UNIVERSITY OF PADOVA

Department of General Psychology

Department of Neuroscience

Master's Degree in Cognitive Neuroscience and Clinical Neuropsychology

Final dissertation:

Multiple System Atrophy: A general overview of clinical, cognitive and behavioral manifestations.

Supervisor:

Prof. Antonini Angelo

Candidate: Lozjana Shametaj

Student ID number: 1230743

Academic Year 2021/2022

Table of contexts:

Abstract

In this review we will look at the general characteristics of Multiple System Atrophy, including pathophysiology, etiopathogenesis and the clinical presentation. Pathophysiology is represented by macroscopic brain alterations, glial and neuronal inclusions widely distributed in the brain and neurodegenerative changes. α-synuclein aggregations in oligodendrocytes, and moreover their spread in the neurons represents the key in explaining the pathogenesis. Even though the disease is considered sporadic, genetic and environmental factors will also be mentioned. Motor symptoms in the clinical presentation can help in differentiating between the clinical subtypes (cerebellar and parkinsonian) while many dimensions reflect the non-motor symptoms. Cognitive dysfunctions (eg visuospatial, attention, memory domains) are present and psychiatric symptoms such as depression and anxiety also exist in MSA. Finally, we will look at the symptomatic treatment and future advances of the disease.

1. Introduction

1.1 History of MSA

Multiple system atrophy (MSA) is an adult-onset, rapidly progressive neurodegenerative disease characterized by parkinsonian, cerebellar, dysautonomic and pyramidal features, in any combination. The predominant features may change during the disease course, and all of them can manifest as the disease progresses. [96]

The defining neuropathology of MSA consists of degeneration of striatonigral(eg striatum and substantia nigra) and olivopontocerebellar (eg inferior olives, pons, cerebellum) structures (I would put some examples of both of these structures in parentheses) accompanied by profuse numbers of distinctive glial cytoplasmic inclusions formed by fibrillized α-synuclein. The lesions are not only limited to these areas, but many other parts can be involved such as central, peripheral, and autonomic nervous systems, reflecting also the multisystem character of MSA. The etiology of the disease is unknown, and just as Parkinson disease (PD) and dementia with Lewy bodies

(DLB), it results from a disturbance of α-synuclein protein and is designated under as an α-synucleinopathy. [102]

In multiple system atrophy, this protein precipitates into the cytoplasmic oligodendroglial inclusions (GCIs) and neuronal inclusions (NCIs), being a hallmark of the disease. The involvement of these GCIs may hold the key to the mechanisms of cell loss, indicating why certain cells are susceptible to whatever causes the disease whilst adjacent cells and structures are spared. Together with the distribution of cell loss, the absence of Lewy bodies distinguishes it from idiopathic Parkinson's disease and the lack of neurofibrillary tangles separates it from Steele-Richardson-Olsewski disease and post-encephaltic Parkinsonism. [66]

In order for a better understanding of the disease, we need to start from the historical roots. The first clinical findings of MSA were recorded in 1900 at the Salpêtrière Hospital in Paris. Two middle-aged sporadic patients were presenting ataxia, dysarthria, akinesia, rigidity, incontinence of urine and brisk reflexes. They both died three years later. The post-mortem autopsy of one of the cases revealed olivo-ponto-cerebellar atrophy. The next demonstration of the disease was seen in 1925, with the symptoms of postural hypotension and anhydrosis (diminished sweating in response to appropriate stimuli). Furthermore, one patient had impotence and the other two showed hyperactive or asymmetric reflexes. The association between autonomic failure and neurologic dysfunction was noticed in one of the patients showing extensor plantar responses. [68]

In 1960 Shy and Drager described fours cases of a neurologic syndrome with orthostatic hypotension. "The full syndrome was described by orthostatic hypotension, urinary and rectal incontinence, loss of sweating, iris atrophy, external ocular palsies, rigidity, tremor, loss of associated movements, impotence, atonic bladder, loss of rectal sphincter tone, fasciculations, wasting of distal muscles, with the evidence of a neuropathic lesion in the electromyogram indicating involvement of the anterior horn." A syndrome designated striatonigral degeneration was firstly described by Adams et al in the early 1960s. The patients had mild autonomic failure and ataxia, and showed lesions in olivopontocerebellar system. (Rehman, H. U, 2001, p. 379)

Until the late, 1960s, olivopontocerebellar atrophy, Shy-drager syndrome and striatonigral degeneration were considered as different entities. It was only until 1969 when Graham

Oppenheimer introduced the term multiple system atrophy (MSA) that the three diseases previously mentioned were reunited into one entity. [68]

Later in 1989, Papp, Kahn and Lantos published their key neuropathological paper describing for the first time oligodendroglial cytoplasmic inclusions (GCIs), which was seen to be present in all cases of sporadic MSA, regardless of the clinical subtype. This finding gave also the pathological underpinning of Shy-Drager syndrome, striatonigral degeneration and olivopontocerebellar atrophy as one disease. [68]

Moreover, in 1998 it was revealed that Lewy bodies and Lewy neurites in Parkinson's disease stained positive with antibodies to α-synuclein. Later the same year, GCIs showed the same characteristic, introducing the umbrella term "α-synucleinopathy" This discovery determined the direction of research regarding MSA pathogenesis. [68]

Regarding the consensus criteria, the attempts to create the diagnostic aspect was influenced by the need of a proper clinical definition for MSA. In 1980 it was extremely difficult to distinguish between MSA, Parkinson disease and Progressive supranuclear palsy (PSP) due to overlapping characteristics. The Majo Clinic criteria defined the clinical diagnosis of MSA by the presence of autonomic failure with striatonigral or olivopontocerebellar involvement. However, further characteristics were needed in order to obtain a better and clearer diagnosis. For example criteria defining autonomic failure either of cardiovascular or urogenital type were needed in order to distinguish between milder forms of autonomic failure and age-related disturbances of the autonomic nervous system. L-dopa responsiveness should have been included in order to distinguish between PD with autonomic failure and MSA patients with isolated or predominant parkinsonian features. Moreover cognitive performance and family history of similar disease should have been included. [96]

Quinn was the first to propose widely accepted clinical criteria for MSA in 1989. The phenotypes of the disease were named striatonigral degeneration (SND type, related to the predominant parkinsonism) and olivopontocerebellar (OPCA type, related to the predominant cerebellar ataxia).Three levels of diagnosis were established; definite, probable and possible MSA. Warning signs (red flags) and exclusion criterion were also added. This was both a major progression but also an indication for future research. A low rate of false-positives was observed by the

clinicopathologic study in the Queen Square Brain Bank for Neurological Disorders while another study demonstrated a greater rate (up to 55%) of false-negatives. [67]

Quinn's data were further rationalized by the first Consensus Criteria for MSA in 1998. MSA-P and MSA-C (parkinsonian and cerebellar variant respectively) were the decided acronyms for the disease subtypes, avoiding possible misleading terms. In the list of exclusion criteria further features were added such as hallucination and family history of a similar disorder. However, despite the progress made in 1998, the amount of information in the three diagnostic levels seemed to be complex and with a lot of added information. [87]

The second consensus statement of MSA diagnosis presented in 2007 the same acronyms of the disease subtypes, but it was added that the predominant motor features can change over time. Moreover, it is not recommended anymore to use the term MSA-mixed due to possible confusion with ataxia and parkinsonism combination. The three diagnostic categories (definite, probable, possible) didn't change while new characteristics were added to the red flags and the non-supporting features. [102]

Definite MSA requires neuropathologic findings of widespread and abundant central nervous system α-synuclein-positive glial cytoplasmic inclusions, associated with neurodegenerative changes in striatonigral or olivopontocerebellar structures. [102]

Probable MSA is described with a disease onset of above 30 years old, characterized by autonomic failure involving urinary incontinence or orthostatic decrease of blood pressure within 3 min of standing by at least 30mm Hg systolic or 15 mmHg diastolic. Probable MSA criteria also include poorly levodopa-responsive parkinsonism (bradykinesia with rigidity, tremor, or postural instability) or a cerebellar syndrome (gait ataxia with cerebellar dysarthria, limb ataxia, or cerebellar oculomotor dysfunction) [102]

Possible MSA is related to a disease with an onset of above 30 years old, represented by parkinsonism (bradykinesia with rigidity, tremor, or postural instability) or cerebellar syndrome (gait ataxia with cerebellar dysarthria, limb ataxia, or cerebellar oculomotor dysfunction) and at least one feature suggesting autonomic dysfunction. One additional feature is necessary to conclude the criteria of possible MSA, and there are many possibilities. Babinski sign with hyperreflexia or stridor can be featured in both possible MSA-P and MSA-C. Furthermore we can

observe in possible MSA-P rapidly progressive parkinsonism, poor response to levodopa, postural instability within three years of motor onset, and atrophy on MRI of the putamen, middle cerebellar peduncle, pons, or cerebellum. Investigational disease markers have also included hypometabolism on FDG-PET in the putamen, brain stem, or cerebellum. Regarding possible MSA-C features, patients can present parkinsonism (bradykinesia and rigidity), atrophy on MRI of the putamen, middle cerebellar peduncle, or pons, hypometabolism on FDG-PET in the putamen, or presynaptic nigrostriatal dopaminergic denervation on SPECT or PET. [102] [98]

Red flags is the term used for the features supporting the disease diagnosis. The features consist of head-neck dystonia; disproportionate antecollis; bent spine (forward, lateral, or both); contractures of the hands or feet; inspiratory sighs; severe dysphonia (the voice sounding involuntarily breathy, raspy, or strained, or is softer in volume or lower pitch); severe dysarthria; new or increased snoring; cold hands and feet; emotional incontinence (pathological laughter or crying); jerky, irregular or postural action tremor. [19]

1.2 Epidemiologic features

Regarding the epidemiologic features of MSA, the estimated mean incidence is 0.6 to 0.7 cases per 100,000 person-years, with a range of 0.1 to 2.4 cases per 100,000 person-years. The estimated point prevalence is 3.4 to 4.9 cases per 100,000 population, increasing to 7.8 per 100,000 among persons older than 40 years of age. [19]In Italy, 4900 prevalent cases have been estimated, 81.5% of these were concentrated in the age band 60-79 years, while only 9.8% and 8.7% represents the age bands 50-59 years and >80 years, respectively. [90]In a study performing in London on computerized records of 15 general practitioners, the age-adjusted prevalence for MSA was 4.4 per 100.000 population. In the first epidemiological study who adopted the new clinical criteria the age adjusted prevalence observed in Gironde, France, was 1.86 per 100.000 population. [89]The incidence regarding sex has been approximately equal. Survival from first symptom (autonomic or motor) averaged 9 to 10 years in one large clinical series, but being shorter in pathologic series. [67]

A regional difference has been seen regarding the disease subtypes. MSA-P has demonstrated to have a predominance in Europe, USA and Korea, while MSA-C has been more frequent in Japan. [75] The study of Sakushima in 2015 was the first large community-based retrospective cohort

survey, which was focused on natural history of MSA. It was seen that MSA-C is more predominant in Japanese individuals (confirming also results from previous studies). Furthermore it was seen based on a Kaplan-Meier survival curve that cerebellar ataxia onset symptoms had a better prognosis than parkinsonian or autonomic failure ($p<0.01$). Additionally, the higher age of onset was associated with poor prognosis. [75] On the other hand the North American study has found that 60% of the MSA patients had MSA-P while only 13% were represented by MSA-C. [87]

MSA patients demonstrate a rapid disease progression at onset, and generally motor and non-motor symptoms worsen within a time frame of 10 years. About 50% of the patients require walking aid within three years of the initiation of motor symptoms while 50% after 5 years require a wheelchair. The time before the patients is bedridden is 6 to 8 years. Causes of death regarding MSA include urosepsis, bronkopneumonia, or sudden death. Older age at onset, a parkinsonian phenotype and early development of severe autonomic failure represent negative prognostic factors, while a cerebellar phenotype and later onset of autonomic failure predict slower disease progression. [19]

There are many reasons for choosing MSA. Firstly, it represents a unique disease due to the fact that in the spectrum of α-synucleinopathies it is the only one with α-synuclein aggregation occurring within oligodendroglial cells. Moreover, the diverse clinical presentation and the constantly changing symptoms lead to a major diagnostic challenge in many fields such as neurology, cardiology, gastroenterology, urology, otolaryngology and sleep medicine. [19] The symptoms of MSA make it harder to distinguish from PDs or idiopathic late-onset cerebellar ataxia until advanced stages of the disease. The lack of knowledge regarding the path towards neurodegeneration makes MSA an enigmatic case, which influences more the research and the need for discovery.

This review will give an overview of Multiple System Atrophy. The neuropathology of MSA will consist of a description related to brain alterations, the histopathology, glial and neuronal inclusions described by their microscopic characteristics and localization. Neurodegeneration will also be explained and the pathological variants of the disease will be also mentioned. The chapter

related to etiopathogenesis will present us with the pathogenesis, going deeper in the explanation regarding α-synuclein in oligodendrocytes followed by genetic and environmental factors. The clinical spectrum of MSA is represented by motor and non-motor features. Moreover the cognitive dysfunction in MSA-P and MSA-C and the differences will be explained. Finally we will see the symptomatic treatment and future perspectives of the disease.

2. Neuropathology of MSA

2.1 Macroscopic changes

Anatomical alterations are seen in both MSA-P and MSA-C brain, as a result of disease progression. The macroscopic changes can be shown in both olivopontocerebellar (OPC) and striatonigral(StrN) systems, where both or one system can be more affected than the other.[1]The macroscopic examination of MSA brain has revealed severe atrophy of the cerebellum, middle cerebellar peduncle and the pontine nuclei associated with mild diffuse cortical atrophy in the frontal lobes [10] The overall brain weight is within the normal range, and the neocortex and limbic structures usually remain intact. Alterations in these brain areas could indicate atypical MSA or MSA with other concomitant neurodegenerative conditions. [45]

Regarding the characteristic MRI brain signal intensity abnormalities, it is included the “hot cross bun” sign, a cruciform hypointensity in the pons that resembles the Easter pastry, and the “putaminal slit” sign, a hyperintense signal in the dorsolateral margin of the putamen, which have a high positive predictive value for the diagnosis of MSA. [61] Patients with early “hot cross bun” sign have shown to be more likely to develop cerebellar symptoms while patients with early bilateral putaminal slit signs mostly develop the parkinsonian variant of MSA. [61]

Coronal sections of the cerebrum of MSA-P brains, have shown atrophy and dark discoloration of the putamen. In extreme cases these changes extend to the caudate nucleus with the subthalamic nucleus remaining microscopically normal. The substantia nigra is pallor and is a more severe feature in SND (striatonigral degeneration) in contrast to the midbrain which does not show major atrophy. [1]

MSA-C variant has demonstrated varying degree of atrophy of the paleo-and neocerebellum, with narrowing of the folia and a discoloration of the cerebellar while matter. The outflow pathways of the cerebellum (superior cerebellar peduncle) and deep cerebellar nuclei are preserved. Moreover in OPCA cases have shown severe atrophy of the pontine base and middle cerebellar peduncle. The ribbon on the inferior olive is usually obscure. Moreover, the spinal cord does not typically show gross changes [1]

VBM studies (voxel-based morphometry) have also proved that basal ganglia and infratentorial volume loss in MSA patients with structural abnormalities in infratentorial brain regions are more expressed in MSA-C than in MSA-P patients. [5]

Patients with MSA-C demonstrate a high frequency and severity of MRI abnormalities (atrophy and hyperintense signal changes) of the middle cerebellar peduncle and pons compared to cerebellar ataxia and extracerebellar features. [5]

Differently from PSP, atrophy of the subthalamic nucleus, midbrain, cerebellar dentate nucleus and superior cerebellar peduncle are not seen in MSA. Lacking of focal atrophy in the superior frontal and motor cortices makes the difference from corticobasal degeneration. [45]

Comparing MSA with PD, it was seen that the average middle cerebellar peduncle was significantly smaller in MSA patients compared to PD and healthy subjects. Furthermore no overlap was seen comparing MSA patients with the other groups. The pons has also demonstrated to be significantly smaller in MSA-P patients compared with PD, PSP, and healthy controls with overlapping individual values. [5]

2.2 Histopathology

The histopathological aspect of MSA is represented by α-synuclein-immunoreactive cellular inclusions, selective neuronal loss and axonal degeneration, myelin pallor and gliosis, microglial activation and astrogliosis. [10]

α-synuclein-immunoreactive inclusions consist of five types: glial cytoplasmic inclusion (GCIs)when the protein is accumulated within oligodendrocytes, glial nuclear inclusions which are less frequent, neuronal cytoplasmic inclusions (NCI) , neuronal nuclear inclusions(NNI)and finally astroglial cytoplasmic inclusions and threads of similar composition.

