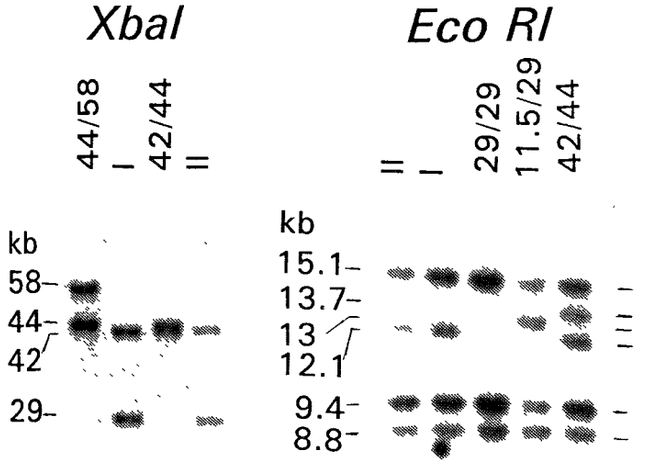
Molecular basis for rational megaprescribing in ultrarapid hydroxylators of debrisoquine

SIR,—About 7% of whites are poor metabolisers of debrisoquine and of several essential groups of drugs (eg, antidepressants, neuroleptics, and antiarrhythmics).[1-3] Poor metabolisers of debrisoquine (metabolic ratio [MR] > 12·6) are homozygous for a defective *CYP2D6* gene and can be identified with genomic DNA and PCR-based allele-specific amplification,[4] which predicts the metabolic phenotype with 92–99% accuracy.[5] Poor metabolisers of debrisoquine attain high plasma concentrations of antidepressants, such as clomipramine, desipramine, and nortriptyline, and may experience adverse reactions even on low doses.[6] Different strategies to treat poor metabolisers with various drugs have been discussed.[3] The problems in treating patients with extremely rapid metabolism have been given much less concern. We have described a woman (I) with depression having an MR for debrisoquine of 0·07 who had to be treated with 500 mg nortriptyline daily (3–5 times the recommended dose) to attain therapeutic plasma levels.[7] Another psychiatric patient (II) with agoraphobia without panic disorder was treated with 150 mg clomipramine daily (normal dose 25–150 mg). Since there was no response, the dose was increased to 225 mg per day to yield plasma concentrations of 150 nmol/l clomipramine and under 100 nmol/l demethylchlomipramine (limit of detection), which are much lower than expected.[8] The patient is now stabilised on 300 mg per day.

We have investigated the molecular genetic basis for the extremely rapid metabolism of our two patients. *Xba*I restriction fragment length polymorphism (RFLP) of genomic DNA isolated from leucocytes revealed a fragment pattern of 42/29 kb in both patients (figure, left). Allele-specific PCR[4] showed absence of the two defective variant *CYP2D6A* and *CYP2D6B* genes in both subjects (data not shown), which indicates that they belong to the "44E genotype" previously described for extensive metabolisers having the *Xba*I 44 kb haplotype.[5] *Eco*RI RFLP analysis showed fragments corresponding to the pseudogenes *CYP2D7P* and *CYP2D8P*[9] as well as to the wt *CYP2D6* gene (figure, right). In addition, a 12·1 kb fragment corresponding to another *CYP2D* gene was seen in both cases. Such a fragment has recently been reported to originate from a functionally active *CYP2D* gene, 12-fold amplified in an inherited manner, which we have termed *CYP2D6L*. Allele-specific PCR analysis for the mutations at 2938C→T and 4268G→C present in this allelic form of *CYP2D6* indicated that both subjects carried the *CYP2D6L* gene. We thus suggest that the origin for the extremely rapid metabolism in our patients is the presence of two different functionally active *CYP2D6* genes in their *CYP2D* locus, causing more enzyme to be expressed. Our data further indicate that it should be possible to screen for this haplotype by allele-specific PCR.



RFLP of *CYP2D6* locus and genomic DNA from two patients (I and II).

Left: *Xba*I RFLP of *CYP2D6* locus in comparison with two subjects of *Xba*I 44/58 kb haplotype and 42/44 kb haplotype.

Right: *Eco*RI RFLP of genomic DNA in comparison with DNA from individuals of the *Xba*I 29/29, 11·5/29, and 42/44 kb haplotypes. Fragments with length of 15·1, 13·7, 9·4, and 8·8 kb correspond to genes *CYP2D7AP, CYP2D7BP, CYP2D6,* and *CYP2D8P,* respectively. Characteristic for DNA of 42 kb haplotype was appearance of *Eco*RI fragment with length 12·1 kb, which was seen in genomic DNA isolated from both patients

Patients who do not respond to normal doses of antidepressants are often switched to other drugs in the same category. This is not rational if the reason for the poor response is metabolic in nature, since both classical tricyclic antidepressants and the novel serotonin uptake inhibitors are metabolised by the same enzyme, *CYP2D6*.[2,3] Ultrarapid metabolism of these drugs may be one reason for therapeutic failure. Studies are in progress to assess the frequency of ultrarapid metabolisers of *CYP2D6* substrates.

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Storage of skin grafts in honey

SIR,—A search for a simple and cheap method of preservation of skin for grafting continues in the developing world. Honey is hypertonic, sterile, and bactericidal.[1-3] Experimental evidence shows good histological preservation of skin grafts in honey.[4]

We had 28 patients between January, 1990, and June, 1991, who required skin grafts at multiple sites. The patients were aged 5–55. 20 patients needed grafting for burns (average burn area 14·2%) and in 8 cases for ulcers. Informed consent was obtained in each case. When healthy granulation tissue appeared in one of the sites, grafting was planned to cover the area. The remaining areas in which grafting could not be done at the same time due to infection or slough were also assessed. Extra grafts were taken for future use to cover the other areas. After grafting at the primary site, the extra grafts were preserved in undiluted unprocessed honey kept at room temperature and at 4°C. The honey was confirmed sterile before use. Subsequently grafting with honey-stored grafts was done after the swab culture was reported sterile. This was done at 2–3 weeks in 8 patients, 4–6 weeks in 5, 7–10 weeks in 7, and 11–12 weeks in 8. The areas that were covered with these grafts were upper limb (6), abdomen (4), lower limb (8), chest (6), and neck and perianal region (4).