iScience, Volume *25*

Supplemental information

A lipid transfer protein ensures nematode cuticular impermeability

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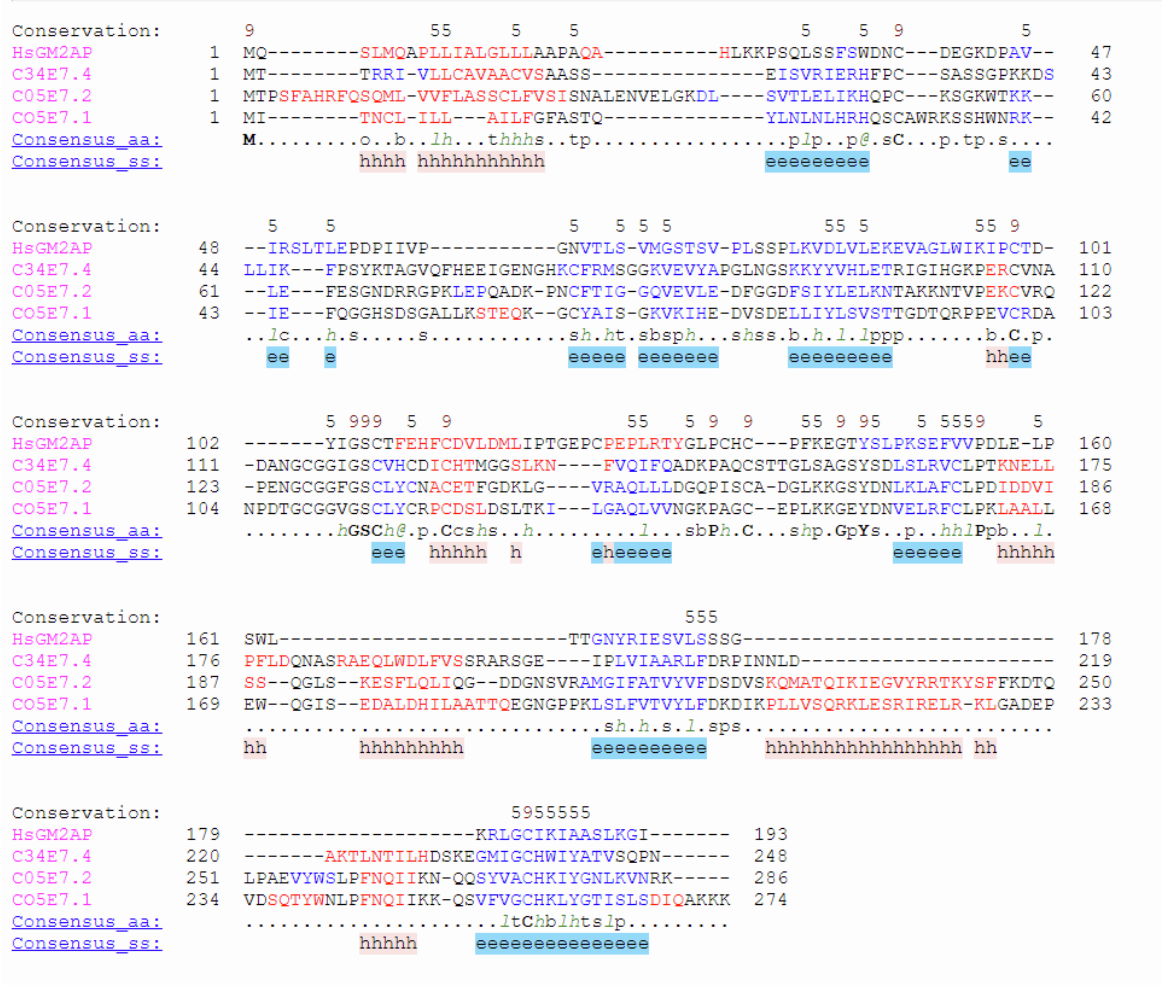
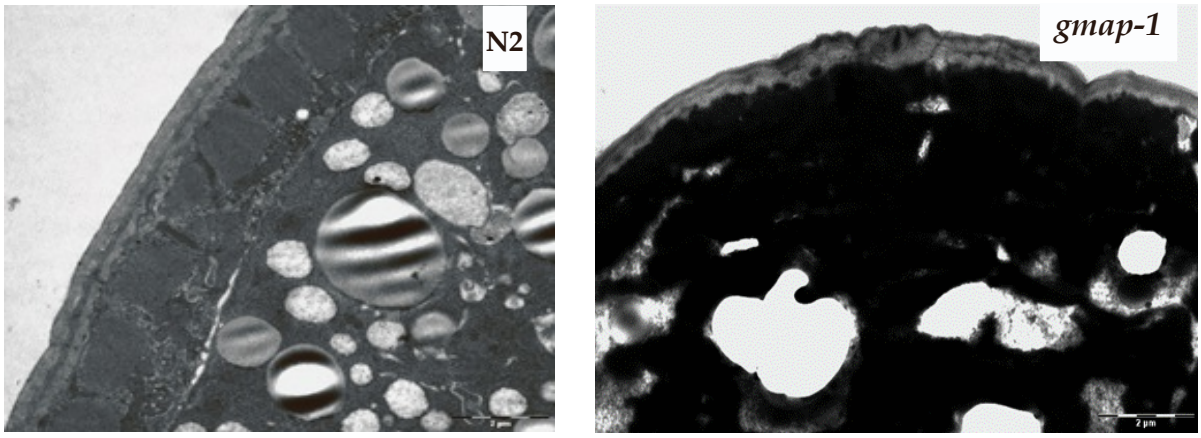
S1 Figure: Alignments of C. elegans GM2AP-like proteins (related to the 1st paragraph of result section).



