國立臺灣大學公共衛生學院流行病學與預防醫學研究所

博士論文

Graduate Institute of Epidemiology and Preventive Medicine

College of Public Health

National Taiwan University

Doctoral Dissertation

維生素 D 缺乏與結核病感染之關聯性:

配對密度病例對照研究

Vitamin D Deficiency and Incident Active Tuberculosis:

A Matched Density Case-Control Study

許孟璇

Meng-Shiuan Hsu

指導教授: 方啓泰 博士

Advisor: Chi-Tai Fang, MD, PhD

中華民國 112 年 5 月

May 2023

doi:10.6342/NTU202301094

國立臺灣大學博士學位論文
口試委員會審定書

論文中文題目

維生素D缺乏與結核病感染之關聯性:

配對密度病例對照研究

論文英文題目

Vitamin D Deficiency and Incident Active Tuberculosis among non-HIV patients:

A Matched Density Case-Control Study

本論文係 **許孟璇** 君(學號 D06849001)在國立臺灣大學流行病學與預防醫學研究所完成之博士學位論文, 於民國112年5月9日承下列考試委員審查通過及口試及格, 特此證明。

口試委員: 方啓泰 (簽名)

(指導教授)

王振源

方以全

郭世隆

王振泰

林菀俞

誌　　謝

我知道念博士班不容易。
我也知道念博士班再加上臨床工作會更辛苦。
我更知道念博士班再加上臨床工作又要兼顧家庭，會是一個不可能的任務。
但，唸碩士班的經驗告訴我，方老師一定可以帶領我完成這個不可能的任務，再加上家人的鼓勵，於是，我毅然決然的報名了博士班的考試。人類因夢想而偉大，不是嗎?

著手進行第一個研究(Ascorbic acid-vitamin B-Corticosteroid in immune-modulation of sepsis/septic shock, 簡稱 ABC study (**Appendix I**)的前置作業就充滿艱辛。台灣目前沒有實驗室可以協助檢測血漿中 vitamin B1, vitamin C 的濃度。 於是我先參考了國外(Mayo Clinic)的檢驗方法，再尋找台灣有串聯式質譜儀的醫院。在此非常感謝**淡水馬偕醫院的醫學研究部生化遺傳研究組的莊志光博士及魏方潔組長**，非常阿莎力的同意協助我進行這個研究，也參考了許多國外文獻來確認檢驗方法，終於解決了這個研究的關鍵步驟，可以開始著手進行研究畫撰寫了。然而，本研究所給予的 vitamin B1 的劑量(200mg q12h)超過現行仿單的建議劑量(100mg q12h), 於是 IRB 又將案子轉送到 TFDA 進行審查，於是經過好幾個月的公文往返，終於通過審查(**Appendix II**)，可以開始收案了。只是沒想到，才收了兩個 cases,竟然爆發了 SARS-COV 2 疫情。加護病房禁止訪客，急診也限制進出，收案進度緩慢。與方老師討論後，決定放棄這個研究案，另起爐灶…於是這 2-3 年的努力付之流水。我也只能安慰自己，至少享受了過程…把這次的經驗可以做為以後寫研究計畫的參考。

在等待 TFDA 審查研究案的同時，我們也沒有浪費時間。在此，要感謝**環衛所的楊婉秀學姊及蔡坤憲教授**。學姊慷慨的將其碩士班的研究資料提供給我進行論文撰寫，而蔡教授則協助後續的論文潤飾。蔡老師修改後的論文，比英修過的還美，連我都不相信這是我寫的了。這一篇論文目前刊登在熱帶醫學期刊排名 Q1 的 PLOS Neglected Tropical Diseases 中(**Appendix III**)，不僅是我莫大的榮幸，同時也讓我跟畢業的距離更接近了。

放棄了 ABC 之後，我立刻開始了 D? 在方老師的建議下，我著手進行 **Vitamin D 與 TB** 的相關研究。本研究可以完成，首先要感謝劉郁芬及林秀卿兩位 TB 個管師，協助收案，常常連下班、假日、過年期間都會被我騷擾。另外，要感謝**心臟科吳彥雯主任及家醫科陳志道主任**，同意讓我收心臟科的病人及預防保健中心的病人做為研究中對照組。特別是心臟科的吳主任，連研究助理(**陳永崢**

doi:10.6342/NTU202301094

助理，簡妘恩助理，羅雅瓊助理，陳佳琪助理)都借我了，對一個沒有研究助理卻想要完成一個 300~400 人的研究來說，真的幫我解決最大的困擾，讓我在專心進行我的臨床業務的同時還能收案。在此感謝心臟科的同事吳彥雯醫師，許榮城醫師，曾炳憲醫師，林恆旭醫師，杜宗明醫師，黃繼正醫師，張藝耀醫師，蔡浩元醫師，江俊賢醫師，羅顯榮醫師，劉芃宏醫師，邱昱偉醫師，李建霖醫師，黃姍惠醫師，廖本智醫師，李愛先醫師，陳運淇醫師等幾位心臟科醫師協助收案，不求回報。也謝謝每一個願意捐 3cc 的血給我進行研究的每一位病人及院內同仁。然而，研究經費有限也是進行研究時一個很現實的問題，非常感謝檢驗醫學部的朱芳業主任的大力協助，讓我僅需負擔試驗藥品的成本就完成了 vitamin D 的檢測，約省了 80%的研究經費。因為博士計畫書口試時，委員希望能再將 pulmonary TB 病人臨床表現的嚴重性與 vitamin D 的關聯再進一步的分析。在此，我要感謝胸腔科鄭世隆主任，在大年初四的下午，大部分的人應該都還在家裏放年假的時候，在醫院裏陪我看了 62 個 TB 病人的 chest images。最後，還要感謝流預所碩士班的鍾子謙學妹協助完成論文中 SAS 統計的部分。

這六年來，事業和學業佔據了我大部分的時間，如果不是有一個默默在背後支持我的團隊，我應該早就放棄了。我稱他們為「地表最強的工具人」團隊，其中，我特別要感謝的是我的**父親大人**。這六年來，他幾乎取代了我在家裏的功能，每天接送我女兒上下課，去補習班，準備三餐…。而這六年也是我的寶貝女兒志君要準備考高中會考及大學學測的關鍵六年，她卻被我整整忽略了六年。今年我們一起畢業了，可她即將遠赴高雄就學，心中對她的虧欠，我一定會用這個暑假好好的補償她。

回想起來，方啓泰**老師**和我的師生緣從台大的感染科 fellow training 到碩士班再到博士班，加起來至少有 12 年了。我不是一個聰明的學生，但老師總是耐心的指導我，記得在準備博班資格考時，方老師還特別播空幫我開了一堂「**傳染病數理模型」的家教課**，讓我成功的跨過了博士班最令人擔心的門檻。真的非常感謝方老師幫我圓了這個背了 20 年的夢想。在這裏要告訴學弟妹們，帶著你堅定的心，跟著方老師的步調走，然後有一天你真的就會到終點的。

最後，要感謝這六年來，所有一路陪我、支持我、挺我走過每一步的人。有你們的鼓勵與幫忙，才造就出今天的我。借一句陳之藩先生的話:「因為要感謝的人太多，那就謝天吧!」

學生 許孟璇 2023/6/7

doi:10.6342/NTU202301094

中文摘要

背景

維生素 D 是正常的先天性和適應性免疫功能對抗結核分枝桿菌的關鍵調節因子。統合分析證據顯示，維生素 D 缺乏會增加結核病感染風險，但實驗設計的不同，使其結論仍存在爭議。本研究旨在研究維生素 D 缺乏是否與台灣的結核病感染有關，並探討造成結核病感染的潛在危險因素。

方法

本研究是一項在台灣，位於東亞北緯 24 度的國家，所進行的性別和年齡匹配的病例對照研究 (1:4)，旨在探索維生素 D 缺乏與非 HIV 的結核病感染患者之間的關係。並用條件式羅吉斯迴歸的方法檢查結核病感染的潛在風險因素。

結果

本研究包括 62 名結核病患者和 248 名對照組病患。總體而言，34.2% (106/310) 的參與者都有維生素 D 缺乏的情形。結核病患者的血漿中平均 25(OH)D 的濃度較對照組顯著為低 (21.25±8.93 ng/ml vs 24.45±8.36 ng/ml，p=0.008)。單變量分析發現，較低的身體質量指數 (22.9±3.3 kg/m^2 vs 25.2±3.9 kg/m^2, P<0.001)，吸煙者 (n=22, 35.5% vs n=43, 17.3%, p=0.002)，有使用酒精濫用的行為 (n=3, 4.8% vs n=1, 0.4%, p=0.026) 和維生素 D 不足 20 ng/ml (n=32, 51.6% vs n=74, 29.8%, p=0.001) 與對照組相比，和結核感染有顯著相關;而患有肝硬化病患則有傾向與結核感染有關 (n=3, 4.8% vs n=2, 0.8%, p=0.056)。多變量分析發現，維生素 D 不足 20 ng/ml(p=0.002,aOR=3.034 (95%信賴區間為 1.510-6.095)，較低的身體質量指數(p<0.001,aOR=0.805 (95%信賴區間為 0.721-0.899)、

doi:10.6342/NTU202301094

有肝硬化患者(p=0.042,aOR=8.992 (95%信賴區間為 1.088-74.344 以及吸煙者(p=0.001, aOR=4.516 (95%信賴區間為 1.853-11.002)和與對照組相比，仍然是結核病感染的獨立危險因子。

結論

與對照個體相比，台灣的非 HIV 結核感染病患者有顯著的維生素 D 缺乏的現象。此外，我們的數據表明，較低的身體質量指數、吸煙者、肝硬化和 25(OH)D < 20 ng/ml 是感染結核病的獨立危險因素。

關鍵詞: 肺結核, 維生素 D 缺乏, 身體質量指數, 吸煙,肝硬化， 25(OH)D

doi:10.6342/NTU202301094

Abstract

Background: Vitamin D is an essential modulator for normal innate and adaptive immune function to combat *Mycobacterium tuberculosis* infection (TB). However, the association between vitamin D deficiency (VDD, i.e., Vitamin D concentration<20 ng/ml) and tuberculosis remain controversial. Previous studies didn't control known important confounders, such as body-mass index (BMI), smoking, and diabetes mellitus (DM), and yielded highly heterogeneous results. This study aimed to investigate whether VDD is associated with TB in Taiwan and to identify potential risk factors for TB.

Methods and Findings: A sex- and age-matched case-control study (1:4) was conducted to explore the association between VDD (<20 ng/ml) and TB among non-HIV patients at a latitude of 24°N in Taiwan. Conditional logistic regression was used to examine potential risk factors. The study included 62 TB patients and 248 controls. Overall, VDD was diagnosed in 34.2% (106/310) of all participants. The mean 25(OH)D level was significantly lower in TB cases compared to control cases (21.25 ± 8.93 ng/ml vs 24.45 ± 8.36 ng/ml, multi-*p*=0.008, aOR=0.95 [95% CI = 0.91-0.99]). Multivariable analysis found that VDD (*p*=0.002), lower body mass index (*p*<0.001), liver cirrhosis (*p*=0.042), and smoking (*p*=0.001) were independent risk factors for TB comparing to the control group. The adjusted odds ratio (aOR) and 95% confidence interval (CI) were 3.034 [95% CI= 1.5-6.1], 0.805 [95% CI= 0.7-0.9], 8.992 [95% CI=1.1-74.3], and 4.516 [95% CI=1.9-11], respectively.

Conclusions: Vitamin D deficiency is an independent risk factor for incident active TB among non-HIV patients. Randomized controlled trials are warranted to examine whether vitamin D supplementation reduced the risk of incident TB in high-risk patients with vitamin D deficiency.

doi:10.6342/NTU202301094

Keywords: Tuberculosis, Vitamin D deficiency, Body mass index, Liver cirrhosis, Smoking, 25(OH)D

doi:10.6342/NTU202301094

目 錄

doi:10.6342/NTU202301094

圖 目 錄

doi:10.6342/NTU202301094

表 目 錄

doi:10.6342/NTU202301094

Background

Tuberculosis is a communicable disease and one of the leading causes of death worldwide. According to the World Health Organization (WHO), there were 10.6 million people fell ill with TB, an increase of 4.5% from 10.1 million in 2020 and 1.6 million TB-related deaths in 2021 [1]. The rising of incidence rate of TB by 3.6% between 2020 and 2021 sets back the aim of the End TB strategy. An innovative approach for TB control is desperate needed.

