THE EFFECT OF SEA GRAPE (*Caulerpa racemosa*) POWDERED DRINK ON OXIDATIVE STRESS AND INFLAMMATION IN DIET-INDUCED OBESITY RAT MODELS

Dwi Santy Damayati[1,4*], Evy Damayanthi[1], Hadi Riyadi[1], I Wayan Teguh Wibawan[2], Ekowati Handharyani[3], Ainulyakin Imlani[5], Caddoumy Zahrel Ben[6], Fahri Sinulingga[6,7]

[1]Department of Nutrition Science, Faculty of Human Ecology, IPB University, Bogor, Indonesia 16680
[2]Department of Animal Infectious Diseases and Veterinary Public Health,
The School of Veterinary Medicine and Biomedical Sciences, IPB University, Bogor, Indonesia 16680
[3]Department of Clinic, Reproduction and Pathology,
The School of Veterinary Medicine and Biomedical Sciences, IPB University, Bogor, Indonesia 16680
[4]Department of Public Health, Faculty of Medicine dan Health Sciences,
Alauddin State Islamic University of Makassar, Gowa, Indonesia 92113
[5]Department of Aquaculture, College of Fisheries, Mindanao State University,
Tawi-Tawi College of Technology and Oceanography, Sanga-Sanga, Bongao Tawi-Tawi, Philippines 7500
[6]Department of Aquatic Product Technology,
Faculty of Fisheries and Marine Sciences, IPB University, Bogor, Indonesia 16680
[7]Fisheries Product Technology Study Program,
Faculty of Agriculture, University of Sriwijaya, Palembang, Indonesia 30662

Submitted: 3 August 2025/Accepted: 30 December 2025
*Correspondence: santy@uin-alauddin.ac.id

How to cite (APA Style 7th): Damayati, D. S., Damayanthi, E., Riyadi, H., Wibawan, I. W. T., Handharyani, E., Imlani, A., Ben, C. Z., & Sinulingga, F. (2026). The effect of sea grape (*Caulerpa racemosa*) powdered drink on oxidative stress and inflammation in diet-induced obesity rat models. *Jurnal Pengolahan Hasil Perikanan Indonesia,* 29(2), 106-118. http://dx.doi.org/10.17844/ms5t6f55

Abstract

The increasing incidence of obesity is a significant public health concern. The accumulation of excessive fat in obese individuals leads to an increase in beta-oxidation and esterification, which in turn causes inflammation and oxidative stress, eventually leading to cell death in the liver. This study aimed to examine the potential of a sea grape powder drink (SGPD) derived from *Caulerpa racemosa* in mitigating inflammation and oxidative stress in the livers of obese rats. This study employed an in vivo research design utilizing a complete randomized design (CRD) with 24 male Wistar rats weighing 150–250 g. The rats were divided into six groups, designated as follows: Standard Ration (SR), high-fat ration (HFR) + CMC, HFR + Orlistat 10.8 mg/kg, HFR + SGPD 1 g, HFR + SGPD 1.5 g, and HFR + SGPD 2 g. The results showed that the HFR + SGPD group exhibited the most pro-nounced obesity. The HFR + SGPD group administered a 2 g dose exhibited a reduction in TNF-α levels compa-rable to that of the RTL + Orlistat group ($p<0.05$). Immunohistochemistry of TNF-α in rat liver elucidated the effect of SGPD on TNF-α-induced liver inflammation. Nevertheless, the effect of SGPD on superoxide dismutase (SOD) and malondialdehyde (MDA) showed only a tendency toward improvement across various treatment groups. The study concluded that SGPD can mitigate liver inflammation in obese rats. SGPD has antioxidant potential, making it a promising therapeutic alternative for obesity.

Keywords: beta-oxidation, MDA, obesity, powder drink, TNF-α

Pengaruh Minuman Bubuk Anggur Laut (*Caulerpa racemosa*) terhadap Stres Oksidatif dan Inflamasi pada Model Tikus Obesitas yang Diinduksi Diet

Abstrak

Insiden obesitas yang terus meningkat menjadi masalah kesehatan masyarakat yang signifikan. Akumulasi lemak berlebih pada individu obesitas menyebabkan peningkatan beta-oksidasi dan esterifikasi, yang pada gilirannya memicu terjadinya inflamasi dan stres oksidatif hingga kematian sel pada hati. Tujuan penelitian ini adalah untuk mengkaji potensi minuman bubuk anggur laut (*sea grape powder drink*/ SGPD) yang berasal dari *Caulerpa racemosa* dalam menurunkan inflamasi dan stres oksidatif pada hati tikus obesitas. Penelitian ini menggunakan desain in vivo dengan rancangan acak lengkap (RAL) yang melibatkan 24 ekor tikus jantan Wistar dengan berat 150–250 g. Tikus dibagi menjadi enam kelompok, yaitu: pakan standar (SR), pakan tinggi lemak (HFR) + CMC, HFR + Orlistat 10,8 mg/kg, HFR + SGPD 1 g, HFR + SGPD 1,5 g, dan HFR + SGPD 2 g. Hasil penelitian menunjukkan bahwa kelompok HFR + SGPD mengalami obesitas paling nyata. Kelompok HFR + SGPD dosis 2 g menunjukkan penurunan kadar TNF-α yang sebanding dengan kelompok HFR + Orlistat (p<0,05). Analisis imunohistokimia TNF-α pada hati tikus menjelaskan efek SGPD terhadap inflamasi jaringan hati yang diinduksi TNF-α. Meskipun demikian, pengaruh SGPD terhadap kadar superoksida dismutase (SOD) dan malondialdehid (MDA) hanya menunjukkan kecenderungan perbaikan pada berbagai kelompok perlakuan. Kesimpulan dari penelitian ini adalah bahwa SGPD dapat menurunkan inflamasi hati pada tikus obesitas. Minuman bubuk anggur laut (SGPD) memiliki potensi antioksidan sehingga menjadikannya produk terapi alternatif yang menjanjikan untuk mengatasi obesitas.

