### APPENDIX

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### **Appendix Figure S1**

Representative transmission electron microscopy images from serial cross sections of the hermaphrodite amphid sensory pore in WT, *rabs-5(ok1513)* and *vps-45(tm0246)* worms. The entire pore is shown in the large panels; individual cilia are shown at higher magnifications in the small panels. Red outline denotes the PCMCs. The schematics show the the section positions (colour coded) and the ultrastructure phenotypes observed. For simplicity, the cartoons of the pores in longitudinal orientation only show 3 out of the 10 channel axonemes. PCMC; periciliary membrane compartment. sMT; singlet microtubules.

### **Appendix Figure S2**

**A.** Representative transmission electron microscopy images from serial cross sections of the male-specific cephalic (CEP and CEM) sensory pore in WT, *rabs-5(ok1513)* and *vps-45(tm0246)* worms. CEM and CEP cilia outlined in pink and yellow, respectively. Neighbouring OLQ cilium outlined in green. PCMC; periciliary membrane compartment. Left images show the entire pore; smaller right-hand images show indicated cilia at higher magnifications. Blue arrowheads denote vesicles in the PCMC of CEM cilia. Yellow arrowheads denote extracellular vesicles in the sensory pore lumen. Schematics show the

CEP, CEM and OLQ cilia and associated pores in longitudinal and cross section, and indicate the ultrastructural phenotypes observed, as well as the section positions (colour coded). Note that whilst the proximal and middle (doublets and singlets; (Silva et al. Curr Biol. 2017 Apr 3; 27(7):968-980)) portions of the mutant CEM cilia are normal, we cannot tell from our images if the distal portion is normal or not in the mutants.

**B.** Representative transmission electron microscopy images from serial cross sections of the hermaphrodite inner labial sensory pore in WT, *rabs-5(ok1513)* and *vps-45(tm0246)* worms. IL1 cilium outlined in dark blue. IL2 cilium outlined in light blue.

**Appendix Table S1** List of *C. elegans* genes and corresponding alleles screened for cilium structure and function defects (**Fig 1A**).

Appendix Table S2 C. elegans strains used in this study.

**Appendix Table S3.** Primer used in this study to genotype by PCR the indicated *C. elegans* mutant alleles.

# Appendix Figure S1, related to Figure 4



WT amphid pore



rabs-5 and vps-45 amphid pore



distal segment



B w

Image: second second

e PCMC vesicles increase

Appendix Figure S2, related to Figure 4

### Appendix Table S1:

List of C. elegans genes and corresponding alleles screened for cilium structure and function defects

