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Production of orange peel-based ingredients for dairy sheep feed on a pilot scale

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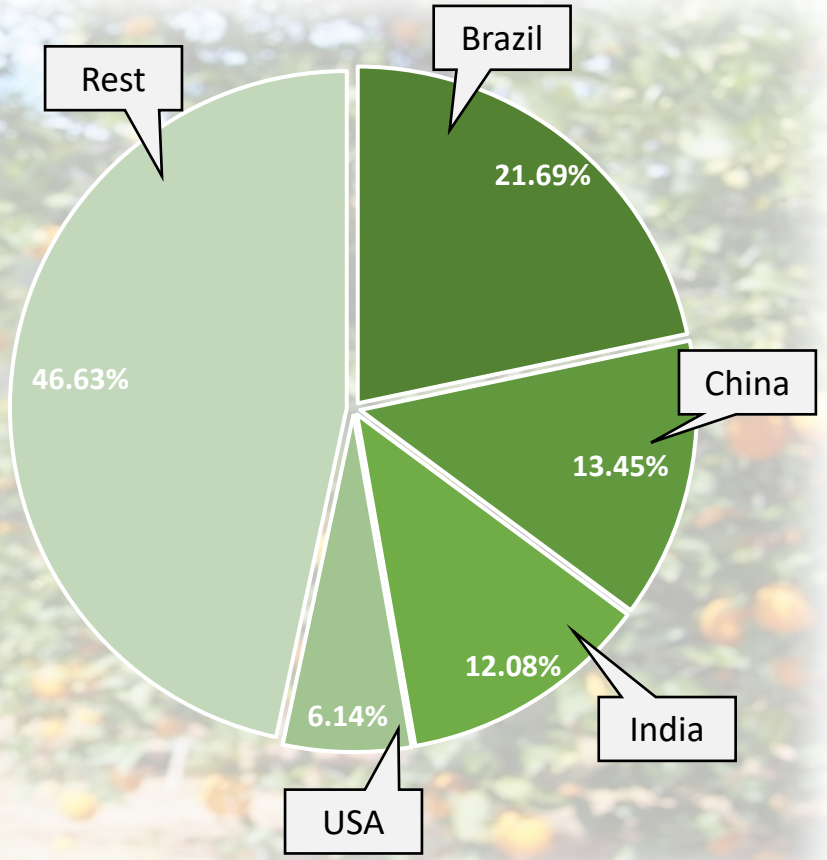
Contents

Oranges

Current Status

Global production of fresh orange:
78.7 million tons

45 – 60% is wasted.



