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REVIEW | *New Investigator Review Award*

The differential role of reactive oxygen species in early and late stages of cancer

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Assi M. The differential role of reactive oxygen species in early and late stages of cancer. *Am J Physiol Regul Integr Comp Physiol* 313: R646–R653, 2017. First published August 23, 2017; doi:10.1152/ajpregu.00247.2017.—The large doses of vitamins C and E and β-carotene used to reduce reactive oxygen species (ROS) production and oxidative damages in cancerous tissue have produced disappointing and contradictory results. This therapeutic conundrum was attributed to the double-faced role of ROS, notably, their ability to induce either proliferation or apoptosis of cancer cells. However, for a ROS-inhibitory approach to be effective, it must target ROS when they induce proliferation rather than apoptosis. On the basis of recent advances in redox biology, this review underlined a differential regulation of prooxidant and antioxidant system, respective to the stage of cancer. At early precancerous and neoplastic stages, antioxidant activity decreases and ROS appear to promote cancer initiation via inducing oxidative damage and base pair substitution mutations in prooncogenes and tumor suppressor genes, such as *RAS* and *TP53*, respectively. Whereas in late stages of cancer progression, tumor cells escape apoptosis by producing high levels of intracellular antioxidants, like NADPH and GSH, via the pentose phosphate pathway to buffer the excessive production of ROS and related intratumor oxidative injuries. Therefore, antioxidants should be prohibited in patients with advanced stages of cancer and/or undergoing anticancer therapies. Interestingly, the biochemical and biophysical properties of some polyphenols allow them to selectively recognize tumor cells. This characteristic was exploited to design and deliver nanoparticles coated with low doses of polyphenols and containing chemotherapeutic drugs into tumor-bearing animals. First results are encouraging, which may revolutionize the conventional use of antioxidants in cancer.

cancer stages; reactive oxygen species; metabolism; genetic mutations; antioxidants

DURING THE AEROBIC METABOLISM, mitochondria splits 95% of oxygen (O_2) into water, leaving 5% of free anion superoxide ($O_2^{\cdot -}$) in the cell, thereby, initiating the generation of a panel of reactive oxygen species (ROS) (8). The term reactive species encompasses O_2, nitrogen, chlorine, bromine, and sulfuric species. They include $O_2^{\cdot -}$, hydrogen peroxide (H_2O_2), hydroxyl radical ($^{\cdot}OH$), and nitric oxide ($NO^{\cdot}$) (23). ROS have been thought of as unstable, highly reactive, and short-living species that indiscriminantly react with proteins, lipids, and DNA to cause oxidative damages. Our current view is that ROS cannot be considered as a single entity, since each species has inherent chemical properties and is produced in different conditions (13, 22). For example, in the presence of ferrous iron (Fe^{2+}), H_2O_2 generates $^{\cdot}OH$ radicals (Fenton reaction) that interact with any biological target to induce oxidative damage. Whereas in physiological conditions in which intracellular Fe^{2+} levels are kept low, H_2O_2 selectively reacts with cysteine and selenocysteine residues of different transcriptional factors (e.g., NF-κB and AP-1) (39), without affecting reduced glutathione (GSH) pool (13), which allows redox biology signaling rather than oxidative damage. H_2O_2 is present in nanomolar range in living cells, its relatively stable structure and its cellular compartmentalization (e.g., plasma membrane and mitochondria) confer to it a determinant role in the activation of signaling pathways controlling proliferation, differentiation, migration and, even, apoptosis (50). This natural balance between life and death response is maintained by the presence of antioxidant enzymes (e.g., superoxide dismutase, catalase, and glutathione peroxidase) and low-molecular-weight antioxidants (e.g., vitamins, polyphenols, and oligo-elements) that buffer the intracellular milieu and maintain ROS at a physiological level. The disruption in the prooxidant/antioxidant balance has been reported in more than 200 clinical disorders (33).

Over the last two decades, the scientific progress has stressed a prominent role for ROS in the pathogenesis of

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neurodegenerative diseases, inflammatory bowel diseases, and cancer (21). Particularly, in cancer, which is the focus of this review, the excessive production of ROS and related oxidative stress (OS), are considered as important molecular hallmarks (38). Indeed, H_2O_2 production is elevated in tumor tissues compared with adjacent normal ones (65). Additionally, high levels of 4-hydroxynonenal (4-HNE) (lipid peroxidation) and 8-oxoguanine (8-oxoG) (oxidative DNA damage) are usually present in the blood and urine of cancer patients and correlate with poor prognosis (61, 73). For more than 20 years, large doses of vitamins C and E and β-carotene have been used as a therapeutic approach to counteract OS- and ROS-dependent mechanisms in cancer. Unfortunately, most of these studies failed and produced very contradictive results that detracted from our attention to the main goal of generating effective ROS inhibitors that work in vivo (4, 5). This controversy could be attributed to the double-faced role of ROS in driving both proliferation and apoptosis in cancer cells (3, 58). However, for an antioxidant strategy to be effective, it must target ROS when they promote proliferation rather than apoptosis. From this point of view, the notion of "cancer stage" should be taken into account, as the concentrations of ROS and, therefore, their role may change with the evolution of the disease (18). This review distinguishes a different role for ROS between early and late stages of cancer, which could improve our understanding of the underpinning molecular mechanisms and help to develop more effective anticancer strategies.

