TOXICITY OF FERRIC OXIDE AND BENZO[A]PYRENE ALONE OR IN COMBINATION IN RESPIRATORY TRACT OF SPRAGUE DAWLEY RATS

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SUMMARY

The association of small quantities of ferric oxide with Benzo[a]Pyrene (BaP) appears to increase in vivo the toxic effect of BaP. The effect of Fe₂O₃ may be mediated by the recruitment of alveolar macrophages. These cells would contribute to the production of toxic and carcinogenic BaP metabolites and would stimulate development of tumors by producing cellular mediators of inflammation. In order to understand the mechanism of the synergic effect, we have instilled male Sprague Dawley rats 3 weeks of age with a single dose: Fe₂O₃ (3 mg) or BaP (5 mg). Combination Fe₂O₃-BaP (3 mg-3 mg) in 200 μl of physiological saline solution. Control group of identical size (treated with physiological saline solutions and untreated) were used for this study. Animals were sacrificed 48 hours after instillation and a bronchoalveolar lavage (BAL) was performed. With each BAL, we have obtained protein measurement, cells were stained with May-Grünwald-Giemsa method and slides were studied with polarised light. The malonaldehyde (MDA) was measured by High Performance Liquid Chromatography. The PMN elastase determination was performed by IMAC (immune-activation) technology. An automated kinetic method for measuring cathepsins B and L was carried out using a fluorogenic substrate: Z-Phe-Arg-AMC, a specific inhibitor E64 and AMC as an internal standard. After a quantitative Dot-Blot of the samples of BAL, an immunodetection of α₁-antitrypsin (α₁AT) was performed. The inhibitory capacity of α₁AT was determined by an enzymatic reaction with porcine pancreatic elastase. We have observed an increased MDA level for rats intoxicated with Fe₂O₃ (123 %), BaP (31 %) and Fe₂O₃ + BaP (56 %). The levels of PMN elastase and cathepsin B and L were increased: Fe₂O₃ (51–58 %), BaP (52–27 %). This effect was not seen for rats intoxicated by Fe₂O₃ + BaP. The free α₁AT was decreased with the three toxics (Fe₂O₃: 44 % – BaP: 42 % – Fe₂O₃: 41 %). The inhibitory capacity of α₁AT was lower in groups of rats instilled with toxics.

Key words: ferric oxide, benzo[a]pyrene, combined exposure, rat

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