ORIGINAL ARTICLE

Open Access



ISSN 2278-1404

Thin layer chromatographic analysis of extracted Fructooligosaccharides from organic waste

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Manuscript: received 18 November, 2018 revised 16 Dec, 2018 accepted 18 Dec, 2018

Abstract

Nutraceuticals and functional foods are increasing in popularity in the recent years. Fructooligosaccharides (FOS) has emerged as functional food ingredient in market and is very well known for its prebiotic properties. Hence, there is a growing need for extensive research on usage and extraction of Fructooligosaccharides. In this study, organic wastes namely onion waste and spoilt wheat have been explored for its potential use in extraction of FOS. The extraction was done using ethanol and optimization was done by considering different initial raw material weights. Thin Layer Chromatography (TLC) confirmed the presence of FOS in the extracts.

Keywords: Nutraceuticals, Fructooligosaccharides, TLC.

1. Introduction

The nutritional and therapeutic benefits of prebiotics have attracted the keen interest of consumers and food processing industry for their use as food ingredients. Nutraceuticals and functional foods (NFF) are increasing in popularity as a tool of the consumers for the management of their health and wellness. Pre-, pro- and symbiotic are an important group of NFF shown to be effective in modulating gastrointestinal diseases and other ailments^[1-2].

The main objective of this study is efficient extraction and optimization of FOS from organic wastes. FOS being a prebiotic offers numerous benefits as already mentioned above. The food industry produces a large amount of onion waste, making it necessary to search for possible ways for their utilization. One way could be to use these 'waste' onions as a new and natural source of high-value functional ingredients, due to the presence of bioactive compounds in onion, which present health benefits ^[2]. Not only onions, spoilt wheat can also be utilized in the same way. Therefore, efficient extraction of FOS has been worked on from onions and spoilt wheat. Ethanol extraction was used to minimize side effects. TLC was carried out for qualitative estimation^[3-4].

1.1 Fructooligosaccharides

Fructo-oligosaccharides (FOS), new alternative sweeteners, constitute 1- kestose, nystose, and 1-beta-

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Department of Biotechnology, R.V College of Engineering, Bangalore-560059, India fructofuranosyl nystose is produced from sucrose by the action of fructosyltransferase from plants, bacteria, yeast, and fungi. FOS has non-cariogenic properties, low caloric values and help gut absorption of ions, bifidus-stimulating functionality and, decrease levels of lipids and cholesterol ^[5-6]. The purified linear fructose oligomers are added to various food products like cookies, yoghurt, infant milk products, desserts, and beverages due to their potential health benefits. Among the prebiotics, fructo-oligosaccharides (FOS) are of major focus due to their properties and great economic potential for sugar industry. Being 0.4–0.6 times sweeter then sucrose, these are being used in pharmaceutical

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1.2 Thin-layer chromatography

industry as artificial sweeteners ^[6-7].

TLC is a chromatography technique that separates non-volatile mixtures. Thin-layer chromatography is performed on a sheet of glass, plastic, or aluminum foil, which is coated with a thin layer of adsorbent material, usually silica gel, aluminum oxide (alumina), or cellulose. This layer is called as the stationary phase.

After addition of the sample on the plate, the mobile phase (a solvent or solvent mixture) is drawn up the plate via capillary action. Because completely different analytes ascend the TLC plate at different rates, separation is achieved ^[8-11].

2. Materials and methods

The analysis performed by TLC plate showed the production of FOS from the dried powder of onion waste and wheat waste. The spots were compared with the known standard values. In the current investigation, a

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standard protocol was followed.

Hence, to determine FOS qualitatively, TLC plates of Silica Gel 60 F254 (Merck, Germany) were used. The plate served as a stationary phase in which at the bottom side, spots of the unknown samples were loaded using a capillary tube.