Kata kunci: beta-oksidasi, MDA, minuman bubuk, obesitas, TNF-α

INTRODUCTION

Obesity is a non-communicable disease that constitutes the triple burden of malnutrition and has become a significant public health concern (Rah *et al.*, 2021). The prevalence of obesity worldwide is estimated to be 1.9 billion adults (WHO 2020) and the age-related decline in the capacity of adipose tissue to release lipids (Arner *et al.*, 2019). In vivo studies have shown that the consumption of high-fat foods triggers inflammatory responses and oxidative stress (Wu *et al.*, 2016). In response to this growing issue, multiple strategies have been implemented to address obesity. Conventional approaches include calorie restriction, which has been found to be less effective over extended periods when not combined with other strategies (Hassan & El-Gharib, 2015). Synthetic drugs such as orlistat, an inhibitor of pancreatic lipase that reduces intestinal fat absorption, and sibutramine, an appetite suppressant, are linked to adverse effects, including hypertension, constipation, headache, dry mouth, and insomnia. Surgical interventions are costly and may cause further complications (Pan *et al.*, 2016). As an alternative, the biomedical industry is developing functional foods and nutraceutical products containing antioxidants and anti-inflammatory compounds derived from seaweed (Lomartire *et al.*, 2021). A previous study showed that *Caulerpa* sp. has mild antioxidant properties when extracted with methanol (Hidayat *et al.*, 2020). Another study optimized the extraction method and showed that antioxidants can be improved by using DES for extraction at 50.50±0.067 mg TE/g dry weight (Satyantini *et al.*, 2025). Antioxidant properties have also been shown to improve liver function (Ezhilarasan & Lakshmi, 2022). One example is the sea grape (*Caulerpa* sp.), which contains bioactive compounds such as terpenoids, polyphenols, and flavonoids that exhibit antioxidant properties and have the potential to improve lipid profiles, reduce inflammation, and combat obesity (Ganesan *et al.*, 2019)

An imbalance between energy input and expenditure results in the accumulation of fat in adipose and other tissues, including the liver and skeletal muscle, leading to the development of chronic inflammation (Vulchi *et al.*, 2023). Adipose tissue is an endocrine organ that plays a role in inducing endocrine, metabolic, and inflammatory signals and contains many immune cells (Karundeng *et al.*, 2014; Vulchi *et al.*, 2023). The continuous accumulation of excessive fat leads to an increase in macrophage immune cells during obesity (Saltiel and Olefsky, 2017).

In addition to the role of adipose tissue, the maintenance of metabolic homeostasis and regulation of inflammation in obese individuals are also dependent on liver function (Saltiel & Olefsky, 2017). Excessive fat accumulation in the circulation results in the entry of fatty acids into the liver, leading to an increase in beta oxidation and esterification. However, this is not balanced by the formation of very low-density lipoprotein (VLDL) as a triglyceride transporter, resulting in the accumulation of fat in the liver (Heeren & Scheja, 2021). The abundance of triglycerides in the liver increases the demand for electron transport chains in the mitochondria, leading to the generation of free radicals and increased oxidative stress. Additionally, the excessive protein synthesis required for lipoprotein transporter formation contributes to the increased production of free oxygen radicals, further exacerbating oxidative stress (Niharika *et al.*, 2017).

An increase in free fatty acids in hepatocytes results in the production of oxidizing derivatives, which contribute to the generation of proinflammatory cytokines, including TNF-α and IL-6. This, in turn, leads to an increase in oxidative stress and the formation of lipid peroxidation products in hepatocyte membranes, which contribute to the development of inflammation and liver fibrosis (Okechukwu *et al.*, 2019). Kupffer cells, which are resident hepatic macrophages, play an important role in energy regulation and can activate inflammatory pathways during obesity (Saltiel & Olefsky, 2017). Elevated levels of inflammatory cytokines, such as TNF-α, are indicative of the progression of fatty liver disease in individuals with obesity (Varra *et al.*, 2025). In vivo studies have shown that a high-fat diet is associated with increased serum TNF-α and malondialdehyde (MDA) levels in obese mice (Wu *et al.*, 2016).

In vivo studies have demonstrated that *Caulerpa lentilifera* can improve inflammatory cells in Wistar rats induced with metabolic syndrome over a 16-week period (Preez *et al.*, 2020). Additional research on RAW 264.7 cells indicated that *Caulerpa lentilifera* extract can downregulate the expression and production of proinflammatory cytokines IL-6 and TNF-α (Yoojam *et al.*, 2021). With such remarkable properties, *Caulerpa* sp. can be maximized as a food product. One study investigated the acceptance of *Caulrepa*-based yoghurt, which yielded favorable results (Setiadi & Husni 2024). Accordingly, the present study sought to investigate the potential of sea grape powder drinks prepared from a sea grape species distinct from those used in previous studies. This study aimed to evaluate the effects of a sea grape (*Caulerpa racemosa*) powder drink on inflammation and oxidative stress in the liver of diet-induced obese rats, as indicated by tumor necrosis factor-alpha (TNF-α), malondialdehyde (MDA), and superoxide dismutase (SOD) levels.

METHODS

Sample Preparation

Sea grapes were procured from ponds situated within Takalar Regency, South Sulawesi Province, Indonesia. The sea grapes were then transformed into sea grape powder using a vacuum evaporator operating at a temperature of 40 °C within the PAU Laboratory of IPB University. The liver was pre-primed using an ELISA kit (BioTek 800 TS, Sigma-Aldrich). TNF-α (RAB0480, USA) was placed on a microplate using a micropipette to analyze TNF-α-mediated inflammation in the rat liver, and the resulting signal was read using an ELISA reader (800 TS). Liver tissue preparation was conducted using a Biocare kit to describe the inflammatory histology in rat liver using the TNF-α immunohistochemistry method, which was then read using an Olympus Digital BX51-P microscope. The Randox RX Monza SD 125 kit was used to prepare rat liver organs before analysis using a spectrophotometer (PerkinElmer).

Evaluation of Antioxidant Properties

The analysis of bioactive compounds from sea grape powder administered to rats consisted of an examination of antioxidants, which consisted of beta-carotene, carotene, phenol, and flavonoid content using a colorimetric method. Beta-carotene analysis was conducted using HPLC (Waters Alliance e2695) at SIG, Bogor, Indonesia. Carotenoid, phenol, and flavonoid contents were analyzed using a spectrophotometer (PerkinElmer) at the Biochemistry Laboratory of the Faculty of Medicine and Health Sciences at Alauddin State Islamic University of Makassar.

Animal Experiment

The experimental research design employed a completely randomized design consisting of six treatment groups. The animal model employed was male Wistar rats (*Rattus norvergicus*), with an initial body weight of 150–250 g and an age of two (2) to two and a half (2.5) months. Rats were procured from PT Biomedical Technology, Indonesia. The sample size was determined using the Federer formula (1966), which yielded 24 rats. The rats were immobilized for 14 days in an environment that was alternately illuminated for12 hours per day. The subjects were provided with a high-fat diet (46% fat) for 60 days. Subsequently, the obese rats were administered SGPD for 21 days. The rats were anesthetized using a combination of ketamine (80 mg/kg bw) and xylazine (5 mg/kg bw) on the day following the conclusion of the treatment phase, after which liver samples were collected to determine oxidative stress, antioxidant activity, and inflammatory markers. This study was approved by the Animal Ethics Committee of the School of Veterinary Medicine and Biomedical Sciences (SKHB), IPB University (reference number: 015/KEH/SKE/VI/2021).

