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"Characterization of the in vitro cosmetic efficacy of an extract derived from a by-product of the agri-food industry"

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1. Introduction

Today, the circular economy plays a crucial role in reducing environmental impact by transforming production waste into reusable resources. In Belgium, some residues have been produced in increasing quantities in recent years and contain numerous high-value-added molecules. The project aims to produce active ingredients from agri-food waste for the cosmetics sector. To achieve this, a by-product biorefining process has been set up. This process has made it possible to recover various fractions that can be used in the cosmetics sector. Only the results obtained for the polyphenolic extract are presented here. In addition to the proof of cosmetic efficacy. The project led to the development of cascade extraction processes, as well as stability and safety studies. Tests were also carried out to integrate the extracts into cosmetic formulas. A theoretical study on scaling up to an industrial scale was also carried out. Polyphenols are one of the families of high-value-added molecules present in the by-product. They demonstrate strong anti-aging activities. Their antioxidant properties, ability to inhibit dermal proteases, and their photoprotective activity have been proved in vitro.

2. Materials and Methods

The polyphenolic extract obtained from the by-products was analyzed in various ways. Its antioxidant properties were determined by DPPH analysis. This is a well-known spectrophotometric method. Enzyme inhibition tests were carried out by measuring tyrosinase inhibition, and conclusions were drawn about the bleaching effect of the extract. Measurement of elastase and collagenase inhibition gave indications of anti-ageing activity. Lastly, anti-inflammatory activity was assessed by measuring lipoxygenase inhibition. Cell culture techniques using fibroblasts were also used. Scratch test Method was used in cell culture to assess cell migration and proliferation. It involves creating a 'scratch' or gap in a monolayer of cells, then monitoring the closure of the wound over time. In order to objectify the anti-aging effect, thanks to cell culture, MTT test was used. It's a colorimetric method used to assess in vitro cell viability, by measuring the metabolic activity of living cells through the reduction of MTT tetrazolium salt to the colored formazan.

3. Results

The results of in vitro analysis are shown below, enzyme inhibition is measured and quantified by spectrophotometry and represented as a percentage.

