WP2 : Applying the ensilage process to macroalgae

Hélène Marfaing (CEVA)
Pierrick Stévant (Møreforsking Ålesund)

PROMAC final meeting November 8th- Ålesund

Introduction

- Context : development of seaweed cultivation in Europe
  - Huge quantity of seaweed in a short period : ≈ 2 months harvesting
  - Challenge of **preservation** of seaweed : degradation can occur rapidly
    - **Drying**
      - Sun-drying : strongly dependent on weather
      - Oven-drying : cost-intensive energy processes
        - » Not economically viable for all applications
    - Freezing?
    - Alternative method with low energy input : preservation by **ensiling**

Ensiling

PROMAC

- Principle : bacterial conversion of water-soluble carbohydrates (WSC) to organic acids under anaerobic conditions
  - Accumulation of acids (mainly **lactic acid**)
  - Reduction of pH
    - pH<4,3 is recommended
  - Inhibition of spoilage microorganisms growth
- Ensiling widely used in terrestrial agriculture
  - Spontaneous anaerobic lactic acid fermentation
- Seaweed ensiling is **challenging**
  - High pH
  - High mineral content : buffering capacity :
  - Low water soluble carbohydrate content (but sugar acohols)
  - Natural presence of lactic acid bacteria ?

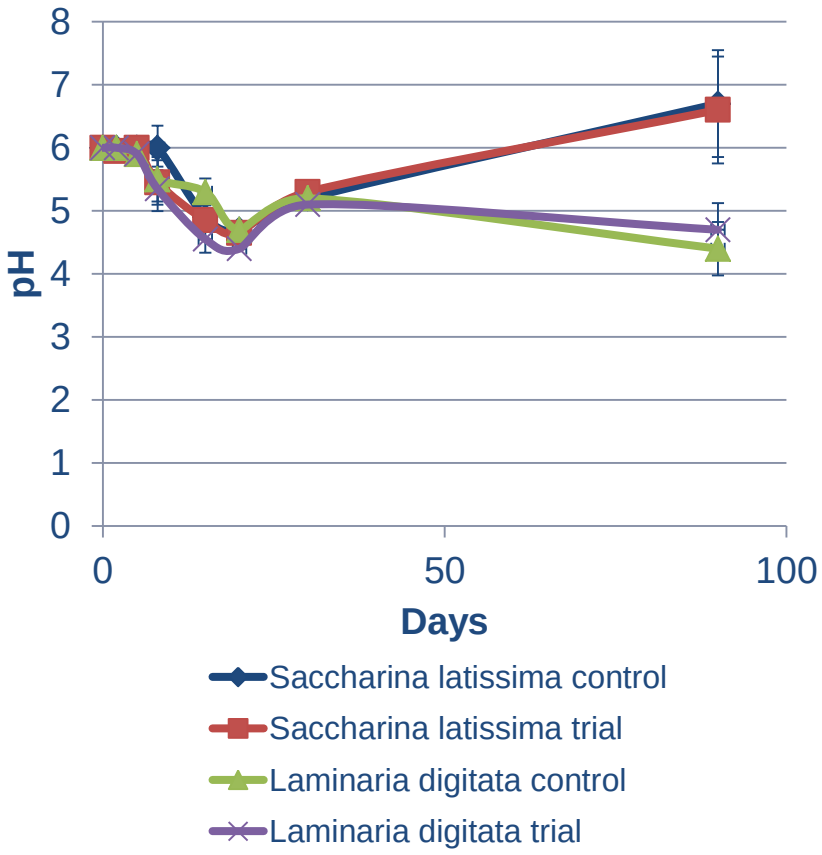
Previous results

- **Preservation of *Laminaria digitata*** through ensilage process is possible at room temperature
  - **Adequate acidification** after 90 days ensilage in anaerobic conditions
  - Production of lactic acid up to 2,4%
  - Consumption of free sugars and glucose chains (and cellulose)
  - **Valuable juice** obtained from ensilage : 3% mannitol and 8% uronic acids

