# Effect of gas composition on quality attributes and microbial population dynamics of fresh-cut iceberg lettuce

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#### Introduction:

- In North-Europa, shelf life of fresh-cut iceberg lettuce is limited to P+8
  - E-MAP: BOPP film → anaerobe condition reached within 3 days
  - Storage temperature: 7°C
  - No chemicals used during sanitation process

 Overall visual quality (OVQ) mainly determines the shelf life of fresh-cut iceberg lettuce

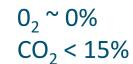




# Challenge: the right gas permeability

 $0_2 = 0\%$  $CO_2 > 15\%$ 







 $0_2 > 0.5\%$  $CO_2 < 15\%$ 



Too low

Increasing gas permeability

Too high

Fermentation / Decay

**Red discoloration** 

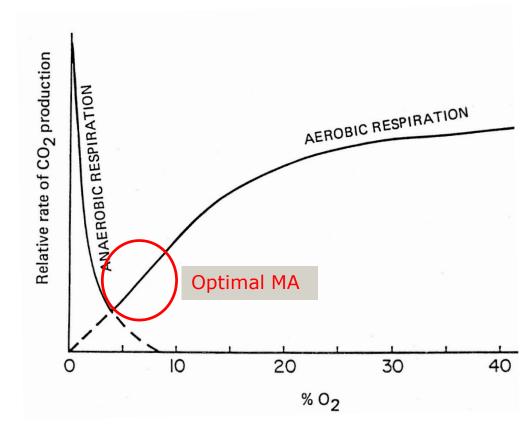




# Equilibrium-Modified Atmosphere Packaging (E-MAP)

- Suitable for product with respiration rate (fruit, vegetables, ornamentals)
- Use respiration rate of fresh product to create MAP

- Slow down activity of product
  - → extend shelf life
- BUT avoid fermentation





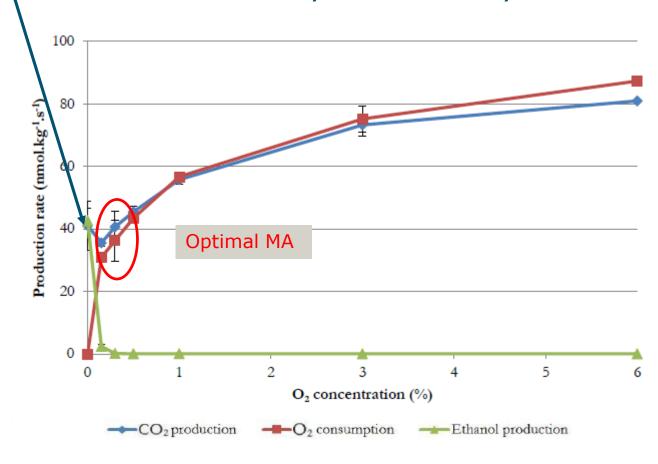


#### Anaerobic E-MAP

Anaerobic gas conditions are reached to avoid pink discoloration (oxidation process)

■ Production of ethanol and acetaldehyde → toxicity?







# Sensorial and microbial quality in MAP

- Shelf life of iceberg lettuce packed under E-MAP is limited by sourness off-odour and tissue collapsing.
- Acetic and lactic acid produced over the time by lactic acid bacteria population (Lactococcus and Leuconostoc spp.)



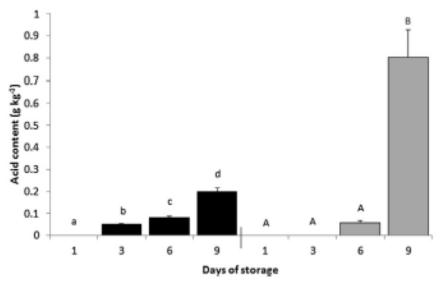




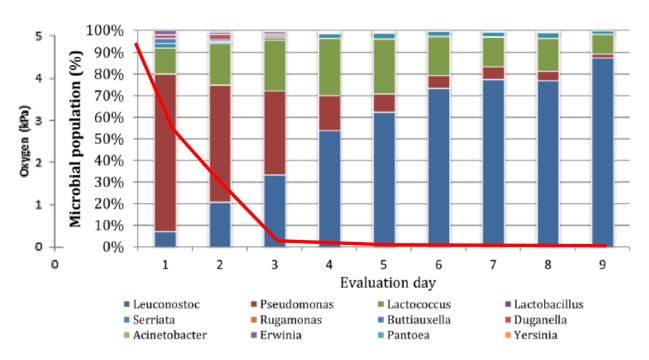




Fig. 4. Acetic acid (black bars) and lactic acid (grey bars) contents of fresh-cut iceberg lettuce stored at  $7\,^{\circ}$ C. Bars are the mean of three replicates  $\pm$  SD. Different small and caps letters indicate statistical differences between the evaluation days for acetic acid and lactic acid contents, respectively (P < 0.05).

# Sensorial and microbial quality in MAP

• Microbial population dynamic follows the dynamic of the gas composition inside the packaging.



