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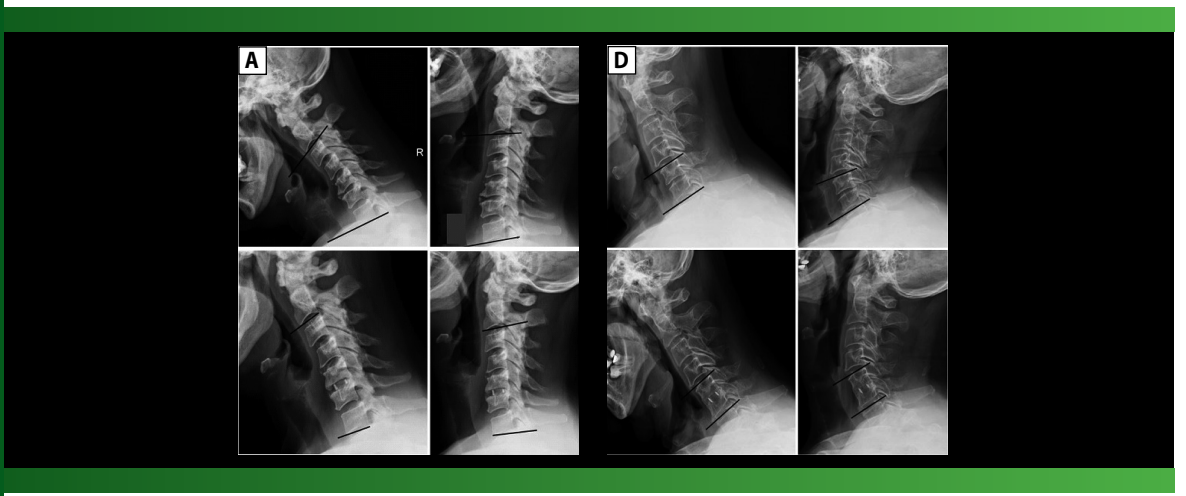

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Cover photo: Gabriela Zapolska et al., Biomechanical evaluation of single- and multi-level anterior cervical discectomy and fusion with polyetheretherketone cages: radiological and clinical outcomes, see figure on page 360.

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Polish Journal of Neurology and Neurosurgery
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Searching for tics

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ABSTRACT

Introduction. In the current edition, Szejko and colleagues describe a subset of patients with Gilles de la Tourette syndrome (GTS) who had dystonic tics (DTs), which occurred more frequently in those with a greater number of tics and likely contribute to impairment.

Clinical reflections. DTs manifest as an abnormal posture that may be difficult to distinguish from other movements, such as dystonia and other tic types. Electromyography is an invaluable tool that can aid clinicians in making this important distinction.

Clinical implications. Accurately diagnosing these movements can significantly impact treatment decisions and contribute to more homogenous research populations.

(*Neurol Neurochir Pol 2019; 53 (5): 315–316*)

Gilles de la Tourette syndrome (GTS) is diagnosed in individuals who have two or more motor tics and at least one phonic tic for at least one year's duration beginning before age 18 [1]. Patients with GTS often have comorbid psychiatric problems that, coupled with tics, can lead to substantial impairment. Research has sought to characterize the phenotypic variability among those with GTS in order to better understand disease mechanisms and identify elements that have the greatest impact on quality of life. Accomplishing these goals will allow clinicians to more effectively tailor treatment to an individual's specific needs. Szejko *et al.* have nicely contributed to this effort and found that 73.9% of patients with GTS developed dystonic tics (DTs) [2]. They also provided a thorough characterization of their cohort of 153 patients with DTs. Their attention to detail highlights the importance of accurately differentiating tics from other movement phenomena, taking an account of one's complete tic repertoire, and correctly identifying types of tics, all of which can have a significant impact on treatment.

Tics are stereotypical movements sometimes described as semivoluntary or unvoluntary to highlight that they are not truly involuntary [3] unlike some other movement phenomena, such as dystonia, which is an involuntary sustained or intermittent co-contraction of muscle agonist and antagonists resulting in abnormal movements and/or postures [4]. Simple tics often have a jerk-like appearance and may be differentiated from other jerking movements by their serotyped nature and other attributes, including a premonitory urge that resolves when the tic is completed. Szejko et al. provide a nice table summarizing characteristics to aid in differentiating tics from dystonia [2]; however, there are limitations to an approach based solely on clinical history and observation as illustrated by the fact that 90% of adults and only 37% of children endorse a hallmark premonitory urge [5, 6]. Moreover, simple motor tics may be misidentified as myoclonus thus leading the clinician to misdiagnosis. EMG is a useful technique that can aid in differentiation among these [7] and should be considered an integral extension of the neurological examination. Measuring burst duration enables one to categorize tics as clonic (< 100ms) or dystonic (> 300ms) [8]. This is a distinction that may escape the sensitivity of clinical observation and only consistently accomplished with EMG.

Clinicians should be aware that individuals with GTS may have hyperkinetic movements in addition to tics, such as tardive chorea or tardive dystonia [9]. Accurately identifying these movements, which may mimic tics, can have a significant impact on treatment decisions, such as whether to increase or decrease the dosage of a dopamine antagonist like aripirazole, which is commonly used to treat tics [10]. The wrong decision could have serious and potentially irreversible consequences given the association between these medications and extrapyramidal side effects. Once an accurate and specific diagnosis of tics is established, appropriate treatments can be pursued. The American

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Academy of Neurology recently published new guidelines for the treatment of tic disorders [11]. Comprehensive Behavioral Intervention for Tics (CBIT) is the mainstay of the nonpharmacological treatment arm while pharmacological options include α-adrenergic agonists, antipsychotics, vesicular monoamine transporter-2 inhibitors, and botulinum toxin injections, the last of which has particular applicability to those with dystonic tics [12]. Botulinum toxin may also be used to treat laryngeal tics [13] and has even been shown to reduce the premonitory urge [14].

To effectively treat their patients, clinicians must compile a patient's tic inventory and determine their impact on one's life. This can be accomplished with various rating scales, such as the Yale Global Tic Severity Scale (YGTSS) [15, 16]. Szejko and colleagues made several interesting observations including the average age of onset of DTs 3.7 years after development of their first tic suggesting that most individuals do not present with DTs [2]. They also showed that the presence of DTs was more common in those with more tics overall. Previous reports have shown co-occurance of tics and dystonia [17] as well as cosegregation of eye-winking tics, frequent eye-blinking and blepharosm with a family [18, 19], raising the question of a shared mechanism. This idea has since been strengthened by identification of a shared mutation within the guanosine triphosphate cyclohydrolase I gene (*GCHI*) in a Danish family with dopa-responsive dystonia and GTS [20].

While the first step in evaluating movement disorders is accurately identifying the movement phenomenon, clinicians must be aware that not all phenomena are 'pure,' i.e. tremor or tics, but can have characteristics of multiple movement phenomena simultaneously, e.g. DTs. Differentiating tics from dystonia as well as correctly identifying the type of tic is necessary for clinicians to provide appropriate treatment options, properly counsel their patients, and to better homogenize patient populations so that higher quality research studies may be conducted.

Conflict of interests / Funding: *None declared.*

Abbreviations

GTS — Gilles de la Tourette syndrome
DTs — Dystonic tics
Cts — Clonic tics
CBIT — Comprehensive Behavioral Intervention for Tics
YGTSS — Yale Global Tic Severity Scale

References

1. American Psychiatric Association. Diagnostic and statistical manual of mental disorders, 5th ed (DSM-5). Washington, DC: American Psychiatric Association. ; 2013.
2. Szejko N, Jakubczyk A, Dunalska A, et al. Dystonic tics in patients with Gilles de la Tourette syndrome. Neurol Neurochir Pol. 2019; 53(5): xxxx
3. Fahn S, Jankovic J, Hallett M. Psychogenic movement disorders. Principles and Practice of Movement Disorders [Chapter 16: Tics and Tourette syndrome]. 2011: 350–379, doi: 10.1016/b978-1-4377-2369-4.00025-1.
4. Albanese A, Bhatia K, Bressman SB, et al. Phenomenology and classification of dystonia: a consensus update. Mov Disord. 2013; 28(7): 863–873, doi: 10.1002/mds.25475, indexed in Pubmed: 23649720.
5. Kwak C, Dat Vuong K, Jankovic J. Premonitory sensory phenomenon in Tourette's syndrome. Mov Disord. 2003; 18(12): 1530–1533, doi: 10.1002/mds.10618, indexed in Pubmed: 14673893.
6. Banaschewski T, Woerner W, Rothenberger A. Premonitory sensory phenomena and suppressibility of tics in Tourette syndrome: developmental aspects in children and adolescents. Dev Med Child Neurol. 2003; 45(10): 700–703, doi: 10.1017/s0012162203001294, indexed in Pubmed: 14515942.
7. Panyakaew P, Cho HJ, Hallett M. Clinical Neurophysiological Evaluation for Simple Motor Tics. Clin Neurophysiol Pract. 2016; 1: 33–37, doi: 10.1016/j.cnp.2016.04.001, indexed in Pubmed: 27777987.
8. Jankovic J, Stone L. Dystonic tics in patients with Tourette's syndrome. Mov Disord. 1991; 6(3): 248–252, doi: 10.1002/mds.870060309, indexed in Pubmed: 1922130.
9. Kompoliti K, Goetz CG. Hyperkinetic movement disorders misdiagnosed as tics in Gilles de la Tourette syndrome. Mov Disord. 1998; 13(3): 477–480, doi: 10.1002/mds.870130317, indexed in Pubmed: 9613740.
10. Janik P, Szejko N. Aripiprazole in treatment of Gilles de la Tourette syndrome - New therapeutic option. Neurol Neurochir Pol. 2018; 52(1): 84–87, doi: 10.1016/j.pjnns.2017.10.015, indexed in Pubmed: 29154107.
11. Pringsheim T, Okun MS, Müller-Vahl K, et al. Practice guideline recommendations summary: Treatment of tics in people with Tourette syndrome and chronic tic disorders. Neurology. 2019; 92(19): 896–906, doi: 10.1212/WNL.0000000000007466, indexed in Pubmed: 31061208.
12. Jankovic J. Botulinum toxin in the treatment of dystonic tics. Mov Disord. 1994; 9(3): 347–349, doi: 10.1002/mds.870090315, indexed in Pubmed: 8041378.
13. Vincent DA. Botulinum toxin in the management of laryngeal tics. J Voice. 2008; 22(2): 251–256, doi: 10.1016/j.jvoice.2006.08.014, indexed in Pubmed: 17056228.
14. Kwak CH, Hanna PA, Jankovic J. Botulinum toxin in the treatment of tics. Arch Neurol. 2000; 57(8): 1190–1193, doi: 10.1001/archneur.57.8.1190, indexed in Pubmed: 10927800.
15. Leckman JF, Riddle MA, Hardin MT, et al. The Yale Global Tic Severity Scale: initial testing of a clinician-rated scale of tic severity. J Am Acad Child Adolesc Psychiatry. 1989; 28(4): 566–573, doi: 10.1097/00004583-198907000-00015, indexed in Pubmed: 2768151.
16. Martino D, Pringsheim TM, Cavanna AE, et al. Members of the MDS Committee on Rating Scales Development. Systematic review of severity scales and screening instruments for tics: Critique and recommendations. Mov Disord. 2017; 32(3): 467–473, doi: 10.1002/mds.26891, indexed in Pubmed: 28071825.
17. Stone LA, Jankovic J. The coexistence of tics and dystonia. Arch Neurol. 1991; 48(8): 862–865, doi: 10.1001/archneur.1991.00530200104028, indexed in Pubmed: 1898264.
18. Elston JS, Granje FC, Lees AJ. The relationship between eye-winking tics, frequent eye-blinking and blepharospasm. J Neurol Neurosurg Psychiatry. 1989; 52(4): 477–480, doi: 10.1136/jnnp.52.4.477, indexed in Pubmed: 2738590.
19. Németh AH, Mills KR, Elston JS, et al. Do the same genes predispose to Gilles de la Tourette syndrome and dystonia? Report of a new family and review of the literature. Mov Disord. 1999; 14(5): 826–831, indexed in Pubmed: 10495045.
20. Romstad A, Dupont E, Krag-Olsen B, et al. Dopa-responsive dystonia and Tourette syndrome in a large Danish family. Arch Neurol. 2003; 60(4): 618–622, doi: 10.1001/archneur.60.4.618, indexed in Pubmed: 12707079.

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Novel emerging treatments for NMOSD

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ABSTRACT

Neuromyelitis optica spectrum disorders (NMOSD) are inflammatory demyelinating diseases of the central nervous system (CNS) that cause optic neuritis, transverse myelitis, and some other CNS syndromes.

Recently, diagnosis and understanding of these diseases has been markedly enhanced by the discovery that serum autoantibodies that target aquaporin-4 (AQP4) are strongly associated with the disease. This spectrum includes also a potential subset of patients with a phenotype of NMOSD who have anti-myelin oligodendrocyte glycoprotein (MOG) antibody. Although steroids and immunosuppressive drugs have been widely used for NMOSD treatment, until recently there was no approved therapy for these diseases. With improved understanding of the pathophysiology of NMOSD, numerous new therapeutic strategies have recently been evaluated. The results of these studies, involving monoclonal antibodies (mAbs) inhibiting terminal complement protein cleavage interfering with interleukin-6 receptor (IL-6 R) signaling and depleting CD19-positive B cells, have been published in recent months. All of these new therapeutics have shown a high degree of efficacy in diminishing NMOSD activity and inhibiting disability progression. At the same time, all these mAbs have demonstrated favorable safety and tolerability profiles, with a limited rate of adverse events. The first of these new drugs, eculizumab, have been approved in USA and Europe for NMOSD treatment within the last couple of months and it is expected that the other novel, effective and safe treatments for NMOSD will be approved in the near future.

Key words: neuromyelitis optica spectrum disorders, monoclonal antibodies, demyelinating diseases

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Introduction

Neuromyelitis optica spectrum disorders (NMOSD) are autoimmune inflammatory disorders of the central nervous system (CNS) characterised by bilateral or rapidly sequential optic neuritis and/or transverse myelitis. Other suggestive presentations include episodes of brainstem symptoms, in particular area postrema clinical syndrome. The disease can result in severe muscle weakness and paralysis, loss of vision, sensory loss, bladder dysfunction, neuropathic pain, and in the most severe cases respiratory failure [1]. The prevalence of NMOSD in Caucasians is relatively low at 0.5–10 per 100,000, but severe and frequently rapid impairment observed in patients can lead to an unfavourable prognosis, including death.

The mechanism of NMOSD is associated with the presence in the serum of patients' IgG1 antibodies against aquaporin-4 (AQP4 Ab), the major water channel present within the CNS specifically on astrocyte endfeets at the blood-brain-barrier (BBB) [2]. This antibody can be found in more than 80% of patients. The discovery of AQP4 Ab has contributed significantly to our understanding of the pathology of NMOSD, also allowing for a much more precise diagnosis. AQP4 Ab appears to have a pathogenic role in the mechanism of NMOSD [3]. Recombinant AQP4 Ab after passive transfer in rats has induced NMOSD-specific immunopathology [4]. Serum AQP4 Ab titers have been shown to correlate with clinical attacks and with the extent of spinal cord lesions on MRI [5, 6]. In addition, serum AQP4 Ab titers have been shown to drop after immunosuppressive treatment, and to remain low during remissions [7]. This antibody, produced by T-helper dependent B cells from the peripheral immune compartment, crosses the blood-brain-barrier (BBB) and interacts with AQP4 on astrocyte endfeet leading to astrocyte oedema and dysfunction. Subsequent inflammation involves complement activation, increased BBB permeability, and a massive influx of neutrophils and eosinophils [8]. Thus, inflammatory lesions in NMOSD are clearly different from CNS inflammation in multiple sclerosis (MS), where T and B cells constitute the majority of invading

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cells. These findings, demonstrating significant mechanistic differences between MS and NMOSD, have allowed for a definitive separation of these two clinical entities.

More recently, another antigen was found to be associated with AQP4 seronegative NMOSD. Antibodies against myelin oligodendrocyte glycoprotein (MOG) were detected in 4–11% of seronegative patients [9]. Unlike AQP4 Ab, anti-MOG Ab did not induce astrocytic pathology. It was found that anti-MOG Ab led to demyelination with limited immune cell infiltration [10]. Anti-MOG Ab almost never co-exists with AQP4 Ab and occurs much more frequently in children than in adult patients.

