

**THE EFFECT OF BEE PROPOLIS AND TABATA
EXERCISE ON IMMUNE RESPONSE, BONE
METABOLISM MARKERS, OXIDANTS-
ANTIOXIDANTS STATUS,
CARDIORESPIRATORY FITNESS, BODY
COMPOSITION AND QUALITY OF LIFE AMONG
PHYSICALLY INACTIVE OVERWEIGHT MEN**

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UNIVERSITI SAINS MALAYSIA

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by

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LIST OF SYMBOLS

%	Percentage
~	Approximation
$10^9 \cdot \mu\text{L}^{-1}$	1 billion cells per microliter
$10^3 \cdot \mu\text{L}^{-1}$	1,000 cells per microliter
α	Alpha
β	Beta
CD3^+	Total T lymphocytes
$\text{CD3}^+\text{CD4}^+$	T-helper
$\text{CD3}^+\text{CD8}^+$	T-cytotoxic
$\text{CD16}^+\text{CD56}^+$	Natural killer
$\text{cells} \cdot \mu\text{L}^{-1}$	Cells per microliter
$^{\circ}\text{C}$	Degrees Celsius
$\text{g} \cdot \text{cm}^{-2}$	Grams per square centimeter
$\text{kg} \cdot \text{m}^{-2}$	Kilograms per square meter
$\mu\text{g} \cdot \text{mL}^{-1}$	Micrograms per milliliter
$\text{pg} \cdot \text{mL}^{-1}$	Picograms per milliliter
$\text{ng} \cdot \text{mL}^{-1}$	Nanograms per milliliter
$\text{mmol} \cdot \text{L}^{-1}$	Millimoles per liter
$\text{MET} \cdot \text{min} \cdot \text{week}^{-1}$	MET per minute per week
$\text{mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$	Milliliters of oxygen per kilogram of body weight per minute
$\text{g} \cdot \text{min}^{-1}$	Gram per minute
mg	Milligram
$\text{mg} \cdot \text{g}^{-1}$ body weight	Milligram per gram body weight
$\text{mg} \cdot \text{kg}^{-1}$	Milligram per kilogram

LIST OF ABBREVIATIONS

1-CTP	Serum C-terminal cross-linked telopeptide of type 1 collagen
6MWD	Six-minute walking distance
ALP	Serum alkaline phosphatase
ANMS	Agenda Nasional Malaysia Sihat
ANOVA	Analysis of Variance
ATP	Adenosine triphosphate
BMD	Bone mineral density
BMI	Body mass index
BMU	Basic multicellular units
CAPE	Caffeic acid phenethyl ester
CD	Cluster of differentiation
CKD	Chronic kidney disease
CONSORT	CONsolidated Standards of Reporting Trials
CRF	Cardiorespiratory fitness
CRP	C-reactive protein
CRT	Circuit resistance training
CVD	Cardiovascular disease
DNA	Deoxyribonucleic acid
ELISA	Enzyme-linked immunosorbent assay
EPOC	Excess post-exercise oxygen consumption
EEP	Ethanol extract of propolis
FBC + DC	Full blood count and differential count
FBG	Fasting blood glucose
GABA	Damma-aminobutyric acid
GPx	Glutathione peroxidase
HIIE	High-intensity interval exercise
HIIT	High-intensity interval training
K ₂ EDTA	Ethylenediaminetetraacetic acid
HbA1c	Haemoglobin A1c.
HR _{max}	Heart rate maximum
LDL-C	Low-density lipoprotein cholesterol

LPS	lipopolysaccharide
IG	Immunoglobulin
IL	Interleukin
IOC	International Olympic committee
IPAQ	International physical activity questionnaire
IT	Interval training
MET	Metabolic equivalent of task
MICT	Moderate-intensity continuous training
MISS	Moderate-intensity steady-state exercise
MOH	Ministry of health
NAFLD	Non-alcoholic fatty liver disease
NCDs	Non-communicable diseases
NK	Natural killer
NPRA	National Pharmaceutical Regulatory Agency
OC	Serum osteocalcin
P90X	Power 90 Extreme
SOD	Superoxide dismutase
TAS	Serum total antioxidant status
TOS	Total oxidant status
TC	Total cholesterol
Th	T-helper
TNF	Tumour necrosis factor
T2DM	Type-2 diabetes mellitus
URTI	Upper respiratory tract infections
PAR-Q	Physical activity readiness questionnaire
QoL	Quality of life
RM	Ringgit Malaysia
ROS	Reactive oxygen species
SPIRIT	Standard Protocol Items: Recommendations for Interventional Trials
VO _{2max}	Maximal rate of oxygen consumption
VO _{2peak}	Peak oxygen uptake
WHO	World Health Organization
WHOQOL-BREF	World Health Organisation Quality of Life Questionnaire: Brief
WHR	Waist-hip-ratio

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**KESAN PROPOLIS LEBAH DAN SENAMAN TABATA TERHADAP
TINDAKBALAS IMUN, PENANDA METABOLISME TULANG, STATUS
OKSIDA-ANTI OKSIDA, KECERGASAN KARDIORESPIRATORI,
KOMPOSISI BADAN, DAN KUALITI HIDUP DALAM KALANGAN
LELAKI BERLEBIHAN BERAT BADAN YANG TIDAK AKTIF FIZIKAL**

