

The Role of β -1.3/1.6-D-Glucan From *Ganoderma lucidum* Mycelium Extract in Ulcerative Colitis

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ABSTRACT

Ulcerative colitis (UC) is an idiopathic inflammatory disease that affects the colon. Current pharmacological modalities to treat UC have various side effects; therefore, there is a demand to develop a new alternative medicine that can reduce side effects and increase drug efficacy. One candidate for alternative therapy is Polysaccharide Peptide which is extracted from *Ganoderma lucidum* mycelium. This Polysaccharide has an active compound of β -1,3/1,6-D-Glucan which has strong immunomodulatory and anti-inflammatory properties. Various studies have reported that *Ganoderma lucidum* polysaccharides can reduce inflammatory markers such as TNF- α , IFN- γ , and IL-17A, which is produced by colonic mucosal inflammation. In addition, β -1,3/1,6-D-Glucan has shown improvements in inflammatory parameters and intestinal immunological barrier function animal studies with artificial colitis and requires further research in humans before clinical applications.

Keywords: inflammatory bowel disease, ulcerative colitis, *Ganoderma lucidum*, herbal

ABSTRAK

Kolitis ulserativa (UC) adalah penyakit inflamasi idiopatik yang mempengaruhi usus besar. Modalitas farmakologis saat ini untuk mengobati UC memiliki berbagai efek samping, oleh karena itu, ada kesempatan untuk mengembangkan pengobatan alternatif baru yang dapat mengurangi efek samping dan meningkatkan khasiat obat. Salah satu kandidat terapi alternatif adalah Polisakarida Peptida yang diekstrak dari miselium *Ganoderma lucidum*. Polisakarida ini memiliki senyawa aktif β -1,3/1,6-D-Glucan yang memiliki sifat imunomodulator dan anti inflamasi yang kuat. Berbagai penelitian telah melaporkan bahwa polisakarida *Ganoderma lucidum* dapat mengurangi penanda inflamasi seperti TNF- α , IFN- γ , dan IL-17A, yang dihasilkan oleh inflamasi mukosa kolon. Selain itu, β -1,3/1,6-D-Glucan telah menunjukkan perbaikan dalam parameter inflamasi dan studi hewan fungsi penghalang imunologi usus dengan kolitis buatan dan memerlukan penelitian lebih lanjut pada manusia sebelum aplikasi klinis.

Kata kunci: penyakit radang usus, kolitis ulserativa, *Ganoderma lucidum*, herbal

INTRODUCTION

Inflammatory bowel disease (IBD) is an idiopathic inflammatory disease characterized by chronic inflammation of the gastrointestinal tract and relapses.¹ IBD is classified into three types, namely ulcerative colitis (UC), Crohn's disease (CD), and intermediate colitis (IC). This classification is based on differences in the pathology and clinical manifestations that appear. The most common clinical manifestations in IBD patients are chronic diarrhea with or without blood, abdominal pain, and weight loss.²

IBD is a disease with a high incidence and prevalence in Europe and North America. The average incidence of ulcerative colitis is 0.55 per 100,000 population.³ The cause of UC is still unknown; however, many studies have found correlations to be a combination between genetic, environmental, diet, and autoimmune disorders.⁴ Treatment of UC consists of diet modification, lifestyle improvement, stress reduction, and medications. Available drugs that can be used to treat IBD symptoms include anti-inflammatory drugs 5-aminosalicylic acid (5-ASA), immunomodulators (azathioprine, mercaptopurine, methotrexate), and biological agents.⁵

An herbal alternative derived from *Ganoderma lucidum* mushroom can be used for adjunctive therapy in patients with ulcerative colitis. The polysaccharides inside the fungus have anti-inflammatory and immunomodulatory characteristics, which can reduce the production of TNF- α , IFN- γ , IL-17A. These inflammation factors are produced by colonic mucosal inflammation in patients with ulcerative colitis; therefore, *Ganoderma lucidum* might be helpful to be used as adjunctive therapy for ulcerative colitis.^{6,7} In addition, a study done by Silva et al stated that *Ganoderma lucidum* could be used as an alternative dietary approach for the prevention of colitis-associated cancer.^{8,9}