The presence of cytoplasmic α-synuclein-immunoreactive GCIs represents the histological hallmark, which is necessary for the postmortem diagnosis of definite MSA. [36] The accumulation of α-synuclein aggregates as a pathological feature in MSA brains is part of synucleinopathies. [54]

The degree of neuronal loss and cellular inclusions in different brain areas corresponds with the MSA motor subtype and the predominance of SND or OPCA. Quantitative analyses of neuronal loss and GCIs revealed a positive correlation between both lesions, indicating that the accumulation of GCIs is likely to be an important factor in neuronal death in MSA. [96]

2.3 Glial and neuronal α-synuclein pathologies

Glial cytoplasmic inclusions are also known as Papp-Lantos inclusions, named after the scientist that discovered them. [1] All types of oligodendrocytes (satellite, interfascicular, perivascular) are vulnerable to GCIs. To localize the inclusions in oligodendroglia cells oligodendroglial markers are used (anhydrase isoenzyme II, Olig 2, Leu-7) [96]

Glial cytoplasmic inclusions are argyrophilic(easily impregnated with silver) and oligodendrolial in origin. These inclusions could be sickle shaped, oval, or conical. The nuclei of the oligodendroglia cell containing GCI has been demonstrated to be slightly larger and lighter compared to normal looking oligodendrocytes. [93]

“GCIs are immunoreactive for ubiquitin, normal adult tau, α- and ß-tubulin and p62. Furthermore double immunolabelling for both α-synuclein and ubiquitin has showed more abundant and extensive staining for α-synuclein. (Wenning, K.G , 2016, p. 32)

GCIs are non-membrane-bound cytoplasmic inclusions composed of filaments and tubules (20-40 nm in diameter) and granular material. These filaments could be twisted or straight: twisted filaments have an alternating width between 5 nm and 18 nm and a periodicity of 70–90 nm; straight filaments have a uniform width of approximately 10 nm. [1] These filaments are different from filamentous oligodendrolial inclusions called coiled bodies found in other diseases such as PSP, corticobasal degeneration and Braak’s argyrophilic disease. [18]

The main component of GCIs is Ser129-phosphorylated α-synuclein fibrils. However other components are detected such as: α- and ß-tubulin 14–3-3 protein and fragments of cellular

organelles (mitochondria, secretory vesicles). This indicates that they are highly complex proteinaceous aggregates. These are components which are found also in Lewy Bodies, however their proportion and ultrastructural feature are different. [10] In the study of D.L Pountney et al in 2005, it was investigated the abundance and effects of αB-crystallin in the formation of GCI. This chaperone protein was discovered to be an important component of GCI, more expressed in Lewy bodies than in Lewy body dementia, thanks to immunohistochemistry and confocal microscopy. [64]

Gallyas-Braak impregnation method and α-synuclein immunohistochemistry have shown to be the most sensitive techniques for revealing the presence of GCIs. [93] On the other hand, other techniques can be used to identify GCIs such as ubiquitin and p62 but they lack specificity of α -synuclein. [1]

GCIs are widely distributed in white and grey matter in the brain and they have been shown to increase with time. [96] GCIs might influence more on the OPC pathology than in the StrN. [60]

The density of GCIs is demonstrated lower in white matter structures with severe myelin pallor and higher in those with mild to moderate myelin loss. [1]

"The highest densities of GCIs in the grey mater (>300/mm 2) are found in the deeper laminae of the primary motor and premotor cortex; dorsolateral areas of the putamen; globus pallidus; subthalamus; substantia nigra pars compacta; pretectal area; pontine base nuclei; vestibular nuclei; motor nuclei of V, VII and XII cranial nerves; pontomedullary reticular centres and intermediolateral column of the spinal cord. In the white mater, GCIs are most numerous beneath the motor cortex, the internal and external capsule, corpus callosum, corticospinal tracts, middle cerebellar peduncle and the cerebellar hemispheric white matter" (Wenning, K.G , 2016, p.26)

In one study by Papp, M.I in 1994, semiquantitative mapping of GCIs and sensitive silver techniques were used in 14 MSA brains and 11 spinal cords of patients with various combinations of the clinical subtypes. The results showed degeneration in all oligodendroglial cell types. Additionally, areas where GCIs-rich structures occurred mostly were the supra-segmental motor systems and supraspinal autonomic systems, and in their target connections. The motor system

includes primary motor and higher motor areas of the cerebral cortex, pyramidal and extrapyramidal systems and cortico-cerebellar connections. [62]

In the study of D.W Dickson et al in 1999, immunostained sections of 7 cases were used (3 men and 4 women; 3 OPCA and 4 SND; average age 62.1- 9.4 years) to identify the neuroanatomical distribution of the density of GCIs. Furthermore immunoblot analysis was used. It was demonstrated that OPCA cases had fewer inclusions than SND cases especially in the brain stem region. The greatest density in both MSA subtypes was demonstrated to be in the basal ganglia (putamen and globus pallidus). [18]

Armstrong and colleagues in 2006 studied the topographical distribution of the pathology in MSA. Glial cytoplasmic inclusions were shown to be randomly distributed or in large clusters in the brain. Furthermore this distribution was different compared to other α-synucleinopathies. This distribution was a demonstration that GCIs are the primary pathological process in the brain. On the other hand neuronal pathology demonstrated to be more selective and affected smaller topographical brain areas. [3]

The study done by Tu P-H et al in 1998, using immunoelectron microscopy, showed that in the white mater α-synuclein-positive GCIs were restricted to oligodendrocytes. Insoluble α-synuclein was accumulated selectively in the white mater with α-synuclein-positive GCIs. Stating from the results of the study, reduction in the solubility of α-synuclein might reflect an important factor to why we have this protein accumulation. [85]

In some regions such as the motor cortex, there can be nothing histologically unremarkable but still a high density of GCIs. This leads to the fact that neurodegeneration is not a prerequisite for developing GCIs. [1]

A positive correlation between neuronal loss and the density of GCIs was seen in StrN and OPC regions indicating a link between neurodegeneration and GCIs in these regions. The substantia nigra was seen to be a region with severe neuronal loss, however it has also demonstrated a relatively low density of GCIs. This might demonstrate that other factors might interfere in neuronal loss or perhaps that region was affected earlier in the disease and has been burnt out. [1]

Glial intranuclear inclusions are also present in MSA brains, however they have a low frequency and due to that, they have a rod-like morphology and are difficult to observe. They are not found

in every MSA case. Brain areas usually showing GNIs are: pontine nuclei, putamen, subthalamic nucleus, arcuate nucleus, subiculum, amygdala, hippocampus, dentate fascia, substantia nigra, inferior olivary nucleus and brainstem reticular formation. [96]

As previously mentioned, neuronal inclusions are found in the MSA brain (in the cytoplasm, nuclei and axons). Cytoplasmic argyrophilic inclusions in neurons were firstly found in the pontine and arcuate nuclei of OPCA patients by Kato and Nakamura. [51]

NCI and NNI share the same features regarding the ultrastructural aspect. [93]

Fibrillary structure resembling microtubules represented the inclusions with a diameter of 24 nm to 40 nm. (Lantos, P. 1994) Osmiophilic' granules (reacting to or staining with osmium tetroxide) covered the outer surface and they were intermingled with a few filaments of 10 nm in diameter. [51]

Homogenous α-synuclein staining in neuronal nuclei and the cytoplasm has been observed in early stages of the disease in addition to GCIs in different parts of the CNS. [36]

The shape of NCI is different in the pontine nucleus, inferior olivary nucleus and dentate nucleus. Pontine nucleus inclusions are represented by round or ovoid shapes, while the inferior olivary nucleus has demonstrated a reniform (resmbing a kidney), crescent-shaped or coarse granular shape. Furthermore, neuronal cytoplasmic inclusions in dentate granule cells have a ring-like or C-shaped inclusions. [93]

NCIs have demonstrated no difference in the frequency regarding: putamen, pontine nucleus and inferior olivary nucleus between SND and OPCA subtypes. This shows that the pathological abnormality is not related to the disease subtypes. [60]

Compared to GCIs, neuronal inclusions are thought to be lower in number and this has led to a less defined topography and frequency. Neuronal inclusions seem to have a similar distribution in the cortical, subcortical, brainstem, cerebellar nuclei. The areas with more frequency are the basis pontis (ventral portion of the pons) and inferior olives. The density of glial cytoplasmic inclusions has shown to be unrelated to neuronal nuclear inclusions. [96]

Other areas with NCIs are putamen, substantia nigra, motor cortex and dentate gyrus. [93]

In the study of D.Cykowski et al in 2015, lateral cuneate showed more intranuclear inclusions. Combined neuronal intranuclear inclusion/neuronal cytoplasmic inclusion were shown in pontine nuclei. Moreover, perinuclear neuronal cytoplasmic inclusions were mostly in pyramidal cells of the isocortical laminae III and v. [17]

A correlation matrix of pathologic severity was calculated between distinct anatomic regions of involvement (striatum, substantia nigra, olivary and pontine nuclei, hippocampus, forebrain and thalamus, anterior cingulate and neocortex, white mater of cerebrum, corpus callosum) and areas not related to the disease such as anterior cingulate cortex, amygdala, entorhinal cortex, basal forebrain and hypothalamus and cerebellar roof nuclei. [17]

Based on the interregional correlations in the pathologic glial and neuronal lesion burden, it revealed a very important finding (rho ≥ 0.6) that a shared mechanism of the disease is suggested between discrete anatomical regions (for example the substantia nigra and frontal white mater) and cell types (neuronal and glial inclusions in frontal cortex and white matter respectively). Based on the data, it was suggested that just like glial inclusions, neuronal pathology is also important in the evolvement of the disease. [17]

NNI compared to NCI have shown to be in higher in number in the pontine nuclei in some MSA cases. This suggested that the formation of NCIs is related to the evolvement of the disease and that the formation of NNIs might be prior to NCIs. [96]

2.4 Distribution of neurodegenerative lesions

Every neurodegenerative disease has the hallmark of selective neuronal loss, followed by reactive changes in astrocytes and microglial. [96]

In the study of Ozawa.T et al (2004), even though the number of GCIs gradually increases with time, neuronal loss occurs sooner and takes place in a rapid manner in the striatonigral region. Moreover they stated that GCIs contribute less to the pathogenesis of neurodegeneration in the StrN than they do in the OPC region, as mentioned previously [60]

In MSA-P, the most affected areas are the dorsolateral caudal putamen and caudate nucleus. Furthermore the dorsolateral region of the substantia nigra (SN) pars compacta (SNpc), globus pallidus and the subthalamic nucleus are also involved. [10]

Additionally, there is a selective loss of medium-sized spiny GABAergic neurons positive for calcineurin and preservation of choline aceryltransferase (CHAT)-positive neurons. [96]

In the study of Sato.K et al (2007), immunohistochemical studies were performed using antibodies to calbindin (CALB) and calcineurin (CaN) as neurochemical markers for striatal medium spiny neurons. It was found that in the caudal and dorsolateral putamen, the medium spiny neurons positive for CALB were diminished, on the other hand CaN-positive neurons were relatively spared in a mosaic pattern**.** An area less affected in MSA is the dorsolateral caudate nucleus. A compartmentalized distribution that corresponded with the striosomal arrangement visualized by Metenkephalin immunostaining was shown by residual CALB-positive neurons. The findings led to the conclusion that regarding the neurodegeneration in MSA-P, there is a compartmental difference in striatal medium spiny neurons. [77]

In the study of Refolo.V et al (2018), progressive striatonigral degeneration was investigated in a transgenic PLP-α-synuclein mouse model of MSA. Motor deficits were shown emerging 6 months and deteriorating up to 18 months of follow up. The motor phenotype corresponded with dopaminergic cell loss, striatal dopaminergic terminal loss and DARPP32-positive medium sized projection neurons. The beginning of the degeneration corresponded with the increase of soluble oligomeric α-synuclein. Furthermore, striatonigral degeneration was linked to abnormal neuroinflammatory responses [70]

“Both striatal outflow pathways are affected according to different immunohistochemical and autoradiographical studies. Encephalin-containing striatal neurons projecting to external globus pallidus which carry dopamine D2 receptors (indirect pathway) and substance P-containing cells projecting to internal globus pallidus and substantia nigra pars reticulata that carry D1 receptors (direct pathway). This leads to striatal degeneration, and differentiation of the posterolateral portions of the external and internal globus pallidus and the ventrolateral substantia nigra pars reticulata.” (Geser, F , 2005 , p. 325)

On a clinical aspect, the SND phenotype demonstrates the most severe bradykinesia confirming that the clinical phenotype is dependent on the distribution of the pathology within the basal ganglia. Furthermore putaminal involvement correlates with poor levodopa response. [60]

“In MSA-C, neuronal loss correlates with Purkinje cell layers of cerebellum, vermis, cerebellar dentate nucleus, basis pontis and the inferior olivary nucleus and less severely with the substantia nigra and locus coeruleus, and with minimal damage to the striatum.” (Wenning, K.G , 2016, p 22)

A “dying back” process is the term for the disproportionate depletion of transverse pontocerebellar fibers from the middle cerebellar peduncles compared to the loss of pontine neurons. (Geser, F , 2005 , p. 325)

In the study of Wenning G. (1996) a morphometric analysis of 20 MSA cases and 8 controls was performed. It was discovered that in MSA cases mean neuronal cell densities were significantly reduced in (medial and dorsal) accessory and principal inferior olives, pontine nuclei, cerebellar vermis and hemispheres. In most cases inferior olives and pontine nuclei were more severely affected than cerebellar Purkinje cells. Purkinje cells demonstrated more degeneration in the vermis rather than in the hemispheres. Furthermore a poor topographic correlation between neuronal cell loss in the inferior olives and cerebellar cortex was seen. A primary degeneration of olivopontine nuclei and cerebellar Purkinje cells in OPCA was suggested based on the results. Comparing OPCA cases with SND subtypes, brain areas such as the inferior olives, pontine nuclei and cerebellar cortex were severely affected in cases with cerebellar subtype. [101]

“Furthermore, Purkinje cells have shown immunoreactivity for GDNF (glial cell line derived neurotrophic factor) while abundant DGNF -positive dendrites were found in some areas of the molecular layer. The data has suggested that GDNF is produced and found in Purkinje cells even in MSA patients and the functional impairment of Purkinje cells in MSA can cause a focal accumulation of GDNF in the dendrites of surviving Purkinje cells.” (Wenning, K.G , 2016 , p.23)

Cerebellar signs are more frequent in the OPCA phenotype, reflecting the distribution of the pathology correlating to the clinical aspect of the patient. A more advanced cerebellar pathology is related to ataxia symptoms. [60]

The dorsal nucleus of vagus (dmX), intermediolateral column of spinal cord, and Onufrowicz nucleus are involved in both MSA subtypes. [36]

In the study of Yoshida M. (2007) the pathological features of 102 MSA cases were evaluated. Regarding the motor neuron system in MSA, quantitative studies of the pyramidal tract showed a significant decrease in the number of small myelinated fibers. Neuronal loss of lower motor neurons has also been reported. Degeneration of the ambigui nuclei is related to laryngeal abductor paralysis and laryngeal muscle atrophy. Indeed, the third lumbar segment in MSA showed a decreased total cell count. Morphometrical analysis of ventral horn cells of the fourth lumbar segment in MSA showed a marked decrease of small neurons in the intermediate zone, and the decrease of large-and medium-sized neurons in the medial and lateral nuclei was relatively mild. Furthermore it was seen a distribution of α-synuclein -positive GCIs in the pyramidal tract, while NCIs and NNIs were seen in large motor neurons of the spinal cord and the hypoglossal nuclei. The involvement of motor neurons is reflected by these findings. Neuronal loss and myelinated fibers in MSA differ from ALS (amyotrophic lateral sclerosis) and control subjects. [105]

Betz cell loss and astrocytosis (increase in the number of astrocytes) in the cerebral cortex has demonstrated to be present in many MSA cases.

Microglial proliferation activity and nonheme iron (Fe31) content in the SNc and GP are more prominent than in DLB (dementia with lewy body) while mesopontine cholinergic neuron involvement(lateral tegmental nuclei) is more severe than in DLB. [36]

Damage to the autonomic system in MSA represents a multidomain autonomic failure. There are lesions regarding the supraspinal site. The brain areas included are: " cholinergic neurons in dorsal vagal nucleus and ventrolateral nucleus ambiguous catecholaminergic neurons of ventrolateral medulla, neurokinin-1 receptor–like immunoreactive neurons in ventrolateral medulla, serotoninergic neurons of the medulla, Edinger-Westphal nucleus and posterior hypothalamus (including the tuberomammillary nucleus), and brainstem pontomedullary reticular formation." (Geser, F , 2005 , p. 327)

In MSA, adrenergic neurons are more susceptible than serotonergic neurons in the medulla Furthermore, the loss of monoaminergic neurons may progress independently from α-synuclein accumulation in MSA. [15]

“Lesions also affect: sympathetic preganglionic neurons in the intermediolateral column of thoracolumbar spinal cord and postganglionic sudomotor denervation is seen in MSA. Mild degeneration of cardiac postganglionic sympathetic fibers can also be possible in MSA, a phenomenon related to α-synuclein pathology in sympathetic ganglia. However this phenomenon has demonstrated to be less severe than in Parkinson disease.” (Jellinger, K. A. 2014 , p. 1726)

Filamentous α-synuclein represent the involvement of the peripheral nervous system in MSA. These aggregates are located in the neurons of sympathetic ganglia and in the cytoplasm of Schwann cells. The reduction of unmyelinated fibers (sensory afferent and postganglionic sympathetic fibers) is demonstrated by sural nerve biopsies. [36]

Regarding white matter, reduced myelin staining in white matter tracts is seen in both striatonigral and olivopontocerebellar regions such as: the external capsule, striatonigral fibers, transverse pontine fibers, middle cerebellar peduncle and cerebellar hemispheric white mater. [96]

In the study of by Nykjær.C et al (2017), widespread microgliosis was seen in MSA brains, without concomitant astrogliosis. Furthermore a presence of significant oligodendroglial degeneration was not demonstrated which leads to the need of future research. [59]

In the study of Wakabayashi.K et al (1998), a postmortem examination revealed major myelin and axonal loss in the frontal and parietal white matter. Ubiquitin-positive oligodendroglial inclusions were found in these cortical and white matter lesions. Motor and cerebral white matter sites demonstrated significant involvement in the MSA disease process and oligodendroglial inclusions contribute to white matter degeneration. [94]

2.5 Pathological variants in MSA

Rare MSA types are represented by the terms: minimal change, preclinical, and incidental. They can be crucial in having a better understanding of the pathogenesis of the disease.

Minimal change is the term that indicates neuronal loss and GCIs restricted to either striatonigral or olivopontocerebellar region. Striatonigral degeneration minimal change is represented by neuronal loss restricted to the substantia nigra and locus coeruleus. In the study of Ling et al (2015) NCIs were seen in the caudate nucleus and substantia nigra (P=0.002) and nucleus raphe obsurus (P=0.04). The findings indicated that a-synuclein pathology can lead to neuronal dysfunction which can cause clinical symptoms before overt neuronal loss in MSA. [53]

On the other hand, minimal change regarding OPCA was reported in the study of Wakabayashi and colleagues (2005). Neuronal loss was restricted to the olivopontocerebellar system, being more severe in the pontine nucleus. Mild neuronal loss was found in: the anterior vermis and inferior olivary nucleus. Glial cytoplasmic inclusions were more in number in the pontine base and cerebellar white matter. NCI were located in the pontine and inferior olivary nuclei. The number of neuronal intranuclear inclusions was much higher than the NCIs. Additionally, α-synuclein accumulation was more severe in the neurites than in the cytoplasm or nucleus. [95]

Preclinical cases of MSA have also been described. A MSA-C preclinical case was described by Kon and colleagues (2013), representing the first of its kind to be reported. A 71 year old Japanese patient without clinical signs of MSA revealed slight gliosis in the pontine base and widespread occurrence of glial cytoplasmic inclusions in the CNS. The greatest abundance was discovered in the pontine base and cerebellar white matter. NCIs and NNIs were mostly located in the pontine and inferior olivary nuclei. [47]

Two other preclinical case were discovered by Fujishiro et al (2008) regarding a 96 year old woman and an 82 year old man. Both cases had widespread of GCIs and NCIs in the central nervous system but no neuronal loss or gliosis in vulnerable brain regions including: the substantia nigra, putamen, inferior olive and pontine base. The pathology demonstrated in both cases was below the one detected with clinically overt patients. However further examinations are needed in order to understand if the presence of GCIs in elderly patients is a neurologically age related α-synucleinopathy or a rare prodromal MSA cases. [22]

A rare MSA case was seen in the study of Aoki and colleagues (2015) where four patients were studied with atypical MSA represented by clinical features of frontotemporal dementia, including two corticobasal syndromes (one with progressive non-fluent aphasia and one with the behavioral

variant of FTD). None of them showed autonomic dysfunction. Frontotemporal atrophy and severe limbic α-synuclein neuronal pathology was demonstrated in all patients. Neuronal inclusions were heterogeneous and included Pick body (inclusion body in a neuron consisting of a disorderly collection of tau protein filaments)similar inclusions. Atypical MSA had significantly more neuronal inclusions in the anteromedial temporal lobe and limbic structures compared to typical MSA. MSA may present clinically and pathologically as a frontotemporal lobar degeneration (FTLD). This might even represent a novel subtype of FTLD associated with α-synuclein. [2]

Cases of Multiple system atrophy have shown that this disease can be presented with conjunctional pathologies.

