Review article

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Proteinaceous intracellular inclusions in neurodegenerative disorders

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Abstract

Neurodegenerative disorders are characterized by accumulation of "toxic", pathologic proteins in brain cells. Mutations in genes coding these proteins result in conformational disturbances of the protein structure and their accumulation and aggregation manifesting at the level of light microscope as various intracellular inclusions. This paper is an attempt of approach cellular mechanisms underlying neurodegenerative disorders with special attention to mechanisms of protein elimination.

Key words: *aggresome, α-synuclein, autophagy, proteasome, ubiquitin, tau protein.*

Introduction

While great attention and much research have been spent on understanding how the cell controls the synthesis of protein, publications dealing with the reverse process, the degradation of proteins, have been considered less important. A great contribution into the recognition of molecular mechanisms allowing cell to regulate the protein content honoured three scientists: Aaron Ciechanover, Avram Hersko and Irvin Rose by Nobel Prize for 2004 in chemistry. Their fundamental discovery of ubiquitin-mediated proteolysis helps to better understand at molecular level cell cycle, mechanisms of DNA repair and transcription, immune response and protein quality control. Defects in these systems have a casual role in many human diseases, including a group of neurodegenerative disorders called cerebral amyloidoses.

Cerebral amyloidoses have traditionally been defined as diseases in which normally soluble proteins physiologically present in cell (Table 1) accumulate as insoluble deposits inside the cell or in extracellular compartment. It is believed that proteinaceous aggregates initiate profound cell dysfunction often leading to its death. Disturbances in protein homeostasis resulting in protein aggregation constitute a common molecular pathomechanism of many neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, corticobasal degeneration, progressive supranuclear palsy, Huntington's disease and several

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Table I. Localization and functions of some intracellular proteins

Protein	Localization	Functions
ubiquitin	cytoplasm and nucleus	• protein labelling and elimination in proteasomes [24,33,74] • sodium-dependent uptake of various neurotransmitters [56]
tau	microtubules	• microtubule stabilization, fast axonal transport [51]
α-synuclein	presynaptic terminals nuclear membrane	• negative regulator of dopaminergic neurotransmission [1] • regulation of cellular lipid metabolism [14] • regulation of vesicle trafficking [59,69]
huntingtin	endoplasmic reticulum microtubules synaptic vesicles mitochondria	• regulation of retrograde axonal transport [22,65,85] • involved in protein degradation [2] • regulation of the iron pathway [36]
parkin	cytoplasm and nucleus	• stimulation of ubiquitin binding to specific substrates [75]

others. In familiar cases, pathological proteins are synthesized as a result of gene mutation while in sporadic cases, proteinaceous aggregates are formed due to pathological posttranslational modification.

In normal conditions, synthesized in ribosomes long polypeptide chains are folded forming three-dimensional structures specific to a certain protein. The folding process requires participation of many various proteins like chaperones and folding catalysts. In this process, polypeptide chains form two essential conformational structures: spiral-resembling structure called helix alpha and fanfold-like structure of beta type (β-pleated sheet). Normally folded proteins are transported to Golgi apparatus and then to other cellular organelles or outside the cell. Misfolded proteins are recognized by special cellular control mechanisms and eliminated.

Protein aggregation

Accumulation of misfolded or damaged proteins plays a key role in pathomechanism of neurodegeneration. But protein accumulation alone is not sufficient to their aggregation. Misfolded proteins are rich in β-pleated sheets of strong hydrophobic properties and have a tendency to self-aggregation. Aggregation is a process similar to crystallization and starts from nucleation – a poorly understood phenomenon leading to formation of a centre of the future aggregate (nucleus). Aggregate nucleus is composed of protein oligomers around which peptide monomers assemble and aggregate forming initially small fibrillar structures called protofibrils and then bigger fibrils. Nucleation and aggregation require energy and are detrimental from the point of view of cell kinetics. Therefore, nucleation and aggregation are slow processes that can explain relatively late onset of symptoms in degenerative diseases [74].

