Vitamin D:

High, K. P., C. Legault, et al. (2002). "Low plasma concentrations of retinol and alpha-tocopherol in hematopoietic stem cell transplant recipients: the effect of mucositis and the risk of infection." Am J Clin Nutr **76**(6): 1358-66.

BACKGROUND: Although vitamin deficiencies are rare in the United States, acute reductions in concentrations of plasma retinol (vitamin A) or alpha-tocopherol (vitamin E) have been associated with impaired immune responses in some clinical settings. OBJECTIVE: The objectives were to determine the plasma concentrations of retinol and alpha-tocopherol in patients undergoing dose-intensive therapy and hematopoietic stem cell transplant and to examine the association of plasma concentrations with clinical outcomes reflecting immunity. DESIGN: **This was an observational trial of 120 consecutive recipients of hematopoietic stem cell transplant and a multivariate analysis of plasma vitamin concentrations, mucositis, infections in the first 30 d, and herpes zoster infections in the first year after hematopoietic stem cell transplant.** RESULTS: Plasma retinol and alpha-tocopherol concentrations declined from baseline to day 7, typically recovering without specific replacement toward baseline by day 14. The severity of mucositis was a strong predictor of low plasma retinol on day 7 (P = 0.001). Eighty-two patients (68%) had at least one plasma retinol concentration < or = 1.05 micro mol/L, a concentration previously determined to be of immunologic significance, during the peritransplant period (day -8 to day 14). Men more frequently acquired herpes zoster than women, and men who developed hyporetinolemia (< or = 1.05 micro mol/L) had a significantly higher risk of herpes zoster (OR: 6.6; 95% CI: 1.5, 29.6). Plasma alpha-tocopherol was not associated with any clinical event measured in this study. CONCLUSION: **Hyporetinolemia is common, particularly in subjects with severe mucositis, and is associated with an increased risk of herpes zoster infection in recipients of hematopoietic stem cell transplant. Additional investigations are required to determine whether these findings indicate a causal relation.**

Vieth, R. (1999). "Vitamin D supplementation, 25-hydroxyvitamin D concentrations, and safety." Am J Clin Nutr **69**(5): 842-56.

For adults, the 5-microg (200 IU) vitamin D recommended dietary allowance may prevent osteomalacia in the absence of sunlight, but more is needed to help prevent osteoporosis and secondary hyperparathyroidism. Other benefits of vitamin D supplementation are implicated epidemiologically: prevention of some cancers, osteoarthritis progression, multiple sclerosis, and hypertension. Total-body sun exposure easily provides the equivalent of 250 microg (10000 IU) vitamin

D/d, suggesting that this is a physiologic limit. Sailors in US submarines are deprived of environmentally acquired vitamin D equivalent to 20-50 microg (800-2000 IU)/d. The assembled data from many vitamin D supplementation studies reveal a curve for vitamin D dose versus serum 25-hydroxyvitamin D [25(OH)D] response that is surprisingly flat up to 250 microg (10000 IU) vitamin D/d. **To ensure that serum 25(OH)D concentrations exceed 100 nmol/L, a total vitamin D supply of 100 microg (4000 IU)/d is required. Except in those with conditions causing hypersensitivity, there is no evidence of adverse effects with serum 25(OH)D concentrations <140 nmol/L, which require a total vitamin D supply of 250 microg (10000 IU)/d to attain**. Published cases of vitamin D toxicity with hypercalcemia, for which the 25(OH)D concentration and vitamin D dose are known, all involve intake of > or = 1000 microg (40000 IU)/d. **Because vitamin D is potentially toxic, intake of >25 microg (1000 IU)/d has been avoided even though the weight of evidence shows that the currently accepted, no observed adverse effect limit of 50 microg (2000 IU)/d is too low by at least 5-fold.**

Hickish, T., D. Cunningham, et al. (1993). "The effect of 1,25-dihydroxyvitamin D3 on lymphoma cell lines and expression of vitamin D receptor in lymphoma." Br J Cancer **68**(4): 668-72.

