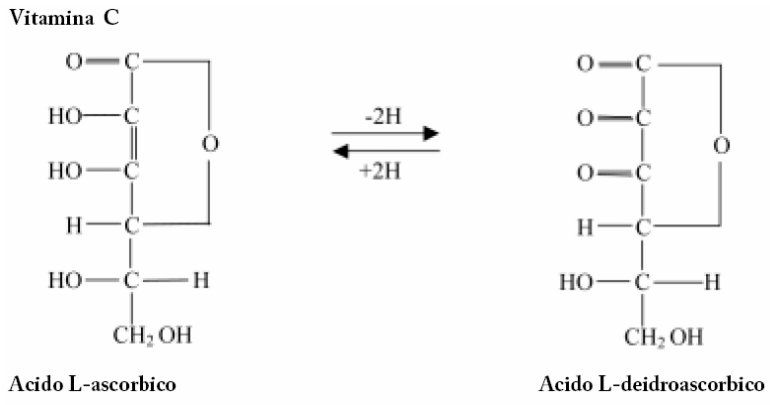
VITAMINA C

IN

MALATTIE LINFOPROLIFERATIVE



Introduzione

Chimicamente la Vitamina C (Vit.C) è definita acido L-ascorbico e presenta strette analogie con gli esosi, cioè i glucidi (zuccheri) con sei atomi di carbonio. La Vit.C è uno dei più importanti agenti riducenti presenti nei tessuti viventi ed è ampiamente distribuita nelle piante. Mentre molti animali sintetizzano la Vit.C, gli esseri umani e pochi altri animali (come i primati non-umani) non sono in grado di sintetizzare Vit.C, che deve quindi essere introdotta con la dieta. La Vit.C è altamente solubile in acqua ed è un forte agente anti-ossidante, che reagisce direttamente con atomi di ossigeno singoli, idrossidi e radicali superossidi (Sauberlich 1994).

La Vit.C viene usata come agente anti-scorbutico, per l'integrità del tessuto connettivo, e può avere un ruolo preventivo e terapeutico in diverse patologie, tra cui le malattie cardiovascolari ed il cancro (Bendich and Langseth 1995).

Diversi meccanismi d'azione dell'attività antineoplastica della Vit.C sono stati riportati (Cameron, Pauling et al. 1979; Head 1998).
La Vit.C previene i danni cellulari indotti da prodotti ossidativi, inclusi i radicali liberi (Padh 1991).
Ci sono evidenze che la Vit.C possa inibire gli effetti carcinogenici prodotti da sostanze mutagene (Aidoo, Lyn-Cook et al. 1994; Lee, Lee et al. 2002).
La Vit.C può inibire il processo metastatico tumorale in diversi modi:

- inducendo la sintesi di collagene (Pinnel, Murad et al. 1987; Peterkofsky 1991);
- inibendo l'azione della ialuronidasi (Cameron and Pauling 1973);
- diminuendo la permeabilità di cellule endoteliali (Utoguchi, Ikeda et al. 1995);
- agendo come un fattore angiostatico sulla proliferazione delle cellule endoteliali (Ashino, Shimamura et al. 2003).

Tutti questi processi contrastano con la capacità delle cellule maligne di invadere gli altri tessuti, prevenendo così il processo di disseminazione neoplastica.

Vari lavori hanno dimostrato che la Vit.C potenzia l'azione antitumorale di diversi farmaci chemioterapici, come il cisplatino, la doxorubicina ed il 5-fluorouracile (Lee and Wurster 1994; Prasad, Hernandez et al. 1994; Kurbacher, Wagner et al. 1996; Nagy, Mucsi et al. 2003). Inoltre, è stato anche riportato che la Vit.C riduce la tossicità di altri agenti chemioterapeutici come l'adriamicina (Fujita, Shinpo et al. 1982; Shimpo, Nagatsu et al. 1991).

Vari studi clinici hanno valutato l'efficacia della somministrazione di Vit.C in pazienti con malattie neoplastiche (Head 1998).

Alcuni studi clinici, condotti in Scozia, hanno riportato che pazienti a cui veniva somministrata la Vit.C avevano un periodo di sopravvivenza medio superiore a quello di pazienti a cui non era data la Vit.C (Cameron and Campbell 1974; Cameron and Pauling 1976; Cameron and Pauling 1978; Cameron 1991).

Anche studi clinici condotti in Giappone hanno confermato l'aumento del periodo di sopravvivenza di pazienti con cancri terminali trattati con Vit.C (Murata, Morishige et al. 1982).

Al contrario, studi clinici condotti negli stati uniti hanno mostrato che la somministrazione di Vit.C non ha benefici in pazienti oncologici (Creagan, Moertel et al. 1979; Moertel, Fleming et al. 1985).

Vitamina C in malattie linfoproliferative

I linfociti umani normali hanno la capacità di concentrare intracellularmente la Vit.C (Levine, Conry-Cantilena et al. 1996), che aiuta a proteggere tali cellule dai danni ossidativi (Ozturk, Mulholland et al. 2001).

Uno studio epidemiologico di fattori che influenzano lo sviluppo di linfoma non-Hodgkin, in uomini e donne in Nebraska (USA), ha trovato una relazione inversa statisticamente significativa tra la quantità di Vit.C, caroteni, verdure ed agrumi consumati e l'incidenza di linfoma non-Hodgkin (Ward, Zahm et al. 1994).

È stata isolata una linea di cellule-T maligne, da un paziente con un linfoma maligno, con la caratteristica di essere sensibile alla Vit.C. Concentrazioni minori di 50 micromol/l uccidevano le cellule nel giro di poche ore (Helgestad, Pettersen et al. 1990).
Sono state successivamente riportate altre linee cellulari di tumori linfocitici, che sono sensibili ad un effetto inibitorio della Vit.C (Kao, Meyer et al. 1993).

La Vit.C ha mostrato anche un'azione di potenziamento della citotossicità di farmaci antineoplastici, in vari tipi di cellule maligne.
Studi in vitro dimostrano che la Vit.C potenzia l'azione del 5-fluorouracile e dell'arsenico triossido in cellule di linfoma (Michel, Dupuy et al. 2003; Nagy, Mucsi et al. 2003).
Anche studi in vivo in topi confermano un effetto sinergico della Vit.C con molecole chemio-terapeutiche in linfomi maligni (Prasad, Giri et al. 1992; Sarna and Bhola 1993).

