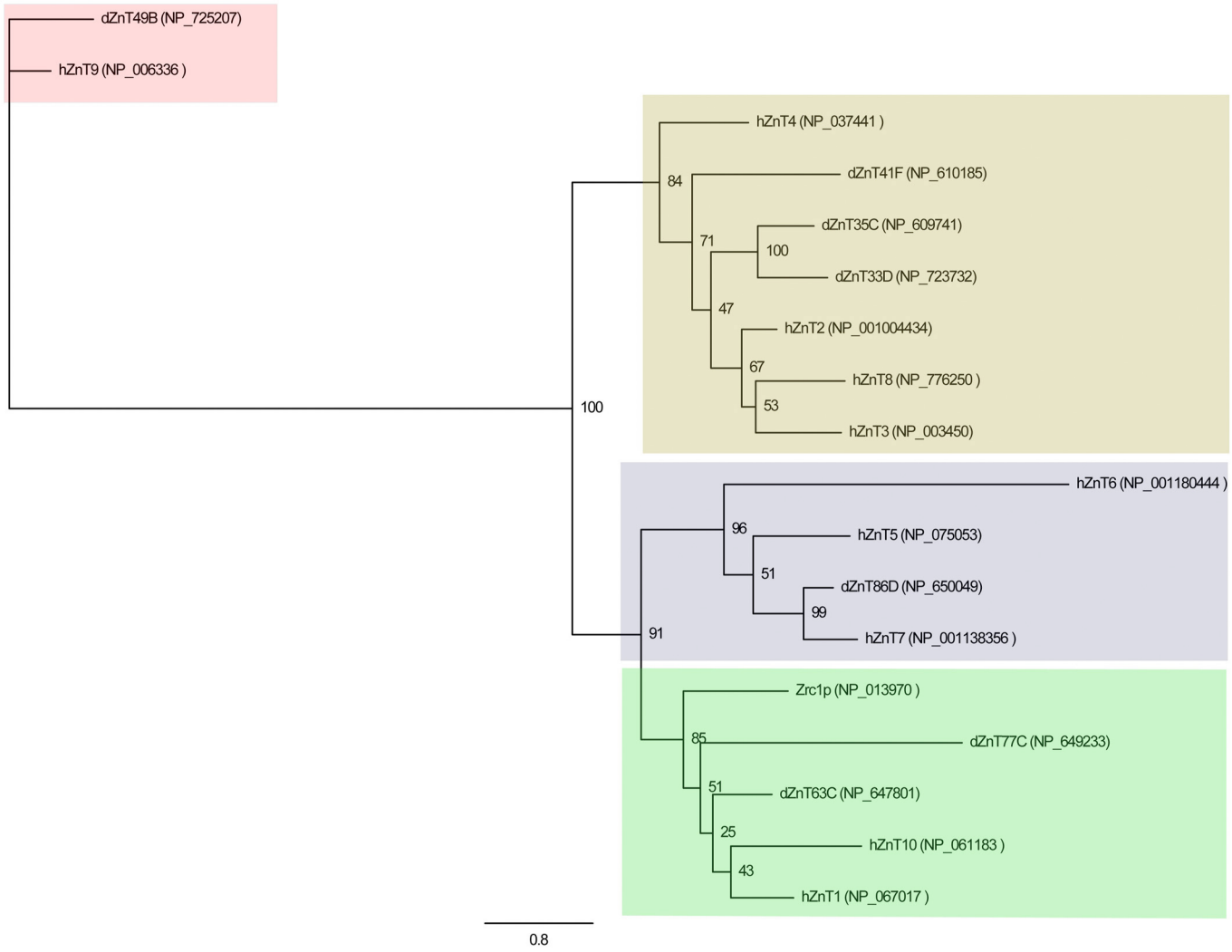

Fig. S1. Amino acid sequence comparison between Drosophila, mammalian and yeast ZnT proteins. Tree was generated using MUSCLE sequence alignment software and displayed with FigTree. The scale bars represent amino acid substitutions per amino acid site. Bootstrap values are included at nodes. Accession numbers are listed for each protein sequence used for the alignment. Proteins that display homology and group into clades are highlighted.

