Supplementary material belonging to:

The genetic architecture underlying body-size traits plasticity over different temperatures and developmental stages in *Caenorhabditis elegans*

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Supplementary Table S1. An overview of the experimental set up and raw data (in pixels) of this study. Per nematodes individual, detailed workflow of the experiment starting from the nematodes line used, replicate per lines, bleaching date and time per nematodes, picture date and time per nematodes, developmental stage and age of the nematodes when the pictures were taken, and raw data of the traits measured (in pixel) are given.

Supplementary Table S2. Calculated raw data (in mm) and the mean value of every traits measured. Conversion of the raw data was done by calculating the conversion constant from pixels to milimiters using ImageJ. Mean values of the trait were calculated per nematodes lines per temperature and developmental stages. All blank cells containing no value were removed.

Supplementary Table S3. Calculated transgressive segregation. Per Strain, stage, temperature, and trait the t-test significance - adjusted for multiple testing by the false discovery rate (FDR) method – is given. When both the N2 and CB4856 parent were significant, a strain was considered to show transgression.

Supplementary Table S4. Calculated broad sense and narrow sense heritability. Per trait, developmental stage and temperature the broad-sense heritability H^2 was calculated (H2_REML), the value for the 0.05 FDR threshold is also given (H2_FDR) as is whether the H^2 was significant (H2_significance). Similar for the narrow-sense heritability h^2 was calculated (h2_REML), the value for the 0.05 FDR threshold is also given (h2_FDR) as is whether the h^2 was significant (h2_significance).

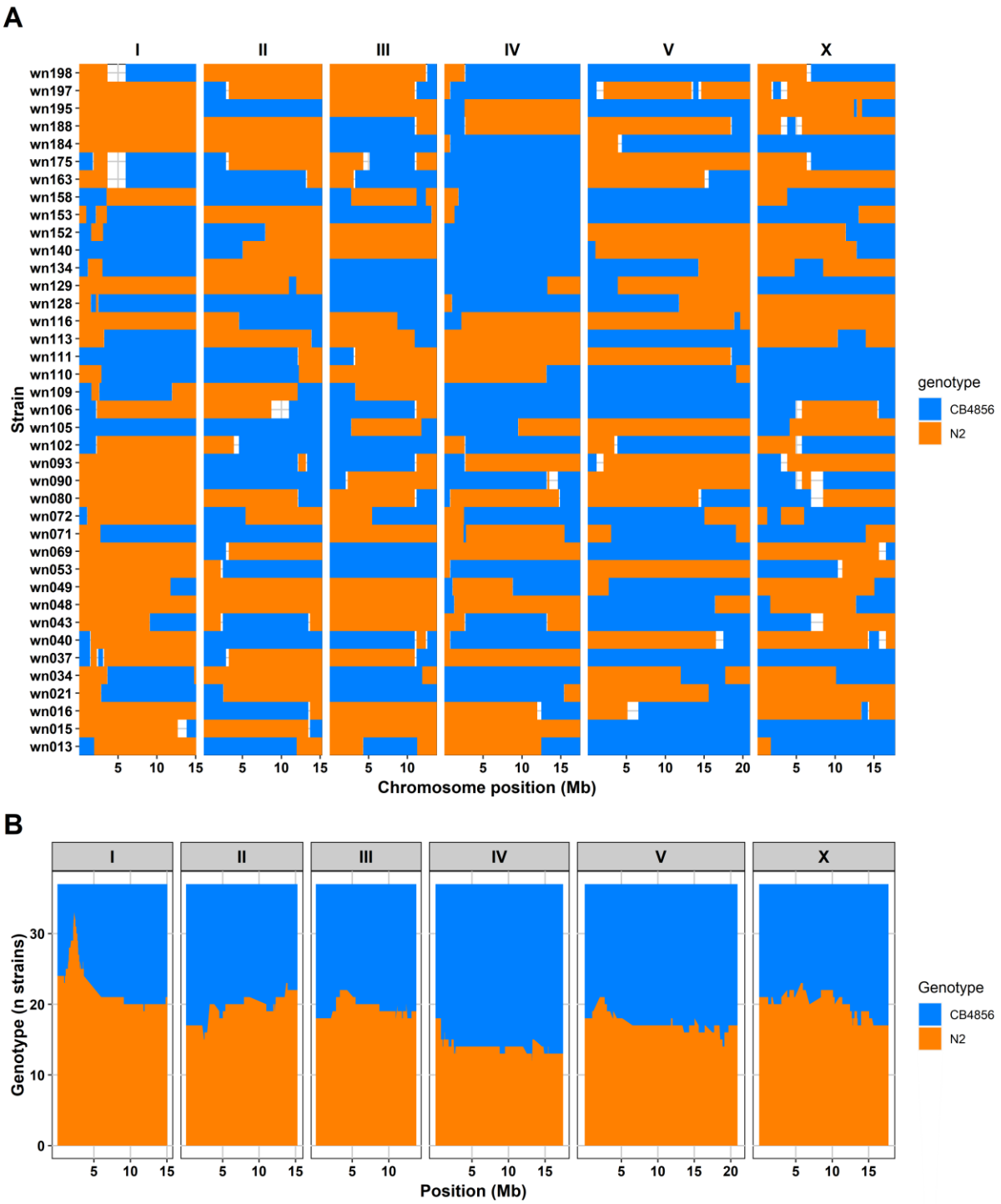
Supplementary Table S5. Analysis of power by simulation of QTL. Ten simulated QTL per marker location was performed that explained 20-80% of the variation, with an increment of 5% (20%, 25%, 30%, ..., 80%). The threshold used was $-\log_{10}(p)$ 3.4 that was derived from 1000x permutation at an FDR 0.05. The number of variance explained, fraction of false QTL, detected QTL, undetected QTL, as well as QTL effect size estimation were given.

