Monitoring solubles mono-, di- and oligosaccharides in Jerusalem artichoke with HPLC-ELSD

Stéphane Lamaudière[1], Bruno Godin[2], Richard Agneessens[2], Thomas Schmit[2], Jean-Pierre Goffart[2], Didier Stilmant[2], Jérôme Delcarte[2] and Patrick A. Gerin[1]

Unit of Bioengineering, Earth & Life Institue, Université catholique de Louvain, Croix du Sud 2, box L7.05.19, B-1348 Louvain-la-Neuve, Belgium (stephane.lamaudiere@uclouvain.be)

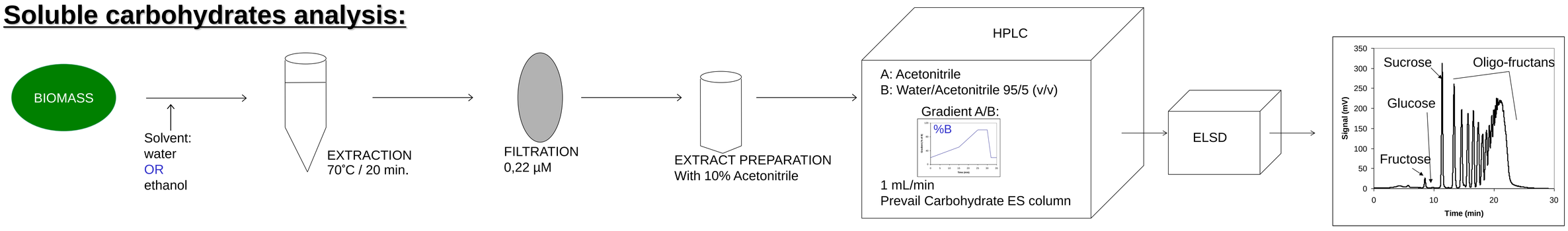
•Walloon Agricultural Research Center, CRA-W, Rue du Serpont, 100, B-6800 Libramont, Belgium

Introduction: Jerusalem artichoke biomass consists mainly of structural polysaccharides, reserve polyfructans and solubles sugars that can be converted by microorganisms to ethanol.

Aim: - To test a method to extract the main mono-, di- and oligo-saccharides present in the biomass and to analyse them with HPLC-ELSD.

- To determine fructose, glucose, sucrose and oligo-fructans availability in the stalks, leaves and tubers.

Soluble carbohydrates analysis:



Extraction: ethanol vs water:

Figure 1 & 2: Chromatogram of mono-, di-, and oligo-saccharides of Jerusalem artichoke tubers after first extraction (▬▬) made with water (Figure 1) or ethanol (Figure 2) at 70˚C/20min. Second extraction (▬▬) with hot water at 70˚C/20min. Duplicate experiment.

The sugars present in the second extract correspond to sugars not extracted in the first extraction.

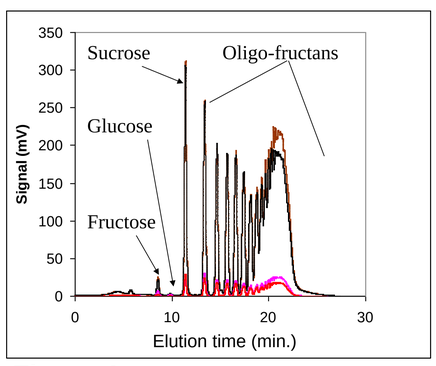


Figure 1

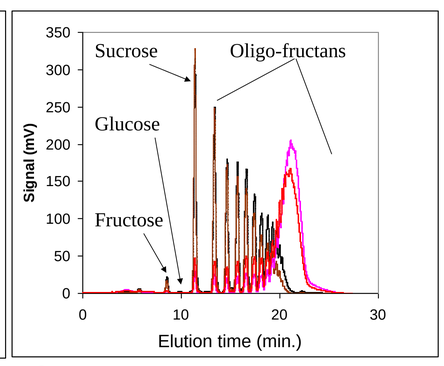


Figure 2

Extraction of mono-, di- and oligo-saccharides with hot water (70˚C/20 min) (Figure 1) is more effective than extraction with ethanol (Figure 2) or methanol (not shown).

Calibration of HPLC-ELSD analysis

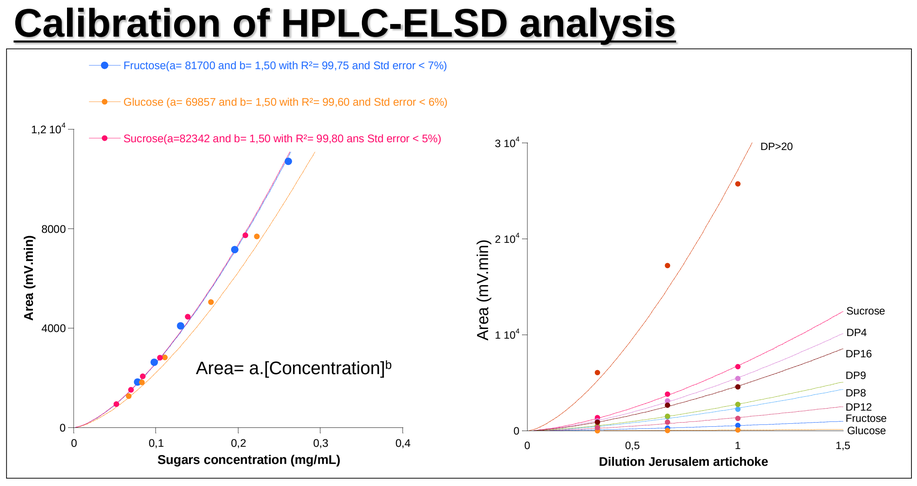


Figure 3: Calibration of fructose, glucose,sucrose and oligo-fructans analysis. Fitting of Area= a.[Concentration]$^{1.5}$. Standard error on "a" is presented.

