Published in final edited form as:

Nutr Cancer. 2011 April ; 63(3): 319–326. doi:10.1080/01635581.2011.535960.

Circulating 25-Hydroxyvitamin-D and Risk of Colorectal Adenomas and Hyperplastic Polyps

Scott V. Adams^{1,2}, Polly A. Newcomb^{1,2}, Andrea N. Burnett-Hartman^{1,2}, Emily White^{1,2}, Margaret T. Mandelson^{2,3}, and John D. Potter^{1,2}

¹ Department of Epidemiology, University of Washington, 1959 NE Pacific St., Box 357236, Seattle, WA, 98195 USA

² Program in Cancer Prevention, Fred Hutchinson Cancer Research Center, 1100 Fairview Ave N, Seattle, WA, 98109 USA

³ Group Health Research Institute, 1730 Minor Ave, Suite 1600, Seattle, WA, 98101 USA

Abstract

Colorectal adenomas are clear precursors of cancer; hyperplastic polyps may also have malignant potential. An inverse association between circulating vitamin D metabolites and adenoma risk has been reported, but less is known about vitamin D and hyperplastic polyps. We conducted a case-control study of adenomas and hyperplastic polyps among 459 members of an integrated health plan evaluated via colonoscopy. Questionnaires provided information on colorectal polyp risk factors, and plasma samples were assayed for 25-hydroxyvitamin-D (25(OH)D). Polytomous regression was used to estimate odds ratios for adenomas (N=149) and hyperplastic polyps (N=85) compared to polyp-free controls (N=225) by tertile of 25(OH)D. An inverse association between 25(OH)D and adenomas was suggested after adjustment for potential confounding factors (comparing upper to lower tertiles, OR[95%CI]: 0.71 [0.38–1.30]). After restriction of the analyses to study participants with no history of polyps, this OR estimate was reduced further (adjusted OR [95%CI]: 0.52 [0.23–1.20]). In comparison, no inverse association between hyperplastic polyps and 25(OH)D was observed among the full study participants (adjusted OR [95%CI]: 1.17 [0.55–2.51]) or among those without prior polyps (adjusted OR [95%CI]: 1.42 [0.55–3.65]). Our study suggests that the established inverse association between circulating 25(OH)D and adenoma may not apply to hyperplastic polyps.

Keywords

Vitamin D; 25-hydroxyvitamin D; colorectal adenoma; hyperplastic polyp; epidemiology; case/control

INTRODUCTION

Adenomatous colorectal polyps (adenomas) are recognized precursor lesions of colorectal cancer (1). Hyperplastic polyps are common lesions that have historically been considered benign, but recent evidence suggests that some hyperplastic polyps may progress to

Corresponding Author & Request for Reprints: Scott V Adams, PhD, Fred Hutchinson Cancer Research Center, 1100 Fairview Ave N, Mailstop M4-B402, Seattle, WA 98109 USA, sadams@fhcrc.org, Tel: +1 206 667 6427, Fax: +1 206 667 7850.

The preliminary results of this study were presented at the American Society of Preventive Oncology meeting, March 2010 (Bethesda, MD).

Competing Interests: The authors have no competing interests.

malignancy along the “serrated polyp” pathway (2–5). Studies of colorectal neoplasia, including studies evaluating the association between vitamin D and colorectal neoplasia, have often used adenomas as an outcome to understand risk factors involved in colorectal cancer initiation. In contrast, hyperplastic polyps were seldom included in such studies.

There are multiple regulatory targets of the vitamin D receptor, suggesting several pathways by which vitamin D could influence risk of colorectal adenomas and cancer, and possibly hyperplastic polyps. These include regulation of cell cycle, bile-acid catabolism, growth-factor signaling, and apoptosis (6, 7). Although diet and supplements are also sources, vitamin D is primarily synthesized in skin in response to exposure to UV radiation, and subsequently converted to 25-hydroxyvitamin-D (25(OH)D) (8). The active form, 1,25-dihydroxyvitamin-D (1,25(OH)₂D) (8, 9), is synthesized intracellularly from 25(OH)D in the kidney and many other tissues (9) including colon (10). Because blood 1,25(OH)₂D content is highly regulated (9), 25(OH)D is recognized as the circulating metabolite most salient to monitoring physiologic levels of vitamin D (11).

Previous observational studies have reported an inverse association between 25(OH)D concentration and risk of colorectal adenomas (12–17) and cancer (18, 19). Randomized controlled trials of vitamin D supplementation and colorectal cancer have had mixed results (16, 20, 21), possibly as a result of differences in supplement dose (22, 23), insufficient follow-up (23), or interactions with exogenous hormone use by female participants (24).

Despite the depth of the literature on vitamin D and colorectal neoplasia, little is known about the relationship between vitamin D and hyperplastic polyps. To our knowledge, no previous study has evaluated the association between hyperplastic polyps and circulating 25(OH)D, and the few studies of hyperplastic polyp risk that included information on vitamin D intake failed to identify any relationship (25, 26). We therefore conducted a clinic-based case-control study to evaluate the association between circulating 25(OH)D and hyperplastic polyps, and to compare it to the association between 25(OH)D and adenomas. Because hyperplastic polyps and adenomas may have unique molecular origins and are believed to develop through different pathways (3, 4), we hypothesized that plasma 25(OH)D may have distinct associations with adenomas and hyperplastic polyps.