Despite the discovery of AQP4 Ab and anti-MOG Ab, there are still patients who meet the clinical diagnostic criteria of NMOSD, but in whose sera the two Abs cannot be detected. The prerequisite of a NMOSD diagnosis requires the presence of optic nerve and spinal cord symptoms [11]. The current 2015 International Consensus Diagnostic Criteria for NMOSD are stratified according to the presence of AQP4 Ab. For seropositive patients, they require the presence of at least one of the core manifestations, whereas for seronegative patients two manifestations including optic neuritis, myelitis or area postrema syndrome are required (Tab. 1). Recently, preliminary results have suggested the role of antibodies against glial fibrillary acidic protein (GFAP) in seronegative NMOSD [12].

Current treatment of NMOSD

Until recently, there was no approved treatment for NMOSD, and patients were restricted to off-label therapies bringing uncertain benefits. Based on its clinical course, NMOSD therapy can be divided into relapse treatment and preventive treatment.

Relapse treatment

For relapse, patients are usually treated with pulsed steroid therapy. Methylprednisolone at a dose of 1g is given intravenously (IV) for 3–5 days but therapy can be extended in a very severe relapse [13]. Relapses that respond poorly to methylprednisolone can be treated with plasma exchange (PLEX), usually 5–7 procedures every other day. PLEX can also be used as a first line therapy for NMOSD relapse [14]. Since the progression of disability in NMOSD is mainly driven by relapses, it is critically important that NMOSD relapse is treated as early as possible and with the most effective procedure.

Shortly after steroid pulsed therapy or PLEX, patients usually are started with immunosuppressive therapies. To protect patients from disease progression before the immunosuppressive drugs start to work, prednisone or prednisolone can be given orally for up to six months. Relapses in NMOSD dependent on anti-MOG Ab usually respond better to steroid treatment than in AQP4 dependent disease [15]. Limited data indicates that intravenous immunoglobulins (IvIg) might show a benefit in NMOSD relapse treatment**.** In one study, the effectiveness of IvIg in the treatment of acute relapses in NMOSD was assessed in a retrospective review of 10 patients unresponsive to treatment with steroids with or without PLEX [16]. Improvement was noted in five of 11 (45.5%) relapses, and the remaining relapses had no further worsening. The study concluded that IvIg may have a role in treating acute NMOSD relapses.

Immunosuppressive treatment

Based on the autoimmune mechanism of NMOSD involving the production of auto-antibodies against AQP4 and MOG, several immunosuppressive agents have been tested in this disease over the past 30 years. Unfortunately, on most occasions the studies only involved a small number of patients and were designed as open label trials. None of these drugs have been formally approved for NMOSD treatment.

Traditionally, Azathioprine (AZT) was widely used as a first line NMOSD treatment. AZT was recommended as

Table 1. IPND 2015 diagnostic criteria for NMOSD

NMOSD with AQP4-Ab
At least one core clinical characteristic plus positive test for AQP4-IgG using best available detection method*
Exclusion of alternative diagnoses
NMOSD without AQP4-Ab
At least two core clinical characteristics occurring as a result of one or more clinical attacks and meeting all of the following requirements:
1. At least one core clinical characteristic must be optic neuritis, acute myelitis with LETM, or area postrema syndrome.
2. Dissemination in space (two or more different core clinical characteristics).
3. Additional MRI requirements, as applicable.
4. Negative tests for AQP4-IgG using best available detection method* or testing unavailable.
5. Exclusion of alternative diagnoses.
Core clinical characteristics: Optic neuritis; acute myelitis; area postrema syndrome (hiccups, nausea and vomiting); acute brainstem syndrome; symptomatic narcolepsy or acute diencephalic clinical syndrome with NMOSD-typical diencephalic MRI lesions; symptomatic cerebral syndrome with NMOSD-typical brain lesions.

*AQP4-IgG serology: cell-based assay is strongly recommended

a first line preventive treatment by the EFNS panel on guidelines on diagnosis and management of NMO [17]. Several small studies have tested the efficacy of AZT alone, or in combination with steroids [18]. Some of these studies showed an approximately 70% reduction in the NMOSD relapse rate and reduced disability after several years of treatment. In a large retrospective review of the medical records of 103 AQP-4 antibody-positive NMOSD patients, 89% (n = 92) had reduced their median annualised relapse rates from 1.5 (IQR 0.6–4.0) to 0 (IQR 0–0.27, $p < 0.00005$) during treatment. Sixty-one per cent (n = 63) remained relapse-free at a median follow-up of 18 months. Neurological function improved or stabilised in 78%. However, the discontinuation rate was 47%, reflecting the poor tolerability of this drug [19].

The other problem with AZT treatment in NMOSD is the delayed mechanism of action of this drug. AZT on average requires 3–6 months to demonstrate its effect on the immune system. Mitoxantrone [20], cyclophosphamide [21] and some other immunosuppressive treatments including methotrexate have demonstrated beneficial effects in NMOSD only in case reports and only in a limited number of patients. In addition, safety profiles of these drugs reduce the enthusiasm for their use in NMOSD. Little more data is available regarding Mycophenolate Mofetil (MMF). In a prospective study including 67 NMOSD seropositive and seronegative patients, MMF reduced the annual relapse rate by 49% and stabilised EDSS in 80% of patients within a 24 month observation period. There was no difference between seropositive and seronegative patients [22]. Several retrospective studies have assessed the efficacy of MMF in AQP4- and MOG- seropositive and double negative patients. In one of these studies [23], the median post-MMF annualised relapse rate was significantly lower than the pre-MMF annualised relapse rate (0.0 *vs* 1.5; $p < 0.001$). EDSS scores also significantly decreased after MMF treatment (3.0 *vs* 2.5; $p = 0.005$). Thirty-five patients (60%) were relapse-free with a median treatment duration of 20 months, and EDSS scores were stabilised or improved in 53 patients (91%). In a similar way to AZT, MMF requires an extended period of time to demonstrate its effect on the immune system, and a substantial number of patients discontinued treatment due to side effects.

Intravenous immunoglobulins

In recent years, intravenous immunoglobulins (IvIg), which have been proven to be effective in some other antibody-mediated autoimmune conditions including inflammatory demyelinating polyneuropathies [24], have attracted significant attention in terms of NMOSD treatment. IvIg have been shown to induce diminished activity on membrane-damaging components of the complement system, B cell activating factor (BAFF), and several other immune mechanisms including interference with antigen recognition, downregulation of cytokine secretion, adhesion molecules expression, and suppression of T-cell activation relevant to NMOSD [25]. Although popular views and practical experience are strong advocates for the use of IvIg in NMOSD, there is relatively little evidence to support these conclusions.

The first study to look into the role of IvIg treatment in NMOSD included eight patients [26]. Five experienced relapsing optic neuritis with or without myelitis, and the remaining three had relapsing longitudinal extensive transverse myelitis (LETM). After a total of 83 infusions (4–21 per patient) and a mean follow-up duration of 19.3 months (6–39 months), it was observed that the mean relapse rate had decreased from 1.8 in the 12 months pre-IvIg therapy to 0.006 during follow-up ($p = 0.0001$), while the mean EDSS score had declined from 3.3 ± 1.3 to 2.6 ± 1.5 ($p = 0.04$). In another study [27], the use of IvIg was evaluated in preventing relapses in patients with NMOSD. Six NMOSD patients who were treated with an IvIg induction dose followed by infusions every 2-3 months were retrospectively analysed. ARR and EDSS pre- and post-IvIg were recorded. The median number of relapses and the median ARR were significantly reduced (8.0 to 1.0 and 0.75 to 0.15; $p < 0.05$) during IvIg treatment. EDSS remained the same during four years of treatment.

Cell depletion therapy

The discovery of a pathological role of autoantibodies against AQP4 and MOG in NMOSD has prompted the application of B cell depletion therapy which might help to eliminate antibody-producing cells and improve treatment of this disease. Rituximab (RTX), a chimeric monoclonal antibody against CD20, a pan-B cell surface marker, has been tested in NMOSD in several small or medium-sized open labelled studies [28, 29]. RTX very efficiently depleted B cells over a duration of 6–9 months. Importantly, the cells from early stages of B cell lineage development and plasma cells were preserved, securing immune homeostasis during RTX treatment.

Most of the RTX studies have shown its profound beneficial effect on ARR and on stabilisation and reduction of disability measured with EDSS. Some of these studies have even shown complete suppression of relapses over a 2–3 year period. A recently published meta-analysis analysed 26 studies, in which differences in the ARR ratio and EDSS score before and after RTX therapy were used as the main efficacy measures [30]. This meta-analysis involved 577 patients. Antibodies against aquaporin-4 were present in 435 (75.39%) patients. The findings suggested diminished mean ARR ratio after rituximab therapy by 1.56 (95% CI, -1.82 to -1.29). No significant correlation was detected between the outcome of ARR ratio change and the following variables: age at onset, duration of disease, follow-up time, dose of infusion or AQP4-IgG serostatus. The findings of this meta-analysis disclosed also a reduction in the mean EDSS score by -1.16 (95% CI, -1.36 to -0.96) during RTX treatment. A total of 330 out of 528 patients (62.9%) achieved relapse-free status. RTX showed acceptable tolerance, and there were no serious safety issues in NMOSD patients treated with RTX. All of these findings have led to increased off label use of RTX in NMOSD in recent years.

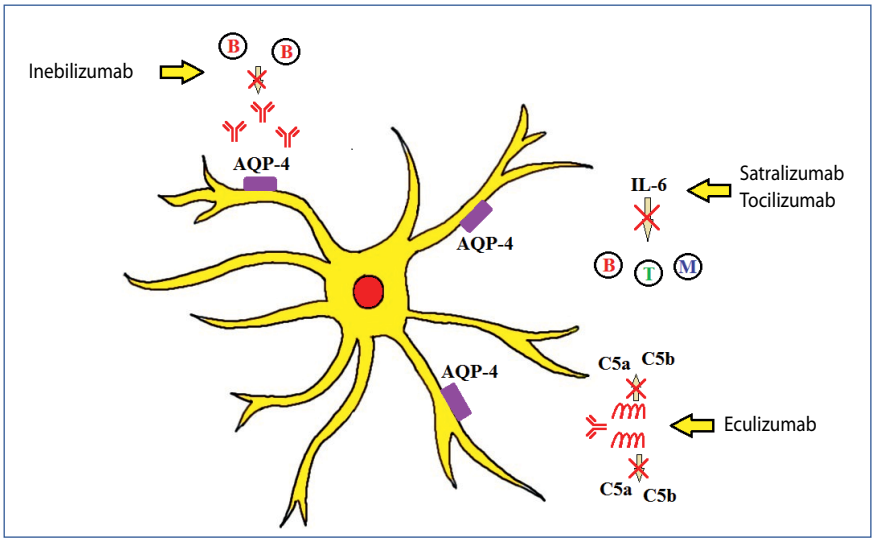

Figure 1. Emerging treatments of NMO

Novel emerging treatments of NMOSD

Progress in the understanding of NMOSD pathologic mechanisms has led to unprecedented attempts to intensify research into the development of new therapies in this disease in recent years. New therapeutic strategies have emerged which involve targeting novel molecules believed to be operating in the pathomechanism of NMOSD. These new strategies include the prevention of complement activation, interfering with IL6 receptor (IL6-R) signalling, and the depletion of AQP-4 and MOG antibodies producing cells (Fig. 1). More importantly, these new strategies were tested in studies designed as double blinded, randomised and controlled phase 2 and 3 trials. Within the last few months, very encouraging results of these studies have become available and have generated renewed hope of combatting this devastating disease. It is expected that the results of these recent studies will allow for the formal approval of all of these therapies for NMOSD in the near future.

Eculizumab

Eculizumab is a humanised monoclonal antibody which inhibits the terminal complement protein C5 and prevents its cleavage into C5a and C5b fragments [31]. Preclinical data indicates that AQP4 Ab triggers the complement cascade [32], which leads to inflammation and the formation of a membrane attack complex. The membrane attack complex is implicated in astrocyte destruction and neuronal injury. C5a expresses proinflammatory activity, and C5b induces the formation of a membrane attack complex. Experimental studies have shown that complement inhibitor efficiently suppresses NMOSD development [33]. Eculizumab has been approved in the USA and Europe for paroxysmal nocturnal haemoglobinuria, atypical haemolytic uraemic syndrome, and myasthenia gravis.

In recently published results of a phase 3, randomised, double-blind, placebo-controlled, time-to-event trial (PREVENT) it was shown that eculizumab significantly suppressed disease activity in 143 AQP4-Ab-positive patients with a moderate to severe course of the disease [34]. Patients were randomised 2:1 to either treatment with eculizumab or a placebo. The drug was administered intravenously at a dose of 900 mg weekly for the first four doses; subsequently patients received a maintenance regimen of 1,200 mg every two weeks until relapse or until the end of the trial. Immunosuppressive treatment used prior to study entry was allowed (with the exception of rituximab) and the group of immunosuppressive concomitant treatment was 108 patients out of the total group of 143. The primary efficacy endpoint was the first adjudicated relapse. The primary endpoint occurred much less often in the eculizumab group (3 of 96 patients – 3%) than in the placebo group (20 of 47 – 43%) (HR, 0.06; 95% CI, 0.02 to 0.20; $p < 0.001$). At 48 weeks, 97.9% of patients receiving eculizumab were relapse-free, compared to 63.2% of patients receiving the placebo. Symptoms of most of the relapses were related to myelitis. Eculizumab was associated with a lower adjudicated annualised relapse rate after adjustment than the placebo, which represented the first secondary end-point, 0.02 and 0.35, respectively ($p < 0.001$). No conclusions could be made regarding the remaining secondary endpoints because the difference between groups for the next endpoint in the hierarchy, which was a change in the EDSS score, was not significant. The lack of between-group differences in disability progression resulted from the trial design precluding follow-up beyond six weeks after a single relapse. This implied that there were no benefits of the drug on disability progression during the short period of the trial. Patients from the eculizumab group had higher rates of upper respiratory tract infection and headache than did patients in

the placebo group: 31 versus 19 events of upper respiratory tract infection per 100 patient-years, and 55 versus 38 headache events per 100 patient-years. One patient in the eculizumab group who was receiving concomitant azathioprine died from pulmonary empyema. Eculizumab increases the risk of meningococcal and encapsulated bacterial infection [35], and all patients received meningococcal vaccination prior to study entry. No cases of meningococcal infection were reported during the trial.

Eculizumab (Soliris) was approved by FDA for NMOSD treatment following an expedited six-month priority review in June 2019. EMA approved Eculizumab for treatment of AQP4-Ab-positive patients with relapsing course of the disease in August 2019.

Tocilizumab

Tocilizumab was the first anti-IL6-R mAb tested in the treatment of NMOSD. The rationale for using tocilizumab in NMO was related to the potential role of IL-6 in the mechanism of NMOSD [36]. This drug has been already used for the treatment of severe rheumatic arthritis patients.

Tocilizumab has been tested in NMOSD in several small open label studies and case reports [37, 38]. These small series have demonstrated a beneficial effect in NMOSD. In one study [39], eight female patients with highly active AQP4-Ab-seropositive NMOSD (n = 6) and NMOSD patients (n = 2) whose disease had been resistant to previous medications including B-cell depletion, were switched to tocilizumab (6–8 mg/kg of body weight per dose). The patients were followed up for 30.9 months after switching to tocilizumab. Two of the eight patients received add-on therapy consisting of monthly corticosteroid pulses or azathioprine. During tocilizumab treatment, the median annualised relapse rate significantly decreased from 4.0 in the year before tocilizumab therapy to 0.4 (p = 0.008), and the median EDSS score significantly decreased, from 7.3 to 5.5 (p = 0.03). Active magnetic resonance imaging lesions were seen in 6/8 patients at tocilizumab initiation and in 1/8 patients at the last magnetic resonance imaging. Three patients remained relapse-free during tocilizumab treatment. The AQP4-Ab titers (p = 0.02) and pain levels (p = 0.02) dropped significantly during tocilizumab treatment. Adverse effects included moderate cholesterol elevation in 6/8 patients, infections in 4/8, deep venous thrombosis in 1/8, and neutropenia in 1/8.