- In non **optimal anaerobic conditions** : *Saccharina latissima* rotting, faecal odours and increase of pH

- No significant effect of pretreatment : washing in seawater/freshwater
  - No washing in further experiments :to keep soluble sugars (mannitol) and salts

Evolution of pH during ensiling of *Laminaria digitata* and *Saccharina latissima*



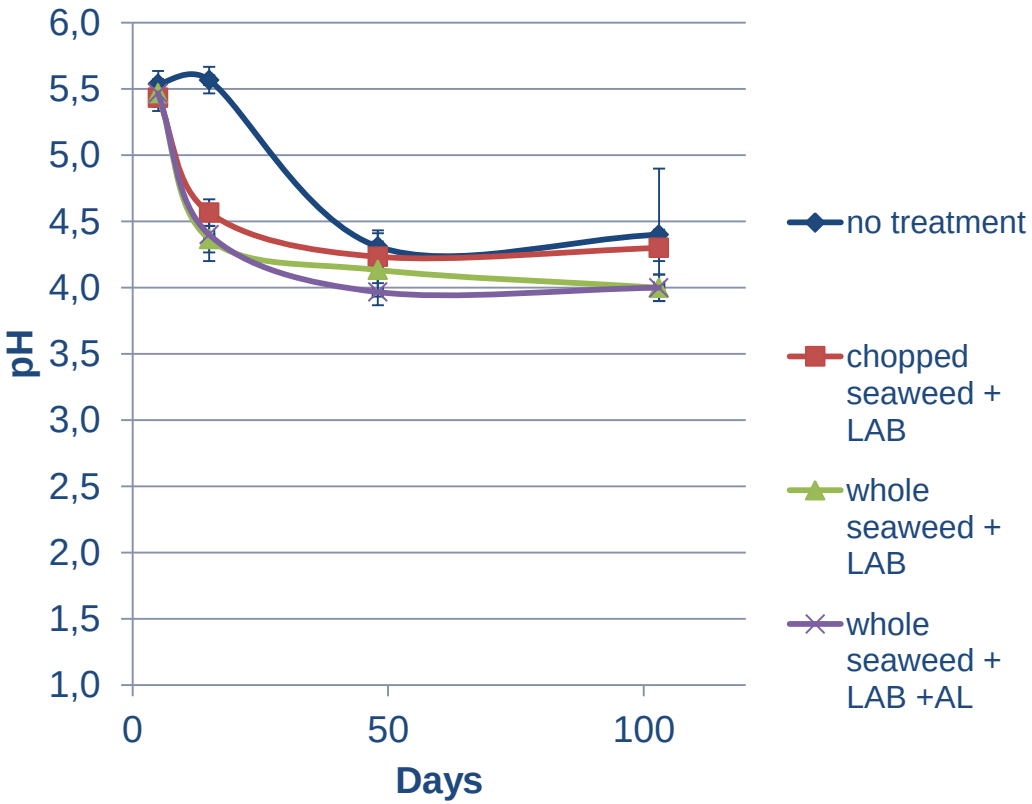
Promac experiment

- Raw materials : *Saccharina latissima* (culture, North Brittany, France)
- Stored in plastic bags under anaerobic conditions for 3 months at room temperature (≈ 18°C)
- Different parameters tested :
  - Reduction of size: chopping seaweed to 2 cm
  - Addition of lactic acid bacteria *Lactobacillus plantarum* (LAB)
  - Addition of polysaccharide degrading enzyme : Alginate lyase (AL)
- Analysis
  - Visual aspect, odour, juice
  - pH
  - Organic acids (lactic, acetic and butyric acids)
  - Polysaccharide content

