Selection of Surrogate Endpoint Biomarkers to Evaluate the Efficacy of Lycopene/Tomatoes for the Prevention/Progression of Prostate Cancer

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EXPANDED ABSTRACT

The use of surrogate endpoint biomarkers (SEBs) is essential to the crafting of cost-effective clinical trials to evaluate not only the efficacy of lycopene/tomatoes but also combinations of bioactive compounds. An SEB can be defined as a measurable biological process or molecule that is closely linked to the progression pathway to invasive cancer and that undergoes change in concert with neoplastic regression (1). The closer the SEB is in the pathway to prostate cancer expression or progression, the more likely its predictive value. High-grade prostate intraepithelial neoplasia (HGPIN), microvesSEL density, and prostate-specific antigen (PSA) have been exploited for these purposes, and their sensitivity and specificity in predicting clinically relevant prostate cancer can be determined. Modulation of a number of signaling pathways distinguishes neoplastic cells, and patterns or combinations of these changes may be more predictive of cancer progression. The sensitivity and specificity of SEBs can be assessed through their incorporation in case-control and longitudinal studies in men at higher risk and men diagnosed with prostate cancer.

The pathways modulated by tomato bioactive ingredients can be determined through tissue culture and animal studies. A number of possible bioactive compounds are contained in tomato. They include ascorbic acid; some unique carotenoids such as lycopene, phytoene, phytofluene, and γ-carotene; and a number of polyphenols such as quercetin, naringenin, and chlorogenic acid. Tomato products also contain small amounts of glucosinolate, tomatine, and the lycopene oxidation product, cyclolycopene, which is found in physiologically important quantities in circulating plasma with high tomato product consumption. Lycopene and quercetin are the likely candidates for prostate cancer mediation. They are predominant compounds in tomato and have been shown in animal studies to reduce cancer. Quercetin attenuates androgen receptor function and cancer cell lipogenesis leading to apoptosis (2,3). These actions have been identified in cell culture at higher than physiological concentrations. Lycopene acts as an antioxidant in vivo but can be a prooxidant also. It inhibits cell cycle progression and promotes apoptosis. It inhibits IGF-1 and androgen signaling and IL-6 expression in the prostate, while upregulating gap junction communication. These pathways were explored to determine the extent of the lycopene effect and the possible predictive value of the pathway for the development of prostate cancer and advanced prostate cancer.

The first pathway evaluated was lycopene’s role in the prevention of oxidative DNA damage. Leukocyte and prostate 8-OH-deoxyguanosine (8OHdG), a DNA damage product, were reduced 21 and 28%, respectively, in men with prostate cancer whose diets were supplemented daily with tomato sauce for 3 wk before prostatectomy (4). Lycopene or tomato supplementation decreased strand breaks in the leukocytes of nonsmokers (5) and reduced by 20% the increase in strand breaks seen in lung cells with ozone exposure (6). How well do some measures of DNA damage predict prostate cancer? Urinary 8OHdG was nonsignificantly 62 and 29% higher in men with prostate cancer but was unrelated to PSA, cancer stage, Gleason score, or prostatectomy (7,8). The presence of 8OHdG in blinded prostate samples did not predict prostate cancer. However, the log of the ratio of the sum of 8OHdG and 8-OH-deoxyadenine divided by the sum of 2 other DNA oxidation products that arrest cell cycle progression, Fapyadenine and Fapyguanine, had high sensitivity (82%) and specificity (93%) for predicting which individuals had prostate cancer (8). Because this was the only candidate SEB modulated by lycopene for which sensitivity and specificity values were determined, this biomarker was chosen as the focus of this review.
where AP is a useful calculation. AP = S(1 – 1/R), where S is the sensitivity and R = [a(a + b)]/[(a + c)(b + d)], where a is the number of subjects with a positive SEB with cancer, b is a positive SEB without cancer, c is a negative SEB with cancer, and d is a negative SEB without cancer (1). The calculation requires a defined cutpoint (0.54 for the DNA damage ratio) and the number of subjects that fall into each category. For our example of the SEB, DNA damage ratio, the AP = 0.67 where 1 signifies all cases passing through the SEB and the pathway it marks, whereas an AP < 0.5 signifies the presence of alternative pathways to cancer. Thus, the DNA damage ratio might be a modestly good SEB, but this was measured in prostate tissue and not in circulating leukocytes where the predictive value might be much weaker. If we are to design an efficient small trial to evaluate lycopene’s ability to modulate the DNA damage ratio, there are a number of other facts we must know. The effect size, based on the lowering of cancer risk, is necessary for the calculation of subject numbers. However, the chosen SEB is not perfectly predictive of cancer. We can estimate the SEB misclassification by calculating the observed relative risk (ORR), which is a correction for the true relative risk of cancer (TRR) because we are using an SEB instead. For example, we will assume that there is a 40% reduction in prostate cancer risk based on population studies for advanced cancer for men in the highest consumption quartiles for tomato products. The TRR would be 0.6. ORR is given by the equation [(1 – TRR * P_c)(1 – S_p) + (S_c * TRR * P_c)]/[P_c(1 – S_p) + (S_c * P_c)], where P_c is the proportion of subjects predicted to develop prostate cancer (1). A reasonable estimate might be 20% for those entering a study with serum PSA > 2.5 µg/L. S_p is the sensitivity (0.82) and S_c is the specificity (0.93) of our SEB of DNA damage ratio. The calculated ORR for our example is 0.72 instead of the 0.60 we would have used to formulate the sample size calculation. We would have been in danger of a nonsignificant phase II trial for efficacy when lycopene was truly able to reduce cancer risk. The positive predictive value of an SEB is highly sensitive to the number of subjects expected to have prostate cancer, so it is important to choose subjects at higher risk although we might miss efficacy for early stages of the carcinogenic process. High specificity minimizes SEB bias, whereas high sensitivity hardly affects it. For example, the ORR only increases to 0.74 when sensitivity is lowered to 0.7.

