

## Captions for Supplemental Figures and Tables

**Figure S1.** A 2D-DIGE analysis of proteomic changes in whole lenses of 2-day-old DKO mice. (A) A 2D gel of WT1 lens proteins labeled with Cy3. (B) A 2D gel of DKO1 proteins labeled with Cy2. Arrowheads indicate the position of spots where different forms of  $\alpha$ A-insert and  $\alpha$ B-crystallin are expected to migrate. Arrows indicate the position where different forms of  $\alpha$ A-crystallin are expected to migrate. Note that the  $\alpha$ A-crystallin and  $\alpha$ B-crystallin bands are absent in the DKO1 gel.

**Figure S2.** Sequence coverage of protein spots identified as (A)  $\beta$ 1-catenin and (B)  $\beta$ B1-crystallin in 2-day-old mouse lenses.

**Figure S3.** Sequence coverage for proteins identified as (A) vimentin and (B)  $\beta$ B2-crystallin in 1- to 2-month-old mouse lens epithelial fractions.

**Figure S4.** Ingenuity networks created for proteins whose abundance was changed in DKO mouse lenses as compared with WT. Biological networks and pathways were generated from input data in Tables 1-3. These three networks represent the position of proteins in different cellular networks. See Figure 5 for an additional four networks generated by the data in Tables 1-3.

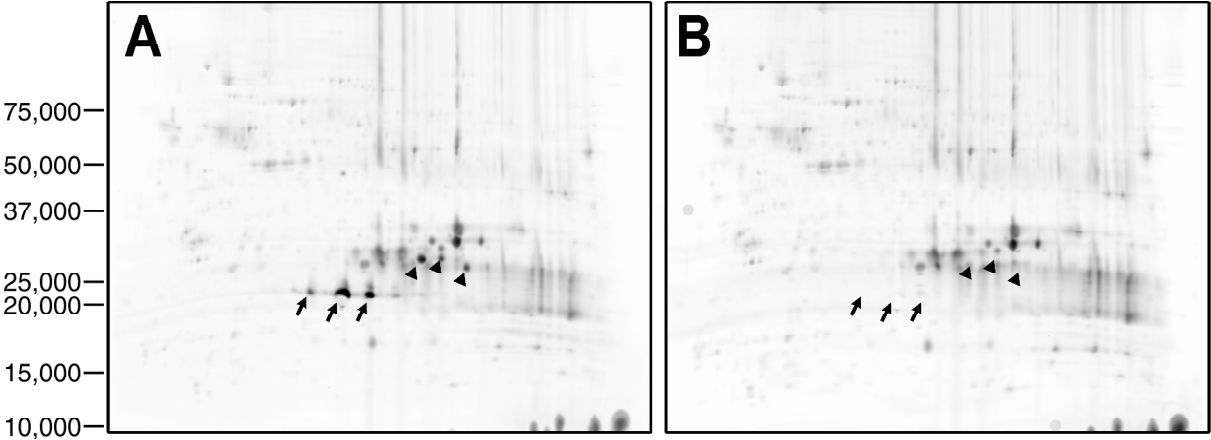
**Figure S5.** Immunoblot analysis of changes in amounts of  $\beta$ B2-crystallin and vimentin in WT vs. DKO lenses. Lens proteins were resolved by SDS-PAGE and analyzed by immunoblotting with antibodies specific for  $\beta$ B2-crystallin (top) and vimentin (bottom). Molecular weight markers (Markers) are shown on the left. Lane 1, WT water-soluble proteins; lane 2, WT water-insoluble proteins; lane 3, DKO water-soluble proteins; lane 4, DKO water-insoluble proteins.

**Figure S6.** GPC analysis of equal protein concentrations (200  $\mu$ g) of water-soluble proteins isolated from WT and DKO lenses. (A) RI profiles for 7-day-old WT and 9-day-old DKO lens proteins. (B) Light scattering profiles for 7-day-old WT and 9-day-old DKO lens proteins. Arrows in A and B indicate new bands observed at 328 kDa in DKO lens proteins. (C) RI profiles for adult WT and adult DKO lens proteins. The arrow indicates a new shoulder observed in the RI profile of adult DKO lens proteins. (D) Light scattering profiles for adult WT and adult DKO lens proteins. The arrow indicates a new light scattering species eluting near the void volume of the DKO lens proteins.

