Integral Valorization of *Artemisia annua L.* for Artemisinin Extraction and Bioactive Co-products Recovery within a One Health Framework

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INTRODUCTION

Malaria remains a major global health challenge, and the emergence of partial artemisinin resistance highlights the need for sustainable and resilient supply chains for artemisinin-based medicines. Artemisinin and its derivatives, obtained from *Artemisia annua L.,* are fast-acting antimalarial compounds used in WHO-recommended artemisinin-based combination therapies (ACTs) [1].

Artemisia annua L. is the primary botanical source of artemisinin. As artemisinin content in A. annua is strongly influenced by genotype, environmental conditions, developmental stage, and harvest timing [2,3], this study evaluates the integrated valorisation of plants cultivated in Extremadura (south-western Spain). The approach combines optimisation of artemisinin extraction with recovery of high-value metabolites from process side-streams, including artemisinin acid, arteannuin B, polymethoxylated flavonoids, and chlorophylls.

By fully characterising and optimising this process, this study reduces waste and maximises plant efficiency. The resulting high-value fractions hold strong potential for pharmaceutical, nutraceutical, veterinary, agri-food, or cosmetic applications.

MATERIALS & METHODS

Seed sourcing and varieties: *Artemisia annua L*. accession seeds originating from China, Vietnam and Switzerland were evaluated for agro-climatic adaptation in the experimental plots of CTAEX (Extremadura, SW Spain)

Extraction and purification: Pilot-scale extractions processed 1,200 kg of pelleted dry leaves using technical-grade acetone under continuous recirculation (40-45°C, 4 h), obtaining a crude oleoresin. The crude extract was redissolved in acetone: water (5:4 v/v), filtered through a Celite bed to remove waxes and lipids, diluted with cold water (10:4v/v), and refrigerated (5°C, 48 h). Pure artemisinin was obtained via organic dissolvent extraction and hot recrystallization.

Chemical analysis and quantification : Artemisinin and its derivatives were measured by HPLC-MS, flavonoid compounds were identified by HPLC-UV, and chlorophylls were quantified by UV-Vis spectrophotometry at their characteristic wavelengths (645 nm and 663 nm).

RESULTS & DISCUSSION

The results show clear variability in metabolite accumulation depending on the agricultural variety and the phenology of the crop, with maximum biomass and artemisinin yields observed when harvesting takes place in the pre-flowering stage or at the onset of flowering (Fig. 1).

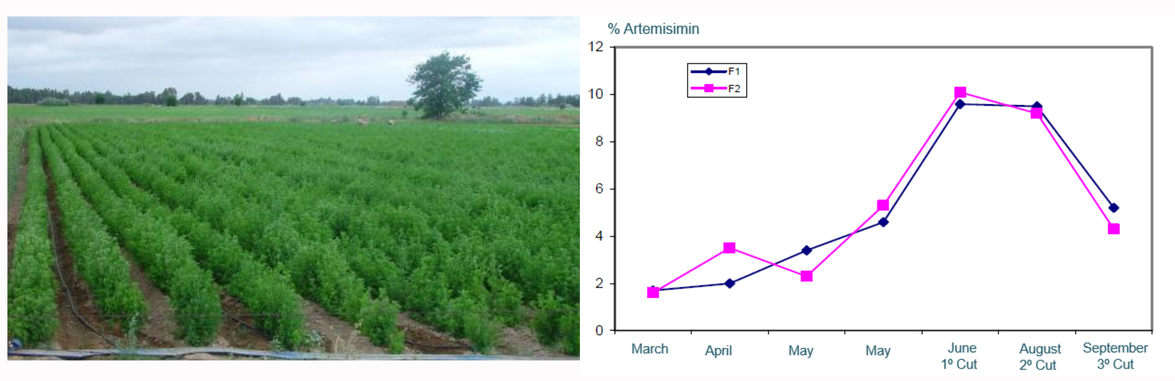

Figure 1. Left: Artemisia annua field crop in Extremadura. Right: Evolution of artemisinin synthesis across the phenological cycle.

Following pelletization and solvent extraction, a primary oleoresin was obtained (6% mass yield, ~5.94% artemisinin content), from which pure artemisinin was successfully isolated via crystallisation as potential high-value pharmaceutical product .(Fig2).