Poiché gli studi clinici sull'efficacia della Vit.C in pazienti neoplastici, con diversi tipi di tumori, hanno dato risultati contrastanti, non sono stati effettuati finora studi clinici mirati in pazienti con leucemie o linfomi, per valutare un possibile effetto benefico della Vit.C.

I dati riportati indicano che la Vit.C può avere un'azione antitumorale in malattie linfoproliferative maligne, soprattutto se considerata all'interno di un regime terapeutico pluri-farmacologico come è quello della Terapia Di Bella.

BIBLIOGRAFIA

Aidoo, A., L. E. Lyn-Cook, et al. (1994). "Ascorbic acid (vitamin C) modulates the mutagenic effects produced by an alkylating agent in vivo." Environ Mol Mutagen **24**(3): 220-8.

Ashino, H., M. Shimamura, et al. (2003). "Novel function of ascorbic acid as an angiostatic factor." Angiogenesis **6**(4): 259-69.

Bendich, A. and L. Langseth (1995). "The health effects of vitamin C supplementation: a review." J Am Coll Nutr **14**(2): 124-36.

Cameron, E. (1991). "Protocol for the use of vitamin C in the treatment of cancer." Med Hypotheses **36**(3): 190-4.

Cameron, E. and A. Campbell (1974). "The orthomolecular treatment of cancer. II. Clinical trial of high-dose ascorbic acid supplements in advanced human cancer." Chem Biol Interact **9**(4): 285-315.

Cameron, E. and L. Pauling (1973). "Ascorbic acid and the glycosaminoglycans. An orthomolecular approach to cancer and other diseases." Oncology **27**(2): 181-92.

Cameron, E. and L. Pauling (1976). "Supplemental ascorbate in the supportive treatment of cancer: Prolongation of survival times in terminal human cancer." Proc Natl Acad Sci U S A **73**(10): 3685-9.

Cameron, E. and L. Pauling (1978). "Supplemental ascorbate in the supportive treatment of cancer: reevaluation of prolongation of survival times in terminal human cancer." Proc Natl Acad Sci U S A **75**(9): 4538-42.

Cameron, E., L. Pauling, et al. (1979). "Ascorbic acid and cancer: a review." Cancer Res **39**(3): 663-81.

Creagan, E. T., C. G. Moertel, et al. (1979). "Failure of high-dose vitamin C (ascorbic acid) therapy to benefit patients with advanced cancer. A controlled trial." N Engl J Med **301**(13): 687-90.

Fujita, K., K. Shinpo, et al. (1982). "Reduction of adriamycin toxicity by ascorbate in mice and guinea pigs." Cancer Res **42**(1): 309-16.

Head, K. A. (1998). "Ascorbic acid in the prevention and treatment of cancer." Altern Med Rev **3**(3): 174-86.

Helgestad, J., R. Pettersen, et al. (1990). "Characterization of a new malignant human T-cell line (PFI-285) sensitive to ascorbic acid." Eur J Haematol **44**(1): 9-17.

Kao, T. L., W. J. Meyer, 3rd, et al. (1993). "Inhibitory effects of ascorbic acid on growth of leukemic and lymphoma cell lines." Cancer Lett **70**(1-2): 101-6.

Kurbacher, C. M., U. Wagner, et al. (1996). "Ascorbic acid (vitamin C) improves the antineoplastic activity of doxorubicin, cisplatin, and paclitaxel in human breast carcinoma cells in vitro." Cancer Lett **103**(2): 183-9.

Lee, K. W., H. J. Lee, et al. (2002). "Preventive effects of vitamin C on carcinogenesis." Lancet **359**(9301): 172.

Lee, Y. S. and R. D. Wurster (1994). "Potentiation of anti-proliferative effect of nitroprusside by ascorbate in human brain tumor cells." Cancer Lett **78**(1-3): 19-23.

Levine, M., C. Conry-Cantilena, et al. (1996). "Vitamin C pharmacokinetics in healthy volunteers: evidence for a recommended dietary allowance." Proc Natl Acad Sci U S A **93**(8): 3704-9.

Michel, L., A. Dupuy, et al. (2003). "Arsenic trioxide induces apoptosis of cutaneous T cell lymphoma cells: evidence for a partially caspase-independent pathway and potentiation by ascorbic acid (vitamin C)." J Invest Dermatol **121**(4): 881-93.

Moertel, C. G., T. R. Fleming, et al. (1985). "High-dose vitamin C versus placebo in the treatment of patients with advanced cancer who have had no prior chemotherapy. A randomized double-blind comparison." N Engl J Med **312**(3): 137-41.

Murata, A., F. Morishige, et al. (1982). "Prolongation of survival times of terminal cancer patients by administration of large doses of ascorbate." Int J Vitam Nutr Res Suppl **23**: 103-13.

Nagy, B., I. Mucsi, et al. (2003). "Chemosensitizing effect of vitamin C in combination with 5-fluorouracil in vitro." In Vivo **17**(3): 289-92.

Ozturk, G., C. W. Mulholland, et al. (2001). "Vitamin C decreases intracellular calcium level in human lymphoid cells." J Physiol Pharmacol **52**(2): 285-92.

Padh, H. (1991). "Vitamin C: newer insights into its biochemical functions." Nutr Rev **49**(3): 65-70.

Peterkofsky, B. (1991). "Ascorbate requirement for hydroxylation and secretion of procollagen: relationship to inhibition of collagen synthesis in scurvy." Am J Clin Nutr **54**(6 Suppl): 1135S-1140S.

Pinnel, S. R., S. Murad, et al. (1987). "Induction of collagen synthesis by ascorbic acid. A possible mechanism." Arch Dermatol **123**(12): 1684-6.

Prasad, K. N., C. Hernandez, et al. (1994). "Modification of the effect of tamoxifen, cis-platin, DTIC, and interferon-alpha 2b on human melanoma cells in culture by a mixture of vitamins." Nutr Cancer **22**(3): 233-45.

