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CUSTOMIZED ENZYME SOLUTIONS

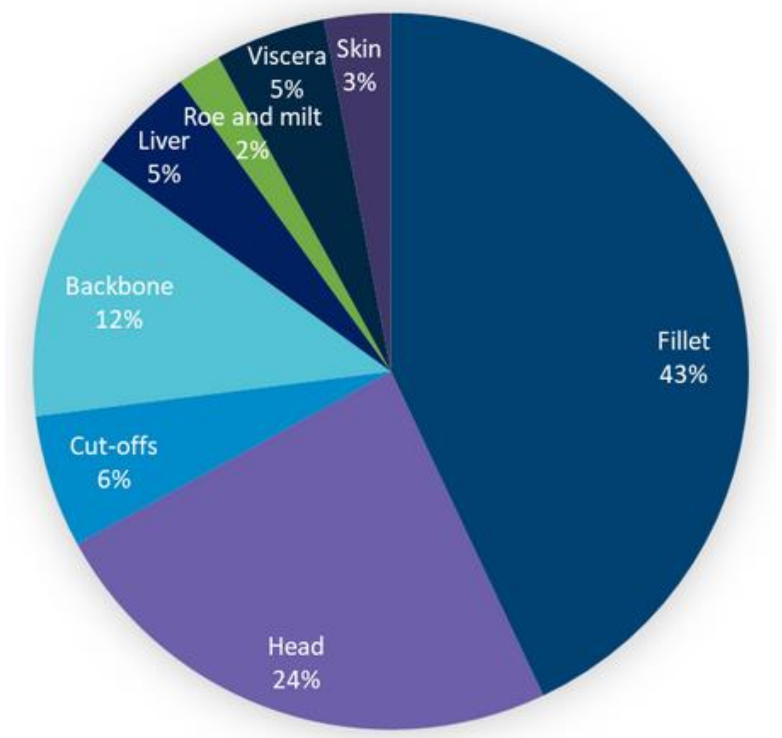
Products & Side Streams from Fish Processing

Collagen peptides produced using enzymatic hydrolysis

Agenda

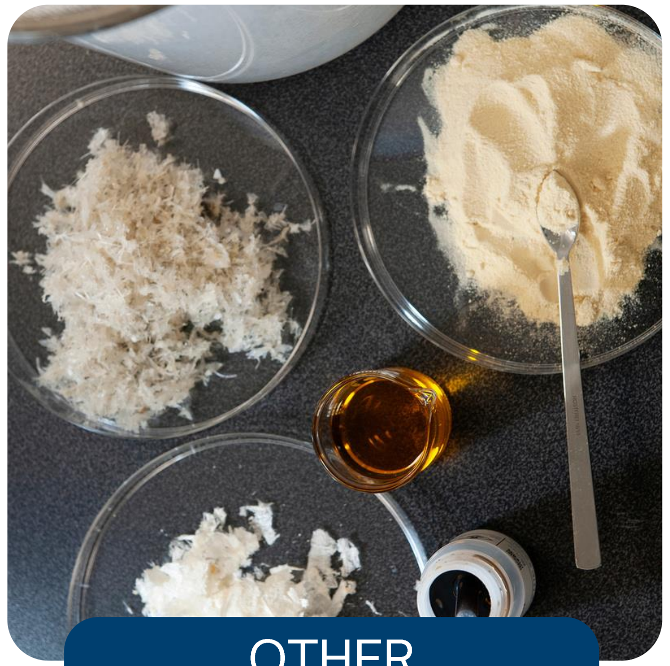
- Fish processing **side streams** and possible **valuable products**
- Collagen, gelatin and **collagen peptides**
- **Process overview** to produce collagen peptides
- Some **laboratory examples**
- From a laboratory example to **industrial scale**

Fish processing generates valuable side streams that can be revalorized into high-valuable products



PRODUCTS & SIDE STREAMS

Products and side streams from shore processed cod (adapted from *Arason et al., 2009*)



OTHER

Potential high-value products:

Protein hydrolysate from cut-offs, viscera, liver and head.

Fish oil from viscera, liver and head.