ABSTRAK

Tujuan kajian ini adalah untuk menyiasat kesan gabungan senaman Tabata dengan suplemen propolis lebah terhadap tindak balas imun, penanda metabolisme tulang, status oksida-antioksidan, kecergasan kardiorespiratori, komposisi badan dan kualiti hidup. Seramai enam puluh lelaki yang tidak aktif secara fizikal dan mempunyai berat badan berlebihan telah direkrut dan dibahagikan secara rawak ke dalam empat kumpulan (n=15): kumpulan kawalan (C), kumpulan propolis lebah (B), kumpulan senaman Tabata (E) dan kumpulan gabungan senaman Tabata propolis lebah (EB). Propolis lebah diambil oleh peserta dalam kumpulan B dan EB pada dos 1000 mg (500 mg/tablet) setiap hari selepas makan selama 12 minggu. Peserta dalam kumpulan E dan EB melakukan senaman Tabata, 3 sesi setiap minggu selama 12 minggu. Ketinggian badan, berat badan, indeks jisim tubuh (BMI), peratusan lemak badan, nisbah pinggang-pinggul (WHR), kecergasan kardiorespiratori dan kualiti hidup peserta diukur pada ujian pra-, tengah- dan pasca-kajian. Selain itu, sampel darah diambil pada ujian pra-, tengah- dan pasca-kajian selepas berpuasa semalaman untuk menganalisis tindak balas imun (jumlah leukosit, neutrofil, monosit, eosinofil, basofil, jumlah limfosit T CD3⁺, T-pembantu CD3⁺CD4⁺, T-sitotoksik CD3⁺CD8⁺, nisbah CD4/CD8, sel pembunuh semula jadi CD16⁺CD56⁺, IL-6, IL-10, nisbah IL-6/IL-10), penanda metabolisme tulang (serum C-terminal cross-linked telopeptide

kolagen jenis 1 (1-CTP), serum fosfatase alkali (ALP), serum osteocalcin (OC)), status oksida-antioksidasi (jumlah status antioksidan (TAS), F2-Isoprostanes) dan faktor risiko kardiovaskular (glukosa darah puasa (FBG), jumlah kolesterol (TC)). Keputusan menunjukkan bahawa pengambilan suplemen propolis lebah sahaja (B) hanya menurunkan secara signifikan ($p < 0.05$) kepekatan 1-CTP pada ujian pasca-kajian berbanding dengan ujian pra-kajian, manakala senaman Tabata sahaja (E) menunjukkan perubahan signifikan dalam peratusan lemak badan (34.12 to 30.62 %) ($p = 0.002$), WHR (0.98 to 0.92) ($p = 0.040$), lemak tepu (14.30 to 9.20) ($p = 0.035$), nisbah CD4/CD8 (0.90 to 1.44) ($p = 0.004$), IL-6 (3.44 to 2.58 pg.mL^{-1}) ($p = 0.018$), IL-10 (2.91 to 3.83 pg.mL^{-1}) ($p = 0.038$), nisbah IL-6/IL-10 (1.18 to 0.67) ($p < 0.01$) dan FBG (6.34 to 5.84 mmol.L^{-1}) ($p = 0.022$). . Gabungan senaman Tabata dan suplemen propolis lebah (EB) menghasilkan perubahan signifikan tambahan ($p < 0.05$) dalam jumlah leukosit leukocytes (7.36 to $6.09 \times 10^9 \cdot \text{uL}^{-1}$) ($p = 0.037$) dan TAS (1.37 to 1.85 mmol.L^{-1}) ($p = 0.038$) pada ujian pasca-kajian berbanding dengan ujian pra-kajian serta peningkatan signifikan dalam IL-10 (2.36 to 4.16 pg.mL^{-1}) ($p = 0.025$) pada ujian tengah berbanding dengan ujian pra-kajian. Sebagai kesimpulan, kedua-dua senaman Tabata yang dilakukan secara berasingan (E) dan gabungan senaman Tabata dengan propolis (EB) memberikan manfaat yang hampir sama dalam meningkatkan tindak balas imun, penanda metabolisme tulang, status pengoksidaan-antioksidasi dan mengurangkan faktor risiko CVD pada lelaki yang tidak aktif secara fizikal dan mempunyai berat badan berlebihan, manakala suplemen propolis lebah sahaja (B) hanya menunjukkan manfaat kepada penanda resorpsi tulang (1-CTP). Menggabungkan propolis selepas senaman Tabata memberikan kesan sinergistik tambahan terhadap jumlah status antioksidasi.

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IMMUNE RESPONSE, BONE METABOLISM MARKERS, OXIDANTS-
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INACTIVE OVERWEIGHT MEN**

ABSTRACT

The purpose of this study is to investigate the combined effects of Tabata exercise with bee propolis supplementation on immune responses, bone metabolism markers, oxidants-antioxidants status, cardiorespiratory fitness, body composition and quality of life. Sixty physically inactive overweight men were recruited were randomised into into four groups (n=15): control group (C), bee propolis group (B), exercise group (E) and combined exercise and bee propolis (EB). Bee propolis was consumed by participants in B and EB groups at a dosage of 1000 mg (2 tablets of 500 mg) daily after meals for 12 weeks. The participants in the E and EB groups performed the Tabata exercise, 3 sessions per week for 12 weeks. Participants' body height, weight, body mass index (BMI), body fat percentage, waist-hip-ratio (WHR), cardiorespiratory fitness and quality of life were measured at pre-, mid- and post-tests. In addition, blood samples were collected at pre-, mid- and post-tests after an overnight fast to analyse immune responses (total leukocytes, neutrophils, monocytes, eosinophils, basophils, total T lymphocytes CD3⁺, T-helper CD3⁺CD4⁺, T-cytotoxic CD3⁺CD8⁺, CD4/CD8 ratio, natural killer CD16⁺CD56⁺, IL-6, IL-10, IL-6/IL-10 ratio), bone metabolism markers (serum C-terminal cross-linked telopeptide of type 1 collagen (1-CTP), serum alkaline phosphatase (ALP), serum osteocalcin (OC)), oxidants-antioxidants status (serum total antioxidant status (TAS), F2-Isoprostanes)

