Down syndrome: searching for the genetic culprits

Eva Lana-Elola1,*, Sheona D. Watson-Scales1,*, Elizabeth M. C. Fisher2 and Victor L. J. Tybulewicz1,‡

Down syndrome (DS) is caused by trisomy of human chromosome 21 (Hsa21) and results in a large number of phenotypes, including learning difficulties, cardiac defects, distinguishing facial features and leukaemia. These are likely to result from an increased dosage of one or more of the ~310 genes present on Hsa21. The identification of these dosage-sensitive genes has become a major focus in DS research because it is essential for a full understanding of the molecular mechanisms underlying pathology, and might eventually lead to more effective therapy. The search for these dosage-sensitive genes is being carried out using both human and mouse genetics. Studies of humans with partial trisomy of Hsa21 have identified regions of this chromosome that contribute to different phenotypes. In addition, novel engineered mouse models are being used to map the location of dosage-sensitive genes, which, in a few cases, has led to the identification of individual genes that are causative for certain phenotypes. These studies have revealed a complex genetic interplay, showing that the diverse DS phenotypes are likely to be caused by increased copies of many genes, with individual genes contributing in different proportions to the variance in different aspects of the pathology.

Introduction
Down syndrome (DS) is a complex set of pathologies caused by an extra copy of human chromosome 21 (Hsa21). DS occurs in about 1 in 750 live births and is the most frequent cause of learning difficulties. Although the underlying genetic cause, trisomy Hsa21, is the same in most individuals with DS, the penetrance of the resulting pathologies is varied (Antonarakis et al., 2004). For example, most individuals with DS have memory and learning difficulties, craniofacial alterations and muscle hypotonia, but only some have congenital heart malformations, leukaemia or gut abnormalities. Furthermore, the severity of the defects is variable. For example, the extent of cognitive impairment varies widely between individuals with DS (Pennington et al., 2003).

The prevailing hypothesis for the genetic causes underlying DS pathology is that individual phenotypes are caused by an extra copy of one or more of the ~310 genes present on Hsa21 (Ensembl release 62, including known and newly identified protein-coding and RNA genes but excluding pseudogenes; http://www.ensembl.org/Homo_sapiens/Location/Chromosome?r=21:1-481298895). Such genes are described as being dosage sensitive, and much effort is being made to identify the dosage-sensitive genes underlying each of the DS phenotypes. The hope is that identification of such genes will lead to a better understanding of the molecular mechanisms underlying the pathologies, and hence to more effective therapy.

The search for these dosage-sensitive genetic culprits has taken advantage of both human and mouse genetics. In humans, rare partial trisomies of Hsa21 have been used to narrow down regions of the chromosome that might contain dosage-sensitive genes. Early studies suggested that a limited region of Hsa21, termed the Down syndrome critical region (DSCR) (Fig. 1), might contain one or more dosage-sensitive genes that contribute to many of the DS phenotypes (McCormick et al., 1989; Rahmani et al., 1989; Korenberg et al., 1990; Sinet et al., 1994). However, further studies that included larger numbers of partial trisomy cases and more-detailed genetic mapping have shown that different regions of Hsa21 contribute to different phenotypes, arguing against a single DSCR (Delabar et al., 1993; Korenberg et al., 1994; Korbel et al., 2009; Lyle et al., 2009). Despite these studies, it is clear that the use of human partial trisomies to identify dosage-sensitive genes is limited by the rarity of partial trisomies, heterogeneity of the specific phenotype and genetic variation between individuals.

Further progress in the search for dosage-sensitive genes underlying DS has been enabled by the generation of several mouse models of DS. Hsa21 shares conserved synteny with orthologous regions on three mouse chromosomes: Mmu10, Mmu16 and Mmu17 (Fig. 1). The first two models that were generated, Ts65Dn and Ts1Cje mice, were animals with duplications of parts of Mmu16 that are orthologous to Hsa21. The Ts65Dn strain carries an extra chromosome that has a region of Mmu16 translocated onto a short segment of Mmu17, and is thus trisomic for ~104 genes on Mmu16 that are orthologous to Hsa21 genes (Fig. 1) (Davisson et al., 1990; Gardiner et al., 2003; Olson et al., 2004a). However, it should be noted that it also has three copies of 19 genes on Mmu17 that are not orthologues of genes on Hsa21, and thus some phenotypes in this strain might not be related to human DS (Li et
By contrast, the Ts1Cje strain has a duplication of a shorter region of Mmu16, encompassing ~81 genes with orthologues on Hsa21 (Fig. 1) (Sago et al., 1998; Gardiner et al., 2003; Olson et al., 2004a). Both strains show some of the features of DS, including learning difficulties, and have thus been used to narrow down the search for dosage-sensitive genes. More recently, Cre-loxP technology has been used to precisely engineer duplications of the Hsa21-orthologous regions. For example, the Ts1Rhr mouse carries a duplication of the region of Mmu16 that is orthologous to the DSCR, and is thus trisomic for ~33 genes with orthologues on Hsa21 (Olson et al., 2004a), whereas the Dp(10)1Yey;Dp(16)2Yey;Dp(17)1Yey ‘triple trisomic’ mouse has duplications of Mmu10, Mmu16 and Mmu17, resulting in trisomy for all ~250 mouse genes that are orthologous to Hsa21 genes (Fig. 1) (Gardiner et al., 2003; Li et al., 2007; Yu et al., 2010b). This latter compound strain is the most complete DS model currently available.

Several years ago, we created a different mouse model of DS, called Tc1, carrying a freely segregating copy of Hsa21 (Fig. 1) (O’Doherty et al., 2005). This chromosome has several deletions, and thus contains ~83% of the genes on Hsa21 (Susan Gribble, V.L.I.T., E.M.C.F. and Nigel Carter, unpublished data). The Tc1 strain has the advantage of allowing the study of the human genes on Hsa21, including a number that are not found in the orthologous
Disease Models & Mechanisms  •  DMM

588

COMMENTARY

Down syndrome: searching for the genes

regions of Mmu10, Mmu16 and Mmu17 (Gardiner et al., 2003). Phenotypic characterisation showed that Tc1 mice have a number of DS-like phenotypes, including learning and memory impairment, cardiac defects, craniofacial abnormalities, and decreased tumour angiogenesis (O’Doherty et al., 2005; Morice et al., 2008; Galante et al., 2009; Reynolds et al., 2010).

