

Advances in Research of the Mongolian Medicine *Physochlaina physaloides* (L.) G. Don

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Abstract *Physochlaina physaloides* (L.) G. Don, a traditional Mongolian medicinal herb, exhibits anti-inflammatory, analgesic, and anti-migraine properties. Its main components include alkaloids, flavonoids, coumarins, and diphenylethanes. Processing methods such as milk soaking reduce toxicity while enhancing efficacy. Pharmacological studies confirm its activities against arthritis, migraine, gastric ulcers, and certain cancers. Quality control remains limited to single-marker analysis, and artificial cultivation requires further optimization. Despite progress, gaps persist in fully elucidating active components, processing mechanisms, and comprehensive quality standards. Future research should integrate multi-omics approaches, establish holistic evaluation systems, and advance clinical translation to support sustainable utilization.

Key words Mongolian medicine, *Physochlaina physaloides* (L.) G. Don, Chemical components, Processing technology, Pharmacological effects, Advances in research

1 Introduction

Physochlaina physaloides (L.) G. Don is a commonly used medicinal material in Mongolian medicine and has been recorded in classical medical works of Mongolian and Tibetan medicine of successive dynasties. In Mongolian medical theory, it has the effects of killing "Nian", reducing swelling, relaxing muscles, and relieving pain. Clinically, it is commonly applied to treat asthma, inflammation, migraine and other diseases. Its medicinal part is the dried root, and its Mongolian medicinal name is Chagan – Tangpuru^[1]. Due to the inherent toxicity of the *P. physaloides*, in Mongolian medical practice, it needs to be processed before being used as medicine to reduce toxicity and enhance therapeutic effects^[2]. In recent years, with the modernization of ethnic medicine, scholars at home and abroad have conducted extensive research on *P. physaloides*, achieving remarkable progress in areas such as the separation and identification of chemical components, the optimization of processing techniques, the interpretation of pharmacological mechanisms, and the establishment of quality standards. Xilintuya reviewed the research progress of *P. physaloides* and pointed out that research was still relatively scarce at that time^[3]. After years of development, the research scope of *P. physaloides* has been continuously expanding and the research depth has been constantly improving. However, no research has systematically integrated its latest achievements so far. Previous reviews, such as that by Xilintuya^[3], provided foundational chemical data but lacked systematic comparisons of processing methods and did not address recent findings on anti-tumor activity, multi-omics mechanisms, or artificial cultivation. The present

review aims to fill these gaps by critically evaluating recent advances, highlighting controversies, and proposing directions for future research. This review updates the latest progress, sorts out academic controversies, analyzes research limitations, and provides targeted suggestions for future studies. Therefore, this paper, in combination with the existing research literature, conducts a comprehensive review of the core research contents of *P. physaloides*, providing theoretical support for its subsequent research and industrial application.

2 Chemical components

There are relatively few reports on the chemical components and their analysis of *P. physaloides*^[4]. Moreover, the chemical components of *P. physaloides* are complex and diverse, mainly including anisodamine, hyoscyamine, scopolamine, coumarins, neorutin, and quercetin trisaccharide, among others. Among them, alkaloids are the core active and toxic components and have also been a research hotspot in recent years. Liu Yonglong *et al.*^[5] first isolated two alkaloids from the roots of *P. physaloides*, namely, anisodamine and hyoscyamine. Relevant studies have verified that *P. physaloides* is rich in anisodamine, and its extraction and separation procedures are relatively simple. Therefore, it can be regarded as an important natural plant resource for obtaining anisodamine. This discovery laid the foundation for the study of the chemical composition of *P. physaloides*, and subsequent scholars have continuously conducted in-depth research on this basis.