and cardiovascular risk factor (fasting blood glucose (FBG), total cholesterol (TC)). Results showed that consumption of bee propolis supplementation alone (B) had only significantly decreased of the 1-CTP concentration (3.65 to 2.12 ng.mL⁻¹) (p = 0.027) at post-test as compared to pre-test while the Tabata exercise alone (E) had significant changed in body fat percentage (34.12 to 30.62 %) (p = 0.002), WHR (0.98 to 0.92) (p = 0.040), visceral fat (14.30 to 9.20) (p = 0.035), CD4/CD8 ratio (0.90 to 1.44) (p = 0.004), IL-6 (3.44 to 2.58 pg.mL⁻¹) (p = 0.018), IL-10 (2.91 to 3.83 pg.mL⁻¹) (p = 0.038), IL-6/IL-10 ratio (1.18 to 0.67) (p < 0.01) and FBG (6.34 to 5.84 mmol.L⁻¹) (p = 0.022). The combination of Tabata exercise and bee propolis supplementation (EB) resulted in additional significant changes in total leukocytes (7.36 to 6.09 10⁹.uL⁻¹) (p = 0.037) and TAS (1.37 to 1.85 mmol.L⁻¹) (p = 0.038) at post-test as compared to pre-test as well as significant increase in IL-10 (2.36 to 4.16 pg.mL⁻¹) (p = 0.025) at the mid-test compared to the pre-test. As a conclusion, both independent Tabata exercise (E) and the combination of Tabata exercise with propolis (EB) yield nearly identical benefits for improving immune response, bone metabolism markers, oxidant-antioxidant status and reducing CVD risk factors in physically inactive overweight men while independent bee propolis supplementation (B) only exhibited benefit for the bone resorption marker (1-CTP). Combining propolis following the Tabata exercise offered addition synergistic effect on serum antioxidant status.

CHAPTER 1

INTRODUCTION

1.1 Background of the study

The modern lifestyle is characterised by sedentary activities and easy access to calorie-dense food, leading to a growing number of physically inactive and overweight individuals (Macia *et al.*, 2021). Malaysia has the highest prevalence of obesity among adults in Southeast Asia (Kyaw *et al.*, 2022) where 54.4% of the adult population was reported to be overweight or obese and 84.0% of the adult population are not active in sports, fitness, and leisure activities (Chong *et al.*, 2023). The prevalence of physical inactivity among Malaysians is higher compared to other Asian countries such as China, India and Hong Kong (Nik-Nasir *et al.*, 2022).

Numerous studies have reported that being physically inactive and overweight may lead to an increased risk of obesity (Kazmi *et al.*, 2022; Silveira *et al.*, 2022), osteoporosis (Gkastaris *et al.*, 2020), immunosuppression (de Frel *et al.*, 2020; Syed *et al.*, 2024), persistent systemic low-grade inflammation (Burini *et al.*, 2020; Khanna *et al.*, 2022), which in turn increase the risk of developing various diseases, especially non-communicable diseases (NCDs) (Felisbino-Mendes *et al.*, 2020; Ejigu *et al.*, 2024), which contribute to an individual's poor health status and quality of life (QoL). These conditions can be life-threatening and, in turn, will increase the risk of developing numerous diseases, including heart disease (Alfaddagh *et al.*, 2020), diabetes (van de Vyer, 2023; Sharif *et al.*, 2021) and hypertension (Zhang *et al.*, 2023; Copur *et al.*, 2023).

High-intensity interval training (HIIT) is an exercise method that alternates short periods of intense anaerobic exercise with a less intense recovery period between

intervals (Atakan *et al.*, 2021). Studies have shown that HIIT is well known to elicit countless health benefits such as increased cardiorespiratory fitness (CRF) within as little as 2 weeks (Martin-Smith *et al.*, 2020), increased immune function (Lee *et al.*, 2023; Souza *et al.*, 2021), increased antioxidant status (Portela *et al.*, 2024; Lu *et al.*, 2021) and improved bone health status (Noll *et al.*, 2021; Alghadir *et al.*, 2025). HIIT is much preferred by many people as it could elicit similar benefits as traditional moderate-intensity continuous training (MICT) but requires less time.

Tabata is a type of HIIT workout developed by Dr. Izumi Tabata in 1996 that is more precise in its interval timing and has shorter recovery periods than traditional HIIT (Tabata., 2019). Each Tabata round involves eight intervals of 20 seconds of intense exercise followed by 10 seconds of rest (Tabata., 2019). Tabata exercise is widely favoured due to its ability to generate superior benefits within a shorter timeframe in several aspects, such as excess post-exercise oxygen consumption (EPOC) (Jung *et al.*, 2019), improved CRF (Men *et al.*, 2023; Martin-Smith *et al.*, 2020; Islam *et al.*, 2020) and improved muscle strength and endurance (Domaradzki *et al.*, 2020).

Consuming supplements may also enhance an individual's health status. Bee propolis is a complex resinous substance collected by bees from various barks of trees, buds, and sprouts of plants, which is subsequently mixed with beeswax and bees' salivary enzymes (Tekiner and Yeşilkaya, 2021). Propolis is produced by bees in the construction and maintenance of the bees' hives and has been traditionally used to fight infections (Ożarowski *et al.*, 2023; Rivera-Yañez *et al.*, 2023), boost immune function (Al-Hariri *et al.*, 2019) and treat osteoporosis (Ekeuku and Chin, 2021). More than 300 chemical compounds have been identified in bee propolis and the main primary biologically active components present in propolis are phenolic compounds (e.g.,

flavonoids, aromatic compounds, terpenes, and essential oil) (Tekiner and Yeşilkaya, 2021). Bee propolis has been extensively studied for its antioxidant (Bhatti *et al.*, 2024), anti-inflammatory (Pahlavani *et al.*, 2020), antiviral (Yosri *et al.*, 2021), anticarcinogenic (Oršolić and Jazvinščak Jembrek, 2022), hepatoprotective (Sahlan *et al.*, 2021; Nna *et al.*, 2021) and anti-microbial (Vargas Sánchez *et al.*, 2020).

