PYCNOGENOL in chronic venous insufficiency

The aim of our study was to investigate the efficacy of Pycnogenol - a French maritime pine bark extract - in the treatment of chronic venous insufficiency (CVI). The study consisted of a double-blind phase - in which 20 patients were recruited and randomly treated with placebo or Pycnogenol (100 mg x 3/day for 2 months) - and an open phase - in which other 20 patients were treated with Pycnogenol at the same dose schedule. In total, 40 patients were enrolled; 30 of them were treated with Pycnogenol and 10 with placebo. Pycnogenol significantly improved the legs’ heaviness and subcutaneous edema; the venous pressure was also significantly reduced by the Pycnogenol treatment, thus adding further clinical evidence to its therapeutic efficacy in patients with CVI. Pycnogenol was effective, probably by either stabilizing the collagenous subendothelial basal membrane or scavenging the free radicals, or by a combination of these activities. Clinically, capillary leakage, perivascular inflammation and subcutaneous edema were all reduced. The safety of use of Pycnogenol is demonstrated by the lack of side effects or changes in blood biochemistry and hematologic parameters. Pycnogenol can be therefore recommended both for prevention and treatment of CVI and related veno-capillary disturbances. Petrassi C, Mastromarino A, Spartera C. Phytomedicine 2000 Oct;7(5):383-8.

Treatment of vascular retinopathies with Pycnogenol

The aim of our study was to investigate the effects of Pycnogenol on the progression of diabetic retinopathy and other vascular retinal disorders. The study consisted of a double-blind phase in which 20 patients were recruited and randomly treated with placebo or Pycnogenol (50 mg x 3/day for 2 months) and an open phase in which another 20 patients were treated with Pycnogenol at the same dose schedule. In total, 40 patients with diabetes, atherosclerosis and other vascular diseases involving the retina were enrolled; 30 of them were treated with Pycnogenol and 10 with placebo. The results demonstrated a beneficial effect of Pycnogenol on the progression of retinopathy. Without any treatment (placebo) the retinopathy progressively worsened during the trial and the visual acuity significantly decreased; on the contrary, the Pycnogenol-treated patients showed no deterioration of retinal function and a significant recovery of visual acuity was also obtained. The fluorangiography showed an improvement of retinal vascularization and a reduced endothelial permeability and leakage in the Pycnogenol, but not in the placebo-treated, patients. The ophthalmoscopy and the electroretinogram (ERG) also confirmed the beneficial effects of Pycnogenol. The mechanism of action of Pycnogenol may be related to its free radical (FR) scavenging, anti-inflammatory and capillary protective activities. It has been suggested that Pycnogenol may bind to the blood vessel wall proteins and mucopolysaccharides and produce a capillary ‘sealing’ effect, leading to a reduced capillary permeability and oedema formation. Spadea L, Balestrazzi E. Phytother Res 2001 May;15(3):219-23.

Pycnogenol inhibits tumor necrosis factor-alpha-induced nuclear factor kappa B activation and adhesion molecule expression in human vascular endothelial cells