Early Stages of Cancer: a Prooncogenic Role for ROS

Oxidative DNA damage and reprogramming of cell metabolism. $^{\cdot}OH$-induced mutations in purines, pyrimidines, and chromatin proteins affect genome stability and dynamics of gene expression and are widely accepted as a cause for cancer initiation (69). One major oxidative DNA damage product is 8-oxoG, well known for inducing adjacent DNA base mutations (67). Most mutations target GC bases and are usually base pair substitutions rather than deletions or insertions (67). A well-understood mechanism is the G-to-T conversion found in the tumor-suppressor gene *TP53* (67). Exciting research found that p53 can act as a stress-sensor protein, and its antioxidant role emerges from its ability to enhance DNA repair and to regulate the expression of a subset of antioxidant genes; thus, loss-of-function mutations in p53 induce a further increase in intracellular ROS, provoke abnormal mitosis, and promote cancer development (52). Additionally, end products of lipid peroxidation like 4-HNE can compromise DNA repair by interfering with cysteine, lysine, and histidine residues of proteins involved in the nucleotide excision repair system (15). The accumulation of 8-oxoG and the deficiency in DNA repair induce mutations in other genes like *RAS*. The oncogenic activation of Ras commonly found in cancer increases the dependency on the Krebs cycle within mitochondria to produce ATP and amino acids, as well as $O_2^{\cdot -}$ and H_2O_2 as main by-products (16). Particularly, glutamine catabolism during the Krebs cycle is essential for tumor cell growth in the presence or absence of glucose (70). As a direct consequence, ATP and ROS are generated by oxidative phosphorylation and the mitochondrial respiratory chain complexes I, II, and III, respectively (70). The regular exposure of mitochondrial DNA (mDNA) to $O_2^{\cdot -}$ and H_2O_2 induces oxidative lesions in genes encoding for complex I, III, IV, and V (47). Accordingly, previous research reported that mutations in complex I, which cause a high production of ROS, enhanced cell proliferation (28), whereas cells null from mDNA did not produce ROS and failed to grow (70). In the same way, MitoQ, an analog of the endogenous mitochondrial antioxidant coenzyme Q10, hampered the proliferation of premalignant mammary cells (48). These observations indicate that ROS-induced nucleic/mitochondrial DNA damage, and metabolic adaptations are crucial for cancer initiation and promotion. Therefore, inhibition of ROS may slow and/or increase the latency of early-stage tumor development (52). As depicted in Table 1, recent preclinical evidence shows beneficial effects of synthetic and dietary antioxidant compounds on colon, liver, lung, and prostate cancer initiation and growth (9, 20, 32, 54, 63).

Inflammation and ROS-dependent prooncogenic signaling pathways. A sustained cellular proliferation in a microenvironment rich in inflammatory cytokines and growth factors promotes cancer development (12). This process is not independent of ROS. Indeed, TNF-α and EGF induce H_2O_2 production from NADPH oxidase located at the plasma membrane (40). Phosphoinositide-3 kinase (PI3K)/Akt is one of the major routes that maintain cancer cell survival via turning on the protein synthesis machinery. H_2O_2 activates the PI3K/Akt pathway, resulting in the phosphorylation and inactivation of Forkhead box3a (FOXO3a) (56). This abolishes the sustained inhibition exerted by FOXO3a on AP-1, leading to its nuclear accumulation (56). AP-1 is a transcriptional factor for miR-21 that promotes carcinogenesis by targeting various tumor suppressors (66). Indeed, AP-1 upregulates the expression of miR21, which, in turn, decreases the levels of numerous tumor suppressors, including phosphatase TENsin homolog and Von Hippel-Lindau (49, 56), well known to induce apoptosis and control cell migration and cytoskeleton remodeling. H_2O_2 may also induce the activation of the MAPK and ERK1/2, which, in turn, provoke NF-κB nuclear accumulation and expression of genes involved in inflammation and extracellular matrix (7). Recent research indicates that under hypoxic conditions, ROS activate hypoxia-inducible factor-1α and related angiogenic gene expression, like vascular endothelial growth factor (10). Therefore, there is a substantial body of evidence indicating that tumor cells redefine a new intracellular level of ROS, superior to normal cells, sufficient to induce proximal signaling that promote proliferation, survival and, subsequent, cancer growth (Fig. 1).

Late Stages of Cancer: a Potential Tumor-Suppressor Role for ROS

Excessive ROS production and metabolic shift toward intracellular antioxidant synthesis. The evolution from a neoplastic state into in situ carcinoma and invasive carcinoma is, respectively, associated with moderate, high, and excessive increase in ROS levels (18). This is particularly due to the increased need of energy blocks that enhances tumor metabolism and the subsequent ROS production (44). Even though ROS are crucial for normal-to-cancerous cell transformation and cancer development, the accumulation of oxidative insults promotes the death of cancer cells (71). For this reason, cancer cells have implemented a mechanism through which some derivatives of the glycolysis circuit are shuttled to the pentose phosphate

Table 1. *Preclinical studies published in the last three years show that antioxidants reduce tumor initiation in the early steps of carcinogenesis, whereas they enhance tumor progression, metastasis, and cachexia in more advanced/invasive stages*

	Cancer Model	AO Compounds or AO Enzymes Inhibitors	AO Starting Time	General Outcomes	Molecular Changes in Tumor Tissue or Metastatic Nodules
Initiation/promotion	MNU-induced hepatocellular carcinoma in mice (61)	Papaya extracts	With MNU-Treatment	↑ Body weight; ↓ Anemia, inflammation and liver dysplasia	↑ SOD, CAT and GPx activity; ↓ DNA damage
	B(*a*)P-induced lung carcinoma in mice (32)	Chrysin	Before B(*a*)P-treatment	↓ Tumor incidence and proliferation of alveolar epithelium	↑ SOD, CAT and GPx activity; ↓ COX-2 and NF-κB expression
	PhIP-induced prostate cancer in mice (9)	α, β, γ, and δ Tocopherol	Before PhiP-treatment	↓ Prostatic intraepithelial neoplasia	↑ PTEN expression; ↓ P-Akt expression and 8-oxoG content
	DMH-induced colon cancer in mice (52)	Quercetin	With DMH-treatment	↓ Colon dysplasia and inflammatory cell infiltration	—
	Low-grade (AH1) prostate tumor-bearing rats (20)	Pomegranate jus	With tumor implantation	↓ Tumor size and proliferation	↑ Bax/Bcl-2 ratio and cleaved caspase-3; ↓ P-ERK and 8-oxoG content
Progression/metastasis/ cachexia	Hepatocellular carcinoma (HepG2) tumor-bearing mice (62)	Diterpenoid Isoforretin A: thioredoxin inhibitor	After tumor injection	↓ Tumor volume	↑ 8-oxoG and DNA fragmentation; ↓ GSH content and thioredoxin activity
	High-grade colon (C26) tumor-bearing mice (2)	Vitamin C and E, quercetin, and curcumin	With tumor injection	↑ Cancer cachexia; ↓ Survival	↑ K_i-67 (proliferation); ↓ 4-HNE content
	Mice bearing human mammary tumors xenografts (25)	Inhibition of glutathione and thioredoxin	After tumor transplantation	↓ Malignant mammary tumor growth	↓ Nrf-2 signaling
	Mice bearing human melanoma xenografts (43)	NAC	After tumor transplantation	↑ Metastasis in blood and visceral organs	Metastatic nodules: ↑ NADPH generation via folate pathway
	Mice Tyr-Cre LSL-BRAF$^{CA/+}$ *PTEN*$^{fl/fl}$ (melanoma) (36)	Trolox and NAC	After tumor induction	↑ Lymph node metastasis	Metastatic nodules: ↑ GSH/GSSG ratio
	Mice iCre LSL-KRAS^{G12D} and mice BRAF^{V600E} (lung cancer) (55)	Vitamin E and NAC	After tumor induction	↑ Lung cancer progression; ↓ survival	↑ BrdU (proliferation); ↓ p53 expression and 8-oxoG content