Animal Feed

During the acclimatization phase, all experimental groups were provided standard feed and water ad libitum. Subsequently, for a period of eight weeks, all groups were provided with a high-fat diet, except the standard feed group, until they reached an index value of greater than 300 g/cm. The rats were divided into three control groups: standard rations (SR), high-fat ration (HFR)+carboxymethyl cellulose (CMC), and HFR+ Orlistat 10.8 mg/kg, as well as HFR, and three treatment groups: HFR+sea grape powder drink (SGPD) 1 g, HFR+SGPD 1.5 g, and HFR+SGPD 2 g. The control group received orlistat orally for 21 days, whereas the treatment groups received sea grape powder orally for the same duration.

Statistical Analysis

The effect of sea grape powder drink on inflammatory data was analyzed using a completely randomized design (CRD) with a single factor and six treatments, with three replications for each treatment. Statistical analyses were performed using Microsoft Excel and SPSS Version 22, and the data are expressed as the mean ± standard error. Differences among the treatment groups were evaluated using one-way analysis of variance (ANOVA). When a significant effect was detected ($p<0.05$), the analysis was followed by Duncan's multiple range test to determine differences between groups.

RESULT AND DISCUSSION

Antioxidant Content

Antioxidants play a crucial role in protecting biological systems from oxidative stress, which is associated with various diseases. In this study, the antioxidant composition of SGPD administered to rats was analyzed to assess its potential protective effects. The results of this analysis are presented in Table 1. Antioxidants play a crucial role in protecting biological systems from oxidative stress, which is associated with various diseases. In this study, the antioxidant composition of SGPD administered to rats was analyzed to assess its potential protective effects. The results of this analysis are presented in Table 1.

The antioxidant properties of *Caulerpa lentilifera*, commonly known as sea grapes, have been extensively studied, revealing its potential as a functional food with significant health benefits. Various extraction methods and conditions have been shown to influence antioxidant activity, with findings indicating that different solvents yield varying levels of bioactive compounds. One study reported that the total phenolic content in methanol extracts was 1.54 mg GAE/g, while flavonoids were only detected in methanol extracts at 1.16 mg QE/g (Diharmi *et al.*, 2024). Another study reported that the total phenolic content of *Caulerpa lentillifera* ranged from 1.30–2.04 mg GAE/g

http://dx.doi.org/10.17844/ms5t6f55

Table 1 Antioxidant compound content of sea grape powder drink

Content (mg/g)	Total
Carotene	1.25±0.29
Phenol	2.86±0.06
Flavonoids	1.38±0.03

Antioxidant assay was performed with three replications

with different treatments (Nguyen *et al.*, 2011) and the total flavonoid content ranged from 1.17–1.47 mg GAE/g (Peerakietkhajorn *et al.*, 2024). The carotene compounds have different contents based on the specific compound in the carotene group. The highest compound yielded in *Caulerpa lentillifera* was β-carotene at 19.5 mg/g, followed by canthaxanthin and astaxanthin, which yielded 14.6 and 3.6 mg/g, respectively (Balasubramaniam *et al.*, 2020).

The antioxidant compounds mentioned above play a crucial role in enhancing liver function by mitigating oxidative stress and promoting hepatoprotection. The liver, a primary detoxifying organ, is susceptible to damage by free radicals generated during metabolic processes. As the primary detoxifying organ, the liver is highly susceptible to damage by free radicals generated during metabolic processes. Antioxidants help restore oxidative balance by neutralizing free radicals, reducing lipid peroxidation, and enhancing intrinsic antioxidant defense systems, such as superoxide dismutase, thereby preventing hepatocyte damage and liver dysfunction (Patil *et al.*, 2014; Casas-Grajales & Muriel, 2015). These compounds have also demonstrated clinical benefits, such as improving graft function and patient survival in liver transplantation by reducing ischemia-reperfusion injury (Patil *et al.*, 2014). Furthermore, natural antioxidants from dietary sources, including fruits and vegetables, have been shown to be effective in supporting liver health and preventing diseases (Gheorghe *et al.*, 2019). However, while antioxidants show promise in improving liver health, caution is warranted due to potential pro-oxidant effects, emphasizing the need for further research to clarify their clinical applications and interactions with other treatments (Das *et al.*, 2024).

Phenolic acids and phenols have been shown to improve liver function through their antioxidant properties. A study demonstrated that phenolic acids protect liver cells from oxidative stress caused by iron overload by activating nuclear factor erythroid-2-related factor 2 (Nrf2), which regulates antioxidant genes (Kose *et al.*, 2023). Another study found that phenolic acids upregulate hepatic antioxidant enzymes, such as superoxide dismutase (SOD) and glutathione peroxidase (GPx), enhancing the liver's defense against oxidative damage (Machado *et al.*, 2023). Additionally, another study suggested that their hydrophilic nature enhances their hepatoprotective effects, making them more bioavailable for liver protection (Kandalintseva *et al.*, 2002). These findings indicate that dietary intake or supplementation with phenolic acids may serve as a natural therapeutic strategy for liver disorders caused by oxidative stress.

The antioxidant mechanisms of flavonoids are similar. Flavonoids exhibit strong antioxidant properties that contribute significantly to liver protection and regeneration. These compounds enhance hepatic antioxidant defenses by increasing the levels of superoxide dismutase (SOD), glutathione peroxidase (GPx), and glutathione-S-transferase, thereby reducing oxidative stress, which is a key factor in liver diseases (Wan & Jiang, 2018). Additionally, flavonoids regulate liver metabolism and inflammatory pathways by inhibiting tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6) and modulating apoptosis and autophagy through the phosphatidylinositol 3-kinase/protein kinase B (PI3K/Akt) signaling pathway. The hepatoprotective effects of flavonoids extend to their role in the nuclear factor erythroid 2-related factor 2 (Nrf2) pathway, where they activate antioxidant response elements (AREs), reducing liver inflammation and fibrosis and promoting mitochondrial function (Sharma *et al.*, 2024). Studies on specific flavonoids, such as those extracted from Buchholzia coriacea, indicate their ability to improve liver biomarkers, reduce oxidative damage, and protect against metabolic disorders, such as diabetes-induced liver stress (Amaralam *et al.*, 2025). Moreover, synthetic flavones, such as 6,3'-dimethoxy flavone (DMF), exhibit hepatoprotective effects against drug-induced toxicity, comparable to standard treatments such as silymarin (Abou Baker, 2022). These findings underscore the potential of flavonoids as therapeutic agents for liver diseases, highlighting their promise for future drug development.

Anti-inflamation Activity

Table 2 illustrates that the administration of HFR + SGPD 2 g influenced the decrease in TNF-α levels in the livers of obese rats. The higher the dosage of sea grapes administered, the greater the decrease, comparable to the effect observed with HFR + Orlistat administration. This finding aligns with the results of another study, which showed that eight weeks of sea grape administration can improve TNF-α parameters in the liver and heart.

