Original Research Paper

Comparison of Compertz and Logistic Models in Estimating the Growth of Leptin-Deficient (*ob/ob*) Mice and Wild-Type Mice Fed an In-House Prepared High-Fat Diet

[1,2]Ah Reum Son, [1,2]Hyunwoong Jo, [1]Kyu Ree Park and [1,2]Beob Gyun Kim

[1]*Department of Animal Science and Technology, Konkuk University, Seoul, Republic of Korea*
[2]*Monogastric Animal Feed Research Institute, Konkuk University, Seoul, Republic of Korea*

Abstract: This experiment was conducted to growth curve models for leptin-deficient (*ob/ob*) and wild-type mice fed a meal-form High-Fat (HF) diet prepared in-house for over 120 weeks. Two hundred and twenty-four sets of Body Weight (BW) and age data were collected from *ob/ob* mice and 485 sets of data were collected from wild-type mice. All animals had free access to the HF diet (34.3% energy from fat) and water. Individual BW and survival rates of mice were measured. To develop growth curves of *ob/ob* mice and wild-type mice fed the HF diet, Gompertz and Logistic growth models were employed. The survival rates of *ob/ob* mice fed the HF diet were lower than those of wild-type mice. Models for estimating growth of *ob/ob* mice fed the HF diet were: BW, $g = 61.36 \times e^{-2.56 \times e^{-0.13t}}$ in Gompertz model (Akaike's Information Criterion, AIC = 1,694); and BW, $g = 60.65 \times (1 + 7.04 \times e^{-0.18t})^{-1}$ in Logistic model (AIC = 1,719). Growth models for wild-type mice fed the HF diet were: BW, $g = 28.94 \times e^{-1.75 \times e^{-0.13t}}$ in Gompertz model (AIC = 3,171); and BW, $g = 28.23 \times (1 + 4.85 \times e^{-0.25t})^{-1}$ in Logistic model (AIC = 3,198). In conclusion, Gompertz models may be more appropriate to estimate the growth of *ob/ob* mouse fed the diet with high-fat concentration.

Keywords: Gompertz Model, Growth Curve, High-Fat Diet, Logistic Model *ob/ob* mice

Introduction

Many obese models used in obesity or obesity-related disease research are gene-specific knockout or transgenic rodents and the obese model animals are often fed High-Fat (HF) diets (Woods *et al*., 2003; Buettner *et al*., 2007). Commercial pellet-type HF diets have been widely used to induce and maintain obesity in mouse models. In many nutrition experiments for testing dietary supplements, on the other hand, meal-form diets are preferred due to the convenience in preparing experimental diets (Niu *et al*., 2017; Elkahoui *et al*., 2018). However, meal-form HF diets for rodents are rarely available.

As elderly population increases, research on obese elderly men and women becomes more and more important (Vioque *et al*., 2007; Sardeli *et al*., 2018). Although young obese mice mostly less than 20 weeks of age are often used in nutrition research, older mice are more appropriate for mimicking adult or elderly human obesity and thus, have been recently used in elderly obesity research (Hunsche *et al*., 2016; Frasca *et al*., 2017). However, information on the survival rate and Body Weight (BW) changes of leptin-deficient (*ob/ob*) mice fed a HF diet for over 6 months is very limited. Therefore, the objectives of the present work were to test a novel meal-form HF diet fed to mice and to develop growth curve models for *ob/ob* and wild-type mice fed a meal-form HF diet for over 120 weeks.

Materials and Methods

Animals and Experimental Diet

Two hundred and twenty-four sets of BW and age data were collected from *ob/ob* mice and 485 sets of data were collected from wild-type mice. Body weight and age ranged from 6.3 to 77.7 g and from 3 to 127 weeks, respectively.

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A HF diet (34.3% energy from fat) was prepared in-house to contain 49% ground corn, 30% soybean meal (crude protein 48%), 20% fat powder and 1% vitamin-mineral premix (Table 1). The fat powder is a protein-coated fat product that was mainly composed of 80% lard, whey and casein. All animals had free access to diet and water. Individual BW of mice was measured with approximately two-week intervals.

Growth Models and Statistical Analysis

For estimating the growth curves for *ob/ob* and wild-type mice, the NLMIXED procedure of SAS (SAS Inst. Inc., Cary, NC, USA) with nonlinear regression was used. The model equation used for the Gompertz model (Gompertz, 1825) is:

$$Wt\ (g) = Ae^{-be^{\wedge}-kt}$$

The model equation used the Logistic model (Robertson, 1908) is:

$$Wt(g) = A(1 + be^{-kt}) - 1$$

For both equations, 3 parameters in the equation were employed to estimate the growth model parameters in which *Wt* is the BW of mice at time *t* (week), *A* is the mature weight, *b* is the growth ratio, *k* is the maturing rate; and *e* is the natural logarithm. Based on the estimated 3 parameters, the age at point of inflection, gain at inflection and weight at inflection were calculated.