Physiological mechanisms of elimination of misfolded/damaged proteins

Ubiquitin-dependent system of proteasomes

Misfolded or damaged proteins are recognized by a 76-amino-acid-long polypeptide called ubiquitin. Since the polypeptide was found in many different tissues and organs, it was given the name from the Latin word *ubique* meaning everywhere. Normally, ubiquitin immunoreactivity in cell is diffuse; however, in various pathological conditions, it concentrates in inclusion bodies. Ubiquitin is involved in the sodium-dependent uptake of various neurotransmitters in the cerebral cortex [56]. It also recognizes pathological proteins and labels them binding covalently to their molecules that is referred as ubiquitination. Since labelled proteins are transported then to special protein-degrading system and destroyed there, ubiquitination may be treated as a molecular ”kiss of death”. The degradation of ubiquitin-labelled pathological proteins to small peptide fragments takes place in cell waste disposers called proteasomes [21,24,33] (Fig. 1). Proteasomes are cylindrical structures containing proteases. They are detected in cytoplasm, nuclei, dendrites, axons and synaptic buttons of various CNS cell types [55]. Ubiquitin -proteasome system

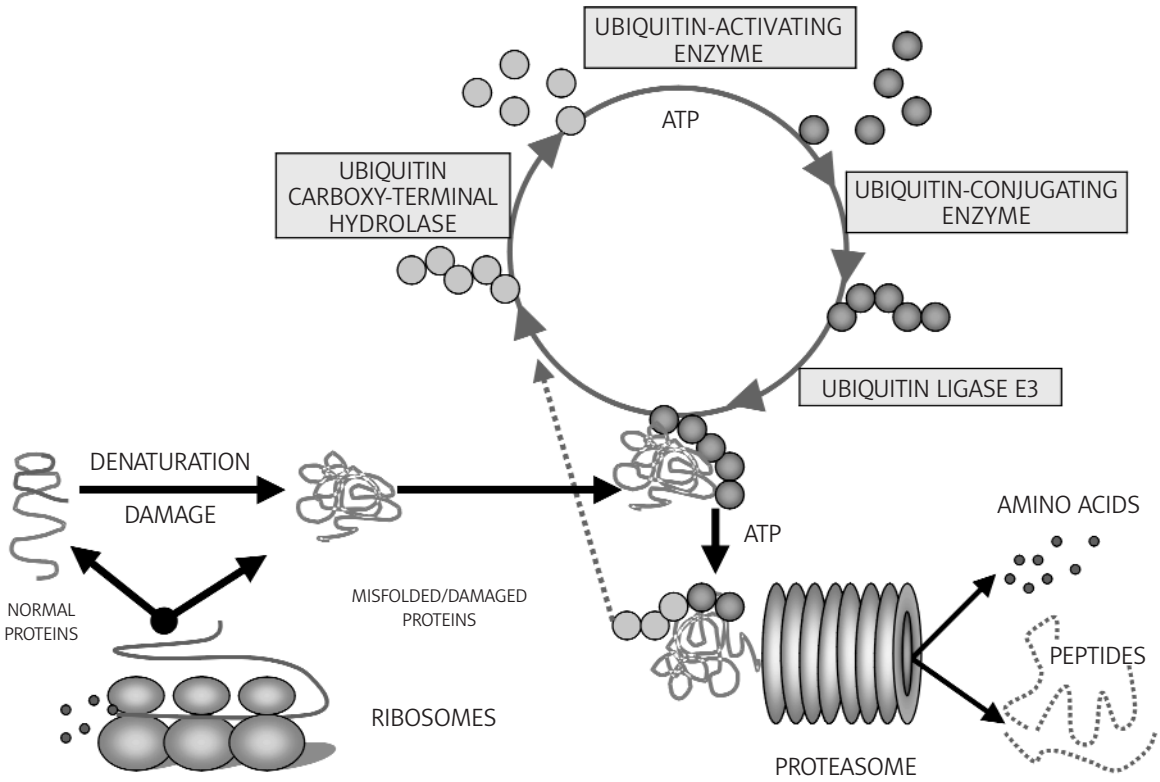

Fig. 1. The ubiquitin-proteasome system

can break-down practically all proteins to several amino-acid-long peptides.

Thanks of the presence of proteasomes the process of the enzymatic degradation of pathological proteins is spatially limited, what prevents other cellular proteins from damage by proteolytic enzymes. In neurodegenerative diseases, one of the possible causes of protein aggregation may be insufficiency of ubiquitin-dependent eliminating mechanisms resulting in accumulation of pathological proteins, dysfunction of the cell and its death [6,78]. Because both ubiquitination of misfolded/damaged proteins and their degradation in proteasomes require energy from ATP, there is a conception that the process of formation of proteinaceous aggregates may be due to the defect of mitochondrial respiratory complexes [24].