Inflammatory Bowel Disease

Inflammatory bowel disease (IBD) is comprised of three major disorders: ulcerative colitis (UC), Crohn's disease (CD), indeterminate colitis (IC). UC and CD have distinct pathologic and clinical characteristics, while IC shares clinical and pathological characteristics between the two. However, all of their pathogenesis remains poorly understood. CD is characterized by transmural inflammation and skips lesion. The transmural inflammatory nature of CD can lead to fibrosis or strictures, which can be manifested as obstructive clinical manifestation, which is not

commonly seen in UC. In addition, a transmural inflammation can result in sinus tracts, giving rise to micro-perforations and fistulae.¹

The term IC has been used to describe cases of inflammatory bowel disease that cannot be classified as UC or CD. However, this term has suffered varying definitions, which in addition to numerous difficulties in diagnosing inflammatory bowel disease has led to much confusion. The term IC should only be used in cases where a colectomy has been performed and the overlapping features of CD and UC do not allow a definitive diagnosis. Over time, most patients remain with a diagnosis of indeterminate colitis or show symptoms similar to UC.^{3,4}

IBD is a disease with a high incidence and prevalence in Europe and North America. The average incidence of UC and CD in Europe was 24.3 and 29.3 per 100,000 people, respectively.⁶ On the contrary, the incidence of IBD is still increasing in developing countries.⁷ In Indonesia, the average incidence of UC is 0.55 per 100,000 people.⁸

Ulcerative colitis

UC is an idiopathic chronic inflammatory which influenced by several risk factors but not limited to age, gender, race, ethnicity, genetic susceptibility, smoking, diet, and usage of several medications (antibiotics, isotretinoin, NSAIDs, and oral contraceptive).¹¹

UC damages the colonic epithelial barrier through several mechanisms, including host immune response, recruitment of leukocytes, and large intestine microflora involvement UC characterized by relapsing and remitting episodes of inflammation on the mucosal layer and can spread from the rectum to the proximal colon; depending on its location, UC can be classified as Ulcerative proctitis, Ulcerative proctosigmoiditis, Left-sided or distal ulcerative colitis, Extensive colitis, and Pancolitis.^{1,4,9}

Common clinical presentations include chronic diarrhea with or without blood for more than four weeks, abdominal pain, weight loss, urgency, tenesmus, and incontinence.⁵ In addition, to these symptoms to diagnose UC, other similar diagnoses such as infective colitis, radiation colitis, parasitic infection, medication associated colitis, therefore, stool examination, history of abdominal/pelvic radiation and drug consumption history must be obtained to exclude these diagnoses.¹² Endoscopy evaluation also has to be done to confirm the diagnosis and assess the extent of the disease. One criterion that can be used to measure the disease severity is Mayo scoring (Figure 1).¹³

Parameters	Subscore, 0-3
Stool Frequency	0 = Normal Numbers of stools for this patient 1 = 1-2 stools more than normal 2 = 3-4 stools more than normal 3 = 5 or more stools more than normal
Rectal Bleeding	0 = No blood seen 1 = Streaks of blood with stool less than one-half of the time 2 = Obvious blood with stool most of the time 3 = Blood alone Passes
Finding on endoscopy	0 = Normal or inactive disease 1 = Mild disease (erythema, decreased vascular pattern, and mild friability) 2 = Moderate disease (marked erythema, lack of vascular pattern, friability, and erosions) 3 = Severe disease (spontaneous bleeding and ulcerations)
Physician's global assessment	0 = Normal 1 = Mild Disease 2 = Moderate disease 3 = Severe disease

Figure 1. Mayo score

The current pharmacological approach aimed to achieve clinical and histological remission by inhibiting inflammation and suppress the immune system's function. Medications that are available including anti-inflammatory drugs (5-aminosalicylic acid and corticosteroids), immunomodulators (azathioprine, mercaptopurine, and methotrexate), and biological agents. A herbal medicine called Polysaccharide Peptide (PsP), derived from the *Ganoderma lucidum* Mycelium Extract, contains an active compound of β -1,3/1,6-D-Glucans. This active compound has an immunomodulatory and anti-inflammatory effect which potentially can help patients with UC. The administration of *Ganoderma lucidum* in CD patients decreased the production of TNF- α , IFN- γ , and IL-17A, which are produced by colonic mucosal inflammation.¹⁰ However, there has yet to be a study exploring the effect of administering β -1,3/1,6-D-Glucan derived from *Ganoderma lucidum* mycelium extraction to UC patients.

