

# **EFFECT OF HIGH SUCROSE DIETS ON CARCASS COMPOSITION IN CONVENTIONAL AND GERM-FREE MICE**

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## **SUMMARY**

1. The effect of dietary sucrose on carcass composition was determined in conventional (CV) and germ-free (GF) mice fed on a control diet and on diets containing 20 and 40% sucrose.

2. Germ-free mice fed the control diet were heavier than their conventional counterparts. CV mice fed on sucrose diets were, however, heavier than their GF counterparts.

3. GF mice fed on high-sucrose diets showed a lower lipid content on carcass than the GF controls and their CV counterparts.

4. CV mice fed the high-sucrose diets were heavier than the conventional animals fed on the control diet.

5. The water content in the carcass of GF mice fed on high sucrose diets was higher than that of controls, as well as their CV counterparts.

6. The present results suggest different lipidogenic effect of dietary sucrose in germ-free and conventional mice.

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## INTRODUCTION

Germ-free (GF) animals are being widely used in studies of specific biomedical problems. Under GF conditions there are important qualitative and quantitative differences regarding animal anatomy, physiology, and metabolism. Utilization of certain nutrients is enhanced in GF animals, decreasing their requirements when compared with those of the conventional (CV) ones. In theory, the GF animals possess a whole set of physiological characteristics which could allow a better digestive utilization of the diet. Even though GF animals have a lower total absorptive gut epithelium than the CV ones (1), the absorption efficiency of the GF epithelium is usually greater than that of CV animals for xylose (2), glucose (3), vitamins (3), minerals (4), fatty acids (5), and amino acids (6). Nevertheless, carefully conducted experiments with rats revealed that the fecal dry matter in GF animals is about double that of their CV counterparts, even though the intake of the former was only 18% higher. As a result, both GF and CV adult rats retained approximately the same energy from ingested food, amounting to 71% of the intake for the CV animal (7). On the other hand, the smaller cardiac output and the lower resting oxygen consumption in GF rats (7) suggest a lower metabolic rate in GF animals.

This work deals with the influence of high sucrose diets on energy metabolism in GF mice as far as lipid production from sucrose is concerned.

## MATERIAL AND METHODS

### *Animals*

The germ-free CFW (LOB) mice used in this experiment were the progeny of a breeding nucleus kindly supplied by Dr. Morris Pollard, LOBUND Laboratory, University of Notre Dame, USA. The animals were shipped in a special sterile container provided with a wet diet in order to minimize hunger and thirst during transportation. The GF animals were maintained in flexible plastic Trexler-type isolators (Standard Safety Co, Palatine, USA) and handled according to established procedures (8). The conventional CFW (LOB) mice were derived from the germ-free mice that had been removed from the isolators and maintained in an open room for several generations. Control of the GF status was performed by collecting samples of feces from GF animals which were inoculated in fluid thioglycollate medium (Difco) and brain heart infusion (Difco) at 37°C and 25°C, as described by Wagner (9). Conventional animals were maintained in the animal room. A photoperiod of 12 hrs d<sup>-1</sup> was used for CV and GF animals.

### *Diets*

Three types of diets were used for both CV and GF animals: control, 20% of sucrose, and 40% of sucrose (Table 1). The powdered mixture was made to a paste by the addition of water and sterilized in "square pak" flasks (American Sterilizer Co, Erie, USA) using a hospital-type autoclave as

**TABLE 1**  
**COMPOSITION OF THE DIETS**

Ingredients (%)	Control diet	Sucrose diets	
		20%	40%
Casein <sup>a</sup>	15.0	16.5	18.5
Sucrose	-	20.0	40.0
Corn meal <sup>b</sup>	69.4	47.6	25.0
Soy bean oil	7.0	7.2	7.9
Lysine	0.2	0.2	0.2
Choline chloride	0.4	0.4	0.4
Vitamin mixture <sup>c</sup>	3.0	3.0	3.0
Mineral mixture <sup>c</sup>	5.0	5.0	5.0
<b>Protein content</b>	<b>16.96</b>	<b>16.78</b>	<b>16.98</b>
<b>Lipid content</b>	<b>9.26</b>	<b>8.97</b>	<b>9.17</b>
<b>Carbohydrate content</b>	<b>54.96</b>	<b>58.59</b>	<b>61.66</b>
<b>Kcal/100 g</b>	<b>371</b>	<b>382</b>	<b>397</b>

a Commercial product containing: protein, 83% and fat, 3.5%.

b Commercial product containing: protein, 6.5%; oil, 2.5%; carbohydrate, 75%; fiber, 0.5% and water, 12.5.

c According to AOAC (1980).

described previously (10). All diets were equally processed and kept sterilized in the "square pak" flask until used to feed CV and GF mice.

### *Experimental Design*

Three groups of CV mice and three groups of GF mice were used in the experiment and fed on the diets described above. In each group, seven to eight 21-day old mice were used with an approximately equal number of males and females. The initial weights varied from 8.0 to 12.5 g. The mice were housed in individual cages. After 19 weeks, each animal was sacrificed under chloroform anesthesia, the whole right hind leg was amputated, and the viscera were removed. Each carcass was weighed and dried for 24 hr at 105°C to constant weight, to obtain water content, and extracted with petroleum ether in a Soxhlet apparatus to determine the fat content (11). The right hind leg was used for nitrogen determination by the Kjeldahl method as recommended by Lachance and Miller (12) as an index of body protein content. Crude protein was expressed as total nitrogen multiplied by 6.25 (11). The differences between the means were analyzed by the Student's "t" test.

