

Background and Purpose

Cynomolgus monkeys are commonly used in non-clinical toxicity studies because they are more anatomically and physiologically homologous to humans compared to other mammals such as rats. Toxicity studies involving monkeys provide valuable data for drug development regarding the potential effects of drugs on humans.

In such studies, weight loss is occasionally observed in monkeys and can be difficult to distinguish from drug-related effects.

This study retrospectively analyzed data from control-group monkeys without drug treatment to investigate the incidence of weight loss and its physiological and pathological characteristics. We also investigated potential improvements through enhanced animal welfare.

Materials and Methods

Animals

Animals in the control groups of 4-week and 13-week studies conducted at the test facility from 2010 to 2022 were retrospectively evaluated.

4-week studies: 46 studies (males: n=206, females: n=196)

13-week studies: 33 studies (males: n=146, females: n=136)

Weight change rate

Weight change rate (%) was calculated with the following formula.

(Final body weight – Initial body weight) × 100 / (Initial-body weight)

Other examinations

Clinical observation, Food consumption, Hematology (CA-510, XT-2000iV), Blood chemistry (TBA-2000FR, TBA-200FR), Organ weight, Necropsy, and Histopathology

Measures to improve animal welfare

Since September 2015, measures to improve animal welfare have been implemented at the test facility, including provision of enrichment items, increased treats, and full-scale group housing before the start of studies. We compared the weight change rate before and after the start of the enrichment program.

Statistical analysis

Weight change rates before and after the enrichment program were statistically analyzed. Variance homogeneity was examined using the F-test; when variances were homogeneous, a t-test was applied, and when heterogeneous, an Aspin–Welch t-test was applied.

