

## 乳癌高発系および低発系マウス(SHN,SLN)におけるレプチンの血中レベルと,体重,臓器重量との関係

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# Serum Leptin Level and Body and Organ Weights in a High and a Low Mammary Tumor Strains of SHN and SLN Mice

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## Summary

Serum level of leptin, a product of the *obese* gene, was determined by EIA test in the 3 month-old female and male SHN and SLN, a high and a low mammary tumor strains of mice, respectively. There was little difference between strains in the female body weight, but SLN was significantly higher than SHN in the male body weight. The absolute weight of the female spleen was significantly higher in SHN than in SLN and it was also the case in the relative liver weight and both the absolute and the relative kidney weights of the male. SHN was lower than SLN in the serum leptin level in either the female or the male and the female was higher than the male in the level of both strains. The correlations between leptin level and body weight were higher in the female than in the male of both strains and the SHN female was further higher than SLN female. The significantly negative correlations were found between the leptin level and the heart weight in the female and the male SHN. The significance of these findings were discussed in relation to mammary tumor risk.

## Introduction

Leptin is a protein product of the *obese* gene, which is secreted from adipocytes and plays an important role in the regulation of body weight and metabolism<sup>1)-4)</sup>. Plasma leptin level is related to the adiposity and is regulated by feeding and fasting<sup>5)-7)</sup>. It is generally considered that obesity is a key factor of several adult diseases in including breast cancer<sup>8)-12)</sup>. Thus, we investigated serum leptin level in a high and a low mammary tumor strains of female and male mice and the correlations between leptin level and body and organ weights.

## Materials and Methods

*Animals and treatments.* A high and a low mammary tumor strains of female and male SHN/Mei and SLN/Mei virgin mice<sup>13),14)</sup> maintained in our laboratory by strict brother × sister mating were used at F86 and F73, respectively. The animals were kept in aluminum cages (18 × 30 × 15 cm) with wood shavings (M size: CLEA JAPAN, Tokyo, Japan), maintained in a windowless animal room, which was air-conditioned (22–24°C and 60–70% relative humidity) and artificially illuminated (14 hours of light from 0500 to 1900 h), and provided with a commercial diet (Lab MR Breeder: Nihon Nosan Kogyo KK, Yokohama, Japan) and tap water *ad libitum*.

All mice were killed on the morning at 3 months of age under the light ether anesthesia. The blood was collected from the trunk, left at room temperature for 6 hours, stored in the refrigera-

tor overnight and centrifuged at  $1,000 \times g$  for 20 min at  $4^{\circ}\text{C}$ , and serum was stored at  $-70^{\circ}\text{C}$  for assay of leptin.

*Organ weights.* At autopsy, heart, liver, spleen and kidneys were immediately removed and weighed.

*Serum leptin level.* The level was determined by the sandwich enzyme immunoassay method (EIA kit; Immuno-Biological Laboratories Co., Ltd, Gunma, Japan).

*Statistics.* The significance of difference in the correlation coefficients and that in each parameter among groups were evaluated by Fisher's z-test and Duncan's Multiple range test, respectively. Probability values less than 5% were considered significant.

## Results

*Body weight and organ weights* (Table I). There was little difference between strains in the female body weight, but SLN was significantly higher than SHN in the male body weight.

The absolute weight of the female spleen was significantly higher in SHN than in SLN and it was also the case in the relative liver weight and both the absolute and the relative kidney weights of the male.

*Serum leptin Level* (Figure 1). SHN was lower than SLN in the serum leptin level in either the female or the male and the female was higher than the male in the level of both strains, while the differences were not always statistically significant due to the large variations of the values.

**Table I** Body and organ weights in each strain (Mean  $\pm$  SEM).

	Female			Male		
	SHN (5)	SLN (5)	p	SHN (4)	SLN (5)	p
Body weight (g)	26.9 $\pm$ 0.9	27.9 $\pm$ 0.7		30.9 $\pm$ 0.7	35.6 $\pm$ 1.0	<0.05
Organ weight*						
Heart	115 $\pm$ 4	116 $\pm$ 9		147 $\pm$ 5	156 $\pm$ 11	
	129 $\pm$ 8	125 $\pm$ 8		143 $\pm$ 5	132 $\pm$ 9	
Liver	1251 $\pm$ 98	1125 $\pm$ 104		1248 $\pm$ 23	1231 $\pm$ 40	
	1390 $\pm$ 82	1214 $\pm$ 114		1213 $\pm$ 48	1039 $\pm$ 19	<0.05
Spleen	78.2 $\pm$ 8.0	62.5 $\pm$ 2.8	<0.05	65.3 $\pm$ 2.6	66.4 $\pm$ 4.2	
	87.1 $\pm$ 8.2	67.5 $\pm$ 4.2		63.6 $\pm$ 3.8	55.8 $\pm$ 2.4	
Kidneys	282 $\pm$ 13	284 $\pm$ 9		475 $\pm$ 14	407 $\pm$ 8	<0.05
	316 $\pm$ 15	307 $\pm$ 11		461 $\pm$ 12	344 $\pm$ 11	<0.01

Number of estimates is in the parentheses.