Table 2 illustrates that the administration of HFR+SGPD 2 g decreased TNF-α levels in the livers of obese rats. The higher the dosage of sea grapes administered, the greater the decrease, comparable to the effect observed with RTL + Orlistat administration. Statistical analysis showed that the HFR + SGPD 2 g treatment produced significantly different results compared to the other treatments ($p < 0.05$), demonstrating better MDA prevention and higher SOD activity. This finding aligns with the results of a study by Preez *et al.* (2020), who showed that eight weeks of sea grape administration can improve TNF-α parameters in the liver and heart. Epidemiological studies have demonstrated a negative correlation between body mass and plasma carotene levels (Gunanti *et al.*, 2014). During obesity, carotene is sequestered in the adipose tissue, leading to a reduction in its plasma concentration (Okechukuwu *et al.*, 2019). Consequently, the body requires carotene intake as an anti-obesity agent, which has been linked to pro-vitamin A and has the potential to mitigate oxidative stress (Hamulka *et al.*, 2023). Table 2 shows that sea grape administration had no discernible effect on MDA and SOD levels across all treatment groups. However, there was a notable tendency for a decrease in MDA levels and an increase in SOD activity in the livers of obese rats. These results differ from those of previous studies, which showed a decrease in MDA levels following the administration of sea grapes for eight weeks in Wistar rats (Julyasih *et al.*, 2017). This can be attributed to the longer treatment duration, which allowed more pronounced differences to emerge.

The sea grape powder drink exhibited anti-inflammatory effects due to the presence of bioactive compounds, namely phenols and carotenes, which can act as antioxidants. A

Table 2 The effect sea grape powder drink on inflammatory (TNF-α) and oxidatives stress (MDA and SOD) markers

Group	TNF-α (pg/ml)	MDA (nmol/mL)	SOD (U/mL)
SR	1758.44±399[a]	4429.75±152[b]	267.72±16[bc]
HFR	1358.85±217[a]	4445.25±408[b]	227.69±8[a]
HFR + Orlistat	1698.95±551[a]	4793.00±646[b]	244.47±22[ab]
HFR + SGPD 1 g	1690.09±115[a]	5953.50±141[a]	263.83±6[bc]
HFR + SGPD 1.5 g	1442.63±153[a]	4603.50±531[b]	250.76±26[ab]
HFR + SGPD 2 g	−125.57±58[b]	3685.00±111[c]	283.18±11[c]
p-value	0.002*	0.241	0.301

Distinct superscript letters within the same column denote statistically significant differences; The inflammatory and oxidative assays were conducted in triplicate.

study revealed that the phenolic derivatives present in *Caulerpa* sp. include gallic acid, catechins, isoquercetin tannic acid, and quercetin (Yoojam *et al.*, 2021). Phenolic compounds have been demonstrated to suppress inflammation and oxidative stress by suppressing the regulation of NFkB gene expression and modulating the control of antioxidative signals through Nrf2 regulation. Additionally, they have been shown to reduce de novo lipogenesis by suppressing the regulation of the SREB1c gene, thereby reducing the accumulation of excess fatty acids and, consequently, lipid peroxidation, which is also a factor causing inflammation (Rodriguez-Ramiro *et al.*, 2016; Lu *et al.*, 2023). Catechins and their derivatives can also reduce oxidative stress directly by functioning as traps and antidotes for free radicals or indirectly by increasing superoxide dismutase (SOD) levels and reducing malondialdehyde (MDA) levels in the liver (Bernatoniene & Kopustinskiene, 2018). Epigallocatechin is a treatment for fatty liver disease because it is a hepatoprotector (Chen *et al.*, 2018).

Additionally, SGPD contains carotene, another antioxidant. Previous studies have demonstrated that carotene can reduce cytochrome P450 activity and the expression of pro-inflammatory cytokines at the mRNA level. Furthermore, it has been shown to impede IkB Kinase (IKK) activity, which is responsible for regulating NFkB activity and downregulating inflammatory genes such as TNF α (Tolares *et al.*, 2019).

Carotene functions as a precursor of retinoic acid, which binds to nuclear receptors, namely, the retinoic acid receptor (RAR) and retinoid X receptor (RXR). These receptors form heterodimers with peroxisome proliferator-activated receptors (PPAR) and other nuclear receptors, thereby playing a role in whole-body glucose regulation. This occurs through two main mechanisms: increased insulin sensitivity and insulin release (Saeed *et al.*, 2017; Harari *et al.*, 2020). Additionally, carotene exerts protective effects through its antioxidant capacity (Jurnalis *et al.*, 2014). Carotene can bind to reactive species, such as singlet oxygen and peroxyl radicals, and has been shown to increase Nrf2 levels (Luisa Bonet *et al.*, 2015).

An imbalance between free radicals and antioxidants results in oxidative stress, characterized by increased MDA levels (Elsayed *et al.*, 2019). The continuous occurrence of oxidative stress reactions results in an elevation of lipid peroxides, which impairs mitochondrial function and triggers the induction of TNF-α (Chernyavskij *et al.*, 2023). Fat accumulation results from an imbalance between the intake, synthesis, degradation, and secretion of fat in the liver. Fatty acids for triacylglycerol synthesis are obtained from de novo formation in the liver, portal blood as free fatty acids, and circulating lipoproteins, especially chylomicrons (Sinulingga *et al.* 2024). Serum-free fatty acids are increased by accelerated lipolysis in the peripheral adipose tissue and visceral fat (Kucera *et al.*, 2014).

Immunohistochemical images obtained at 40x magnification of liver tissue demonstrated the expression of TNF-α in hepatocytes A and B, as indicated by brown coloration within the cell nucleus and cytoplasm. Tumor necrosis factor-α (TNF-α) is mainly produced by the monocyte-macrophage lineage in the liver, with Kupffer cells representing the predominant population in this lineage (Jiang *et al.*, 2016). The appearance of brown coloration, indicative of a positive immunoreactive result, is a consequence of the immunohistochemical (IHK) staining process. During this process, antigen in the form of the TNF-α cytokine in the cell cytoplasm binds to the primary antibody (Rat Anti TNF-α) and is then labeled by secondary antibodies.

Once the binding process was complete, the diaminobenzidine (DAB) substrate was added, resulting in the production of a brown coloration indicative of the presence of the cytokine (TNF-α) within the cell cytoplasm. The expression level of TNF-α in liver tissue is a crucial marker for evaluating both the normal and diseased states of hepatocytes (Mohallem *et al.*, 2021). Tumor necrosis factor-α (TNF) is an endogenous soluble molecule that causes necrosis in solid tumors.

TNF is a major inflammatory marker cytokine linked to the immunopathogenesis of several autoimmune diseases. TNF causes hepatocyte death and necroptosis, as well as hepatic inflammation, regeneration, autoimmunity, and the progression of hepatocellular cancer in the liver (Tiegs & Horst, 2022).

