AP&T Alimentary Pharmacology and Therapeutics

# Higher 25-hydroxyvitamin D levels are associated with greater odds of remission with anti-tumour necrosis factor-α medications among patients with inflammatory bowel diseases

R. W. Winter, E. Collins, B. Cao, M. Carrellas, A. M. Crowell & J. R. Korzenik

Division of Gastroenterology and Hepatology, Brigham and Women's Hospital, Boston, MA, USA.

**Correspondence to:**
Dr R. W. Winter, Division of Gastroenterology and Hepatology, Brigham and Women's Hospital, 75 Francis Street, Boston, MA 02115, USA.
E-mail: rwinter1@partners.org

**Publication data**
Submitted 22 February 2016
First decision 26 March 2016
Resubmitted 28 September 2016
Resubmitted 16 December 2016
Resubmitted 18 December 2016
Accepted 19 December 2016

*The Handling Editor for this article was Professor Jonathan Rhodes, and it was accepted for publication after full peer-review.*

## SUMMARY

### Background
Vitamin D has been linked to disease activity among patients with inflammatory bowel diseases (IBD). Prior investigation has also suggested that vitamin D levels may affect duration of therapy with anti-tumour necrosis factor-α (anti-TNF-α) medications among patients with IBD.

### Aim
To evaluate the relationship between vitamin D levels and odds of reaching remission while on an anti-TNF-α medication.

### Methods
A total of 521 IBD patients enrolled in the Brigham and Women's IBD Centre database were eligible for inclusion. Patients treated with anti-TNF-α therapy who had vitamin D levels drawn within 6 months prior or 2 weeks after initiation of anti-TNF-α medication and who had reported remission status at 3 months were included. A logistic regression model adjusting for age, gender, IBD diagnosis, anti-TNF-α medication (infliximab vs. adalimumab) and first or subsequent anti-TNF-α medication was used to identify the effect of vitamin D level on initial response to anti-TNF-α therapy.

### Results
A total of 173 patients were included in the final analysis. On logistic regression, patients with normal vitamin D levels $n = 122$ at the time of anti-TNF-α medication initiation had a 2.64 increased odds of remission at 3 months compared to patients with low vitamin D levels $n = 51$ when controlling for age, gender, diagnosis, type of anti-TNF-α medication and first or subsequent anti-TNF-α medication (OR = 2.64, 95% CI = 1.31–5.32, $P = 0.0067$).

### Conclusions
These findings suggest that vitamin D levels may influence initial response to anti-TNF-α medication and that low vitamin D levels may pre-dispose patients to decreased odds of remission.




doi:10.1111/apt.13936

## INTRODUCTION

Vitamin D has been suggested to play a role in the pathogenesis of inflammatory bowel diseases (IBD) and other immune-mediated diseases.[1] A high prevalence of vitamin D deficiency has been reported among patients with inflammatory bowel diseases, including Crohn's disease and ulcerative colitis (UC).[2] Nearly 50% of patients with IBD had a vitamin D deficiency in one study, with 11% considered to have a severe deficiency.[3] Similarly, the Nurses' Health Study showed higher 25-OH vitamin D levels were associated with a lower risk of a subsequent development of IBD.[4] Among individuals with newly diagnosed IBD, 78% had insufficient levels, making it difficult to determine if vitamin D deficiency is a result or contributing cause to the development or exacerbation of disease.[5] Because vitamin D is absorbed in the small intestine, inflammation of the bowel might account for lower vitamin D values. Other factors that may increase the risk for vitamin D deficiency specifically among patients with IBD include decreased oral intake due to disease, increased excretion, increased catabolism or decreased exposure to sunlight.[2, 6, 7] Vitamin D may also support innate immune function.[1]

It has been suggested that vitamin D may have a role in influencing the clinical course of Crohn's disease and UC. While there are insufficient data regarding treatment of IBD with vitamin D, studies have shown that patients who are vitamin D deficient are also likely to have complications of IBD including increased risk of hospitalisation, stricturing disease, surgical resection, pancolitis in UC and use of steroids earlier after diagnosis.[8, 9] In addition, higher vitamin D levels have been associated with a higher quality of life among patients with IBD, as measured by the short IBD questionnaire.[10] Compared to placebo, treatment with vitamin D was associated with decreased risk of relapse.[11]

