Materials and methods

Data collection from published data

I explored the literature reporting absolute values of cell and nuclear volumes and visualizing the nucleus and/or whole cell using Google Scholar, Google Image and PubMed. When the cell or nuclear volumes were reported only on a graph, I estimated approximately values from the graph by measuring relative position of target data against the scales on the graph. When values in the cell or nuclear volume were not reported, I measured the major and minor axes of the nucleus or cell by using ImageJ from microscopic images and estimated the absolute length (*L*) and width (*W*) information based on the listed scale bar. I utilized the images, which could be detected for nucleus and/or whole cell visualized by staining using specific dyes or antibodies against DNA, nuclear membrane, nucleoplasmic proteins or cytoplasmic proteins, live-imaging with fluorescent-fused proteins or electron microscopic imaging. For simplifying the volume estimation, I excluded cells with complicated shape (non-spherical) such as adhesive culture cells due to inaccurate prediction and difficulty in estimating the height. To estimate the nuclear volume (*NV*), I calculated the volume as $NV = L/2 \times (W/2)^2 \times PI \times 4/3$ by assuming an oval sphere. In case of multiple nuclei inside the cell such as two pronuclei in the metazoan one-cell stage embryos, I used the total nuclear volume. To estimate the cell volume (*CV*), I used different equations for the predicted 3-dimentional cell shape as $CV = L/2 \times (W/2)^2 \times PI \times 4/3$ or $(L/2)^2 \times W/2 \times PI \times 4/3$ for oval-sphere-shaped cells (e.g. *Caenorhabditis elegans* embryos), $CV = (W/2)^2 \times PI \times L$ for cylindrical cells (e.g. fission yeast), $CV = L^2 \times W$ or $L \times W^2$ for cuboid cells (e.g. plant root tip cells surrounding rigid cell wall). After estimating the multiple data of nuclear, and cell volumes from the individual literatures, the averaged values were used for analyses and averaged values with standard deviation were shown in graphs. All data used in this study are listed in the electric supplementary material (Tables S1 and S2). The data were divided into three or four categories as multicellular eukaryotes (subdivided with heterotrophic and phototrophic organisms in some cases), unicellular eukaryotes and prokaryotes (including one archaea bacterium). The genome content is assumed as situations after DNA replication and 4N (4-fold of genome size) in eukaryotes and 2N in prokaryotes if the cell cycle information is unavailable. When the cell cycle is described as before S phase (like G1 phase), I assume the

reduced genome content as 2N in eukaryotes. When the genome size in the species remains unanalyzed, the genome content is estimated from the C-values.

Data analysis

For showing graph of scaling relationship, all or categorized data were plotted and fitted with a power-law regression line using Excel software (Microsoft). For understanding the strength of correlation, the coefficient of determination (R^2) was calculated using Excel software. The calculation of standard deviations (S.D.) and statistical difference against scaling exponent were done by regression analysis tool with two-sided Student's *t* test in Excel software using the $\log_{10}$ transformed datasets.

For comparing the correlation with genomic content and exclude spontaneous variation in cell size regardless genome content, I used a single data point in each species as only female gamete, oocyte, in multicellular eukaryotes and data under more normal growth condition without any perturbation of genes in unicellular organisms.