Prasad, S. B., A. Giri, et al. (1992). "Use of subtherapeutical dose of cisplatin and vitamin C against murine Dalton's lymphoma." Pol J Pharmacol Pharm **44**(4): 383-91.

Sarna, S. and R. K. Bhola (1993). "Chemo-immunotherapeutical studies on Dalton's lymphoma in mice using cisplatin and ascorbic acid: synergistic antitumor effect in vivo and in vitro." Arch Immunol Ther Exp (Warsz) **41**(5-6): 327-33.

Sauberlich, H. E. (1994). "Pharmacology of vitamin C." Annu Rev Nutr **14**: 371-91.

Shimpo, K., T. Nagatsu, et al. (1991). "Ascorbic acid and adriamycin toxicity." Am J Clin Nutr **54**(6 Suppl): 1298S-1301S.

Utoguchi, N., K. Ikeda, et al. (1995). "Ascorbic acid stimulates barrier function of cultured endothelial cell monolayer." J Cell Physiol **163**(2): 393-9.

Ward, M. H., S. H. Zahm, et al. (1994). "Dietary factors and non-Hodgkin's lymphoma in Nebraska (United States)." Cancer Causes Control **5**(5): 422-32.

Annu Rev Nutr. 1994;14:371-91.

Pharmacology of vitamin C.

Sauberlich HE.

A better understanding of the functions of ascorbic acid would help clarify the magnitude of the influence of this vitamin on health-related conditions. Many of the purported benefits require confirmation as well as a knowledge of the mechanism of action. The majority of investigations of the association of vitamin C with various types of cancer, with cardiovascular risk, and with cataract formation were epidemiologic studies. Often it was not possible to discern whether the apparent protective effect was due to vitamin C, vitamin E, or carotene, or to a combined effect of these nutrients or of additional factors. Human intervention trials may provide definitive and quantitative assessments of the role of vitamin C in health maintenance. We need to gain a more thorough understanding of the interactions of vitamin C with other nutrients, such as vitamin E and carotenoids, in order to appreciate the role of vitamin C in disease prevention. Investigators are increasingly recognizing the diverse functions of vitamin C in the body in addition to its role in collagen synthesis. However, the functional consequences of these many important roles of vitamin C remain essentially unknown. Excluding scurvy, the health consequences of inadequate vitamin C status are not well characterized. Nonetheless, epidemiologic evidence suggests a role for vitamin C in cancer and heart disease as well as in a number of other diseases.

J Am Coll Nutr. 1995 Apr;14(2):124-36.

The health effects of vitamin C supplementation: a review.

Bendich A, Langseth L.

A comprehensive review of the literature indicates that populations with long-term consumption of higher than RDA levels of vitamin C (> or = 60 mg/day) from foods and/or supplements have reduced risks of cancer at several sites, cardiovascular disease, and cataracts. The safety of higher than RDA intakes of vitamin C is confirmed in eight placebo-controlled, double-blind studies and six non-placebo clinical trials in which up to 10,000 mg of vitamin C was consumed daily for up to 3 years. There are no clinical data which suggest that vitamin C's enhancement of non-heme iron absorption in individuals with low iron status could be a critical factor in the possible increased risk of heterozygous hemochromatosis-related cardiovascular disease. In fact, the cumulative data do not confirm that iron status is related to risk of cardiovascular disease. Moreover, higher than RDA intakes of vitamin C have been associated with several indices of lowered cardiovascular disease risk including increases in HDL, and decreases in LDL oxidation, blood pressure and cardiovascular mortality.

Cancer Res. 1979 Mar;39(3):663-81.

Ascorbic acid and cancer: a review.

Cameron E, Pauling L, Leibovitz B.

Host resistance to neoplastic growth and invasiveness is recognized to be an important factor in determining the occurrence, the progress, and the eventual outcome of every cancer illness. The factors involved in host resistance are briefly reviewed, and the relationship between these factors and ascorbic acid metabolism is presented in detail. It is shown that many factors involved in host resistance to neoplasia are significantly dependent upon the availability of ascorbate.

Altern Med Rev. 1998 Jun;3(3):174-86.

Ascorbic acid in the prevention and treatment of cancer.

Head KA.

Proposed mechanisms of action for ascorbic acid (ascorbate, vitamin C) in the prevention and treatment of cancer include enhancement of the immune system, stimulation of collagen formation necessary for "walling off" tumors, inhibition of hyaluronidase which keeps the ground substance around the tumor intact and prevents metastasis, prevention of oncogenic viruses, correction of an ascorbate deficiency often seen in cancer patients, expedition of wound healing after cancer surgery, enhancement of the effect of certain chemotherapy drugs, reduction of the toxicity of other chemotherapeutic agents such as Adriamycin, prevention of free radical damage, and neutralization of carcinogenic substances. Scottish as well as Japanese studies have pointed to the potential benefit of high dose vitamin C for the treatment of "terminal" cancer. Mayo Clinic studies, however, have contradicted the Scottish and Japanese findings, resulting in accusations of methodological flaws from both sides. Numerous epidemiological studies have pointed to the importance of dietary and supplemental ascorbate in the prevention of various types of cancer including bladder, breast, cervical, colorectal, esophageal, lung, pancreatic, prostate, salivary gland, stomach, leukemia, and non-Hodgkin's lymphoma.

Nutr Rev. 1991 Mar;49(3):65-70.

Vitamin C: newer insights into its biochemical functions.

Padh H.

Ever since the discovery of vitamin C (ascorbic acid), scientists have been intrigued as to how ascorbic acid deficiency can lead to the diverse symptoms exhibited in scurvy. Only in recent years has it been appreciated that ascorbic acid has important functions in many cellular reactions and processes in addition to its role in collagen synthesis. The few such reactions that are understood at the molecular level make it apparent that ascorbic acid does not directly participate in enzyme-catalyzed conversion of substrate to product. Instead, the vitamin regenerates prosthetic metal ions in these enzymes in their required reduced forms. This is in agreement with other antioxidant functions of vitamin C, e.g., scavenging of free radicals. Ascorbate and other antioxidant nutrients are presumed to play a pivotal role in minimizing the damage from oxidative products, including free radicals. This protective function is twofold: the already-oxidized groups in prosthetic centers of enzymes are reduced and the oxidants and free radicals are removed.