SKIN

- 80% protein
- 3% fat
- 5-15% ashes

Potential high-value product:
Collagen peptides

BACKBONE

- 22% protein
- 3.5% fat
- 75% ashes

Potential high-value product:
Collagen peptides

Collagen, gelatin, and collagen peptides

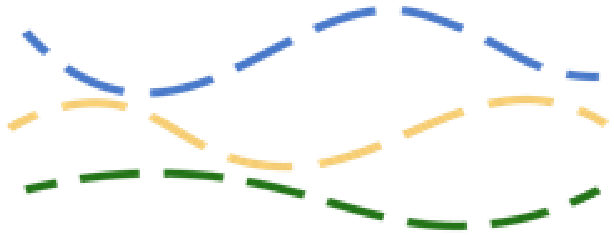
Collagen

- Most abundant protein in the human body (30% of total protein)
- Main building block for connective tissues

Gelatin

- Multifunctional protein

Collagen peptides



- Reduced viscosity, enhanced solubility → Eased production and formulation processes
- Improved digestibility and bioavailability → Better product

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Process Overview

Cleaning the raw material & pre-treatments

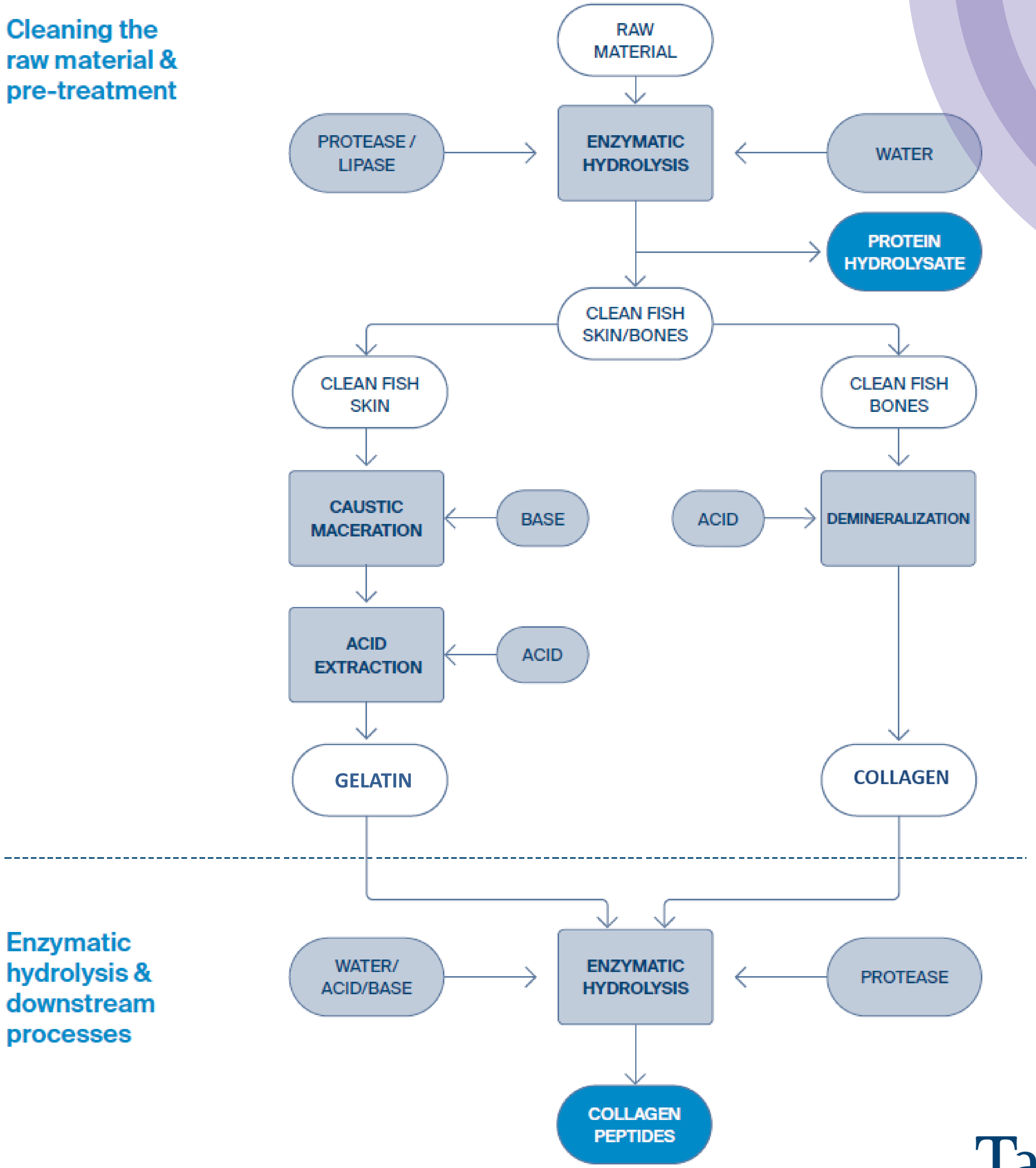
Used to remove residual meat, oils, or other non-collagenous components. Also used to open up the collagen structure.