Tau-positive granules have been detected in α-synuclein filaments in different studies. In the study of Nagaishi M. and colleagues (2010) the tau-positive granules were detected in GCIs in the neurodegenerative region, more specifically in the putamen and internal capsule. Tau accumulation was of three or four-repeat, which was recognized thanks to specific antibodies. [57]

Another study by Homma T. et al (2020) demonstrated the presence of tau positive granular glia in the frontal and temporal white matter in MSA, which was associated with longer disease duration. The tau isoform of AT8-positive granular glia in the cerebral white matter showed three-repeat. Also in the putamen just like in the previous study the presence of tau with GCIs was seen with the isoform of three-repeat tau and four-repeat tau. [31]

Moreover, Piao and colleagues (2001) showed by a double-labeling immunofluorescence study that the NCIs in the dentate gyrus and amygdaloid nucleus, and the GCIs in the frontal and temporal white matter expressed both α-synuclein NACP-5 and phosphorylated tau AT8 epitopes. Double-immunolabeling electron microscopy of the NCIs in the dentate gyrus and the GCIs in the temporal white matter demonstrated labeling of their constituent granule associated filaments with NACP-5 and some of them were labeled with AT8. The mechanisms of abnormal tau and accumulation in NCIs and GCIS are still unknown. [63]

Concomitant Alzheimer's disease (AD) in MSA has shown to be a rare phenomenon For example this occurrence was mentioned in two cases in the study of Terni and colleagues in 2006 with α-synuclein inclusions present in glial and neuronal cytoplasm in the brainstem, amygdala and hippocampal formation. [84]

Only a few reports have mentioned the presence of Lewy bodies in MSA. A subset of tau 14-3-3 protein has been detected in GCIs, in addition with other components with are present in Lewy body. These components represent the hallmark of PD and dementia with Lewy body. [35]

In the study of Koga and colleagues (2020) LBD (Lewy body disease) was observed in 11 MSA patients: seven were brainstem type, three were transitional type, and one was diffuse. The last 4 had an intermediate or high likelihood of DLB. Two patients had neuronal loss in the substantia nigra, but not in striatal or olivocerebellar systems with widespread glial cytoplasmic inclusions (consistent with minimal change MSA). In these cases LBD was considered the primary pathology while MSA was the coincidental. APOEε4 allele frequency was not different between MSA+LBD and MSA without LBD. [46]

3. Etiopathogenesis

3.1 Pathophysiology

α-synuclein is a 140 amino-acid protein that was discovered in synaptic vesicles in the presynaptic nerve terminals of neurons and has demonstrated to interact with membranes in vitro and in vivo. It is highly abundant in the brain and also present in other tissues, including red blood cells. [54]

The presence of α-synuclein deposition in GCIs is investigated in different studies. An important factor leading to this accumulation is P25α (tubulin polymerization-promoting protein; TPPP) which has an important role in stabilizing microtubules, projections of mature oligodendrocytes and ciliary structures. Moreover, it is thought to be important in the differentiation of oligodendrocytes as it is expressed in mature myelinating oligodendrocytes but is absent or expressed at low levels in the precursor stages [1]

TPPP/p25α colocalizes with a myelin basic protein (MBP), but this colocalization is lost in MSA. [40] “P25α relocates within oligodendroglia compartments away from the myelin sheath towards the soma, where it promotes enlargement.” (Wenning, G. K ,2008, p. 242)

With the redistribution of p25α, a significant reduction in the total level of MBP is seen, with an increase of its degradation products. These alterations indicate a chain of events where the normal cellular function of p25α in myelin is impeded and the stability of MBP is decreased by early

pathogenic signals. As a consequence, the translocation of p25α in oligodendroglia cell bodies may lead to the deposition and fibrillation of α-synuclein. [100]

In the paper of Kaji and colleagues in 2020, the autophagic degradation of α-synuclein was mentioned. The overexpression of p25α in differentiated PC12 cells (cell line derived from a pheochromocytoma of the rat adrenal medulla) interferes with the autophagic degradation of α-synuclein by preventing the fusion of autophagosomes with lysosomes and enhances the secretion of α-synuclein into the medium. Moreover, pharmacologic and genetic inhibition of autophagy causes significant accumulation of endogenous and exogenously applied α-synuclein in oligodendroglia cells. Regarding the general autophagy defect in MSA brains, it is considered a prodromal condition predisposing the individual to the emergence of GCIs. [40]

The relevance of iron in MSA is also an important element to be mentioned. "The oligodendrocytes contain high amounts of iron, which together with GCI pathology make a contribution towards MSA pathogenesis. Iron converts native α-synuclein into a β-sheet conformation and influences its aggregation either directly or via increasing levels of oxidative stress. Interestingly α-synuclein possesses ferrieductase activity and α-synuclein expression underlies iron mediated translational control via RNA stem loop structures. Despite a correlation between progressive putaminal atrophy and iron accumulation as well as clinical decline, it remains unclear whether pathologic iron accumulation in MSA is a secondary event in the cascade of neuronal degeneration rather than a primary cause." (Kaindlstorfer, C , 2018 , p.1253)

Oligodendroglia cells have not expressed α-synuclein mRNA in controls and MSA cases, indicating the ectopic origin of these cells. [80]

α-synuclein in oligodendrocytes has undergone post-translational modifications (oxidative modifications, nitration, and phosphorylation at serine 129) which lead to the contribution of oxidative stress in MSA in alpha-synuclein fibril formation in vitro. [96]

When compared to PD and DLB, MSA shows higher levels of α-synuclein in the detergent-soluble fraction of brain samples from the pons and white mater compared to controls, but detergent insoluble α-synuclein was not seen. [9]

Most of the soluble α-synuclein is seen in areas with few GCIs, indicating an altered solubility preceding the formation of GCIs. An increase in soluble monomeric alpha-synuclein can be

transformed into abnormally modified, insoluble, filamentous alpha-synuclein aggregation, leading to neurodegeneration. [38]

It is suggested that in MSA pathology the spread of α-synuclein through the CNS (central nervous system) is spatiotemporal, possibly by prion-like propagation of α-synuclein aggregates between synaptically connected areas. The study of Bernis and colleagues in 2015 investigated this topic and proved the prion-like propagation of α-synuclein. "Mice injected with brain extracts from patients with MSA and probable LBD contained hyperphosphorylated alpha-synuclein that also seeded aggregation of recombinant human wild-type alpha-synuclein in a Thioflavin T binding assay." (Bernis, M. E , p. 1) The intercellular transmission of α-synuclein might be possible through mechanisms such as: endocytosis, direct penetration, micropinocytosis, pore formation, nanotube, tunneling or diffusion. [92]

It was suggested that GCIs-like pathology has a main role in the pathogenesis of the disease inducing neurodegeneration associated with secondary aggregation of α-synuclein in axons or mitochondrial dysfunction based on transgenic models [96] The idea of a primary oligodendrogliopathy is related to the fact that myelin degeneration is widespread in MSA and p25a co-localizes with α-synuclein in GCIs. Axonal dysfunction and neurodegeneration in MSA comes from affected oligodendrocytes either through direct physical contact or through humoral and cellular effects leading to an inflammatory response resulting in death. This is a case that needs further investigation. [37]

On the other hand, we know that α-synuclein aggregates are also present in the neuronal cytoplasm, nuclei and neurites. Based on the fact of α-synuclein immunoreactivity of NNIs (not in NCIs) it has given the idea that abnormal α-synuclein aggregates in neuronal nuclei can interfere with neuronal nuclear function. [96]

There is evidence of neuron to oligodendroglia transfer of α-synuclein. Oligodendroglia cells have demonstrated to take up recombinant α-synuclein monomers, oligomers, and to a lesser extent, fibrils in vitro in a concentration and time-dependent manner. [72]

Neuron to neuron transmission of α-synuclein is also possible via endocytosis, leading to nuclear fraction and caspase 3 activation based on a PD study. [37]

Based on the presence of α-synuclein in oligodendroglial and neuronal inclusions in MSA, two parallel processes of the disease are suggested: one process is related to oligodendroglia-myelin axis degeneration (as in a primary oligodendrogliopathy), and the other process is α-synuclein accumulation in the nerve cells (as a neuronal synucleinopathy). [37]

However, not only oligodendrocytes and neurons are mentioned as cells responsible for α-synuclein propagation. Astrocytes have demonstrated to produce α -synuclein in advanced stages of the disease. Astrocytes express either wild-type or mutant (A53T or A30P) human α-synuclein, which are propagated and accumulated α-synuclein prions. Furthermore filamentous and granular forms of the accumulated protein in the astrocytes were discovered. [48]

Based on different evidence regarding the possibility of propagation, it was shown that α-synuclein aggregates transmission is represented by neuron to neuron transmission, neuron to astroglial and oligodendroglial cells. Furthermore there is the α-synuclein aggregates transmission from oligodendroglial to astroglial cells, influencing neuronal dysfunction, apoptosis and neuroinflammation. [88]

The transmission of α-synuclein from the neurons to the extracellular environment was described by the process of exocytosis, while endocytosis is the process regarding α-synuclein uptake from neurons, oligodendrocytes and astrocytes. Further speculation is needed regarding how this protein accumulation in neurons gets transformed in the extracellular environment. [88]

3.1.1 Oxidative stress and mitochondrial dysfunction

Other mechanisms related to α-synuclein aggregation are oxidative stress (OS) and mitochondrial dysfunction.

“Oxidative stress is associated with increased production of reactive oxygen species (ROS), such as oxygen (O2)-derived free radicals, the hydroxyl radical (OH) and nonradical derivatives of O2 such as hydrogen peroxide (H2O2), or a significant decrease in the activity of antioxidant defenses. Increased production or inadequate clearance of ROS can result in high levels of ROS, which can

then damage many components of the cell, including proteins, lipids and DNA." (Ubhi, K, 2011 , p. 584)

Reactive oxygen species derived from mitochondria dysfunction in the presence of toxins, and it can contribute to the pathogenesis of MSA. Oxidative damage of α-synuclein has been detected in GCIs. [81]

Moreover, mitochondrial toxin 3-nitropropionic acid (3NP) administration has altered levels of nitrated and oxidized α-synuclein in the MBP transgenic (tg) mouse model, influencing the global levels of phosphorylated or total α-synuclein. 3NP administration had also exaggerated neurological deficits in the MBP human α-synuclein tg mice, resulting in widespread degeneration and behavioral impairment. [86]

A possible genetic risk basis for the association of oxidative stress with MSA pathogenesis has also been mentioned. The candidate genes involved in oxidative stress are: CCAAT or enhancer-binding protein b; sequestosome 1, SQSTM1; cysteinyl-tRNA synthetase; solute carrier family 1A4, SLC1A4; and eukaryotic translation initiation factor 4E-binding protein 1, EIF4EBP1.[87]

3.1.2 Neuroinflammation

Neuroinflammation is defined as the dynamic response that involves changes in glial cell morphology, number, function and concomitant production of signaling molecules. Microglial and astrocytes are responsible for persistent intra-and extracellular imbalances in neurodegenerative diseases. In MSA neuroinflammation has correlated with the density of inclusions, disease duration, and inflammatory markers. [92]

Since α-synuclein can be released from degenerating neurons, it contributes to abnormal protein accumulation leading to neuroinflammation. It is predicted that neuroinflammation can also contribute to α-synuclein aggregation due to the release of cytokines and the shift to a pro-inflammatory environment. [92]

In MSA astrocytes have demonstrated high reactivity, and astrogliosis can lead to oxidative stress and neurotoxicity (similar to PD/DLB). [20]

Pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF-a) and interleukin-1beta (IL-1b), are released by microglia cells under the influence of α-synuclein. This dependency from

α-synuclein fibrils has been shown also by the release of pro-inflammatory cytokines by microglial BV2. [29]

In the study of Hoffman and colleagues in 2018, neuroinflammation was demonstrated in the white mater which coincided with the high number of α-synuclein aggregations. At the pre-symptomatic stage, increase in neuroinflammation has been detected in regions with a significant α-synuclein protein aggregation number, such as the corpus callosum and the striatum. Furthermore, the inflammatory response was restricted to myeloid cells (being highly proliferative and showing an activated, phagocytic phenotype). On the other hand grey matter with fewer α-synucleinopathy remained unaffected. Additionally, severe astrogliosis was seen only in the grey matter. An early crosstalk between neuroinflammation and oligodendrocytes containing α-synuclein inclusions leading to an immune response locally restricted to white matter regions in MSA was suggested based on the data. [28]

3.1.3 Working hypothesis for the pathogenesis of MSA

Based on the data obtained by many studies, a hypothesis regarding the pathogenesis of the disease was established. Important elements discovered such as overexpression of α-synuclein, toxic stressors and microglial derived neuroinflammation are important factors for the pathogenesis of the disease.

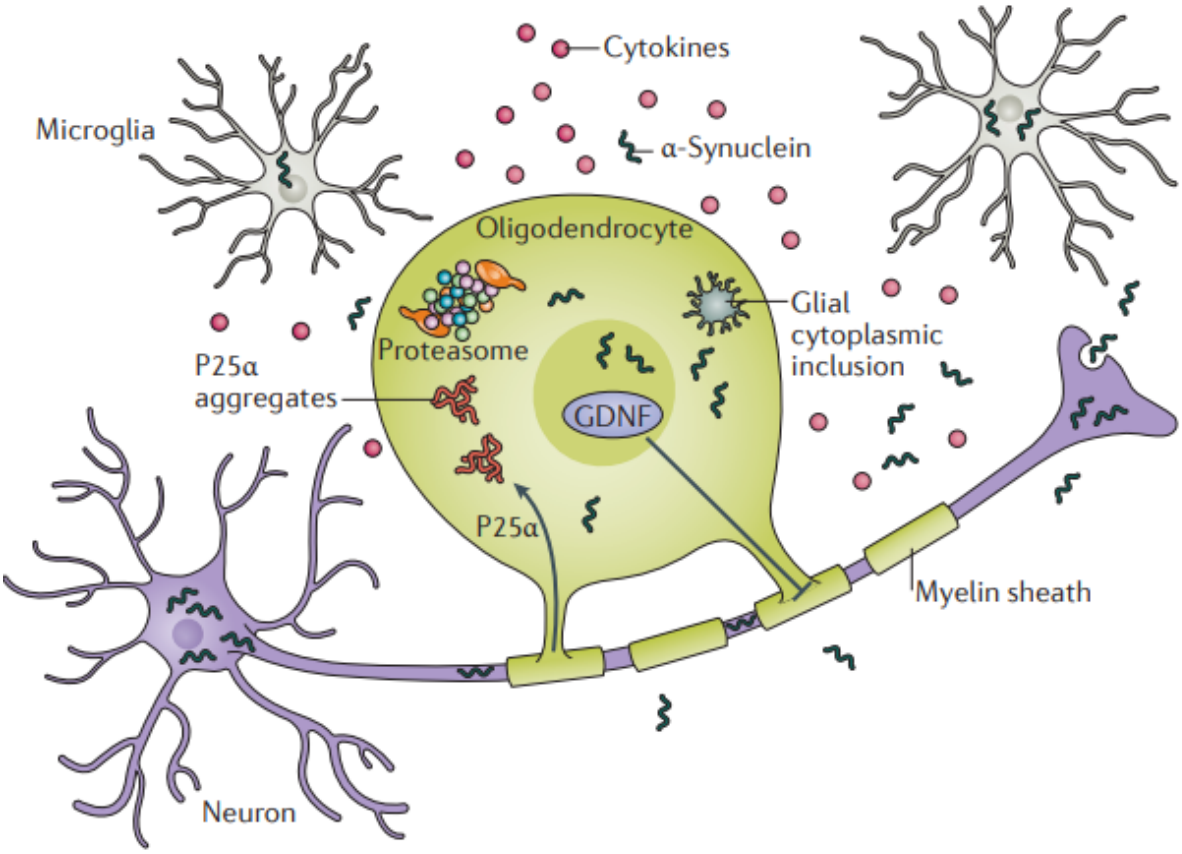

Figure 1-The hypothesis for MSA pathogenesis [49]

What researchers have been trying to understand is the pathway from α-synuclein aggregation, which is described to initiate in the oligodendroglial cells, leading to the death of other neurons. Oligodendrocytes are known for myelinating axons, and expressing neurotrophic factors (including glial-derived neurotrophic factor, GDNF; brain-derived neurotrophic factor, BDNF; and insulin-like growth factor 1, IGF-1 which are responsible for the maintenance and survival of neuronal populations). [87]

The accumulation of α-synuclein in oligodendrocytes, resulting in neuronal death is thought to be the result of the altered communication between oligodendrocytes and neurons, probably due to the perturbation of the neurotrophic support leading to neurodegeneration. A post mortem study has shown MSA subjects with lower GDNF levels in the white matter of the frontal cortex and to a lesser degree in the cerebellum compared to controls. A transgenic mice model has shown a drecreased in GDNF protein expression in total brain lysates. These results have indicated that α-

synuclein expression in oligodendrocytes can impact the trophic support provided by oligodendrocytes for neurons, contributing to neurodegeneration. [87]

The hypothesis for the MSA pathogenesis depicted in figure 1, indicates that oligodendrocyte precursor cells mature and myelinate axons, and microglia is in a quiescent state. P25a is located in the myelinating processes of oligodendrocytes and α-synuclein in axons and synapses of neurons. The earliest stages of MSA pathogenesis are thought to include the overexpression and mis-localization of α-synuclein. This aberrant α-synuclein can result in p25a redistribution from the myelin to the oligodendrocyte cell soma, which is known to be associated with myelin dysfunction and an increase in cell soma size. This degenerative process and the direct effects of aberrant of α-synuclein can lead to the activation of microglia. [1]

α-synuclein undergoes fibril formation, promoting later on GCIs, which was also enhanced by p25α relocation, and these processes lead to oligodendrocyte dysfunction, with the retraction of myelinating processes. [103]

This is an indication that myelin homeostasis disruption is an early event in the pathophysiology cascade. [49]

Moreover, the myelin degeneration can leave the axons vulnerable to damage by proinflammatory molecules derived from activated microglia. The presence of GCIs leads to disorders of cellular functions and death of oligodendrocytes. [96]

This can contribute to a neuroinflammatory cascade and these dysfunctional oligodendrocytes can release the misfolded α-synuclein in the extracellular space; this misfolded α-synuclein can be taken by neighboring neurons to form neuronal cytoplasmic inclusions. Moreover loss of oligodendroglial neurotrophic support, neuroinflammation and neuronal dysfunction due to the presence of α-synuclein can promote neuronal death and subsequent reactive astrogliosis. Toxic α-synuclein species can then spread in a prion-like fashion to other functionally connected brain areas, leading to the multisystem neuronal involvement called multiple-system atrophy. (Fanciulli, A, 2015 , p.251-252)

3.2 Genetic and environmental factors

MSA is considered to be a sporadic disease, however different studies have shown a correlation between genes and MSA cases. Moreover an interaction between genes and environmental factors can contribute to the pathophysiological cascade of MSA.