How vitamin D levels may impact response to other therapies has been minimally explored. It has been previously shown that vitamin D deficient patients with IBD on anti-TNF-α therapy had earlier cessation of anti-TNF-α therapy compared to those with normal vitamin D levels.[12] We performed a retrospective study to determine whether vitamin D levels were associated with achieving remission among patients with IBD who received anti-TNF-α therapy.

## METHODS

### Data source and study population

Patients were identified through the Brigham and Women's Hospital Crohn's and Colitis Centre database, an electronic database including patient data from the years 1998–2015. Vitamin D levels from all dates were requested for all patients entered in the database. Vitamin D levels included results not only from the laboratories at Brigham and Women's and its partner institutions, but also from outside laboratories.

There were 521 patients in the database and 384 patients enrolled in the Brigham and Women's Inflammatory Bowel Disease database who received anti-TNF-α therapy. All of the patients in the database were at least 18 years old. All patients with a diagnosis of IBD (including Crohn's disease, ulcerative colitis and indeterminate colitis) who were enrolled in the patient database at the Brigham and Women's IBD Centre were initially eligible for inclusion. Two hundred and three patients that were treated with anti-TNF-α therapy who also had vitamin D levels drawn within 6 months prior or 2 weeks after initiation of anti-TNF-α medication were included in this single-centre, retrospective cohort study. Patients starting on anti-TNF-α medication for the first time and also those starting a second or third anti-TNF-α medication were included. The main predictor was vitamin D level, defined as low (below the normal range for the vitamin D lab assay) or normal. This approach using dichotomised levels of vitamin D was utilised because a number of different vitamin D assays with varying scales were included in the database. Due to the limitations of outside lab assays, we were unable to classify vitamin D levels as normal, insufficient and deficient.

### Variables and outcomes

For all study patients, age, gender, subtype of IBD diagnosis, vitamin D level (as low or normal), type of anti-TNF-α medication, first, second or third anti-TNF medication, treatment with combination or monotherapy, C-reactive protein (CRP), smoking status and Montreal classification were recorded. All patients included in this study were prescribed either infliximab or adalimumab. Combination therapy was defined as anti-TNF-α therapy and an immunomodulator. IBD diagnosis was classified as Crohn's disease or ulcerative colitis. There were only five patients who were prescribed a third anti-TNF, and thus, patients were noted to have prior exposure, defined by starting a second or third anti-TNF medication, or no prior exposure if the anti-TNF was the first medication in this class ever prescribed. C-reactive protein values were dichotomised to reflect flare, defined by elevated CRP, or no flare, defined by CRP within the normal range. Because vitamin D

results included levels from laboratory assays with different reference ranges, vitamin D level was classified as low or normal based on the reference lab's normal range. The lower cut-off for normal vitamin D levels ranged from 9 to 33 ng/mL depending on the assay. Dates of plasma 25(OH) vitamin D levels and dates of initiation of anti-TNF-α therapy were recorded.

Our primary outcome was remission at 3 months after initiation of anti-TNF-α therapy. Remission was assessed by categorisation of the clinical evaluation recorded by the treating physician in the medical record and was based on the physician's assessment of the patient's status using a patient's reported symptoms and review of the medical chart. Response at 3 months was recorded as remission, incomplete response, no response, allergic reaction or unknown. All patients with incomplete response or no response were considered not to be in remission.

### Statistical analysis

All analyses were performed using SAS 9.4 (SAS Institute, Cary, NC, USA).

All variables were individually compared to the outcome of remission using chi-square or Fisher's exact test. Regardless of significance, the variables vitamin D status (defined as low vs. normal), age, gender, IBD diagnosis and anti-TNF-α medication were included in the regression model. Any variable that was significant on univariate analysis was also included in the final regression model. A logistic regression model adjusting for age, gender, IBD diagnosis, anti-TNF-α medication (infliximab vs. adalimumab) and prior anti-TNF exposure was used to identify the effect of vitamin D level on initial response to anti-TNF-α therapy. A $P$ <0.05 in the final regression model was considered statistically significant.