Although extensive research has been conducted on the Tabata exercise, only a limited number of studies have specifically investigated its impact on selected immune parameters, oxidant-antioxidant status, bone metabolism markers, and QoL. This results in fragmented findings and a dearth of conclusive evidence regarding its efficacy and mechanisms of action. Furthermore, there are very limited human studies investigating the beneficial effects of bee propolis supplementation despite the numerous properties it possesses. Hence, the novelty of this study is to investigate the possible mechanism of action of the integrative effects of bee propolis supplementation and Tabata exercise.

1.2 Problem statement

According to Martin *et al.* (2024), who reported findings from the World Obesity Atlas 2024, Malaysia is expected to face a significant rise in both adult and childhood obesity. The report predicts that adult obesity will rise by 3.7% annually from 2020 to 2035, while child obesity rates will increase by 3.8% per year over the same period (Al-Haroni *et al.*, 2024). This prevalence is alarming given that Malaysia has the highest prevalence of obesity among adults in Southeast Asia (Kyaw *et al.*, 2022) where 54.4% of the adult population was reported to be overweight or obese and 84.0% of the adult population are not active in sports, fitness, and leisure activities (Chong *et al.*, 2023).

Consequently, this health crisis has precipitated a severe economic burden. According to (Isamail *et al.* (2024) and Chandran *et al.* (2021) who reported findings from the Ministry of Health (MOH) Malaysia and the World Health Organization (WHO), the government of Malaysia has spent RM 9.65 billion annually for healthcare allocation to cover the expenditure of hospitalisation, medical tests, medications, and primary consultations for NCDs, particularly cardiovascular disease (CVD), diabetes and cancer. The increasing prevalence of NCDs in Malaysia is putting a significant strain on the country's health system and is the main factor in the loss of productivity in the country, highlighting the urgent need for effective strategies to address and manage these chronic conditions.

Although the benefits of regular exercise and supplementation are well-established (Adji *et al.*, 2022; Prasertsri *et al.*, 2022; Zamani *et al.*, 2023), adherence to traditional exercise guidelines remains low. A primary barrier to physical activity is the lack of time due to occupational demands, modern lifestyle constraints and marital status (Ahmad *et al.*, 2021; Safi *et al.*, 2022). To address this time constraint barrier, HIIT, specifically the Tabata protocol, has gained global attention for producing physiological benefits similar to MICT but with significantly less time commitment (Atakan *et al.*, 2021; Alwi *et al.*, 2024). However, a notable research gap exists, while Tabata exercise has been reported to improve CRF (Lu *et al.*, 2023; Padkao and Prasertsri, 2025), reports on Tabata exercise on QoL, specifically on immune function, antioxidants, and bone metabolism markers in physically inactive overweight men remain limited and underexplored.

Furthermore, lifestyle interventions are often most effective when paired with nutritional support. However, there is a considerable degree of uncertainty among

individuals about which supplements are most beneficial for their health. This is due to contradictory findings reported on certain supplements, which result from different study designs and protocols employed in each study. Bee propolis has been studied for its medicinal properties (Yosri *et al.*, 2021; Pahlavani *et al.*, 2020). Yet, the current body of literature on bee propolis presents a significant limitation as the majority of findings are derived from animal models (Nna *et al.*, 2021) and *in vitro* studies (González-Serrano *et al.*, 2023). Critically, there is a scarcity of evidence examining the combined synergistic effects of Tabata exercise and bee propolis on immune and oxidative markers in humans. There are only a few human studies with a different type of population, parameters measured, and study design (Kanazashi *et al.*, 2023; Sajjadi *et al.*, 2023; Zakerkish *et al.*, 2019).

Therefore, this study aims to bridge these knowledge gaps by determining whether bee propolis supplementation and Tabata exercise interventions are able to improve QoL by positively modulating immune function, oxidants-antioxidants status, bone metabolism markers, CRF, CVD risk factors and body composition in physically inactive overweight men. By isolating and combining these variables, this research seeks to identify if the combination of Tabata exercise and bee propolis supplementation could produce superior health outcomes compared to either intervention alone, providing a novel, evidence-based strategy for managing obesity and health risks in this specific demographic.

1.3 Research questions

- I. What are the effect of bee propolis supplementation and Tabata exercise intervention on immune response (total leukocytes, neutrophils, monocytes, eosinophils, basophils, total T lymphocytes CD3⁺, T-helper CD3⁺CD4⁺, T-cytotoxic CD3⁺CD8⁺, CD4/CD8 ratio, natural killer CD16⁺CD56⁺, Interleukin-6, Interleukin-10, IL-6/IL-10 ratio) among physically inactive overweight men?
- II. What are the effect of bee propolis supplementation and Tabata exercise intervention on bone metabolism markers (Serum C-terminal cross-linked telopeptide of type 1 collagen [1-CTP], serum alkaline phosphatase [ALP] and serum osteocalcin [OC]) among physically inactive overweight men?
- III. What are the effect of bee propolis supplementation and Tabata exercise intervention on oxidants-antioxidants status (serum total antioxidant status [TAS] and F2-isoprostanes) among physically inactive overweight men?
- IV. What are the effect of bee propolis supplementation and Tabata exercise intervention on cardiorespiratory fitness (CRF) (six-minute walking distance [6MWD], estimated maximal rate of oxygen consumption [VO_{2max}]) among physically inactive overweight men?
- V. What are the effect of bee propolis supplementation and Tabata exercise intervention on body composition (body weight, body mass index [BMI], body fat percentage, waist-hip-ratio [WHR]) among physically inactive overweight men?
- VI. What are the effect of bee propolis supplementation and Tabata exercise intervention on cardiovascular disease (CVD) risk factors (total cholesterol

[TC] and fasting blood glucose [FBG]) among physically inactive overweight men?

- VII. What are the effect of bee propolis supplementation and Tabata exercise intervention on quality of life (QoL) (WHOQOL-BREF) among physically inactive overweight men?
- VIII. Are the combination effects of Tabata exercise and bee propolis supplementation significantly better than Tabata exercise only or bee propolis supplementation only?