S1 Figure: Alignment of GMAP-1 protein sequences of *C. elegans* paralogues to the human GM2AP. Protein sequences were retrieved from WormBase or Uniprot and aligned using PROMALS3D **(A)**. HsGM2AP = *Homo sapiens (Uniprot ID,* P17900*)*, C34E7.4 = *Caenorhabditis elegans* (GMAP-1)*,* C05E7.2 = *C. elegans* (GMAP-3)*,* C05E7.1 = *C. elegans* (GMAP-2*)*. Sequence color representation; red = predicted alpha helices, blue = predicted beta strands; Consensus Structure (ss) symbols: e = beta strand, h = alpha helix. Consensus amino acid symbols: conserved amino acids = bold and uppercase letters; aliphatic (I, V, L): *l*; aromatic (Y, H, W, F): @; hydrophobic (W, F, Y, M, L, I, V, A, C, T, H): *h*; alcohol (S, T): o; polar residues (D, E, H, K, N, Q, R, S, T): p; tiny (A, G, C, S): t; small (A, G, C, S, V, N, D, T, P): s; bulky residues (E, F, I, K, L, M, Q, R, W, Y): b; charged (D, E, K, R, H): c. Bold residues in the consensus sequence represent greater than 80% consensus. Numbers in the first row represent a level of conservation above 4.

