The Effects of Vitamin C on Cancer:

A Cellular and Epidemiological Perspective

Amy C. McLaughlin

A Senior Thesis submitted in partial fulfillment
of the requirements for graduation
in the Honors Program
Liberty University
Spring 2013

Acceptance of Senior Honors Thesis

This Senior Honors Thesis is accepted in partial
fulfillment of the requirements for graduation from the
Honors Program of Liberty University.

_____

Jeffrey L. Lennon, Ph.D.
Thesis Chair

_____

David Titcomb, D.P.T.
Committee Member

_____

Stephen J. Bell, M.A.
Committee Member

_____

James H. Nutter, D.A.
Honors Director

_____

Date

Abstract

While vitamin C has been proven to benefit the immune system during acute infections, there are now many studies to support the findings that vitamin C may also contribute to more effective chemotherapy and lessened damage on the body as a result of chemotherapy. This thesis investigates the impact of vitamin C on many components of cancer such as C-reactive protein, interleukins, reactive oxygen species, and many types of antioxidants and examines vitamin C's ability to inhibit or promote these agents' functions in the body. In addition, this thesis evaluates the ability of vitamin C to keep tumor cells from developing, enhance the antioxidant abilities of other cellular components including vitamin E, lessen the carcinogenic environment produced by chronic inflammation, and diminish the effects of chemotherapy. This thesis concludes by comparing various methods of vitamin C administration and their varied levels of effectiveness in cancer treatment.

The Effects of Vitamin C on Cancer: A Cellular and Epidemiological Perspective

For the past four decades, dietitians and physicians have been promoting the intake of vitamin C to fight rhinoviruses and influenza, assist in nutrient digestion, and maintain stability in bone and connective tissue (Morgan, 2012; National Center for Biotechnology Information, 2012). As an antioxidant, vitamin C can impede the spread of many foreign agents in the body. Because of the effectiveness of vitamin C on many physical processes, especially those having to do with illness, researchers have begun to wonder what other diseases vitamin C could be used to treat. Recently, researchers have devoted a great deal of time to studying the effects of vitamin C on cancer—its effects on cancer cells, natural body cells, and its impact on chemotherapy treatment.

The purpose of this thesis is to determine whether vitamin C is effective in impeding cancer growth and lessening the negative effects of chemotherapy on the body. This will be determined by examining several epidemiological studies on cancer and the effect of vitamin C on the malignant and natural cells on the subjects in each study. Viewing the processes of vitamin C from a cellular level will provide detailed information on its relationship to cancer, chemotherapy, and natural body cells.

Definition of Terms

For the purpose of this study, some terms need to be defined. Ascorbic acid, the chemical name for vitamin C, contains the chemical structure $C_6H_8O_6$ (Hendrickson, 2011). Ascorbate, on the other hand, contains the chemical formula $C_6H_7O_6$. Because the two molecules are so close in structure, they are often used interchangeably, which will be the case in this thesis as well

(Hendrickson, 2011). When oxidation is mentioned in this thesis, it refers to the removal of one of a cell's electrons as it is placed on another cell (Suhail et al., 2012).

In regards to inflammation, four terms need to be defined. Any foreign substance that triggers an immune response is known as an antigen (Merriam Webster Dictionary, 2013). Cytokines are non-antibody proteins that help to produce an immune reaction in response to their interaction with certain antigens (The American Heritage Medical Dictionary, 2007). Chemokines are a type of cytokine that trigger the actions of lymphocytes, and interleukins are another type of cytokine that control the actions of T-cells (The American Heritage Medical Dictionary, 2007).

Vitamin C as an Antioxidant

Vitamin C is an antioxidant, which means that it can "lose a hydrogen atom and…form a relatively stable ascorbate free radical" (Suhail et al., 2012, p. 25). In some situations, however, it can act as a prooxidant, where it accepts a hydrogen atom instead of giving one (Seifried, McDonald, Anderson, Greenwald, & Milner, 2003). Whether vitamin C acts as an antioxidant or a prooxidant depends on its oxidative state (2003).

When other types of cells are oxidized, they often release free radicals such as reactive oxygen species (ROS) (Ratnam, Ankola, Bhardwaj, Sahana, & Ravi Kumar, 2006). While ROS are typically not harmful on their own, they can be converted into more destructive oxidative species and these, when they bind to a natural body cell, can cause the cell's DNA to mutate (Ratnam et al., 2006). This mutation process causes the cell's structure to be damaged and can result in neoplastic transformation (Lee, Lee, Surh, & Lee, 2003), the process by which non-cancerous natural body cells are converted into cancerous tumors (Mosby's Medical Dictionary,

2009). As an antioxidant, ascorbic acid can neutralize these free radicals and keep them from damaging natural cells (Kim et al., 2010; Ratnam et al., 2006).

Two of the most abundant free radicals that cause this type of damage are oxidized versions of glutathione, a non-enzymatic peptide that typically acts as an antioxidant, and MDA, a type of lipid (Suhail et al., 2012). Because ascorbic acid can keep glutathione from becoming oxidized, it can keep the levels of non-oxidized glutathione high in the body, thereby reducing the damage caused by this free radical in its oxidative state. In fact, in Suhail et al.'s study they measured the activity of several different enzymes that scavenge, or neutralize, free radicals. After treating twenty breast cancer patients with both chemotherapy and vitamin C and another twenty with chemotherapy alone, Suhail et al. found that individuals who also supplemented with vitamin C treatment had higher levels of free radical enzymes like superoxide dismutase (SOD), catalase, glutathione-S-transferase, and glutathione reductase, than those who took chemotherapy alone (2012). Lamson et al. found similar results in their study: "A number of human cancers demonstrate low levels of intracellular antioxidant enzymes (catalase, glutathione peroxidase) and smaller antioxidant molecules (glutathione, vitamin E, vitamin C, and vitamin A)" (2010, p.245).

According to Singh & Bhat, "The SODs represent the major cellular defense system against superoxide radicals" (2012, p. 2601). In their study, Singh & Bhat determined the levels of protein expression of SOD1, SOD2, and SOD3. They performed this study by administering vitamin C, vitamin E, BHA, or a combination of the three to female rats with breast cancer. While after 240 days of treatment the levels of SOD3 protein expression had increased in the rats treated with vitamin C and BHA, it had decreased in the rats taking vitamin E. They concluded

that “antioxidants vitamin C (Vit C) and BHA can prevent E2-induced breast cancer in female ACI rats” (p. 2601).

