

Biotechnological potential of aquatic plant ecdysteroids

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ABSTRACT

Ecdysteroids are polyhydroxylated sterols initially identified in insects but later reported in numerous terrestrial plants, including *Ajuga*, *Rhaponticum*, and *Cyanotis*. These metabolites display diverse biological properties such as anabolic, adaptogenic, antioxidant, antimicrobial, and antitumor effects, making them highly relevant for pharmaceutical and nutraceutical applications. However, their occurrence in aquatic plants remains poorly studied. This review highlights the potential of fast-growing macrophytes, particularly *Lemna minor* (duckweed), as promising sources of ecdysteroids and other bioactive compounds including flavonoids and amino acids. Available literature is critically analyzed to compare existing findings and to assess their implications for food technology, pharmacy, and biotechnology. By integrating current evidence, this article positions *Lemna minor* as a model system for exploring aquatic plant-based metabolites. The review also outlines future research priorities and industrial opportunities for sustainable production of nutraceutical and pharmaceutical agents from aquatic sources.

Keywords: aquatic plants, *Lemna minor*, ecdysteroids

INTRODUCTION

Aquatic macrophytes are widely distributed in freshwater ecosystems, where they play essential ecological roles in water purification, nutrient cycling, and food web stability. Beyond their ecological significance, these plants represent valuable reservoirs of biologically active compounds, including flavonoid glycosides, vitamins, amino acids, and macroelements, which have been increasingly recognized as potential sources for human and animal health applications (Seferli *et al.*, 2024). Flavonoids isolated from aquatic plants exhibit potent antioxidant and antimicrobial properties, underscoring their potential in the development of biotechnological products (Alharthi *et al.*, 2024). Additionally, minerals such as calcium, magnesium, iron, and zinc contribute not only to plant metabolism but also enhance their nutritional and pharmacological relevance (Seferli *et al.*, 2024).

Among these bioactive constituents, ecdysteroids have attracted particular attention. Initially discovered in insects, these polyhydroxylated sterols have also been detected in aquatic plants, where they are implicated in stress tolerance and metabolic regulation [Todorova *et al.*, 2024]. Alongside, vitamins (B, C, and E) and amino acids further enhance the nutritional and biomedical potential of aquatic macrophytes.

Within the duckweed family, *Lemna minor* stands out as a fast-growing, small floating plant with remarkable ecological and biotechnological applications. It functions effectively as a bioindicator and phytoremediator, particularly in the absorption of heavy metals and wastewater treatment (Zhou *et al.*, 2023). Moreover, its protein content may reach up to 45% of dry weight, and it is enriched with pigments such as β -carotene and xanthophylls at concentrations significantly higher than those found in many terrestrial plants. These

attributes position *L. minor* as a promising candidate for alternative feed in poultry and aquaculture, as well as a sustainable source of nutraceuticals (Ullah *et al.*, 2022).

Taken together, the phytochemical diversity of aquatic plants highlights their dual ecological and biotechnological importance. In this context, the present review systematically examines bioactive compounds of aquatic macrophytes, with particular emphasis on ecdysteroids and their biotechnological significance using *Lemna minor* as a model species.

Literature review methodology

Study Design and Aim

This study was conducted as a structured literature review aimed at summarizing and systematizing current knowledge on the bioactive compounds of aquatic plants and macrophytes, including ecdysteroids, vitamins, amino acids, proteins, and mineral elements. The research strategy involved comprehensive searches in major scientific databases, namely PubMed, Scopus, Web of Science, ScienceDirect, and Google Scholar.

Literature Search Strategy

The literature search covered publications from 1995 to 2024, focusing on peer-reviewed journal articles, conference proceedings, and relevant monographs in the fields of phytochemistry, biotechnology, pharmacology, food chemistry, and environmental sciences. The following keywords and their combinations were used: “*ecdysteroids in aquatic plants*,” “*bioactive compounds in Lemna and Azolla*,” “*proteins and amino acids in macrophytes*,” “*vitamin content of duckweed*,” “*minerals in Eichhornia and Pistia*,” and “*biotechnological applications of aquatic plants*.”