To map dosage-sensitive genes in these trisomic models, the mice are being crossed with strains bearing small deletions, which reduces the number of copies of some of the trisomic genes from three to two. If such a cross rescues a phenotype, this indicates the presence of one or more dosage-sensitive genes in the deleted interval. For example, the Ms4Yah strain, which bears a deletion of the region of Mmu10 that is orthologous to Hsa21 (Fig. 1), was crossed with the Tc1 strain and showed that trisomic genes in this interval do not contribute to learning defects (Duchon et al., 2010). Such mapping crosses can be continued with ever-smaller deletions, until eventually the individual dosage-sensitive genes can be identified by crossing the mouse model with a ‘knockout’ for the relevant gene.

In this review, we present recent progress in the identification of dosage-sensitive genes underlying DS phenotypes that has resulted from the use of both human and mouse genetics.

Learning, memory, and brain development and function

All individuals with DS exhibit some form of learning and memory impairment, which varies in severity (Lott and Dierssen, 2010). Studies of mouse models have indicated that defects in neurogenesis, synaptogenesis, synaptic transmission and cell signalling pathways could all contribute to this phenotype, potentially by causing excessive inhibitory neurotransmission (Belichenko et al., 2004; Kleschevnikov et al., 2004; Harashima et al., 2006; Siarey et al., 2006; Belichenko et al., 2007; Chakrabarti et al., 2007; Morice et al., 2008). Indeed, chronic application of γ-aminobutyric acid (GABA) antagonists to dampen the action of inhibitory neurons can reverse the cognitive deficit in DS mouse models (Fernandez et al., 2007). The analysis and treatment of cognitive defects in mouse models of DS is discussed in detail in the accompanying Perspective featured in this issue of Disease Models & Mechanisms (Das and Reeves, 2011).

Studies of individuals with partial trisomy of Hsa21 demonstrated that multiple regions in Hsa21 contribute to cognitive deficits, indicating that several genes and pathways are involved in this phenotype (Korbel et al., 2009; Lyle et al., 2009). Both studies identified the importance of the proximal portion of 21q. In addition, Lyle et al. identified a contributing region from 37.94 Mb to 38.64 Mb (Lyle et al., 2009), whereas Korbel et al. noted that trisomy of the most telomeric 4.6 Mb of the chromosome resulted in the lowest IQ levels (Korbel et al., 2009). By contrast, analysis of mouse crosses showed that genes on the regions of Mmu10 and Mmu17 that are orthologous to the most telomeric 4.6 Mb of Hsa21 do not contribute to defects in learning and memory (Duchon et al., 2010; Yu et al., 2010a). Indeed, in the Ts1Yah strain, trisomy of 12 mouse genes located on a region of Mmu17 that is orthologous to Hsa21 resulted in improved outcomes in the Morris water maze (a test used to assess spatial learning), and, compared with euploid control mice, the Ts1Yah and Dp(17)1Yey strains show increased hippocampal long-term potentiation (LTP; a form of synaptic plasticity that is thought to be the physiological basis of learning and memory) (Pereira et al., 2009; Yu et al., 2010a). For a more detailed discussion of synaptic plasticity, see Box 1 in the accompanying Perspective (Das and Reeves, 2011).

The evidence connecting trisomy of the DSCR with neurological defects is mixed. Studies of humans with partial trisomy of Hsa21 showed that the DSCR region was not required in three copies to cause intellectual disability, although the data do not exclude the possibility that it contributes to the phenotype (Korbel et al., 2009; Lyle et al., 2009). Analysis of mouse models with or without three copies of the DSCR showed that trisomy of this region was necessary but not sufficient to cause a defect in the Morris water maze test (Olson et al., 2007). However, a separate study using the novel-object-recognition test, a different assay of learning and memory, concluded that trisomy of the DSCR was sufficient to result in cognitive defects (Belichenko et al., 2009). More work is needed to resolve the basis for these distinct conclusions. Nonetheless, a number of genes located in the DSCR have been proposed as candidate dosage-sensitive genes that might contribute to DS-associated brain phenotypes, including DYRK1A, SIM2, DSCAM and KCNJ6.

DYRK1A is expressed in the developing and adult nervous system (Hammerle et al., 2008) and can inhibit cell proliferation and promote premature neuronal differentiation, potentially by regulating NOTCH signalling (Yabut et al., 2010; Hammerle et al., 2011). Together with DSCR1 (also known as RCAN1), a protein also encoded by a gene on Hsa21, DYRK1A inhibits the nuclear translocation of the NFAT family of transcription factors. Thus, it has been proposed that, in DS, excess inhibition of the NFAT pathway might contribute to both neuronal and cardiac defects (Arron et al., 2006). Transgenic mice containing a 180 kb yeast artificial chromosome (YAC) that includes DYRK1A, or mice overexpressing Dyrk1a alone, showed severe learning difficulties and spatial memory deficits (Smith et al., 1997; Altafaj et al., 2001; Ahn et al., 2006). Extracellular hippocampal recordings in DYRK1A transgenic mice demonstrated altered induction of LTP and long term depression (LTD; the selective weakening of synapses during the learning and memory process), although the changes in both processes were in the opposite direction to that seen in other more complete trisomic mouse models, namely Ts65Dn and Ts1Cje (Kleschevnikov et al., 2004; Ahn et al., 2006). Nevertheless, these studies demonstrate that DYRK1A might affect the balance between excitatory and inhibitory transmission. Surprisingly, small human segmental duplications indicate that trisomy of DYRK1A does not lead to severe reductions in IQ levels (Korbel et al., 2009).

The Drosophila single minded transcription factor is a master developmental regulator (Nambu et al., 1991). SIM2, the human orthologue of single minded, is expressed in the developing human brain (Rachidi et al., 2005). Transgenic mice overexpressing Sim2 demonstrate mild learning and memory impairments (Ema et al., 1999); however, no defect was seen in a bacterial artificial chromosome (BAC) transgenic mouse in which Sim2 is expressed from its endogenous promoter, thereby achieving a physiologically more relevant level of overexpression (Chrast et al., 2000). Interestingly, SIM2 has been shown to repress Drebrin expression by directly binding to its promoter (Ooe et al., 2004). Drebrin is known to affect dendritic spine structure and neuritogenesis, and is decreased in the cortex of patients with Alzheimer’s disease (AD).
and DS (Hayashi et al., 1996; Hayashi and Shirao, 1999; Shim and Lubec, 2002; Geraldo et al., 2008).

Down syndrome cell adhesion molecule (DSCAM) inhibits dendrite branching when overexpressed in hippocampal neurons in vitro (Alves-Sampaio et al., 2010). NMDA receptor signalling, an important component of learning and memory, leads to local translation of DSCAM protein within neuronal dendrites, a process that is likely to contribute to synaptic plasticity. Interestingly, this local translation of DSCAM is lost in neurons from the Ts1Cje mouse, which contains three copies of Dscam (Alves-Sampaio et al., 2010).

