# Morphological profiling by cell painting in human neural progenitor cells classifies hit compounds in a pilot drug screen for Alzheimer's disease

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## 1 Abstract (351 words)

Alzheimer's disease (AD) accounts for 60-70% of dementia cases making it the most common form. Current treatments are partially effective, and novel treatments undergoing testing currently have questionable safety/efficacy. Therefore, there is a need to apply alternative strategies for AD drug discovery. Drug repositioning/repurposing can identify already approved compounds that may be effective. Age is the greatest risk factor for AD, and a demographic shift towards a more aged society means the number of AD cases is set to increase. In addition to age, genetic variants are important for defining risk-level for AD. Increased AD risk has been repeatedly linked to variants in *SORL1* (encodes the protein SORLA) from genome-wide association studies. Variants that confer loss of, or decreased, SORLA expression are also linked with AD. Deletion of *SORL1* is associated with enlarged endosomes in neural progenitor cells and neurons consistent with the role of SORLA (encoded by *SORL1*) in the endolysosomal pathway. Therefore image-based phenotyping may identify features characteristic of *SORL1* deletion. Here, an automated morphological profiling assay (known as Cell Painting) was applied to wild-type and *SORL1*$^{-/-}$ (and therefore SORLA-depleted) NPCs. This was used to determine the phenotypic response of *SORL1*$^{-/-}$ NPCs to compound treatment from a preliminary small library drug screen (TargetMol, 330 compounds; 100nM, 300nM or 1μM). Using morphological profiling as a multiparametric, image-based phenotyping method, we showed distinct phenotypic signatures for for *SORL1*$^{-/-}$ NPCs compared to to isogenic wild-type controls and predicted 16 FDA-approved compounds that reverse the morphological signatures. Bortezomib, ixazomib and

gemcitabine HCl reversed *SORL1*$^{-/-}$ after 100nM, 300nM or 1μM treatment, and further 13 FDA-approved compounds were effective at either 100nM, 300nM or 1μM. Network pharmacology analysis reveals the 16 compounds interact with X known targets and were predicted to target a further X proteins. These were enriched in X biological pathways, including ….. The findings suggest that image-based phenotyping by morphological profiling distinguished between *SORL1*$^{-/-}$ NPCs and wild-type isogenic controls, and predicted treatment responses that rescue *SORL1*$^{-/-}$-associated cellular signatures suggesting high-content image analysis is a viable option for drug repurposing and perhaps drug discovery in human neural cell models of Alzheimer's disease.