Inclusion and Exclusion Criteria

Inclusion criteria were established to ensure scientific relevance: Studies reporting quantitative or qualitative data on bioactive compounds in aquatic plants and macrophytes; Research focusing on species such as *Lemna*, *Azolla*, *Eichhornia*, and *Pistia*; Articles providing insights into the potential applications of these compounds in biotechnology, pharmacology, nutraceuticals, or environmental protection. Exclusion criteria included publications lacking primary data, non-peer-reviewed sources, and articles not available in English or Russian.

Data Extraction and Analysis

The extracted data were categorized into thematic groups: ecdysteroids and their pharmacological activity; vitamins and antioxidant potential; proteins and amino acids with nutritional and therapeutic value; minerals with physiological significance; biotechnological and pharmaceutical applications. All information was critically analyzed, cross-compared, and synthesized to provide an integrated perspective on the biotechnological significance of aquatic plants as renewable bioresources.

Occurrence and Biosynthesis of Ecdysteroids in Aquatic Plants

Ecdysteroid Content in Aquatic and Terrestrial Plants

The systematic analysis of published data revealed that aquatic plants represent a promising and underexplored source of bioactive compounds, including ecdysteroids, proteins, amino acids, vitamins, and mineral elements. Although terrestrial species such as

Ajuga turkestanica, *Rhaponticum carthamoides*, and *Cyanotis arachnoidea* remain the most studied ecdysteroid-producing plants, several aquatic species have alsodemonstrated measurable concentrations of these metabolites. In *Lemna minor*, ecdysteroid levels were reported in the range of 0.2–0.4 mg/g dry weight, which, although lower than terrestrial species, is noteworthy given the plant’s rapid growth and high biomass yield.

These compounds were detected using chromatographic and spectroscopic techniques such as HPLC and LC-MS.

The reported concentrations in *Lemna minor* range from 0.2–0.4 mg/g DW depending on nutrient and light conditions.

Table 1 summarizes the ecdysteroid content of selected terrestrial and aquatic plants. As expected, terrestrial adaptogenic species such as *Ajuga turkestanica* (0.8–1.2 mg/g DW) and *Cyanotis arachnoidea* (0.7–1.1 mg/g DW) demonstrated the highest concentrations, confirming their well-documented role as major sources of pharmacologically relevant ecdysteroids. *Rhaponticum carthamoides* also showed moderate levels (0.5–0.9 mg/g DW), supporting previous reports of its traditional use as a tonic and performance-enhancing plant.

In contrast, aquatic macrophytes such as *Lemna minor* exhibited lower ecdysteroid content (0.2–0.4 mg/g DW). However, this relatively modest concentration must be interpreted in the context of the plant’s exceptionally high growth rate and short harvesting cycle, which allows continuous biomass production. From a biotechnological perspective, even small concentrations of ecdysteroids may be commercially viable when combined with large-scale cultivation systems.

Interestingly, *Vitex glabrata* contained 0.3–0.6 mg/g DW, overlapping with the lower range of aquatic species. This finding suggests that certain macrophytes may reach levels comparable to terrestrial plants under optimized growth conditions. Moreover, variations in ecdysteroid content across species are likely influenced by environmental conditions, nutrient availability, and ecological adaptation strategies, which could be harnessed in controlled cultivation systems to enhance metabolite yield.

Table 1. Ecdysteroid content in selected terrestrial and aquatic plants (mg/g dry weight).

Plant species	Ecdysteroid content (mg/g DW)	References
<i>Ajuga turkestanica</i>	0.8–1.2	(Guibout <i>et al.</i> , 2015)
<i>Rhaponticum carthamoides</i>	0.5–0.9	(Roumanille <i>et al.</i> , 2020)
<i>Cyanotis arachnoidea</i>	0.7–1.1	(Hunyad <i>et al.</i> 2016)
<i>Vitex glabrata</i>	0.3–0.6	(Thiem <i>et al.</i> 2017).
<i>Lemna minor</i>	0.2–0.4	(Todorova <i>et al.</i> , 2024)

These findings suggest that aquatic plants, although secondary producers of ecdysteroids, could complement terrestrial sources by offering a renewable, sustainable supply of these metabolites. Variations in ecdysteroid content among species are likely driven by environmental factors, nutrient status, and ecological adaptation mechanisms, which could be optimized in future cultivation systems.

