SUPPLEMENTARY INFORMATION FOR CDD-16-0342

SUPPLEMENTARY FIGURE LEGENDS

Figure S1. Spatial expression patterns of genes in the critical region for brain abnormalities.

a. Gene expression of *CAPN8*, *CAPN2* and *TP53BP2* across human tissues. Data exported from the Genotype Tissue Expression project (GTEX). Brain regions are coloured yellow and highlighted by the red line. RPKM, reads per kilobase of transcript length per million mapped reads.

b. Regions of the prenatal human brain with the highest *TP53BP2* expression. Data mined from the Allen Human Brain Atlas, z-scores shown. **Right**: bar plot of z-scores from regions of the developing brain, combining the data from all sub-regions for each region.

Figure S2. Location of chromosomal deletions and duplications of newly described patients.

a. Genomic location of copy number change in patient DNA ascertained by chromosomal microarray. Position of *TP53BP2* is shown with a black arrow. The data are shown are for the following:

- **i.** Case 1 (DECIPHER 263059) – location of deleted region indicated by a yellow line.
- **ii.** Case 2 (DECIPHER 279742) – location of deleted region indicated by probes outside of the marked diploid region.
- **iii.** Case 3 (DECIPHER 257458) – location of deleted region indicated by a yellow line.
- **iv.** Case 4 (DECIPHER 271297) - location of deleted region indicated by red shading.
- **v.** Case 5 (DECIPHER 266948) – location of deleted region indicated by probe positions (red markers unmatched by blue markers).
- **vi.** Case 6 (DECIPHER 257256) - location of deleted region indicated by orange shading.
- **vii.** Case 7 - location of deleted region indicated by probe positions outside the diploidy interval delimited by two lines.

Figure S3. Intraventricular haemorrhage and other abnormalities in *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos.

a. Intraventricular haemorrhage (white arrows) in *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos in Balb/c and C57BL/6 backgrounds.

b. Haemorrhage in lateral ventricle (red arrow) in a Balb/c *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo as seen by microCT. Blood is also seen in other CSF spaces in other *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos.

c. Spinal canal stenosis: open (left) vs closed (right) spinal canal (red arrow) in E14.5 wild type vs E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo, respectively. HREM images shown.

d. Haemorrhage inside spinal canal in E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo vs clear spinal canal in wild type E14.5 embryo (red arrows). HREM images shown.

e. Cyst-like structures found in the spinal column of E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo (red arrows), while the spinal canal (below) is clear in the wild type littermate (single red arrow). HREM images shown.

Figure S4. Details on CNS phenotypes of *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos.

a. Exencephaly in E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ Balb/c embryo: lateral ventricular zone tissue protrudes outside the head, axial (**center**) and sagittal (**right**) views. MicroCT images shown.

b. Gross opening of the neural tube as seen in E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo with spina bifida (black arrow). MicroCT images shown.

c. Gender genotyping of *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos with NTD phenotype. Embryos positive for *ZFY* are male.

Figure S5. Few genes that cause NTDs in mice lie in regions whose deletion is associated with human NTDs

a. Positions of genes in the human genome that are orthologous to genes that cause NTDs when mutated in mice. Each gene is indicated as a black line; coloured dots mark the associated NTD phenotype in mice. Cytogenetic chromosomal bands are marked in grey and white; regions with

heterochromatin in light blue. The location of bands the deletion of which is associated with NTDs in humans marked with red lines and labelled.

b. Focus on the 1q4 region, which is significantly associated with NTDs in humans. Genes are marked that cause NTDs when deleted in mice.