Supplemental figures

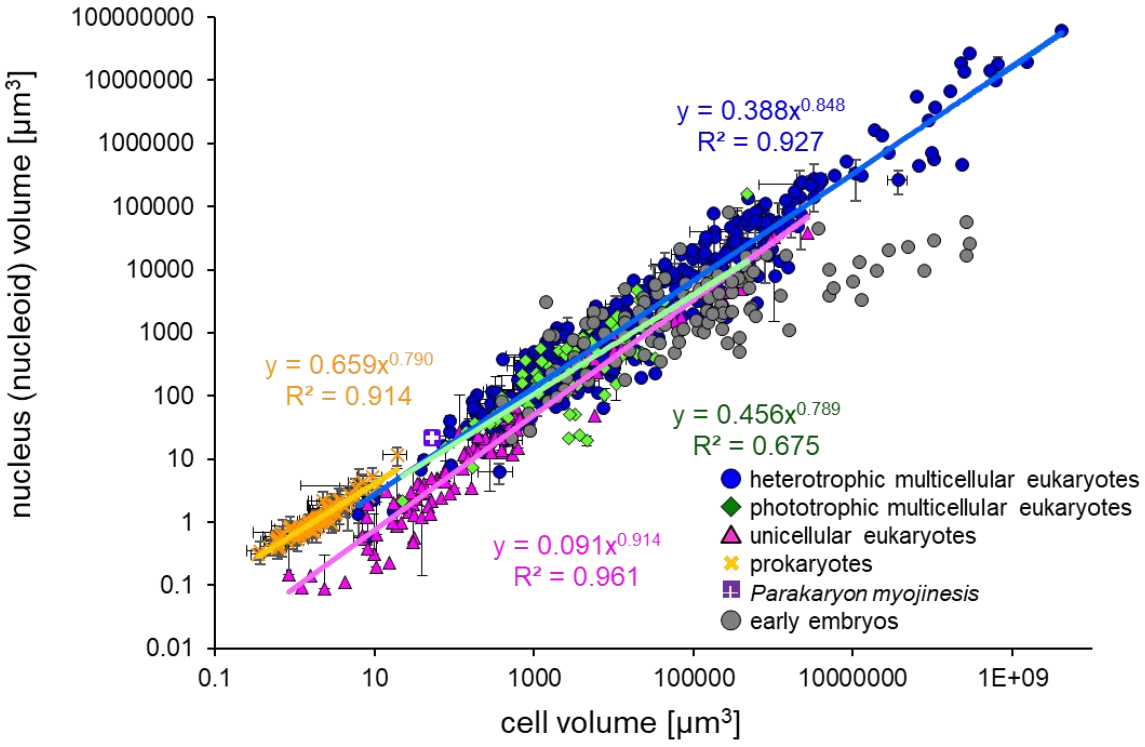

Fig. S1: Size scaling of nucleus volume with cell volume. (A) Size scaling of nucleus volume with cell volume in all data of various cell types. Data of heterotrophic multicellular eukaryotes (blue circles), phototrophic multicellular eukaryotes (green diamonds), unicellular eukaryotes (pink triangles), prokaryotes (orange crosses), and *Parakaryon myojinesis* (purple cross) are identical to Fig. 1. Data from early embryos in muliticellular eukaryotes are represented as grey circles.

