

Original Article

Effect of 1, 25 (OH)₂ Vitamin D₃ on Serum Levels of Inflammatory Cytokines in Patients Undergoing Coronary Artery Bypass Grafting

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Abstract

Background: A number of preventive or therapeutic strategies have been proposed to compensate for the inflammatory process during adult cardiac surgeries; none has been considered definitive. 1,25 (OH)₂ vitamin D₃ is a type of physiologic hormones with immunomodulatory effects. This study assessed the effect of oral vitamin D on serum levels of interleukin-1 (IL-1), IL-6 and tumor necrosis factor alpha (TNF- α) in adult coronary artery bypass grafting (CABG).

Materials and methods: In a double blind placebo controlled clinical trial, with a sample size of 80 patients, randomly assigned in either the placebo group (including 40 patients; placebo pills) or the intervention group (i.e. 40 patients; vitamin D pearls), CABG surgery with cardiopulmonary bypass was performed. Serum cytokines (IL-1, IL-6, and TNF- α) were evaluated preoperatively and during the first postoperative day.

Results: Serum TNF- α level was similar in the two groups; however, postoperative levels of IL-1 and IL-6 were significantly lower in vitamin D group.

Conclusion: The current study demonstrated that oral vitamin D supplements could augment immunomodulatory effects in adult patients undergoing cardiac surgical procedures (mainly CABG).

Keywords: Vitamin D, Cardiopulmonary Bypass, Cytokine, Inflammation

Please cite this article as: Fani K, Shadnoush M, Jahangirifard A, Foroughi M, Jelveh-Moghaddam HA, Dabbagh A. Effect of 1,25 (OH)₂ Vitamin D₃ on Serum Levels of Inflammatory Cytokines in Patients Undergoing Coronary Artery Bypass Grafting. J Cell Mol Anesth. 2018;3(3):98-102.

Introduction

Coronary artery diseases are among the most common health problems with a significant burden of disease among adults worldwide. Coronary artery bypass grafting (CABG) is among the most common non-medical treatment modalities all over the world.

However, CABG is not complication free. One of the most important untoward effects of CABG is the clinical presentations of the background inflammation during cardiopulmonary bypass, a mechanical substitute for heart and lung during the bypass period. A number of preventive or therapeutic strategies have been proposed to compensate for the inflammatory

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process; none have been considered definitive (1-4).

1,25 (OH)₂ Vitamin D₃ is a type of physiologic hormones. However, a number of other effects of vitamin D have been quoted including its immunomodulatory effects (5-7). The main anti-inflammatory effect of vitamin D is exerted through increasing tolerogenicity properties of dendritic cells; which in turn, will affect helper T cells, conducting them towards anti-inflammatory effects (8, 9). Though the effects of T cells on pro-inflammatory and anti-inflammatory cytokines has been studied extensively, the role of prophylactic vitamin D on these cytokines in patients undergoing CABG is not studied yet (5-7, 10-12).

In this research, the effect of oral vitamin D supplements in adult patients undergoing CABG, on serum levels of interleukin-1 (IL-1), IL-6 and tumor necrosis factor alpha (TNF- α) is compared with placebo.

Methods

This study was a double blind placebo controlled clinical trial; the aim of study was to determine the effect of 1,25 (OH)₂ vitamin D₃ administered before operation in elective CABG on postoperative serum inflammatory cytokines. The study was registered in the Iranian Clinical Trial Registry with this code: IRCT201502022804N7. This study started from March 2015 and ended after 24 months.

The patients entered the study after taking informed written consent and matching for inclusion and exclusion criteria. Sample size was determined after a power analysis (power = 0.8, β = 0.2, α = 0.05) using sample size software: PASS 2005; NCSS, LLC; UT, USA. A final sample size of 80 patients were calculated as the study sample size; which were randomly assigned in either the placebo group (including 40 patients who received placebo pills as mentioned later) or the intervention group (i.e. 40 patients; who received vitamin D as mentioned later). In the intervention group, each patient received 1 vitamin D pearl each day (each pearl being 50000 units); these pearls started 3 days before surgery. The pearls were made by Dana Pharma Co, Tabriz, Iran. All patients were anesthetized, operated on and handled in based on the institutional clinical protocols described

earlier in other studies (2,13-15).