Formation of enclosed proteinaceous deposits – aggresomes

An interesting hypothesis concerning the pathomechanism of formation and elimination of pathological proteinaceous aggregates has been published lately in *The Lancet Neurology* [62]. It originally referred to formation of Lewy bodies but can have wider implications because presence of Lewy bodies is not only limited to Parkinson's disease. According to this hypothesis, proteinaceous aggregates are locked in specific cytoprotective cellular structures called aggresomes. Aggresomes are formed at the centrosome – a perinuclear organelle participating in regulation of microtubule assembly and organisation. Centrosomes play a principle role in mitotic cell division, but according to recent studies, they are also involved in elimination of misfolded/damaged proteins [47]. Accumulated in cell "unwanted" proteins are actively transported along microtubules to the centrosomes. Simultaneously, ubiquitin and various active factors participating in protein elimination also accumulate in the centrosome. Deposits of proteinaceous aggregates and ubiquitin-dependent components of the eliminating mechanisms in the centrosome can be visible on electron microscope as

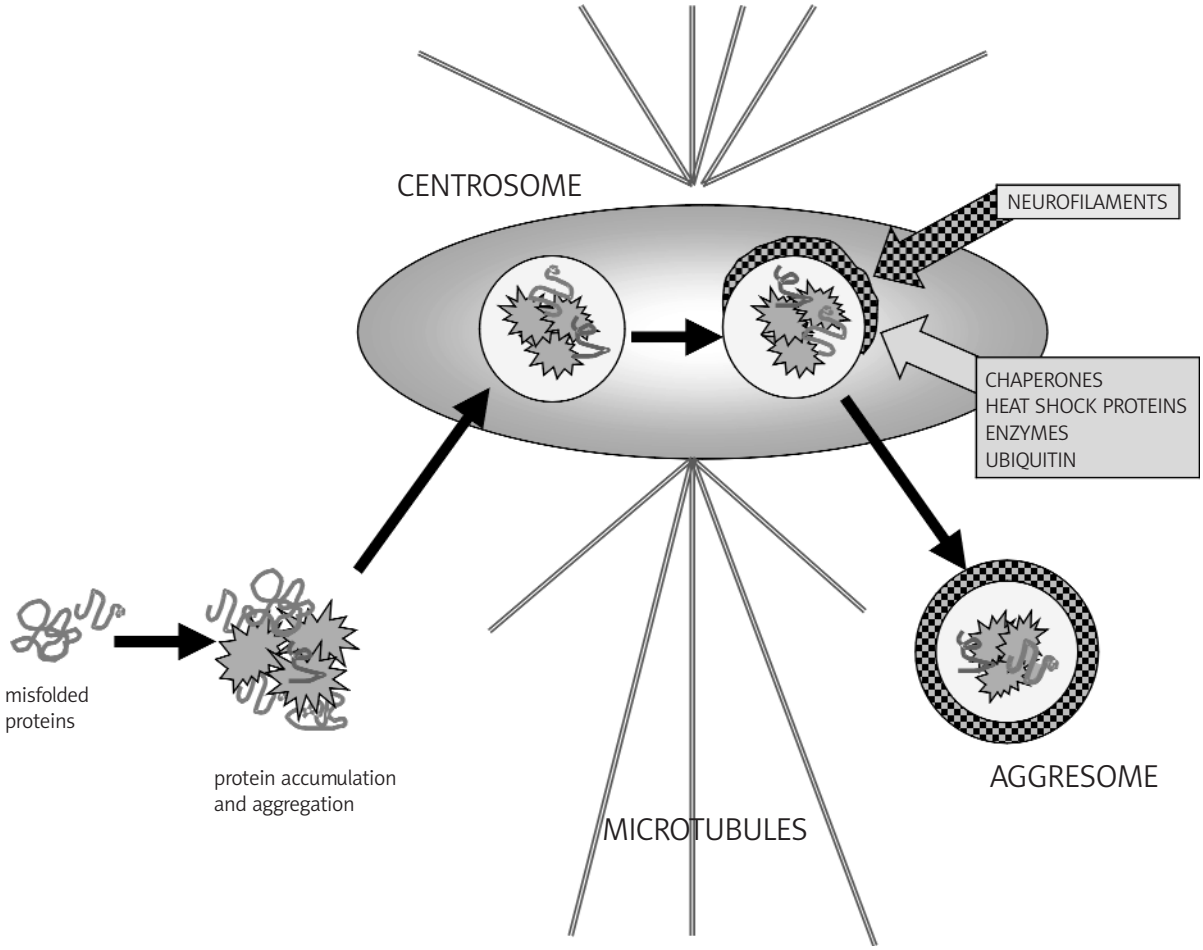

Fig. 2. Formation of aggresome

electron-dense areas in the central part of the cell. Then, these deposits are surrounded by intermediate filaments forming a structure referred as aggresome (Fig. 2). Aggresomes can be eliminated from the cell on the way of autophagy or can "survive" as insoluble cytoplasmic inclusions.

