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CICHORIUM INTYBUS (BEEKHE KASNI): A COMPREHENSIVE ANALYSIS OF TRADITIONAL, PHARMACOLOGICAL, AND NUTRITIONAL ASPECTS

Original Research

Hamda Tanzeem Khan[1]*, Sidra Siddiqui[2], Syed Tahir Ali[2], Sana Siddiqui[2], Muhammad Akhlaq[1], Muhammad Khaleeq Alum[3]
[1]Office of Research Innovation and Commercialization, Hamdard university Karachi, Pakistan.
[2]Faculty of Pharmacy Hamdard University Karachi, Pakistan.
[3]Government Emerson College University Multan, Pakistan.
Corresponding Author: Hamda Tanzeem Khan, Office of Research Innovation and Commercialization, Hamdard university Karachi, Pakistan.
Hamda.tanzeem@hamdard.edu.pk
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ABSTRACT

Background: Cichorium intybus L., commonly known as chicory, is a perennial herb of the Asteraceae family with a long-standing history of use in traditional medicine. Originating from Africa and parts of Eurasia, it has been used to treat various health conditions, including liver disorders, digestive ailments, and inflammatory diseases. Its high inulin content has gained significant attention for its role in managing blood glucose levels, particularly benefiting individuals with diabetes.

Body: This review explores the traditional applications, phytochemical composition, pharmacological activities, and nutritional value of Cichorium intybus. The plant exhibits a wide range of therapeutic effects, including antimicrobial, anthelmintic, antimalarial, hepatoprotective, antidiabetic, gastroprotective, anti-inflammatory, analgesic, antioxidant, and antitumor activities. Key bioactive compounds, such as sesquiterpene lactones concentrated in the roots, have been identified as potential agents for drug development. Despite its established safety profile, concerns regarding the toxicity of certain compounds at high doses highlight the need for regulated consumption.

Conclusion: While preclinical studies support its therapeutic potential, clinical trials remain limited but suggest possible benefits in managing joint pain and promoting cardiovascular health. The plant's adaptability and inulin yield position it as an economically valuable crop for sustainable agriculture. Further research is necessary to validate traditional uses, explore its pharmacological potential, and develop standardized medicinal applications.

Keywords: Antidiabetic, Bioactive compounds, Hepatoprotective agents, Inulin, Sesquiterpene lactones, Sustainable agriculture, Tumor inhibitors.

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INTRODUCTION

Cichorium intybus L., commonly referred to as chicory, is a herbaceous perennial plant belonging to the Asteraceae family. Native to regions of Africa and Eurasia, it has long been recognized for its medicinal, nutritional, and industrial significance. With its tall, erect growth reaching up to one meter and its fleshy taproot extending approximately 75 cm, chicory has a rich history of cultivation that dates back to ancient Egyptian civilization. Historically, it served multiple purposes, including use as a coffee substitute, a remedial herb, a leafy vegetable, and animal fodder. In modern contexts, the plant has attracted scientific interest due to its versatile applications and notable phytochemical composition, particularly its high inulin content(1, 2). The industrial cultivation of *Cichorium intybus* has expanded significantly, primarily driven by its capacity to produce inulin, a fructan-type polysaccharide found predominantly in the plant's roots. Inulin is a non-digestible carbohydrate that offers a range of health benefits, especially for individuals managing diabetes, as it does not raise blood glucose levels. This has made chicory-derived inulin a valuable ingredient in the food and pharmaceutical industries, contributing to the growing market for functional foods and dietary supplements. The ability of chicory to withstand harsh environmental conditions during both vegetative and reproductive stages further enhances its viability as a sustainable crop for large-scale cultivation(3, 4).