Environ Mol Mutagen. 1994;24(3):220-8.

Ascorbic acid (vitamin C) modulates the mutagenic effects produced by an alkylating agent in vivo.

Aidoo A, Lyn-Cook LE, Lensing S, Wamer W.

Recent reports suggest that ascorbic acid (vitamin C) inhibits tumorigenesis as well as exerts a protective effect against mutagenesis in vitro; however, there is no information on its ability to affect gene mutations induced in vivo. In this study, we have investigated the antimutagenic effects of ascorbic acid on the frequency of 6-thioguanine-resistant (6-TGr) T-lymphocytes produced in Fischer 344 rats dosed with the direct-acting alkylating agent, N-ethyl-N-nitrosourea (ENU). The frequency of 6-TGr T-lymphocytes from the spleen measured five weeks after ENU treatment indicated that ENU produced a substantial mutagenic response. Pretreatment and/or post-treatment of rats with ascorbic acid administered in the drinking water appeared to inhibit the response, but the inhibition was statistically significant only when data from the various dosing schedules were pooled. In addition, there was no clear dose-dependency to the inhibitory effect of ascorbic acid. To further evaluate the time effects of the vitamin supplement on ENU mutagenicity, rats were exposed to the mutagen together with ascorbic acid, which was given continuously for the entire duration of the experiment. At specific times after ENU treatment, the frequency of 6-TGr T-cells was determined in lymphocytes isolated from the spleen and the thymus. Time-dependent increases in the frequency of 6-TGr T-cells were observed with ENU treatment; ascorbic acid significantly reduced the ENU-mediated mutagenic responses, most dramatically in the spleen at weeks 6 and 8 ($P < 0.0001$), and to a lesser extent in the thymus ($P < 0.01$ at week 6 and $P < 0.006$ at week 8). Our data suggest that ascorbic acid intake affects the in vivo mutagenicity of ENU, a direct-acting mutagen/carcinogen, and that the reported inhibitory effects of the antioxidant on carcinogenesis may be partially mediated by its effects on mutagenesis. Although it is difficult to extrapolate from rodent studies to humans, the results presented suggest an explanation for epidemiological data that link vitamin C ingestion with decreased cancer risk.

Arch Dermatol. 1987 Dec;123(12):1684-6.

Induction of collagen synthesis by ascorbic acid. A possible mechanism.

Pinnel SR, Murad S, Darr D.

L-Ascorbic acid stimulates procollagen synthesis in cultured human skin fibroblasts without appreciably altering noncollagen protein synthesis. The effect is unrelated to intracellular degradation of newly synthesized procollagen. Levels of mRNA for pro alpha 1(I), pro alpha 2(I), and pro alpha 1(III), measured by hybridization with the corresponding cDNA probes, are elevated in the presence of ascorbic acid, whereas the level of mRNA for fibronectin is unchanged. Levels of functional mRNA for procollagen, measured in a cell-free translation assay, are specifically increased in the presence of ascorbic acid. Thus, ascorbic acid appears to control the expression of three different procollagen genes, each of which is located on a separate chromosome. It is proposed that intracellularly accumulated procollagen in ascorbate deficiency may lead to a translational repression of procollagen synthesis. Ascorbic acid may relieve this block by promoting hydroxyproline formation and, consequently, secretion of procollagen from the cell. The increased level of procollagen mRNA under the influence of ascorbic acid may be secondary to increased synthesis of procollagen polypeptides; the control point may be gene transcription or mRNA degradation.

Am J Clin Nutr. 1991 Dec;54(6 Suppl):1135S-1140S.

Ascorbate requirement for hydroxylation and secretion of procollagen: relationship to inhibition of collagen synthesis in scurvy.

Peterkofsky B.

Vitamin C deficiency is associated with defective connective tissue, particularly in wound healing. Ascorbate is required for hydroxylation of proline residues in procollagen and hydroxyproline stabilizes the collagen triple helical structure. Consequently, ascorbate stimulates procollagen secretion. However, collagen synthesis in ascorbate-deficient guinea pigs is decreased with only moderate effects on proline hydroxylation. Proteoglycan synthesis, which does not require ascorbate, also is decreased and both effects are correlated with the extent of weight loss during scurvy. Fasting, with ascorbate supplementation, produces similar effects. Both functions are inhibited in cells cultured in sera from either scorbutic or starved guinea pigs and inhibition is reversed with insulin-like growth factor (IGF)-I. The inhibitor appears to consist of two IGF-binding proteins induced during vitamin C deficiency and starving and may be responsible for in vivo inhibition of collagen and proteoglycan synthesis.

J Cell Physiol. 1995 May;163(2):393-9.

Ascorbic acid stimulates barrier function of cultured endothelial cell monolayer.

Utoguchi N, Ikeda K, Saeki K, Oka N, Mizuguchi H, Kubo K, Nakagawa S, Mayumi T.

The macromolecular permeability of cultured bovine aortic, bovine venous, and human umbilical vein endothelial cell monolayers was decreased significantly in culture medium containing L-ascorbic acid (Asc Acid; 0.01-0.1 mM) and L-ascorbic acid 2-phosphate (Asc 2-P). Dithiothreitol, which shows reducing activity equivalent to that of Asc Acid, did not affect endothelial permeability. Asc Acid induced a sixfold increase in collagen synthesis by the endothelial cells. The coexistence of L-azetidine 2-carboxylic acid, an inhibitor of collagen synthesis, attenuated the effect of Asc 2-P in a dose-dependent manner. Another collagen synthesis inhibitor, ethyl-3,4-dihydroxybenzoate, also inhibited collagen synthesis and increased endothelial permeability. The decrease in permeability of the endothelial monolayer was dependent on a reduction of the permeability coefficient of the endothelial monolayer. These findings indicate that endothelial barrier function is stimulated by Asc Acid via an increase in collagen synthesis.

Angiogenesis. 2003;6(4):259-69.

Novel function of ascorbic acid as an angiostatic factor.

Ashino H, Shimamura M, Nakajima H, Dombou M, Kawanaka S, Oikawa T, Iwaguchi T, Kawashima S.