It is believed that aggresome formation can represent a protective mechanism allowing cell to neutralize "unwanted" proteins through their isolation. Inhibition of this process due to disturbances in microtubular transport of the pathological proteins may be one of the factors leading to cell degeneration.

Lysosomal degradation (autophagy)

Cytoplasmic components of the cell are degradated within lysosomes by three ways:

microautophagy, chaperone-mediated autophagy and macroautophagy [19,45]. In mammalian cells, microautophagy has not been well characterises. The second cellular mechanism of protein elimination, chaperone-mediated autophagy, involves a highly specific subset of cytosolic proteins with a motif recognized by chaperones from the heat shock protein family [17,19]. Following binding of the chaperone-substrate complex to a lysosomal membrane receptor, substrate proteins are translocated inwards the lysosome for degradation by hydrolases [19]. Degradation by chaperone-mediated autophagy is one of the mechanisms of α-synuclein elimination [18].

Macroautophagy is the major non-specific pathway for general turnover of cytoplasmic components. In this process, portion of cytoplasm

Table II. Localization of intracellular inclusions in neurodegenerative diseases

Disease	neuronal inclusions		oligodendrocyte inclusions	astrocyte inclusions
	intracytoplasmic	intranuclear		
amyotrophic lateral sclerosis	+	–	+	+
Alzheimer's disease	+	–	+	–
corticobasal degeneration	+	–	+	–
familial encephalopathy with neuroserpin inclusion bodies	+	–	–	–
Huntington's disease	+	+	–	–
Lewy body disease	+	–	–	+
neuronal intranuclear inclusion disease	–	+	–	–
multiple system atrophy	+	+	+	+
Parkinson's disease	+	–	+	+
Pick's disease and other frontotemporal dementias	+	–	+	+
progressive supranuclear palsy	+	–	+	+
spinocerebellar ataxias	+	–	+	+

are sequestrated within double-membrane vesicles known as autophagic vacuoles. Delivery of autophagic vacuole to lysosome depends on microtubules [46]. Then, they are docked and fused with the lysosome and their content breakdown. Elevated level of macroautophagy was found in Parkinson's disease [4].

Neurotoxicity of proteinaceous aggregates

In published lately research papers, there was emphasized that not proteinaceous aggregates are responsible for cell degeneration, but their intermediate structural forms. Large proteinaceous deposits are probably non-active or play a cytoprotective role through the isolation of misfolded/damaged proteins and prevention them from interaction with other components of the cell. Neurotoxic influence on the cell can exert protofibrils and soluble protein oligomers [2]. They probably influence functions of ionic channels and cause an increase of the cell membrane permeability [15,48]. They can also affect other cell structures and intracellular signalling pathways disturbing an exchange of molecular signals. An example of the last type of disturbances is inhibition of the transcriptional activity by mutated huntingtin in Huntington's disease [27].

In most of the degenerative diseases pronounced loss of neurons is observed. Damaged cells containing "unwanted" proteins are eliminated by the organism probably on the way of apoptosis [4]. The process makes it possible to remove pathological cells without triggering an inflammatory reaction and damaging to neighbouring cells.

Intracellular proteinaceous inclusions in neurodegenerative disorders

In neurodegenerative diseases, intracellular inclusions can be found in cytoplasm, nucleus and/or processes of neurons, oligodendrocytes and astrocytes (Table 2).

Neuronal intracytoplasmic inclusions

- **Lewy bodies (LB)**

LB are neuronal cytoplasmic inclusions characteristic of Parkinson's disease, but they were also found in several other neurodegenerative disorders such as Lewy body dementia [76], some forms of prion and Alzheimer's diseases [30], Down's syndrome [70], infantile neuroaxonal

dystrophy [35], neurodegeneration with brain iron accumulation type 1 (formerly Hallerworden-Spatz syndrome) [29,61] and subacute sclerosing panencephalitis [31]. Single LB were observed also in 4-13% of healthy elderly population [51] and described as incidental Lewy body.