Enzymatic hydrolysis

Enzyme choice depends on the requirements of the final product. Several parameters need to be controlled for an optimal process: pH, temperature, stirring, etc.

Downstream and final processes

To obtain the final product, separation processes are normally required, for the product to be concentrated and dried.



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Collagen peptides production yields using enzymatic hydrolysis

Skin		Backbone
19%	**Total Yield** Collagen peptides obtained (dry) / Raw material (dry)	16%
87%	**Enzymatic Hydrolysis Step** Collagen peptides obtained (dry) / Pre-treated material (dry)	98%
22%	**Pre-treatment Step** Pre-treated material (dry) / Raw material (dry)	17%

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Some laboratory examples

Insights for industrial processes

Demineralization of Tilapia fish bones as a pre-treatment for collagen hydrolysis

BACKGROUND

- Collagen is inside a hydroxyapatite structure, rich in calcium, and it is insoluble.

$$(Ca)_{10}(PO_4)_6(OH)_2$$

- When adding an acid (e.g. HCl) the ions are converted into other soluble salts.

$$(Ca)_{10}(PO_4)_6(OH)_2 + 20\ \mathbf{HCl}$$

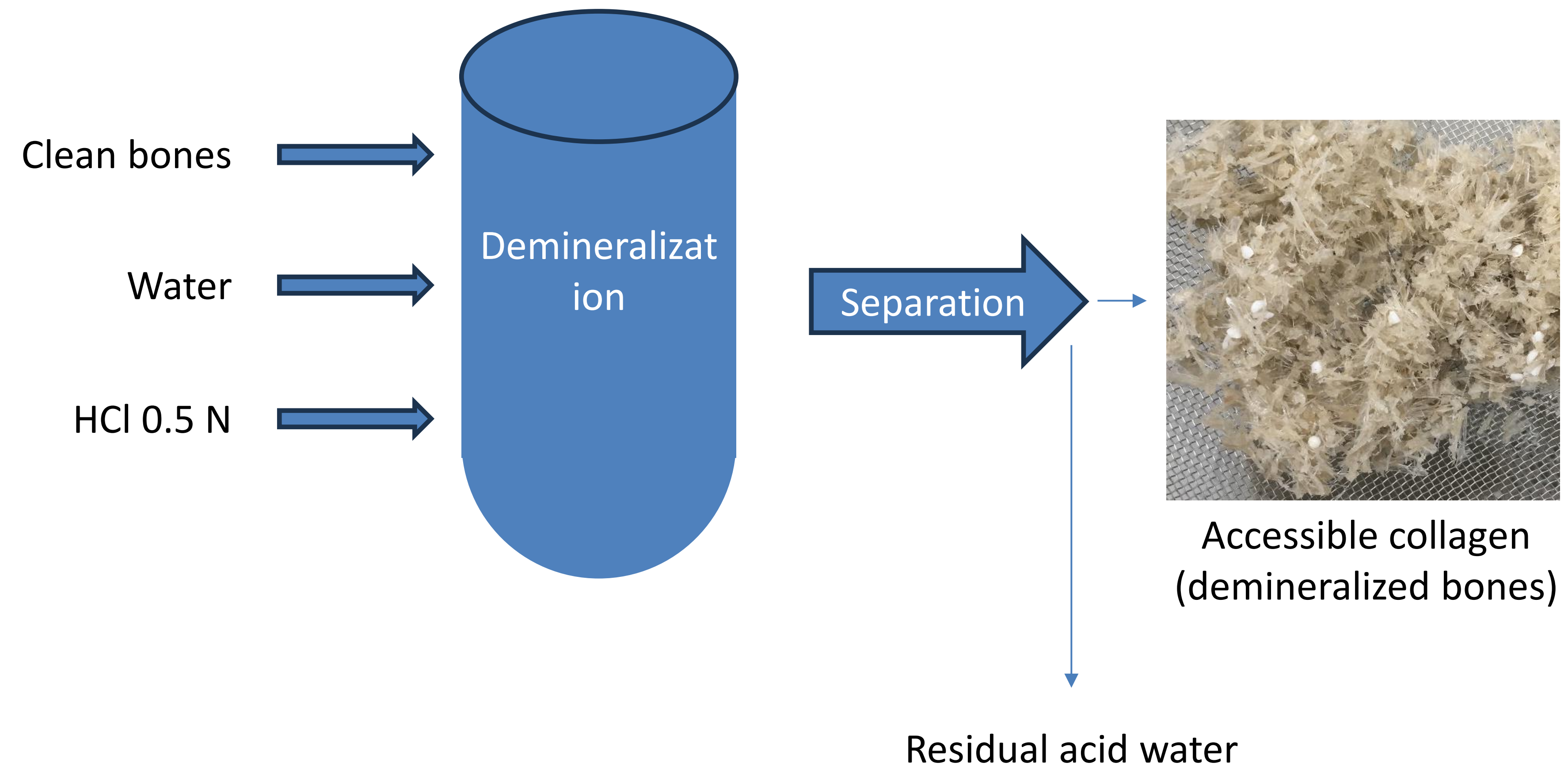
$$\Downarrow$$

$$10\ CaCl_2 + 6\ H_3PO_4 + 2\ H_2O$$

- **Problem**: <u>too much acid</u> can also hydrolyze the protein that we want to remain for later in the process

TESTS

- **How much time is needed to demineralize the calcium matrix?**
- Variable parameter:
  - Demineralization time: 5 min, 10 min, 30 min, 1h, 6h, 12h and, 24h.
- Fix parameters:
  - Temperature: RT
  - Acid used: HCl
  - Concentration: 0.5 N
  - Solvent to mass ratio: 25:1
  - Stirring speed: 300 rpm
- Measurements:
  - Residual calcium determination
  - Protein quantification