It is known that TB is a highly infectious disease, which can be transmitted by inhalation of a minimal dose (1-5 bacilli) of *Mycobacterium tuberculosis*, and one in ten infected persons may develop active TB [2]. An individual's susceptibility to developing active TB is associated with multiple factors, including environmental, genetic, socioeconomic, and nutritional factors. Current evidence reveals that genetic variability can influence an individual's susceptibility to active TB [3]. Many studies have focused on the polymorphisms of different gene candidates [4] , with a particular interest in the gene for the vitamin D receptor (VDR) [5-9]. The findings support the role of vitamin D in TB pathophysiology.

Though vitamin D is a key modulator for innate and adaptive immunity against *Mycobacterium tuberculosis*. [10-14], the association between vitamin D deficiency and tuberculosis remain controversial. Previous studies did not control known important confounders, such as body mass index (BMI), smoking, diabetes mellitus (DM) and yielded highly heterogenous results [15-18]. Studies from populations in Indonesia [19], China [20], West Africa [21], and South Korea [22] showed no significant association between vitamin D deficiency and TB. On the other hand, reports from London [23], South Africa [24], Pakistan [25], and India [26] showed a significant association between lower vitamin D levels and susceptibility to TB.

doi:10.6342/NTU202301094

Taiwan is a highly prosperous industrialized country with universal health coverage and social welfare. Herein, we conducted a high-powered case-control study (1:4) to explore the association between vitamin D deficiency and tuberculosis in non-HIV patients.

doi:10.6342/NTU202301094

Patients and Methods

Study design:

The investigation was conducted at Far Eastern Memorial Hospital, New Taipei City (latitude 24°N), Taiwan. Far Eastern Memorial Hospital is a tertiary medical center with an outpatient service volume of more than 6,500 persons per day. In the past 3 years, around one hundred patients per year have undergone anti-TB regimens at our hospital.

TB patients were recruited from September 15, 2021, to September 14, 2022. TB cases were selected from patients aged over 20 with microbiological evidence of TB, including those who were newly diagnosed with TB during the study period and those who had been diagnosed with TB before the study initiation but were still receiving anti-TB medication treatment during the study period. Four controls were recruited for each TB case, matched based on sex and age, by screening outpatients from cardiovascular outpatient departments, Integrated Preventive Health Center, and healthcare members for eligible subjects from September 15, 2021 to December 14, 2022. Eligible controls were excluded from participating in the study if they had received vitamin D supplements within one month. All participants with human immunodeficiency virus (HIV) infection and pregnancy were also excluded. To control the sunlight exposure level, only participants living in the northern area of Taiwan were enrolled.

Ethical Statement

This investigation was approved by the Research Ethics Review Committee of Far Eastern Memorial Hospital (IRB number 110133-E), and written informed consent was obtained from all participants before enrollment.

doi:10.6342/NTU202301094

Data collection

Demographic information, including sex, age, body mass index, and place of residence, was recorded. All participants underwent a detailed medical history, including underlying diseases, alcohol consumption, and smoking history. To define smoking status, we grouped patients into either smokers (i.e., those who smoked cigarettes at the time of study enrollment or had previously smoked) or non-smokers (i.e., those who had never smoked or had smoked < 100 cigarettes in their lifetime) [27]. Additional clinical data in the TB group was collected, including TB sites, duration of anti-TB regimens, grading of acid-fast bacilli (AFB) stain [28], time to AFB and culture negative conversion, chest x-ray (CXR), or chest computer tomography (CT) findings on the day of diagnosis.

Vitamin D concentration and status definition

Total circulating serum 25(OH)D was measured by the Cobas® e 801 Module by Roche Diagnostics according to the manufacturer's protocols. The level of 25(OH)D was expressed as ng/mL (to convert ng/mL to nmol/L, multiply by 2.49). We defined the serum 25(OH)D level into four categories based on the Endocrine Society Clinical Practice Guidelines with modification into four categories [29]. Briefly, vitamin D "deficiency" (VDD) was considered when the serum 25(OH)D was < 20 ng/ml. Further, "severe deficiency" was considered as serum 25(OH)D < 10 ng/mL and "moderate deficiency" was considered as the serum 25(OH)D level between 10 ng/mL to 20 ng/ml. Serum 25(OH)D levels of 20 to 30 ng/mL were considered "insufficiency". Serum vitamin 25(OH)D ≧ 30 ng/mL was considered "sufficiency".

Statistical analysis

Statistical analyses were performed using SAS 9.4 (SAS Institute, Cary, North Carolina, USA). Categorical variables were expressed as percentages, and continuous variables were expressed as

doi:10.6342/NTU202301094

mean ± standard deviation. A Chi-square test was used to compare two proportions. Fisher's exact test was used when any value in the cells of the contingency table was smaller than five. Differences between groups for continuous data were compared using Student's t-test. Conditional logistic regression was used to analyze the risk factors, and the exact method was used. All variables with $P < 0.10$ in the univariable analysis and the risk factors mentioned in previous literature were included in the hypothesis-driven model, and stepwise selection was used during the multivariable analyses. All statistical tests were two-tailed, and $P < 0.05$ was considered statistically significant.

doi:10.6342/NTU202301094

Results

TB Patients

During the study period from September 15, 2021, to September 14, 2022, a total of 153 patients were notified to the Taiwan CDC (Centers for Disease Control) with newly diagnosed tuberculosis, and 73 patients who were diagnosed before our study period remained in our hospital for tuberculosis treatment (Fig 1A). Patients who were younger than 20 (n=6), without microbiology evidence (n=53), patients who refused (n=64), those who had taken vitamin D within one month (n=2), those who died (n=24), or those who transferred to another hospital before recruitment (n=15) were excluded from our study. Finally, 41 patients were recruited for our study. On the other hand, 73 patients were diagnosed before the initiation of our study but were still receiving anti-TB medication during the study period. Fifty-two patients who didn't meet the criteria were excluded, and finally, 21 patients were recruited. In total, 62 TB-infected patients were included in our study, with an average age of 59 years old, and 71% (n=44) were male (table 1). All patients came from northern Taiwan (Keelung to the north of Hsinchu) to eliminate the influence of sunlight exposure. Most patients had pulmonary TB (59 out of 62, 95%), and only 3 patients had extrapulmonary TB, including one with TB meningitis and the other two with TB lymphadenitis of the neck. The average 25(OH)D concentration of the 62 patients was 21.25±8.93 (5.3-45.8) ng/mL, and among them, 32 (51.6%) patients had VDD (i.e., 25(OH)D concentration less than 20 ng/ml). Other baseline demographic characteristics and underlying medical diseases were shown in Table 1.

These 32 TB patients with VDD cases had an average level of 14.34±3.79 (range 5.3-19.9) ng/ml, including four (6.5%) had severe VDD with levels below 10 ng/mL, while 28 cases (45.2%) had moderate VDD with levels between 10 and 20 ng/ml (table 2). Since 95% of TB patients had pulmonary infection, we further compared the differences of clinical manifestation in patients with

doi:10.6342/NTU202301094

or without VDD. Around half of patients had positive AFB stain in sputum (55% vs 53%, respectively) and most of them had high grading (4+) (41% vs 33%, p=0.647). Nearly 60% of patients had cavitation on CXR (58% vs 57%). Half of patients with VDD (55.6%) had multiple lobar involved; while only 31.3% of patients without VDD had the same findings but no significant difference in statistics analysis (p=0.154). Furthermore, the duration of negative conversion of sputum, either AFB or culture, didn't have significant differences in patients with or without VDD.

Twenty-two patients in TB group (22 out of 62, 35.5%) had their 25(OH)D measured within 30 days (i.e., 11 ± 8 days) of the initiation of the anti-TB regimens; the other 40 patients had been treated for more than one month with anti-TB regimens (i.e., 101 ± 46 days) at the time of 25(OH)D measurement (Table 3). There was no difference in the measured 25(OH)D level between those who received anti-TB drugs for less or more than 30 days (22 ± 8.8 ng/ml (11.1-45.8) vs 21 ± 9.1ng/ml (5.3-37.7), p=0.85). In addition, there was no association between the vitamin D concentration and the duration of anti-TB treatment (r=-0.013, p=0.92) (supplement 1).

Control group

Between Sep 15, 2021, and December 14, 2022, a total of 257 cases were included in the control group. Among them, 154 patients (60%) came from the cardiovascular outpatient department, 35 (13.6%) patients came from the Integrated Preventive Health Center, and 68 (26.5%) patients came from voluntary health-care members (Fig 1B). Six patients were excluded because three patients' age did not match and another three patients were under vitamin D supplementation. Finally, 248 patients were recruited in our control group, including 176 men and 72 women with an average age of 58.6±14.5 (range 20-89) years old. Other clinical characteristics of age- and sex-matched participants in the control group are shown in Table 1.

doi:10.6342/NTU202301094

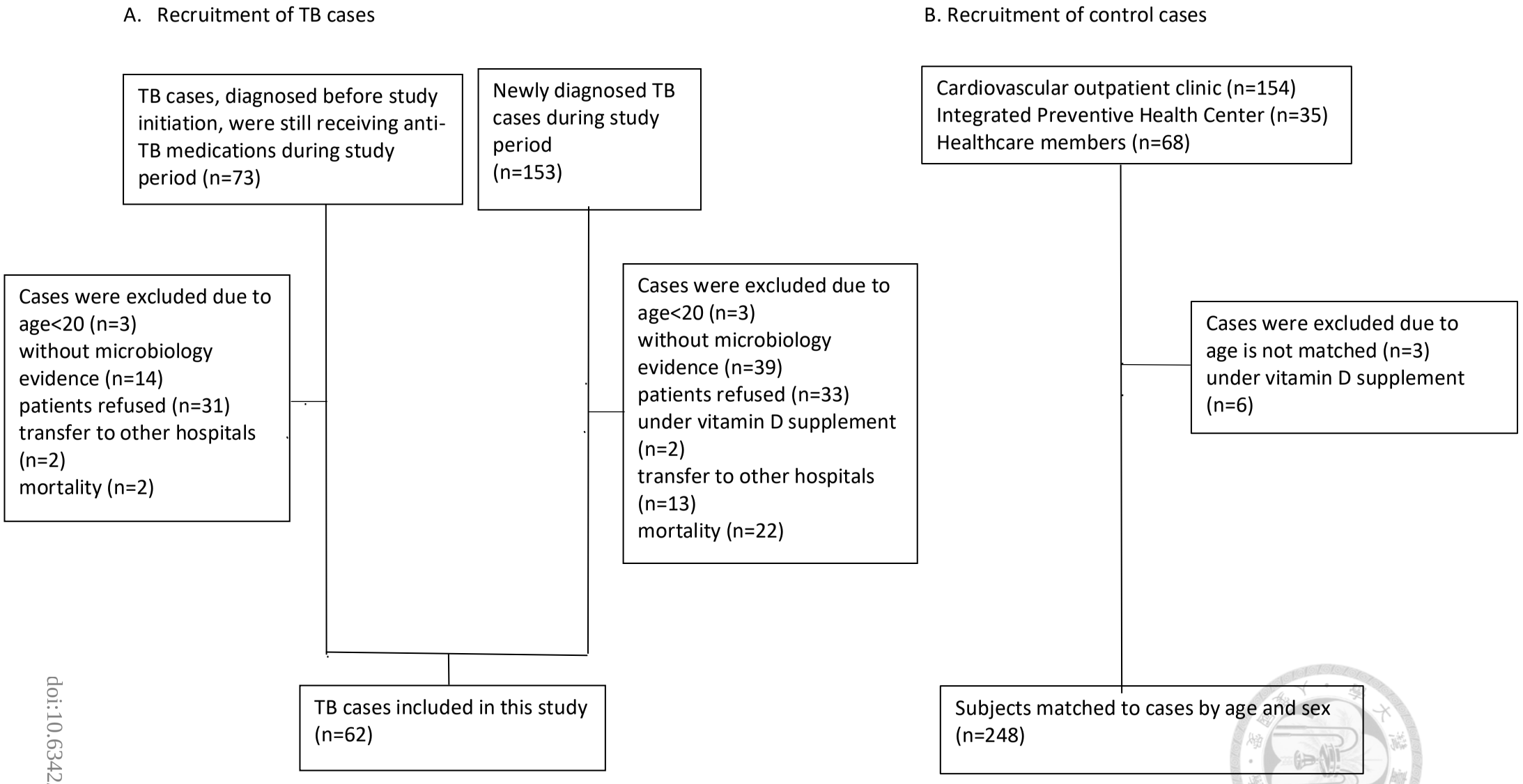


Fig 1. Summary of the recruitment process included the (A) TB cases and (B) control cases.