LB were found not only in cytoplasm of neurons but also in neurite terminals [10,54] which probably constitute a place of LB formation [54]. LB are composed of various components which can be divided into four groups on the ground of immunohistochemical findings: (1) structural fibrillar elements, (2) proteins being cell answer to LB, (3) enzymes and (4) cytosol proteins absorbed by forming LB [66]. One of the LB components is ubiquitin. The presence of ubiquitin in LB suggests that these inclusions can be a structural manifestation of cytoprotective mechanisms eliminating damaged cellular elements [52].

Another important compound of LB is α-synuclein detected for the first time in 1988 in synaptic endings of the electric eel *Torpedo californica* [53]. Alpha-synuclein is a protein of presynaptic terminals and nuclear membrane (the name syn-nuclein originates from names of these two structures). Aggregation of α-synuclein and formation of insoluble proteinaceous complexes are detrimental to development of LB [80]. In Parkinson's disease, LB formation can be associated with dysfunction of ubiquitin-proteasome pathway due to inhibition of mitochondrial complex I [24]. According to other hypothesis, LB can be a form of aggresomes and can delay damage to neurons [62].

Probably, there are several molecular mechanisms of α-synuclein elimination from the cell. Some studies failed to show alteration of intracellular α-synuclein level by proteasomal inhibition [3,72] while others revealed that lysosomal inhibitors increased the protein level [49,82]. These data suggest that there are some alternate forms of α-synuclein degradation and one of them can be autophagy. In synucleinopathies, the pathologic α-synuclein may bound to the receptor for chaperone-mediated autophagy on the lysosomal membrane and blocks it inhibiting both their own degradation and that of other substrates [17].

Except α-synuclein, LB contain also other important protein – parkin. Normal parkin has got properties of ubiquitin ligase (Fig. 1) and stimulates binding of misfolded proteins to ubiquitin before their degradation in proteasomes. Thanks to that, parkin inhibits degeneration of nerve cells and prevents them from apoptosis [24,75]. Parkin dysfunction was detected in autosomal recessive juvenile form of Parkinson's disease.

- **Lewy neurites (LN)**

Except LB, other morphological hallmark of Parkinson's disease and Lewy body dementia are LN. In immunohistochemical reactions, these degenerating neuronal terminals demonstrate reactivity to ubiquitin, α-synuclein and some isoforms of tau protein. Electron microscopy revealed intermediate filaments as a major component of LN. In Parkinson's disease, LN are found in the same structures as LB. In Lewy body dementia, characteristic localization of LN is hippocampus although they are also found in amygdala, basal forebrain, substantia nigra, pedunculo-pontine and raphe nuclei and neocortex [references in: 40]

- **Neurofibrillary tangles (NFT)**

NFT are neuronal intracytoplasmic fibrillar inclusions made up largely of tau protein. Tau protein is a compound of microtubules – fibrillar cell structures responsible for lengthening of neurites, quick axonal transport and stabilization of microtubules. Functions of the tau protein are regulated at two levels: at the level of mature cell through tau phosphorylation and at the genetic level decisive for its synthesis. Phosphorylated in excess tau protein or insufficient tau phosphorylation makes the protein less stable and diminishes its ability to bind to microtubules. Regulation of tau protein functions at the genetic level is reached on the way of synthesis of its isoforms of different affinity to microtubules and different structural stability [50].

In tau disorders, two kinds of mutations were detected: missense mutations disturbing tau phosphorylation and intronic mutations influent on the synthesis of tau isoforms. Both types of mutations cause disturbances in tau functions and its destabilization and aggregation visible at the light microscope in the form of fibrillar intracellular inclusions [50].

NFT are hallmark of tauopathies, a group of diseases characterized by extensive accumulation of tau protein. The group involves such neurodegenerative disorders as Alzheimer's disease, some forms of prion diseases and frontotemporal dementias, corticobasal degeneration and progressive

supranuclear palsy. NFT immunoreactive with specific isoform of tau protein, but not with beta-amyloid, were observed in amyotrophic lateral sclerosis [85].