1.4 Research objectives

1.4.1 Main objectives

To investigate the 12-week effect of bee propolis supplementation and Tabata exercise intervention on immune response, bone metabolism markers, oxidants-antioxidants status, cardiorespiratory fitness, body composition measurements and quality of life at pre-, mid- and post-test in physically inactive overweight men.

1.4.2 Specific objectives

- I. To compare the changes in immune response (total leukocytes, neutrophils, monocytes, eosinophils, basophils, total T lymphocytes CD3⁺, T-helper CD3⁺CD4⁺, T-cytotoxic CD3⁺CD8⁺, CD4/CD8 ratio, natural killer CD16⁺CD56⁺, IL-6, IL-10, IL-6/IL-10 ratio) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.

- II. To compare the changes in bone metabolism markers (1-CTP, ALP, OC) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- III. To compare the changes in oxidants-antioxidants status (TAS, F2-Isoprostanes) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- IV. To compare the changes in cardiorespiratory fitness (CRF) (6MWD, VO_{2max}) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- V. To compare the changes in body composition (body weight, BMI, body fat percentage, visceral fat, WHR) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- VI. To compare the changes in cardiovascular (CVD) risk factors (total cholesterol [TC] and fasting blood glucose [FBG]) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.

- VII. To compare the changes in quality of life (QoL) (WHOQOL-BREF) at pre-, mid- and post-test following the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- VIII. To investigate whether the 12-week combined intervention of Tabata exercise and bee propolis supplementation elicits significantly greater improvements compared to Tabata exercise or bee propolis supplementation alone across pre-, mid-, and post-test measurements.

1.5 Research hypotheses

- I. There is a significant difference in immune response (total leukocytes, neutrophils, monocytes, eosinophils, basophils, total T lymphocytes CD3⁺, T-helper CD3⁺CD4⁺, T-cytotoxic CD3⁺CD8⁺, CD4/CD8 ratio, natural killer CD16⁺CD56⁺, IL-6, IL-10, IL-6/IL-10 ratio) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- II. There is a significant difference in bone metabolism markers (1-CTP, ALP, OC) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- III. There is a significant difference in oxidants-antioxidants status (TAS, F2-Isoprostanes) at pre-, mid- and post-test between the 12-week combined bee

propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.

- IV. There is a significant difference in cardiorespiratory fitness (CRF) (6MWD, $\text{VO}_{2\text{max}}$) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- V. There is a significant difference in body composition (body weight, BMI, body fat percentage, visceral fat, WHR) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- VI. There is a significant difference in CVD risk factors (TC, FBG) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- VII. There is a significant difference in the quality of life (QoL) (WHOQOL-BREF) at pre-, mid- and post-test between the 12-week combined bee propolis supplementation and Tabata exercise, Tabata exercise only and bee propolis supplementation only in physically inactive overweight men.
- VIII. There is a significant difference between the 12-week combined intervention of Tabata exercise and bee propolis supplementation, Tabata exercise or bee propolis supplementation alone across pre-, mid-, and post-test measurements.

1.6 Research significance

This study aims to provide in-depth knowledge of the effect of Tabata exercise and bee propolis supplementation, areas where comprehensive human data are currently lacking. While Tabata is known to enhance CRF, its impact on immune function, oxidant-antioxidant status, and bone health remains underexplored. Furthermore, bee propolis was reported to have numerous medicinal properties, but to date, human studies on its effects on improving QoL (Davoodi *et al.*, 2022; Sajjadi *et al.*, 2023), immune function (Brätter *et al.*, 1999; Conte *et al.*, 2021), antioxidant (Diniz *et al.*, 2020; Anvarifard *et al.*, 2023) and bone health (Matsumoto *et al.*, 2022) have not been reported.

Furthermore, the synergistic potential of combining Tabata exercise with bee propolis remains unknown. This randomised control study may identify the possible differences between the respective groups as it focuses on physically inactive overweight males. This approach not only advances scientific understanding of lifestyle and nutritional interactions but also targets a high-risk demographic often overlooked in performance-based research. The majority of local studies in Malaysia and international research have primarily concentrated on investigating the impact of Tabata exercise on body composition and CRF, and there is a noticeable gap in the literature regarding its evaluation on immune response, oxidant-antioxidant status, and bone health. Consequently, this research addresses a significant gap in the literature by shifting the focus from standard body composition metrics to a broader evaluation of physiological health markers in humans.

Furthermore, the health policies in Malaysia were proposed to address the rising trend of NCDs, which incur high costs to be treated (MOH, 2024). This study aligned

with the government's National Agenda for a Healthy Malaysia (ANMS) policies for a healthier nation (ANMS, 2025; IHBR, 2025). Findings from this study might benefit society in terms of promoting an active lifestyle amidst the demands of busy working life and improving awareness of the burden of NCDs. Findings from this study could also contribute towards promoting time-saving and effective exercise programs to enhance an individual's health and quality of life. Thus, this intervention offers a viable solution to address common barriers such as time constraints and high workloads among adult males. Ultimately, establishing an effective, accessible protocol for this population may contribute to reduced healthcare burdens, improved workforce productivity, and broader economic stability.

1.7 Operational definitions

Summary of the main terms for the study's variables based on research objectives

- Bee propolis A complex resinous substance collected by bees from various barks of trees, buds, and sprouts of plants, which is subsequently mixed with beeswax and bees' salivary enzymes (Tekiner and Yeşilkaya, 2021).
- High-intensity interval training (HIIT) An exercise method that alternates short periods of intense anaerobic exercise with a less intense recovery period between intervals (Atakan *et al.*, 2021).
- Tabata A type of HIIT workout that is more precise in its interval timing and has shorter recovery periods, 20-seconds of intense short-bursts exercise followed by 10-seconds of rest periods (Tabata, 2019).

