Dementia in mouse models of neurodegeneration

C Janus*

Abstract

Introduction
Mouse models of human neurodegenerative diseases have recently dominated the field of translational research. These models make it possible to dissect the complexity of Alzheimer’s disease and to assess the pathogenic impact of individual factors in vivo. However, expression of the complex cognitive phenotype of human dementia in a mouse presents a formidable challenge limiting the translational potential of these models to only pre-symptomatic stages of disease. This review provides a brief discussion of issues pertaining to the design and interpretation of behavioural studies in translational research using mouse models of neurodegeneration.

Discussion
Dementia is an age-progressing phenomenon encompassing impairments in multiple memory systems, including abstract thinking and verbal communication. Mouse models of Alzheimer’s disease usually replicate only a subset of the full spectrum of brain pathology and manifest only mild cognitive dysfunctions. In order to improve the translational interpretation of the results generated in pre-clinical studies, it is important to establish the relevance of observation in a model’s phenotypes and pathological changes in specific brain regions to facets of human dementia and its underlying neural circuitry.

Unfortunately, none of the existing models manifests mnemonic dysfunction comparable to dementia.

Conclusion
Over-expression in the mouse of multiple copies of human genes implicated in a disease triggers complex developmental adjustments in the mouse genotype, manifested by great diversity of behavioural phenotypes. Only some of these phenotypes might represent biomarkers relevant to dementia. Meta-analysis of multiple behavioural screens should reveal subsets of behaviours that compromise ethologically relevant mouse phenotypes with underlying neuropathology that would be functionally relevant to the genetic design of the model. Translation of these results to a relevant clinical stage of a disease should be carried out within the framework and ramifications of the model.

Introduction
Sir Francis Bacon wrote, ‘a man is but what he knoweth’. This phrase defines one of our most valued traits in being humans, and in this respect, neurodegenerative diseases are devastating age-associated disorders that compromise and eventually destroy our cognitive functions, ‘de-humanising (dementia)’ our lives. Cognition is loosely defined with divergent meanings in different disciplines. Despite its inaccessibility as a research object and intrinsic complexity, an increasing number of psychologists, neuroscientists and geneticists study cognitive abilities in humans and animals. We hope that these multidisciplinary research efforts will increase our knowledge of neural mechanisms underlying cognitive abilities; why they decline in many neurological disease; and how we can model human dementia experimentally. In this respect, the main goal of translational research is to develop animal models, which will adequately represent cognitive profiles relevant to homologous systems in humans.

Behavioural genetics provides ample evidence that variability in cognitive behaviour is to a lesser or larger part genetically determined. General cognitive ability in humans, or ‘g factor’, is derived from the performance in a battery of mnemonic tests. The development of the equivalent test battery in animals would significantly improve translational aspect of research focusing on modelling dementia. The manipulation of genes and resultant changes in learning and memory should help elucidate molecular and cellular mechanisms crucial for coding, consolidation and memory recall. Deficits in these processes are the hallmark of mental retardation, senile dementias and dementias accompanying neurodegeneration; however, the underlying mechanisms might differ between diseases. Knowing the genetic contribution to cognition is also essential to determine the extent the environmental modifications can influence mnemonic behaviour. Such knowledge may be invaluable in lifestyle modification strategies to potentially prevent or ameliorate the severity of progressing dementias. The aim of this review was to discuss dementia in mouse models of neurodegeneration.

Discussion
The author has referenced some of his own studies in this review. The protocols of these studies have been approved by the relevant ethics committees related to the institution in...
which they were performed. Animal care was in accordance with the institution guidelines.

Genetic approaches to cognition in translational research

Behavioural molecular genetics investigate whether a specific gene is essential for or modifies cognitive function in mice. This reverse-genetics approach involves the manipulation of specific genes and the comparison of behaviour of genetically engineered mice with their wild-type control littermates. Gene targeting or transgenesis can disable, reduce or increase expression of genes. While the so-called constitutive mutant mice carry the mutation in all cells and throughout their life, conditional mutant mice carry the mutation only in a part of the brain and/or only during an experimentally restricted time in development. Furthermore, knock-in methods allow the mouse endogenous gene to be replaced with a wild type or mutated form of human gene thus further humanising the model.

