Dietary Nitrate and Reductive Polyphenols May Potentiate the Vascular Benefit and Alleviate the Ulcerative Risk of Low-Dose Aspirin

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Abstract

The recent revelation that daily low-dose aspirin not only lowers risk for vascular events, but also can notably decrease risk for a range of adenocarcinomas, decreasing total cancer mortality by about 20%, makes it highly desirable to implement this protective stategy on a population-wide basis. Nonetheless, the fact that low-dose aspirin approximately doubles risk for serious gastrointestinal bleeding may impede health authorities from recommending its use by people judged to be at low cardiovascular risk. Nitric oxide (NO) exerts gastroprotective effects by boosting blood flow and mucus production in the gastric mucosa – effects which demonstrably oppose the pro-ulcerative impact of aspirin and other NSAIDS. A nitrate-rich diet, as well as ingestion of reductive catechol-bearing polyphenols, can collaborate in promoting NO generation in gastric juice, and they are protective in rodent models of gastric ulceration. Moreover, a high-nitrate diet, as well as certain reductive polyphenols such as epicatechin and quercetin, can exert platelet-stabilizing effects complementary to those of aspirin, and act in other ways to preserve vascular health. Hence, diets rich in nitrate and reductive polyphenols have the potential to amplify the vascular-protective benefits of low-dose aspirin, while diminishing its pro-ulcerative risk. Low-dose aspirin may be more unequivocally recommendable within the context of such a dietary strategy.

Low-Dose Aspirin for Cancer Prevention and Vascular Protection

Low-dose aspirin administration has been demonstrated to significantly and quite cost-effectively decrease risk for thrombotic episodes (MI, stroke, TIAs) in at-risk subjects, in both primary and secondary prevention trials.[1-5] This phenomenon reflects the fact that, even in low doses, aspirin stabilizes platelets by inducing permanent inhibition of platelet cyclooxygenase, a source of pro-aggregatory thromboxane. Moreover, recent meta-analyses examining long-term health outcomes in subjects who participated in controlled trials of daily low-dose aspirin, have concluded that those randomized to receive aspirin were 20% less likely to have died from cancer in the 20 years following their trial entry.[6, 7] This protection appeared to be confined to protection from adenocarcinomas; mortality from adenocarcinomas was 34% less likely in those receiving aspirin. These findings are particularly impressive in that none of the examined studies was longer than 9 years in duration – though, as could be expected, participants in longer-term studies tended to achieve greater protection from cancer. Partial inhibition of cox-2 in pre-neoplastic tissues is a likely explanation for the cancer protection afforded by aspirin.[7] Daily administration of aspirin appears to be crucial for this benefit, as no cancer prevention has been observed in studies in which aspirin was administered on alternate days.[8]

In light of low-dose aspirin's ability to reduce mortality from both vascular events and cancer to a very notable degree, it is tempting to recommend this measure not only for older subjects at risk for thrombosis, but for most healthy adults as well (excluding subjects who are aspirin-allergic or who have rare bleeding diatheses). However, oral aspirin, even in low doses, has a propensity to damage the gastroduodenal mucosa and increase risk for gastrointestinal bleeding; this fact may constrain health

authorities from recommending aspirin use for subjects deemed to be at low cardiovascular risk. Recent meta-analyses estimate that a year of low-dose aspirin therapy will induce major gastrointestinal bleeding (requiring hospitalization) in one subject out of 833; low-dose aspirin approximately doubles risk for such bleeds at any given age.[9, 10] Like other NSAIDS, aspirin traumatizes the gastrointestinal mucosa by inhibiting cyclooxygenase activity, preventing the production of prostaglandins which promote effective mucosal blood flow and stimulate production of the protective mucous lining.[11] Loss of mucosal prostaglandin activity also leads to an influx of activated neutrophils which exacerbate the tissue injury, in part via oxidative stress.[12-14]

It is well known that concurrent proton pump inhibitor therapy can notably decrease risk for NSAID-induced gastrointestinal bleeding. Individuals known to be at elevated risk for gastrointestinal bleeding are well advised to avail themselves of this protection when using low-dose aspirin or other NSAIDS.[15, 16] However, proton pump inhibitors are not devoid of risks of their own,[17] and it would be of questionable wisdom and practicality to put a high proportion of the adult population on these drugs. This essay proposes a more physiological strategy for providing gastroprotection that could be expected to complement the impact of low-dose aspirin on platelet function and vascular risk.