TNF-α Immunohistochemistry

Inflammation plays a crucial role in metabolic disorders, including obesity-induced liver dysfunction. Tumor necrosis factor-alpha (TNF-α) is a key pro-inflammatory cytokine involved in hepatic inflammation, and its expression can be assessed using immunohistochemistry. The presence of TNF-α in liver tissue was indicated by brown staining in the nucleus and cytoplasm of the cells. A reduction in TNF-α expression suggests a potential anti-inflammatory effect of the treatment. In this study, the effect of sea grape powder on TNF-α expression in rat liver tissue was evaluated, and the results are shown in Fig. 1.

Figure 1 illustrates the effect of sea grape powder on TNF-α expression in rat liver tissue, as observed through immunohistochemical analysis. TNF-α presence was indicated by brown staining within the nucleus and cytoplasm, reflecting the level of inflammation. In the control group (a), a strong brown coloration was observed, suggesting high TNF-α expression and significant inflammation. Similarly, the commercial treatment groups (b and c) exhibited noticeable TNF-α staining, although with some reduction compared to the control. In contrast, the sea grape treatment groups (d, e, and f) showed visibly lower brown staining intensity, indicating a potential anti-inflammatory effect. The reduction in TNF-α expression suggests that sea grape powder may mitigate inflammation in diet-induced obese rats, consistent with its known bioactive properties. These findings support the potential of sea grape powder as a functional food ingredient for managing inflammation.

TNF-α is a proinflammatory cytokine released by Kupffer cells, which are magrophage (m1) caused by fat overload in the liver (Su *et al.*, 2018). In the liver, KCs respond to two main types of stimuli: intrahepatic danger-associated molecular patterns (DAMPs), which are released by infiltrated and damaged hepatocytes, and gut-derived bacterial antigens, also known as pathogen-associated molecular patterns (PAMPs), which travel from the gut to the liver owing to a damaged gut epithelial barrier (Vachliotis & Polyzos, 2023). A significant

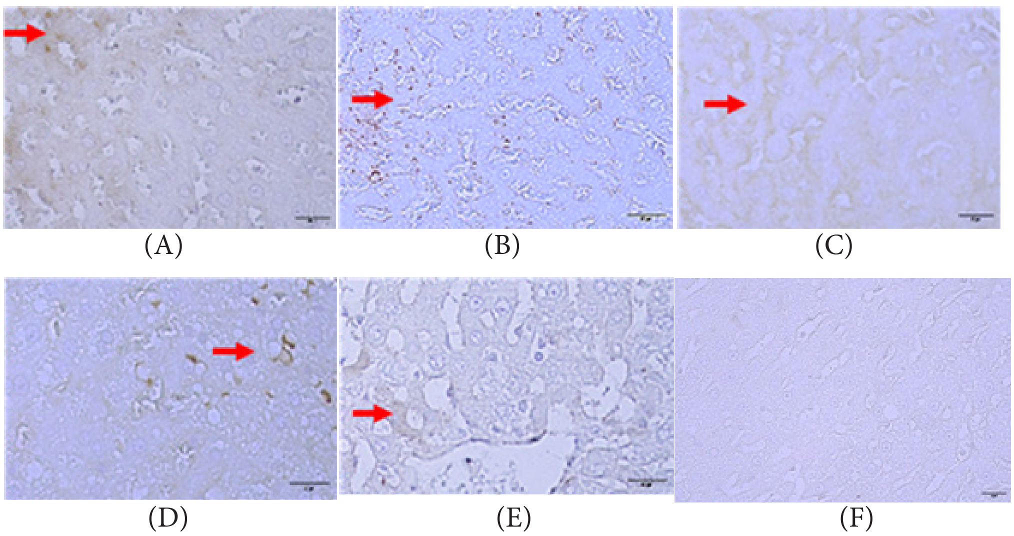

Figure 1 Illustrates the impact of sea grape powder (*Caulerpa racemosa*) on TNF-α immunohistochemistry in rat liver tissue. The groups are as follows: RS group (A); RTL+CMC group (B); RTL+Orlistat 10.8 mg/kg group (C); RTL+SGPD 1 g group (D); RTL+SGPD 1.5 g group (); and RTL+SGPD 2 g group (F). The red arrows indicate the presence of TNF-α.

signaling mechanism for the transcription of TNF-α, as well as other cytokines and chemokines, DAMPs, and PAMPs, is binding to Toll-like receptors (TLRs) on the surface of KCs and activating the nuclear factor-kappa B (NF-κB) intracellular pathway (Liu *et al.*, 2017). TNF-α can induce the accumulation of reactive oxygen species (ROS) in the liver, potentially resulting in hepatic damage (Yang, 2023).

The results of this study showed no visible expression of TNF-α in Figures 1F hepatocyte tissue. These results suggest that SGPD exerts an anti-inflammatory effect. According to the findings of earlier studies, SPGD possesses antioxidant properties and has the potential to serve as an alternative treatment for obesity (Damayati *et al.*, 2023). Antioxidants have emerged as promising therapeutic agents because they mitigate oxidative stress and modulate inflammatory pathways (Bhol *et al.*, 2024). Antioxidants reduce inflammation by inhibiting the expression of tumor necrosis factor-alpha (TNF-α), a pro-inflammatory cytokine. This inhibition occurs at the transcriptional level by blocking the activation of NF-κB, which regulates TNF-α transcription (Mucha *et al.*, 2021). Green seaweed can also inhibit the release of cytokines and inflammatory mediators, such as AMPK, mTOR, IL4, and TNF-α (Prayogo *et al.*, 2024).

The results of this study are consistent with those of a previous study, which showed that the administration of lycopene can downregulate the expression of TNF-α, thereby reducing fatty liver infiltration and improving histopathological changes, with the degree of improvement depending on the administered dose (Jiang *et al.*, 2016). Lycopene is a carotene-derived antioxidant that is both acyclic and nonpolar (Bin-Jumah *et al.*, 2022). It is postulated that the increased levels of antioxidant enzymes and decreased lipid peroxide content observed in the presence of lycopene are important mechanisms by which this compound prevents the development of liver damage caused by a high-fat diet (Jiang *et al.*, 2016).

CONCLUSION

Sea grape (*Caulerpa racemosa*) powder drink (SGPD) supplementation effectively reduced inflammation and oxidative stress in the liver of diet-induced obese rats. The HFR + SGPD group, which received 2 g of SGPD, showed the most significant reduction in TNF-α levels from 1,758.44 to -125.57 pg/mL, exceeding the effect observed in the SR group. This group also demonstrated the greatest improvement in oxidative stress markers, with SOD activity increasing from 267.72 to 283.18 U/mL and MDA levels decreasing from 4,429.75 to 3,685.00 nmol/mL. These findings suggest that SGPD supplementation can reduce liver inflammation and oxidative stress associated with obesity, supporting its potential application as a functional food for obesity-related liver disorders.

