

Elucidation of protein-polphyhenol interactions in *Moringa oleifera* leaves during protein extraction using an *in-vitro* digestion model

Introduction

Moringa oleifera is a green plant from the sub-Himalayan tract of India, Bangladesh and Afghanistan

Interesting new source of protein

- Grows in both humid and dry conditions
- Good production yield
- Alternative to animal protein
- Up to 30% of proteins (dry matter)

Source of many bioactives

- Glucosinolates (e.g. glucomoringin)
- Polyphenols (e.g. quercetin and kaempferol)

Polyphenols provide good antioxidant capacity which help to prevent noncommunicable diseases

But they may also bind to the proteins and this

- Limits the protein extraction yield
- May precipitate the proteins
- Reduces the protein digestibility

Objectives:

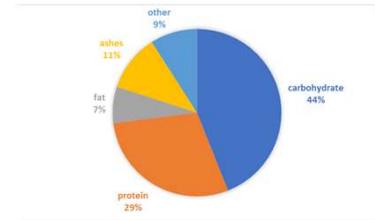
- Use an *in-vitro* digestion system to assess impact on protein digestibility
- Find which incubation parameters lead to digestibility improvement



Moringa oleifera growing regions

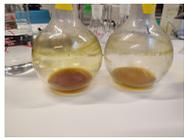


Moringa oleifera leaves and powder



Composition of Moringa leaves dry matter [%]

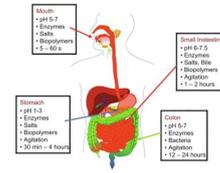
Methodology



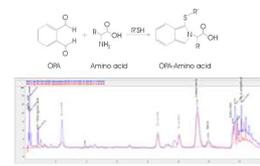
Final polyphenol extract



Schematic representation of the protein polyphenol mix



In-vitro digestion system



Used machines (spectrophotometer and HPLC-DAD) and the OPA reaction as well as the HPLC outcome with identification of the six different polyphenols

Polyphenol extraction from *Moringa oleifera* leaves

- Isolation
- Purification
- Quantification

Incubation of the *Moringa oleifera* polyphenols extract with

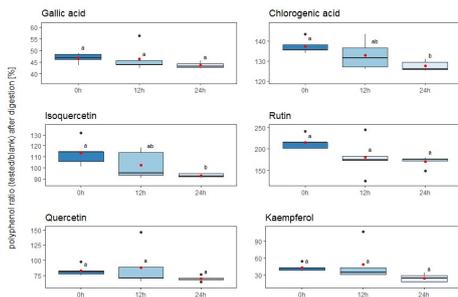
- Bovine serum albumin
- Rubisco
- Moringa oleifera* protein extract
- Variation of different incubation parameters

Use of a micro scale *in-vitro* digestion model for the assessment of the digestibility of the treated and non-treated samples with simulation of the different digestion phases

Assessment of the digested samples by

- Measuring the release of amino acids
- Measuring the polyphenol concentrations

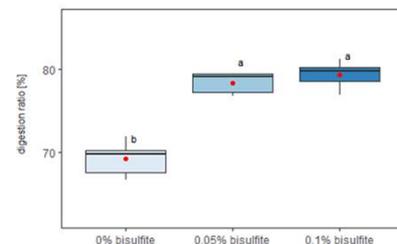
Results



Impact of the digestion on the polyphenol ratios of the mix between *Moringa oleifera* protein extract and polyphenols, after different incubation times

- The incubation at 4 and 40 °C improved the digestibility and the polyphenol preservation for all mixes.
 - Polyphenoloxidases are less active or partially deactivated in those conditions
- Addition of sodium bisulfite at a concentration of at least 0.05% improved protein digestibility.
 - This salt provides an antioxidant capacity that limits the polyphenol oxidation
- Over time the polyphenol concentration decreases independantly of the protein type.

- Over time two trends are visible:
 - Animal protein (BSA) digestibility increased
 - Plant based proteins digestibility decreased
- Plant based proteins were less impacted by polyphenols than animal proteins
 - Difference of structure
 - Difference of treatment



Impact of the digestion on the digestion ratios of the mix between BSA protein and polyphenols, after incubation with addition of different sodium bisulfite concentrations

Conclusion

- Possible to improve the digestibility
- Method optimisation required to obtain clearer trends