METHODS

Study Approval

This research protocol was approved by the Institutional Review Boards of the Fred Hutchinson Cancer Research Center and Group Health.

Study Population and Case Definitions

Participants were part of a clinic-based study of biomarkers of colorectal neoplasia risk conducted at Group Health, a large integrated healthcare delivery system in western Washington state, as described by Chia *et al* (27). Of 738 participants evaluated for colorectal disease by colonoscopy enrolled in the parent study, 94 patients were newly diagnosed with colorectal cancer, familial adenomatous polyposis, ulcerative colitis, or Crohn’s disease, making them ineligible for this study of polyps. Of the remaining 644 participants, 559 donated a blood sample at the index colonoscopy. We also excluded 49 with incomplete data, and a further 51 with self-reported previous diagnosis of colorectal cancer, ulcerative colitis, familial adenomatous polyposis, or Crohn’s disease.

A total of 149 adenoma cases, with or without hyperplastic polyps; 85 hyperplastic polyp cases; and 225 controls without evidence of polyps at colonoscopy were included in this study. Participants included in the adenoma case group had at least one pathologically

confirmed adenomatous polyp detected at colonoscopy. Of 149 adenoma cases included in this study, 44 also had hyperplastic polyps. Participants included in the hyperplastic polyp case group had one or more pathologically confirmed hyperplastic polyps, but no adenomas, detected at colonoscopy.

Data and Specimen Collection

In brief, consenting participants completed a self-administered questionnaire detailing personal and family medical history along with demographic, lifestyle, and some dietary factors, and provided a blood sample prior to colonoscopy (27). Samples were fractionated and stored at -70°C from collection until aliquoted for this study.

Measurement of 25-Hydroxyvitamin-D Concentrations

25(OH)D concentration in stored plasma samples was assessed at the University of Washington Clinical Nutrition Research Unit, by isotope-dilution liquid chromatography-tandem mass spectrometry (28) on a Micromass Quattro *micro* API tandem quadrupole system (Waters Corp., Milford, MA). This method measures two forms of 25(OH)D, 25(OH)D₂ and 25(OH)D₃, with detection limits of 1ng/mL for each form. Coefficients of variation measured on in-house standards were less than 8.5% when tested at 10 and 30ng/mL (25(OH)D₃) or 15.3 and 36.5 ng/mL (25(OH)D₂). For this analysis, the sum of 25(OH)D₂ and 25(OH)D₃ above the detection limit are reported as 25(OH)D concentration. All samples had quantifiable concentrations of 25(OH)D₃; 274 results for 25(OH)D₂ below detection were assigned a value of zero.

Statistical Analysis

We calculated multivariable-adjusted odds ratios (OR) and confidence intervals for adenoma and hyperplastic polyp risk using polytomous (multinomial) regression analyses. Two models of the association between 25(OH)D and polyps were examined. First, total 25(OH)D concentrations (ng/mL) were divided into tertiles using cut-points from the entire control group. In a second model, total 25(OH)D concentration was included in units of 10 ng/mL as a continuous variable with a single linear term. Wald tests comparing coefficients across polytomous equations were used to assess the statistical significance of associations between 25(OH)D; polytomous models allow comparison of model coefficients between each case group and the controls, as well as directly between case groups.

Covariates evaluated included age (years), sex, body mass index (BMI, kg/m^2), self-reported previous polyps (yes, no, unknown), season of blood draw (May–October; November–April), physical activity (metabolic equivalent (MET) hours per week, categorized into four groups), cigarette smoking history (never-smoker; smoker for 20 or fewer pack-years, smoker for more than 20 pack-years), current supplement use (yes or no), ethnicity (Non-Hispanic white, other), prior endoscopy (never, ever), and self-reported previous cancer (other than colorectal). A base model including the potential confounders age, sex, previous polyp diagnosis, BMI, cigarette smoking history, and season of blood draw was constructed based on previously published studies. Ethnicity, prior endoscopy, and previous (non-colorectal) cancer diagnosis, when added singly to this base model, did not change odds ratios for adenoma or hyperplastic polyps by more than 10%; for this reason these variables were excluded. To the base model, the exogenous sources of 25(OH)D, physical activity and supplement use, were added to assess whether further adjustment was needed despite direct measurement of 25(OH)D. Results after adjustment for the base model, physical activity and supplement use are referred to as “fully adjusted.”

Adjustment factors, except age and BMI (kg/m^2), were coded as categorical variables. Age and BMI were incorporated into statistical models as continuous variables with a single

linear term. Physical activity quartile was included as a grouped-linear term. Participants who indicated current use of multivitamins, calcium supplement, or vitamin D supplements at least once weekly were identified as “supplement users.” Calcium supplement users were included as “supplement users” because calcium and vitamin D are often combined in supplements.

Two additional sub-analyses were performed. First, to estimate the independent associations of 25(OH)D₂ and 25(OH)D₃ with polyps, 25(OH)D₂ and 25(OH)D₃ concentrations were both included in an additional polytomous model as continuous linear variables, along with the same covariates as in the fully adjusted primary analysis.

