

Research**Vitamin D blood level in nasopharyngeal carcinoma of the Minangkabau ethnic group****Sukri Rahman*, Hirowati Ali**, Beni Indra*****

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ABSTRACT

Background: One of the most common head and neck malignancy is nasopharyngeal cancer. The etiology of nasopharyngeal carcinoma (NPC) is not yet known with certainty, but is suspected to be an interaction of multiple factors. NPC is closely related to Epstein-Barr virus (EBV) infection which interacts with genetic susceptibility and environmental factors. Various studies have published the connection of vitamin D with the risk of various cancers, but very little research has reported the association with NPC. **Purpose:** To assess the correlation of vitamin D plasma level and NPC in the Minangkabau ethnic group. **Method:** This was a cross-sectional study comparing plasma levels of vitamin D [25(OH)] in new NPC patients of the Minangkabau ethnic who were treated at Dr.M.Djamil Hospital, Padang, with healthy control plasma. **Result:** The average vitamin D plasma level in the NPC group was higher than in the control group. Vitamin D levels in this study varied in both cases and controls. In the NPC group, vitamin D levels varied from 14.64 to 75.56 ng/ml. Most of the NPC group had normal vitamin D levels (69.6%), and in controls, vitamin D levels also varied from 6.06 to 73.96 ng/ml, and most of them were also with normal levels (52.2%). There was no statistically significant difference between cases and controls ($p>0.05$). **Conclusion:** This study found no significant correlation between the plasma level of vitamin D and the incidence of NPC in the Minangkabau ethnic group.

Keywords: vitamin D, 25(OH)D, nasopharyngeal carcinoma**ABSTRAK**

Latar belakang: Karsinoma nasofaring (KNF) sampai saat ini belum diketahui etiologinya secara pasti, namun diduga merupakan interaksi dari multi faktor. Terjadinya KNF dianggap berhubungan erat dengan infeksi virus Epstein-Barr (EBV) yang berinteraksi dengan kerentanan genetik dan faktor lingkungan. Berbagai penelitian telah melaporkan hubungan vitamin D dengan risiko terjadinya kanker, namun masih sangat sedikit penelitian tentang hubungan vitamin D dengan KNF. **Tujuan:** Untuk mengetahui korelasi kadar vitamin D dengan KNF pada etnik Minangkabau. **Metode:** Studi potong lintang membandingkan kadar vitamin D [25(OH)] plasma pasien baru KNF etnik Minangkabau yang berobat di Rumah Sakit Dr.M.Djamil, Padang dengan plasma kontrol sehat. **Hasil:** Didapatkan rerata kadar vitamin D pada kelompok KNF lebih tinggi dibandingkan dengan kelompok kontrol. Kadar vitamin D pada penelitian ini bervariasi, pada kasus maupun pada kontrol. Pada kelompok KNF kadar vitamin D bervariasi dari 14,64 sampai 75,56 ng/ml. Sebagian besar kelompok KNF memiliki kadar vitamin D yang normal (69,6%), dan pada kontrol kadar vitamin D juga bervariasi dari 6,06 sampai 73,96 ng/ml, dan sebagian besarnya juga dengan kadar normal (52,2%). Tidak terdapat perbedaan yang bermakna secara statistik antara kasus dan kontrol ($p>0,05$). **Kesimpulan:** Tidak didapati korelasi bermakna antara kadar vitamin D plasma dengan insidens KNF pada etnik Minangkabau.

Kata kunci: vitamin D, 25(OH)D, karsinoma nasofaring

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INTRODUCTION

Cancer is a leading cause of mortality and one of the major public health concerns at the international level. Nasopharyngeal carcinoma (NPC) is one of the most common malignancy of the head and neck in Indonesia, although the global incidence of NPC is scarce. NPC has a unique geographic distribution pattern. Knowledge about the incidence and mortality of this disease and its distribution in terms of geographical area is necessary because NPC has the highest mortality in special areas such as in South China, and in South East Asia including Indonesia. The incidence rate of NPC in Indonesia is 6.2/100,000.¹

Based on data from Dharmais Cancer Central Hospital in 2007, NPC was found to be the most frequent carcinoma in male.² In West Sumatra, the data from Dr.M. Djamil General Hospital revealed that NPC was the highest malignancy in head and neck.³

The etiology of NPC has yet to be defined, but it was assumed as multifactorial; Epstein-Barr virus (EBV) infection as the main etiology, which interact with genetic predisposition, and environmental factors. Further research of NPC etiology, as well as other new factors, could potentially reduce the impact of this disease.^{4,5}