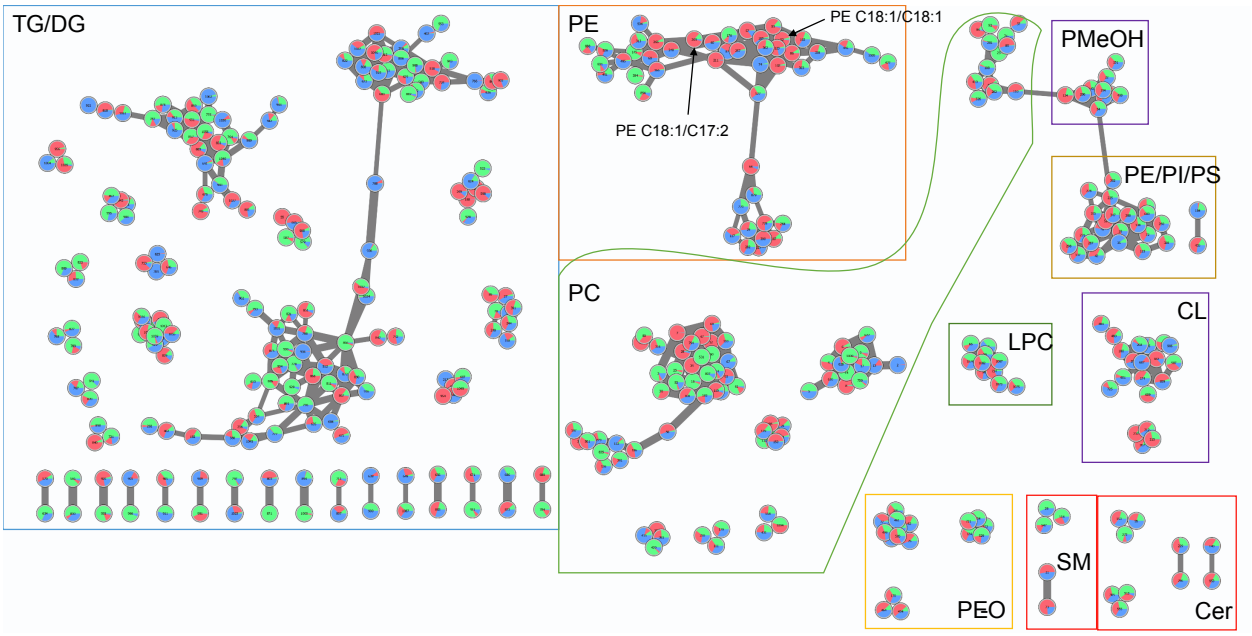
S2 Figure: Electron microscopy analysis of N2 and *gmap-1* (related to Figure 3).



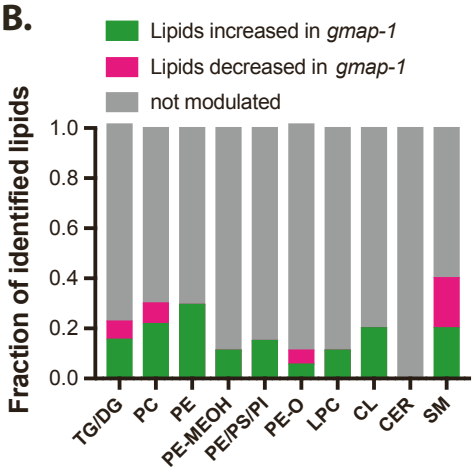
S2 Figure: Electron microscopy analysis of N2 and *gmap-1*. Worms were fixed with glutaraldehyde and formaldehyde in cacodylate buffer prior to osmium staining.

S3 Figure: Lipidomic analysis of the cuticle (related S1 Table).

A.



B.



S3 Figure: (A) Molecular network of lipids extracted from the external layer of the *C. elegans* cuticle and organized by MetGem 1.3.6. Each node represents one lipid identified by LC-MS/MS operated in the positive mode. Clusters are labeled according to the lipids identified in each of them. The relative surface of the node is divided according to the abundance of this lipid in each genotype: green: N2; Red: *gmap-1*; and Blue: *gmap-1* rescued in hypodermis. Two of the most modulated lipids are indicated: PE C18:1/C17:2 and PE C18:1/C18:1 that are increased in the cuticle of *gmap-1* (red) compared to N2 (green) and *gmap-1* rescued in hypodermis (blue). **(B)** For each lipid family, we identified the number of nodes/lipids regulated >30% in *gmap-1* compared to N2 and *gmap-1* rescued in hypodermis. The fraction of nodes upregulated, downregulated or not modulated is represented for each lipid family.