Tocilizumab development for NMOSD treatment has been delayed since another anti-IL6-R mAb, satralizumab, was designed to improve pharmacokinetics of IL6-R binding and provide better efficacy (see below). However, at the recent ECTRIMS meeting in Stockholm, the results were presented of an investigator-initiated study (Tango) in China [40]. This was a randomised, open-label, parallel-group study comparing tocilizumab with azathioprine. Patients (n = 118) were randomly assigned 1:1 to receive 8 mg/kg intravenous tocilizumab monthly or 2–3 mg/kg oral azathioprine daily. Treatment was administered in conjunction with a gradual discontinuation of the previous treatments, followed by monotherapy for 12 months; 85% of patients were seropositive for AQP-4. The primary endpoint was the time to first relapse. After a mean observation period of 48 weeks, the percentages of relapse-free patients were 91.5% in the tocilizumab group and 67.8% in the azathioprine group (HR = 0.32, 95% CI 0.14–0.70, p = 0.004). Sustained reduction in disability was more likely among patients treated with tocilizumab than in patients with azathioprine (HR = 0.34, 95% CI 0.13–0.90, p = 0.03). Serum levels of anti-AQP4-ab were reduced significantly, by 42% with tocilizumab compared to 15% with azathioprine (p = 0.03). In the azathioprine group, there was a higher frequency of lymphopenia (46% *vs* 7%) and anaemia (27% *vs* 41%) than in the tocilizumab group. During the Tango study, two patients died (one in each arm): in tocilizumab because of a severe recurrence of NMOSD, and in azathioprine because of meningoencephalitis. In both groups, there was an increased number of patients with elevated transaminases (31% and 46%) respectively for tocilizumab and azathioprine.

Satralizumab

Satralizumab is a humanised IgG2 subtype recombinant anti-IL6-R monoclonal antibody [41]. IL-6 is a pro-inflammatory pleiotropic cytokine produced by a large number of cell types, including T and B lymphocytes, monocytes and fibroblasts [42]. IL-6 plays a role in several immunopathological processes such as T-cell activation, induction of immunoglobulin secretion, and enhancing macrophage activity, and it has been implicated in the mechanism of NMOSD [43]. IL-6 specifically contributes to the development of autoimmunity by promoting the generation of Th17 cell lineage [44]. Th17 cells are recognised as the primary T cell sub-population contributing to the development of autoimmune conditions. IL-6 has been found to be significantly elevated in the serum and cerebrospinal fluid of patients with NMOSD; it induces AQP4-Ab production by plasmablasts, and thus represents a novel therapeutic target for NMOSD.

The molecule of satralizumab was designed to improve pharmacokinetics of anti-IL6-R Ab by applying so-called 'antibody recycling technology' [45]. This technology leads to increased dissociation of anti-IL6-R Ab from IL6-R within the acidic environment of the endosome, while maintaining its binding affinity to IL6-R in plasma. Thus, in the endosome acidic environment, IL6-R Ab after degradation of IL6-R is dissociated from this complex and can again bind another IL6-R in the plasma, increasing its efficacy of IL6-R elimination.

Satralizumab efficacy in NMOSD was evaluated in 83 patients in a randomised, double-blind, phase 3 study (SAkuraSky) [46]. AQP4-positive patients represented 66.3% and AQP4-negative patients 33.7%. In this study, satralizumab was compared to a placebo as an add-on to baseline treatment with a stable dose of immunosuppressive and/or corticosteroids.

Subjects were randomised to satralizumab (120 mg s.c.) or placebo administered at weeks 0, 2, 4, and Q4W thereafter. The primary endpoint was time to first protocol-defined relapse (PDR), adjudicated by a clinical endpoint committee. Pre--specified subgroup analyses included assessing the response to treatment by AQP4-Ab serostatus, baseline treatment, and region. Satralizumab showed a 79% risk reduction of PDR compared to placebo in the NMOSD AQP4-Ab positive subgroup (HR, 0.21; 95% CI, 0.06-–0.75). At weeks 48 and 96, the percentages of relapse-free patients were 91.5% (95% CI, 69.6%–97.8%) and 91.5% (95% CI, 69.6%–97.8%) with satralizumab and 59.9% (95% CI, 36.3%–77.3%) and 53.3% (95% CI, 29.3%–72.4%) with placebo, respectively. However for the NMOSD AQP4-Ab negative patients, satralizumab showed a risk reduction of PDR that was significantly lower compared to the AQP4 Ab positive group, 34% compared to the placebo group (HR, 0.66; 95% CI, 0.20–2.23), and the percentages of relapse-free patients at weeks 48 and 96 were 84.4% (95% CI, 50.4%–95.9%) and 56.3% (95% CI, 24.2%–79.2%) with satralizumab, and 75.5% (95% CI, 41.6%–91.4%) and 67.1% (95% CI, 34.2%–86.2%) with placebo, respectively.

More recently, results have become available for satralizumab monotherapy in NMOSD. In the SAkuraStar study, the efficacy and safety of satralizumab was compared to placebo for relapse prevention in patients with NMOSD [47]. In this phase 3, double-blind, placebo-controlled study, 95 patients were randomised 2:1 to satralizumab (120 mg s.c.) or placebo, administered at weeks 0, 2, 4 and every four weeks thereafter. Unlike the SAkuraSky study, concomitant immunosuppressant medications were prohibited. Patients with AQP-4 antibodies represented 65.1% in the satralizumab group and 71.9% in the placebo group. All patients had had ≥ 1 documented relapse, including first attack, in the year prior to screening. The primary endpoint was time to first protocol-defined relapse (PDR) adjudicated by a clinical endpoint committee. Satralizumab monotherapy significantly reduced the risk of PDR by 55% compared to placebo (HR 0.45; 95% CI 0.23–0.89; p = 0.018). The percentages of relapse-free patients at week 48 were 76.1% in the satralizumab group, and 61.9% in the placebo group. At week 96, these values were 72.1% and 51.2%, respectively. As in the SAkuraSky study, in the group of patients without AQP-4 Ab, satralizumab showed significantly less effect on the proportion of relapse-free compared to the AQP-4 Ab positive patients, and the difference between satralizumab and placebo was not significant. Satralizumab was well tolerated, and similar proportions of patients in the satralizumab and placebo groups experienced adverse events. Rates of serious infections were similar between groups. No deaths or anaphylactic reactions were observed with satralizumab or placebo treatment.

The clear difference in satralizumab efficacy between AQP4 Ab-positive patients and AQP4 Ab-negative patients observed in the SAkuraSky and SAkuraStar studies will require further analysis.

Inebilizumab

Inebilizumab is a humanised mAb of IgG1 subtype directed against the extracellular B cell marker CD19 leading to depletion of a broad range of B cells, including autoantibody--secreting plasmablasts and CD19-expressing plasma cells [48]. Inebilizumab induces a cytotoxic T-lymphocyte response and a strong antibody dependent cell cytotoxicity (ADCC) against B cells.

Inebilizumab was evaluated in a phase 3, double-blind, randomised, placebo-controlled trial (N-MOmentum) in 231 NMOSD patients, both AQP4 Ab-positive (91%) and AQP4 Ab-negative (9%) [49]. Enrollment of AQP4 Ab-negative patients required approval of an eligibility committee that confirmed the entry criteria. Participants were randomised 3:1 to either treatment with inebilizumab, 600 mg iv in two doses of 300 mg each two weeks apart, or a placebo, with no further doses occurring after day 15. Concurrent treatment with other immune suppressants was prohibited.

This means that inebilizumab was the first monotherapy tested in NMOSD free from the confounding influence of other background immunosuppressive treatments. B cells were depleted within approximately four weeks of treatment initiation, and this depletion was sustained throughout the randomised, controlled period of the study. The patients were followed for 28 weeks, after which time the blinded control period was stopped early for efficacy. The primary outcome measure was time to first adjudicated relapse. Following the blinded period, patients were given the option of entering an open-label extension period, in which they received 300 mg of inebilizumab every six months. Inebilizumab met the primary efficacy endpoint with a 77% reduction in the risk of developing an NMOSD relapse when compared to placebo in AQP4-Ab seropositive patients after 28 weeks of treatment (HR: 0.227; p < 0.0001).

A similar effect on relapse risk (73% reduction) was observed in the total inebilizumab-treated patient population, inclusive of AQP4-Ab seronegative patients, (HR: 0.272; p < 0.0001). At 28 weeks, at the end of the randomised--controlled period, 89% of AQP4-Ab seropositive patients treated with inebilizumab were relapse-free, versus 58% in the placebo group. Inebilizumab had also met most of the secondary endpoints. Reduction of disability worsening measured with EDSS in the inebilizumab-treated patients was significantly lower, 15.5%, than in the placebo group where it was 33.9%, (p = 0.0049). In inebilizumab-treated patients, the reduction in NMOSD-related hospitalisations was significantly lower, 5.7% of patients, versus placebo 14.3% (p = 0.01). In this study, MRI was also used as a secondary endpoint and showed a reduction in the frequency of cumulative total active MRI lesions in inebilizumab--treated patients (45.4% patients) versus placebo (57.1 %) (p = 0.0034). Visual acuity, another secondary endpoint, did not demonstrate a statistically significant difference

between inebilizumab-treated patients and placebo. Inebilizumab demonstrated favourable safety and tolerability profiles, with an adverse event rate similar to that of the placebo. The rate of infusion-related reactions was low in both arms. The rates of serious and/or ≥ Grade 3 severity adverse events were similar in the inebilizumab (10.3%) and placebo (14.3%) groups. Two deaths occurred in the open-label period: one related to a severe NMOSD relapse, and the other related to a brain event of unclear aetiology without a definite diagnosis.

Based on data from the pivotal N-MOmentum study, the FDA has granted Breakthrough Therapy Designation (BTD) for the development of inebilizumab for the treatment of NMOSD.

Breakthrough Therapy Designation is designed to expedite the development and regulatory review of medicines intended to treat a serious condition that have shown encouraging early clinical results which may demonstrate substantial improvement on a clinically significant endpoint over available medicines.

Future perspectives

The pathogenic role of AQP4-Ab in NMOSD is associated with the production of AQP4-Ab by peripheral plasma cells, which can enter the CNS and bind to AQP4 on perivascular astrocytes. This binding initiates activation of the terminal complement complex and the induction of inflammatory lesion formation. Impaired blood-brain barrier function allows for a massive influx of neutrophils and eosinophils into the CNS of NMOSD patients. Neutrophil counts are elevated in CSF in about 60% of NMOSD patients during relapse, and about 20% during remission [50]. Eosinophils are also present in the CSF of NMOSD patients. Degranulation of these cells and the release of several toxic proteins and enzymes provide a direct mechanism of damage to astrocytes, followed by oligodendrocyte injury and neuronal death [51].

Future NMOSD therapies will need to address the inhibition of AQP4-Ab and prevention of neutrophils and eosinophils activation leading to CNS infiltration.

In vitro and *in vivo* studies have demonstrated that AQP4--Ab deglycosylation or cleavage reduce the complement--dependent cytotoxicity and the antibody-dependent cell--mediated cytotoxicity, leading to decreased astrocyte damage and reduced development of inflammation [52]. Similarly, IgG--degrading enzyme produced by *Streptococcus pyogenes* efficiently cleaved AQP4-Ab in mice *in vivo* and greatly reduced lesion formation in an experimental mice model of NMOSD [53]. The bacteria-derived endoglycosidases may target AQP4-Ab and reduce its pathogenicity by inhibition of the AQP4-Ab binding to AQP4 and preventing NMOSD pathology. Another strategy to inhibit interaction between AQP4-Ab with AQP-4 protein involved Aquaporumab which is a synthetic IgG that competes with AQP4-Ab for AQP 4 binding [54]. In contrast to pathogenic AQP4-Ab, the mutated Fcγ portion of Aquaporumab does not activate the antibody-dependent complement and cell-dependent mediated cytotoxicity. Its competitive inhibition of AQP4-Ab binding depends also on the greater affinity of aquaporumab to the AQP4 protein, compared to that of pathological AQP4-Ab. Its efficacy to compete with AQP4-Ab binding has already been proven in a preclinical study [55].

The dominant presence of neutrophils in inflammatory infiltrates of the CNS, and the proven role of these cells in NMOSD lesions formation, points at their inhibition as a new strategy of NMOSD treatment.

This concept was supported by findings that intracerebral injection of AQP-4 Ab in neutropenic mice induced less inflammation and demyelination than in mice with normal neutrophil counts [56]. The potential utility of neutrophil protease inhibitors might have particular significance in this regard [57]. It has already been shown that Sivelestat, an inhibitor of neutrophil elastase, demonstrated a beneficial effect in animal models of NMOSD, as evidenced by reduced NMOSD lesion formation [56]. Sivelestat was applied intraperitoneally or intracerebrally either alone or in combination with cathepsin G inhibitor. In addition to its inhibition of proteolytic activity, Sivelestat also reduced the production of inflammatory cytokines and suppressed neutrophil-induced capillary permeability and leukocyte kinetics in other conditions [58].

Eosinophil infiltration is another prominent feature of NMOSD lesions, and eosinophils have been found to be elevated in the CSF of NMOSD patients [59]. Accordingly, hypoeosinophilic mice showed diminished potential for NMOSD development. Eosinophil inhibition, either by anti-IL-5 or gene depletion, led to reduced lesion severity in experimental models of NMOSD.

These findings confirm the involvement of eosinophils in NMOSD's pathogenesis, and suggests the therapeutic utility of eosinophil-targeted drugs [60]. The inhibition of eosinophil degranulation has demonstrated promising results in animal models of NMOSD. The histamine H1 receptors antagonists have been shown to influence eosinophil activity. Cetirizine, a selective antagonist of the H1 receptor, was administered orally before and during AQP4-Ab intracerebral injection and significantly reduced eosinophil infiltrates and lesion formation in mice. These results prompted the testing of cetirizine in a pilot, open-label, add-on trial to standard therapy for 16 NMOSD patients. Cetirizine was administered at a dose of 10 mg daily. After one year of treatment, ARR was reduced fourfold in these patients [61]. Thus, antihistaminic drugs affecting eosinophilic function might be beneficial as an add-on therapy in NMOSD treatment.

Another future strategy for NMOSD treatment might be associated with an attempt to enhance apoptotic death of plasma cells. This strategy might reduce the number of cells producing pathogenic AQP4-Ab and demonstrate benefit for NMOSD patients.

Table 2. Results of primary findings for novel NMOSD treatment

Eculizumab	Inebilizumab	Satralizumab	Tocilizumab (SAkuraSky) (SakuraStar)	
Mechanism of action	Anti-C5	Anti-CD19	Anti-IL6-R Ab recycling	Anti-IL6-R
Patients, number	143	230	83 95	118
Status AQP-4 Ab				
– positive	143	212	55 62	100
– negative	0	18	28 33	18
Placebo	+	+	+ +	AZT
Concomitant				
Immunosuppression	+	-	+ -	+/-
Relapse free	96.1%	87.6%	91.5%(AQP+)	91.5%
	(96 w)	(28 w)	72.1% (AQP+) (96 w)	(48 w)
Relapse reduction	93.1%	73%	79% (AQP+)	NA
(HR)	(0.06)	(0.272)	74% (AQP+) (0.21)	
Disability risk reduction	NS	0.371	NA	0.34
(OR)			NA	

HR — hazard ratio; OR — odds ratio; W — weeks; NS — non significant; NA — not available

Bortezomib is a selective inhibitor of the 26S proteasome subunit leading to enhanced cell death. Bortezomib has been tested in an open-label study including five NMOSD AQP4--Ab-positive patients. All patients were refractory to previous therapies, and two of them were resistant to RTX. Four of these five patients remained stable or improved within 12 months of the study. Patients treated with bortezomib had a lower plasma cell count and diminished levels of serum AQP4-Ab [62].

Conclusions

Recent years have witnessed unprecedented progress in the understanding and treatment of NMOSD (Tab. 2). 2019 has been called the Year of NMOSD. The discovery of the pathogenic role of AQP4-Ab and anti-MOG Ab has allowed the targeting of the basic immune mechanism of this disease.

The exciting findings of the recent randomised and controlled trials have provided a realistic hope that the era of unproved therapies in NMOSD will come to the end.

The findings from studies with novel drugs targeting complement activation, interfering with IL6-R activation and depleting antibody-producing plasma cells, should soon bring about a new and effective treatment of this devastating disease. These results are very welcome for NMOSD patients who have been relegated to off-label therapies with uncertain benefits for many years. The approval of new therapies for NMOSD will undoubtedly prove to be another breakthrough in modern neurology.