Endothelial permeability is increased by vascular endothelial cell growth factor and decreased by antioxidants. Whether or not l-ascorbic acid (Asc), which decreases endothelial permeability by stimulating the endothelial barrier function, is anti-angiogenic (angiostatic) remains unknown. We examined the role of Asc on angiogenesis using two assay systems. At first, the potential role of Asc on four steps of angiogenesis was investigated in cultured bovine microvascular endothelial cells. Asc inhibited the formation of vessel-like tubular structures of endothelial cells cultured on Matrigel; however, it did not decrease the activity of plasminogen activator (PA), which creates the space into which vascular vessels extend. Furthermore, even at high concentrations, Asc did not inhibit either the proliferation or migration of endothelial cell cultures. Secondly, whether Asc inhibited in vivo angiogenesis or not was studied on chick chorioallantoic membrane (CAM) during the 4-6 days of embryogenesis when neovascularization is rapid. It also revealed that angiogenesis was dose-dependently inhibited by Asc from 0.5 micro mol/CAM with half-maximal inhibition at 2.5 micro mol/CAM. Because it was previously reported that the endothelial barrier function decreases permeability via the stimulation of collagen synthesis induced by Asc, we treated CAM with the inhibitor of collagen synthesis, l-azetidine 2-carboxylic acid (AzC). This compound partially attenuated the angiostatic function of Asc on CAM. To understand the involvement of an antioxidant activity in the angiostatic function of Asc, we further examined the effect of glutathione (GSH), which is an endogenous antioxidant, on angiogenesis in CAM and endothelial cells. GSH inhibited CAM angiogenesis, as well as the formation of vessel-like tubular structures of endothelial cell cultures on Matrigel. Both Asc and GSH inhibited hydrogen peroxide (H(2)O(2)) induced tubular morphogenesis. These findings suggest that Asc affects angiogenesis through both its antioxidant properties and the stimulation of collagen synthesis. As the angiostatic activity of Asc may be one of the many effects involved in host resistance to the growth or invasiveness of solid cancer, it may be useful as a supplementary therapy in various angiogenic diseases.

Cancer Lett. 1996 Jun 5;103(2):183-9.

Ascorbic acid (vitamin C) improves the antineoplastic activity of doxorubicin, cisplatin, and paclitaxel in human breast carcinoma cells in vitro.

Kurbacher CM, Wagner U, Kolster B, Andreotti PE, Krebs D, Bruckner HW.

Utilizing a microplate ATP bioluminescence assay, two human breast carcinoma cell lines, MCF-7 and MDA-MB-231, were tested against doxorubicin (DOX), cisplatin (DDP), and paclitaxel (Tx) alone and in combination with ascorbic acid (Vit C). In both cell lines, Vit C exhibited cytotoxic activity at high concentrations (i.e. 10(2)-10(3) microM). Both cell lines also were resistant to DOX. MCF-7 was found to be DDP-resistant, MDA-MB-231 was moderately sensitive to DDP. Both cell lines were strongly sensitive to Tx. Vit C both at non-cytotoxic (1 microM) and moderately cytotoxic concentrations (10(2) microM) improved the cytotoxicity of DOX, DDP, and Tx significantly. Combination effects between Vit C and DDP or Tx were partly synergistic and partly additive or subadditive whereas a consistent synergism was found between Vit C and DOX. The mechanisms by which Vit C potentiates the cytostatics studied are yet unclear and should be evaluated further.

Nutr Cancer. 1994;22(3):233-45.

Modification of the effect of tamoxifen, cis-platin, DTIC, and interferon-alpha 2b on human melanoma cells in culture by a mixture of vitamins.

Prasad KN, Hernandez C, Edwards-Prasad J, Nelson J, Borus T, Robinson WA.

The effect of a mixture of vitamins in modifying the efficacy of commonly used drugs in the treatment of human melanoma has not been studied. Vitamin C and d-alpha-tocopheryl succinate (alpha-TS) alone reduced the growth of human melanoma (SK-30) cells in culture, whereas beta-carotene (BC), 13-cis-retinoic acid (RA), or sodium selenite alone was ineffective. RA caused morphological changes, as evidenced by flattening of cells and formation of short cytoplasmic processes. A mixture of four vitamins (vitamin C, BC, alpha-TS, and RA) was more effective in reducing growth of human melanoma cells than a mixture of three vitamins. The growth-inhibitory effect of cis-platin, decarbazine, tamoxifen, and recombinant interferon-alpha 2b was enhanced by vitamin C alone, a mixture of three vitamins (BC, alpha-TS, and RA), and a mixture of four vitamins (vitamin C, BC, alpha-TS, and RA) that contained 50 micrograms/ml of vitamin C. These data show that a mixture of three or four vitamins can enhance the growth-inhibitory effect of currently used chemotherapeutic agents on human melanoma cells.

Cancer Lett. 1994 Apr 1;78(1-3):19-23.

Potentiation of anti-proliferative effect of nitroprusside by ascorbate in human brain tumor cells.

Lee YS, Wurster RD.

The interactions of nitric oxide (NO) and ascorbate were explored on the control of growth of human brain tumor cells. Sodium nitroprusside, a NO-generating agent, inhibited the growth of SK-N-MC human neuroblastoma cells in a dose-dependent manner. The growth inhibitory effect of nitroprusside was potentiated by sodium ascorbate and inhibited by hemoglobin. Ascorbate-induced potentiation was also observed in U-373 MG human astrocytoma cells. In both tumor cell lines, this potentiation was blocked by catalase, suggesting that hydrogen peroxide may be involved in the potentiation mechanism. In astrocytoma cells, mannitol or deferoxamine also reversed ascorbate-induced potentiation, indicating involvement of hydroxyl radical. These results suggest that the combined treatment with nitroprusside and ascorbate may be a valuable therapeutic strategy for brain tumors.

Cancer Res. 1982 Jan;42(1):309-16.

Reduction of adriamycin toxicity by ascorbate in mice and guinea pigs.

Fujita K, Shinpo K, Yamada K, Sato T, Niimi H, Shamoto M, Nagatsu T, Takeuchi T, Umezawa H.