AO, antioxidants; DMH, dimethylhydrazine; MNU, *N*-methyl-*N*-nitrosourea; B(*a*)P, benzo(*a*)pyrene; PhiP-P, 2-amino-1-methyl-6-phenylimidazo [4,5-b] pyridine; NAC, *N*-acetylcysteine; PTEN, phosphatase TENsin homolog; SOD, superoxide dismutase; CAT, catalase; GPx, glutathione peroxidase.

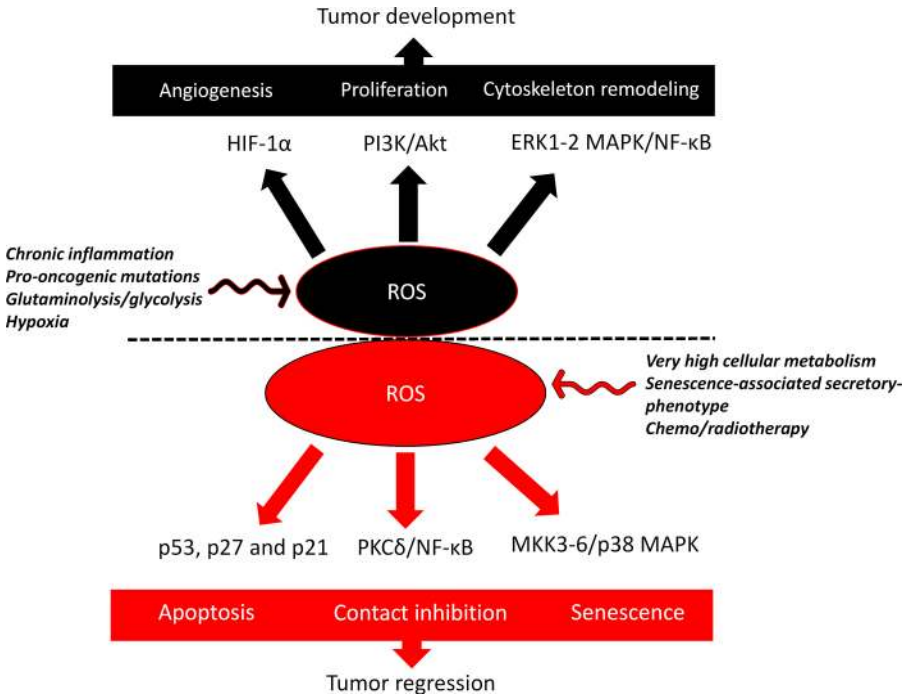

Fig. 1. The double-faced role of reactive oxygen species (ROS) in cancer. Figure summarizes main data cited in the text. High levels of ROS, resulting from abnormal cellular metabolism and inflammation, promote tumor proliferation, vascularization, and metastasis, while an excessive amount of ROS is likely to induce senescence and/or apoptosis.

pathway to synthesize NADPH and regenerate GSH (51). Consequently, NADPH and GSH buffer OS and allow cells to survive. Mechanistically, malignant cancer cells produce excessive levels of H_2O_2 near the toxicity threshold; thus, giving an antioxidant like *N*-acetylcysteine (NAC) reduces OS and promotes proliferation (35). Panieri and Santoro (44) have recently reviewed and listed the impact of some agents targeting the tumor antioxidant defense. They found that depleting NADPH and GSH enhanced the susceptibility of the malignant colon, ovarian, and lung cancer cells to ROS-mediated apoptosis (44). This is in line with our research and that of other laboratories, showing that vitamins C and E, NAC, and quercetin accelerated cancer progression, metastasis, and cachexia development in rodents, while inhibitors of thioredoxin and GSH induced tumor regression (2, 25, 36, 45, 57, 64) (Table 1).

Clinically, the expression and activity of antioxidant enzymes, as well as GSH content, are increased in malignant tumors comparing to adjacent normal tissue (30, 46), and a higher ratio of both levels (malignant/normal) is associated with poor survival (30). Interestingly, in melanoma, breast, and ovary cancers, the expression and activity of antioxidant enzymes in tumor tissue appear to decrease during neoplastic stages but increase in more advanced and malignant stages (6, 17, 27, 34, 53, 55) (Table 2). Similarly, inflamed tissue, a precursor condition for cancer development, exhibits a decrease in antioxidant capacities comparing to cancerous tissue in which antioxidants are highly present (60). Globally, clinical observations and preclinical data suggest a tumor-protective role for antioxidants in late stages of cancer.