Microbial population dynamics of natural flora growing on fresh-cut iceberg lettuce packed under MA conditions and stored at  $7^{\circ}$ C. Microbial species are expressed as percentages of the total microbial load per evaluation day. Values are means (n=3).



### Hypothesis:

- Can the microbial population dynamics be controlled by adjusting the headspace gas composition?
  - Lactic acid bacteria stays in lag phase when under pressure of Pseudomonas spp.
    - Pseudomonas spp. are strictly aerobe microorganisms
  - Pseudomonas spp. stay predominant in the microbial population dynamics when oxygen is still available
- → Hypothesis: Lactic acid bacteria population growth can be inhibited by applying a semi-anaerobe gas conditions





#### Material and method

 Compare fresh-cut iceberg lettuce (sample of 200g) stored at 7°C for a total period of 8 days

- Two storage conditions:
  - E-MAP: commercial packaging → polypropylene bags (670 cm², 30μm thickness, initially flushed with 6kPa O₂ and 11kPa CO₂)
  - CA: fresh-cut lettuce stored in glass-jar, continuously flushed with humidified gas mix made of 0.3-0.5kPa O<sub>2</sub> and 12-14kPa CO<sub>2</sub>





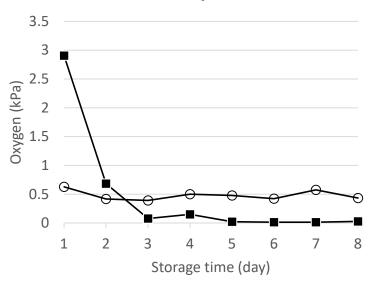


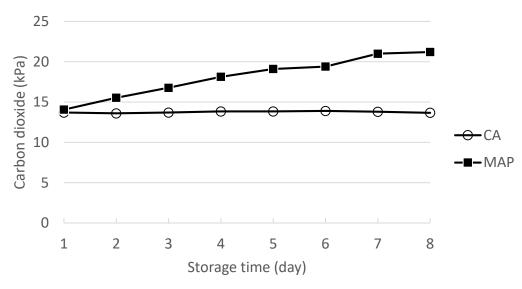




#### Gas content

- Fresh-cut lettuce consumes the oxygen and produces CO<sub>2</sub> --> respiration rate
  - In MAP: Anaerobe condition reached after 3 days of storage and CO<sub>2</sub> content build up into packaging
  - In CA: O<sub>2</sub> and CO<sub>2</sub> content are controlled by the flushing set-up





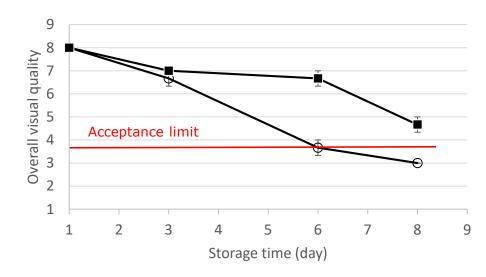


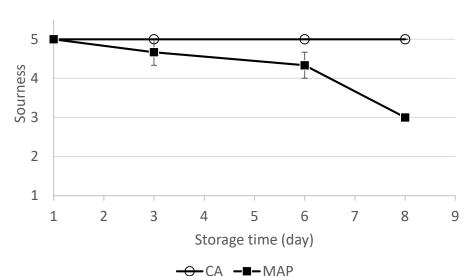


# OVQ + sensorial quality

 Low quality score for CA storage due to discoloration (oxidation process)

No sourness development when stored under semianaerobe condition.







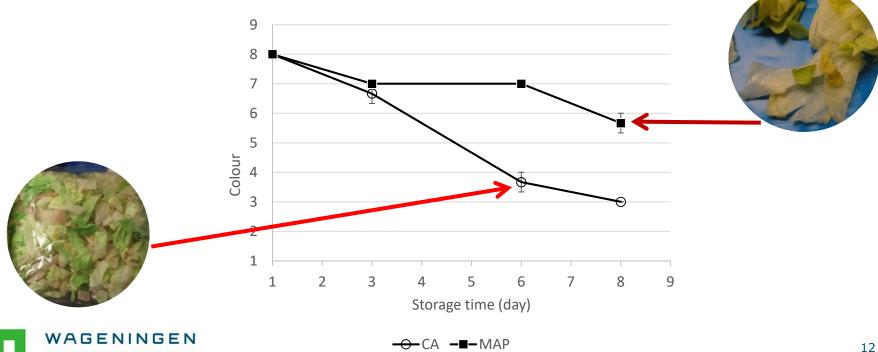


#### Discoloration

Pink discoloration results of oxidation processes occurring under semi-anaerobe condition