The effect of ascorbate in reducing Adriamycin toxicity has been examined in mice and guinea pigs. Ascorbate had no effect on the antitumor activity of Adriamycin in mice inoculated with leukemia L1210, but it significantly prolonged the life of mice and guinea pigs treated with Adriamycin. Adriamycin elevated lipid peroxide levels in serum and liver, and ascorbate prevented the elevation. The significant prevention of Adriamycin-induced cardiomyopathy by ascorbate was proved by means of electron microscopy. The earliest alterations of dilation of the sarcoplasmic reticulum and transverse tubular system and the appearance of a large number of cytoplasmic fat droplets, which were seen in cardiac tissue from guinea pigs receiving Adriamycin, were apparently reduced in animals that were treated with ascorbate.

Am J Clin Nutr. 1991 Dec;54(6 Suppl):1298S-1301S.

Ascorbic acid and adriamycin toxicity.

Shimpo K, Nagatsu T, Yamada K, Sato T, Niimi H, Shamoto M, Takeuchi T, Umezawa H, Fujita K.

Adriamycin (ADR) is effective against a wide range of human neoplasms. However, its clinical use is compromised by serious cardiac toxicity, possibly through induction of peroxidation in cardiac lipids. Ascorbic acid, a potent antioxidant, was examined for effect in reducing ADR toxicity in mice and guinea pigs. Ascorbic acid had no effect on the antitumor activity of ADR in mice inoculated with leukemia L1210 or Ehrlich ascites carcinoma, but it significantly prolonged the life of animals treated with ADR. ADR elevated lipid peroxide levels in mouse heart, and ascorbic acid prevented the elevation. The significant prevention of ADR-induced cardiomyopathy in guinea pigs by ascorbic acid was proved by electron microscopy. Ascorbic acid and the derivatives may delay general toxicity of ADR and also prevent the cardiac toxicity. The results also suggest the clinical efficacy of the combined treatment of ADR and ascorbic acid or the derivatives.

Proc Natl Acad Sci U S A. 1976 Oct;73(10):3685-9.

Supplemental ascorbate in the supportive treatment of cancer: Prolongation of survival times in terminal human cancer.

Cameron E, Pauling L.

Ascorbic acid metabolism is associated with a number of mechanisms known to be involved in host resistance to malignant disease. Cancer patients are significantly depleted of ascorbic acid, and in our opinion this demonstrable biochemical characteristic indicates a substantially increased requirement and utilization of this substance to potentiate these various host resistance factors. The results of a clinical trial are presented in which 100 terminal cancer patients were given supplemental ascorbate as part of their routine management. Their progress is compared to that of 1000 similar patients treated identically, but who received no supplemental ascorbate. The mean survival time is more than 4.2 times as great for the ascorbate subjects (more than 210 days) as for the controls (50 days). Analysis of the survival-time curves indicates that deaths occur for about 90% of the ascorbate-treated patients at one-third the rate for the controls and that the other 10% have a much greater survival time, averaging more than 20 times that for the controls. The results clearly indicate that this simple and safe form of medication is of definite value in the treatment of patients with acvanced cancer.

Med Hypotheses. 1991 Nov;36(3):190-4.

Protocol for the use of vitamin C in the treatment of cancer.

Cameron E.

A protocol for the use of vitamin C in the treatment of cancer, developed over a number of years in Vale of Leven Hospital, Scotland, is presented. Clinical experience has shown this protocol to be both safe and efficient. It need not be followed 'to the letter', but provides general guidance to physicians unfamiliar with this therapeutic approach. It recommends that all cancer patients treated in this fashion be given an initial course of intravenous ascorbate followed by a maintenance oral dose to be continued indefinitely thereafter. The importance of continuous as opposed to intermittent administration is emphasized.

Int J Vitam Nutr Res Suppl. 1982;23:103-13.

Prolongation of survival times of terminal cancer patients by administration of large doses of ascorbate.

Murata A, Morishige F, Yamaguchi H.

Clinical trials administering supplemental ascorbate to terminal cancer patients were conducted at two hospitals in Japan. During the period 1973-1977 there were 99 patients with terminal cancer at the Fukuoka Torikai Hospital. The average times of survival after the date of designation as terminal were 43 days for 44 low-ascorbate patients and 246 days for 55 high-ascorbate patients. Three of the high-ascorbate patients were still alive, their average survival being 1550 days, on April 1, 1980. Similar effectiveness of ascorbate was also observed at the Kamioka Kozan Hospital. There were 31 patients with terminal cancer during the period 1975-1979. The average survival times were 48 days for 19 control patients and 115 days for 6 high-ascorbate patients. One of the high-ascorbate patients was still alive, his survival being 215 days. In addition to the increase in survival times, the administration of large doses of ascorbate seemed to improve the quality of life.

N Engl J Med. 1979 Sep 27;301(13):687-90.

Failure of high-dose vitamin C (ascorbic acid) therapy to benefit patients with advanced cancer. A controlled trial.

Creagan ET, Moertel CG, O'Fallon JR, Schutt AJ, O'Connell MJ, Rubin J, Frytak S.

One hundred and fifty patients with advanced cancer participated in a controlled double-blind study to evaluate the effects of high-dose vitamin C on symptoms and survival. Patients were divided randomly into a group that received vitamin C (10 g per day) and one that received a comparably flavored lactose placebo. Sixty evaluable patients received vitamin C and 63 received a placebo. Both groups were similar in age, sex, site of primary tumor, performance score, tumor grade and previous chemotherapy. The two groups showed no appreciable difference in changes in symptoms, performance status, appetite or weight. The median survival for all patients was about seven weeks, and the survival curves essentially overlapped. In this selected group of patients, we were unable to show a therapeutic benefit of high-dose vitamin C treatment.

N Engl J Med. 1985 Jan 17;312(3):137-41.

High-dose vitamin C versus placebo in the treatment of patients with advanced cancer who have had no prior chemotherapy. A randomized double-blind comparison.

Moertel CG, Fleming TR, Creagan ET, Rubin J, O'Connell MJ, Ames MM.

It has been claimed that high-dose vitamin C is beneficial in the treatment of patients with advanced cancer, especially patients who have had no prior chemotherapy. In a double-blind study 100 patients with advanced colorectal cancer were randomly assigned to treatment with either high-dose vitamin C (10 g daily) or placebo. Overall, these patients were in very good general condition, with minimal symptoms. None had received any previous treatment with cytotoxic drugs. Vitamin C therapy showed no advantage over placebo therapy with regard to either the interval between the beginning of treatment and disease progression or patient survival. Among patients with measurable disease, none had objective improvement. On the basis of this and our previous randomized study, it can be concluded that high-dose vitamin C therapy is not effective against advanced malignant disease regardless of whether the patient has had any prior chemotherapy.