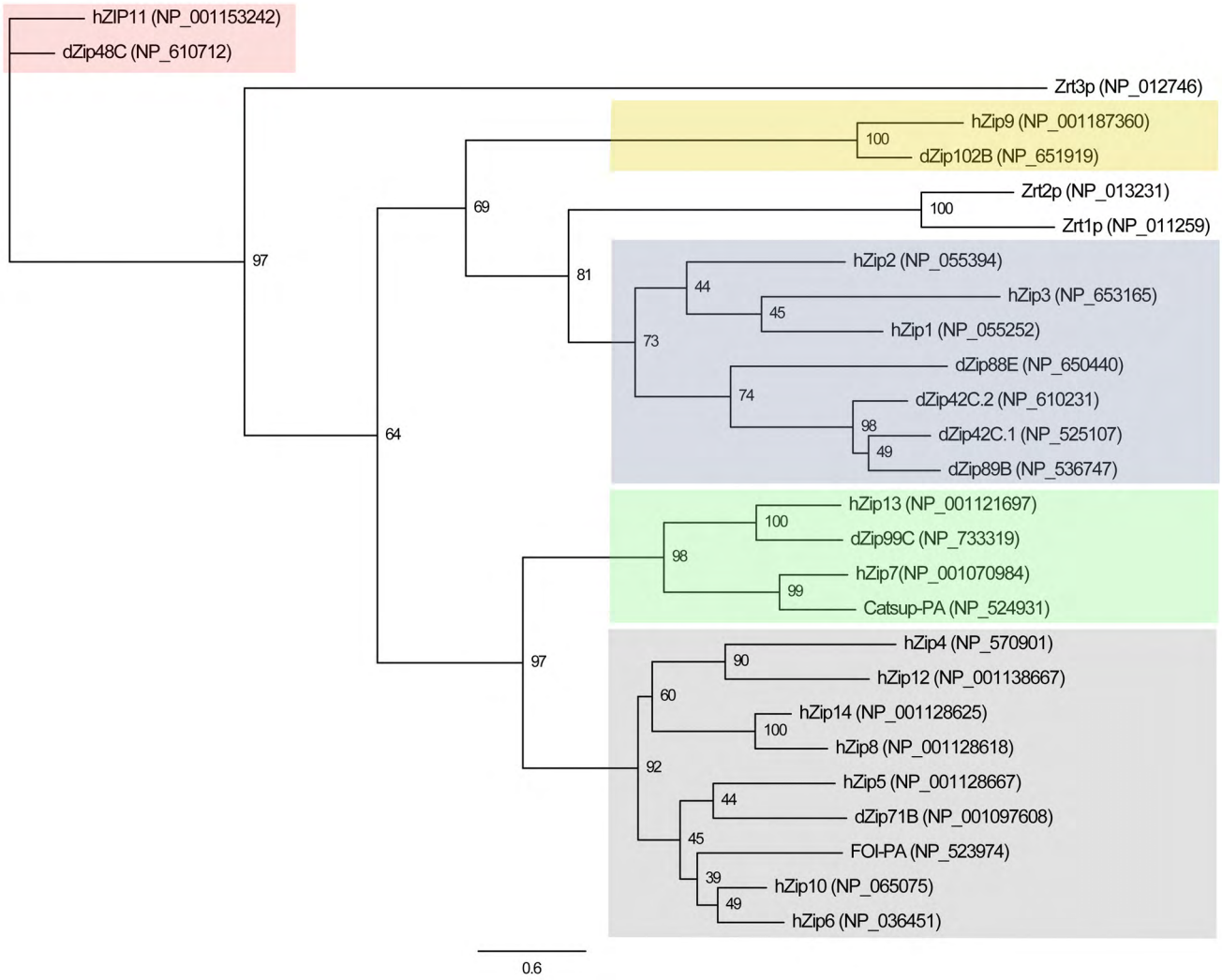

Fig. S2. Amino acid sequence comparison between Drosophila, mammalian and yeast Zip proteins. Tree was generated using MUSCLE sequence alignment software and displayed with FigTree. The scale bars represent amino acid substitutions per amino acid site. Bootstrap values are included at nodes. Accession numbers are listed for each protein sequence used for the alignment. Proteins that display homology and group into clades are highlighted.