C. elegans gene	allele	human orthologue	
arf-1.2	ok796	ARF1	ADP ribosylation factor 1
asm-2	gk293929	SMPD1	sphingomyelin phosphodiesterase 1
cav-1	ok2089	CAV1	caveolin 1
cav-2	hc191	CAV2	caveolin 2
dgk-1	ok1462	DGKQ	diacylglycerol kinase theta
eea-1	ok1040	EEA1	early endosome antigen 1
gta-1	ok517	GABAT	4-aminobutyrate aminotransferase
hyl-1	ok976	CERS6	ceramide synthase 6
hyl-2	ok1766	CERS4	ceramide synthase 4
M01E5.2	ok2552	MTG2	mitochondrial ribosome associated GTPase 2
mec-2	e75	STOM	stomatin
mtm-6	ok330	MTMR6	myotubularin related protein 6
mtm-9	ok3523	MTMR9	myotubularin related protein 9
mtr-4	ok2642	SKIV2L2	Ski2 like RNA helicase 2
nex-1	gk148	ANX13	annexin A13
nex-2	ok764	ANX7	annexin A7
nex-3	gk385	ANX5	annexin A5
nex-4	gk102	ANX6	annexin A6
num-1	bc365	NUMBL	NUMB like, endocytic adaptor protein
prmn-1	gk545	PROM1	prominin 1
rab-10	ok1494	RAB10	RAB10
rabn-5	ok1555	RABPT5	rabaptin-5, RAB GTPase binding effector protein 1
rabs-5	ok1513	RABSN	rabenosyn, RAB effector
rabx-5	tm1215	RABGEF1	rabex5, RAB guanine nucleotide exchange factor 1
ric-19	ok833	ICA1	islet cell autoantigen 1
ric-4	md1088	SNAP25	synaptosome associated protein 25
ric-8	md1909	RIC8A	RIC8 guanine nucleotide exchange factor A
rme-1	b1045	EHD1	EH domain containing 1
rme-2	b1008	LRP2	LDL receptor related protein 2
rme-6	b1014	GAPVD1	GTPase activating protein and VPS9 domains 1
rme-8	b1023	DNAJC13	DnaJ heat shock protein family (Hsp40) member C13
sax-7	ok1244	NRCAM	neuronal cell adhesion molecule
stn-2	ok2417	SNTG2	syntrophin gamma 2
syx-7	gk338314	STX12	syntaxin 12
tat-3	gk819359	ATP10A	ATPase phospholipid transporting 10A
tat-6	ok1984	ATP9B	ATPase phospholipid transporting 9B
tin-9.1	ok2194	TIMM9	translocase of inner mitochondrial membrane 9
tmem-135	ok1646	TMEM135	transmembrane protein 135
unc-1	e94	STOM	stomatin
unc-24	e138	STOML1	stomatin like 1
unc-44	e362	ANK1	ankyrin 1
vps-45	tm0246	VPS45	vacuolar protein sorting 45 homolog
vps-54	ok1463	VPS54	VPS54, GARP complex subunit
wdfy-2	ok3592	WDFY2	WD repeat and FYVE domain containing 2

#### Appendix Table S2: C. elegans strains used in this study

N2 (Bristol) him-5(e1490) osm-5(p813) bbs-8(nx77) arf-1.2(ok796) asm-2 (gk293929) cav-1(ok2089) cav-2(hc191) dgk-1(ok1462) eea-1(ok1040) gta-1 (ok517) hyl-1(ok976) hyl-2(ok1766) M01E5.2 (ok2552) mec-2(e75) mtm-6 (ok330) mtm-9 (ok3523) mtr-4 (ok2642) nex-1(gk148) nex-2(ok764) nex-3(gk385) nex-4(gk102) num-1(bc365) prmn-1(gk545) rab-10(ok1494) rabn-5(ok1555) rabs-5(ok1513) rabx-5(tm1215) ric-19(ok833) ric-4(md1088) ric-8(md1909) rme-1(b1045) rme-2(b1008) rme-6(b1014) rme-8(b1023) sax-7(ok1244) stn-2(ok2417) syx-7 (gk338314) tat-3(gk819359) tat-6(ok1984) tin-9.1(ok2194) tmem-135 (ok1646) unc-1(e94) unc-24(e138) unc-44(e362) vps-45(tm0246) vps-54 (ok1463) wdfy-2(ok3592) N2;oqEx306[rabs-5p::gfp + unc122p::dsRed] rabs-5(ok1513;oqEx307[rabs-5p::rabs-5 + unc122p::dsRed] vps-45 (tm0246); oqEx308[arl-13p::vps45 + unc122p::dsRed] rabs-5(ok1513);oqEx401[arl-13p::rfp::rabs-5(WT) + unc122p::dsRed] rabs-5(ok1513);oqEx402[arl-13p::rfp::rabs-5(ΔZN) + unc122 p::dsRed] rabs-5(ok1513);oqEx403[arl-13p::rfp::rabs-5(ΔFYVE) + unc122p::dsRed] rabs-5(ok1513;oqEx404[arl-13p::rfp::rabs-5(ΔRBD) + unc122p::dsRed] N2;oqEx405[arl-13p::rfp::rabs-5(WT) + unc122p::gfp] N2;oqEx406[arl-13p::rfp::rabs-5(ΔZN) + unc122p::gfp]

N2;oqEx407[arl-13p::rfp::rabs-5(ΔFYVE) + unc122p::gfp]