Various researches had studied the role of vitamin D in reducing cancer risk, or enhancing cancer therapy outcome. Several accumulating data indicate that vitamin D deficiency raises the risk of developing cancer.⁶⁻⁸

The role of vitamin D in the development of malignancy became extensive studies. Some studies on several kinds of cancer got a consistent result which revealed that

the low-level vitamin D was associated with higher risk and worse prognosis for certain malignancy, such as the breast, prostate, and colorectal carcinoma.⁶ Several studies reported that vitamin D could inhibit proliferation, angiogenesis, migration and invasion of cancer cells, and the differentiation and cell apoptosis. Besides, the synthesis of prostaglandin and Wnt/b-catenin signaling are also influenced by vitamin D.^{6,7} It is written in American Cancer Society (ACS) guideline for diet and physical activity for cancer prevention, the importance of vitamin D supplementation and monitoring.⁸

Vitamin D also has an anti-inflammatory effect, and potent modulator of the adaptive immune system, so it has an important aspect in counteract various malignancy disorders, especially malignancy caused by virus infection. A laboratory study had shown that EBV activity is influenced by vitamin D, thus vitamin D carries a potential protection effect against EBV infection. Yet, it was still undetermined that vitamin D could inhibit NPC occurrence.⁹

This study was conducted to assess the correlation of vitamin D level and NPC in the Minangkabau ethnic group.

METHOD

This was a cross-sectional study comparing plasma levels of vitamin D [25(OH)] in new NPC patients of the Minangkabau ethnic who were treated at Dr. M. Djamil Hospital, Padang with healthy control plasma. The plasma sample was tested for 25(OH)D level.

There were 23 NPC cases and 23 healthy control subjects participated in this study. This research was approved by the Research

Ethical Commission of Medicine Faculty Universitas Andalas Padang, Indonesia (No.569/UN.16.2/KEP-FK/2022).

Antecubital vein blood samples were taken from 23 NPC patients and 23 healthy controls, collected and stored at the Biomedic Laboratory of Faculty of Medicine Universitas Andalas; to be examined the serum level of 25(OH)D. The samples were in plasma form, stored in aliquot temperatured -80 °C. The examination of 25(OH)D was performed using ELISA Diagnostic Biochemistry Canada (DBC) 25-Hydroxyvitamin D (DBC, London, Ontario Canada).

In this study, vitamin D level was determined by 25-hydroksyvitamin D (25(OH)D) level. The cross-section was set upon the result of earlier research of vitamin D and NPC, which divided the vitamin D level into 3 groups i.e. <37.5 nmol/L (deficient), 37.5-<75 nmol/L (insufficient), and ≥75 nmol/L (adequate); or <15 ng/ml, 15-<30, and ≥30 nmol/L.^{4,5} This study denominated the unit using ng/ml.^{10,11}

Data was analyzed using t-test for median values to evaluate the difference

between NPC groups, and using Chi-square test for control, with statistical significance level $p < 0.05$.

RESULT

Patient characteristics

The subject of study were 23 NPC cases and 23 healthy controls, in similar age (with mean difference of 3 years) and gender. The clinical and pathological characteristics of NPC were presented in Table 1. The Table 1 revealed that majority NPC patients were male, mean age 46.6 ± 12.89 years. The highest NPC cases were advanced stage (91.3%) i.e. stadium III and IV 13.0% and 78.3%, respectively. The highest pathological type was undifferentiated carcinoma (69.6%), and there was no keratinized carcinoma type.

Level of plasma 25(OH)D

The examination result of vitamin D level using ELISA was varied in the NPC, as well in control groups. In the NPC group from 14.64 to 75.56 ng/ml with median 33.62 ng/ml, while in the control group was 6.065 to 73.959 ng/ml with median 30.784 ng/ml.

Table 1. Characteristics

Characteristics	Nasopharyngeal carcinoma f (%)	Control f (%)
Gender		
Male	15 (65.2)	15 (65.2)
Female	8 (34.8)	8 (34.8)
Age (years), median±SD	46.6±12.89	46.2±13.72
20-29	3 (13.0)	3 (13.0)
30-39	2 (8.7)	3 (13.0)
40-49	9 (39.1)	8 (34.8)
50-59	4 (17.4)	4 (17.4)
≥60	5 (21.7)	5 (21.7)
Stadium		
I	0 (0.0)	
II	2 (8.7)	
III	3 (13.0)	
IV	18 (78.3)	

Pathology type	
Keratinizing squamous cell ca (WHO I)	0 (0.0)
Non-keratinizing differentiated squamous cell carcinoma (WHO II)	7 (30.4)
Undifferentiated non-keratinizing squamous cell carcinoma (WHO III)	16 (69.6)

Table 2. Level of vitamin D

Group	Vitamin D level (ng/ml)		p
	Median	SD	
NPC	38.992	14.7495	0.346
Control	34.084	19.8219	

Based on Table 2 the vitamin D median of NPC cases was higher (38.992 ± 14.7495) than control (34.084 ± 19.8219). Statistically there was no significant correlation ($p > 0.05$).

