Characterization of a mouse model of the 22q11.2 microdeletion syndrome

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Background

The 22q11.2 microdeletion is a copy number variant (CNV) that confers a strongly increased risk of schizophrenia (Odds Ratio ≈30). It also predisposes to mild intellectual disability, ADHD, autism spectrum disorders and certain peripheral phenotypes. Aspects of 22q11 biology have been examined in several mouse models. To further investigate the impact of this microdeletion, we have generated a mouse model (*Df(h22q11)/+*), and characterized it in a broad behavioral test battery.

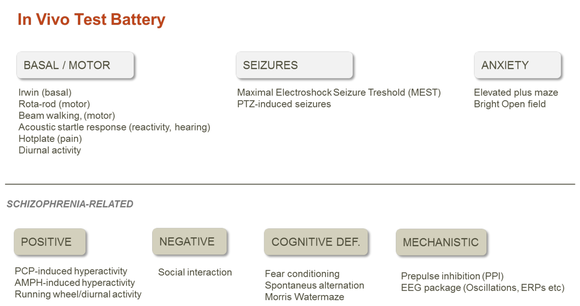

Figure 1. Behavioral domains investigated in *Df(h22q11)/+* mice

Generation of *Df(h22q11)/+* mice

The *Df(h22q11)/+* mouse line was generated by Taconic Artemis (Köln, Germany). Two targeting vectors were generated using bacterial artificial chromosome clones from the C57BL/6J RPCI-23 bacterial artificial chromosome library and transfected into TaconicArtemis C57BL/6N Tac embryonic stem cell line. The first vector introduced a loxP site upstream of the Gpr89 gene. The second vector introduced a loxP site downstream of Prkab2. Homologous recombinant clones were isolated and the 800 kilobase region on mouse chromosome 3 (Figure 2) between the loxP sites was removed using in vitro Cre-mediated recombination. Hemizygotic embryonic stem cells were injected into blastocysts isolated from impregnated BALB/c female mice and transferred to pseudopregnant NMRI female mice. Chimeric male pups were selected by coat color and mated with wild-type C57BL/6 female mice. Finally, a chimera with germline transmission was selected for expansion breeding.

Animals were bred by mating wild-type C57BL/6N female mice with hemizygotic (*Df(h22q11)/+* male mice to avoid any placental or maternal care effects of the deletion. After weaning at 3 weeks, tail biopsies were collected for polymerase chain reaction based genotyping. Mice were then group housed (two wild-type mice and two hemizygotes from the same litter per cage). Experiments presented here were performed in 6-17 week old *Df(h22q11)/+* male mice .

All studies were carried out in accordance with Danish legislation, granted by the animal welfare committee, appointed by the Danish Ministry of Food, Agriculture and Fisheries–Danish Veterinary and Food Administration.

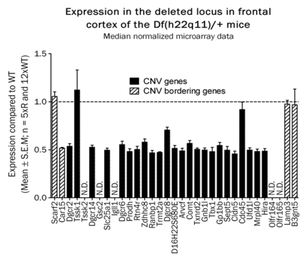

Figure 2. Expression changes in tissue from frontal cortex measured by microarray in 14-week-old *Df(h22q11)/+* mice compared with their wild-type littermates. N.D.=not detected

Decreased fertility / in utero survival

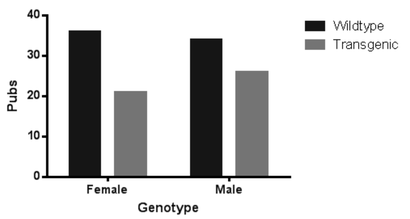

Figure 3. Birth ratios of *Df(h22q11)/+* pups. Both male and females pups were born at lower than expected ratios: Female hemizygotes=0.37 (95% CI:0.2550-0.4985); Male hemizygotes=0.40 (95% CI: 0.3173-0.4924). N=117.

Changes in acoustic startle response

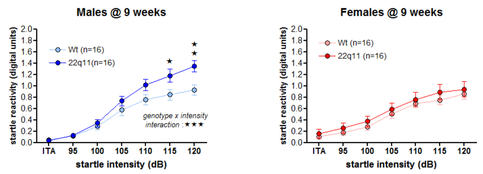

Figure 4. Acoustic startle response (ASR) amplitude following various pulse intensities in male (left) and female (right) *Df(h22q11)/+* mice. Male hemizygous mice show significantly increased ASR at higher pulse intensities. Bonferoni post hoc comparison following Mixed model two-way ANOVA. n=16 mice/group.

Decreased prepulse inhibition

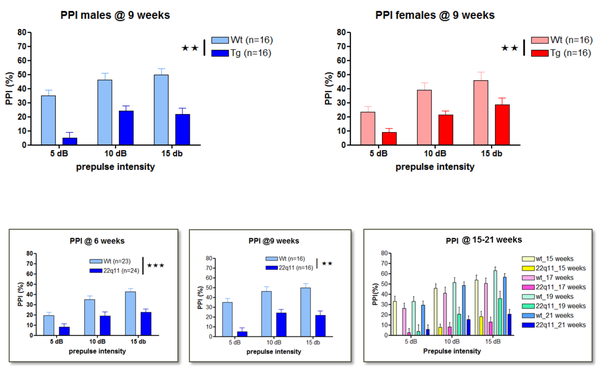

Figure 5. *Top:* Basal prepulse inhibition (PPI) in male (left) and female (right) *Df(h22q11)/+* mice. Average PPI of 5,10 and 15dB prepulse intensities. Bottom: PPI in male mice at different ages. ★★=p<0.01 effect of genotype following Mixed model 2-way ANOVA.. n=16-24 mice/group.