**Figure S7.** Immunoblot analysis of WT and DKO lens protein fractions. Column fractions were collected from gel permeation chromatography separation of water-soluble lens proteins. Lanes 1-5 indicate fractions eluting at retention volumes of 12, 13, 15, 16, and 18 ml. Note that in 9-day-old DKO lens proteins, a band at 328 kDa was detected with a  $\beta$ -crystallin antibody (lane 2). These fractions did not show reactivity with a  $\gamma$ -crystallin antibody (not shown).

**Figure S8.** (A) Primers used to assess vimentin and  $\beta$ B2-crystallin transcript levels in the lens. (B) PCR conditions for vimentin and  $\beta$ B2-crystallin real-time PCR. (C) Amounts of  $\beta$ B2-crystallin and vimentin gene transcripts in WT and DKO lenses were measured by quantitative PCR analysis then normalized to that of the GAPDH transcripts. Ten lenses were pooled for each analysis.

Supplemental Figure S1



Supplemental Figure S2

**A**

sp|Q02248|CTNB1\_MOUSE (100%), 85,470.9 Da

Catenin beta-1 OS=Mus musculus GN=Ctnnb1 PE=1 SV=1

5 unique peptides, 5 unique spectra, 5 total spectra, 49/781 amino acids (6% coverage)

MATQADLMEL	DMAMEPDRKA	AVSHWQQQSY	LDSGIHSGAT	TTAPSLSGKG
NPEEEDVDTS	QVLYEWEQGF	SQSFTQEQVA	DIDGQYAMTR	AQRVRAAMFP
ETLDEGMQIP	STQFDAAHPT	NVQR <b>LAEPSQ</b>	<b>MLK</b> HAVVNL I	NYQDDAELAT
RAIPELTK <b>LL</b>	<b>NDEDQVVVNK</b>	AAVMVHQLSK	KEASRHAIMR	<b>SPQMVSAIVR</b>
TMQNTNDVET	ARCTAGTLHN	LSHHREGLLA	IFKSGGIPAL	VKMLGSPVDS
VLFYAITTLH	NLLHLHQEGAK	MAVRLAGGLQ	KMVALLNKTN	VKFLAITTDC
LQILAYGNQE	SKL I I L A S G G	PQALVNIMRT	YTYEKLWTT	SRVLKVL SVC
SSNKPAIVEA	GGMQALGLHL	TDPSQRLVQN	CLWTLRNLSD	AATKQEGMEG
LLGTLVQLLG	SDDINVVTCA	AGILSNLTCN	NYKNKMMVCQ	VGGIEALVRT
VLRA GDREDI	TEPAICALRH	LTSRHQEAEM	AQNAVRLHYG	LPVVVKLLHP
PSHWPLIKAT	VGLIRNLALC	PANHAPLREQ	GAIPR <b>LVQLL</b>	<b>VR</b> AHQDTQRR
TSMGGTQQDF	VEGVRMEFIV	EGCTGAIHII	ARDVHNRIVI	RGINTIPIEV

**B**

sp|Q9WVJ5|CRBB1\_MOUSE (100%), 28,002.3 Da

Beta-crystallin B1 OS=Mus musculus GN=Crybb1 PE=2 SV=3

4 unique peptides, 4 unique spectra, 4 total spectra, 64/250 amino acids (26% coverage)

