A clue to Alzheimer's disease in Down syndrome: a summary of my PhD thesis

by M. Florencia Iulita*

Alzheimer's disease is a common medical problem in older adults with Down syndrome. Given that the brain changes that lead to Alzheimer's disease develop decades before signs of dementia manifest, studies focusing on the early mechanisms of Alzheimer's onset might offer valuable clues for diagnosis and treatment. The recent discovery of a group of molecules responsible for the health of memory brain cells, and their dysregulation in Alzheimer's disease and Down syndrome brains, could provide new insights for the development of new biomarkers or the identification of novel therapeutic targets.

Alzheimer's disease and Down syndrome: a common link

Alzheimer's disease, the most common cause of dementia in the senior population, affects millions of people worldwide. Its most distinctive symptoms include the loss of memories, changes in judgement, thinking and behaviour, followed by the progressive loss of functional independence. Given the increasing life expectancy in our modern society, the number of Alzheimer patients is projected to continue rising, unless we find a cure or are able to diagnose it and treat it earlier.

The same is true in Down syndrome. Thanks to better medical care and improvements in social programs, life expectancy is now higher for individuals with Down syndrome, many of whom live thriving and healthy lives well into their 60s and even 70s. While this is certainly a most positive outcome, it also implies that they are now faced with the health challenges that arise during aging, including a higher risk of Alzheimer's disease.

Alzheimer's disease can be one of the main medical problems in older adults with Down syndrome. The reason behind this is that extra copy of chromosome 21, which harbours a gene known as *amyloid precursor protein* (referred as APP), from where a protein called *amyloid-beta* is produced. This is the very same 'sticky' protein that forms aggregates (known as *amyloid plaques*) outside brain cells in Alzheimer's disease patients (Figure 1). Although researchers have found that other genes in chromosome 21 are also important determinants of the cognitive difficulties faced by

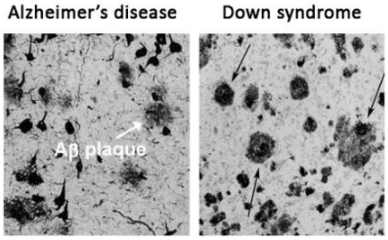

Figure 1. Microscope images of amyloid-beta (Aβ) plaques (arrows) in the brain of an Alzheimer's disease patient (left) and in an individual with Down syndrome (right). Images from links 1 & 2 (see Links for further reading), credit to Dr. Dale Bredesen and Dr. Elizabeth Head.

* Florencia Iulita
Département de Neurosciences, Université de Montréal, Montréal
florencia-iulita@mail.mcgill.ca

individuals with Down syndrome during aging, the brain build-up of amyloid-beta represents the most direct link with Alzheimer's disease. Indeed, microscopic examinations of amyloid plaques in Down syndrome and Alzheimer brains are almost indistinguishable (Figure 1). The main difference is that while the prevalence of Alzheimer's disease is as low as 5-6% in 50-60 year-old adults in the general population, it can be as high as 60-80% in adults with Down syndrome of the same age (see S&S Bulletin 2: Alzheimer's disease in adults with Down syndrome - a challenge).

When does Alzheimer's disease begin?

Traditionally, we tend to think of Alzheimer's disease in terms of its associated memory problems, which in fact reflects how dementia is diagnosed in the clinic. However, scientific advances in the past decades have revealed that the brain changes that lead to Alzheimer's disease begin to develop many years –even decades- before signs of memory loss and dementia manifest. In other words, there is a lag between the onset of Alzheimer's pathology (the changes that build-up in the brain) and its symptoms, such that the disease evolves 'silently' for years before it becomes symptomatic. This phase is known as *pre-clinical Alzheimer's disease.*

The silent progression of Alzheimer's disease is not unique to the elderly; it also happens in Down syndrome. This has significant implications. First, it means that if we were able to find indicators or *biomarkers* that signal that this 'silent' process has started, then we would have the opportunity to intervene earlier. Second, it highlights that in addition to representing a population that would greatly benefit from new Alzheimer treatments, studies in Down syndrome can also teach us about the early changes that occur in the brain as Alzheimer's disease develops.

But where to start?