REFERENCES

Abou Baker, D.H. (2022). An ethnopharmacological review on the therapeutical properties of flavonoids and their mechanisms of actions: A comprehensive review based on up-to-date knowledge. *Toxicology Reports*, 9, 445–469. https://doi.org/10.1016/j.toxrep.2022.03.011

Amaralam, C., Achi, K., Oyedemi, O., Aloh, S., & Ajah, O. (2025). Effects of flavonoid extract from wonderful kola (*Buchholzia coriacea*) seed on antioxidant and liver function biomarkers of fructose-fed streptozotocin-induced type 2 diabetic albino rats. *GSC Biological and Pharmaceutical Sciences*, 30(1), 163–168. https://doi.org/10.30574/gscbps.2025.30.1.0001

Arner, P., Bernard, S., Appelsved, L., Fu, K.Y., Andersson, D.P., Salehpour, M., Thorell, A., Rydén, M., & Spalding, K.L. (2019). Adipose lipid turnover and long-term changes in body weight. *Nature Medicine*, 25(9), 1385–1389. https://doi.org/10.1038/s41591-019-0565-5

Balasubramaniam, V., Chelyn, L.J., Vimala, S., Fairulnizal, M.N.M., Brownlee, I.A., & Amin, I. (2020). Carotenoid composition and antioxidant potential of *Eucheuma* *denticulatum*, *Sargassum polycystum* and *Caulerpa lentillifera*. *Heliyon*, 6(8), 1–8. https://doi.org/10.1016/j.heliyon.2020.e04654

Bernatoniene, J., & Kopustinskiene, D.M. (2018). The role of catechins in cellular responses to oxidative stress. *Molecules*, 23(4), 1–11. https://doi.org/10.3390/molecules23040965

Bhol, N.K., Bhanjadeo, M.M., Singh, A.K., Dash, U.C., Ojha, R.R., Majhi, S., Duttaroy, A.K., & Jena, A.B. (2024). The interplay between cytokines, inflammation, and antioxidants: mechanistic insights and therapeutic potentials of various antioxidants and anti-cytokine compounds. *Biomedicine & Pharmacotheraphy*, 178, 117177. https://doi.org/10.1016/j.biopha.2024.117177

Bin-Jumah, N.M., Nadeem, M.S., Gilani, S.J., Mubeen, B., Ullah, I., Alzarea, S.I., Ghoneim, M.M., Alshehri, S., Al-Abbasi, F.A., & Kazmi, I. (2022). Lycopene: A natural arsenal in the war against oxidative stress and cardiovascular diseases. *Antioxidants*, 11(2), 232. https://doi.org/10.3390/antiox11020232

Casas-Grajales, S., & Muriel, P. (2015). Antioxidants in liver health. *World Journal of Gastrointestinal Pharmacolology and Therapeutics*, 6(3), 59–72. https://doi.org/10.4292/wjgpt.v6.i3.59.

Chen, C., Liu, Q., Liu, L., Hu, Y.Y., & Feng, Q. (2018). Potential biological effects of epigallocatechin-3-gallate on the treatment of nonalcoholic fatty liver disease. *Molecular Nutrition Food Research*, 62(1), 1–11. https://doi.org/10.1002/mnfr.201700483.

Chernyavskij, D.A., Pletjushkina, O.Y., Kashtanova, A.V., Galkin, I.I., Karpukhina, A., Chernyak, B.V., Vassetzky, Y.S., & Popova, E.N. (2023). Mitochondrial oxidative stress and mitophagy activation contribute to TNF-dependent impairment of myogenesis. *Antioxidants*, 12(3), 602. https://doi.org/10.3390/antiox12030602

Coronel, J., Pinos, I., & Amengual, J. (2019). β-carotene in obesity research: Technical considerations and current status of the field. *Nutrients*, 11(4), 1–19. https://doi.org/10.3390/nu11040842

Damayati, D.S., Damayanthi, E., Riyadi, H., Wibawan, I.W.T., & Handharyani, E. (2023). The analysis of antioxidant capacities and sensory in sea grapes (*Caulerpa racemosa*) powdered drink as a therapeutic obesity. *Amerta Nutrition*, 7(2), 175–184. https://doi.org/10.20473/amnt.v7i2.2023.175-184

Das, S.K., Nerune, S.M., & Das, K.K. (2024). Antioxidant therapy for hepatic diseases: A double-edged sword. *Journal of Basic and Clinical Physiology and Pharmacolology*, 35(1–2), 7–14. https://doi.org/10.1515/jbcpp-2023-0156

Diharmi, A., Edison, Ilza, M., Dahlia, Saputra, R. (2024). Aktivitas antioksidan, total fenolik, flavonoid dan saponin anggur laut (*Caulerpa lentillifera*) diekstrak dengan pelarut yang berbeda polaritas. *Agrointek*, 18(3), 761–768. http://dx.doi.org/10.21107/agrointek.v18i3.12240

Elsayed Azab, A., A Adwas Almokhtar., Ibrahim Elsayed, A.S., A Adwas, A., Ibrahim Elsayed Ata Sedik., & Quwaydir, F.A. (2019). Oxidative stress and antioxidant mechanisms in human body. *Journal of Applied Biotechnology & Bioengineering*, 6(1), 43–47. https://doi.org/10.15406/jabb.2019.06.00173

Ezhilarasan, D., & Lakshmi, T. (2022). A molecular insight into the role of antioxidants in nonalcoholic fatty liver diseases. *Oxidative Medicine and Cellular Longevity*, 5, 1–15. https://doi.org/10.1155/2022/9233650

Ganesan, A.R., Tiwari, U., & Rajauria, G. (2019). Seaweed nutraceuticals and their therapeutic role in disease prevention. *Food Science and Human Wellness*, 8(3), 252–263. https://doi.org/10.1016/j.fshw.2019.08.001

Gheorghe, G., Stoian, A.P., Găman, M.-A., Socea, B., Neagu, T.P., Stănescu, A., Bratu, O., Mischianu, D., Suceveanu, A., & Diaconu, C. (2019). The benefits and risks of antioxidant treatment in liver diseases. *Revista de Chimie*, 70(2), 651–655. http://dx.doi.org/10.37358/

RC.19.2.6977

Gunanti, I.R., Marks, G.C., Al-Mamun, A., & Long, K.Z. (2014). Low serum concentrations of carotenoids and vitamin E are associated with high adiposity in Mexican-American children. *Journal of Nutrition*, 144(4), 489–495. https://doi.org/10.3945/jn.113.183137