MSQAAK <b>ASAT</b>	<b>TAVNPGPDGK</b>	GK <b>GAPSTGPA</b>	<b>PAPGPTVPA</b>	<b>SVPRPAAK</b> VG
DLPPGSYRLI	<b>VFEQENFQGR</b>	RVEFSGECLN	LGDRGFDRVR	SLIVVSGPWV
AFEQSAFRGE	MFVLEKGEYP	RWDTWTSSYR	SDRLMSFRPI	RMDSQEHKIC
LFEGANFKGN	TMEIQEDDVP	SLWVYGFCDR	VGSITVSGGT	WVGYQYPGYR
<b>GYQYLLEPGD</b>	<b>FR</b> HWNEWGAF	QPQMQAVRRL	RDRQWHQEGC	FPVLTAEPPK

Supplemental Figure S3

**A**

sp|P20152|VIME\_MOUSE (100%), 53,688.8 Da

Vimentin OS=Mus musculus GN=Vim PE=1 SV=3

34 unique peptides, 35 unique spectra, 45 total spectra, 311/466 amino acids (67% coverage)

MSTRSVSSSS	YRRMFGGSGT	SSRPSSNR	SY	VTTSTR	TYSL	GSALRPSTSR
SLYSSSPGGA	YVTRSSAVRL	RSSVPGVRL		QDSVDFSLAD		AINTEFKNTR
TNEKVELQEL	NDRFANYIDK	VRFLEQQNKI		LLAELEQLKG		QGKSRLGDLY
EEEMRELRRQ	VDQLTNDKAR	VEVERDNLAE		DIMRLREKLQ		EEMLQREEAE
STLQSFQRQDV	DNASLARLDL	ERKVESLQEE		IAFLKKLHDE		EIQELQAQIQ
EQHVQIDVDV	SKPDLTAALR	DVRQQYESVA		AKNLQEAEEW		YKSKFADLSE
AANRNNDALR	QAKQESNEYR	RQVQSLT	CEV	DALKGTNESL		ERQMREMEEN
FALEAANYQD	TIGRLQDEIQ	NMKEEMARHL		REYQDLLNVK		MALDIEIATY
RKLLEGEESR	ISLPLPTFSS	LNLRETNLES		LPLVDTHSKR		TLLIKTVETR
DGQVINETSQ	HHDDLE					

**B**

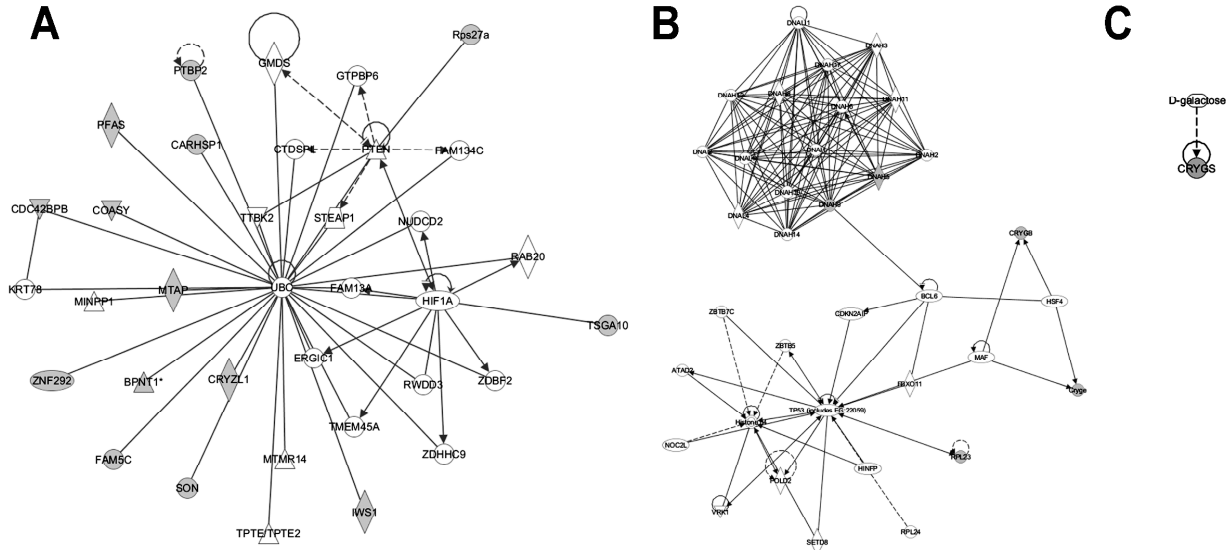
sp|P62696|CRBB2\_MOUSE (100%), 23,380.6 Da