doi:10.6342/N

12301094

Table 1. Clinical characteristics of sex-and age-matched TB and control cases

	TB (n=62)	Control (n=248)	Univariable *p* value
Age (years)	58.5±14.3 (21~88)	58.6±14.5 (20~89)	0.955
Male sex	44 (71%)	176 (71%)	1
Body mass index(kg/m^2)	22.9±3.3 (14.7~30.1)	25.2±3.9 (16.2~40.7)	<0.001
Habitat in Taiwan	North (100%)	North (100%)	1
Diabetes mellitus	16 (25.9%)	55 (22.2%)	0.54
Steroid use	0 (0%)	2 (0.8%)	0.48
End stage renal disease	0 (0%)	1 (0.4%)	1.0
Liver cirrhosis	3 (4.8%)	2 (0.8%)	0.056
Congestive heart failure	2 (3.2%)	16 (6.5%)	0.330
Smoking	22 (35.5%)	43 (17.3%)	0.002
Alcohol use behavior	3 (4.8%)	1 (0.4%)	0.026
25(OH)D level (ng/ml)	21.25±8.93 (5.3~45.8)	24.45±8.36 (6.3~52.2)	0.008
25(OH)D <20 ng/mL	32 (51.6%)	74 (29.8%)	0.001

doi:10.6342/NTU202301094

Table 2. Comparison of vitamin D level <20 and >= 20 ng/ml in TB patients

	Vitamin D concentration <20 ng/ml (n=32)	Vitamin D concentration ≧20 ng/ml (n=30)	*p* value
Absolute vitamin D concentration (ng/ml)	14.34±3.79 (5.3~19.9)	28.62±6.6 (20~45.8)	<0.0011
Age (years)	56.66±16.20 (29~88)	60.47±11.89 (21~80)	0.298
Male	23 (72%)	21 (70%)	0.871
BMI (kg/m^2)	22.37±3.46 (14.72~29.23)	23.41±3.08 (17.89~30.05)	0.22
Diabetes mellitus	9 (28%)	6 (20%)	0.455
Pulmonary tuberculosis	31 (97%)	28 (93.3%)	0.607
sputum AFB stain positive/ grading in mass	17 (55%)/4+ (n=7, 41%)	15 (53%)/4+ (n=5, 33%)	0.622/0.647
cavitation on CXR/multiple lobes	18 (58%)/10 (55.6%)	16 (57%)/5 (31.3%)	0.818/0.154
negative conversion of sputum culture (days)	37.57±22.26 (n=28*, 6~148)	42.92±34.35 (n=25**, 13~109)	0.5
negative conversion of sputum AFB (days)	33.25±18.23 (n=16†, 10~55)	75.21±100.08 (n=14‡, 6~360)	0.145

*: Three patients' sputum culture didn't have microbiology evidence and TB was diagnosed by lung tissue biopsy in two patients and bronchial washing fluid in one patient. **: Three patients' sputum culture didn't have microbiology evidence and TB was diagnosed by lung tissue biopsy in two patients and bronchial washing fluid in one patient. †: One patient's sputum AFB was not yet negative conversion. ‡: One patient's sputum AFB was not yet negative conversion.

doi:10.6342/NTU202301094

Table 3. Comparison of the TB patients with receiving anti-TB medication over and less than 30 days

	<=30 days (n=22)	>30 days (n=40)	*p* value
Duration between anti-TB regimen and vitamin D test (days)	10.5±7.87 (0~21)	100.86±45.45 (36~370)	<0.001
Age (years)	58.77±13.9 (37~85)	58.4±14.7 (21~88)	0.912
Male	16 (73%)	28 (70%)	1
Pulmonary tuberculosis	21[*] (95.5%)	38[†] (95%)	1
BMI (kg/m^2)	22.13±3.73	23.3±3.01	0.188
Vitamin D concentration (ng/ml)	21.96±8.84 (11.1~45.8)	20.86±9.06 (5.3~37.7)	0.647
Proportion of vitamin D concentration <20 ng/ml	11 (50%)	21 (52.5%)	0.851

doi:10.6342/NTU202301094

Clinical Characteristics of Age- and Sex-Matched TB and Control Cases

Overall, vitamin D deficiency (VDD was diagnosed in 34.2% (106/310) of all participants. In univariable analysis (TB cases vs control cases), as expected, the BMI was significantly lower in TB cases (22.9±3.3 (14.7-30.1) kg/m2) than in control cases (25.2±3.9 (16.2-40.7) kg/m2) (p< 0.001) (Table 1). Additionally, patients with TB had a significantly higher proportion of smokers (35.5% vs 17.3%, *p*=0.002), alcohol use behavior (4.4% vs 0.6%, *p*=0.026), and moderate to severe VDD (i.e., 25(OH)D < 20 ng/dL) compared to control cases (n=32, 51.6% vs n=74, 29.8%, *p*=0.001). The absolute 25(OH)D concentration was also significantly lower in TB patients than in the subjects of the control group (21.25±8.93 (5.3-45.8) vs 24.45±8.36 (6.3-52.2), *p*=0.008) (Fig 2). There was a borderline significantly higher proportion of liver cirrhosis among the TB cases (4.8%) compared to control cases (0.8%) (*p*=0.056). There were no significant differences in underlying diseases such as diabetes mellitus (26% vs 22%, *p*=0.54), steroid use (0% vs 0.8%, *p*=0.48), end-stage renal disease (0% vs 0.4%, *p*=1.0), and congestive heart failure (3.2% vs 6.5%, *p*=0.33).

doi:10.6342/NTU202301094

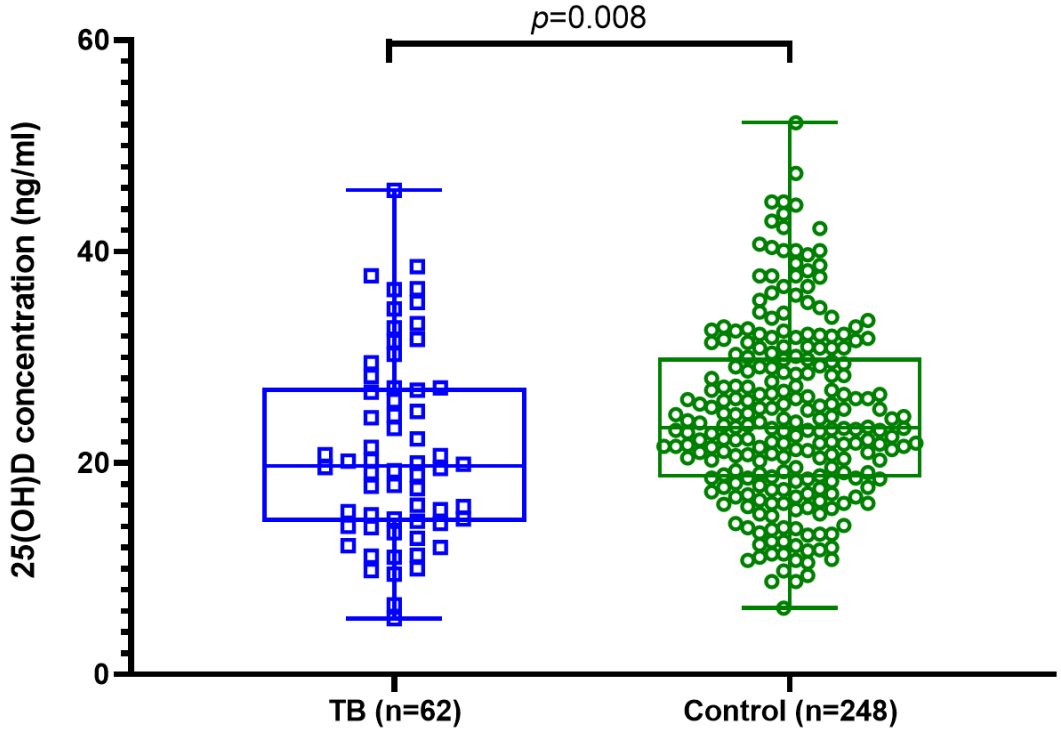


Fig 2. The distribution of individual 25(OH)D level of TB and control cases

doi:10.6342/NTU202301094

In multivariable analysis, vitamin D < 20 ng/ml (p=0.002), lower BMI (p<0.001), liver cirrhosis (p=0.042), and smoking (p=0.001) remained significant risk factors for patients with TB infection compared to those who were not infected with TB. The adjusted odds ratio (aOR) and 95% confidence interval [CI] of the four independent risk factors were 3.03 [95% CI=1.51-6.10], 0.81 [95% CI=0.72-0.90], 8.99 [95% CI=1.09-74.34], and 4.52 [95% CI=1.85-11.00], respectively (Table 4). When we replaced VDD by absolute 25(OH)D concentration, the four factors remained significant (supplement 2). The correlation between BMI and vitamin D level was significant low (r=0.133, p=0.02, supplement 3). In addition, the association of vitamin D concentration with TB risk presented a linear, not an U-shape, (supplement 4).

doi:10.6342/NTU202301094

Table 4.
Multivariable analysis of sex- and age-matched TB and control cases with the items of ratio of 25(OH)D <20 ng/mL, body mass index, liver cirrhosis, smoking, alcohol use disorders in the model

	p-value	Adjusted odd ratio	95% confidence interval
Ratio of 25(OH)D <20 ng/mL	0.002	3.034	1.510~6.095
Body mass index	<0.001	0.805	0.721~0.899
Liver cirrhosis	0.042	8.992	1.088~74.344
Smoking	0.001	4.516	1.853~11.002
Alcohol use behavior	0.138	7.23	0.513~98.404

doi:10.6342/NTU202301094

Discussion

Worldwide surveillance reports have shown that VDD is a common health problem, with 37.3% of the population having a mean value below 20 ng/ml [30, 31]. We also found that 34.2% of our study population has VDD. In addition, our present study demonstrated that the absolute serum 25(OH)D level is significantly lower in TB cases than in control cases (p=0.008, aOR=0.95 [95% CI=0.91-0.99]). Moreover, severe to moderate vitamin D deficiency was more prevalent in TB cases than in control cases (univariable p= 0.008, multivariable p= 0.002, aOR=3.034), which hinted when the serum 25(OH)D level fell below a critical point (i.e., <20 ng/ml), the risk of TB increases threefold. Our study concluded that after adjusting for effects of BMI, smoking and DM, VDD remain an independent risk factor for TB.

In India, Jaimni V et al. reported that patients with VDD were associated with more severe symptoms, higher sputum smear positivity, and extensive lesions on CXR images [32]. Though the sample size was not powered to compare differences in our study outcome, we noted a higher ratio of AFB 4+ (41% vs 33%, p=0.647) and multiple lobar cavitation in chest image manifestation (55.6% vs 31.3%, p=0.154) in TB patients with VDD than those without VDD. Large scale study may be needed to prove this association.

Our study found that the measured vitamin D level did not differ significantly between TB patients who had been treated with anti-TB drugs lesser than 30 days and those for 30 days or more (21.96 vs 20.86 ng/mL, p=0.647). The same findings were noted in Huang S J's meta-analysis, which revealed anti-TB treatment did not affect the 25(OH)D level and TB patients who completed anti-TB treatment still had lower levels of 25(OH)D than non-TB controlled cases [16]. We suggested that VDD is a risk factor rather than a consequence for TB.

doi:10.6342/NTU202301094

Chronic undernutrition is prevalent in low- to middle-income countries, such as sub-Saharan Africa and Southern Asia, where TB is endemic due to the secondary immunodeficiency attributed to malnutrition [33]. BMI is a commonly used indicator to measure nutritional status, and lower BMI has been found to be associated with TB in several studies. In Ethiopia, Workineh M et al. reported significantly lower BMI in the TB group compared to the control group (mean BMI 17.4 kg/m2 vs 21.4 kg/m2, $p < 0.001$) [34]. In India, Jaimni V et al. reported the same finding that the TB group had a significantly lower BMI than the control group (19.4 kg/m2 vs 24.0 kg/m2, $p < 0.0001$) [32]. In Korea, Hong JY et al. also noted this finding that the BMI of the TB group was lower than the control group (20.65 kg/m2 vs 23.65 kg/m2, $p < 0.001$) [35]. Our study also found a lower BMI in TB cases compared to control cases (22.9 kg/m2 vs 25.2 kg/m2, univariable $p < 0.001$, multivariable $p < 0.001$). Although different ethnicities and countries have different BMI statuses, lower BMI was clearly associated with TB. Reflecting lower BMI as a risk factor for TB, Bueno H et al. reported that increasing a 2600 kcal/day diet for adults with a BMI of 16-18.4 kg/m2 would be a cost-effective strategy in reducing TB incidence and mortality in India [36]. Undernutrition can negatively influence the phagocytosis, antigen presentation, activation of Th1 response, and granulomatous formation processes that target *M. tuberculosis*. It is not surprising to find that lower BMI is a risk factor for TB [33].