References

1. Flanagan EP, Weinshenker BG. Neuromyelitis optica spectrum disorders. Curr Neurol Neurosci Rep. 2014; 14(9): 483, doi: 10.1007/s11910-014-0483-3, indexed in Pubmed: 25027264.
2. Lennon VA, Wingerchuk DM, Kryzer TJ, et al. A serum autoantibody marker of neuromyelitis optica: distinction from multiple sclerosis. Lancet. 2004; 364(9451): 2106–2112, doi: 10.1016/S0140-6736(04)17551-X, indexed in Pubmed: 15589308.
3. Hinson SR, Pittock SJ, Lucchinetti CF, et al. Pathogenic potential of IgG binding to water channel extracellular domain in neuromyelitis optica. Neurology. 2007; 69(24): 2221–2231, doi: 10.1212/01.WNL.0000289761.64862.ce, indexed in Pubmed: 17928579.
4. Bennett JL, Lam C, Kalluri SR, et al. Intrathecal pathogenic anti-aquaporin-4 antibodies in early neuromyelitis optica. Ann Neurol. 2009; 66(5): 617–629, doi: 10.1002/ana.21802, indexed in Pubmed: 19938104.
5. Takahashi T, Fujihara K, Nakashima I, et al. Anti-aquaporin-4 antibody is involved in the pathogenesis of NMO: a study on antibody titre. Brain. 2007; 130(Pt 5): 1235–1243, doi: 10.1093/brain/awm062, indexed in Pubmed: 17449477.
6. Jarius S, Aboul-Enein F, Waters P, et al. Antibody to aquaporin-4 in the long-term course of neuromyelitis optica. Brain. 2008; 131(Pt

11): 3072–3080, doi: 10.1093/brain/awn240, indexed in Pubmed: 18945724.
7. Papadopoulos M, Verkman AS. Aquaporin 4 and neuromyelitis optica. The Lancet Neurology. 2012; 11(6): 535–544, doi: 10.1016/s1474-4422(12)70133-3.
8. Bradl M, Reindl M, Lassmann H. Mechanisms for lesion localization in neuromyelitis optica spectrum disorders. Curr Opin Neurol. 2018; 31(3): 325–333, doi: 10.1097/WCO.0000000000000551, indexed in Pubmed: 29465432.
9. Sato DK, Callegaro D, Lana-Peixoto MA, et al. Distinction between MOG antibody-positive and AQP4 antibody-positive NMO spectrum disorders. Neurology. 2014; 82(6): 474–481, doi: 10.1212/WNL.0000000000000101, indexed in Pubmed: 24415568.
10. Ikeda K, Kiyota N, Kuroda H, et al. Severe demyelination but no astrocytopathy in clinically definite neuromyelitis optica with anti-myelin-oligodendrocyte glycoprotein antibody. Mult Scler. 2015; 21(5): 656–659, doi: 10.1177/1352458514551455, indexed in Pubmed: 25257613.
11. Wingerchuk DM, Banwell B, Bennett JL, et al. International Panel for NMO Diagnosis. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. Neurology. 2015; 85(2): 177–189, doi: 10.1212/WNL.0000000000001729, indexed in Pubmed: 26092914.
12. Sechi E, Morris PP, McKeon A, et al. Glial fibrillary acidic protein IgG related myelitis: characterisation and comparison with aquaporin-4-IgG myelitis. J Neurol Neurosurg Psychiatry. 2019; 90(4): 488–490, doi: 10.1136/jnnp-2018-318004, indexed in Pubmed: 30032117.
13. Abboud H, Petrak A, Mealy M, et al. Treatment of acute relapses in neuromyelitis optica: Steroids alone versus steroids plus plasma exchange. Mult Scler. 2016; 22(2): 185–192, doi: 10.1177/1352458515581438, indexed in Pubmed: 25921047.
14. Watanabe S, Nakashima I, Misu T, et al. Therapeutic efficacy of plasma exchange in NMO-IgG-positive patients with neuromyelitis optica. Mult Scler. 2007; 13(1): 128–132, doi: 10.1177/1352458506071174, indexed in Pubmed: 17294622.
15. Reindl M, Waters P. Myelin oligodendrocyte glycoprotein antibodies in neurological disease. Nat Rev Neurol. 2019; 15(2): 89–102, doi: 10.1038/s41582-018-0112-x, indexed in Pubmed: 30559466.
16. Elsone L, Panicker J, Mutch K, et al. Role of intravenous immunoglobulin in the treatment of acute relapses of neuromyelitis optica: experience in 10 patients. Mult Scler. 2014; 20(4): 501–504, doi: 10.1177/1352458513495938, indexed in Pubmed: 23986097.
17. Sellner J, Boggild M, Clanet M, et al. EFNS guidelines on diagnosis and management of neuromyelitis optica. Eur J Neurol. 2010; 17(8): 1019–1032, doi: 10.1111/j.1468-1331.2010.03066.x, indexed in Pubmed: 20528913.
18. Kimbrough DJ, Fujihara K, Jacob A, et al. GJCF-CC&BR. Treatment of Neuromyelitis Optica: Review and Recommendations. Mult Scler Relat Disord. 2012; 1(4): 180–187, doi: 10.1016/j.msard.2012.06.002, indexed in Pubmed: 24555176.
19. Elsone L, Kitley J, Luppe S, et al. Long-term efficacy, tolerability and retention rate of azathioprine in 103 aquaporin-4 antibody-positive neuromyelitis optica spectrum disorder patients: a multicentre retrospective observational study from the UK. Mult Scler. 2014; 20(11): 1533–1540, doi: 10.1177/1352458514525870, indexed in Pubmed: 24647557.
20. Cabre P, Olindo S, Marignier R, et al. Aegis of French National Observatory of Multiple Sclerosis. Efficacy of mitoxantrone in neuromyelitis optica spectrum: clinical and neuroradiological study. J Neurol Neurosurg Psychiatry. 2013; 84(5): 511–516, doi: 10.1136/jnnp-2012-303121, indexed in Pubmed: 23138769.
21. Awad A, Stüve O. Cyclophosphamide in multiple sclerosis: scientific rationale, history and novel treatment paradigms. Ther Adv Neurol Disord. 2009; 2(6): 50–61, doi: 10.1177/1756285609344375, indexed in Pubmed: 21180630.
22. Montcuquet A, Collongues N, Papeix C, et al. NOMADMUS study group and the Observatoire Français de la Sclérose en Plaques (OFSEP). Effectiveness of mycophenolate mofetil as first-line therapy in AQP4-IgG, MOG-IgG, and seronegative neuromyelitis optica spectrum disorders. Mult Scler. 2017; 23(10): 1377–1384, doi: 10.1177/1352458516678474, indexed in Pubmed: 27885065.
23. Huh SY, Kim SH, Hyun JW, et al. Mycophenolate Mofetil in the Treatment of Neuromyelitis Optica Spectrum Disorder. JAMA Neurology. 2014; 71(11): 1372, doi: 10.1001/jamaneurol.2014.2057.
24. Hughes RAC, Donofrio P, Bril V, et al. ICE Study Group. Intravenous immune globulin (10% caprylate-chromatography purified) for the treatment of chronic inflammatory demyelinating polyradiculoneuropathy (ICE study): a randomised placebo-controlled trial. Lancet Neurol. 2008; 7(2): 136–144, doi: 10.1016/S1474-4422(07)70329-0, indexed in Pubmed: 18178525.
25. Dalakas M. IVIg in other autoimmune neurological disorders: current status and future prospects. J Neurol. 2008; 255 Suppl 3: 12–16, doi: 10.1007/s00415-008-3004-y, indexed in Pubmed: 18685921.
26. Magraner MJ, Coret F, Casanova B. The effect of intravenous immunoglobulin on neuromyelitis optica. Neurologia. 2013; 28(2): 65–72, doi: 10.1016/j.nrl.2012.03.014, indexed in Pubmed: 22841880.
27. Viswanathan S, Wong AHY, Quek AML, et al. Intravenous immunoglobulin may reduce relapse frequency in neuromyelitis optica. J Neuroimmunol. 2015; 282: 92–96, doi: 10.1016/j.jneuroim.2015.03.021, indexed in Pubmed: 25903734.
28. Cabre P, Mejdoubi M, Jeannin S, et al. Francophone Society of Multiple Sclerosis and OFSEP investigators. Treatment of neuromyelitis optica with rituximab: a 2-year prospective multicenter study. J Neurol. 2018; 265(4): 917–925, doi: 10.1007/s00415-018-8771-5, indexed in Pubmed: 29455361.
29. Alldredge B, Jordan A, Imitola J, et al. Safety and Efficacy of Rituximab: Experience of a Single Multiple Sclerosis Center. Clin Neuropharmacol. 2018; 41(2): 56–59, doi: 10.1097/WNF.0000000000000268, indexed in Pubmed: 29389745.
30. Gao F, Chai B, Gu C, et al. Effectiveness of rituximab in neuromyelitis optica: a meta-analysis. BMC Neurol. 2019; 19(1): 36, doi: 10.1186/s12883-019-1261-2, indexed in Pubmed: 30841862.
31. Brachet G, Bourquard T, Gallay N, et al. Eculizumab epitope on complement C5: Progress towards a better understanding of the mechanism of action. Mol Immunol. 2016; 77: 126–131, doi: 10.1016/j.molimm.2016.07.016, indexed in Pubmed: 27497837.
32. Soltys J, Liu Y, Ritchie A, et al. Membrane assembly of aquaporin-4 autoantibodies regulates classical complement activation in neuromyelitis optica. J Clin Invest. 2019; 129(5): 2000–2013, doi: 10.1172/JCI122942, indexed in Pubmed: 30958797.
33. Yao X, Verkman AS. Complement regulator CD59 prevents peripheral organ injury in rats made seropositive for neuromyelitis optica immunoglobulin G. Acta Neuropathol Commun. 2017; 5(1): 57, doi: 10.1186/s40478-017-0462-4, indexed in Pubmed: 28750658.
34. Pittock SJ, Berthele A, Fujihara K, et al. Eculizumab in Aquaporin-4-Positive Neuromyelitis Optica Spectrum Disorder. N Engl J Med. 2019; 381(7): 614–625, doi: 10.1056/NEJMoa1900866, indexed in Pubmed: 31050279.

35. Benamu E, Montoya JG. Infections associated with the use of eculizumab: recommendations for prevention and prophylaxis. Curr Opin Infect Dis. 2016; 29(4): 319–329, doi: 10.1097/QCO.0000000000000279, indexed in Pubmed: 27257797.
36. Horellou P, Wang M, Keo V, et al. Increased interleukin-6 correlates with myelin oligodendrocyte glycoprotein antibodies in pediatric monophasic demyelinating diseases and multiple sclerosis. J Neuroimmunol. 2015; 289: 1–7, doi: 10.1016/j.jneuroim.2015.10.002, indexed in Pubmed: 26616865.
37. Kieseier BC, Stüve O, Dehmel T, et al. Disease amelioration with tocilizumab in a treatment-resistant patient with neuromyelitis optica: implication for cellular immune responses. JAMA Neurol. 2013; 70(3): 390–393, doi: 10.1001/jamaneurol.2013.668, indexed in Pubmed: 23599943.
38. Araki M, Matsuoka T, Miyamoto K, et al. Efficacy of the anti-IL-6 receptor antibody tocilizumab in neuromyelitis optica: a pilot study. Neurology. 2014; 82(15): 1302–1306, doi: 10.1212/WNL.0000000000000317, indexed in Pubmed: 24634453.
39. Ringelstein M, Ayzenberg I, Harmel J, et al. Long-term Therapy With Interleukin 6 Receptor Blockade in Highly Active Neuromyelitis Optica Spectrum Disorder. JAMA Neurol. 2015; 72(7): 756–763, doi: 10.1001/jamaneurol.2015.0533, indexed in Pubmed: 25985228.
40. Zhang C, Zhang M, Qiu W, et al. Tocilizumab versus Azathioprine in highly relapsing neuromyelitis optica spectrum disorders (TANGO): a head-to-head comparative study. Multiple Sclerosis J. 2019; 25(S2): 44.
41. Kaplon H, Reichert JM. Antibodies to watch in 2018. MAbs. 2018; 10(2): 183–203, doi: 10.1080/19420862.2018.1415671, indexed in Pubmed: 29300693.
42. Ayzenberg I, Kleiter I, Schröder A, et al. Interleukin 6 receptor blockade in patients with neuromyelitis optica nonresponsive to anti-CD20 therapy. JAMA Neurol. 2013; 70(3): 394–397, doi: 10.1001/jamaneurol.2013.1246, indexed in Pubmed: 23358868.
43. Uchida T, Mori M, Uzawa A, et al. Increased cerebrospinal fluid metalloproteinase-2 and interleukin-6 are associated with albumin quotient in neuromyelitis optica: Their possible role on blood-brain barrier disruption. Mult Scler. 2017; 23(8): 1072–1084, doi: 10.1177/1352458516672015, indexed in Pubmed: 27682231.
44. Kimura A, Naka T, Kishimoto T. IL-6-dependent and -independent pathways in the development of interleukin 17-producing T helper cells. Proc Natl Acad Sci U S A. 2007; 104(29): 12099–12104, doi: 10.1073/pnas.0705268104, indexed in Pubmed: 17623780.
45. Igawa T, Ishii S, Tachibana T, et al. Antibody recycling by engineered pH-dependent antigen binding improves the duration of antigen neutralization. Nat Biotechnol. 2010; 28(11): 1203–1207, doi: 10.1038/nbt.1691, indexed in Pubmed: 20953198.
46. de Se, Kleiter I, Fujihara K, et al. et al.. A double-blind placebo-controlled study of satralizumab (SA237), a recycling anti-IL-6 receptor monoclonal antibody, as add-on therapy for neuromyelitis optica spectrum disorder (NMOSD). MS Journal 2018, 24: issue 2 suppl. : abstract.
47. Bennett, B. Greenberg, A. Traboulsee, Efficacy of satralizumab as monotherapy in pre-specified subgroups of SAkuraStar, a double-blind placebo-controlled Phase 3 clinical study in patients with neuromyelitis optica spectrum disorder (NMOSD)J.L. Multiple Sclerosis J. 2019; 25(S2): 44.
48. Chen D, Gallagher S, Monson NL, et al. Inebilizumab, a B Cell-Depleting Anti-CD19 Antibody for the Treatment of Autoimmune Neurological Diseases: Insights from Preclinical Studies. J Clin Med. 2016; 5(12), doi: 10.3390/jcm5120107, indexed in Pubmed: 27886126.
49. Cree BAC, Bennett JL, Kim HoJ, et al. N-MOmentum study investigators. Inebilizumab for the treatment of neuromyelitis optica spectrum disorder (N-MOmentum): a double-blind, randomised placebo-controlled phase 2/3 trial. Lancet. 2019; 394(10206): 1352–1363, doi: 10.1016/S0140-6736(19)31817-3, indexed in Pubmed: 31495497.
50. Jarius S, Paul F, Franciotta D, et al. Cerebrospinal fluid findings in aquaporin-4 antibody positive neuromyelitis optica: results from 211 lumbar punctures. J Neurol Sci. 2011; 306(1-2): 82–90, doi: 10.1016/j.jns.2011.03.038, indexed in Pubmed: 21550068.
51. Michael BD, Elsone L, Griffiths MJ, et al. Post-acute serum eosinophil and neutrophil-associated cytokine/chemokine profile can distinguish between patients with neuromyelitis optica and multiple sclerosis; and identifies potential pathophysiological mechanisms - a pilot study. Cytokine. 2013; 64(1): 90–96, doi: 10.1016/j.cyto.2013.07.019, indexed in Pubmed: 23941778.
52. Tradtrantip L, Ratelade J, Zhang H, et al. Enzymatic deglycosylation converts pathogenic neuromyelitis optica anti-aquaporin-4 immunoglobulin G into therapeutic antibody. Ann Neurol. 2013; 73(1): 77–85, doi: 10.1002/ana.23741, indexed in Pubmed: 23055279.
53. Tradtrantip L, Asavapanumas N, Verkman AS. Therapeutic cleavage of anti-aquaporin-4 autoantibody in neuromyelitis optica by an IgG-selective proteinase. Mol Pharmacol. 2013; 83(6): 1268–1275, doi: 10.1124/mol.113.086470, indexed in Pubmed: 23571414.
54. Verkman AS, Phuan PW, Asavapanumas N, et al. Biology of AQP4 and anti-AQP4 antibody: therapeutic implications for NMO. Brain Pathol. 2013; 23(6): 684–695, doi: 10.1111/bpa.12085, indexed in Pubmed: 24118484.
55. Verkman AS, Smith AJ, Phuan PW, et al. The aquaporin-4 water channel as a potential drug target in neurological disorders. Expert Opin Ther Targets. 2017; 21(12): 1161–1170, doi: 10.1080/14728222.2017.1398236, indexed in Pubmed: 29072508.
56. Saadoun S, Waters P, MacDonald C, et al. Neutrophil protease inhibition reduces neuromyelitis optica-immunoglobulin G-induced damage in mouse brain. Ann Neurol. 2012; 71(3): 323–333, doi: 10.1002/ana.22686, indexed in Pubmed: 22374891.
57. Herges K, de Jong BA, Kolkowitz I, et al. Protective effect of an elastase inhibitor in a neuromyelitis optica-like disease driven by a peptide of myelin oligodendroglial glycoprotein. Mult Scler. 2012; 18(4): 398–408, doi: 10.1177/1352458512440060, indexed in Pubmed: 22343184.
58. Domon H, Nagai K, Maekawa T, et al. Neutrophil Elastase Subverts the Immune Response by Cleaving Toll-Like Receptors and Cytokines in Pneumococcal Pneumonia. Front Immunol. 2018; 9: 732, doi: 10.3389/fimmu.2018.00732, indexed in Pubmed: 29922273.
59. Muroishi T, Sakai K, Yanase D, et al. Serum anti-aquaporin-4 antibody-positive neuromyelitis optica spectrum disorder presenting as acute eosinophilic encephalomyelitis. J Clin Neurosci. 2018; 48: 93–94, doi: 10.1016/j.jocn.2017.10.074, indexed in Pubmed: 29137920.
60. Zhang H, Verkman AS. Eosinophil pathogenicity mechanisms and therapeutics in neuromyelitis optica. J Clin Invest. 2013; 123(5): 2306–2316, doi: 10.1172/JCI67554, indexed in Pubmed: 23563310.
61. Katz Sand I, Fabian MT, Telford R, et al. Open-label, add-on trial of cetirizine for neuromyelitis optica. Neurol Neuroimmunol Neuroinflamm. 2018; 5(2): e441, doi: 10.1212/NXI.0000000000000441, indexed in Pubmed: 30426035.
62. Zhang C, Tian DC, Yang CS, et al. Safety and Efficacy of Bortezomib in Patients With Highly Relapsing Neuromyelitis Optica Spectrum Disorder. JAMA Neurol. 2017; 74(8): 1010–1012, doi: 10.1001/jamaneurol.2017.1336, indexed in Pubmed: 28692708.