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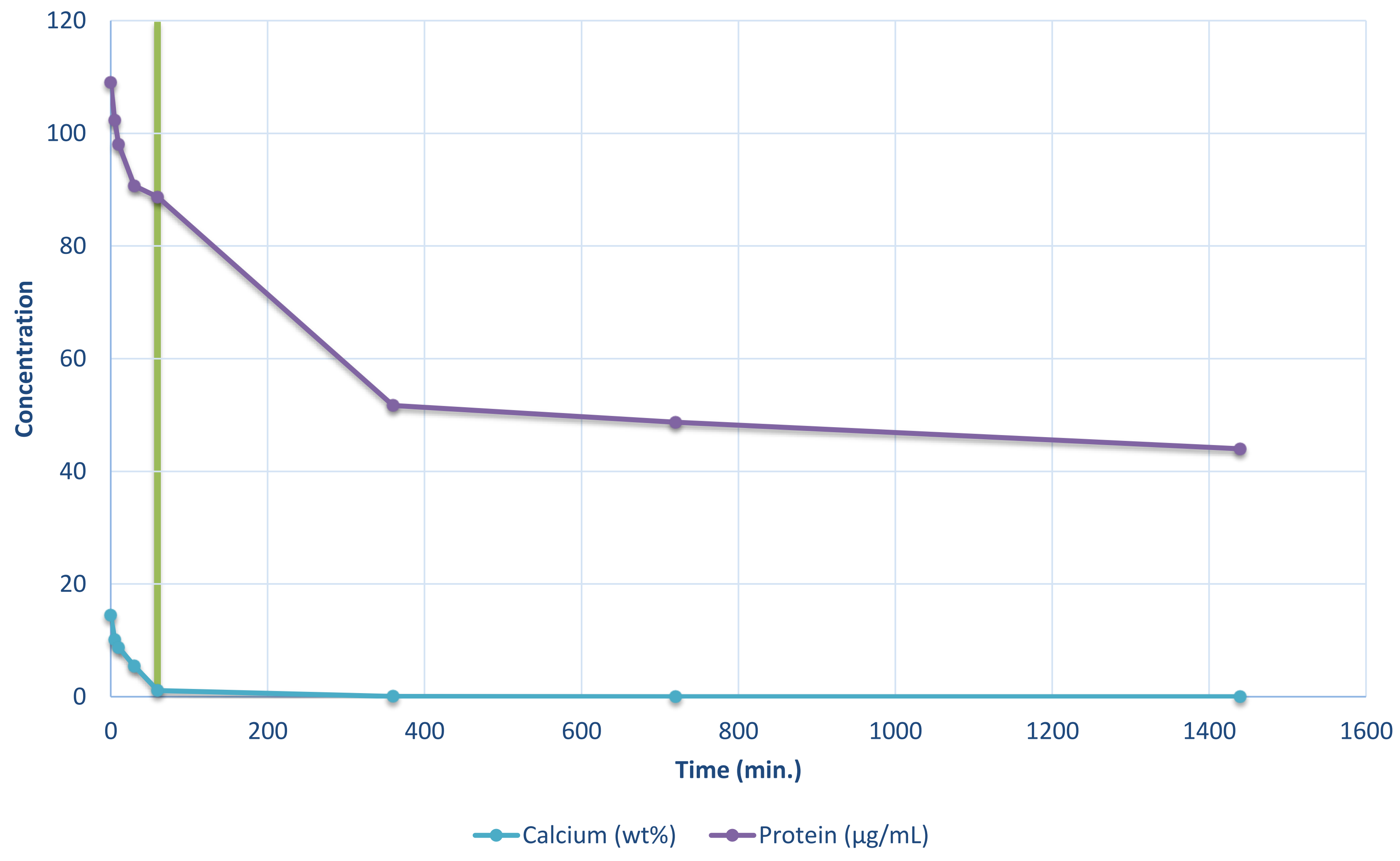
RESIDUAL CALCIUM DETERMINATION & PROTEIN QUANTIFICATION IN THE BONES



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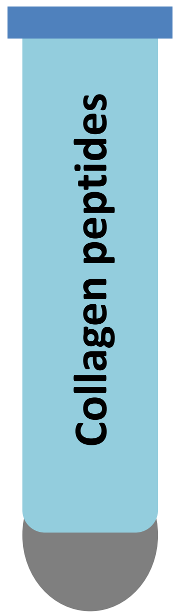
Enzymatic hydrolysis of demineralized fish bones to obtain collagen peptides