Ganoderma lucidum

Ganoderma lucidum is a fungus that has long been used in China as herbal medicine to prevent and treat various diseases. It is assigned to the family of ganodermataceae, which describe as polypore basidiomycetous fungi having a double-walled basidiospore. Fungus of this genus has a laccate surface linked with the presence of thick-walled pilocystidia embedded in an extracellular melanin matrix *Ganoderma lucidum*, and several related fungus species have been known for more than 4000 years as a medicine for various diseases in Japan and China. However, in the wild, these mushrooms have

irregular distribution and therefore only accessible to nobilities in the past. Despite its scarcity, there is an increasing demand for medicinal purposes; hence many attempts have been made to increase the production of these mushrooms. A streamlined process, strict quality control and specific growth parameters (temperature and ph) has allowed these products become a commodity product and can be used for worldwide.

In Japan, this herbal formula is known as Reishi, while in China and Korea, it is known as *Ling Chu* or *Ling Zhi*.¹⁵ In Indonesia, *Ganoderma lucidum* has been cultivated, extracted, and studied in animals or humans for medicinal purposes.¹⁶ *Ganoderma lucidum* has been studied in various clinical trials for the last 30 years. Several studies have shown that *Ganoderma lucidum* polysaccharides can modulate immune function both *in vivo* and *in vitro* studies. It can modulate the immune system by enhancing antigen-presenting cells (APC), mononuclear phagocyte system, humoral immunity, and cellular immunity function.^{15,18} Specifically, the active compound of β -1,3/1,6-D-Glucan from the polysaccharide peptide (PsP) extract has strong immunomodulatory and anti-inflammatory properties.¹ This compound allows binding to immune cell receptors and thus acts as an immunomodulator.¹

The active compound from *Ganoderma lucidum* has the following characteristics (Figure 2): (1) The 1D-NMR spectrogram profile is similar to the β -Glucan USP (United States Pharmacopeia) with the reference standard for β -Glucan: Category number 1048288, lot number F0K129; (2) Molecular weight >3.755 kilo Daltons, with a purity of 54%; (3) Complex branches with a bond ratio of 2:1 between the main β -1,3 chain and the β -1,6 branch; (4) Large molecular weight and branching of the β -1,3/1,6-D-Glucan complexes show high potency as potent immunomodulators.³

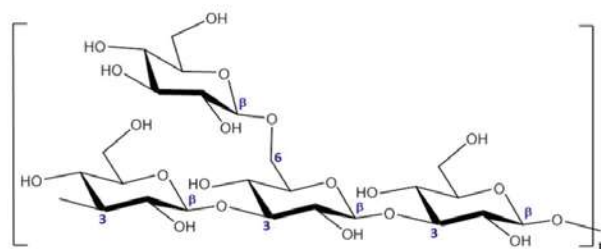


Figure 2. Chemical Structure of β -1,3/1,6-D-Glucan *Ganoderma lucidum*

Immuno modulatory properties of β -1,3/1,6-D-Glucan from *Ganoderma lucidum*

The β -1,3/1,6-D-Glucan stimulates the immune system through several mechanisms including,

activation and proliferation of B cells, inducing the release of T cells from TNF- α and interferon- γ , increasing the activation and maturation of immature dendritic cells, increasing the differentiation and maturation of macrophages. These effects are mediated through different cascades (Figure 3).¹⁸ Jin et al also showed that the *Ganoderma lucidum* polysaccharide could significantly regulate the expression of occludin, nuclear factor- κ B p65 (NF- κ B p65), secretory immunoglobulin A (SIgA) in the ileum, increase the levels of interferon- γ (IFN- γ), interleukin-2 (IL-2), IL-4, and reduce serum diamine oxidase (DAO) levels. Thus, the *Ganoderma lucidum* polysaccharides can be used as agents to regulate intestinal barrier function.¹⁹