## RESULTS

We reviewed vitamin D levels of 384 patients who were treated with anti-TNF-α therapy in our electronic medical record. Patients were excluded if they had an adverse reaction or unknown response to anti-TNF-α medication, or if they had vitamin D levels drawn outside the window of 6 months prior or 2 weeks after initial dose of anti-TNF-α medication. Two hundred and three patients met these criteria. Sixty-three patients (31%) had a low vitamin D level and 140 patients (69%) had normal vitamin D levels. Among the patients with low vitamin D, 42 (67%) had Crohn's disease and among patients with normal vitamin D, 95 (68%) had Crohn's Disease. There were no statistically significant differences between the groups who had low vs. normal vitamin D levels (Table 1).

For 30 patients, remission status was unknown and therefore 173 patients were included in the final univariable and multivariable analyses. In the univariable analyses, we did not observe any significant associations between age, gender, subclass of IBD, type of anti-TNF-α medication, administration of combo- or monotherapy, smoking status, elevated CRP or Montreal classification and the odds of remission; however, there was a significant association between vitamin D status and remission and between prior anti-TNF exposure and remission (Table 2).

On logistic regression, we observed a significant association with remission after 3 months of anti-TNF-α therapy and normal vitamin D levels (OR = 2.64, 95% CI = 1.31–5.32, $P$ = 0.0067) (Table 3). Age, gender, diagnosis, type of anti-TNF-α medication and prior anti-TNF exposure were not independently associated with remission status after 3 months of anti-TNF-α therapy.

## DISCUSSION

In this single-centre, retrospective study, we found a significant association between vitamin D status and remission of IBD with anti-TNF-α medication.

These results show a significant association between vitamin D levels and rates of remission among patients with IBD. Specifically, patients who had low vitamin D levels prior to treatment had decreased odds of being in remission after 3 months of treatment with anti-TNF-α therapy. This observation may be supported by prior evidence suggesting associations between vitamin D levels and disease severity as well as complications of IBD. In addition, murine studies have shown that vitamin D is associated with decreased severity of colitis,[13] and also that oral intake of vitamin D improves colitis.[14] In humans, a randomised control trial showed decreased risk of relapse with treatment with vitamin D compared to placebo. While results did not reach significance, Jorgensen *et al.* showed that patients with Crohn's disease who were in remission were less likely to experience clinical relapse at 1 year if they took vitamin D.[11] Ananthakrishnan *et al.* showed that normalising vitamin D levels in patients with Crohn's disease who previously were vitamin D deficient results in decreased risk of IBD-related surgery.[8] Similar studies have shown decreases in peripheral inflammatory markers and Crohn's disease activity index among patients taking vitamin D supplementation.[15]