Industrial Orange peel waste

Practices and Valorization

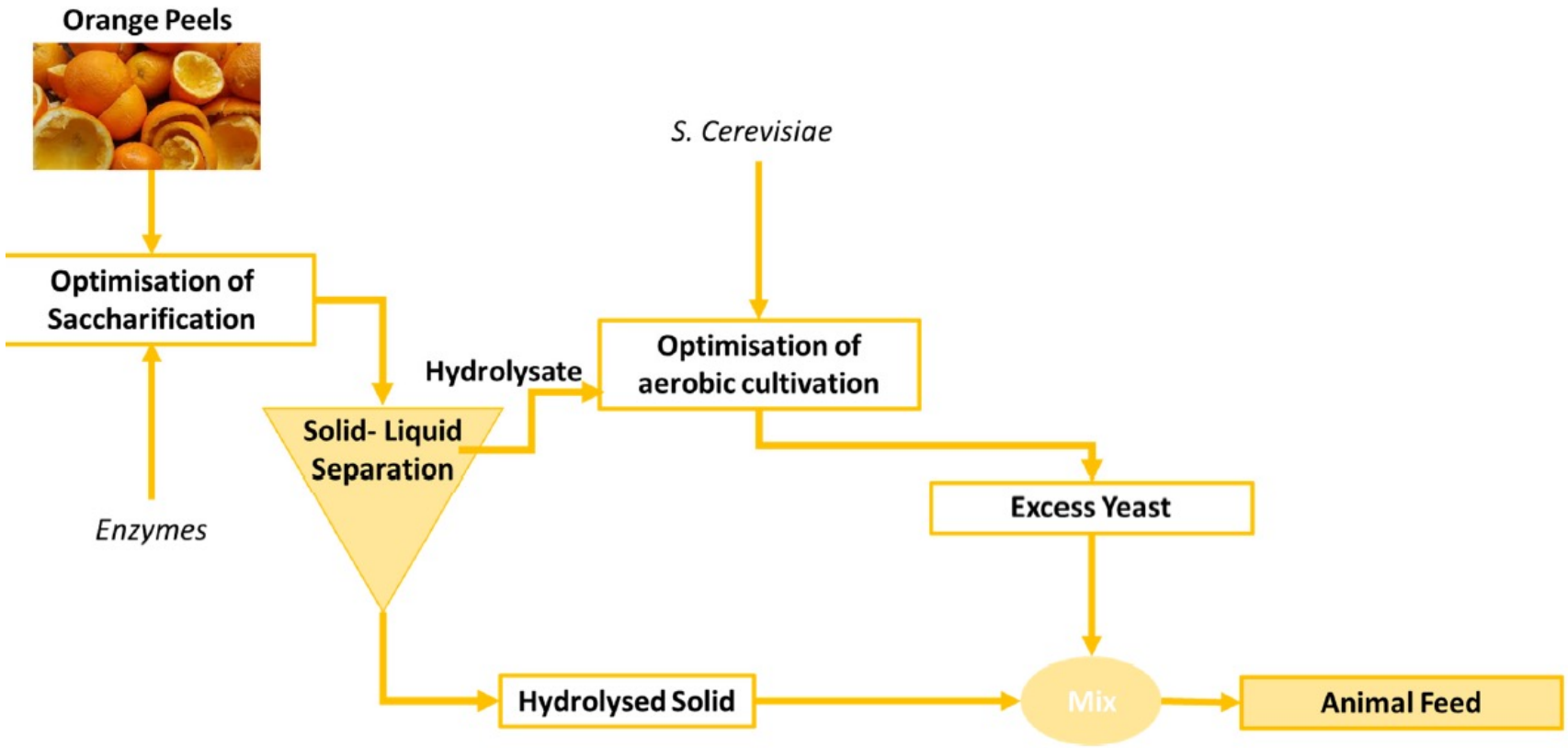
Orange Content

Pectin
Cellulose
Hemicellulose
Lignin
Essential Oils
Polyphenols

Possible products

Sugars
Flavors
Butanol
Bioethanol
Lactic Acid
Citric Acid
Antioxidants
Green Solvents
Enzyme Production
Pharma Products
Single Cell Protein