- Immune response The complex biological processes through which the body detects and defends itself against harmful agents such as pathogens (e.g., bacteria, viruses, fungi), foreign substances, or abnormal cells (e.g., cancer cells) (Agrati *et al.*, 2023).
- Bone metabolism markers A biochemical indicator used to assess the activity of bone remodeling, which is the continuous process of bone resorption (breakdown) and bone formation (Aggarwal *et al.*, 2023).
- Oxidants Molecules that have the ability to accept or donate electrons from other molecules in a chemical reaction, a process known as oxidation, which can result in oxidative stress formation (Chaudhary *et al.*, 2023).
- Antioxidants Molecules that can inhibit the oxidation of other molecules by donating an electron, thereby preventing oxidative damage (Ali *et al.*, 2020).
- Cardiorespiratory fitness (CRF) The efficiency and capacity of the cardiovascular and respiratory systems to supply oxygen to muscles during prolonged physical activity, typically measured by maximal oxygen uptake (VO_{2max}) (Raghuveer *et al.*, 2020).
- Quality of life (QoL) A broad multidimensional concept that encompasses an individual's overall well-being, including physical,

psychological, and social functioning (Aqtam *et al.*, 2023).

- Overweight A condition in which an individual has more body weight than is considered healthy for a given height with a body mass index (BMI) ranging from 25 to 29.9 kg.m⁻², defined by the World Health Organization (WHO) (Haam *et al.*, 2023).
- Physically Inactive An individual whose energy expenditure from physical activity is less than 600-MET.min.week⁻¹ (Shristi *et al.*, 2024)

CHAPTER 2

LITERATURE REVIEW

2.1 High-intensity interval training (HIIT): Tabata

High-intensity interval training (HIIT) is a form of interval training alternating short periods of intense anaerobic exercise with less intense recovery periods (Cao *et al.*, 2025). Ataka *et al.* (2021) defined HIIT as consisting of relatively brief bouts of vigorous exercise performed at a high relative intensity, typically at or above 90% of VO_{2max} , exceeding 75% of maximal power output, or at a minimum running speed sufficient to elicit VO_{2max} . The perceived exertion during such efforts generally falls within the "hard" to "very hard" range, corresponding to a score of 6 or higher on the 10-point Borg scale or 15 or higher on the 6 to 20 scale.

There are numerous types of HIIT protocol, such as Power 90 Extreme (P90X), Tabata, Spinning and CrossFit (Knapik, 2015). Tabata is more specific in its interval timing and has shorter recovery periods compared to the traditional HIIT and was developed by Dr. Izumi Tabata and his colleagues at the National Institute of Fitness and Sports in Japan in 1996 (Tabata, 2019). The original Tabata protocol was conducted on a cycle ergometer with a 4-minutes supramaximal effort at 170% of VO_{2max} for 4 times per week, with 7-8 rounds of 20-seconds exercise and 10-seconds of downtime HIIT intervals (Tabata, 2019).

This original protocol was implemented on the Japanese Olympic speed skating team and the team showed notable achievements at the 1998 Nagano Winter Olympics where the Japanese speed skater Hiroyasu Shimizu won the gold medal in the men's 500 meters, setting an Olympic record with a combined time of 1:11.35 over two races (IOC, 2025). These accomplishments suggest that implementing the Tabata protocol

may have contributed to the improved performance of Japanese speed skaters during this period.

2.1.1 Variation of Tabata training modalities

While the original Tabata protocol used cycling as the modality (Tabata *et al.*, 1996), contemporary adaptations often utilise bodyweight and functional movements, constructing a more accessible and versatile. Exercise movements like burpees, jump squats, push-ups, and mountain climbers are commonly integrated into bodyweight Tabata circuits.

For instance, Bibi *et al.* (2024) investigated the physiological effects of Tabata exercise on cardiometabolic and cardiorespiratory parameters in transgender individuals in Multan. This study did not implement the original Tabata protocol (170% of VO_{2max}) and employed with only three exercised-induced stress movements (pot jumps, high knees and jumping jacks) at 85 to 95% heart rate maximum (HR_{max}) with a total of 7 to 8 bouts of 20/10 seconds exercise/recovery bouts. This protocol led to significant improvements in systolic and diastolic blood pressure, resting heart rate, and oxygen saturation, demonstrating the effectiveness of Tabata-style HIIT in enhancing cardiometabolic and respiratory function

Noorbakhsh and Roshan (2023) also investigated the effects of 8 weeks of Tabata HIIT program combined with nanocurcumin supplementation on inflammation and cardiorespiratory health in overweight elderly women. The Tabata protocol was based on body weight-based exercise training programme (air squats, high knees, push-ups on knees, mountain climbers, standard plank, butt kickers, split jumps, and skaters) at an average intensity of 80-90% of HR_{max} of 20-second effort interval. Results showed significant improvements in inflammatory markers (including IL-6 and $TNF-\alpha$) and enhanced cardiorespiratory function, indicating that Tabata-style HIIT, when combined

with antioxidant-rich nanocurcumin, offers a promising strategy for improving health outcomes.

Twelve weeks of Tabata-style functional HIIT was also employed by Lu *et al.* (2023) to assess its effects on cardiometabolic health in female university students. This study only included four functional movements (jumping jacks, high knees, squat jumps, and mountain climbers) at 90% HR_{max} cut-off point. Despite the simplicity and low volume of the protocol, with only four exercises repeated cyclically, the intervention led to significant improvements in cardiorespiratory fitness, body fat reduction, insulin sensitivity, and lipid profiles compared to the control group. These findings highlight that even low-volume Tabata protocols, when performed consistently, can yield meaningful health benefits.

Murawska-Cialowicz *et al.* (2020) also conducted a 40-minute HIIT Tabata training consisting of eight repetitions of exercises (squat jumps, back extensions, crunches, push-ups, triceps dips, side crunches, military press with medicine ball and chin-ups) at 90 to 100% HR_{max} for 8 weeks. The results showed a significant 29.7% increase in serum irisin levels, both aerobic and anaerobic performance improvements, and a reduction in body fat percentage.