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Botulinum toxin type A as an alternative way to treat trigeminal neuralgia: a systematic review

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ABSTRACT

Introduction. Trigeminal neuralgia (TN) is one of the most common neurological diseases involving the orofacial region. It affects mainly the older population, usually after the age of 60, and more commonlywomen. It involves the fifth cranial nerve and manifests as paroxysmal, unilateral, severe, shock-like or knife-like pain of from a second to two minutes' duration. Usually pain attacks arise spontaneously, but they can also be precipitated by triggers such as cold weather, brushing teeth or shaving. The ICHD-3 classification divides TN into classical, secondary and idiopathic. Current treatment includes pharmacological and surgical methods. Anticonvulsants, such as carbamazepine and oxcarbazepine, are the first line therapy. Microvascular decompression is the most common and most effective way to treat TN surgically. However, none of these methods is free from complications. Moreover, 25–50% of patients became refractory to drug therapy. Some studies have shown that a new therapy that uses a Botulinum toxin type A can be a safe and effective way to treat trigeminal neuralgia.

Methods. Literature from the PubMed base and the Main Medical Library from the last 18 years was analysed. Forty-three items were obtained; after verification, seven articles were included.

Aim of the study. To look at current guidelines about treating trigeminal neuralgia with Botulinum Toxin type A in patients who are refractory to drug therapy or who do not want to undergo surgical treatment.

Conclusion. BoNT-A therapy is a safe and effective method of treating trigeminal neuralgia.

Key words: trigeminal neuralgia, neuropathic pain, trigeminal nerve, botulinum toxin

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Introduction

Trigeminal neuralgia (TN) is one of the most common neurological pains involving the orofacial region. Epidemiological studies have shown that approximately 4 to 28.9/100,000 persons worldwide experience TN. It affects mainly the older population, usually after the age of 60, and is more common in women than in men [1, 2]. It involves the fifth cranial nerve and manifests as paroxysmal, unilateral, severe, shock-like or knife-like pain lasting between a second and two minutes. Usually pain attacks arise spontaneously, but they can also be precipitated by triggers such as cold weather, brushing teeth or shaving [3].

IHS Classification ICHD-3 divides TN into classical, secondary and idiopathic.

There are two types of classical TN: purely paroxysmal (without persistent background facial pain); and with concomitant continuous pain (with persistent background facial pain). Classical TN is mainly caused by vascular compression.

The classification distinguishes three types of secondary TN: TN attributed to multiple sclerosis, TN attributed to a space-occupying lesion (e.g. brain tumour) and TN attributed to a cause other than those described above.

Idiopathic TN divides in two: purely paroxysmal and TN with concomitant continuous pain [4, 5]

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TN can be treated pharmacologically and non-pharmacologically. Anticonvulsants such as carbamazepine and oxcarbazepine are the first-line therapy in patients with trigeminal neuralgia. Unfortunately, about 25–50% of patients become refractory to drug therapy and require higher doses during the therapy. Also, the risk of side effects increases with prolonged use. Patients who are refractory to drugs or who do not tolerate side effects may undergo surgical procedures, i.e. microvascular decompression, gamma-knife radiosurgery, partial sensory rhizotomy, and percutaneous radiofrequency thermocoagulation, which comprise the second-line therapy in TN. But all of these surgical methods are associated with multiple postoperative complications [6, 7]

Some studies have shown that there is a new therapy that uses a Botulinum toxin type A (BTX-A or BoNT-A), a natural neurotoxin derived from the bacterium *Clostridium botulinum* [8, 9]. It inhibits the release of acetylcholine at neuromuscular junctions, causing relaxation of the muscle as well as pain-modulating neurotransmitters, which is why it has been studied for its effectiveness in treating neuropathic pain [10]. BTX-A is commonly used in cosmetic surgery, and in recent years also in medicine to manage conditions such as blepharospasm, hemifacial spasm, chronic migraine, post-stroke spasticity and many other types of headache [11–15].

The present review was undertaken to give an overview of the available evidence that BoNT-A has a therapeutic effect on TN.

Methods

The literature from the PubMed base and the Main Medical Library from the last 18 years was analysed. Key words included: trigeminal neuralgia, botulinum toxin, neuropathic pain, and trigeminal nerve. Forty-three items were obtained; after verification, seven articles were reviewed with attention to the efficacy and safety of Botulinum toxin Type A therapy in trigeminal neuralgia. The works were set out in a tabular form considering the number of patients and the BoNT-A therapy used to reduce the pain in TN patients: this is Table 1. Different types of toxin which were used in the evaluated studies are set out in Table 2.

Results

Wu et al. [16] performed a randomised, double-blind, placebo-controlled study to investigate the efficacy, safety and tolerability of BoNT-A in the treatment of trigeminal neuralgia. A total of 42 patients were randomly divided into two groups: 22 in the BoNT-A group and 20 in the placebo group. They were followed-up over 12 weeks. Intradermal and/or submucosal injections were performed along the area of pain distribution. A total of 75 U of BoNT-A were given into 15 injection sites. Similarly, patients from the placebo group received an equal volume of 0.9% saline. The object of the study was to estimate the degree of pain relief, which was based on the visual analogue scale (VAS), plus the frequency of pain attacks per day. Patients with a ≥ 50% reduction in mean VAS score at the endpoint of the study were considered to be responders. Out of 42 patients, 21 and 19 patients from the BoNT-A and placebo groups, respectively, completed the study. Fifteen (68.18%) patients from the BoNT-A group and three (15.00%) from the placebo group responded to treatment. Moreover, 17 (77.27%) patients from the BoNT-A and four (20.00%) patients from the placebo group reported a significant or very significant improvement in symptoms. Attack frequency was also reduced in the BoNT-A patients. All of these differences were found to be statistically significant. The authors concluded that Botulinum toxin type-A may be a novel, safe and efficient strategy for trigeminal neuralgia treatment.

Zúñiga et al. [17] treated 36 patients with a subcutaneous injections that contained 1 ml of 0.9% saline or 50 U of BOTOX, depending on the group of patients. Cases with third branch of involved also received either 10 U of BOTOX or placebo into the masseter muscle. This randomised, double-blind, placebo-controlled study was performed to evaluate the tolerability, safety and efficacy of BoNT-A treatment in classical TN. Patients were assigned to two groups, 20 of them to receive BOTOX, and 16 to receive a placebo. Drugs were administered in various sites, 1 cm apart from one another, according to the path of the involved branch. This was always performed by the same physician using the same technique. The aim of the study was to evaluate the reduction in pain severity and attack frequency. Pain was assessed with the visual analogue scale (VAS). All patients completed the study. The initial mean VAS scores was 8.85 and 8.19 for BoNT-A and placebo patients, respectively. One month after intervention, the differences in mean VAS values in both groups were compared, and reported as nonsignificant (VAS 5.05 *vs* 6.06 in BoNT-A and placebo group, respectively). A significant decrease in the number of paroxysms was observed among the patients treated with BOTOX. Two months after the injections, a significant reduction in mean VAS scores in the BoNT-A group was observed and this was maintained until the end of the three-month follow-up period. At the endpoint of the study, the mean VAS score for patients treated with BOTOX was 4.75, and 6.94 for those treated with a placebo. Also, the frequency of attacks was significantly lower than at the beginning in the BoNT-A group (29.1 *vs* 7.1 paroxysms per day). These results were statistically significant. The authors concluded that Botulinum toxin type A is useful in acute treatment of classical TN, and when added to current drug therapy may cause a significant decrease in pain expression. Eighteen patients completed the study.

Shehata et al. [18] carried out a randomised, double-blind, placebo-controlled study. They treated 20 patients with intractable idiopathic TN, which was defined as a failure to achieve 50% reduction in pain intensity (quantified by VAS) and/or paroxysm frequency during the previous three months. The aim

Table 1. Summary of study characteristics

Author, country, year	Title	Placebo group	BoNT-A group	BoNT-A therapy	Results	Conclusion
Wu et al., China, 2012	Botulinum Toxin Type A for the treatment [...]	n = 20	n = 22	I.d. and/or s.m.; 75 U into 15 sites	40 patients completed the study. 17/22 patients from BoNT-A group reported significant or very significant improvement of symptoms *vs* 4/20 patients from placebo group.	BoNT-A may be an efficient, safe and novel strategy for TN treatment.
Zůňiga et al., Argentina, 2013	Acute Treatment [...]	n = 16	n = 20	50 U of BOTOX; $V_{1/2}$ - s.c., according to the path of branches; V_3 - i.m.	Reduction of attack frequency from 1st month and pain intensity from 3rd month.	BoNT-A as an addition to drug therapy and in acute treatment.
Shehata et al., Egypt, 2013	Botulinum Toxin--Type A [...]	n = 10	n = 10	$V_{1/2}$ - s.c., 'follow the pain' method and trigger areas; V_3 - i.m. into posterior part of the masseter; 5 U per site	Significant pain intensity and attack frequency reduction in BoNT-A group *vs* placebo group during 12-week follow-up.	BoNT-A can relieve pain in patients with intractable TN.
Li et al., China, 2014	Therapeutic effect of Botulinum toxin-type [...]		$n_{\leq 5\ U} = 43$ $n_{50\text{-}100\ U} = 32$ $n_{\geq 100\ U} = 13$	Inj. in facial area and trigger points, 2.5–5 U per point; doses from 25 to 170 U	No significant difference in BoNT-A efficacy between groups. Effects of treatment decreased after 3 months.	Maintenance of the therapeutic effect was related to a reduction of the VAS score after the first inj. and not related to different dosages.
Zhang et al., China, 2014	Two doses of botulinum [...]	n = 28	$n_{25\ U} = 27$ $n_{75\ U} = 29$	I.d. and/or s.m. inj. according to patient's expression of pain; 25 U or 75U at 20 sites	80 patients completed the study. No significant differences between BoNT-A groups in terms of VAS score reduction.	Lower dose and high dose are similar in efficacy in short-term.
Zhang et al., China, 2017	Single-dose Botulinum Toxin Type A [...]		$n_{s\text{-}d} = 50$ $n_{r\text{-}d} = 50$	I.d. and/or s.c., 1.25–5U per site; Single-doses from 70 to 100 U; Repeated-doses from 100 to 140 U, 50–70 U per time; 2 weeks break between inj.	Significant decrease in VAS scores in both groups. Duration of efficacy significantly longer in single-dose group.	Repeated dosing has no advantage over single dosing of BoNT-A.
Liu et al., China, 2018	Efficacy and Safety of Botulinum Toxin [...]		$n_{\geq 80y.o.} = 14$ $n_{< 6y.o.} = 29$	I.d. and/or s.m. inj. guided by patient's perception and trigger zones; doses from 45 to 150 U in older and from 30 to 200 U in younger patients.	Significant VAS score reduction in both groups.	Effective and safe treatment for idiopathic TN in older patients at doses similar to those used in younger patients.

Table 2. Types of botulinum toxin

Author, title, country, year	Type of botulinum toxin	Dilution
Wu et al., Botulinum Toxin Type A for the treatment [...]; China, 2012	HengLi® (Onabotulinumtoxin type A); 100 U of Clostridium botulinum type A neurotoxin complex, 5mg gelatin, 25mg dextran, and 25mg saccharose (Lanzhou Biological Products Institute, China)	100 U of BoNT-A in 2 ml 0.9% saline
Zůñiga et al., Acute Treatment [...]; Argentina, 2013	BOTOX® (Onabotulinumtoxin type A)	100 U of BoNT-A in 2 ml 0.9% saline
Shehata et al., Botulinum Toxin-Type A [...]; Egypt, 2013	BOTOX® (Onabotulinumtoxin type A)	100 U of BoNT-A in 2 ml 0.9% saline
Li et al., Therapeutic effect of Botulinum toxin-type [...]; China, 2014	HengLi® (Onabotulinumtoxin type A); 100 U of Clostridium botulinum type A neurotoxin complex, 5mg gelatin, 25mg dextran, and 25mg saccharose (Lanzhou Biological Products Institute, China)	100 U of BoNT-A in 2 ml 0.9% saline
Zhang et al., Two doses of botulinum [...]; China, 2014	HengLi® (Onabotulinumtoxin type A); 100 U of Clostridium botulinum type A neurotoxin complex, 5mg gelatin, 25mg dextran, and 25mg saccharose (Lanzhou Biological Products Institute, China)	Depending on group: 25 U of BoNT-A in 1 ml 0.9% saline or 75 U of BoNT-A in 1 ml 0.9% saline
Zhang et al., Single-dose Botulinum Toxin Type A [...]; China, 2017	HengLi® (Onabotulinumtoxin type A); 100 U of Clostridium botulinum type A neurotoxin complex, 5mg gelatin, 25mg dextran, and 25mg saccharose (Lanzhou Biological Products Institute, China)	100 U of BoNT-A in 2 ml 0.9% saline
Liu et al., Efficacy and Safety of Botulinum Toxin [...]; China, 2018	HengLi® (Onabotulinumtoxin type A); 100 U of Clostridium botulinum type A neurotoxin complex, 5mg gelatin, 25mg dextran, and 25mg saccharose (Lanzhou Biological Products Institute, China)	100 U of BoNT-A in 4 ml 0.9% saline

of the study was to evaluate the efficacy, safety and tolerability of BoNT-A in patients who were refractory to first-line drug therapy. Patients were randomised into either the BoNT-A or the placebo group, 10 subjects in each group. Subcutaneous injections were performed using the 'follow the pain' method. Also, botulinum toxin was injected into the posterior part of the masseter muscle if the mandibular root of the trigeminal nerve was affected. Each injection site received 5 U of BoNT--A. The overall doses ranged from 40 U to 60 U (mean ± SD of 48 ± 5.87 U). All of the patients completed the 12-week study period. Significant reductions in both efficacy measures, i.e. pain VAS scores and attack frequency, were observed in week 2 and were maintained over the follow-up period. At the endpoint, mean VAS scores were decreased by 6.5 and 0.3 for BoNT-A and placebo, respectively. The authors concluded that subcutaneous BoNT-A injections can be an effective treatment option in cases with intractable TN.