1,25(OH)2D3 promotes differentiation and has an antiproliferative effect in a variety of cell lines derived from the immunohaematopoetic system. alpha-Calcidol which is metabolised to 1,25(OH)2D3 has been shown to produce tumour regression in follicular low grade non-Hodgkin's lymphoma (NHL) and the dose limiting toxicity is hypercalcaemia. The cellular action of 1,25(OH)2D3 is mediated by binding to an intracellular protein, the vitamin D receptor (VDR). We have evaluated the activity of 1,25(OH)2D3 and its non-calcaemogenic analogue MC903 in the SU-DHL4 and SU-DUL5 B cell lines which carry the 14;18 translocation characteristic of follicular NHL, and also the expression of **the VDR in a range of B cell NHLs.** Both agents induced differentiation and had an antiproliferative effect on the SU-DHL4 and SU-DUL5 cell lines. However this occurred at a relatively high concentration (10(-7) M) which exceeds the physiological concentration of 1,25(OH)2D3 by approximately 10(3)-10(4)-fold. Expression of the VDR was low in each cell line and in the low grade lymphoma tumour samples. **To account for the observed clinical response to 1 alpha OHD3 (alpha-calcidol) in follicular NHL a network is suggested whereby 1,25(OH)2D3 modulates the activity of CD4+T cells which have previously been shown to promote follicle centre cell proliferation. Vitamin D3 analogues may enable serum levels to be achieved which produce a direct action on follicular**

lymphoma cells without disturbing calcium metabolism.

Mawer, E. B., J. Walls, et al. (1997). "Serum 1,25-dihydroxyvitamin D may be related inversely to disease activity in breast cancer patients with bone metastases." J Clin Endocrinol Metab **82**(1): 118-22.

1,25-dihydroxyvitamin D (1,25-(OH)2D) stimulates differentiation and controls proliferation in breast cancer cells. The role of endogenous 1,25-(OH)2D and its relation to PTH related protein (PTHrP) during the progression of breast cancer is not known; we therefore investigated these hormones in two studies. In a cross-sectional study of patients with breast cancer at different stages of disease, serum 1,25-(OH)2D levels (mean +/- SE) were highest in early disease (102 +/- 3.7 pmol/L), fell in normocalemic patients with bone metastases (52 +/- 5.3 pmol/L; $P < 0.01$), and were lowest in hypercalcemic patients (33 +/- 5.6 pmol/L; $P < 0.001$). PTHrP was detectable in the serum of only one normocalcemic patient with progressive metastases but was present in 11 of the 12 hypercalcemic patients, thus PTHrP did not stimulate 1,25-(OH)2D synthesis. In a 6-month longitudinal study of normocalcemic patients with bone metastases undergoing hormonal therapy, serum 1,25-(OH)2D concentrations fell in patients whose disease progressed ($P = 0.0056$), but remained constant in those who were stable or responded to treatment. These changes in 1,25-(OH)2D preceded clinical signs of progression and predicted disease response. In the progressive group, five of whom died during the study, 1,25-(OH)2D decreased between the initial and final samples, PTH fell significantly from 24.8 to 13.5 ng/L ($P = 0.025$), serum calcium rose from 2.27 to 2.39 mmol/L ($P = 0.017$), and the urinary calcium/creatinine ratio rose from 0.37 to 0.68 ($P = 0.046$). PTH and 1,25-(OH)2D were significantly correlated in the final samples from this group, Spearman's rank correlation = 0.80, $P = 0.022$. The results indicate that normocalcemia in these patients is maintained, at the expense of suppressing PTH and 1,25-(OH)2D, in the face of increased calcium released from lytic lesions in bone. Loss of the antiproliferative effects of 1,25-(OH)2D may then permit more rapid secondary growth of the tumor.

Prasad, K. N., J. Edwards-Prasad, et al. (1993). "Vitamins regulate gene expression and induce differentiation and growth inhibition in cancer cells. Their relevance in cancer prevention." Arch Otolaryngol Head Neck Surg **119**(10): 1133-40.