Alzheimer's disease is a complex disorder, where not only amyloid-beta builds-up and disturbs the communication between nerve cells (neurons) in the brain; there are also aggregates of another protein known as *tau* that develop in parallel, as well as inflammation and loss of neurons. A key feature that distinguishes Alzheimer's disease from other brain disorders, such as Parkinson's or ALS (amyotrophic lateral sclerosis) is the selective vulnerability of a group of neurons, known as *cholinergic neurons of the basal forebrain*, which have key roles in the processes of learning, memory and attention. These neurons are peculiar because in order to remain healthy they depend on the continuous supply of a "nutrient" -or the proper term would be a *neurotrophin*- called *nerve growth factor* (*NGF)* throughout life. If NGF levels are compromised, these neurons can become sick and die.

Some years ago, a group of researchers led by neuroscientist Dr. Claudio Cuello from McGill University, Canada, described the mechanisms by which NGF is produced, released and processed in the brain – a finding that challenged older discoveries which suggested that all these events occurred inside the cell (1). They found that NGF is released from neurons in its immature form, and that it is processed into its mature molecule via the concerted action of a group of proteins that operate outside the neurons. They coined the term *NGF metabolic pathway* to refer to NGF and its associated markers.

The discovery of the NGF metabolic pathway further allowed them to examine its status in the brains of elderly individuals who had died of Alzheimer's disease. They found a severe increase in the immature form of NGF, which was due to a failed processing into its mature form, in addition to an enhanced degradation of the NGF molecule (2). Such findings suggested an improper production of NGF in Alzheimer brains and could explain the selective vulnerability of cholinergic neurons in Alzheimer's disease. Thus, a next question emerged. If Down syndrome and Alzheimer's disease share the same pathology, would NGF dysfunction also occur in Down syndrome?

My PhD thesis research

I joined Dr. Cuello's laboratory at McGill in 2009, to investigate the early stages of Alzheimer's pathology and its development in Down syndrome, as part of my doctoral studies. With Dr. Cuello, we embarked on a comprehensive project to examine whether individuals with Down syndrome who suffered from Alzheimer's dementia exhibited deficits in the metabolism of NGF, similar to those reported in the brains of Alzheimer's disease patients. To answer this question, we teamed with Down syndrome experts Dr. Jorge Busciglio at the University of California, Irvine, Dr. Thomas Wisniewski at the University of New York and Dr. Lotta Granholm at the Medical University of South Carolina.

We found that the brains of individuals with Down syndrome and Alzheimer's dementia showed the same pattern of alterations in NGF metabolism seen in elderly subjects with Alzheimer's disease; mainly, an elevation of the NGF immature molecule along with disturbed levels of the proteins involved in its maturation and degradation. The changes were pronounced in the frontal cortex (Figure 2), a region of the brain that is important for judgement, problem-solving and reasoning, and one of the earliest to develop amyloid plaques in Down syndrome. With the help of our colleagues, we further detected the compromise in NGF production and its increased degradation in a widely used mouse model of Down syndrome, as well as in Down syndrome fetal brain cells grown in culture (4).

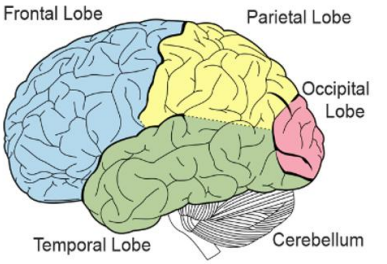

Figure 2. Schematic illustration of the different brain regions, known as lobes. Image from (3)

The results of this work strengthened the evidence of an NGF metabolic disconnection in Alzheimer's disease and Down syndrome, and provided a novel rationale to explain the preferential degeneration of basal forebrain cholinergic neurons in these conditions. Given that NGF alterations were found in fetal brain cells, it also suggested that NGF metabolic deficits might occur at earlier stages, before dementia manifests; a possibility warranting future investigations. A more extensive overview of this work is published in specific scientific reviews (5, 6).

Where next?

Considering that NGF metabolic deficits occur in the brains of individuals with Down syndrome and Alzheimer's disease, we next inquired whether these changes could be reflected elsewhere outside the brain, for example in body fluids such as blood. If this were true, this knowledge could open the door for the development of new screening and diagnostic tools for clinical application.

In subsequent studies done in collaboration with Dr. Filippo Caraci from the University of Catania, Italy, we revealed significant changes in markers of NGF metabolism and inflammation in plasma samples collected from adults with Down syndrome with and without Alzheimer's dementia. Interestingly, these markers appeared altered in subjects who did not yet manifest dementia symptoms. We also found associations between the levels of NGF markers and cognitive decline, which was measured over a two-year period (7). More extensive studies in larger cohorts are planned to validate these findings.