In *in vivo* studies done in mice, most β -Glucan was detected in the stomach and duodenum 5 minutes after administration, and the number rapidly decreased during the first 30 minutes. A significant amount of β -Glucan then enters the proximal intestine shortly after consumption, and the concentration rises rapidly in the ileum.^{1,2}

Indications of *Ganoderma lucidum* extract

Ganoderma lucidum extract can be used in the prevention of atherosclerosis. In laboratory studies, it has been shown that it can arrest the development of tumor cells at different points of the cell cycle through the regulation of different signals, for example, breast at G0/G1 phase, lung at G1 phase, liver at G1/G2 phase bladder at G2 phase and so on. It also can boost the immune system by increasing GLPP immunomodulators in bacterial/viral infections and modulate immune function in autoimmune diseases such as UC.^{15,24-27}

The effectiveness of *Ganoderma lucidum* polysaccharide peptides (PsP) as an immunomodulator and for anti-inflammation

Lull C et al demonstrated that the *Ganoderma lucidum* polysaccharide has anti-inflammatory and immunomodulatory effects by: decreasing IL-1 β , TNF- α , and IL-6; decreasing the production of antibodies; increasing the proliferation of rat spleen lymphocytes; increasing the activation, proliferation, and differentiation of B lymphocytes; increasing the production of immunoglobulin.²⁵

Another *in vitro* study by Wang SY et al, *Ganoderma lucidum* polysaccharides, can suppress IL-1 β , TNF- α , and IL-6 in macrophage culture. In addition, *Ganoderma lucidum* polysaccharides have immunomodulatory and anti-tumor effects.²⁸

Role *Ganoderma lucidum* Mycelium Extract on Ulcerative Colitis

Ganoderma lucidum polysaccharides, especially β -1,3/1,6-D-Glucan, is expected to act as adjunctive therapy for IBD patients primarily due to their immunomodulatory and anti-inflammatory effects on patients with UC. A study conducted on CD patients showed that administration of *Ganoderma lucidum* decreased the production of TNF- α , IFN- γ , and IL-17A, which are produced by colonic mucosal inflammation.¹⁰ Wei B et al found that the *Ganoderma lucidum* polysaccharide can suppress Th17 cell response in mice with artificial colitis. This study reported that the *Ganoderma lucidum* polysaccharide significantly increased the survival rate, improved bowel length shortening, weight loss, histopathological score, and DAI score in mice with artificial colitis.