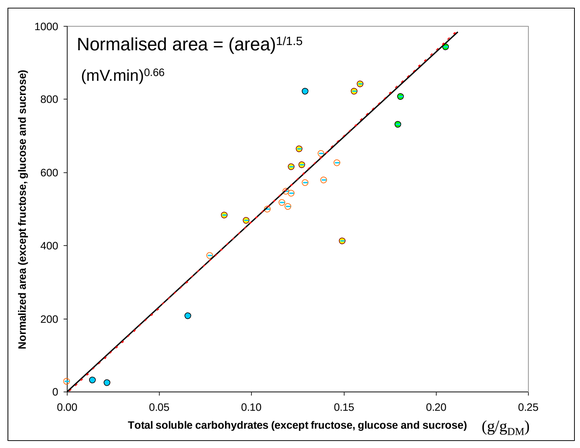
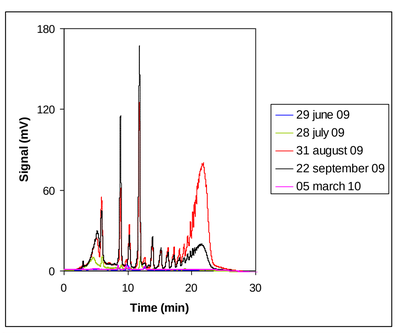
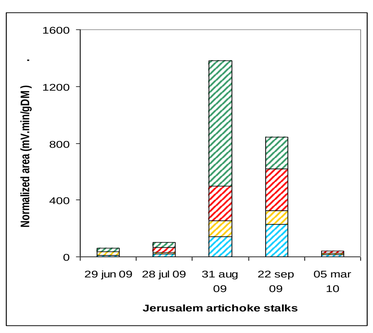


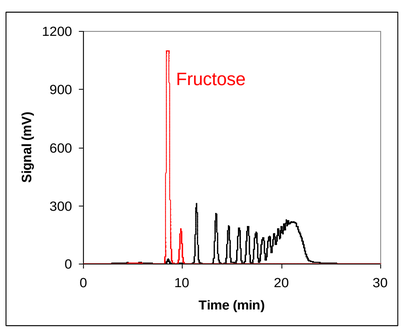
Figure 4: Correlation between oligofructans normalised peak area (HPLC-ELSD) and total soluble carbohydrate (Luff-Shorll method). For consistency, fructose, glucose and sucrose were subtracted from total soluble carbohydrate.

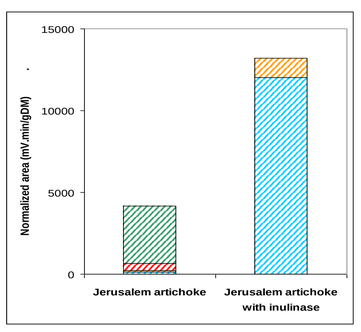
- ELSD response: Area = a.[concentration]^{b}
- « b » can be set to 1.5
- Standard error of « a » < 7%
- Fitting R^2 is > 98%
- a linear correlation seem exists with area and Luff method for oligo-fructans:
  - Normalized area = a'.[TSC] with a'=4685 (R^2= 0.84)
    (TSC total soluble carbohydrates (mg/g_{DM}))

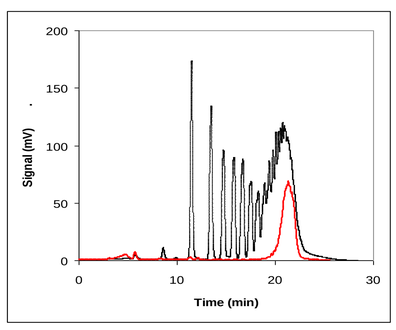
Monitoring of Jerusalem artichoke's soluble saccharides











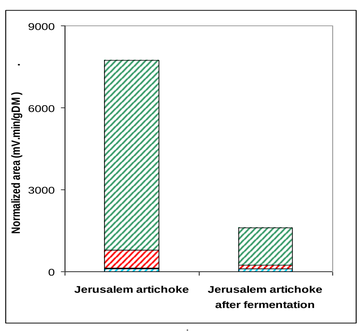


Figure 5a : Soluble sugar concentrations of stems and leaves is maximum at the end of august with fructose 7.2 mg/g_{DM}, glucose 7.4 mg/g_{DM}, sucrose 13.8 to 15.5 mg/g_{DM} and oligofructans 129 mg/g_{DM}. Oligo-fructans possibly translocate to the tuber in september.

Figure 5b : Inulinase hydrolyses tuber poly-fructans to fructose. Fructose concentration increases from 8.3 to 670 mg/g_{DM}.

▬ before inulinase treatment or fermentation

▬ after inulinase treatment or fermentation

Oligo-fructans
Sucrose
Glucose
Fructose

Figure 5c : *S. cerevisiae* was able to ferment tuber mono- and di-saccharides as well as the oligo-fructans up to a DP of 11. Hydrolysis was not complete for higher DP.

Interpretation and Conclusion

- Response of HPLC-ELSD method are Area = a x [Concentration]^{b} where b can be set to 1.5
- The developed method is sustable to monitor mono-, di-, and oligo-saccharides in Jerusalem artichoke

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