\* The top and the bottom rows are for the absolute (mg) and the relative (mg/30 g body weight) weights, respectively.

Serum Leptin Level in a High and a Low Mammary Tumor Strains of Mice

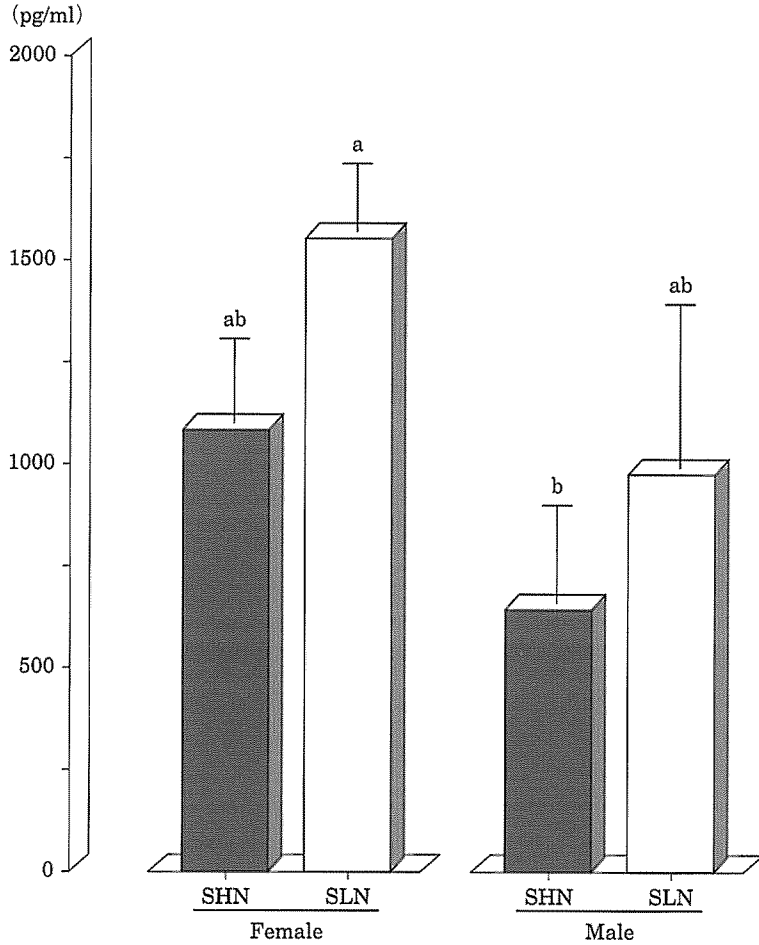


Fig. 1 Serum leptin level in each strain (Mean  $\pm$  SEM). <sup>a,b</sup>Values with different superscripts are different at  $P < 0.05$  or  $0.01$ . The number of estimates is 4 in SHN male and 5 in all others.

Table II Correlation coefficients\* between serum leptin level and organ weights in each strain.

	Female			Male		
	SHN (5)	SLN (5)	Pooled (10)	SHN (4)	SLN (5)	Pooled (9)
Heart	-0.609	0.500	0.084	-0.747	0.149	0.086
	-0.933 <sup>a</sup>	0.335	-0.378	-0.995 <sup>b</sup>	0.175	-0.114
Liver	0.633	0.100	0.152	-0.484	-0.203	-0.270
	0.334	-0.075	-0.129	-0.516	-0.319	-0.383
Spleen	0.254	-0.509	-0.235	-0.677	-0.064	-0.147
	-0.059	-0.649	-0.472	-0.640	-0.075	-0.303
Kidneys	0.131	-0.072	0.075	0.780	-0.817	-0.299
	-0.558	-0.549	-0.552	0.465	-0.485	-0.338

Number of estimates is in the parentheses.

\* The top and the bottom rows are for the coefficients with the absolute (mg) and the relative (mg/30 g body weight) weights, respectively.

<sup>a</sup> or <sup>b</sup> significant at  $p < 0.05$  or  $0.01$ .