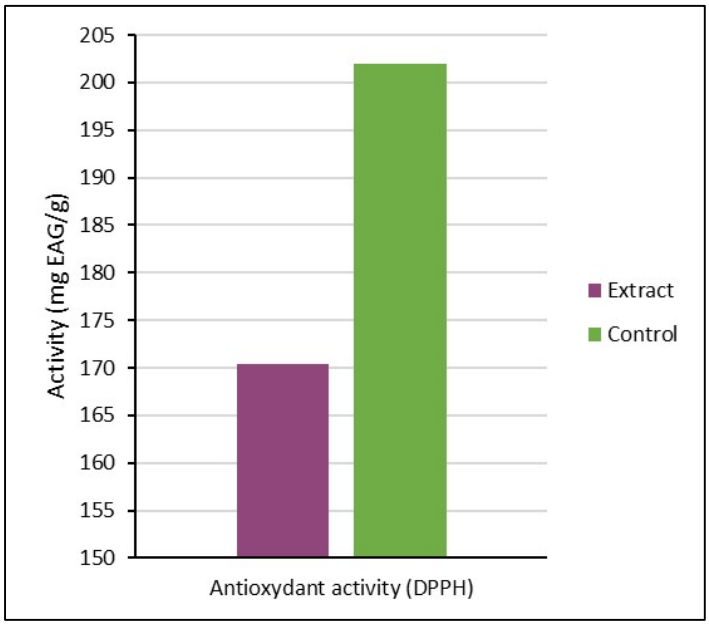

Figure 1. Antioxidant activity comparison

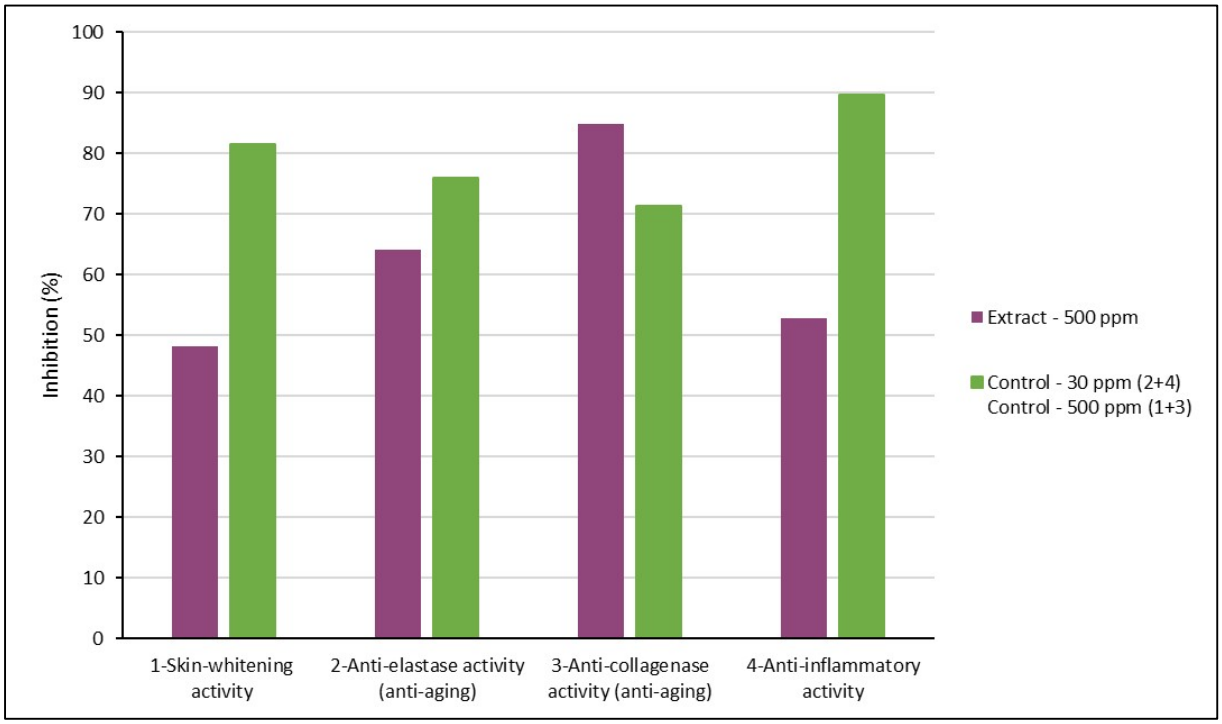

Figure 2. Inhibition activity comparison at different concentrations.

For the MTT Test, the cells were treated with the extract at concentrations ranging from 250 to 1000 ppm. The control was left untreated and cultured with standard culture medium.

Sample absorbance was measured at 570 nm to detect the concentration of formazan in the wells and assess cell viability.