Brown discoloration is a result of tissue collapsing and

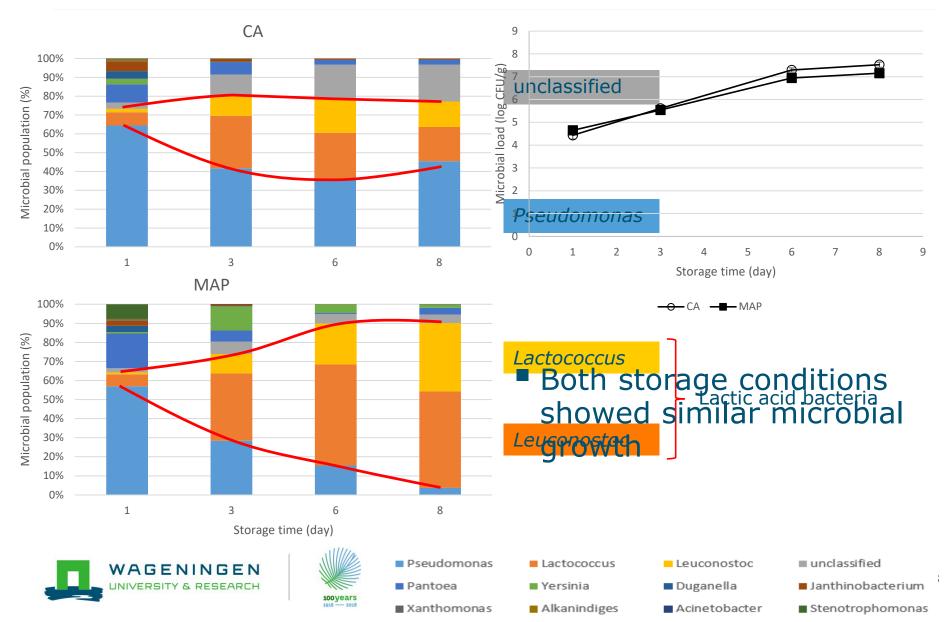
anaerobe process





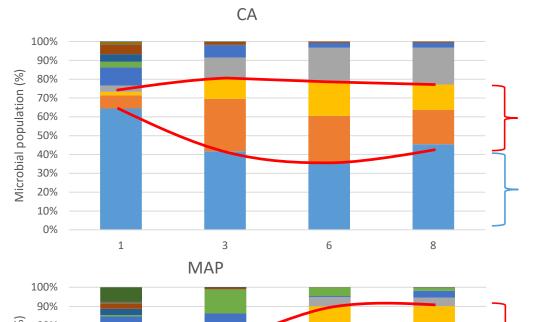
# Microbial population dynamics

(16S rDNA amplicon mass sequencing)

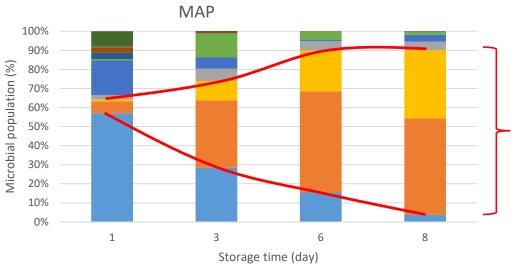


### Microbial population dynamics

(16S rDNA amplicon mass sequencing)



 CA: Lactic acid bacteria remains under pressure by aerobe bacterial population (Pseudomonas)



 Higher proportion of lactic acid bacteria causes off-odour (sourness)





■ Pseudomonas
■ Pantoea

years
■ Xanthomonas



■ Leuconostoc
■ Duganella
■ Acinetobacter

unclassifiedJanthinobacteriumStenotrophomonas

#### Conclusion

- Storing fresh-cut iceberg lettuce under CA condition (semi-anaerobe gas conditions) limits the growth of lactic acid bacteria:
  - --> No sourness odour development
  - --> But did not avoid discoloration of lettuce tissue (oxidation process)
- MAP creates anaerobe gas condition within 3 days of storage:
  - --> Correlation between the anaerobe condition, the growth of lactic acid bacteria and the sourness quality decay





# New insight & recommendations

- Packaging concept affects:
  - Activity of fresh-cut product (respiration rate, physiological and enzymatic processes)
  - Microbial population dynamic
- Use of more permeable packaging may help to control the growth of lactic acid bacteria, but extra hurdles (ex: Heat shock, mutant lettuce variety, ...) should be combined to avoid discoloration (due to semi-anaerobe gas condition)





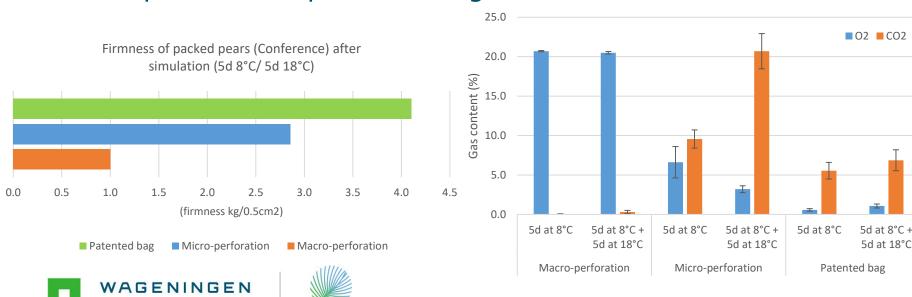
# Dynamic packaging concept

New packaging material allows to store fresh-product at several storage temperatures: gas transmission rate of packaging follow similar rate that product activity



Patent: PCT/EP2019/067393

Pears stay longer firm and green when packed into patented bags
 Conference pears



# Thank you for your attention

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