Supplementary Table S6. QTL table of the body-size traits across different temperatures and developmental stages. Per trait in temperature and developmental stage combination, the QTL detection was done by calculating the $-\log_{10}(p)$ score and significance was determined if the QTL $-\log_{10}(p)$ value is bigger than the FDR (0.05) at $-\log_{10}(p)$ of 3.4 based on permutation analysis. Per QTL in developmental stage and temperature combination, the position of QTL in chromosome, QTL position in base pairs, QTL left region, QTL right region, QTL effect, and the R^2 value of each QTL are given.

Supplementary Table S7. Calculated broad sense and narrow sense heritability of plasticity. Plasticity per trait, developmental stage and temperature ranges the broad-sense heritability

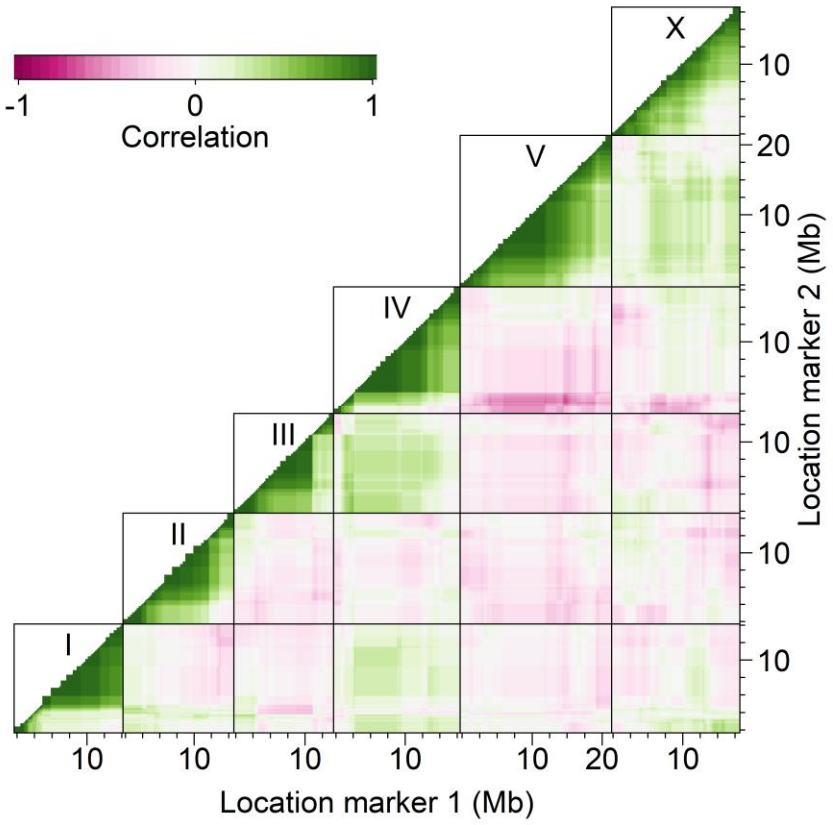
H^2 was calculated (H2_REML), the value for the 0.05 FDR threshold is also given (H2_FDR) as is whether the H^2 was significant (H2_significance). Similar for the narrow-sense heritability h^2 was calculated (h2_REML), the value for the 0.05 FDR threshold is also given (h2_FDR) as is whether the h^2 was significant (h2_significance).

Supplementary Table S8. Plasticity QTL table of the body-size traits across different temperatures changes and developmental stages. Per trait and developmental stage, the temperature change, the position of QTL in chromosome, QTL position in base pairs, QTL left region, QTL right region, QTL effect, and the R^2 value of each QTL are given. The QTL detection was done by calculating the $-\log_{10}(p)$ score and significance was determined if the QTL $-\log_{10}(p)$ value is bigger than the FDR (0.05) at $-\log_{10}(p)$ of 3.4 or 4.1 (for temperature range 24ºC to 26ºC).

