



Effect of the neonicotinoid pesticide clothianidin at a no-observed-adverse-effect-level (NOAEL) dose on maternal behavior in pregnant mice and their female offspring

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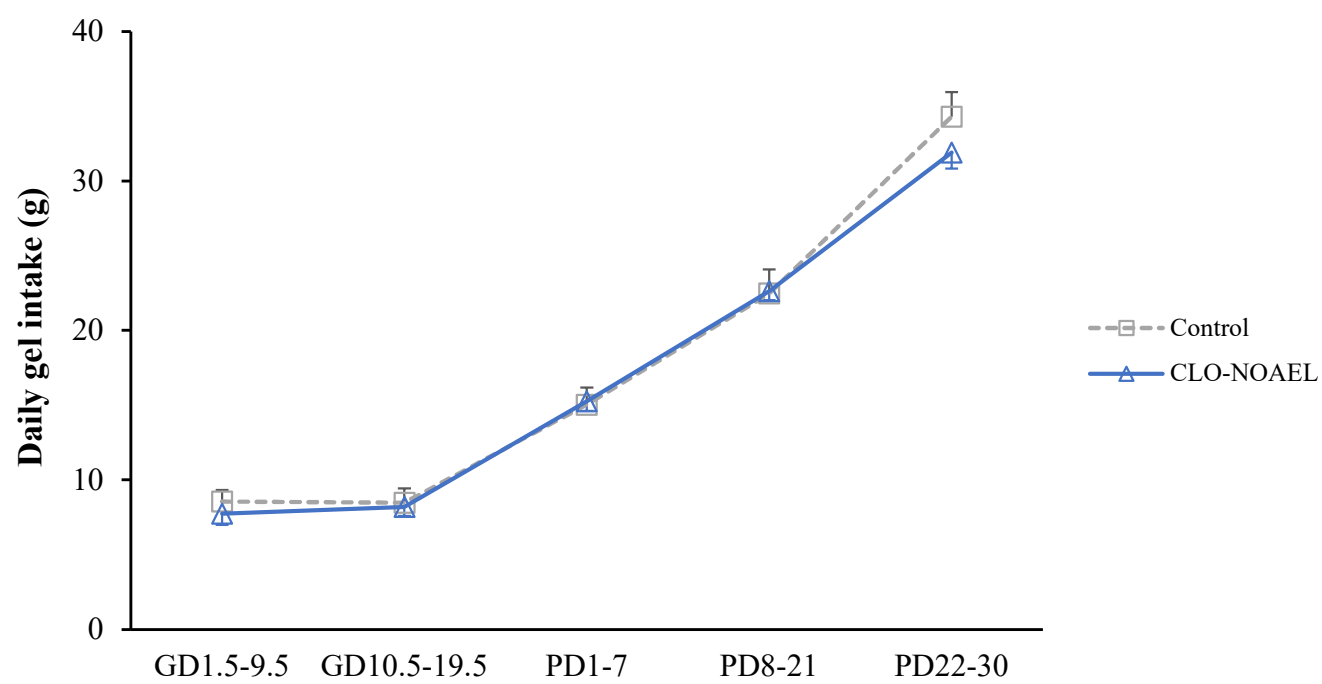
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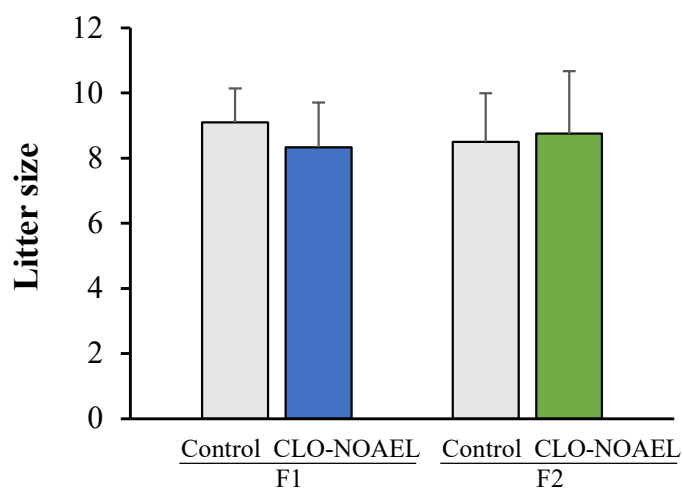
Supplementary Fig. 1