Second, we investigated the association between total 25(OH)D concentration and polyps according to the site of the lesion within the colon and rectum. Cases were categorized as “distal” if one or more lesions were present in the rectum, rectosigmoid junction, or descending colon; or as “proximal” if the lesion(s) were found in the cecum, ascending, or transverse colon. Each case was placed in one or more categories: proximal adenoma, distal adenoma, proximal hyperplastic polyp, and distal hyperplastic polyp. Participants with both adenomas and hyperplastic polyps entered in the site-specific analysis only in the distal and/or proximal adenoma groups. Thus for these participants, hyperplastic polyps were not included in the sub-site analysis. Information on the site of the lesion was unavailable for 21 participants with adenoma(s), and 18 participants with hyperplastic polyp(s). Multivariable logistic regression was employed to estimate associations between total 25(OH)D and each of four case categories (proximal adenoma, distal adenoma, proximal hyperplastic, distal hyperplastic) compared to the entire control group, fully adjusted as for the primary analysis.

Statistical analysis was completed using Stata versions 10 and 11 (StataCorp LP, College Station, TX).

RESULTS

Mean plasma 25(OH)D concentrations in adenoma cases (23.1 ng/mL) and in hyperplastic polyp cases (24.2 ng/mL) were slightly lower than those in controls (24.9 ng/mL) (Table 1). Notably higher 25(OH)D concentrations were found in blood drawn from participants with BMI below 25 kg/m², higher reported levels of physical activity, non-Hispanic whites, and samples drawn between June and October. Current regular users (one or more times per week) of dietary supplements (multivitamins, calcium supplements, or vitamin D supplements) also showed higher plasma 25(OH)D concentration.

Compared to controls, both adenoma and hyperplastic polyp cases were more likely to be male, older, report a previous diagnosis of a polyp, or have a history of cigarette smoking (Table 1). Participants diagnosed with only hyperplastic polyps were more likely to have a history of cigarette use, to be obese (BMI > 30 kg/m²), to have donated blood between November and May, and to report supplement use than either adenoma cases or controls. Ethnicity and previous diagnosis with non-colorectal cancer (not shown) did not differ by case status.

In the entire study population, adjusted ORs (Table 2) suggested an inverse association between plasma 25(OH)D and adenoma (OR, upper tertile compared to lowest [95%CI]: 0.66 [0.36–1.19]). After further adjustment for current supplement use and physical activity, this relationship was largely unchanged (OR [95%CI]: 0.71 [0.38–1.30]). Results with 25(OH)D as a continuous, linear variable were similar.

In contrast, a direct association between plasma 25(OH)D and hyperplastic polyps (Table 2; adjusted OR [95%CI], upper tertile compared to lowest: 1.39 [0.69–2.81]) was suggested.

Additional adjustment for supplement use and physical activity attenuated this estimate (OR [95%CI]: 1.17 [0.55–2.51]).

Previous diagnosis of a polyp modestly influenced the association between 25(OH)D and adenoma (Table 2). Among adenoma cases and controls reporting no previous diagnosis with a polyp, the inverse association of 25(OH)D with adenoma was stronger (fully adjusted OR [95%CI] for third tertile compared to first tertile: 0.52 [0.23–1.20]). Restriction in this manner among hyperplastic polyp cases also moved adjusted estimates away from 1 (fully adjusted OR [95%CI]: 1.42 [0.55–3.65]). Comparison of base-model estimates with fully adjusted estimates, however, suggested the possibility of some confounding by supplement use and physical activity among participants without previous polyps; the observed change in OR with adjustment was largely due to inclusion of supplement use in the statistical model. The difference in the OR estimates for adenoma and hyperplastic polyps, comparing the upper tertile of 25(OH)D to the lowest, approached statistical significance for base-model adjusted estimates and fully adjusted estimates ($p=0.03$ and 0.08 by Wald tests, respectively; Table 2).

The anatomic site of the polyp was considered by assigning each polyp to either the distal or proximal colon based on information in the colonoscopy clinical records. Among all adenoma cases, distal adenomas ($n=96$) were slightly more strongly associated with 25(OH)D concentrations than proximal adenomas ($n=58$) (fully adjusted OR [95%CI] per 10ng/mL 25(OH)D, distal: 0.76 [0.53–1.09]; proximal: 0.93 [0.61–1.42]), and when analysis was restricted to participants without a history of polyps (fully adjusted OR [95%CI] per 10ng/mL 25(OH)D, distal: 0.65 [0.38–1.11]; proximal: 0.87 [0.48–1.57]). Among all hyperplastic polyp cases, a smaller difference was observed between distal ($n=52$) and proximal ($n=19$) lesions (fully adjusted OR [95%CI] per 10ng/mL 25(OH)D, distal: 0.96 [0.61–1.49]; proximal: 0.98 [0.56–1.73]), and after restriction to participants without previous polyps (OR [95%CI] per 10ng/mL 25(OH)D, distal: 0.99 [0.52–1.90]; proximal: 1.01 [0.52–1.95]).