Kenj6 (GIRK2; G protein-activated inward rectifying potassium channel subunit 2) is overexpressed in the hippocampus of the Ts65Dn mouse model (Harashima et al., 2006), leading to increased GIRK channel density in hippocampal neurons and increased GIRK current in response to inhibitory GABA<sub>β</sub> signalling (Best et al., 2007). It has been hypothesised that an increased dosage of Kenj6 together with Dyrk1a might explain the synaptic phenotype of Ts1Rhr mice (Belichenko et al., 2009).

Several genes outside the DSCR have also been implicated in the neurological phenotypes of DS. The Olig1 and Olig2 genes encode transcription factors that have been implicated in neurogenesis and oligodendrogenesis (Takebayashi et al., 2000; Lu et al., 2002; Zhou and Anderson, 2002). Analysis of the Ts65Dn mouse model demonstrated that reduction of both Olig genes from three to two copies corrected the overproduction of inhibitory interneurons and the associated increase in inhibitory neurotransmission in the forebrain, although it is not known whether this resulted in improved behavioural performance (Chakrabarti et al., 2010). This latter study is notable in that it is one of only a few in which specific dosage-sensitive genes contributing to DS phenotypes have been directly identified using the ‘gold standard’ assay of reducing the copy number of the gene from three to two (Table 1).

Synaptotagmin 1 (SYN1) dephosphorylates phosphatidylinositol (4,5)-bisphosphate [PtdIns(4,5)<sub>2</sub>], a key lipid involved in normal neurotransmission. PtdIns(4,5)<sub>2</sub> metabolism is altered in the brains of Ts65Dn mice, a defect that is normalised by reducing the SYN1 gene from three to two copies (Voronov et al., 2008). In addition, transgenic mice overexpressing SYN1 show defects in learning and memory (Voronov et al., 2008).

Finally, application of inhibitors to decrease the levels of APP (amyloid precursor protein) metabolites in Ts65Dn mice improves their learning and memory, suggesting that trisomy of APP contributes to neurological phenotypes of DS (Netzer et al., 2010). However, analysis of human segmental trisomies argues against a major role for APP in these defects (Rovelet-Lecrux et al., 2006; Korbel et al., 2009).

**Neurodegeneration**

DS is characterised by the early onset of the neuropathological features of AD and the eventual onset of dementia. A strong candidate for a dosage-sensitive gene contributing to this phenotype is APP, because proteolysis of APP generates amyloid-β (Aβ), the main constituent of amyloid plaques in AD brains, and mutations or duplications of APP have been associated with early onset AD (Goate et al., 1991; Rovelet-Lecrux et al., 2006; McNaughton et al., 2010).

Degeneration of basal forebrain cholinergic neurons (BFCNs) has been seen both in humans with AD (Lehericy et al., 1993) and in the Ts65Dn mouse (Holtzman et al., 1996). Neurons from Ts65Dn mice have a defect in retrograde transport of the neurotrophic factor nerve growth factor (NGF) to the cell soma, and this might contribute to their degeneration (Cooper et al., 2001). Notably, reduction of the dose of APP from three to two copies partially rescues the defective NGF retrograde transport and BFCN degeneration in Ts65Dn mice (Salehi et al., 2006). By contrast, overexpression of APP alone results in defective NGF transport but does not cause BFCN degeneration (Cataldo et al., 2003; Salehi et al., 2006). Together, these studies suggest that increased dosage of APP contributes to BFCN degeneration in DS, but is not sufficient, implying that other Hsa21 genes contribute to the phenotype.

One such gene might be Dyrk1a, whose protein product has been shown to phosphorylate APP. Consistent with this, transgenic mice overexpressing Dyrk1a demonstrate higher levels of phospho-APP and Aβ (Ryoo et al., 2008). In addition, Dyrk1a might also contribute to the AD-like pathology by phosphorylating Tau, leading to further phosphorylation by glycogen synthase kinase 3β and subsequent aggregation of Tau into neurofibrillary tangles (Liu et al., 2008). Studies of human partial Hsa21 trisomies are consistent

| Phenotype                                  | Candidate dosage-sensitive genes | References                        |
|--------------------------------------------|-----------------------------------|-----------------------------------|
| Learning, memory, brain development        | Olig1<sup>a</sup>, Olig2<sup>a</sup> | Chakrabarti et al., 2010          |
|                                            | Dyrk1a                            | Smith et al., 1997; Altufaj et al., 2001; Ahn et al., 2006 |
|                                            | Sim2                              | Ema et al., 1999                  |
|                                            | Dscam                             | Alves-Sampaio et al., 2010        |
|                                            | Synj1                             | Voronov et al., 2008              |
|                                            | App                               | Netzer et al., 2010               |
| Neurodegeneration                          | App<sup>a</sup>                   | Salehi et al., 2006               |
|                                            | Dyrk1a                            | Liu et al., 2008                  |
| Motor control                              | App                               | Trazzi et al., 2011               |
|                                            | Dyrk1a                            | Altufaj et al., 2001              |
| Cardiac defects                            | Dscam                             | Korbel et al., 2009               |
|                                            | Slc19a1                           | Locke et al., 2010                |
|                                            | Col6a1                            | Davies et al., 1994; Davies et al., 1995 |
| Leukaemia                                  | Ets2, Erg                         | Rainis et al., 2005; Stankiewicz and Crispino, 2009 |
| Reduction in solid tumours                 | Ets2<sup>a</sup>                  | Sussan et al., 2008               |
|                                            | Dscr1<sup>a</sup>                 | Baek et al., 2009                 |
| Craniofacial alterations                   | Ets2                             | Sumarsono et al., 1996            |

Table 1. Candidate dosage-sensitive genes causing DS phenotypes

<sup>a</sup> Genes for which a reduction from three to two copies reverses the phenotype in a mouse model.

<sup>b</sup> Only genes for which there is direct evidence that increased dose or allelic variation (Slc19a1, Col6a1) of the gene induces a phenotype have been included.
with a role of increased dosage of APP but not of DYRK1A in the early onset AD seen in DS (Korbel et al., 2009).

Motor control and hypotonia
Neonates with DS present with muscle hypotonia and many individuals with DS demonstrate some form of motor impairment, often described as clumsiness or deficits in fine motor control (Morris et al., 1982; Shumway-Cook and Woollacott, 1985; Spano et al., 1999; Moldrich et al., 2007). However, in contrast to cognitive and cardiac defects, these motor and muscle phenotypes in DS have not been documented in detail in terms of either penetrance or severity. Studies of human trisomic patients have mapped two regions on Hsa21 that are required in three copies to cause hypotonia: an interval from 37.4 Mb to 38.4 Mb and a second region from 46.5 Mb to 21qter (Lyle et al., 2009).