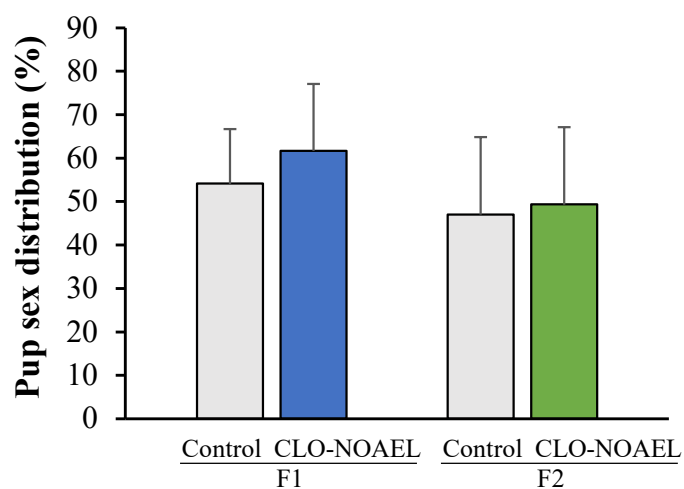
Daily gel intake in F0 mothers. Variations in intake among individuals were not significant. Values are mean \pm SD (n=6-10 mice each). CLO: clothianidin, NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 2