Analysis of the independent effects 25(OH)D₂ and 25(OH)D₃ showed that, included as separate continuous linear terms in the same model, the fully adjusted OR for the association between 25(OH)D₃ and adenoma among patients without previous polyps was 0.77 [0.54–1.09] per 10ng/mL; for 25(OH)D₂ the corresponding OR was 1.03 [0.51–2.12]. However, the highest measured 25(OH)D₂ concentration was 21.6 ng/mL, much lower than the highest 25(OH)D₃ concentration of 60.2 ng/mL, and lower than approximately one third of all 25(OH)D₃ concentrations. No difference was observed between the association of hyperplastic polyps with 25(OH)D₂ (fully adjusted OR [95%CI] per 10ng/mL: 1.06 [0.44–2.54]) or 25(OH)D₃ (fully adjusted OR [95%CI] per 10ng/mL: 1.06 [0.70–1.60]).

DISCUSSION

In this colonoscopy-based case-control study we observed the suggestion of an inverse association between plasma 25(OH)D concentration and colorectal adenomas, but not hyperplastic polyps. When we restricted analysis to participants who reported no previous diagnoses of polyps, the inverse association became stronger for adenomas. These results might be expected if previous polyps indicate a predisposition to polyp formation that attenuates an inverse relationship between vitamin D and adenomas. In contrast, restriction in this manner had less impact on the estimate of the association between hyperplastic polyps and 25(OH)D.

To our knowledge, this is the first study to examine the association of circulating vitamin D metabolites with both colorectal hyperplastic polyps and adenomas in the same population.

Recent meta-analyses (17) of previous studies (12–16) have supported an inverse relationship between circulating vitamin D metabolites and first occurrence of adenomas; a similar association has been observed for frank colorectal cancer (18, 29). Vitamin D has not been widely studied as a risk factor for hyperplastic polyps; the only two studies of which we are aware examining crude dietary intake of vitamin D, without direct measurement of circulating vitamin D metabolites, found no association (25, 26).

This study has several strengths, including the performance of colonoscopy on all participants, so that we can be confident that controls were, in fact, polyp-free at the time of this study. Secondly, blood sampling and questionnaire responses were obtained prior to colonoscopy and resulting diagnosis, so that participation rates, recall, and 25(OH)D were unlikely to be affected by case-control status. We were able to identify both adenomas and hyperplastic polyps in the same population, and our results reproduce the well-characterized relation between 25(OH)D and adenoma (17), increasing our confidence in the findings.

Another important strength is the direct assessment of circulating concentration of the salient vitamin D metabolite, 25(OH)D, with a mass spectrometry-based method that has high sensitivity and specificity for 25(OH)D (28) and is considered the “gold-standard” for clinical assessment of 25(OH)D (30). This method can also distinguish 25(OH)D₂ and 25(OH)D₃ (28, 30). Vitamin D₃ is produced in the human body as a result of exposure to UV radiation, is the most common form of vitamin D in supplements, is found in foods such as fish, and is most commonly added to milk (8, 31, 32). Vitamin D₂ is a component of some supplements, for example, those obtained by physician prescription in the US, and may be present in small amounts in some foods such as mushrooms (31–33). We observed that 25(OH)D₂ levels above 5.1 ng/mL were only found in participants who indicated taking supplements, and that 25(OH)D₃ concentrations increased with supplement use and higher levels of physical activity (data not shown). When 25(OH)D₂ and 25(OH)D₃ were separately accounted for in a single regression model, 25(OH)D₃ was inversely associated with adenomas but not hyperplastic polyps, while 25(OH)D₂ was not associated with either polyp. However, the restricted range of 25(OH)D₂ concentrations hampered our ability to conclude that only 25(OH)D₃, and not 25(OH)D₂, is likely to be inversely associated with adenoma risk.

In a sub-analysis, we divided each case group between two subgroups according to the site of polyp(s). The suggested inverse association between 25(OH)D and adenoma was stronger for adenomas in the distal colon and rectum than for proximal adenomas. Many previous studies of 25(OH)D and adenomas have been sigmoidoscopy-based, and therefore, unable to examine in detail differences in association with respect to lesion location throughout the colorectum (12, 14, 15). The results of other studies that did examine lesion site have been mixed. A stronger association of 25(OH)D with distal lesions was not observed in at least two studies (13, 34), but was reported in another study based on dietary intake of vitamin D (35).

The limitations of this study include its cross-sectional nature. We assayed 25(OH)D in plasma collected at the time of colonoscopy and assessed its relationship with polyps identified at the same colonoscopy. Although there is little reason to suspect that the presence of polyps alters circulating 25(OH)D, we cannot know whether measured 25(OH)D reflects 25(OH)D levels prior to polyp formation. However, recent longitudinal studies of 25(OH)D levels support the validity of a single 25(OH)D measurement as a marker of long-term 25(OH)D status (36, 37).

The study sample comprised largely non-Hispanic whites, which may limit the application of these results to more diverse populations. We observed that 25(OH)D concentrations

were lower among non-whites in this study sample, consistent with nationally representative samples (38). Although vitamin D receptor polymorphisms might vary across ethnicities (39), the functional relevance of these polymorphisms in relation to 25(OH)D and colorectal polyps remains uncertain.