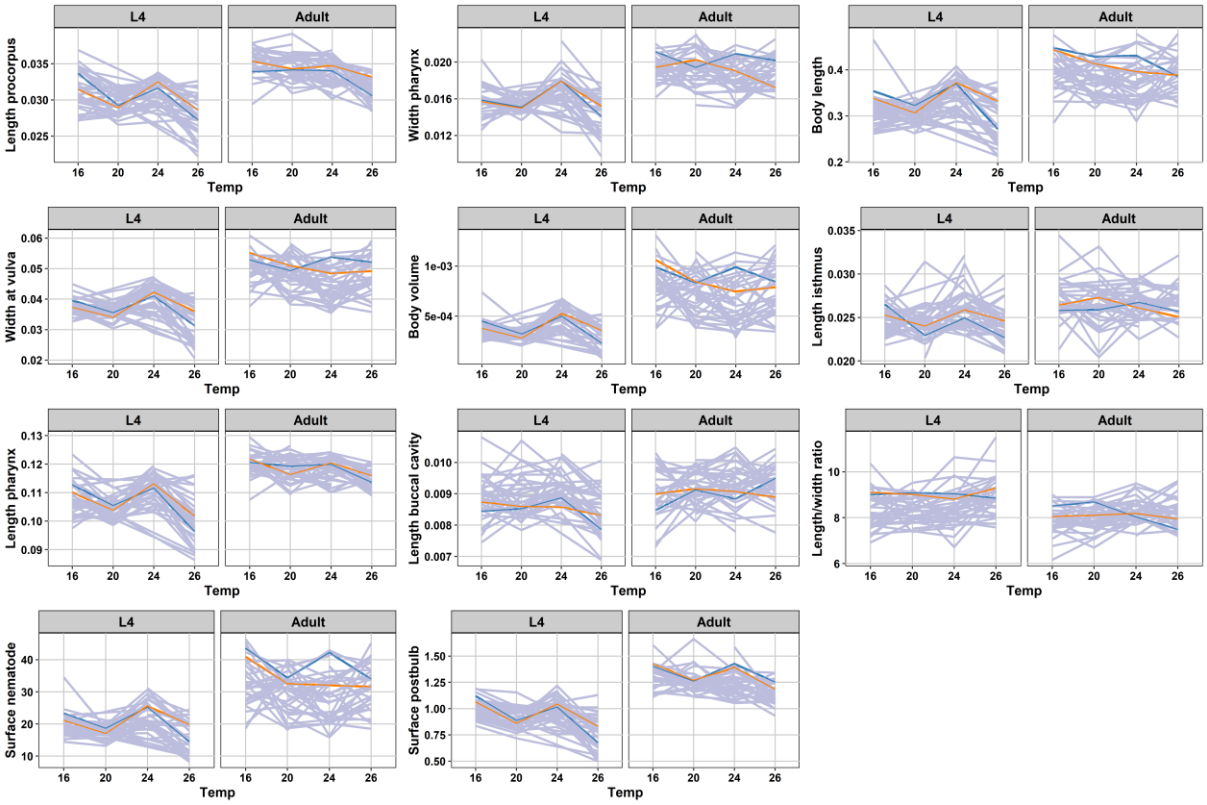


Supplementary Figure S1. (A) Genetic map of the 40 RILs population. The x-axis indicates the genomic position with separate chromosomes indicated at the top of the graph. The strains of the RILs are depicted n y-axis. The RILs were genotyped by PCR-based genotyping. Orange color represents N2 genotype while CB4856 is represented by blue. Regions with uncertain genotyped are indicated by white. (B) Genotype distribution of the 729 markers along the six chromosomes. Orange represent N2 background while CB4856 was depicted in blue. The proportion of alleles frequencies that formed the RILs genetic background were roughly equal over most of the genome (Figure S2). On the left arm of chromosome I however, there was a skew of allele frequencies which we have been expected as we used the

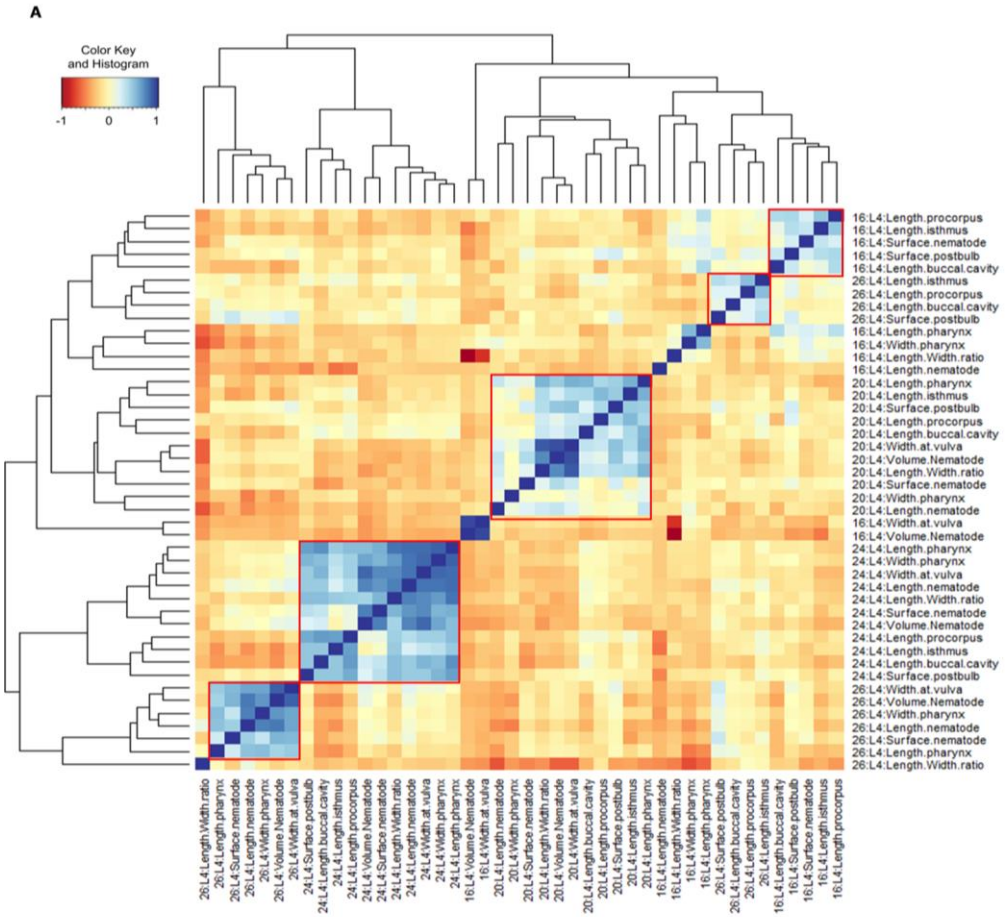
same RILs panel developed by (Li *et al*., 2006). This skew is the result of the genetic incompatibility between *zeel-1* and *peel-1* allele from N2 parent. The strains that are not protected by N2-provided *zeel-1* gene would be killed by the toxic effect from the N2-provided *peel-1* gene (Seidel *et al*., 2011).