The transcriptional regulatory protein nuclear factor kappa B (NF-kappa B) participates in the control of gene expression of many modulators of inflammatory and immune responses, including vascular cell adhesion molecule-1 (VCAM-1) and intercellular adhesion molecule-1 (ICAM-1). The heightened expression of these adhesion molecules has been reported to play a critical role in atherosclerosis, inflammation, ischemic vascular disorders, diabetes, and cancer metastasis. In the present study, we investigated the effect of pycnogenol, an antioxidant phytochemical, on the activation of NF-kappa B and the induction of VCAM-1 and ICAM-1 in tumor necrosis factor (TNF)-alpha-treated human umbilical vein endothelial cells (HUVECs). Gel-shift analysis of HUVEC demonstrated that pretreatment with pycnogenol exhibited a concentration-dependent suppression of TNF-alpha-induced activation of NF-kappa B. Induction of VCAM-1 and ICAM-1 surface expression by TNF-alpha was dose-dependently reduced by pycnogenol. TNF-alpha significantly increased the release of superoxide anion and hydrogen peroxide from HUVECs. Pycnogenol dose-dependently inhibited their release. The ability of pycnogenol to inhibit NF-kappa B activation and VCAM-1 and ICAM-1 expression suggests that this phytochemical may play an important role in halting or preventing the atherogenic process. Peng Q, Wei Z, Lau BH. Cell Mol Life Sci 2000 May;57(5):834-41.
Breast cancer is the second leading cause of cancer death in women in the United States. The 1999 Cancer Facts and Figures, published by the American Cancer Society, estimates that almost 43,700 women and men will die of breast cancer in the United States. In this study, we compared the response of human breast cancer cells (MCF-7) and normal human mammary cells (MCF-10) to apoptosis in the presence of pycnogenol. Pycnogenol is a mixture of flavonoid compounds extracted from the bark of pine trees. MCF-7 and MCF-10 cells were plated out in culture dishes and grown in medium containing 0, 40, or 80 micrograms pycnogenol/ml culture medium. Cells were harvested at confluence, incubated with DAPI for 15 min and viewed microscopically for evidence of apoptosis. Apoptosis is detectable by morphology, chromatin condensation, nuclear DNA fragmentation, DNA strand breakage or apoptotic bodies. DAPI is a DNA-binding fluorescent dye used to visualize DNA fragmentation. Apoptosis, as detected by DAPI staining, was significantly higher in MCF-7 cells treated with pycnogenol than the untreated cells. The presence of pycnogenol did not significantly alter the number of apoptotic cells in MCF-10 samples. These results suggest that pycnogenol selectively induced death in human mammary cancer cells (MCF-7) and not in normal human mammary MCF-10 cells. Huynh HT, Teel RW. Anticancer Res 2000 Jul-Aug;20(4):2417-20.

Pine bark extract reduces platelet aggregation

The effects of long-term consumption of the bioflavonoid mixture, French maritime pine bark extract (Pycnogenol(R)), were assessed on aggregation of platelets from cigarette smokers and nonsmokers. Previously we showed that a single dose of Pycnogenol(R) reduced platelet aggregation in cigarette smokers in a dose-response fashion. Cigarette smoking increased platelet reactivity aggregation when measured 2 h after smoking the first cigarette of the day. Blood was collected immediately before and 5 min after smoking three cigarettes each. Smoking increased platelet aggregation (1.17 +/- 0.04). However 200 mg Pycnogenol(R)/day, taken 3 h prior to first cigarette for the day for 2 months, significantly (p <.0023) reduced smoke-induced platelet aggregation (0.98 +/- 0.05) to the level of nonsmokers. In a group of 19 nonsmokers, platelet aggregation was measured during in vitro stimulation by platelet aggregation factor (PAF) after 4 or 8 weeks of 200 mg/day of Pycnogenol(R) consumption. Platelet aggregation was significant when induced in vitro by PAF. However, Pycnogenol(R) consumption did not change platelet aggregation, suggesting that Pycnogenol(R)'s regulation of aggregation is by another mechanism. Thromboxane A2 (TXA2) is increased in smokers by release from platelets and rapidly becomes thromboxane B2 (TxB2). Smoking increased TxB2, which was prevented by Pycnogenol(R), lowering TxB2 levels to those of nonsmokers. However, Pycnogenol(R) had no effect on the lower levels of TxB2 in nonsmokers. These observations suggest that Pycnogenol(R) supplementation reduces a risk factor for cardiovascular diseases, that is, platelet aggregation in smokers. The bioflavonoids in Pycnogenol(R) reduced platelet aggregation stimulated by tobacco smoke. Araghi-Niknam M, Hosseini S, Larson D, Rohdewald P, Watson RR. Integr. Med. 2000 Mar 21;2(2):73-77.