Proc Natl Acad Sci U S A. 1996 Apr 16;93(8):3704-9.

Vitamin C pharmacokinetics in healthy volunteers: evidence for a recommended dietary allowance.

Levine M, Conry-Cantilena C, Wang Y, Welch RW, Washko PW, Dhariwal KR, Park JB, Lazarev A, Graumlich JF, King J, Cantilena LR.

Determinants of the recommended dietary allowance (RDA) for vitamin C include the relationship between vitamin C dose and steady-state plasma concentration, bioavailability, urinary excretion, cell concentration, and potential adverse effects. Because current data are inadequate, an in-hospital depletion-repletion study was conducted. Seven healthy volunteers were hospitalized for 4-6 months and consumed a diet containing <5 mg of vitamin C daily. Steady-state plasma and tissue concentrations were determined at seven daily doses of vitamin C from 30 to 2500 mg. Vitamin C steady-state plasma concentrations as a function of dose displayed sigmoid kinetics. The steep portion of the curve occurred between the 30- and 100-mg daily dose, the current RDA of 60 mg daily was on the lower third of the curve, the first dose beyond the sigmoid portion of the curve was 200 mg daily, and complete plasma saturation occurred at 1000 mg daily. Neutrophils, monocytes, and lymphocytes saturated at 100 mg daily and contained concentrations at least 14-fold higher than plasma. Bioavailability was complete for 200 mg of vitamin C as a single dose. No vitamin C was excreted in urine of six of seven volunteers until the 100-mg dose. At single doses of 500 mg and higher, bioavailability declined and the absorbed amount was excreted. Oxalate and urate excretion were elevated at 1000 mg of vitamin C daily compared to lower doses. Based on these data and Institute of Medicine criteria, the current RDA of 60 mg daily should be increased to 200 mg daily, which can be obtained from fruits and vegetables. Safe doses of vitamin C are less than 1000 mg daily, and vitamin C daily doses above 400 mg have no evident value.

J Physiol Pharmacol. 2001 Jun;52(2):285-92.

Vitamin C decreases intracellular calcium level in human lymphoid cells.

Ozturk G, Mulholland CW, Hannigan BM.

Human lymphocytes have low levels of many antioxidant enzymes however they are know to concentrate vitamin C. Cell injury, including oxidative stress effects, is associated with calcium influx so the influence of vitamin C on the maintenance of calcium levels in leukocytes was studied. Incubation of Molt-3 human lymphoblastoid cells with physiologically relevant concentrations of vitamin C and the calcium ionophore A23187 reversed the calcium influx and increased nuclear protein level associated with the ionophore alone. It is concluded that intracellular vitamin C can inhibit calcium influx into leukocytes so helping to minimise cell damage.

Cancer Causes Control. 1994 Sep;5(5):422-32.

Dietary factors and non-Hodgkin's lymphoma in Nebraska (United States).

Ward MH, Zahm SH, Weisenburger DD, Gridley G, Cantor KP, Saal RC, Blair A.

Little is known about dietary factors and non-Hodgkin's lymphoma (NHL) risk, although high intakes of animal protein and milk have been associated with NHL in two previous studies. As part of a population-based case-control study of agricultural and other risk factors for NHL in eastern Nebraska (USA), we examined the self- and proxy-reported frequency of consumption of 30 food items by 385 White men and women with NHL and 1,432 controls. Animal protein intake was not associated significantly with the risk of NHL, however, there was a nonsignificantly elevated risk of NHL among men with high milk consumption. Vitamin C, carotene, citrus fruit, and dark green vegetable intakes were inversely significantly related to the risk of NHL for men, but not for women. Among men, the odds ratios for the highest quartiles of both vitamin C and carotene intake were 0.6 (95% confidence intervals = 0.3-1.0). There were no meaningful differences in the associations of nutrient intakes and NHL risk between B- and T-cell lymphomas and histologic types. Risks for low intakes of vitamin C and carotene were greater among men and women with a family history of cancer, particularly a history of lymphatic or hematopoietic cancer among first-degree relatives.

Eur J Haematol. 1990 Jan;44(1):9-17.

Characterization of a new malignant human T-cell line (PFI-285) sensitive to ascorbic acid.

Helgestad J, Pettersen R, Storm-Mathisen I, Schjerven L, Ulrich K, Smeland EB, Egeland T, Sorskaard D, Brogger A, Hovig T, et al.

A new malignant human T-cell line-labelled PFI-285-has been isolated from a boy with malignant lymphoma. Morphologically, the cells had characteristics of malignant lymphoid cells. The cells presented surface antigens as early cortical lymphocytes and proliferated non-adherently as single cells, independent of T-cell growth factor (IL-2), in liquid culture. The cells had undetectable levels of receptors for IL-2, were not clonogenic in soft agar, but did form tumors in nude mice. Their establishment and continuous growth in vitro was dependent on the number of cells inoculated and on the growth medium used. Cytogenetic alteration, HTLV-1 or reverse transcriptase activity were not detected. The production of known T-cell derived lymphokines such as IL-2, B-cell growth factor(s), alpha-interferon or granulocyte/macrophage colony stimulating or inhibiting factor(s) was not detected. The cells had 5-8% natural killer (NK)-cell activity against NK-cell sensitive target cells (K562) and were not sensitive for NK cells. A most unusual characteristic was the pronounced sensitivity of the cells to ascorbic acid. Concentrations down to 50 mumol/l killed the cells within hours.

Cancer Lett. 1993 Jun 15;70(1-2):101-6.

Inhibitory effects of ascorbic acid on growth of leukemic and lymphoma cell lines.

Kao TL, Meyer WJ 3rd, Post JF.