DS-associated motor defects have been reproduced in mouse models (O’Doherty et al., 2005; Galante et al., 2009), and both humans with DS and mouse trisomic models exhibit reduced numbers of granule neurons in the cerebellum, a part of the brain that is important for fine motor control (Baxter et al., 2000; Olson et al., 2004b; O’Doherty et al., 2005). Reduced cerebellar neurogenesis might be due to defective sonic hedgehog (SHH) signalling in neuronal precursors (Roper et al., 2006) caused by increased levels of APP, leading to elevated expression of the SHH receptor patched 1, an inhibitor of the SHH signalling pathway (Trazzi et al., 2011).

Transgenic mice overexpressing Dyrk1a (TgDyrk1a) have a delay in neuromotor development, which leads to impaired performance in motor tasks (Altafaj et al., 2001). Interestingly, intrastral injections of viral vectors expressing shRNA against Dyrk1a into TgDyrk1a mice rescued this defect (Ortiz-Abalia et al., 2008). By contrast, a BAC transgenic mouse strain overexpressing Dyrk1a showed no significant motor deficits, suggesting that the motor phenotypes in TgDyrk1a mice might be a consequence of Dyrk1a overexpression at levels well above those seen in DS (Ahn et al., 2006). Alternatively, the differences in phenotype might be due to the use of mouse Dyrk1a versus human Dyrk1a, or to effects of the transgene insertion sites.

One possible cause of motor deficits in DS could be defects in synapse morphology and vesicle recycling at the neuromuscular junction (NMJ). In support of this, overexpression of the Drosophila homologues of Itsn1, Synj1, and Dscrei in transgenic flies resulted in locomotor defects and impaired vesicle recycling at the NMJ, suggesting that these three genes, along with APP and Dyrk1a, are candidates for the dosage-sensitive genes underlying motor defects in DS (Chang and Min, 2009).

Cardiac defects
A total of 40-61% of individuals with DS present with congenital heart defects (CHDs), a major cause of high morbidity or infant mortality in individuals with DS (Yang et al., 2002; Vis et al., 2009). The most common heart malformation in DS is the atrioventricular septal defect (AVSD), which is considered a hallmark of DS: the incidence of AVSD is 1000-fold increased in individuals with DS compared with the non-DS population (Freeman et al., 1998; Torfs and Christianson, 1998).

Analysis of humans with partial trisomy of Hsa21 led to the identification of a 2.82 Mb region from Dscam to CBS that is sufficient to cause CHDs (Korbel et al., 2009). By contrast, other studies found that genetic variation of a more telomeric gene, Col6a1, which is outside this region, was associated with CHDs in trisomy 21 (Davies et al., 1994; Davies et al., 1995). Col6a1 encodes the α1 chain of collagen VI, which is expressed at a higher level in the atrioventricular cushions in the hearts of fetuses with DS compared with those of euploid controls (Gittenberger-de Groot et al., 2003). Genes on Hsa21 as well as on other chromosomes can affect the susceptibility to CHDs in DS. Mutations in the cell adhesion molecule Crel1d (encoded by the Crel1d gene on Hsa3) have been postulated as a risk factor for AVSD in the euploid population, and contribute to AVSD in DS (Maslen, 2004). Another study demonstrated that polymorphisms in two folate pathway genes, Slc19a1 and Mthfr (located on Hsa21 and Hsa1, respectively), are associated with AVSD in DS (Locke et al., 2010).

Using high resolution episcopic microscopy (HREM) and three-dimensional (3D) modelling, a range of cardiac defects, including AVSDs similar to those seen in humans with DS, were found in Tc1 mouse embryos (Dunlevy et al., 2010). Cardiac defects have also been reported in the Ts65Dn and Dp(16)1Yey mouse models of DS, but not in Dp(10)1Yey or Dp(17)1Yey mice, indicating that dosage-sensitive genes for CHDs lie on Mmu16 (Li et al., 2007; Williams et al., 2008; Yu et al., 2010b). However, AVSDs were not reported in either the Ts65Dn or the Dp(16)1Yey mouse models. This might reflect trisomy of different sets of genes in these mice compared with Tc1 mice and/or genetic background differences. Alternatively, the difficulty of detecting AVSDs by routine methods might have led to defects being missed in Ts65Dn or Dp(16)1Yey mice; thus, it would be very interesting to apply the HREM technique to embryos from these strains as well.

Korbel et al. combined results from the human segmental trisomy data together with data from mouse models (O’Doherty et al., 2005; Li et al., 2007), and proposed a region of 1.77 Mb from Dsca to Znf295 as the ‘heart critical region’ (Korbel et al., 2009). Because Dscam is the only gene in this region that is known to be expressed in the heart, the authors suggested Dscam as a candidate dosage-sensitive gene for CHDs in DS. However, no cardiac malformations are seen in the Ts1Rhr strain, which is trisomic for 33 genes in the Dscr, including Dscam (Fig. 1), demonstrating that three copies of Dscam are not sufficient to cause the DS heart phenotype in mice (Dunlevy et al., 2010; Liu et al., 2011). A recent study showed that duplication of a 5.43 Mb region of Mmu16 from Tiam1 to Kcnj6 in the Dp(16)2Yey strain was sufficient to cause cardiac defects (Fig. 1), thereby again excluding Dscam and further narrowing down the region containing causative dosage-sensitive genes (Liu et al., 2011).

Leukaemia
Compared with the non-DS population, individuals with DS have an 18-fold increased risk of developing leukaemia (Hasle et al., 2000). In particular, DS is associated with a 500-fold increased risk of acute megakaryoblastic leukaemia (AMKL). Interestingly, DS-AMKL is always associated with a stereotypical mutation in the X-linked Gata1 gene, which leads to synthesis of a truncated protein termed Gata1s (Wechsler et al., 2002). This has led to the suggestion that the generation of AMKL requires cooperation between trisomy Hsa21 and the Gata1s mutation, together with additional mutations (Hitzler and Zipursky, 2005). Human partial...
trisomies have mapped an 8.35 Mb region of Hsa21 from RUNX1 to CBS as contributing to AMKL (Korbel et al., 2009). This region includes the RUNX1, ERG and ETS2 genes, all of which have been hypothesised to play a role in DS-AMKL (Hitzler and Zipursky, 2005; Rainis et al., 2005; Stankiewicz and Crispino, 2009).