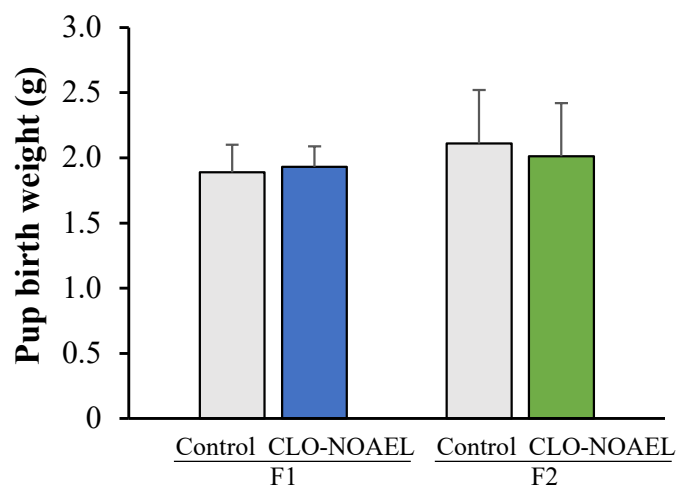
A



B

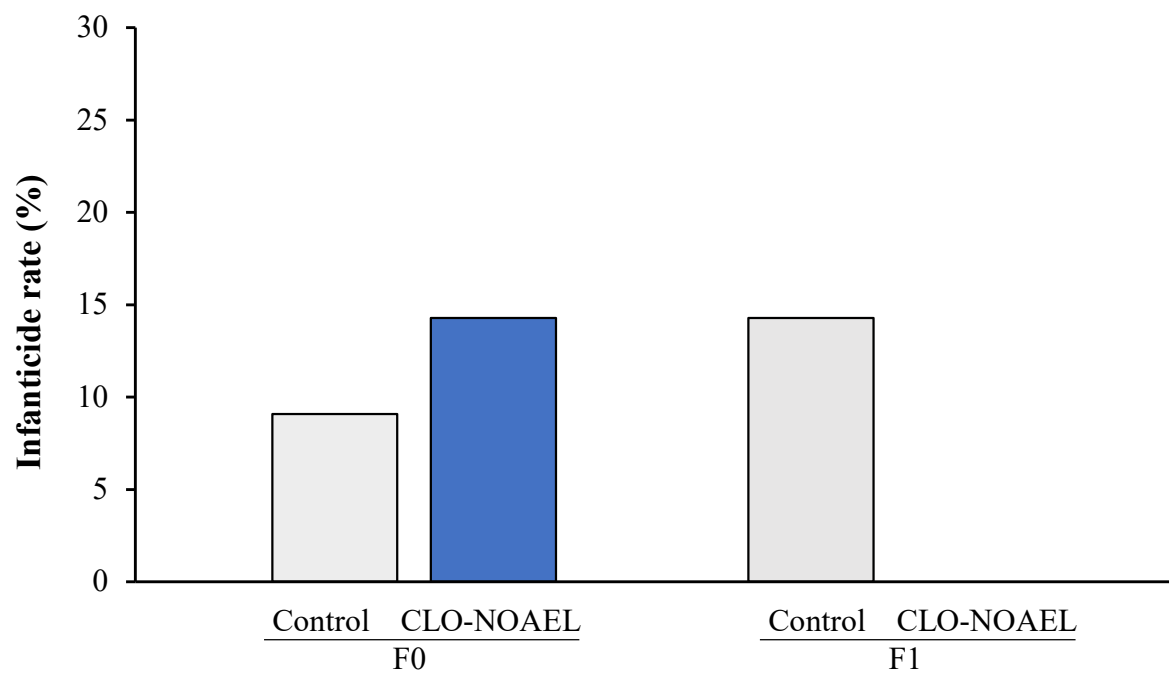


C



(A) Litter size, (B) pup sex distribution, and (C) pup birth weight in F1 and F2 pups. Clothianidin (CLO) had no significant effect on each parameter. Values are mean \pm SD: 60 pups in F0 Control, 36 in F0 CLO-NOAEL, 36 in F1 Control, and 48 in F1 CLO-NOAEL. NOAEL: no-observed-adverse-effect level.