In addition, we were unable to assess intake of calcium, possibly an important co-factor with 25(OH)D in colorectal polyp development (40–42). Therefore, some of the association we observe between vitamin D and colorectal adenomas might be due to differences in calcium levels rather than vitamin D levels. However, vitamin D increases absorption of ingested calcium (32), an action that might mediate part of the association between 25(OH)D and adenoma or hyperplastic polyps (40–42). Therefore, complete adjustment for calcium status may be inappropriate when estimating the association between 25(OH)D and polyps, and our inability to assess calcium intake is unlikely to substantially impact the interpretation of our results.

Notably, adjustment for reported physical activity level and current use of multivitamins, vitamin D supplements, or calcium supplements only weakly attenuated the association between circulating 25(OH)D and adenoma (Table 2). Adjustment for activity and supplement use resulted in decreases of the ORs between 25(OH)D and hyperplastic polyps, particularly among participants without a history of prior polyps. This was mainly attributable to adjustment for supplement use, and only a small portion was related to adjustment for physical activity. This suggests that although physical activity and supplement use are both sources of 25(OH)D, only supplement use is a potentially important confounder of the association between 25(OH)D and hyperplastic polyps in this study sample, after adjustment for the other variables in our base model. Because of limited data we were unable to more fully adjust for supplement use, for example, by including dose information. Although such additional adjustment might further attenuate the observed association between 25(OH)D and hyperplastic polyps, it is unlikely to fully account for the observed qualitative differences in comparison to the association between 25(OH)D and adenomas.

Emerging molecular evidence suggests that a subset of colorectal cancers displaying high microsatellite instability and CpG-island methylation may progress from hyperplastic polyps along the “serrated polyp” pathway (3–5), distinct from the more common adenoma-cancer pathway (1). Epidemiologic studies, however, have generally found few differences between risk factors for hyperplastic polyps and adenomas; the most consistently observed differences are a stronger association between cigarette smoking and hyperplastic polyps than adenomas, and a younger mean age of individuals with hyperplastic polyps (25, 26, 43–46).

The results of this study provide preliminary evidence that vitamin D status may be another risk factor that differs between adenomas and hyperplastic polyps, suggesting that vitamin D and the molecular processes regulated by the vitamin D receptor might provide some insights into etiologic differences. In particular, the regulatory actions of the vitamin D receptor on the *Wnt*/β-catenin signaling pathway, which is critical to adenoma development, has been suggested to mediate the apparent protective effect of vitamin D against adenomas and colorectal cancer (47–50). Although speculative, our results are consistent with a model in which the *Wnt*/β-catenin pathway is less relevant to the development of hyperplastic polyps, and perhaps also to the progression of hyperplastic polyps to sessile serrated adenomas and colorectal cancer.

Acknowledgments

Funding: This study was supported by a National Cancer Institute grant (P01 CA74184), training funds to ANBH (T32 CA09168-32), and additional research funding and support for SVA from a National Institute of Health, National Cancer Institute Cancer Prevention Training Grant (R25 CA094880).

The authors thank Thomas Laha and Andrew Hoofnagle at the University of Washington Clinical Nutrition Research Unit (Seattle, WA) for performing the 25(OH)D assays.

References

1. Kinzler KW, Vogelstein B. Lessons from hereditary colorectal cancer. *Cell*. 1996; 87:159–70. [PubMed: 8861899]
2. Jass JR. Hyperplastic-like polyps as precursors of microsatellite-unstable colorectal cancer. *Am J Clin Pathol*. 2003; 119:773–5. [PubMed: 12817423]
3. O'Brien MJ. Hyperplastic and serrated polyps of the colorectum. *Gastroenterol Clin North Am*. 2007; 36:947–68. [PubMed: 17996799]
4. Jass JR. Classification of colorectal cancer based on correlation of clinical, morphological and molecular features. *Histopathology*. 2007; 50:113–30. [PubMed: 17204026]
5. East JE, Saunders BP, Jass JR. Sporadic and syndromic hyperplastic polyps and serrated adenomas of the colon: Classification, molecular genetics, natural history, and clinical management. *Gastroenterol Clin North Am*. 2008; 37:25–46. [PubMed: 18313538]
6. Bouillon R, Eelen G, Verlinden L, Mathieu C, Carmeliet G, et al. Vitamin D and cancer. *J Steroid Biochem Mol Biol*. 2006; 102:156–62. [PubMed: 17113979]
7. Bouillon R, Carmeliet G, Verlinden L, van Etten E, Verstuyf A, et al. Vitamin D and human health: Lessons from vitamin D receptor null mice. *Endocr Rev*. 2008; 29:726–76. [PubMed: 18694980]
8. DeLuca HF. Overview of general physiologic features and functions of vitamin D. *Am J Clin Nutr*. 2004; 80:1689S–96S. [PubMed: 15585789]
9. Holick MF. Sunlight and vitamin d for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease. *Am J Clin Nutr*. 2004; 80:1678S–88S. [PubMed: 15585788]
10. Tangpricha V, Flanagan JN, Whitlatch LW, Tseng CC, Chen TC, et al. 25-hydroxyvitamin D-1 α -hydroxylase in normal and malignant colon tissue. *Lancet*. 2001; 357:1673–4. [PubMed: 11425375]
11. Hollis BW. Assessment of vitamin d status and definition of a normal circulating range of 25-hydroxyvitamin D. *Curr Opin Endocrinol Diabetes Obes*. 2008; 15:489–94. [PubMed: 18971676]
12. Peters U, Hayes RB, Chatterjee N, Shao W, Schoen RE, et al. Circulating vitamin d metabolites, polymorphism in vitamin d receptor, and colorectal adenoma risk. *Cancer Epidemiol Biomarkers Prev*. 2004; 13:546–52. [PubMed: 15066918]
13. Peters U, McGlynn KA, Chatterjee N, Gunter E, Garcia-Closas M, et al. Vitamin D, calcium, and vitamin D receptor polymorphism in colorectal adenomas. *Cancer Epidemiol Biomarkers Prev*. 2001; 10:1267–74. [PubMed: 11751444]
14. Levine AJ, Harper JM, Ervin CM, Chen YH, Harmon E, et al. Serum 25-hydroxyvitamin D, dietary calcium intake, and distal colorectal adenoma risk. *Nutr Cancer*. 2001; 39:35–41. [PubMed: 11588900]
15. Platz EA, Hankinson SE, Hollis BW, Colditz GA, Hunter DJ, et al. Plasma 1,25-dihydroxy- and 25-hydroxyvitamin D and adenomatous polyps of the distal colorectum. *Cancer Epidemiol Biomarkers Prev*. 2000; 9:1059–65. [PubMed: 11045788]
16. Grau MV, Baron JA, Sandler RS, Haile RW, Beach ML, et al. Vitamin D, calcium supplementation, and colorectal adenomas: Results of a randomized trial. *J Natl Cancer Inst*. 2003; 95:1765–71. [PubMed: 14652238]
17. Wei MY, Garland CF, Gorham ED, Mohr SB, Giovannucci E. Vitamin D and prevention of colorectal adenoma: A meta-analysis. *Cancer Epidemiol Biomarkers Prev*. 2008; 17:2958–69. [PubMed: 18990737]