Animal Feeding



Industrial Orange peel waste

Analysis Method

Van Soest Analysis Method

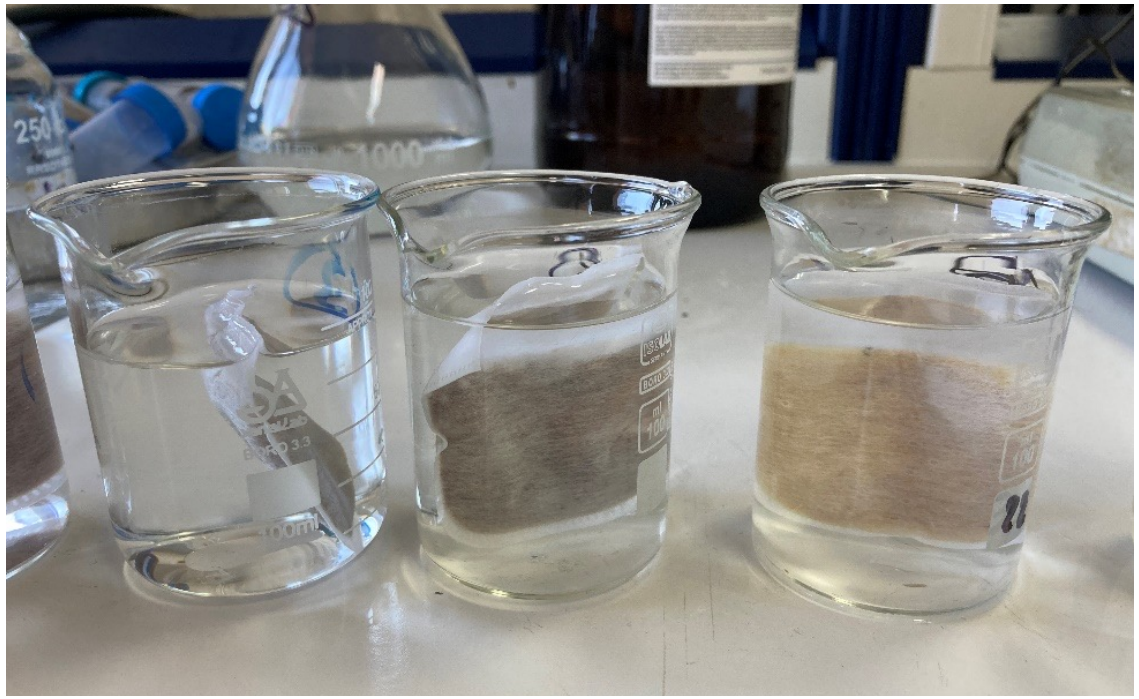
↳ Applicable for grains, animal feeds, forages, and all fiber-bearing materials.

↳ Calculated Index:

- Neutral Detergent fiber (NDF)
- Acid Detergent Fiber (ADF)
- Acid Detergent Lignin (ADL)

↳ ADF is considered a reliable Index for the digestibility of animal feed.

- Max 40-50% for Ruminants and
- Max 20-30% for Monogastric animals



Industrial Orange peel waste

Physicochemical Characterisation

	Feedstock
Total Solids , TS (%)	91.85 ± 1.65
Moisture (%)	8.15 ± 1.65
Volatile Solids, VS (% d.b.)	91.35 ± 1.78
Ash (% d.b.)	8.65 ± 1.78
Water-Soluble Solids, WSS (% d.b.)	35.99 ± 1.98
Oils (% d.b.)	0.25 ± 0.01
Cellulose (% d.b.)	17.47 ± 2.12
Hemicellulose (% d.b.)	30.70 ± 4.46
Acid Insoluble Residue, AIL (% d.b.)	10.70 ± 1.11
Acid Soluble Lignin, ASL (% d.b.)	1.06 ± 0.08
Total Kjeldahl Nitrogen, TKN (% d.b.)	1.38 ± 0.04
Protein (% d.b.)	8.63 ± 2.39
NDF (% d.b.)	34.10 ± 3.21
ADF (% d.b.)	24.80 ± 3.43
ADL (% d.b.)	6.20 ± 2.16

Experimental Protocol

Saccharification at 50°C, followed up with aerobic cultivation

1st Scale up

2nd Scale up

<u>Factorial experiment</u>

Process Variables:

- ➢Pectinase Dosage (Pectinex)
- ➢Cellulose Dosage (CellicCTec3)
- ➢Solid Loading

✓Saccharification yield

<u>4L Reactor</u>

Variable testing:

- ➢Nutrients addition
- ➢pH adjustment
- ➢Yeast Dosage

✓Single Cell Protein

<u>Pilot Trials</u>

Optimal conditions of the previous experiment

Measurements:

- ➢Sugars
- ➢Ethanol
- ➢TKN
- ➢ADF

Experimental Protocol

Factorial Bench Experiments Results

Optimisation of Saccharification phase at 50°C

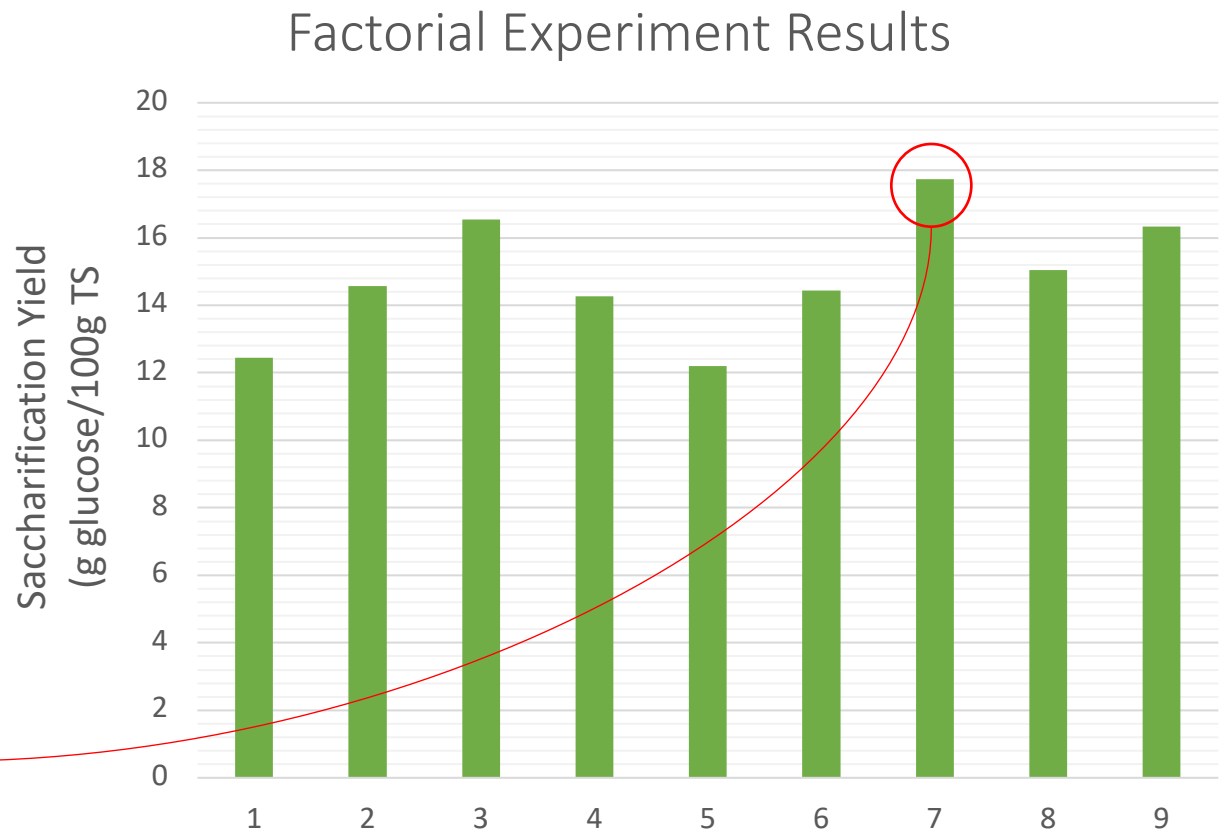
Parameter	Low Level (-)	High Level (+)	Center
Pectinex (µL/g solid)	25	75	50
CellicCTec3 (µL/g solid)	25	75	50
Solid Loading (%)	2.5	7.5	5

Optimum conditions

- Pectinex: 75 µL/g TS
- CellicCTec3: 75 µL/g TS
- Solid Loading: 2.5 %
- Temperature: 50°C

Results:

- Average Saccharification Yield: 14.84%
- Highest Saccharification Yield: 17.73%