Supplementary Figure S2. Correlation plot between markers. The markers were arranged to conform their physical position starting from telomere until the arms of chromosome. The correlation ranges from -1 (purple) to 1 (green) where strong correlation indicated a linkage between markers. The genotyping was done using 729 SNPs marker generated previously from low coverage sequencing of the parental strains (N2 and CB4856) (Thompson et al., 2015). To validate the genetic map for QTL mapping, we conducted a correlation analysis of the markers to detect linkage over the map. The markers are arranged to represent their physical position in chromosome.



Supplementary Figure S3. The reaction norms of *C. elegans* body-size traits plasticity across different temperature and developmental stages. The x-axis represents temperatures used while y-axis represent the mean value of the individual strains in their respective traits (in mm, except for body volume in mm3 and surface area in mm2). Please note that the y-axis is not identical between traits. Both parents are depicted in blue (CB4856) and orange (N2), while the RILs are grey. Lines are the traits mean value (N = 3 for RILs, and N= 9 to 12 for parents).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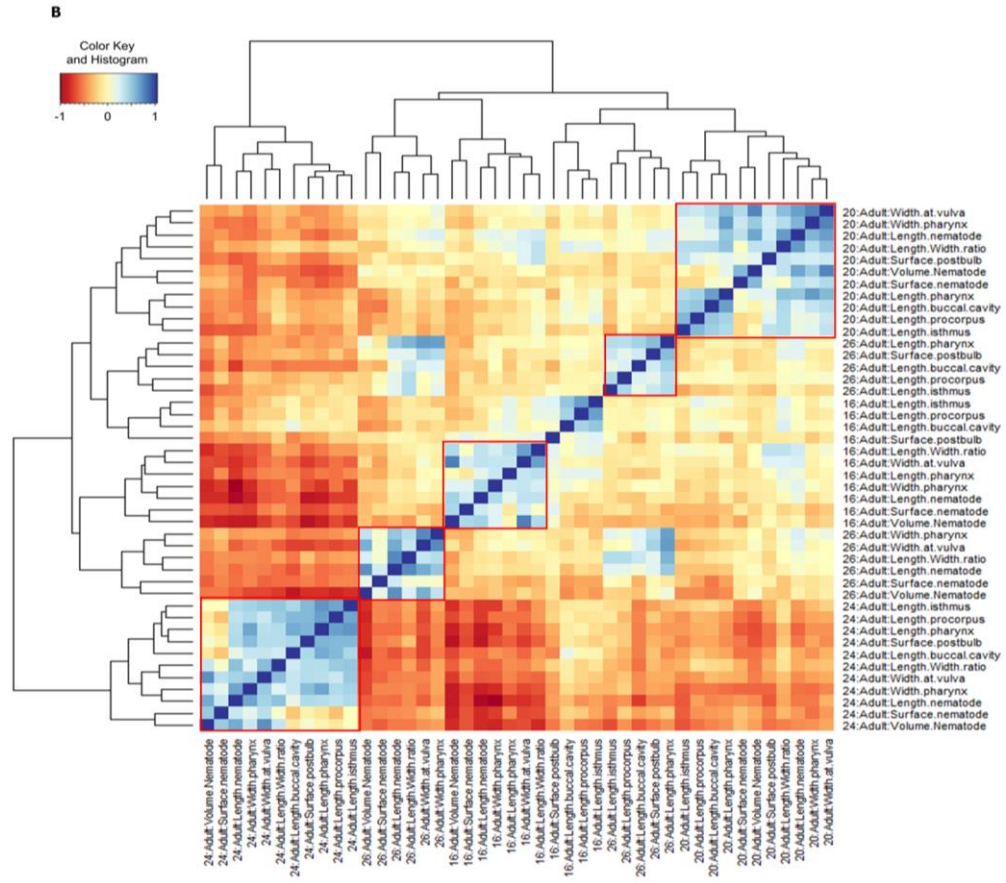
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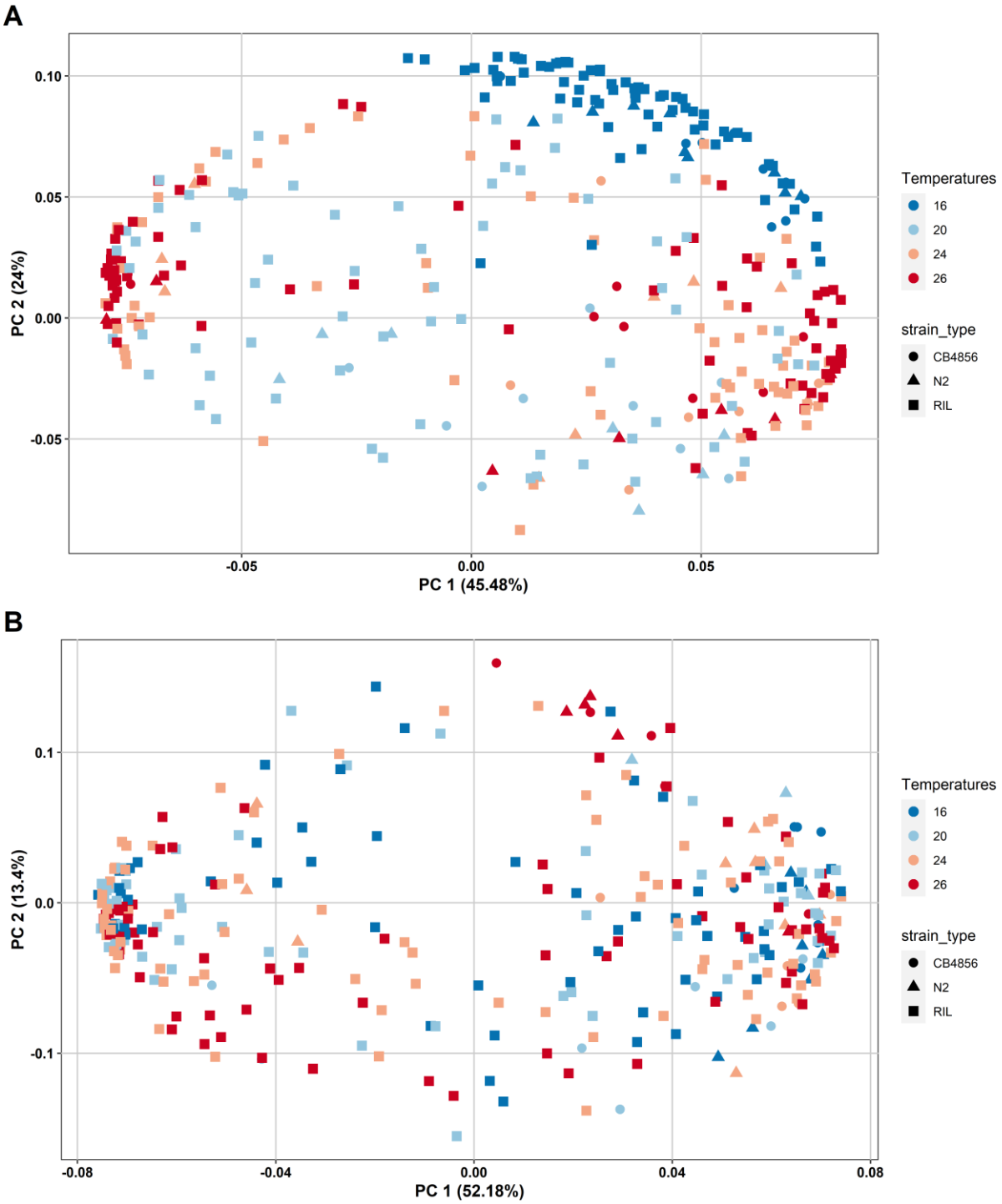
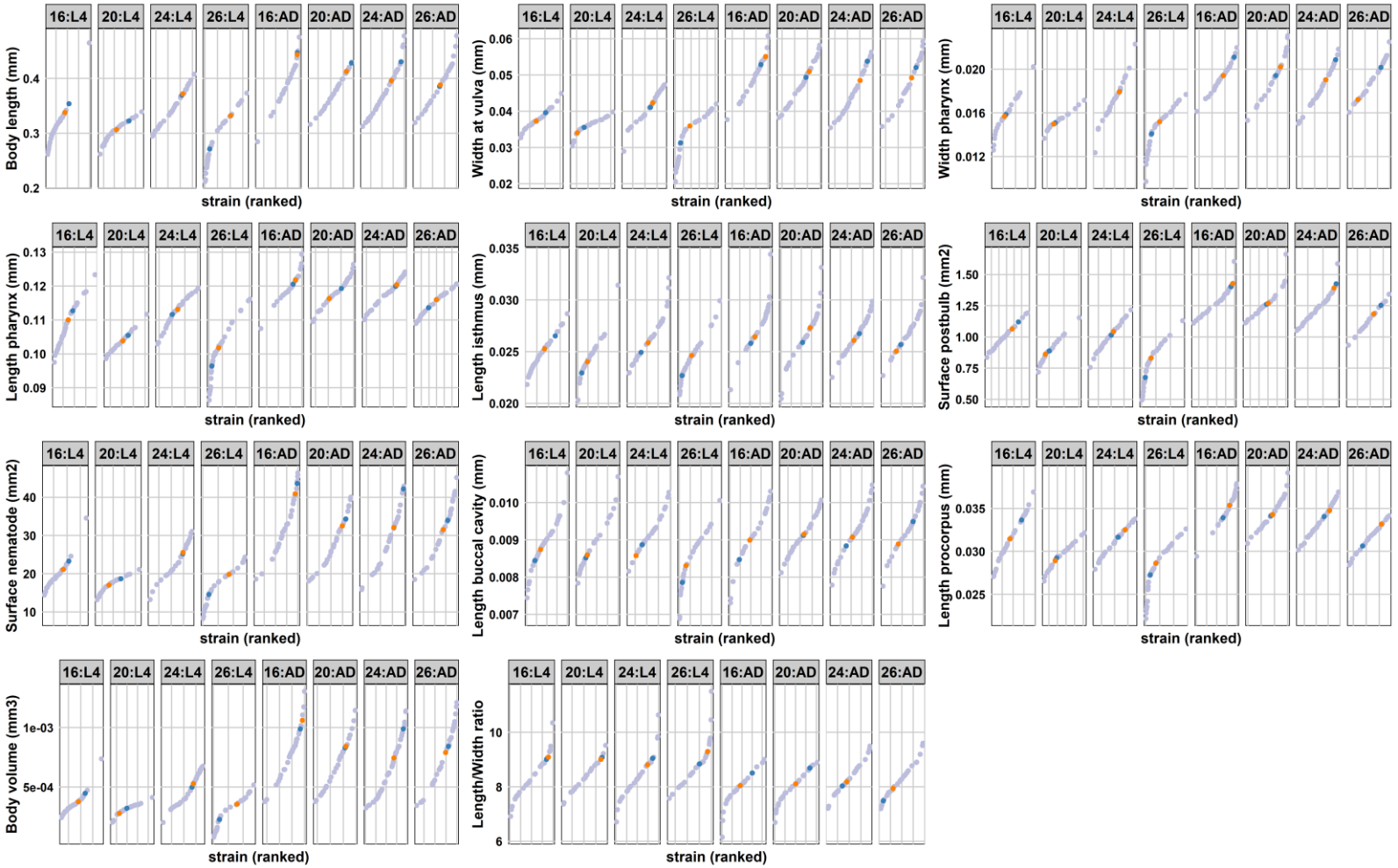
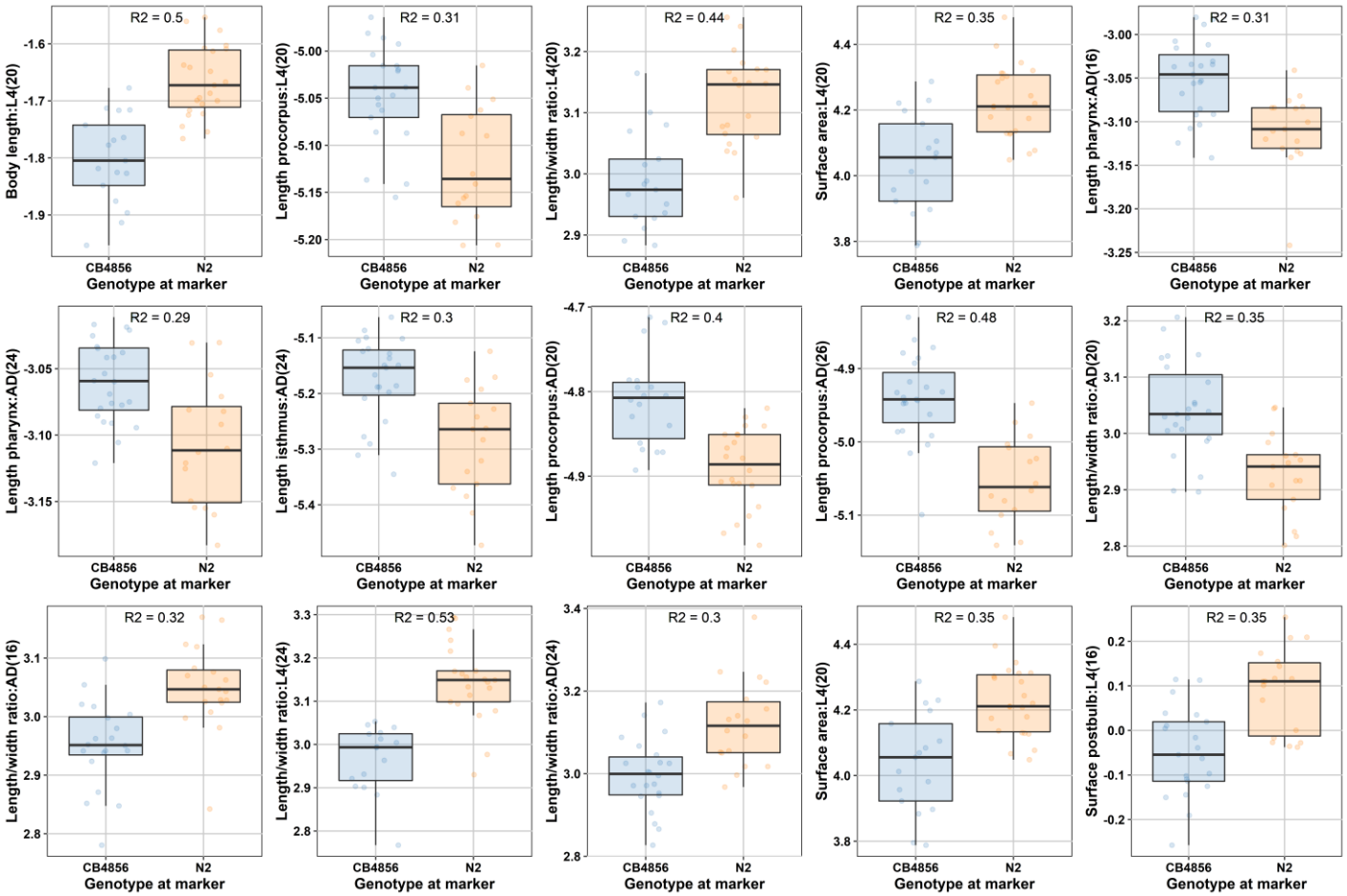
Supplementary Figure S4. Correlation analysis of body-size traits in the RIL population. (A) correlation heatmap of body-size traits of all RILs in L4 stage. There were three five strong positively correlated clusters which were temperature specific. (B) correlation heatmap of body-size traits of all RILs in adult stage. There were five strong cluster of positive correlation of worm grown in 20^{o}C and 24^{o}C.