Vitamin C has been suggested and disputed as an anti-cancer agent. Previous in vitro studies using either primary cell cultures from cancer patients or tumor cell lines have suggested that tumor cells with different lineages may have different sensitivities to ascorbic acid. In this study we report characterization of the effects of ascorbic acid on growth of two ascorbic acid sensitive and one ascorbic acid resistant lymphocyte tumor cell lines. The cytotoxic effects of ascorbic acid on the sensitive cell lines were time and dosage dependent. Furthermore, the energy state of the ascorbic acid sensitive cells was affected by the presence of ascorbic acid before the cells became apparently non-viable, as demonstrated by 31P nuclear magnetic resonance spectroscopy. The existence of these lymphocyte cell lines with varying sensitivities to ascorbic acid may provide a useful model system for further understanding of vitamin C action on cancer cells.

In Vivo. 2003 May-Jun;17(3):289-92.

Chemosensitizing effect of vitamin C in combination with 5-fluorouracil in vitro.

Nagy B, Mucsi I, Molnar J, Varga A, Thurzo L.

The antiproliferative effect and apoptosis-inducing action of 5-fluorouracil (5-FU) in combination with vitamin C were tested in vitro against the chemosensitive mouse lymphoma, the chemoresistant HEp-2 and a human lung fibroblast cell line. Vitamin C itself had no antiproliferative effect on the fibroblasts, but increased the anticancer effect of 5-FU dose-dependently. In the case of the chemoresistant cell line, only a high concentration of vitamin C increased the cytotoxicity of 5-FU. A combination of 5-FU and vitamin C exerted a significantly enhanced apoptotic effect on the mouse lymphoma cell line, whereas for the HEp-2 cell line this effect was less marked and was achieved only at a high concentration of vitamin C. These findings suggest that the administration of a high dose of vitamin C in combination with 5-FU chemotherapy enhances the chemoresponsiveness of cancer cells and serves as a potential sensitizer, especially in chemo-resistant cell lines. One of the mechanisms by which vitamin C potentiates cytostatics could be apoptosis induction.

J Invest Dermatol. 2003 Oct;121(4):881-93.

Arsenic trioxide induces apoptosis of cutaneous T cell lymphoma cells: evidence for a partially caspase-independent pathway and potentiation by ascorbic acid (vitamin C).

Michel L, Dupuy A, Jean-Louis F, Sors A, Poupon J, Viguier M, Musette P, Dubertret L, Degos L, Dombret H, Bachelez H.

Arsenic trioxide (As2O3) displays apoptogenic properties against various types of hematopoietic malignancies. We investigated the effects of As2O3 on the viability of the cutaneous T cell lymphoma cell lines HuT-78, SeAx, and Myla, and of peripheral blood mononuclear cells from patients with Sezary syndrome, by using propidium iodide and annexin-V staining, terminal deoxyuridine triphosphate nick end labeling (TUNEL), cell cycle analysis, mitochondrial transmembrane potential (delta psi(m)) alterations, cytochrome c release, and detection of processed caspase-3. We also report in vivo effects of As2O3 in two patients with cutaneous T cell lymphoma. The results show that As2O3 induces apoptosis of cutaneous T cell lymphoma lines and of Sezary cells from patients in a time- and concentration-dependent manner in vitro, as demonstrated by annexin-V staining, mitochondrial depolarization, and DNA fragmentation. Ascorbic acid 100 microM potentiated As2O3-induced Sezary cell death, whereas interferon-alpha had no synergistic effect. As2O3-induced Sezary cell death involves activation of caspase-3, cleavage of poly(ADP-ribose)polymerase, and cytochrome c release, but was only partially inhibited by the pancaspase inhibitor Z-VAD.fluoromethylketone. Finally, As2O3 was administered to two patients with cutaneous T cell lymphoma, allowing us to obtain a partial response in one case, whereas stability was observed in the second patient. These results demonstrate that As2O3 synergizes with ascorbic acid to induce Sezary cell death at clinically achievable concentrations, through a caspase-partially independent pathway, and provide a rationale for further in vivo studies addressing the therapeutic efficacy of As2O3 in cutaneous T cell lymphoma patients.

Arch Immunol Ther Exp (Warsz). 1993;41(5-6):327-33.

Chemo-immunotherapeutical studies on Dalton's lymphoma in mice using cisplatin and ascorbic acid: synergistic antitumor effect in vivo and in vitro.

Sarna S, Bhola RK.

Combined effects of ascorbic acid (vitamin C) and cisplatin on the growth of Dalton's lymphoma in C3H/He mice was investigated. Chemotherapy with sub-therapeutical dose (3 mg/kg) enabled to increase the survival time of the tumor bearing mice without any tumor free survivors. Ascorbic acid enhances the antitumor effect of cisplatin in vivo resulting in 60/70 day survivors along with tumor free survivors. Ascorbic acid also enhances the efficacy of low dose of cisplatin (5 micrograms/ml) in vitro. Tumor cells incubated with cisplatin and ascorbic acid, when injected into normal mice, exhibited inhibited growth resulting in an increased life span of tumor bearing mice and tumor free survivors. Inoculation of tumor cells incubated with cisplatin (5 micrograms/ml) and different concentrations (25 or 50 micrograms/ml) of ascorbic acid resulted in 30% tumor free mice which was not observed when concentration of cisplatin increased to 10 micrograms/ml in the medium. A possible cause of the enhancement of cisplatin-induced tumor growth inhibition may be the modulation of permeability of tumor cell membrane by ascorbic acid which increases the uptake of cisplatin into tumor cells, making less efficient the DNA repair machinery due to increased efficiency of adduct formation in DNA molecule. Possibly, this effect of ascorbic acid renders cisplatin more effective as an antitumor agent.

Pol J Pharmacol Pharm. 1992 Jul-Aug;44(4):383-91.

Use of subtherapeutical dose of cisplatin and vitamin C against murine Dalton's lymphoma.

Prasad SB, Giri A, Arjun J.

The antitumor activity of subtherapeutical dose of cisplatin and vitamin C combinations was studied against murine Dalton's lymphoma in vivo. The sequence-dependent synergistic antitumor effect of vitamin C and cisplatin was shown to lead to the regression of the tumor resulting in a significant increase in the host survivals with tumor free hosts. Decrease in tumor pH noted in the treated tumor bearing mice and involvement of host's immune system could be an important step in this sequence-dependent antitumor activity of vitamin C and cisplatin.