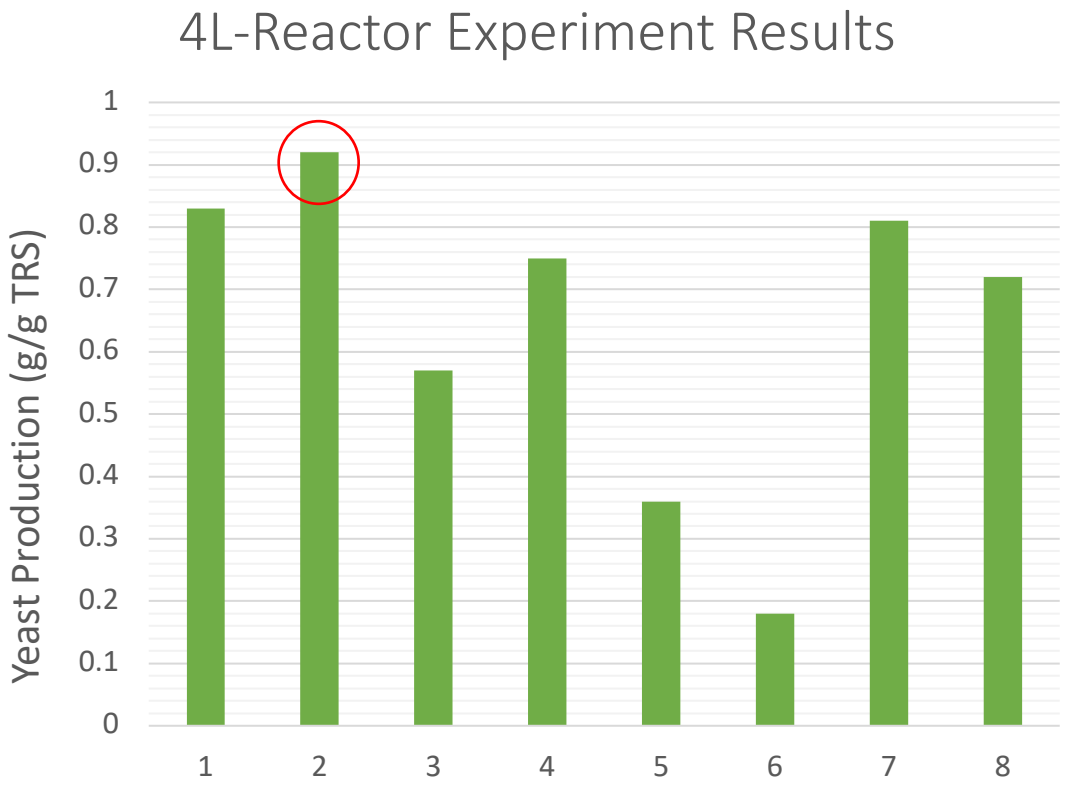


Experimental Protocol

4L-Reactor Experiments

1st Scale-up: Optimisation of Aerobic Cultivation phase

Parameter	Low Level (-)	High Level (+)
Nutrients Addition	No	Yes
Yeast Loading (w/w)	0.005	0.020
pH Adjustment	No	Yes



Experimental Protocol

4L-Reactor Experiments

1st Scale-up: Optimisation of Aerobic Cultivation phase

Results:

- ✓ Glucose level after saccharification: **13.42 g/L**
- ✓ Nitrogen level after cultivation: **1.50 % d.b. or 9.39 % protein content**

Optimum conditions

- ✓ Nutrients Addition: Yes
- ✓ pH Adjustment: No
- ✓ Yeast: 0.020 w/w
- ✓ Temperature: 25^{o}C

Experimental Protocol

Pilot Trials

2nd Scale-up: Pilot Trials

Trials conducted at optimum conditions:

- ✓ Pectinex: 25 μL/g TS
- ✓ CellicCTec3: 25 μL/g TS
- ✓ Solid Loading: 2.5 %
- ✓ Yeast: 0.020 w/w
- ✓ Temperature: 50^{o}C
- ✓ Residence Time: 24 h
- ✓ pH: 5 (No adjustment)

Results:

- ✓ The up-scaling does not seem to affect the process.