Pharmacologically, various parts of the plant—roots, leaves, and flowers—contain a diverse array of bioactive compounds, including sesquiterpene lactones, polyphenols, coumarins, and flavonoids. These phytochemicals have demonstrated anti-inflammatory, antioxidant, hepatoprotective, and antimicrobial properties in numerous preclinical studies. Despite its widespread use in traditional medicine across multiple cultures, the European Pharmacopoeia and other official pharmacopoeias have yet to provide a comprehensive definition or standardized guidelines for its medicinal applications. Nevertheless, empirical evidence continues to support the therapeutic potential of chicory, warranting further investigation through rigorous scientific inquiry(4, 5). The global dispersal of *Cichorium intybus* has facilitated its incorporation into diverse traditional medicine systems. In various cultures, chicory has been employed to address ailments such as digestive disorders, liver dysfunction, and inflammatory conditions. While much of its traditional use is anecdotal, contemporary research has begun to validate these claims, offering scientific substantiation for its ethnopharmacological relevance. Notably, the concentration of bioactive compounds is most prominent in the root, making it a focal point for both scientific research and industrial exploitation(5-7).

Despite its extensive history of use and growing recognition in modern pharmacology, certain gaps persist in understanding the full therapeutic potential of *Cichorium intybus*. Specifically, standardized dosing guidelines, clinical efficacy trials, and mechanistic studies remain underdeveloped areas of research. Moreover, variations in phytochemical content due to environmental and genetic factors pose challenges for consistent therapeutic application(8, 9). Given its historical significance, pharmacological potential, and growing commercial importance, a comprehensive review of *Cichorium intybus* is both timely and necessary. This review aims to consolidate existing knowledge on the plant's traditional uses, phytochemical composition, and pharmacological activities. By critically analyzing recent scientific advancements and identifying gaps in the current research landscape, this analysis seeks to provide a clearer understanding of chicory's therapeutic promise. Ultimately, the objective of this review is to establish a foundation for future research and facilitate the integration of *Cichorium intybus* into evidence-based medical practices(10, 11).

MAIN BODY

Traditional Uses of *Cichorium intybus*

Medicinal plants have played a pivotal role in healthcare systems across diverse cultures, with traditional societies relying on indigenous flora for treating a wide range of ailments. Among these, *Cichorium intybus*—commonly known as chicory—holds a significant place due to its diverse therapeutic applications, documented over centuries through empirical observation and cultural inheritance. This enduring reliance underscores the profound traditional knowledge embedded in its use, often passed down through generations by trial and error(12, 13). The use of chicory as a medicinal herb dates back to ancient Egyptian civilization, where it was valued for its purported curative properties. Over time, its use has expanded across various geographical regions, with each culture integrating the plant into their unique medicinal systems. Across its native habitats and regions where it has been introduced, chicory extracts have been utilized

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to address a wide spectrum of health issues. For instance, in South Africa, despite being classified as a weed, chicory is incorporated into teas that are traditionally used for liver health, while its syrup serves as a restorative tonic for infants, believed to possess detoxifying and invigorating properties(14, 15).

In Turkey, the leaves of chicory are commonly prepared as ointments intended to promote wound healing, whereas decoctions of the roots and leaves are used to treat kidney stones and alleviate urinary disorders. In India, chicory has found extensive application in managing liver-related ailments, including jaundice and liver enlargement, as well as conditions like diabetes and rheumatism. Furthermore, the plant's traditional use extends to Afghanistan, where its aqueous root extracts have historically been used as a remedy for malarial fevers, a practice supported by modern findings that highlight its antimalarial compounds such as lactucopicrin and lactucin(15, 16). In various regions of Europe, such as Italy, Poland, and Serbia, chicory has been employed in the form of teas and decoctions to aid digestion, purify the blood, and alleviate symptoms related to hepatic disorders. In Bosnia and Herzegovina, its aerial parts are used in decoctions for their antiseptic, spasmolytic, and antihyperlipidemic properties, while in Bulgaria, it serves as a hypoglycemic agent and digestive stimulant(16).