At present, 15 components have been isolated from *P. physaloides*^[6], including flavonoids (neorutin, rutin, quercetin-3- β -D-glucopyranosyl- α -L-rhamnopyranosyl-4- β -D-glucopyranoside, quercetin-3- β -D-glucopyranosyl-(1 $\rightarrow$ 6)- α -L-rhamnopyranosyl-7- α -L-rhamnopyranoside, quercetin-3- β -D-glucopyranosyl-6- β -L-rhamnopyranosyl-4- β -D-glucopyranoside, and quercetin-3- β -D-glucopyranoside) and coumarins (7-hydroxycoumarin, 6,8-dimethoxycoumarin-7-O- β -D-glucoside, 7-methoxycoumarin, 6,7,8-

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trimethoxycoumarin, 6-methoxycoumarin-7-O- β -D-glucoside, and 8-hydroxy-6,7-dimethoxycoumarin). Ultrasonic extraction process of total flavonoids from *P. physaloides* has been optimized to improve the yield of flavonoids, and the obtained total flavonoids possess obvious cytotoxic activity against HeLa cervical cancer cells^[7]. Diphenylethane compounds are a new type of component discovered in *P. physaloides*. Hubis Halatu *et al.*^[8] isolated three diphenylethane compounds from the chloroform extract of *P. physaloides* by conventional silica gel column chromatography combined with reversed-phase preparative High Performance Liquid Chromatography (HPLC). The three compounds were identified as 1-(3-hydroxyphenyl)-2-(3-hydroxy-5-methoxyphenyl) ethane, 1-(3-hydroxyphenyl)-2-(3,5-dimethoxyphenyl) ethane, and 1-(3-hydroxyphenyl)-2-(3,5-dihydroxyphenyl) ethane. All of them were isolated from *P. physaloides* for the first time. Zang *et al.*^[9] investigated the chemical components of the whole plant of *P. physaloides*, and isolated 8 alkaloids, 14 flavonoids, 2 coumarins and 1 cyclic sesquiterpene compound. Current chemical research shows obvious unbalanced tendencies. Most studies focus on alkaloids, whereas flavonoids, coumarins and newly identified diphenylethanes are only structurally determined, with insufficient exploration of their pharmacodynamic targets and metabolic mechanisms. In addition, inconsistent extraction solvents and parameters across different studies lead to incomparable component data, resulting in fragmented and unsystematic research results. For example, while Liu *et al.* used ethanol for alkaloid extraction, later studies employed methanol, making direct quantitative comparisons difficult. Standardizing extraction protocols is thus a prerequisite for building a unified chemical profile. Among them, several compounds were obtained from this plant for the first time, which further enriched the chemical composition profile of *P. physaloides*.

3 Processing technology

Mongolian medicinal processing is guided by the theories of Mongolian medicine. According to clinical needs, traditional processing methods are adopted to ensure the safety and efficacy of Mongolian medicinal materials. These methods summarize the practical experience of Mongolian medicine application^[10]. *P. physaloides* possesses definite toxicity. In Mongolian clinical application, it must be processed for detoxification before medicinal use. Common processing methods include milk soaking, goat milk soaking, roasting, paste processing. Different processing methods exert significant effects on its chemical components and pharmacological efficacy. In recent years, scholars have optimized the processing technology of *P. physaloides* by means of orthogonal experiments and established standardized processing procedures. Bao Tingting *et al.*^[11] used LC-MS to analyze the chemical components of raw and paste processed products of *P. physaloides*. A total of 73 components were identified, among which 48 were common to both samples, 15 were new compounds produced after processing, 10 were lost after processing, 15 showed an increase in content, and