In the Gompertz model, equations for the age at point of inflection (t_i), body weight at inflection (Wt_i) and gain at inflection ($\delta Wt/\delta t$) are:

$$t_i = (\log_e b)/k$$
$$Wt_i = A/e$$
$$dy_i/dt_i = kbWt_i e^{-kt}$$

Table 1: Ingredients composition and calculated gross energy contents of high-fat diet

Item	High-fat diet
Ingredient, %	
Ground corn	49
Soybean meal (crude protein, 48%)	30
Fat powder (ether extract, 80%)	20
Vitamin-mineral premix[a]	1
Gross energy, kcal/kg	4,891

[a]Provided the following quantities per kg of complete diet: vitamin A, 50,000 IU; vitamin D$_3$, 8,000 IU; vitamin E, 100 U; vitamin K, 10.0 mg; thiamin, 9.8 mg; riboflavin, 20.0 mg; pyridoxine, 9.8 mg; vitamin B$_{12}$, 0.12 mg; pantothenic acid, 75.0 mg; folic acid, 2.20 mg; niacin, 124 mg; biotin, 0.12 mg; Cu, 50 mg as copper sulfate; Fe, 536 mg as iron sulfate; I, 10.0 mg as potassium iodate; Mn, 250 mg as manganese sulfate; Se, 0.76 mg as sodium selenite; Zn, 626 mg as zinc oxide; butylated hydroxytoluene, 100 mg

In the Logistic model, equations for the t_i, Wt_i and $\delta Wt / \delta t$ are:

$$t_i = (\log_e b)/k$$
$$Wt_i = A/2$$
$$dy_i/dt_i = Abke^{kti}/(e^{kti} + b)^2$$

The inflection point, second derivative of function, means where the slope of the growth curve becomes maximum.

Results

The *ob/ob* mice gained more weight compared with the wild-type mice, and a hair loss was found on the neck area of *ob/ob* mice (Fig. 1). The survival rate of the *ob/ob* mice was lower than that of the wild-type mice (Fig. 2).

The average mature BW of *ob/ob* mice estimated from the Gompertz and Logistic models was approximately 61.0 g (Table 2; Fig. 3 and 4). The average mature BW of wild-type mice estimated from the models was approximately 28.6 g. The age at point of inflection, BW at inflection and gain at inflection are presented in Table 3. The average BW at inflection of *ob/ob* and wild-type mice estimated from the models was approximately 26.5 and 12.4 g, respectively.

Discussion

It has been reported that the obesity of *ob/ob* mice can be visually identified after 4 weeks of age (Kennedy *et al*., 2010), and physical differences between the *ob/ob* and wild-type mice were also observed in the present study. Moreover, the wild-type mice had more sleek hairs and active movements than *ob/ob* mice in this study. This observation is supported by previous studies that reported critical role of leptin in hair cycle (Sumikawa *et al*., 2014; Tasaki *et al*., 2015; Sasaki *et al*., 2018). After depilation, elongation of telogen was observed in *ob/ob* mice compared with the wild-type mice (Tasaki *et al*., 2015). Moreover, the *ob/ob* mice and mice fed the high-fat diet suffered delayed wound closure (Seitz *et al*., 2010).