So far, three mouse models of DS have been tested for leukaemia (Kirsammer et al., 2008; Carmichael et al., 2009; Alford et al., 2010). None showed leukaemia, but all three reported alterations in megakaryopoiesis. Tc1 mice have macrocytic anaemia, and older animals show splenomegaly with increased megakaryopoiesis and erythropoiesis, but they do not develop acute leukaemia, even when crossed with a strain expressing GATA1S (Alford et al., 2010). Both Ts65Dn and Ts1Cje mice also display macrocytic anaemia, whereas only Ts65Dn mice present with a myeloproliferative disorder (MPD) with increased megakaryopoiesis and erythropoiesis (Kirsammer et al., 2008; Carmichael et al., 2009). These results suggest that one or more of the 23 genes that are in three copies in Ts65Dn but not in Ts1Cje are responsible for the MPD, whereas the trisomic region shared between the two mouse models contains a gene(s) that contributes to the macrocytic anaemia.

The Hsa21-encoded RUNX1 transcription factor plays a key role in megakaryopoiesis and haematopoietic stem cell maintenance (Okuda et al., 1996), and hence has been proposed to contribute to the predisposition to AMKL when present in three copies. However, mouse studies showed that reducing Runx1 from three to two copies did not affect MPD in Ts65Dn mice, arguing against a key role for this gene (Kirsammer et al., 2008).

**Solid tumours**

Individuals with DS are at lower risk of developing almost all types of malignant solid tumours (Yang et al., 2002), indicating that trisomy 21 protects from tumour growth. This observation has led to the suggestion that Hsa21 encodes tumour suppressor genes. A cross of the ApcMin mouse model for colorectal cancer with either the Ts65Dn or Ts1Rhr mouse resulted in fewer tumours in the trisomic mice compared with euploid controls. Conversely, when ApcMin mice were crossed with Ms1Rhr (which have segmental monosomy for the same 33-gene region that is triplicated in the Ts1Rhr strain), a significant increase in the number of tumours was observed compared with controls, demonstrating the influence of gene dosage (Sussan et al., 2008). This effect was mapped to the Ets2 gene, which, when present in three copies, inhibits tumour formation and, conversely, when reduced to one copy, results in increased tumour rates (Sussan et al., 2008). Similar studies using the NPCis mouse model – which develops lymphomas, sarcomas and carcinomas at high frequency – also found that the Ts65Dn genotype delayed tumour development (Yang and Reeves, 2011). However, in this case, no dosage effect of Ets2 was found.

Interestingly, recent data have suggested that reduced solid tumour growth in individuals with DS might also be due to decreased angiogenesis (Baek et al., 2009; Reynolds et al., 2010). Ectopically implanted tumour lines grew more slowly in Ts65Dn mice compared with in euploid controls, indicating that tumour growth was suppressed in the trisomic mice owing to effects in the supporting stroma (Baek et al., 2009). Histological examination of the tumours showed that they had reduced microvessel density, suggesting that Ts65Dn mice have a defect in tumour angiogenesis. The Hsa21-encoded DSCR1 protein had previously been shown to inhibit angiogenesis induced by vascular endothelial growth factor (VEGF) by attenuating NFAT signalling, and was thus a candidate gene for this effect. Indeed, reduction of the dosage of Dscr1 from three to two copies resulted in increased tumour volume and tumour angiogenesis. Another recent study using the Tc1 mouse also proposed that reduced angiogenesis in DS accounts for decreased tumour growth (Reynolds et al., 2010). In this case, the reduced angiogenesis was shown to be due to increased doses of Admats1, Erg, Jam2 and Pttg1ip. It is interesting to note that the Tc1 model does not have three copies of Dscr1, so this gene cannot account for the reduced angiogenesis in Tc1 mice. In contrast to these two reports, the cross of Ts65Dn to the NPcis mouse model of cancer showed no effect on tumour angiogenesis, showing that this is not a universal mechanism to explain the lower incidence of solid tumours in DS (Yang and Reeves, 2011). Taken together, these studies indicate that the reduced frequency of tumours in DS might be due to increased dosage of several genes, with effects both in the tumour cells and in the supporting stroma.

**Craniofacial alterations**

Characteristic facies is one of the few phenotypes seen in all individuals with DS, and is the result of an underlying craniofacial dysmorphology, which includes reduced skull size, flattened occiput (posterior portion of the head), brachycephaly (shortened front-to-back diameter of the skull), small midface and reduced size of the maxilla and the mandible (Richtsmeier et al., 2002). Human partial trisomies have mapped areas of Hsa21 contributing to craniofacial abnormalities, but a discrete region has not been identified (Lyle et al., 2009). Bone morphometric analysis of Ts65Dn mice revealed a pattern of craniofacial anomalies similar to those seen in individuals with DS, including a smaller flattened face, brachycephaly, a flattened occiput and a smaller mandible (Richtsmeier et al., 2000). Subsequent analysis showed that Ts1Cje mice have craniofacial alterations very similar to those seen in Ts65Dn mice (Richtsmeier et al., 2002), indicating that the trisomic region in Ts1Cje mice (Sod1-Znf295) is sufficient to produce the craniofacial phenotype. Overexpression in mice of one of the genes in this interval, Ets2, led to skeletal abnormalities that resembled those in humans with DS (Sumarsono et al., 1996). However, a reduction of the copy number of the Ets2 gene from three to two did not rescue the craniofacial phenotype of Ts65Dn mice, demonstrating that trisomy of this gene is not required for the phenotype (Hill et al., 2009).

Analysis of Ts1Rhr mice, which are trisomic for the DSCR, showed that they had rostrocaudally elongated skulls and larger mandibles than euploid littermates, a phenotype opposite from that seen in individuals with DS and in Ts65Dn mice, demonstrating that trisomy of the DSCR is not sufficient to cause the DS craniofacial phenotype (Olson et al., 2004a). When Ts65Dn mice were crossed to Ms1Rhr to reduce the DSCR from three to two copies, the brachycephaly phenotype was rescued but the overall pattern of dysmorphology was similar to that seen in Ts65Dn mice. Together, these results demonstrate a complex interplay of genetic effects of genes both within and outside the DSCR contributing both positively and negatively to the craniofacial phenotypes.
**Congenital gut disorders**

Congenital gut disorders have an increased incidence in DS. Patients with DS constitute ~12% of all cases of Hirschprung's disease, and duodenal stenosis (DST) and imperforate anus (IA) are 260 and 33 times more likely to occur in DS, respectively (Torfs and Christianson, 1998; Korbel et al., 2009). Hirschprung’s disease arises when a portion of the colon is not innervated correctly by the enteric nervous system; alterations in ~10 non-Hsa21 genes have thus far been linked to this disease (Amiel et al., 2008). Analysis of human segmental trisomies has defined a 13 Mb critical region for Hirschprung’s disease that contains the DSCAM gene, which is expressed in the neural crest cells that give rise to the enteric nervous system (Korbel et al., 2009). No other Hsa21 genes have been implicated so far. An overlapping critical region was described for DST and IA (Korbel et al., 2009).