18. Gorham ED, Garland CF, Garland FC, Grant WB, Mohr SB, et al. Optimal vitamin D status for colorectal cancer prevention: A quantitative meta analysis. *Am J Prev Med.* 2007; 32:210–6. [PubMed: 17296473]
19. Yin L, Grandi N, Raum E, Haug U, Arndt V, et al. Meta-analysis: Longitudinal studies of serum vitamin D and colorectal cancer risk. *Aliment Pharmacol Ther.* 2009; 30:113–25. [PubMed: 19392870]
20. Wactawski-Wende J, Kotchen JM, Anderson GL, Assaf AR, Brunner RL, et al. Calcium plus vitamin D supplementation and the risk of colorectal cancer. *N Engl J Med.* 2006; 354:684–96. [PubMed: 16481636]
21. Lappe JM, Travers-Gustafson D, Davies KM, Recker RR, Heaney RP. Vitamin D and calcium supplementation reduces cancer risk: Results of a randomized trial. *Am J Clin Nutr.* 2007; 85:1586–91. [PubMed: 17556697]
22. Holick MF. Calcium plus vitamin D and the risk of colorectal cancer. *N Engl J Med.* 2006; 354:2287–8. author reply 2287–8. [PubMed: 16723623]
23. Giovannucci E. Calcium plus vitamin D and the risk of colorectal cancer. *N Engl J Med.* 2006; 354:2287–8. author reply 2287–8. [PubMed: 16729368]
24. Ding EL, Mehta S, Fawzi WW, Giovannucci EL. Interaction of estrogen therapy with calcium and vitamin D supplementation on colorectal cancer risk: Reanalysis of women's health initiative randomized trial. *Int J Cancer.* 2008; 122:1690–4. [PubMed: 18092326]
25. Morimoto LM, Newcomb PA, Ulrich CM, Bostick RM, Lais CJ, et al. Risk factors for hyperplastic and adenomatous polyps: Evidence for malignant potential? *Cancer Epidemiol Biomarkers Prev.* 2002; 11:1012–8. [PubMed: 12376501]
26. Lieberman DA, Prindiville S, Weiss DG, Willett W. Risk factors for advanced colonic neoplasia and hyperplastic polyps in asymptomatic individuals. *Jama.* 2003; 290:2959–67. [PubMed: 14665657]
27. Chia VM, Newcomb PA, Lampe JW, White E, Mandelson MT, et al. Leptin concentrations, leptin receptor polymorphisms, and colorectal adenoma risk. *Cancer Epidemiol Biomarkers Prev.* 2007; 16:2697–703. [PubMed: 18086776]
28. Maunsell Z, Wright DJ, Rainbow SJ. Routine isotope-dilution liquid chromatography-tandem mass spectrometry assay for simultaneous measurement of the 25-hydroxy metabolites of vitamins D2 and D3. *Clin Chem.* 2005; 51:1683–90. [PubMed: 16020493]
29. Huncharek M, Muscat J, Kupelnick B. Colorectal cancer risk and dietary intake of calcium, vitamin D, and dairy products: A meta-analysis of 26,335 cases from 60 observational studies. *Nutr Cancer.* 2009; 61:47–69. [PubMed: 19116875]
30. Vogeser M. Quantification of circulating 25-hydroxyvitamin D by liquid chromatography-tandem mass spectrometry. *J Steroid Biochem Mol Biol.* 2010
31. Yetley EA. Assessing the vitamin D status of the us population. *Am J Clin Nutr.* 2008; 88:558S–564S. [PubMed: 18689402]
32. Holick MF. Vitamin D deficiency. *N Engl J Med.* 2007; 357:266–81. [PubMed: 17634462]
33. Ko JA, Lee BH, Lee JS, Park HJ. Effect of uv-b exposure on the concentration of vitamin D2 in sliced shiitake mushroom (*lentinus edodes*) and white button mushroom (*agaricus bisporus*). *J Agric Food Chem.* 2008; 56:3671–4. [PubMed: 18442245]
34. Fedirko V, Bostick RM, Goodman M, Flanders WD, Gross MD. Blood 25-hydroxyvitamin D3 concentrations and incident sporadic colorectal adenoma risk: A pooled case-control study. *American journal of epidemiology.* 2010; 172:489–500. [PubMed: 20650953]
35. Oh K, Willett WC, Wu K, Fuchs CS, Giovannucci EL. Calcium and vitamin D intakes in relation to risk of distal colorectal adenoma in women. *Am J Epidemiol.* 2007; 165:1178–86. [PubMed: 17379616]
36. Jorde R, Sneve M, Hutchinson M, Emaus N, Figenschau Y, et al. Tracking of serum 25-hydroxyvitamin D levels during 14 years in a population-based study and during 12 months in an intervention study. *Am J Epidemiol.* 2010; 171:903–8. [PubMed: 20219763]
37. Hofmann JN, Yu K, Horst RL, Hayes RB, Purdue MP. Long-term variation in serum 25-hydroxyvitamin D concentration among participants in the prostate, lung, colorectal, and ovarian