**Table 1 |** Study population characteristics by vitamin D status

| Characteristic | Low vitamin D ($n$ = 63) | Normal vitamin D ($n$ = 140) |
|---|---|---|
| Mean age* | 37.39 (12.31 s.d.) | 38.72 (14.71 s.d.) |
| Gender | | |
| Female | 42 (66.67) | 98 (70.00) |
| Male | 21 (33.33) | 42 (30.00) |
| Diagnosis | | |
| Crohn's disease | 42 (66.67) | 95 (67.86) |
| Ulcerative Colitis | 21 (33.33) | 45 (32.14) |
| Medication | | |
| Infliximab | 41 (65.08) | 75 (53.57) |
| Adalimumab | 22 (34.92) | 65 (46.43) |
| Combination therapy | | |
| Monotherapy | 60 (95.24) | 130 (92.86) |
| Combination | 3 (4.76) | 10 (7.14) |
| Prior anti-TNF exposure | | |
| None (first anti-TNF) | 48 (76.19) | 110 (78.57) |
| Prior exposure (second or third anti-TNF) | 15 (23.81) | 30 (21.43) |
| C reactive protein | | |
| Normal | 16 (25.40) | 40 (28.57) |
| Elevated | 40 (63.49) | 80 (57.14) |
| Unknown | 7 (11.11) | 20 (14.29) |
| Smoking status | | |
| Never smoker | 42 (66.67) | 98 (70.00) |
| Current smoker | 4 (6.35) | 13 (9.29) |
| Former smoker | 17 (26.98) | 29 (20.71) |
| Montreal classification: behaviour of Crohn's disease | | |
| Nonstricturing, nonpenetrating | 21 (52.50) | 30 (31.58) |
| Stricturing | 9 (22.50) | 36 (37.89) |
| Penetrating | 10 (25.00) | 27 (28.42) |
| Unknown | 0 (0.00) | 2 (2.11) |
| Montreal classification: location of Crohn's disease | | |
| Ileal | 10 (24.39) | 20 (21.05) |
| Colonic | 8 (19.51) | 18 (18.95) |
| Ileal and colonic | 23 (56.10) | 55 (57.89) |
| Isolated upper disease | 0 (0.00) | 1 (1.05) |
| Unknown | 0 (0.00) | 1 (1.05) |
| Montreal classification: behaviour of perianal Crohn's disease | | |
| No perianal disease | 26 (63.41) | 54 (58.06) |
| Perianal disease | 15 (36.59) | 38 (40.86) |
| Unknown | 0 (0.00) | 1 (1.08) |
| Montreal classification: extent of UC | | |
| Ulcerative proctitis† | 1 (4.76) | 1 (2.38) |
| Left-sided colitis‡ | 4 (19.05) | 15 (35.71) |
| Extensive colitis§ | 16 (76.19) | 26 (61.90) |

All values are presented as number (percentage) unless indicated otherwise.

This table compares the baseline characteristics of the study population among those with normal and low vitamin D levels. There were no differences in gender, diagnosis, medication, combination or monotherapy, anti-TNF exposure, CRP level, smoking status or Montreal classification between the two subsets of the population.

* Age in years (s.d.).

† Limited to rectum.

‡ Distal to splenic flexure.

§ Disease proximal to splenic flexure.

**Table 2 |** Univariate analysis of association between variables and odds of remission on anti-TNF-α medication

| | Remission ($n$ = 97) | No remission ($n$ = 76) | $P$ value |
|---|---|---|---|
| Age* | 39.99 (14.06) | 36.61 (13.97) | 0.1181 |
| Gender | | | |
| Female | 65 (67.01) | 55 (68.42) | 0.8713 |
| Male | 32 (32.99) | 24 (31.58) | |
| Diagnosis | | | |
| Crohn's disease | 62 (63.92) | 54 (71.05) | 0.3340 |
| Ulcerative colitis | 35 (36.08) | 22 (28.95) | |
| Medication | | | |
| Infliximab | 57 (58.76) | 40 (52.63) | 0.4436 |
| Adalimumab | 40 (41.24) | 36 (47.37) | |
| Combination therapy | | | |
| No combination therapy | 90 (92.78) | 71 (93.42) | 1.0000 |
| Combination therapy | 7 (7.22) | 5 (6.58) | |
| Prior anti-TNF exposure | | | |
| None (first anti-TNF) | 82 (84.54) | 52 (68.42) | **0.0118** |
| Prior exposure (second or third anti-TNF) | 15 (15.46) | 24 (31.58) | |
| C reactive protein | | | |
| Normal | 28 (28.87) | 20 (26.32) | 0.9239 |
| Elevated | 56 (57.73) | 46 (60.53) | |
| Unknown | 13 (13.40) | 10 (13.16) | |
| Smoking status | | | |
| Never smoker | 65 (67.01) | 54 (71.05) | 0.7333 |
| Current smoker | 8 (8.25) | 7 (9.21) | |
| Former smoker | 24 (24.74) | 15 (19.74) | |
| Vitamin D status | | | |
| Normal vitamin D | 77 (79.38) | 45 (59.21) | **0.0045** |
| Low vitamin D | 20 (20.62) | 31 (40.79) | |
| Montreal classification: behaviour of Crohn's disease | | | |
| Nonstricturing, Nonpenetrating | 23 (37.10) | 20 (38.46) | 0.5069 |
| Stricturing | 22 (35.48) | 16 (30.77) | |
| Penetrating | 15 (24.19) | 16 (30.77) | |
| Unknown | 2 (3.23) | 0 (0.00) | |
| Montreal classification: location of Crohn's disease | | | |
| Ileal | 16 (25.81) | 10 (18.87) | 0.5755 |
| Colonic | 12 (19.35) | 10 (18.87) | |
| Ileal and colonic | 33 (53.23) | 32 (60.38) | |
| Isolated upper disease | 0 (0.00) | 1 (1.89) | |
| Unknown | 1 (1.61) | 0 (0.00) | |
| Montreal classification: behaviour of perianal Crohn's disease | | | |
| No perianal disease | 35 (58.33) | 29 (54.72) | 0.5674 |
| Perianal disease | 24 (40.00) | 24 (45.28) | |
| Unknown | 1 (1.67) | 0 (0.00) | |
| Montreal classification: extent of UC | | | |
| Ulcerative proctitis† | 2 (6.06) | 0 (0.00) | 0.5147 |
| Left-sided colitis‡ | 10 (30.30) | 7 (33.33) | |
| Extensive colitis§ | 21 (63.64) | 14 (66.67) | |

