97 **PRACTICAL INTERPRETATION OF THE REVISED OECD GUIDELINES FOR GENOTOXICITY TESTING. THE RECOMMENDATIONS OF THE INDUSTRIAL GENOTOXICITY GROUP.** Jones E[1], Henderson LM[2], Clements J[3]. [1]AstraZeneca Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, SK10 4TJ, UK. [2]Unilever Research, Port Sunlight Laboratory, Quarry Road East, Bebington, Wirral, Merseyside, L63 3JW, UK. [3]Covance Laboratories Ltd, Otley Road, Harrogate, North Yorks, HG3 1PY, UK.

Revised guidelines for the core genotoxicity assays were published by the OECD in 1998. As with all regulatory guidelines there are areas where legitimate alternatives are allowed. However, there are also points which are open to a number of interpretations. The Industrial Genotoxicity Group (affiliated to UKEMS) convened a panel of experienced genetic toxicologists to review the guidelines and, where alternatives or uncertainty existed, to make recommendations on best scientific practice to aid in consistent interpretation of the guidelines. The review covered the five core assays: bacterial mutation; in vitro cytogenetics, mammalian cell gene mutation (with particular emphasis on the L5178Y TK+/- assay); erythrocyte micronucleus; in vivo UDS. General points covered by the review included: presentation and use of historical control data; definition of maximum dose level; evidence of absorption/tissue exposure in in vivo assays; justification for use of a single sex in in vivo assays; strategy for repeat assays in in vitro assays; testing of insoluble materials. Assay specific recommendations included the following topics. Bacterial mutation assay: strategy for anti-bacterial compounds; choice of strains. In vitro cytogenetics assay: measurement of polyploidy; test design; cell cycle measurement. Mammalian cell gene mutation assay: colony size data for L5178Y; design of repeat test. Erythrocyte micronucleus test: choice of dose route. Liver UDS assay: method used for cytoplasmic counts; repair criteria. The details of the panel's recommendations will be given in this presentation to enable them to be discussed within the genetic toxicology community.

98 **ALTERATIONS OF CANCER-RELATED GENES IN TUMOR CELL LINES DERIVED FROM CADMIUM-TRANSFORMED BALB/C-3T3 CELLS.** Joseph P[1], Muchnok TK[1], Zhou G[1], Ong T[1], Whong WZ[1]. [1]Health Effects Laboratory Division, National Institute for Occupational Safety and Health, Morgantown, WV 26505.

The International Agency for Research on Cancer (IARC) has classified cadmium as a human carcinogen. A large number of workers are potentially exposed to this metal in various occupational settings. Considering the lack of information regarding the molecular mechanism(s) of cadmium carcinogenesis, studies were conducted to investigate the changes in copy number and expression of various cancer-related genes as a possible mechanism for carcinogenesis due to exposure to cadmium. BALB/c-3T3 cells were morphologically transformed with cadmium and the transformed cells were subcutaneously injected into nude mice to develop tumors. Cell lines established from these tumors were used in the present study. Differential PCR and RT-PCR were employed to determine changes in copy number and expression of p53, p16, K-ras, c-myc, c-fos, c-jun, c-sis and erbB1. Tumor cell lines showed a decrease in gene copy number and expression of p16 and erbB1 compared to the control BALB/c-3T3 cells. Gene copy number and gene expression of c-fos and c-jun were significantly higher in the tumor cell lines compared to those of the control cells. In spite of a significant increase in gene copy number of K-ras in tumor cells, no change was observed in its expression. Neither the copy number nor the expression of c-myc exhibited any significant change in the tumor cell lines. These results indicate that the observed changes in gene copy number and gene expression of the cancer-related genes may be at least partly responsible for the cellular transformation induced by cadmium.

99 **ASSOCIATION BETWEEN TUMORS DEVELOPED FROM BERYLLIUM-TRANSFORMED CELLS AND CHANGES IN CANCER-RELATED GENES.** Joseph P[1], Klishis ML[1], Muchnok TK[1], Ong T[1], Whong WZ[1]. [1]Health Effects Laboratory Division, National Institute for Occupational Safety and Health, Morgantown, WV 26505.