Vitamin D level was categorized into 3 groups i.e. deficient (<15 ng/ml), insufficient (15 - <30), and adequate (≥ 30 ng/ml). In Table

3, the NPC group, 4.3% had vitamin D <15 ng/ml, while the control group, 21.7% had vitamin D level <15 ng/ml. For statistical analysis, the category was grouped in two i.e. <30 ng/ml and ≥ 30 ng/ml. Statistically there was no correlation between vitamin D level and NPC incidence.

Table 3. The correlation of vitamin D level and NPC

Vitamin D level (ng/ml)	Group		p
	NPC f (%)	Control f (%)	
<15	1 (4.3)	5 (21.7)	0.365
15 - <30	6 (26.1)	6 (26.1)	
≥ 30	16 (69.6)	12 (52.2)	
Total	23 (100)	23 (100)	

DISCUSSION

In the last decade, the role of micronutrient in malignancy growth had become one of core topics in medical research. Multiple studies have assessed whether vitamin D could prevent cancer, or if it could improve cancer treatment outcomes. Several studies reported that high concentration of vitamin D in the circulation could decrease the risk of certain types of cancer including cancer of the head and neck,¹²⁻¹⁵ but other studies reported different results, stating there was no correlation between vitamin D level and the incidence of malignancy.¹⁵

Primarily, vitamin D is synthesized in the human skin after exposure to ultraviolet B (UVB) from sun ray. The secondary source of vitamin D is from diet or food supplement.¹⁶

Vitamin D obtained from sun exposure, food, and supplements, is biologically inactive, and must undergo activation through two consecutive enzymatic hydroxylation reactions to form 25-hydrox vitamin D, [25(OH)D], the major circulating vitamin D metabolite which is then converted to its active form (1.25-dihydroxyvitamin D) in the kidney and other tissues.¹⁷

This research discovered that the average vitamin D level in the NPC group was higher than in the control group. Vitamin D level in this study varied in both cases and controls. In the NPC group, vitamin D level varied from 14.64 to 75.56 ng/ml; most of the NPC group had normal vitamin D levels (69.6%), and in controls, vitamin D level also varied from 6.06 to 73.96 ng/ml, and most of them were also within normal levels (52.2%). This study found no correlation between vitamin D level and NPC in Minangkabau ethnic group.

Mai et al.¹⁰ also reported that there was no correlation between NPC and the circulating vitamin D, but they found that vitamin D level was lower in NPC patients. Some cancer studies found the relation of low level vitamin D with carcinoma incidence. This was assumed because of the role of vitamin D and its metabolites to reduce many types of cancer by inhibiting tumor angiogenesis, stimulating mutual adherence of cells, and enhancing intercellular communication through gap junction; thereby strengthening the inhibition of proliferation that results from tight physical contact with adjacent cells within a tissue.¹⁸

Several studies had shown that vitamin D decreased cancer risk through regulation of cell proliferation, cellular differentiation, induction of apoptosis, and inhibition of angiogenesis.¹⁰

NPC has strong association with Epstein-Barr virus (EBV). Although there was no correlation between vitamin D and EBV, vitamin D deficiency has been associated with tumor necrosis factor-alpha (TNF- α), which in turn has been associated with EBV in NPC incidence.¹⁰

Vitamin D is closely related to UVB exposure, where studies on mortality of various cancer in China showed inverse relation with sun rays, but not for NPC. Only for cancer of the nasopharynx did both mortality and incidence rise with increasing UVB. Possibly it was because of NPC specific

geographic distribution, and influenced by other environment factors.¹⁹

Even though many researches had found that vitamin D reduced risk of cancer or mortality, but the biological mechanism between vitamin D exposure and NPC risk remained unclear.¹⁰

This study concluded that there was no correlation between vitamin D level and NPC incidence in Minangkabau ethnic group.