All values are presented as number (percentage) unless indicated otherwise.

This table displays results of univariate analyses, showing association between vitamin D status, gender, diagnosis, medication, combination or monotherapy, anti-TNF exposure, CRP level, smoking status and Montreal classification with odds of remission.

Bold values are statistically significant.

* Age in years (s.d.).

† Limited to rectum.

‡ Distal to splenic flexure.

§ Disease proximal to splenic flexure.

**Table 3 |** Odds of remission at 3 months

| Variable | OR (95% CI) | *P* value |
|---|---|---|
| Normal vitamin D | 2.64 (1.31–5.32) | **0.0067** |
| Age | 1.01 (0.99–1.04) | 0.3366 |
| Male | 1.20 (0.61–2.38) | 0.5983 |
| Diagnosis of Crohn's | 0.78 (0.39–1.54) | 0.4697 |
| Medication (Adalimumab) | 0.91 (0.43–1.93) | 0.8061 |
| First anti-TNF | 2.22 (0.93–5.32) | 0.0729 |

This table displays results of a logistic regression model showing the association between vitamin D status and odds of remission while controlling for age, gender, diagnosis, type of anti-TNF medication prescribed and exposure to prior anti-TNF therapy.

Vitamin D can be acquired either by endogenous production or through consumption of foods containing vitamin D. Vitamin D is produced when the skin comes in contact with ultraviolet light, and can also be found in egg yolks, as well as certain seafood and dairy products.[6] Some foods such as orange juice may be supplemented with Vitamin D which is absorbed in the small intestine.[9] The vitamin D that is produced or absorbed is metabolised in the liver to produce 25-hydroxyvitamin D (25-OH vitamin D), which is activated by the kidney to form 1,25 dihydroxyvitamin D. The primary circulating form of vitamin D is 25-OH vitamin D, which is often measured clinically to determine vitamin D sufficiency.[2]

The pathophysiology of how vitamin D may affect clinical response is not entirely clear. Prior studies have suggested that vitamin D is associated with innate immune function.[1] Vitamin D has been reported to affect multiple cells involved in the immune process, including monocytes, dendritic cells, B and T cells.[16] Vitamin D inhibits B and T cell proliferation, affects T cell maturation and is involved in the induction of T regulatory cells, which may result in increased production of anti-inflammatory cytokines and decreased production of inflammatory cytokines.[16, 17] Whether vitamin D deficiency is a marker of severity of disease, which itself may affect rates of remission, or whether vitamin D interacts with the immune system and therefore affects severity of inflammation and remission rates is unknown.