Results

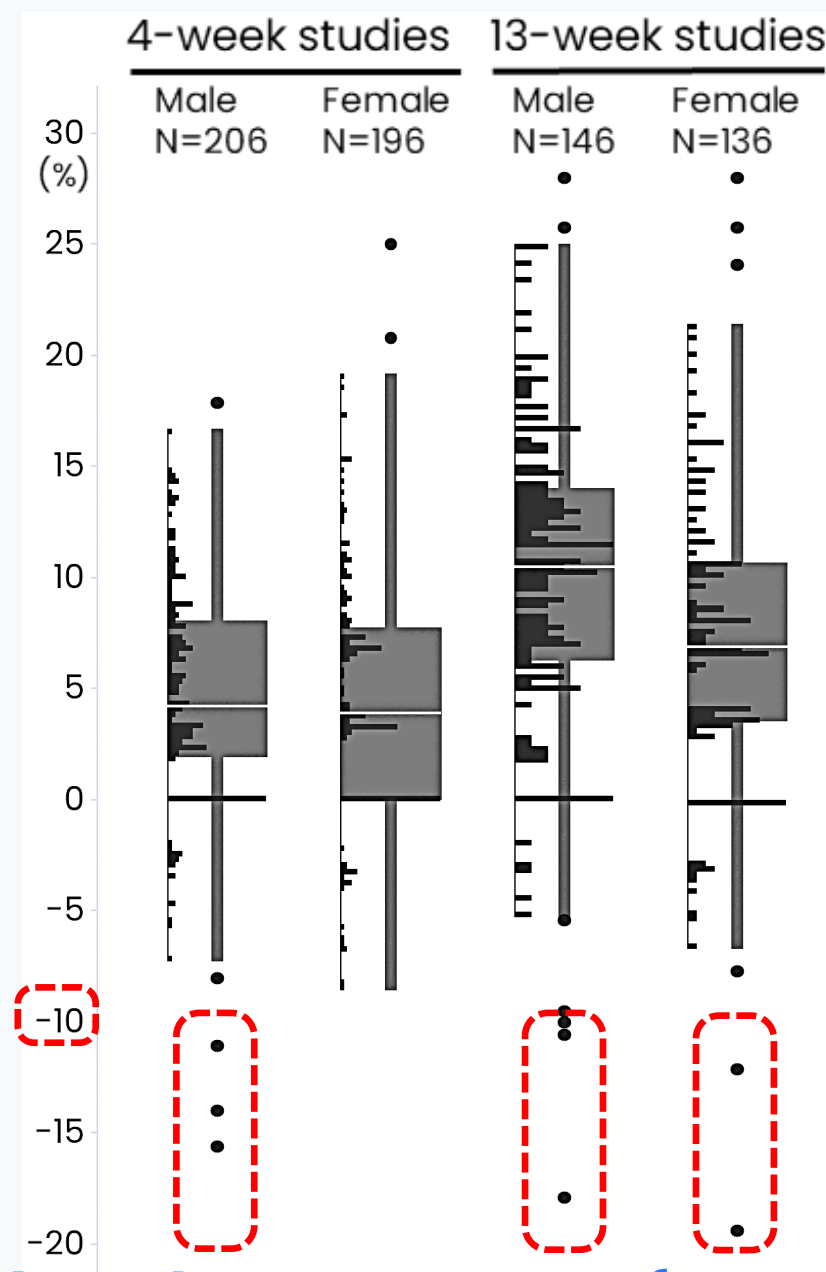


Fig. 1. Weight change rate from the start of the acclimation period to the end of the dosing period
Across the studies, 3 males (3/402 animals, 2/46 studies) in the 4-week studies and 3 males and 2 females (5/282 animals, 4/33 studies) in the 13-week studies showed a weight change rate below –10%.

Details are in Fig. 2, 3, and Table 1.