REFERENCE

1. MahdaviFar N, Ghoncheh M, Mohammadian-Hafshejani A, Khosravi B, Salehiniya H. Epidemiology and Inequality in the Incidence and Mortality of Nasopharynx Cancer in Asia. *Osong Public Heal Res Perspect.* 2016; 7(6):360-72.
2. Adham M, Kurniawan AN, Muhtadi AI, Roezin A, Hermani B, Gondhowiardjo S, et al. Nasopharyngeal carcinoma in Indonesia: Epidemiology, incidence, signs, and symptoms at presentation. *Chin J Cancer.* 2012; 31(4):185-96.
3. Faiza S, Rahman S, Asri AA. Karakteristik Klinis dan Patologis Karsinoma Nasofaring di Bagian THT-KL RSUP Dr.M.Djamil Padang. *J Kesehat Andalas.* 2016; 5(1):90-6.
4. Teow S-Y, Yap H-Y, Peh S-C. Epstein-Barr Virus as a Promising Immunotherapeutic Target for Nasopharyngeal Carcinoma Treatment. *J Pathog.* 2017; 2017(2017):1-10.
5. Tsang CM, Tsao SW. The role of Epstein-Barr virus infection in the pathogenesis of nasopharyngeal carcinoma. *Virol Sin.* 2015;30(2):107-21.
6. Feldman D, Krishnan AV, Swami S, Giovannucci E, Feldman BJ. The role of vitamin D in reducing cancer risk and progression. *Nat Rev Cancer [Internet].* 2014; 14(5):342-57.
7. McDonnell SL, Baggerly CA, French CB, Baggerly LL, Garland CF, Gorham ED, Hollis BW, Trump DL, Lappe JM. Breast cancer risk markedly lower with serum 25-hydroxyvitamin D concentrations ≥ 60 vs < 20 ng/ml (150 vs 50 nmol/L): Pooled analysis of two randomized trials and a prospective cohort. *PLoS One.* 2018; 13(6):e0199265.

8. Rock CL, Thomson C, Gansler T, Gapstur SM, McCullough ML, Patel A V., et al. American Cancer Society guideline for diet and physical activity for cancer prevention. *CA Cancer J Clin.* 2020; 70(4):245-71.
9. Disanto G, Meier U, Giovannoni G, Ramagopalan S V. Vitamin D: A link between Epstein-Barr virus and multiple sclerosis development? *Expert Rev Neurother.* 2011; 11(9):1221-4.
10. Mai ZM, Ngan RKC, Ng WT, Lin JH, Kwong DLW, Yuen KT, et al. Low vitamin D exposure and risk of nasopharyngeal carcinoma: Observational and genetic evidence from a multicenter case-control study. *Clin Nutr.* 2021; 40(9):5180-188.
11. Mai ZM, Lin JH, Ngan RKC, Kwong DLW, Ng WT, Ng AWY, et al. Solar ultraviolet radiation and vitamin D deficiency on epstein-Barr virus reactivation: Observational and genetic evidence from a nasopharyngeal carcinoma-endemic population. *Open Forum Infect Dis.* 2020; 7(10):ofaa426.
12. Amrein K, Scherkl M, Hoffmann M, Neuwersch-Sommeregger S, Köstenberger M, Tmava Berisha A, et al. Vitamin D deficiency 2.0: an update on the current status worldwide. *Eur J Clin Nutr.* 2020; 74(11):1498-513.
13. Skaaby T, Husemoen LLN, Thuesen BH, Pisinger C, Jørgensen T, Roswall N, et al. Prospective population-based study of the association between serum 25-hydroxyvitamin-D levels and the incidence of specific types of cancer. *Cancer Epidemiol Biomarkers Prev.* 2014; 23(7):1220-9.
14. Izreig S, Hajek M, Edwards HA, Mehra S, Sasaki C, Judson BL, et al. The role of vitamin D in head and neck cancer. *Laryngoscope Investig Otolaryngol.* 2020; 5(6):1079-88.
15. Young MRI, Xiong Y. Influence of vitamin D on cancer risk and treatment: Why the variability? *Trends Cancer Res.* 2018;13:43-53.
16. Ángel Gil, Julio Plaza-Díaz, María Dolores Mesa; Vitamin D: Classic and Novel Actions. *Ann Nutr Metab.* 2018; 72 (2): 87-95.
17. Muller DC, Hodge AM, Fanidi A, Albanes D, Mai XM, Shu XO, et al. No association between circulating concentrations of vitamin D and risk of lung cancer: an analysis in 20 prospective studies in the Lung Cancer Cohort Consortium (LC3). *Ann Oncol.* 2018; 29(6):1468-1475.
18. Garland CF, Garland FC, Gorham ED, Lipkin M, Newmark H, Mohr SB, et al. The role of vitamin D in cancer prevention. *Am J Public Health.* 2006;96(2):252-61.
19. Chen W, Clements M, Rahman B, Zhang S, Qiao Y, Armstrong BK. Relationship between cancer mortality/incidence and ambient ultraviolet B irradiance in China. *Cancer Causes Control.* 2010; 21(10):1701-9.