These mutation-induced deficits in synaptic plasticity are evaluated in various mnemonic tests. However, performance in limited number of mnemonic tests cannot be uncritically equated with overall cognitive function and concomitant non-cognitive deficits might often cloud the interpretation of results. Therefore, combining mnemonic and non-mnemonic tasks in a test battery should significantly improve the validity of results. The matter is further complicated with the selection of tasks, their design and procedures that are still highly variable from laboratory to laboratory. The multitude of genes affecting learning and memory mirrors the complexity of synaptic processes. These genes code for proteins involved in exocytosis, hormones, receptors, signalling cascades, transcription and translation, and various membrane bound proteins such as cell adhesion molecules or postsynaptic density proteins; each of these processes can be affected by the expression of transgenes implicated in human disease.

Neurodegenerative diseases: genetics, pathology and mouse models

Dementias with neurodegeneration are characterised by a progressive decline in mental function that results from loss of the underlying neuronal architecture. The regional specificity and pathological hallmarks of each disease manifest behaviourally in patients as distinct, although sometimes partially overlapping, clinical entities. Alzheimer’s disease (AD) is one of the most prominent disorders characterised by progressive memory loss. The identification of gene mutations implicated in familial forms of AD (FAD) opened a new research field for disease modelling. FAD mutations are missense mutations in the genes encoding amyloid-β precursor protein (APP), presenilin 1 (PS1) and PS2. Although, no mutations directly associated with neurofibrillary tangles (NFTs) have been identified in AD, the accumulation of hyperphosphorylated forms of the microtubule-associated protein tau (MAPT) in NFT is the most obvious pathological hallmark of AD and other tauopathies. NFTs are ubiquitously present in a variety of other dementias such as frontotemporal dementia with Parkinsonism linked to chromosome 17, Pick’s disease, progressive supranuclear palsy, argyrophilic grain dementia, Creutzfeldt–Jakob disease, Down’s syndrome, Gerstmann–Sträussler–Scheinker disease, Hallervorden–Spatz disease, tangle-only dementia and other tauopathies. The discovery of FAD and over 30 mutations in MAPT made these genes powerful candidates for modelling of neurodegeneration in a mouse.

Pathological diagnosis of AD is based on the presence of extracellular senile amyloid beta (Aβ) plaques and intracellular NFTs found throughout specific brain regions. The bulk of the fibrillar Aβ consists of a 42 amino acid peptide (Aβ42), cleaved from APP by the γ-secretase proteolytic complex. There is strong human genetic evidence linking increased production of the highly amyloidogenic Aβ42 protein relative to the Aβ40 form to the disease. Both Aβ42 and a shorter form Aβ40 typically co-localise in amyloid plaques, and according to amyloid hypothesis, trigger a cascade of pathological events in AD (Figure 1). Studies using mouse models over-expressing human APP mutations refined the hypothesis and showed that cognitive decline might develop...
before the onset of widespread Aβ plaque deposition in the brain. These studies suggest that soluble Aβ assemblies, rather than deposited aggregated Aβ, may underlie the memory deficits15,16. It is important to stress that abnormal phosphorylation of tau protein and the formation of NFTs is not present in mouse models of AD-like amyloidosis (Table 1). The absence of this vital hallmark of neurodegenerative diseases in APP transgenic models creates a serious limitation in the translational power of these models.

The first successful mouse model replicating major hallmarks of AD was characterised more than a decade ago by Games and his colleagues17. The ability to study the effect of expressing human mutated genes linked to AD and other tauopathies in mouse models have proven to be extremely informative18. Although, mice never developed AD-like pathology spontaneously during their life span, the mouse brain develops many AD-like pathologies when exposed to high levels of human Aβ. Some abnormalities, including loss of calbindin in dentate gyrus19 or the relation between transforming growth factor and cerebral amyloid angiopathy20, were first identified in mouse models and subsequently identified in AD patients. Multigenic mice co-expressing genetic factors implicated in pathology of AD revealed synergisms between Aβ and apolipoprotein E421, Aβ and tau22, and Aβ and α-synuclein23. Translational research using these models also led to major advances in the development and testing of new therapeutic strategies24,25, many of which were consequently tested in clinical trials26,27.

Dementia in a mouse model

Dementia is the major clinical hallmark that signals symptomatic stage of AD28. Disturbance in other systems are often concurrent to progressing dementia29. It is likely that similar broad spectrum of behavioural changes occurs in APP mouse models, with only some being directly related to modelled brain pathology (Table 1). The goal of translational pre-clinical research is to identify these behavioural systems in a mouse.