Figure S6. Abnormalities in *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos and heart function in *Trp53bp2*$^{\Delta 3/\Delta 3}$ adult mice.

a. Phenotypes detected in E13.5 and E14.5 *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos by visual/light microscopy inspection, microCT and HREM, and phenotypes detected in E13.5 and E14.5 *Trp53bp2*$^{\Delta 3/+}$ embryos by microCT. In the case of BALB/c embryos, eye abnormalities were difficult to determine visually and therefore quantified for a small number of embryos. CNS, central nervous system.

b. Heart function in *Trp53bp2*$^{\Delta 3/\Delta 3}$ vs wild type adult BALB/c mice examined by echocardiography. LV is left ventricular, VTI is velocity time integral. Values are mean ± SEM.

c. Small and elongated 1st pair of spinal ganglia (pair of red arrows) in a *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo; spinal canal stenosis (single red arrow). HREM images shown.

d. Significantly enlarged trigeminal ganglia (red arrows) in *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryo; HREM images shown.

Figure S7. Abnormalities shown by adult *Trp53bp2*$^{\Delta 4/+}$ mice

a. Birth rates of wild type, *Trp53bp2*$^{\Delta 4/+}$ and *Trp53bp2*$^{\Delta 4/\Delta 4}$ mice and embryos in heterozygous-heterozygous matings.

b. Results from the Open field test performed on *Trp53bp2*$^{\Delta 4/+}$ mice (denoted *Trp53bp2*$^{+/-}$) vs wild type controls (*Trp53bp2*$^{+/+}$): distance travelled in the peripheral part of the arena (left), average speed in the periphery (center) and % time spent in the center of the arena. P values were obtained by IMPC's Mixed Model framework, linear-mixed effects model. N. s., not significant.

c. Forelimb and hindlimb grip strength normalised by body weight for *Trp53bp2*$^{\Delta 4/+}$ mice (denoted *Trp53bp2*$^{+/-}$) vs wild type controls (*Trp53bp2*$^{+/+}$). P values were obtained by IMPC's Mixed Model framework, linear-mixed effects model. *, $p < 0.05$.

d. Tibia length (left panel) and bone density (right panel) as determined by X-ray and Dual Energy X-ray Absorptiometry, respectively, in *Trp53bp2*$^{\Delta 4/+}$ mice (denoted *Trp53bp2*$^{+/-}$) vs wild type controls (*Trp53bp2*$^{+/+}$). P values were obtained by IMPC's Mixed Model framework, linear-mixed effects model. *, $p < 0.05$; **, $p < 0.01$; n. s., not significant.

e. Glucose tolerance measured by the Intraperitoneal glucose tolerance test in *Trp53bp2*$^{\Delta 4/+}$ mice (denoted *Trp53bp2*$^{+/-}$) vs wild type controls (*Trp53bp2*$^{+/+}$). P values were obtained by IMPC's Mixed Model framework, linear-mixed effects model; **, $p < 0.01$; ***, $p < 0.001$; ****, $p < 0.0001$.

SUPPLEMENTARY TABLES

Supplementary Table S1

Summary of new and previously published cases of 1q41q42 microdeletions, and novel cases of 1q41q42 microduplications. [a]hypertonia; [b]clinodactyly of 5th digits; ID – intellectual disability; LV – lateral ventricles; N/S – not specified; N/A – not applicable or available.