The mechanism by which vitamin D is thought to affect disease severity is via the TNF pathway. Froicu *et al.* showed that $1,25(OH)_2D_3$ down-regulates many genes associated with TNF-α.[14] This finding suggests that vitamin D and anti-TNF-α medications may work together to decrease TNF-α, supporting a potential role of vitamin D in treatment of IBD. The pathophysiology regarding how vitamin D deficiency affects anti-TNF-α response requires further clarification. Vitamin D has also been showed to induce expression of the nucleotide-binding oligomerisation domain (NOD2) gene, a susceptibility gene associated with Crohn's disease, and has been suggested to be a potential immune modulator in vitro.[18, 19] However, further studies clarifying the interaction between vitamin D, NOD2 and disease course are warranted.

These findings may have clinical implications as we have shown that vitamin D levels are associated with initial response to anti-TNF-α therapy. We are aware of one prior study that showed an association between vitamin D levels and duration of anti-TNF-α therapy,[12] but larger studies investigating remission rates among patients with low vitamin D levels are warranted. If our findings are confirmed in larger studies, recommendations for clinical management of IBD may include therapy with vitamin D. Further study regarding optimal timing and dosing of vitamin D supplementation should be clarified in future investigations. Furthermore, because of a variety of assays used in our patients, we dichotomised results as below normal or normal. We were not able to explore whether higher levels – even within normal – may improve outcomes.

This study has several limitations. First, this was a single-centre undersized study that was in part limited because it was conducted at a tertiary care centre. Larger multicentre studies would be preferable. Many patients were excluded because anti-TNF-α treatment was initiated prior to referral and exact start dates of therapy were unknown. A prospective trial monitoring vitamin D levels prior to initiating therapy would enable us to better study the association of vitamin D deficiency and response to anti-TNF-α therapy. Second, response to medication was determined at 3 months after initiation of therapy. Long-term maintenance of therapy is also of interest. Effect of vitamin D level on long-term response to anti-TNF-α therapy is of interest, but was not feasible with the available data. Third, our patient populations were dichotomised as low vitamin D or normal vitamin D. Because patients were referred from many medical practices, vitamin D levels were measured in laboratories with many different vitamin D assays. We were not able to categorise vitamin D levels as normal, insufficient or deficient, as assays from many laboratories were included, and they did not all provide classification of vitamin D levels. We recommend future studies using vitamin D as a continuous variable to further characterise the relationship between levels and response. In addition, only 203 of our patients had

vitamin D levels measured, and fewer had levels at the time of initiation of anti-TNF-α therapy. Some patients did not have vitamin D levels drawn at initiation of the anti-TNF agent because at the time they started the medication, routine assessment of vitamin D status was not standard practice. It is possible that physicians were more likely to check vitamin D levels in patients with more active disease. Last, we were not able to account for vitamin D supplementation, as many of our patients are advised to take over the counter vitamin D and this information was not available in the database. However, even with supplementation, improvement in vitamin D levels may not be seen for a number of months, and likely did not affect short-term outcomes.

In conclusion, these findings suggest that vitamin D levels are associated with initial response to anti-TNF-α medication and that low vitamin D levels may pre-dispose patients to decreased odds of remission, though no causal association has yet been shown. As anti-TNF-α medications are commonly prescribed to patients with IBD, further clarifying the role of vitamin D and its association with response to anti-TNF-α therapy is recommended and larger cohort trials are warranted.

## AUTHORSHIP

*Guarantor of the article*: Rachel Winter.

*Author contributions*: Rachel Winter was involved in the study design, data collection and analysis and writing and editing of the manuscript. Emily Collins, Bonnie Cao, Madeline Carrellas and Anne Marie Crowell were involved in data collection. Joshua Korzenik was involved in study design and editing of the manuscript.

All authors approved the final version of the manuscript.

## ACKNOWLEDGEMENTS

*Declaration of personal interests*: None.

*Declaration of funding interests*: Dr Winter has received support from the National Institutes of Health [grant number T32 DK007533-29]. Funding for this project was provided by Takeda Pharmaceuticals U.S.A., Inc.