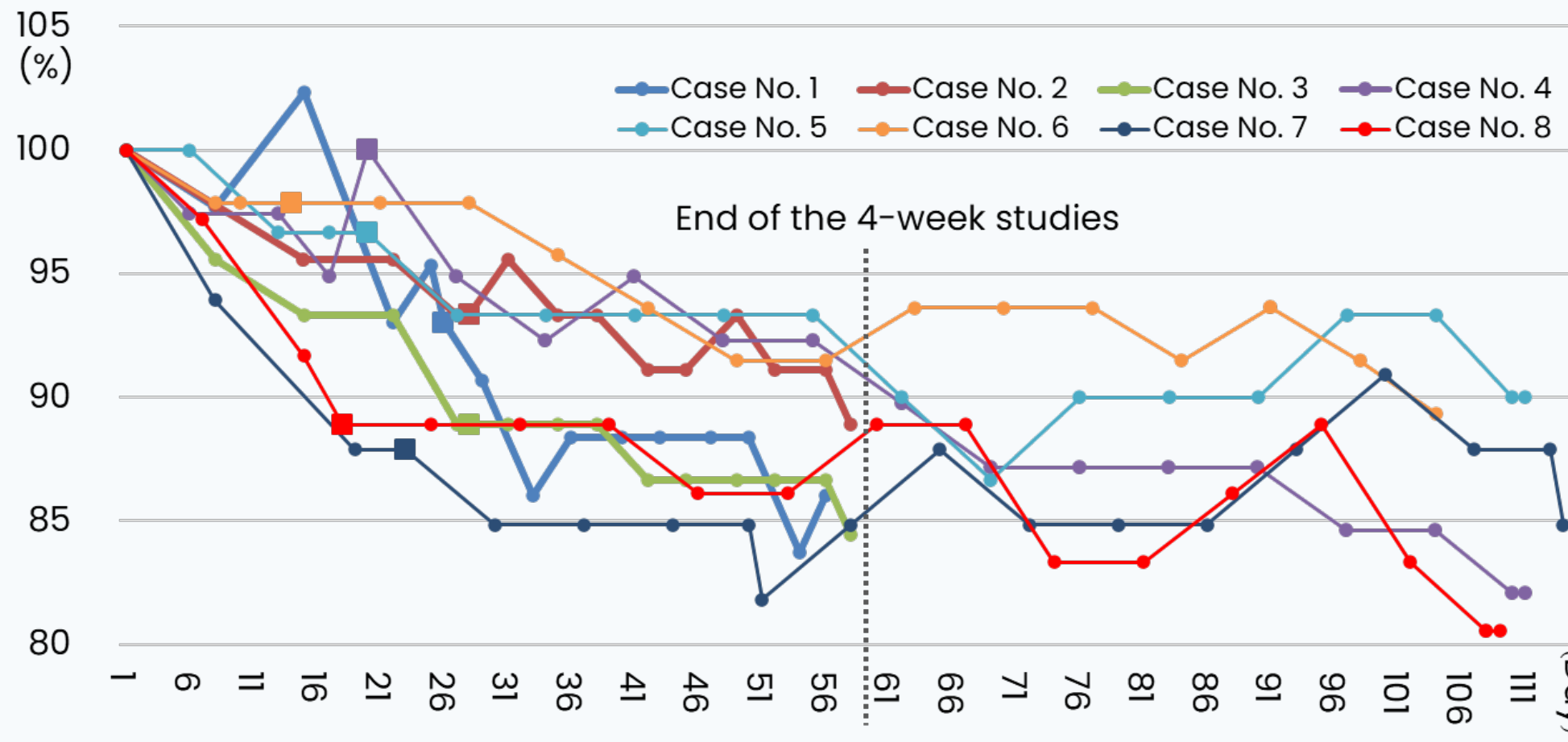


Fig. 2. Weight transition in animals with weight loss

■: Initial dosing day

Case Nos. 1–3: 4-week studies

Case Nos. 4–8: 13-week studies

Weight at the start of acclimation was set to 100%, and the horizontal axis indicates days from Day 1 of acclimation.

In both the 4- and 13-week studies, animals showed rapid weight loss up to the end of the 4-week period, after which weight changes plateaued or declined slowly in the 13-week studies.

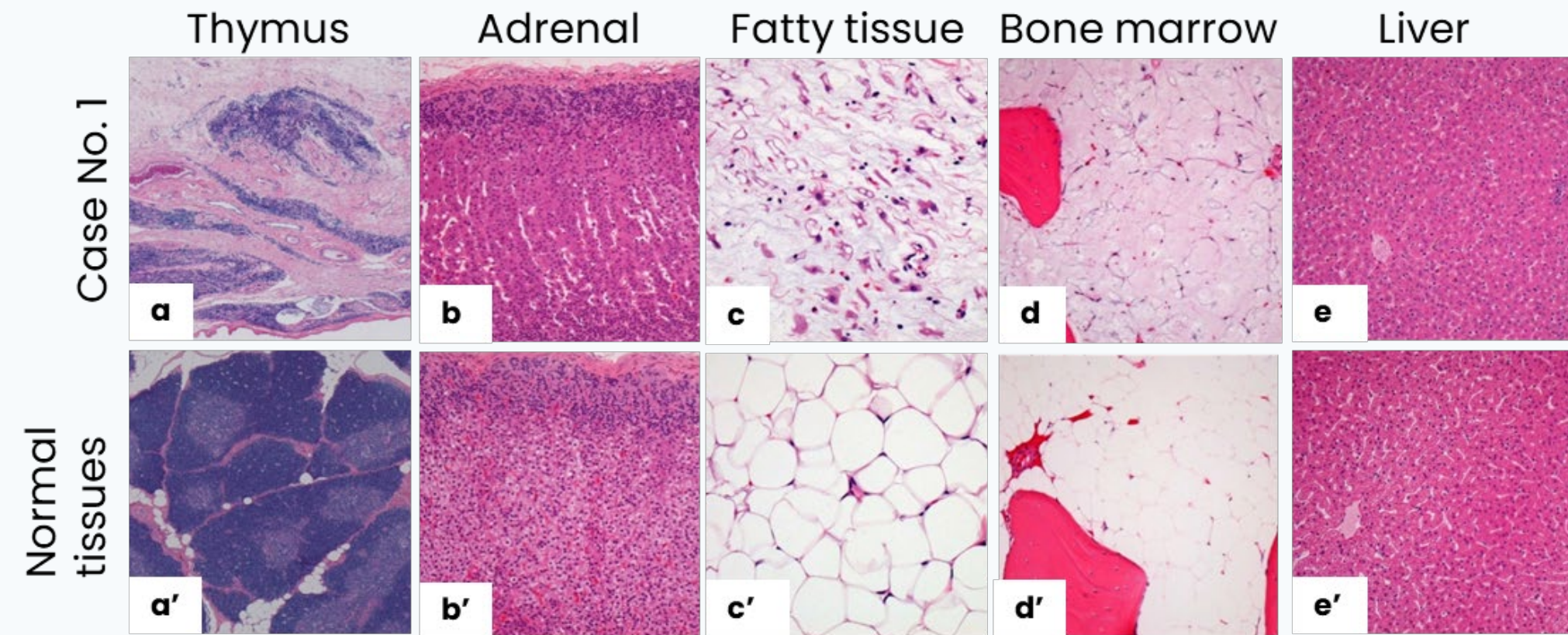


Fig. 3. Histopathological changes of Case No. 1

a: Atrophy

b: Reduction in cytoplasmic lipid

c: Atrophy

d: Gelatinous atrophy

e: Decrease in glycogen in hepatocytes

The changes observed in the 4-week studies resembled drug-induced immunosuppression and secondary stress- or wasting-related findings typically seen in toxicity studies. In contrast, such changes were not observed in weight-loss cases in the 13-week studies.