Hamulka, J., Sulich, A., Górnicka, M., & Jeruszka-Bielak, M. (2023). Changes in plasma carotenoid concentrations during the AntioxObesity weight reduction program among adults with excessive body weight. *Nutrients*, 15(23), 4890. https://doi.org/10.3390/nu15234890

Harari, A., Coster, A.C.F., Jenkins, A., Xu, A., Greenfield, J.R., Harats, D., Shaish, A., & Samocha-Bonet, D. (2020). Obesity and insulin resistance are inversely associated with serum and adipose tissue carotenoid concentrations in adults. *The Journal of Nutrition*, 150(1), 38–46. https://doi.org/10.1093/jn/nxz184

Hassan, H.A., & El-Gharib, N.E. (2015). Obesity and clinical riskiness relationship: therapeutic management by dietary antioxidant supplementation—a review. *Applied Biochemistry and Biotechnology*, 176(3), 647–669. https://doi.org/10.1007/s12010-015-1602-6

Heeren, J., & Scheja, L. (2021). Metabolic-associated fatty liver disease and lipoprotein metabolism. *Molecular Metabolism*, 50, 101238. https://doi.org/10.1016/j.molmet.2021.101238

Hidayat, T., Nurjanah, N., Jacoeb, A. M., & Putera, B. A. (2020). Aktivitas antioksidan *Caulerpa* sp. segar dan rebus. *Jurnal Pengolahan Hasil Perikanan Indonesia*, 23(3), 566–575. https://doi.org/10.17844/jphpi.v23i3.33869

Jiang, W., Guo, M.H., & Hai, X. (2016). Hepatoprotective and antioxidant effects of lycopene on non-alcoholic fatty liver disease in rat. *World Journal of Gastroenterology*, 22(46), 10180–10188. https://doi.org/10.3748/wjg.v22.i46.10180

Julyasih, K.S.M., & Wirawan, I. (2017). Potential effect of macro alga *Caulerpa* sp. and *Gracilaria* sp. extract lowering malondialdehyde level of wistar rats fed high cholesterol diet. *International Journal of Biosciences and Biotechnology*, 5(1), 71–79. https://doi.org/10.24843/IJBB.2017.v05.i01.p06

Jurnalis, Y.D., Sayoeti, Y., & Elfitrimelly, E. (2014). Peran antioksidan pada *non alcoholic fatty liver disease* (NAFLD). *Jurnal Kesehatan Andalas*, 3(1), 15–20. https://doi.org/10.25077/jka.v3i1.18

Kandalintseva, N.V., Dyubchenko, O.I., Terakh, E.I., Prosenko, A.E., Shvarts, Y.Sh., & Dushkin, M.I. (2002). Antioxidant and hepatoprotector activity of water-soluble 4-propylphenols containing hydrophilic groups in alkyl chains. *Pharmaceutical Chemistry Journal*, 36, 177–180. https://doi.org/10.1023/A:1019876403624

Karundeng, R., & Wangko, S. (2014). Jaringan lemak putih dan jaringan lemak coklat. *Jurnal Biomedik*, 6(3), 8–16. https://doi.org/10.35790/jbm.6.3.2014.6328

Kose, T., Sharp, P.A., & Latunde-Dada, G.O. (2023). Phenolic acids rescue iron-induced damage in murine pancreatic cells and tissues. *Molecules*, 28(10), 4084. https://doi.org/10.3390/molecules28104084

Kucera, O., & Cervinkova, Z. 2014. Experimental models of non-alcoholic fatty liver disease in rats. *World Journal of Gastroenterology*, 20(26), 8364–8376. https://doi.org/10.3748/wjg.v20.i26.8364

Liu, T., Zhang, L., Joo, D., & Sun, C. (2017). NF-κB signaling in inflammation. *Signal Transduct Target Therapy*, 2, 17023. https://doi.org/10.1038/sigtrans.2017.23

Lomartire, S., Marques, J.C., & Gonçalves, A.M.M. (2021). An overview to the health benefits of seaweeds consumption. *Marine Drugs*, 19(6), 1-24. http://doi.org/10.3390/md19060341

Lu, Y., Zhang, C., Song, Y., Chen, L., Chen, X., Zheng, G., Yang, Y., Cao, P., & Qiu, Z. (2023). Gallic acid impairs fructose-driven de novo lipogenesis and ameliorates hepatic steatosis via AMPK-dependent suppression of SREBP-1/

ACC/FASN cascade. *European Journal of Pharmacology*, 940, 175457. https://doi.org/10.1016/j.ejphar.2022.175457

Luisa Bonet, M., Canas, J.A., Ribot, J., & Palou, A. (2015). Carotenoids and their conversion products in the control of adipocyte function, adiposity and obesity. *Archives of Biochemistry and Biophysics*, 572, 112–125. https://doi.org/10.1016/j.abb.2015.02.022

Machado, I.F., Miranda, R.G., Dorta, D.J., Rolo, A.P., & Palmeira, C.M. (2023). Targeting oxidative stress with polyphenols to fight liver diseases. *Antioxidants*, 12(6), 1212. https://doi.org/10.3390/antiox12061212

Mohallem, R., & Aryal, U.K. (2021). Quantitative proteomics and phosphoproteomics reveal TNF-α-mediated protein functions in hepatocytes. *Molecules*, 26(18), 5472. https://doi.org/10.3390/molecules26185472

Mucha, P., Skoczyńska, A., Małecka, M., Hikisz, P., & Budzisz, E. (2021). Overview of the antioxidant and anti-inflammatory activities of selected plant compounds and their metal ions complexes. *Molecules*, 26(16), 4886. https://doi.org/10.3390/molecules26164886

Nguyen, V.T., Ueng, J.-P., & Tsai, G.-J. (2011). Proximate composition, total phenolic content, and antioxidant activity of seagrape (*Caulerpa lentillifera*). *Journal of Food Science*, 76(7), 950–958. http://dx.doi.org/10.1111/j.1750-3841.2011.02289.x

Niharika, S., Sarah, T.A., Naga Chalasan., Ryan, M., Anderson., & Mirmira aghavendra, G. (2017). Molecular mechanisms of nonalcoholic fatty liver disease: Potential role for 12-lipoxygenase niharika. *Journal of Diabetes and its Complications*, 34(14), 1389–1395. https:/doi.org/10.1016/j.jdiacomp.2017.07.014.Molecular

Okechukwu, G., Nweke, O., Nwafor, A., Godson, A., Kenneth, E., & Ibegbu, A. (2019). Beta (β)-carotene-induced effects on the hepato-biochemical. *Jordan Journal of Biological Sciences*, 12(3), 283–288.

