Alzheimer’s disease (AD) is the most common form of age-related dementia, characterized by progressive memory loss and cognitive disturbance. Two pathological hallmarks are thought to be crucial in the disease: (1) amyloid plaques, primarily composed of the amyloid-β protein (Aβ), liberated from the amyloid precursor protein (APP) via serial cleavage by β- and γ-secretase, and (2) neurofibrillary tangles, composed of filamentous accumulations of aggregated hyperphosphorylated tau (reviewed in Tanzi and Bertram, 2005).

Familial, early-onset (<60 years), autosomal-dominant forms of AD (FAD) can be caused by fully penetrant mutations in the genes APP, presenilin 1 (PSEN1), and presenilin 2 (PSEN2). Sporadic AD (sAD) is the more common form of the disease, and usually involves late onset owing to multifactorial genetic and environmental risk factors (reviewed in Bertram et al., 2010). In a large twin study, the contribution of heritability to sAD was estimated to be as high as 80% (Gatz et al., 2006). The best-established genetic risk factor is the e4 allele of the gene encoding apolipoprotein E (APOE) (reviewed in Tanzi and Bertram, 2005).

Although postmortem studies have provided a wealth of knowledge on the pathological hallmarks of AD, postmortem brain samples often represent the end stage of the disease and therefore offer few if any clues regarding the etiology and molecular mechanisms underlying pathogenesis. Mouse models expressing FAD mutations have contributed invaluably to our current understanding of AD; however, they fail to fully recapitulate AD pathogenesis in humans as none exhibit β-amyloid-driven tau tangle formation. In contrast, genetic studies indicate that in humans, AD is a β-amyloid-induced tauopathy. Fundamental species-specific differences in genome and protein composition between mice and humans likely preclude the recapitulation of bona fide AD pathological events in animal models. While immortalized cell lines and primary rodent cell cultures have been useful for elucidating the molecular mechanism of AD pathogenesis, they do not adequately represent the environmental features of native neurons.

A previous study has shown that human fibroblasts can be reprogrammed, by transient expression of a small number of genes, into induced pluripotent stem cells (iPSCs) that functionally and phenotypically resemble embryonic stem cells (Takahashi and Yamanaka, 2006). Two recent studies, Yagi et al. (2011) and Israel et al. (2012), provide proof of principle that iPSCs can be used to model patient-specific AD pathology in vitro, recapitulating several pathological features of AD. Yagi et al. (2011) generated iPSCs from fibroblasts of FAD patients carrying mutations in PSEN1 (A246E; Sherrington et al., 1995) or PSEN2 (N141I; Levy-Lahad et al., 1995). Neurons differentiated from these iPSCs displayed elevated ratios of Aβ42 to Aβ40, a hallmark feature of FAD with presenilin or APP mutations (reviewed in Tanzi and Bertram, 2005). Israel et al. (2012) generated iPSC lines from six individuals, including two sAD patients (sAD1/sAD2), two FAD patients (harboring a duplication of APP; APPΔp), and two age-matched nondemented controls. Virtually all iPSC lines were shown to differentiate into neurons that form functional synaptic contacts, exhibit normal electrophysiological activity, and express GABAergic and glutamatergic neuronal markers. The iPSC-derived neurons from the APPΔp patients and the sAD2 patient exhibited significantly higher levels of secreted Aβ40 relative to control neurons, whereas the sAD1 iPSC-derived neurons did not.

Unfortunately, the authors were not able to provide data regarding the ratio of Aβ42 to Aβ40, as Aβ42 secretion was below the detection sensitivity of their system. The Aβ42/Aβ40 ratio is of particular relevance, since Aβ42 is considered the toxic aggregate-prone species and is the main component of β-amyloid plaques in AD. However, it should be noted that the number of neurons for each assay was relatively small and in most cases, the neurons generated may not have been fully mature. As such, future studies will need to measure Aβ42/Aβ40 ratios during extended culture periods, and preferably with larger numbers of neurons.

Although both APPΔp and sAD2 neurons were also shown to have significantly higher levels of phospho-Tau (p-Tau), abnormal tau protein accumulation or tangle formation was not observed. In future studies, it will be crucial to assess whether sAD and/or FAD iPSC-derived neurons can be induced to form actual neurofibrillary tangles, and whether treatments of iPSC-derived neurons with exogenous Aβ affect the biochemical dynamics of tau proteins. Interestingly, APPΔp and sAD2 neurons also had increased levels of active glycogen synthase kinase-3β (GSK-3β), a kinase that phosphorylates tau. Despite these cellular changes in Aβ, tau, and GSK-3β, iPSC-derived neurons from AD patients did not display synaptic loss, a feature correlated with the severity of dementia in AD. Whether this lack of synaptic pathology may be due to the limited culture duration is unclear, and will need to be further investigated.
Perhaps one of the most interesting findings in the Israel et al. (2012) study was that treatment with β-secretase inhibitors, but not γ-secretase inhibitors, reduced levels of p-Tau and active GSK-3β. This would argue that β-secretase-derived products of APP besides Aβ may affect downstream tau pathology. However, caution would be advised before firmly drawing such a conclusion because γ-secretase inhibitors can adversely affect neuronal differentiation and, perhaps, exert their own neurotoxicity, leading to tauopathy. Nonetheless, if this intriguing result were confirmed, it could challenge the amyloid cascade hypothesis that posits that Aβ is the sole initiator of tauopathy.

Collectively, Israel and colleagues have shown that iPSC technology can be used to observe phenotypes of AD patients, not only for FAD (Qiang et al., 2011; Yagi et al., 2011), but also for sAD cases. However, it is important to note that only one of the two sAD cell lines recapitulated the APPDp Aβ phenotype. Nonetheless, the observed phenotypic similarity between APPDp and at least one sAD patient suggests that the mechanism underlying autosomal-dominant forms of FAD may be relevant to the pathogenesis of sAD. Clearly, larger numbers of samples will be needed to more comprehensively investigate the phenotypic heterogeneity of sAD in future studies. Finally, tau-related changes were observed concomitant with the Aβ phenotype, indicating that human iPSC-derived neurons could prove particularly useful for more effectively assessing the functional relationship between Aβ and tau in AD pathogenesis.

Since the discovery of iPSCs, scientists have been excited by the potential of these cells for modeling diseases, screening drugs, and even being an option in cell replacement therapy. However, there remain numerous open questions regarding iPSC technology, including which cell lines serve as the best controls for AD-iPSCs. Are iPSCs from age/gender-matched healthy individuals or family members regardless of age/gender, or patient-derived iPSCs in which AD mutations have been converted to wild-type, the most appropriate? It is also fair to question whether a cell culture that contains only one or two cell types successfully represents AD, which is a complex disease involving both neuronal and glial activity. As such, future studies should be aimed at investigating cell-autonomous and non-cell-autonomous effects in a more diverse set of differentiated cell types generated from iPSCs. Particularly because nonneuronal cells are involved in Aβ clearance and inflammation, iPSC-derived glia from AD patients may be differentially affected. Finally, it is not clear whether genetically engineered iPSC-derived neurons are true representatives of primary human neurons because they exhibit gene expression patterns more similar to human fetal brain tissue (Israel et al., 2012), whereas AD affects the adult elderly brain. Despite these challenges, iPSC technology provides an innovative strategy for the study of AD and should greatly facilitate efforts to develop therapies for the treatment and prevention of this devastating disorder.

REFERENCES