**Concluding remarks**

For most of the last century, the study of the molecular genetics of DS was an undertaking for only the very dedicated, because the resources available for gene mapping were mostly limited to the few known human partial trisomy cases. In the mid-1990s, two new mouse models, Ts65Dn and Ts1Cje, showed that dosage sensitivity could be recapitulated in the mouse, giving rise to phenotypes similar to those seen in human DS. Crucially, these two mouse strains demonstrated that it was possible to use phenotype-genotype comparisons to map causative dosage-sensitive genes, thus establishing the concept of a mouse mapping panel.

These trisomic mouse strains, in combination with gene knockouts and YAC or BAC transgenics, provided new hope for the identification of individual dosage-sensitive genes responsible for different phenotypes. However, the scale of the problem remained immense, given that researchers were attempting to assay an entire chromosome, albeit the smallest autosome, for the effects of individual causative genes. The DNA sequence of Hsa21 generated a list of genes on the chromosome and is an essential tool for the understanding of DS, but it did not give insight into dosage sensitivity (Hattori et al., 2000). Nevertheless, in the last 10 years, the field has again taken a step forward with the advent of chromosome engineering to create mapping panels of trisomic animals that span entire stretches of the human chromosome or the orthologous mouse regions. Thus, it is now realistic to use the resources of mouse genetics to identify the dosage-sensitive genes involved in DS. Results so far suggest that, although some dosage-sensitive genes might have strong effects on their own, it is likely that many of the phenotypes associated with DS are due to complex effects of multiple Hsa21 genes.

An even more challenging aim is to understand the genetic basis of the large variation in DS phenotypes, which is probably caused largely by allelic variation in genes both on Hsa21 and on other chromosomes. In the next 10 years, genome-wide association studies of DNA biobanks from people with DS will be used to identify more candidate genes, and to dissect the effects of individual alleles and combinations of alleles both on Hsa21 and on other chromosomes. These are studies that must be carried out internationally and require high levels of cooperation between different countries to collect samples from individuals with DS and to carry out standardised clinical and psychometric analysis of the participants. There is a huge challenge ahead to create these banks and agree on tests, but already the international community is mobilising and starting to tackle the problem.

This is an exciting time to be involved in dissecting the molecular genetics of DS because we now have the tools to find the individual dosage-sensitive genes and thus to understand the biological effects of trisomy. There is also synergy with human genomic studies that are highlighting the extraordinary variability in copy number that is present in all humans, and hence the importance of gene dosage to everyone, not just to people with DS.

Finally, the molecular genetic study of DS is a challenging endeavour that has given us new insights into euploid and aneuploid biology, but how is the identification of dosage-sensitive genes relevant to people with DS? The disorder is complex, but new therapies that tackle aspects of the syndrome are already being developed by applying knowledge of the dosage-sensitive genes, combined with investigations of their biology (Das and Reeves, 2011). These range from therapies currently in clinical trials to test their ability to enhance learning and memory in people with DS, to novel therapies to combat DS-associated AD. The next 10 years of DS research look very promising indeed.

**ACKNOWLEDGEMENTS**

This work was supported by a grant from the Wellcome Trust and by the AnEUploidy integrated project (EU Framework 6). In addition, V.L.J.T. was funded in part by the Medical Research Council (program number U117527522).

**COMPETING INTERESTS**

The authors declare that they have no competing or financial interests.

**REFERENCES**

Ahn, K. J., Jeong, H. K., Choi, H. S., Ryoo, S. R., Kim, Y. J., Goo, J. S., Choi, S. Y., Han, J. S., Ha, I. and Song, W. J. (2006). DYRK1A BAC transgenic mice showed altered synaptic plasticity with learning and memory defects. Neurobiol. Dis. 22, 463-472.

Alfold, K. A., Slender, A., Vanes, L. L., Li, Z., Fisher, E. M., Nitizic, D., Orkin, S. H., Roberts, I. and Tybulewicz, V. L. (2010). Perturbed hematopoiesis in the Tc1 mouse model of Down syndrome. Blood 115, 2928-2937.

Altafaj, X., Dierssen, M., Baamonde, C., Marti, E., Visa, J., Guimera, J., Oset, M., Gonzalez, J. R., Florez, J., Fillat, C. et al. (2001). Neurodevelopmental delay, motor abnormalities and cognitive deficits in transgenic mice overexpressing Dyrk1a (minibrain), a murine model of Down's syndrome. Hum. Mol. Genet. 10, 1915-1923.

Alves-Sampaio, A., Troca-Marim, J. A. and Montesinos, M. L. (2010). NMDA-mediated regulation of DSCAM dendritic local translation is lost in a mouse model of Down's syndrome. J. Neurosci. 30, 13357-13348.

Amiel, J., Sprott-Emison, E., Garcia-Barcelo, M., Lantieri, F., Burzynski, G., Borrego, S., Pelet, A., Arnold, S., Miao, X., Griseri, P. et al. (2008). Hirschsprung disease, associated syndromes and genetics: a review. J. Med. Genet. 45, 1-14.

Antonarakis, S. E., Lyle, R., Dermitzakis, E. T., Reymond, A. and Deutsch, S. (2004). Chromosome 21 and down syndrome: from genomics to pathophysiology. Nat. Rev. Genet. 5, 725-738.

Arron, J. R., Winslow, M. M., Polleri, A., Chang, C. P., Wu, H., Gao, X., Neilson, J. R., Chen, L., Heit, J. J., Kim, S. K. et al. (2006). NFAT dysregulation by increased dosage of DSCR1 and DYRK1A on chromosome 21. Nature 441, 595-600.

Baek, K. H., Zaslavsky, A., Lynch, R. C., Britt, C., Okada, Y., Siarey, R. J., Lensch, M. W., Park, I. H., Yoon, S. S., Minami, T. et al. (2009). Down's syndrome suppression of tumour growth and the role of the calcineurin inhibitor DSCR1. Nature 459, 1126-1130.

Baxter, L. L., Moran, T. H., Ristschmeier, J. T., Troncoso, J. and Reeves, R. H. (2000). Discovery and genetic localization of Down syndrome cerebellar phenotypes using the Ts65Dn mouse. Hum. Mol. Genet. 9, 195-202.