ROS-mediated tumor suppression routes. Mitochondrial dysfunction and ROS accumulation are important proapoptotic events occurring in cancer cells (71). They lead to the expression of the cell cycle inhibitor p53, cell cycle arrest at the G2/M phase, DNA fragmentation, and subsequent apoptosis induction (71). Early in vivo observations demonstrated that ROS can trigger apoptosis through increasing lipid, protein, and DNA oxidation within the tumor itself (72). Recent studies have confirmed that chemotherapeutic agents, like cyclophosphamide, increase ROS-induced lipid peroxidation and apoptosis in sarcoma-180 tumor tissues (29). Mitochondria-dependent apoptosis is the most studied path. Excessive levels of ROS induce the oxidation of cardiolipin (phospholipids), which, in

Table 2. *Clinical studies demonstrating that the expression/activity of antioxidant enzymes and GSH content increase in malignant tumors comparing to benign hyperplasia or precursor lesions*

Type of Cancer	Patient Number	Malignant Versus Benign or Precursor Events	Antioxidant Defense	Detection Method
Thyroid (51)	34	Carcinoma vs. adenoma	↑ SOD, CAT, and GPx activity	Colorimetric method
Ovary (17)	26	Malignant vs. benign	↑ GSH content	Colorimetric method
Breast (17)	26	Malignant vs. benign	↑ GSH and GPx activity	Colorimetric method
Prostate (6)	27	Cancerous vs. neoplastic	↑ CuZnSOD expression	IHC
Breast (34)	50	Stage III vs. stage I/II	↑ SOD, CAT, and GPx activity	Colorimetric method
Skin (53)	36	Melanoma vs. actinic keratosis precancerous lesions	↑ CuZnSOD, MnSOD, and CAT expression	IHC
Pancreas (58)	13	Cancerous vs. pancreatitis	↑ MnSOD expression	WB
Ovary (27)	29	Carcinoma vs. benign cystadenoma	↑ CuZnSOD and MnSOD expression	WB and IHC

GSH, glutathione; IHC, immunohistochemistry; WB, Western blot.

turn, constitute a docking platform for the formation of tBid/Bax pore at the outer mitochondria membrane, allowing mitochondria-to-cytoplasm exit of cytochrome *c* (31). Then, cytochrome *c* forms with apoptotic protease activating factor 1 and procaspase-9 a complex called "apoptosome," which once activated, cleaves executioner caspases 3 and 7 to promote DNA fragmentation and apoptosis induction (11). Senescence constitutes another form of tumor regression, as it disables the proliferating capacity of cancer cells without inducing cell death (1). Indeed, the increase in ROS amounts induces the activation of MKK3/6 and its downstream p38-MAPK, which, in turn, phosphorylates p53 at three different serine residues Ser-33, Ser-37, and Ser-46 (24). Activated p53 then promotes the expression and stabilization of $p16^{INK4\alpha}$, $p14/p19^{ARF}$, and p21 to induce replicative senescence and irreversible cell cycle arrest at phase G1 (14, 59). Additionally, ROS are able to maintain cancer cells in a senescent state through the activation of NF-κB and subsequent release of IL-6/IL-8, a phenomenon known as senescence-associated secretory phenotype (68). To date, research in this field indicates that MKK/p38-MAPK is the main ROS-dependent pathway triggering senescence (Fig. 1).

Nanoformulation of Polyphenols in the Development of Potential Drug Carriers for Selective Tumor Targeting

Naturally, the above-mentioned mechanisms underline the need for a therapeutic approach that substitutes the simple antioxidant treatment and allows a "smart" targeting of a tumor. Besides the classical role of polyphenols in the activation of the Keap1-Nrf2 system, their ability to modulate important signaling pathways and, subsequent, cellular growth has raised the question about their utility in cancer therapy (41). However, like other nutritional and antioxidant compounds, the use of polyphenols in a supplementation setting is not without risk, as there is a clear dose-dependent functionality. For example, low doses of green tea extracts are beneficial, while high doses are likely to aggravate carcinogenesis and hepatotoxicity (42). Additionally, past clinical studies have taught us that unspecific systemic supplementation is complex to control and predict its outcomes, because of a multitude of factors like, the optimal dose, bioavailability, comorbidity, self-prescription, and patient's responsiveness (4). For these reasons, this paragraph focuses on the biochemical and biophysical features of a green tea extract called, epigallocatechin gallate (EGCG), which could be exploited in a new context to increase the effectiveness of standard anticancer therapies. EGCG is an interesting candidate, as it possesses hydrophobic characteristics that facilitate the interaction with lipid rafts on the plasma membrane to promote endocytosis (43). Additionally, EGCG may interact with specific membrane receptors highly expressed by tumor cells, such as the laminin receptor, which confers to it a higher affinity for cancerous cells compared with normal ones (37). These observations have introduced EGCG to nanomedicine with the hope that it will provide a more selective drug delivery system in cancer. Indeed, gold and polymeric nanoparticles (approved by the U.S. Food and Drug Administration), as well as liposomes, have been coated with low levels of EGCG and injected into animals bearing melanoma and bladder and prostate tumors (19). EGCG provoked increased cellular uptake and subsequent accumulation of EGCG-coated nanoparticles in tumor tissue (19, 37). More interestingly, EGCG nanoparticles enhanced the ability of the chemotherapeutic agent cisplatin to induce tumor regression and prolong lifespan in animals with malignancy (62). Therefore, chemotherapeutic agents encapsulated in EGCG-coated nanoparticles may represent a promising option to increase treatment efficiency and reduce related side effects.

Perspectives and Significance

The biological functions exerted by ROS appear to be dependent on the stage of tumor. As depicted in Fig. 2, precancerous stages, especially in melanoma and ovary and breast cancer, are associated with a decrease in antioxidant defense. The subsequent accumulation of intracellular ROS leads to oxidative DNA damage and mutations into prooncogenes and tumor-suppressor genes, which promote cancer development. Systematic reviews and meta-analysis are still needed to confirm that the decrease in antioxidant activity occurs in a general manner during the initiation of different