Another component linked to the production of SOD3 is nuclear factor erythroid 2-related factor 2 (NRF2), which according to Singh & Bhat (2012), is “the master regulator of the antioxidant response” and can assist in the production of SOD3 (p. 2601). In addition to measuring the amounts of SOD3 present after the end of the treatment period, Singh & Bhat also measured the amount of NRF2 to determine if vitamin C had an influence on the amount of NRF2 produced. While NRF2 protein expression decreased with vitamin E treatment, it significantly increased with vitamin C and BHA treatment.

Although SOD3 has not been as well researched as SOD1 and SOD2, recent studies indicate that it is most likely linked to preventing DNA damage (Singh & Bhat, 2012). Vitamin C’s ability to increase the levels of SOD3 and NRF2 (which triggers SOD3) in rats with cancer could produce the same effects in epidemiological trials. Based on this study, vitamin C could be beneficial in increasing the rates of SODs in the body, and indirectly increasing the ability of the body to fight tumor cells (Singh & Bhat, 2012).

Lipid peroxidation, the process by which electrons are removed from cell membrane lipids (Northwestern University), is another oxidative process that can cause the development of cancer cells (Mohan & Priya, 2009). According to Mohan & Priya (2009),“lipid peroxidation mediated by free radicals is considered to be the major mechanism of cell membrane destruction and cell damage” (p. 1). In their study, Mohan & Priya (2009) tested thirty-eight patients with ovarian cancer and compared them to thirty-eight controls of similar age. Compared to the controls, the participants with ovarian cancer had lower levels of antioxidants such as ascorbic

acid, GSH, catalase, and vitamin E, and higher levels of lipid peroxidation. These increased levels of lipid peroxidation indicate that more oxidative stress was present in the cells of subjects with cancer. In Suhail et al.'s study (2012), the levels of lipid peroxidation were higher in the forty breast cancer patients who did not receive treatment than in the forty controls. In addition, levels of lipid peroxidation (MDA) were lower in those who received vitamin C and E supplementation than those who received just chemotherapy. Based on these findings, other researchers have determined that consistently high doses of vitamin C could allow the inhibition of lipid peroxidation in cancer patients (Lee et al., 2003).

Vitamin C's effectiveness as an antioxidant is also demonstrated in another study, in which dietary and supplemental intakes of vitamins A, C, and E of 288 healthy controls were compared to 144 women of the same age with cervical cancer. Based on a statistical analysis of the study, "the subjects in the highest quartiles of dietary vitamin A, β-carotene, and vitamin C intakes had statistically significantly lower cervical cancer risk than those in the lowest quartiles" (Kim et al., 2010, p. 183). Other studies also support the idea that vitamin C is inversely associated with lowered cancer risk (Kim et al., 2010).

Dietary Effectiveness of Vitamin C

In addition to vitamin C supplementation, a study measuring the amount of vitamin C in the Italian's natural diet indicates that 130 or more milligrams of vitamin C can also be effective when consumed as part of a daily diet (Bidoli et al., 2009). Evaluating the effectiveness of dietary vitamin C is important because the body can absorb a greater percentage of dietary vitamin C than it can from a supplement (University of Maryland Medical Center, 2011).

One study evaluating this data included 1294 men with prostate cancer and 1451 controls in various regions of Italy. Surveys of their diets were taken to determine how much vitamin C and E they consumed regularly. There was a significant relationship between high intake of vitamin E and lower prostate cancer risk, but only a slight relationship between the same intake of vitamin C and lower cancer risk. However, since the study took place in Italy, most of the participants were consuming a Mediterranean diet high in vitamin E, but not in vitamin C. If they had been eating a diet which incorporated higher levels of vitamin C, the correlation of vitamin C to lowered cancer risk would likely have been stronger (Bidoli et al., 2009). Another study investigating this topic found similar results. After measuring the levels of vitamin C and zinc in 155 Indians suffering from laryngeal cancer, Kapil et al. concluded that vitamin A had the strongest impact on lowering laryngeal cancer risk but that "the people with the highest level of vitamin C intake were at a low risk for laryngeal cancer" as well (Kapil et al., 2003, p.69).

Another example of dietary vitamin C impeding cancer growth is in a study of Iraqi citizens and the incidence of esophageal cancer. The researchers compared the amount of vitamins that the participants consumed daily in their diets with their risk of developing esophageal cancer (Malekshah et al., 2010). At the end of the study, Malekshah et al. determined that the region in Iraq which featured the highest number of cases of esophageal cancer also consumed the lowest amount of vitamins in their diets than all the other regions measured. In addition, the women in the study displayed more cases of esophageal cancer than men, and the percentage of vitamin C intake below RDA was 68% among the women compared to only 55% among the men (Malekshah et al., 2010).

The Effect of Vitamin C on Tumor Cells

Vitamin C's Interaction with p53

Many studies have been performed on ascorbate, a component of vitamin C, and its effect on tumor cells (Kim et al., 2012). Besides directly damaging the chemical make-up of tumor cells, vitamin C can also assist other chemicals in this process and prevent the damage to natural cells that occurs during chemotherapy In fact, Kim et al. (2012) found that "pharmacological [.5 mM and higher] concentrations of ascorbate selectively kill cancer cells but not normal cells, a characteristic of an ideal cancer drug" (p. 1607).

A key element in tumor cells is p53, a transcription factor that modulates stresses to DNA. It can thwart the growth of tumors, halt a tumor cell in the middle of its metabolic cycle, and cause tumor cells to die (Kim et al., 2012). In a recent study, the researchers tested levels of p53 on two types of tumor cells while adding the same amount of ascorbate to the two tumor cells. Next, they measured the levels of p53 in each tumor cell and compared them to the effectiveness of ascorbate in damaging each tumor cell. When this study was performed in vitro, the tumor cells that had higher levels of p53 responded better to the cytotoxicity caused by ascorbate than the cells with lower levels of p53. In both types of tumor cells, the results were similar. This experiment was repeated in vivo on live mice, with similar results: tumors that did not have p53 present increased in size three times as much as the tumor cells that had p53 present (Kim et al., 2012).