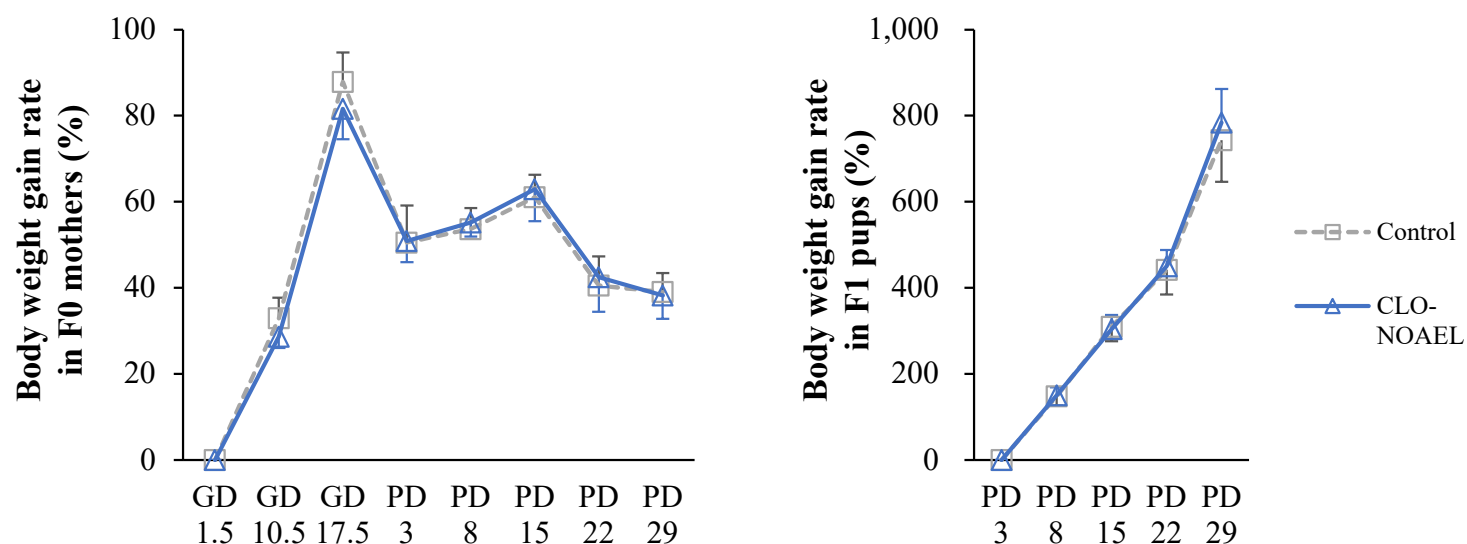
Supplementary Fig. 3



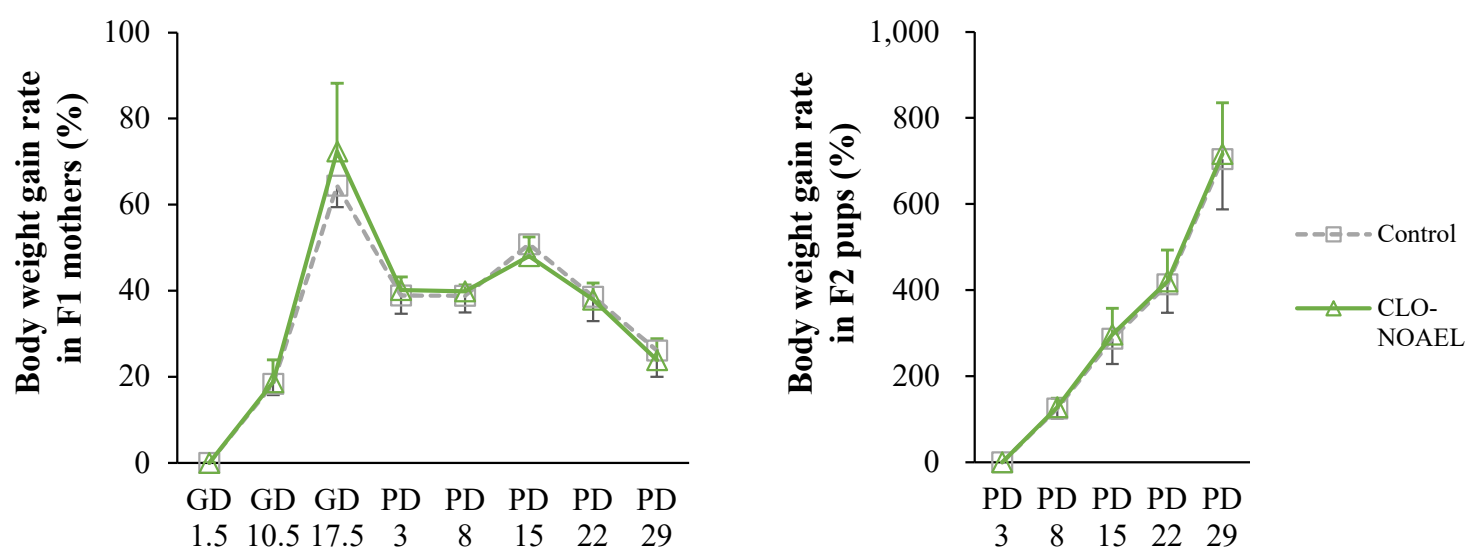
Infanticide rate, calculated by dividing the number of infanticide cases by the total number of deliveries and multiplied by 100. There was one case of infanticide in each group in F0 mothers (Control group: 1/11, CLO-NOAEL group: 1/7), and there was one case of infanticide only in the Control group in F1 mothers (Control group: 1/7, CLO-NOAEL group: 0/8). CLO: clothianidin, NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 4