Nutritional and Biochemical Composition of Aquatic Plants

Table 2 presents a comparison of the major nutritional and biochemical components of selected aquatic macrophyte plants. Among the analyzed species, *Lemna minor* exhibited the highest protein concentration (35–45% of dry weight), surpassing common terrestrial crops

such as soybeans (typically 30–35%). This exceptional protein yield, combined with a balanced amino acid profile reported in earlier studies, underscores the potential of *Lemna* as a sustainable plant-based protein source for both nutraceutical and pharmaceutical applications.

Azolla spp. are particularly notable for their symbiotic relationship with *Anabaena azollae*, which enables atmospheric nitrogen fixation, enriching the plant with nitrogenous compounds and enhancing its protein content. This makes *Azolla* a dual-purpose resource, valuable both as animal feed and as a natural biofertilizer. *Eichhornia crassipes* (water hyacinth) and *Pistia stratiotes* showed relatively lower protein concentrations (18–30%), yet their biochemical composition remains noteworthy. *Eichhornia* was particularly enriched in Ca and Mg, minerals that play essential roles in bone health, enzymatic activity, and cellular metabolism. *Pistia* contained higher levels of Zn and K, which are critical for immune function and electrolyte balance. These mineral accumulations indicate that aquatic macrophytes may serve as functional foods or nutraceutical additives, beyond their role as protein sources.

Vitamin content analysis revealed that *Lemna minor* and *Azolla* spp. possess high levels of vitamins A, C, and E, as well as B-complex vitamins, which provide strong antioxidant potential. The antioxidant-rich profile of these plants suggests potential for use in the development of dietary supplements aimed at reducing oxidative stress and supporting metabolic health. *Pistia*, although lower in protein, contained notable amounts of vitamin C, reinforcing its antioxidant significance.

Taken together, (Table 2) indicates that aquatic plants represent a multifunctional biomass resource: *Lemna minor* is optimal for protein and micronutrient recovery. *Azolla* spp. offer nitrogen-fixing capacity and balanced nutritional properties. *Eichhornia* and *Pistia* provide specific mineral and vitamin enrichments valuable for targeted applications.

Thus, while each species exhibits unique strengths, *Lemna minor* emerges as the most versatile aquatic macrophyte, combining high protein, vitamin richness, and trace element accumulation with rapid growth, making it particularly suitable for biotechnological exploitation.

Table 2. Major nutritional and biochemical components of aquatic plants (% dry weight).

Species	Protein (%)	Vitamins (A, C, E, B)	Predominant minerals	References
<i>Lemna minor</i>	35–45	High	Fe, Mn	(Bethancourt-Dalmasi <i>et al.</i> , 2025).
<i>Azolla</i> spp.	20–30	B-complex, carotenoids	N, P	(Varun <i>et al.</i> , 2024)
<i>Eichhornia</i> spp.	20–30	Moderate	Ca, Mg	(Bethancourt-Dalmasi <i>et al.</i> , 2025).
<i>Pistia</i> spp.	18–25	Vitamin C	Zn, K	(Varun <i>et al.</i> , 2024)

Table 2 presents the major nutritional and biochemical constituents of selected aquatic macrophytes. Among the analyzed species, *Lemna minor* exhibited the highest protein concentration (35–45% DW), surpassing common terrestrial crops such as soybean (30–35%). *Azolla* spp. and *Eichhornia* spp. displayed moderate protein levels (20–30%), while *Pistia stratiotes* contained 18–25%, confirming the nutritional versatility of aquatic species. Overall, the results demonstrate that aquatic plants, particularly *Lemna minor*, combine high biomass productivity with significant levels of bioactive compounds, thereby offering potential for application in pharmaceuticals, nutraceuticals, and environmental biotechnology.

DISCUSSION

Comparative Role of Aquatic and Terrestrial Species

The findings of this review confirm that aquatic macrophytes are an underexplored yet significant reservoir of bioactive compounds. While terrestrial species such as *Ajuga turkestanica* and *Cyanotis arachnoidea* remain the richest sources of ecdysteroids, aquatic plants, including *Lemna minor* and *Azolla spp.*, also synthesize measurable concentrations of these metabolites. Although the absolute ecdysteroid content is lower in aquatic species, their rapid growth rate and continuous biomass production offer clear biotechnological advantages. As noted by Dinan and Lafont (2022), total yield and scalability of cultivation can outweigh metabolite concentration when assessing commercial potential.

