Supplementary Figure Legends

Fig. S1. Alignment of *dPZLF*, PLZF and EOR-1.

(a) The BTB/POZ domain of dPLZF is similar to the domain of human PLZF, *Drosophila* kelch and *C. elegans* EOR-1. Conserved regions are boxed. Shaded areas indicate identical residues. **(b)** Alignment of zinc finger motifs regions of dPLZF, PLZF and EOR-1. The C_2H_2 zinc-finger motifs are underlined. Cysteines and histidines are indicated with dots.

Fig. S2. High level of *dPLZF* mRNA expression can be detected in embryo, larva and pupa stages but low in adult stage.

(a) RT-PCR analysis of *dPLZF* expression in wild-type flies showed a single band of 1 kb transcript. Total RNA from various developmental stages of *Drosophila* was reverse transcribed, and PCR amplification was performed for 35 cycles with specific primers for *dPLZF* or ribosomal protein (*rp49*). *rp49* was used as a loading control. **(b)** Real-time PCR analysis of *dPLZF* expression in various developmental stages. Adult was used as a standard of relative expression.

Fig. S3. Genetic phenotypes by double expressed dPLZF induced the extra wing veins under *en*-Gal4 or MS1096-Gal4. (a) Double expression of dPLZF in *en*-Gal4 flies produced several extra wing veins. **(b)** Double expression of dPLZF in MS1096-Gal4 flies produced the strong extra wing veins. We checked individually more than 200 *Drosophila* wings. Flies were grown at 25℃.

Fig. S4. Genetic phenotypes by Draf/ERK reduced the interaction with dPLZF RNAi or dPLZF haplodeficiency.

(a) Wing of wild type fly **(b)** Overexpression of dPLZF in MS1096-Gal4 flies produced several extra wing veins. **(c)** dPLZF overexpression phenotype was mostly suppressed by dPLZF RNAi. **(d)** Quantification of flies with extra wing vein phenotype for indicated genotypes (from (a) to (c)). We checked more than 200 *Drosophila* wings for each genotype. **(e)** Wing of Draf overexpression fly using *vg*-Gal4. **(f, g)** Draf activity was completely suppressed by *dPLZF* haplodeficiency. **(h)** Wing of ERK^{s} overexpression fly using *en*-Gal4. **(i, j)** ERK^{s} activity was completely suppressed by *dPLZF* haplodeficiency. **(k)** Quantification of flies with extra wing vein phenotype for indicated genotypes (from (e) to (j)). We checked more than 200 *Drosophila* wings for each genotype. Flies were grown at 25℃.

Fig. S1.

a


dPLZF 28 QRRTGQFCDLLLELDSDDSLSLSVHFCVLAAQSQFINSNQKQQQFSIH-----NP-
PLZF 27 MRLAGTLCDVVIMVDS---QEFHAHRTVLACTSKMFEILFH--------R----N-S
EOR-1 35 QRKTGRFCDVEIVVQN---KSFAAHRNILAAHSPYFDAIFK--------YCKIT--K
kelch 180 MRKQKQLCDVILVADD---VEIHAHRMVLASCSPYFYAMFT--------SFEESR-Q
dPLZF 78 LKITIRNFSCTQCLHTIVDFFYEDLVSVSKEHELHFRELAQILAVTEL
PLZF 68 QHYTL-DFLSPKTFQQILEYAYTATLQAKAEDLDDLLYAAEILEIEYL
EOR-1 79 EQLTI-NSKSPQVFELFLNYMYSGTVIIDRSSVSELLRFANNFLIVKL
kelch 225 ARITL-QSVDARALELLIDYVYTATVEVNEDNVQVLLTAANLLQLTDV