Based on these results, p53 has been shown to increase cellular oxidative stress, which helps to destroy cancer cells or at least to inhibit their growth, especially when combined with the oxidative properties of ascorbate. Ascorbate cytotoxicity seems to be more effective in cells

with higher levels of p53 because p53 is an agent that moderates oxidative stress. Because natural body cells have lower levels of p53 than tumor cells, ascorbate does not damage these cells as much (Kim et al, 2012). The reactivation of p53 seems to maintain the cytotoxicity of ascorbate more consistently in cancer treatments. The effectiveness of vitamin C in destroying tumor cells when combined with p53, and the relatively small damage that it causes to natural cells, makes it a viable area of therapy to be tested in the battle against tumor cells (Kim et al., 2012).

Vitamin C's Interaction with Hydrogen Peroxide

Kim et al. continued their study by measuring the amount of hydrogen peroxide that ascorbate produces. Hydrogen peroxide, which can be made from the transformation of superoxide radicals (Singh & Bhat, 2012), has been reported to damage tumor cells' DNA (Quillin, 2005). When ascorbate was added to cancer cells to determine if ascorbate increases the amount of hydrogen peroxide produced, the amount of hydrogen peroxide increased in one type of cancer cell, HCT116+/+, but not in the other, HCT116-/-. In general, hydrogen peroxide increased more in HCT116-/- cells that also had p53 than in those that did not (Singh & Bhat, 2012). Similarly, in *Beating Cancer with Nutrition,* Quillin (2005) found that vitamin C produces a large amount of hydrogen peroxide.

However, a concern of hydrogen peroxide is that it could also damage natural body cells, producing mutations that could lead to cancer (Lee, Lee, Surh, & Lee, 2003). Two cellular-level processes can keep this from happening. One is that, although vitamin C can trigger the production of greater amounts of hydrogen peroxide to attack cancer cells, it can also indirectly reduce the production of hydrogen peroxide through its interaction with free ferric iron.

According to Lee et al, free ferric iron triggers the production of hydrogen peroxide, which can quickly mutate cancer cells and cause them to become malignant. However, in their in vitro study of vitamin C and cancer, Lee et al. found that vitamin C controls levels of free ferric iron, therefore preventing it from producing hydrogen peroxide and causing mutations. Lee et al. conclude that "vitamin C was found to predominantly reduce oxidative damage in vivo even in the presence of iron" (Lee et al., 2003, p. 1075).

Secondly, the properties of hydrogen peroxide can prove useful in cancer treatments because natural cells neutralize hydrogen peroxide in a way that tumor cells cannot (Quillin, 2005). In the body's natural cells, the enzyme catalase interacts with excess hydrogen peroxide and neutralizes it, preventing cell damage (Quillin, 2005; Singh & Bhat, 2012). However, cancer cells do not contain catalase, so excess hydrogen peroxide goes uninhibited, causing more damage (Quillin, 2005). This process can also be repeated with peroxidase, another enzyme that extracts hydrogen peroxide from natural cells (Singh & Bhat, 2012).

Vitamin C's Interaction with Reactive Oxygen Species

In addition to its interaction with p53 and hydrogen peroxide, ascorbate can also activate reactive oxygen species (ROS) which can damage DNA and cause cell death, although the extent to which ROS is beneficial in causing tumor cell death has been difficulty to study (Mamede et al., 2011; National Cancer Institute). According to Inoue et al. (2009), the amount of ROS present in a cell is critical to its ability to cause apoptosis, or cell death. While some of the results varied, many of the results gathered in Inoue et al.'s study of ROS assistance in tumor cell death showed that proteins such as p21 and p53 were more effective in their destruction of tumor cells when coupled with ROS (2009). Because ascorbate as an antioxidant tends to increase the

effectiveness of p53, higher levels of ROS in the cell could also make ascorbate more effective. According to Mamede et al. (2011), when ascorbate was combined with ROS scavenging agent NAC, the cells responded more strongly to the ascorbate than they did with the ascorbate alone.

Another study validating the influence of vitamin C on tumor damage was an in vivo study on rats completed by Velanganni & Balasundaram (2010). In their study, the researchers injected a group of rats with p-dimethylaminoazobenzene (DAB), which resulted in the development of hepatoma, or liver cancer, in the rats. For six months, the researchers administered vitamin E, L-ascorbic acid, and vitamin A to the variable group. When the effects of the vitamins on the two groups were measured, the liver size did not enlarge as greatly in the rats treated with vitamins as in the ones who were not treated with vitamins. In addition, the DNA and RNA multiplied less in the tumors of the vitamin-treated rats than in the rats not treated with vitamins: DNA increased by 4.2 milligrams and RNA by 7.22 milligrams in the untreated rats, but only by 1.8 and 4.32 milligrams respectively in the treated rats (Velanganni & Balasundaram, 2010).

The Effect of Vitamin C on Inflammation

Inflammation can be a major factor in cancer development (Federico, Morgillo, Tuccillo, Ciardiello, & Loguercio, 2007). According to Mikirova, Casciari, Rogers, & Taylor (2012), inflammation not only affects the growth of tumors, but also facilitates “tumour proliferation, angiogenesis, metastasis, and resistance to therapy” (p. 189). This facilitation is due to increasing levels of free radicals involved in inflammation, and their ability to decrease immune function (Kim et al., 2010).

Many types of cancer, including colon carcinoma and liver cancer, result from chronic inflammatory diseases (Aggarwal, Vijayalekshmi, & Sung, 2009; Federico et al., 2007). In the case of acute infections, vitamin C helps to produce inflammation so that the infection can be contained (Aggarwal et al., 2009). This occurs when vitamin C triggers more T-lymphocytes to be produced, which in turn causes more cytokines and immunoglobulins to be produced (Hartel, Strunk, Bucsky, & Schultz, 2004). However, a different process occurs when inflammation is present long-term (Aggarwal et al., 2009).