The following items were considered as inclusion criteria: patients 40-75 years of age, elective CABG surgery with cardiopulmonary bypass (CPB), constant surgeon, no history of diabetes mellitus or Chronic hepatic failure (impaired Liver Function Test) or chronic kidney disease causing serum Creatinine >2mg/dL before anesthesia, no history of pulmonary hypertension (Pulmonary artery pressure below 30 mmHg).

Exclusion criteria: new onset of underlying diseases especially renal or central nervous system (CNS) injury, modification in the course of surgery (changing from on pump to off pump CABG), altered level (elevated or decreased) of Vit D₃ (out of normal range) in perioperative period, any complications during surgery, pulmonary artery pressure increased >30 mmHg, ejection fraction of left ventricle decreased <30% in any time perioperatively, repeated operation during early postoperative period, any unwanted complication in the operation period, re-starting CPB after weaning from bypass, pulmonary artery pressure >30 during perioperative period, re-operation in the postoperative period.

Serum cytokines (IL-1, IL-6, and TNF- α) were evaluated preoperatively as baseline and also during the 1st day after the operation; all measurements were carried out by ELISA kits. For assessment of blood levels of cytokines and vitamin D, preoperative samples were taken 5 days before surgery; postoperative blood samples were taken 6 hours after intensive care unit (ICU) entry. Blood samples were centrifuged at 2500g for 10 minutes at 4°C; afterwards, samples were frozen to -18°C and then, transferred to -70°C immunology laboratory. At the end of sampling, they were thawed and used for ELISA assessments of IL-1, IL-6 and TNF- α (2, 4, 7, 15).

Results

The results of this study demonstrated no difference between the two groups regarding the age, sex, time of surgical operation, time of aortic clamp and time of cardiopulmonary bypass, presented in detail in Table 1. In addition, preoperative levels of vitamin D before starting supplemental vitamin D administration was not statistically different between the two groups.

The results of cytokine assays by ELISA are demonstrated in Table 2. There was no difference between the groups regarding serum TNF- α levels; including both preoperative and postoperative levels. However, postoperative serum levels of IL-1 and IL-6 were significantly lower in vitamin D group; meanwhile, preoperative serum levels of IL-1 and IL-6 had no statistically significant difference between the two groups.

effect, the results demonstrated improved cytokine profile in patients receiving preoperative oral vitamin D supplements; with decreased serum levels of proinflammatory cytokines and increased serum levels of anti-inflammatory cytokines.

These results are in favor of anti-inflammatory effects of vitamin D; however, there are not any considerable previous studies besides our study, which have assessed the effects of oral vitamin D

Table 1: Basic variables in the two study groups

Vitamin D group	Placebo group	P value
Gender		
Male		
22	19	P value for Chi Square>0.05
Female		
18	21	
Weight (Kg)		
87 \pm 9	90 \pm 8	P value>0.05
Pre-operative Left Ventricular Ejection Fraction in Percent		
41 \pm 8	42 \pm 10	P value>0.05
Age in years (Mean \pm SD)		
53 \pm 8	55 \pm 9	P value>0.05
Aortic Clamp Time in minutes (Mean \pm SD)		
59 \pm 9	61 \pm 11	P value>0.05
Cardiopulmonary Bypass Time (CPB time) in minutes (Mean \pm SD)		
104 \pm 11	99 \pm 14	P value>0.05

Table 2: Postoperative Cytokine Levels in the two Study Groups

Vitamin D group	Placebo group	P value
TNF- α (pg/mL)		
12.2 \pm 2.8	11.4 \pm 3.1	>0.05 (non-Significant)
IL 1 (pg/mL)		
13.2 \pm 3.2	23.2 \pm 5.7	0.001
IL6 (pg/mL)		
43.1 \pm 11.2	87.4 \pm 41.1	0.02

Discussion

The current study showed that oral supplements of vitamin D in adult patients undergoing CABG leads to lower serum levels of IL-1 and IL-6; however, this study failed to demonstrate the effects of oral vitamin D supplements on serum levels of TNF- α .

The results of our study demonstrated the role of preoperative oral vitamin D supplements on the serum cytokine levels of patients undergoing CABG with cardiopulmonary bypass. In this study, with this presumption that vitamin D has an immunomodulatory

supplements on serum cytokine levels of adult cardiac surgical patients. On the other hand, there are a large number of studies, which have assessed the role of vitamin D in critical care setting and in patients with clinical major problems. Moraes *et al.*, demonstrated that low vitamin D levels on admission play an important role as an independent risk factor predicting mortality in critical care setting (16). In addition, de Haan *et al.*, reviewed, in a systematic review, 14 observational studies including 9,715 critically ill patients; they concluded that vitamin D deficiency could lead to increased susceptibility for severe

infections and mortality in those patients with critical clinical conditions and receiving critical care (17). In another systematic review and meta-analysis, Upala *et al.*, found that Vitamin D deficiency is significantly associated with an increased susceptibility to sepsis (18). Though these studies have assessed the role of vitamin D in major outcomes of an important group of critical care patients, none of them has assessed serum levels of cytokines in such patients.