Belichenko, P. V., Masliah, E., Kleschevnikov, A. M., Villar, A. J., Epstein, C. J., Salehi, A. and Mobley, W. C. (2004). Synaptic structural abnormalities in the Ts65Dn mouse model of Down Syndrome. J. Comp. Neurol. 480, 281-298.

Belichenko, P. V., Kleschevnikov, A. M., Salehi, A., Epstein, C. J. and Mobley, W. C. (2007). Synaptic and cognitive abnormalities in mouse models of Down syndrome: exploring genotype-phenotype relationships. J. Comp. Neurol. 504, 329-345.

Belichenko, N. P., Belichenko, P. V., Kleschevnikov, A. M., Salehi, A., Reeves, R. H. and Mobley, W. C. (2009). The “Down syndrome critical region” is sufficient in the
Geraldo, S., Khanzada, U. K., Parsons, M., Chilton, J. K. and Gordon-Weeks, P. R. (2008). Targeting of the F-actin-binding protein drebrin by the microtubule plus-tip protein EB3 is required for neurotrophin responsiveness. Nat. Cell Biol. 10, 1181-1189.

Gittenberger-de Groot, A. C., Bartram, U., Oosthoek, P. W., Bartelings, M. M., Hoogaars, W. A., Poelmans, R. F., Jongsma, E. J. and Aerts, J. S. (2003). Collagen type VI expression during cardiac development and in human fetuses with trisomy 21. Anat. Rec. A Discov. Mol. Cell Evol. Biol. 275, 1109-1116.

Goate, A., Chartier-Harlin, M. C., Mullan, M., Brown, J., Crawford, F., Fidani, L., Giuffra, L., Haynes, A., Irving, N., James, L. et al. (1991). Segregation of a missense mutation in the amyloid precursor protein gene with familial Alzheimer’s disease. Nature 349, 704-706.

Hammerle, B., Elalinde, C. and Tejedor, F. J. (2008). The spatio-temporal and subcellular expression of the candidate Down syndrome gene Mbn/Dyrk1A in the developing mouse brain suggests distinct sequential roles in neuronal development. Eur. J. Neurosci. 27, 1061-1074.

Hammerle, B., Ulin, E., Guimera, J., Becker, W., Guillomot, F. and Tejedor, F. J. (2011). Transient expression of Mbn/Dyrk1A couples cell cycle exit and differentiation of neuronal precursors by inducing p27kip1 expression and suppressing NOTCH signaling. Development 138, 2543-2554.

Harashima, C., Jacobowitz, D. M., Witta, J., Borke, R. C., Best, T. K., Siarey, R. J. and Galdrizzi, D. (2006). Abnormal expression of the G-protein-activated inwardly rectifying potassium channel 2 (GIRK2) in hippocampus, frontal cortex, and substantia nigra of Ts65Dn mouse: a model of Down syndrome. J. Comp. Neurol. 494, 815-833.

Hasle, H., Clemmensen, I. H. and Mikkelsen, M. (2000). Risks of leukaemia and solid tumours in individuals with Down’s syndrome. Lancet 355, 165-169.

Hattori, M., Fujiyama, A., Taylor, T. D., Watanabe, H., Yada, T., Park, H. S., Toyoda, A., Ishii, K., Tokotyi, Y., Choi, D. K. et al. (2000). The DNA sequence of human chromosome 21. Nature 405, 311-319.

Hayashi, K. and Shirao, T. (1999). Change in the shape of dendritic spines caused by overexpression of drebrin in cultured cortical neurons. J. Neurosci. 19, 3918-3925.

Hayashi, K., Ishikawa, R., Ye, L. H., He, X. L., Takata, K., Kohama, K. and Shirao, T. (1996). Modulatory role of drebrin on the cytoskeleton within dendritic spines in the substantia nigra of Ts65Dn mouse: a model of Down syndrome.

Hill, C. A., Sussan, T. E., Reeves, R. H. and Richtmeier, J. T. (2008). Complex contributions of Ets2 to craniofacial and thymus phenotypes of trisomic “Down’s syndrome” mice. Am. J. Med. Genet. A 149A, 2158-2165.

Hitzer, J. K. and Zipursky, A. (2000). Origins of leukaemia in children with Down syndrome. Nat. Rev. Cancer 5, 11-20.

Holtzman, D. M., Santucci, D., Killbride, J., Chuang, C., Fontana, D. J., Daniels, S. E., Johnson, R. M., Chen, K., Sun, Y., Carlson, E. et al. (1996). Developmental abnormalities and age-related neurodegeneration in a mouse model of Down syndrome. Proc. Natl. Acad. Sci. USA 93, 13333-13338.

Kirsammer, G., Jiliani, S., Liu, H., Davis, E., Gurbuxani, S., Le Beau, M. M. and Crispino, J. D. (2008). Highly penetrant myeloproliferative disease in the Ts65Dn mouse model of Down syndrome. Blood 111, 767-775.

Kleshevnikov, A. M., Belichenko, P. V., Villar, A. J., Epstein, C. J., Malenka, R. C. and Mobley, W. C. (2004). Hippocampal long-term potentiation suppressed by increased inhibition in the Ts65Dn mouse, a genetic model of Down syndrome. J. Neurosci. 24, 8153-8160.

Korbel, J. O., Tirosh-Wagner, T., Urban, A. E., Chen, X. N., Kasowski, M., Dai, L., Grubert, F., Erdmann, C., Gao, M. C., Lange, K. et al. (2009). The genetic architecture of Down syndrome phenotypes revealed by high-resolution analysis of human segmental trisomies. Proc. Natl. Acad. Sci. USA 106, 12031-12036.

Korenberg, J. R., Kawashima, H., Pulst, S. M., Ikeuchi, T., Ogasawara, N., Yamamoto, K., Schonberg, S. A., West, R., Allen, L. and Klemens, E. M. et al. (1990). Molecular definition of a region of chromosome 21 that causes features of the Down syndrome phenotype. Am. J. Hum. Genet. 47, 236-246.

Korenberg, J. R., Chen, X. N., Schipper, R., Sun, Z., Gonsky, R., Gerwehr, S., Carpenter, N., Daumer, C., Dignan, P., Disteche, C. et al. (1994). Down syndrome phenotypes: the consequences of chromosomal imbalance. Proc. Natl. Acad. Sci. USA 91, 4900-4907.

Korinth, S., Hirsch, E. C., Cervera-Pierot, P., Hersh, L. B., Bakchine, S., Plette, F., Duycquaerts, C., Hauw, J. J., Javoy-Agid, F. and Agid, Y. (1993). Heterogeneity and selective deprivation of cholinergic neurons in the basal forebrain of patients with Alzheimer’s disease. J. Comp. Neurol. 330, 35-51.