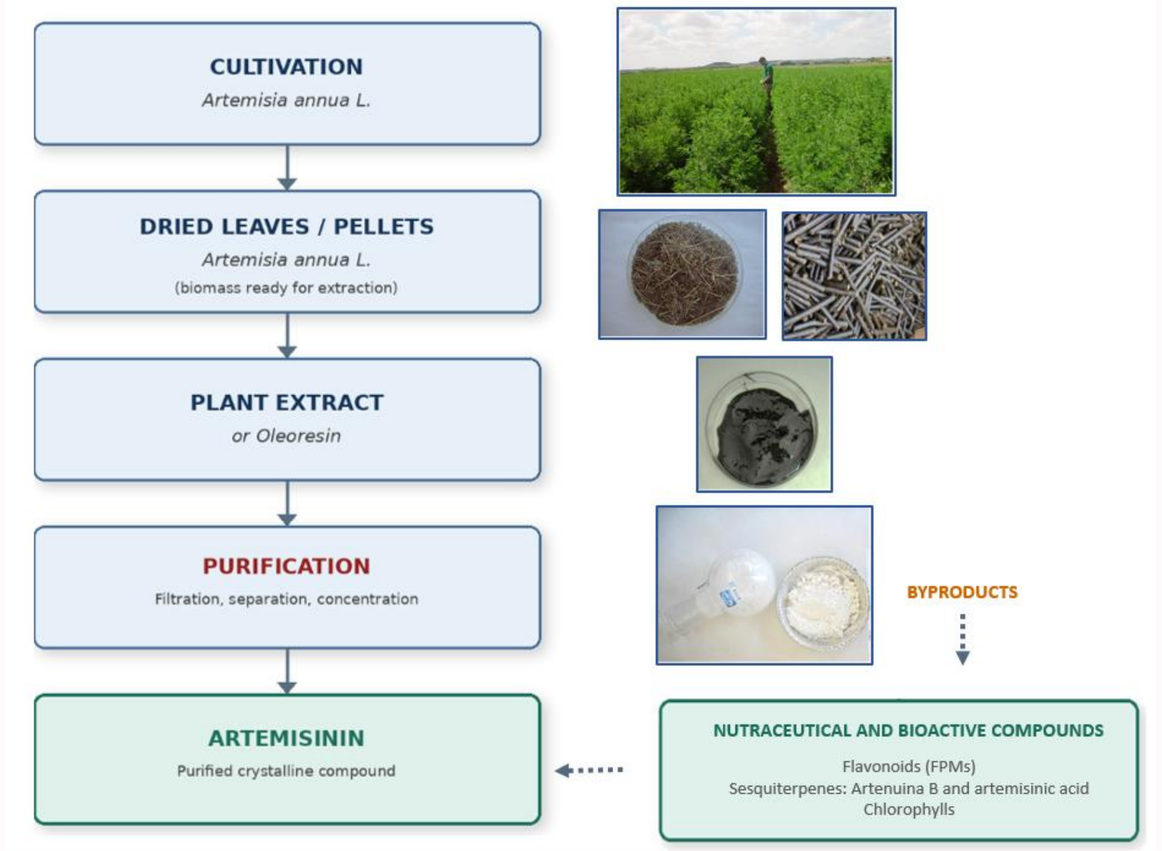

Figure 2 . Extraction of artemisinin and bioactive compounds with nutraceutical / phytotherapeutic potential

Analysis of process by-products confirmed the efficient recovery of multiple high-value compounds, including arteannuin B, artemisinin acid, polymethoxylated flavonoids, and chlorophylls. These findings demonstrate that optimizing extraction parameters while simultaneously recovering high-value side-stream metabolites maximizes total plant biomass utilization and minimizes process waste.

CONCLUSIONS

Bioactive metabolite accumulation is highly dependent on the cultivar, phenological stage, and climatic conditions, highlighting the critical importance of selecting suitable varieties, cultivation locations, and optimal harvest timing to maximize bioresource recovery.

Pure **artemisinin** was successfully isolated, meeting the quality standards required for pharmaceutical-grade applications, particularly in WHO-recommended artemisinin-based combination therapies (ACTs). Additionally, these fractions show strong potential for nutraceutical formulations and veterinary applications, including antiparasitic, boosting immunity and treatments specifically targeting leishmaniasis. Process side-streams yielded valuable secondary metabolites with significant phytotherapeutic, pharmaceutical, cosmetic and agrifood potential. Notably, **artemisinic acid** serves as a key intermediate for the semi-synthetic production of artemisinin, while recovered **polymethoxylated polyphenols** may offer synergistic benefits that enhance antimalarial efficacy. Furthermore, **chlorophylls** offer direct applications as natural colorants (E140/E141) and functional pigments in animal feed and hygiene products, whereas **arteannuin B** also, a precursor of artemisinin, exhibits promising experimental antifungal, anti-inflammatory, and anti-tumour activities.

Overall, the integrated biorefinery model represents a promising **One Health strategy** by linking human health, animal health, and sustainability within a single *Artemisia annua* value chain. The recovery of pharmaceutical-grade artemisinin could support the production of artemisinin-based combination therapies (ACTs) and veterinary treatments, while the co-extraction of bioactive fractions creates opportunities for the future development of pharma, veterinary, nutraceutical, cosmetic, food and functional-feed applications. Although these results remain preliminary, the study demonstrated the potential of a biorefinery to improve profitability and minimizes process waste in the processing of *A. annua*.

LITERATURE CITED

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