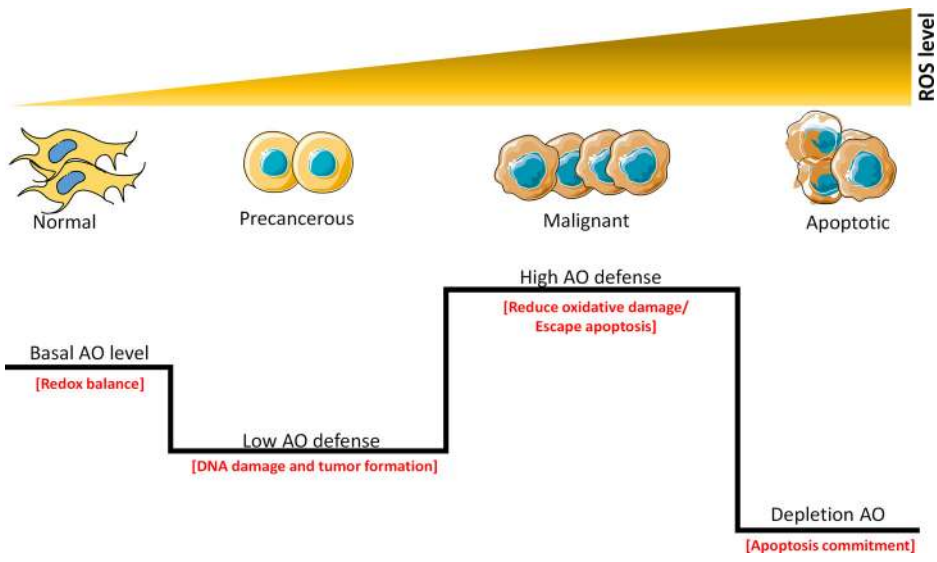

Fig. 2. Antioxidants reduce cancer initiation but enhance progression/invasiveness during carcinogenesis. Precancerous conditions, like inflamed or neoplastic tissue, are associated with a decrease in antioxidant response, allowing ROS to induce further DNA damage/mutations and tumor development. Whereas in later stages, antioxidant activity increases to limit excessive intratumor oxidative damage and help cancer cells to escape apoptosis. In both cases, intracellular antioxidants are modulated to promote tumor cells' survival.

types of cancer. In later stages, when cancer acquires a malignant and invasive phenotype, the activity of antioxidant enzymes and the intracellular pool of NADPH and GSH increase to limit oxidative damage caused by excessive ROS generation; thus, antioxidants at late stages of cancer are likely to help tumor cells in escaping ROS-induced oxidative insults and apoptosis. These molecular explanations are in line with the current literature, indicating that antioxidants should be avoided in patients with advanced stages of cancer and/or undergoing radiotherapy (26). After the completion of anticancer treatments, nutritional support could be useful for patients with systemic deficiencies to alleviate symptoms like mucositis, fibrosis, nephrotoxicity, and ototoxicity.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author.

AUTHOR CONTRIBUTIONS

M.A. conceived and designed research; M.A. analyzed data; M.A. prepared figures; M.A. drafted manuscript; M.A. edited and revised manuscript; M.A. approved final version of manuscript.