Unfortunately, there are insufficient published data to perform these same calculations for putative SEBs on the pathways that lycopene or tomato appear to modulate. Studies of the insulin-like growth factor-1 (IGF-1) pathway appear to have produced the most data, and the AP and TRR can be calculated with a little effort by communication with authors of published articles. Lycopene modestly downregulates IGF-1 expression in healthy rat lateral prostate lobe and rat prostate tumors (9,10). There was a >60% increase in plasma IGFBP-3 and 20% lower IGF-1/IGFBP-3 in ferrets, and the effect was more dramatic in smoking ferrets (11). For each incremental serving of tomato products, men without prostate cancer had a 31% lower plasma IGF-1 concentration (12). On the cancer prediction side, IGF-1 or IFG binding protein 3 (IGFBP-3) or their ratio appear to be predictive for advanced cancer and, perhaps, tumor volume, but are not markers for screening (13–16). Confounding the distinction of early cancer is the observation that IGF-1 is higher and IGFBP-3 is lower in cases of benign prostatic hyperplasia (BPH) (17).

Many investigators have reported growth inhibition with lycopene in numerous prostate cell lines. The effect appears to be through cell cycle arrest and apoptosis and has been shown at physiological concentrations. Downregulation of cyclin D1 and D2 expression may be the mediator, but this has only been shown in cell lines other than prostate (18,19). Lycopene oxidation products may be more bioactive than lycopene (20). Tomato supplementation increased apoptotic index in cancer regions in men with prostate cancer, but most of this was attributable to a huge increase in 5 of the 28 subjects evaluated. Bcl-2 expression was not affected (21). Apoptotic bodies are almost always present in cancer and HGPIN but absent in BPH and normal tissue. Apoptotic index increases with Gleason score. It is unlikely that a further increase stimulated by lycopene would be distinguishable, Bcl-2, an inhibitor of apoptosis, is always expressed in BPH and normal tissue and occasionally expressed in cancer tissue (22), making it an unlikely candidate SEB. There is hope that biomarkers of the earlier stages of apoptosis may become useful.

There is a fair amount of circumstantial evidence that lycopene or tomatoes are anti-inflammatory; for example, lycopene attenuates macroscopic and microscopic markers of inflammation in induced colitis (23). The interest in the inflammation pathway comes from the finding that IL-6 expression is downregulated in tumors generated in the Dunning rat model (9) and IL-6 as well as a number of other inflammatory marker genes in normal rats (10) with lycopene supplementation. The 2 most measured biomarkers of inflammation are serum IL-6 and C-reactive protein (CRP). Serum IL-6, TNFα, and CRP distinguished metastatic cancer but not localized cancer and prediagnostic CRP was not associated with subsequent risk of cancer (24–26).

CONCLUSIONS

At this point only DNA damage and the IGF pathways have SEBs that are both modulated by lycopene and somewhat predictive of prostate cancer. The natural products in tomatoes such as lycopene and quercetin show only mild regulation of pathways involved in carcinogenesis at physiologically relevant concentrations. Both compounds appear to modulate a number of pathways. Therefore, the use of SEBs in multiple pathways that are prone to modulation either on the road to carcinogenesis or in the progression/regression of existing prostate cancer is a reasonable strategy. Specificity can be enhanced using a combination of SEBs to determine sensitivity and specificity. The organization of any research program to evaluate the efficacy of lycopene/tomatoes in the prevention or progression of prostate cancer should begin by determining the ability to modulate various pathways using appropriate SEBs for the pathway in humans. The calculation of AP for each SEB would aid comparisons. Then synergies with compounds that modulate complementary pathways should be explored before designing clinical trials using cancer or cancer progression or closely related SEBs as outcome measures. The evaluation of the efficacy and safety of using lycopene/tomatoes in combination with existing therapies such as androgen ablation and radiation is urgently needed.

LITERATURE CITED


