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Introduction

uropean chestnut (Castanea sativa, angiosperm family Fagaceae) has a eastern Mediterranean origin and actually is a significant tree in agricultural and forestry economy. For many centuries, chestnut fruits represented one of the most important food resources of rural areas.

Chestnut world production is estimated in 1.1 million tons (FAO data). Italy, Portugal, Greece, and France represent the main European producers, with a global production of about 12%. A great part of production is used by agro-industries for fresh consumption, industrial processing (shelling, freezing and peeling), and for various food products such as chestnut purée and marron-glacé.



Chestnut manufacture generate a large quantity of wastewaters (ChW) that needs to be properly treated due to organic load content. Several processes are available for wastewater treatment, but until now no efficient and eco-friendly systems have been assessed. The peeling process produces a huge amount of solid residues (ChS), representing about the 10-15% in weight of the chestnut production, that are very rich in biomolecules with antioxidant/antiradical activity. Nowadays, the need to replace synthetic antioxidants used in the pharmaceutical, cosmetic and food industries has promoted the research of new sources of natural antioxidant molecules. The undervalued products obtained from the agriculture and forestry could be used as an inexpensive supply of such compounds.

Research Aim

Development of low-cost efficient technologies for ChW and ChS treatment that lead to the recovery of valuable by-products and energy generation, through anaerobic digestion process, thus reducing the negative impacts of wastes in environment and increasing the competitiveness of chestnut companies.

Materials and Methods

The wastewater and the solid residues were collected from a chestnut processing industry. Several analytical and chromatograph methods, described elsewhere [1] were used to characterize chestnut effluents (ChW). The energy potential value of the wastewaters were estimated using the anaerobic digestion as a process providing the effluent treatment and energy recovery, considering cogeneration (CHP) use.

The extraction of bioactive compounds was carried out on chestnut shells (ChS) using different solvents (water, methanol or ethanol with HCl) and temperatures (20 ÷ 100 °C) in a solid/liquid ratio ranging from 1:10 to 1:40 (w/v).

Total phenols content was determined by the Folin-Ciocalteu method and the results were expressed as mg of gallic acid equivalents per gram of dry matter [2]. Ortho-diphenols and flavonoids amounts were also determined and the values were showed as mg of caffeic acid and catechin equivalents per gram of dry matter respectively.

Antiradical activity was evaluated by a modification of the method described by von Gadow et al. [3] and it is defined as the amount of antioxidant (expressed as μg of total polyphenols) necessary to decrease the initial DPPH concentration by 50 % (EC $_{50}$ = Efficient Concentration).

Results

Effluents coming from chestnut processing industries are toxic to biological degradation mainly due to acid pH and phenols content (Table 1). Anaerobic digestion have been recognized as an effective treatment option to deal with high polluted and/or unbalanced and toxic substrates [1]. From the energetic point of view (Table 2), 1 m3 of chestnut wastewater, digested anaerobically, can potentially provide a energy amount of about 10 kWh. By applying cogeneration system, approximately 3 kWh electricity and 5 kWh of thermal energy will be available

On the other hand, extraction process utilizing environmental friendly solvents (water and alcohols) allows to obtain high added value biomolecules from the solid manufacture wastes. Table 1 reports the chestnut wastewater chemical/physical parameters and Table 2 the energy potential of chestnut wastewaters.

The extraction methods for shells residues were compared considering the biomolecules contents (data not showed). The best result was obtained utilizing hot water at 100 °C in as solid/liquid ratio of 1:40 (data not showed). The ortho-diphenol fraction, provided with the highest antioxidant power among the phenolic molecules, corresponded to 80% of the total phenols. Some interesting phenolic compounds (as Rutin, Gallic, Ellagic, p-Coumaric and Ferulic acid) with antiradical activity were identified in the extracts. The chestnut shells extracts showing highest phenolic amount were also tested for antioxidant properties and their activity was evaluated through DPPH assay (Table 3).

Table 1 Parameters	Chestnut wastewater (Chw)
pН	4.54
Total COD (kg m ⁻³)	5.60 ± 0.00
T5 (kg m ⁻³)	3.36 ± 0.14
VS (kg m ⁻³)	3.00 ± 0.14
Ashes (g L ⁻¹)	0.11 ± 0.04
TSS (kg m ⁻³)	2.69 ± 0.37
VSS (kg m ⁻³)	2.58 ± 0.42
Total Nitrogen (g m ⁻³)	174.72 ± 3.96
Ammonium ion (N g m-3)	7.00 ± 0.00
Total phenols (g GAE/m ⁻³)	60 ± 0.002
Conductivity (mS cm ⁻¹⁾	1.08 ± 0.06
Color (Abs 390nm)	1.58 ± 0.12

Table 2 1m3 - Chestnut wastewaters	Energy available CHP
Methane volume (m3)	1,0
energy (kWh)	9,92
Electricity (kWh)	3,37
thermal energy (kWh)	4,96

Table 3 Biomolecules	Chestnut Wastewater	Chestnut Shells
Total phenols	0.060 ± 0.002 (mg GAE/mL)	17.68 ± 1.09 (mg GAE/g DM)
Ortho- diphenols	0.048 ± 0.003 (mg CAE/mL)	8.29 ± 0.30 (mg CAE/g DM)
Fiavonoids	0.012 ± 0.001 (mg CE/mL)	7.36 ± 0.32 (mg CE/g DM)
Antiradical Activity (EC ₅₀)	8.50 ± 0.14	68.00 ± 3.50

CAE and CE = Gallic acid, Caffeic acid and Catechin alents; EC₈₀ = Efficient Concentration

Future Plans

Further studies are needed to assess the best extraction method useful in recovering the highest amount of bioactive compounds from wastes of Castanea sativa and to obtain the highest yield in biogas/methane production.

Conclusions

The best extraction method for chestnut shells was the water at boiling point and ortho-diphenolic content reproduced the same trend of those obtained for total phenols

- Chemical and biological parameters were performed by identification and quantification of phenolics and activity bioassays
- Only environmental friendly solvents (water and alcohols) were utilized in this research and so they could be safely used for applications in the food and cosmetic industries.
- Anaerobic digestion of ChW allows to treat the effluent and recover its energy potential though biogas/methane production. Values about 10 kWh 1 m³ of ChW can be obtain.