A credible model should exhibit deficits in testing paradigms addressing memory systems relevant to mouse biology. The extent of age-related behavioural impairment would eventually encompass non-cognitive systems due to significantly progressing neuropathology. Although, this may raise operational complexities related to interpretations of results related to cognitive decline confounded by the late emergence of impairments in non-cognitive systems, drug screens might reveal which deficits in a model are ameliorated by a treatment at a given stage of pathology. The independent confirmation and replication of the results in several laboratories is desirable30; however, it may not be easily obtainable, even in well-established behavioural laboratories under supervision of experienced researchers31. Idiosyncratic differences, difficult to control and standardise, exist between various labs and animal colonies, including expertise of technical personnel and differences in handling methods32, which often make tests replication difficult. Robust phenotypes obtained in less labile tests (in which data collection is based on motor response to strong sensory inputs) are usually replicable within tolerable margin. More labile phenotypes based on emotional or social behaviours may be strongly affected by differences between laboratories33, or in less efficiently managed animal colonies34.

Testing cognition in a mouse

Despite similarities between murine and human brain, it can be challenging to draw definitive parallels between cognitive function in humans and mouse models. In order to maximise the alignment, we should first target tests that implicate cognitive systems that are well conserved across mammalian species and have clearly delineated function and neuroanatomy. To this end, spatial navigation meets this assumption. The system is highly conserved in mammals35, and its neuroanatomical structure and underlying synaptic plasticity have been intensively studied36. The involvement of the hippocampus in spatial memory in humans has also been demonstrated; patients with temporal lobe damage showed severe impairments in learning and recall of spatial locations and in solving spatial maze tasks37. However, there are some caveats in studying spatial navigation in mouse models. Mainly, the performance of mice in behavioural tests not only depends on cognitive propensity of a particular inbred strain or hybrid genetic background38 or even different suppliers39 but it may also be seriously confounded by the presence of idiosyncratic for particular strains deleterious mutations. Retinal degeneration (rd) caused by an autosomal recessive mutation (Pde6brd), which results in rapid age-progressing degeneration of rods and cones40, is most infamous, especially in visually guided spatial navigation test. Although nearly 20% of all inbred mouse strains carry this mutation, including C3H, FVB and SJL strains, which often contribute to hybrid genetic background of mouse models of neurodegeneration, the choice of a strain or hybrid genetic background of a model is crucial with respect to specific planned cognitive screens.

Mouse models of Alzheimer’s disease-like pathology and anti-amyloid beta therapy

Although the mouse models provided a powerful tool for investigating Aβ-centric therapeutics, the successes of active and passive anti-Aβ immunisation in preventing and
clearing parenchymal amyloid in these models were not, however, replicated in clinical trials that had to be prematurely halted when subset of patients developed meningoencephalitis. Furthermore, recent phase 3 trials testing two more refined anti-Aβ antibodies: bapineuzumab (the humanised IgG1 antibody of the mouse monoclonal antibody 3D6) and solanezumab (the humanised IgG1 antibody of the mouse monoclonal antibody 266) reported negative results. Also, attempts focusing on the lowering of the production of toxic Aβ42 species through inhibition of γ-secretase were unsuccessful. The phase 3 trial evaluating efficacy of semagacestat, a small-molecule γ-secretase inhibitor had to be stopped following the results that patients receiving the drug showed worse performance than patients receiving placebo. These negative results likely indicated that either the treatment was administered too late in the course of the disease, or Aβ alone is the wrong target for an effective treatment of AD. Both hypothetical explanations have some support from recent mouse studies. First, the significance of cognitive decline in a mouse model, based on limited number of tests without reporting the amount of variance accounted for by genotype and/or treatment effects, is bound to seriously bias translational application of the results to clinical trials. The cognitive impairment manifested in the models of amyloidosis is usually mild, likely reflecting stages of disease that precede the diagnosis of mild cognitive impairment (Table 1). It might be argued that the life span of a mouse is too short for the disease to progress in a manner analogous to its clinical stages. If such assumption is correct, the translation of results obtained from mouse models to symptomatic AD patients might present high experimental risk. Second, our recent research, using novel BRI2-Aβ mouse models that express Aβ40,

