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#### Review

## Fructooligosaccharides and fructans analysis in plants and food crops



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#### ABSTRACT

Because fructooligosaccharides (FOS) and fructans are increasingly important in food and nutrition sciences, separation and analysis tools, in particular with the development of analytical chemistry, are slowly but rationally being developed for their separation and identification in plants, crops, and food products. Several chromatographic and other methods have been described to assess FOS and fructans, however, most of these methods are technically complicated, time-consuming and expensive. Although modern techniques have evolved tremendously, FOS and fructans analyses involve multiple and/or sequential extractions, chemical or biochemical polymers hydrolysis, and multiple chromatographic and/or other analytical runs to quantify these polymers. This paper aims to review and describe the different chromatographic techniques and other methods that are used to separate, analyze and quantify FOS and fructans © 2013 Elsevier B.V. All rights reserved.

#### **Contents**

1.	Introduction		54
2.	FOS and fructans chemistry and structural composition.		55
3.	Analytical techniques of f	nalytical techniques of fructans and FOS	
	3.1. Chromatography		57
	3.1.1. Thin layer	r chromatography (TLC)	57
		romatography	
	3.1.3. Gas chron	natography	58
	3.1.4. Chromato	ography coupled to mass spectrometry	58
3.2	3.2. Other techniques.	3.2. Other techniques	
4.	How then to conduct FOS	How then to conduct FOS and fructans analysis?	
5.		Conclusion	
	Acknowledgements	cknowledgements	
References			60

### 1. Introduction

Over the past three decades during the development of analytical chemistry, interest in measuring fructan oligo- and polymers has been stimulated by new findings on nutritional benefits. These polymers are being used as foods ingredients and are commonly incorporated as dietary fibers in many food products. Several procedures were initially described to assess the levels of fructooligosaccharides (FOS) and fructans in plants, crops and food products, however, most of these methods of analysis are

modern techniques have evolved tremendously during the last three decades, analyses of FOS and fructans involve multiple and/or sequential extractions, chemical (acidic) or biochemical (enzymatic) polymers hydrolysis, and multiple chromatographic and/or analytical runs to quantify FOS and fructan-derived hexoses [1,2]. On the other hand, highly polymerized fructans and long chain FOS (DP > 12) cannot be identified using these modern techniques because of (i) the high DP, (ii) absence of high DP standards (DP > 5), and (iii) the difficulty in separating these high polymers. Thus, one of the most described method used to assess fructans is posthydrolysis measurement of FOS or fructans chains to fructose (and eventually glucose), but this introduces the problem of indepen-

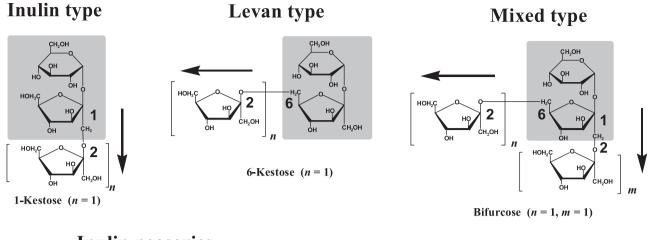
dently removing, or measuring, sucrose, fructose and glucose [2].

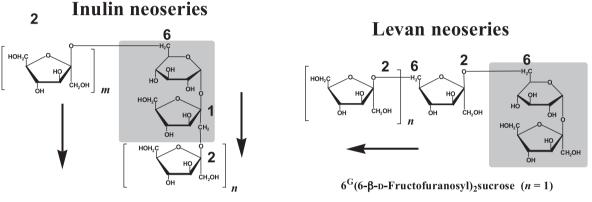
technically complicated, time-consuming and expensive. Although

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1<sup>F</sup>, 6<sup>G</sup>-Di-β-D-Fructofuranosylsucrose (m = 1, n = 1)

Fig. 1. Molecular structures of the different types on fructooligosaccharides found in higher plants.

Currently, most of the approaches employed in the analysis of FOS and fructans are based on utilizing a multistage enzymatic digestion of the sample following the analysis of mono and disaccharides by high pressure liquid chromatography (HPLC). After enzymatic or chemical hydrolysis of the sample, the quantities of fructose and glucose released are measured, and FOS and fructans contents are estimated from the quantity of the released glucose and fructose.

FOS and fructans have a history of two centuries, and some review articles have reported on some historical aspects but have included very little on the general history of fructans research [3–5]. Prior to the contemporary science of FOS and fructans, ancient people have been using fructans-containing plants not only as a source of food, but also as animal feed and medicine. The most common and oldest vegetables used were onion and garlic which were widely used in ancient Egypt in their rituals. These two plants have been included as ones of the numerous symbols of the religious rites. Nevertheless, the modern and present history of fructans began really with their discovery by the scientist Rose (1804). Since the turn of the 20th century considerable development ensued when the scientist Edelman proposed a mechanism concerning their metabolism in higher plants. More recently, FOS and fructans research has known considerable progress, in particular with the boom of molecular biology tools. Thus, the scope of FOS and fructans research has been moved from being a basic to an applied science. At present time, FOS and fructans are not only considered as food ingredients but as foods, and are found in more than 500 food products resulting in significant daily consumption. Because the science of nutrition itself has changed, FOS and fructans are now considered as functional foods [6,7]. In addition, they are nowadays used as feed additive for poultry in the USA and Japan. This passionate history of FOS and fructans concerning their safety and health benefits continues to evoke the interest of scientists who are discovering daily their potential as food and food ingredients.

Fructans are defined as any polymer where one or more fructosyl-fructose linkage constitutes the majority of the linkages [2,7], and this polymeric material includes other even smaller oligomers such as the disaccharide, inulobiose. Although the definition of FOS and fructans may or may not consider molecules containing p-glucosyl substituents, the terms oligomer and polymer are often used to distinguish between materials which can be specifically characterized and those which cannot [8].

#### 2. FOS and fructans chemistry and structural composition

In nature, FOS and fructans are widely distributed in the plant kingdom, and are present in monocotyledons, dicotyledons and in green algae. Depending on their source, FOS and fructans differ in both molecular structure and weight. They may be classified into three main types: the inulin group, the levan group and the branched group. The inulin group consists of materials that possess mostly or exclusively  $(2 \rightarrow 1)$  fructosyl-fructose linkages. Levan is any material that contains mostly or exclusively  $(2 \rightarrow 6)$  fructosyl-fructose linkages. The branched group contains both  $(2 \rightarrow 1)$  and  $(2 \rightarrow 6)$  fructosyl-fructose linkages in significant amounts (e.g. graminan from Gramineae).

From a chemical point of view, FOS and fructans polymers, are polyfructosylsucroses of varying molecular size built on a sucrose starter unit and are biochemically designated by  $1^F$  (1- $\beta$ -D-fructofuranosyl) $_n$  sucrose oligomers where n may vary from 0 to up to 15 for FOS, and up to 200 for fructans. Contrary

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