32 showed a decrease. These changes in chemical components provide the material basis for the detoxification and efficacy enhancement of *P. physaloides* during processing, and lay an important foundation for clarifying its processing mechanism. Yue Ying *et al.*^[12] determined the contents of scopolamine and hyoscyamine in *P. physaloides* samples collected from six regions: Ewenki, Manzhouli, Xiwu Banner, Xinyou Banner, Xinzuo Banner, and Yakeshi. The results showed that the total alkaloid content in raw materials and two types of processed products was highest in Xinzuo Banner and lowest in Ewenki. Milk processing induces opposite alkaloid variation trends among different producing areas, yet the underlying reasons remain unexplained. This geographical disparity suggests that the chemical response to milk processing may be modulated by intrinsic factors such as soil composition or harvest time, which warrants systematic investigation through controlled cultivation experiments. The effects of soil, climate and harvest period on alkaloid transformation during processing are still unclear. Moreover, orthogonal parameters from different research teams are inconsistent, making optimized processing conditions difficult to replicate, which limits the industrial application of existing technological achievements. After the raw materials were processed with milk, only the samples from Ewenki showed an increase in total alkaloid content, while those from other regions decreased. When the raw materials were prepared into extracts, the total alkaloid content increased in all regions, with a more significant increase compared to milk-processed samples. These results indicate that alkaloid contents of *P. physaloides* vary greatly among different producing areas, and both milk processing and extract preparation can change its alkaloid levels. The regulatory effect of milk processing differs across regions, whereas extract preparation can significantly increase alkaloid content. This clearly demonstrates that Mongolian medicinal processing can effectively regulate the active ingredient levels of medicinal materials. Bao Minglan *et al.*^[13] took anisodamine and hyoscyamine as comprehensive evaluation indexes to optimize the processing technology. An $L_9(3^4)$ orthogonal experimental design was adopted to optimize the paste preparation method and milk soaking processing method of *P. physaloides*. The results showed that the optimal procedure for the paste preparation was adding 8-fold water to the raw materials and boiling three times for 30 min each time. The optimal conditions for the milk soaking method were: adding 4.5-fold milk to the medicinal materials, soaking for 4 h, and drying at 40 °C for 2.5 h. This study provides a scientific basis for the standardized processing procedure of *P. physaloides*. Wuyunga *et al.*^[14] optimized the goat milk soaking-stewing method and the roasting method of *P. physaloides*. The optimal conditions for goat milk soaking-stewing were as follows: adding 5.5-fold goat milk to the medicinal materials, soaking for 4 h, and drying at 100 °C for 2.5 h. For the roasting method, the optimal process was to crush the medicinal materials into fragments of 1–3 cm and roast at 150 °C for 60 min. This study further expands the processing technology options for *P. physaloides*.

The effects of processing on the chemical components and pharmacological efficacy of *P. physaloides* are the core of research on its processing mechanism. Song Hongchun *et al.*^[15] found that after milk processing and decoction, the content of atropine alkaloids in processed samples was lower than that in raw materials. The contents of atropine sulfate in the raw material, milk-processed product, and decocted product were 4.875, 4.505, and 3.740 mg/g, respectively. This variation provides an important material basis for the detoxification effect of processing on *P. physaloides*. Studies have shown that after milk processing, the contents of anisodamine, hyoscyamine, and scopolamine in *P. physaloides* decrease significantly. Meanwhile, seven new compounds, including scopolamine lactone, rutin, and vanillic acid, are generated. These newly formed compounds may be closely related to the detoxification and efficacy-enhancement mechanism of *P. physaloides*^[2]. In addition, Bao Minglan *et al.*^[16] also found that the contents of scopolamine and hyoscyamine increased markedly in the paste-processed products of *P. physaloides*. Such products can be used for the treatment of toxic shock and as raw materials for the extraction of scopolamine. Moreover, milk-processed products exhibit enhanced analgesic effects, which have potential development value as preparations for migraine treatment.

Hou Yaxing *et al.*^[17] adopted electronic tongue technology to analyze the taste differences among raw materials, milk-processed products and extracts of *P. physaloides*. The results showed significant differences in astringency, bitterness and umami taste among the three samples. The extracts exhibited the strongest astringency and umami, while milk-processed products had the most prominent bitterness. Such taste changes are intrinsically associated with the pharmacological efficacy variations of *P. physaloides* after processing. This provides a new perspective for exploring the processing mechanism of *P. physaloides*. Sha Yi *et al.*^[18] established a quality evaluation model for *P. physaloides* based on 1H-NMR technology. The model can effectively distinguish the compositional differences among samples processed by different methods, offering a new approach for processing technology optimization and quality control of *P. physaloides*.

4 Pharmacological effects

With the development of modern pharmacological research methods, the pharmacological effects of *P. physaloides* have been further explored continuously. It has been confirmed that this herb possesses multiple pharmacological activities, including anti-inflammatory, analgesic, anti-arthritis, anti-migraine, gastric mucosa-protective and anti-tumor effects. Its mechanism is closely related to the regulation of metabolic pathways, gene expression and intestinal flora *in vivo*.