Ultimately, our hope is that the investigation of NGF metabolism in Down syndrome could assist the diagnosis of Alzheimer's disease in this population. Although clinicians are aware of the high risk of Alzheimer's disease in Down syndrome, a diagnosis of dementia has to be carefully differentiated from other potential conditions that could produce similar symptoms (e.g. normal decline associated with aging, depression, infections, etc.). Knowing that NGF and its related markers are affected in Alzheimer's disease and Down syndrome brains, and that these changes can be measured in blood, we could use this information to strengthen a suspected dementia diagnosis in parallel to traditional screening instruments. Going even further, if additional studies confirm that NGF dysfunction occurs before dementia symptoms emerge, these patients could be closely monitored and be offered the possibility to participate in clinical trials of new drugs to prevent or delay the onset of Alzheimer's disease. In other words, having new markers that could help clinicians perform more accurate diagnosis may allow earlier detection and earlier treatment when disease-modifying drugs become available.

Links for further reading

- Amyloid-beta and Alzheimer's disease. By Robert O'Brien, January 28 2015 http://sage.buckinstitute.org/amyloid-beta-and-alzheimers-disease/
- Studying Down syndrome might help us understand Alzheimer's disease better. By Elizabeth Head and Frederick Schmitt, May 5 2015 https://theconversation.com/studying-down-syndrome-might-help-us-understand-alzheimers-disease-better-36118
- M. Florencia Iulita PhD dissertation: *Studying Alzheimer's Disease Pre-clinical Stages: Insights from Down's Syndrome*

and Transgenic Animal Models
http://digitool.library.mcgill.ca/webclient/StreamGate?folder_id=0&dvs=1531778542761~626

- Alzheimer's Disease in People with Down Syndrome
https://www.nia.nih.gov/health/alzheimers-disease-people-down-syndrome
- Alzheimer's Disease & Down syndrome: A Practical Guidebook for Caregivers
http://www.ndss.org/wp-content/uploads/2017/11/NDSS_Guidebook_FINAL.pdf

Acknowledgements

My biggest thanks goes to my mentor Dr. A. Claudio Cuello for all his guidance and support during my studies, and for the continued collaboration. I am most grateful to our colleagues Jorge Busciglio, Thomas Wisniewski, Lotta Granholm and Filippo Caraci for the opportunity to work together. I have my deepest appreciation to the families and individuals with Down syndrome who enthusiastically participated in our plasma study. I would also like to thank the Trisomy 21 Research Society for the recognition of my PhD thesis, and for the opportunity to write this piece. Finally, I would like to thank Lisi Flores Aguilar for revising this text and for the always inspiring discussions about Down syndrome research.

References

1. Bruno MA, Cuello AC (2006) Activity-dependent release of precursor nerve growth factor, conversion to mature nerve growth factor, and its degradation by a protease cascade. *Proc Natl Acad Sci U S A* 103, 6735-6740.
2. Bruno MA, Leon WC, Fragoso G*, et al.* (2009) Amyloid beta-induced nerve growth factor dysmetabolism in Alzheimer disease. *J Neuropathol Exp Neurol* 68, 857-869.
3. Http://controlmind.info/human-brain/structure-of-the-brain
4. Iulita MF, Do Carmo S, Ower AK*, et al.* (2014) Nerve growth factor metabolic dysfunction in Down's syndrome brains. *Brain* 137, 860-872.
5. Iulita MF, Cuello AC (2014) Nerve growth factor metabolic dysfunction in Alzheimer's disease and Down syndrome. *Trends Pharmacol Sci* 35, 338-348.
6. Iulita MF, Cuello AC (2016) The NGF Metabolic Pathway in the CNS and its Dysregulation in Down Syndrome and Alzheimer's Disease. *Curr Alzheimer Res* 13, 53-67.
7. Iulita MF, Ower A, Barone C*, et al.* (2016) An inflammatory and trophic disconnect biomarker profile revealed in Down syndrome plasma: Relation to cognitive decline and longitudinal evaluation. *Alzheimers Dement* 12, 1132-1148.

Take home message

- Alzheimer's disease is a common medical complication in older adults with Down syndrome
- The changes that lead to Alzheimer's disease develop silently over many years until symptoms emerge
- Deficits in NGF metabolism explain the preferential degeneration of cholinergic neurons, both in Alzheimer's disease and Down syndrome
- Analysis of NGF markers in body fluids may help identify new biomarkers for early dementia detection