	1q41q42 microdeletion postnatal					1q41q42 microdeletion prenatal		1q41q42 microdeletion											
	Case 1 (DECIPHER 263059)	**Case 2 (DECIPHER 279742)**	**Case 3 (DECIPHER 257458)**	**Case 4 (DECIPHER 271297)**	**Case 5 (DECIPHER 266948)**	**Case 6 (DECIPHER 257256)**	**Case 7**	**Shaffer 1-7**	**Rosenfeld 8-9,12-13**	**Mazzeu 1, 2**	**Rice 2006**	**Slavotinek 2009**	**Filges 2010**	**Spreiz 2014**	**Christensen 2012**	**Wat 2011**	**Jun et al, 2013**	**Au et al, 2014**	**Rosenfeld 10,11,14**
Age at diagnosis	2y	7y	9y	9y	7m	Prenatal	Prenatal	N/A	N/A	N/A	10m	NS	10m	8y	N/S	2y	2m	15y	N/A
Gender	Male	Male	Male	Female	Female	Male	Female	N/A	N/A	N/A	Male	Male	Female	Female	N/S	Male	Female	Female	N/A
Deletion/duplication coordinates (hg19)	1:222,075,911-228,894,743	1:222,694,309-224,069,984	1:223,820,857-224,076,362	1:221,654,334-224,660,615	1:222,694,079-227,147,000	1:222,732,707-224,034,322	1:221,484,568-229,116,929	N/A	N/A	N/A	1:221,911,605-227,293,265	1:221,288,725-230,748,012	1:223,828,382-229,256,492	1:222,015,102-225,414,828	1:223300702-226350463	1:223,076,895-225,311,293	1:223,104,211-223,287,570	1:224,233,297-224,820,132	N/A
Size of deletion	6.8 Mb	1.38 Mb	2.21 Mb	3.0 Mb	4.45 Mb	1.30 Mb	7.6 Mb	N/A	N/A	N/A	5.38 Mb	9.46 Mb	5.43 Mb	3.4 Mb	N/S	2.2 Mb	0.18 Mb	0.59 Mb	N/A
***TP53BP2* deleted?**	YES	YES	YES	YES	YES	YES	YES	7/7	YES (4/4)	YES (2/2)	YES	YES	YES	YES	YES	YES	NO	NO	NO
Inheritance	*de novo*	paternal	*de novo*	N/A	*de novo*	maternal	N/A	N/A	N/A	*de novo*	N/A	*de novo*	*de novo*	*de novo*	*N/A*	*de novo*	maternal	N/A	N/A
Brain MRI analysed?	N/A	N/R	N/R	YES	YES	N/A	YES	2/6	YES	N/A	N/R	N/A	N/A	YES	N/R	YES	YES	YES	N/R
Developmental delay/ID	+	+	+	+	+	N/A	N/S	6/6	4/4	2/2	+	N/S	+	+	N/S	+	N/S	+	2/2
Behaviour problems	N/S	+	-	+	N/S	N/S	N/S	2/6	1/4	0/1	-	N/S	-	-	N/S	-	N/S	-	1/2
Hypotonia	a	-	-	-	+	N/S	N/S	1/6	4/4	1/1	+	N/S	-		N/S	+	N/S	-	2/2
Seizures	-	-	-	+	-	N/A	N/A	5/6	3/4	0/2	+	N/S	+	+	N/S	+	+	+	0/2
Brain abnormalities	+	N/A	N/A	+	+	+	+	2/6	4/4	2/2	+	N/S	+	-	+	+	-	-	0/2
Enlarged/dysmorphic LVs	+	N/A	N/A	+	+	+	+	3/6	4/4	1/2	-	N/S	+	-	+	+	-	-	0/1
Corpus callosum abnormalities	+	N/A	N/A	+	+	N/A	+	1/2	2/4	1/2	-	N/S	+	-	+	-	-	-	0/1
Encephalocele	-	-	-	-	-	+	-	0/7	0/4	0/2	-	-	-	-	-	-	-	-	0/3
Microcephaly	+	-	-	-	-	N/S	N/S	4/7	2/4	0/2	-	N/S	-	-	N/S	+	N/S	-	1/2
Growth retardation	-	-	-	-	-	N/S	N/S	3/5	1/4	2/2	+	N/S	-	-	N/S	-	N/S	+	1/2
Craniofacial abnormalities	+	-	+	+	+	N/A	N/A	7/7	4/4	2/2	+	+	+	+	+	+	-	+	2/2
Unusual iris	-	-	-	-	N/S	N/S	N/S	N/A	1/4	0/2	+	N/S	-	N/A	N/S	-	N/S	-	0/2
Strabismus	-	-	-	-	N/S	N/S	N/S	1/6	1/4	1/1	+	N/S	-	-	N/S	-	N/S	-	1/2
Full or tented lips	+	-	+	+	N/S	N/S	N/S	4/7	2/4	2/2	+	N/S	+	+	N/S	-	N/S	+	1/2
Congenital diaphragmatic hernia	+	-	-	-	N/S	N/S	N/S	2/7	0/4	0/2	-	+	-	-	N/S	+	N/S	-	0/3
Congenital heart defects	-	-	-	-	N/S	-	N/S	1/7	2/4	2/2	-	+	+	-	+	-	N/S	-	1/3
Pectus deformity	-	-	-	-	N/S	N/S	N/S	1/7	0/4	0/2	+	N/S	-	-	N/S	-	N/S	-	0/2
Male genital abnormalities	-	-	-	-	N/A	+	N/S	0/5	1/3	1/2	+	+	N/A	N/S	N/S	+	N/S	N/A	0/1
Limb shortening	-	-	-	-	N/S	N/S	N/S	2/7	0/4	2/2	-	N/S	-	-	N/S	-	N/S	-	0/2
Clubfoot	-	-	-	-	N/S	N/S	N/S	3/7	1/4	1/2	-	+	-	-	N/S	-	N/S	-	0/3
Short fingers	-	-	-	-	b	N/S	N/S	2/7	0/4	2/2	-	N/S	-	-	N/S	-	N/S	-	0/2
Nail hypoplasia	-	-	+	-	N/S	N/S	N/S	3/7	1/4	1/2	-	N/S	+	+	N/S	-	N/S	+	0/2
Pelger-Huet anomaly		-	-	-	N/S	N/S	N/S	1/2	1/4	N/S	N/S	N/S	+	N/S	+	N/S	N/S	N/S	0/1