Anti-inflammatory and analgesic effects are the most fundamental pharmacological properties of *P. physaloides*. Lin Yuncai *et al.*^[19] further verified that *P. physaloides* decreased in toxicity after milk processing, while retaining good analgesic and anti-in-

flammatory efficacy, thereby achieving the purpose of toxicity reduction and efficacy enhancement. Zhang Hongyu *et al.*^[20] drew a similar conclusion, confirming that both raw and milk-processed samples exhibit significant analgesic and anti-inflammatory effects. Li Xiaopeng *et al.*^[21] conducted a comparative study on the anti-inflammatory and analgesic activities of *P. physaloides* before and after processing. The results showed that both raw and processed products could significantly reduce acetic acid-induced writhing responses, elevate the pain threshold in hot-plate tests, and markedly alleviate xylene-induced ear edema as well as acetic acid-induced increased peritoneal permeability in mice. There was no significant difference in efficacy between raw and processed samples.

Anti-arthritis and anti-migraine effects are important directions for the clinical application of *P. physaloides*. Yue Ying *et al.*^[6] established a rat migraine model induced by nitroglycerin. The study indicated that milk-processed *P. physaloides* could significantly reduce serum levels of calcitonin gene-related peptide (CGRP), NO and ET-1, increase β -EP and 5-HT contents in brain tissue, improve pathological damage of the hippocampus, regulate intestinal flora structure in migraine rats, and modulate the relative abundance of related intestinal microbial families. It exerts anti-migraine action by regulating pathways such as secondary metabolite biosynthesis, amino acid metabolism and the tricarboxylic acid cycle. Bao Tingting *et al.*^[12] established a rat adjuvant arthritis model using Freund's complete adjuvant, and found that the paste-processed product of *P. physaloides* showed obvious therapeutic effects. It could significantly reduce paw swelling, decrease serum inflammatory indices including RF, IL-1 β , MMP-9 and IgG, and improve the pathological morphology of synovial tissue. Its mechanism is related to the regulation of metabolomic and proteomic pathways.

In recent years, the gastrointestinal protective and anti-tumor effects of *P. physaloides* have also attracted extensive attention. Wei *et al.*^[22] isolated two novel compounds, physochloside B and physochloside C, from *P. physaloides* extract. These compounds exhibited significant cytotoxicity against Hep-G2 and HGC-27 cell lines. They could induce tumor cell apoptosis and cell cycle arrest by inhibiting the Wnt/ β -catenin pathway, with relatively low toxicity to normal cells, providing a new reference for the anti-tumor research of *P. physaloides*. In addition, total flavonoids of *P. physaloides* extracted by optimized ultrasound method can significantly inhibit the proliferation of HeLa cervical cancer cells^[7]. Furthermore, combined with network pharmacology, pharmacokinetics and experimental verification, relevant studies have confirmed that *P. physaloides* can exert therapeutic effects on chronic atrophic gastritis by regulating core targets and key signaling pathways^[23]. Li S *et al.*^[24] established rat models of chronic gastritis and gastric ulcer, and found that *P. physaloides* has a significant protective effect on gastric mucosal injury. Its mechanism is associated with the activation of the PPAR/ $\text{NF-}\kappa\text{B}$ signaling pathway.

5 Quality control

Quality control is crucial to ensuring the safety and effectiveness of the clinical application of *P. physaloides*. In recent years, scholars have established a series of scientific and reliable quality control methods focusing on the qualitative identification and quantitative determination of *P. physaloides*, providing a scientific basis for the quality evaluation of its medicinal materials.

In terms of qualitative identification, Tian Xiangyang *et al.*^[25] used microscopic identification and Thin Layer Chromatography (TLC) methods to conduct qualitative analysis on *P. physaloides*. They found that the medicinal material has obvious microscopic identification characteristics, clear TLC spots and good separation, which can effectively distinguish *P. physaloides* from other similar herbs. Wang Qinghu *et al.*^[26] established a thin-layer chromatographic identification method for *P. physaloides*. They used coumarin-7-O- β -D-glucoside and scopolamine as reference substances and adopted different developing agents, and clear characteristic spots could be observed under the corresponding conditions, which makes it easy to identify the authenticity of *P. physaloides*. Bao Minglan *et al.*^[16] used thin-layer chromatography to qualitatively identify the raw products and different processed products of *P. physaloides*, with atropine as the reference substance. The results showed that all samples from different origins presented characteristic spots, which can be used to identify the authenticity of *P. physaloides* and its processed products. The team also established an HPLC method to quantitatively determine the contents of scopolamine, hyoscyamine and anisodamine in *P. physaloides* and its processed products. This method is accurate and has good repeatability, and can be used for the quality evaluation of *P. physaloides* and its processed products^[12]. Wang Qinghu *et al.*^[26] established a multi-index content determination method, which simultaneously determined the contents of coumarin-7-O- β -D-glucoside, 6-methoxy-coumarin-7-O- β -D-glucoside, scopolamine and hyoscyamine in *P. physaloides*, providing a scientific basis for the comprehensive quality evaluation of *P. physaloides*.

