PAE treatment significantly lowered the number of adenomas in the small intestine by 38% and 40% (avg ± standard error of the mean (SE) control: 64.7. ± 7.2 vs. CAJ: 40.3 ± 5.0, P = 0.011, and PAE: 38.9 ± 2.9, P = 0.007), whereas tumor numbers in the colon were not affected (avg ± SE control: 1.3 ± 0.3, CAJ: 1.8 ± 0.4, PAE: 0.9 ± 0.2). Adenomas were stronger decreased in the middle and the distal part of the SI than in the proximal part. Further, particularly numbers of medium and large adenomas were significantly reduced by both interventions compared to control. Intestinal cell proliferation, determined by immuno-histochemical staining of proliferating cell nuclear antigen, was not influenced by either intervention. Hematocrit values as an indication of intestinal bleeding were negatively correlated with total tumor numbers in both intervention groups (r = -0.82), whereas a positive correlation was observed between spleen weights and adenoma numbers (r = 0.91). Both parameters were slightly ameliorated by the treatments, with significant effects observed with PAE intervention. Overall, our study suggests that CAJ and PAE should be further investigated as part of a prevention strategy for hereditary and sporadic colorectal cancer.

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Cancer chemopreventive and anti-angiogenic activities of xanthohumol from hop (Humulus lupulus L.)

C. Gerhauser[1], R. Hussong[1], E. Bertl[1], K. Klimo[1], N. Frank[1], H. Bartsch[1], H. Becker[2]. [1]*German Cancer Research Center, Toxicology and Cancer Risk Factors, Heidelberg, Germany;* [2]*University of Saarland, Pharmacognosy and Analytical Phytochemistry, Saarbruecken, Germany*

Hop (Humulus lupulus L.) is a good source of phenolic constituents in beer. We investigated a hop extract in a series of test systems indicative of cancer chemopreventive potential and isolated the chalcone xanthohumol (XN) as a most interesting lead structures with high chemopreventive potential at the initiation, promotion and progression stage of carcinogenesis. Although hop is commonly linked with phytoestrogenic effects, we identified XN as a pure estrogen antagonist. Interestingly, XN also affected the generation of estrogens by inhibition of the enzymatic activity of aromatase, which converts testosterone into estrogen. In an uterotrophy assay with prepubertal rats, XN treatment (100 mg/kg bw/day) lowered unstimulated as well as ethinylestradiol-induced uterine weights by about 30%. XN did not cause any adverse effect on female reproduction and on the development of offspring when given either for four weeks prior to or during mating, gestation and nursing. Treatment of male rats prior to mating however significantly (p = 0.027) increased the sex ratio of male to female offspring. Inhibition of angiogenesis represents an innovative approach to cancer chemoprevention. We investigated the angiostatic potential of XN in a human in vitro anti-angiogenic assay premised on the principle of wound healing. We observed dose-dependent reduction of newly formed capillary growth in a concentration range of 0.5 to 10μM. Further mechanistic investigations were performed with HMEC-1, an immortalized human microvascular endothelial cell line. XN effectively inhibited migration of HMEC-1 cells after wounding in a wound closure assay (half-maximal inhibitory concentration IC50=0.03μM). Also, XN effectively inhibited tube-formation on basement membrane matrix at 1, 5 and 10μM, respectively, whereas at 0.1μM, some tubes started to form within the incubation period of 6 h. These effects were only partly due to inhibition of HMEC-1 proliferation, as XN inhibited cell growth with an IC50 of 6.4μM. Xanthohumol also inhibited the transcription of hypoxia-inducible genes under hypoxic conditions. Subcutaneous application of XN (1mg/g body weight) for 14 days to SCID mice bearing human MX-1 breast tumor xenografts significantly reduced the tumor size in treated animals by 82%. Concomitantly, we observed a 30% reduction of tumor-induced neovascularization.

Based on these results, chemopreventive and therapeutic activities of XN will be further investigated.