The lower survival rates of *ob/ob* mice compared with wild-type mice agreed with previous studies. Ren *et al*. (2010) reported reduced survival rates of the *ob/ob* mice compared with wild-type mice. In addition, the survival rate began to decline from 50 weeks of age *ob/ob* mice in the present study. This result agreed with Harrison and Archer (1987) reporting that the *ob/ob* mice showed high mortality after 52 weeks of age.
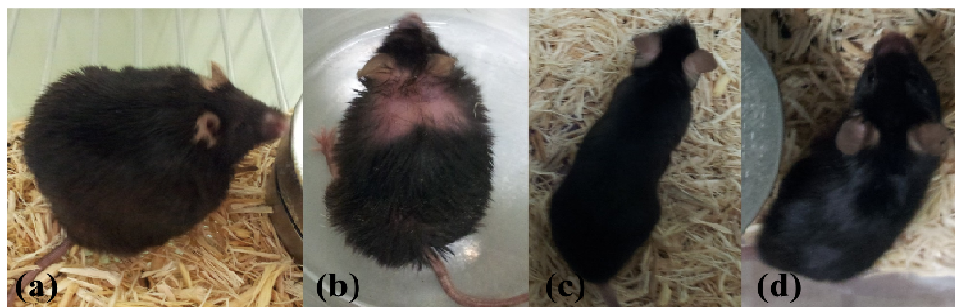
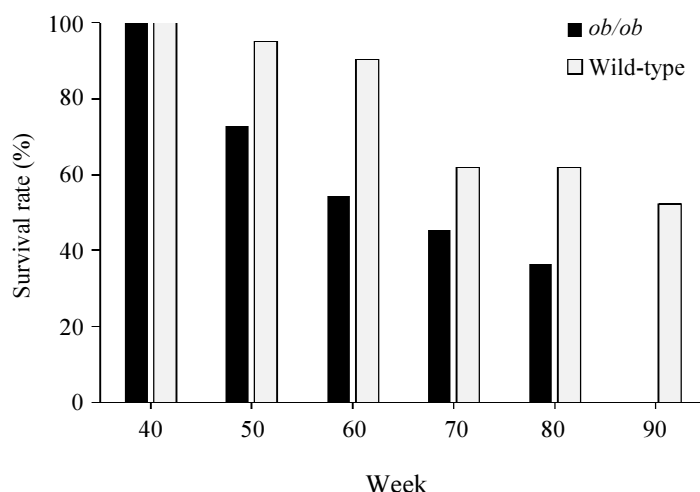**Fig. 1:** Leptin-deficient (*ob/ob*) mice (a and b) and wild-type mice (c and d) fed a high-fat diet

Fig. 2: The survival rates of *ob/ob* and wild-type mice fed a high-fat diet

Table 2: Estimates of growth curve parameters for leptin-deficient (*ob/ob*) and wild-type mice using Gompertz and Logistic models as a function of age (week)

Genotype	Growth model	Parameters[a]			
		$A\pm SE$	$b\pm SE$	$k\pm SE$	AIC[b]
ob/ob	Gompertz[c]	61.36±0.66	2.56±0.17	0.13±0.01	1,694
	Logistic[d]	60.65±0.65	7.04±0.88	0.18±0.01	1,719
Wild-type	Gompertz	28.94±0.36	1.75±0.15	0.13±0.02	3,171
	Logistic	28.23±0.33	4.85±1.31	0.25±0.06	3,198

[a]A = mature body weight; b = growth rate; k = maturing rate; SE= Standard Error
[b]Akaike's information criterion
[c]Gompertz model: body weight, $g = Ae^{-be^{-kt}}$
[d]Logistic model: body weight, $g = A(1+be^{-kt})^{-1}$

Table 3: Characteristics at inflection point on growth curves using Gompertz and Logistic models for leptin-deficient (*ob/ob*) and wild-type mice as a function of age

Genotype	Growth model	Age at point of inflection[a] (week)	Body weight at inflection[b] (g)	Gain at inflection[c] (g/week)
ob/ob	Gompertz	7.22	22.57	2.93
	Logistic	10.70	30.33	2.77
Wild-type	Gompertz	4.19	10.65	1.43
	Logistic	6.31	14.11	1.76