In long-term diseases, inflammatory cells produce a large number of oxygen free radicals which stay in the body for a longer period of time than they would in a short-term illness (Madhuri, Vani, Syed, & Alshatwi, 2011). This process is a result of an antigen interacting with an inflammatory cell, causing reactive oxygen species to be released into the body (Madhuri et al., 2011). Because many studies now support the idea that reactive oxygen species aid in cancer cells development, the treatment of inflammation via vitamin C therapy could prove vital in inhibiting cancer cell growth (Madhuri et al., 2011). In instances of chronic inflammation, Vitamin C has been shown to impede the release of several types of cells involved in inflammation, including C-reactive proteins, interleukin-6, tumor necrosis factor-alpha (TNF-□), and certain types of reactive oxygen species (Mikrova et al., 2012; Hartel et al., 2004; Federico et al., 2007).

According to Mikirova et al. (2012), a cell unit called C-reactive protein (CRP) accompanies inflammation. It is produced when high levels of the cytokine IL-6 are present, and can be used to identify how much of an infection is present. The authors indicate that “there are particularly strong negative correlations between CRP levels and cancer survival in a wide

variety of cancer types" (p. 190). Based on these findings, Mikirova et al. (2012) measured the amount of CRP before and after 45 patients were treated with intravenous vitamin C therapy. The patients had various types of cancer, including prostate cancer, breast cancer, pancreatic cancer, lung cancer, thyroid cancer, and B-cell lymphoma. Seventy-five percent of the patients experienced a reduction in the amount of CRP compared to their levels of CRP before beginning intravenous vitamin C therapy (IVC). In addition, half of the patients who had a high level of CRP before beginning treatment experienced a reduction in their level of CRP after the IVC.

In a study completed by Hartel, Strunk, Bucsky, & Schultz (2004), which measured how much the production of intracytoplasmic cytokine production was affected by vitamin C, the monocytes that produce interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-□) were inhibited from producing these types of cytokines after doses of vitamin C were administered (Hartel et al., 2004). Because TNF-□, IFN-□, IL-1, IL-6, and IL-8 are all cytokines that work together to create an immune response, without the assistance of each type of cytokine, the immune response is not as strong (Hartel et al., 2004). In addition, based on the findings of Mikirova et al. (2012), a decreased production of IL-6 would result in a decreased production of CRP, and therefore can benefit the cancer patient's prognosis.

Two other cell units involved in the inflammation process are reactive oxygen intermediates (ROI) and reactive nitrogen intermediates (RNI) (Federico et al., 2007). These free radicals are two intermediate factors in the metabolic process which can damage a portion of cells either directly or indirectly. Normally ROI and RNI are reabsorbed during the metabolic process through the actions of antioxidants such as superoxide dismutase, catalase, glutathione peroxidase, and glutathione (GSH), which keep the body in homeostasis and therefore unaffected

by excess ROI and RNI free radicals. However, when this system of homeostasis gets out of balance and more ROI and RNI are being produced than are being absorbed, which results in oxidative and nitrosative stress. This stress can cause the apoptosis or regeneration of normal body cells as well as trigger mutations that lead to the development of tumor cells (Federico et al., 2007; Lee et al., 2003). Because ROI and RNI recruit additional inflammatory cells, such as cytokines, which have harmful effects on natural body cells, they can also indirectly contribute to further cell damage (Federico et al., 2007).

Similar to its effects on other reactive oxygen species, high doses of vitamin C have been shown to neutralize the oxidative effects of ROI and RNI in the body, inhibiting these free radicals' ability to cause DNA damage (Lee et al., 2003). According to Kapil et al. (2004), "[vitamin C] may prevent cancer through its ability to scavenge and reduce nitrate, thereby reducing the substrate for the reaction with secondary amines to form nitrosamines" (p. 69). With its ability to scavenge free radicals and cause chemical reactions to occur among them, vitamin C can effectively inhibit processes that destroy natural DNA, of which RNI are a part (Lee et al., 2003).

In addition to the damage that chronic inflammation can cause on natural body cells, the components of chronic inflammation can also provide an environment for tumor growth (Federico et al., 2007). One of the factors found to be active in most cancers is nuclear factor-kB (NFkB), which according to Aggarwal et al. is "a nuclear factor that binds to the enhancer region of the kB chain of immunoglobulin in B cells" (2009, p. 425). NFkB regulates process such as inflammation, angiogenesis, and metastasis, and in turn can be activated by factors that trigger

inflammation, such as carcinogens, hyperglycemia, and radiation. Cytokines such as TNF and IL-1 also activate NFkB (Aggarwal et al., 2009).

Although it is typically involved in the growth process of most cancers, NFkB could actually assist in protecting natural cells against cancer (Lee et al., 2003; Owuor & Kong, 2002). In small amounts, NFkB contributes to the detoxification of antioxidants, but can also contribute to cancer formation in larger amounts (Lee et al., 2003). In their study, Lee et al. postulated that if ascorbate could replace NFkB completely in this detoxification process, the damaging effects of high levels of NFkB would no longer be a concern (2003).

Vitamin C and Chemotherapy

Lessened Damage on Natural Cells' DNA

In several instances, vitamin C has been shown to lessen the damage on natural cells while a cancer patient is undergoing chemotherapy. In a study completed by Madhuri et al. (2011), 80 patients with gastric cancer were treated with both chemotherapy and vitamin C treatment to determine if the number of changes to natural lymphocytic chromosomes was less in those who had taken vitamin C therapy and chemotherapy compared to those who had undergone chemotherapy alone. Patients who underwent vitamin C therapy as well as chemotherapy were found to have much smaller amounts of chromosomal aberrations than those who took chemotherapy alone. Antunes et al. produced similar results when they tested the number of chromosomal aberrations after chemotherapy with and without vitamin C: those who received vitamin C therapy in addition to chemotherapy had a lower number of chromosomal aberrations than those who did not (Madhuri et al., 2011). In another study which investigated a similar protocol with breast cancer patients, the researchers found that the patients who were

administered both vitamin C & E therapy (VCE) and chemotherapy had lower levels of DNA damage than those who were administered chemotherapy alone (Suhail et al., 2012).

