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O-アセチル転移酵素遺伝子を特異的に破壊したエームス試験菌株の作製

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目的：*O*-アセチル転移酵素（OAT）は，ニトロアレーンや芳香族アミン由来の*N*-ヒドロキシアリルアミンの代謝活性化に関わる酵素である．*oat*遺伝子を特異的に破壊したエームス試験菌株を作製し，既存の*oat*欠損株(TA100/1,8-DNP, TA98/1,8-DNP_6)と感受性を比較した．

方法：プレライゲーション法を用い，染色体上の*oat*遺伝子をクロラムフェニコールアセチル転移酵素遺伝子(*cat*)と置換することで，*oat*遺伝子破壊株を作製した．*S. typhimurium* TA1535, TA1538の*oat*遺伝子破壊株をYG7125, YG7129と命名し，更にpKM101を導入した株をYG7126, YG7130とした．

結果と考察：YG7126株とYG7130株は，1,8-dinitropyrene(1,8-DNP), 1-nitropyrene, Glu-P-1, IQに対し既存の*oat*欠損株とほぼ同様の感受性を示した．既存の*oat*欠損株は変異原処理により得られた菌株なので，複数の遺伝子に変異が起きている可能性があるが，今回の結果から変異原の活性化に関しては*oat*遺伝子のみが欠損しているものと考えられる．一方，2-nitrofluorene(2-NF)に対するYG7126株とYG7130株の感受性は，親株と既存の*oat*欠損株との中間の値を示した．*cat*遺伝子を含むプラスミドを既存の*oat*欠損株に導入し，2-NFに対する感受性を調べると，プラスミド導入株が3-10倍高い感受性を示した．この結果から，クロラムフェニコールアセチル転移酵素が2-NFの活性化に関与することが示唆された．

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Influence of donor genotype on sister chromatid exchange induction in cultured human lymphocytes by styrene and styrene-7,8-oxide

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Styrene is converted in the liver to styrene-7,8-oxide (SO), a reactive epoxide which can be detoxified by microsomal epoxide hydrolase (mEH; hydrolysis to styrene glycol)or by glutathione *S*-transferase (GST; conjugation with glutathione). As inherited differences in the activity of mEH and GST may influence individual susceptibility to styrene, we studied the importance of the known genetic polymorphisms of mEH gene (*EPHX*) and *GSTM1* and *GSTT1* genes on the *in vitro* genotoxicity of SO and styrene in 72-h whole-blood lymphocyte cultures (48-h treatment started 24 h after culture initiation) of donors representing different genotypes.

The $His_{113}Tyr$ polymorphism in *EPHX* (suggested to result in decreased mEH activity) did not influence SCE induction by SO in lymphocyte cultures of 16 donors. The results may indicate that the mEH activity of blood cells is too low to significantly affect SO metabolism *in vitro* or that the polymorphism does not result in high enough difference in mEH activity. In fact, erythrocytic GSTT1 may be more important than mEH in detoxifying SO in whole-blood cultures. We showed earlier that *GSTT1* genotype influences SCE induction by SO in cultured human lymphocytes. The importance of glutathione conjugation *in vitro* was further supported by our results with styrene which induced a higher SCE response in cultures of donors lacking both *GSTT1* and *GSTM1* genes in comparison with donors having both genes.

These findings suggest that *GST* polymorphisms may influence SO genotoxicity in blood cells and possibly in blood-forming organs. However, a major part of styrene metabolism in humans *in vivo* is thought to occur in the liver where the mEH pathway (eventually leading to the main urinary products of styrene, mandelic acid and phenyl glyoxylic acid) is clearly more prominent than glutathione conjugation. Thus, individual differences in mEH activity are expected to be important for SO genotoxicity *in vivo*.