REFERENCES

1. **Acosta JC, Gil J.** Senescence: a new weapon for cancer therapy. *Trends Cell Biol* 22: 211–219, 2012. doi:10.1016/j.tcb.2011.11.006.
2. **Assi M, Derbré F, Lefeuvre-Orfila L, Rébillard A.** Antioxidant supplementation accelerates cachexia development by promoting tumor growth in C26 tumor-bearing mice. *Free Radic Biol Med* 91: 204–214, 2016. doi:10.1016/j.freeradbiomed.2015.12.019.
3. **Assi M, Rébillard A.** The Janus-faced role of antioxidants in cancer cachexia: new insights on the established concepts. *Oxid Med Cell Longev* 2016: 9579868, 2016. doi:10.1155/2016/9579868.
4. **Bjelakovic G, Nikolova D, Gluud LL, Simonetti RG, Gluud C.** Antioxidant supplements for prevention of mortality in healthy participants and patients with various diseases. *Sao Paulo Med J* 133: 164–165, 2015. doi:10.1590/1516-3180.20151332T1.
5. **Bjelakovic G, Nikolova D, Simonetti RG, Gluud C.** Antioxidant supplements for prevention of gastrointestinal cancers: a systematic review and meta-analysis. *Lancet* 364: 1219–1228, 2004. doi:10.1016/S0140-6736(04)17138-9.
6. **Bostwick DG, Alexander EE, Singh R, Shan A, Qian J, Santella RM, Oberley LW, Yan T, Zhong W, Jiang X, Oberley TD.** Antioxidant enzyme expression and reactive oxygen species damage in prostatic intraepithelial neoplasia and cancer. *Cancer* 89: 123–134, 2000. doi:10.1002/1097-0142(20000701)89:1<123::AID-CNCR17>3.0.CO;2-9.
7. **Cao L, Liu J, Zhang L, Xiao X, Li W.** Curcumin inhibits H_2O_2-induced invasion and migration of human pancreatic cancer via suppression of the ERK/NF-κB pathway. *Oncol Rep* 36: 2245–2251, 2016. doi:10.3892/or.2016.5044.
8. **Chance B, Erecinska M, Wagner M.** Mitochondrial responses to carbon monoxide toxicity. *Ann NY Acad Sci* 174: 193–204, 1970. doi:10.1111/j.1749-6632.1970.tb49786.x.
9. **Chen JX, Liu A, Lee MJ, Wang H, Yu S, Chi E, Reuhl K, Suh N, Yang CS.** δ- and γ-tocopherols inhibit phIP/DSS-induced colon carcinogenesis by protection against early cellular and DNA damages. *Mol Carcinog* 56: 172–183, 2017. doi:10.1002/mc.22481.
10. **Choi JE, Kim JH, Song NY, Suh J, Kim DH, Kim SJ, Na HK, Nadas J, Dong Z, Cha YN, Surh YJ.** 15-Deoxy-Δ(12,14)-prostaglandin J2 stabilizes hypoxia inducible factor-1α through induction of heme oxygenase-1 and direct modification of prolyl-4-hydroxylase 2. *Free Radic Res* 50: 1140–1152, 2016. doi:10.1080/10715762.2016.1219352.
11. **Circu ML, Maloney RE, Aw TY.** Disruption of pyridine nucleotide redox status during oxidative challenge at normal and low-glucose states: implications for cellular adenosine triphosphate, mitochondrial respiratory activity, and reducing capacity in colon epithelial cells. *Antioxid Redox Signal* 14: 2151–2162, 2011. doi:10.1089/ars.2010.3489.
12. **Coussens LM, Werb Z.** Inflammation and cancer. *Nature* 420: 860–867, 2002. doi:10.1038/nature01322.
13. **Davies MJ.** Protein oxidation and peroxidation. *Biochem J* 473: 805–825, 2016. doi:10.1042/BJ20151227.
14. **Deng Q, Liao R, Wu BL, Sun P.** High-intensity *ras* signaling induces premature senescence by activating p38 pathway in primary human fibroblasts. *J Biol Chem* 279: 1050–1059, 2004. doi:10.1074/jbc.M308644200.
15. **Feng Z, Hu W, Tang MS.** Trans-4-hydroxy-2-nonenal inhibits nucleotide excision repair in human cells: a possible mechanism for lipid peroxidation-induced carcinogenesis. *Proc Natl Acad Sci USA* 101: 8598–8602, 2004. doi:10.1073/pnas.0402794101.
16. **Gao P, Tchernyshyov I, Chang TC, Lee YS, Kita K, Ochi T, Zeller KI, De Marzo AM, Van Eyk JE, Mendell JT, Dang CV.** c-Myc suppression of miR-23a/b enhances mitochondrial glutaminase expression and glutamine metabolism. *Nature* 458: 762–765, 2009. doi:10.1038/nature07823.
17. **Ghalia AA, Rabboh NA, el Shalakani A, Seada L, Khalifa A.** Estimation of glutathione S-transferase and its Pi isoenzyme in tumor tissues and sera of patients with ovarian cancer. *Anticancer Res* 20, *2B*: 1229–1235, 2000.
18. **Gorrini C, Harris IS, Mak TW.** Modulation of oxidative stress as an anticancer strategy. *Nat Rev Drug Discov* 12: 931–947, 2013. doi:10.1038/nrd4002.
19. **Granja A, Pinheiro M, Reis S.** Epigallocatechin gallate nanodelivery systems for cancer therapy. *Nutrients* 8: 307, 2016. doi:10.3390/nu8050307.
20. **Gueritat J, Lefeuvre-Orfila L, Vincent S, Cretual A, Ravanat JL, Gratas-Delamarche A, Rannou-Bekono F, Rebillard A.** Exercise training combined with antioxidant supplementation prevents the antiproliferative activity of their single treatment in prostate cancer through inhibition of redox adaptation. *Free Radic Biol Med* 77: 95–105, 2014. doi:10.1016/j.freeradbiomed.2014.09.009.
21. **Halliwell B.** Free radicals and antioxidants: updating a personal view. *Nutr Rev* 70: 257–265, 2012. doi:10.1111/j.1753-4887.2012.00476.x.
22. **Halliwell B.** Oxidative stress and cancer: have we moved forward? *Biochem J* 401: 1–11, 2007. doi:10.1042/BJ20061131.
23. **Halliwell B.** Oxygen and nitrogen are pro-carcinogens. Damage to DNA by reactive oxygen, chlorine and nitrogen species: measurement, mechanism and the effects of nutrition. *Mutat Res* 443: 37–52, 1999. doi:10.1016/S1383-5742(99)00009-5.
24. **Han J, Sun P.** The pathways to tumor suppression via route p38. *Trends Biochem Sci* 32: 364–371, 2007. doi:10.1016/j.tibs.2007.06.007.
25. **Harris IS, Treloar AE, Inoue S, Sasaki M, Gorrini C, Lee KC, Yung KY, Brenner D, Knobbe-Thomsen CB, Cox MA, Elia A, Berger T, Cescon DW, Adeoye A, Brüstle A, Molyneux SD, Mason JM, Li WY, Yamamoto K, Wakeham A, Berman HK, Khokha R, Done SJ, Kavanagh TJ, Lam CW, Mak TW.** Glutathione and thioredoxin antioxidant pathways synergize to drive cancer initiation and progression. *Cancer Cell* 27: 211–222, 2015. doi:10.1016/j.ccell.2014.11.019.
26. **Harvie M.** Nutritional supplements and cancer: potential benefits and proven harms. *Am Soc Clin Oncol Educ Book* 34: e478–e486, 2014. doi:10.14694/EdBook_AM.2014.34.e478.
27. **Hu Y, Rosen DG, Zhou Y, Feng L, Yang G, Liu J, Huang P.** Mitochondrial manganese-superoxide dismutase expression in ovarian cancer: role in cell proliferation and response to oxidative stress. *J Biol Chem* 280: 39485–39492, 2005. doi:10.1074/jbc.M503296200.
28. **Ishikawa K, Takenaga K, Akimoto M, Koshikawa N, Yamaguchi A, Imanishi H, Nakada K, Honma Y, Hayashi J.** ROS-generating mitochondrial DNA mutations can regulate tumor cell metastasis. *Science* 320: 661–664, 2008. doi:10.1126/science.1156906.
29. **Jana S, Patra K, Sarkar S, Jana J, Mukherjee G, Bhattacharjee S, Mandal DP.** Antitumorigenic potential of linalool is accompanied by modulation of oxidative stress: an in vivo study in sarcoma-180 solid tumor model. *Nutr Cancer* 66: 835–848, 2014. doi:10.1080/01635581.2014.904906.