Beryllium is used extensively in various industrial applications and many workers are potentially exposed to toxic levels of this metal. Even though laboratory studies have documented the carcinogenic potential of beryllium in experimental animals, the underlying molecular mechanisms are poorly understood. In order to elucidate the molecular mechanisms of beryllium-induced tumorigenesis, we have studied the various cancer-related genes that were altered as a result of cellular transformation induced by beryllium. BALB/c-3T3 cells, morphologically transformed with beryllium, were subcutaneously injected into nude mice to develop tumors. DNA and RNA isolated from cell lines derived from the tumors were used to determine the alterations of various cancer-related genes, such as p53, p16, K-ras, c-myc, c-fos, c-jun, c-sis and erbB1. Alterations of the copy number and expression of the genes were studied by differential PCR and RT-PCR, respectively. Both the gene copy number and expressions of K-ras, c-fos and c-jun were higher in the tumor cells compared to those of the control Balb/c-3T3 cells. In contrast, p16 exhibited a decrease in both the gene copy number and expression in the tumor cells compared to those of the control cells. These results suggest that beryllium-induced tumorigenesis is associated with alterations of various cancer-related genes, and these alterations may play a role in beryllium-induced tumorigenesis.

100 **NEW *E. COLI* STRAINS EXPRESSING HUMAN OR RAT P450 ENZYMES -- APPLICATION TO PROBLEMS IN MOLECULAR TOXICOLOGY AND MUTATION RESEARCH.** Josephy PD[1], Batty S[1], Kelly J[2], Goulden E[2], Evans DH[2]. [1]Chemistry and Biochemistry, University of Guelph, Guelph, Ontario, Canada N1G 2W1. [2]Molecular Biology and Genetics, University of Guelph, Guelph, Ontario, Canada N1G 2W1.

Recombinant enzymes can replace the hepatic "S9" preparations which are conventionally used as metabolic activation systems in bacterial mutagenicity tests. Here, we report new applications of this strategy to problems in molecular toxicology. *E. coli* strain DJ702 expresses recombinant human P450 1A2 and P450 reductase (on a pBR322-derived plasmid) and S. typhimurium acetyl CoA: arylamine N-acetyltransferase (on a pACYC184-derived plasmid) and carries a *lacZ* frameshift allele as a reversion mutation assay target. (i) We have constructed strains isogenic to DJ702, but expressing rat P450 1A2 (DJ710) or human P450 1B1 (DJ711), and compared the responses of these strains to aromatic amine carcinogens. In mutagenicity assays with MeIQ (2-amino-3,4-dimethylimidazo[4,5-*f*]quinoline), DJ710 gives fewer revertants than does DJ702, even though the rat enzyme is expressed at a high level. This finding may have implications for the extrapolation to humans of the results of carcinogenesis studies with rodents. (ii) We report further studies of variants of the human 1A2 coding sequence. Pools of mutants are generated by random mutagenesis of the 1A2 sequence carried in DJ702; active variants are picked from these pools by screening clones for MeIQ-dependent mutagenicity. (iii) We have constructed a new vector, pDJ99 (and a corresponding *E. coli* tester strain, DJ720) expressing all three enzymes required for arylamine activation on a single plasmid. (Supported by grants from NSERC Canada and MRC Canada)

National Library of Medicine PubMed

PubMed Nucleotide Protein Genome Structure PopSet

Search PubMed for Environmental and Molecular Mutagenesis Go Clear

Limits Preview/Index History Clipboard

Display Summary Save Text Order Add to Clipboard

Entrez PubMed

PubMed Services

Related Resources

1 : [No authors listed] Related Articles

Environmental Mutagen Society 31st annual meeting. New Orleans, Louisiana, USA. April 8-13, 2000. Abstracts.

Environ Mol Mutagen. 2000;35 Suppl 31:9-79. No abstract available.

PMID: 10709568; UI: 20174351

Revised: April 3, 2000.

Disclaimer | Write to the Help Desk

NCBI | NLM | NIH

An International Journal Specializing in Environmental Mutagenesis

Volume 35
Supplement 31
2000

Environmental and Molecular Mutagenesis

WILEY-LISS
ISSN: 0893-6692