Experimental Protocol

Pilot Trials - Hydrolysis and Cultivation

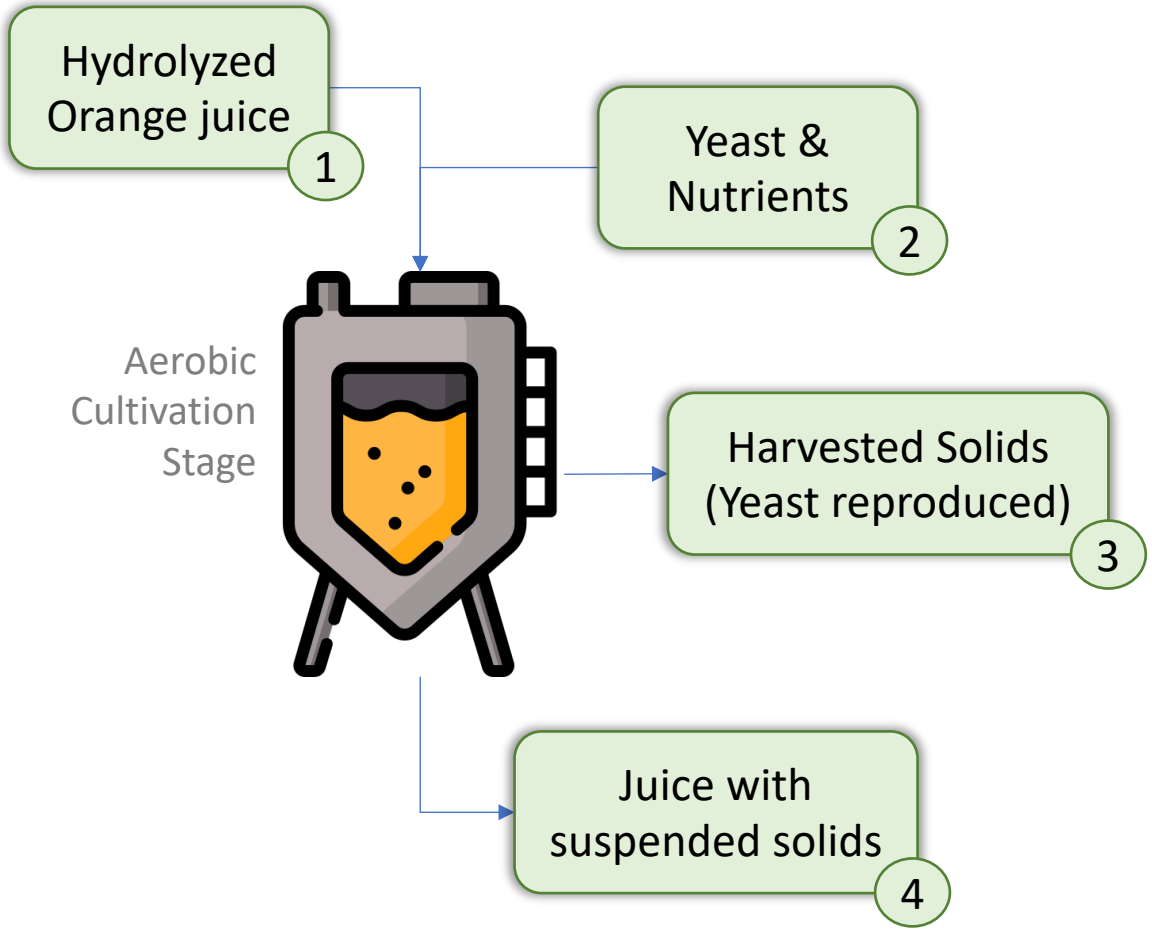
1st Step: Enzymatic Hydrolysis

Hydrolysed Orange Peels

2nd Step: Aerobic Cultivation

Analysis of Results

Pilot Trials



Input: 1 2

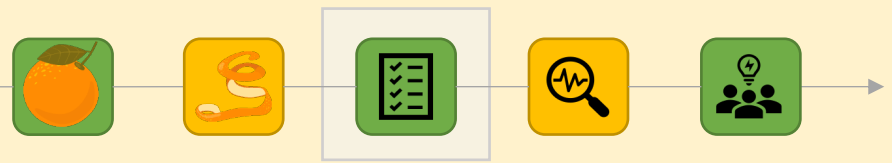
- **TN_1** in Juice
- **TKN_1** of suspended solids in juice
- **TKN_2** of Yeast
- **TN_2** of Nutrients (NH_4SO_4)