Several studies were evaluated for the purpose of assessing the efficacy of different doses and methods in botulinum toxin type A therapy. Li et al. [19] performed a study to investigate the long-term effects and safety of BoNT-A in TN treatment. Eighty-eight subjects with one-branch classical TN were included. Patients were asked to report the expression of pain (measured by VAS) and attack frequency. A patient's overall response to treatment was evaluated over the course of a 14-month follow-up period. The main goal of the study was to estimate the influence of different doses on the therapeutic effect of BoNT-A. Injections were given in the trigger areas at 15–20 points (0.5 cm depth, 15 mm separation, 2.5–5 U per point). Forty-three cases received ≤ 50 U, 32 cases received from 50 U to 100 U, and 13 cases received ≥ 100 U of BoNT-A. Minimal and maximal doses were 25 U and 170 U, respectively. All 88 patients showed an improvement in symptoms within two weeks. At two months, the percentage of patients in whom treatment was effective had reached 100%. After three months, the effectiveness of BoNT-A therapy decreased gradually. The authors concluded that there were no significant differences in effectiveness of treatment or attack frequency between the different dose groups at identical time points between oneand 14 months. Moreover, the maintenance of therapeutic effect was not related to different dosages, but was related to the reduction of VAS score after the first set of injections.

Zhang et al. [20] included 84 patients into a randomised, double-blind, placebo-controlled study to assess the efficacy and safety of two different doses of BoNT-A in TN therapy. All patients suffered from classical TN. Patients were randomly divided into three groups: placebo (n = 28), BoNT-A_{25U} (n = 27), and BoNT-A_{75U} (n = 29), and were followed-up during an 8-week period. Patients received placebo or BoNT-A via intradermal and/or submucosal injections, which were guided by the individual patient's experience of pain. These doses were injected into 20 sites. The purpose of the study was to define

an effective dose of BoNT-A. Pain severity, attack frequency, overall response to treatment, and proportion of responders (≥ 50% reduction in mean pain score from baseline to endpoint) were assessed during the study. 32.1% of patients from the placebo group, 70.4% of patients from the BoNT-A_{25U} group, and 86.2% of patients from the BoNT-A_{75U} group responded to treatment. The VAS scores in the BoNT-A patients reduced significantly one week after the first injections, and were maintained until the end of the study. Pain symptoms vere much or very much improved in 66.7% and 75.9% of patients from the 25 U and the 75 U group, respectively. Only 32.1% of patients who received the placebo reported the same improvement as in the BoNT-A groups. These results showed that both doses were effective in TN treatment, but there were no significant differences between the groups. Thus, the authors concluded that 25 U and 75 U doses are similar in efficacy over a short period of time.

Zhang et al. [21] compared two different methods of BoNT-A administration: single-dose and repeated-dose therapy. They conducted a open-label trial which included 100 patients with classical TN. Fifty patients received an intradermal and/or a submucosal single injection of 70–100 U of BoNT-A at the site of pain. The other 50 patients received two sets of injections using the aforesaid method. The initial injection contained 50–70 U of BoNT-A. Two weeks later, another set of injections was given with an equal volume of BoNT-A. The drug was given in 15–25 sites, with 0.1 cm depth and 15 mm separation between sites. Each point received 1.25–5 U of BoNT-A. Every patient was interviewed for severity of pain (VAS score) and rate of TN occurrence. After a 6-month follow-up, 44 patients from the single-dose group and 37 patients from the repeated-dose group completed the study. Mean VAS scores at the baseline and the endpoint for the single-dose method were 8.26 ± 1.68 and 3.02 ± 3.29, respectively. In the repeated-dose method, mean VAS score at the baseline was 7.98 ± 1.60, which had decreased by the endpoint to 4.32 ± 3.61. In both groups, VAS scores and drug response rates were not significantly different. However, the duration of efficacy in those patients who underwent single-dose therapy was significantly longer than that of the repeated-dose patients. The authors conluded that repeated dosing has no advantage over single dosing of BoNT-A, and that single dosing may be the best choice in TN therapy. Multiple injections should be considered for patients who respond poorly to the first set of injections. Dosing should be adjusted for the individual patient.

A new study, which assessed the efficacy and safety of BoNT-A for treating idiopathic TN in patients ≥ 80 y.o., appeared last year. Liu et al. [22] selected 43 patients who were divided into two groups: ≥80 years old (n = 14) and < 60 years old (n = 29). Prior to treatment, the median pain scores (measured by VAS) in both older and younger patients were 8.5 and 8.0, respectively. BoNT-A was given in similar doses in all groups. Total doses ranged from 30 U to 200 U (mean ± SD of 71.80 ± 33.14 U) and from 45 U to 150 U (91.30 ± 25.64 U) in the < 60 y.o. and the ≥ 80 y.o. patients, respectively. One month after treatment, median VAS scores in older (4.5) and younger (4.0) were significantly lower than the corresponding baseline values. The authors proved that BoNT-A injections are an effective and safe therapy for idiopathic TN in elderly patients at doses similar to those used in younger patients.

The use of BoNT-a in the treatment of TN is confirmed not only by the abovementioned studies, but also by many other publications that were not included in our review.

Discussion

TN is characterised by episodes of spontaneous pain or a triggered intense facial pain that lasts for a short duration [23]. According to the current guidelines of the American Academy of Neurology and the European Federation of Neurological Societies (AAN-EFNS), carbamazepine (CBZ) and oxcarbazepine are recommended as first-line drugs for treating patients with trigeminal neuralgia. Drugs such as baclofen, lamotrigine, phenytoin, topiramate and pimozide may also be considered [24]. However, high dosages and the long-term use of CBZ are not free from complications. These mainly take the form of eliciting drowsiness, dizziness, diplopia, leukopenia, hyponatremia and disturbances of the vitamin D metabolism [25]. The most common adverse effects of oxcarbazepine are usually related to the central nervous system and digestive system, including fatigue, drowsiness, diplopia, dizziness, nausea, vomiting and rashes [26]. If the pharmacological treatment fails, there are surgical options that can alleviate the pain in TN, e.g. microvascular decompression, percutaneous radiofrequency rhizotomy, percutaneous glycerol rhizotomy, balloon compression and gamma knife radiosurgery. Microvascular decompression is the most effective treatment for classical TN: it may provide initial pain control in the range of 80.3% to 96.0%. However, this procedure is not as safe as it might seem to be. The most common postoperative complications are an up to 22% rate of trigeminal nerve deficit, less than 11% of facial weakness, about 7% of hearing loss, and sometimes aseptic meningitis, cerebrospinal fluid leakage and anaesthesia dolorosa [27]. Based on the this knowledge, it is safe to say that there is a need to find a new treatment for TN patients.

The use of BTX-A in trigeminal neuralgia was first reported in 2002 by Michaeli et al. who successfully treated a patient with a hemifacial spasm associated with TN [28]. The main action of BoNT-A is to inhibit the release of acetylcholine at the neuromuscular junction. The mechanism of potential analgesic effect of BoNT-A is still unclear. It can inhibit the peripheral sensitisation of nociceptive fibres, hence reducing central sensitisation by inhibiting the release of glutamate and substance P. Moreover, BTX-A significantly reduces the high expression of TRPA1, TRPV1 and TRPV2 in the spinal trigeminal nucleus, what can directly modulate central sensitisation and exerts an antinociceptive function [29, 30].

A new study performed by Yang et al. revealed that the antinociceptive effects of BoNT-A are connected with the inhibition of upregulated Nav isoform 1.7 in Gasserian ganglion [31]. More research needs to be done to thoroughly understand how BoNT-A relieves the pain in TN.

The studies included in our review promote the use of BoNT-A. This topic enjoys great interest in China, and therefore most of the articles chosen for this review are publications from that particular country. However, it should be noted that attempts to treat trigeminal neuralgia with BoNT-A have also been undertaken in other countries, e.g. Norway, USA, Germany, Iran and Malaysia [32–37].

The percentage of patients who have responded to treatment has ranged from 68% to as much as 100%. Attack frequency and pain severity were predominantly reduced one week and two weeks after injections, respectively. Treatment was effective for about 3-6 months. The most commonly used injection method was multiple intradermal and/or subcutaneous injections in the painful area. However, other methods, such as injections into the masseter muscle or injections above and below the zygomatic arch, should also be considered [38–40]. Doses of BoNT-A have been determined empirically and ranged from 25U to 200 U. In addition, this therapy is not free from complications. All of the reported adverse reactions (e.g. facial asymmetry, oedema, pain, haematoma) were mild and transient.

Despite the above-mentioned information, which highlights the positive effect of BoNT-A treatment, a few points need to be discussed.

The first question is the duration of patient follow-up. Wu et al., Zúñiga et al. and Zhang et al. performed studies with follow-ups of eight or 12 weeks or three months. Such periods are too short to assess the long-term effects of BoNT-A in TN, especially in those studies that set out to compare the effects of different doses. Long-term trials are needed to correctly evaluate whether a high dose of BoNT-A has any advantages over lower doses.

Secondly, was the baseline therapy kept unchanged after BoNT-A treatment? There was no information about the anticonvulsants that patients received after injections. In addition, each patient took different doses or medications before and during BoNT-A therapy, what may have played a role in reducing the primary endpoints, such as pain severity and attack frequency. What is more, Li et al. did not provide any information about the medicines their patients were taking. In Wu et al.'s trial, one patient changed his medications during the BoNT-A therapy. We suggest that doses and medications should be uniform so as to clearly assess the efficacy of a new treatment strategy.

Another problem is that follow-up visits have been carried out by some authors for much longer than one week. Liu et al. and Zuniga et al. estimated patient response to treatment every one or three months after BoNT-A administration, respectively. Such a long period of time made it impossible to detect the early disappearance of pain and any reduction in the number of paroxysms per day.

In the study performed by Shehata et al., the blind phase was not maintained after the first follow-up visit. Despite the correct method of randomisation and blinding, facial asymmetry occurred in some patients and led to failure in the blinded phase. For these reasons, the investigator's impressions could be biased.

Liu et al. included only 14 patients aged 80 or older. For comparison, a group with patients younger than 60 years old compared 29 cases. This number of patients is not sufficient to evaluate whether older patients can receive the same doses as younger ones. A study with more elderly patients is needed.

Finally, Zakrzewska JM [41] reported that Wu et al.'s study is not a high-quality evidence of BoNT-A's effectiveness in trigeminal neuralgia therapy. This is mainly because of too little information being provided about the blinding phase and insufficient measures regarding selection, which can capture all aspects of pain experience.

Conclusions

To conclude, this review suggested that Botulinum toxin Type A injections are a safe and effective treatment option for patients with trigeminal neuralgia, and may be offered before surgery or for those unwilling to undergo surgery or where drug treatment has failed. Despite the possibility of occurrence of transient and mild adverse reactions, it represents a propitious risk-to-benefit ratio. However, future studies are necessary to determine the optimal dosages and injection schemes for BoNT-A treatment, the duration of therapeutic efficacy, and indications as to when repeated injections are needed.

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References

1. Dieleman JP, Kerklaan J, Huygen FJ, et al. Incidence rates and treatment of neuropathic pain conditions in the general population. Pain. 2008; 137(3): 681–688, doi: 10.1016/j.pain.2008.03.002, indexed in Pubmed: 18439759.
2. Koopman JS, Dieleman JP, Huygen FJ, et al. Incidence of facial pain in the general population. Pain. 2009; 147(1-3): 122–127, doi: 10.1016/j.pain.2009.08.023, indexed in Pubmed: 19783099.
3. Nurmikko TJ, Jensen TS. Trigeminal neuralgia and other facial neuralgias. In: Olesen J, Goadsby PJ, Ramadan NM, Tfelt-Hansen P, Welch KMA, eds. The Headaches. 3rd ed. Philadelphia: Lippincott Williams & Wilkins; 2006; p. 1053-62; [chapter. ; 38.
4. Headache Classification Committee of the International Headache Society (IHS). The International Classification of Headache Disorders, 3rd edition (beta version). Cephalalgia. 2013; 33(9): 629–808, doi: 10.1177/0333102413485658, indexed in Pubmed: 23771276.