Rare cases exist presenting a **family history of MSA**. Two cases were presented with familial MSA of unknown genetic background by Itoh and colleagues in 2014. The report is related to two affected siblings in one Japanese family (one MSA-C case and another MSA-P+C). An autosomal recessive inheritance represented this family due to the fact that both men and women were affected in successive generations. Advanced neural degeneration was seen with α-synuclein-immunoreactive inclusions distributed in more brain areas than the typical sporadic MSA. No multiplication of the SNCA gene or mutations in COC2 gene was identified and no molecules regulating α-synuclein that might have been involved in these familial MSA cases was yet discovered. [33]

In another study, four Japanese families were identified, in which multiple siblings were affected with MSA. One consanguineous marriage was observed (parents being first-degree cousins). Among eight patients, one of them had definite MSA (first family from the consanguineous marriage), five had probable MSA (from the first, second, third, and fourth family) and two had possible MSA (from the third and the fourth family). The most frequent phenotype was MSA-P, seen in five patients. One patient demonstrated MSA-C phenotype while two other patients had MSA-P+C phenotype. The families were expected for an autosomal recessive inheritance trait due to an ascertainment bias for multiplex families (X^2=1.53, P=.22) “The clinical phenotypes were concordant between the affected siblings in the three families (first, second, and third family). Furthermore six patients demonstrated pontine atrophy with cross sign or slit-like signal change at the posterolateral putaminal margin or both on brain MR imaging. No mutations in the SNCA gene were found in the family members.” (Hara, K , 2007, p.545-546)

An 84 year old woman from US presented with MSA. The patient’s two sisters also presented MSA symptoms showing the possibility for an autosomal dominant linkage. The study lacks SNCA gene testing and a conclusive autopsy was not obtained. [30]

The possibility for a hereditary component in MSA was seen in two successive generations in a German family. One of the affected individuals deceased at the age of 82 and had definite MSA (showed at postmortem examination). No mutation was revealed sequencing the entire region of the α-synuclein gene, moreover exon 3 and exon 4 demonstrated no increased SNCA dosage by quantitative PCR. Spinocerebellar ataxia was excluded and no mutation in the SNCA gene or a gene dosage effect was seen in the deceased patient. [104]

The family concerned comes from the western part of Japan. There are 18 living members in three generations, without any history of consanguinity α-synuclein is encoded by the **SNCA gene** located on 4q22.1, and it was demonstrated to be a cause of PD. (Katzeff, J. S , 2019) This gene was seen as an important candidate for MSA due to the fact that families with SNCA gene multiplications can present with an MSA phenotype. [1]
Testing if PD and MSA share a common genetic etiology, a SNP (single nucleotide polymorphism) study was done of the 385 most associated SNPs in a genome-wide association study of PD in 431 MSA cases and 3974 control subjects. The 10 most significant SNPs were then replicated in an additional 108 cases and 537 controls. SNPs at the SNCA locus were significantly associated with risk for increased risk for the development of MSA. How the identified SNCA risk haplotype influences the development of MSA is still unclear. The gene dosage measurements in MSA patients have not revealed SNCA duplications or triplications, however slight alterations in gene expression could be possible. This idea came from the fact that duplications and triplications of SNCA in autosomal dominant PD families leads to GCI-like inclusions and clinical features of MSA. [82] Further studies are needed in order to understand the proper mechanism regarding this gene. [83]

MAPT is located on chromosome 17q21.31 and encodes the protein tau. Based on evidence it is stated that tau can be involved in MSA pathogenesis. Even though tau is present in GCIs, it is unclear whether this is due to tau playing an active role in MSA pathogenesis or if this occurs during the disease process. [41]

The full MAPT haplotype diversity in MSA patients has been investigated tagging SNPs. In the study of Labbé in 2016, two protective haplotypes were identifies, H2 ($p=0.024$) and H1E ($p=0.014$). Also two rare risk haplotypes were discovered H1x ($P=0.030$) and H1J ($p=0.021$). Moreover, a greater size effect was in the MSA-C compared to MSA-P for H2. [50]

MAPT has also been shown to be clearly associated with PD indicating that MAPT may be a general risk factor for synucleinopathies. [1]

"COQ2 encodes the enzyme coenzyme-Q2 polyprenyltransferase in the biosynthetic pathway of coenzyme Q10. Deficiencies in coenzyme Q10 cause mitochondrial dysfunction, oxidative stress and reduced ATP synthesis." Several studies have investigated polymorphisms in COQ2 associated with MSA. (Katzeff, J. S , 2019, p.2)

The COQ2 p.S146N substitution has been previously reported as a pathogenic mutation in primary CoQ10 deficiency (including infantile multisystem disorder) in a recessive manner. This variant was found as the third primary Coq10 deficiency mutation observed in an MSA case (p.R387X and p.R197H). This indicates that in the heterozygous state it might possibly increase susceptibility to MSA.

In the study of Zhao et al (2015), it was seen that COQ2 V393A polymorphism was associated with increased risk of MSA in Han Chinese patients, as well as other East Asian populations. The same phenomenon had been previously seen in Japanese patients. [108]

However, other analysis failed to replicate these results in Caucasian populations. In the study of Procopio et al (2019), genetic screening of COQ2 gene was done in 100 MSA Italian patients. No pathological mutation was found, indicating that COQ2 is not a genetic risk factor for MSA in Italian population. [65]

Plasma coenzyme Q10 levels has been demonstrated to be lower in MSA patients compared to controls, giving evidence of the pathological link between COQ2 and MSA. Q10 levels showed to be lower in the cerebellum, demonstrating also increase in mitochondrial dysfunction and oxidative stress compared to controls. Importantly, these alterations happened in MSA cases in the absence of any COQ2 variant associated with MSA. [41]

Further evidence is needed to better understand the link between COQ2 and MSA pathogenesis, regarding also the ethnic groups. [41]

Occupational-environmental factors might contribute to MSA neurodegeneration. Patients with MSA had significantly more potential exposure to metal dusts and fumes, plastic monomers and additives, organic solvents, and pesticides than controls. [58]

In another study, one MSA patients had been exposed to high concentrations of malathion, diazinon, and formaldehyde, while the other patients with MSA had well documented high exposures to agents including n-hexane, benzene, methyl isobutyl ketone, and pesticides. Extensive advanced glial changes were seen in the MSA cases, including GCIs in deep cerebellar white matter, brainstem, cortex (superior frontal insula) and putamen, with notable cell loss and depigmentation of the substantia nigra and locus coeruleus. [26]

In the study of Zhou and colleagues (2016), they mentioned environmental factors such as farming and smoking as contributing factors to the occurrence of the disease in Chinese patients but not the progression of it. [109] Moreover, it has been indicated that smoking and farming influence MSA risk but individually, so they do not interact with each other.

Differently from previous mentions, in a case control study of French patients, MSA was not associated with exposure to pesticides, solvents, and other toxins neither to occupations, except plant and machine operators and assemblers (OR =10.0 [2.1– 47.5]). Also low education level was more frequent in MSA cases than in controls. Furthermore the risk of MSA increased with number of years in the occupation. (P=0.004). [91]

4. Clinical presentation

4.1 Motor features of MSA-P and MSA-C

In the sixth decade of life the first MSA-related symptoms are developed. Disability progresses rapidly while life expectancy is reduced. The medial survival has been from six to nine years from symptom onset. MSA-P and MSA-C can be distinguished clinically in the motor aspect, however they might reflect also an overlap, demonstrating a mixed phenotype. [49] Parkinsonian features predominate in 80% of patients regarding MSA-P subtype while cerebellar ataxia represents the main motor feature in 20% of patients for MSA-C subtype. Both clinical subtypes have similar survival rate but MSA-P patients demonstrate a more rapid functional deterioration than MSA-C. [98]

The parkinsonian subtype is characterized by progressive akinesia and rigidity, with jerky postural tremor, while tremors at rest, for example “pill-rolling” are seen less frequently. Orofacial or craniocervical dystonia accompanied by a typical quivering high-pitched dysarthria is also seen. At the beginning of the disease postural instability is seen, and the recurrent falls are not a typical symptom for MSA compared to PSP. [98]In the early stages of the disease is it difficult to distinguish MSA-P and PD due to overlapping characteristics such as rest tremor or asymmetrical akinesia and rigidity. [98] Progressive degeneration in the striatum is related to poor response or lack of response to Levodopa, which is a mandatory criteria for MSA-P, however 83% of patients may reflect effective results from dopaminergic replacement therapy. Unfortunately these effects are not permanent. Only a few MSA cases continue to show levodopa responsiveness. Furthermore levodopa withdrawal might show sometimes irreversible, symptomatic worsening in nonresponsive cases. Among responsive cases, they might show drug induced involuntary movements. Dyskinesias mostly of dystonic type in MSA, involve the craniocervical region, even though limb or generalized choreic dyskinesias may occur in 20% of levodopa-responsive patients. [96]

“Cerebellar ataxia predominates in the motor presentation of the cerebellar subtype of MSA. Cerebellar features consist of a wide-based gait, uncoordinated limb movements, action tremor, and spontaneous, gaze-evoked, or positional downbeat nystagmus. Spasticity or pyramidal weakness should cast doubts on a diagnosis of MSA, but generalized hyperreflexia, as well as Babinski sign, may occur in 30 to 50% of cases. Abnormal postures, including bent spine and disproportionate antecollis (severe forward neck flexion that interferes with eating, speaking, and vision) as well as hand or foot dystonia, are associated with the motor presentation of MSA in 16 to 42% of patients. Recurrent falls, dysphonia (voice-tone changes), dysarthria (difficulty in articulating words), drooling, and dysphagia are defining features of advanced disease.” (Fanciulli , A , 2015, p.258)

4.2 Non-motor features

The central and peripheral autonomic networks are affected in MSA, that correspond with cardiovascular, respiratory, urogenital, gastrointestinal and sudomotor disruptions. Some of these disturbances can precede motor feature onset. [34]

The most common pre-motor symptoms are genitourinary (erectile dysfunction, bladder dysfunction), orthostatic hypotension (OH) and REM sleep behavioral disorder. These symptoms can precede motor features by up to six year or more.

The presence of **urogenital symptoms** is described as an early event, preceding other neurological symptoms (including autonomic features) by several years. [34]

Bladder dysfunction in MSA is represented by frequency and urgency which is similar to PD characteristics, however in MSA there is also the presence of a constant urge with continuous leakage, usually occurring in advanced stages of the disease. Furthermore early urinary incontinence with involuntary partial or total bladder emptying is a typical MSA feature [12] Erectile dysfunction is almost universal among men with MSA preceding other non-motor and motor features. Regarding female patients, sexual dysfunction is mostly related to genital sensitivity. The presence of glial cytoplasmic inclusions in Onuf's and inferior intermediolateral sacral nuclei corresponds with bladder, rectal and sexual dysfunction. [12]

Orthostatic hypotension (OH) represents the main feature **of cardiovascular autonomic failure** in clinically established MSA. OH is explained by an orthostatic fall in systolic blood pressure of >20 mmHg or in diastolic blood pressure of >10 mmHg. MSA patients can remain asymptomatic despite the drop in systolic and diastolic blood pressure. The symptomatic state is characterized by syncope, light-headedness (dizziness), weakness, nausea, tremulousness, headache, or "coat-hanger pain" (pain in the neck and shoulder region) on standing. [12]

OH symptoms in MSA patients usually occur after the onset of erectile dysfunction and urinary symptoms. OH is caused by insufficient noradrenergic neurotransmission with diminished release of noradrenaline from sympathetic vasomotor neurons. MSA cases have shown primary central noradrenergic failure with relatively spared peripheral innervation. This lesion site represents the difference between MSA and PD. The hypotensive effects of levodopa treatment, fluid depletion, infections or physical deconditioning can worsen the OH intolerance [49]

OH symptoms are found in 50 % of MSA patients in their initial disease stage. MSA-C variant has demonstrated more frequency and severity regarding OH symptoms compared to MSA-P. Furthermore post-prandial hypotension and supine and nocturnal hypertension accompany orthostatic hypotension in half of patients with MSA. [96]

Gastrointestinal symptoms in MSA include dysphagia, constipation, and diarrhea. It is as common as in PD, they have the same severity and functional test abnormalities. What is typical to MSA is that sacral Onuf's nucleus is affected early in the disease progression, leading to common fecal incontinence reflecting weak anal sphincter muscles. [73]

Regarding the mechanism of constipation and fecal incontinence in MSA, MSA patients demonstrate weak abdominal strain, smaller rectal contraction on defecation, and larger anal contraction on defecation (paradoxical sphincter contraction on defecation). Except for the sphincter denervation and weakness in MSA the mechanism described shows similarities to the one of PD. Constipation in MSA most probably results from slow colonic transit, decreased phasic rectal contraction, and weak abdominal strain, and fecal incontinence results from weak anal sphincter due to denervation. The responsible sites for these dysfunctions seem to be both central and peripheral nervous systems that regulate the LGIT (lower grastrointestinal tract). [74]

Dysphagia demonstrates a frequent and disabling symptom in MSA, with an occurrence of 5 years of motor onset, representing an additional diagnostic feature. It can lead to aspiration pneumonia, which is a common cause of death. Abnormalities of the oral and pharyngeal phases of swallowing, esophageal dysfunction and aspiration occur in MSA and worsen as the disease progresses. Dysphagia is also associated with poor survival in MSA, unfortunately effective treatments for it are lacking and only compensatory strategies are available such as diet modification, swallowing maneuvers head postures etc. [8]

Regarding MSA subtypes, their difference has been seen in the early disease stages. In MSA-C, swallowing dysfunction represents the oral phase. The delayed bolus transport from the oral cavity to the pharynx is explained by the cerebellar dysfunction causing disturbed coordination of the tongue, while the pharyngeal phase was not affected at onset. Regarding MSA-P the pharyngeal phase has demonstrated to be more impaired at the initiation of the disease. After the disease progression in MSA-P swallowing dysfunction worsens compared to MSA-C, and progressive

worsening of dysphagia in MSA-C seems similar to PD. MSA-P patients require diet modifications earlier than MSA-C, and no significant difference was found between the subtypes regarding the latency of tube feeding onset. [8]

Features of **respiratory involvement** include **stridor**, **sleep related breathing disorders** and **respiratory insufficiency** which are listed as additional and supportive characteristics of the new consensus diagnostic criteria.

Regarding stridor, it occurs during sleep or during both sleep and wakefulness. Based on different clinical studies, stridor prevalence in MSA ranges from 12% to 42% and it is similar in MSA-C and MSA-P, it is presented as an initial manifestation of MSA. Furthermore, involuntary sighs or gasping are also frequent respiratory symptoms regarding probable MSA. [16]

Sleep disorders in MSA include REM sleep behavior disorder (RBD), excessive daytime sleepiness, reduced and fragmented sleep and sleep related breathing disorders. RBD (also being a red flag) has shown to be the most common symptom (affecting 90% – 100% of MSA patients). RBD and stridor during sleep can be initial symptoms of the disease, and might occur several years before the motor and dysautonomic onset. Regarding MSA subtypes, it was seen that sleep disorders occur in both MSA-P and MSA-C. (Ferini-Strambi, L, 2012, p. 467)

“RBD is represented by vigorous and injurious behavior related to vivid, action filled, and violent dreams during nocturnal REM sleep and REM sleep without atonia (RWA). According to the second edition of the International Classification of Sleep Disorders (ICSD), a clinical diagnosis of RBD can only be made when a patient displays violent, potentially violent or sleep-disruptive dream-enactment behavior along with RWA, as determined by a polysomnogram (PSG).” (Ferini-Strambi, L, 2012, p. 467)

RBD occurs in α-synucleinopathies such as PD and MSA. Brainstem regions are responsible for the occurrence of RBD and are also involved in the primary pathology of MSA and PD.

It is mentioned that acetylcholine and monoamine containing brainstem nuclei are responsible for REM sleep and its atonia. Regarding MSA cases and these acting structures of sleep, loss of cholinergic REM-on mesopontine neurons in the setting of loss of noradrenergic locus ceruleus neurons and preservation of rostral raphe serotonergic neurons, may contribute to REM sleep abnormalities. [24]

It is also suggested that RBD is part of the wide spectrum of dopamine deficiency disorders. Decreased nigrostriatal dopaminergic projections in idiopathic RBD is an example of this. Furthermore in MSA patient's severity of RBD has correlated with the loss of monoaminergic innervation of the striatum. Also neuronal loss is seen in the substantia nigra (SN) and the locus coeruleus in an individual with idiopathic RBD. [24]

"Nigrostriatal projections contribute to RBD through downstream connections, mainly with the PPN (pedunculopontine), which displays reciprocal connections with the SN and plays a major role in the regulation of REM sleep. In MSA, RBD could either be the result of basal ganglia dysfunction leading to secondary dysfunction of the PPN, or could be caused by primary dysfunction of the PPN or other key brainstem caudal structures associated with basal ganglia pathology through a temporal and topographic sequence of pathology similar to that found in PD. According to animal studies the GABAergic projections from the SN pars reticulate to the PNN could also be involved in the control of REM atonia." (Ghorayeb, I, 2005, p.167)

Sleep related breathing disorders have shown to be a frequent major problem in MSA. The most common symptoms are nocturnal stridor and obstructive sleep apnea (OSA) which often occur in conjunction despite their diverse pathophysiology.