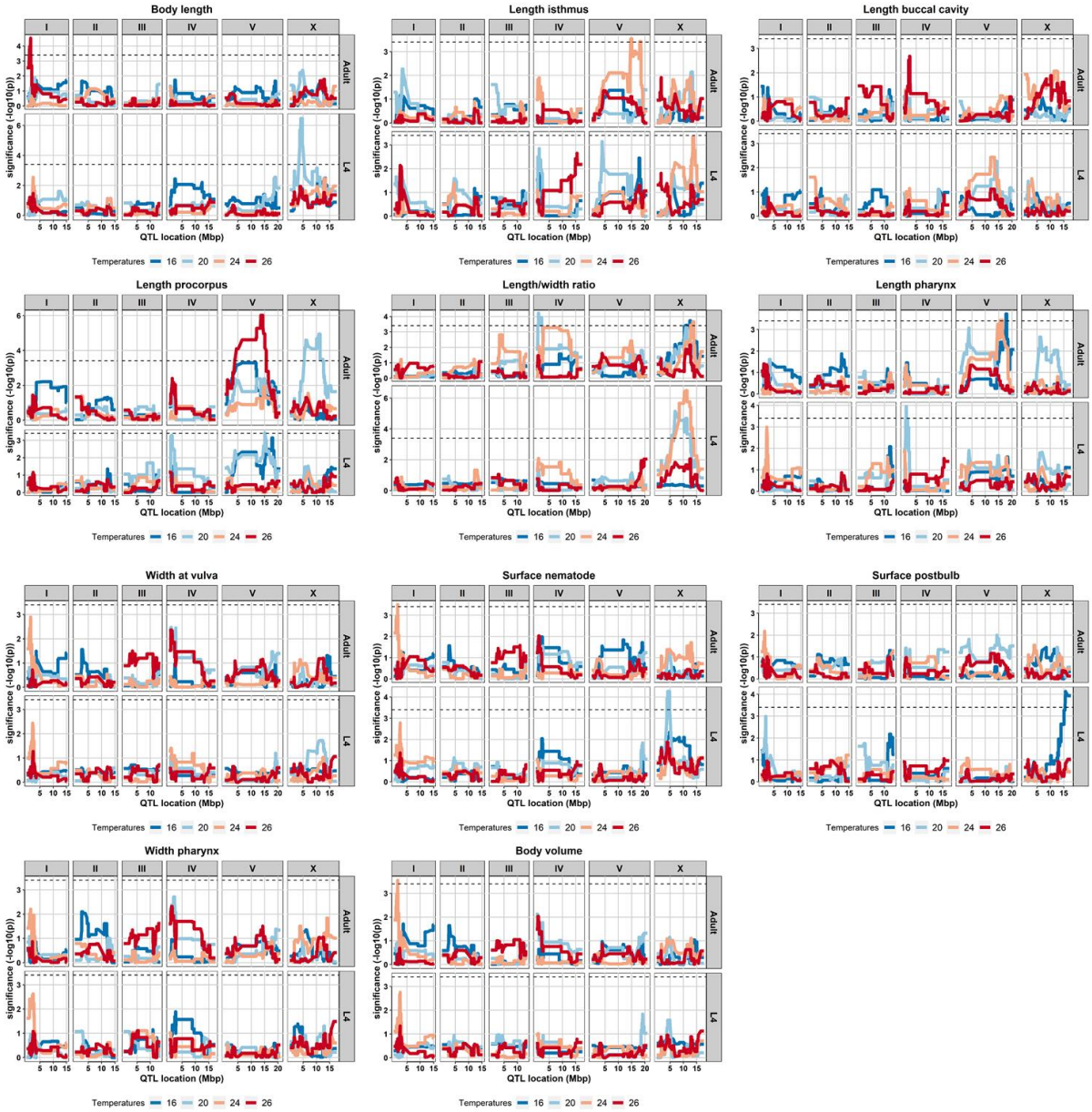
Supplementary Figure S5. Principal component analysis of *C. elegans* body-size at (A) L4 stage and (B) adult stage: the first principal component (PC1) versus the second principal component (PC2) of the body-size traits from four different temperatures (16^{o}C, 20^{o}C, 24^{o}C, and 26^{o}C) and genetic backgrounds (N2, CB4856, and RILs).