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HMG CoA reductase inhibitors decrease the risk of pancreatic cancer in US veterans: longer use translated to higher protection

V. Khurana[1], G. Caldito[2], J.S. Barkin[3]. [1]*Overton Brooks VA Medical Center, Gastroenterology, Shreveport, LA, United States of America;* [2]*Lousiana State University HSC, Biometry, Shreveport, LA, United States of America;* [3]*Mt. Sinai Medical Center / University of Miami, Gastroenterology, Miami Beach, FL, United States of America*

Background: HMG CoA reductase inhibitors (Statins) are commonly used cholesterol-lowering agents that are noted to suppress tumor growth in cell cultures and several animal models. The anti-tumor effects of statins are exerted by its antiproliferative, proapoptotic, anti-invasive and radio sensitizing properties. Statins induced apoptosis is predominantly mediated through depletion of geranylgeranylated proteins. Anti invasive effects are mediated by RhoA inactivation.

Methods: US Veterans Health Administration (VHA) is organized into 21 administrative regions called Veterans Integrated Service Networks (VISN). VISN 16 or the South Central (US) VA Health Care Network provides health care treatment to >1.4 million veterans in an eight state region. The network, an integrated health care system, includes ten medical centers, 33 community-based outpatient clinics, seven nursing homes, and two domiciliary. The data was queried from Oct 1998 to June 2004, using a retrospective case control design. Statistical analysis was performed using SAS software version 9.0 (Chicago, IL). Multiple logistic regression analysis was used with calculation of odds ratios and 95% confidence intervals. The data was adjusted for age, race, gender, BMI, smoking, alcohol use and diabetes. Patients were placed in the Statin user group if they were using statin prior to the diagnosis of pancreatic cancer.

Results: Of the 483,733 patients in the study, 163,662 (33.8%) were on statins and 475 (0.1%) patients had a primary diagnosis of pancreatic cancer. Statin use of more than 6 months was associated with a risk reduction of pancreatic cancer of 67% (adjusted OR, 0.33; 95% CI, 0.26 to 0.41; p value <0.01). A dose response relationship was noted between statin use duration and pancreatic cancer with 80% risk reduction (adjusted OR, 0.20; 95% CI, 0.33 to 0.71; p value, <0.01) with use of statin for more than 4 years. Furthermore, the protective effect of statin was seen across different age and racial groups, and was irrespective of the presence of diabetes or alcohol use. The protective benefit was not seen in smokers.

Conclusions: Statins appear to be protective against the development of pancreatic cancer and the magnitude of the effect correlates with the duration of statin usage.

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Circumvention of DNA methylation and histone deacetylation causes re-expression of TSGs leading to apoptosis of breast Ca and chemoprevention of subsequent metastasis

J. Giannos[1], P. Lambrinos[2], N. Alexandropoulos[3]. [1]*PHSA, Dept. of Oncology, Athens, Greece;* [2]*PF, Dept. of Oncology, Athens, Greece;* [3]*GR, Clinical Genetics & Biochemistry, Athens, Greece*