- cancer screening trial. *Cancer Epidemiol Biomarkers Prev.* 2010; 19:927–31. [PubMed: 20332255]
38. Ginde AA, Liu MC, Camargo CA. Demographic differences and trends of vitamin D insufficiency in the us population, 1988–2004. *Archives of Internal Medicine.* 2009; 169:626–632. [PubMed: 19307527]
  39. Uitterlinden AG, Fang Y, Van Meurs JB, Pols HA, Van Leeuwen JP. Genetics and biology of vitamin D receptor polymorphisms. *Gene.* 2004; 338:143–56. [PubMed: 15315818]
  40. Chia VM, Newcomb PA. Calcium and colorectal cancer: Some questions remain. *Nutr Rev.* 2004; 62:115–20. [PubMed: 15098858]
  41. Wallace K, Baron JA, Cole BF, Sandler RS, Karagas MR, et al. Effect of calcium supplementation on the risk of large bowel polyps. *J Natl Cancer Inst.* 2004; 96:921–5. [PubMed: 15199111]
  42. Baron JA, Beach M, Mandel JS, van Stolk RU, Haile RW, et al. Calcium supplements for the prevention of colorectal adenomas. Calcium polyp prevention study group. *N Engl J Med.* 1999; 340:101–7. [PubMed: 9887161]
  43. Shrubsole MJ, Wu H, Ness RM, Shyr Y, Smalley WE, et al. Alcohol drinking, cigarette smoking, and risk of colorectal adenomatous and hyperplastic polyps. *Am J Epidemiol.* 2008; 167:1050–8. [PubMed: 18304959]
  44. Wallace K, Grau MV, Ahnen D, Snover DC, Robertson DJ, et al. The association of lifestyle and dietary factors with the risk for serrated polyps of the colorectum. *Cancer Epidemiol Biomarkers Prev.* 2009; 18:2310–7. [PubMed: 19661090]
  45. Potter JD, Bigler J, Fosdick L, Bostick RM, Kampman E, et al. Colorectal adenomatous and hyperplastic polyps: Smoking and n-acetyltransferase 2 polymorphisms. *Cancer Epidemiol Biomarkers Prev.* 1999; 8:69–75. [PubMed: 9950242]
  46. Ji BT, Weissfeld JL, Chow WH, Huang WY, Schoen RE, et al. Tobacco smoking and colorectal hyperplastic and adenomatous polyps. *Cancer Epidemiol Biomarkers Prev.* 2006; 15:897–901. [PubMed: 16702367]
  47. Palmer HG, Gonzalez-Sancho JM, Espada J, Berciano MT, Puig I, et al. Vitamin D(3) promotes the differentiation of colon carcinoma cells by the induction of E-cadherin and the inhibition of beta-catenin signaling. *J Cell Biol.* 2001; 154:369–87. [PubMed: 11470825]
  48. Deeb KK, Trump DL, Johnson CS. Vitamin d signalling pathways in cancer: Potential for anticancer therapeutics. *Nat Rev Cancer.* 2007; 7:684–700. [PubMed: 17721433]
  49. Byers S, Shah S. Vitamin d and the regulation of wnt/beta-catenin signaling and innate immunity in colorectal cancer. *Nutr Rev.* 2007; 65:S118–20. [PubMed: 17867386]
  50. Egan JB, Thompson PA, Vitanov MV, Bartik L, Jacobs ET, et al. Vitamin D receptor ligands, adenomatous polyposis coli, and the vitamin D receptor foki polymorphism collectively modulate beta-catenin activity in colon cancer cells. *Mol Carcinog.* 2009

Table 1

Mean values (standard deviation) of plasma 25-hydroxyvitamin-D (25(OH)D) concentration according to selected characteristics of study participants.