Gastroprotective Potential of Dietary Nitrate and Reductive Polyphenols

Nitric oxide (NO) has an impact on the gastrointestinal mucosa similar to that of prostaglandins – it boosts mucosal blood flow, stimulates mucus production, and inhibits influx of activated neutrophils.[18, 19] Although endothelial and neural NO synthase activity in mucosal tissue functions physiologically to protect it from trauma and ulceration, there is recent evidence that salivary nitrite also can provide significant protection in this regard, in light of the fact that the acidic gastric milieu enables conversion of nitrite to NO; physiological concentrations of ascorbic acid in gastric secretions stimulate this reaction via electron donation.[20] Salivary nitrite is notably increased by a nitrate-rich diet, as a portion of absorbed nitrate is secreted in saliva and reduced to nitrite by commensal oral bacteria (an effect prevented by anti-bacterial mouthwashes).[21] Lundberg and colleagues have demonstrated that, in an *in vivo* rat model, exposure of the gastric mucosa to acidified nitrite-rich human saliva boosts mucosal blood flow and increases the thickness of the protective mucus layer.[18] In subsequent studies, these researchers showed that a nitrate-rich diet protected the gastric mucosa of rats challenged with diclofenac (oral or intravenous) or taurocholate.[22, 23] These observations prompted the speculation that nitrate-rich diets might lessen risk for peptic ulceration in humans. Indeed, such a speculation appears consistent with epidemiology demonstrating that peptic ulceration is less common in people who use nitrovasodilator drugs.[24, 25]

The efficiency with which gastric nitric acid is converted to NO within the stomach is greatly enhanced by consumption of a wide range of reductive polyphenols possessing catechol groups.[26-33] These agents can donate a single electron to nitric acid, generating NO and a water molecule; in the process, they become resonance-stabilized semi-quinone radicals.[28] They can then donate a further electron to become quinones, undergo addition reactions with other semi-quinone radicals, or react to form adduct products with NO metabolites. Commonly occurring compounds with this property include (epi)catechin, epicatechin-3-O-gallate, quercetin, chlorogenic acid, caffeic acid, oleuropein, cyanidin, and procyanidins, which are richly supplied by foods and drinks such as tea, coffee, raw cocoa, red wine, grapes, apples, berries, olives, and a host of others. There is a considerable literature demonstrating that a range of

reductive polyphenols – or plant extracts which feature these compounds – can prevent or aid healing of gastric ulcers in rats.[34-44] Gastric production of NO likely contributes to this protection since, even in the context of a low-nitrate diet, metabolism of endogenously produced NO gives rise to a meaningful level of salivary nitrite.

Evidently, a high nitrate diet including frequent servings of foods rich in catechol-bearing polyphenols – and/or complemented with nutraceutical supplements featuring such polyphenols – will induce generation of a large amount of NO in the stomach, thereby promoting effective mucosal blood flow and production of gastric mucus. Arguably, consumption of such foods, drinks, or supplements at the same time as mini-dose aspirin is administered would provide the most potent protection – albeit, inasmuch as aspirin permanently inhibits cyclooxygenase, its impact on the gastric mucosa will be fairly durable, so consumption of these agents at other times of the day will likely amplify the protection achieved.

Complementary Platelet-Stabilizing and Vascular-Protective Effects

These findings are particularly intriguing in light of evidence that dietary nitrate also has platelet-stabilizing activity.[45, 46] Indeed, it is well known that NO, either per se or as spontaneously generated S-nitrosothiols, exerts physiological anti-aggregatory and anti-adhesive effects that are partially though not wholly mediated by cGMP, and that are complementary to the anti-aggregatory effects of aspirin; indeed, NO-donor aspirin derivatives have long been studied as potential anti-thrombotic agents.[47-52] Moreover, chronic low-dose aspirin therapy somehow compromises the ability of beta-agonists to activate platelet NO synthase – an effect which may lessen aspirin's utility for platelet stabilization.[53] There is growing evidence that nitrate-rich diets, by increasing plasma levels of nitrite, promote the generation of NO in tissues – most notably in tissues that are hypoxic and acidotic; this conversion of nitrite to NO is catalyzed by deoxyheme proteins or xanthine oxidoreductase.[46, 54-56] Two clinical studies have demonstrated that nitrate-rich diets inhibit platelet function *ex vivo*.[45, 46] Epidemiologically, diets rich in green leafy vegetables – rich natural sources of nitrate - are associated with decreased risk for MI or stroke – an effect which, in part, may reflect NO-mediated platelet stabilization (albeit NO and dietary nitrate also have important anti-atherosclerotic and anti-hypertensive effects – and provide protection from ischemia-reperfusion injury).[57-62]

