



# Nutrient Synergy – A Specific Formulation of Nutrients Containing Lysine, Proline, Ascorbic Acid, and Epigallocatechin Gallate Inhibits Matrix Metalloproteinases Activity and Invasion Potential of Human Cancer Cell Lines (2002)

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## Abstract

### **Introduction:**

One of the hallmarks of cancer is its ability to invade and metastasize to distal organs. Matrix metalloproteinases (MMPs) have been identified as key players in tumor invasion and metastasis. Excessive MMP secretion has been regarded as an index of malignancy, which leads to the degradation of extracellular matrix. Current treatment protocols with chemotherapy and/or radiation, although beneficial, are toxic and have the potential to destroy healthy cells as well. Our approach has been to develop strategies to inhibit cancer development, progression and metastasis using naturally occurring nutrients, which are relatively non-toxic. Lysine and proline are building blocks of collagen fibers that stabilize connective tissue by inhibiting the enzymatic digestion of collagen fibers. Vitamin C is essential for production of collagen and acts as a powerful antioxidant by scavenging free radicals and thereby protects cells from damage. Epigallocatechin gallate (EGCG) is a green tea extract with antioxidant and anticancerogenic properties. It prevents cancer cell invasion by inhibiting MMPs. It is postulated that the combination of these nutrients would exert a very potent synergistic anticancer activity. Based on the above hypothesis, Nutrient Synergy (NS), a mixture of nutrients such as lysine, proline, ascorbic acid and EGCG, was formulated by Dr. Rath Research Institute.

### **Objective:**

In the present study, we investigated the effect of NS on MMP expression, matrix invasion potential and cell proliferation in several human cancer lines those of skin (melanoma), breast (MDA MB-231) and liver (Hep G2). We also studied the effects of NS on normal human dermal fibroblast (NHDF) and on a co-culture of melanoma and NHDF cells.

**Methods and Materials:**

MMP expression was studied by zymography, extracellular matrix invasion by using reconstituted basement membrane (Matrigel) and cytotoxicity/cell proliferation by MTT assay.

**Results:**

NS inhibited the expression of both MMP-2 and MMP-9 in a dose dependent fashion. The expression of both MMP -2 and -9 were significantly inhibited with a concentration of 100 µg/ml of NS and virtually undetectable with a concentration of 1000 µg/ml. NS used at 10 and 100 µg/ml concentrations did not significantly affect cells viability and at 1000 µg/ml it showed cytotoxicity at the range of 10-40% depending on the cell type. The invasion of melanoma cells through Matrigel matrices was inhibited by 20% and 100% at 10 and 50 µg/ml respectively. Similar invasion of MDA MB-231 was also reduced by 50%, 60% and 95% at 10, 50 and 100 µg/ml respectively. When melanoma cells were co-cultured with NHDF cells, NS inhibited the invasion by 30% and 100% with 10 and 50 µg/ml, respectively.

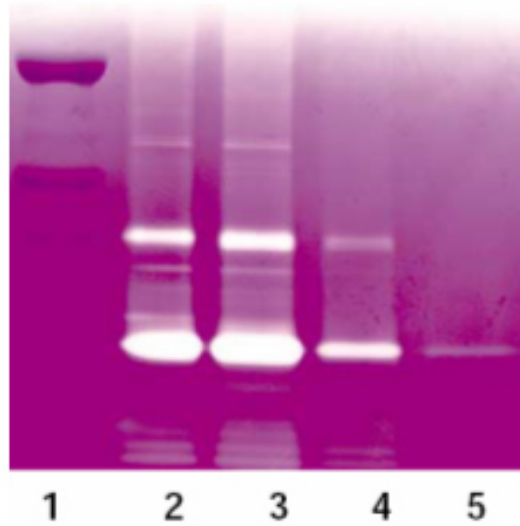
**Conclusions:**

Thus, these results demonstrate that NS is very effective for several cancer cell lines and also in co-culture in inhibiting the expression of MMPs and preventing cellular invasion through Matrigel. These observations revealed that NS may provide a natural therapeutic basis, which makes it a valuable and promising candidate for the treatment of human cancers. Currently, experiments are in progress to evaluate the efficacy of NS in a clinical setting.

**Comment:**

These results demonstrated that the synergistic effect of ascorbic acid, lysine, proline, and epigallocatechin gallate significantly inhibited metastasis potential of human melanoma, breast and liver cancer cells by inhibiting the expression of MMPs and Matrigel invasion, suggesting this non-toxic agent as a promising candidate for the treatment of human cancers.

### Effect of Nutrient Synergy on MMP Expression in Melanoma Cell A2058



1. Markers, 2. Untreated, 3-5. Nutrient Synergy  
10, 100, 1000 µg/ml

### Effect of Nutrient Synergy (NS) on Matrigel Invasion and Migration by Melanoma Cells