Increased PCP sensitivity after

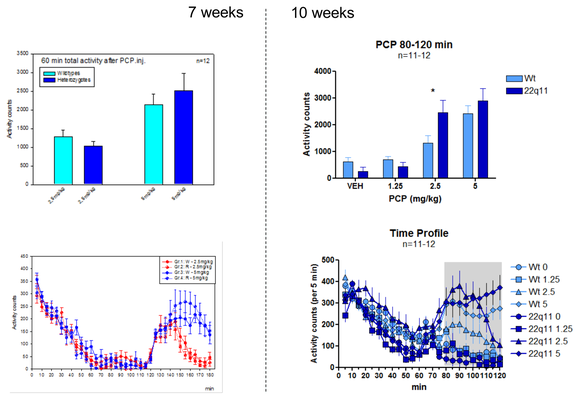

Figure 6. PCP-induced hyperactivity pre-pubertal (left) and post-pubertal (right) *Df(h22q11)/+* mice. *Bottom panel:* Activity counts divided in 5 min bins. *Top panel:* Total activity counts. n= 11-12 mice/group

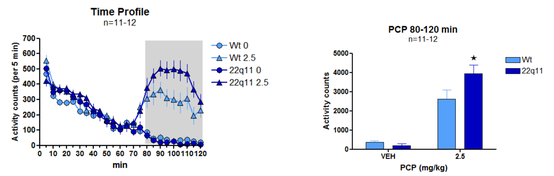

Figure 7. Replication of PCP-induced hyperactivity in new post-pubertal batch. n= 10-12 mice/group

Trend for impaired spatial ref.

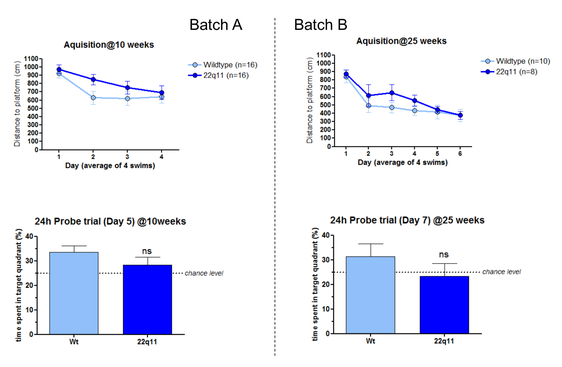

Figure 8. Morris Watermaze performance in 10 week old (left) and 25 week old (right) male *Df(h22q11)/+* mice. n=8-16 mice /group

Slower learning but normal performance in T-maze assay

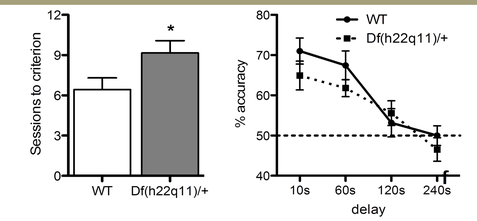

Figure 9. Performance of Df(h22q11)/+ and WT littermates T-maze assay. Df(h22q11)/+ mice required more sessions to acquire T-maze alternation criterion (a). No effect of genotype on tests of variable delays (b). Broken line represents random responding.

Higher accuracy in trial-unique non-match to sample (TUNL) touchscreen

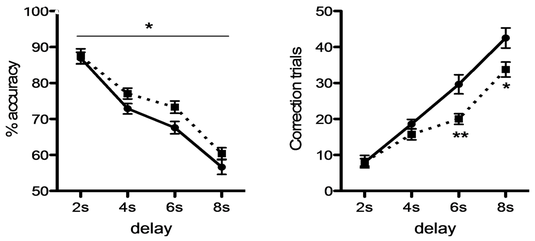

Figure 10. Performance of Df(h22q11)/+ and WT littermates on touchscreen assays. (a-b) TUNL. The Df(h22q11)/+ showed higher accuracies (a) and required fewer correction trials (b) at longer delays.

Summary

- *Df(h22q11)/+* mice did not differ from wildtypes in most behavioral assays investigated, and showed no gross changes in brain anatomy (data not shown)
- *Df(h22q11)/+* mice have decreased birth ratios suggesting lower in utero survival
- *Df(h22q11)/+* mice have changed auditory processing both pre- and post-puberty, as indicated by increased ASR and decreased PPI. The reported hearing deficits in another 22q11 mouse model (*Fuchs et al 2013*) may complicate the interpretation of these data.
- *Df(h22q11)/+* mice show increased hyperactivity following phencyclidine (but not amphetamine) administration in a novel environment. This phenotype debuts after puberty. Together with the PPI data it supports face validity for positive symptoms of schizophrenia
- *Df(h22q11)/+* mice have decreased performance in some cognitive assays, but unaltered and increased performance in others.
- Further characterization of these mice is ongoing within the NEWMEDS consortium, including extensive electrophysiological assessment

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