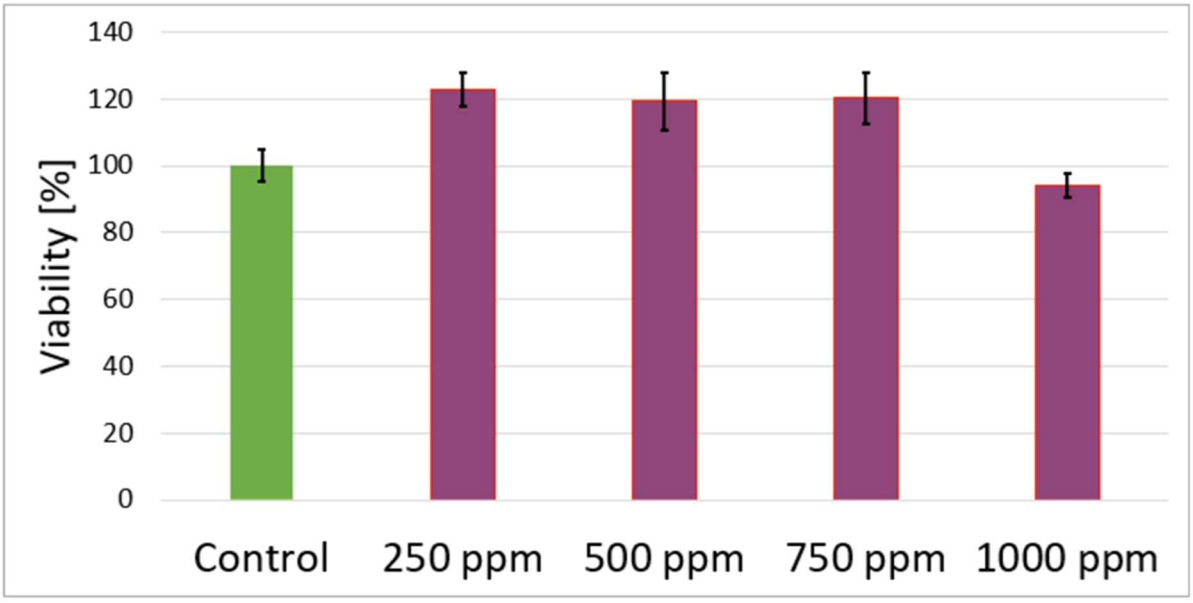

Figure 3. Cell viability for different concentrations of the natural polyphenolic extract.

The control is in green and the polyphenolic extracts in purple. Thanks to this analysis t, the cytotoxic concentration can also be determined.

Finally, scratched cells were treated with the extract at concentrations ranging from 250 to 1000 ppm.

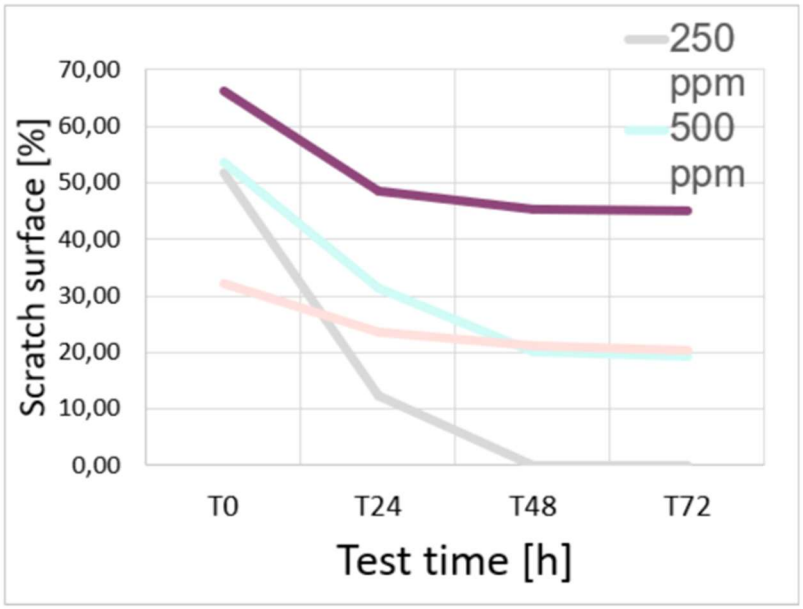

Figure 4. Wound area evolution over time.