In progressive supranuclear palsy, NFT in a form of globose or flame shaped inclusions were found in basal ganglia, oculomotor nucleus and substantia nigra. They are similar to NFT in Alzheimer's disease and may be distinguished from them by the paucity of ubiquitin immunoreactivity [8,16]. Hitherto existing research have not revealed any mutation in the tau gene in progressive supranuclear palsy. It indicates that structural disturbances of tau protein have a character of posttranslational modifications and can be caused by excessive tau phosphorylation making the protein resistant to proteolytic degradation in proteasomes [79].

Corticobasal degeneration is another neurodegenerative disorder in which NFT are found. They are wispy and mostly globose (so-called corticobasal bodies) and immunoreactive to tau protein. Dominant location of the pathology is cerebral cortex. Corticobasal degeneration was first described in 1968 by Rebeiz et al as "corticodentatonigral degeneration with neuronal achromasia" [71]. The term "achromasia" referred to characteristic appearance of cortical neurons which were enlarged, pale and with lack of Nissl substance. Such cells were later named ballooned neurons and are analogous to Pick's cells in Pick's disease [26]. In corticobasal degeneration, except ballooned cells and NFT, conglomerates of degenerated astroglial processes called astrocytic plaques are seen.

• Pick's bodies (PB)

PB are round, argyrophilic intraneuronal inclusions characteristic of Pick's disease – a rare cerebral amyloidosis associated with clinical syndrome referred as frontotemporal dementia. Clinically diagnosed frontotemporal dementia is a hallmark of a group of neurodegenerative diseases also called frontotemporal dementias involving – except Pick's disease-motor-neuron disease-associated dementia, frontal lobe degeneration, corticobasal degeneration and frontotemporal dementia with parkinsonism linked to chromosome 17. There are still many controversies if some of frontotemporal dementias are separate entities or only a form of the same disease [43,60]. Pick's disease, corticobasal degeneration and frontotemporal dementia with parkinsonism belong also to the larger group of diseases called tauopathies.

Pick's disease is characterized by frontotemporal atrophy with neuronal loss, astrocytosis and neuropil microvacuolation, mainly in the II cortical layer. At light microscopy, except PB, swollen enlarged neurons called ballooned cells and cytoplasmic fibrillar inclusions in neurons and glial cells are found. Morphological picture of Pick's disease is similar to other frontotemporal dementias making neuropathologists much diagnostic difficulties. In human CNS six form of tau protein are generated. Since molecular profiles of tau aggregates are different and disease specific [34] immunohistochemistry may help in differential diagnosis [9].

In corticobasal degeneration, rounded intraneuronal inclusions similar to PB were found but opposite to Pick's disease, not in dentate gyrus [44]. Both types of inclusions contain aberrantly phosphorylated tau protein, but each of them demonstrates a distinct isoform pattern on Western blots [25].

• Hirano bodies (HB)

These rarely seen intraneuronal inclusions have a form of eosinophilic rods or round cytoplasmic deposits. They were found in Parkinson-dementia complex of Guam [37], Alzheimer's disease [58], Pick's disease [73] and in healthy elderly individuals. Although HB preferentially occur in the neuronal soma and neurites, occasionally they can be seen as small inclusions, intermingled with neurofibrillary tangles and in association with senile plaques [58].

Immunologically, HB are positive to actin and actin-associated proteins (tubulin, vinculin, alpha-actinin) and tau protein but not to ubiquitin [28].

• Collins bodies (CB)

CB are intraneuronal inclusions found in autosomal dominant disease causing a progressive dementia and called familial encephalopathy with neuroserpin inclusion bodies (FENIB) [11]. The disease is a result of point mutation in the gene coding serine protease inhibitor – neuroserpin. Neuroserpin and other serpins belong to a family of proteins that inhibit serine proteinases – proteolytic enzymes involved in many various physiological functions such as among others, formation of

synapses and neuronal plasticity [57]. Mutation in neuroserpin gene results in synthesis of unstable neuroserpin protein that readily aggregates into intraneuronal inclusions. Inclusions are PAS-positive and diastase-resistant but distinctly different from any previously described entity, including Lewy bodies and Pick bodies. CB are distributed throughout the cerebral hemispheres but are significantly more numerous in cerebral cortex and substantia nigra.

Neuronal intranuclear inclusions

Neuronal intranuclear inclusions are rare histological findings observed only in a few neurodegenerative diseases. They are the common neuropathology for a group of inherited disorders caused by expansion of CAG triplets coding amino acid glutamine. Neuronal intranuclear inclusions contain one of proteins rich in polyglutamine repeats such as huntingtin, ataxins, atrophin or androgen receptor. They were observed in the brains of patients with spinocerebellar ataxia type 7 [39], 3 and 1 [23], dentato-rubro-pallido-luysian atrophy [23] and Huntington's disease.