A

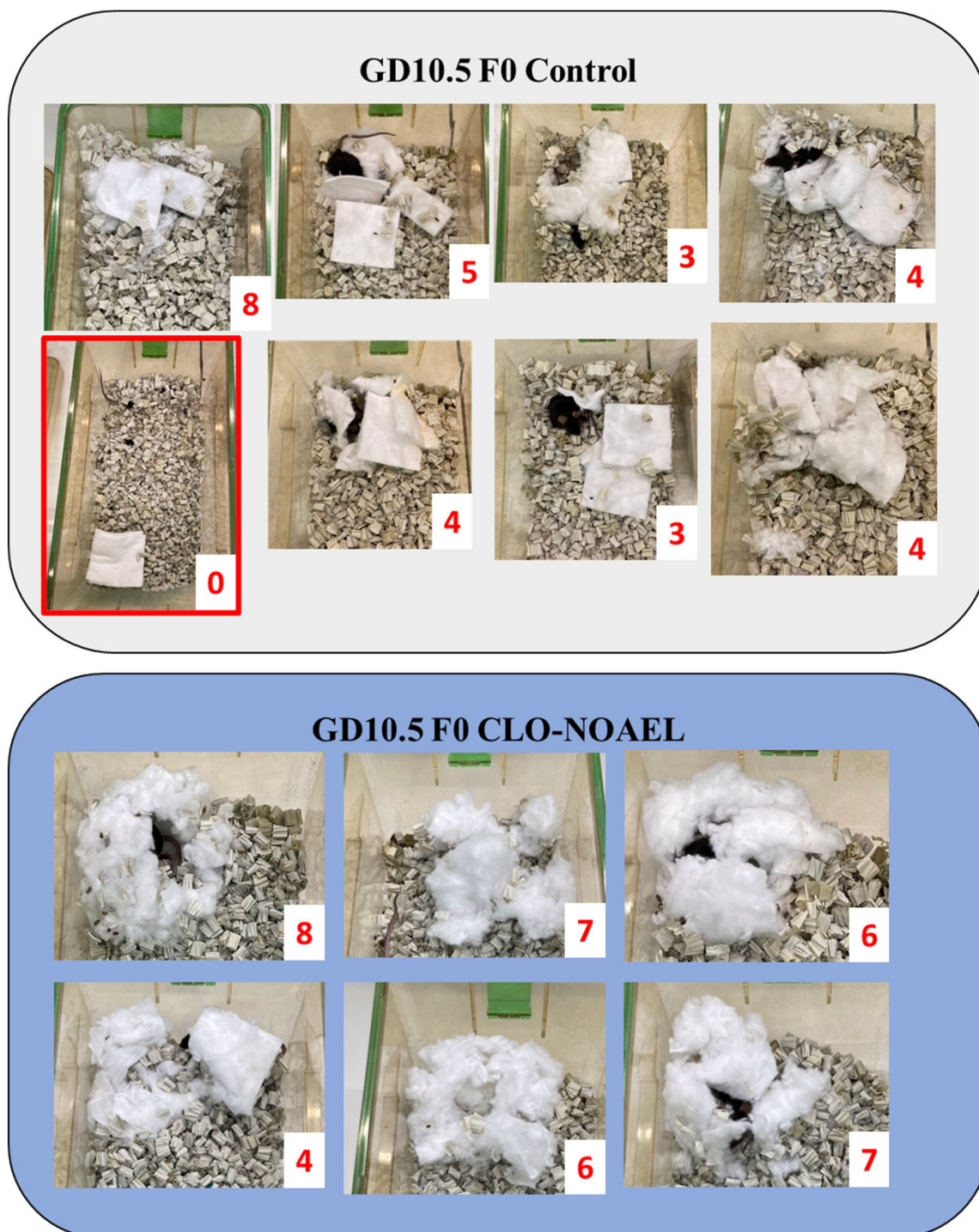


B



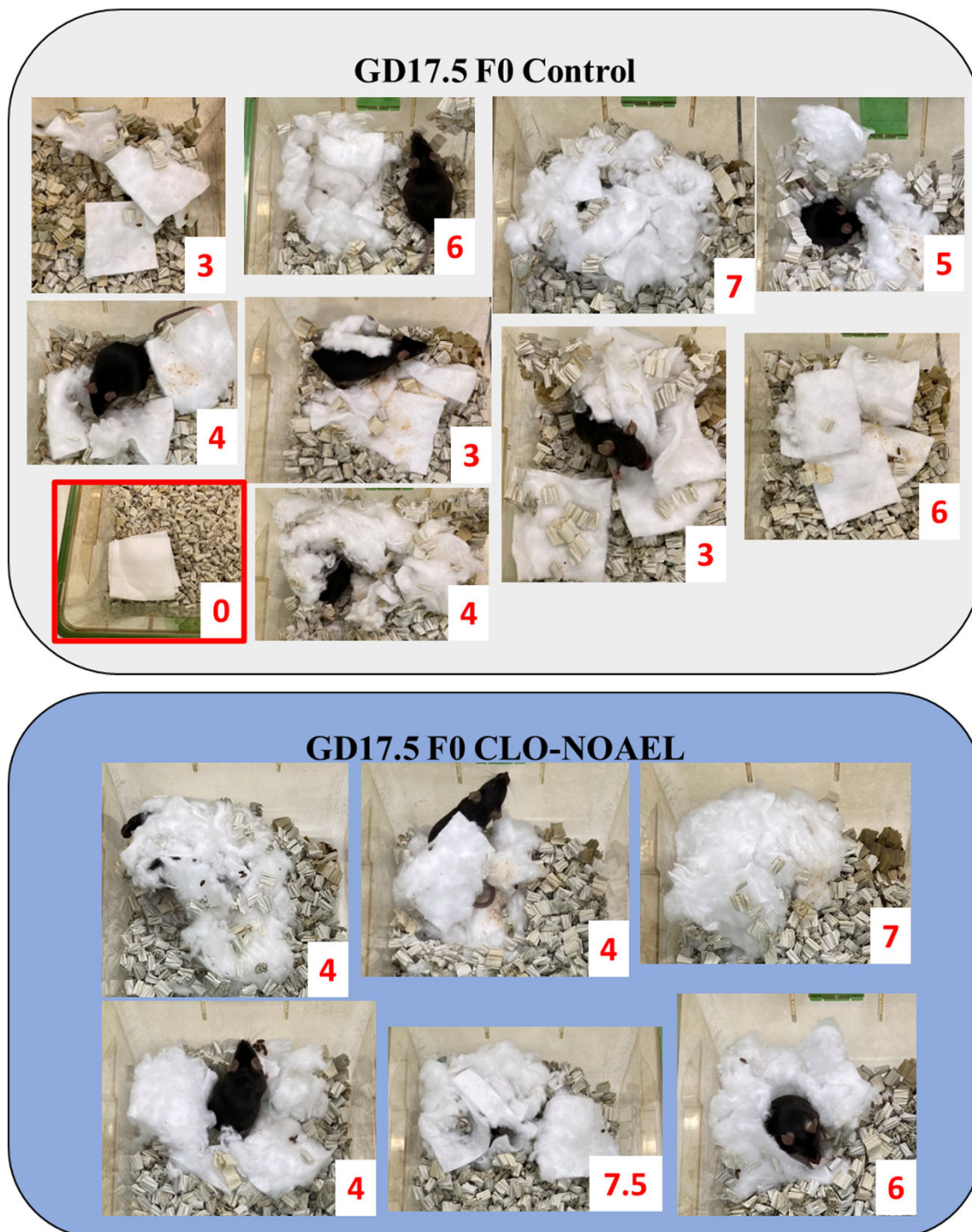
Body weight gain rate. No effects of clothianidin (CLO) were observed in either (A) F0 mothers and F1 pups or (B) F1 mothers and F2 pups. Values are mean \pm SD (F0 Control: 10 mothers & 60 pups, F0 CLO-NOAEL: 6 mothers & 36 pups, F1 Control: 6 mothers & 36 pups, F1 CLO-NOAEL: 8 mothers & 48 pups). NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 5



