30B09-1

TRANSITIONAL DOWNREGULATION OF THE CYP1B1 GENE BY PROMOTER CpG METHYLATION IN COLON CANCER CELLS
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Cytochrome P450 1B1 (CYP1B1) and 1A1 (CYP1A1), dioxin-inducible members of the CYP family, are associated with carcinogenesis in hormone-mediated cancers (breast, endometrial, ovarian, and prostate). Although aberrant expression of these enzymes is also observed in different types of cancers (independent of hormone response), their roles in carcinogenesis are not yet fully understood. We examined DNA methylation status of CYP1B1 and CYP1A1 genes in 7 colorectal cancer cell lines and 40 pairs of normal and cancerous tissues of the colorectum. By bisulfite-modified DNA direct sequencing, CYP1B1 gene promoter methylation was detected in 2 (29%) cell lines (SW48 and Caco-2) and 2 (5%) cancerous tissues, but not in normal tissues. Treatment of the cell lines with 5-aza-2'-deoxycytidine revealed a clear increase in the CYP1B1 mRNA levels in SW48 and Caco-2 cells, while amount of methylated alleles decreased. Interestingly, only HT29 cells showed a clear increase in CYP1A1 mRNA, although there were no apparent differences in CYP1A1 gene methylation status among the 7 cell lines. None of these cell lines showed significant change in mRNA levels of AhR (aryl hydrocarbon receptor) and ARNT (AhR nuclear translocator), which are known to directly activate CYP1 transcription. This observation suggested that CYP1B1, but not CYP1A1, expression was downregulated by promoter methylation rather than decreased expression of AhR and/or ARNT. In conclusion, CpG methylation of the CYP1B1 promoter region in some colorectal cancers is involved in the regulation in CYP1B1 gene during cancer progression. Moreover, cancers with aberrant CYP1B1 expression might show altered response to procarcinogen metabolism and chemotherapy.

30B09-2

EFFECT OF DIETARY CONSTITUENTS ON INDUCTION OF CYTOCHROME P450 IN NEONATAL RAT LIVER
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In a previous study, we observed the developmental changes of liver cytochrome P450 (CYP) activities in rat, showing that the activities were extremely low in prenatal days (PND) 1~7 and then increased gradually. As CYP1As activities were significantly increased around weaning, we thought that the development of CYP activity could be affected by diet. The present study investigated the comparative effects of two diets on the ontogeny of hepatic cytochrome P450 (CYP) in neonatal rats. A regular cereal-based diet (MF; Oriental Yeast Co. Japan) or a purified diet (AIN-93G; Clea Japan, Inc.), according to the recommendations of the American Society of Nutrition, were fed to pregnant Wistar rats. Their offspring were sacrificed at PND 14, 21, 28, 35, and the liver microsomal CYP activities were assayed. Alkylresorufin-O-dealkylase activities (EROD: CYP1A1, MROD:CYP1A2, BROD:CYP2B) and alkylxoy-4-(trifluoromethyl)coumarin-O-dealkylase activities (BFCD: CYP3A, MFCD: CYP2C) were measured. The expression of CYP isoforms was determined by Western blotting.

The neonatal body weight gain in the pure diet (AIN-93G) group was slightly larger than that of the normal diet group. The CYP activities were extremely low at PND 14 and PND 21 in both diet groups. In the normal diet (MF) group, the activities were remarkably elevated at PND28, whereas the pure diet (AIN-93G) group showed lower CYP activities than those of the normal diet group and still showed low activities at PND35. These results suggest that regular cereal-based diets contain inducers of CYP.