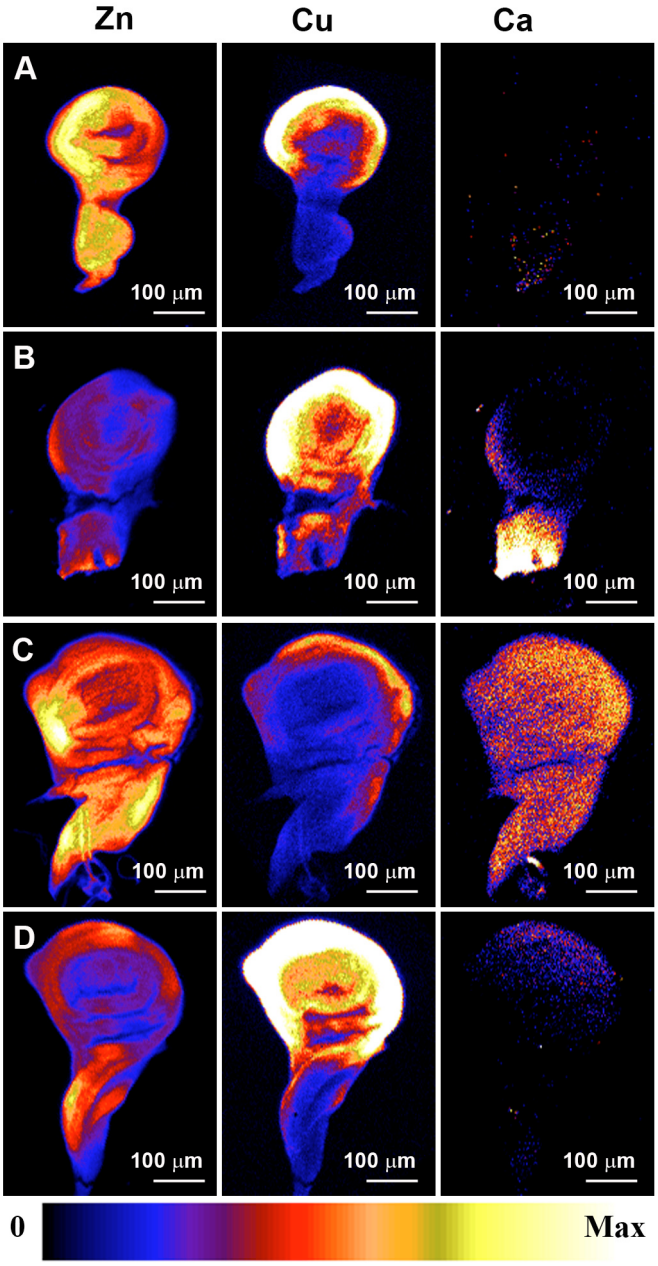

Fig. S3. Manipulation of zinc transporter gene expression alters spatial distribution of metals *in vivo*. XRF analysis of third instar wing imaginal discs, showing distribution of zinc (left panel), copper (middle panel) and calcium (right panel). Distribution is displayed as a heat map with the relative concentration shown at the bottom of the figure. For imaginal discs shown in panels A–D element visualization is normalized so that maximum values are as follows: zinc 264 ng cm^{-2}, copper 59 ng cm^{-2}, calcium 264 ng cm^{-2}. Genotypes are: (A) Wild-type control disc; (B) *pannier>dZip42C.1*^{flag}; (C) *pannier>dZnT63C*^{RNAi}; (D) *pannier>dZip42C.1*^{eGFP}*;dZnT63C*^{RNAi}.

Table S1. Summary of *Drosophila* zinc transporter genes, isoforms, mammalian homologues and expression patterns