TABLE 2

INFLUENCE OF SUCROSE LEVELS IN THE DIET ON FINAL BODY WEIGHT (FBW), BODY WEIGHT GAIN (BWG) AND CARCASS COMPOSITION OF CONVENTIONAL (CV) AND GERM-FREE (GF) MICE (NUMBER OF ANIMALS IN PARENTHESIS)

Group	FBW g ± SEM	BWG g ± SEM	Carcass			
			Weight g ± SEM	Moisture % ± SEM	Lipid % ± SEM	Protein % ± SEM
<i>Control</i>						
CV (7)	22.8 ± 1.1	13.1 ± 1.0	18.5 ± 0.9	57.1 ± 0.9	16.2 ± 1.4	22.1 ± 0.8
GF (8)	28.2 ± 1.3 <sup>a</sup>	18.1 ± 1.3 <sup>a</sup>	23.2 ± 1.2 <sup>a</sup>	57.4 ± 2.3	20.7 ± 2.2	21.8 ± 0.2
<i>20% sucrose</i>						
CV (8)	31.0 ± 1.8 <sup>b</sup>	19.8 ± 1.6 <sup>b</sup>	26.1 ± 1.7 <sup>b</sup>	48.0 ± 1.5 <sup>b</sup>	30.0 ± 2.2 <sup>b</sup>	22.0 ± 0.7
GF (7)	25.5 ± 1.5 <sup>a</sup>	14.4 ± 1.1 <sup>a</sup>	21.0 ± 1.3 <sup>a</sup>	64.5 ± 1.5 <sup>ab</sup>	13.1 ± 1.6 <sup>ab</sup>	25.1 ± 1.9
<i>40% sucrose</i>						
CV (7)	32.9 ± 2.3 <sup>b</sup>	20.6 ± 2.4 <sup>b</sup>	28.0 ± 1.6 <sup>b</sup>	50.1 ± 2.3 <sup>b</sup>	28.8 ± 2.4 <sup>b</sup>	19.8 ± 0.2
GF (7)	27.6 ± 2.1 <sup>a</sup>	17.1 ± 1.7	22.3 ± 1.8 <sup>a</sup>	65.2 ± 2.0 <sup>ab</sup>	12.7 ± 2.8 <sup>ab</sup>	24.2 ± 1.1

a Statistical difference ( $p < 0.05$ ) between CV and GF in the same group.

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## RESULTS AND DISCUSSION

Table 2 shows that, in GF mice fed the control diet, final body weight, body weight gain, and carcass weight were significantly higher than in the CV mice. These results are conflictant with those of Levenson and Tennant (13) who reported that carcass fat content was higher in CV than in GF rats. The results in Table 2 suggest that GF mice utilized better the control diet which made them heavier than CV mice, although no alteration in the proportion of the carcass constituents was observed. The CV mice fed on sucrose diets were, however, heavier than their GF counterparts and the CV mice fed on the control diet. Final body weight, body weight gain, and carcass weight were statistically similar in GF mice fed either on the control diet or on the sucrose diets.

Table 2 also indicates that sucrose content of diet had a greater lipogenic effect for CV mice than for their GF counterparts. Fat content was significantly lower in the carcass of the GF animals fed on the high sucrose diets. It should be pointed out that there was a significant negative correlation between moisture content and fat content in the carcass of both CV and GF mice fed on the high sucrose diets. In GF animals the apparently, but not statistically different, higher protein content or perhaps carbohydrate content (not measured) in the carcass, could be responsible for the higher water content. Previous studies showed that in GF rats, the activities of liver glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase were 60% and 80%, respectively, of those of their CV counterparts (14). In addition to this reduction in the activity of rate-controlling enzymes of the hexosemonophosphate shunt, the activity of *alpha*-glycerophosphate deshydrogenase was also reduced in GF animals (15). These data suggested that production of NADPH might be decreased in GF rodents and that this might be reflected in a reduced potential of the liver to synthesize fatty acids from carbohydrate catabolism. In the absence of an outlet for acetyl CoA via fatty acid synthesis, an increase in the amount of citrate transferred to the cytoplasm can be expected. This excess citrate could fall in the circulation and then into the urine. In fact, Gustafsson and Norman (16) found five times more citrate in urine of GF rats than in their CV controls.

The present investigation designed to study the effect of high-sucrose diets on body weight, body weight gain, and carcass composition in CV and GF mice, indicated a greater lipogenic effect of the high-sucrose diets only in the CV animals. This discrepancy between CV and GF mice shows that GF animals offer an excellent tool in the studies of the influence of the intestinal microflora on carbohydrate metabolism.

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## RESUMO

1. O efeito da sacarose na dieta sobre a composição da carcassa foi determinado em camundongos convencionais (CV) e isentos de germes (GF) alimentados com ração controle e com rações contendo 20 e 40% de sacarose.

2. Os camundongos GF na ração controle foram mais pesados do que os CV. Contudo, os camundongos CV alimentados com dieta com sacarose foram mais pesados que os GF.

3. Camundongos isentos de germes (GF) alimentados com dieta rica em sacarose mostraram um conteúdo lipídico menor na carcassa em relação aos GF alimentados na ração controle e os outros CV.

4. Camundongos convencionais (CV) na dieta rica em sacarose foram mais pesados do que os animais CV na dieta controle.

5. O conteúdo aquoso na carcassa dos camundongos GF na dieta rica em sacarose foi maior do que nos controles GF e nos CV.

6. Esses resultados sugerem um efeito lipídico diferente da sacarose alimentar nos camundongos GF e CV.

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