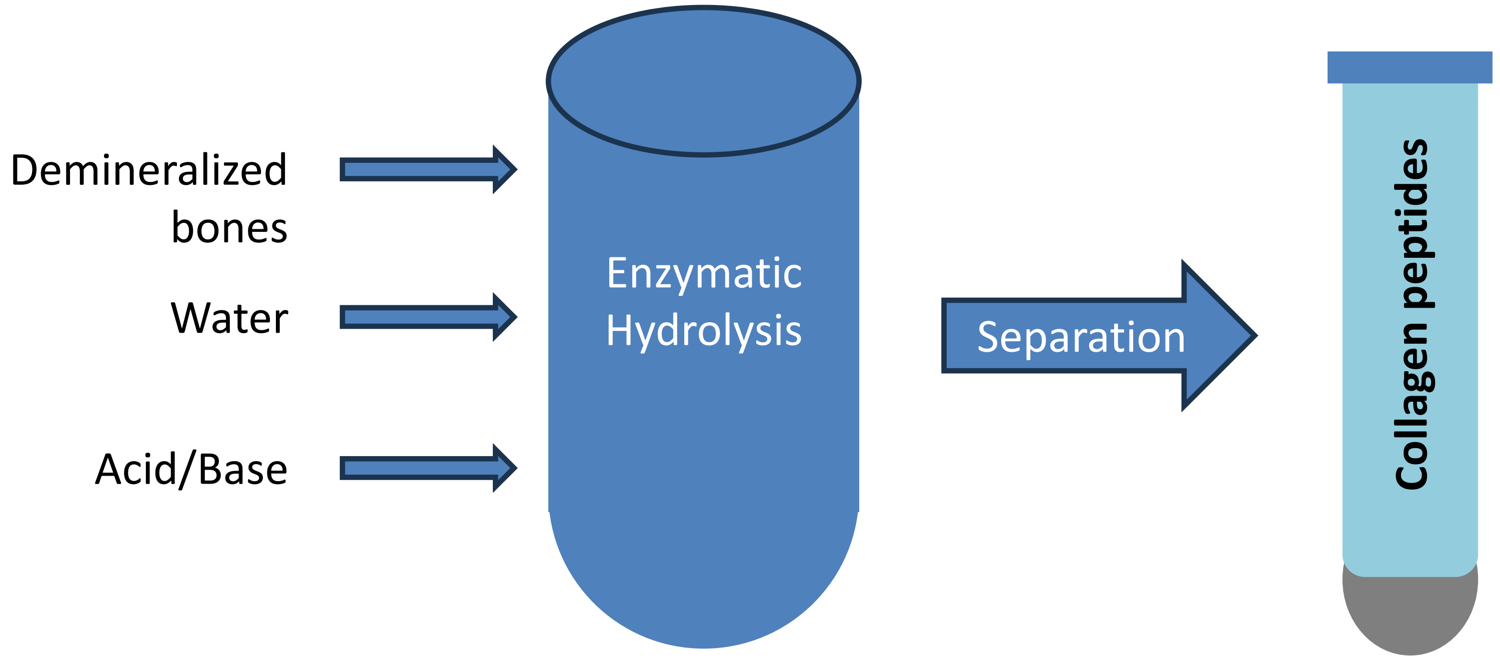
BACKGROUND

- Collagen is now exposed to be hydrolyzed.
- Enzymes will hydrolyze the collagen to collagen peptides, so it will be recovered in the water-soluble fraction.



TESTS

- **How much enzyme is needed?**
- **How much hydrolysis time is needed?**
- Variable parameters
  - Enzyme dosage
  - Hydrolysis time
- Fixed parameters
  - Water-to-mass ratio
  - Temperature
  - pH
  - Enzyme used
  - Stirring speed
- Measurements:
  - Yield (%) – protein recovered in the soluble fraction compared to the protein in the demineralized bones.



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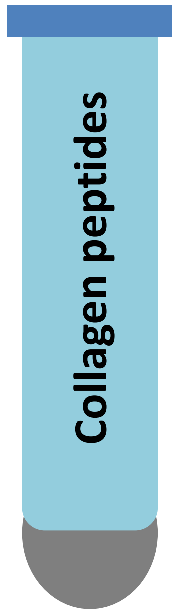