Stridor reflects the upper airway obstruction at the level of the glottic aperture in the larynx due to partial or complete vocal cord abduction restriction. The high-pitched sound of stridor during sleep occurring during inspiration distinguishes it from snoring. However MSA patients with stridor can also have concomitant upper airway obstruction at the level of oropharynx leading to snoring and having a mix of stridor and snoring characteristics. Stridor during sleep in MSA occur in 13%-19% of patients. Using PSG (polysomnography) with synchronized audiovisual recording, stridor during sleep was seen in 30% to 42% of cases of variable intensity. On the other hand OSA occurs around 15% to 37% of cases. (Ferini-Strambi, L, 2012, p. 467)

Patients with stridor during sleep are at increased risk of developing OSA. With disease progression stridor can occur during daytime, indicating a severe glottis stenosis. In MSA stridor during wakefulness can lead to respiratory failure and death and stridor during sleep has been associated with low survival rate and higher rates of sudden death during sleep. (Ferini-Strambi, L, 2012, p. 467)

Nocturnal akinesia of the upper airway muscle can participate in the pathogenesis of sleep-related breathing disorders in MSA. Degeneration of the nucleus ambigus leading to vocal cord abductor atrophy and paralysis causing inspiratory stridor MSA was previously hypothesized. However studies based on EMG (electromyography) showed that stridor is due to sustained tonic activity similar to dystonia in adductor vocal cord muscles during inspiration leading to laryngeal narrowing and to the development of inspiratory flow limitation. (Ghorayeb, I, 2005, p.167)

Impaired ventilatory response to hypoxia and depletion of serotonergic and cholinergic neurons from key structures implicated in the control of respiratory function, especially during sleep can influence the dysfunction of respiratory control, leading to respiratory problems and accelerating the harmful role of stridor and apnea.

Based on fMRI (functional magnetic resonance imaging) evidence on cholinergic involvement is was seen that the OSA severity correlated with thalamic cholinergic deficit due to degeneration of brainstem cholinergic neurons originating from the pedunculopontine (PPN) and laterodorsal tegmental (LTD) nuclei. [24]

Sleep onset and maintenance insomnia is frequently reported by MSA patients. Sleep fragmentation and reduced sleep efficiency has been reported by polysomnographic studies. Regarding sleep fragmentation it is present in both clinical subtypes. In MSA-P it is caused by rigidity and bradykinesia with inability to turn over or to rise towards the bathroom. In MSA-C sleep fragmentation is caused due to poor motor coordination of cerebellar origin. Reduced and fragmented sleep is also influenced by urinary dysfunction [32].In a clinical questionnaire-based study it was seen that 70% of MSA patients showed complains of sleep disorder compared with 51% of PD patients. 53% of patients have shown complains **of sleep fragmentation**, followed by early waking (33%) and insomnia (20%) with no significant difference in PD. [79] Sleep disturbance is related to some brainstem nuclei in MSA. Sleep complaints in MSA patients are related to longer disease, disease severity, longer dopaminergic treatment and depression. [32]

Excessive daytime sleeping (EDS) has been reported in 28% of MSA patients, showing weak correlation with disease severity and no correlation with the amount of dopaminergic treatment. [79] This phenomenon in MSA seems to be more related to poor sleep efficiency and sleep-disordered breathing.

Restless leg syndrome (RLS) is a sensorimotor disorder, represented by uncomfortable sensations in the legs developing at rest that compel the person to move. RLS can contribute to sleep fragmentation and reduced sleep efficiency. This phenomenon in MSA occurred in 28% of cases, occurring more in frequency compared to PD cases (14%) and the control groups (7%). Furthermore restless leg syndrome did not correlate with dopaminergic treatment. [79] RLS has a higher prevalence in MSA-P patients compared to MSA-C. Moreover, two MSA-cases exhibited severe Parkinsonism. [25]

Other autonomic features are anhidrosis or hypohidrosis occurring frequently in MSA patients. It is explained that a preganglionic sudomotor dysfunction can contribute to the loss of sweating. The sudomotor dysfunction occurs frequently in MSA, however comparing the two subtypes it is seen more commonly and severely in MSA-P. More specifically MSA-P patients have abnormal thermoregulatory sweat test compared to MSA-C (98% versus 90%, P=0.006) and a higher mean percentage of anhidrosis (57%) compared to the cerebellar type (48%; P=0.033). The preganglionic pattern of sweat loss is common in MSA, however pre and postganglionic abnormalities can coexist. Regarding the temporal aspect, the frequency of postganglionic sudomotor abnormalities increases over time. [14]

“Features such as heat tolerance, impaired vasomotor sympathetic skin responses and skin temperature regulation have been mentioned in MSA. Additionally signs such as cold, ducky, violaceous hands, with poor circulatory return after blanching by pressure is seen in MSA. The cold-hand symptom is listed in the “red flag” features of the disease”. (Wenning, K.G , 2016, p.104)

Pupillomotor abnormalities have also been documented in MSA. Moreover between the disease subtypes there were no significant differences. The pupillography has shown no difference related to pupil diameter in darkness between MSA-P and MSA-C with a tendency of lower pupil diameter in darkness in MSA-C patients, which demonstrated to be significant when compared with healthy controls ($P < 0.018$). Also pupil light reflexes has revealed no significant differences between MSA-P and MSA-C patients. [78]

Pain complains in MSA are seen in 47% of MSA patients, musculoskeletal type in 64%, sensory type in 28%, and dystonic type in 21%. [56]

The overall pain prevalence has shown to be higher in MSA-P 63% (120/190) compared to MSA-C 41% (67/164). The greater involvement of the basal ganglia in MSA-P compared to MSA-C can influence the difference in pain prevalence. Moreover, MSA-P patients may have their head drop which can lead to musculoskeletal neck pain. Mobility impairment was a phenomenon more pronounced in MSA-C rather than MSA-P. [69]

MSA patients at an early stage demonstrate a reduced heat pain sensitivity in these patients compared to healthy controls and PD. Furthermore, no significant difference in electrical pain sensitivity and spinal nociception was detected between MSA patients and healthy controls or PD patients between MSA-C and MSA-P. MSA and PD patients didn't differ clinically regarding the prevalence and severity of chronic pain syndrome, however PD patients demonstrated an increase spinal nociception compared to healthy controls (HC). It is thought that pain sensitivity and the frequency of complains can increase over years with disease progression in MSA. [56]

The reduced heat sensitivity observed in MSA when compared to HC can indicate a frontal-executive dysfunction. Another explanation is related to a reduced somatosensory temporal discrimination in MSA patients and an alteration of the dorsal striatum is considered to contribute to this finding. [56]

Regarding spinal nociception, it is represented by an increasing trend rather than a reduced sensitivity with time in MSA patients. This comes also from the fact that MSA-P patients demonstrate increased spinal nociception at advanced stages of the disease. Based on this fact, it is stated that the difference in this disease is in the timing of the development of the symptoms, and not in the final characteristic. [69]

Emotional incontinence (part of the red flag features) is described as an exaggerated or inappropriate laughter, crying, or both, without an apparent motivation stimulus or in response to stimuli that would generally not elicit such an emotional response. Patients show difficulty controlling their emotional expression according to contextual information. This symptom represents one feature of the so-called pseudobulbar palsy, which develops after the degeneration of the corticobulbar tract. [107] In the study of Zhang L. and colleagues in 2021, emotional incontinence was a common feature in the disease. The younger age and greater disease severity were associated with the presence of emotional incontinence in both MSA-P and MSA-C. The

frequency of emotional incontinence in MSA was 12.7% (59/465), in MSA-P 12.0% (25/208), and 13.2% (34/257) in MSA-C. [107]

4.3 Cognitive impairment

Cognitive impairment in MSA is listed as a non-supporting feature of MSA mentioned in the DSM-IV. Moreover, previous studies of the disease were mostly based on infratentorial and subcortical structures, while the cortex pathology wasn't a main target of investigation. With further research, recent studies brought light to the cognitive dysfunctions in MSA and their anatomical correlates, leading to the possibility that cortical degeneration might influence the cognitive dysfunction in MSA. [44]

In the study of Kawai Y. and colleagues in 2008, cognitive impairment in MSA was investigated with the use of neuropsychological tests and SPECT (single-photon emission computed tomography). In the study, MSA patients demonstrated visuospatial and constructional dysfunction (Block design), impairment of verbal fluency, dysexecutive sundrome (Rule Shift Cards test) and depression (Hospital Anxiety and Depression). Moreover the Mini-Mental Sate Examination (MMSE) score used for overall assessment of cognition was not reduced, so it wasn't considered useful by the examiners. [42]

Comparing the two subtypes, MSA-P patients demonstrated cognitive dysfunctions in: visuospatial construction, verbal fluency, dysexecutive syndrome and depression. MSA-P patients showed hypo-perfusion in the medial frontal cortices and dorsolateral prefrontal cortex, and the severity of cognitive impairment was significantly correlated with hypo-perfusion in the dorsolateral prefrontal cortex. Moreover the results of the Block Design, phonemic and semantic fluency, and the Rule Shift Card tests significantly correlated with brain perfusion in the prefrontal cortices[42]On the other hand MSA-C patients showed visuospatial and constructional dysfunction (assessed by Block Design task) and depression that correlated with the perfusion in the prefrontal cortex and cerebellum.

To conclude on this study MSA-P patients show more severe and widespread cognitive dysfunctions compared to MSA-C patients. [42]

Another study investigated the cognitive deficits in MSA-C with the use of surface-based morphometry and region-of-interest cortical thickness, while performing also the neuropsychological examination. Significant cortical thinning was seen in the fronto-temporo-parietal regions along with volume reduction in subcortical structures with shape changes.

Moreover cerebellar volume had no significant effect on cortical and subcortical volumes. The severity of atrophic changes in the bilateral thalamus, the left cerebellum and the left pericalcarine gyrus significantly correlated with attentional (demonstrated on the digit span forward test), executive (shown on COWAT; the semantic and phonemic version, and STROOP test) and visuospatial (shown in the Rey Osterrieth Complex figure test) dysfunctions. Based on the results it was stated that cognitive impairment in MSA-C can result from functional disruption of the corticostriatal and pontocerebellar circuit mediated by primary cortical, cerebellar or thalamic pathology. [52]

Cognitive dysfunction in MSA-P was investigated by Kim J. and colleagues in 2015 by the use of the Seoul Neuropsychological Screening Battery, containing neuropsychological tests with standardized and validated tests for a variety of cognitive functions. Moreover they analyzed morphological changes using cortical thickness analysis, voxel-based morphometry (VBM) and cerebellar volumetry.

The cerebellar Volumetric Analysis showed that the mean cerebellar volume of MSA-P patients was significantly smaller than the control group. ($p < 0.005$)The Cortical Thickness Analysis showed two clusters exhibiting cortical thinning; the right paracentral lobule and the parahippocampal gyrus ($p < 0.001$, uncorrected). Moreover, the VBM analysis revealed significant gray-matter atrophy in MSA-P, in the bilateral basal ganglia, cerebellum, and temporal and frontal areas. [44]

Among the cortical thinning clusters, atrophy in the right parahipocampal gyrus showed a significant correlation with the participant's scores on the Digit Symbol Test (for attention) and TMT-A (Trail Making Test part-A). Moreover, among the clusters identified by VBM analysis, MoCA-K(Korean version of the Montreal Cognitive Assessment) (used for the general mental state) was associated with atrophy in the right basal ganglia, frontal area, cerebellum and left

temporal area. Praxis (language and related functions) correlated with the inferior frontal area. A significant correlation was found between SVLT (Seoul Verbal Learning Test) (test for learning and memory function, results being significant for immediate and delayed memory in this study) scores and changes in the bilateral temporal areas and cerebellum. The COWAT (Controlled Oral Association Test) (test for frontal and executive functions) scores correlated with morphological alterations in the bilateral basal ganglia, right cerebellum and frontal area. Finally, the TMT-B (Trail Making Test part-B) was significantly associated with atrophy in the right frontal area. [44]

The results indicated that cognitive impairment in MSA-P is influenced by cortical and cerebellar degeneration in addition to striatal pathology**.** [44]

Similar results were seen in the study of Caso F. in 2019 regarding MSA-P patients with the use of the neuropsychological evaluation and 1.5 MRI brain scan. Almost 46% of the patients showed mild cognitive impairment involving mainly attentive-executive and memory domains. Apathy and depression were found in half of MSA-P patients. Significant cortical thinning was seen in the fronto-temporal-parietal regions with atrophy of the periaqueductal gray matter, left cerebellar hemisphere, left pallidum and bilateral putamen. Cortical thinning in temporal regions correlated with the global cognitive status and memory impairment while grey matter cerebellar atrophy correlated with motor deficits. MSA-P patients demonstrated the multidomain cognitive impairment with more prominent cortical damage in anterior more than posterior brain regions and grey mater volume reduction in subcortical structures. [11]

In the study of Santagelo G. and colleagues in 2020, cognitive and neuropsychiatric symptoms in MSA-P and MSA-C were investigated in cross sectional studies. These patients underwent a neuropsychological battery and questionnaires assessing depression and apathy, and after one year they performed the same tests again.

At the initial assessment both subtypes demonstrated a worse performance regarding repetition abilities, executive and attention functions, compared to healthy controls. These results were found on the CAT (Constructional Apraxia Test), phonological and semantic fluency task, immediate and delayed copy of (ROCF) Rey-Osterrieth complex figure test, all tasks of Trail making test and all repetition tasks. [76]

Moreover the Dunn's test revealed that MSA-P patients performed worse on semantic fluency compared to MSA-C patients (repetition), and both clinical subtypes did not differ on any cognitive test.

Depression did not influence cognitive scores. Regarding the functional autonomy from the Dunn's test it was seen that both subtypes had a lower number of preserved ADL (activities of daily living) and IADL (instrumental activities of daily living) and no difference between MSA-P and MSA-C.

Furthermore, both subtypes showed depression in BDI-II (Beck depression inventory) and apathy in AES (Apathy evaluation scale), and no difference was seen between the subtypes.

"After one year, it was seen worsening in spatial planning and psychomotor speed in MSA-C group while MSA-P demonstrated significant worsening in memory, spatial planning, repetition abilities and functional autonomy. Furthermore, the prevalence of apathy increased in both subtypes, while the prevalence of depression was reduced in MSA-C and remained relatively consistent in MSA-P." (Santangelo, G , 2020 , p.67)

These findings indicated that with time MSA-P shows a wide-range worsening while MSA-C shows a decline in processing speed. [76]

Depression and anxiety in MSA, was investigated in a Chinese population by the use of the Hamilton Depression Rating Scale-24 items and Hamilton Anxiety Rating Scale. 62.0% of the patients showed mild depression and 71.7% of them showed anxiety. The severity of depression was associated with lower educational years (P=.024), longer disease duration (P<.001), and disease severity y (P<.001). The severity of anxiety was associated with an increased disease duration (P<.001), disease severity (P=.013), and orthostatic hypotension (P=.005).[106]

Hallucinations are defined as a sensory perception of visual, auditory, tactile or olfactory nature in the absence of an external stimulus. Regarding MSA we can find this characteristic in the non-supporting features since it is seen to be a rare condition for both subtypes. Only a few cases have been described with visual hallucinations as a secondary symptom after the neuropsychological changes in MSA. Pathologically confirmed MSA hallucinations were found in 9.5% of the patients in a study, with predominance of mild-to moderate visual and auditory symptoms. [6]

5. Conclusion

In the introduction we saw that MSA has undergone an evolvement during time regarding the main findings of the disease and the diagnostic criteria. The first clinical presentation of MSA was reported in 1900. It was named in 1969 by Graham Oppenhneimer. Moreover year 1998 was important in showing that Multiple system Atrophy is part of "α-synucelinopathies" due to the positively stained α-synuclein. The first diagnostic criteria was updated into the second consensus criteria in 2007.

We started by going over the pathophysiology, where Brain alterations showed by the macroscopic examination reflected the multisystem character of MSA. Atrophy of the cerebellum, middle cerebellar peduncle and the pontine nuclei associated with mild diffuse cortical atrophy in the frontal lobes. Moreover these anatomical alterations are seen in both disease subtypes (OPCA and SND). The histopathology of MSA is represented by α-synuclein-immunoreactive inclusions, selective neuronal loss and axonal degeneration, myelin pallor and gliosis, microglial activation and astrogliosis. Glial cytoplasmic inclusions (Papp-Lantos inclusions) are non-membrane bound cytoplasmic inclusions composed of filaments and tubules-in the brain increase with time duration, and they influence more the OPCA pathology rather than SND. They are thought to be a primary pathological process in the brain due to the random distribution. Basal ganglia has shown the highest density of GCIs in both clinical subtypes. The positive correlation of neuronal loss and the density of GCIs shows the link between neurodegeneration and these inclusions. Moreover glial intranuclear inclusions have a low frequency in the brain. Neuronal cytoplasmic inclusions and neuronal nuclear inclusions share the same ultrastructural aspect, with fibrillar structures resembling microtubules with inclusions Neuronal inclusions are seen in early disease stages, but compared to GCIs they are lower in number, and they show a similar distribution. The basis pontis and inferior olives have the highest frequency of neuronal inclusions. Neurodegeneration is widely distributed in striatonigral degeneration and olivopontocerebellar atrophy.

Localizations/ α-synuclein inclusions

GCIs	GNIs	NCIs	NNIs
Primary motor cortex	Pontine nuclei	Basis pontis and inferior olives(highest frequency)	Lateral cuneate
Basal ganglia : putamen and globus pallidus (greatest density),	Putamen	cortical, subcortical, brainstem, cerebellar nuclei (similar distribution)	Pontine nuclei
Substantia nigra	Subthalamic nucleus	Putamen, substantia nigra	
Suprasegmental motor system	Arcuate nucleus	Motor cortex	
Supraspinal autonomic system	Substantia nigra	Dentate gyrus	
Vestibular nuclei	Limbic system	Lateral cuneate	
Cranial nerves (V,VII,XII)	Inferior olivary nucleus	Striatum	
Brainstem	Reticular formation	Hippocampus	
		Forebrain	
		Thalamus	
	Brainstem	Neocortex	
		White mater of Cerebrum	
		Corpus callosum	

Table 1: Distribution of glial and neuronal inclusions.

