Protein recovery from animal by-products in tannery and rendering industries for biostimulant applications in agriculture

Msc. Mayra Lacruz Asaro
Advanced materials and Technology
Footwear Technological Centre- INESCOP

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1. PROBLEM AND IMPACT
Around 60%* of each animal is converted into food for human consumption.

REMAINING 40% becomes animal by-products (ABPs) transformed by the rendering industry into high added value products.

12 MT material/year are processed

High-protein wastewater

2.85 MT PAPs and edible proteins

From 1000 kg of raw skin, 400 kg are fleshing waste with 30 m³ of effluent wastewater

*328 million livestock European

Fat Processors and Renderers Association (EFPRA), 2021 and Kanagaraj, 2015
Animal by-products applications

According European Regulation Nº1069/2009
By-products valorisation into PAP

ABPs waste were previously pressure-sterilised through a method 1 transformation in compliance with Regulation (EC) No 1069/2009

High-protein wastewater category 3 from rendering industry

Flesh liming category 3 from tanning industry

Meat and Bone meal category 2 from rendering industry
Production of advanced biodiesel from animal wastes using supercritical technologies
Protein recovery and recycling from animal by-products processes

BYPROTVAL

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5 m3
2. VALORISATION METHOD
ENZYMATIC HYDROLYSIS FOR RECOVERY PROTEINS AS BIOSTIMULANTS*

Preparation and characterization**

**pH, ash, moisture content, total and free aminoacids, organic material and nitrogen. Total nitrogen, ammonium nitrogen and organic nitrogen.

Method 1***

Method 5***

Proteolytic enzyme with endo and exo-activity

Mechanical separation

PROTEIN SOLUTION ****

*Regulation (EU) No 142/2011
***Regulation (EU) 1069/2009
****Regulation (EU) 2019/1009
# COMPARISON OF THE PARAMETERS CORRESPONDING TO DIFFERENT ANIMAL BY-PRODUCT USED

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Biostimulant obtained from Cat 2 PAP</th>
<th>Biostimulant obtained from Cat 3 wastewater</th>
<th>Biostimulant obtained from Fleshings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein yield (%)</td>
<td>87.50</td>
<td>97.22</td>
<td>78.18</td>
</tr>
<tr>
<td>Total nitrogen (%)</td>
<td>7.43</td>
<td>7.54</td>
<td>8.05</td>
</tr>
<tr>
<td>Ammonia nitrogen (%)</td>
<td>0.22</td>
<td>0.625</td>
<td>0.625</td>
</tr>
<tr>
<td>Organic nitrogen (%)</td>
<td>5.13</td>
<td>7.41</td>
<td>7.41</td>
</tr>
<tr>
<td>Urea nitrogen (%)</td>
<td>0.008</td>
<td>0.05</td>
<td>0.05</td>
</tr>
<tr>
<td>Total amino acids (% in dry matter)</td>
<td>70.68</td>
<td>60.72</td>
<td>68.06</td>
</tr>
<tr>
<td>Free amino acids (%) in solution</td>
<td>3.53</td>
<td>5.09</td>
<td>4.67</td>
</tr>
<tr>
<td>Dry matter (%)</td>
<td>57.52</td>
<td>56.74</td>
<td>67.25</td>
</tr>
<tr>
<td>Organic matter (%)</td>
<td>53.38</td>
<td>55.85</td>
<td>77.85</td>
</tr>
<tr>
<td>Ashes (% in dry matter)</td>
<td>7.8</td>
<td>7.3</td>
<td>10.5</td>
</tr>
<tr>
<td>Density (kg/L)</td>
<td>1.24</td>
<td>1.23</td>
<td>1.25</td>
</tr>
<tr>
<td>pH</td>
<td>5.6</td>
<td>5.7</td>
<td>5.7</td>
</tr>
</tbody>
</table>
AMINOACID PROFILE CORRESPONDING TO HYDROLYSED PRODUCT OBTAINED FROM DIFFERENT ANIMAL BY-PRODUCT

Hydrolysed protein from cat 2 PAP
Hydrolysed protein cat 3 Wastewater
Hydrolysed protein Lime Fleshings

% Total Aminoacid (g AA/100 g of Protein)

Aspartic acid, Glutamic acid, Serine, Histidine, Glycine, Threonine, Arginine, Alanine, Tyrosine, Valine, Methionine, Phenylalanine, Isoleucine, Leucine, Lysine, trans-Hyp, cis-Hyp, Proline
FREE AMINOACID PROFILE CORRESPONDING TO HYDROLYSED PRODUCT OBTAINED FROM DIFFERENT ANIMAL BY-PRODUCT