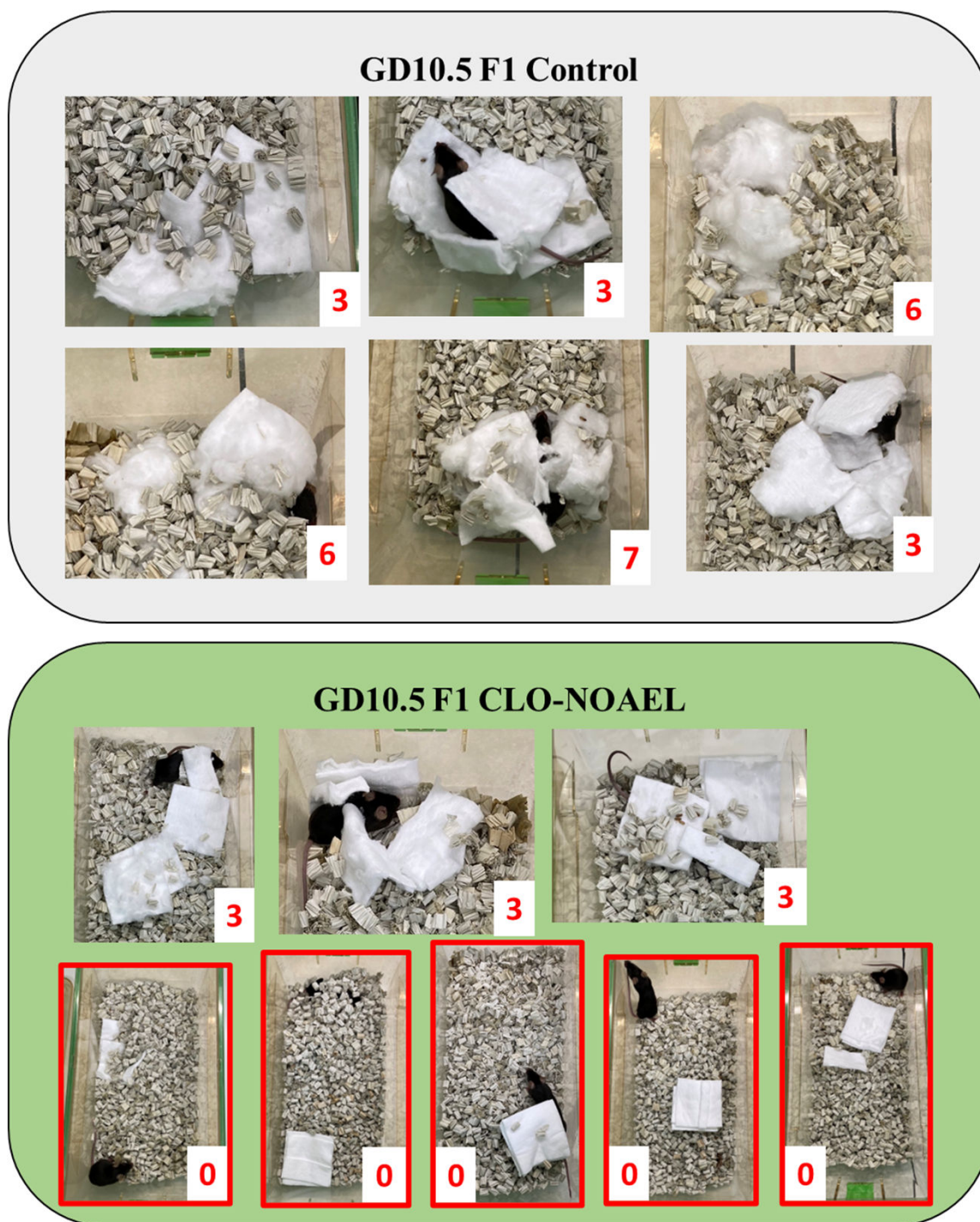
Nest-building behavior in F0 mothers at gestational day (GD) 10.5. The CLO-NOAEL group showed more careful nest-building than the Control group. The numbers in the figure indicate nest-building scores. Scores of 0 are indicated by a red frame. CLO: clothianidin, NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 6