These protocols highlight the practicality of Tabata training using bodyweight exercises. The 20/10-second work/rest Tabata protocol and the 4-minute bout make it easy to organize when creating workouts that have multiple bouts (Evangelista *et al.*, 2022). This adaptability of Tabata-style workouts that require no equipment and their diversity in high intensity, duration and exercise movements make Tabata protocols suitable in modern fitness culture that prioritises minimalist training that can be performed anywhere (Scoubeau *et al.*, 2022).

2.1.2 HIIT/Tabata exercise for the physically inactive and overweight population

Many researchers have focused on finding optimal methods to combat obesity and overweight. The prevalence of physical inactivity among Malaysians is higher compared to other Asian countries such as China, India and Hong Kong (Nik-Nasir *et al.*, 2022). According to Martin *et al.* (2024), who reported findings from the World Obesity Atlas 2024, Malaysia is expected to face a significant rise in both adult and childhood obesity. The report predicts that adult obesity will rise by 3.7% annually from 2020 to 2035, while child obesity rates will increase by 3.8% per year over the same period (Al-Haroni *et al.*, 2024). Malaysia is currently living amidst an NCDs crisis and calling for urgent interventions to improve physical activity levels (Khoo *et al.*, 2020).

The time factor has become a very significant obstacle to the involvement of a person in a routine exercise programme (Healy *et al.*, 2024). Tabata exercise has emerged in recent years as a time-efficient means, as little as 4-minutes to improve cardiorespiratory fitness in as little as 2 weeks in adults (Martin-Smith *et al.*, 2020). Numerous evidence continues to support HIIT and Tabata as powerful methods to increase physical activity levels across diverse populations, especially overweight populations, and as scalable for different fitness levels. These training modalities remain at the forefront of efficient exercise strategies for time-constrained individuals.

The results of a few studies have indicated that Tabata exercise may be appropriate for the physically inactive and overweight population (Imanudin and Sultoni, 2017; Domaradzki *et al.*, 2020; Nur *et al.*, 2021; Zuo *et al.*, 2025). A local study conducted by Noor *et al.* (2021) implemented 6 weeks of Tabata workouts on twelve physically inactive individuals reported a positive immune cell response without significant changes in body weight, BMI, or body fat percentage. These findings suggest

that Tabata-style HIIT is a safe and effective method to improve immune responses in physically inactive individuals.

Bellicha *et al.* (2021) conducted a comprehensive overview of 12 systematic reviews encompassing 149 studies to assess the impact of exercise training on weight loss, body composition, and weight maintenance in adults with overweight or obesity. The analysis revealed that HIIT is a safe and feasible exercise modality for this population, leading to significant improvements in body composition and cardiorespiratory fitness. Notably, HIIT protocols can be adapted to individual fitness levels, allowing overweight individuals to perform exercises at intensities appropriate for their capabilities, thereby minimizing the risk of injury and enhancing adherence.

Zuo *et al.* (2025) demonstrated that 34 overweight female college students can safely and effectively perform Tabata-style HIIT, leading to improvements in both body composition and cognitive function after 8 weeks of intervention. These findings highlight that HIIT could be a scientific basis for the application of HIIT in the health management of the overweight population. Even though the protocol is very demanding and implemented at high intensity, previous research suggested that chronic exposure to such training may elevate enjoyment and the dropout rate in the study of overweight young women was relatively low (6.67%) (Li *et al.*, 2022).

Besides, Logan *et al.* (2014) reported a high adherence rate (90%) among inactive volunteer adolescents upon completion of the regimen. Tabata is perceived to be more enjoyable than moderate-intensity continuous training (MICT) in the obese population, which may improve adherence to exercise and promote health benefits over a longer period. Vella *et al.* (2017) also reported a high exercise adherence (93.40%) among 17 overweight and obese young adults who performed independent HIIT for

twelve sessions. This finding indicates that HIIT is not only effective but also enjoyable, with high unsupervised adherence rates among overweight and obese adults.

For physically inactive individuals, the Tabata exercise intensity can be scaled down by adjusting movement types (e.g., bodyweight squats, modified jumping jacks, or brisk cycling) while still preserving the high-intensity work interval (Domaradzki *et al.*, 2020). During high-effort intervals, the body undergoes rapid adenosine triphosphate (ATP) breakdown and accumulates lactate. (Smith *et al.*, 2023). One of the key features that makes Tabata suitable is the brief but sufficient recovery period between high-intensity intervals that allows partial replenishment of ATP and phosphocreatine stores (Tabata, 2019). This recovery window reduces fatigue accumulation and enables participants to maintain performance throughout the session.

Therefore, the short duration and customisable nature of Tabata lower psychological barriers to exercise, encouraging better adherence in the overweight population. These rates may indicate that the Tabata exercise protocol is tolerable and positively accepted for intervention in the physically inactive overweight population and might be a useful tool for combating the problem of being overweight while avoiding adverse effects (long-term exercise adherence). Table 2.1 summarise previous findings on HIIT/ exercise in physically inactive and overweight population,

Table 2.1 Summary of previous studies on the effects of HIIT on overweight and physically inactive populations

Author (Year)	Population (N)	Intervention	Key Findings	Adherence/ Feasibility
Martin-Smith et al. (2020)	Healthy overweight/ obese adults (Review of 18 studies)	Various HIIT protocols	Improved cardiorespiratory fitness (CRF).	Time- efficient strategy.