- Improve nutrient uptake
- Salinity stress mitigation
- High temperatures effect mitigation
- Improve chlorophyll production

<table>
<thead>
<tr>
<th>Amino Acid</th>
<th>Cat 3 Wastewater</th>
<th>Cat 2 PAP</th>
<th>Lime fleshings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspartic acid</td>
<td>5</td>
<td>10</td>
<td>20</td>
</tr>
<tr>
<td>Glutamic acid</td>
<td>10</td>
<td>15</td>
<td>25</td>
</tr>
<tr>
<td>Serine</td>
<td>15</td>
<td>20</td>
<td>25</td>
</tr>
<tr>
<td>Histidine</td>
<td>20</td>
<td>25</td>
<td>30</td>
</tr>
<tr>
<td>Glycine</td>
<td>25</td>
<td>30</td>
<td>35</td>
</tr>
<tr>
<td>Threonine</td>
<td>30</td>
<td>35</td>
<td>40</td>
</tr>
<tr>
<td>Arginine</td>
<td>35</td>
<td>40</td>
<td>45</td>
</tr>
<tr>
<td>Alanine</td>
<td>40</td>
<td>45</td>
<td>50</td>
</tr>
<tr>
<td>Tyrosine</td>
<td>45</td>
<td>50</td>
<td>55</td>
</tr>
<tr>
<td>Valine</td>
<td>50</td>
<td>55</td>
<td>60</td>
</tr>
<tr>
<td>Methionine</td>
<td>55</td>
<td>60</td>
<td>65</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>60</td>
<td>65</td>
<td>70</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>65</td>
<td>70</td>
<td>75</td>
</tr>
<tr>
<td>Leucine</td>
<td>70</td>
<td>75</td>
<td>80</td>
</tr>
<tr>
<td>Lysine</td>
<td>75</td>
<td>80</td>
<td>85</td>
</tr>
<tr>
<td>trans-hyp</td>
<td>80</td>
<td>85</td>
<td>90</td>
</tr>
<tr>
<td>cis-hyp</td>
<td>85</td>
<td>90</td>
<td>95</td>
</tr>
<tr>
<td>Proline</td>
<td>90</td>
<td>95</td>
<td>100</td>
</tr>
</tbody>
</table>

% Free Amino acid (1g of AA/100g of protein)
3. VALIDATION AND RESULTS
PROTEIN PRODUCT VALIDATION, IN VITRO GERMINATION BIOASSAYS

Study of the growth of Chinese cabbage and lettuce seeds by dosing different concentrations of biostimulant solution. Control sample of Chinese cabbage (a). Growth of Chinese cabbage using a dosage of 0.15 (b). Lettuce growth using a dosage of 0.15 (c). Source: own elaboration, INESCOP.

Seeds growth by 19-42% with the optimal biostimulant dilution concentration in the range of 0.05-0.2%. ISO standard 16086-2 2012.
VALIDATION OF HYDROLYSED PRODUCT FROM WASTEWATER AND PAP AS A BIOSTIMULANT

Improvement in growth by germination (%) vs. % Biostimulant dilution for:
- Lettuce wastewater
- Chinese Cabbage Wastewater
- Chinese Cabbage PAP
- Lettuce PAP
VALIDATION OF HYDROLYSED PRODUCT FROM LIME FLESHING AS A BIOSTIMULANT
4. CONCLUSION
CONCLUSION

- High content of total amino acids is observed in the range of 60-70%, with the results of the products corresponding to category 2 PAPs being higher than those of category 3 wastewater and fleshings.
- The products obtained show a noticeably high protein recovery, in the range of 87-97%, depending on the animal by-product used.
- The best germination growth results were obtained with a low concentration of hydrolysed protein (from fleshing) although the improvement in growth was not considerably higher than with PAP cat 2.
- Valorisation of these animal by-products to produce biostimulants could be replicated and implemented in other waste management plants.
BIOECONOMY CONTRIBUTIONS

Energy-efficiency
Enhancing the potential of renewable energies, saving energy and costs.

Optimising water consumption
Reduction of at least 96% of water consumption by industrial processes.

Intersectoral collaboration
For resource-efficient and high value applications of ABPs category 2 and 3.

Reducing production and waste disposal costs, increasing the profitability of organic waste value creation by recovering up to 100 tonnes of protein per year.

Resource-efficient process
Reduction of 35% emissions in CO2 and chemical substances, and wastewater reduction.

Increased competitiveness
Opportunity for the tannery and rendering industry sectors to close the loop and resource optimization.

Introduce new functionalised bioproducts that will be able to meet current market demands on bio-based products in the agriculture industry.
Σας ευχαριστώ

Project Manager in Advanced materials and Technology
Footwear Technological Centre- INESCOP

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