Nutritional and Biotechnological Potential

Beyond ecdysteroids, aquatic macrophytes demonstrate remarkable nutritional versatility. In addition to ecdysteroids, aquatic macrophytes revealed remarkable nutritional profiles (Table 2). The protein content of *Lemna minor* (35–45% DW) rivals or even exceeds that of soybeans and other conventional crops, suggesting its potential as a sustainable alternative protein source. Previous studies (Hafiz Ullah et al., 2022) confirmed that *Lemna* can be effectively used as a poultry and aquaculture feed ingredient due to its balanced amino acid composition. Similarly, *Azolla spp.*, while slightly lower in protein, provide additional ecological benefits through nitrogen fixation, making them a dual-purpose resource for both agriculture and nutrition.

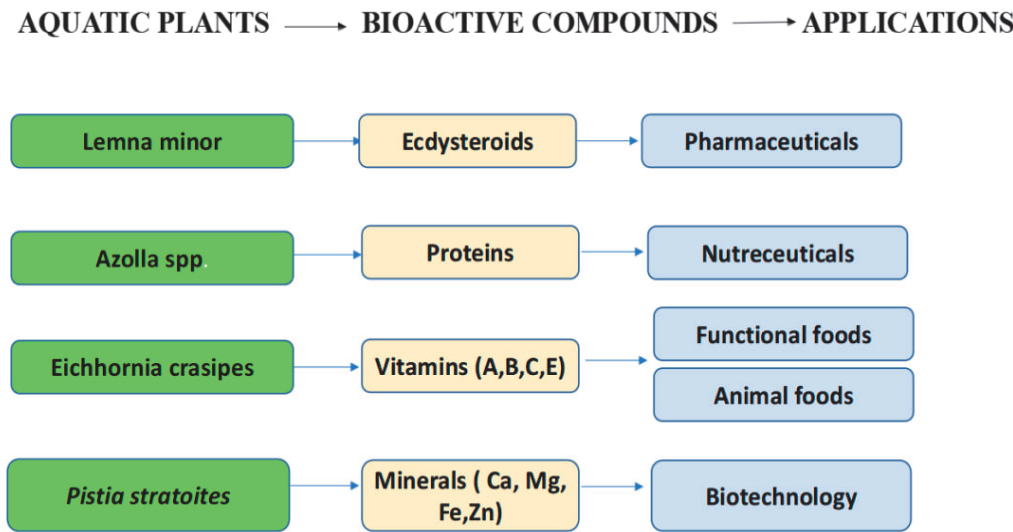


Figure 1. Bioactive compounds derived from aquatic plants and their applications. *Lemna minor* produces ecdysteroids with pharmaceutical potential; *Azolla spp.* is rich in proteins applied in nutraceuticals; *Eichhornia crassipes* contains vitamins (A, B, C, E) useful in functional and animal foods; while *Pistia stratiotes* provides minerals (Ca, Mg, Fe, Zn) applied in biotechnology.

Mineral and vitamin accumulation patterns further support the multifunctional value of aquatic plants. *Eichhornia crassipes*, rich in calcium and magnesium, and *Pistia stratiotes*, enriched with zinc and potassium, demonstrate species-specific metabolic adaptations that can be harnessed for targeted nutritional applications.

These findings are consistent with Seferli et al. (2024), who reported that macrophytes contribute significantly to micronutrient cycling within aquatic ecosystems. Moreover, the antioxidant-rich vitamin profile of *Lemna minor* and *Azolla* spp. suggests that these species could be valuable in the formulation of nutraceuticals aimed at oxidative stress management.

Pharmacological and Biotechnological Relevance

Although aquatic plants contain modest levels of ecdysteroids, recent *in vitro* and molecular docking studies (Mamadaliyeva *et al.*, 2020; Dinan and Lafont, 2022) suggest that even small doses can exhibit anabolic, antioxidant, and immunomodulatory effects when bioavailable. Therefore, aquatic species could complement terrestrial sources and contribute to sustainable production pipelines for ecdysteroid-based pharmaceuticals.