N2;oqEx408[arl-13p::rfp::rabs-5(ΔRBD) + unc122 p::gfp] N2;oqEx409[arl-13p::vps-45::gfp + unc122 p::dsRed] N2;oqEx410[arl-13p::gfp::rab-5 + unc122p::dsRed] N2;oqEx411[arl-13p::wdfy-2::gfp + unc122p::dsRed] N2;oqEx412[arl-13p::rfp::eea-1 + unc122p::gfp] N2;oqEx413[arl-13p::dpy-23::gfp + unc122p::dsRed] N2;oqEx414[arl-13p::gfp::cav-1 + unc122p::dsRed] him-5(e1490;oqEx415[pkd-2p::rfp::rabs-5(WT) +unc122p::dsRed] him-5(e1490;oqEx416[pkd-2p::rfp::cav-1 +unc122p::dsRed] him-5(e1490;oqEx417[pkd-2p::wdfy-2::rfp +unc122p::dsRed] N2;oqEx418[str-2p::jbts-14::gfp + str-2::rfp + unc122p::gfp] N2;oqEx204[arl-13p::gfp::rab-5(Q78L) + rol-6(su1006)] N2;oqEx205[arl-13p::gfp::rab-5(S33N) + rol-6(su1006)] him-5(e1490);myls1[pkd-2::gfp + unc-122p::gfp] IV N2;oqEx58[arl-13p::arl-13::gfp + rol-6(su1006)] N2;oyEx[srbc-66::gfp + unc122 p::dsRed] N2;nxEx[tram-1::tdTomato + bbs-8p::mks-2::gfp + rol-6(su1006)] lin-15(n765;kyls141[osm-9::gfp + lin-15(+)] N2;nxEx289[rpi-2::gfp + xbx-1::tdTomato + rol-6(su1006)] N2;kyls104[str-1p::gfp] N2;mnls17[osm-6p::osm-6::gfp]

primer name	primer sequence	allele genotyped
cav-1_F	AGGCGAAGTTACATTAGGCTG	cav-1(ok2089)
cav-1_R	GCACTCTTTCTACGCAATCG	cav-1(ok2089)
cav-1_Pn	TGTGTCAGTAGGGAAATCGC	cav-1(ok2089)
eea-1_F	ATTTAAGCGGAACAGCCTCA	eea-1(b1040)
eea-1_R	CTTCCAATTCTCTTGCTCG	eea-1(b1040)
eea-1_Pn	GAAATCAGAGGGAATCCGG	eea-1(b1040)
rabs-5_F	GATTGGAGCAACTGGTTCAG	rabs-5(ok1513)
rabs-5_R	ATGATCCGTTTGTATTGTATCATC	rabs-5(ok1513)
rabs-5_Pn	CGGTAAAATTTCATCAGGAG	rabs-5(ok1513)
rabx-5_F	CGCTTAAGGAGAGTGGTGA	rabx-5(tm1215)
rabx-5_R	ATTCTTCGCAAAATCCTGAC	rabx-5(tm1215)
rabx-5_Pn	CTCGCCATGCTTATCTGAC	rabx-5(tm1215)
rme-1_F	CTTATCCGGAGAAAAGCAAA	rme1(b1045)
rme-1_R	TACCATGAAGCAGTCGACC	rme1(b1045)
rme-1_Pn	GAATGATCCCAAATAGACACG	rme1(b1045)
rme-6(WT)_F	GCAATCATCAGTCCGC	rme-6(b1014)
rme-6(b1014)_F	GGCAATCATCAGTCCGT	rme-6(b1014)
rme-6_R	TTTCTAAAACTGACGTCCCTC	rme-6(b1014)
vps-45_F	AATGCAAATTCTCCTGCTCT	vps-45(tm0246)
vps-45_R	TTCAACTGCAACAATAGCG	vps-45(tm0246)
vps-45_Pn	ACAGTGTCATGCGCATTT	vps-45(tm0246)
wdfy-2_F	ATGATAGATCCGTTCGCCTG	wdfy-2(ok3592)
wdfy-2_R	AGAAGGGGCAGAGCAAAC	wdfy-2(ok3592)
wdfy-2_Pn	GAAAGTGGTCGGATTGAGAC	wdfy-2(ok3592)

## Appendix Table S3\_Primer used for genotyping the indicated mutant allele