b

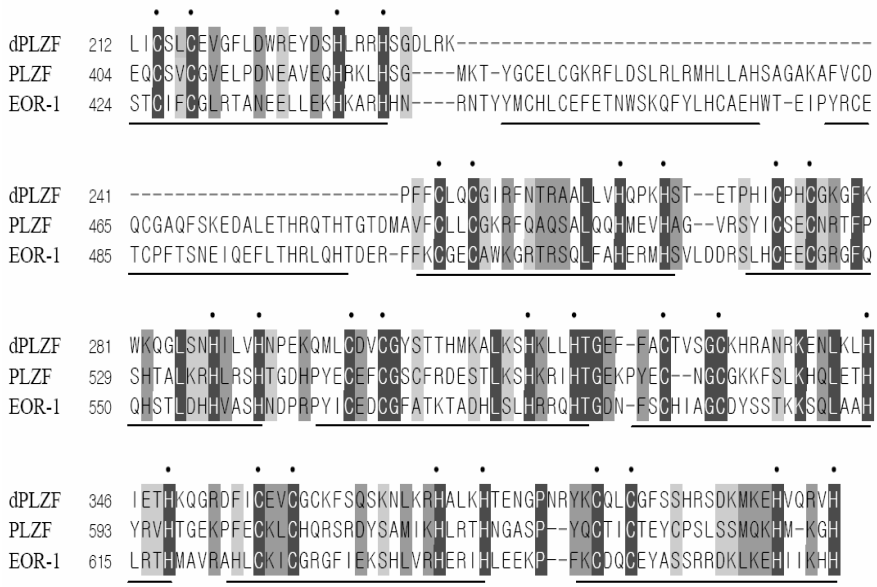

dPLZF 212 LICSLCEVGFLDWREYDSHLRRHSGDLRK----------
PLZF 404 EQCSVCGVELPDNEAVEQHRKLHSG----MKT-YGCELCGKRFLDSLRLRMHLLAHSAGAKAFVCD
EOR-1 424 STCIFCGLRTANEELLEKHKARHHN----RNTYYMCHLCEFETNWSKQFYLHCAEHWT-EIPYRCE
dPLZF 241 ----------PFFCLQCGIRFNTRAALLVHQPKHST--ETPHICPHCGKGFK
PLZF 465 QCGAQFSKEDALETHRQTHTGTDMAVFCLLCGKRFQAQSALQQHMEVHAG--VRSYICSECNRTFP
EOR-1 485 TCPFTSNEIQEFLTHRLQHTDER-FFKCGECAWKGRTRSQLFAHERMHSVLDDRSLHCEECGRGFQ
dPLZF 281 WKQGLSNHILVHNPEKQMLCDVCGYSTTHMKALKSHKLLHTGEF-FACTVSGCKHRANRKENLKLH
PLZF 529 SHTALKRHLRSHTGDHPYECEFCGSCFRDESTLKSHKRIHTGEKPYEC--NGCGKKFSLKHQLETH
EOR-1 550 QHSTLDHHVASHNQPRPYICEDCGFATKTADHLSLHRRQHTGDN-FSCHIAGCDYSSTKKSQLAAH
dPLZF 346 IETHKQGRDFICEVCGCKFSQSKNLKRHALKHTENGPNRYKCQLCGFSSHRSDKMKEHVQRVH
PLZF 593 YRVHTGEKPFECKLCHQRSRDYSAMIKHLRTHNGASP--YQCTICTEYCPSLSSMQKHM-KGH
EOR-1 615 LRTHMAVRAHLCKICGRGFIEKSHLVRHERIHLEEKP--FKCDQCEYASSRRDKLKEHIIKHH

Fig. S2.

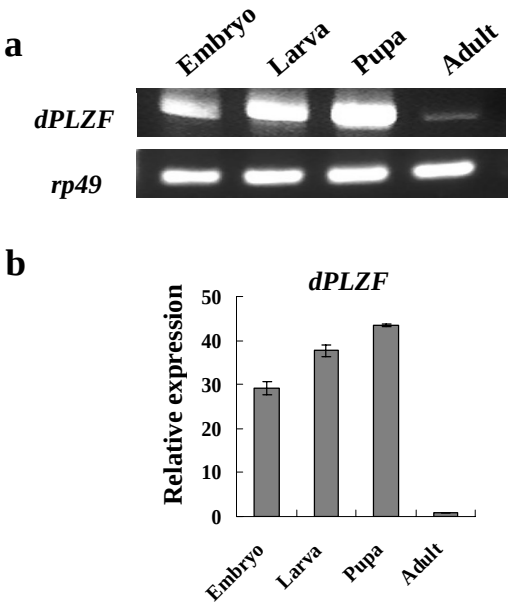
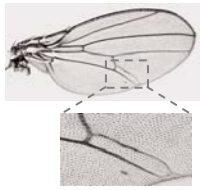

a
Embryo
Larva
Pupa
Adult
dPLZF
rp49
b
dPLZF
Relative expression
50
40
30
20
10
0
Embryo
Larva
Pupa
Adult