The following table summarizes the traditional applications of chicory across different regions:

Region	Traditional Uses	Part Used	Preparation
Afghanistan	Treatment of malarial fever	Root	Aqueous extract
India	Liver disorders, diabetes, jaundice, rheumatism, gout	Whole plant, Root	Decoction
South Africa	Liver tonic, jaundice treatment, infant restorative	Leaves, stems, roots	Tea, syrup
Turkey	Kidney stones, wound healing, urinary disorders	Roots, leaves	Decoction, ointment
Bosnia & Herzegovina	Hepatic disorders, antiseptic, spasmolytic, antihyperlipidemic	Aerial parts	Decoction
Italy	Blood pressure, digestive aid, antispasmodic	Leaves, roots	Decoction
Pakistan	Diabetes treatment	Roots	Decoction
Serbia	Digestive aid, antidiarrheal, liver support	Flower, roots	Infusion, decoction

These traditional uses underscore the global recognition of chicory as a valuable medicinal herb. Despite regional variations in preparation methods and specific applications, the commonality lies in the plant's role in promoting digestive health, supporting liver function, and providing relief from metabolic disorders(17).

Pharmacological Activities of *Cichorium intybus*

The therapeutic potential of *Cichorium intybus* is rooted in its complex phytochemical composition, with various studies exploring its pharmacological activities. Despite its widespread traditional use, scientific investigations remain relatively nascent, with much of the existing research focusing on hydro-alcoholic extracts derived from different parts of the plant(18).

Antimicrobial Properties

Chicory has demonstrated significant antimicrobial activity, particularly against oral pathogens such as *Actinomyces naeslundii*, *Streptococcus mutans*, and *Prevotella intermedia*. These effects are primarily attributed to organic acids present in the plant's extracts, including succinic acid, oxalic acid, quinic acid, and shikimic acid. These compounds not only inhibit bacterial adhesion and biofilm formation but also facilitate the detachment of dead cells from surfaces, thereby disrupting established biofilms(19). Further studies have shown that extracts from chicory seeds effectively combat harmful pathogens like *Candida albicans*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Escherichia coli*. Root extracts have also demonstrated varying degrees of antimicrobial activity against *Salmonella typhi*, *Bacillus subtilis*, and *Micrococcus* species. Additionally, the guaianolide-rich root extract has exhibited antifungal efficacy against dermatophytes such as *Trichophyton tonsurans*, *Trichophyton rubrum*, and *Trichophyton violaceum*. These findings suggest that chicory holds promise as a natural antimicrobial agent, particularly in an era where resistance to conventional antibiotics is becoming increasingly prevalent(19).

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Anthelmintic Potential

Chicory's anthelmintic properties have been extensively studied in grazing animals, revealing its ability to reduce gastrointestinal nematode infections. Research suggests that sesquiterpene lactones and condensed tannins contribute significantly to this antiparasitic effect. Studies conducted on sheep and cattle have demonstrated a reduction in parasitic loads, especially in the case of *Haemonchus contortus*, a nematode responsible for severe infections in ruminants. The larval motility assays revealed a dose-dependent reduction in the movement of both gastrointestinal nematodes and deer lungworms, providing strong evidence for chicory's antiparasitic efficacy in livestock management(20).

Antimalarial Activity

Traditional accounts from Afghanistan have highlighted the use of chicory root infusions in treating malarial fever. Modern pharmacological research has validated these claims, identifying sesquiterpene lactones such as lactucopicrin and lactucin as active antimalarial compounds. These constituents have demonstrated significant inhibitory effects on *Plasmodium falciparum*, the parasite responsible for the most severe form of malaria. Their ability to suppress parasitic growth at relatively low concentrations highlights the potential of chicory as a complementary therapy in malaria management(21).

Hepatoprotective Effects

The hepatoprotective properties of *Cichorium intybus* have been extensively documented, particularly in traditional medicine systems such as Ayurveda. Chicory is a key component of polyherbal formulations like Liv-52 and Jigrine, which are used to treat liver dysfunctions. Clinical trials have shown that these formulations can significantly lower hepatic enzyme levels, including aspartate aminotransferase (AST) and alanine aminotransferase (ALT), in patients with liver cirrhosis and other hepatic conditions(22). Experimental studies using animal models have further substantiated chicory's liver-protective effects. Administration of hydro-methanolic seed extracts has been shown to mitigate hepatic damage induced by acetaminophen and carbon tetrachloride, reducing enzyme levels associated with liver toxicity. Moreover, phenolic compounds such as esculetin and guaianolide sesquiterpene glycosides derived from chicory have demonstrated hepatoprotective effects by preventing oxidative damage and reducing hepatic steatosis(23).