Moreover, to the extent that catechol-bearing polyphenols are absorbable, at least some of them – quercetin and epicatechin have received particular attention in this regard – can act directly on vascular endothelium to stimulate the activity of the endothelial NO synthase.[63, 64] This explains why foods containing these compounds (such as cocoa) have anti-hypertensive potential[65-68] – and may rationalize epidemiology associating high dietary consumption of flavonols (of which quercetin is the most prominent) with lower cardiovascular risk.[69, 70] The mechanism involved in this effect is still somewhat obscure, but may involve oxidant-mediated activation of certain membrane potassium channels which promote membrane hyperpolarization.[71, 72] Not unlikely, the propensity of these compounds to donate a single electron may account for their abilities to generate NO in the stomach *and* to activate endothelial NO synthase. Evidently, increased endothelial NO synthase activity will tend to stabilize platelets, while acting in a number of other complementary ways to preserve vascular health. Indeed, ingestion of grape juice, red wine, raw cocoa, and supplemental quercetin has been shown to inhibit platelet function *ex vivo* in humans. [73-82]

Practical Implications for Health Protection

It is therefore straightforward to propose that low-dose aspirin regimens may be notably safer in individuals ingesting nitrate-rich diets – particularly if these diets are also rich in catechol-bearing polyphenols. And such diets are likely to complement the platelet-stabilizing efficacy of aspirin. Whether such diets might afford protection from aspirin or NSAID-induced *intestinal* ulceration is not yet clear, though some S-nitrosothiols generated in the stomach might be expected to reach the duodenum, and nitrite in the duodenal circulation might give rise to NO when this circulation is compromised. Encouragingly, a recent study finds that a nitrate-rich diet prevents influx of leukocytes into the small intestinal mucosa in rodents gavaged with diclofenac.[83] Hence, there is reason to suspect that the protection from NSAID damage afforded by dietary nitrate may extend to the small intestine as well.

Dark green leafy vegetables – most notably spinach and collard greens – as well as beets and beet juice, are among the richest dietary sources of nitrate.[84, 85] Since it may be unrealistic to expect people to eat ample servings of these every day, it is fortunate that supplemental potassium nitrate is quite inexpensive, and could be used to "buffer" nitrate intake on days when dietary nitrate would otherwise be suboptimal. Greater protection might be achieved if aspirin is ingested in conjunction with drinks high in catechol-bearing polyphenols – notably tea, coffee, raw cocoa, red wine, grape juice, apple juice, among others. Nutraceutical polyphenol or ascorbic acid supplements could also be employed for this purpose. Consuming these drinks or supplements multiple times daily, while maintaining a nitrate-rich diet, might afford the greatest protection to the gastric mucosa. One very simple way to implement this idea would be to produce a capsule featuring both potassium nitrate and an appropriate polyphenol – such as quercetin – that could be taken with meals several times daily.

Within this dietary context, it might be decidedly safer and more feasible to recommend low-dose aspirin supplementation to a high proportion of the population – thereby achieving notable reductions not only in vascular events, but also cancer mortality. And this dietary advice would be highly protective for vascular health in its own right.

In regard to cancer risk and nitrate exposure, it should be noted that, whereas nitrite-preserved processed meats have been linked to increased cancer risk in epidemiology, nitrate of plant origin by-and-large has not been linked to cancer risk, and indeed has been associated with decreased risk in some studies.[18] Generation of nitrosamines in heated processed meats may largely mediate the cancer risk associated with consumption of such meat. However, rat studies suggest that amplified gastric generation of NO in the context of reflux esophagitis may give rise to cytotoxic concentrations of peroxynitrite at the gastroesophageal junction and thereby promote tissue transformation analogous to Barrett's esophagus;[86-89] these findings suggest that dietary strategies which induce increased gastric NO production could prove to be contraindicated in those with reflux esophagitis. Nonetheless, frequent consumption of green leafy vegetables and ascorbate-rich foods consistently correlates *negatively* with risk for esophageal adenocarcinoma in epidemiological studies.[90]

Finally, it should be noted that various effective antioxidant measures – which arguably should be incorporated into nutraceutical preventive health regimens for a host of other reasons – may also have potential for decreasing the risk for ulceration associated with aspirin use, in light of the role which neutrophil-mediated oxidative stress plays in this pathology. Indeed, some years ago, pre-administration of a nitroxide drug with superoxide quenching and dismutating activity was shown to provide profound

protection from gastric ulceration in rats challenged with aspirin or indomethacin.[91] Although this agent has never been developed for clinical use, astaxanthin, taurine, high-dose folate, and heme-oxygenase-1 inducers have subsequently been shown to provide partial protection in rodent models of gastric ulcer induction.[92-98] Spirulina, which has never been tested in gastric ulcer models, may also have potential in this regard, as its key phytochemical phycocyanobilin can inhibit NADPH oxidase, the source of neutrophil-derived oxidants.[99] Such measures, which, like low-dose aspirin, have potential for vascular protection and cancer prevention, might nicely complement NO-boosting strategies in minimizing risk for aspirin-provoked peptic ulceration.

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