[96], [62], [18], [60], [93], [17]

	Neurodegeneration
MSA-P	Dorsolateral caudal putamen and caudate nucleus, dorsolateral region of SNpc, globus pallidus, subthalamic nucleus Loss of medium-sized spiny GABAergic neurons Dopaminergic cell loss Both striatal outflow pathways affected (direct and indirect pathway)
MSA-C	Loss of purkinje cell layer of vermis, cerebellar dentate nucleus, basis pontis, Inferior olivary nucleus Cerebellar cortex
Other areas (found in both subtypes)	Dorsal nucleus of vagus (dmX), intermediolateral column of spinal cord, and Onufrowicz nucleus. Pyramidal tract (decrease in the number of small myelinated fibers), lower motor neurons, ambigui nuclei, ventral horn cells.
Autonomic nervous system	Supraspinal site(neurons in vagal nucleus and ambiguous nucleus, Edinger-Westphal nucleus and posterior hypothalamus) , sympathetic preganglionic neurons in the intermediolateral column of thoracolumbae spinal cord and postganglionic sudomotor denervation, mild degeneration of cardiac postganglionic sympathetic fibers
Peripheral nervous system	Reduction of unmyelinated fibers (sensory afferent and postganglionic sympathetic sympathetic fibers)
White mater	Reduced myelin staining in striatonigral and olivopontocerebellar regions. Myelin and axonal loss in frontal and parietal white mater.

Table 2: Distribution of neurodegenerative lesions

[10], [96], [77], [70], [23], [101], [36], [105], [15], [59], [94]

Pathological variants of MSA are represented by minimal change, preclinical and incidental MSA. Frontotemporal atrophy and severe limbic α -synuclein neuronal pathology are indicators for rare MSA. This disease has been presented also with tau-positive granules, AD's doesn't frequently co-occur with MSA while Lewy body disease has been presented in a few cases.

The accumulation of α-synuclein has been described by the relocation of p25α (TPPP) (the important stabilizer of myelin integrity) within the oligodendrocytes. Another explanation was that p25α (TPPP) in differentiated PC12 cells interferes with autophagic degradation of α-synuclein. Moreover, the presence of iron in oligodendrocytes can influence the aggregation directly or through increasing levels of oxidative stress converting native α-synuclein into a β-sheet conformation. An altered solubility of α-synuclein can precede the formation of GCIs, transforming this protein into insoluble oligomers. The presence of α-synuclein is of an ectopic nature since the oligodendroglial cells do not express α-synuclein mRNA. The spread of α-synuclein is explained by the prion-like propagation of these aggregates between synaptically connected areas. Oligodendroglial-myelin axis degeneration and accumulation of α-synuclein in the nerve cells are two parallel process suggested for the spread of the protein. Another form of transmission is suggested by astrocytes that seem to produce this protein at advanced stages of the disease. Oxidative stress is mentioned to increase the production of reactive oxygen species (ROS) damaging components of the cells such as protein, lipids and DNA. Mitochondrial toxin 3-nitropropionic acid (3NP) might alter α-synuclein. Moreover, the inflammatory cytokines are released by microglial cells under the influence of α-synuclein. The working hypothesis of MSA explained that p25α relocalization in the oligodendroglial soma is an early event, followed by cellular swelling and abnormal uptake and overexpression of α-synuclein by oligodendroglia. α -synuclein and p25α promote phosphorylation precipitate into glial cytoplasmic inclusions, which hinder neuronal trophic support and induce microglial activation. Misfolded α-synuclein released by dysfunctional oligodendrocytes can be taken up by neurons to form NCI. Neuroinflammation, loss of neurotrophic support and neuronal dysfunction due to α-synuclein inclusions simultaneously contribute to neuronal death in striatonigral, olivopontocerebellar and central autonomic pathways [19]. MSA is considered a sporadic disease, however familial cases are seen, and genes such as SNCA, MAPT, and COQ2 correlate with MSA in some studies. Some

environmental factors mentioned are: metal dusts, fumes, plastic monomers and additives, organic solvents, malathion, diazinon, formaldehyde, n-hexane, benzene, methyl isobutyl ketone and pesticides.

Motor symptoms can help distinguish between the two clinical subtypes but an overlap can be possible. Both clinical subtypes have a similar survival rate but the parkinsonian variant shows a more rapid functional deterioration rate. The most common autonomic features are: genitourinary (erectile and bladder dysfunction), orthostatic hypotension (OH) and REM sleep behavior disorder. Interestingly, MSA-P had more prevalence in the non-motor symptoms of restless leg syndrome, anhidrosis, pain, while MSA-C patients showed more prevalence for OH and had lower pupil diameter in darkness.

Motor features		**Non-motor features**
MSA-P	**MSA-C**	
Akinesia and rigidity Jerky postural tremor Orofacial or craniocervical dystonia High-pitched dysarthria Poor response to levodopa Drug induced involuntary movements	Wide-based gait Uncoordinated limb movements Action tremor Downbeat nystagmus Generalized hyperreflexia Babinski sign Abnormal posture Recurrent falls Dysarthria	**Urogenital** Bladder dysfunction (frequency and urgency) Urinary retention Involuntary partial or total bladder emptying Nocturia Erectile dysfunction in men, sexual dysfunction in women **Cardiovascular** Orthostatic hypotension Post-prandial hypotension Nocturnal hypertension **Gastrointestinal symptoms** Dysphagia, constipation, diarrhea, constipation, dysphagia **Respiratory involvement** Stridor, sleep related breathing disorders, respiratory insufficiency **Sleep disorders** Reduced and fragmented sleep, RBD, sleep related breathing disorders, excessive daytime sleepiness, restless leg syndrome **Pain**

		musculoskeletal, sensory , dystonic. **Other(**emotional incontinence, anhidrosis pupilomotor abnormalities)

Table 3. Motor and non-motor symptoms

[49], [98], [96], [19], [34], [12], [49], [73], [74], [8], [16], [24], [79], [32], [25], [14], [78], [56], [69], [107]

The presence of cognitive dysfunctions is represented in recent studies. The study of Kawai in 2008 demonstrated that MSA-P patients have a more widespread and severe cognitive dysfunction regarding visuospatial construction, verbal fluency, dysexecutive syndrome and depression correlating with hypo-perfusion in the dorsolateral prefrontal cortex. MSA-C patients have demonstrated visuospatial and constructional dysfunction and depression correlating with perfusion in the prefrontal cortex and cerebellum. [42] However, another study showed that MSA-C also represents a widespread cognitive dysfunction in attention, executive, and visuospatial correlating with functional disruption of the corticostriatal and pontocerebellar circuit.[52] The study of Kim J . in 2015 demonstrated that MSA-P patients represent cognitive dysfunctions in attention, learning and memory (immediate and delayed) and frontal executive functions correlating with thinning in the neocortex, cerebellum, and striatum.[44] Similar results were seen in the study of Caso F. in 2019 where MSA-P patients showed mild cognitive impairment related to attentive-executive and memory domains correlating with fronto-temporal-parietal atrophy.[11] In the study of Santagelo G. in 2020 MSA patients were re-assessed after a year for cognitive dysfunction and psychiatric symptoms. At the initial assessment both subtypes showed a worse performance on repetition abilities, executive and attention compared to controls. MSA-P patients performed worse on semantic fluency compared to MSA-C patients. After a year MSA-C subtype showed worsening in processing speed while MSA-P patients showed a wide-range worsening

(memory, spatial planning, repetition abilities and functional autonomy) Apathy increased in both subtypes while the prevalence of depression was reduced in MSA-C while in MSA-P patients remained relatively constant [76]. Moreover, the presence of hallucinations is seen in only a few MSA cases.

6. Discussion

At present, only symptomatic therapy is available for MSA, including pharmacologic and non-pharmacologic approaches. The symptoms that have treatment options are parkinsonism OH, sleep disorders, neurogenic bladder and constipation.

The parkinsonian variant of MSA demonstrates features of tremor, rigidity, bradykinesia and postural instability. This subtype is partially responsive to levodopa. “Around a third of MSA-P patients benefit from levodopa therapy, but with a mean duration of 3.5 years. Levodopa remains a first-line therapy with trial up to 2g daily dose recommended for at least 3 months.” (Burns, M. R , 2020 , p. 1587) Levodopa withdrawal can be presented with deterioration in some patients, even in nonresponsive cases which can lead to discontinuation of the treatment. [7]

OH exacerbation is a main concern regarding levodopa treatment. In these cases the standing BP needs to be recorded at baseline before the drug usage is initiated and recorded during time. [13] Moreover initiation and titration of medications fighting against low BP can be used. Levodopa usage should also be reduced in cases when it influences symptoms such as dyskinesias and facial dystonia. [13] The poor response to levodopa reflects the loss of striatal dopamine receptors and downstream pathology of the striatopallidal projections. [7]

Dopamine agonists are used as a second choice, however they are not an optimal choice due to the increased side effects. Amantadine up to 300 mg in 3 divided doses is also an alternative treatment, however further evidence is needed regarding this option. Moreover, non-pharmacological approaches including physical, occupational, and speech therapy are necessary and important therapies in MSA. Deep brain stimulation should not be included as a treatment option due to lack

of improvement of the symptoms. For dystonia symptoms, botulin toxin injections can be helpful. Treatment of cervical dystonia, in particular anterocollis, leads to possible risk due to underlying dysphagia. [7]

Ataxia, falls cerebellar ataxia, fall risks and imbalance do not improve from medication. Use of sedative drugs or muscle relaxants should be excluded from the daily use to improve the balance. Physical therapy can improve motor symptoms based on the patient's needs and strategies should be considered in order of avoiding falls (which has the risk of fractures). Bisphosphonates or similar drugs needs to be considered, while vitamin D would be also a helpful option. [13]

Treatment of OH is focused on the increase of intravasal volume and the reduction of volume shift to lower body parts when changing into upright position. Postural hypotension is exacerbated after prolonged recumbency, mealtime, and physical exertion. Other factors that worsen postural hypotension are alcohol, coughing, defecation and heat. The severity of the symptoms determines the treatment of the patients, if it will be pharmacologic, non-pharmacologic, or a combination of both. [99]

Non-pharmacologic treatment includes: high-salt diet, sufficient fluid intake, reducing postprandial hypotension by spreading the total carbohydrate intake by consuming more frequent smaller meals per days, and custom made elastic body garments. "A head-up tilt at night reduces hypertensive cerebral perfusion pressure and increases intravasal volume up to 1 liter within a week, which improves hypotension in the morning. This is achieved by an increased secretion of renin due to reduced perfusion pressure and reduced atrial natriuretic hormone because of lower atrial pressure. This approach is successful in particular in combination with the mineralocorticoid fludrocortisone, which further supports sodium retention." (Wenning, G. K , 2005, p. 72)

Related to pharmacologic treatment, midodrine is used, however it can exacerbate urinary retention. Phenylpropanolamine (in low doses) or yohimbine or indomethacin (in moderate doses) can also be used. l-threodihydroxy-phenylserine (l-threo-DOPS) seems to be a promising drug. Somatostatin analogue, octreotide, have demonstrated to be helpful in postprandial hypotension because it inhibits release of vasodilatory gastrointestinal peptides and does not worsen nocturnal hypertension. [97]

MSA patients having neurogenic bladder symptoms should be screened regularly for urinary tract infections. Medications for urinary urgency (anticholinergics, mirabergon) are for hyperactive bladders and may worsen hypoactive bladder symptoms. It is suggested that postvoid residual urine should be checked before the prescription of the medication. Retention of more than 100 mL of urine suggests a hypoactive bladder and medications cannot manage this problem. Management of symptoms include timed voiding to reduce overflow incontinence. Clean intermittent catheterization, indwelling catheterization, or suprapubic catheterization are also recommended. [13]

Treatment of sexual dysfunction needs to be evaluated individually in every MSA patient. Sildenafil citrate can be effective in treating erectile dysfunction in these patients, however careful usage is suggested due to it's cardiovascular side effects. Erectile dysfunction can also be improved by oral yohimbine or by intracarvernosal injection of papaverine or a penis implant. [99]

Regarding REM sleep behavior disorder (RBD) the most used medications are clonazepam and melatonin. Unfortunately clonazepam can worsen comorbid sleep apnea. Zopiclone is suggested in these cases with less severe adverse effects. Melatonin has shown less side effects compared to clonazepam. Adverse effects mentioned are: hallucinations, daytime sleepiness and morning headache. Since RBD is manifested with acting of violent dreams during sleep, the treatment consists of the safety of the patient and the bed partner. This includes organizing the sleeping room in a way that the patients cannot use objects to harm themselves or the partner. Moreover an alarm system called The Posey Sitter Select has been invented in order of providing a calming instruction to the patient to return back to sleep. (Ferini-Strambi, L, 2012, p. 469)

Stridor is treated with tracheostomy or continuous positive airway pressure (CPAP). "Tracheostomy bypassing the vocal cord obstruction completely eliminates stridor both during sleep and wakefulness. CPAP abolishes nocturnal stridor because it increases the glottic aperture separating the vocal cords and reducing the downward displacement of the larynx. An increased survival in MSA patients with stridor may be obtained both with CPAP and tracheostomy". Lack of knowledge exists for treatments such as botulinum toxin and laryngeal surgery but they have shown to increase the risk of bronchial aspiration (which leads to death). (Ferini-Strambi, L, 2012, p. 469)

Tracheotomy and vocal cord surgery are traditional treatments regarding stridor, while botulinum has not been investigated enough. Moreover, CPAP is also an effective treatment for the elimination of obstructive sleep apnea, however adapting in severe disease stages is difficult.

For dysphagia, there are no known specific effective procedures regarding treatment. Even though there is a high prevalence of oral symptoms, the treatment strategies are not recommended since no study has addressed them systematically or investigated a potential therapeutic approach. Compensatory treatment was selected. Modifications of the texture of liquids and food or of bolus volume can be done. Postural adjustment by chin down movement (chin-tuck maneuver) helps the patients when attempting to swallow. However this procedure is not applicable for patients with antecollis. Furthermore, there is the supraglottic swallow maneuver (a technique aiming to close the vocal cords and the supraglottic structures to protect the upper airways well in advance of the bolus arriving). Finally, the enhancement of oral care and proper positioning of the patient's head, both after meals and during sleep can be done (reduces the risk of aspiration pneumonia).[8]

"Constipation can be very difficult to treat in patients with MSA. Satisfying results are likely to be achieved if alimentary measures are combined with regular administration of osmotic bulking agents." (Fanciulli , A , 2015 , p.259)

The speech findings in MSA include dysarthria with combinations of hypokinetic, ataxic and spastic components. Important elements for an improved communication are speech examination and therapy. Severe dysarthria at the initial course of the disease is an indication of the disease. [13]

For neuropathic pain, gabapentin or pregabalin is suggested. In individuals with muscle contraction pain associated with dystonia (cervical region or limbs) targeted botulinum toxin injections can be effective. Involvement of palliative care specialists of pain medicine specialists can be helpful in patients whom pain is interfering the quality of life. [13]

Therapeutic guidance for depression and anxiety are lacking. Selective serotoninergic reuptake inhibitors (SSRIs) are prescribed, that are more effective than other medications. The combination of this medication with psychotherapy is a common clinical approach recommended. [7]

Regarding cognitive dysfunction, acetylcholinesterase inhibitors such as donepezil or rivastigmine are often employed with modest benefit. [7] The cognitive and psychiatric features of MSA should be explored in the daily clinical practice. [76]

Treatment of emotional incontinence needs more focus. Treatments such as (SSRIs) dextromethorphan/quinidine, tricyclic antidepressants, noradrenergic reuptake inhibitors, novel antidepressants, and dopaminergic agents may be useful for emotional incontinence. Further advancement are needed in the assessment of the symptoms (for example assessment of severity). [107]

The pharmacological symptomatic treatments described have not evolved much with time. "The major care innovation is related to a multidisciplinary care team approach, bringing together health care expertise in neurology, urology, cardiology, gastroenterology, speech therapy, physiotherapy, occupational therapy, palliative care and others needed for the management of the complex spectrum of motor and non-motor symptoms endured by MSA patients." The symptomatic treatment provides a temporary and not complete benefit to some individuals which leads to the need of further studies in order of including novel targets and methodological improvements. (Meissner, W. G , 2019, p. 1635)

Studies for therapeutic alternatives should focus on different aspects. The mechanism of α-synuclein accumulation in oligodendrocytes and neurons and this protein propagation are important elements leading to drug discovery. Unfortunately, the knowledge on how this protein is accumulated in oligodendrocytes is still unknown, making more challenging the possibility of finding suitable targets for preventing α-synuclein aggregation in early stages. Approaches limited to reducing α-synuclein accumulation in oligodendrocytes may not be enough to stop or delay the progression of the disease. Moreover, the neuronal pathology in MSA needs further exploration, since the main focus on MSA has mostly been α-synuclein in oligodendrocytes. The disease modification can also consist of a reduction in neuronal expression and release of α-synuclein which would also reduce the amount of α-synuclein in oligodendrocytes. Moreover, supporting and repairing neuronal function by restoring trophic support (such as BDNF), myelination, and the use of regenerative therapies, should be considered as therapeutic alternatives for MSA. [88]

The main target for neuroprotective treatment until today has been α-synuclein due to the understanding of the disease mechanisms. Regarding preclinical trials, the European ARTEMIS consortium is testing the effect of four complementary strategies targeting α-synuclein in models of MSA. These strategies are tested alone and could be more efficient in combination. VX-765, a caspase-1 inhibitor prodrug, and anle138b, a small molecule targeting oligomeric α-synuclein, have demonstrated effects on motor behavior, α-synuclein burden, and other neuropathological readouts in transgenic MSA mice. Other compounds, targeting α-synuclein (eg, CLRO1, MPLA, transcription factor EB, active immunization) and other disease mechanisms (such as NaPB, exenatide, benztropine) have demonstrated positive results regarding MSA preclinical models. Beyond active immunization, other compounds in the future can also play a role in the clinical development. These compounds can be AFF1 (corresponds to PD01 in clinical trials), Anle138b, CD5-D5 and lenalidomide, CLR01, Neurosin etc. (Meissner, W. G , 2019 , p.1634)

Clinical trials for neuroprotection have not shown significant effects. Only one study has demonstrated positive effects on the progression of UMSARS (Unified MSA rating scale) scores performed placebo-controlled intravenous and intra-arterial injections of mesenchymal stem cells (MSCs). This study showed limitations due to the single-center design, the exclusive recruitment of patients with MSA-C and the safety concerns due to the small ischemic lesions on MRI in one-third of the participants. (Meissner, W. G , 2019 , p.1634)

The lack of an effective disease-modifying therapy for MSA, leads to the need for an early and accurate diagnosis which is important for the optimal management of the patients and for the development of therapeutic strategies. [43] An issue regarding the diagnosis of MSA is differentiating it from other neurodegenerative diseases such as Parkinson's disease, progressive supranuclear palsy (PSP) and dementia with Lewy body (DLB). The heterogeneity of these diseases and the presence of variants makes it more challenging to give a reliable diagnosis. Moreover, there is still the need of diagnostic biomarkers for MSA. [43]

Identifying early biomarkers that reflect the underlying disease process or progression would not only help in the early diagnosis, but also for designing future clinical trials that assess compounds with putative disease-modifying or neuroprotective properties. Modern neuroimaging techniques

have improved clinical accuracy in distinguishing MSA and PD. [38] Skin punch biopsies for the demonstration of α-synuclein aggregations in Schwann or other cells in peripheral nerves in MSA is under discussion regarding its diagnostic validity. Studies on fluid markers has suggested that combination of several CSF fluid biomarkers can lead to a more successful outcome compared to using one marker. A combination of the light chain of neurofilaments, metabolites of the catecholamine pathway (dopamine and norepinephrine) and proteins (α-synuclein, DJ-1, and total tau) may represent the most useful markers today. Combining DJ-1, total-tau (t-tau) and phosphorylated tau (p-tau) demonstrated high sensitivity (82%) and specificity (91%) for the distinction between MSA and PD or controls. The combination of NFL and FLT3 ligand with DJ-1, t-tau protein in CSF had the best result for distinguishing MSA from PD and controls. This combination could indicate a potential biomarker for the differential diagnosis between MSA and PD. "Moreover, the results of proteomics for biomarker discovery and of miRNA expression need validation using independent technologies." The research of biomarkers for MSA will need to continue. (Jellinger, K. A , 2016 , p.564)

Interestingly, machine learning and multimodal techniques can also help in the diagnosis of MSA. Extracting volumes of the cerebellum, brain stem, and putamen from T1-weighted images seems to be an optimal way of distinguishing between MSA and PD. In a study, all patients with a MSA diagnosis were correctly classified by MRI volumetry, while the accuracy of consensus diagnostic criteria was only 65%. Moreover, adding the volume of the middle cerebellar peduncle to the classifier algorithm improved more the distinction between MSA and PD. Unsupervised deep-learning methods on multimodal MRI findings also provided consistent and coherent clusters of participants. [55]

Finally, using new technologies such as genome wide association studies (GWASs) could help to validate risk factors and identify new disease-associated genes, leading towards disease mechanisms and potential therapeutic targets. [1]

To conclude, it is important to go beyond the symptomatic treatment in MSA. A better understanding of the mechanism of the disease will lead to new treatment strategies. Preclinical and clinical studies should continue in order of giving new insights. Identifying early biomarkers

will help with a better clinical diagnosis and in designing future clinical trials. Moreover the evolvement of machine learning will also help in advancing the diagnosis. The continuation of genetic studies will also give new findings in the future.