Emerging Research Trends and Knowledge Gaps

Despite the promising pharmacological activities associated with *Cichorium intybus*, several gaps remain in the current body of research. Much of the existing data is derived from preclinical studies, with limited clinical trials conducted on human subjects. Additionally, variations in phytochemical content due to environmental factors, soil composition, and cultivation practices present challenges in standardizing chicory-based treatments. Future research should focus on conducting large-scale clinical trials to validate the efficacy of chicory in treating specific health conditions and establishing standardized dosing guidelines(23). The growing interest in natural remedies and plant-based therapeutics underscores the need for continued exploration of chicory's pharmacological potential. By bridging the gap between traditional knowledge and modern scientific validation, chicory can be positioned as a valuable therapeutic agent in contemporary healthcare practices(24).

Antidiabetic Activity

Cichorium intybus has garnered significant scientific attention for its antidiabetic properties, aligning with its traditional use in managing metabolic disorders. Various studies have highlighted the plant's hypoglycemic and hypolipidemic effects, particularly in animal models induced with diabetes using streptozotocin, a compound known for its ability to damage insulin-producing cells. In experiments involving male Sprague Dawley rats, ethanolic extracts of the whole plant significantly lowered blood glucose levels during oral glucose tolerance tests. At a dosage of 125 mg/kg body weight, treated animals exhibited marked reductions in both blood triglycerides and cholesterol levels. Moreover, the activity of hepatic glucose-6-phosphatase—a key enzyme involved in glucose metabolism—was notably reduced in treated diabetic rats compared to untreated controls(25). Further investigations into the aqueous extracts of chicory seeds provided additional evidence supporting its antidiabetic efficacy. In experimental models involving male Wistar albino rats, chicory treatment ameliorated weight loss typically associated with early- and late-stage diabetes. Treated animals displayed improved glucose tolerance, reduced fasting glucose levels, and increased insulin production, particularly in early-stage diabetes. Chicory also normalized several biochemical markers, including alanine aminotransferase (ALT), triacylglycerol, total cholesterol, and glycosylated hemoglobin levels, suggesting a broad-spectrum metabolic benefit(25). The beneficial effects of *Cichorium intybus* extend beyond glucose regulation. Studies have demonstrated that its administration in diabetic rats led to lowered malondialdehyde levels—an indicator of oxidative stress—alongside increased glutathione levels and restored anticholinesterase activity. This suggests that chicory

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possesses antioxidant properties that may mitigate complications associated with diabetes-induced oxidative damage. Additionally, chicoric acid, a bioactive compound found in chicory, has shown promise as an insulin-sensitizing agent, enhancing glucose uptake by muscle cells and stimulating insulin secretion from pancreatic β-cells(26).

Gastroprotective and Anti-inflammatory Activities

The gastroprotective effects of *Cichorium intybus* are deeply rooted in traditional Turkish medicine, where decoctions of its roots have been historically used to prevent ulcerogenesis. Experimental studies using ethanol-induced ulcer models in Sprague-Dawley rats revealed that chicory root decoctions administered prior to ethanol exposure prevented over 95% of ulcer formation, highlighting its potent antiulcerogenic potential(27). In terms of anti-inflammatory activity, ethyl acetate extracts of chicory have been evaluated for their ability to suppress cyclooxygenase-2 (COX-2) expression, a key enzyme involved in inflammatory responses. Studies using human colon cancer cell lines (HT-29) demonstrated that chicory extracts inhibited the production of prostaglandin E2 in a dose-dependent manner, effectively reducing inflammation mediated by tumor necrosis factor (TNF). This suggests that chicory could be a valuable therapeutic agent for managing chronic inflammatory conditions and related cancers(27).

