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Sex as a major determinant of gene expression in tissues of mice exposed to arsenate.

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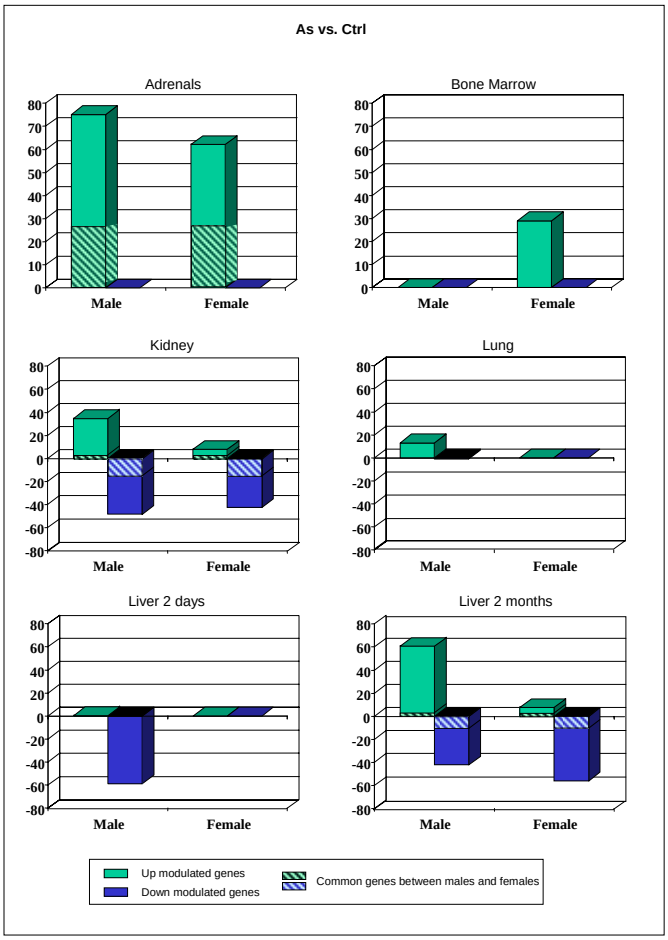
Introduction

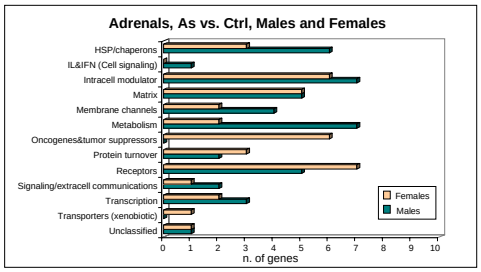
Inorganic arsenic, frequently found as contaminant of ground water used for drinking purposes in many areas of the world, is a well-known potent human toxicant and carcinogen. Chronic exposure to inorganic arsenic has been associated with cancer of skin, lung, bladder and kidney and, probably, liver. The mechanism of arsenic action *in vivo* is poorly understood, in particular in relation to dose, type of tissue and gender.
To elucidate tissue- and gender dependent biological responses in the genome of mice, we have used cDNA macroarrays for investigation on the expression of 1185 **cancer-related genes** in mice after exposure to arsenate in drinking water.

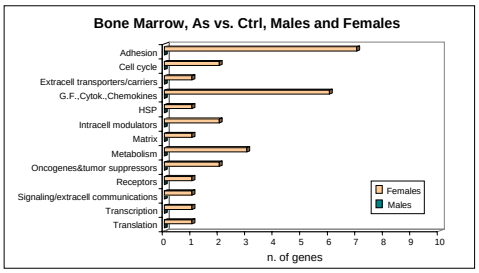
Materials and Methods

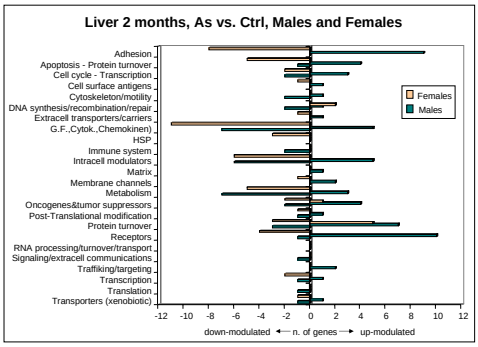
√ **Experimental animals***:* male and female CD-1 mice.
√**Treatment***:* Female adult mice were treated with arsenate in drinking water (1 mg As/L) for 10 days before mating and during the gestation. Separate groups of arsenic exposed males and females offspring were exposed for 2 months to 1 mg As/L of additional arsenate (As). Control male and female mice without any treatment were also analysed (Ctrl).
√ **Total RNA was extracted** from tissues using RNeasy Qiagen kit and 1 µg was converted into [^{33}P]-labelled cDNA using Super Script III Reverse Transcriptase (Invitrogen) and ^{33}P-dATP (Amersham), Mouse Cancer 1.2 CDS primer mix (Atlas™, Clontech, U.S.A.).
√**cDNA Hybridization** on Mouse Cancer 1.2 Array (Atlas™, Clontec, U.S.A.) membranes (16 hours at 50°C).
√**Image Analysis***:* After acquisition by Cyclone instrument (Packard Camberra Instruments, U.S.A.), the images were analyzed by Atlas Image software (Atlas™).
√ **Data Analysis***:* Significance Analysis of Microarrays (SAM).

Results









Conclusions

Continuous exposures of mice to arsenate in drinking water modulate the gene expression in tissues. Interestingly, there were remarkable sex differences: male and female mice show completely different changes in the expression of cancer-related genes.
The main gene functional families modulated, were covering a wide range of biochemical and physiological regulations, like cell cycle modulation, cell adhesion, apoptosis, xenobiotic metabolism, DNA repair, protein turnover and proto-oncogens.
This result demonstrates important gene-environmental interactions: the molecular mechanisms triggered by arsenic levels frequently experienced following exposure via drinking water, are totally different in males and females.
The results obtained using cancer-related genes will be compared with the profiles of over 30.000 genes using the Applied Biosystems expression Array System, to clarify the sex-specific gene pathways.