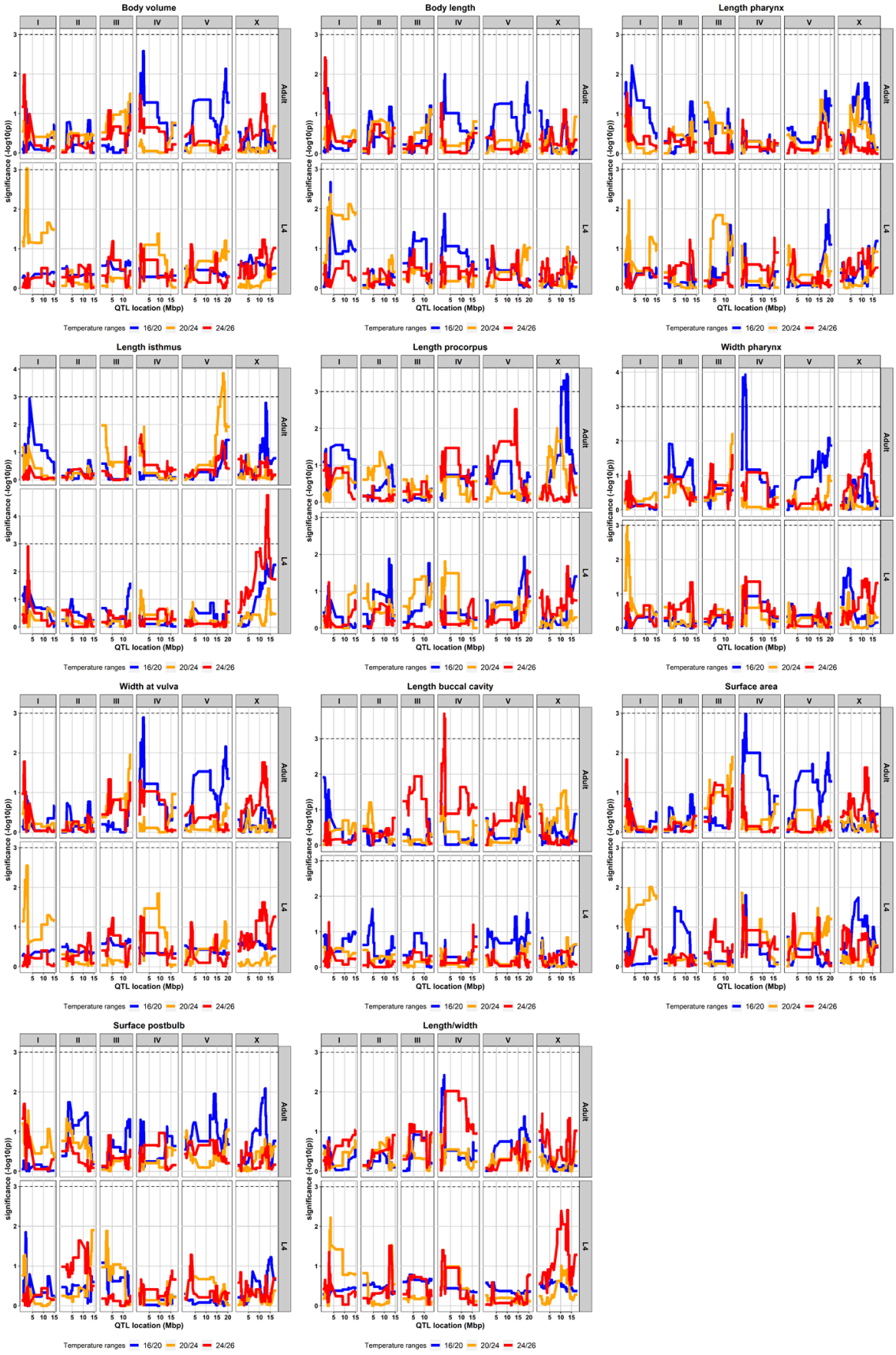
Supplementary Figure S6. The body size parameters of *C. elegans* across 40 RILs and two parental strains. The average traits are expressed as percentage of sum of each traits (x-axis), followed by the mean value of the traits (y-axis). Please note that the y-axis is not identical between traits. The 40 RILs are indicated with grey, the parental N2 in orange, and the parental CB4856 in blue. Points are the traits mean value (N = 3 for RILs, and N= 9 to 12 for parents).