Analgesic and Sedative Effects

The analgesic properties of *Cichorium intybus* are primarily attributed to its sesquiterpene lactones, including lactucopicrin, lactucin, and 11,13-dihydrolactucin. In animal models, these compounds exhibited significant pain-relieving effects in tail-flick and hot-plate tests, with lactucopicrin demonstrating comparable efficacy to ibuprofen at a lower dose. Additionally, these compounds have shown sedative properties in mice, as evidenced by decreased spontaneous locomotor activity, suggesting a dual role as both analgesic and sedative agents(28).

Antioxidant Activity

The antioxidant potential of *Cichorium intybus* has been extensively documented through various biochemical assays. Studies have demonstrated its ability to scavenge free radicals, particularly using DPPH (2,2-diphenyl-1-picrylhydrazyl) assays. Polyphenolic fractions derived from chicory roots exhibited significant radical-scavenging activity, comparable to standard antioxidants like Trolox(29). Chicory's antioxidant action extends to cellular systems, where it has been shown to inhibit lipid peroxidation in rat hepatocyte membranes induced by carbon tetrachloride, thereby protecting against oxidative damage. Furthermore, studies involving bacterial cultures exposed to oxidative stress confirmed chicory's ability to enhance bacterial growth resilience, suggesting broader biological relevance for its antioxidant properties. Red chicory phenolics, in particular, have shown comparable efficacy in neutralizing synthetic radicals, further supporting the plant's potential in mitigating oxidative stress-related diseases(29).

Tumor-Inhibitory Activity

Emerging evidence points to the potential antitumor activity of *Cichorium intybus*. Ethanolic extracts of its roots have been shown to significantly inhibit Ehrlich ascites carcinoma in mice, extending their lifespan by approximately 70%. In vitro studies have demonstrated the ability of chicory leaf macerates to suppress the proliferation of amelanotic melanoma cell lines (C32). Furthermore, magnolialide, a sesquiterpene lactone isolated from chicory roots, has exhibited cytotoxic effects against multiple cancer cell lines while simultaneously promoting the differentiation of leukemia cells. These findings suggest that chicory possesses compounds capable of targeting various cancer pathways, warranting further investigation for its potential use as an adjunct in cancer therapy(29).

Antiallergic and Immunomodulatory Activities

Chicory's potential as an antiallergic agent has been validated in both in vitro and in vivo models. Aqueous extracts of the plant have been shown to suppress mast cell-mediated allergic responses, including systemic anaphylaxis and passive cutaneous anaphylaxis in rodents. The treatment significantly reduced blood histamine levels while increasing intracellular cyclic AMP levels, highlighting its ability to stabilize mast cells and mitigate allergic reactions(29). Additionally, *Cichorium intybus* exhibits immunomodulatory effects. Studies have revealed that ethanol extracts of its roots can counteract ethanol-induced immunotoxic effects in animal models by improving various immunity markers such as delayed-type hypersensitivity responses, natural killer (NK) cell activity, and macrophage function. These findings indicate that chicory holds promise in boosting immune responses, particularly under immunosuppressive conditions(30).

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Other Pharmacological Properties

Beyond its well-documented activities, *Cichorium intybus* exhibits a variety of additional pharmacological effects. For instance, chicoric acid has demonstrated vasorelaxant effects in isolated rat aortic strips, potentially aiding in the management of hypertension. Additionally, its dichloromethane extracts have shown strong anticholinesterase activity, indicating potential applications in managing neurodegenerative conditions like Alzheimer's disease(31). The plant also contains β-sitosterol, a compound recognized for its wound-healing capabilities, which functions through anti-inflammatory and antioxidant pathways while inhibiting enzymes such as hyaluronidase and collagenase. This highlights chicory's multifaceted role in promoting tissue repair and reducing inflammation in wound management(31).