Huntington's disease is an autosomal dominant disorder manifesting as progressive dementia and extrapyramidal motor system disturbances in a form of chorea. Pathogenesis of the disease is connected with a mutation in the gene encoding protein huntingtin. Physiologically, huntingtin is present in the cytoplasm of most of cells in the body. In brain, its expression is predominantly observed in neurons. Huntingtin has no great structural homology with other human proteins, so determination of its normal function has proved difficult. Many data indicate that the protein is important for cell survival and participates in normal axonal transport [22,65,85], protein degradation [2] and iron metabolism [36].

In Huntington's disease, rich in glutamine mutant huntingtin is demonstrated as fibrillar inclusions in neuronal cytoplasm, nucleus and neurites [85]. These inclusions are especially numerous in cortical neurons. Except huntingtin, they also contain ubiquitin.

There are several hypotheses concerning the pathomechanism of degenerative process in Huntington's disease. One of them assumes that observed in the disease dysfunction of striatal neurons can be due to lack of beneficial influence of neurotrophic factors synthesized by cortical neurons. Formation of pathological aggregates of mutant huntingtin in cortical neurons not only can cause the decrease of neurotrophic factor synthesis but also blocks their axonal transport to striatal neurons [2]. Another potential cause of the neuronal dysfunction and degeneration in Huntington's disease can be disturbances in transcription process due to binding of mutant huntingtin with transcription factors and various intracellular signalling proteins vital to the normal neuron functions [22,27].

Intranuclear inclusions are especially numerous in a rare familial or sporadic degenerative disease called neuronal intranuclear inclusion disease (NIID) and characterized by progressive ataxia. In NIID, inclusions were found only in nucleus and intensely immunopositive to SUMO-1 [67], a protein which acts similarly to ubiquitin. SUMO-1 covalently conjugates to other proteins and targets them to the nuclear regions responsible for nuclear proteasomal degradation.

Argyrophilic neuronal intranuclear inclusions were also described in multiple system atrophy [12]. Their compounds are abnormal filamentous form of α-synuclein.

Glial cell inclusions

Glial inclusions (GI) have received little scientific attention compared to the abundance of information dealing with intraneuronal aggregates. At light microscopic level, most of GI are various in size and shape argyrophilic intracytoplasmic inclusions displacing the glial nucleus into an eccentric position. Only a few authors described GI in glial nuclei [64]. Ultrastructurally, GI are composed of a meshwork of randomly arranged, loosely packed filaments. Most of them are immunopositive to ubiquitin and β-crystallin, some of them to tau protein or heat shock proteins [13].

GI inclusions are located in oligodendroglial cells or/and astrocytes and may have a characteristic form dependently on type of the "host" glial cell.

Oligodendroglial inclusions

Oligodendroglial cytoplasmic inclusions have been observed in: parkinsonism-dementia complex

of Guam and amyotrophic lateral sclerosis [63,85], Parkinson's disease [81], spinocerebellar ataxia type 1 [32] and type 2 [68], Alzheimer's disease, Pick's disease, dementia with argyrophilic grains, corticobasal degeneration, progressive supranuclear palsy, subacute sclerosing panencephalitis and multiple system atrophy [references in: 68].

• Sickle- or flame-shaped argyrophilic glial inclusions

Oligodendroglial inclusions resembling neurofibrillary tangles are especially numerous and widespread in sporadic cases of multiple system atrophy – a neurodegenerative disorder clinically characterized by parkinsonism, autonomic dysfunctions and cerebellar disturbances. Under microscopic examination of brains with multiple system atrophy, two kinds of inclusions were found: argyrophilic intranuclear and intracytoplasmic deposits in oligodendrocytes and filamentous inclusions in neurons. Ultrastructurally, GI consist of loosely assembled fibrils and granular material which can be fragments of cellular organelles such as mitochondria or secretory vesicles [7]. Main proteinaceous component of intracellular inclusions is α-synuclein. Except α-synuclein, other proteins such as ubiquitin, tau and various cytoskeletal proteins were found. The presence of inclusions immunopositive to α-synuclein allows to include multiple system atrophy to synucleinopathies – a diverse group of neurodegenerative illnesses sharing a common pathologic inclusions composed of insoluble α-synuclein aggregates.