Pan, H., Gao, Y., & Tu, Y. (2016). Mechanisms of body weight reduction by black tea polyphenols. *Molecules*, 21(12), 1–24. https://doi.org/10.3390/molecules21121659

Patil, V.S., Suryakar, A.N., & Naik, P. (2014). Beneficial role of antioxidants during liver transplantation. *Al Ameen Journal of Medical Sciences*, 7(3), 224–228

Peerakietkhajorn, S., Worakit, W., Moukamnerd, C., & Tipbunjong, C. (2024). Proximate composition, phytochemical analysis and toxicity assessment of extracts of *Caulerpa lentillifera* using autoclave- and microwave-assisted extractions. *Sains Malaysiana*, 53(2), 335–345. http://doi.org/10.17576/jsm-2024-5302-08

Prayogo, E.W., Sholikhah, I., Suciati, S., Dej-adisai, S., & Widyowati, R. (2024). Systematic review of green seaweed *Caulerpa racemosa* as an anti-inflammatory agent: Current insights and future perspectives. *Jurnal Farmasi dan Ilmu Kefarmasian Indonesia*, 11(2), 156–173. https://doi.org/10.20473/jfiki.v11i22024.156-173

Preez, R., Majzoub, M.E., Thomas, T., Panchal, S.K., & Brown, L. (2020). *Caulerpa lentillifera* (Sea Grapes) improves cardiovascular and metabolic health of rats with diet-induced metabolic syndrome. *Metabolites*, 10, 2–18. https://doi.org/10.3390/metabo10120500

Rah, J.H., Melse-boonstra, A., Agustina, R., Van Zutphen, K.G., & Kraemer, K. (2021). The triple burden of malnutrition among adolescents in Indonesia. *Food and Nutrition Bulletin*, 42, 4–8. https://doi.org/10.1177/03795721211007114

Rodriguez-Ramiro, I., Vauzour, D., & Minihane, A.M. (2016). Polyphenols and non-alcoholic fatty liver disease: Impact and mechanisms. *Proceeding of the Nutrition Society*, 75(1), 47–60. https://doi.org/10.1017/S0029665115004218

Saeed, A., Dullaart, R.P.F., Schreuder, T.C.M.A., Blokzijl, H., & Faber, K.N. (2017). Disturbed vitamin A metabolism in non-alcoholic fatty liver disease (NAFLD). *Nutrient*, 10(29), 1–25. https:/doi.org/10.3390/nu10010029

Saltiel, A.R., & Olefsky, J.M. (2017).

Inflammatory mechanisms linking obesity and metabolic disease. *The Journal of Clinical Investigation*, 127(1), 1–4. https://doi.org/10.1172/JCI92035. systems

Satyantini, W. H., Izzata, A. K., Patmawati, P., Tjahjaningsih, W., Kurnia, K. A., & Md Yasin, I. S. (2025). Antioxidant and antibacterial activities of *Caulerpa racemosa* extract with a combination of solvents and variation of maceration time. *Jurnal Pengolahan Hasil Perikanan Indonesia*, 28(11), 981–992. https://doi.org/10.17844/wkb2jq46

Setiadi, M. K., & Husni, A. (2024). Aktivitas antioksidan dan tingkat penerimaan konsumen yoghurt yang diperkaya rumput laut *Caulerpa lentillifera*. *Jurnal Pengolahan Hasil Perikanan Indonesia*, 27(5), 417–430. https://doi.org/10.17844/jphpi.v27i5.53538

Sharma, R., Majee, C., Mazumder, R., Padhi, S., Khan, F., & Saxena Pal, R. (2024). Insight into the regulation of Nrf2/Keap1 pathway by flavonoids as an approach for treatment of liver diseases: A review. *Indian Journal of Pharmaceutical Education and Research*, 58(1), 40–57. https://doi.org/10.5530/ijper.58.1s.4

Sinulingga, F., Trilaksani, W., & Setyaningsih, I. (2024). Karakteristik fisikokimia tablet berbasis mikrokapsul minyak mata tuna dan spirulina. *Jurnal Pengolahan Hasil Perikanan Indonesia*, 27(1), 1-15. https://doi.org/10.17844/jphpi.v27i1.49473

Su, L., Li, N., Tang, H., & *et al*. (2018). Kupffer cell-derived TNF-α promotes hepatocytes to produce CXCL1 and mobilize neutrophils in response to necrotic cells. *Cell Death & Disease*, 9(323), 1–15. https://doi.org/10.1038/s41419-018-0377-4

Tiegs, G., & Horst, A.K. (2022). TNF in the liver: Targeting a central player in inflammation. *Seminars in Immunopathology*, 44, 445–459. https://doi.org/10.1007/s00281-022-00910-2

Tolares, E., Alonso, G., & Castón, P. (2019). Nutritional importance of carotenoids and their effect on liver health: A review. *Antioxidants*, 8(7), 1-23. https://doi.org/10.3390/antiox8070229

Vachliotis, I.D., & Polyzos, S.A. (2023). The role of tumor necrosis factor-alpha in the pathogenesis and treatment of nonalcoholic fatty liver disease. *Current Obesity Reports*, 12(3), 191–206. https://doi.org/10.1007/s13679-023-00519-y

Varra, F. N., Varras, M., Varra, V. K., & Theodosis-Nobelos, P. (2025). Mechanisms linking obesity with non-alcoholic fatty liver disease (NAFLD) and cardiovascular diseases (CVDs): The role of oxidative stress. *Current Issues in Molecular Biology*, 47(9), 766. https://doi.org/10.3390/cimb47090766

Vulchi, J., Suryadevara, V., Mohan, P., Kamalanathan, S., Sahoo, J., Naik, D., *et al*. (2023). Obesity and metabolic dysfunction-associated fatty liver disease: Understanding the intricate link. *Journal of Translational Gastroenterology*, 1(2), 74–86. https://doi.org/10.14218/JTG.2023.00043

Wan, L., & Jiang, J.-G. (2018). Protective effects of plant-derived flavonoids on hepatic injury. *Journal of Functional Foods*, 44, 283–291. https://doi.org/10.1016/j.jff.2018.03.015

WHO. (2020). Obesity and overweight. Retrieved from www who int. https://doi.org/10.1016/j.med.2020.07.010

Wu, P., Zhang, F., Dai, Y., Han, L., & Chen, S. (2016). Serum TNF-α, GTH and MDA of high-fat diet-induced obesity and obesity resistant rats. *Saudi Pharmaceutical Journal*, 24(3), 333–336. https://doi.org/10.1016/j.jsps.2016.04.011

Yang, Q. (2023). The role of reactive oxygen species (ROS) in the nandrolone-decanoate-induced liver fibrosis. *Theoretical and Natural Science*, 20(1), 262–267. https://doi.org/10.54254/2753-8818/20/20230788

Yoojam, S., Ontawong, A., Lailerd, N., Mengamphan, K., & Amornlerdpison, D. (2021). The enhancing immune response and anti-inflammatory effects of *Caulerpa lentillifera* extract in raw 264.7 cells. *Molecules*, 26(19), 1–12. https://doi.org/10.3390/molecules26195734