Ensiling : Promac results

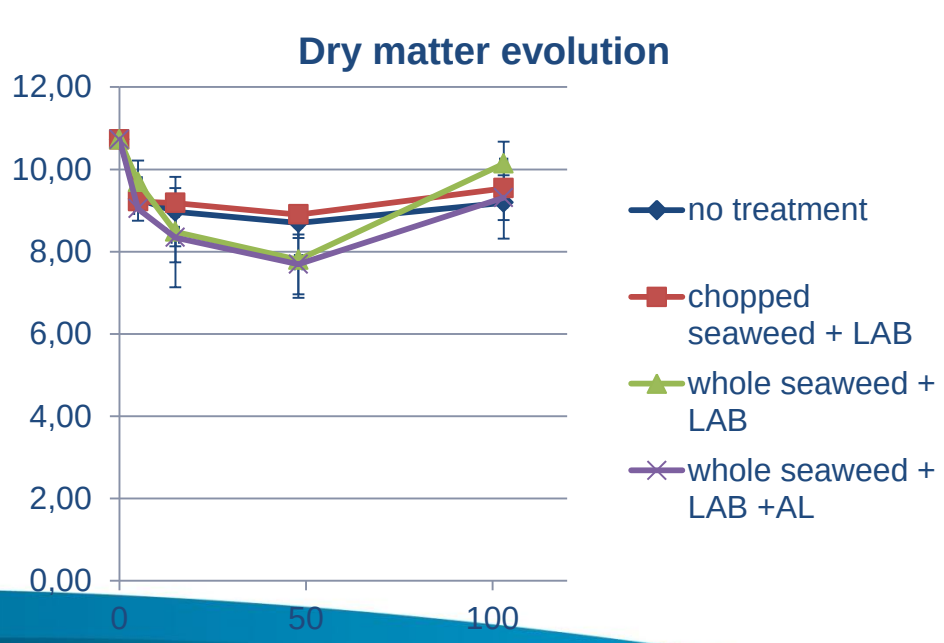
- **Significant acidification is observed** after **20 days ensilage** in anaerobic conditions **with addition of lactic acid bacteria** (*Lb plantarum*)
- **Without addition of lactic acid bacteria** (control) pH is significantly reduced after 48 days of silage (and similar to other)
- Pretreatment (chopping) or addition of alginate lyase are not necessary

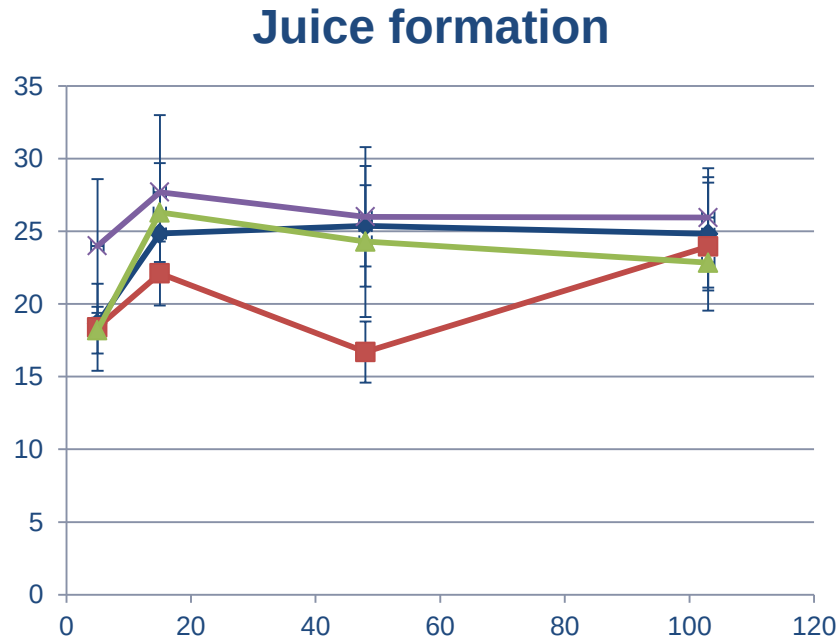
Evolution of pH during ensiling of *Saccharina latissima* under different conditions