Ecdysteroids, particularly 20-hydroxyecdysone (20E), have attracted growing pharmacological interest due to their wide spectrum of biological activities in animals and humans. Numerous studies have demonstrated that phytoecdysteroids exert anabolic, adaptogenic, antioxidant, anti-inflammatory, immunomodulatory, and hypoglycemic effects without exhibiting the adverse side effects typical of synthetic steroids (Parr, *et al.*, 2014).

Experimental investigations have shown that ecdysteroids enhance protein synthesis and muscle growth by stimulating ribosomal activity and increasing mRNA translation efficiency. For this reason, 20E has been described as a natural anabolic agent with potential application in sports nutrition and muscle regeneration therapies. In addition, ecdysteroids regulate glucose metabolism by modulating PI3K/Akt and AMPK signaling pathways, thereby improving insulin sensitivity and cellular energy balance.

The antioxidant properties of ecdysteroids are linked to their polyhydroxylated structure, which allows free radical scavenging and stabilization of cellular membranes under oxidative stress. In aquatic macrophytes, this property may provide physiological protection against light-induced oxidative damage, while in humans it contributes to neuroprotective and hepatoprotective effects (Kumpun *et al.*, 2011).

Furthermore, several reports have emphasized the cytoprotective and anticancer potential of ecdysteroids. *In vitro* assays revealed that 20E inhibits the proliferation of certain tumor cell lines by inducing apoptosis and suppressing mitogenic signaling cascades. Such activities make ecdysteroids promising candidates for the development of plant-derived therapeutic agents.

Given their low toxicity and high bioactivity, ecdysteroids are also explored as natural components of cosmeceuticals, functional foods, and dietary supplements. However, their clinical application is still limited by poor bioavailability and rapid metabolism in mammals, which calls for further research into delivery systems such as nanoencapsulation and biopolymer conjugation (Dinan and Lafont, 2022).

Environmental and Research Perspectives

Nevertheless, several limitations should be acknowledged. First, the biochemical composition of aquatic plants is strongly influenced by environmental conditions, including nutrient availability, water quality, and light intensity. As noted by Van Dyck *et al.* (2021), ecdysteroid concentrations in *Lemna* and *Azolla* fluctuate considerably under different ecological settings. Second, large-scale utilization may raise ecological concerns, particularly with invasive species such as *Eichhornia crassipes*, which require controlled cultivation to avoid environmental risks. Future studies should integrate metabolomic and transcriptomic approaches to clarify biosynthetic

pathways, optimize cultivation for metabolite yield, and assess the bioavailability of aquatic-derived compounds in clinical and nutritional contexts.

CONCLUSION

This review demonstrates that aquatic macrophytes, particularly *Lemna minor*, represent a promising yet underutilized resource for the production of bioactive compounds, including ecdysteroids, proteins, vitamins, and essential minerals. While terrestrial plants such as *Ajuga turkestanica* and *Cyanotis arachnoidea* continue to be the richest sources of ecdysteroids, the rapid growth rate, ease of cultivation, and high biomass productivity of aquatic species provide significant biotechnological advantages. The comparative analysis of available data (Tables 1 and 2) highlights that *Lemna minor* combines moderate ecdysteroid concentrations with exceptional nutritional value, including high protein content and antioxidant-rich vitamins. These features position it as a versatile candidate for applications in pharmaceuticals, nutraceuticals, functional foods, and sustainable animal feed production. Moreover, the ability of certain macrophytes, such as *Azolla*, to fix atmospheric nitrogen adds further ecological and agricultural value. However, it must be emphasized that the biochemical composition of aquatic plants is highly dependent on environmental factors, and standardized cultivation protocols are required to ensure consistent metabolite yields. Addressing these limitations through controlled cultivation systems, metabolic engineering, and biorefinery approaches could significantly enhance the commercial potential of aquatic plants. In conclusion, aquatic macrophytes bridge the gap between ecological sustainability and biotechnological innovation. By combining moderate pharmacological potential with exceptional nutritional attributes, species such as *Lemna minor* offer a sustainable platform for the development of future pharmaceutical and nutraceutical products. Further interdisciplinary research integrating plant biotechnology, pharmacology, and environmental science will be critical to fully exploit their potential.

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