Although several hypotheses for human carcinogenesis have been proposed, the specific genetic changes that cause normal cells to become cancer cells have not been identified**. In spite of uncertainties regarding the mechanisms of carcinogenesis, several vitamins such as beta-**

carotene and vitamins A, C, and E, which can reduce the risk of cancer, have been identified, using animal and in vitro models of carcinogenesis. These studies have led to a hypothesis that the supplemental intake of these vitamins may reduce the risk of cancer. This hypothesis in humans can be tested only by intervention trials that are in progress. Prospective and retrospective case-controlled experimental designs are not suitable for testing the above hypothesis. The fact that some vitamins induce cell differentiation and/or growth inhibition in tumor cells in culture suggests that the use of these vitamins in cancer prevention has a cellular basis. **In addition to having a direct effect on tumor cells, vitamins such as alpha-tocopheryl succinate and beta-carotene enhance the effect of other agents that induce differentiation in tumor cells. Some vitamins like beta-carotene, retinoic acid, alpha-tocopheryl succinate, and vitamin D also regulate the expressions of certain oncogenes and cellular genes.** These are exciting new functions of vitamins that nobody could have predicted only a few years ago.

Whelan, R. L., K. D. Horvath, et al. (1999). "Vitamin and calcium supplement use is associated with decreased adenoma recurrence in patients with a previous history of neoplasia." Dis Colon Rectum **42**(2): 212-7.

INTRODUCTION: Although some have suggested that certain vitamins or calcium supplements may reduce adenoma recurrence, our own prior retrospective study found no such effects. The purpose of this case-control study was to further investigate whether regular vitamin or calcium supplement intake influenced the incidence of recurrent adenomatous polyps in patients with previous neoplasia who were undergoing follow-up colonoscopy. METHODS: This study enrolled 1,162 patients who underwent colonoscopy by one of three surgeons at Columbia-Presbyterian Medical Center in New York City between March 1993 and February 1997. Of these patients 448 (250 males) had a previous diagnosis of colorectal neoplasia (cancer, adenomas, or dysplasia). Of these, 183 (40.8 percent) had an adenoma at the index colonoscopy. Information was collected on personal and family history of colonic diseases, cigarette smoking, medication, and vitamin and micronutrient supplement usage on a questionnaire that was completed by the patients before the colonoscopy. Odds ratios were obtained by unconditional logistic regression analysis, adjusting for age and gender, and used adenoma recurrence at index colonoscopy as the outcome. RESULTS: The mean interval between colonoscopic examinations was 37 months for the recurrent adenoma group and 38 months for the nonrecurrent group of patients (P = not significant). In this case-control study we found a protective effect for the use of vitamin supplements in general (any vitamin) on the recurrence of adenomas (odds ratio, 0.41; 95 percent

confidence interval, 0.27-0.61). Specifically, this protective effect was observed for the use of multivitamins (odds ratio, 0.47; 95 percent confidence interval, 0.31-0.72), vitamin E (odds ratio, 0.62; 95 percent confidence interval, 0.39-0.98), and for calcium supplementation (odds ratio, 0.51; 95 percent confidence interval, 0.27-0.96). Nonsignificant protective effects were noted for carotene/vitamin A, vitamin D, and vitamin C. CONCLUSIONS: **The use of multivitamins, vitamin E, and calcium supplements were found to be associated with a lower incidence of recurrent adenomas in a population of patients with history of previous colonic neoplasia.** Prospective, randomized trials are needed to better assess the impact of these agents and to determine whether the use of these supplements is associated with a protective effect against recurrent adenomas.

Ehinger, M., G. Bergh, et al. (1996). "Expression of the p53 tumor suppressor gene induces differentiation and promotes induction of differentiation by 1,25-dihydroxycholecalciferol in leukemic U-937 cells." Blood **87**(3): 1064-74.