Study design	4-week studies			13-week studies			
Study No.	A	B	C	C	D	E	F
Case No.	1	2	3	4	5	6	7
Country of origin/sex/age	Vietnam/male/4 years old	Vietnam/male/6 years old	Vietnam/male/7 years old	China/male/4 years old	China/male/4 years old	Vietnam/male/4 years old	Vietnam/female/4 years old
Number of days of rearing	56 days	58 days	58 days	110 days	110 days	104 days	109 days
Weight change	4.3 kg to 3.7 kg	4.5 kg to 4.0 kg	4.5 kg to 3.8 kg	3.9 kg to 3.2 kg	3.0 kg to 2.7 kg	4.7 kg to 4.2 kg	3.3 kg to 2.8 kg
Weight change rate	–14.0%	–11.1%	–15.6%	–18.0%	–10.0%	–10.6%	–15.1%
Clinical sign	–	–	–	Crouching posture (Days 85–86) ^{c)} Diarrhea and loose stool (Days 81–92)	–	–	–
Food consumption ^{a)}	100 g/100 g	100 g/100 g	96.5 g/100 g	100 g/100 g	95.5 g/100 g	100 g/100 g	97.7 g/100 g
Hematology ^{b)}	Hemoglobin concentration ↓ (11.8 g/dL) Hematocrit ↓ (32.2 %)	–	Neutrophil ↑ (14.02×10 ³ /μL)	N.E.	N.E.	Red blood cell ↓ (4.9×10 ³ /μL) Hemoglobin concentration ↓ (12.5 g/dL) Hematocrit ↓ (36.0%)	Neutrophil ↑ (25.26×10 ³ /μL) Red blood cell ↓ (4.49×10 ³ /μL) Hemoglobin concentration ↓ (11.1 g/dL) Hematocrit ↓ (35.9%)
Blood chemistry	Glucose ↓ (49 mg/dL)	Glucose ↓ (66 mg/dL)	–	N.E.	N.E.	Inorganic phosphorus ↓ (2.6 mg/dL)	–
Organ weight	Thymus ↓ (0.63 g)	Thymus ↓ (0.28 g)	Thymus ↓ (0.80 g)	N.E.	N.E.	–	–
Necropsy	Thymus: Small	–	–	–	–	–	–
Histopathology ^{d)}	Thymus, Lymph nodes, Salivary glands: Atrophy Femur bone marrow: Gelatinous atrophy Sternal bone marrow: Gelatinous atrophy, Decrease in hematopoietic cells ^{e)} Fatty tissue: Atrophy Adrenal gland: Reduction in cytoplasmic lipid Liver: Decrease in glycogen in hepatocytes	Thymus: Atrophy Femur bone marrow: Gelatinous atrophy Fatty tissue: Atrophy ^{e)} Pancreas: Atrophy	Thymus: Atrophy	–	–	–	–

Table 1. Summary of various tests on monkeys with weight change rate of less than –10%

–: No noteworthy findings, ↑: High value, ↓: Low value

a) Mean daily food consumption/feeding amount. The weight loss occurred despite adequate food intake.

c) Hypoglycemia was suspected, and the animal was given a glucose injection, after which it showed improvement. The day of the initial dosing was designated as Day 1.

b) Tendency of anemia was noted.