Critical Evaluation and Knowledge Gaps

Despite the broad range of pharmacological activities attributed to *Cichorium intybus*, several gaps persist in the current literature. Many of the existing studies rely heavily on animal models or in vitro systems, with limited clinical trials conducted in human populations. Standardization of extraction methods and dosage forms remains another major challenge, particularly considering variations in phytochemical content due to environmental and genetic factors(12). Emerging research trends suggest a growing interest in isolating specific bioactive compounds from chicory for targeted therapeutic applications. However, the long-term safety, efficacy, and potential interactions with conventional pharmaceuticals remain underexplored. Addressing these gaps through rigorous clinical investigations and standardization of chicory-based products could pave the way for its integration into evidence-based medical practice(20).

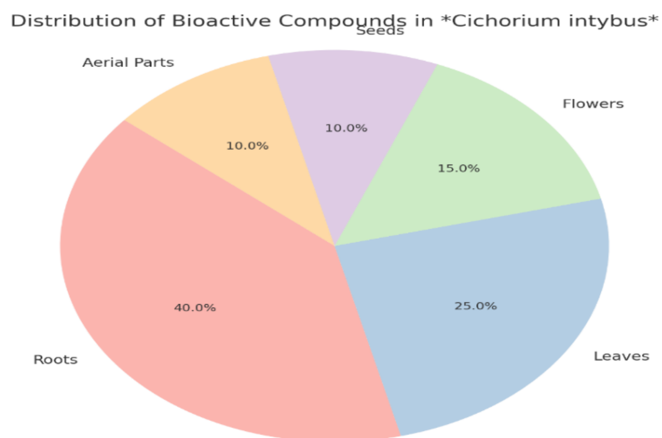

Figure 2 Distribution of Bioactive Compound in Cichorium Intybus

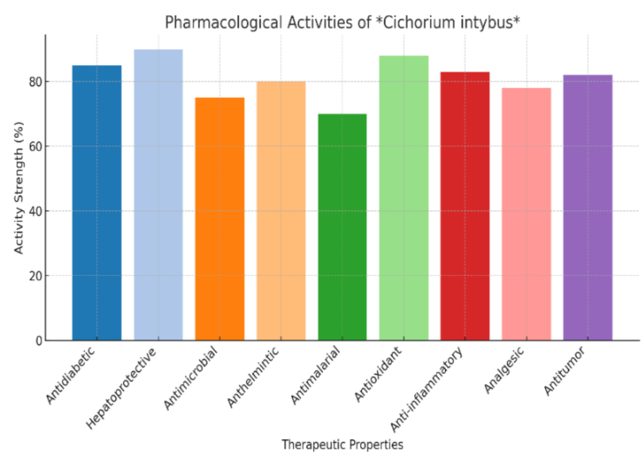

Figure 1 Pharmacological Activities of Cichorium Intybus

DISCUSSION

The exploration of *Cichorium intybus* has revealed a diverse pharmacological profile, aligning closely with its extensive history of traditional use. The plant has demonstrated significant antidiabetic, hepatoprotective, antimicrobial, anti-inflammatory, antioxidant, and antitumor properties, all of which validate its longstanding presence in various traditional medicinal systems. The antidiabetic potential, supported by multiple animal model studies, suggests that chicory could be a valuable adjunct in managing metabolic disorders. These studies consistently reported reductions in blood glucose levels, improved insulin sensitivity, and normalized lipid profiles, highlighting chicory's relevance in addressing the growing global burden of diabetes(6). Beyond its metabolic benefits, chicory's hepatoprotective effects have been well-documented through its ability to regulate hepatic enzyme levels and protect against chemically induced liver damage. These findings resonate with its traditional use in herbal formulations for liver support, offering scientific validation for its role in modern hepatology. Similarly, antimicrobial research has confirmed chicory's efficacy against a broad range of pathogens, including drug-resistant strains, indicating potential as an alternative or complementary treatment in an era of rising antibiotic resistance(9).