Ensiling : Promac results

- **No significant decrease of seaweed dry matter**
- **Tendancy of dry matter reduction** after 48 days ensilage for chopped *Saccharina* with lactic acid bacteria then “reabsorption” of juice ?
- **Young blades** of *Saccharina latissima* : low production of exsudate

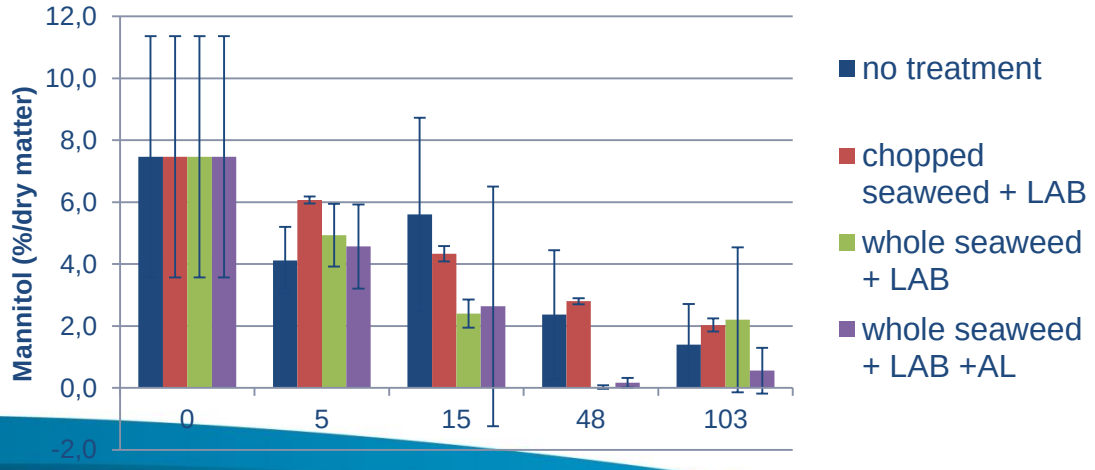




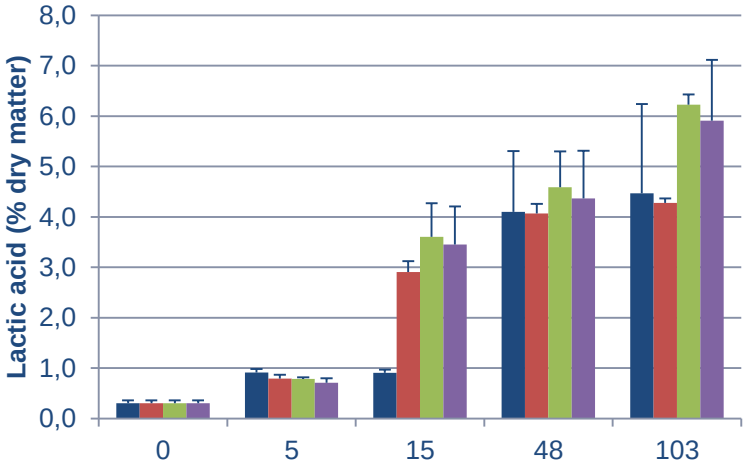
Ensiling : Promac results

- **Consumption of available sugars** during silage : mainly **mannitol** (low level of laminarin)
- **Production of organic acids**:
  - lactic acid in all trials. Highest level in seaweed with LAB : up to 6%/dry matter
  - acetic acid only in the control : up to 1,4%/dry matter
- No modification of alginate content