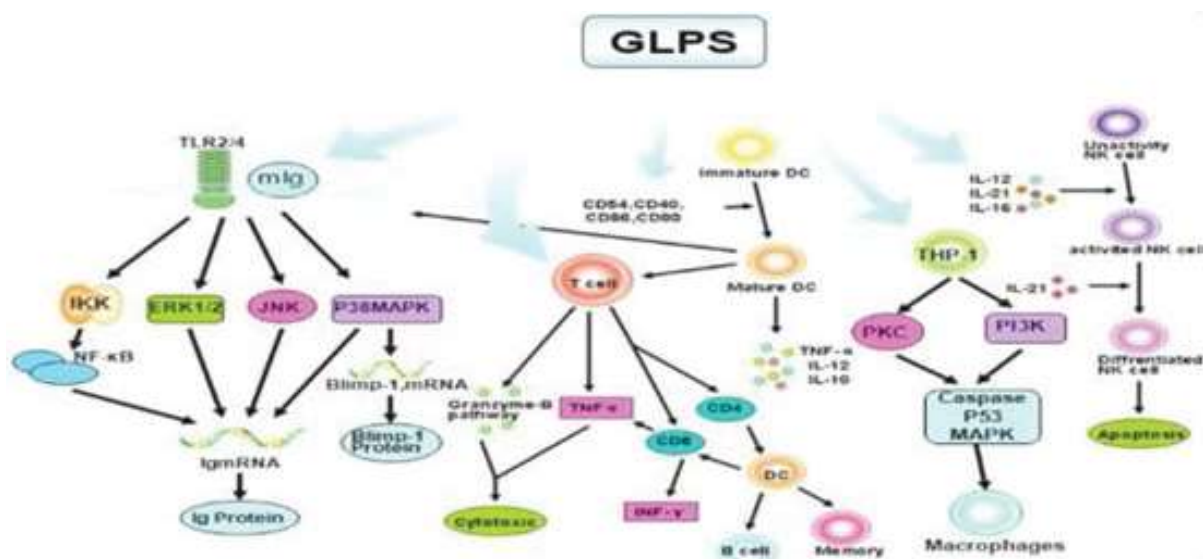


Figure 3. *Ganoderma lucidum* polysaccharide peptide as an immunomodulatory

Ganoderma lucidum polysaccharides also significantly suppressed the secretion of TNF- α , IL-1 β , IL-6, IL-17A, and IL-4, and the number of Th17 cells, B cells, NK cells, and NKT cells in the lamina propria lymphocytes.²⁷ Sliva D et al demonstrated that *Ganoderma lucidum* polysaccharides could prevent carcinogenesis in mice with artificial colitis.²⁹ In this study, *Ganoderma lucidum* polysaccharides significantly suppressed focal hyperplasia and tumor formation.

Ganoderma lucidum can prevent the shortening of the colon and reduce macrophages' infiltration in the colon tissue. The administration of *Ganoderma lucidum* also significantly reduced the regulation of PhIP/DSS-dependent expression of cytokines D1, COX-2, CYP1A2, and CYP3A4 in the colonic tissue. Xie J et al demonstrated that *Ganoderma lucidum* extract given to mice with artificial colitis showed improved colitis due to regulation of the gut microbiota and gene expression of colon epithelial cells.²⁹

The effect of *Ganoderma lucidum* as an immunomodulator was also reported by Bhardwaj N et al to suppress inflammation, allergic response and enhance other immunological capabilities.³⁰ Nie Y et al reported the Role of non-starch polysaccharides, including polysaccharides derived from *Ganoderma lucidum*, to treat IBD.³¹ In a study performed in Mount Sinai hospital, the United States of America, on CD patients aged 8-19 years, the *Ganoderma lucidum* extract in the form of Triterpene ganoderic acid C1 (GAC1) can inhibit TNF- α production and other proinflammatory cytokines and improve the colonic mucosa of CD, which was inflamed due to the NF- κ B.¹⁰

Thus, *Ganoderma lucidum* polysaccharides are beneficial for patients with UC through several mechanisms. Decreasing the production of several inflammatory factors such as TNF- α , IFN- γ , and IL-17A, maintain intestinal homeostasis by modulating the immunological response to artificial colitis through suppression of Th17 response and regulate the function of the intestinal immunologic barrier in mice with artificial colitis by suppressing the secretion of inflammatory factors and adaptive immune response in lamina propria lymphocytes. Moreover, it also prevents carcinogenesis by suppressing hyperplasia and tumor formation in animal studies with artificial colitis, which in UC might be beneficial because the patient has a higher risk of developing colon cancer. Other mechanisms which *Ganoderma lucidum* might beneficial for patient with UC are, prevention of colon shortening as the end-stage disease of UC by reducing

inflammation on the colon, regulation of normal flora in the colon, therefore, providing a homeostasis environment for the colon to recover and reducing the allergic and inflammation response which is the primary mechanism of UC

However, to date, no studies have been conducted on UC patients. In Indonesia, a study by Farikh A et al. in Malang explore the administration of β -1,3/1,6-D-Glucan derived from *Ganoderma lucidum* reported clinically meaningful results 90 days after administration in patients with cardiovascular problems.³³

CONCLUSION

The β -1,3/1,6-D-Glucan from *Ganoderma lucidum* mycelium extract is a potentially effective herbal remedy for UC patients. Studies have shown that, in animals, the herbal remedy is useful as an anti-inflammatory alternative and as an immunomodulator. However, further researches are needed in humans to confirm such beneficial effects.

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