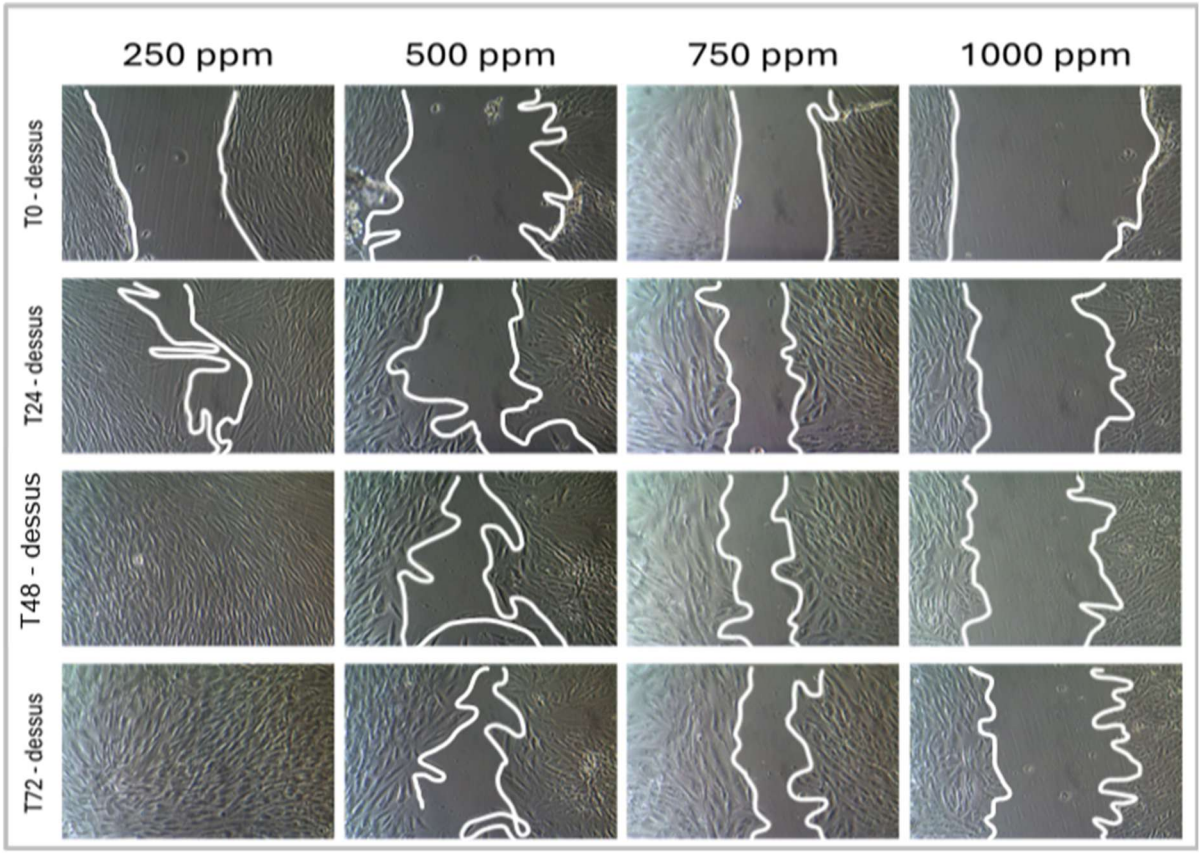

Figure 5. Visual evolution of the wound area over time.

4. Discussion

The antioxidant capacity measured by DPPH was not as good as that obtained with the control. However, other tests have demonstrated the anti-ageing effect that can be correlated with antioxidant capacity. The results give good skin-whitening properties with a percentage >40% for tyrosinase inhibition. Despite its low antioxidant capacity, the anti-ageing effect is confirmed by the elastase and collagenase inhibition with a percentage >60%. Finally, anti-inflammatory was measured thanks to lipoxygenase inhibition. This inhibition shows a percentage >50%, the effects are satisfying. Figures 1 and 2 show the interest of the extract for cosmetic applications.

Figure 3 shows that the cell viability increased between 250 and 750 ppm by 120 %. This proves the anti-ageing effect of the extract studied. Thanks to tests at higher concentrations, the cytotoxicity limit was set at 1000 ppm. Lower concentrations should be used in future formulations. It proves stimulation of cell migration and proliferation.

The last two figures present scratch test results. The best results were obtained with the 250 ppm extract, where the cell space was filled after 48 hours. This concentration will be the best option for final formulation.

5. Conclusion

The developed polyphenolic extract shows promising results across various cosmetic tests, highlighting its potential as an active ingredient in the cosmetic industry. Its origin from an agro-industrial byproduct further enhances its appeal as a sustainable option.