# Response of rats to grapefruit flavonoids and inulin fed singly or in combination\*

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#### ABSTRACT

Rats of an initial body weight of 124 g were fed for 28 days on a casein diet unsupplemented or supplemented with grapefruit flavonoids (FL) 0.3% or inulin (IN) 5 and 10%, given singly or in combination. Feed intake and weight gain were registered and protein digestibility and N retention were determined. At the end of the experiment blood was sampled and concentrations of cholesterol (total and HDL), triacylglycerols (TAG), glucose, Mg, Ca and P were determined. FL fed either alone or with IN depressed feed intake, and FL+IN10 also body gain. The supplements did not affect feed conversion or protein digestibility, N utilization was or tended to be higher in rats fed IN singly or with FL. FL did not affect the total cholesterol level while IN fed alone tended to decrease, and fed with FL, significantly decreased total cholesterol. Both supplements tended to decrease TAG concentration.

KEY WORDS: flavonoids, oligosaccharides, nutritive value, rats

#### INTRODUCTION

Hypolipidemic effects are attributed to many plant substances, including flavonoids and other polyphenols (Kuo et al., 2005), and to poorly digestible saccharides with prebiotic properties (Delzenne et al., 2002). The results of some works indicate, however, that both types of substances can decrease the nutritional value of diets. Polyphenols, particularly condensed tannins, can decrease the digestibility and biological value of protein (Shahkhialili et al., 1990) while poorly digestible poly- and oligosaccharides can reduce the energy value of a diet (Delzenne et al., 2002). Therefore, the effects of the combined use of both substances on animal performance and gastrointestinal tract metabolism, may be of interest.

<sup>\*</sup> Supported by the State Committee for Scientific Research, Grant No. 3P06T 046 24

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The aim of the experiment was to investigate the nutritional and physiological effects of separate and combined supplementation of a rat diet with grapefruit flavonoids and inulin.

#### MATERIAL AND METHODS

Commercial preparations of grapefruit flavonoids from Cintamani-Poland and Frutafil-Inulin Tex (ORAFTI) were used. Flavonoid extract (FL, 0.3%) and inulin (IN, 5 and 10%) were added to a standard casein diet (control) as single (diet FL, IN5 and IN10) or combined (diet FL-IN5 and FL-IN10) additives.

The experiment was performed for 28 days on 48 male Wistar rats of initial body weight 123.8 g . Rats were housed in individual cages and fed *ad libitum*. Feed intake and body weight were recorded. During the last 5 days faeces and urine were collected, N retention was calculated as % of N intake and apparent biological value (BV) as % of N absorbed. At the end of the experiment rats were anaesthetized with urethane and blood was sampled. Total (TC) and HDL cholesterol, triacylglycerols (TAG), glucose, magnesium, calcium and phosphorus concentrations in serum were determined using Alpha Diagnostics sets. Results were subjected to one-way variance analysis, significance of differences was determined by Duncan's multiple range test at the significance level P<0.05.

#### RESULTS

A tendency towards lower feed intake and body gain on all diets supplemented with FL and IN, fed singly and in combination, was observed (Table 1) but feed intake was significantly affected only by feeding the FL and FL-IN10 diet, and body gain, by the FL-IN10 diet. Feed/body weight gain was not affected. Protein digestibility did not differ among the groups, while N retention % tended to be higher in all supplemented groups, the difference between IN10 and control being significant. A similar tendency was observed in

	Diet							
	control	FL	$IN_5$	IN <sub>10</sub>	FL-IN <sub>5</sub>	FL-IN <sub>10</sub>	Pooled SEM	
Diet intake, g	439a	401bc	419ab	405abc	412abc	381°	5.00	
Body weight gain, g	$150.4^{a}$	139.3ab	$147.7^{a}$	$141.1^{ab}$	145.8a	$128.7^{b}$	2.01	
Feed/body gain, g/g	2.92	2.88	2.84	2.87	2.83	2.96	0.02	
Protein digestibility, %	93.4	93.0	91.8	93.0	92.8	92.9	0.26	
N retention, %	59.1 <sup>b</sup>	$60.6^{ab}$	61.5ab	65.2a	$61.4^{ab}$	63.1ab	0.35	
BV, %	63.5°	$65.2^{abc}$	$66.6^{abc}$	69.95ª	$66.0^{\mathrm{abc}}$	$67.7^{ab}$	0.39	

Table 1. Growth performance and protein utilization

the utilization of nitrogen absorbed, which was significantly higher in groups IN10 and FL-IN10 than in control rats.