Output: 3 4

- **TKN_3** of Harvested Solids
- **TN_4** in Juice
- **TKN_4** of suspended solids in the juice

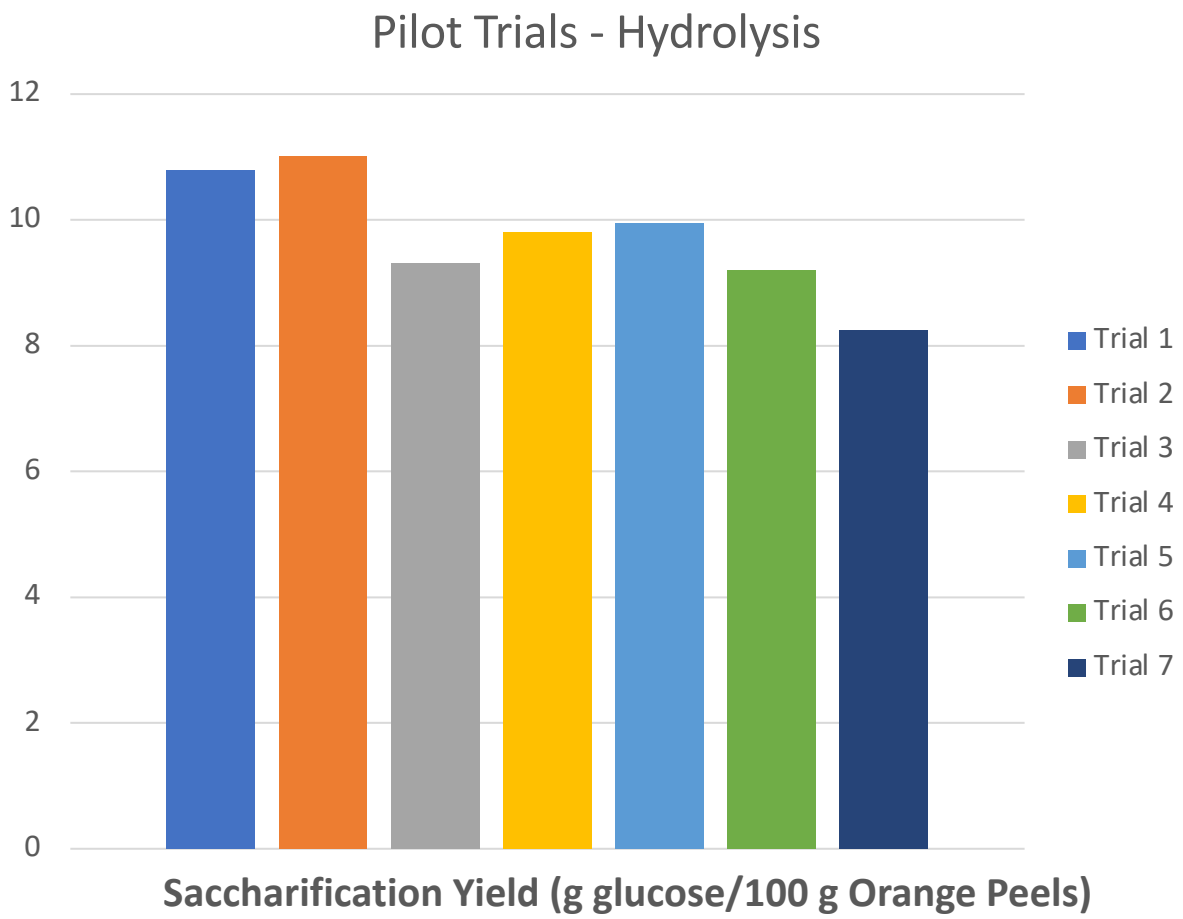
Notes:

- Mass Balance Closure.
- The TKN of Harvested Solids should be higher than the suspended solids in the hydrolyzed juice.