A small number of such GI may be seen in corticobasal degeneration [20].

Lately described in spinocerebellar ataxia type 2 GI were variably in size and shape and localized in oligodendroglial cells in the dentate nucleus, cerebellar white matter, transverse fibers of pons and inferior olivary nucleus. These cytoplasmic inclusions revealed immunoreactivity to ubiquitin. Unlikely to similar inclusions observed in multiple system atrophy, they were immunonegative to α-synuclein and tau protein [68].

Oligodendroglial inclusions positive to α-synuclein and negative to tau protein were seen in Parkinson's disease [81]. They were found not only in regions showing neuronal loss and gliosis, but also in areas without these morphological changes, such as cerebral cortex and white matter, striatum, globus pallidus, thalamus, cerebellum and spinal cord [38].

• Coiled bodies (CB)

CB are oligodendroglial deposits having a form of cytoplasmic filamentous inclusions. They appear as a bundle of filaments wrapped (coiled) around glial nucleus and extended towards cell processes. In immune reactions CB are positive to tau protein and negative to α-synuclein. Such kind of morphological changes was observed in Alzheimer's disease, Pick's disease, dementia with argyrophilic grains, corticobasal degeneration, progressive supranuclear palsy, subacute sclerosing panencephalitis [references in: 68] and amyotrophic lateral sclerosis [85].

Astroglial inclusions

In some neurodegenerative diseases GI were observed also in astroglial cells, where they can have a form of tufted or thorn-shaped astrocytes.

• Tufted astrocytes

Tufted degenerating astrocytes are the most characteristic morphological features of progressive supranuclear palsy [77]. They were also found in amyotrophic lateral sclerosis [85], Pick's disease [83], Parkinson's disease [81], Lewy body dementia [5] and cases of parkinsonism after Economo encephalitis [41]. Tufted astrocytes have a form of conglomerates or tufts of thin radiating fibers with an astrocyte-appearing nucleus in the centre of the tuft. They are immunoreactive with antibodies against abnormal tau protein.

• Thorn-shaped astrocytes

Thorn-shaped astrocytes typically are located in the subpial region and show much more cytoplasmic staining then tufted astroglial cells. They have no apparent disease specificity and were found in many neurodegenerative disorders such as Pick's disease, progressive supranuclear palsy, Alzheimer's disease, Lewy body disease, parkinsonism after Economo encephalitis, dementia pugilistica and in healthy elderly individuals.

• One more different type of astroglial deposits, granular hazy inclusions, were seen in astrocytes in parkinsonism-dementia complex of Guam and amyotrophic lateral sclerosis [63].

Table III. "Toxic" proteins as a component of different intracellular inclusions in neurodegenerative diseases

"Toxic" protein	Diseases
α-synuclein	Alzheimer's disease Lewy body disease multiple system atrophy Parkinson's disease
tau	amyotrophic lateral sclerosis Alzheimer's disease corticobasal degeneration Huntington's disease Lewy body disease Parkinson's disease Pick's disease and other fronto-temporal dementias progressive supranuclear palsy spinocerebellar ataxias
ubiquitin	Alzheimer's disease amyotrophic lateral sclerosis Huntington's disease Lewy body disease multiple system atrophy Parkinson's disease Pick's disease and other fronto-temporal dementias progressive supranuclear palsy (controversial data)
neuroserpin	familial encephalopathy with neuroserpin inclusion bodies
polyglutamine rich proteins (huntingtin, atrophin, ataxin, androgen receptor)	Huntington's disease spinocerebellar ataxias

It is noteworthy that some GI except ubiquitin and tau protein contain also heat shock proteins [13]. The presence of heat shock proteins in GI suggests that these proteinaceous intracellular deposits may be formed as a result of disturbances in cellular eliminating mechanisms using these proteins. One of such mechanisms is chaperone-mediated autophagy.

Conclusions

Various intracellular inclusions different in shape and size are described in many neurodegenerative diseases. None of them is pathognomonic for any of the diseases. The presence of the same type of inclusions in different neurological disorders and their similar immunoreactivity (Table 3) is probably due to disturbances in the same molecular mechanisms of protein elimination. Moreover, final structure of protein aggregates are similar to each other independently of primary amino acid sequence and protein conformational structure [42]. It indicates that inclusion formation may be an unspecific phenomenon not only dependent on molecular properties of the certain protein.

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