5. Wu M, Fu X, Ji Y, et al. Microvascular Decompression for Classical Trigeminal Neuralgia Caused by Venous Compression: Novel Anatomic Classifications and Surgical Strategy. World Neurosurg. 2018; 113: e707–e713, doi: 10.1016/j.wneu.2018.02.130, indexed in Pubmed: 29510278.
6. Di Stefano G, Truini A, Cruccu G. Current and Innovative Pharmacological Options to Treat Typical and Atypical Trigeminal Neuralgia. Drugs. 2018; 78(14): 1433–1442, doi: 10.1007/s40265-018-0964-9, indexed in Pubmed: 30178160.
7. Obermann M. Treatment options in trigeminal neuralgia. Therapeutic Advances in Neurological Disorders. 2010; 3(2): 107–115, doi: 10.1177/1756285609359317.
8. Piovesan EJ, Teive HG, Kowacs PA, et al. An open study of botulinum--A toxin treatment of trigeminal neuralgia. Neurology. 2005; 65(8): 1306–1308, doi: 10.1212/01.wnl.0000180940.98815.74, indexed in Pubmed: 16247065.
9. Allam N, Brasil-Neto JP, Brown G, et al. Injections of botulinum toxin type a produce pain alleviation in intractable trigeminal neuralgia. Clin J Pain. 2005; 21(2): 182–184, indexed in Pubmed: 15722812.
10. Guo BL, Zheng CX, Sui BD, et al. A closer look to botulinum neurotoxin type A-induced analgesia. Toxicon. 2013; 71: 134–139, doi: 10.1016/j.toxicon.2013.05.011, indexed in Pubmed: 23747735.
11. Safarpour Y, Jabbari B. Botulinum toxin treatment of pain syndromes –an evidence based review. Toxicon. 2018; 147: 120–128, doi: 10.1016/j.toxicon.2018.01.017.
12. Bendtsen L, Sacco S, Ashina M, et al. Guideline on the use of onabotulinumtoxinA in chronic migraine: a consensus statement from the European Headache Federation. J Headache Pain. 2018; 19(1): 91, doi: 10.1186/s10194-018-0921-8, indexed in Pubmed: 30259200.
13. Aschehoug I, Bratbak DF, Tronvik EA. Long-Term Outcome of Patients With Intractable Chronic Cluster Headache Treated With Injection of Onabotulinum Toxin A Toward the Sphenopalatine Ganglion - An Observational Study. Headache. 2018; 58(10): 1519–1529, doi: 10.1111/head.13398, indexed in Pubmed: 30216444.
14. Rosales RL, Efendy F, Teleg ESa, et al. Botulinum toxin as early intervention for spasticity after stroke or non-progressive brain lesion: A meta-analysis. J Neurol Sci. 2016; 371: 6–14, doi: 10.1016/j.jns.2016.10.005, indexed in Pubmed: 27871449.
15. Santamato A, Cinone N, Panza F, et al. Botulinum Toxin Type A for the Treatment of Lower Limb Spasticity after Stroke. Drugs. 2019; 79(2): 143–160, doi: 10.1007/s40265-018-1042-z, indexed in Pubmed: 30623347.
16. Wu CJ, Lian YJ, Zheng YK, et al. Botulinum toxin type A for the treatment of trigeminal neuralgia: results from a randomized, double-blind, placebo-controlled trial. Cephalalgia. 2012; 32(6): 443–450, doi: 10.1177/0333102412441721, indexed in Pubmed: 22492424.
17. Zúñiga C, Piedimonte F, Díaz S, et al. Acute treatment of trigeminal neuralgia with onabotulinum toxin A. Clin Neuropharmacol. 2013; 36(5): 146–150, doi: 10.1097/WNF.0b013e31829cb60e, indexed in Pubmed: 24045604.
18. Shehata HS, El-Tamawy MS, Shalaby NM, et al. Botulinum toxin-type A: could it be an effective treatment option in intractable trigeminal neuralgia? J Headache Pain. 2013; 14: 92, doi: 10.1186/1129-2377-14-92, indexed in Pubmed: 24251833.
19. Li S, Lian YJ, Chen Y, et al. Therapeutic effect of Botulinum toxin-A in 88 patients with trigeminal neuralgia with 14-month follow-up. J Headache Pain. 2014; 15: 43, doi: 10.1186/1129-2377-15-43, indexed in Pubmed: 24952600.
20. Zhang H, Lian Y, Ma Y, et al. Two doses of botulinum toxin type A for the treatment of trigeminal neuralgia: observation of therapeutic effect from a randomized, double-blind, placebo-controlled trial. J Headache Pain. 2014; 15: 65, doi: 10.1186/1129-2377-15-65, indexed in Pubmed: 25263254.
21. Zhang H, Lian Y, Xie N, et al. Single-dose botulinum toxin type a compared with repeated-dose for treatment of trigeminal neuralgia: a pilot study. J Headache Pain. 2017; 18(1): 81, doi: 10.1186/s10194-017-0793-3, indexed in Pubmed: 28799056.
22. Liu J, Xu YY, Zhang QL, et al. Efficacy and Safety of Botulinum Toxin Type A in Treating Patients of Advanced Age with Idiopathic Trigeminal Neuralgia. Pain Res Manag. 2018; 2018: 7365148, doi: 10.1155/2018/7365148, indexed in Pubmed: 29849847.
23. Yadav YR, Nishtha Y, Sonjjay P, et al. Trigeminal Neuralgia. Asian J Neurosurg. 2017; 12(4): 585–597, doi: 10.4103/ajns.AJNS_67_14, indexed in Pubmed: 29114270.
24. Gronseth G, Cruccu G, Alksne J, et al. Practice parameter: the diagnostic evaluation and treatment of trigeminal neuralgia (an evidence-based review): report of the Quality Standards Subcommittee of the American Academy of Neurology and the European Federation of Neurological Societies. Neurology. 2008; 71(15): 1183–1190, doi: 10.1212/01.wnl.0000326598.83183.04, indexed in Pubmed: 18716236.
25. Keränen T, Sivenius J. Side effects of carbamazepine, valproate and clonazepam during long-term treatment of epilepsy. Acta Neurol Scand Suppl. 1983; 97: 69–80, indexed in Pubmed: 6424398.
26. Fang S, Gong ZC. [Adverse effects of oxcarbazepine]. Zhongguo Dang Dai Er Ke Za Zhi. 2015; 17(4): 414–419, indexed in Pubmed: 25919567.
27. Bick SKB, Eskandar EN. Surgical Treatment of Trigeminal Neuralgia. Neurosurg Clin N Am. 2017; 28(3): 429–438, doi: 10.1016/j.nec.2017.02.009, indexed in Pubmed: 28600016.
28. Micheli F, Scorticati MC, Raina G. Beneficial effects of botulinum toxin type a for patients with painful tic convulsif. Clin Neuropharmacol. 2002; 25(5): 260–262, indexed in Pubmed: 12410057.
29. Aoki KR. Evidence for antinociceptive activity of botulinum toxin type A in pain management. Headache. 2003; 43 Suppl 1: S9–15, indexed in Pubmed: 12887389.
30. Wu C, Xie N, Lian Y, et al. Central antinociceptive activity of peripherally applied botulinum toxin type A in lab rat model of trigeminal neuralgia. Springerplus. 2016; 5: 431, doi: 10.1186/s40064-016-2071-2, indexed in Pubmed: 27104119.
31. Yang KY, Kim MJ, Ju JS, et al. Antinociceptive Effects of Botulinum Toxin Type A on Trigeminal Neuropathic Pain. J Dent Res. 2016; 95(10): 1183–1190, doi: 10.1177/0022034516659278, indexed in Pubmed: 27418174.
32. Lunde HM, Torkildsen Ø, Bø L, et al. Botulinum Toxin as Monotherapy in Symptomatic Trigeminal Neuralgia. Headache. 2016; 56(6): 1035–1039, doi: 10.1111/head.12791, indexed in Pubmed: 26992044.
33. Borodic GE, Acquadro MA. The use of botulinum toxin for the treatment of chronic facial pain. J Pain. 2002; 3(1): 21–27, indexed in Pubmed: 14622850.
34. Herrero Babiloni A, Kapos FP, Nixdorf DR. Intraoral administration of botulinum toxin for trigeminal neuropathic pain. Oral Surg Oral Med Oral Pathol Oral Radiol. 2016; 121(6): e148–e153, doi: 10.1016/j.oooo.2016.03.013, indexed in Pubmed: 27181448.
35. Burmeister J, Holle D, Bock E, et al. Botulinum neurotoxin type A in the treatment of classical Trigeminal Neuralgia (BoTN): study protocol for a randomized controlled trial. Trials. 2015; 16: 550, doi: 10.1186/s13063-015-1052-z, indexed in Pubmed: 26634453.
36. Bohluli B, Motamedi MH, Bagheri SC, et al. Use of botulinum toxin A for drug-refractory trigeminal neuralgia: preliminary report. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2011; 111(1): 47–50, doi: 10.1016/j.tripleo.2010.04.043, indexed in Pubmed: 20674409.

37. Ngeow WC, Nair R. Injection of botulinum toxin type A (BOTOX) into trigger zone of trigeminal neuralgia as a means to control pain. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2010; 109(3): e47–e50, doi: 10.1016/j.tripleo.2009.03.021, indexed in Pubmed: 20219585.
38. Wu C, Xie N, Liu H, et al. A new target for the treatment of trigeminal neuralgia with botulinum toxin type A. Neurol Sci. 2018; 39(3): 599–602, doi: 10.1007/s10072-017-3171-7, indexed in Pubmed: 29086125.
39. Türk Börü Ü, Duman A, Bölük C, et al. Botulinum toxin in the treatment of trigeminal neuralgia: 6-Month follow-up. Medicine (Baltimore). 2017; 96(39): e8133, doi: 10.1097/MD.0000000000008133, indexed in Pubmed: 28953646.
40. Volcy M, Tepper SJ, Rapoport AM, et al. Botulinum toxin A for the treatment of greater occipital neuralgia and trigeminal neuralgia: a case report with pathophysiological considerations. Cephalalgia. 2006; 26(3): 336–340, doi: 10.1111/j.1468-2982.2005.00959.x, indexed in Pubmed: 16472343.
41. Zakrzewska JM. Botulinum toxin for trigeminal neuralgia–do we have the evidence? Cephalalgia. 2012; 32(15): 1154–5; author reply 1156, doi: 10.1177/0333102412459577, indexed in Pubmed: 22990690.

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Dystonic tics in patients with Gilles de la Tourette syndrome

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ABSTRACT

Clinical rationale for the study. Gilles de la Tourette syndrome (GTS) is a childhood onset disorder characterised by motor and vocal tics. Different types of motor tics may occur in GTS, including dystonic tics (DTs). Although DTs have been recognised as part of GTS symptomatology, little is known about their risk factors or about how often and at what age they appear in affected individuals.

Aim of the study. The aim of our study was to investigate lifetime prevalence and clinical correlations of DTs in a Polish cohort of GTS patients.

Material and methods. We performed a prospective, one-registration study in a cohort of 207 consecutive ambulatory patients (mean age: 16.5 ± 9.4 years, 131 children, 162 males) with GTS. Duration of GTS was 9.0 ± 8.0 years (range: 1–39 years). DTs were diagnosed during the interview. DTs were defined as slower and lasting longer than typical clonic tics, abnormal dystonia-like movements that led to a sustained, but not fixed, posture.

Results. DTs occurred at some point in the lifetime of 73.9% (n = 153) of patients. The prevalence of DTs in adults and children was almost the same (p = 0.963). Age at onset of DTs was 9.9 ± 5.2 years with the most frequent onset in children (7–11 years, 74.4%, n = 64), followed by adolescence (12–18 years; 17.4%, n = 15) and adulthood (≥ 18 years, 8.1%, n = 7). DTs occurred 3.7 ± 4.2 years after tic onset. On average, patients suffered from 1.8 ± 1.7 types of DTs. The most frequent manifestations of DTs were: eyes (tightening resembling blepharospasm 84.3%, n = 129 and oculogyric crisis 45.8%, n = 70), trunk (dystonic postures 59.5%, n = 91), jaw (bruxism 34.6%, n = 53), neck (30.7%, n = 47), upper limb (26.1%, n = 40), and foot (20.9%, n = 32). Multivariate logistic regression analysis showed significant associations of DTs with the total number of simple, and the total number of complex, tics.

Conclusions and clinical implications. DTs are early and frequent symptoms of GTS. They tend to localise in the facial area. DTs occur more frequently in individuals with a higher number of tics and probably add to the global impairment caused by tics.

Key words: Gilles de la Tourette syndrome, dystonic tics, simple tics, complex tics, dystonia

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Introduction

Gilles de la Tourette syndrome (GTS) is a childhood onset disorder characterised by motor and vocal tics. In up to 90% of patients, psychiatric comorbidities also occur. Different types of motor tics have been recognised as part of GTS symptomatology. Tics are generally brief. They can be categorised as clonic (less than 100 ms) or dystonic and tonic (more than 300 ms). Dystonic tics (DTs) are less common than clonic tics and are characterised by a repetitively abnormal posture similar to dystonia (e.g. torticollis). In tonic tics, there is a relatively long duration of contraction (in e.g. tension of abdominal muscles) but without exhibiting any abnormal postures.

DTs are known as a part of GTS symptomatology [1], but studies dedicated exclusively to DTs in GTS are scarce. Two reports describing this phenomenon were published by Jankovic and Stone in 1991 [2] and 1994 [3]. In the first study, 156 GTS patients were included and 57% of them had DTs.

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The authors examined the patients not only clinically but in some cases, when DTs were present at the time of examination, they also used surface electromyography (EMG). The group was divided into two subgroups i.e. only clonic tics, and both clonic and dystonic tics. The patients with DTs had more head trauma, were suffering more frequently from attention deficit hyperactivity disorder (ADHD), and had more relatives who had tics. The next study dedicated to DTs was published by Jankovic in 1994 and focused on the treatment of DTs with botulin toxin. However, this study included just 10 patients, five with DTs that involved the eyes and five involving the neck muscles. This study focused merely on the effectiveness of botulin toxin.

A slightly different entity was described recently by Erro et al. [4]. They presented 11 patients with adult-onset primary DTs, which seems to be an intermediate disorder between tics and dystonia because it bears some characteristics of tics (localisation) and some of dystonia (sensory gestures, progression to dystonia in one case).

Finally, there have been two publications regarding co-existing tics and dystonia [5, 6]. Stone and Jankovic [5] described nine patients who developed dystonia additionally to their vocal and motor tics. Importantly, these patients were not treated with neuroleptics to exclude possible tardive dystonia. However, the authors underlined that the co-existence of both diseases could be mere chance due to the very small sample. Twenty years later, in 2011, Damásio et al. [6] analysed a group of 224 patients with tics; 20 of them developed dystonia. The group with dystonia was clearly distinguished from the patients without it, and the authors characterised the syndrome of a primary condition of tics associated with persistent focal/segmental dystonia. The subjects with dystonia had later age of onset, lesser severity of tics, and lower frequency of associated features such as depression, obsessive compulsive symptoms (OCS) and ADHD.

Clinical rationale for the study

Although DTs have been recognised as part of GTS symptomatology, nonetheless little is still known about how often and at what age they appear in affected individuals, or if their occurrence is related to any clinical factors. Moreover, previous reports were only based on very small samples. Our study aimed to answer all of the above questions and confirm previous findings in a larger group of patients.

Material and methods

Study participants

Our cohort of GTS cases comprised 207 consecutive ambulatory patients aged 5–50 years (mean age: 16.5 ± 9.4 years; 162 males). The subjects were evaluated from 2013 to 2018. 131 children (62.6%, mean age: 10.4 ± 3.1 years), and 76 adults (mean age: 27.2 ± 7.4 years) were enrolled. The mean age at tic onset was 6.4 ± 2.7 years. Duration of GTS was 4.9 ± 3.0 years (range: 0–13) in children and 18.3 ± 7.3 years (range: 6–39) in adults. 167 (80.7 %) patients had at least one psychiatric co-morbidity. To assess current tic severity, we used the Yale Global Tic Severity Scale (YGTSS) [7, 8]. The worst lifetime tic severity was investigated and each patient was asked to determine whether their peak tic severity was mild, moderate, or severe. The total number of tics was measured using the YGTSS: for each simple or complex vocal and motor tic, the patient received one point. The patients were evaluated for the clinical diagnosis of GTS and co-morbid mental disorders according to the Diagnostic and Statistical Manual of Mental Disorders (DSM-IV-TR). We questioned in order to determine different types of tics experienced by patients. OCS was diagnosed if obsessions and compulsions were egosyntonic, as opposed to egodystonic symptoms which characterise obssesive-compulsive disorder (OCD). The diagnosis of co-morbid mental disorders was also established on the basis of earlier psychiatric examinations that had been performed before the patient had been evaluated. This allowed us to address psychiatric disorders that are usually diagnosed in childhood (e.g. ADHD or oppositional defiant disorder) and also those for which symptoms were not yet present in adult patients at the time of examination. However, this applied only to a few subjects. We counted the total number of psychiatric disorders for each patient (defined according to DSM-IV-TR, the following disorders: ADHD, OCD, depression, anxiety disorder, conduct disorder, oppositional defiant disorder, pervasive developmental disorder, and learning disorder). Only the mental diseases classified in DSM were added to the total number of psychiatric disorders. All the patients were referred to neurologists experienced in movement disorders and were personally interviewed by the author of the study (PJ). The study was designed as a one-time registration study, and no new clinical data obtained on follow-up visits was included in the analysis.

The collection of clinical data from patients with GTS was approved by the Ethics Committee of the Medical University of Warsaw (KB/2/2007).

Definition and differentiation of DTs

DTs were defined as sustained contractions of muscles that led to abnormal, short-lasting, unfixed postures. We explained to the patients the differences between jerk-like movements (clonic tics) and slower and sustained DTs to facilitate the differentiation between those two movements. DTs were mainly differentiated into clonic tics, dystonia and tonic tics. As described by Jankovic [9], clonic tics are brief and jerk-like, involve only a single muscle or a group of muscles, and last less than 100 ms. Tonic tics consist of isometric muscle tension, which is typically brief tension of abdominal or limb muscles that lasts more than 300 ms. Finally, dystonia is abnormal posturing that ends in fixation, cannot be controlled voluntarily, and sometimes resolves after some gestures (*gestes*

antagonistes). Patients and/or their caregivers were asked by the interviewer whether they had experienced such symptoms in the past or were experiencing them currently. We defined DTs as a current symptom when it had been present during the previous seven days, analogically to tic evaluation according to the YGTSS. In some cases, DTs were also witnessed at the time of examination or the diagnosis was supported by a video recording that had been provided by the subject or her/his family, or was recorded during the clinical evaluation. We additionally collected information about the anatomical region affected by DTs, and showed the patients examples of characteristic movements localised in each region (e.g. strong contraction of eyelid muscles resembling blepharospasm, slow head turning toward the arm similar to torticollis). As indicated by Ella et al. [10] and following Jankovic and Stone [2], we used the term 'bruxism' to describe DTs involving the mandibular region.

Statistical analysis

The statistical analyses were performed using STATISTICA ver 13.1 and SPSS ver 25 software. Normality of distribution was assessed using the Shapiro-Wilk test. For parametric variables, data was presented as arithmetic means and standard deviations (mean ± SD). For non-parametric variables, data was presented as median and quartiles (25; 75). Categorical variables were presented as frequencies (percentages). The parametric data was compared by an independent *t*-test and the nonparametric data by the Mann-Whitney U-test, as appropriate, and the categorical data was compared by the Fisher's exact test (two-sided).

In both analyses, comparisons between groups were considered significantly different when a two-tailed test was $p < 0.05$. All variables that were significant in the primary analyses were entered into a logistic regression analysis in order to determine risk factors for DTs in GTS patients. In addition, gender and age were entered into the multivariate model as control variables.

Results

DTs occurred at some point in the lifetime of 73.9% (n = 153) of patients, in 57 adults and 96 children. In 41.2% of patients (n = 63), DTs had only been present in the past and not at the time of evaluation. The prevalence of DTs in adults and children was almost the same (p = 0.963, 75%, n = 57 *vs.* 73.2%, n = 96, respectively). Age at onset of DTs was known only in 57% (n = 86) of patients, with the most frequent onset in children (7–11 years, 74.4%, n = 64), followed by adolescence (12–18 years; 17.4%, n = 15) and adulthood (≥ 18 years, 8.1%, n = 7). The age of onset was known in 23 adults (23/57, 40.4%) and in 63 children (63/96, 65.6%). Mean age of DTs onset was 9.9 ± 5.2 years compared to 6.4 ± 2.8 years for GTS onset. On average, patients suffered from 1.8 ± 1.7 types of DTs. The most frequent localisations of DTs were: eyes (tightening resembling blepharospasm 84.3%, n = 129 and oculogyric crisis 45.8%, n = 70), trunk (dystonic postures 59.5%, n = 91), jaw (bruxism 34.6%, n = 53), neck (30.7%, n = 47), upper limb (26.1%, n = 40), and foot (20.9%, n = 32).