The plant's antioxidant and anti-inflammatory activities underscore its role in mitigating chronic diseases associated with oxidative stress and inflammation. Studies involving both in vitro and in vivo models have demonstrated chicory's ability to neutralize free radicals, reduce lipid peroxidation, and inhibit key inflammatory pathways. This suggests a promising role in preventing or managing conditions such as cardiovascular diseases and neurodegenerative disorders(11). Toxicological evaluations of *Cichorium intybus* have

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established a favorable safety profile, with no significant mutagenic or cytotoxic effects observed in various models. Sub-chronic toxicity studies have further reinforced its safe use at appropriate dosages, supporting its inclusion in both dietary and therapeutic applications. However, the findings from toxicity assessments highlight the importance of monitoring dosage levels to prevent potential adverse effects, particularly from high concentrations of secondary metabolites(14).

Despite the promising pharmacological evidence, human clinical trials remain limited and largely exploratory. The few pilot studies available suggest potential benefits in conditions like osteoarthritis and cardiovascular health, but the small sample sizes and short durations restrict the generalizability of these findings. Larger, well-designed clinical trials are necessary to validate the therapeutic efficacy of chicory in human populations and establish standardized dosage guidelines(5, 16). The cultivation of *Cichorium intybus* also presents significant implications for sustainability and commercial viability. Its high inulin content, adaptability to diverse soil conditions, and resilience to environmental stressors make it an attractive crop for industrial applications. The potential for inulin extraction, animal feed production, and use in functional foods further expands its economic importance. However, factors such as soil composition, nutrient availability, and harvest timing can influence inulin yield and phytochemical content, necessitating optimized agricultural practices to ensure consistent quality and output(16).

While the current body of literature highlights the plant's therapeutic promise, several limitations must be acknowledged. Much of the existing research is based on preclinical studies, with a lack of robust clinical evidence to support widespread medical use. Additionally, variations in extraction methods, plant parts used, and phytochemical concentrations complicate the comparison of results across studies. Standardization of research protocols and cultivation practices would help address these discrepancies and promote consistency in outcomes(25). The integration of findings from traditional knowledge and modern pharmacological research underscores the multifaceted therapeutic potential of *Cichorium intybus*. Its broad-spectrum efficacy, coupled with a favorable safety profile, highlights the importance of further research to bridge the gap between ethnomedicine and evidence-based clinical practice. Future studies should focus on large-scale clinical trials, standardization of extraction techniques, and exploration of potential drug-herb interactions. These efforts would provide a stronger foundation for incorporating chicory into mainstream therapeutic regimens and maximizing its health benefits in modern medicine(28).

CONCLUSION

Cichorium intybus has demonstrated a rich history of use, extending from ancient civilizations to modern scientific inquiry. Historically valued as a medicinal plant, coffee substitute, and occasional animal feed, its diverse nutritional profile, including proteins, carbohydrates, and essential minerals, highlights its significance in both traditional and modern health applications. The inulin content of chicory root, recognized for its role in promoting overall well-being, continues to be a focal point for dietary and pharmaceutical innovations. While traditional knowledge has long acknowledged its therapeutic potential, recent research has begun to validate these claims, uncovering its antioxidant, antidiabetic, hepatoprotective, and antimicrobial properties. However, gaps remain in fully understanding the pharmacological effects of many of its bioactive compounds, and further studies are needed to explore its clinical relevance. Additionally, safety considerations, particularly for individuals sensitive to plants in the Asteraceae family, warrant careful attention. Emerging research on its role as a biomonitor for environmental contaminants adds a new dimension to its potential applications but also raises concerns about its safe consumption. Moving forward, integrating traditional wisdom with rigorous scientific investigation will be essential to unlocking the full potential of *Cichorium intybus* as a versatile resource in medicine, nutrition, and sustainable agriculture.

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AUTHOR CONTRIBUTIONS

Author	Contribution
Hamda Tanzeem Khan*	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Sidra Siddiqui	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published
Syed Tahir Ali	Substantial Contribution to acquisition and interpretation of Data Has given Final Approval of the version to be published
Author	**Contribution**
Sana Siddiqui	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Muhammad Akhlaq	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Muhammad Khaleeq Alum	Substantial Contribution to study design and Data Analysis Has given Final Approval of the version to be published

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