In addition, Dai Qing *et al.*^[27] used a microplate reader to detect the total alkaloid content of *P. physaloides* before and after processing. With scopolamine hydrobromide as the reference substance and bromocresol green as the chromogenic agent, this method is simple, low-cost and capable of high-throughput detection. The test results showed that the total alkaloid content of *P. physaloides* increased from 0.29% to 0.46% after processing, which provides new ideas for the quality control of *P. physaloides*. Zhang Runxiang *et al.*^[28] established an HPLC method for the determination of scopolamine content in the Mongolian medicinal material *P. physaloides*. This method is accurate and has good repeatability, which can be used for the quality control of the Mongolian medicinal material *P. physaloides*.

In conclusion, certain progress has been made in the current research on the quality control of *P. physaloides*, but there are still significant deficiencies; the existing methods mainly focus on

the quantification of single components, lacking multi-dimensional comprehensive quality evaluation techniques such as HPLC fingerprinting and DNA barcoding. No systematic research has been conducted on the quality differences of *P. physaloides* in different harvest periods and different medicinal parts, and the current single and one-sided evaluation system cannot reflect the "overall quality" characteristics of Mongolian medicine, nor can it ensure the consistency of quality among different origins and batches of *P. physaloides*. Current quality evaluation systems adopt single-index detection modes suitable for chemical drugs, which cannot match the holistic evaluation concept of Mongolian medicine. Qualitative methods such as microscopic identification and TLC rely heavily on subjective experience without digital quantitative indicators. Inconsistent detection standards and the absence of unified quality thresholds for different harvesting periods and medicinal parts hinder unified quality assessment.

It is therefore urgent to establish a multi-dimensional and comprehensive quality evaluation system.

6 Resource cultivation and conservation

As a rare and endangered Mongolian medicinal herb, *P. physaloides* is facing a gradual decline in its wild resources year by year. Coupled with its large clinical dosage, artificial cultivation has become an important approach to ensure the sustainable utilization of its resources. In 1986, Guo Fuxing *et al.*^[29] carried out studies on stem tip culture of *P. physaloides*. By using MS medium supplemented with different concentrations of NAA and BA, they provided technical references for tissue culture and rapid propagation of *P. physaloides*. Siqinbatuer *et al.*^[30] pointed out that *P. physaloides*, together with *Astragalus membranaceus* and *Acanthopanax purpureus*, belongs to the rare and endangered Mongolian medicinal herbs. Due to the reduction of wild resources and varietal degradation of cultivated populations, its survival is under serious threat. To solve this problem, the research team collected wild *P. physaloides* seeds, adopted various methods to break seed dormancy and relieve hard seed coat characteristics, and explored its germination mechanism. A 2 ha standardized planting demonstration area for *P. physaloides* was established in the Hunshandake Sandy Land of Xilingol League, Inner Mongolia. Meanwhile, a local medicinal material protection zone was set up, and measures such as no-till reseeding and transplanting were adopted to restore the wild population diversity of *P. physaloides*. These efforts provide a practical basis for the resource protection and artificial cultivation of this medicinal herb. Zang Erhuan *et al.*^[31] determined the optimal germination conditions for *P. physaloides* seeds: soaking in 100 mg/L GA₃ solution for 24 h, with the average seed germination rate reaching the highest value of 42.2%. Meanwhile, based on combined metabolomic and transcriptomic analysis, this study explored the effects of drought stress on the accumulation of scopolamine alkaloids in *P. physaloides*.

Existing resource protection measures are fragmented and short-term. The existing standardized planting demonstration area

is limited to small-scale trials without mature large-scale promotion techniques. Wild population restoration lacks long-term dynamic monitoring data, and systematic germplasm breeding research is absent, making it difficult to establish a sustainable resource utilization mechanism.