d) See Fig 3

e) Including visceral and subcutaneous fat

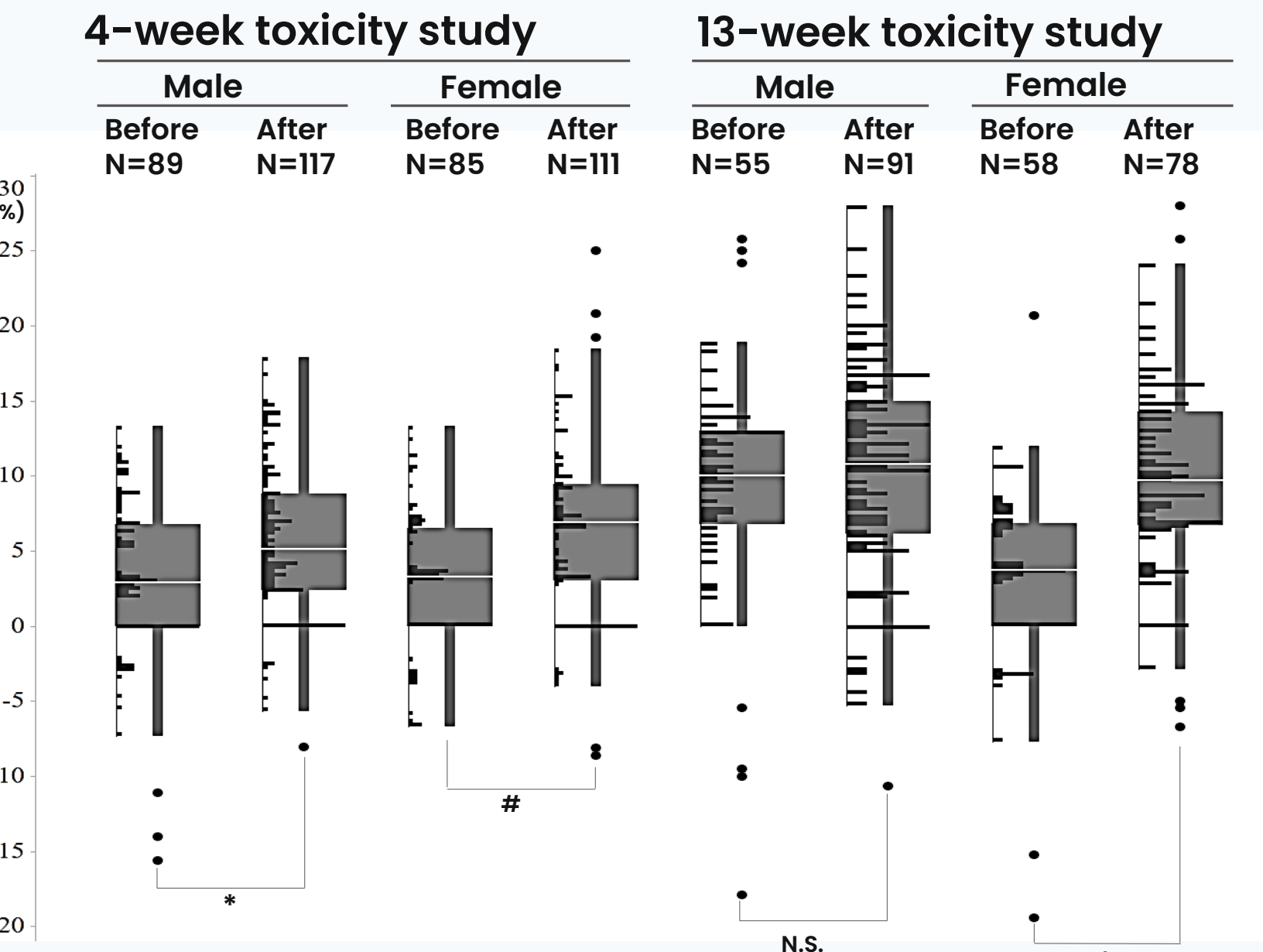


Fig. 4. Comparison of weight change rate before and after the start of the enrichment program

*=P<0.05 (Student t test), #=P<0.05 (Aspin Welch t test), N.S.: Not significant

Vertical axis: Weight change rate (%)

A statistically significant increase in the weight change rate was noted in both sexes in the 4-week studies and in females in the 13-week studies after the enrichment program compared with before its implementation.

Discussion

1. Histopathology

Stress-related histopathological changes were observed only in the 4-week studies, likely due to **acute** and high stress loads with rapid weight loss. In contrast, the 13-week studies involved milder, **chronic** stress with gradual weight loss, which may not have produced detectable histopathological changes.

2. Clinical Pathology

In the 4-week studies, 2 animals showed low blood glucose and 1 showed anemia tendencies. In the 13-week studies, 1 animal each exhibited low inorganic phosphorus or hypoglycemia-like posture, and 2 showed anemia tendencies.

These findings suggest impaired nutrient absorption or altered nutrient metabolism.

3. Animal Welfare

Improved animal welfare reduced the occurrence of incidental weight loss, **confirming the value of enrichment programs.**

These findings suggest that mild stress factors, together with individual differences in stress sensitivity, may contribute to incidental weight loss in cynomolgus monkeys.