7. Bibliography

[1] Ahmed, Z., Asi, Y. T., Sailer, A., Lees, A. J., Houlden, H., Revesz, T., & Holton, J. L. (2012). The neuropathology, pathophysiology and genetics of multiple system atrophy. *Neuropathology and Applied Neurobiology*, *38*(1), 4-24. https://doi.org/10.1111/j.1365-2990.2011.01234.x

[2] Aoki, N., Boyer, P. J., Lund, C., Lin, W. L., Koga, S., Ross, O. A., Weiner, M., Lipton, A., Powers, J. M., White, C. L., & Dickson, D. W. (2015). Atypical multiple system atrophy is a new subtype of frontotemporal lobar degeneration : frontotemporal lobar degeneration associated with α-synuclein. *Acta Neuropathologica*, *130*(1), 93-105. https://doi.org/10.1007/s00401-015-1442-z

[3] Armstrong, R., Cairns, N., & Lantos, P. (2006). Multiple system atrophy (MSA) : Topographic distribution of the α-synuclein-associated pathological changes. *Parkinsonism & Related Disorders*, *12*(6), 356-362. https://doi.org/10.1016/j.parkreldis.2006.02.005

[4] Bernis, M. E., Babila, J. T., Breid, S., Wüsten, K. A., Wüllner, U., & Tamgüney, G. (2015). Prion-like propagation of human brain-derived alpha-synuclein in transgenic mice expressing human wild-type alpha-synuclein. *Acta Neuropathologica Communications*, *3*(1). https://doi.org/10.1186/s40478-015-0254-7

[5] Brooks, D. J., & Seppi, K. (2009). Proposed neuroimaging criteria for the diagnosis of multiple system atrophy. *Movement Disorders*, *24*(7), 949-964. https://doi.org/10.1002/mds.22413

[6] Burghaus, L., Eggers, C., Timmermann, L., Fink, G. R., & Diederich, N. J. (2011). Hallucinations in Neurodegenerative Diseases. *CNS Neuroscience & Therapeutics*, *18*(2), 149-159. https://doi.org/10.1111/j.1755-5949.2011.00247.x

[7] Burns, M. R., & McFarland, N. R. (2020). Current Management and Emerging Therapies in Multiple System Atrophy. *Neurotherapeutics*, *17*(4), 1582-1602. https://doi.org/10.1007/s13311-020-00890-x

[8] Calandra-Buonaura, G., Alfonsi, E., Vignatelli, L., Benarroch, E. E., Giannini, G., Iranzo, A., Low, P. A., Martinelli, P., Provini, F., Quinn, N., Tolosa, E., Wenning, G. K., Abbruzzese, G., Bower, P., Antonini, A., Bhatia, K. P., Bonavita, J., Pellecchia, M. T., Pizzorni, N.,. . . Kaufmann, H. (2021). Dysphagia in multiple system atrophy consensus

statement on diagnosis, prognosis and treatment. *Parkinsonism & Related Disorders*, *86*, 124-132. https://doi.org/10.1016/j.parkreldis.2021.03.027

[9] Campbell, B. C. V., McLean, C. A., Culvenor, J. G., Gai, W. P., Blumbergs, P. C., Jäkälä, P., Beyreuther, K., Masters, C. L., & Li, Q. X. (2008). The solubility of α-synuclein in multiple system atrophy differs from that of dementia with Lewy bodies and Parkinson's disease. *Journal of Neurochemistry*, *76*(1), 87-96. https://doi.org/10.1046/j.1471-4159.2001.00021.x

[10] Campese, N., Fanciulli, A., Stefanova, N., Haybaeck, J., Kiechl, S., & Wenning, G. K. (2021). Neuropathology of multiple system atrophy : Kurt Jellinger`s legacy. *Journal of Neural Transmission*, *128*(10), 1481-1494. https://doi.org/10.1007/s00702-021-02383-3

[11] Caso, F., Canu, E., Lukic, M. J., Petrovic, I. N., Fontana, A., Nikolic, I., Kostic, V. S., Filippi, M., & Agosta, F. (2019). Cognitive impairment and structural brain damage in multiple system atrophy-parkinsonian variant. *Journal of Neurology*, *267*(1), 87-94. https://doi.org/10.1007/s00415-019-09555-y

[12] Colosimo, C. (2011). Nonmotor presentations of multiple system atrophy. *Nature Reviews Neurology*, *7*(5), 295-298. https://doi.org/10.1038/nrneurol.2011.5

[13] Coon, E. A., & Ahlskog, J. E. (2021). My Treatment Approach to Multiple System Atrophy. *Mayo Clinic Proceedings*, *96*(3), 708-719. https://doi.org/10.1016/j.mayocp.2020.10.005

[14] Coon, E. A., Fealey, R. D., Sletten, D. M., Mandrekar, J. N., Benarroch, E. E., Sandroni, P., Low, P. A., & Singer, W. (2016). Anhidrosis in multiple system atrophy involves pre- and postganglionic sudomotor dysfunction. *Movement Disorders*, *32*(3), 397-404. https://doi.org/10.1002/mds.26864

[15] Coon, E. A., Schmeichel, A. M., Parisi, J. E., Cykowski, M. D., Low, P. A., & Benarroch, E. E. (2016). Medullary neuronal loss is not associated with α-synuclein burden in multiple system atrophy. *Movement Disorders*, *31*(12), 1802-1809. https://doi.org/10.1002/mds.26798

[16] Cortelli, P., Calandra-Buonaura, G., Benarroch, E. E., Giannini, G., Iranzo, A., Low, P. A., Martinelli, P., Provini, F., Quinn, N., Tolosa, E., Wenning, G. K., Abbruzzese, G., Bower, P., Alfonsi, E., Ghorayeb, I., Ozawa, T., Pacchetti, C., Pozzi, N. G., Vicini, C.,.

. . Meissner, W. G. (2019). Stridor in multiple system atrophy. *Neurology*, *93*(14), 630-639. https://doi.org/10.1212/wnl.0000000000008208

[17] Cykowski, M. D., Coon, E. A., Powell, S. Z., Jenkins, S. M., Benarroch, E. E., Low, P. A., Schmeichel, A. M., & Parisi, J. E. (2015). Expanding the spectrum of neuronal pathology in multiple system atrophy. *Brain*, *138*(8), 2293-2309. https://doi.org/10.1093/brain/awv114

[18] Dickson, D. W., Liu, W. K., & Yen, S. H. (1999). WIDESPREAD ALTERATIONS IN ALPHA-SYNUCLEIN IN MULTIPLE SYSTEM ATROPHY. *Journal of Neuropathology and Experimental Neurology*, *58*(5), 554. https://doi.org/10.1097/00005072-199905000-00191

[19] Fanciulli, A., & Wenning, G. K. (2015). Multiple-System Atrophy. *New England Journal of Medicine*, *372*(3), 249-263. https://doi.org/10.1056/nejmra1311488

[20] Fellner, L., & Stefanova, N. (2012). The Role of Glia in Alpha-Synucleinopathies. *Molecular Neurobiology*, *47*(2), 575-586. https://doi.org/10.1007/s12035-012-8340-3

[21] Ferini-Strambi, L., & Marelli, S. (2012). Sleep Dysfunction in Multiple System Atrophy. *Current Treatment Options in Neurology*, *14*(5), 464-473. https://doi.org/10.1007/s11940-012-0189-2

[22] Fujishiro, H., Ahn, T. B., Frigerio, R., DelleDonne, A., Josephs, K. A., Parisi, J. E., Eric Ahlskog, J., & Dickson, D. W. (2008). Glial cytoplasmic inclusions in neurologically normal elderly : prodromal multiple system atrophy ? *Acta Neuropathologica*, *116*(3), 269-275. https://doi.org/10.1007/s00401-008-0398-7

[23] Geser, F., Colosimo, C., & Wenning, G. K. (2005) . Multiple system atrophy. Neurogenerative Disease, 623-662. doi:10.1017/cbo9780511544873.044

[24] Ghorayeb, I., Bioulac, B., & Tison, F. (2005). Sleep disorders in multiple system atrophy. *Journal of Neural Transmission*, *112*(12), 1669-1675. https://doi.org/10.1007/s00702-005-0348-7

[25] Ghorayeb, I., Dupouy, S., Tison, F., & Meissner, W. G. (2014). Restless legs syndrome in multiple system atrophy. *Journal of Neural Transmission*, *121*(12), 1523-1527. https://doi.org/10.1007/s00702-014-1232-0

[26] Hanna, P. A., Jankovic, J., & Kirkpatrick, J. B. (1999). Multiple System Atrophy. *Archives of Neurology*, *56*(1), 90. https://doi.org/10.1001/archneur.56.1.90

[27] Hara, K., Momose, Y., Tokiguchi, S., Shimohata, M., Terajima, K., Onodera, O., Kakita, A., Yamada, M., Takahashi, H., Hirasawa, M., Mizuno, Y., Ogata, K., Goto, J., Kanazawa, I., Nishizawa, M., & Tsuji, S. (2007). Multiplex Families With Multiple System Atrophy. *Archives of Neurology*, *64*(4), 545. https://doi.org/10.1001/archneur.64.4.545

[28] Hoffmann, A., Ettle, B., Battis, K., Reiprich, S., Schlachetzki, J. C. M., Masliah, E., Wegner, M., Kuhlmann, T., Riemenschneider, M. J., & Winkler, J. (2019). Oligodendroglial α-synucleinopathy-driven neuroinflammation in multiple system atrophy. *Brain Pathology*, *29*(3), 380-396. https://doi.org/10.1111/bpa.12678

[29] Hoffmann, A., Ettle, B., Bruno, A., Kulinich, A., Hoffmann, A. C., von Wittgenstein, J., Winkler, J., Xiang, W., & Schlachetzki, J. C. (2016). Alpha-synuclein activates BV2 microglia dependent on its aggregation state. *Biochemical and Biophysical Research Communications*, *479*(4), 881-886. https://doi.org/10.1016/j.bbrc.2016.09.109

[30] Hohler, A., & Singh, V. (2012). Probable hereditary multiple system atrophy–autonomic (MSA–A) in a family in the United States. *Journal of Clinical Neuroscience*, *19*(3), 479-480. https://doi.org/10.1016/j.jocn.2011.06.018

[31] Homma, T., Mochizuki, Y., Tobisawa, S., Komori, T., & Isozaki, E. (2020). Cerebral white matter tau-positive granular glial pathology as a characteristic pathological feature in long survivors of multiple system atrophy. *Journal of the Neurological Sciences*, *416*, 117010. https://doi.org/10.1016/j.jns.2020.117010

[32] Iranzo, A. (2007). S47.C Sleep and breathing in multiple system atrophy. *Sleep Medicine*, *8*, S45. https://doi.org/10.1016/s1389-9457(07)70178-8

[33] Itoh, K., Kasai, T., Tsuji, Y., Saito, K., Mizuta, I., Harada, Y., Sudoh, S., Mizuno, T., Nakagawa, M., & Fushiki, S. (2014). Definite familial multiple system atrophy with unknown genetics. *Neuropathology*, *34*(3), 309-313. https://doi.org/10.1111/neup.12092

[34] Jecmenica-Lukic, M., Poewe, W., Tolosa, E., & Wenning, G. K. (2012). Premotor signs and symptoms of multiple system atrophy. *The Lancet Neurology*, *11*(4), 361-368. https://doi.org/10.1016/s1474-4422(12)70022-4

[35] Jellinger, K. A. (2007). More frequent Lewy bodies but less frequent Alzheimer-type lesions in multiple system atrophy as compared to age-matched control brains. *Acta Neuropathologica*, *114*(3), 299-303. https://doi.org/10.1007/s00401-007-0227-4

[36] Jellinger, K. A. (2014). Neuropathology of multiple system atrophy : New thoughts about pathogenesis. *Movement Disorders*, *29*(14), 1720-1741. https://doi.org/10.1002/mds.26052

[37] Jellinger, K. A., & Lantos, P. L. (2010). Papp–Lantos inclusions and the pathogenesis of multiple system atrophy : an update. *Acta Neuropathologica*, *119*(6), 657-667. https://doi.org/10.1007/s00401-010-0672-3

[38] Jellinger, K. A., & Wenning, G. K. (2016). Multiple system atrophy : pathogenic mechanisms and biomarkers. *Journal of Neural Transmission*, *123*(6), 555-572. https://doi.org/10.1007/s00702-016-1545-2

[39] Kaindlstorfer, C., Jellinger, K. A., Eschlböck, S., Stefanova, N., Weiss, G., & Wenning, G. K. (2018). The Relevance of Iron in the Pathogenesis of Multiple System Atrophy : A Viewpoint. *Journal of Alzheimer's Disease*, *61*(4), 1253-1273. https://doi.org/10.3233/jad-170601

[40] Kaji, S., Maki, T., Ishimoto, T., Yamakado, H., & Takahashi, R. (2020). Insights into the pathogenesis of multiple system atrophy : focus on glial cytoplasmic inclusions. *Translational Neurodegeneration*, *9*(1). https://doi.org/10.1186/s40035-020-0185-5

[41] Katzeff, J. S., Phan, K., Purushothuman, S., Halliday, G. M., & Kim, W. S. (2019). Cross-examining candidate genes implicated in multiple system atrophy. *Acta Neuropathologica Communications*, *7*(1). https://doi.org/10.1186/s40478-019-0769-4

[42] Kawai, Y., Suenaga, M., Takeda, A., Ito, M., Watanabe, H., Tanaka, F., Kato, K., Fukatsu, H., Naganawa, S., Kato, T., Ito, K., & Sobue, G. (2008). Cognitive impairments in multiple system atrophy : MSA-C vs MSA-P. *Neurology*, *70*(Issue 16, Part 2), 1390-1396. https://doi.org/10.1212/01.wnl.0000310413.04462.6a

[43] Kim, H. J., Jeon, B. S., & Jellinger, K. A. (2015). Diagnosis and differential diagnosis of MSA : boundary issues. *Journal of Neurology*, *262*(8), 1801-1813. https://doi.org/10.1007/s00415-015-7654-2

[44] Kim, J. S., Yang, J. J., Lee, D. K., Lee, J. M., Youn, J., & Cho, J. W. (2015). Cognitive Impairment and Its Structural Correlates in the Parkinsonian Subtype of Multiple System Atrophy. *Neurodegenerative Diseases*, *15*(5), 294-300. https://doi.org/10.1159/000430953

[45] Koga, S., & Dickson, D. W. (2017). Recent advances in neuropathology, biomarkers and therapeutic approach of multiple system atrophy. *Journal of Neurology, Neurosurgery & Psychiatry*, *89*(2), 175-184. https://doi.org/10.1136/jnnp-2017-315813

[46] Koga, S., Li, F., Zhao, N., Roemer, S. F., Ferman, T. J., Wernick, A. I., Walton, R. L., Faroqi, A. H., Graff-Radford, N. R., Cheshire, W. P., Ross, O. A., & Dickson, D. W. (2020). Clinicopathologic and genetic features of multiple system atrophy with Lewy body disease. *Brain Pathology*, *30*(4), 766-778. https://doi.org/10.1111/bpa.12839

[47] Kon, T., Mori, F., Tanji, K., Miki, Y., & Wakabayashi, K. (2013). An autopsy case of preclinical multiple system atrophy (MSA-C). *Neuropathology*, *33*(6), 667-672. https://doi.org/10.1111/neup.12037

[48] Krejciova, Z., Carlson, G. A., Giles, K., & Prusiner, S. B. (2019). Replication of multiple system atrophy prions in primary astrocyte cultures from transgenic mice expressing human α-synuclein. *Acta Neuropathologica Communications*, *7*(1). https://doi.org/10.1186/s40478-019-0703-9

[49] Krismer, F., & Wenning, G. K. (2017). Multiple system atrophy: insights into a rare and debilitating movement disorder. *Nature Reviews Neurology*, *13*(4), 232-243. https://doi.org/10.1038/nrneurol.2017.26

