Neurobiology of Disease 77 (2015) 257–265

Contents lists available at ScienceDirect

Neurobiology of Disease

journal homepage: www.elsevier.com/locate/ynbdi

From molecule to molecule and cell to cell: Prion-like mechanisms in amyotrophic lateral sclerosis

Leslie I. Grad, Sarah M. Fernando, Neil R. Cashman *

Department of Medicine (Neurology), Brain Research Centre, University of British Columbia, 2211 Wesbrook Mall, Vancouver BC, Canada, V6T 2B5

ARTICLE INFO

Article history:
Received 6 January 2014
Revised 13 August 2014
Accepted 9 February 2015
Available online 17 February 2015

Keywords:
Amyotrophic lateral sclerosis
Propagated protein misfolding
Protein aggregation
Prion-like
Intercellular transmission
Exosomes
Superoxide dismutase 1 (SOD1)
TAR DNA-binding protein 43 (TDP-43)

ABSTRACT

Prions, self-proliferating infectious agents consisting of misfolded protein, are most often associated with aggressive neurodegenerative diseases in animals and humans. Akin to the contiguous spread of a living pathogen, the prion paradigm provides a mechanism by which a mutant or wild-type misfolded protein can dominate pathogenesis through self-propagating protein misfolding, and subsequently spread from region to region through the central nervous system. The prion diseases, along with more common neurodegenerative disorders such as Alzheimer's disease, Parkinson's disease and the tauopathies belong to a larger group of protein misfolding disorders termed proteinopathies that feature aberrant misfolding and aggregation of specific proteins. Amyotrophic lateral sclerosis (ALS), a lethal disease characterized by progressive degeneration of motor neurons is currently understood as a classical proteinopathy; the disease is typified by the formation of inclusions consisting of aggregated protein within motor neurons that contribute to neurotoxicity. It is well established that misfolded/aggregated proteins such as SOD1 and TDP-43 contribute to the toxicity of motor neurons and play a prominent role in the pathology of ALS. Recent work has identified propagated protein misfolding properties in both mutant and wild-type SOD1, and to a lesser extent TDP-43, which may provide the molecular basis for the clinically observed contiguous spread of the disease through the neuroaxis. In this review we examine the current state of knowledge regarding the prion-like properties of proteins associated with ALS pathology as well as their possible mechanisms of transmission.

Contents

Introduction

The identification of prions, or infectious proteins, over thirty years ago (Diener et al., 1982; Prusiner, 1982) as the transmissible agent responsible for a myriad of brain wasting diseases such as scrapie in

* Corresponding author.
E-mail address: neil.cashman@vch.ca (N.R. Cashman).
Available online on ScienceDirect (www.sciencedirect.com).

http://dx.doi.org/10.1016/j.nbd.2015.02.009
0969-9961/

sheep, bovine spongiform encephalopathy (BSE), also known as mad cow disease in cattle, and the devastating human diseases kuru and Creutzfeldt–Jakob disease (CJD), among others was a landmark development in the fields of infectious disease and protein biochemistry. The transmission of disease between organisms without the requirement of nucleic acid was deemed heretical for its time, and even now the concept faces occasional bouts of controversy. Since then, the singularity of pathological mammalian prion protein (PrP) as the sole example of a true infectious protein has come under constant challenge; from earlier studies in yeast (Patino et al., 1996) and other fungi (Uptain and Lindquist, 2002) to more recent work in human neurodegenerative diseases (Aguzzi, 2009; Guest et al., 2011). As such, aberrant protein misfolding and aggregation are a part of the pathobiology of numerous relatively common neurodegenerative disorders including Parkinson's disease (PD), the tauopathies and Alzheimer's disease (AD), known collectively as proteinopathies. Specific proteins in these diseases have been identified that appear to emulate the propagating protein mechanism of prions: amyloid-β (Aβ) in AD (Eisele et al., 2010; Meyer-Luehmann et al., 2006), α-synuclein in PD (Luk et al., 2012, 2009), and tau protein in AD, frontotemporal dementia (FTD) and other dementias (Clavaguera et al., 2009; Frost et al., 2009; Guo and Lee, 2011).

The level of experimental evidence describing the prion-like activities implicated in these diseases varies considerably. While evidence of a prion-like mechanism in AD and PD has populated the literature for over a decade, a relative newcomer to the prion paradigm is amyotrophic lateral sclerosis (ALS). When one examines the clinical pathology and epidemiology of ALS, its inclusion into the family of prion-like proteinopathies is quite reasonable. For example, the most significant clinical feature of ALS is its relentless, but contiguous spread through the neuroaxis; pathology starts at one or more focal points (Sekiguchi et al., 2014) of onset, possibly beginning in cortical neurons (Braak et al., 2013), and spreads in a spatiotemporal fashion through adjacent neuroanatomical regions (Ravits et al., 2007; Ravits and La Spada, 2009). Given the similarities in progression and anatomical spread among neurodegenerative proteinopathies, including the classical prion diseases, a likely mechanism to account for this observation that has gained traction in recent years is the region to region prion-like spread of propagated protein misfolding. In addition, this mechanism also accounts for how a mutant or even wild-type protein can dominate pathogenesis of a phenotypically diverse disease such as ALS, akin to when the normal cellular isoform of prion protein (PrP^{C}) converts to its pathologically dominant form PrP^{Sc} in prion disease. To date at least two proteins associated with the pathobiology of ALS have demonstrated multiple aspects of prion-like activity: SOD1 and TDP-43. This review attempts to summarize the known experimental evidence for prion-like activity of these proteins, and comment on their possible mechanisms of intercellular spread.