Li, Z., Yu, T., Morishima, M., Pao, A., LaDuca, J., Conroy, J., Nowak, N., Matsui, S., Shiraiashita, I. and Yu, V. E. (2007). Duplication of the entire 22.9 Mb human chromosome 21 syntenic region on mouse chromosome 16 causes cardiovascular and gastrointestinal abnormalities. Hum. Mol. Genet. 16, 1359-1366.

Liu, C., Morishima, M., Yu, T., Matsui, S. I., Zhang, L., Fu, D., Pao, A., Costa, A. C., Gardiner, K. J., Cowell, J. K. et al. (2011). Genetic analysis of Down syndrome-associated heart defects in mice. Hum. Genet. [Epub ahead of print].
Locke, A. E., Dooley, K. J., Tinker, S. W., Cheong, S. Y., Feingold, E., Allen, E. G., Freeman, S. B., Torfs, C. P., Cua, C. L., Epstein, M. P. et al. (2010). Variation in folate pathway genes contributes to risk of congenital heart defects among individuals with Down syndrome. Genet. Epidemiol. 34, 613-623.

Lott, I. T. and Dierssen, M. (2010). Cognitive deficits and associated neurological complications in individuals with Down's syndrome. Lancet Neurol. 9, 623-633.

Lu, Q. R., Sun, T., Zhu, Z., Ma, N., Garcia, M., Stiles, C. D. and Rowitch, D. H. (2002). Common developmental requirement for Olig function indicates a motor neuron/oligodendrocyte connection. Cell 109, 759-769.

Lyle, R., Bena, F., Gagos, S., Gehrig, C., Lopez, G., Schinzel, A., Lespinasse, J., Bottani, A., Dahoun, S., Taine, L. et al. (2009). Genotype-phenotype correlations in Down syndrome identified by array CGH in 30 cases of partial trisomy and partial monosomy chromosome 21. Eur. J. Hum. Genet. 17, 454-466.

Maslen, C. L. (2004). Molecular genetics of atrioventricular septal defects. Curr. Opin. Cardiol. 19, 205-210.

McCormick, M. K., Schinzel, A., Petersen, M. B., Stetten, G., Driscoll, D. J., Cantu, E. S., Tranebaerjag, L., Mikkelsen, M., Watkins, P. C. and Antonarakis, S. E. (1989). Molecular genetic approach to the characterization of the “Down syndrome region” of chromosome 21. Genomics 5, 325-331.

McNaughton, D., Knight, W., Guerreiro, R., Ryan, N., Lowe, J., Poulter, M., Morris, A. F., Vaughan, S. E. and Vaccaro, P. (2010). Syndrome mice.

McNaughton, D., Knight, W., Guerreiro, R., Ryan, N., Lowe, J., Poulter, M., Morris, A. F., Vaughan, S. E. and Vaccaro, P. (2010). Syndrome mice.

McNaughton, D., Knight, W., Guerreiro, R., Ryan, N., Lowe, J., Poulter, M., Morris, A. F., Vaughan, S. E. and Vaccaro, P. (2010). Syndrome mice.

Morris, A. F., Vaughan, S. E. and Vaccaro, P. (2010). Syndrome mice.
Forthcoming DMM articles

For articles published online ahead of print see http://dmm.biologists.org/content/early/recent
Sign up for email alerts (eToC) at http://dmm.biologists.org/cgi/alerts/etc

Review Articles

- COMMENTARY: Animal models for Gaucher disease research. Tamar Farfel-Becker, Einat B. Vitner and Anthony H. Futerman
- COMMENTARY: Modeling psychiatric disorders through reprogramming. Kristen J. Brennand and Fred H. Gage
- COMMENTARY: Studying synthetic lethal interactions in the zebrafish system. Vinita A. Hajeri and James F. Amatruda
- PERSPECTIVE: Modeling tumor invasion and metastasis in Drosophila. Wayne O. Miles, Nicholas J. Dyson and James A. Walker

Research Articles

- A high level of liver-specific expression of oncogenic Kras \( V_{12} \) drives robust liver tumorigenesis in transgenic zebrafish. Anh Tuan Nguyen, Alexander Emelyanov, Chor Hui Vivien Koh, Jan M. Spitsbergen, Siew Hong Lam, Sinnakaruppan Mathavan, Serguei Parinov and Zhiyuan Gong
- Fetal overgrowth in the Cdkn1c mouse model of Beckwith-Wiedemann syndrome. Simon J. Tunster, Mathew Van de Pette and Rosalind M. John
- The feelgood mutation in zebrafish dysregulates COPII-dependent secretion of select extracellular matrix proteins in skeletal morphogenesis. David B. Melville, Mercedes Montero-Balaguer, Daniel S. Levic, Kevin Bradley, Jeffrey R. Smith, Antonis K. Hatzopoulos and Ela W. Knapik
- Harmonin (Ush1c) is required in zebrafish Müller glial cells for photoreceptor synaptic development and function. Jennifer B. Phillips, Bernardo Blanco-Sanchez, Jennifer J. Lentz, Alexandra Tallafuss, Kornnika Khanobdee, Srijan Ganpathy, Zachary A. Jacobs, Philip F. Han, Monalisa Mishra, David S. Williams, Bronya J. Keats, Philip Washbourne and Monte Westerfield
- Development and validation of a yeast high-throughput screen for inhibitors of \( \alpha B_{42} \) oligomerization. Sei-Kyoung Park, Scott D. Pegan, Andrew D. Mesecar, Lisa M. Jungbauer, Mary Jo LaDu and Susan W. Liebman
- Abca12-mediated lipid transport and Snap29-dependent trafficking of lamellar granules are crucial for epidermal morphogenesis in a zebrafish model of ichthyosis. Qiaoli Li, Michael Frank, Masashi Akiyama, Hiroshi Shimizu, Shiu-Ying Ho, Christine Thisse, Bernard Thisse, Eli Sprecher and Jouini Uitto

Research Reports

- A high-sugar diet produces obesity and insulin resistance in wild-type Drosophila. Laura Palanker Musselman, Jill L. Fink, Kirk Narzinski, Prasanna Venkatesh Ramachandran, Sumitha Sukumar Hathiramani, Ross L. Cagan and Thomas J. Baranski
- The inflammatory bowel disease (IBD) susceptibility genes NOD1 and NOD2 have conserved anti-bacterial roles in zebrafish. Stefan H. Oehlerls, Maria Vega Flores, Chris J. Hall, Simon Swift, Kathryn E. Crosier and Philip S. Crosier