Gene	Synonyms	Isoforms	Mammalian homologue	Sequence similarity	Temporal expression profile	Tissue expression profile
cg11163 (FBgn0025693)	dZnT41F	4	hZnT2 hZnT3 hZnT8	I: 36.5%, S: 61.0% I: 43.0%, S: 64.8% I: 46.9%, S: 72.7%	All stages (moderately high – high)	Larval and adult mid/hindgut and malphigian tubules, female spermatheca
cg17723 (FBgn005432)	dZnT1, dZnT63C	5	hZnT1 hZnT10	I: 40.5%, S: 57.6% I: 38.3%, S: 57.3%	All stages (moderately high – high)	Larval malphigian tubules and salivary glands
cg31860 (FBgn0051860)	dZnT33D	2 *	hZnT2 hZnT3 hZnT8	I: 44.1%, S: 63.2% I: 41.2%, S: 62.2% I: 40.3%, S: 59.7%	Pupae and adult male (moderate)	Adult testis
cg3994 (FBgn0028516)	dZnT35C	2 *	hZnT2 hZnT3 hZnT8	I: 49.7%, S: 68.4% I: 37.0%, S: 53.9% I: 36.2%, S:54.4%	All stages (moderate – moderately high)	Malphigian tubules
cg5130 (FBgn0037000)	dZnT77C	2	hZnT1 dZnT10	I: 38.0%, S: 59.9% I: 28.8%, S: 52.2%	00–06 and 18–24h embryo and L3 stage (moderately high)	Malphigian tubules and male accessory gland
cg6672 (FBgn0037875)	dZnT86D	1	hZnT5 HznT6 hZnT7	I: 48.7%, S:65.7% I: 61.3%, S:74.9%	All stages (moderate – high)	Salivary glands, adult maphigian tubules, female spermatheca, male accessory gland
cg8632 (FBgn0033762)	dZnT49B	2	hZnT9	I: 53.4%, S: 71.9%	All stages (moderate) 00–04h embryo and adult females 5–30 days (high)	Ubiquitous expression
cg4334 (FBgn0038312)	dZip88E	1	hZip3 hZip1 hZip2	I: 34.5%, S:52.6% I: 28.3%, S: 47.2% I: 31.9%, S: 52.9%	12–24h embryo and L3 larval stage (moderately high)	Larval salivary gland
cg9430 (FBgn0033097)	dZip42C.2	1	hZip3 hZip1 hZip2	I: 33.1%, S: 56.8% I: 27.2%, S: 50.7% I: 31.4%, S: 60.3%	adults <5 days (moderate)	Adult midgut
cg9428 (FBgn0033096)	Zinc/iron regulated transporter-related protein 1 (dZip1), dZip42C.1	1	hZip3 hZip1 hZip2	I:30.8%, S:46.8% I: 28.7%, S: 47.7% I: 39.5%, S: 61.2%	00–06h embryo and L1 to early L3 larval stage (v.low) adult (v.low) 30 day old adult female (low)	Larval midgut and trachea, low ubiquitous expression
cg6898 (FBgn0038412)	Zinc/iron regulated transporter-related protein 3 (dZip3), dZip89B	1	hZip3 hZip1 hZip2	I:35.5%, S: 58.7% I: 30.4%, S: 51.8% I: 38.3%, S: 60.2%	14h embryo onwards (moderate – moderately high)	Larval/adult hindgut and malphigian tubules, adult head
cg6817 (FBgn0024236)	fear of intimacy (foi)	1	hZip6 hZip10	I: 33.9%, S: 51.9% I: 35.4%, S: 55.9%	00–10h and 14–20h embryo (moderately high – high) L1 and prepupae (moderately high)	Ubiquitous expression
cg7816 (FBgn0039714)	dZip99C	9	hZip13	I: 46.2%, S: 59.8%	All stages (moderately high – high)	Larval and adult midgut, testis and male accessory gland
cg10449 (FBgn0002022)	catchecolamines up (catsup)	1	hZip7	I: 52.0%, S: 65.3%	All stages (moderately high – high)	Ubiquitous expression
cg10006 (FBgn0036461)	dZip71B	1	hZip5	I: 35.9%, S: 51.6%	14h embryo onwards (moderate – moderately high)	Larval and adult malphigian tubules
cg2177 (FBgn0039902)	dZip102B	3 *	hZip9	I: 53.2%, S: 70.9%	All stages (moderate – moderately high)	Larval/adult malphigian tubules, larval fat bodies, adult crop, male accessory gland
cg13189 (FBgn0033665)	dZip48C	1	hZip11	I: 54.3%, S: 66.0%	All stages (moderate – moderately high)	Larval and adult malphigian tubules

Putative zinc transport genes identified in the *Drosophila* genome, associated mammalian homologues and expression profiles. Blastp identified 17 putative zinc transport genes in the *Drosophila* genome. Many of these genes have not been characterized; however, synonyms have been provided for all putative zinc transport genes listed. Most isoforms differ in the 5′ and 3′ UTRs; however, isoforms that are alternatively spliced within coding regions are denoted with an asterisk. Many *Drosophila* zinc transport proteins have several possible mammalian homologues. Temporal expression profiles were sourced from modENCODE whilst tissue expression profiles were obtained from the FlyAtlas Anatomical Expression Data set. Only the highest temporal and spatial expression data are included.

Table S2. Oligonucleotides used to clone *Drosophila* zinc transporter genes

Zinc transporter	Forward primer (5′–3′)	Reverse primer (5′–3′)
dZnT41F	ATGCCAAAATATCAGAAGCT	GACTCTGTTGTAGGTTCCAATTG
dZnT63C	ATGGCCAAGTACTCGGGC	AACCAAATCGCTCTCGGCG
dZnT33D	ATGAGTAAAAAATCGCTT	TTCAGATCCCGAGTGATTCT
dZnT35c	ATGTCCAGGAACGAGGATACAC	CTTCTCGGGCACATTGCAC
dZnT77C	ATGGTAAAGGATATTCTCCAGCGG	ATCGCTGCTCGTTGGCCC
dZnT86D	ATGATACCGCTGTCGCTGT	AACATAATCCAACTGTATGTAGAT
dZip88E	ATGAGCTTGCCAACCGAC	ATCCTCATCAGAAAGATA
dZip42C.2	ATGGTAGACCAACACTTAATCG	TTTGGGCTTTTTAGTATGC
dZip42C.1	ATGAGCGCTACCGCAA	GGAACAGGTTAGGCTGTCA
dZip89B	ATGAATCAAACGCAAGTAAATAATTTCC	GGCGTCCTTCTTGGGGTG
foi	ATGGCGCGTCACATAATG	GTGCGCGTGCTGGTG
dZip99C	ATGACCACGAACAGCAGCTTC	GTGTTCGAATAGCATGGTCATCAC
Catsup	ATGGCCAAACAAGTGGCTGA	CTCGAACTTGGCGATAACGATCATT