For mobile phase, a solution of Acetonitrile and Water was prepared (85:15). After the complete run by the mobile phase, the plate was dried at 100°C for 10mins. In order to observe the spots, a staining reagent of 0.2% orcinol (mixture of methanol and Sulphuric acid in the ratio 90:10) was sprayed onto the plate and was dried at 100°C for 10mins.

Finally, the plate was observed under Long UV and the various spots were marked and Rf values were calculated [8-11].

Table 1: Distance travelled by the spots and the corresponding Rf values

Point	Distance travelled (in cm)	Rf value
01.1	2.9	0.41
01.2	4.4	0.62
01.3	5.6	0.8
01.4	6.6	0.94
02.1	2.9	0.41
O2.2	4.5	0.64
O2.3	5.5	0.78
02.4	6.5	0.92
W1.1	2.8	0.4
W1.2	3.7	0.53
W1.3	5.5	0.78
W2.1	2.9	0.41
W2.2	3.7	0.53
W2.3	5.4	0.77

Table 2: Standard Rf values^[11-13]

Rf	Substance	
0.17	Lactose	
0.26	Maltose	
0.38	Sucrose	
0.42	Galactose	
0.44	Glucose	
0.47	Mannose	
0.51	Fructose	
0.53	Arabinose	
0.66	Xylose	
0.69	Ribose	
0.74 Rhamnose		

3. Results and discussion

Thin layer chromatography was performed to confirm the presence of FOS in the extracts. FOS as mentioned, consists mostly of 3 types 1-kestose, nystose, and fructofuranosyl nystose. In both wheat and onion extracts, 1-kestose and fructofuranosyl nystose was detected which confirms FOS being present in the extract ^[13-15].

Table 3: Standard Rf values[11-13]

Standard	Rf value
1-kestose	0.54
Nystose	0.48
Fructofuranosyl nystose	0.39

Table 4: Distance travelled by the spots and the corresponding Rf values

Point	Distance travelled (in cm)	Rf value
01.1	2.9	0.41
01.2	4.4	0.62
01.3	5.6	0.8
01.4	6.6	0.94
O2.1	2.9	0.41
O2.2	4.5	0.64
O2.3	5.5	0.78
O2.4	6.5	0.92
W1.1	2.8	0.4
W1.2	3.7	0.53
W1.3	5.5	0.78
W2.1	2.9	0.41
W2.2	3.7	0.53
W2.3	5.4	0.77

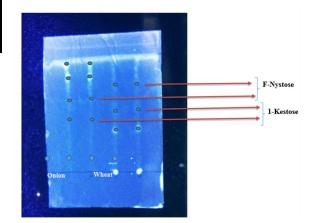


Fig 1: TLC Plate observed under Long UV

4. Conclusion

In the study carried out, it can be concluded that Fructooligosaccharides is indeed present in the organic wastes of onions and wheat. Since the method of extraction was by the means of ethanol, it proved to be a less time-consuming procedure and easier work conditions were required. From the TLC Analysis, it was very much evident of the extraction of FOS. Upon further purification by strategies like ultra filtration and charcoal purification, pure samples of FOS can be obtained^[15-18]. The other characterization technique such as FTIR (Fluorescence Transformed Infrared

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Spectroscopy), MS (Mass spectrometry) analysis and HPLC (High Performance Liquid Chromatography) analysis can further test the purity of the extract. The study has proven that the onions are a good source of antioxidants with the highest capacity found in the outer sections. Therefore, onion waste could be used to produce functional ingredients with important heath benefiting properties, due to the presence of bioactive compounds. Wheat too, is one of the major sources of food-waste that is generated^[19-20].

Acknowledgment

The authors listed in this paper wish to express their appreciation to the RSST trust Bangalore for their continuous support and encouragement. As a corresponding author, I also express my sincere thanks to all other authors whose valuable contribution and important comments made this manuscript to this form.

Conflict of interest

The authors listed in this paper have no conflict of interest known best from our side. There was also no problem related to funding. All authors have contributed equally with their valuable comments which made the manuscript to this form.

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