Beta-crystallin B2 OS=Mus musculus GN=Crybb2 PE=1 SV=2

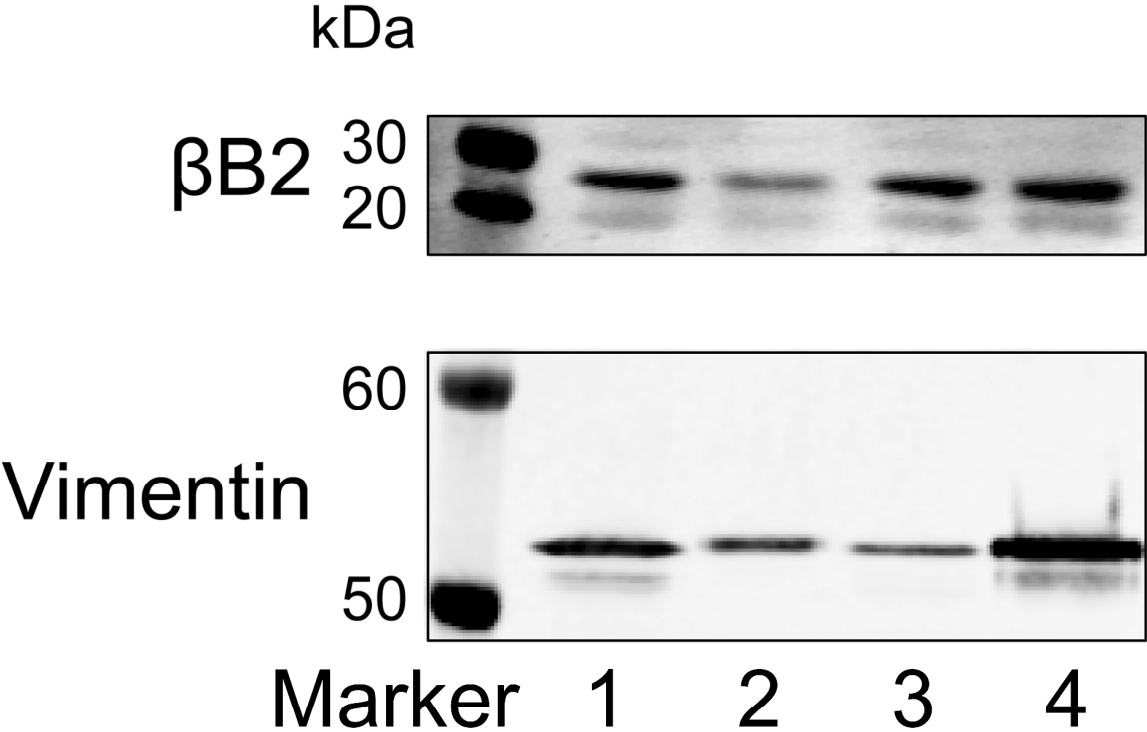
9 unique peptides, 9 unique spectra, 13 total spectra, 90/205 amino acids (44% coverage)

MASDHQTQAG	KPQPLNPKII	IFEQENFQGH	SHELSGPCPN	LKETGMEK	AG
SVLVQAGPWV	GYEQANCKGE	QFVFEKGEYP	RWDSWTSSR	TDSLSSLRPI	
KVDSQEHKII	LYENPNFTGK	KMEIVDDDVP	SFH AHGYQEK	VSSVRVQSGT	
WVGYPYQYR	GLQYLLEKGD	YKDNSDFGAP	HPQVQSVRRI	RDMQWHQRGA	
FHPSS					