## REFERENCES

1. Holick MF. Sunlight and vitamin D for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease 1–4. *Am J Clin Nutr* 2004; **80**: 1678S–88S.
2. Mouli V, Ananthakrishnan AN. Review article: vitamin D adn inflammatory bowel diseases. *Aliment Pharmacol Ther* 2014; **39**: 125–36.
3. Ulitsky A, Ananthakrishnan AN, Naik A, *et al.* Vitamin D deficiency in patients with inflammatory bowel disease: association with disease activity and quality of life. *JPEN J Parenter Enteral Nutr* 2011; **35**: 308–16.
4. Ananthakrishnan AN, Khalili H, Higuchi LM, *et al.* Higher predicted vitamin D status is associated with reduced risk of crohn's disease. *Gastroenterology* 2012; **142**: 482–9.
5. Leslie WD, Miller N, Rogala L, Bernstein CN. Vitamin D status and bone density in recently diagnosed inflammatory bowel disease: the Manitoba IBD Cohort Study. *Am J Gastroenterol* 2008; **103**: 1451–9.
6. Holick MF. Vitamin D deficiency. *N Engl J Med* 2007; **357**: 266–81.
7. Rosen CJ, Vitamin D. Insufficiency. *N Engl J Med* 2011; **364**: 248–54.
8. Ananthakrishnan AN, Cagan A, Gainer VS, *et al.* Normalization of plasma 25-hydroxy vitamin D is associated with reduced risk of surgery in Crohn's disease. *Inflamm Bowel Dis* 2013; **19**: 1921–7.
9. Reich KM, Fedorak RN, Madsen K, Kroeker KI. Vitamin D improves inflammatory bowel disease outcomes: basic science and clinical review. *World J Gastroenterol* 2014; **20**: 4934–47.
10. Hlavaty T, Krajcovicova A, Koller T, *et al.* Higher vitamin D serum concentration increases health related quality of life in patients with inflammatory bowel diseases. *World J Gastroenterol* 2014; **20**: 15787–96.
11. Jørgensen SP, Agnholt J, Glerup H, *et al.* Clinical trial: vitamin D3 treatment in Crohn's disease - a randomized double-blind placebo-controlled study. *Aliment Pharmacol Ther* 2010; **32**: 377–83.
12. Zator ZA, Cantu SM, Konijeti GG, *et al.* Pretreatment 25-hydroxyvitamin D levels and durability of anti-tumor necrosis factor-α therapy in inflammatory bowel diseases. *JPEN J Parenter Enteral Nutr* 2014; **38**: 385–91.
13. Zhu Y, Mahon B, Froicu M, Cantorna M. Calcium and 1 alpha,25-dihydroxyvitamin D3 target the TNF-alpha pathway to suppress experimental inflammatory bowel disease. *Eur J Immunol* 2005; **35**: 217–24.
14. Froicu M, Cantorna MT. Vitamin D and the vitamin D receptor are critical for control of the innate immune response to colonic injury. *BMC Immunol* 2007; **8**: 5.
15. Miheller P, Muzes G, Hritz I, *et al.* Comparison of the effects of 1,25 dihydroxyvitamin D and 25 hydroxyvitamin D on bone pathology and disease activity in Crohn's disease patients. *Inflamm Bowel Dis* 2009; **15**: 1656–62.
16. Aranow C. Vitamin D and the immune system. *J Investig Med* 2011; **59**: 881–6.
17. Calton EK, Keane KN, Newsholme P, Soares MJ. The impact of Vitamin D levels on inflammatory status: a systematic review of immune cell studies. *PLoS ONE* 2015; **10**: 1–12.
18. Wang TT, Dabbas B, Laperriere D, *et al.* Direct and indirect induction by 1,25-dihydroxyvitamin D3 of the NOD2/CARD15-defensin β2 innate immune pathway defective in crohn disease. *J Biol Chem* 2010; **285**: 2227–31.
19. Dionne S, Calderon MR, White JH, *et al.* Differential effect of vitamin D on NOD2- and TLR-induced cytokines in Crohn's disease. *Mucosal Immunol* 2014; **7**: 1405–15.