In addition to chromosomal aberrations, 500 mg doses of vitamin C can also protect against the harmful effects of 8-oxoguanine, an oxidized form of DNA, on natural DNA. 8-oxoguanine has been found to be mutagenic to natural cells' DNA (Lee et al., 2003). In a study by Podmore et al. (1998), the researchers found that the cancer patients who took 500 milligrams of vitamin C daily experienced a decrease of 8-oxoguanine concentrations compared to those who were not taking vitamin C supplements (Lee et al., 2003). In another study results were similar: there was a negative correlation between vitamin C dosage and the level of 8-oxo-deoxyguanosine, which can also be harmful to DNA (Lee et al., 2003).

Lessened Side Effects of Chemotherapy

Besides preventing direct damage to the DNA caused by chemotherapy, vitamin C can also maintain antioxidant levels in the body after a chemotherapy treatment (Ratnam et al., 2006). This could make vitamin C a better choice for chemotherapy treatment than cisplatin, which is currently one of the most effective chemotherapeutic agents used to treat cancerous tumors (Weijl et al., 2004). While it has many positive attributes including its ability to effectively attack cancer cells without many negative side effects, one of cisplatin's side effects is the extent to which it decreases plasma concentrations of antioxidants in the body, a result that is almost non-existent when vitamin C is used (Ratnam et al., 2006).

In the study performed by this group of researchers, Weijl et al. administered supplements of vitamins C and E with selenium to some patients, and placebos to others. The study consisted of 48 subjects who had cancers such as testicular cancer, osteosarcoma,

gastrointestinal cancer, melanoma, urogenital cancer, and head and neck cancer. Twenty-five of the study participants received the supplemental treatment while 23 received the placebo, and each group had an equal number of cancer types. When the groups reached the nadir day of cycle 1, both groups' plasma concentrations of antioxidants were lower than when they had begun. However, after the nadir day of cycle 1, the levels of antioxidants in patients taking supplements began to rise, while the levels in the placebo group did not (Weijl et al., 2004).

A part of Weijl et al.'s (2004) study that fails to show correlation is the fact that after chemotherapy was terminated, the levels of antioxidants in the two groups were not statistically different. However, because 64% of the group taking the supplements failed to take their supplements consistently, these results could be skewed. The authors believed that if that group had been more consistent in taking their supplements, the results would have been found more significant.

When the results of Weijl et al.'s (2004) study are combined with the information already known about the benefits of antioxidants on tumor damage and protection of natural cells, further benefits of vitamin C in cancer treatment can be seen. Because vitamin C can also stay in the body in small amounts and be replenished quickly during chemotherapy treatments, the body does not have to fight the cancer for long before new waves of antioxidants can assist in the cancer-fighting process once again.

Madhuri et al. (2011) report another aspect in which vitamin C can curb the effects of chemotherapy on further cancer growth. In their study they measured vitamin C intake in cancer patients and the amount of chromosomal aberrations in those patients' cells. While Madhuri et al. state that normally unstable chromosome order can be a principal cause of cancer cell formation,

both in the beginnings of cancer as well as in metastasis, the results of their study indicate that vitamin C may be effective in decreasing the number of chromosomal changes that occur in the cell.

The Effectiveness of Vitamin C in Combination with Vitamin E

While vitamin C has been shown to be effective on its own, a study by Kim et al. found that vitamin C can increase its effects by working in cohesion with vitamin E. According to Kim et al. (2010), "Vitamin C…has an important function in sparing vitamin E, which is a lipid soluble antioxidant" (p. 188). Because vitamin C spares vitamin E, it can help direct the activity and processes that this vitamin performs, which are often similar to the effects vitamin C could perform on its own (2010). Many studies claim that vitamin C working synergistically with vitamin E can produce more effective damage to tumor cells than can vitamin C alone (Greenlee et al., 2012; Kim et al., 2010; Weijl et al., 2004).

In a study by Greenlee et al. (2012), the results of a synergism between vitamin C and vitamin E were supportive of this idea. In their study, Greenlee et al. evaluated 2264 women with breast cancer. Of the 2264 women, 81% received at least one form of antioxidant supplement, 40% took vitamin C supplements, 48% took vitamin E supplements, and 70% used multivitamin supplements, which typically contained both vitamin E and vitamin C. Greenlee et al. found that the women who took vitamin C supplements in combination with vitamin E supplements tended to have lower rates of all-cause mortality and a lower risk of the breast cancer recurring than those who did not take vitamin C supplements.

Effective Doses of Vitamin C for Cancer Cell Cytotoxicity

In studies performed in vitro, in vivo, and in live population samples, vitamin C has been shown effective in inhibiting the growth of tumors and protecting the body's own cells. In many of the studies the patients only experienced the effectiveness of vitamin C at a specific dosage (Hartel et al., 2004; Huang, Helzlsouer, & Appel, 2000; Kim et al., 2012; Lee et al., 2003). Because the effectiveness of vitamin C is just beginning to be demonstrated in cancer treatment, only a small percentage of all of the studies on this topic have been able to determine effective dosages. However, more studies have been performed monitoring the activity of vitamin C against harmful cytokines, damage to the DNA of natural cells, and modulation of transcription factors, in which researchers have been able to determine effective dosages (Hartel et al., 2004; Lee et al., 2003; Podmore et al., 1998).

Some researchers claim that patients who take doses of 500 or more milligrams of vitamin C a day actually experience damage to their natural cells' DNA (Lee et al., 2003). However, Huang et al. concluded that any damage that vitamin C could cause at a dose of 500 milligrams a day would not be enough to cause neoplastic transformation (2000). In another study, vitamin C supplementation at the levels of 5000 milligrams a day was not enough to cause DNA damage or cancer formation (Lee et al., 2003). According to Hartel et al., who completed a study about vitamin C and its effects on the long-term inflammation process, a twenty milliMolar concentration of ascorbate kept IL-6 and TNF- □ from being produced, while leaving other levels of interleukins unaffected (2004). While these studies have demonstrated specific doses at which vitamin C is effective in specific situations, Kim et al. (2012) indicate that it is hard to get

a high enough amount of ascorbate through oral dosing, but intravenous doses, since they are higher, can be effective in causing cancer cell death (National Institutes of Health, 2011).