7 Conclusions and prospects

In recent years, considerable progress has been achieved in the research of *P. physaloides*, and in-depth studies have been carried out on its medicinal value across multiple research fields. In terms of chemical components, researchers have isolated and identified a variety of compounds from *P. physaloides* by modern separation and identification technologies, mainly including alkaloids, coumarins, flavonoids and diphenylethanes. Meanwhile, the effects of processing methods on the types and contents of chemical components have been clarified, laying a solid foundation for further studies on the material basis of efficacy, processing mechanism and quality control. In the field of processing technology, relevant studies have continuously optimized various traditional processing methods of *P. physaloides*, and explored the integration of traditional and modern processing techniques. The detoxification and efficacy-enhancement mechanisms during processing have been preliminarily elucidated, providing theoretical and practical support for the standardization and normalization of its processing technology. With regard to pharmacological effects, *in vivo* and *in vitro* experiments have confirmed that *P. physaloides* possesses extensive biological activities, such as anti-inflammatory, analgesic, anti-arthritis, anti-migraine, gastric protective and anti-tumor effects. The molecular mechanisms underlying these pharmacological activities have been gradually revealed, which further validates its traditional medicinal value and provides experimental evidence for new clinical application directions. In quality control research, qualitative identification and quantitative determination methods for *P. physaloides* raw materials and processed products have been established, and preliminary quality standards have been formulated. These can meet the basic quality control requirements and provide a preliminary specification for its further development and clinical application.

Although relevant research on *P. physaloides* has achieved the above progress, there are still some deficiencies urgently needing to be addressed from the perspective of industrial development and in-depth exploration of its medicinal value. First, the medicinal components of *P. physaloides* have not been fully clarified, and the molecular mechanisms underlying some pharmacological effects remain to be further explored. In particular, research on its novel pharmacological activities such as anti-tumor and gastric protective effects is still in its initial stage, and their functional targets and regulatory pathways have not been fully elucidated. Second, research on its processing mechanism lacks systematicity. The detoxification and efficacy-enhancement mechanisms of traditional characteristic processing methods such as milk processing are still unclear. Moreover, the types and contents of

newly generated compounds during processing, as well as their correlation with pharmacological efficacy, need further verification and clarification. Third, the quality control system remains to be improved. At present, a comprehensive multi-index and multi-method quality evaluation model has not been well established. Research on quality differences among different producing areas and different processed products is insufficient, which makes it difficult to comprehensively reflect the overall quality characteristics of *P. physaloides*. Fourth, artificial cultivation techniques still need further optimization. There is still much room for improvement in the yield and quality stability of artificially cultivated *P. physaloides*. A long-term mechanism for sustainable resource utilization has not yet been formed, which cannot meet the needs of large-scale industrial development.

Based on the current research foundation and existing deficiencies, future research on *P. physaloides* should focus on the following key directions to promote the exploitation of its medicinal value and industrial development: First, in-depth research on chemical components should be strengthened. Relying on modern separation, purification and structural identification technologies, new active components of *P. physaloides* that have not yet been discovered should be explored, and the core pharmacological material basis should be further clarified to provide support for subsequent related research. Second, the processing mechanism should be thoroughly explored. By integrating multi-omics technologies such as metabolomics and proteomics, the regulatory mechanisms of processing techniques on the chemical composition, content and efficacy of *P. physaloides* should be systematically elucidated, especially the scientific connotation of traditional Mongolian medicinal processing methods. Third, the quality control system should be improved. The limitation of single-component evaluation should be broken, and a multi-index and intelligent comprehensive quality evaluation method should be established to realize comprehensive quality control of *P. physaloides* medicinal materials, processed products and related preparations, thereby enhancing the scientificity and practicality of quality standards. Fourth, artificial cultivation techniques should be optimized. The effects of environmental factors such as light, water and soil on the growth and development of *P. physaloides* and the accumulation of medicinal components should be thoroughly explored, standardized and normalized cultivation models should be established to improve the stability of yield and quality, and a long-term mechanism for sustainable resource utilization should be constructed. Fifth, clinical application research should be enhanced. By integrating modern clinical diagnosis systems with Mongolian medical theories, we will conduct systematic clinical trials to validate its therapeutic efficacy and support the development of standardized Mongolian patent medicines.

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