Procyanidins extracted from Pinus maritima (Pycnogenol): scavengers of free radical species and modulators of nitrogen monoxide metabolism in activated murine RAW 264.7 macrophages

Nitrogen monoxide (NO) has diverse physiological roles and also contributes to the immune defense against viruses, bacteria, and other parasites. However, excess production of NO is associated with various diseases such as arthritis, diabetes, stroke, septic shock, autoimmune, chronic inflammatory diseases, and atherosclerosis. Cells respond to activating or depressing stimuli by enhancing or inhibiting the expression of the enzymatic machinery that produce NO. Thus, maintenance of a tight regulation of NO production is important for human health. Phytochemicals have been traditionally utilized in ways to treat a family of pathologies that have in common the disregulation of NO production. Here we report the scavenging activity of pycnogenol (the polyphenols containing extract of the bark from Pinus maritima) against reactive oxygen and nitrogen species, and its effects on NO metabolism in the murine macrophages cell line RAW 264.7. Macrophages were activated by the bacterial wall components lipopolysaccharide (LPS) and interferon (IFN-gamma), which induces the expression of large amounts of the enzyme nitric oxide synthase (iNOS). Preincubation of cells with physiological concentrations of Pycnogenol significantly decreased NO generation. It was found that this effect was due to the combination of several different biological activities, i.e., its ROS and NO scavenging activity, inhibition of iNOS activity, and inhibition of iNOS-mRNA expression. These data begin to provide the basis for the conceptual understanding of the biological activity of Pycnogenol and possibly other polyphenolic compounds as therapeutic agents in various human disorders. Virgili F, Kobuchi H, Packer L. Free Radic Biol Med 1998 May;24(7-8):1120-9.

Activity of monomeric, dimeric, and trimeric flavonoids on NO production, TNF-alpha secretion, and NF-kappaB-dependent gene expression in RAW 264.7 macrophages

Flavonoids are potent antioxidants and have been associated with lowering the risk of cardiovascular diseases. In this study, the effect of flavonoids (monomers, dimers and a trimer) as well as French maritime pine bark extract, Pycnogenol, on NO
production, tumor necrosis factor-alpha (TNF-alpha) secretion and nuclear factor (NF)-kappaB activity was compared. Monomers and dimers repressed NO production, TNF-alpha secretion and NF-kappaB-dependent gene expression induced by interferon gamma, whereas the trimeric procyanidin C2 and Pycnogenol enhanced these parameters. In addition, in unstimulated RAW 264.7 macrophages, both procyanidin C2 and Pycnogenol increased TNF-alpha secretion in a concentration- and time-dependent manner. These results demonstrate that procyanidins act as modulators of the immune response in macrophages. Park YC et al. *FEBS Lett* 2000 Jan 14;465(2-3):93-7.

**Immunomodulation by pycnogenol in retrovirus-infected or ethanol-fed mice**

Pycnogenol is a commercial mixture of bioflavonoids that exhibits antioxidative activity. The effects of dietary pycnogenol on immune dysfunction in normal mice as well as those fed ethanol or infected with the LP-BM5 murine retrovirus were determined. The ethanol consumption and retrovirus infection caused abnormalities in the function and/or structure of a broad array of cells involved in humoral and cellular immunity. Pycnogenol enhanced in vitro IL-2 production by mitogen-stimulated splenocytes if its production was suppressed in ethanol-fed or retrovirus-infected mice. Mitogenesis of splenocytes did not show a significant change in mice treated with pycnogenol. It reduced the elevated levels of interleukin-6 produced in vitro by cells from retrovirus infected mice and IL-10 secreted by spleen cells from mice consuming ethanol. Natural killer cell cytotoxicity was increased with pycnogenol treatment. Cheshier JE, Ardestani-Kaboudanian S, Liang B, Araghiniknam M, Chung S, Lane L, Castro A, Watson RR. *Life Sci* 1996;58(5):PL.