Fig. S3.

a ***en*-Gal4>2X *dPLZF***



b **MS1096-Gal4>*2X dPLZF***

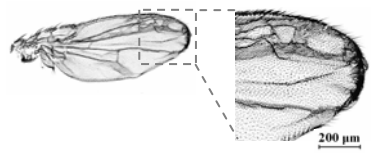
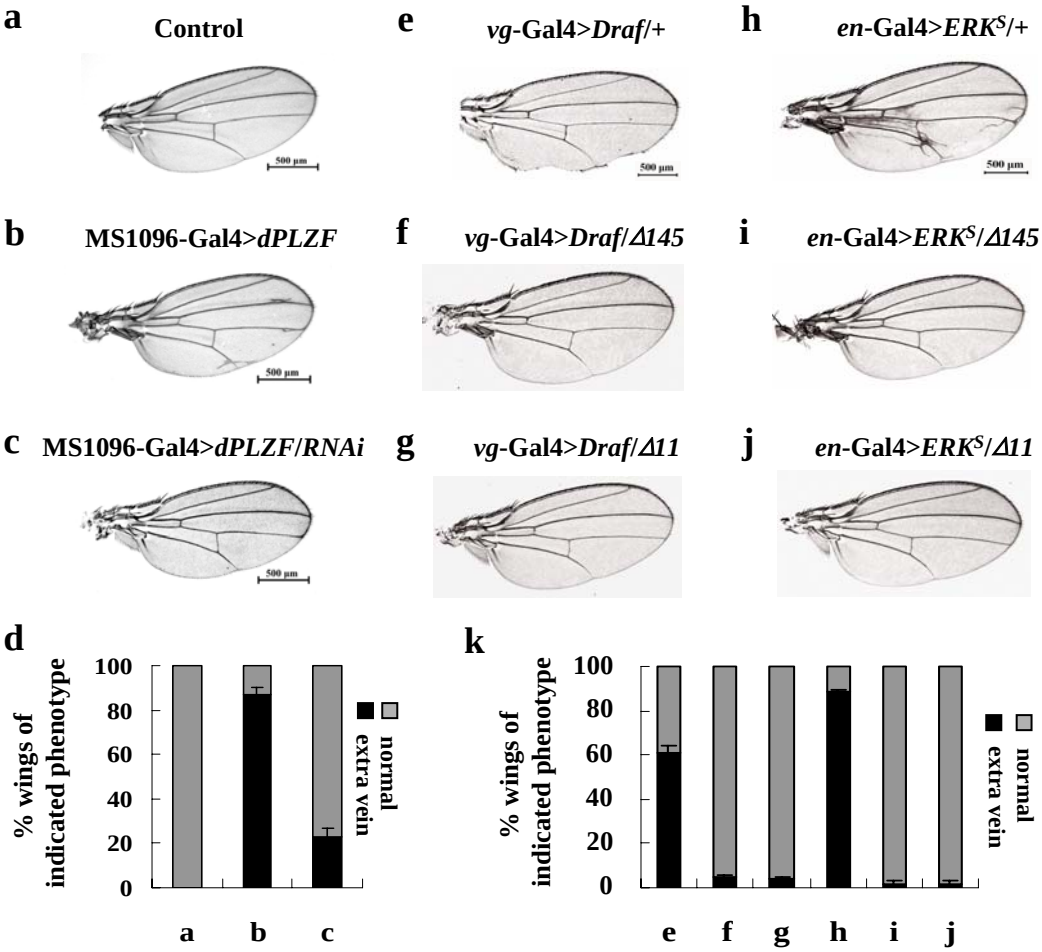


Fig. S4.


a
Control
e
vg-Gal4>Draf/+
h
en-Gal4>ERK^S/+
b
MS1096-Gal4>dPLZF
f
vg-Gal4>Draf/Δ145
i
en-Gal4>ERK^S/Δ145
c
MS1096-Gal4>dPLZF/RNAi
g
vg-Gal4>Draf/Δ11
j
en-Gal4>ERK^S/Δ11
500 μm
d
% wings of indicated phenotype
100
80
60
40
20
0
extra vein
normal
a
b
c
k
% wings of indicated phenotype
100
80
60
40
20
0
extra vein
normal
e
f
g
h
i
j