30. **Janssen AM, Bosman CB, van Duijn W, Oostendorp-van de Ruit MM, Kubben FJ, Griffioen G, Lamers CB, van Krieken JH, van de Velde CJ, Verspaget HW.** Superoxide dismutases in gastric and esophageal cancer and the prognostic impact in gastric cancer. *Clin Cancer Res* 6: 3183–3192, 2000.
31. **Kagan VE, Tyurin VA, Jiang J, Tyurina YY, Ritov VB, Amoscato AA, Osipov AN, Belikova NA, Kapralov AA, Kini V, Vlasova II, Zhao Q, Zou M, Di P, Svistunenko DA, Kurnikov IV, Borisenko GG.** Cytochrome *c* acts as a cardiolipin oxygenase required for release of proapoptotic factors. *Nat Chem Biol* 1: 223–232, 2005. doi:10.1038/nchembio727.
32. **Kasala ER, Bodduluru LN, Barua CC, Madhana RM, Dahiya V, Budhani MK, Mallugari RR, Maramreddy SR, Gogoi R.** Chemopreventive effect of chrysin, a dietary flavone against benzo(*a*)pyrene induced lung carcinogenesis in Swiss albino mice. *Pharmacol Rep* 68: 310–318, 2016. doi:10.1016/j.pharep.2015.08.014.
33. **Kohen R, Nyska A.** Oxidation of biological systems: oxidative stress phenomena, antioxidants, redox reactions, and methods for their quantification. *Toxicol Pathol* 30: 620–650, 2002. doi:10.1080/01926230290166724.
34. **Kumaraguruparan R, Subapriya R, Viswanathan P, Nagini S.** Tissue lipid peroxidation and antioxidant status in patients with adenocarcinoma of the breast. *Clin Chim Acta* 325: 165–170, 2002. doi:10.1016/S0009-8981(02)00292-9.
35. **Laurent A, Nicco C, Chéreau C, Goulvestre C, Alexandre J, Alves A, Lévy E, Goldwasser F, Panis Y, Soubrane O, Weill B, Batteux F.** Controlling tumor growth by modulating endogenous production of reactive oxygen species. *Cancer Res* 65: 948–956, 2005.
36. **Le Gal K, Ibrahim MX, Wiel C, Sayin VI, Akula MK, Karlsson C, Dalin MG, Akyürek LM, Lindahl P, Nilsson J, Bergo MO.** Antioxidants can increase melanoma metastasis in mice. *Sci Transl Med* 7: 308re8, 2015. doi:10.1126/scitranslmed.aad3740.
37. **Lu YC, Luo PC, Huang CW, Leu YL, Wang TH, Wei KC, Wang HE, Ma YH.** Augmented cellular uptake of nanoparticles using tea catechins: effect of surface modification on nanoparticle-cell interaction. *Nanoscale* 6: 10297–10306, 2014. doi:10.1039/C4NR00617H.
38. **Luo J, Solimini NL, Elledge SJ.** Principles of cancer therapy: oncogene and non-oncogene addiction. *Cell* 136: 823–837, 2009. doi:10.1016/j.cell.2009.02.024.
39. **Marinho HS, Real C, Cyrne L, Soares H, Antunes F.** Hydrogen peroxide sensing, signaling and regulation of transcription factors. *Redox Biol* 2: 535–562, 2014. doi:10.1016/j.redox.2014.02.006.
40. **Meitzler JL, Antony S, Wu Y, Juhasz A, Liu H, Jiang G, Lu J, Roy K, Doroshow JH.** NADPH oxidases: a perspective on reactive oxygen species production in tumor biology. *Antioxid Redox Signal* 20: 2873–2889, 2014. doi:10.1089/ars.2013.5603.
41. **Mileo AM, Miccadei S.** Polyphenols as modulator of oxidative stress in cancer disease: new therapeutic strategies. *Oxid Med Cell Longev* 2016: 6475624, 2016. doi:10.1155/2016/6475624.
42. **Murakami A.** Dose-dependent functionality and toxicity of green tea polyphenols in experimental rodents. *Arch Biochem Biophys* 557: 3–10, 2014. doi:10.1016/j.abb.2014.04.018.
43. **Nakayama T, Hashimoto T, Kajiya K, Kumazawa S.** Affinity of polyphenols for lipid bilayers. *Biofactors* 13: 147–151, 2000. doi:10.1002/biof.5520130124.
44. **Panieri E, Santoro MM.** ROS homeostasis and metabolism: a dangerous liason in cancer cells. *Cell Death Dis* 7: e2253, 2016. doi:10.1038/cddis.2016.105.
45. **Piskounova E, Agathocleous M, Murphy MM, Hu Z, Huddlestun SE, Zhao Z, Leitch AM, Johnson TM, DeBerardinis RJ, Morrison SJ.** Oxidative stress inhibits distant metastasis by human melanoma cells. *Nature* 527: 186–191, 2015. doi:10.1038/nature15726.
46. **Portakal O, Ozkaya O, Erden Inal M, Bozan B, Koşan M, Sayek I.** Coenzyme Q10 concentrations and antioxidant status in tissues of breast cancer patients. *Clin Biochem* 33: 279–284, 2000. doi:10.1016/S0009-9120(00)00067-9.
47. **Ralph SJ, Rodríguez-Enríquez S, Neuzil J, Saavedra E, Moreno-Sánchez R.** The causes of cancer revisited: "mitochondrial malignancy" and ROS-induced oncogenic transformation—why mitochondria are targets for cancer therapy. *Mol Aspects Med* 31: 145–170, 2010. doi:10.1016/j.mam.2010.02.008.
48. **Rao VA, Klein SR, Bonar SJ, Zielonka J, Mizuno N, Dickey JS, Keller PW, Joseph J, Kalyanaraman B, Shacter E.** The antioxidant transcription factor Nrf2 negatively regulates autophagy and growth arrest induced by the anticancer redox agent mitoquinone. *J Biol Chem* 285: 34447–34459, 2010. doi:10.1074/jbc.M110.133579.
49. **Ren Y, Zhou X, Liu X, Jia HH, Zhao XH, Wang QX, Han L, Song X, Zhu ZY, Sun T, Jiao HX, Tian WP, Yang YQ, Zhao XL, Zhang L, Mei M, Kang CS.** Reprogramming carcinoma associated fibroblasts by AC1MMYR2 impedes tumor metastasis and improves chemotherapy efficacy. *Cancer Lett* 374: 96–106, 2016. doi:10.1016/j.canlet.2016.02.003.
50. **Rhee SG.** Cell signaling. H_2O_2, a necessary evil for cell signaling. *Science* 312: 1882–1883, 2006. doi:10.1126/science.1130481.
51. **Ros S, Santos CR, Moco S, Baenke F, Kelly G, Howell M, Zamboni N, Schulze A.** Functional metabolic screen identifies 6-phosphofructo-2-kinase/fructose-2,6-biphosphatase 4 as an important regulator of prostate cancer cell survival. *Cancer Discov* 2: 328–343, 2012. doi:10.1158/2159-8290.CD-11-0234.
52. **Sablina AA, Budanov AV, Ilyinskaya GV, Agapova LS, Kravchenko JE, Chumakov PM.** The antioxidant function of the p53 tumor suppressor. *Nat Med* 11: 1306–1313, 2005. doi:10.1038/nm1320.
53. **Sadani GR, Nadkarni GD.** Role of tissue antioxidant defence in thyroid cancers. *Cancer Lett* 109: 231–235, 1996. doi:10.1016/S0304-3835(96)04484-9.
54. **Saleem TH, Attya AM, Ahmed EA, Ragab SM, Ali Abdallah MA, Omar HM.** Possible protective effects of quercetin and sodium gluconate against colon cancer induction by dimethylhydrazine in mice. *Asian Pac J Cancer Prev* 16: 5823–5828, 2015. doi:10.7314/APJCP.2015.16.14.5823.
55. **Sander CS, Hamm F, Elsner P, Thiele JJ.** Oxidative stress in malignant melanoma and non-melanoma skin cancer. *Br J Dermatol* 148: 913–922, 2003. doi:10.1046/j.1365-2133.2003.05303.x.
56. **Saxena A, Tammali R, Ramana KV, Srivastava SK.** Aldose reductase inhibition prevents colon cancer growth by restoring phosphatase and tensin homolog through modulation of miR-21 and FOXO3a. *Antioxid Redox Signal* 18: 1249–1262, 2013. doi:10.1089/ars.2012.4643.
57. **Sayin VI, Ibrahim MX, Larsson E, Nilsson JA, Lindahl P, Bergo MO.** Antioxidants accelerate lung cancer progression in mice. *Sci Transl Med* 6: 221ra15, 2014. doi:10.1126/scitranslmed.3007653.
58. **Schieber M, Chandel NS.** ROS function in redox signaling and oxidative stress. *Curr Biol* 24: R453–R462, 2014. doi:10.1016/j.cub.2014.03.034.
59. **Serrano M, Lin AW, McCurrach ME, Beach D, Lowe SW.** Oncogenic ras provokes premature cell senescence associated with accumulation of p53 and p16INK4a. *Cell* 88: 593–602, 1997. doi:10.1016/S0092-8674(00)81902-9.
60. **Shen J, Person MD, Zhu J, Abbruzzese JL, Li D.** Protein expression profiles in pancreatic adenocarcinoma compared with normal pancreatic tissue and tissue affected by pancreatitis as detected by two-dimensional gel electrophoresis and mass spectrometry. *Cancer Res* 64: 9018–9026, 2004. doi:10.1158/0008-5472.CAN-04-3262.
61. **Sheridan J, Wang LM, Tosetto M, Sheahan K, Hyland J, Fennelly D, O'Donoghue D, Mulcahy H, O'Sullivan J.** Nuclear oxidative damage correlates with poor survival in colorectal cancer. *Br J Cancer* 100: 381–388, 2009. doi:10.1038/sj.bjc.6604821.
62. **Singh M, Bhatnagar P, Mishra S, Kumar P, Shukla Y, Gupta KC.** PLGA-encapsulated tea polyphenols enhance the chemotherapeutic efficacy of cisplatin against human cancer cells and mice bearing Ehrlich ascites carcinoma. *Int J Nanomedicine* 10: 6789–6809, 2015. doi:10.2147/IJN.S79489.
63. **Somanah J, Ramsaha S, Verma S, Kumar A, Sharma P, Singh RK, Aruoma OI, Bourdon E, Bahorun T.** Fermented papaya preparation modulates the progression of *N*-methyl-*N*-nitrosourea induced hepatocellular carcinoma in *Balb/c* mice. *Life Sci* 151: 330–338, 2016. doi:10.1016/j.lfs.2016.03.026.
64. **Sun X, Wang W, Chen J, Cai X, Yang J, Yang Y, Yan H, Cheng X, Ye J, Lu W, Hu C, Sun H, Pu J, Cao P.** The natural diterpenoid isoforretin A inhibits thioredoxin-1 and triggers potent ROS-mediated antitumor effects. *Cancer Res* 77: 926–936, 2017. doi:10.1158/0008-5472.CAN-16-0987.
65. **Szatrowski TP, Nathan CF.** Production of large amounts of hydrogen peroxide by human tumor cells. *Cancer Res* 51: 794–798, 1991.
66. **Talotta F, Cimmino A, Matarazzo MR, Casalino L, De Vita G, D'Esposito M, Di Lauro R, Verde P.** An autoregulatory loop mediated by miR-21 and PDCD4 controls the AP-1 activity in RAS transformation. *Oncogene* 28: 73–84, 2009. doi:10.1038/onc.2008.370.