In this study, we prove that DNA methylation is very important in the chemoprevention of metastasis of breast cancer. Generally,promoter hypermethylation is an epigenetic alteration which causes repression of gene transcription in breast cancer cells. We examine tumor suppressor genes,candidate tumor suppressor genes and other genes involved in transcription factors, cell differentiation, apoptosis, growth inhibitory signals, metabolism, antiproliferative signals, replicative senescence,angiogenesis, cell adhesion, tissue invasion, metastasis and other important regulatory proteins which have been transcriptionally inactivated due to aberrant methylation of promoter region CpG islands. We obtain metastatic tumour cells from breast cancer patients and they were implanted in immunosuppressive animals. We obtained DNA from serum, ductal lavage fluid and tumor cells excised by surgery from animals with implanted breast cells and we analysed the specimens with RLGS, MCA-RDA, MS-AP-PCR, DMH, Microarray and Gene re-expression analysis, Bisulfite sequencing, Southern blot, Immunoprecipitation, Western blot and IHC. We analyze the specimens before and after monotherapy with vinorelbine and combined regimen composed of demethylating agent hydralazine and Vinorelbine. Before treatment, we observed promoter hypermethylation and repression of transcription of the following genes: p16 (INK4a/CDKN2A), p73, p15 (INK4b/CDKN2B), p14ARF, CCND2, SFN, RARb2, HIN-1, BRCA1, GSTP1, FABP3, HOXA5, p21WAF1/CIP1/SDI1, E-cadherin (CDH1), TIMP3, MGMT, APC, RASSF1A, NOEY2 (ARHI), RARb2, MDGI, P27KIP1, Gelsolin, 14-3-3 Sigma (Stratifin), Mad, HMLH1, Nm23-H1, 3-OST-2, Maspin, HIC1, MDG1 and Rb. Results remained stable after monotherapy with vinorelbine. In contrast, after combined treatment with Vinorelbine and hydralazine, we observed inhibition of DNA methylation resulting in activation of gene expression which was accompanied by acetylation of histones. This caused decondensation of chromatin allowing access to endogenous proapoptotic endonucleases. Furthermore, hydralazine re-expressed genes relevant for irreversible apoptotic cell death, type D2. Ki-67 exhibited inhibition of tumour proliferation and MTT assay exhibited inhibition of metabolic activity. TEM and SEM exhibited zeiosis, cytoplasmic and nuclear condensation leading to karyorexis with endolytic cleavage of the DNA into small oligonucleosomal fragments which were phagocytosed by macrophages and adjacent tumor cells. MRI exhibited eradication of tumor sites and no sign of metastatic sites. Concluding, the combined administration of Hydralazine and Vinorelbine is responsible for the re-expression of genes characterised by DNA methylation and histone deacetylation leading to eradication of breast cancer cells and offering a promising new chemopreventive option against metastasis of breast cancer.

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Protection against UV-light-induced skin carcinogenesis by topical or dietary administration of broccoli sprout extracts as a source of sulforaphane in SKH-1 high-risk mice

A.T. Dinkova-Kostova[1], S.N. Jenkins[2], J.W. Fahey[3], L. Ye[4], S.L. Wehage[5], P. Talalay[6]. [1]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America;* [2]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America;* [3]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America;* [4]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America;* [5]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America;* [6]*Johns Hopkins University, Pharmacology and Molecular Sciences, Baltimore, MD, United States of America*

The increase in skin cancer incidence is expected to persist due to ozone depletion, increased sun exposure, and longer life expectancy. The effects of UV light contributing to tumor formation are at least three types: (i) inflammation; (ii) direct DNA damage, and (iii) generation of reactive oxygen species. Phase 2 enzymes and glutathione protect against electrophiles and oxidants. Their induction is an efficient strategy for protection against cancer. Sulforaphane, an isothiocyanate isolated from broccoli guided by a bioassay for phase 2 inducer activity, is one of the most potent naturally occurring inducers known. The plant contains a precursor of sulforaphane, the glucosinolate glucoraphanin. Upon injury, glucoraphanin comes in contact with the otherwise compartmentalized myrosinase that catalyzes its hydrolysis to sulforaphane. We tested the hypothesis that topical or dietary administration of broccoli sprout extracts as a source of sulforaphane could protect against the development of skin tumors in mice that were rendered high-risk by chronic exposure to UV light (30 mJ/cm2/session twice a week for 20 weeks). Two extracts (equivalent to 0.3 mmol [low dose] or 1.0 mmol [high dose] sulforaphane, respectively) were applied topically to groups of 30 mice once a day, 5 days a week, for 11 weeks, at which time point all the control animals had tumors. There was a 50% reduction in tumor burden, incidence, and multiplicity in the animals receiving the high dose of protector. Tumor incidence and multiplicity did not differ between the low dose-treated and the control groups, however the low dose treatment provided a substantial reduction in the overall tumor burden. Two extracts (equivalent to 10 mmol/day [low dose] and 50 mmol/day [high dose] glucoraphanin) were incorporated in the diet and the animals were fed for 15 weeks, at which time point 93% of the control mice had tumors. Tumor incidence was reduced by 25% and 35% in the animals receiving the low dose and the high dose of glucoraphanin, respectively. Even greater was the effect of treatment on tumor multiplicity which was reduced by 47% and 72%, respectively: Thus, while the animals in the control group had on the average of 4.3 tumors per mouse, the number of tumors per mouse was 2.3 for the low dose and 1.2 for the high