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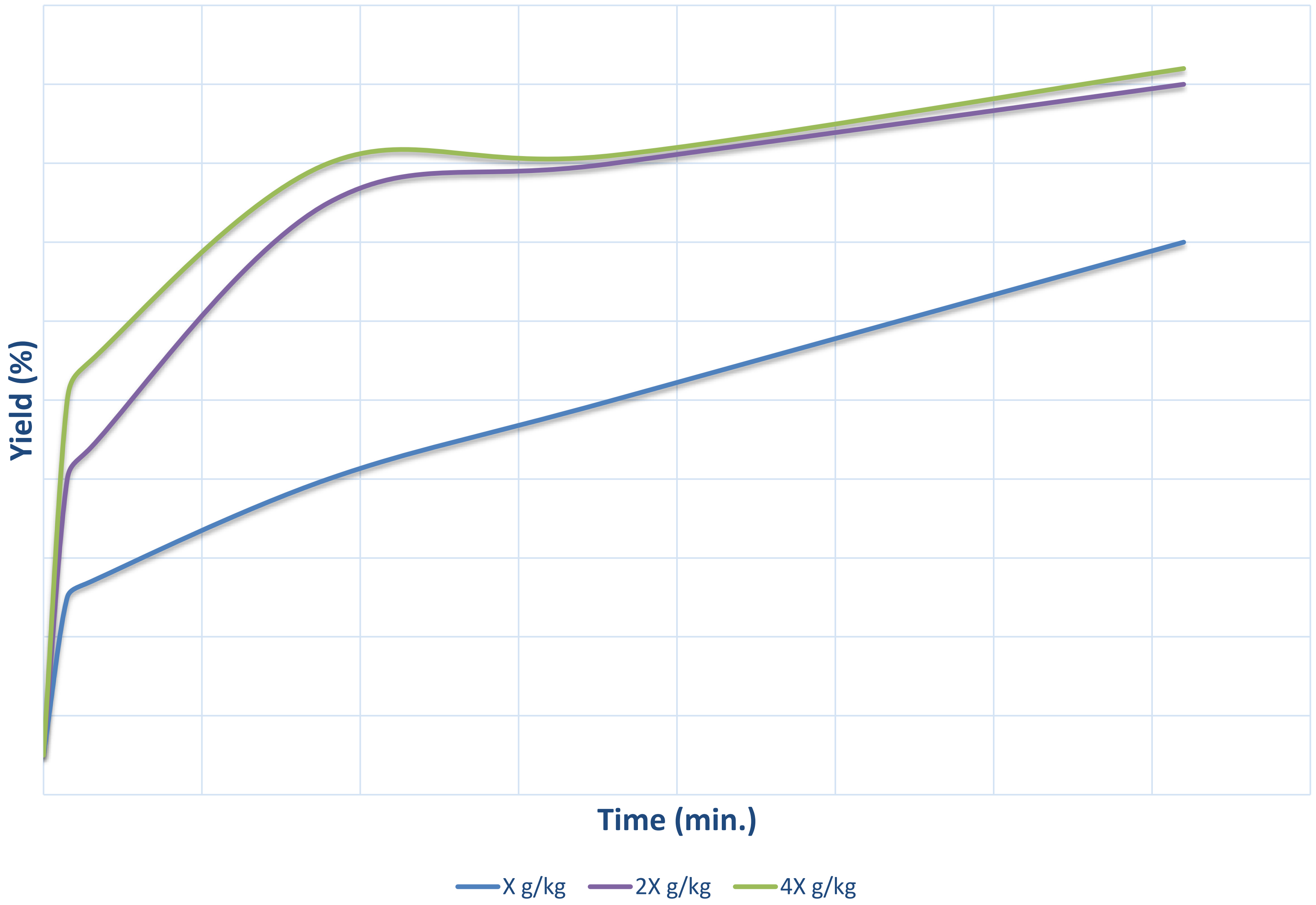
RESULTS



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From laboratory tests to industrial scale

Pre-treatment

Possible parameters to test:

- Cleaning the raw material
- Making the protein accessible (different types of pre-treatments)
- Etc.

Enzymatic hydrolysis

Possible parameters to test:

- Enzyme type
- Enzyme dosage
- Temperature
- pH
- Hydrolysis time

Downstream processes

Possible parameters to test:

- Separation treatments
- Purifying treatments
- Etc.

All parameters adjusted to the raw material, the process possibilities and the desired final product

Are you ready?

Tailorzyme® develops enzyme solutions tailored to each customer's specific substrate and process requirements. Working closely with customers, supporting both food and non-food applications.

Raw material characteristics

What is your raw material?
What is the composition of the raw material?
What is the pH of the raw material?

About the process

What do you currently do with your raw material?
What is your current process?
What volume of raw material do you have?
When/where are you going to produce?

About the product

What is your final product and target customer?
Is flavour important for your final product?
Is colour important for your final product?
Is there a molecular size requirement?
Should the final product be vegan/organic/etc.?

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