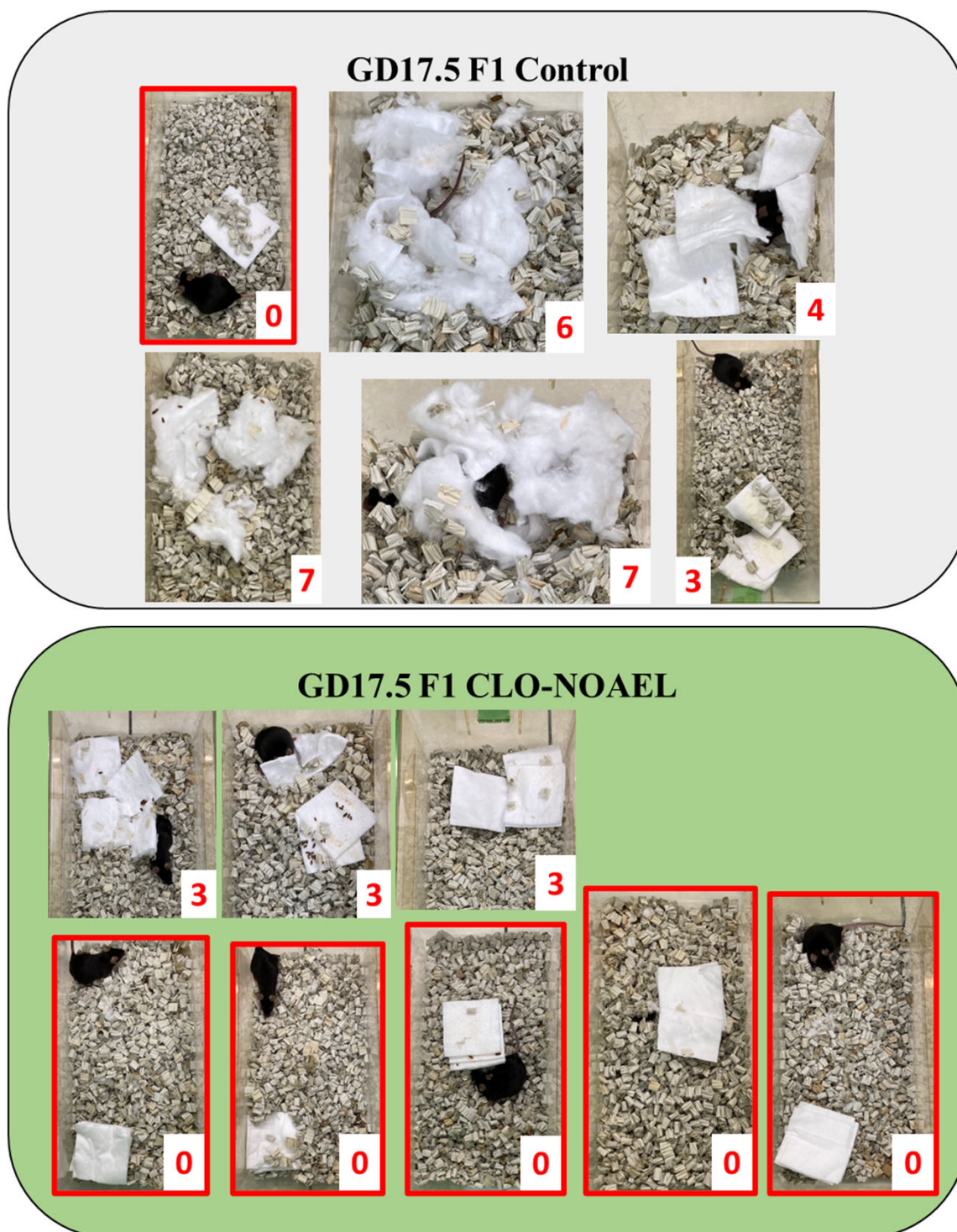
Nest-building behavior in F0 mothers at gestational day (GD) 17.5. The CLO-NOAEL group showed more careful nest-building than the Control group. The numbers in the figure indicate nest-building scores. Scores of 0 are indicated by a red frame. CLO: clothianidin, NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 7



Nest-building behavior in F1 mothers at gestational day (GD) 10.5. Some mothers in the CLO-NOAEL group showed no nest-building behavior at all. The numbers in the figure indicate nest-building scores. Scores of 0 are indicated by a red frame. CLO: clothianidin, NOAEL: no-observed-adverse-effect level.

Supplementary Fig. 8



Nest-building behavior in F1 mothers at gestational day (GD) 17.5. Some mothers in the CLO-NOAEL group showed no nest-building behavior at all. The numbers in the figure indicate nest-building scores. Scores of 0 are indicated by a red frame. CLO: clothianidin, NOAEL: no-observed-adverse-effect level.