S4 Figure: GMAP-1 Purification analysis (related to Method details).

```
atg acg acc cga aga atc gta ctg ttg tgt gcc gtt gcc gcg tgc gtc tca gcc gcc tcc
 M   T   T   R   R   I   V   L   L   C   A   V   A   A   C   V   S   A   A   S

tcg gag atc tct gtc cgc atc gaa aga cac ttc cca tgc tca gct agt tca ggt cca aag
 S   E   I   S   V   R   I   E   R   H   F   P   C   S   A   S   S   G   P   K

aaa gat tct ttg ctc atc aag ttc cca tca tac aaa acc gcc gga gtt caa ttc cac gaa
 K   D   S   L   L   I   K   F   P   S   Y   K   T   A   G   V   Q   F   H   E

gaa att gga gag aat gga cac aaa tgc ttc aga atg tcc ggt ggc aaa gtt gag gtt tat
 E   I   G   E   N   G   H   K   C   F   R   M   S   G   G   K   V   E   V   Y

gcc cca gga cta aat ggc tcc aag aaa tac tat gtg cat ctt gag aca aga atc gga atc
 A   P   G   L   N   G   S   K   K   Y   Y   V   H   L   E   T   R   I   G   I

cac gga aag cca gaa cga tgc gta aat gct gat gct aat ggg tgt gga gga atc gga tca
 H   G   K   P   E   R   C   V   N   A   D   A   N   G   C   G   G   I   G   S

tgt gtt cac tgc gac atc tgt cac act atg gga gga tct ctc aag aac ttt gtt caa atc
 C   V   H   C   D   I   C   H   T   M   G   G   S   L   K   N   F   V   Q   I

ttc caa gcc gat aag cca gct caa tgc tct acc acg gga ctc tcg gca gga tca tac agc
 F   Q   A   D   K   P   A   Q   C   S   T   T   G   L   S   A   G   S   Y   S

gac ttg tcc ctc cgc gtg tgc ctt cca acc aaa aac gaa ctt ctc cca ttc ctc gat caa
 D   L   S   L   R   V   C   L   P   T   K   N   E   L   L   P   F   L   D   Q

aac gcc tct cgt gcc gag cag ctc tgg gat ctc ttt gtc agc tcc cgt gct cgt tcc ggt
 N   A   S   R   A   E   Q   L   W   D   L   F   V   S   S   R   A   R   S   G

gag atc cca ctt gtc att gcc gcc cgt ctt ttc gat aga cca atc aat aat ttg gat gcc
 E   I   P   L   V   I   A   A   R   L   F   D   R   P   I   N   N   L   D   A

aag aca ttg aac act att ctt cac gac tcg aag gaa gga atg atc gga tgc cac tgg atc
 K   T   L   N   T   I   L   H   D   S   K   E   G   M   I   G   C   H   W   I

tac gcg acg gtg tcc cag cca aac gaa ttc tct aga gaa aac ctg tac ttc caa gcg cac
 Y   A   T   V   S   Q   P   N   E   F   S   R   E   N   L   Y   F   Q   A   H

cat cac cat cac cat cat cac tga
 H   H   H   H   H   H   H   -

TGAACTAGTGCCTGCAGTCTCGACA
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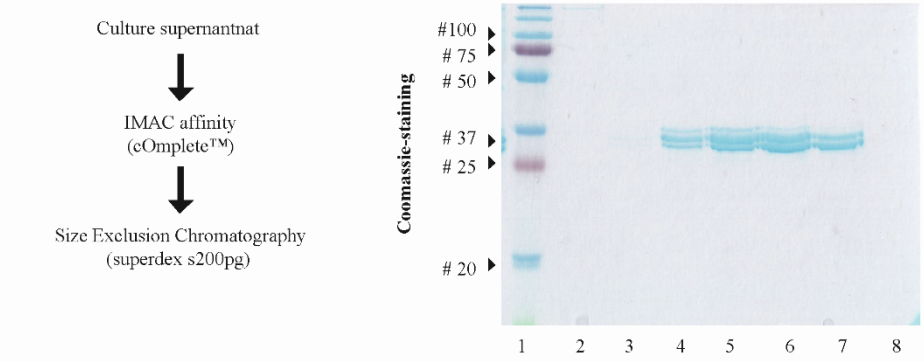
B)



S4 Figure: (**A)** Coding region and translated amino acid sequence of GMAP-1. (**B)** SDS-PAGE analysis of 8X His purified GMAP-1. Elution was performed using increasing concentrations of imidazole including 35 mM (lanes 2 and 3), 100 mM (Lanes 4 and 5) and 500 mM (lanes 6-8). Upper band represents glycosylated species while lower band represents de-glycosylated species.