The deteriorating effects of smoking on TB have previously been documented. In a meta-analysis, Bates MN et al. produced evidence that smoking is a risk factor for TB [37]. Sitas F et al. further reported that smoking was a major risk of death for TB in South Africa [38]. Khan AH et al. also concluded that the smoker group had a higher risk for TB mortality and treatment failure in Malaysia [27]. Using the same smoking definition as Khan AH et al., our data noted a modest increased risk for TB in smokers than non-smokers (35.5% vs 17.3%, univariable $p = 0.002$, multivariable $p = 0.001$).

doi:10.6342/NTU202301094

Despite no outcome data being noted in this study, smoking cessation programs should be provided to all TB patients to prevent treatment failure.

The high incidence of TB in patients with cirrhosis has been described in many previous studies. A cohort study from Denmark (1977-1993) noted an incidence of TB in patients with liver cirrhosis of 168.6 per 100,000 person-years, which was higher than 8 per 1,000,000 person-years in the general population [39]. Two other studies from India also showed a 5-15 times higher prevalence rate of TB in the liver cirrhosis population than in the general population [40, 41]. Patients with liver cirrhosis may have reticuloendothelial system dysfunction and become more susceptible to TB and extra-pulmonary TB [42, 43]. When facing a patient with liver cirrhosis, TB should always be considered as a possible diagnosis.

A high burden of diabetes mellitus (DM) among TB patients has been reported, with a global median prevalence of 16% [44]. In addition, DM is considered a risk factor for TB with a two- to four-fold increased risk of active TB compared to non-DM patients [45]. Diabetes patients show significantly reduced interferon-γ production, which is an essential step in inhibiting the growth of *M. tuberculosis* [46]. In addition, lower expression of interferon-γ has been found to be associated with higher *M. tuberculosis* loads in diabetic mice compared to control mice [47]. The impaired innate immune response in DM patients is a known risk factor for increasing the susceptibility to M. tuberculosis infection [48]. In our study, both the TB and control groups had a relatively high proportion of DM patients (26.7% vs 21.1%, p=0.427). The enrollment of control cases from our medical outpatient clinics, rather than the general population, provided an explanation. According to the surveillance by the Health Promotion Administration in Taiwan from 2017 to 2020, the prevalence of diabetes was only 11% in Taiwan [49]. In this study, 21.1% of control cases had DM, which explains why our study found only a higher proportion of DM in TB patients (26.9%), but not a significant risk factor

doi:10.6342/NTU202301094

for TB.

Some limitations should be mentioned. First, the participants in this study were enrolled from a single center. The relatively small sample size prevented us from finding a dose-dependent relationship between 25(OH)D levels and the clinical manifestations of TB. Second, we did not enroll HIV-positive cases, and no TB patients had MDR-TB. Therefore, the conclusions cannot be extrapolated to HIV-positive and MDR-TB patients. Third, most of the subjects in the control group (60%) came from a cardiovascular clinic. However, currently, no evidence shows that cardiovascular diseases are associated with high 25(OH)D levels that would result in bias in our study [50, 51]. Fourth, we did not obtain the dietary history of participants, a factor that is difficult to quantify but is also important to the status of 25(OH)D.

In conclusion, this high-powered, 1:4 sex- and age-matched case-control study found that, compared to control individuals, TB patients had significantly lower 25(OH)D levels. Particularly, VDD (<20 ng/ml) is more prevalent among TB patients. Furthermore, our data suggest that lower BMI, smoking, liver cirrhosis, and 25(OH)D levels <20 ng/ml are independent risk factors for TB in non-HIV patients in Taiwan, at a geographic location of latitude 24°N.

doi:10.6342/NTU202301094

Reference

1. WHO. Global tuberculosis report 2022. Available from: https://wwwwhoint/teams/global-tuberculosis-programme/tb-reports/global-tuberculosis-report-2022.
2. Jilani TN AA, Zafar Gondal A, et al. Active Tuberculosis. Available from: https://wwwncbinlmnihgov/books/NBK513246/. Treasure Island (FL): StatPearls Publishing; 2023 Jan-.
3. Chocano-Bedoya P, Ronnenberg AG. Vitamin D and tuberculosis. Nutrition reviews. 2009;67(5):289-93. Epub 2009/04/24. doi: 10.1111/j.1753-4887.2009.00195.x. PubMed PMID: 19386033.
4. Fernando SL, Saunders BM, Sluyter R, Skarratt KK, Goldberg H, Marks GB, et al. A polymorphism in the P2X7 gene increases susceptibility to extrapulmonary tuberculosis. American journal of respiratory and critical care medicine. 2007;175(4):360-6. Epub 2006/11/11. doi: 10.1164/rccm.200607-970OC. PubMed PMID: 17095747.
5. Babb C, van der Merwe L, Beyers N, Pheiffer C, Walzl G, Duncan K, et al. Vitamin D receptor gene polymorphisms and sputum conversion time in pulmonary tuberculosis patients. Tuberculosis (Edinburgh, Scotland). 2007;87(4):295-302. Epub 2007/04/24. doi: 10.1016/j.tube.2007.03.001. PubMed PMID: 17449323.
6. Bellamy R, Ruwende C, Corrah T, McAdam KP, Thursz M, Whittle HC, et al. Tuberculosis and chronic hepatitis B virus infection in Africans and variation in the vitamin D receptor gene. The Journal of infectious diseases. 1999;179(3):721-4. Epub 1999/02/10. doi: 10.1086/314614. PubMed PMID: 9952386.
7. Lewis SJ, Baker I, Davey Smith G. Meta-analysis of vitamin D receptor polymorphisms and pulmonary tuberculosis risk. The international journal of tuberculosis and lung disease : the official journal of the International Union against Tuberculosis and Lung Disease. 2005;9(10):1174-7. Epub 2005/10/19. PubMed PMID: 16229232.
8. Selvaraj P, Chandra G, Jawahar MS, Rani MV, Rajeshwari DN, Narayanan PR. Regulatory role of vitamin D receptor gene variants of Bsm I, Apa I, Taq I, and Fok I polymorphisms on macrophage phagocytosis and lymphoproliferative response to mycobacterium tuberculosis antigen in pulmonary tuberculosis. Journal of clinical immunology. 2004;24(5):523-32. Epub 2004/09/11. doi: 10.1023/b:Joci.0000040923.07879.31. PubMed PMID: 15359111.
9. Wilbur AK, Kubatko LS, Hurtado AM, Hill KR, Stone AC. Vitamin D receptor gene polymorphisms and susceptibility M. tuberculosis in native Paraguayans. Tuberculosis (Edinburgh, Scotland). 2007;87(4):329-37. Epub 2007/03/06. doi: 10.1016/j.tube.2007.01.001. PubMed PMID: 17337247.
10. Pfotenhauer KM, Shubrook JH. Vitamin D Deficiency, Its Role in Health and Disease, and Current Supplementation Recommendations. The Journal of the American Osteopathic Association. 2017;117(5):301-5. doi: 10.7556/jaoa.2017.055. PubMed PMID: 28459478.
11. Christakos S, Ajibade DV, Dhawan P, Fechner AJ, Mady LJ. Vitamin D: metabolism. Endocrinology and metabolism clinics of North America. 2010;39(2):243-53, table of contents. doi: 10.1016/j.ecl.2010.02.002. PubMed PMID: 20511049; PubMed Central PMCID: PMC2879391.
12. Chun RF, Liu PT, Modlin RL, Adams JS, Hewison M. Impact of vitamin D on immune function: lessons learned from genome-wide analysis. Front Physiol. 2014;5:151. Epub 2014/05/06. doi: 10.3389/fphys.2014.00151. PubMed PMID: 24795646; PubMed Central PMCID: PMCPMC4000998.
13. Liu PT, Stenger S, Tang DH, Modlin RL. Cutting edge: vitamin D-mediated human antimicrobial activity against Mycobacterium tuberculosis is dependent on the induction of

doi:10.6342/NTU202301094

cathelicidin. J Immunol. 2007;179(4):2060-3. Epub 2007/08/07. doi: 10.4049/jimmunol.179.4.2060. PubMed PMID: 17675463.
14. Liu PT, Stenger S, Li H, Wenzel L, Tan BH, Krutzik SR, et al. Toll-like receptor triggering of a vitamin D-mediated human antimicrobial response. Science. 2006;311(5768):1770-3. doi: 10.1126/science.1123933. PubMed PMID: 16497887.
15. Zeng J, Wu G, Yang W, Gu X, Liang W, Yao Y, et al. A serum vitamin D level <25nmol/l pose high tuberculosis risk: a meta-analysis. PloS one. 2015;10(5):e0126014. Epub 2015/05/06. doi: 10.1371/journal.pone.0126014. PubMed PMID: 25938683; PubMed Central PMCID: PMCPMC4418705.
16. Huang SJ, Wang XH, Liu ZD, Cao WL, Han Y, Ma AG, et al. Vitamin D deficiency and the risk of tuberculosis: a meta-analysis. Drug design, development and therapy. 2017;11:91-102. Epub 2017/01/18. doi: 10.2147/dddt.S79870. PubMed PMID: 28096657; PubMed Central PMCID: PMCPMC5207333.
17. Aibana O, Huang C-C, Aboud S, Arnedo-Pena A, Becerra MC, Bellido-Blasco JB, et al. Vitamin D status and risk of incident tuberculosis disease: A nested case-control study, systematic review, and individual-participant data meta-analysis. PLOS Medicine. 2019;16(9):e1002907. doi: 10.1371/journal.pmed.1002907.
18. Kafle S, Basnet AK, Karki K, Thapa Magar M, Shrestha S, Yadav RS. Association of Vitamin D Deficiency With Pulmonary Tuberculosis: A Systematic Review and Meta-Analysis. Cureus. 2021;13(9):e17883. Epub 2021/10/19. doi: 10.7759/cureus.17883. PubMed PMID: 34660082; PubMed Central PMCID: PMCPMC8504877.
19. Grange JM, Davies PD, Brown RC, Woodhead JS, Kardjito T. A study of vitamin D levels in Indonesian patients with untreated pulmonary tuberculosis. Tubercle. 1985;66(3):187-91. Epub 1985/09/01. doi: 10.1016/0041-3879(85)90035-2. PubMed PMID: 4049530.
20. Chan TY, Poon P, Pang J, Swaminathan R, Chan CH, Nisar M, et al. A study of calcium and vitamin D metabolism in Chinese patients with pulmonary tuberculosis. The Journal of tropical medicine and hygiene. 1994;97(1):26-30. Epub 1994/02/01. PubMed PMID: 8107170.
21. Wejse C, Olesen R, Rabna P, Kaestel P, Gustafson P, Aaby P, et al. Serum 25-hydroxyvitamin D in a West African population of tuberculosis patients and unmatched healthy controls. The American journal of clinical nutrition. 2007;86(5):1376-83. Epub 2007/11/10. doi: 10.1093/ajcn/86.5.1376. PubMed PMID: 17991649.
22. Koo HK, Lee JS, Jeong YJ, Choi SM, Kang HJ, Lim HJ, et al. Vitamin D deficiency and changes in serum vitamin D levels with treatment among tuberculosis patients in South Korea. Respirology (Carlton, Vic). 2012;17(5):808-13. Epub 2012/03/28. doi: 10.1111/j.1440-1843.2012.02172.x. PubMed PMID: 22449254.
23. Sita-Lumsden A, Lapthorn G, Swaminathan R, Milburn HJ. Reactivation of tuberculosis and vitamin D deficiency: the contribution of diet and exposure to sunlight. Thorax. 2007;62(11):1003-7. Epub 2007/05/29. doi: 10.1136/thx.2006.070060. PubMed PMID: 17526677; PubMed Central PMCID: PMCPMC2117124.
24. Martineau AR, Nhamoyebonde S, Oni T, Rangaka MX, Marais S, Bangani N, et al. Reciprocal seasonal variation in vitamin D status and tuberculosis notifications in Cape Town, South Africa. Proceedings of the National Academy of Sciences of the United States of America. 2011;108(47):19013-7. Epub 2011/10/26. doi: 10.1073/pnas.1111825108. PubMed PMID: 22025704; PubMed Central PMCID: PMCPMC3223428.
25. Iftikhar R, Kamran SM, Qadir A, Haider E, Bin Usman H. Vitamin D deficiency in patients with tuberculosis. Journal of the College of Physicians and Surgeons--Pakistan : JCPSP. 2013;23(10):780-3. Epub 2013/10/31. PubMed PMID: 24169384.