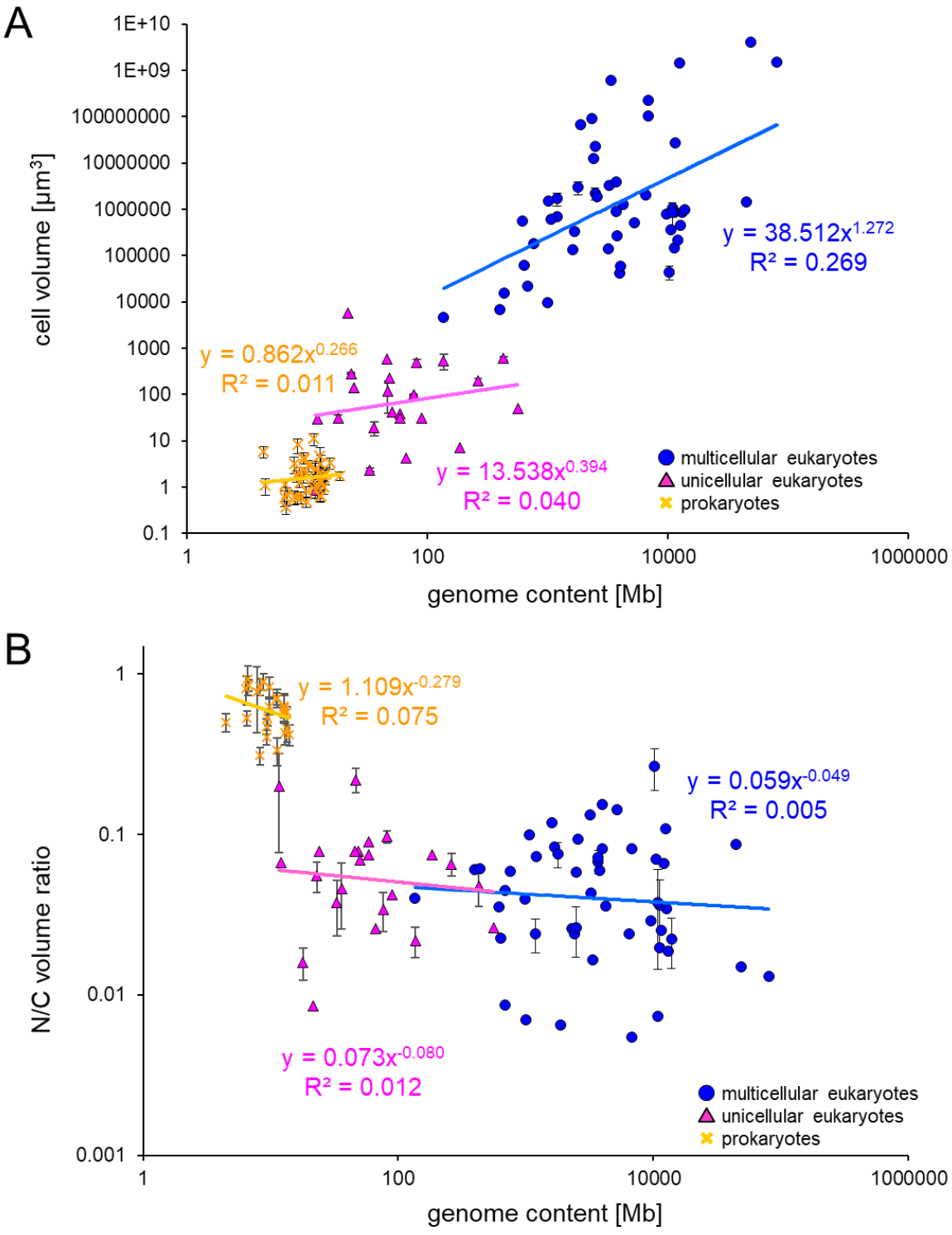

Fig. S2: Scaling relationships with genome content. (A) Size scaling of cell volume with genomic contents in data only including female gamete, which is fully grown oocyte, in multicellular organisms (blue; N = 50) and wild type cells with normal growth condition in unicellular organisms (eukaryotes: pink, N = 23; prokaryotes: orange, N = 40). (B) Size scaling of N/C volume ratio with genomic contents in data only including female gamete in

multicellular eukaryotes and cells under normal growth condition in unicellular organisms. Each species with each DNA ploidy represents one symbol with error bar (S.D.). Data in each category are fitted with a power-law regression line.

Supplemental tables

Table S1. Mean values of GC, CV, NV, N/C volume ratio, and GC/NV ratio

Mean value, standard deviation (S.D.), and sample number (N) of parameters in each condition or cell type were shown. Phylum, species name, cell type (and/or growth condition), and references of images for measurements or numerical data was also shown. The procedures of measurement were categorized as A: genome size listed in literature; B: genome size predicted from C-value, no data: could not find any data for genome size; C: volume calculated as an oval ($V = L/2 \times (W/2)^2 \times PI \times 4/3$ or $= (L/2)^2 \times (W/2) \times PI \times 4/3$; D: volume calculated as a cylindrical shape ($V = (W/2)^2 \times PI \times L$); E: volume calculated as a cuboid ($CV = L^2 \times W$ *or* $CV = L \times W^2$); F: value listed in the literature; G: value calculated from the listed parameter in the literature or estimated from the graph; H: value calculated from our estimated value; ND: no data. Light blue, orange, and green colours in tables represent data from early embryonic development in multicellular eukaryotes (excluded from Figs. 1–3), female gametes (fully-grown oocytes) of multicellular eukaryotes and unicellular organisms under normal growth condition (for Fig. 2B), and erythrocytes (for Fig. 2C), respectively.

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Table S2. All collected data of CV, NV, N/C volume ratio, and GC/NV ratio in each condition

All measured or collected values for parameters were shown in each growth condition or cell type. NV/CV-R and GC/NV-R represent N/C volume ratio and GC/NV ratio, respectively.

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