[a]Age at point of inflection= $(\log_e b)/k$
[b]Body weight at inflection for Gompertz model =A/e; body weight at inflection for Logistic model =$A/2$
[c]Gain at inflection for Gompertz model = $kbWt_i e^{-kt}$; gain at inflection for Logistic model = $Abke^{kti}/(e^{kti} + b)^2$
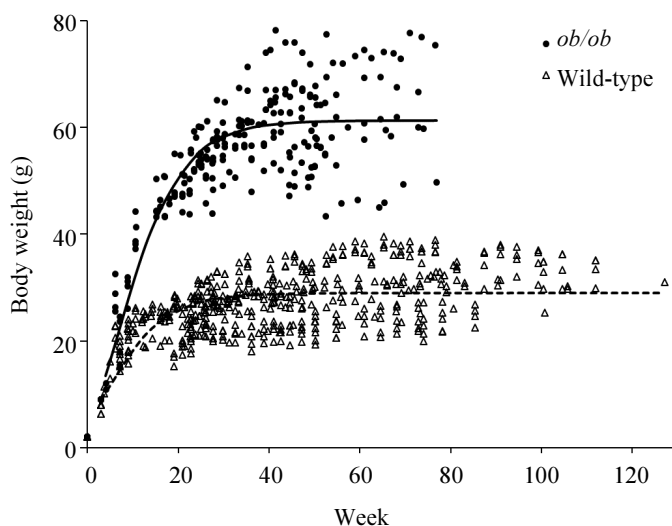
Fig. 3: Gompertz models of estimated Body Weight (BW) growth of mice fed a high-fat diet as a function of age: BW of leptin-deficient (*ob/ob*) mice, $g = 61.36 \times e^{-2.56 \times e^{-0.13t}}$ with Akaike's Information Criterion (AIC) = 1,694; BW of wild-type mice, $g = 28.94 \times e^{-1.75 \times e^{-0.13t}}$ with AIC = 3,171
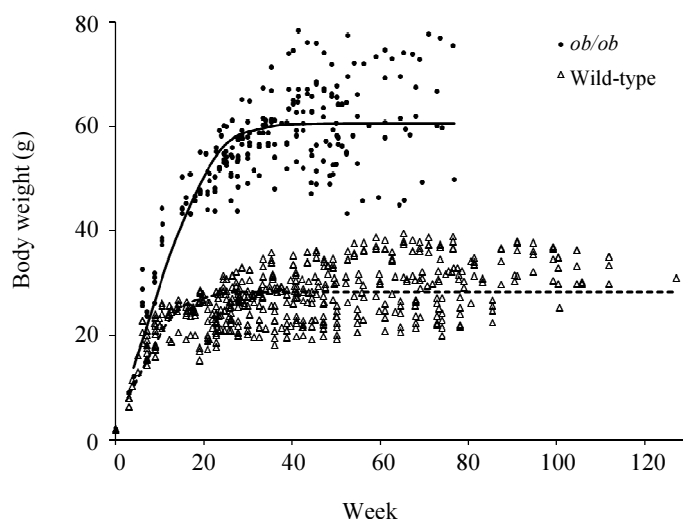

Fig. 4: Logistic models of estimated Body Weight (BW) growth of mice fed high-fat diets as a function of age: BW of leptin-deficient (*ob/ob*) mice, $g = 60.65 \times e^{-7.04 \times e^{-0.18t}}$ with Akaike's Information Criterion (AIC) = 1,717; BW of wild-type mice, $g = 28.23 \times e^{-4.85 \times e^{-0.25t}}$ with AIC = 3,198

The greater mature BW of *ob/ob* mice than wild-type mice was observed in the present study. In both modes, the growth rate of the *ob/ob* mice was also greater compared with that of the wild-type mice. While the *ob/ob* mice reach the maximum BW of 60 to 70 g at 28 to 32 weeks, the lean-type mice reach the maximum BW of 30 to 40 g at 12 to 16 weeks (Hedrich, 2012). Greater energy and nutrient intake may have contributed to the rapid growth rate of *ob/ob* mice used in this study. Murphy *et al*. (2010) reported that the *ob/ob* mice gained more weight compared with lean mice, resulted from

greater accumulative energy intake in the *ob/ob* mice. Moreover, a previous study reported that increases in weight gain and feed intake of the *ob/ob* mice fed HF diets containing corn oil or beef tallow were observed compared with the *ob/ob* mice fed a low-fat diet (Mercer and Trayhurn, 1987). In addition, the magnitude of increase was greater in the *ob/ob* mice than lean mice. Generally, the model with a lower value for Akaike's information criterion can be considered as a more appropriate model (Gbangboche *et al*., 2008). Based on the values for Akaike's information criterion, the

Gompertz model was more appropriate for estimating BW of *ob/ob* and wild-type mice.

In the present study, the age at point of inflection, BW at inflection and gain at inflection were greater in the *ob/ob* mice compared with the wild-type mice. The greater BW and rapid growth rate of *ob/ob* mice compared with wild-type mice have also been reported in the literature (Harris *et al.*, 1998; Sainsbury *et al.*, 2002; Sun *et al.*, 2006; Seitz *et al.*, 2010).

Conclusion

In conclusion, the Gompertz models are more appropriate for estimating the growth of mice fed a high-fat diet compared with Logistic models. The suggested growth curves and survival rate data for leptin-deficient mice fed a high-fat diet can be used for designing long-term experiments or planning to use elderly obesity models. Additionally, the high-fat diet formula for meal-form feeds provided in the present work is useful in nutrition experiments employing leptin-deficient mice. Further research is warranted to develop growth models for other model animals for obesity or nutrition research.

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Author's Contributions

Ah Reum Son: Conducted the animal experiment and drafted most of the manuscript.

Hyunwoong Jo: Performed statistical analysis and critically revised statistical part of the manuscript.

Kyu Ree Park: Assisted animal care and data recording and prepared figures and tables.

Beob Gyun Kim: Supervised the experimental work and manuscript preparation and revised the manuscript.

Ethics

The authors declared no ethical issues.

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