### Table 1 Pathological phenotypes manifested by transgenic mouse models of AD-like amyloidosis

<table>
<thead>
<tr>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Brain pathology</strong></td>
<td></td>
</tr>
<tr>
<td>Ageing- and region-specific Aβ deposition, neurotic dystrophy and reactive gliosis due to over-expression of human genes implicated in AD</td>
<td>Overt neurodegeneration is not observed in brain regions affected in AD. Synapse loss is either missing or only within amyloid plaques</td>
</tr>
<tr>
<td>Amyloid deposits were apoE-positive and were surrounded by activated astrocytes similar to AD amyloid pathology</td>
<td>No tau pathology and accumulation of NFTs</td>
</tr>
<tr>
<td><strong>Additional genetic manipulations are needed to impact other brain structures relevant to AD, such deletion of the CD45 gene, reduction of reelin expression, over-expression of mutant tau genes linked to other neurodegenerative diseases</strong></td>
<td></td>
</tr>
<tr>
<td>Tg mice manifest weakly activated microglia, which expressed low levels of the complement receptor CD11b</td>
<td>AD lesions had strongly activated microglia, which expressed high levels of CD11b</td>
</tr>
<tr>
<td>Immunostaining for complement proteins is weak in Tg mice</td>
<td>Immunostaining for complement proteins is very strong in AD deposits</td>
</tr>
<tr>
<td><strong>Additional AD pathologies such as aberrant neuronal reentry into the cell cycle and hyper-accumulation of lysosomes and lysosome-associated autophagic vacuoles are not present</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Behavioural impairment</strong></td>
<td></td>
</tr>
<tr>
<td>Age-progressing cognitive tests (variable between lines and tests) AD patients and hAPP Tg mice both show navigational deficits that are closely related to impairments of the hippocampus and entorhinal cortex</td>
<td>Only anterograde amnesia studied, retrograde amnesia not addressed. Many tests reveal only weak, albeit significant impairment. Severe dementia is not present</td>
</tr>
<tr>
<td>Spontaneous non-convulsive seizure activity in cortical and HP networks comparable to AD seizures</td>
<td></td>
</tr>
</tbody>
</table>

Apo, apolipoprotein; Tg mice, transgenic mice; AD, Alzheimer’s disease; HP, hippocampus; NFT, neurofibrillary tangles.

The list presents the advantages of the available models and how close they replicate the pathology of human AD, as well as their ramifications in modelling the full spectrum of the disease. References to original work are provided to phenotypes not discussed in the text.

Licensee OA Publishing London 2014. Creative Commons Attribution License (CC-BY)
Aβ42 or both Aβ40/Aβ42 peptides in the secretory pathway without the presence of high levels of the human APP protein, revealed no decline in established cognitive tests that demonstrate mnemonic impairment in APP transgenic mice.56. These results were confirmed independently by conditional suppression of human APP transgene at the stage of florid Aβ pathology in an APP mouse model, which restored cognitive function, despite the abundant presence of Aβ plaques48.

**Conclusion**

Finding a cure that will prevent or dramatically slow down the progression of dementia in AD is a major unmet medical need. Mouse models expressing FAD and/or MAPT mutations present a useful experimental tool for delineating many pathogenic factors required for driving disease progression. Despite the large body of evidence there are still major gaps in our knowledge regarding the relevance of mnemonic and non-mnemonic impairments seen in the mouse models to human dementia. Though it is generally acknowledged that these models exhibit a variety of behavioural alterations, including deficits in learning and memory, the temporal and quantitative relationship of these deficits to underlying pathologies is not consistent from model to model. It is no doubt that presently available models have limited utility in replicating advanced stages of dementia, where both tangible pathology and neurodegeneration almost certainly contribute to severe neuronal loss. The AD research community faces many unanswered questions, including: (1) the possible interaction between Aβ and APP derivatives to mediate cognitive function, (2) the existence of specific toxic species of Aβ and/or tau that trigger the decline in cognitive function or (3) the efficacy of available potential therapeutic agents in mediating cognitive recovery. Some answers to these questions have already been obtained in studies using mouse models. Other problems related to modulation of tau metabolism, autophagic function, cell cycle-related genes and kinases, and other potential pathogenic processes could only be addressed in studies using relevant mouse models. These future studies should elucidate the triggering mechanisms that initiate the massive neurodegeneration and consequent cognitive and behavioural symptoms of AD. The identification of mechanisms that drive progression of AD from its antecedent to its clinical phases beyond amyloid deposition may be important targets for novel disease modifying treatment strategies. Presently available mouse models seem to represent the wrong tool for the job of testing symptomatic patients, but at present it is the best and only tool we have.

**Abbreviations list**

Aβ, amyloid beta; AD, Alzheimer’s disease; APP, amyloid-β precursor protein; HP, hippocampus; MAPT, microtubule-associated protein tau; NFT, neurofibrillary tangles; PS, presenilin.

**References**

19. Palop JJ, Jones B, Kekonius L, Chin J, Yu GQ, Raber J, et al. Neuronal deple- tion of calcium-dependent proteins in the dentate gyrus is tightly linked to...


