

Figure S1. Contact-dependent mate recognition allows *C. elegans* males to distinguish species, sex, and developmental stages. Related to Figure 1.

(A) Schematic diagram of male retention assay. (B) Schematic diagram of hermaphrodites with gonad in black, vulva in green, and secreted pheromones in blue. Indicated mutants with defective hermaphrodite-specific cues or tissues are listed and tested in (C). (C) Percentage of male leavers in male retention assay with indicated mutant hermaphrodites or males. (D) Genus of indicated species tested in (E). (E-F) Percentage of male leavers in male retention assay with indicated mutant hermaphrodites or *C. remanei* females. One-way ANOVA with Bonferroni correction and *p* values are indicated. (G-H) Percentage of male leavers in male retention assay with *C. elegans* hermaphrodites at indicated developmental stages and ages. Two-way ANOVA and one-way ANOVA with Bonferroni correction and *p* values are indicated at indicated developmental stages and ages. Two-way ANOVA and one-way ANOVA with Bonferroni correction and *p* values are indicated. For male retention assay, each dot represents the percentage of leavers in a population containing 15-20 males as one replica, and n indicates the number of total animals tested in all replicas. Error bar = S.E.M.



Figure S2. Masculinization and feminization of hypodermis alter body shape. Related to Figure 2.

Fluorescent and DIC images of masculinized hermaphrodites and feminized males by expressing

FEM-3 and TRA-2(IC), respectively. Scale bar = 20 μ m. Arrow, ray. Arrowhead, ray-like

structure



Figure S3. A screen for candidate genes involved in male retention. Related to Figure 2.

(A, B) Percentage of male leavers in male retention assay with mutant hermaphrodites in indicated genotypes. For male retention assay, each dot represents the percentage of leavers in a population containing 15-20 males as one replica, and n indicates the number of total animals tested in all replicas. Error bar = S.E.M. One-way ANOVA with Bonferroni correction and p values are indicated.



Figure S4. A particular group of collagens is necessary for hermaphrodites to retain males. Related to Figure 3.

(A, B) Percentage of male leavers in male retention assay with mutant hermaphrodites in indicated genotypes. (C) Quantification of body length of indicated sex or genotypes at the adult stage. (D) Percentage of male leavers in male retention assay with mutant hermaphrodites or wild-type hermaphrodites treated by RNAi. (E) Quantification of body length of indicated sex or genotypes at the L4 stage. (F) Percentage of *C. elegans* male leavers in male retention assay with *C. briggsae* hermaphrodites in indicated genotypes. For male retention assay, each dot represents the percentage of leavers in a population containing 15-20 males as one replica, and n indicates the number of total animals tested in all replicas. Error bar = S.E.M. One-way ANOVA with Bonferroni correction and *p* values are indicated.



Figure S5. Collagens and surface proteins are generally not required for evoking contact response. Related to Figure 4.

(A) Contact responses of wild-type males with purified cuticles from indicated genotypes. (B)Contact responses of fake worms or *P. pacificus* with purified cuticles for *dpy-7* hermaphrodites.Chi-squared test and *p* values are indicated.



Figure S6. Interior body substances do not induce male avoidance behaviors. Related to Figure 5. (A) Bright-field images of non-bursting mutant hermaphrodites, including the *dpy-5* and *dpy-13* mutants. Scale bar = 1 mm. (B) A schematic diagram of extraction of body substance from the ruptured worms. (C) Percentage of male leavers in male retention assay with hermaphrodites treated with or without supplement of supernatant. (D) Bright-field images of hermaphrodites fixed by 30 minutes/24 hours PFA. We show the body bending pushed by an eyelash to demonstrate different body stiffness. Scale bar = 1 mm. For male retention assay, each dot represents the percentage of leavers in a population containing 15-20 males as one replica, and n indicates the number of total animals tested in all replicas. Error bar = S.E.M. One-way ANOVA with Bonferroni correction and *p* values are indicated.