STUDIES ON INTESTINAL TRACT GLUCOSINOLATE CONTENT, XENOBIOTIC METABOLIZING ENZYMES AND THYROID STATUS IN GERM-FREE AND CONVENTIONAL RATS FED RAPESEED MEAL

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ABSTRACT

Rats housed in conventional or germ-free environments were fed a rapeseed meal diet for four weeks. Intact and hydrolysed glucosinolate concentrations were determined in intestinal contents and related to diet concentrations using chromic oxide as a marker. Intact glucosinolate disappearance was evident in the stomach of both conventional and germ-free animals. Further disappearance occured in the lower GI tract of the conventional animals but not in the germ-free animals. This effect was reflected in differences in organ weight, thyroid status and liver xenobiotic metabolizing enzyme activities.

INTRODUCTION

Rapeseed meal contains high quality protein but its use in animal diets is limited by its content of glucosinolates (Bell 1993). In a review of the effects of glucosinolates Duncan (1992) indicated that numerous studies have demonstrated the gross effects of glucosinolates but data on specific digestive and systemic influences is not available. In this regard, a study was conducted to follow the fate of ingested glucosinolates in the GI tract of animals and to evaluate systemic effects by measuring organ weights, thyroid status and tissue xenobiotic metabolizing enzymes. Conventional and germ-free animals were used to assess the influence of intestinal microflora on the responses to dietary glucosinolates.

EXPERIMENTAL

Fate of ingested glucosinolates

Twelve rats maintained in a conventional environment and twelve rats housed in a germ-free environment were fed a rapeseed meal (cv. Darmor) diet for four weeks at which time all rats were killed and intestinal contents were obtained. Glucosinolate

concentrations in intestinal contents and feces were adjusted for disappearance of diet dry matter with the aid of a dietary marker (0.25% chromic oxide). Both total intact glucosinolates and indole glucosinolates showed a marked disappearance from stomach contents in conventional and germ-free animals. The majority of intact glucosinolates disappeared in the lower part of the GI tract in conventional animals but remain virtually unchanged in the germ-free animals indicating a marked effect of intestinal flora on intact glucosinolates. Hydrolysed glucosinolates were at a very low level or not detected in the lower part of the GI tract. The data indicates a limited possibility for absorption of hydrolysed intact glucosinolates in germ-free rats in contrast to a high possibility in conventional animals.

Table 1. Intact and hydrolysed glucosinolate[1] concentrations (μmoles/g) in the GI tract and feces of conventional and germ-free rats fed a rapeseed meal diet.

Item	Conventional				Germ-free			
	Intact		Hydrolysed		Intact		Hydrolysed	
	Total	Indole	OZT	SCN	Total	Indole	OZT	SCN
Diet	23.6	3.1	0.03	1.0	23.6	3.1	0.03	1.0
Stomach	8.4	1.6	0.07	2.3	10.7	1.8	0.08	2.1
S. intestine	5.6	0.7	0.02	0.0	9.1	1.2	0.04	0.0
Cecum	0.1	0.0	0.02	0.0	12.2	1.1	0.05	0.0
Colon	0.1	0.0	0.02	0.0	10.0	1.1	0.03	0.0
Feces	0.1	0.0	0.01	0.0	8.6	0.6	0.03	0.0

[1]Total and oxazolidinethione (OZT) determined by GLC; indole and SCN determined indirectly as bound (indole) or free (SCN) thiocyanate ion.

Body weight gain, organ weights and thyroid status

Body weight gain, organ weights and plasma thyroid hormone levels were determined at the termination of the four week experimental period (Table 2).

Table 2. Body weight gain, organ weights and plasma thyroid hormone levels of conventional and germ-free rats fed a rapeseed meal diet for four weeks.[1]

Environment	Weight gain	Organ weight			Plasma T4
	g	Liver g/100g BW	Kidney g/100g BW	Thyroid mg/100g BW	nmol/l
Conventional	85^{b}	5.54^{a}	1.00^{a}	8.4^{a}	42.59^{b}
	±4	±0.08	±0.01	±0.2	±1.32
Germ-free	101^{a}	4.50^{b}	0.85^{b}	6.6^{b}	50.03^{a}
	±3	±0.06	±0.01	±0.2	±2.04

[1]Mean ±standard error. [ab]Means with different superscripts are significantly different ($P<0.01$).

Rats maintained in the germ-free environment gained more body weight and the relative liver, kidney and thyroid weights were lower than those of conventional animals. The elevated organ weights in the conventional animals may be attributed to the influence of intestinal microflora on dietary glucosinolates. Plasma T_4 levels were depressed in the conventional animals but no difference in comparson to the germ-free rats was evident for T_3 (mean = 1.96 nmole/l plasma).

Liver xenobiotic metabolizing enzymes

The activities of liver xenobiotic metabolizing enzymes were compared between conventional and germ-free animals to assess the influence of dietary glucosinolates as modified by the presence of intestinal microorganism (Table 3). Dietary glucosinolates in conventional animals did not result in a significant depletion of cytochrome P450 as had been shown previously (Nugon-Baudon et al. 1990) but an induction of the activities of glutathionine-S-transferase and UDP-glucuronosyltransferase as shown previously was evident. It may be suggested that glucosinolate-derived products absorbed from the GI tract were responsible for the effects observed.

Table 3. Xenobiotic metabolizing enzyme levels in livers of conventional and germ-free rats fed a rapeseed meal diet for four weeks[1]

Environment	Cytochrome P450 nmol/mg protein	Glutathione-S-transferase μmol/min/mg protein	UDP-glucuronosyl-transferase nmol/min/mg protein
Conventional	0.48 ±0.04	1.20[a] ±0.08	0.71[a] ±0.02
Germ-free	0.60 ±0.03	0.67[b] ±0.04	0.26[b] ±0.02

[1]Mean ± standard error. Means with different superscripts differ significantly ($P<0.01$).

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