Supplementary Figure S7. Description of genotype at the peak marker locations of the signficant QTL. X-axis represents parental genotype at the peak marker location of the QTL while y-axis represents the corresponding body-size traits. Blue box visualizes CB4856 and orange bos is N2. R^2 shows how much the variation of the body-size traits can be expalined by genetic variation in the chromosome.



Supplementary Figure S8. QTL plot of the body-size traits. The QTL analysis were performed across four temperatures (16^{o}C, 20^{o}C, 24^{o}C, 26^{o}C) and two developmental stages (L4 and adult). X-axis displays genomic position in the chromosome corresponding to the box above the line while y-axis represents the LOD score. Box in the right graph show the developmental stages. Red line represents QTL at 16^{o}C, green line at 20^{o}C, blue line at 24^{o}C, and purple line at 26^{o}C. Red-dash line represents LOD threshold (1000x permutation-based significant LOD with FDR = 0.05).



Supplementary Figure S9. All plasticity QTL mappings for body-size traits in the 40 RILs. The x-axis represents the position of the QTL in million base pairs (Mbp) for each

chromosome and y-axis displays significance of the association based on a single marker model. The threshold $-\log_{10}(p) = 3.0$ is indicated with a horizontal dashed line. Colours indicate the plasticity comparisons.

References

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