Table 2.1, Continued

Author (Year)	Population (N)	Intervention	Key Findings	Adherence/ Feasibility
Noor et al. (2021)	Physically inactive adults (N = 12)	6 weeks Tabata	Positive immune cell response. No significant changes in Body Weight, BMI, or Body Fat %.	Safe and effective for immune modulation.
Bellicha et al. (2021)	Adults with overweight/obesity (Review of 149 studies)	Various HIIT protocols	Significant improvements in body composition and CRF.	Safe and feasible modality; adjustable intensity minimizes injury risk.
Zuo et al. (2025)	Overweight female college students (N = 34)	8 weeks Tabata-style HIIT	Improvements in body composition and cognitive function.	Safe and effective for health management.
Li et al. (2022)	Overweight young women (N = 60)	12 weeks high- intensity training	Chronic exposure may elevate enjoyment.	Low dropout rate (6.67%).
Vella et al. (2017)	Overweight and obese young adults (N = 17)	12 sessions (Independent HIIT)	Effective management of weight/health.	High exercise adherence (93.40%).

Note. N; Number of participants. BMI; Body Mass Index, HIIT; High-intensity Interval Training.

2.2 Bee propolis supplementation

2.2.1 Overview of bee propolis

Although honey and propolis are both valuable apicultural products, they differ fundamentally in origin, composition, and function. Honey is widely recognized as a nutritional energy source derived principally from floral nectar (Sharaf et al., 2025). In contrast, bee propolis is not a food source but a functional defense substance served as a complex resinous substance collected by bees from various barks of trees, buds, and sprouts of plants, which is then mixed with beeswax and bees' salivary enzymes (Tekiner and Yeşilkaya, 2021). Propolis is in sticky-gummy form and is also known as 'bee glue' (Hossain *et al.*, 2022). The colour is normally yellow, green, and brown, but depends according to the source of resin and the age of the propolis (Tekiner and Yeşilkaya, 2021). Table 2.2 summarises the scientific comparison between propolis and honey.

Table 2.2 Summary of the comparison between propolis and honey

Aspect	Honey	Propolis
Source	Collected by bees from floral nectar, processed with enzymes into a carbohydrate-rich syrup.	Collected by bees from plant resins, mixed with wax, pollen, and salivary enzymes to form a sticky resin used to seal the hive.
Main Composition	~80% sugars (fructose, glucose), ~17% water, small amounts of amino acids, enzymes, vitamins, and polyphenols.	~50% resins (flavonoids, phenolic acids, terpenes), ~30% waxes, ~10% essential oils, ~5% pollen, and other organic compounds.
Typical Use	Raw honey, syrup.	Extracts, tinctures, tablets, nanoparticles, or supplements.
Form		

Table 2.2. Continued

Aspect	Honey	Propolis
Cost	Relatively cheaper, mass-produced.	More expensive, limited harvest and higher concentration of bioactives.
Primary Function (in hive)	Energy source for bees (nutrition).	Hive defense: antibacterial sealant protecting against microbes and environmental toxins.
Primary Bioactive Components	Phenolic acids (e.g., caffeic acid), flavonoids (quercetin, galangin), hydrogen peroxide (from glucose oxidase).	Flavonoids (chrysin, pinocembrin, galangin, quercetin), caffeic acid phenethyl ester (CAPE), artemisinic acid, cinnamic acids. - Potent antioxidant (CAPE inhibits ROS generation and activates Nrf2 pathway) - Strong anti-inflammatory (inhibits COX-2, NF- κ B, and cytokine release) - Immunomodulatory (enhances macrophage and lymphocyte activity) - Antimicrobial, antifungal, antiviral.
Mechanism of Action (Biological)	- Moderate antioxidant (scavenges ROS) - Anti-inflammatory through NF- κ B inhibition - Mild antimicrobial (due to hydrogen peroxide and acidity)	

Note. COX-2; Cyclooxygenase-2, NF- κ B; Nuclear Factor kappa-light-chain-enhancer of activated B cells, Nrf2; Nuclear factor erythroid 2-related factor 2, ROS; Reactive oxygen species.

Bees use propolis primarily as a structural and antimicrobial agent to maintain environmental stability within the colony. Propolis is used to cover the beehive wall structure (i.e. seal cracks, reinforce hive walls, and smooth internal surfaces) and to protect the bee colony from infection (Nader *et al.*, 2021). Its strong antimicrobial

properties help protect the hive from pathogenic microorganisms, including bacteria, fungi, and viruses, thereby contributing to colony health and hygiene (Salatino, 2022). Moreover, propolis is used to embalm and isolate intruders or foreign objects too large to remove from the hive, effectively preventing decomposition and disease spread (Wagh, 2013).

Propolis has been used widely for various purposes, such as in dental care (i.e. mouth rinse) (Otreba *et al.*, 2022; Ahangari *et al.*, 2024), used against respiratory tract infections in children (Yuksel and Akyol, 2016; Ożarowski and Karpiński, 2023) and used in dermatology and skin care (Kurek-Górecka *et al.*, 2020; El-Sakhawy *et al.*, 2023). Wagh (2013) reported that propolis was listed as an official drug in the London Pharmacopoeias of the 17th century and Zhu *et al.*, (2011) also reported that propolis was listed as a new registered medicine in the Chinese Pharmacopeia in the year 2005.

2.2.2 Chemical composition of bee propolis

The chemical composition of bee propolis is very complex and diverse. Although the composition of bee propolis varies depending on the plant sources from which it is derived, most samples share a similar overall chemical profile (Ahangari *et al.*, 2018; Hossain *et al.*, 2022). The primary constituents of propolis typically include approximately 50% resin, 30% wax, 10% essential oils, 5% pollen, and 5% other organic substances (Wagh, 2013; Vitor *et al.*, 2023).

More than 300 chemical compounds have been identified in bee propolis and the main primary biologically active components present in propolis are phenolic compounds (e.g., flavonoids, aromatic compounds, terpenes, and essential oil) (Ahangari *et al.*, 2018; Tekiner and Yeşilkaya, 2021). Figure 2.1 represents the chemical structure of propolis isolated compounds. Flavonoid compounds are abundant in propolis and play a significant role in the bioactivity of propolis (Hossain *et al.*, 2022).