IN VITRO GASTROINTESTINAL STABILITY AND BIOACCESSIBILITY OF GLUCOSINOLATES FROM SELECTED PLANTS OF THE ORDER BRASSICALES

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INTRODUCTION

Glucosinolates (GSLs), compounds found in *Brassica* vegetables, provide various human health benefits, such as anticancer, antimicrobial, antihyperglycemic, and anti-inflammatory activities. Therefore, it is extremely important to evaluate the rate of stability and bioaccessibility of GSLs after digestive process. The aim of this study was to determine the GSLs in the plant seeds of various mustard plants belonging to the order Brassicales, as well as their stability and bioaccessibility after two simulated digestion methods – an *in vitro* method based on the use of commercial enzymes from the stomach and small intestine, and an *ex vivo* method with human digestive juices from the stomach and small intestine.

MATERIALS AND METHODS

Various mustard seeds (*Sinapis alba* L., *Brassica juncea* L., and *Brassica nigra* L.) were purchased commercially and ground to a fine powder using a coffee grinder. Intact GSLs were extracted from ca. 100 mg of each ground plant parts with a EtOH/H_2O (70:30 v/v) at 80 °C to inactivate myrosinase activity followed by ultrasound treatment. The two-phase *in vitro* digestion method includes simulated digestion in the stomach and small intestine with commercial digestive enzymes, and *ex vivo* digestion method with collected human gastric and duodenal enzymes. The extracts were purified on an ion-exchange column. After sulfatase (*Helix pomatia*) addition, the desulfo-GSL were eluted with water. The identification of GSLs, their gastrointestinal *in vitro* and *ex vivo* stability and bioaccessibility were determined using UHPLC-DAD-MS/MS.

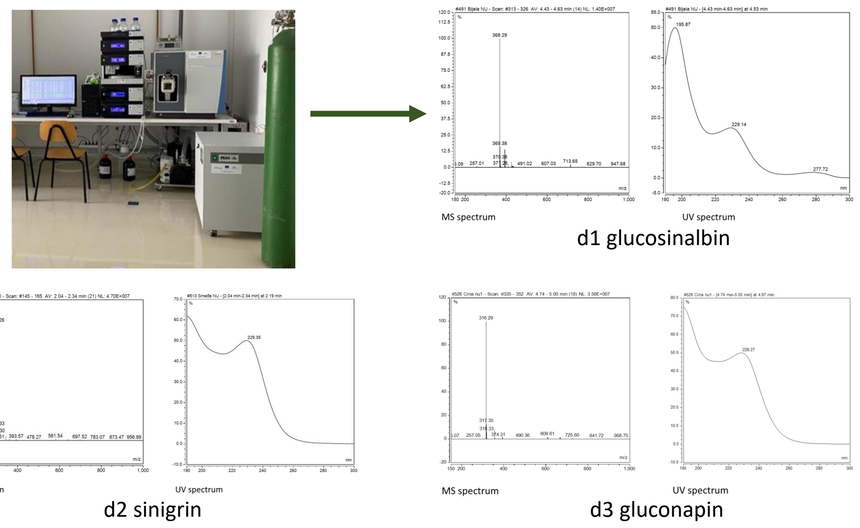

Figure 1. MS and UV spectra of the main desulfo-GSLs in various mustard seeds

Table 1. GSL contents in various mustard seeds (*S. alba, B. juncea*, and *B. nigra*)

GSLs present in mustard seeds	Content (μmol/g DW)		
	S. alba	*B. juncea*	*B. nigra*
Glucosinalbin	142.33	/	/
Sinigrin	/	53.56	12.40
Gluconapin	/	/	42.30
4-Hydroxyglucobrassicin	/	1.00	1.60
Total	142.33	54.56	56.30

GSL glucosinolate
DW dry weight

Table 2. Bioaccessibility of GSLs from various mustard seeds after two-phase *in vitro* and *ex vivo* digestion methods

Digestion phases	Bioaccessibility (%)					
	Various mustard seeds					
	S. alba 1	*B. juncea* 2	*B. juncea* 3	*B. nigra* 2	*B. nigra* 3	*B. nigra* 4
In vitro gastric phase	74.94	63.51	97.00	88.52	83.07	79.39
In vitro intestinal phase	54.04	35.36	75.00	23.59	14.65	28.49
Ex vivo gastric phase	85.58	93.38	~100.00	97.24	84.38	~100.00
Ex vivo intestinal phase	80.26	41.44	35.00	41.18	15.25	64.48

1 Glucosinalbin
2 Sinigrin
3 4-Hydroxyglucobrassicin
4 Gluconapin

DISCUSSION AND CONCLUSION

Biologically active compounds found in vegetables consumed daily, are increasingly the focus of scientific research. Glucosinalbin, sinigrin, and gluconapin are the main GSLs present in three different mustard seeds: *S. alba, B. juncea*, and *B. nigr*a. After the *in vitro* and *ex vivo* intestinal phases, the GSL contents were significantly reduced compared to the *in vitro* and *ex vivo* gastric phases. The absorption of GSLs in the digestive system is of great importance precisely because of the biological activities exhibited by their degradation products such as antiproliferative, antimicrobial, anti-inflammatory, and antihyperglycemic activities.

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