Some case studies support the effectiveness of intravenous doses of vitamin in cancer treatment. For example, in a case study of a 49-year-old man with a bladder tumor, the man was treated with 30 grams of intravenous vitamin C for three months, two times a week. After three months, he continued to receive 30 grams of intravenous vitamin C once every 1-2 months for four years. Nine years after being diagnosed with the bladder tumor, this man was cancer-free and did not exhibit signs of relapse or metastasis; ordinarily, this type of tumor would have quickly metastasized into other parts of the body.

Similarly, in another case study, a 66-year-old woman with lymphoma received 15 grams of vitamin C intravenously twice a week for nine months, and then received the same amount every 2-3 months for the next year. While she did receive radiation treatments in addition to vitamin C therapy, she denied chemotherapy for the whole treatment phase. Additional lymph growths initially appeared. However, two years later the growths had all been eliminated and the woman was cancer-free (Padayatty et al., 2006).

While specific dosage is important, combining vitamin C with other types of chemotherapeutic drugs can also make the vitamin C more effective. In their study measuring how p-53 affects the ability of vitamin C to damage tumor cells, Kim et al. (2012) concluded that although vitamin C could cause significant damage to tumor cells, “a single treatment of ascorbate was not enough for complete regression of the tumor size” (p. 1614). In fact, the authors suggested combining vitamin C therapy with another type of chemotherapeutic treatment so that the ascorbate could be more effective than it would be on its own (2012).

Some studies do not support the effectiveness of vitamin C in treatment. In a study by Lee et al., the researchers claimed that a dose of 20 milligrams of vitamin C per day is not beneficial in chemoprevention because the vitamin C could actually trigger the growth of tumor cells. However, the authors came to this conclusion based on the results of an in vitro study. The results would likely have been different in vivo, because of the assistive actions of glutathione peroxidase and catalase in neutralizing excess free radicals and curbing cancer growth (Lee et al., 2003).

Another limitation of using vitamin C for cancer treatment is the length of time that vitamin C stays in the person's body. According to Ratnam et al. (2006), "approximately 73% of ascorbic acid is [naturally] removed from the body in less than 24 hours" (p. 98). In patients with compromised kidney function, vitamin C cannot be reabsorbed effectively, and even in healthy patients, it is difficult to maintain vitamin C levels in an individual's system orally, even if the dose is as high as 1000 milligrams (Ratnam et al., 2006). To compensate for this problem, Ratnam et al. explain that some forms of vitamin C have now been manufactured to be absorbed in a time-release manner. Also, many studies have indicated that intravenous doses can be effective, because of their quick absorption rates (National Institutes of Health, 2011).

While most researchers have found that high doses of vitamin C positively benefit the body, a few researchers have found that high doses of vitamin C could actually cause DNA damage in natural cells (Lee et al., 2003). However, the unique properties of vitamin C as a natural supplement show that it may be able to act differently upon natural cells than it does upon tumor cells. For example, although vitamin C triggers a cascade of reactions that allow hydrogen peroxide to damage tumor cells, natural cells have the enzyme catalase to protect them

from this type of damage (Quillin, 2005). Because artificial chemotherapy agents are not able to differentiate between natural and tumor cells, vitamin C may still be a better alternative to some of these artificial drugs. In addition, Madhuri et al. (2011) concluded that the patients who received vitamin C therapy experienced less damage to their lymphocytic DNA than those who did not. While more studies need to be performed that conclude the negative effects of vitamin C on natural cells to be insignificant, current studies such as those by Kim et al. (2012), Lee et al. (2003), Padayatty et al. (2006), and Ratnam et al. (2006) vitamin C as a worthwhile choice for supplemental treatment.

Non-Supportive Arguments Regarding the Effectiveness of Vitamin C in Cancer Treatment

While many studies have been performed emphasizing the positive effects of vitamin C in cancer treatment, others indicate that this may not be the case. A study performed by Zhang et al. measured the amounts of supplements, including vitamin C supplements, taken among 132,837 participants in China and compared them to these participants' liver cancer risk. Instead of the inverse correlation between vitamin C supplementation and cancer risk found in many of the other studies, Zhang et al. (2012) found a positive correlation between those who took vitamin C supplements and those who had a high risk of liver cancer. However, out of those who took vitamin C and had a high risk of liver cancer, many had a family history of liver disease or were regular smokers.

Unfortunately, Zhang et al.'s (2012) study is not the only one to conclude non-supportive results for effectiveness of vitamin C in cancer treatment. A study by Kirsh et al. consisted of 29,361 men who were evaluated based on their intake of vitamin supplements and whether they

developed prostate cancer during the eight-year monitoring process. Of the total participants, 56% used a multivitamin or single vitamin supplement: 52% took vitamin E, 42% took beta-carotene, and 51% took vitamin C. At the end of the eight-year study, the relationship between taking vitamin C supplements and development of prostate cancer was not significant. However, Kirsh et al. state that a wide variety of studies on the same topic show that there is at least a relationship between supplement intake and lowered cancer risk in some subgroups of the population.

Conclusion

While 50 years ago hardly any research had been performed to determine the effectiveness of vitamin C on cancer treatment, there are many studies today that evaluate various aspects of vitamin C's relationship with tumor cells and neoplastic processes. The results of several studies indicate that vitamin C can be effective in multiple levels of cancer treatment. As an antioxidant, vitamin C can keep harmful free radicals from damaging natural DNA and causing neoplastic transformation (Ratnam et al., 2010). Its synergistic relationship with free radical enzymes such as superoxide dismutase, catalase, glutathione-S-transferase, and glutathione reductase allow greater work to be accomplished than could be accomplished with a single antioxidant, and its synergistic relationship with vitamin E allows the spared vitamin E to exercise greater antioxidant abilities than it could without vitamin C (Suhail et al., 2012; Kim et al., 2010; Weijl et al., 2004; Greenlee et al., 2012).

With regards to transcription factors, ascorbate could possibly replace NFkB in the process of detoxifying antioxidants to prevent tumor growth (Lee et al., 2003), and has been shown to elevate the levels of NRF2 in the body, which through a cascade process help to

neutralize harmful free radicals (Singh & Bhat, 2012). Vitamin C can protect natural cells from chromosomal damage (Madhuri et al., 2011) and help to destroy cancer cells by activating hydrogen peroxide (Quillin, 2005).