doi:10.6342/NTU202301094

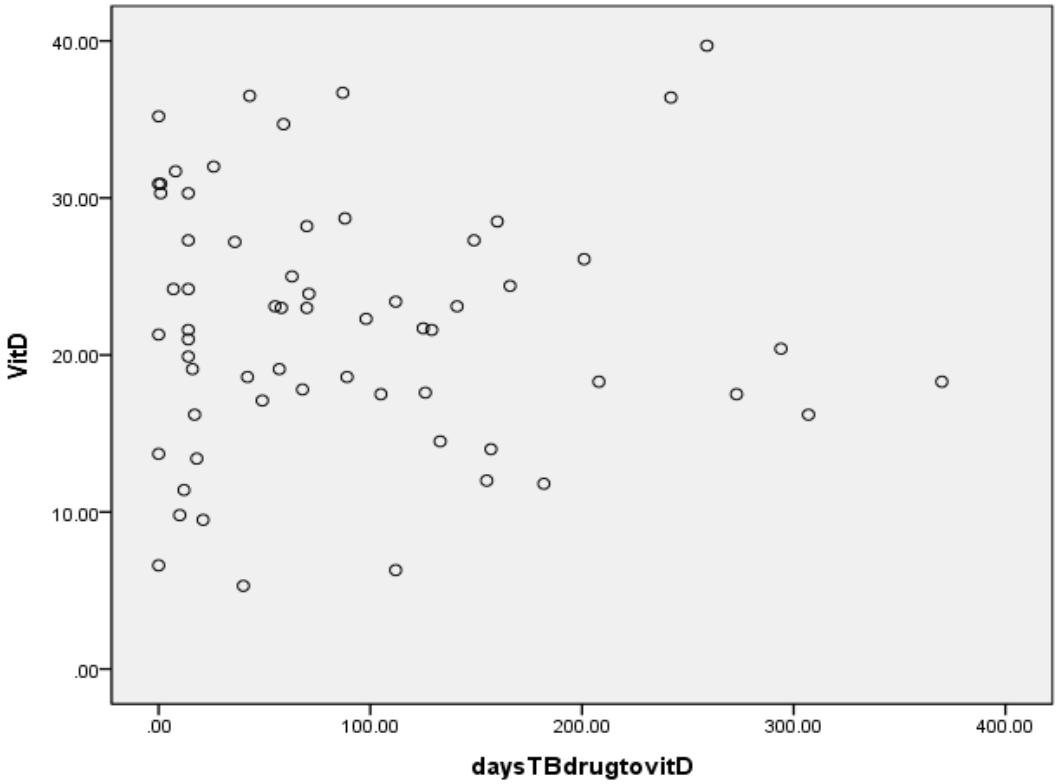
26. Joshi L, Ponnana M, Penmetsa SR, Nallari P, Valluri V, Gaddam S. Serum vitamin D levels and VDR polymorphisms (BsmI and FokI) in patients and their household contacts susceptible to tuberculosis. Scandinavian journal of immunology. 2014;79(2):113-9. Epub 2013/11/14. doi: 10.1111/sji.12127. PubMed PMID: 24219580.
27. Khan AH, Sulaiman SAS, Hassali MA, Khan KU, Ming LC, Mateen O, et al. Effect of smoking on treatment outcome among tuberculosis patients in Malaysia; a multicenter study. BMC Public Health. 2020;20(1):854. Epub 2020/06/06. doi: 10.1186/s12889-020-08856-6. PubMed PMID: 32498682; PubMed Central PMCID: PMCPMC7273674.
28. Revised international definitions in tuberculosis control. The international journal of tuberculosis and lung disease : the official journal of the International Union against Tuberculosis and Lung Disease. 2001;5(3):213-5. Epub 2001/05/01. PubMed PMID: 11326818.
29. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. The Journal of clinical endocrinology and metabolism. 2011;96(7):1911-30. doi: 10.1210/jc.2011-0385. PubMed PMID: 21646368.
30. Hilger J, Friedel A, Herr R, Rausch T, Roos F, Wahl DA, et al. A systematic review of vitamin D status in populations worldwide. The British journal of nutrition. 2014;111(1):23-45. Epub 2013/08/13. doi: 10.1017/s0007114513001840. PubMed PMID: 23930771.
31. Palacios C, Gonzalez L. Is vitamin D deficiency a major global public health problem? The Journal of steroid biochemistry and molecular biology. 2014;144 Pt A:138-45. Epub 2013/11/19. doi: 10.1016/j.jsbmb.2013.11.003. PubMed PMID: 24239505; PubMed Central PMCID: PMCPMC4018438.
32. Jaimni V, Shasty BA, Madhyastha SP, Shetty GV, Acharya RV, Bekur R, et al. Association of Vitamin D Deficiency and Newly Diagnosed Pulmonary Tuberculosis. Pulmonary Medicine. 2021;2021:5285841. doi: 10.1155/2021/5285841.
33. Sinha P, Davis J, Saag L, Wanke C, Salgame P, Mesick J, et al. Undernutrition and Tuberculosis: Public Health Implications. The Journal of infectious diseases. 2019;219(9):1356-63. doi: 10.1093/infdis/jiy675. PubMed PMID: 30476125; PubMed Central PMCID: PMC6941617.
34. Workineh M, Mathewos B, Moges B, Gize A, Getie S, Stendahl O, et al. Vitamin D deficiency among newly diagnosed tuberculosis patients and their household contacts: a comparative cross-sectional study. Archives of public health = Archives belges de sante publique. 2017;75:25. Epub 2017/06/24. doi: 10.1186/s13690-017-0195-7. PubMed PMID: 28638616; PubMed Central PMCID: PMCPMC5474861.
35. Hong JY, Kim SY, Chung KS, Kim EY, Jung JY, Park MS, et al. Association between vitamin D deficiency and tuberculosis in a Korean population. The international journal of tuberculosis and lung disease : the official journal of the International Union against Tuberculosis and Lung Disease. 2014;18(1):73-8. Epub 2013/12/25. doi: 10.5588/ijtld.13.0536. PubMed PMID: 24365556.
36. Bueno H, Bernal JL, Jimenez-Jimenez V, Martin-Sanchez FJ, Rossello X, Moreno G, et al. The Clinical outcomes, healthcare resource utilization, and related costs (COHERENT) model. Application in heart failure patients. Revista espanola de cardiologia. 2022;75(7):585-94. doi: 10.1016/j.rec.2021.08.009. PubMed PMID: 34688580.
37. Bates MN, Khalakdina A, Pai M, Chang L, Lessa F, Smith KR. Risk of tuberculosis from exposure to tobacco smoke: a systematic review and meta-analysis. Archives of internal medicine. 2007;167(4):335-42. doi: 10.1001/archinte.167.4.335. PubMed PMID: 17325294.
38. Sitas F, Urban M, Bradshaw D, Kielkowski D, Bah S, Peto R. Tobacco attributable deaths in South Africa. Tobacco control. 2004;13(4):396-9. doi: 10.1136/tc.2004.007682. PubMed PMID: 15564624; PubMed Central PMCID: PMC1747967.

doi:10.6342/NTU202301094

39. Thulstrup AM, Mølle I, Svendsen N, Sørensen HT. Incidence and prognosis of tuberculosis in patients with cirrhosis of the liver. A Danish nationwide population based study. Epidemiology and infection. 2000;124(2):221-5. Epub 2000/05/17. doi: 10.1017/s0950268899003593. PubMed PMID: 10813146; PubMed Central PMCID: PMCPMC2810904.
40. Baijal R, Praveenkumar HR, Amarapurkar DN, Nagaraj K, Jain M. Prevalence of tuberculosis in patients with cirrhosis of liver in western India. Tropical doctor. 2010;40(3):163-4. Epub 2010/05/19. doi: 10.1258/td.2010.090463. PubMed PMID: 20478985.
41. Saigal S, Nandeesh H, Agarwal S, Misra A, Jain S, Sarin S. High prevalence and profile of tuberculosis in chronic liver disease patients. Gastroenterology. 1998;(114):A38.
42. Bonnel AR, Bunchorntavakul C, Reddy KR. Immune dysfunction and infections in patients with cirrhosis. Clinical gastroenterology and hepatology : the official clinical practice journal of the American Gastroenterological Association. 2011;9(9):727-38. Epub 2011/03/15. doi: 10.1016/j.cgh.2011.02.031. PubMed PMID: 21397731.
43. Cho YJ, Lee SM, Yoo CG, Kim YW, Han SK, Shim YS, et al. Clinical characteristics of tuberculosis in patients with liver cirrhosis. Respirology (Carlton, Vic). 2007;12(3):401-5. Epub 2007/06/02. doi: 10.1111/j.1440-1843.2007.01069.x. PubMed PMID: 17539845.
44. Workneh MH, Bjune GA, Yimer SA. Prevalence and associated factors of tuberculosis and diabetes mellitus comorbidity: A systematic review. PloS one. 2017;12(4):e0175925. Epub 2017/04/22. doi: 10.1371/journal.pone.0175925. PubMed PMID: 28430796; PubMed Central PMCID: PMCPMC5400500.
45. Al-Rifai RH, Pearson F, Critchley JA, Abu-Raddad LJ. Association between diabetes mellitus and active tuberculosis: A systematic review and meta-analysis. PloS one. 2017;12(11):e0187967. Epub 2017/11/22. doi: 10.1371/journal.pone.0187967. PubMed PMID: 29161276; PubMed Central PMCID: PMCPMC5697825.
46. Stalenhoef JE, Alisjahbana B, Nelwan EJ, van der Ven-Jongekrijg J, Ottenhoff TH, van der Meer JW, et al. The role of interferon-gamma in the increased tuberculosis risk in type 2 diabetes mellitus. European journal of clinical microbiology & infectious diseases : official publication of the European Society of Clinical Microbiology. 2008;27(2):97-103. doi: 10.1007/s10096-007-0395-0. PubMed PMID: 17962984.
47. Yamashiro S, Kawakami K, Uezu K, Kinjo T, Miyagi K, Nakamura K, et al. Lower expression of Th1-related cytokines and inducible nitric oxide synthase in mice with streptozotocin-induced diabetes mellitus infected with Mycobacterium tuberculosis. Clinical and experimental immunology. 2005;139(1):57-64. doi: 10.1111/j.1365-2249.2005.02677.x. PubMed PMID: 15606614; PubMed Central PMCID: PMC1809276.
48. Kumar Nathella P, Babu S. Influence of diabetes mellitus on immunity to human tuberculosis. Immunology. 2017;152(1):13-24. doi: 10.1111/imm.12762. PubMed PMID: 28543817; PubMed Central PMCID: PMC5543489.
49. Taiwan Ministry of Health Welfare. National Health Insurance Expenditure Statistics Report (2018). Available from: https://www.nhi.gov.tw/Nhi_E-LibraryPubWeb/Periodical/P_Detail.aspx?CPT_TypeID=5&CP_ID=234.
50. Danik JS, Manson JE. Vitamin d and cardiovascular disease. Current treatment options in cardiovascular medicine. 2012;14(4):414-24. Epub 2012/06/13. doi: 10.1007/s11936-012-0183-8. PubMed PMID: 22689009; PubMed Central PMCID: PMCPMC3449318.
51. Cosentino N, Campodonico J, Milazzo V, De Metrio M, Brambilla M, Camera M, et al. Vitamin D and Cardiovascular Disease: Current Evidence and Future Perspectives. Nutrients. 2021;13(10). Epub 2021/10/24. doi: 10.3390/nu13103603. PubMed PMID: 34684604; PubMed Central PMCID: PMCPMC8541123.

doi:10.6342/NTU202301094

Supplement 1.
Duration of TB treatment and vit D concentration.