Mannitol consumption



Acid lactic production



Ongoing research

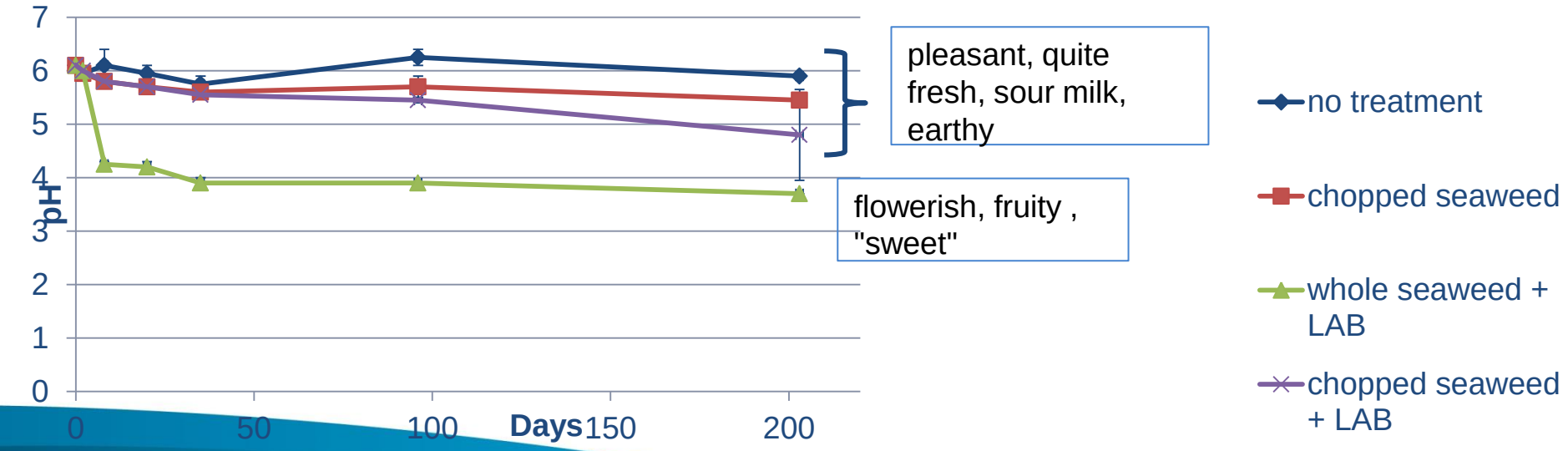
✓ Ensiling experiments objectives (Idealg project)

- To confirm the industrial feasability of ensilage
  - applying ensilage process over a long period at room temperature
- To characterize the impact of ensiling on alginate integrity
  - alternative process to industrial storage of fresh biomass in chemical solution?
- To identify the different bacteria consortia (DNA analysis) on seaweeds during ensiling storage
  - Evaluation of interactions or competitions between natural flora and inoculant

Ongoing research

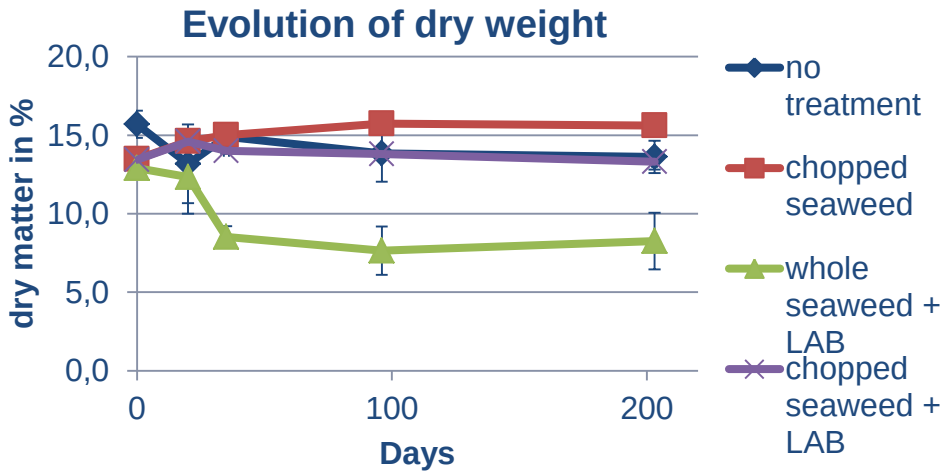
- ✓ **Adequate acidification** obtained after 10 days ensilage in anaerobic conditions for whole seaweed and addition of lactic acid bacteria
  - ✓ rapid growth and predominant presence of LAB
- ✓ **Acidification is maintained over 6 month**
- ✓ Even with no acidification, odour and visual aspect are good for fermented *L. digitata*
- ✓ Pretreatment (chopping) is not necessary