Total cholesterol was significantly decreased only in animals fed on the FL-IN10 diet as compared with controls, but it was significantly lower in all groups fed on diets containing inulin (singly or in combination) than on the diet containing flavonoids (FL diet) (Table 2). Neither HDL cholesterol, TAG nor glucose were affected by the diet. Among minerals, only the Ca concentration was higher on diet IN10 than on IN5 and FL-IN5; P and Mg concentrations were uniform.

	Diet							
	control	FL	$IN_5$	$IN_{10}$	FL-IN <sub>5</sub>	FL-IN <sub>10</sub>	SEM	
TC, mg/dL	125.0ab	136.4a	110.6 <sup>bc</sup>	112.6bc	110.5 <sup>bc</sup>	99.4°	3.27	
HDL-C, mg/dL	43.2	40.9	43.7	49.4	40.9	44.2	1.13	
TAG, mg/dL	279	233	239	232	223	243	9.28	
Glucose, mg/dL	241	238	239	230	252	238	7.50	
Mg, mg/dL	2.87	3.13	3.06	3.12	2.92	3.07	0.06	
Ca, mg/dL	9.81ab	$9.83^{ab}$	$9.66^{b}$	10.43a	$9.57^{b}$	$9.97^{ab}$	0.09	
P, mg/dL	4.97	5.08	5.20	5.27	5.07	5.17	0.15	

Table 2. Biochemical indices of rat serum

#### DISCUSSION

In our study, grapefruit flavonoids decreased feed intake and tended to decrease body weight gain. In the experiment of Kuo et al. (2005) feeding tea polyphenols had no effect on diet intake but depressed growth rate. Inulin fed singly also tended to deteriorate growth performance which is in agreement with the results of Houdijk et al. (1998) but in contradiction with other findings. The worst results were found when animals were fed flavonoids and 10% inulin in combination. The similar efficiency of feed utilization in all groups may indicate that the nutritional value of diets was not affected by flavonoids or inulin fed either singly or in combination. This conclusion is confirmed by the absence of negative effects on protein digestibility and by a tendency towards higher protein retention. This is not in agreement with the results of Shahkhialili et al. (1990) who found that feeding polyphenols depressed protein digestibility and utilization. These discrepancies may be attributed to differences in the structure of the polyphenols used in different experiments.

Grapefruit flavonoids fed alone did not induce hypolipidaemic and hypocholesterolaemic effects, probably because of a too low dietary level, while such a tendency observed in animals fed on inulin is in agreement with the findings of Delzenne et al. (2002). The effect of inulin was more pronounced when it was fed with flavonoids.

The similar serum concentrations of minerals despite slightly lower intake of diets containing inulin seem to confirm the suggestion that inulin stimulates absorption of minerals from the gastrointestinal tract (Ohta et al., 1998).

#### CONCLUSIONS

Responses of rats to dietary flavonoids and inulin occurred to some extent when these additives were fed either singly or in combination. Flavonoids fed singly negatively affected feed intake, while fed with inulin (10%), decreased feed intake and weight gain. Inulin fed singly tended to decrease the serum cholesterol concentration, while fed with flavonoids, decreased TC considerably.

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#### STRESZCZENIE

## Reakcje szczurów na flawonoidy grejpfruta i inulinę dodawane do diety pojedynczo lub łącznie

Szczury o początkowej masie ciała 124 g żywiono przez 28 dni dietą kazeinową bez dodatków (kontrolna) i dietami uzupełnionymi ekstraktem flawonoidów grejpfruta (FL) w ilości 0,3% oraz inuliną (IN) w ilości 5 i 10%, dodawanymi osobno lub razem. Rejestrowano spożycie paszy i masę ciała zwierząt oraz oznaczono strawność białka i retencję azotu. Po zakończeniu doświadczenia pobrano krew i oznaczono zawartość cholesterolu (TC), trójglicerydów(TAG), glukozy, Mg, Ca i P. FL podawane osobno lub z 10% IN obniżały spożycie paszy, a FL+IN10 także przyrosty masy ciała. Żaden z dodatków nie wpłynął na strawność białka, natomiast IN sama oraz w połączeniu z FL powodowała poprawę wykorzystania N strawionego. Poziom cholesterolu nie zmienił się pod wpływem FL, obniżył nieistotnie pod wpływem IN, a istotnie pod wpływem FL+IN10. Obydwa dodatki w sposób nieistotny powodowały obniżenie TAG.