SECONDARY LIPID OXIDATION PRODUCTS IN FOOD AND COLON CANCER

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Introduction

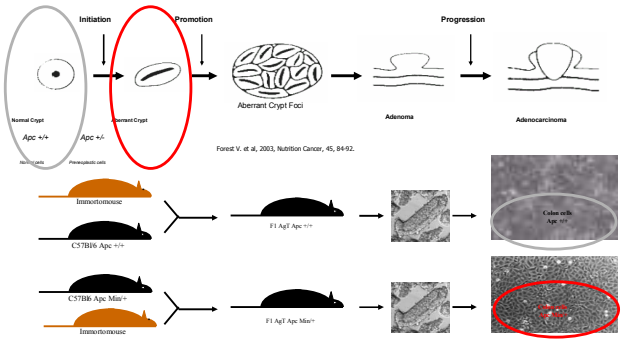
The recent dose-response meta-analysis of epidemiological studies by Norat *et al.* (1) suggests that red meat and processed meat intakes are associated with increased risks of colorectal cancer. The World Cancer Fund International recommends a limited intake of red meat (max: 300g /week) and to avoid processed meat (WCRF 2007).

In a previous work, we have shown that dietary haem and red meat intake in a low-calcium diet with 5% safflower oil promote colon carcinogenesis in rats (2). Oxidative stress and lipid peroxidation are likely the causes of the promotion, as haem effect was inhibited by antioxidants or olive oil . 4-hydroxynonenal (HNE) is considered as the major end product of the oxidation of n-6 polyunsaturated fatty acids. HNE is a cytotoxic and genotoxic compound. It is also found in foodstuffs in concentrations reaching 500 ppm in food fried in thermally oxidized oils (3). The present study was designed to investigate the effects of diets containing haem and n-6 polyunsaturated fatty acids, on the urinary excretion of the major HNE metabolite (DHN-MA, 1,4dihydroxynonene mercapturic acid) and on preneoplastic lesions (MDF, mucin depleted foci). As HNE was shown to be present un colon lumen after feeding those heme rich diets, the cytotoxic and genotoxic effect of HNE on immortalized mouse colon cells lines was investigated too. Those cells were wild type cells (Apc +/+) or Apc mutated cells (Apc min/+). Apc mutation is an early event in human colon carcinogenesis.

Materials and Methods

•In vivo study: different diets (60% casein, chicken, beef or black pudding) on Fisher 344 female rats

•In vitro study : on Apc +/+ or Apc min/+ immortalized mouse colon cell lines



Conclusion

•Excretion of HNE urinary metabolite is associated with heme-rich diets. HNE was present in those diets (not shown).

•Do secondary lipid oxidation products coming from food lipids play a role in colon cancer ?

They have a promoting effect *in vitro* with selection of already mutated cells under luminal « peroxidizing » conditions. In vivo?

Mechanism(s)? Apoptosis? DNA damage? Different biotransformations?

What about 4-hydroxyhexenal (HHE) coming from the oxidation of n-3 fatty acids, malondialdehyde coming from fatty acids bearing more than two insaturations?

Do they reach other tissues, what are the effects on other tissues? : Metabolism and biodisposition need to be studied.

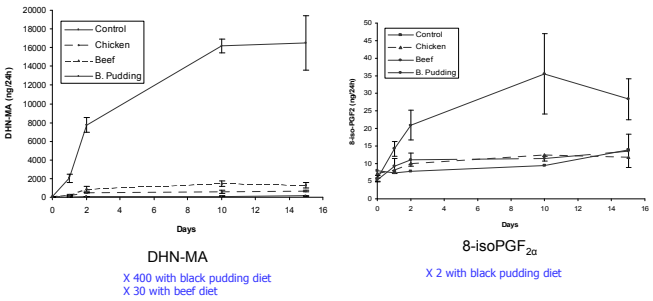
• DHN-MA : a biomarker of food associated with increased colon cancer risk?

Useful but reflects at the same time endogenous+food+ luminal lipid peroxidation

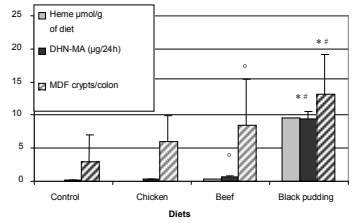
HNE in fecal waters or HNE adducts in colon cells could be pertinent

RESULTS

•In vivo study: different diets (60% casein, chicken, beef or black pudding) on Fisher 344 female rats

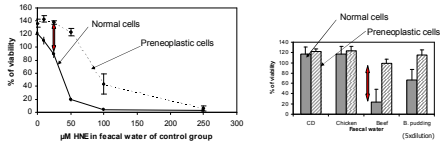


Urinary excretion of DHN-MA (major HNE metabolite) and 8-isoPGF2α



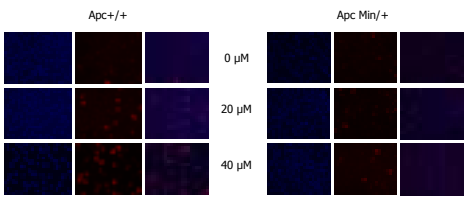
Relationship between heme content of the diet, urinary DHN-MA and colon cancer promotion in rats

•In vitro study : on Apc +/+ or Apc min/+ immortalized mouse colon cell lines

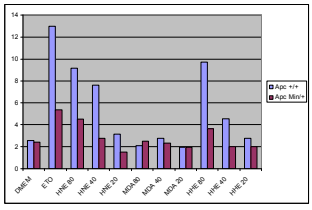


Cytotoxic effect of HNE (left) or of fecal extracts from rats fed the in vivo study diets (right) on colon cell lines using MTT assay

Gamma-H2Ax is a marker of double-strand beaks in genomic DNA



H2AX phosphorylation is dose-dependently increased with HNE and this effect is more pronounced in wild type cells/Apc mutated cells



4-hydroxyalkenals have a differential genotoxic effect on wild type cells compared to Apc mutated cells

Genotoxic effect of HNE (top) or HNE and other lipid peroxidation products (bottom) on colon cell lines using H2AX phosphorylation assay

References: (1) Norat, T., Lukanova, A., Ferrari, P. & Riboli, E. (2002) Meat consumption and colorectal cancer risk: dose-response meta-analysis of epidemiological studies. International Journal of Cancer **98:** 241-256.

(2) Pierre, F., Tache, S., Petit, C. R., Van der Meer, R. & Corpet, D. E. (2003) Meat and cancer: hemoglobin and hemin in a low-calcium diet promote colorectal carcinogenesis at the aberrant crypt stage in rats. Carcinogenesis **24:** 1683-1690.

(3) Seppanen, C.M. and Csallany, A.S. (2004) Incorporation of the toxic aldehyde 4-hydroxy-2-trans-nonenal into food fried in thermally oxidized soybean oil. JAOCS 81,1137-1141.