Evolution of pH during 203 days *Laminaria digitata* ensilage

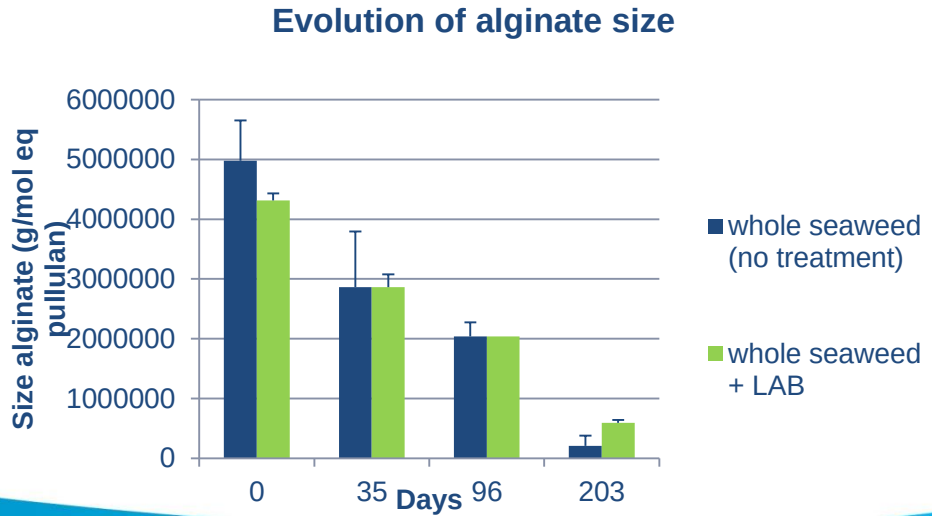


Ongoing research

- ✓ **Trial with inoculant :** lactic acid bacteria *(Lb. plantarum*)
  - ✓ Significant **decrease of dry matter**
  - ✓ **Consumption** of **mannnitol** and production of **lactic acid**
  - ✓ **Reduction of alginate content**



- ✓ **Impact of ensiling** on alginate integrity
  - ✓ **Reduction of size alginate with or without inoculant**
  - ✓ Strong impact for alginate producers



Ongoing research

- Preservation of *Laminaria digitata* through ensilage process is possible at room temperature for at least 6 months
- Addition of lactic acid bacteria is necessary to facilitate the process and obtain good acidification and lactic acid production
  - Lactic acid bacteria >> marine bacteria
- Preserving *Laminaria digitata* through ensilage could lead to changes in the biomass texture, in alginate extraction yield and in the alginate molecular weight
  - reduction of alginate size
  - Strong impact for alginate stakeholders

Applying the ensilage process to macroalgae

PROMAC

Take-home messages

✓ Preservation of *Saccharina latissima* and *Laminaria digitata* through ensilage process is possible at room temperature.

✓ Differences after ensiling between different species : biochemical composition, seasonal variation, composition of the natural bacteria present

- ✓ addition of lactic acid bacteria not always necessary to obtain good acidification (pH <4,3) and lactic acid production.

✓ Impact of ensilage process on alginate content and molecular weight in *Laminaria digitata*

✓ Ensiling process : useful to preserve seaweeds but also to develop new products for food, feed and agriculture applications

PROMAC

Thank you for your attention
Thanks to CEVA's team

Seaweed cultivation
Tristan le Goff

Pilot trials
Yves Lelong

Laboratory analysis
Nathalie Viller
Nelly le Pen