	Controls			Adenomas			Hyperplastic Polyps		
	N	%	Mean 25(OH)D (ng/mL)	SD	N	(%)	Mean 25(OH)D (ng/mL)	SD	SD
All participants	225		24.9	10.0	149		23.1	8.9	85
Sex									
Male	92	41%	25.0	9.1	72	48%	22.5	8.4	41
Female	133	59%	24.8	10.7	77	52%	23.6	9.3	44
Age(y)									
<50	49	22%	25.7	13.0	18	12%	25.5	8.7	12
50-64	106	47%	24.6	8.5	64	43%	22.4	8.7	41
64-79	70	31%	24.8	10.0	67	45%	23.0	9.1	32
Current Supplement Use *									
No	74	33%	21.3	11.5	55	37%	18.6	7.0	20
Yes	151	67%	26.7	8.7	94	63%	25.7	8.8	65
Physical Activity (MET-hrs/wk)									
<3.5	56	25%	19.6	8.5	46	31%	20.3	8.4	16
3.5-11.4	56	25%	24.5	8.1	39	26%	23.3	8.0	31
11.5-23.5	56	25%	26.7	11.2	25	17%	26.3	9.4	19
>23.5	57	25%	28.7	9.9	39	26%	24.0	9.2	19
Previous Polyp Diagnosis									
No	180	80%	25.2	10.2	63	42%	22.6	9.1	52
Yes	45	20%	23.7	9.3	86	58%	23.4	8.7	33
Month of Blood Draw									
November-May	108	48%	21.7	9.7	79	53%	20.5	7.8	51

23.6 10.6

	Controls			Adenomas			Hyperplastic Polyps		
	N	%	Mean 25(OH)D (ng/mL)	SD	N	(%)	Mean 25(OH)D (ng/mL)	SD	SD
June–October	117	52%	27.9	9.4	70	47%	26.0	9.1	25.0 10.0
Body Mass Index									
<25.0	76	34%	28.8	10.6	46	31%	25.0	6.8	26.0 11.2
25–29.9	95	42%	24.2	9.3	60	40%	23.7	9.5	26.2 10.0
30+	54	24%	20.8	8.6	43	29%	20.1	9.2	20.1 8.9
Cigarette Smoking History									
Never Smoker	97	43%	24.0	9.4	64	43%	24.3	8.6	26.4 11.2
≤20 pack years	92	41%	26.8	10.7	48	32%	23.6	8.4	23.8 10.1
>20 pack years	36	16%	22.4	9.5	37	25%	20.3	9.6	22.4 9.7
Ethnicity/Race									
Non-Hispanic White	197	88%	25.1	10.1	132	89%	23.5	8.8	24.9 10.5
Other	28	12%	23.8	9.5	17	11%	20.0	8.6	17.7 6.9

* Use of multivitamins, calcium, or vitamin D supplements at least once per week.

Table 2

Odds ratios and 95% CI for the relation of plasma 25(OH)D concentration and risk of colorectal adenomas or hyperplastic polyps.

Controls			Adenomas			Hyperplastic Polyps		
All Participants	N	OR*	(95% CI)	OR**	(95% CI)	N	OR*	(95% CI)
Total 25(OH)D (ng/mL)								
≤20.5	74	58	Ref	Ref		29	Ref	
20.6–28.9	76	54	0.90 (0.52–1.55)	0.97 (0.55–1.71)		27	1.04 (0.53–2.01)	0.88 (0.44–1.77)
>28.9	75	37	0.66 (0.36–1.19)	0.71 (0.38–1.30)		29	1.39 (0.69–2.81)	1.17 (0.55–2.51)
per 10 ng/mL	225	149	0.88 (0.69–1.12)	0.91 (0.70–1.18)		85	1.10 (0.81–1.49)	1.01 (0.71–1.42)
No Previous Polyps								
All Participants	N	OR*	(95% CI)	OR**	(95% CI)	N	OR*	(95% CI)
Total 25(OH)D (ng/mL)								
≤20.5	60	28	Ref	Ref		17	Ref	
20.6–28.9	59	21	0.81 (0.40–1.65)	0.85 (0.41–1.78)		17	1.35 (0.59–3.08)	1.25 (0.53–2.94)
>28.9	61	14	0.50 (0.22–1.10) ^a	0.52 (0.23–1.20) ^b		18	1.57 (0.65–3.76) ^a	1.42 (0.55–3.65) ^b
per 10 ng/mL	180	63	0.77 (0.56–1.07) ^c	0.80 (0.57–1.13) ^d		52	1.13 (0.78–1.63) ^c	1.06 (0.67–1.60) ^d

* Adjusted for age, sex, previous polyp diagnosis (as appropriate), season of blood draw, BMI, history of cigarette use (Never smoker, ≤20 pack years, >20 pack years).

** Additional adjustment for supplement use, and physical activity level (linear by quartile)

Wald test of the null hypothesis that these coefficients are equal:

^a p=0.03;

^b p=0.08;

^c p=0.08;

^d p=0.26