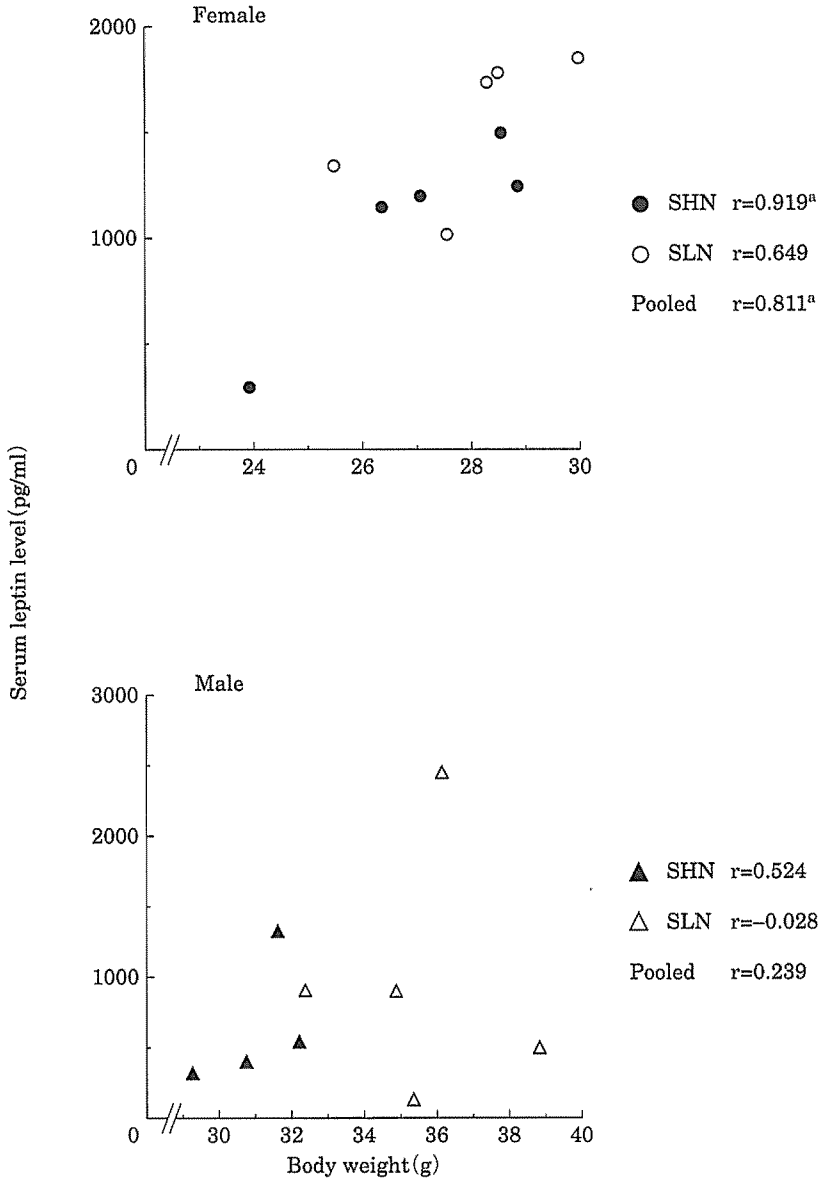


Fig. 2 Correlation coefficients between serum leptin level and body weight in each strain. <sup>a</sup>Significant at P<0.05.

*Correlation coefficients between serum leptin level and body and organ weights.* The correlations between the serum leptin level and the body weight were higher in the female than in the male in each strain, and further higher in SHN female than in SLN female (Figure 2).

The significantly negative correlations between the serum leptin level and relative heart weight were observed in both sexes of SHN strain (Table II).

## Discussion

In this study, the correlations between the body weight and the serum level of leptin, a product of the *obese* gene, were higher in the female than in the male in each strain, and further higher in SHN female than in SLN female. This would support the epidemiological hypotheses that the body weight, which largely reflects obesity, is related to the risk of human breast cancer, whereas the causality between them is still controversial<sup>15)</sup>.

In both the female and the male, SLN was lower and higher than SHN in some organ weights and the serum leptin levels, respectively. These are mostly due to the fact that the female and male SLN become obese after 5–6 months of age<sup>13)</sup>. The higher leptin level in the female than in the male in both strains may also reflect that the former is generally more obese than the latter.

The significantly negative correlations between leptin level and the relative heart weight were obtained in both sexes of SHN strain. The significance of the results is unclear, however, a possibility would be plausible; if the heart weight is an index of its function, the heavier the weight, the more active the blood circulation or the metabolism and this would result in the reduction of the tumor risk.

Finally, serum leptin level may provide valuable information on obesity, which would contribute to the study of several diseases.

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### 乳癌高発系および低発系マウス (SHN, SLN) におけるレプチンの 血中レベルと、体重、臓器重量との関係

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#### 要 約

3ヶ月齢の乳癌高発および低発系 (SHN および SLN) 雌雄マウスにおける、肥満遺伝子産生物質レプチンの血中レベルを EIA 法で測定し、体重および臓器重量との相関を求めた。雌雄とも SLN では SHN よりレプチンレベルは高く、いくつかの臓器重量は低かった。レプチンレベルと体重の間の相関 (正) は両系統とも雌が雄より高く、とくに SHN の雌で高かった。SHN では雌雄ともレプチンレベルと体重 30 g 当たり心臓重量との間に負の相関が認められた。