There are great number of studies assessing the effects of vitamin D on immune system and cardiovascular system (5-7). However, the effect of preoperative vitamin D pearls on patients undergoing CABG with CPB has not been studied yet. There are 4 studies assessing the relationship between serum vitamin D levels and postoperative atrial fibrillation (11, 12, 19, 20). In a systematic review and meta-analysis, Weymann *et al.*, demonstrated the role of C reactive protein (CRP), IL-6, IL-8 and IL-10 in predicting postoperative occurrence of atrial fibrillation (21). On the other hand, Sheane *et al.*, assessed the relationship between MicroRNA-21 expression and vitamin D deficiency in coronary artery disease and demonstrated that there may be an association (10). However, Ruiz-Núñez *et al.*, demonstrated poor pre-operative intakes of fruits, vegetables, vitamin D, etc. in patients undergoing elective CABG; they also proposed the role of inflammatory cytokines as a probable related factor in such patients; however, they did not evaluate inflammatory cytokines (22).

The above studies and other similar ones have found a relationship between inflammatory cytokine levels and cardiac surgery; some have assessed a number of clinical outcomes too; however, none have assessed the effect of supplementary oral vitamin D on postoperative cytokine levels; which is a specific feature for this study.

There are however, a number of limitations in our study. Our blood samples were limited regarding the frequency of measurements; however, increasing the number of measurements and assessing the area under curve for each cytokine may have newer results. We assessed IL-1, IL-6 and TNF- α ; however, results that are more comprehensive could be gained if we had assessed more cytokines, especially in more time intervals.

Conclusion

The current study demonstrated that oral vitamin D supplements could augment immunomodulatory effects in adult patients undergoing cardiac surgical procedures (mainly CABG).

Acknowledgment

None.

Conflicts of Interest

The authors declare that they have no conflict of interest.

References

1. Rajaei S, Dabbagh A. Risk Factors for Postoperative Respiratory Mortality and Morbidity in Patients Undergoing Coronary Artery Bypass Grafting. *Anesth Pain Med.* 2012;2(2):60-5.
2. Foroughi M, Rahimian H, Dabbagh A, Majidi M, Hekmat M, Beheshti M, et al. Postoperative N-terminal pro-brain natriuretic peptide level in coronary artery bypass surgery with ventricular dysfunction after perioperative glucose-insulin-potassium treatment. *J Cardiothorac Vasc Anesth.* 2012;26(4):631-6.
3. Faritout ZS, Aghdaie N, Yazdani F, Azarfari R, Dabbagh A. Perioperative risk factors for prolonged mechanical ventilation and tracheostomy in women undergoing coronary artery bypass graft with cardiopulmonary bypass. *Saudi J Anaesth.* 2011;5(2):167-9.
4. Aryana P, Rajaei S, Bagheri A, Karimi F, Dabbagh A. Acute Effect of Intravenous Administration of Magnesium Sulfate on Serum Levels of Interleukin-6 and Tumor Necrosis Factor-alpha in Patients Undergoing Elective Coronary Bypass Graft With Cardiopulmonary Bypass. *Anesth Pain Med.* 2014;4(3):e16316.
5. Rajaei S, Akbari Sene A, Norouzi S, Berangi Y, Arabian S, Lak P, et al. The relationship between serum Vitamin D level and premenstrual syndrome in Iranian women. *International journal of reproductive biomedicine (Yazd, Iran).* 2016;14(10):665-8.
6. Rajaei S, Dabbagh A. The molecular mechanisms of Vitamin D effects on alleviating premenstrual syndrome pain. *J Cell Mol Anesth.* 2017;2(1):30-6.
7. Rajaei S, Mirahmadian M, Jeddi-Tehrani M, Tavakoli M, Zonoobi M, Dabbagh A, et al. Effect of 1,25(OH)₂ Vitamin D₃ on cytokine production by endometrial cells of women with repeated implantation failure. *Gynecol Endocrinol.* 2012;28(11):906-11.
8. Zhou Q, Qin S, Zhang J, Zhon L, Pen Z, Xing T. 1,25(OH)₂D₃ induces regulatory T cell differentiation by influencing the VDR/PLC-gamma1/TGF-beta1/pathway. *Molecular immunology.* 2017;91:156-64.
9. Xie Z, Chen J, Zheng C, Wu J, Cheng Y, Zhu S, et al. 1,25-