[50] Labbé, C., Heckman, M. G., Lorenzo-Betancor, O., Murray, M. E., Ogaki, K., Soto-Ortolaza, A. I., Walton, R. L., Fujioka, S., Koga, S., Uitti, R. J., van Gerpen, J. A., Petersen, R. C., Graff-Radford, N. R., Younkin, S. G., Boeve, B. F., Cheshire, W. P., Low, P. A., Sandroni, P., Coon, E. A.,. . . Ross, O. A. (2016). MAPT haplotype diversity in multiple system atrophy. *Parkinsonism & Related Disorders*, *30*, 40-45. https://doi.org/10.1016/j.parkreldis.2016.06.010

[51] Lantos, P. L., & Papp, M. I. (1994). Cellular pathology of multiple system atrophy : a review. *Journal of Neurology, Neurosurgery & Psychiatry*, *57*(2), 129-133. https://doi.org/10.1136/jnnp.57.2.129

[52] Lee, M. J., Shin, J. H., Seoung, J. K., Lee, J. H., Yoon, U., Oh, J. H., Jung, D. S., & Kim, E. J. (2015). Cognitive impairments associated with morphological changes in cortical and subcortical structures in multiple system atrophy of the cerebellar type. *European Journal of Neurology*, *23*(1), 92-100. https://doi.org/10.1111/ene.12796

[53] Ling, H., Asi, Y. T., Petrovic, I. N., Ahmed, Z., Prashanth, L. K., Hazrati, L. N., Nishizawa, M., Ozawa, T., Lang, A., Lees, A. J., Revesz, T., & Holton, J. L. (2015). Minimal change multiple system atrophy : An aggressive variant ? *Movement Disorders*, *30*(7), 960-967. https://doi.org/10.1002/mds.26220

[54] Marques, O., & Outeiro, T. F. (2012). Alpha-synuclein : from secretion to dysfunction and death. *Cell Death & Disease*, *3*(7), e350. https://doi.org/10.1038/cddis.2012.94

[55] Meissner, W. G., Fernagut, P., Dehay, B., Péran, P., Traon, A. P., Foubert-Samier, A., Lopez Cuina, M., Bezard, E., Tison, F., & Rascol, O. (2019). Multiple System Atrophy : Recent Developments and Future Perspectives. *Movement Disorders*, *34*(11), 1629-1642. https://doi.org/10.1002/mds.27894

[56] Mylius, V., Pee, S., Pape, H., Teepker, M., Stamelou, M., Eggert, K., Lefaucheur, J. P., Oertel, W., & Möller, J. C. (2016). Experimental pain sensitivity in multiple system atrophy and Parkinson's disease at an early stage. *European Journal of Pain*, *20*(8), 1223-1228. https://doi.org/10.1002/ejp.846

[57] Nagaishi, M., Yokoo, H., & Nakazato, Y. (2010). Tau-positive glial cytoplasmic granules in multiple system atrophy. *Neuropathology*, *31*(3), 299-305. https://doi.org/10.1111/j.1440-1789.2010.01159.x

[58] Nee, L. E., Gomez, M. R., Dambrosia, J., Bale, S., Eldridge, R., & Polinsky, R. J. (1991). Environmental— occupational risk factors and familial associations in multiple system atrophy : A preliminary investigation. *Clinical Autonomic Research*, *1*(1), 9-13. https://doi.org/10.1007/bf01826052

[59] Nykjaer, C. H., Brudek, T., Salvesen, L., & Pakkenberg, B. (2017). Changes in the cell population in brain white matter in multiple system atrophy. *Movement Disorders*, *32*(7), 1074-1082. https://doi.org/10.1002/mds.26979

[60] Ozawa, T. (2004). The spectrum of pathological involvement of the striatonigral and olivopontocerebellar systems in multiple system atrophy : clinicopathological correlations. *Brain*, *127*(12), 2657-2671. https://doi.org/10.1093/brain/awh303

[61] Palma, J. A., Norcliffe-Kaufmann, L., & Kaufmann, H. (2018). Diagnosis of multiple system atrophy. *Autonomic Neuroscience*, *211*, 15-25. https://doi.org/10.1016/j.autneu.2017.10.007

[62] Papp, M. I., & Lantos, P. L. (1994). The distribution of oligodendroglial inclusions in multiple system atrophy and its relevance to clinical symptomatology. *Brain*, *117*(2), 235-243. https://doi.org/10.1093/brain/117.2.235

[63] Piao, Y. S., Hayashi, S., Hasegawa, M., Wakabayashi, K., Yamada, M., Yoshimoto, M., Ishikawa, A., Iwatsubo, T., & Takahashi, H. (2001). Co-localization of α-synuclein and phosphorylated tau in neuronal and glial cytoplasmic inclusions in a patient with multiple system atrophy of long duration. *Acta Neuropathologica*, *101*(3), 285-293. https://doi.org/10.1007/s004010000292

[64] Pountney, D. L., Treweek, T. M., Chataway, T., Huang, Y., Chegini, F., Blumbergs, P. C., Raftery, M. J., & Gai, W. P. (2005). αB-Crystallin is a major component of glial cytoplasmic inclusions in multiple system atrophy. *Neurotoxicity Research*, *7*(1-2), 77-85. https://doi.org/10.1007/bf03033778

[65] Procopio, R., Gagliardi, M., Brighina, L., Nicoletti, G., Morelli, M., Ferrarese, C., Annesi, G., & Quattrone, A. (2019). Genetic mutation analysis of the COQ2 gene in Italian patients with multiple system atrophy. *Gene*, *716*, 144037. https://doi.org/10.1016/j.gene.2019.144037

[66] Quinn, N. (1989). Multiple system atrophy--the nature of the beast. *Journal of Neurology, Neurosurgery & Psychiatry*, *52*(Suppl), 78-89. https://doi.org/10.1136/jnnp.52.suppl.78

[67] Quinn, N. (2004). Multiple System Atrophy. Primer on the Autonomic Nervous System, 290-292. doi:10.1016/b978-012589762-4/50079-7

[68] Quinn, N. (2015). A short clinical history of multiple system atrophy. *Clinical Autonomic Research*, *25*(1), 3-7. https://doi.org/10.1007/s10286-014-0265-7

[69] Rana, A. Q., Qureshi, A. R., Siddiqui, O., Sarfraz, Z., Rana, R., & Shtilbans, A. (2018). Prevalence of pain in atypical parkinsonism : a systematic review and meta-analysis. *Journal of Neurology*, *266*(9), 2093-2102. https://doi.org/10.1007/s00415-018-9049-7

[70] Refolo, V., Bez, F., Polissidis, A., Kuzdas-Wood, D., Sturm, E., Kamaratou, M., Poewe, W., Stefanis, L., Angela Cenci, M., Romero-Ramos, M., Wenning, G. K., & Stefanova, N. (2018). Progressive striatonigral degeneration in a transgenic mouse model

of multiple system atrophy : translational implications for interventional therapies. *Acta Neuropathologica Communications*, *6*(1). https://doi.org/10.1186/s40478-017-0504-y

[71] Rehman, H. U. (2001). Multiple system atrophy. *Postgraduate Medical Journal*, *77*(908), 379-382. https://doi.org/10.1136/pmj.77.908.379

[72] Reyes, J. F., Rey, N. L., Bousset, L., Melki, R., Brundin, P., & Angot, E. (2013). Alpha-synuclein transfers from neurons to oligodendrocytes. *Glia*, *62*(3), 387-398. https://doi.org/10.1002/glia.22611

[73] Sakakibara, R. (2021). Gastrointestinal dysfunction in movement disorders. *Neurological Sciences*, *42*(4), 1355-1365. https://doi.org/10.1007/s10072-021-05041-4

[74] Sakakibara, R., Odaka, T., Uchiyama, T., Liu, R., Asahina, M., Yamaguchi, K., Yamaguchi, T., Yamanishi, T., & Hattori, T. (2004). Colonic transit time, sphincter EMG, and rectoanal videomanometry in multiple system atrophy. *Movement Disorders*, *19*(8), 924-929. https://doi.org/10.1002/mds.20165

[75] Sakushima, K., Nishimoto, N., Nojima, M., Matsushima, M., Yabe, I., Sato, N., Mori, M., & Sasaki, H. (2015). Epidemiology of Multiple System Atrophy in Hokkaido, the Northernmost Island of Japan. *The Cerebellum*, *14*(6), 682-687. https://doi.org/10.1007/s12311-015-0668-6

[76] Santangelo, G., Cuoco, S., Picillo, M., Erro, R., Squillante, M., Volpe, G., Cozzolino, A., Cicarelli, G., Barone, P., & Pellecchia, M. T. (2020). Evolution of neuropsychological profile in motor subtypes of multiple system atrophy. *Parkinsonism & Related Disorders*, *70*, 67-73. https://doi.org/10.1016/j.parkreldis.2019.12.010

[77] Sato, K., Kaji, R., Matsumoto, S., Nagahiro, S., & Goto, S. (2007). Compartmental loss of striatal medium spiny neurons in multiple system atrophy of parkinsonian type. *Movement Disorders*, *22*(16), 2365-2370. https://doi.org/10.1002/mds.21732

[78] Schmidt, C., Herting, B., Prieur, S., Junghanns, S., Schweitzer, K., Globas, C., Schöls, L., Reichmann, H., Berg, D., & Ziemssen, T. (2008). Autonomic dysfunction in different subtypes of multiple system atrophy. *Movement Disorders*, *23*(12), 1766-1772. https://doi.org/10.1002/mds.22187

[79] Stanzani-Maserati, M., Gallassi, R., Calandra-Buonaura, G., Alessandria, M., Oppi, F., Poda, R., Sambati, L., Provini, F., & Cortelli, P. (2014). Cognitive and Sleep Features

of Multiple System Atrophy : Review and Prospective Study. *European Neurology*, *72*(5-6), 349-359. https://doi.org/10.1159/000364903

[80] Stefanova, N., & Wenning, G. K. (2016). Review : Multiple system atrophy : emerging targets for interventional therapies. *Neuropathology and Applied Neurobiology*, *42*(1), 20-32. https://doi.org/10.1111/nan.12304

[81] Stefanova, N., Reindl, M., Neumann, M., Haass, C., Poewe, W., Kahle, P. J., & Wenning, G. K. (2005). Oxidative Stress in Transgenic Mice with Oligodendroglial α-Synuclein Overexpression Replicates the Characteristic Neuropathology of Multiple System Atrophy. *The American Journal of Pathology*, *166*(3), 869-876. https://doi.org/10.1016/s0002-9440(10)62307-3

[82] Stemberger, S., Scholz, S. W., Singleton, A. B., & Wenning, G. K. (2011). Genetic players in multiple system atrophy : unfolding the nature of the beast. *Neurobiology of Aging*, *32*(10), 1924.e5-1924.e14. https://doi.org/10.1016/j.neurobiolaging.2011.04.001

[83] Sturm, E., & Stefanova, N. (2014). Multiple System Atrophy : Genetic or Epigenetic ? *Experimental Neurobiology*, *23*(4), 277-291. https://doi.org/10.5607/en.2014.23.4.277

[84] Terni, B., Rey, M. J., Boluda, S., Torrejón-Escribano, B., Sabate, M. P., Calopa, M., van Leeuwen, F. W., & Ferrer, I. (2007). Mutant ubiquitin and p62 immunoreactivity in cases of combined multiple system atrophy and Alzheimer's disease. *Acta Neuropathologica*, *113*(4), 403-416. https://doi.org/10.1007/s00401-006-0192-3

[85] Tu, P. H., Galvin, J. E., Baba, M., Giasson, B., Tomita, T., Leight, S., Nakajo, S., Iwatsubo, T., Trojanowski, J. Q., & Lee, V. M. Y. (1998). Glial cytoplasmic inclusions in white matter oligodendrocytes of multiple system atrophy brains contain insoluble ? -synuclein. *Annals of Neurology*, *44*(3), 415-422. https://doi.org/10.1002/ana.410440324

[86] Ubhi, K., Lee, P. H., Adame, A., Inglis, C., Mante, M., Rockenstein, E., Stefanova, N., Wenning, G. K., & Masliah, E. (2009). Mitochondrial inhibitor 3-nitroproprionic acid enhances oxidative modification of alpha-synuclein in a transgenic mouse model of multiple system atrophy. *Journal of Neuroscience Research*, *87*(12), 2728-2739. https://doi.org/10.1002/jnr.22089

[87] Ubhi, K., Low, P., & Masliah, E. (2011). Multiple system atrophy : a clinical and neuropathological perspective. *Trends in Neurosciences*, *34*(11), 581-590. https://doi.org/10.1016/j.tins.2011.08.003

[88] Valera, E., & Masliah, E. (2018). The neuropathology of multiple system atrophy and its therapeutic implications. *Autonomic Neuroscience*, *211*, 1-6. https://doi.org/10.1016/j.autneu.2017.11.002

[89] Vanacore, N. (2005). Epidemiological evidence on multiple system atrophy. *Journal of Neural Transmission*, *112*(12), 1605-1612. https://doi.org/10.1007/s00702-005-0380-7

[90] Vanacore, N., Bonifati, V., Fabbrini, G., Colosimo, C., de Michele, G., Marconi, R., Nicholl, D., Locuratolo, N., Talarico, G., Romano, S., Stocchi, F., Bonuccelli, U., de Mari, M., Vieregge, P., & Meco, G. (2001). Epidemiology of multiple system atrophy. *Neurological Sciences*, *22*(1), 97-99. https://doi.org/10.1007/s100720170064

[91] Vidal, J. S., Vidailhet, M., Elbaz, A., Derkinderen, P., Tzourio, C., & Alpérovitch, A. (2008). Risk factors of multiple system atrophy : A case-control study in French patients. *Movement Disorders*, *23*(6), 797-803. https://doi.org/10.1002/mds.21857

[92] Vieira, B. D. M., Radford, R. A., Chung, R. S., Guillemin, G. J., & Pountney, D. L. (2015). Neuroinflammation in Multiple System Atrophy : Response to and Cause of α-Synuclein Aggregation. *Frontiers in Cellular Neuroscience*, *9*. https://doi.org/10.3389/fncel.2015.00437

[93] Wakabayashi, K., & Takahashi, H. (2006). Cellular pathology in multiple system atrophy. *Neuropathology*, *26*(4), 338-345. https://doi.org/10.1111/j.1440-1789.2006.00713.x

[94] Wakabayashi, K., Ikeuchi, T., Ishikawa, A., & Takahashi, H. (1998). Multiple system atrophy with severe involvement of the motor cortical areas and cerebral white matter. *Journal of the Neurological Sciences*, *156*(1), 114-117. https://doi.org/10.1016/s0022-510x(98)00018-5

[95] Wakabayashi, K., Mori, F., Nishie, M., Oyama, Y., Kurihara, A., Yoshimoto, M., & Kuroda, N. (2005). An autopsy case of early ("minimal change") olivopontocerebellar atrophy (multiple system atrophy-cerebellar). *Acta Neuropathologica*, *110*(2), 185-190. https://doi.org/10.1007/s00401-005-1029-1

[96] Wenning, G. K., & Fanciulli, A. (2016b). *Multiple System Atrophy* (Softcover reprint of the original 1st ed. 2014 éd.). Springer.

[97] Wenning, G. K., & Stefanova, N. (2009). Recent developments in multiple system atrophy. *Journal of Neurology*, *256*(11), 1791-1808. https://doi.org/10.1007/s00415-009-5173-8

[98] Wenning, G. K., Colosimo, C., Geser, F., & Poewe, W. (2004). Multiple system atrophy. *The Lancet Neurology*, *3*(2), 93-103. https://doi.org/10.1016/s1474-4422(03)00662-8

[99] Wenning, G. K., Geser, F., & Poewe, W. (2005). Therapeutic strategies in multiple system atrophy. *Movement Disorders*, *20*(S12), S67-S76. https://doi.org/10.1002/mds.20543

[100] Wenning, G. K., Stefanova, N., Jellinger, K. A., Poewe, W., & Schlossmacher, M. G. (2008). Multiple system atrophy : A primary oligodendrogliopathy. *Annals of Neurology*, *64*(3), 239-246. https://doi.org/10.1002/ana.21465

[101] Wenning, G. K., Tison, F., Elliott, L., Quinn, N. P., & Daniel, S. E. (1996). Olivopontocerebellar pathology in multiple system atrophy. *Movement Disorders*, *11*(2), 157-162. https://doi.org/10.1002/mds.870110207

[102] Wenning, G., Gilman, S., & Seppi, K. (2008). Second consensus statement on the diagnosis of multiple system atrophy. *Aktuelle Neurologie*, *35*(S 01). https://doi.org/10.1055/s-0028-1086654

[103] Wong, J. H., Halliday, G. M., & Kim, W. S. (2014). Exploring Myelin Dysfunction in Multiple System Atrophy. *Experimental Neurobiology*, *23*(4), 337-344. https://doi.org/10.5607/en.2014.23.4.337

[104] Wullner, U., Schmitt, I., Kammal, M., Kretzschmar, H. A., & Neumann, M. (2008). Definite multiple system atrophy in a German family. *Journal of Neurology, Neurosurgery & Psychiatry*, *80*(4), 449-450. https://doi.org/10.1136/jnnp.2008.158949

[105] Yoshida, M. (2007). Multiple system atrophy : α-synuclein and neuronal degeneration. *Neuropathology*, *27*(5), 484-493. https://doi.org/10.1111/j.1440-1789.2007.00841.x

[106] Zhang, L. Y., Cao, B., Zou, Y. T., Wei, Q. Q., Ou, R. W., Zhao, B., Wu, Y., & Shang, H. F. (2017). Depression and anxiety in multiple system atrophy. *Acta Neurologica Scandinavica*, *137*(1), 33-37. https://doi.org/10.1111/ane.12804

[107] Zhang, L., Cao, B., Wei, Q. Q., Ou, R., Zhao, B., Yang, J., Wu, Y., & Shang, H. (2021). Pathological laughter and crying in multiple system atrophy with different subtypes : Frequency and related factors. *Journal of Affective Disorders*, *283*, 60-65. https://doi.org/10.1016/j.jad.2020.12.096

[108] Zhao, Q., Yang, X., Tian, S., An, R., Zheng, J., & Xu, Y. (2015). Association of the COQ2 V393A variant with risk of multiple system atrophy in East Asians : a case–control study and meta-analysis of the literature. *Neurological Sciences*, *37*(3), 423-430. https://doi.org/10.1007/s10072-015-2414-8

[109] Zhou, L., Jiang, Y., Zhu, C., Ma, L., Huang, Q., & Chen, X. (2016). Oxidative Stress and Environmental Exposures are Associated with Multiple System Atrophy in Chinese Patients. *Canadian Journal of Neurological Sciences / Journal Canadien des Sciences Neurologiques*, *43*(5), 703-709. https://doi.org/10.1017/cjn.2016.261