Not all studies on vitamin C have yielded positive results. However, even studies like Zhang et al.'s and Kirsh et al.'s indicate limitations in their studies that may have kept the relationship between vitamin C and lowered cancer risk from being stronger. While the effect of vitamin C is not definitive, many studies support the view that vitamin C can be beneficial in cancer treatment. One of the most important findings in these studies is that vitamin C can cause damage to tumor cells without significantly harming natural cells (Quillin, 2005).

By compiling the results of all of these studies and performing more research to determine vitamin C's effectiveness as a chemotherapeutic agent, researchers could begin to determine stronger theories that would allow them to effectively recommend vitamin C as part of cancer therapy. Based on these results, oncologists could begin to prescribe vitamin C therapy as a primary form of treatment. While the extent to which vitamin C can affect tumors and the dosage of vitamin C most useful to cancer treatment has yet to be precisely determined, the strong evidence of vitamin C's benefits, combined with its abilities to keep natural cells from harm, makes vitamin C a valuable supplement for cancer treatment.

References

Aggarwal, B.B., Vijayalekshmi, R.V., & Sung, B. (2009). Targeting inflammatory pathways for prevention and therapy of cancer: Short-term friend, long-term foe. *Clinical Cancer Research, 15,* 425-230. doi:10.1158/1078-0432.CCR-08-0149

Antigen. *The Merriam Webster dictionary.* [Def. 7] (n.d.) Retrieved March 26, 2013 from http://www.merriam-webster.com/medical/antigen

Bidoli, E., Talamini, R., Zucchetto, A., Bosetti, C., Negri, E., Lenardon, O., &…La Vecchia, C. (2009). Dietary vitamins E and C and prostate cancer risk. *Acta Oncologica, 48*(6), 890-894. doi:10.1080/02841860902946546

Chemokine. *The American heritage medical dictionary.* [Def. 4]. (n.d.). Retrieved February 1, 2013, from http://medical-dictionary.thefreedictionary.com/chemokine

Cytokine. *The American heritage medical dictionary*. [Def. 3]. (n.d.). Retrieved February 1, 2013, from http://medical-dictionary.thefreedictionary.com/cytokine

Dugdale, D.C., & Chen, Y.B. (2012, June 5). *Chemotherapy*. Retrieved February 1, 2013 from http://www.nlm.nih.gov/medlineplus/ency/article/002324.htm

Federico, A., Morgillo, F., Tuccillo, C., Ciardiello, F., & Loguercio, C. (2007). Chronic inflammation and oxidative stress in human carcinogénesis. *International Journal of Cancer, 121*(11), 2381-2386.

Goodman, M.T., Kiviat, N., McDuffie, K., Hankin, J.H., Hernandez, B., Wilkens, L.R., &…Killeen, J. (1998). The association of plasma micronutrients with the risk of cervical dysplasia in Hawaii. ***Cancer Epidemiology, Biomarkers & Prevention,*** *7*(6), 537-544.

Greenlee, H., Kwan, M.L., Kushi, L.H., Song, J., Castillo, A., Weltzien, E., &…Caan, B.J. (2012). Antioxidant supplement use after breast cancer diagnosis and mortality in the Life After Cancer Epidemiology (LACE) cohort. *Cancer (0008543X)*, *118*(8), 2048-2058. doi:10.1002/cncr.26526

Hartel, C., Strunk, T., Bucsky, P., & Schultz, C. (2004). Effects of vitamin C on intracytoplasmic cytokine production in human whole blood monocytes and lymphocytes. *Cytokine, 27*(4-5), 101-106.

Hendrickson, K. (2011). Is L-ascorbate the same as ascorbic acid? Retrieved February 9, 2013 from http://www.livestrong.com/article/370080-is-l-ascorbate-the-same-as-ascorbic-acid/

Huang, H.Y., Helzlsouer, K.J., and Appel, L.J. (2000). The effects of vitamin C and vitamin E on oxidative DNA damage: results from a randomized controlled trial. *Cancer Epidemiology, Biomarkers & Prevention, 9*(7), 647-652.

Inoue, T., Kato, K., Kato, H., Asanoma, K., Kuboyama, A., Uekoka, Y., &…Wake, N. (2009). Level of reactive oxygen species induced by p21Waf1/CIP1 is critical for the determination of cell fate. *Cancer Science, 100*(7), 1275-1283. doi:10.1111/j.1349-7006.2009.01166.x

Interleukin. *The American heritage medical dictionary*. [Def. 5]. (n.d.). Retrieved February 1, 2013, from http://medical-dictionary.thefreedictionary.com/interleukin

Kapil, U.U., Singh, P.P., Shukla, N.K., Dwivedi, S.S., Pathak, P.P., & Singh, R.R. (2003). Association of vitamin A, vitamin C and zinc with laryngeal cancer. *Indian Journal of Cancer, 40*(2), 67-70.

Kim, J., Lee, S.D., Chang, B., Jin, D.H., Jung, S., Park, M.Y., Han, Y., &….Lee, M.S. (2012). Enhanced antitumor activity of vitamin C via p53 in cancer cells. *Free Radical Biology & Medicine, 53*(8), 1607-1615. doi: 10.1016/j.freeradbiomed.2012.07.079

Kim, J., Mi Kyung, K., Jae Kwan, L., Jae-Hoon, K., Sung Kyong, S., Eun-Seop, S., &…Young Mi, Y. (2010). Intakes of vitamin A, C, and E, and B-carotene are associated with risk of cervical cancer: A case-control study in Korea. *Nutrition & Cancer, 62*(2), 181-189. doi: 10.1080/01635580903305326

Kirsh, V.A., Hayes, R.B., Mayne, S.T., Chatterjee, N., Subar, A.F., Dixon, L., &…Peters, U. (2006). Supplemental and dietary vitamin E, betacarotene, and vitamin C intakes and prostate cancer risk. *JNCI: Journal of the National Cancer Institute, 98*(4), 245-254. doi:10.1093/jnci/djj050

Lamson, D.W., Yu-Huan, G., Plaza, S.M., Brignall, M.S., Brinton, C.A., & Sadlon, A.E. (2010). The vitamin C:K3 system—enhancers and inhibitors of the anticancer effect. *Alternative Medicine Review,15*(4), 345-351.