An introduction to ALS

ALS, also known as Lou Gehrig's disease in the United States, is a rapidly progressive fatal neuromuscular condition characterized by degeneration of the upper and lower motor neurons causing progressive paralysis and spasticity that affects the muscles of mobility, speech, swallowing and respiration (Bradley, 2009; Cleveland and Rothstein, 2001), although cognitive function is usually spared with only 5% of patients developing frank frontotemporal dementia (FTD) (Phukan et al., 2007). It is the most common form of motor neuron disease worldwide and has a global incidence of about two in 100,000. Half of affected individuals die within 3 years, and less than 20% survive for more than 5 years (Strong and Rosenfeld, 2003). The aetiology of ALS is unknown; however, similar to other neurodegenerative diseases such as AD and CJD, the disease can be divided into two categories, sporadic and familial. 90–95% of ALS cases are sporadic (SALS) where only some predisposing gene mutations have been identified, such as the ataxin-2 repeat expansions (Elden et al., 2010). The remainder of cases are familial (FALS) (Haverkamp et al., 1995), which are predominantly associated with Mendelian-inherited mutations in genes encoding Cu/Zn superoxide dismutase (SOD1), TAR-DNA binding protein 43 (TDP-43), fused in sarcoma/translocated in liposarcoma (FUS/TLS), and C9ORF72, but have been associated with mutations in other genes as well (ALS Mutation Database, 2007; Deng et al., 2011; Stewart et al., 2012; Wu et al., 2012). Clinically, both categories are very similar suggesting that a common downstream pathogenic mechanism, regardless of disease origin, may lie at the heart of the disease (Kabashi et al., 2007).

ALS is considered a protein misfolding disorder, based on its neuropathology, and as such is classified as a proteinopathy, similar to other neurodegenerative diseases. The post-mortem pathology of ALS patients typically features loss of motor neurons in the brain stem and ventral horn of the spinal cord accompanied by astrocyte activation and proliferation of microglia (Philips and Robberecht, 2011). Spinal cord histology often reveals abnormal accumulations of ubiquitinated proteinaceous inclusions in motor neurons and neural accessory cells, which are thought to be the result of the aggregation of misfolded protein. Protein misfolding can be triggered by a multitude of factors, including genetic mutation, oxidation, aberrant post-translational modification, dysfunctional proteostasis and seeded polymerization. In FALS cases where a SOD1 mutation is identified, the primary component of these protein inclusions is SOD1 itself (Kato et al., 2000). In sporadic cases where SOD1 mutations have been excluded there is recent evidence supporting the presence of misfolded SOD1-containing inclusions as well (Bosco et al., 2010; Forsberg et al., 2010; Grad et al., 2014; Pokrishevsky et al., 2012), although this observation is not one of consensus (Kerman et al., 2010). A more universally agreed observation is that TDP-43 is a primary component of cytosolic inclusions in the vast majority of SALS, in combination with its depletion from the nucleus, where the majority of native TDP-43 normally resides (Neumann et al., 2006).

Pathological SOD1 in ALS

Mutations in the gene encoding SOD1, a ubiquitously-expressed free-radical scavenging enzyme, were the first genetic cause of ALS to be identified (Rosen, 1993) and are implicated in ~20% of all FALS cases. The primary function of SOD1 is the conversion of the highly toxic free-radical superoxide to water and hydrogen peroxide (Beckman and Koppenol, 1996). To date, over 150 different disease-causing SOD1 mutations have been identified (ALS Mutation Database, 2007; Andersen et al., 2003). Despite the intrinsic stability of the native SOD1 enzyme, the majority of these mutations induce misfolding and subsequent aggregation. SOD1 aggregation occurs through a mechanism by which the highly-stable native SOD1 homodimer is disrupted producing misfolded monomer intermediates that can be incorporated into higher-order oligomeric structures (Rakhit et al., 2004, 2007). However, genetic mutation is not the only way to destabilize, misfold and aggregate SOD1. Aberrant oxidation or post-translational modification of wild-type (WT) SOD1 has been observed to mimic the aggregation-prone effects of mutant SOD1 *in vitro* (Casoni et al., 2005; Rakhit et al., 2004, 2002) in a concentration-dependent manner (Rakhit et al., 2004). There is increasing evidence that all types of ALS, including non-SOD1-linked familial and sporadic cases are associated with SOD1 misfolding, oxidation and aggregation (Matias-Guiu et al., 2008; Synofzik et al., 2012). Inclusions containing aggregated SOD1 have been detected in spinal cord tissues from both FALS and SALS patients (Chou et al., 1996a, 1996b; Shibata et al., 1994) in addition to biochemical, genetic and immunological evidence of misfolded SOD1 in cases of SOD1-excluded SALS (Bosco et al., 2010; Broom et al., 2008; Forsberg et al., 2010; Grad et al., 2014; Gruzman et al., 2007; Pokrishevsky et al., 2012). Misfolded SOD1 is therefore a prime