Leukemic U-937 cells, which lack normal p53, were stably transfected with a temperature-sensitive mutant of p53 to investigate the consequences for growth and differentiation. On induction of wild-type p53 activity at the permissive temperature, some of these cells underwent maturation as judged by the capacity for oxidative burst and the appearance of monocyte related cell surface molecules. **Moreover, wild-type p53-expressing cells were more sensitive than p53-negative control cells to induction of differentiation by 1,25-dihydroxycholecalciferol;** a twofold to fourfold increase of the fraction of cells showing signs of terminal maturation was observed when wild-type p53-expressing cells were incubated with 1,25-dihydroxycholecalciferol at concentrations that only slightly affected control cells. Whereas wild-type p53 activity per se induced maturation of certain cells, other underwent cell death judging from the reduced capability to exclude trypan blue and the appearance of fragmented DNA in flow cytometric analysis. The p53-induced cell death could be inhibited by incubation with 1,25-dihydroxy-cholecalciferol, but not all-trans retinoic acid. **Thus, 1,25-dihydroxycholecalciferol, seemed to increase the survival of wild-type p53-expressing cells and to cooperate with wild-type p53 to induce differentiation.** The data imply that p53-mediated maturation in U-937 cells depends on optimal regulation of signals for differentiation, survival and proliferation, and suggest a role for p53 in the differentiation induction of leukemic cells.

Gullberg, U., K. Peetre, et al. (1986). "Differentiation induction in myeloid leukemic cells." Med Oncol Tumor Pharmacother **3**(2): 55-61.

Work based on immortalised leukemic cell lines indicates that the maturation arrest in leukemia can be reversible. Successful differentiation

induction would mean restoring the link between proliferation and differentiation. Human cell lines such as the promyelocytic HL-60 and the monoblastic U-937 can be induced to mature by incubation with a wide variety of agents, e.g. phorbol diesters, retinoic acid and 1,25-dihydroxycholecalciferol. In addition, mitogen-stimulated lymphocytes and some T-lymphocyte lines produce a polypeptide called the differentiation-inducing factor (DIF), which mediates maturation of HL-60 into macrophage-like cells with resulting proliferation inhibition. DIF also displays a primary growth inhibitory effect on certain subclones of the cell lines as well as on fresh clonogenic cells from patients with acute myeloid leukemia and on normal granulocyte-macrophage progenitors. **Our data indicate that there is more than one way to induce differentiation in leukemia but final common pathways may exist. Complementary, synergistic, maturation effects are seen between some agents, which may become of clinical utility.**

Olsson, I., U. Gullberg, et al. (1983). **"Induction of differentiation of the human histiocytic lymphoma cell line U-937 by 1 alpha,25-dihydroxycholecalciferol."** Cancer Res **43**(12 Pt 1): 5862-7.

Some clones of the human histiocytic lymphoma line, U-937, were induced to differentiate into monocyte-like cells with loss of plating efficiency in agar by incubation with 0.1 to 10 nM 1 alpha,25-dihydroxycholecalciferol [1,25(OH)2D3]. At 1 nM, 40% of the cells of one sensitive clone exhibited differentiation after 2 days of incubation judging from assays for phagocytosis and capacity to reduce nitroblue tetrazolium. Induction appeared to occur by binding of the cholecalciferol to a specific cytoplasmic and/or nuclear receptor for 1,25(OH)2D3. However, the presence of this receptor was not sufficient for differentiation, since one clone which contained the receptor did not respond with differentiation upon addition of 1,25(OH)2D3. Differentiation induction did not require DNA synthesis but was blocked by agents which inhibit RNA or protein synthesis. It was also blocked by the calcium ionophore A 23187. **A synergistic inducing effect was seen between 1,25(OH)2D3 and retinoic acid.** In addition, the U-937 cells could be primed by a short incubation with 1,25(OH)2D3 to respond, with maturation, to the addition of agents which increase the intracellular level of cyclic adenosine 3':5'-monophosphate, such as prostaglandin E2, cholera toxin, and N6,O2'-dibutyryl adenosine 3':5'-monophosphate and which alone did not induce differentiation. Priming does not depend on the normal rate of RNA or protein synthesis, since it was not significantly inhibited by actinomycin D, cordycepin, or cycloheximide. It remains to be determined if unoccupied receptors for 1,25(OH)2D3 are present in fresh leukemia cells and if such cells can sometimes be induced to differentiate upon addition of cholecalciferol.