Analysis of Results

Pilot Trials



Saccharification yield of Hydrolysis:

$$Y_{Sacch.} = \frac{Glucose\ produced\ (g)}{Total\ Solids\ (g)} * 100$$

Conclusions:

1. The results were successful and repeatable.
2. The yield was between 8 to 11 g/ 100g TS.

Notes:

1. Sugars were measured using an HPLC method.
2. Saccharification yields were verified by polysaccharides' degradation.

Analysis of Results

Pilot Trials: Process Diagram

Feedstock
Orange peels: 28 kg
Moisture: 85 %
TKN: 1.1 %d.b.
NDF: 29.6 %d.b.
ADF: 15.6 %d.b.
ADL: 4.4 %d.b.
1

Enzymatic hydrolysis
Solid Loading: 7.5 %w/w
pH: 3.1
Glucose: 1.8 g/L
TRS: 5.7 g/L
TN: 0.5 g/L

Separated Solids
Orange Peels: 0.9 kg
TKN: 1.7 %d.b.
NDF: 38.5 %d.b.
ADF: 31.5 %d.b.
ADL: 6.4 %d.b.
2

Yeast & Nutrients
Yeast: 0.1 g/L
$MgSO_4$: 0.4 g/L
KH_2PO_4: 5 g/L
NH_4SO4: 2 g/L
4

Aerobic Cultivation
Solid Loading: 1.2 %w/w
pH: 3.7
Glucose: 4.7 g/L
TRS: 5.8 g/L
TN: 0.3 g/L

Harvested Solids
Harvested Solids: 0.9 kg
TKN: 3.7 %d.b.
NDF: 28.9 %d.b.
ADF: 21.4 %d.b.
ADL: 2.8 %d.b.
5

Hydrolyzed Juice
pH: 3.2
Glucose: 7.9 g/L
TRS: 12.2 g/L
TN: 0.5 g/L
Solid Loading: 2.6% w/w
3

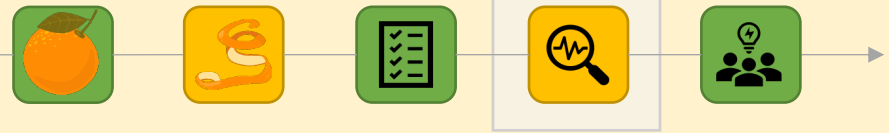
Fermented Orange Juice
pH: 3.8
Glucose: 1.8 g/L
TRS: 3.4 g/L
TN: 0.3 g/L
Solid Loading: 1.7 %w/w
6

Analysis of Results

Pilot Trials: Nitrogen Mass Balance

Nitrogen Mass Balance					
Time (h)	**Nitrogen in Orange Juice (g)**	**Nitrogen Content in Nutrients (g)**	**Nitrogen Content in Yeast (g)**	**Nitrogen in Solids (g)**	**Total (g)**
0	15.4 ± 0.77	21.2 ± 0.11	0.4 ± 0.18	9.6 ± 0.82	46.6 ± 0.34
48	16.0 ± 0.92	0.0 ± 0.0	30.8 ± 1.31		46.8 ± 0.45

- Verified by Mass Balance Closure.
- Rich in protein end products.
- ADF increases and remains below the indigestible range.



Conclusions

- Valorisation of Industrial Organic Waste Stream
- Elevated Protein Content
- Advanced and Digestible Animal Feed
- Validation of the end product for animal feed

Thank you for your attention!

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