The correlation between the vitamin D concentration and the duration of anti-TB treatment show low negative association but not significant due to small sample size.
(r=-0.013, p=0.92)

doi:10.6342/NTU202301094

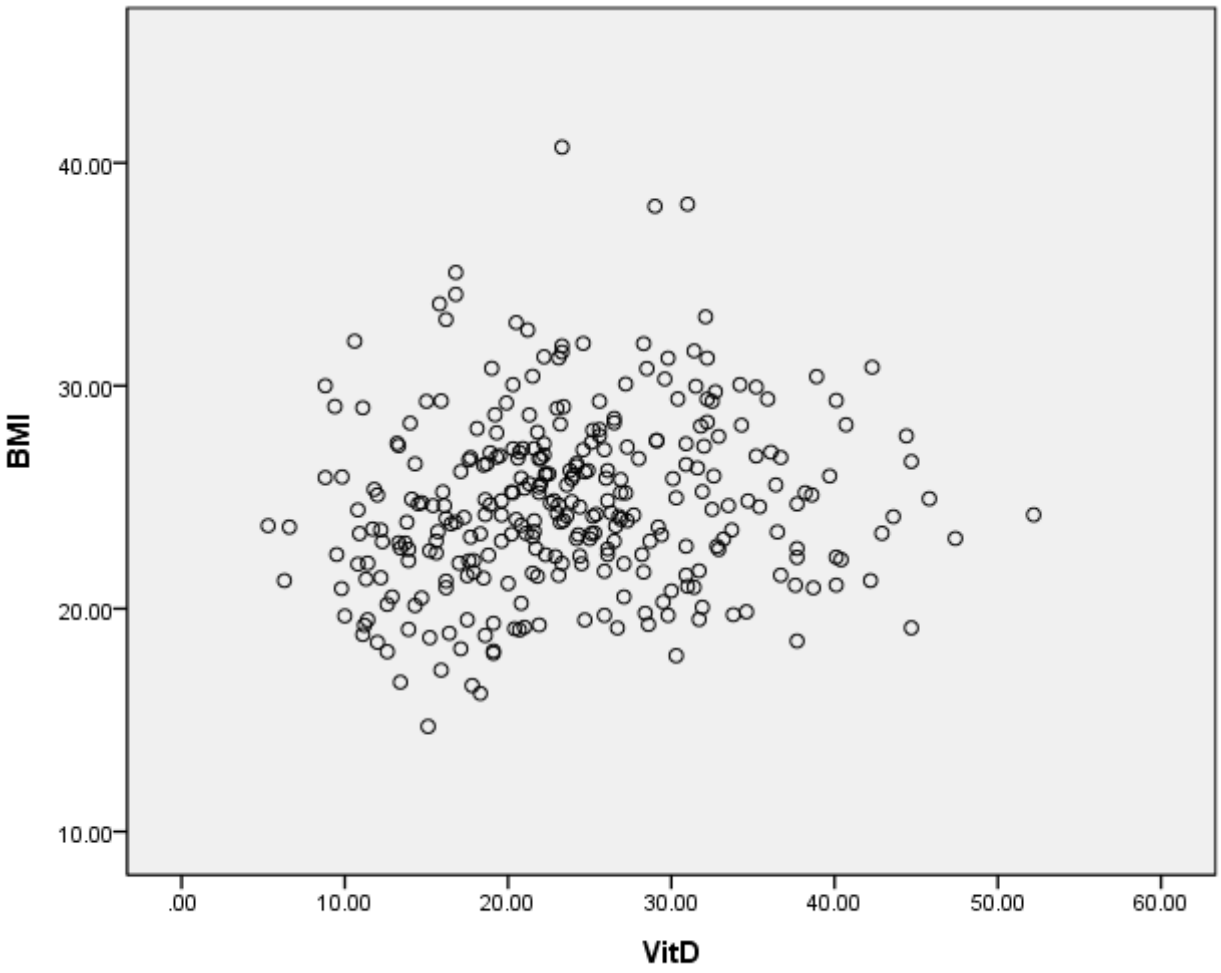
Supplement 2.
Multivariable analysis of sex- and age-matched TB and control cases with the items of absolute 25(OH)D concentration (ng/mL), body mass index, liver cirrhosis, smoking, alcohol use disorders in the model

	p-value	Adjusted odd ratio	95% confidence interval
25(OH)D concentration (ng/mL)	0.008	0.948	0.911~0.986
Body mass index	<0.001	0.790	0.725~0.905
Liver cirrhosis	0.018	11.368	1.513~85.408
Smoking	0.001	4.237	1.77~10.143
Alcohol use behavior	0.101	10.482	0.632~173.95

doi:10.6342/NTU202301094

Supplement 3.

The association of BMI and vitamin D concentration showed a significant low correlation (r=0.133, p=0.019)



doi:10.6342/NTU202301094

Supplement 4.
Multivariable analysis of sex- and age-matched TB and control cases with the items of 4 categories of 25(OH)D concentration (<10ng/ml, 10-20ng/ml, 20-30ng/ml, >30ng/ml), body mass index, liver cirrhosis, smoking, alcohol use disorders in the model

	p-value	Adjusted odd ratio	95% confidence interval
25(OH)D concentration (<10 ng/mL)	0.026	**3.741**	1.169~11.968
25(OH)D concentration (10~20 ng/mL)	0.180	**1.634**	0.797~3.352
25(OH)D concentration (10~30 ng/mL)	0.888	**1.056**	0.493~2.266
25(OH)D concentration (>30 ng/mL)	0.074	-	-
Body mass index	<0.001	0.867	0.801~0.939
Liver cirrhosis	0.027	4.137	1.174~14.577
Smoking	0.002	2.328	1.370~3.957
Alcohol use behavior	0.209	2.169	0.649~7.253

doi:10.6342/NTU202301094

亞東紀念醫院

人體試驗/研究案件 計畫書

一、計畫中文名稱：維生素 C, 維生素 B1 及類固醇做為嚴重敗血症及敗血性休克病人免疫調節的治療成效: 一個隨機對照研究
二、主要主持人：許孟璇
三、協同/共同主持人：張厚台, 沈士雄
四、研究背景說明：(包括學理根據及有關文獻報告，文獻不需全文印出，列於第十二點即可)
儘管醫療的進步，嚴重敗血症和敗血性休克仍然是加護病房 (ICU) 死亡率的主要原因[1-3]。根據流行病學調查，世界各國的嚴重敗血症死亡率約在 20％～60％之間[4-6]。除了死亡外，嚴重的敗血症和感染性休克也會導致一些長期或短期的嚴重併發症，如腎衰竭或呼吸衰竭，導致需要長期血液透析或呼吸機依賴。在台灣，北部某醫學中心的 SICU (外科重症加護病房) 中，感染性休克患者的 28 天死亡率約為 65％。另一個在 2006 年的研究還顯示多器官功能衰竭發生率為 27.6％，住院死亡率為 30.8％[7]。
嚴重敗血症的治療有三個部分：感染控制，血液動力學穩定和調節敗血症反應。先前許多臨床試驗中，嚐試阻斷炎症連鎖反應，如利用皮質類固醇，抗內毒素抗體，腫瘤壞死因子 (TNF) 拮抗劑，白細胞介素-1 受體拮抗劑，等等，到目前為止沒有任何一個藥物被證明有效的 [8,9]。因此，一種安全，有效，可用的治療方法是我們目前最迫切需要。
硫胺素 (Thiamin, vitamin B1)是丙酮酸脫氫酶(pyruvate dehydrogenase),α-酮戊二酸脫氫酶 (alpha-ketoglutarate dehydrogenase) 和轉酮醇酶 (transketolase)的關鍵輔因子。這三種酶在克雷伯氏循環(Krebs Cycle)都是必需要的，以完成有氧呼吸，防止乳酸的產生。以前的研究發現，硫胺素缺乏的現象在感染性休克和其他重症中是普遍存在的[10-12]。一項前鋒性研究也證實，在硫胺素缺乏的敗血性休克患者，在使用硫胺素 24 小時後，血液中的乳酸值會顯著降低[13]。
HYPRESS (氫化可體松預防敗血性休克) 研究未能證實敗血性休克患者輸注氫化可體松的效果[14]。維生素 C 是一種有效的抗氧化劑，可以直接清除氧自由基，還可以恢復其他細胞抗氧化劑，並在維持內皮功能和微循環流動方面發揮作用[15,16]。儘管以前的研究證明氫化可體松和維生素 C 單獨使用時，對敗血性休克患者的臨床結果影響不大[17,18]。維生素 C 和氫化可體松在敗血性休克中有許多重疊和協同的病理生理效應。這兩種藥物都是兒茶酚胺合成所必需的，並且可以增加血管加壓藥的敏感性[15,19,20]。這兩種藥物都可以下調促炎介質的產生，增加內皮細胞和上皮細胞之間的緊密連接，保持內皮功能和微循環血流[21-23]。 **Marik 等在 *CHEST*** 上發表了他們的研究 (2017 年 6 月)，結果發現維生素 B1，維生素 C 和氫化可體松對嚴重敗血症和敗血症性休克的益處[24]。然而，由於本研究的樣本量小和一些研究上的偏差可能會混淆研究結果[25]。在此，我們希望透過隨機對照試驗來了解「維生素 C，維生素 B1 和氫化可體松」在嚴重敗血症和敗血症性休克患者中的協同調節作用，以調節病患因敗血症產生的免疫反應。

研究終點

主要終點是出院的存活率。次要終點包括血管加壓藥治療的持續時間，急性腎損傷 (AKI) 患者腎臟出現洗腎治療的需求及維持的時間，ICU 住院時間 (LOS) 以及 72 小時內 APACH 評分和 SOFA 評分的變化。

doi:10.6342/NTU202301094

五、研究目的：

透過隨機對照試驗來了解「維生素 C，維生素 B1 和氫化可體松」在嚴重敗血症和敗血症性休克患者中的協同調節作用，以調節病患因敗血症產生的免疫反應。

六、研究方法：

(一)選擇標準與人數。

(a)受試者收案數：亞東醫院，共收案 80 人

(b)納入條件：年齡相等及大於 20 歲以上，預期壽命超過 3 天以上，有嚴重敗血症和敗血症性休克的患者來院(急診或病房)，經病患或家屬同意後，將會隨機分配到對照組或研究組，比例為1：1。隨機方式則使用亂數產生器(https://lab.25sprout.com/nrprnd/)，自 1 到 80 隨機產生一個數字，凡遇單數則為研究組 (A-B-C group)，雙數則為對照組 (placebo group)。根據 **Surviving Sepsis Campaign: International Guidelines for Management of Sepsis and Septic Shock: 2016**。嚴重敗血症和敗血症性休克之收案定義 (詳見個案報告表見附件) 。

(1) 嚴重敗血症(含以下兩個或以上條件):

包括: 發燒 (口腔溫度> 38°C [> 100.4°F]) 或體溫過低 (<36°C)，呼吸急促 (> 24 次呼吸/分鐘)，心動過速 (心率> 90 次/分)，白血球增多 (> 12,000 /μL) 或白血球減少 (<4000 /μL) 或> 10% band form 並且 SOFA score 為 2 分或以上

(2) 敗血症性休克

在足夠灌流情形下，動脈收縮壓 ABP 仍<90 mmHg 至少一小時，或需要血管加壓藥來維持收縮壓 ≥90 mmHg (or MBP>=70)

(c)排除條件：懷孕的患者，已知維生素 C，維生素 B 或氫化可體松 (或其他等效產品) 過敏的患者將被排除在外。其他，如: 已是慢性植物人狀態、預期壽命小於 28 天，已經長期接受血液透析或腹膜透析，接受器官移植後長期使用免疫抑制劑者，在使用 ECMO，嚴重頭部外傷或腦出血，重度心衰竭(NYHA class III-IV)或 6 星期內發生過急性心肌梗塞，四週內曾被 CPR 過，已診斷為 HIV/AIDS 之病患，體重超過 150kg，三級以上燒燙傷，因手術併發症所造成的休克，已經簽署"不急救同意書"之病患，也將排除在外。

(d)招募方式：由主持人或協同主持人協同研究護理師在亞東醫院急診或病房，直接向受試者或其法定代理人說明，以取得同意。

(e)研究對象同意之方式：患者或患者家屬或法定監護人需簽立「同意書」。

(二)研究設計：(若為問卷研究，應註明問卷回收方式；若為病歷回溯應註明回溯期間)

(a) 研究進行方式：本實驗為一個開放性試驗。**研究組** (維生素 C-維生素 B1-氫化可體松) 患者將接受靜脈注射硫胺素 (200mg，溶於 0.9%生理鹽水中，並且每 12 小時輸注一次，持續 4 天或入住 ICU< 4 天者，則持續治療至離開 ICU)，維生素 C (1.5g 混合於 100-mL，0.9%生理鹽水溶液中，並且每 6 小時輸注一次，持續 4 天或直至 ICU 出院)，合併氫化可體松每 6 小時 50mg (或其他同等產品)，持續 4 天後，得由臨床醫師依敗血症治療常規予以調整劑量[24]。在**對照組**中，患者之治療方式將由主治醫師依敗血症治療常規予以治療。