67. **Tudek B, Winczura A, Janik J, Siomek A, Foksinski M, Oliński R.** Involvement of oxidatively damaged DNA and repair in cancer development and aging. *Am J Transl Res* 2: 254–284, 2010.
68. **Wang P, Han L, Shen H, Wang P, Lv C, Zhao G, Niu J, Xue L, Wang QJ, Tong T, Chen J.** Protein kinase D_1 is essential for Ras-induced senescence and tumor suppression by regulating senescence-associated inflammation. *Proc Natl Acad Sci USA* 111: 7683–7688, 2014. doi:10.1073/pnas.1310972111.
69. **Waris G, Ahsan H.** Reactive oxygen species: role in the development of cancer and various chronic conditions. *J Carcinog* 5: 14, 2006. doi:10.1186/1477-3163-5-14.
70. **Weinberg F, Hamanaka R, Wheaton WW, Weinberg S, Joseph J, Lopez M, Kalyanaraman B, Mutlu GM, Budinger GR, Chandel NS.** Mitochondrial metabolism and ROS generation are essential for Kras-mediated tumorigenicity. *Proc Natl Acad Sci USA* 107: 8788–8793, 2010. doi:10.1073/pnas.1003428107.
71. **Wu CL, Huang AC, Yang JS, Liao CL, Lu HF, Chou ST, Ma CY, Hsia TC, Ko YC, Chung JG.** Benzyl isothiocyanate (BITC) and phenethyl isothiocyanate (PEITC)-mediated generation of reactive oxygen species causes cell cycle arrest and induces apoptosis via activation of caspase-3, mitochondria dysfunction and nitric oxide (NO) in human osteogenic sarcoma U-2 OS cells. *J Orthop Res* 29: 1199–1209, 2011. doi:10.1002/jor.21350.
72. **Xie K, Huang S, Dong Z, Gutman M, Fidler IJ.** Direct correlation between expression of endogenous inducible nitric oxide synthase and regression of M5076 reticulum cell sarcoma hepatic metastases in mice treated with liposomes containing lipopeptide CGP 31362. *Cancer Res* 55: 3123–3131, 1995.
73. **Young O, Crotty T, O'Connell R, O'Sullivan J, Curran AJ.** Levels of oxidative damage and lipid peroxidation in thyroid neoplasia. *Head Neck* 32: 750–756, 2010. doi:10.1002/hed.21247.