Lee, K.W., Lee, H.J., Surh, Y.J., & Lee, C.Y. (2003). Vitamin C and cancer chemoprevention: reappraisal. *American Journal of Clinical Nutrition, 78*(6), 1074-1078.

Madhuri, K., Vani, K., Syed, R., & Alshatwi, A.A. (2011). Role of ascorbic acid as an antioxidant in gastric cancer patients in south Indian population. *International Journal of Pharmacy and Pharmaceutical Sciences, 3*(4), 179-181.

Malekshah, A., Kimiagar, M., Pourshams, A., Yazdani, J., Majd, S., Goglani, G., &…Malekzadah, R. (2010). Vitamin deficiency in Golestan Province, northern Iran: A high risk area for esophageal cancer. *Archives of Iranian Medicine (AIM) 13*(5), 391-394.

Mamede, A., Tavares, S., Abrantes, A., Trindade, J., Maia, J., & Botelho, M. (2011).The role of vitamins in cancer: A review. *Nutrition & Cancer, 63*(4), 479-494. doi:10.1080/01635581.2011.539315

Mikirova, N., Casciari, J., Rogers, A., & Taylor, P. (2012). Effect of high-dose intravenous vitamin C on inflammation in cancer patients. *Journal of Translational Medicine, 10*(1), 189-198. doi:10.1186/1479-5876-10-189

Mohan, S., & Priya, V. (2009). Changes in lipid peroxidation, glutathione, ascorbic acid, vitamin E and antioxidant enzymes in patients with ovarian cancer. *Acta Medica Academica, 38*(1), 1-5.

Morgan, Shelly. (2010). Vitamin C and the common cold. Retrieved January 13, 2013 from http://www.livestrong.com/article/301309-vitamin-c-the-common-cold/

National Cancer Institute. (n.d.) Reactive oxygen species. Retrieved January 18, 2013 from http://www.cancer.gov/dictionary?cdrid=687227

National Institutes of Health. (2011, June 24). Dietary supplement fact sheet: Vitamin C. Retrieved October 29, 2012 from http://ods.od.nih.gov/factsheets/VitaminC-QuickFacts/

Neoplastic transformation. *Mosby's medical dictionary.* [Def.6] (n.d.) Retrieved February 7, 2013, from http://medicaldictionary.thefreedictionary.com/neoplastic+transformation

Northwestern University. *Lipid peroxidation.* [Def. 8] (n.d.) Retrieved April 2, 2013, from http://groups.molbiosci.northwestern.edu/holmgren/Glossary/Definitions/Def-L/lipid_peroxidation.html

Owuor, E.D. & Kong, A.N.T. (2002). Antioxidants and oxidants regulated signal transduction pathways. *Biochemical Pharmacology, 64*(10), 765-770.

Padayatty, S.J., Riordan, H.D., Hewitt, S.M., &…Katz, A. (2006, March 28). Intravenously administered vitamin C as cancer therapy: Three cases. *Canadian Medical Association Journal, 174*(7), 937-942.

Podmore, I.D., Griffiths, H.R., Herbert, K.E., Mistry, N., Mistry, P., & Lunec, J. (1998). Vitamin C exhibits pro-oxidant properties. *Nature, 392*(6676), 559.

Quillin, P. (2005). *Beating Cancer with Nutrition.* Carlsbad, CA: Nutrition Times Press.

Ratnam, D.V., Ankola, D.D., Bhardwaj, V., Sahana, D.K., & Ravi Kumar, M.N.V. (2006). Role of antioxidants in prophylaxis and therapy: a pharmaceutical perspective. *Journal of Controlled Release, 113*(3), 189-207.

Seifried, H.E., McDonald, S.S., Anderson, D.E., Greenwald, P., & Milner, J.A. (2003). The antioxidant conundrum in cancer. *Cancer Research, 63*(15), 4295-4298.

Singh, B., & Bhat, H.K. (2012). Superoxide dismutase 3 is induced by antioxidants**,** inhibits oxidative DNA damage and is associated with inhibition of estrogen-induced breast cancer. *Carcinogenesis, 33*(12), 2601-2610.

Skin toxicity from external beam radiation therapy in breast cancer patients: protective effects of Resveratrol, Lycopene, and vitamin C and anthocianin (Ixor). (2012). *Radiation Oncology, 7*(1), 12-17. doi:10.1186/1748-717X-7-12

Suhail, N., Bilal, N., Khan, H. Y., Hasan, S., Sharma, S., Khan, F., Mansoor, T. and Banu, N. (2012). Effect of vitamins C and E on antioxidant status of breast-cancer patients undergoing chemotherapy. *Journal of Clinical Pharmacy and Therapeutics, 37*, 22–26. doi: 10.1111/j.1365-2710.2010.01237.x

University of Maryland Medical Center. (2011). *Vitamin C (ascorbic acid).* Retrieved April 2, 2013, from http://www.umm.edu/altmed/articles/vitamin-c-000339.htm.

Velanganni, A.J., & Balasundaram, C.C. (2010). Biochemical investigations on the cancer therapy potential of antioxidant vitamins A, C, and E, and their analogues on p-dimethylaminoazobenzene-induced hepatoma in rats. *Journal of Cell & Tissue Research, 10*(2), 2191-2199.

Weijl, N.I., Elsendoorn, T.J., Lentjes, E.G.W.M., Hopman, G.D., Wipkink-Bakker, A., Zwinderman, A.H., Cleton, F.J., & Osanto, S. (2004). Supplementation with antioxidant micronutrients and chemotherapy-induced toxicity in cancer patients treated with cisplatin-based chemotherapy: A randomised, double-blind, placebo-controlled study. *European Journal of Cancer, 40*(11), 1713-1723.

Zhang, W., Shu, X.O., Li, H., Yang, G., Cai, H., Ji, B.T., Gao, J., Gao, Y.T., Zheng, W., & Xiang, Y.B. (2012). Vitamin intake and liver cancer risk: A report from two cohort studies in China. *Journal of the National Cancer Institute, 104*(15), 1174-1182. doi:10.1093/jnci/djs277