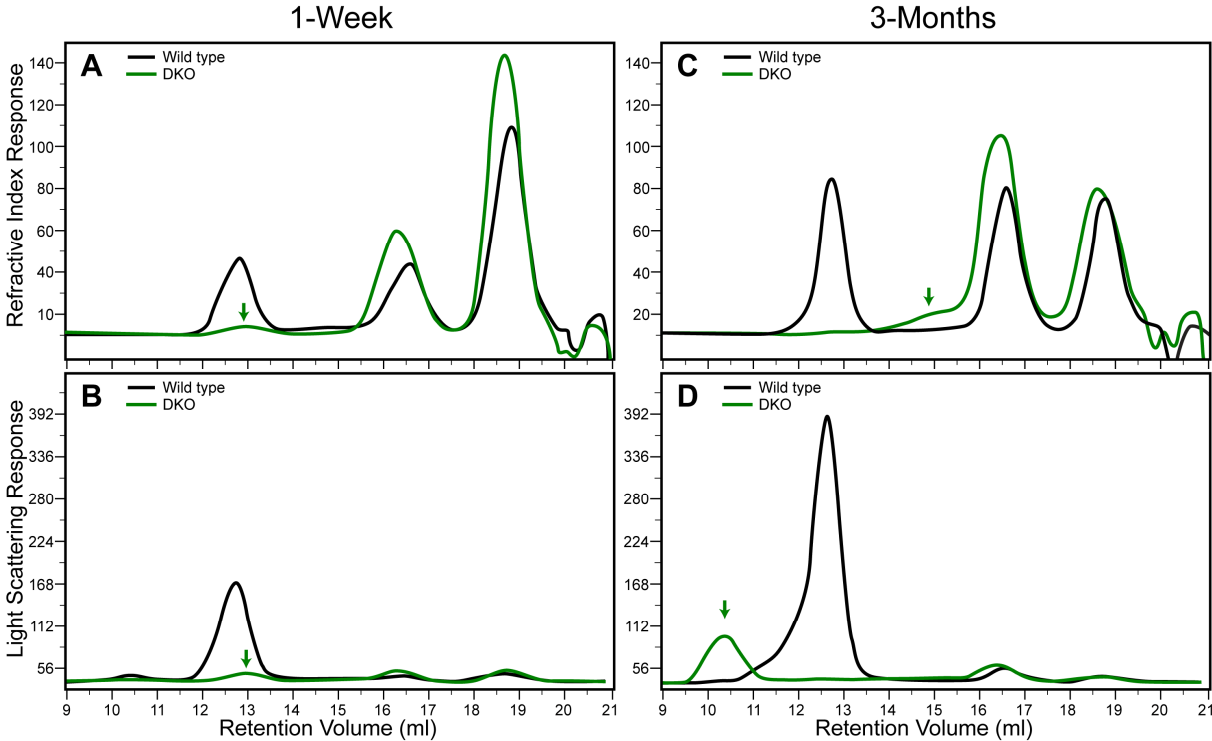
Supplemental Figure S4



Supplemental Figure S5



Supplemental Figure S6



Supplemental Figure S7





## Supplemental Figure S8

### (A) Primers and experimental conditions used in PCR analysis of vimentin and $\beta$ B2-crystallin in mouse lenses.

Vimentin:

Forward primer 5' -ATC GAT GTG GAC GTT TCC AAG CCT -3'

Reverse primer 5' -ATC CAC TTC ACA GGT GAG TGA CTG -3'

These primers generated a 228 bp PCR product

$\beta$ B2-crystallin:

Forward primer 5' -AAC AGG AGA ACT TCC AGG GCC ATT -3'

Reverse primer 5' -ACT TTG ATG GGC CTC AGA GAG CTG -3'

These primers generated a 238 bp PCR product

### (B) PCR Conditions.

Step 1, 94°C, 2 minutes

Step 2, 94°C, 30 seconds

Step 3, 62°C, 30 seconds

Step 4, 68°C, 1 minute

Step 5, go to step 2, 34 times

Step 6, 68°C, 7 minutes

Step 7, 4°C,  $\infty$