(b) 受試者之追蹤(含安全性監視指標)及退出條件：追蹤病患至出院。住院期間，將追蹤病患的血壓(包括血管加壓藥治療的持續時間)，腎臟功能(包括是否出現洗腎治療的需求)，住院時以及 96 小時內 APACH 評分和 SOFA 評分的變化。根據 Surviving Sepsis

doi:10.6342/NTU202301094

Campaign: International Guidelines for Management of Sepsis and Septic Shock: 2016。研究期間可併用升壓藥物，抗生素，壓力性潰瘍預防藥物等，其他經主治醫師評估後，認為所需的治療，如 continuous renal replacement therapy 或 ECMO。但禁止併用其他免疫調節藥物，例如：calcineurin inhibitor，Cyclosporine，Azathioprine、Mycophenolate，高/低劑量糖皮質類固醇，單株抗體－標靶製劑，如 Daclizumab 等等。

(b)-1 研究期間，受試者若撤回同意書，發生嚴重藥物不良反應（例如，對研究藥物過敏反應），或在研究期間經主治醫師判斷，需使用額外使用口服或針劑的維生素 C 或維生素 B1，或其他上述禁止併用之藥物（例如：calcineurin inhibitor，Cyclosporine，Azathioprine、Mycophenolate，高/低劑量糖皮質類固醇，單株抗體－標靶製劑，如 Daclizumab 等等），則需退出本研究。

(b)-2 藥物可能導致之副作用及其處理方式:

- Vitamin B1 (硫胺素): 是水溶性維生素，在大劑量使用時，可能會出現過敏性休克。多餘的分量完全排出體外，不會貯留在人體中。若有過敏現象，將立即停藥，並給予抗組織胺及/或類固醇。
- Vitamin C (抗壞血酸): 在大劑量下會發生下痢等現象。將給予止瀉藥處理
- Hydrocortisone (氫化可體松(類固醇)): 短期內血糖上升。將檢測血糖，並給予適當劑量的降血糖藥物控制。

我們將追蹤藥物之副作用從開使用研究藥物起直到(第一劑藥物後)14 天。

(c) 血液採樣和數據收集

收集患者的臨床資訊，包括年齡，性別，潛在合併症，體重指數（BMI），嚴重敗血症和敗血症性休克原因的初步診斷，呼吸器的需求，血管加壓藥，腎臟替代治療，經驗性和治療性抗生素(種類及時間)，血糖、白蛋白、制酸劑之使用以及使用之管路等等。在實驗室數據則包括，在**收案時**和**第 4 天**的 **WBC 數，乳酸值，肌酸酐和血液培養結果，血清中維生素 C，維生素 B1 值和皮質類固醇值**。

每次抽血量約 6cc。3cc 置於紫頭採血管，將先貼上病患之研究編碼，立即放置於-80℃冰箱;另 3cc 置於綠頭採血管，經初步離心後，取上清液，以鋁箔紙包裹避光並貼上病患之研究編碼，放置於-80℃冰箱進行保存。在 3~6 個月內，將檢體放入檢體盒(內含冰寶，以維持 4℃)中以宅急配送到淡水馬偕醫院遺傳醫學部。我們也記錄病患的住院和住 ICU 天數和預後結果。

doi:10.6342/NTU202301094

Study calendar

procedure	Screening/ baseline	Cycle 1				Post-therapy follow-up
		Day 1	Day 2	Day 3	Day 4	Day 5~14
DM, ESRD, COPD, LC CHF, steroid use,smoking Malignancy, HIV/AIDS…						
BT/RR/BP						
SOFA, APACH score						
WBC, Hb, Cr, sugar, alb, Cortisone, ACTH, lactic acid						
Vit C, Vit B1 drug conc.						
Concomitant medications (antacid agent, abx)						
Septic workup						
Adverse effect (allergy, diarrhea, high blood sugar)						
Response evaluation (need and duration of H/D, inotropic agent, ICU staying duration, Survival, length of hospital staying)						

doi:10.6342/NTU202301094

(d) 治療效果評估及統計分析方法：**主要終點**是出院的存活率。**次要終點**包括血管加壓藥治療的持續時間，急性腎損傷（AKI）患者腎臟出現洗腎治療的需求，ICU 住院時間（LOS）以及 72 小時內 APACH 評分和 SOFA 評分的變化。

統計分析方法

根據之前 Marik 等在 CHEST（2017 年 6 月）上所發表的研究顯示可減少嚴重敗血症及敗血症性休克 32%的死亡率。因此，本實驗欲達到 80%檢力，以及過去五年本院內科加護病房的平均死亡率為 40%的前提下，若真能減少 30% 死已率，則目前預估每組所需人數為 40 人(含 10%的 drop-out)。在第一階段(2019/01~2020/12)收案完成後，會請統計專家審查，並決定是否進行第二階段的收案。

進入實驗之受試者，將以 stratified permuted block randomization:先根據病人依 APACH II score(內已含年齡變項)進行分層: APACH II score ≧ 15 及< 15 兩層。We set a block of 4, 2 patients in a block would be assigned to the control and 2 to the ABC group and the ordering of those 4 assignments would be randomized by random number table. 受試者如使用至少一次藥物者，將進入 modified intent-to-treat (mITT) 分析; 若完成所有研究藥物者將被納入 per-protocol 的分析。

Statistical analysis

Statistical analyses was performed using SPSS 19.0 for Windows (SPSS, Chicago, Illinois). Statistical significance of continuous variables was determined using a non-parametric test (Mann-Whitney U test) and categorical variables was analyzed using Fisher's exact test. All tests were two-tailed and a value of $p < 0.05$ was considered statistically significant. Continuous variables were reported as mean ± standard deviation (SD). A logistic regression model was used for univariate and multivariate analyses to determine the independent risk factors of mortality. The variables included in the multivariate analysis were selected based on the results of the univariate analysis, previous study results, and possible biological association.

(e) 問卷或其他研究相關資料：**(**個案報告表見附件**)** 。

doi:10.6342/NTU202301094

（f）檢驗結束後，檢體將以同樣方式送回，放置於亞東醫院 6 樓的共同研究室檢體冰箱。

doi:10.6342/NTU202301094

(三)研究期限與進度：(應包含結果分析之時間)。

期限	完成之工作項目	工作人員預期可獲得之訓練	研究貢獻
2019/01~2020/12	Collect clinical data and do preliminary analysis	1. 收集臨床資料 2. analysis data	Preliminary results
2020/01/2022/12	Summarize the final results	1. 學習統計軟體的應用 2. 撰寫研究報告	Final results

(四)研究人力、工作職責及相關設備需求：(例如需有幾名研究助理或特殊儀器設備幾台)

類別	姓名	服務單位	職稱	在本計畫內擔任之工作性質、項目及範圍
計畫主持人	許孟璇	內科部 一般醫學內科	主任	研究設計、實驗討論、問題解決。 篩選個案，向病患/家屬解釋研究內容，並簽署同意書。 至研究藥局領藥 將檢體進行初步離心 完成 case report form。
協同主持人	張厚台	重症醫學部	主任	實驗討論、問題解決。向病患/家屬解釋研究內容，並簽署同意書。
協同主持人	沈士雄	急診醫學部	主治醫師	實驗討論、問題解決。向病患/家屬解釋研究內容，並簽署同意書。
研究人員	葉怡珍	內科部 一般醫學內科	研究護士	協助說明、簽署同意書 至研究藥局領藥 將離心後的檢體，以宅急配送至淡水馬階醫院
研究人員	吳美玉	感染管制中心	研究技術員	將檢體進行初步離心，以宅急配送至淡水馬階醫院

七、研究材料處理(請務必說明保存地點及可接觸之人員)

(一)紙本、電子資料/檢體於研究進行中之蒐集處理：不會無故病人個資攜出院外。原則以公務電腦執行業務，需使用個人電腦時，資料加密。

(二)紙本、電子資料/檢體於研究結束後之後續處理、保存方式、保存年限(應說明受試者身分是否以編碼識別或去連結方式，或如何隱去研究參與者身分之詳細作法)：研究結束後，紙本、電子資料/檢體，將以未去連結方式由計畫主持人/亞東紀念醫院保存 10 年。此紙本、電子資料/檢體僅計畫主持人許孟璇醫師及協同主持人張厚台醫師，沈士雄醫師及其授權的研究助理/技術員可使用。

(三) 檢體: 將放放置檢體盒中，保管於亞東醫院共研冰箱中。檢體盒鑰匙將由計畫主持人及研究助理保管。

八、受試者權益維護：

(一)可能引起的損害及其救濟措施：

(一)-1 可能引起的損害: Vitamin B1 (硫胺素): 是水溶性維生素，多餘的分量完全排出體外，不會貯留在人體中。在大劑量使用時，可能會出現過敏性休克。若有過敏現象，將立即

doi:10.6342/NTU202301094

停藥，並給予抗組織胺及/或類固醇。Vitamin C (抗壞血酸): 在大劑量下會發生下痢等現象，將給予止瀉藥處理。Hydrocortisone (氫化可體松): 短期內血糖上升。將檢測血糖，並給予適當劑量的降血糖藥物控制。

(一)-2 救濟措施: 1、如依本研究所訂試驗計畫而引發之身體、心理上之不良反應、副作用或傷害，本醫院及主持人將提供受試者專業醫療照顧及醫療諮詢。受試者不必負擔治療不良反應或損害之必要醫療費用。2、除法定賠償及醫療照顧外，本研究不提供其他形式之賠償或補償。3、受試者不會因為簽署本同意書，而喪失在法律上的任何權利。4、本研究未投保責任保險。

(二)心理方面可能產生之傷害與處理方式：本研究較少造成病患心理方面之傷害。如因資料外洩而造成病患的心理傷害，將照會精神科醫師進行心理輔導。

(三) 受試者退出本實驗之條件: 對所使用藥物出現過敏反應，因其他原因而必需使用與本試驗同成份之藥物

九、研究成果

(一)預期成果及主要效益：

利用「維生素 C，維生素 B1 和氫化可體松」在嚴重敗血症和敗血症性休克患者中的協同免疫調節作用來減少患者的死亡率(40%)。

(二)研究成果之歸屬及利用：研究成果將以論文或海報形式發表，歸屬於研究主持人及亞東醫院。

十、研究經費需求及其來源：研究經費需求 (560,000)及其來源由院內計畫支付。

項目名稱	說明	金額
人事費用-研究護士	10,000*12 月=120,000	120,000
護理部協助研究	80 cases*100*2 times	16,000
藥品費用 (vitamin B1, vitamin C, hydrocortisone, normal saline)	ABC group: Vitamin B1 (100mg/2ml/amp): 20(元/vial)*2(支/次)*2 (次/天) * 4 days * 40 cases= 12,800 Vitamin C (500 mg/5ml/amp): 8(元/vial)*3 (vials/次)*4 (次/天)*4 days*40 cases= 15,360 Hydrocortisone (100mg/amp): 37(元/vial)*1(支/次)*4 (次/天) * 7 days * 80 cases=85,000 Normal saline (500ml/bottle): 1 bottle/day*31 (元/btl)*4 days*80 cases*2= 20,000	133,160
臨床試驗藥局藥品保管費	1 次	10,000
耗材	包含包管路用鋁箔紙，輸液管路、紙張文具、注射及抽血用針具，冰寶，冰桶，試管，保溫箱，及其他耗材	100,000
檢驗 (vitamin B1 & vitamin C, cortisone ,	1000*2 times*80 cases (需外送淡水馬階醫院，含快遞交通費，檢驗技術費)	160,000

