O$^6$ETHYLGUANINE IS NOT THE ONLY ETHYLATED ADDUCT RESPONSIBLE FOR THE INDUCTION OF MUTATIONS IN SYRIAN HAMSTER EMBRYO (SHE) CELLS. A.J.de Kok[1], A.D.Tates[1], J.W.I.M. Simons[1] and L.den Engelse[2]. [1]State University of Leiden and [2]The Netherlands Cancer Institute, Amsterdam, The Netherlands.

The possible correlations between cytotoxic, mutagenic and clastogenic damage induced by N-ethyl-N-nitrosourea (ENU) and specific ENU-induced DNA-lesions were investigated in SHE cells. Immediately after treatment with ENU or after 3 resp. 6 days confluent holding (CH) recovery, cells were either reseeded at low density to assay biological effects, or the DNA was isolated to determine ethylated adducts. To reduce cell turnover during CH to a minimum, medium with low serum (0.5%) was used. Immunological analysis indicated a limited capacity of SHE cells to repair O$^6$EtG ($t_{\frac{1}{2}}$ ± 144 hr). HPLC analysis confirmed this result and also revealed that 7-EtG and 3-EtA disappear rapidly from DNA ($t_{\frac{1}{2}}$ 55 resp. 72 hr). The amount of TpT-ethyl-triester remained unchanged. During a 6 day CH-period relative survival increased (from 25 to 75%), the frequency of ouabain resistant mutants (OUA$^r$) remained constant, whereas for thioguanine resistance it decreased (to 70%), as did chromosome aberration and SCE frequencies (both to 40%). Comparison of elimination kinetics suggest a correlation between O$^6$EtG and mutations at the HGPRT-locus, but not at the Na-K-ATPase-locus. Therefore we conclude that O$^6$EtG can be involved in the induction of TG$^r$, but that in the induction of OUA$^r$ other, more persistent lesions may be involved as well.

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NUCLEOLAR ACTIVITY DURING RAT HEPATOCARCINOGENESIS. A.Deleener[1], M.Kirschvolders[1], H.Barbason[2], J.de Gerlache[3] and M.Lans[3]. [1]Vrije Universiteit Brussel, Brussels, Belgium; [2]Universiteit de Liège, Liège, Belgium; [3]Universiteit Catholoque de Louvain, Brussels, Belgium.

Male Wistar rats were treated in two different ways and compared with untreated rats.
1) They were treated chronically for 2-6 weeks with diethylnitrosamine (DENA; 80 mg/l in drinking water) followed or not by an incubation period with phenobarbital as promotor.
2) They were treated with an acute dose of diethylnitrosamine (200 mg/kg dissolved in 0.9% NaCl solution), followed by a selection procedure for initiated cells with 2-AAF and a necrogenic dose of $CCl_4$. Afterwards they were incubated or not with phenobarbital. Hepatocytes were isolated and their nucleolar activity, identified by a $AgNO_3$ staining (AgNOR), was estimated by two different and complementary approaches: a NOR-cytophotometrical and a NOR-cytomorphological analysis. The results are:
1) in the non-nodular parenchymal cells no significant influence of the carcinogenic treatment was observed, considering the average silver staining intensity per cell,
2) although the cytophotometrical estimations do not differ between control and treated rats, it was demonstrated that during hepatocarcinogenesis DENA and phenobarbital modify in different ways the nucleolar activity as expressed by morphological changes of silver staining.

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GENOTOXICITY OF THE ANTITUMOUR ANTIBIOTIC TALLYSOMYCIN IN ASPERGILLUS NIDULANS. N.Demopoulos[1] and A.Kappas.[2] [1]Department of Genetics, University of Patras, Patras, Greece; [2]Biology Department, Nuclear Research Center "Democritus", Athens, Greece.

Tallysomycin, an antibiotic compound structurally related to bleomycin, shows also an antitumour activity like bleomycins and phleomycins. We have shown that bleomycin induced both point mutations and mitotic crossing-over in the ascomycete Aspergillus nidulans (Mutation Res. 102, 51-57, 1982). In this work we investigated the genetic activity of tallysomycin in the methionine independent reversion system of A.nidulans for suppressor mutations and the system based upon the para-sexual cycle of the fungus for mitotic segregation. We found that tallysomycin at the concentrations of 2, 4, 6, 8 and 16 µg/ml, which inhibited the germination of treated conidia from 30% to 90%, induced 59, 93, 127, 165 and 368 revertants per $10^6$ conidia respectively. Regarding mitotic segregation we found that tallysomycin at the very low concentrations of 0.025, 0.05, 0.075, 0.1 and 0.2 µg/ml, with an inhibitory effect on the colony growth up to 50%, increased the number of mitotic segregants 311%, 423%, 510%, 528% and 607% respectively over the control. The interesting finding is that the mechanism responsible for this segregation was non-disjunction of chromosomes at mitosis and not mitotic crossing-over as in the case of bleomycin.