- dihydroxyVitamin D₃ -induced dendritic cells suppress experimental autoimmune encephalomyelitis by increasing proportions of the regulatory lymphocytes and reducing T helper type 1 and type 17 cells. *Immunology*. 2017;152(3):414-24.
10. Sheane BJ, Smyth P, Scott K, Aziz R, Buckley M, Lodge E, et al. An Association between MicroRNA-21 Expression and Vitamin D Deficiency in Coronary Artery Disease. *MicroRNA (Sharjah, United Arab Emirates)*. 2015;4(1):57-63.
11. Gode S, Aksu T, Demirel A, Sunbul M, Gul M, Bakir I, et al. Effect of Vitamin D deficiency on the development of postoperative atrial fibrillation in coronary artery bypass patients. *J Cardiovasc Thorac Res*. 2016;8(4):140-6.
12. Emren SV, Aldemir M, Ada F. Does Deficiency of Vitamin D Increase New Onset Atrial Fibrillation after Coronary Artery Bypass Grafting Surgery? *Heart Surg Forum*. 2016;19(4):E180-4.
13. Payani N, Foroughi M, Dabbagh A. The Effect of Intravenous Administration of Active Recombinant Factor VII on Postoperative Bleeding in Cardiac Valve Reoperations; A Randomized Clinical Trial. *Anesth Pain Med*. 2015;5(1):e22846.
14. Shahzamani M, Ghanavati A, Froutagheh AN, Foroughi M, Rahimian H, Shahsanaei A, et al. Carvedilol compared with metoprolol on left ventricular ejection fraction after coronary artery bypass graft. *J Perianesth Nurs*. 2011;26(6):384-7.
15. Dabbagh A, Bastanifar E, Foroughi M, Rajaei S, Keramatinia AA. The effect of intravenous magnesium sulfate on serum levels of N-terminal pro-brain natriuretic peptide (NT pro-BNP) in elective CABG with cardiopulmonary bypass. *J Anesth*. 2013;27(5):693-8.
16. Moraes RB, Friedman G, Wawrzyniak IC, Marques LS, Nagel FM, Lisboa TC, et al. Vitamin D deficiency is independently associated with mortality among critically ill patients. *Clinics (Sao Paulo, Brazil)*. 2015;70(5):326-32.
17. de Haan K, Groeneveld AB, de Geus HR, Egal M, Struijs A. Vitamin D deficiency as a risk factor for infection, sepsis and mortality in the critically ill: systematic review and meta-analysis. *Crit Care*. 2014;18(6):660.
18. Upala S, Sanguankeo A, Permpalung N. Significant association between Vitamin D deficiency and sepsis: a systematic review and meta-analysis. *BMC anesthesiology*. 2015;15:84.
19. Shadvar K, Ramezani F, Sanaie S, Maleki TE, Arbat BK, Nagipour B. Relationship between plasma level of Vitamin D and post operative atrial fibrillation in patients undergoing CABG. *Pakistan journal of medical sciences*. 2016;32(4):900-4.
20. Cerit L, Kemal H, Gulsen K, Ozcem B, Cerit Z, Duygu H. Relationship between Vitamin D and the development of atrial fibrillation after on-pump coronary artery bypass graft surgery. *Cardiovascular journal of Africa*. 2017;28(2):104-7.
21. Weymann A, Popov AF, Sabashnikov A, Ali-Hasan-Al-Saegh S, Ryazanov M, Tse G, et al. Baseline and postoperative levels of C-reactive protein and interleukins as inflammatory predictors of atrial fibrillation following cardiac surgery: a systematic review and meta-analysis. *Kardiologia polska*. 2018;76(2):440-51.
22. Ruiz-Nunez B, van den Hurk GH, de Vries JH, Mariani MA, de Jongste MJ, Dijck-Brouwer DA, et al. Patients undergoing elective coronary artery bypass grafting exhibit poor pre-operative intakes of fruit, vegetables, dietary fibre, fish and Vitamin D. *The British journal of nutrition*. 2015;113(9):1466-76.