Supplementary Table S2. Neuroradiological analysis of 1q41q42 microdeletion patients

Patient	DECIPHER number	*TP53BP2* deleted?	Brain MRI description
Case 1	263059	deleted	Ventricles are obviously dysmorphic with absent corpus callosum. There is a monoventricle, thalamic fusion and holoprosencephaly with a grey matter bridge extending from the frontal lobe towards the occiput. Posteriorly there is some midline differentiation into 2 occipital lobes; microcephaly
Case 2	279742	deleted	N/A
Case 3	257458	deleted	N/A
Case 4	271297	deleted	Lateral ventricles more parallel appearance at their superior aspect; relatively hypoplastic corpus callosum
Case 5	266948	deleted	Generalised cerebral atrophy, especially over the frontotemporal region with large ventricles, thin corpus callosum
Case 6	257256	deleted	Occipital encephalocele
Case 7	-	deleted	Agenesis of corpus callosum, prominent midline interhemispheric fluid, minor cerebellar hypoplasia
Jun et al, 2013	-	not deleted	Normal
Au et al, 2014	-	not deleted	Normal
Rosenfeld 10	-	not deleted	Normal
Rosenfeld 11	-	not deleted	N/A
Rosenfeld 14	-	not deleted	Normal
Shaffer 1	-	deleted	Normal
Shaffer 2	-	deleted	Mild prominence of lateral ventricles (2y); The ventricular system is top-normal in size, shape and configuration (9y)
Shaffer 3	-	deleted	1992: nonspecific increase in ventricular size; 2006: lateral ventricles somewhat prominent for age; restricted diffusion identified to suggest acute ischemia. Dysmorphic lateral ventricles with left LV being slightly larger than right; The trigones and posterior parts of the bodies of both lateral ventricles are more parallel than usual and slightly more bulbous, this appearance resembles minor colpocephaly which is a configuration associated with agenesis of the corpus callosum. The more lateral aspects of the posterior part of the body of the corpus callosum may be somewhat hypoplastic, leading to the observed ventricular dysmorphism.
Shaffer 4	-	deleted	Malformation of lateral ventricles, pointed frontal horns, fourth ventricle larger than normal; small cerebellum (CT), gyral malformations; malformations of sulcation, particularly around central sulcus
Shaffer 5	-	deleted	Normal
Shaffer 6	-	deleted	Enlarged ventricles prenatally
Shaffer 7	-	deleted	N/A
Rosenfeld 8	-	deleted	Dysmorphism of lateral ventricles, hypoplastic corpus callosum; retrocerebellar and prepontine arachnoid cysts; displacement of the pons away from the clivus by CSF material, mild cortical atrophy, prominent peripapillar atrophy, mega cisterna magna
Rosenfeld 9	-	deleted	Prominent subarachnoid space; Inferior right parietal lobe punctuate focus of abnormal susceptibility; slightly hypoplastic corpus callosum
Rosenfeld 12	-	deleted	Mild thinning of the periventricular white matter, ventricle size mildly prominent; Delayed myelination
Rosenfeld 13	-	deleted	Wide cisterna magna; mild cerebellar atrophy or hypoplasia; dysmorphic appearance of lateral ventricles with a relatively parallel configuration and slight effacement of the supra and lateral aspects of the bodies of the lateral ventricles; somewhat hypoplastic brainstem
Rice	-	deleted	Hypomyelination
Mazzeu 1	-	deleted	Left brain hemiatrophy with cortical dysplasia, possible microgyria at right
Mazzeu 2	-	deleted	Agenesis of corpus callosum, delayed myelination
Slavotinek	-	deleted	N/A
Filges 2010	-	deleted	Agenesis of corpus callosum, colpocephaly
Spreiz 2014	-	deleted	Normal; common incidental anatomical variant of the ventricles in the form a cavum septum pellusidum.
Christensen 2012	-	deleted	Agenesis of corpus callosum, somewhat small cerebellar vermis
Wat 2011	-	deleted	Severe asymmetrical cerebral atrophy with *ex vacuo* dilation of the lateral ventricle