doi:10.6342/NTU202301094

lactic acid)		
論文英文編修、統計、投稿相關費用		50,000
	合　　計	**589,160**

十一、利益衝突：本計畫為主持人自行發起之學術研究，經費來源為亞東紀念醫院，無可能衍生之商業利益問題。

doi:10.6342/NTU202301094

十二、參考文獻或國內外文獻報告

1. Harrison DA, Welch CA, Eddleston JM: The epidemiology of severe sepsis in England, Wales and Northern Ireland, 1996 to 2004: secondary analysis of a high quality clinical database, the ICNARC Case Mix Programme Database. Critical care (London, England) 2006, 10(2):R42.
2. Finfer S, Bellomo R, Lipman J, French C, Dobb G, Myburgh J: Adult-population incidence of severe sepsis in Australian and New Zealand intensive care units. Intensive care medicine 2004, 30(4):589-596.
3. Huang CT, Tsai YJ, Tsai PR, Yu CJ, Ko WJ: Epidemiology and Outcome of Severe Sepsis and Septic Shock in Surgical Intensive Care Units in Northern Taiwan. Medicine 2015, 94(47):e2136.
4. Kaukonen KM, Bailey M, Suzuki S, Pilcher D, Bellomo R: Mortality related to severe sepsis and septic shock among critically ill patients in Australia and New Zealand, 2000-2012. Jama 2014, 311(13):1308-1316.
5. Gaieski DF, Edwards JM, Kallan MJ, Carr BG: Benchmarking the incidence and mortality of severe sepsis in the United States. Critical care medicine 2013, 41(5):1167-1174.
6. Kadri SS, Rhee C, Strich JR, Morales MK, Hohmann S, Menchaca J, Suffredini AF, Danner RL, Klompas M: Estimating Ten-Year Trends in Septic Shock Incidence and Mortality in United States Academic Medical Centers Using Clinical Data. Chest 2017, 151(2):278-285.
7. Shen HN, Lu CL, Yang HH: Epidemiologic trend of severe sepsis in Taiwan from 1997 through 2006. Chest 2010, 138(2):298-304.
8. Hotchkiss RS, Moldawer LL, Opal SM, Reinhart K, Turnbull IR, Vincent J-L: Sepsis and septic shock. Nature reviews Disease primers 2016, 2:16045-16045.
9. Richard S. Hotchkiss MD, and Irene E. Karl, Ph.D.: <physio and treatment sepsis NEJM review.pdf>. n engl j med january 9, 2003, 348(2).
10. Costa NA, Gut AL, de Souza Dorna M, Pimentel JA, Cozzolino SM, Azevedo PS, Fernandes AA, Zornoff LA, de Paiva SA, Minicucci MF: Serum thiamine concentration and oxidative stress as predictors of mortality in patients with septic shock. Journal of critical care 2014, 29(2):249-252.
11. Donnino MW, Carney E, Cocchi MN, Barbash I, Chase M, Joyce N, Chou PP, Ngo L: Thiamine deficiency in critically ill patients with sepsis. Journal of critical care 2010, 25(4):576-581.
12. Lima LF, Leite HP, Taddei JA: Low blood thiamine concentrations in children upon admission to the intensive care unit: risk factors and prognostic significance. The American journal of clinical nutrition 2011, 93(1):57-61.
13. Donnino MW, Andersen LW, Chase M, Berg KM, Tidswell M, Giberson T, Wolfe R, Moskowitz A, Smithline H, Ngo L et al: Randomized, Double-Blind, Placebo-Controlled Trial of Thiamine as a Metabolic Resuscitator in Septic Shock: A Pilot Study. Critical care medicine 2016, 44(2):360-367.
14. Keh D, Trips E, Marx G, Wirtz SP, Abduljawwad E, Bercker S, Bogatsch H, Briegel J, Engel C, Gerlach H et al: Effect of Hydrocortisone on Development of Shock Among Patients With Severe Sepsis: The HYPRESS Randomized Clinical Trial. Jama 2016, 316(17):1775-1785.
15. May JM, Harrison FE: Role of vitamin C in the function of the vascular endothelium. Antioxidants & redox signaling 2013, 19(17):2068-2083.
16. Wilson JX: Mechanism of action of vitamin C in sepsis: ascorbate modulates redox signaling in endothelium. BioFactors (Oxford, England) 2009, 35(1):5-13.
17. Fowler AA, 3rd, Syed AA, Knowlson S, Sculthorpe R, Farthing D, DeWilde C, Farthing CA, Larus TL, Martin E, Brophy DF et al: Phase I safety trial of intravenous ascorbic acid in patients with severe sepsis. Journal of translational medicine 2014, 12:32.
18. Sprung CL, Annane D, Keh D, Moreno R, Singer M, Freivogel K, Weiss YG, Benbenishty J, Kalenka A, Forst H et al: Hydrocortisone therapy for patients with septic shock. N Engl J Med

doi:10.6342/NTU202301094

2008, 358(2):111-124.
19. Marik PE: Critical illness-related corticosteroid insufficiency. Chest 2009, 135(1):181-193.
20. Wilson JX: Evaluation of vitamin C for adjuvant sepsis therapy. Antioxidants & redox signaling 2013, 19(17):2129-2140.
21. Fisher BJ, Kraskauskas D, Martin EJ, Farkas D, Puri P, Massey HD, Idowu MO, Brophy DF, Voelkel NF, Fowler AA, 3rd et al: Attenuation of sepsis-induced organ injury in mice by vitamin C. JPEN Journal of parenteral and enteral nutrition 2014, 38(7):825-839.
22. Fisher BJ, Seropian IM, Kraskauskas D, Thakkar JN, Voelkel NF, Fowler AA, 3rd, Natarajan R: Ascorbic acid attenuates lipopolysaccharide-induced acute lung injury. Critical care medicine 2011, 39(6):1454-1460.
23. Zhou G, Kamenos G, Pendem S, Wilson JX, Wu F: Ascorbate protects against vascular leakage in cecal ligation and puncture-induced septic peritonitis. American journal of physiology Regulatory, integrative and comparative physiology 2012, 302(4):R409-416.
24. **Marik PE, Khangoora V, Rivera R, Hooper MH, Catravas J: Hydrocortisone, Vitamin C, and Thiamine for the Treatment of Severe Sepsis and Septic Shock: A Retrospective Before-After Study. Chest 2017, 151(6):1229-1238.**
25. Kalil AC, Johnson DW, Cawcutt KA: Vitamin C Is Not Ready for Prime Time in Sepsis but a Solution Is Close. Chest 2017, 152(3):676.
26. Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, Bellomo R, Bernard GR, Chiche JD, Coopersmith CM et al: The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). Jama 2016, 315(8):801-810.
27. Howell MD, Davis AM: Management of Sepsis and Septic Shock. Jama 2017, 317(8):847-848.

doi:10.6342/NTU202301094

正本

檔　　號：
保存年限：

衛生福利部　函

機關地址：11558 台北市南港區忠孝東路六段488號
傳　　真：
聯絡人及電話：張先生02-27877450
電子郵件信箱：

220
新北市板橋區南雅南路2段21號
受文者：醫療財團法人徐元智先生醫藥基金會亞東紀念醫院

發文日期：中華民國108年6月24日
發文字號：衛授食字第1080015698號
速別：普通件
密等及解密條件或保密期限：
附件：藥品臨床試驗應注意事項1份

主旨：有關貴院檢送許孟璇醫師主持之「PanNoble (Ascorbic acid) Injection 250 mg/mL、Thiamine hydrochloride Injection 50 mg "ASTAR"、Hydrocortisone Injection 100 mg "CYH"」供學術研究用藥品臨床試驗計畫（計畫編號：107133-F）乙案，經核，本部原則同意試驗進行，惟本部得於試驗施行期間，依最新之科學發展，通知修正本試驗。隨函檢送藥品臨床試驗應注意事項1份，詳如說明段，請查照。

說明：

一、復貴院108年5月27日亞人體字第1080527006號函。

二、案內試驗申請人/試驗委託者為醫療財團法人徐元智先生醫藥基金會亞東紀念醫院，本部同意之計畫書版本日期為：Version: 7，Date: 20190408。

三、本部同意貴院受試者同意書版本日期如下：2019/04/08 v.7, FEMH-107133-F。

四、對上述內容如有疑義，請與承辦人周彤聯絡，電話：(02)8170-6000#525，E-mail: tchou818@cde.org.tw。

正本：醫療財團法人徐元智先生醫藥基金會亞東紀念醫院
副本：全國藥物不良反應通報中心、財團法人醫藥品查驗中心

部長陳時中

6-25

藥品臨床試驗應注意事項

一、請依107年1月23日衛授食字第1061412167號公告，有關「台灣藥物臨床試驗資訊網」之相關規定，上網登錄公開之資訊，並依107年3月29日衛授食字第1071401881號函定期更新試驗計畫資訊。

二、本藥尚屬臨床試驗用藥，為加強本藥之不良作用監視，請依據「藥品優良臨床試驗準則」第106條之規定，受試者發生任何嚴重不良反應事件，試驗主持人應立即通知試驗委託者，試驗委託者獲知未預期之死亡或危及生命之嚴重藥品不良反應，應於獲知日起七日內通報全國藥物不良反應通報中心，有違者，將列入GCP查核時之考量。

三、本試驗請依全民健康保險法第51條規定：「人體試驗不在保險給付範圍」，故臨床試驗期間醫療費用不應由健保支付。

四、醫師或藥商逕自發表藥品臨床試驗結果予一般媒體者，依下列原則辦理：

(一)如藥品未上市前逕自發表藥品臨床試驗結果予一般媒體，應予個案認定是否屬藥品廣告。

(二)若試驗結果發表於醫學會議或學術性醫學刊物，則依醫療法第87條第2項規定，不視為醫療廣告。

(三)醫院於一般報章雜誌發佈試驗結果，若涉及招徠醫療業務，則依違反醫療法第86條規定論處。

(四)若藥商直接於報章雜誌或產品發表會發布藥品名稱、廠牌及療效，則該藥商違反藥事法第68條第三款，並依藥事法92條規定處新台幣20萬元以上500萬元以下罰鍰。

五、本試驗主持人應任用合格之試驗相關人員，確保其對計畫有充分之瞭解，被授權之責任與工作並應留下書面紀錄。

六、本試驗應經由醫院之人體研究倫理審查委員會同意後始准執行，如醫院核准之計畫與本部核准內容不儘相同，應申請計畫變更並經核准後始可執行。

七、人體研究倫理審查委員會對人體臨床試驗之設計與執行，應進行必要之查核與監督，以確保臨床試驗之品質及安全；並依據「藥品優良臨床試驗準則」之規定，善盡保護受試者之責任。

八、如依「多國多中心藥品臨床試驗計畫審查程序」者，計畫內容變更時，應檢附相關資料及該公告程序第三點文件，於向同公告程序第二點所列國家申請變更案之同時，同步函送本部核備，若經查有延遲通報乙事，將依延遲時間暫停行使「多國多中心藥品臨床試驗計畫審查程序」之權益。

九、有關後續受試者同意書變更案，請向本部公告委託之機構/法人申請。

學術研究績效表

2023/05/01

姓名：許孟璇 **職稱：台大流行病學研究所博五**

一、近五年內之研究成果

序號	論文資料	1. 突破性之創見 2. 對學術發展、社會、經濟等面向之影響
1	Neglected human Rickettsia felis infection in Taiwan: A retrospective seroepidemiological survey of patients with suspected rickettsioses. Yang WH*, **Hsu MS***, Shu PY, Tsai KH, Fang CT. **PLoS Negl Trop Dis.** 2021 Apr 19;15(4):e0009355. doi: 10.1371/journal.pntd.0009355. eCollection 2021 Apr. (***Co-first authors with equal contribution**) (2021JIF=4.781；R/C= 2/24，Q1 tropical medicine)	欲了解 *R. felis* 在台灣的流行病學，臨床表徵。
2	Role of nasal swab culture in guiding antimicrobial therapy for acute cellulitis in the era of community-acquired methicillin-resistant Staphylococcus aureus: A prospective study of 89 patients. **Hsu MS**, Liao CH, Fang CT. **J Microbiol Immunol Infect.** 2019 Apr 10. pii: S1684-1182(18)30459-6. doi: 10.1016 (2021JIF=10.276; R/C= 15/95，Q1 INFECTIOUS DISEASES	改善蜂窩性組織炎病人的治療效果，減少住院天數。

序號	學會海報
2017 ICC	Epidemiology and clinical features of cellulitis in adult with and without Staphylococcus aureus nasal colonization
2020 IDST	Role of nasal swab culture in guiding antimicrobial therapy for acute cellulitis in the era of community-acquired methicillin-resistant Staphylococcus aureus: A prospective study of 89 patients. **Hsu MS**, Liao CH, Fang CT. **J Microbiol Immunol Infect.** 2019 Apr 10. pii: S1684-1182(18)30459-6. doi: 10.1016 (2021JIF=10.276; R/C= 15/95，Q1 INFECTIOUS DISEASES
2022 IDST	Severe eosinophilia with acute hemiparesis
2023 IDST	Vitamin D Deficiency and Incident Active Tuberculosis among non-HIV patients: A Matched Density Case-Control Study

doi:10.6342/NTU202301094