Supplementary Table S3. **Genotype and NTD frequencies of mice derived from *Trp53bp2* heterozygous matings in mixed 129Sv-C57BL/6 ('mixed') and pure C57BL/6 ('B6') backgrounds.** +/+ denotes *Trp53bp2*$^{+/+}$, Δ3/+ denotes *Trp53bp2*$^{\Delta 3/+}$, Δ3/Δ3 denotes *Trp53bp2*$^{\Delta 3/\Delta 3}$. Distribution of +/+, +/Δ3 and Δ3/Δ3 genotypes does not differ significantly from a 1:2:1 ratio among embryos ($\chi^2 = 1.78$, $p > 0.05$, Chi-squared test), whereas there is a significant deviation from this ratio among postnatal mice ($\chi^2 = 198.9$, $p < 0.001$, Chi-squared test).

[a]Percentage of *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos amongst all genotypes.

[b]Percentage of NTDs amongst *Trp53bp2*$^{\Delta 3/\Delta 3}$ embryos only.

STAGE	**+/+**	**+/Δ3**	**Δ3/Δ3** [a]	TOTAL	NTDs [b]
Prenatal mixed					
E9.5	6	27	16 (33%)	49	2 (13%)
E10.5	12	28	17 (30%)	57	4 (24%)
E11.5	28	57	22 (21%)	107	2 (9%)
E12.5	5	21	8 (24%)	34	0
E13.5	10	18	10 (26%)	38	2 (20%)
E14.5	4	5	4 (31%)	13	0
E15.5	21	44	18 (22%)	83	5 (28%)
E16.5	11	21	14 (30%)	46	3 (21%)
E17.5	6	20	7 (22%)	33	0
E18.5	5	7	10 (32%)	22	1 (10%)
TOTAL	112	260	127 (26%)	499	19 (15%)
Postnatal mixed					
P5-P30	353	644	68 (6.4%)	1065	0
Prenatal B6					

E14.5	14	34	13 (21%)	61	6 (46%)