In univariate analyses, patients with DTs suffered from more severe tics, had more simple and complex tics, and more psychiatric comorbidities (ADHD, OCD, depression, anxiety disorder, conduct disorder, pervasive developmental disorder, oppositional defiant disorder, and learning disorder). Multivariate logistic regression analysis showed only significant associations of DTs with total number of simple and total number of complex tics. The results of univariate und multivariate analysis are shown in Table 1.

Discussion

Our study is one of the very few to have reported about DTs in GTS. Previous reports date back to the 1990s and include smaller study samples. In our sample, DTs were found in the majority of patients (73.9%), they started mainly in childhood and adolescence, were located in the face, and correlated with the total number of tics.

In the publication by Jankovic and Stone from 1991 [2], the prevalence of DTs was lower than in our group. This could be related to a different methodology. Jankovic and Stone used additionally EMG in some cases to determine the exact duration of the tic. Clonic tics last less than 100 ms while dystonic last more than 300 ms. However, they used this measurement only in some patients and did not report whether the amount of patients with DTs had changed after using surface EMG. We can only suspect that the amount of patients categorised as DTs+ in our sample could be liable to change if one were to apply surface EMG. Moreover, Jankovic and Stone used DSM III criteria to diagnose GTS and their sample was smaller than ours (156 *vs.* 207). In the study by Jankovic and Stone the patients with DTs had more ADHD and head trauma. In our sample, univariate analysis showed that patients with DTs were more affected by tics as well as having more psychiatric comorbidities, but multivariate regression confirmed only a relationship with the number of motor and vocal tics.

The age at onset was known in only 57% of patients. Unsurprisingly, it was better known in children (65.6%) than in adults (40.4%). This could be related to the fact that usually DTs start in childhood or adolescence and that therefore some adults could not recall when DTs had started due to the long interval between the onset of DTs and the clinical evaluation. Additionally, in children with DTs, the medical history was always supported by information given by at least one parent.

Although we did not use objective measurements to assess impairment due to tics, we believe that DTs may add to global impairment, which was confirmed by correlating with the YGTSS. We speculate that also a correlation between the total number of tics and lifetime peak tic severity indicates that such patients have more tics and, in consequence, this can also

Table 1. Characteristics and associations of DTs

General comparison between DTs+ and DTs- groups			
Variable	**GTS patients (n = 203)**		
	Patients with DTs (n = 153)	**Patients without DTs (n = 54)**	**p**
Age at evaluation [years] [median] (IQR)	14 (9–22)	11 (9–23)	0.60
Gender (male/female)	124/29	38/16	0.102
YGTSS [median] (IQR)	50 (31–65)	35 (21–49)	**0.0004**
Total number of simple tics [mean ± SD]	13.73 ± 4.76	9.36 ± 3.92	**0.000**
Total number of complex tics [median] (IQR)	8 (5–13)	3 (1–6)	**0.000**
Number of mental disorders [median] (IQR)	2 (1–3)	1 (0–2)	0.002
Medication for tics [at evaluation]	n = 67 (43.8%)	n = 21 (38.9%)	0.531
Peak tic severity [median] (IQR)	2 (2–3)	2 (1–2)	0.000
Family history of tics or GTS	n = 35 (22.9%)	n = 13 (24.1%)	0.934
Family history of OCD or OCS	n = 52 (34.0%)	n = 15 (27.8%)	0.276
Logistic regression model			
		Multivariate Analysis	
Variable		**OR (95% CI)**	**p**
Age		1.019 (0.972–1.068)	0.429
Gender		0.755 (0.315–1.813)	0.529
Peak tic severity		1.509 (0.716–3.180)	0.279
YGTSS		0.974 (0.907–1.046)	0.472
Total number of simple tics		1.174 (1.042–1.323)	0.008
Total number of complex tics		1.248 (1.082–1.440)	0.002
Number of mental disorders		0.854 (0.629–1.160)	0.312

n — number of patients; IQR — interquartile range; SD — standard deviation; OR — odds ratio; CI — confidence interval; DTs — dystonic tics; GTS — Gilles de la Tourette Syndrome; YGTSS — Yale Global Tic Severity Scale; OCD — obsessive-compulsive disorder; OCS — obsessive-compulsive symptoms; p — DTs+ *vs.* DTs-; p value < 0.05 are shown in bold. For non-parametric variables, data is presented as median and interquartile range. Categorical variables are presented as frequencies (percentages)

aggravate global impairment in some patients. Nevertheless, patients with DTs did not receive more frequently medication for tics.

Similarly to other studies, also in our group, DTs tended to localise in the head and neck area. This localisation is also typical for clonic tics. Interestingly, in a group of patients with co-existing dystonia and tics described by Damásio et al., the most frequent was cervical dystonia [6].

DTs must be differentiated from primary dystonia. The main difference is that while DTs are characterised by a temporarily sustained posture, in dystonia the abnormal posturing is fixed. Also, the dystonia is constant, with a fixed pattern of movement, while DTs wax and wane in intensity and localisation, as do all tics in GTS. Helpful clues based on clinical observations that could help to differentiate DTs from dystonia are summarised in Table 2.

Table 2. Distinguishing features between dystonic tics and dystonia

Parameter	DTs	Dystonia
Male predominance	+	-
Co-occurrence of phonic and clonic tics	+	-
Suppressibility	+	-
Growing inner tension during suppression	+	-
Premonitory urge	+	-
Waxing and waning course	+	-
Amelioration in adulthood	+	-
Geste antagoniste	-	+
Fixed posture	-	+
Unchanged pattern	-	+

DTs — dystonic tics

None of the patients in our sample had a history of dystonia. Nevertheless, Damásio et al. [6] detected 20 cases of dystonia (8.9%) in his sample of 224 patients with tics. The group of patients included in this study was very heterogenous, some of the patients had tic onset in childhood, some in adulthood, plus patients with GTS but also chronic tic disorder were enrolled. This could possibly bias the results. Previously, Stone and Jankovic [5] presented nine cases (5%) of GTS and dystonia. It seems to be, therefore, a quite rare phenomenon, but one that should be taken into consideration. Nevertheless, the authors mention that those numbers could be biased as only severely affected patients are treated in Movement Disorders Centers, where their study was conducted.

The second disorder that can resemble DTs is tardive dyskinesia, especially tardive dystonia. Tardive dyskinesia is a disorder characterised by involuntary movements, typically of the orofacial muscles and also of the extremities and other muscle groups. The condition is associated with exposure to dopamine receptor blocking agents, including antipsychotics that are commonly used in the treatment of tics. Among psychiatric patients, the risk of developing tardive dyskinesia is quite high, especially when treated with first generation antipsychotics and when this treatment is long-lasting. Correll et al. reported the annual prevalence of tardive dyskinesia as being as high as 3.9% for first generation antipsychotics and 5.5% for second generation antipsychotics [11]. Müller-Vahl and Krüger [12] analysed records of 521 GTS patients treated with neuroleptics. None of them developed tardive dyskinesia, but most patients were treated with neuroleptics for less than a year. Although tardive dyskinesia seems to be a quite rare side effect in GTS, it should be considered. To distinguish tardive dystonia from DTs we should consider the exposure to neuroleptics. Tardive dystonia usually persists for longer, even after discontinuation of the offending drug. DTs are tics, so their intensity changes, and they can even disappear suddenly. DTs are not permanent, but there are periods of remission and relapse. Furthermore, slow tapering should improve, while sudden dose reduction may aggravate, drug-induced dystonia.

The next type of tics that should be taken into consideration in a differential diagnosis of DTs are tonic tics. Similarly to DTs, tonic tics are slow, but they do not lead to sustained posturing. Typical tonic tics are located in the abdominal or limb muscles. It is often unclear whether the subject is suffering from DTs or tonic tics, and it is crucial to explain the difference to the patient. However, we did not find any correlation between DTs and tonic tics ($p = 0.103$).

Finally, Erro et al. [4] suggested that patients who develop DTs during adulthood represent a totally different group which they named adult-onset primary dystonic tics. From their database of 253, they found 21 with DTs and 10 of them developed DTs after the age of 18 as the first manifestation of an unspecified tic disorder. Some of those movements were evaluated posteriorly to dystonia, so the authors suggested that adult onset primary dystonic tics represent a transient entity between tics and dystonia. In our sample, only seven patients had DTs onset in adulthood (7/153, 4.6%). To the best of our knowledge, none of them developed dystonia or had other features that resembled dystonia such as *geste antagoniste*.

Clinical implications/future directions

DTs are frequent and early symptoms in GTS and should be considered by clinicians. Therefore, we believe that our study has an important clinical value. DTs normally appear in the facial region. DTs occur in patients who have more tics and they probably aggravate the impairment caused by tics. They should be differentiated from primary focal/segmental dystonia, tonic tics and tardive dystonia. We should therefore take into account that patients with DTs may require more clinical attention due to greater impairment. As indicated by Jankovic [3], botulin toxin could also be used in the treatment of DTs.

Limitations

The biggest limitation of our study is the recall bias. Many adult patients did not remember the presence or the age of onset of DTs in their childhood, and only in some cases was it possible to obtain this additional information from a family member. We also did not use superficial EMG to make the assessment more objective. Another objective method, video recording, was available only in some cases in our study. The design of our one-time registration study may have also influenced the prevalence of DTs. Moreover, we used different methods of data collection from children as opposed to adults. Most clinical information regarding children was provided by their parents, whereas adults reported themselves.

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References

1. Cath DC, Hedderly T, Ludolph AG, et al. ESSTS Guidelines Group. European clinical guidelines for Tourette syndrome and other tic disorders. Part I: assessment. Eur Child Adolesc Psychiatry. 2011; 20(4): 155–171, doi: 10.1007/s00787-011-0164-6, indexed in Pubmed: 21445723.
2. Jankovic J, Stone L. Dystonic tics in patients with Tourette's syndrome. Mov Disord. 1991; 6(3): 248–252, doi: 10.1002/mds.870060309, indexed in Pubmed: 1922130.
3. Jankovic J. Botulinum toxin in the treatment of dystonic tics. Mov Disord. 1994; 9(3): 347–349, doi: 10.1002/mds.870090315, indexed in Pubmed: 8041378.
4. Erro R, Martino D, Ganos C, et al. Adult-Onset Primary Dystonic Tics: A Different Entity? Mov Disord Clin Pract. 2014; 1(1): 62–66, doi: 10.1002/mdc3.12005, indexed in Pubmed: 30363833.

5. Stone LA, Jankovic J. The coexistence of tics and dystonia. Arch Neurol. 1991; 48(8): 862–865, doi: 10.1001/archneur.1991.00530200104028, indexed in Pubmed: 1898264.
6. Damásio J, Edwards MJ, Alonso-Canovas A, et al. The clinical syndrome of primary tic disorder associated with dystonia: a large clinical series and a review of the literature. Mov Disord. 2011; 26(4): 679–684, doi: 10.1002/mds.23484, indexed in Pubmed: 21506147.
7. Leckman JF, Riddle MA, Hardin MT, et al. The Yale Global Tic Severity Scale: initial testing of a clinician-rated scale of tic severity. J Am Acad Child Adolesc Psychiatry. 1989; 28(4): 566–573, doi: 10.1097/00004583-198907000-00015, indexed in Pubmed: 2768151.
8. Stefanoff P, Wolańczyk T. [Validity and reliability of Polish adaptation of Yale Global Tic Severity Scale (YGTSS) in a study of Warsaw schoolchildren aged 12-15]. Przegl Epidemiol. 2005; 59(3): 753–762, indexed in Pubmed: 16433318.
9. Jankovic J. Phenomenology and classification of tics. Neurologic Clinics. 1997; 15(2): 267–275, doi: 10.1016/s0733-8619(05)70311-x.
10. Ella B, Ghorayeb I, Burbaud P, et al. Bruxism in Movement Disorders: A Comprehensive Review. J Prosthodont. 2017; 26(7): 599–605, doi: 10.1111/jopr.12479, indexed in Pubmed: 27077925.
11. Correll CU, Schenk EM. Tardive dyskinesia and new antipsychotics. Curr Opin Psychiatry. 2008; 21(2): 151–156, doi: 10.1097/YCO.0b013e3282f53132, indexed in Pubmed: 18332662.
12. Müller-Vahl KR, Krueger D. Does Tourette syndrome prevent tardive dyskinesia? Mov Disord. 2011; 26(13): 2442–2443, doi: 10.1002/mds.23894, indexed in Pubmed: 21956454.

RESEARCH PAPER

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Evaluating reflexive saccades and UDPRS as markers of Deep Brain Stimulation and Best Medical Treatment improvements in Parkinson's disease patients: a prospective controlled study

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ABSTRACT

Introduction. To date, there has been no clear evidence regarding the evaluation of saccades as a monitoring tool of motor impairment in Parkinson's disease (PD) Subthalamic Nucleus Deep Brain Stimulation (STN-DBS) patients. The aim of this study was to evaluate the long-term impact of STN-DBS and pharmacological treatment on reflexive saccades' (RS) parameters and UPDRS alterations.

Material and methods. The DBS group consisted of 20 PD patients who underwent bilateral STN-DBS. The Postoperative (POP) group consisted of 14 post-DBS patients. The Best Medical Therapy (BMT) group consisted of 20 patients on pharmacotherapy only. RS parameters and the UPDRS scale were measured during three visits in four phases of treatment (i.e. BMT-ON/OFF, DBS--ON/OFF).

Results. The significant UPDRS III and UPDRS. Total improvements were observed in all three study groups ($p < 0.05$), but RS latency improvement was stated only in the DBS group in the DBS-ON phase ($p < 0.05$). A significant correlation between RS latency increase and UPDRS III score worsening was found in all study groups, with the most evident effect in the UPDRS III ON phase ($p < 0.05$).

Conclusion. RS parameters correlated with UPDRS III outcomes during the postoperative period in DBS-STN patients. Therefore, saccadic evaluation may be a good biomarker of the patient's response to surgical and/or pharmacological treatment.

Key words: Parkinson's disease, BMT, DBS, reflexive saccades, marker

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Introduction

Deep brain stimulation (DBS) has become the standard surgical procedure in Parkinson's disease (PD) patients, particularly in advanced stages of the disease and with complications after levodopa therapy. The subthalamic nucleus (STN) is the most often chosen localisation because of the impact on most of the motor symptoms, particularly tremor, bradykinesia and rigidity [1–3] as well as because of the possibility of decreasing the daily levodopa dose [4].

STN is a part of the saccadic system, the impairment of which influences other structures implicated in the generation of saccades, provoking alterations of saccadic movements [5–7]. PD patients present abnormalities in random saccades, reflexive saccades as well as antisaccades or smooth pursuit movements [8–10]. STN-DBS has also been shown to have

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an observable impact on saccades' parameters such as latency, velocity, amplitude, gain or accuracy [11–17]. Levodopa or dopamine agonist treatment has also been shown to have a possible influence on saccadic movements, but changes in ON-Levodopa state are not clear [18–23], which makes the existing evidence more conflicting. Nevertheless, reflexive saccades' (RS) evaluation is a simple method of assessing the possible influence of DBS or pharmacotherapy on the saccadic system, which can be due to alteration or alleviation of the balance between direct and indirect dopamine pathways [24, 25].

All randomised studies comparing the quality of life of PD patients after STN-DBS implantation to the group of PD patients treated with best medical therapy (BMT) in advanced [26–28] or early PD [29] have revealed clinically meaningful improvements of PDQ-39 score and UPDRS-II, UPDRS-III Stim ON evaluation in DBS patients as compared to the BMT-group in a 6-month [26, 27], 12-month [28], or 24-month [29] re-assessment. None of the randomised trials [26–29] has compared the mean change in UPDRS-III score between the BMT-group and DBS patients in a full OFF phase (BMT-OFF/DBS-OFF) after DBS implantation — all of the studies have evaluated the BMT-OFF/DBS-ON phase only.

The aim of this study was to evaluate the impact of bilateral STN-DBS and pharmacological treatment on changes in reflexive saccades' (RS) parameters and the UPDRS scale [30] in four phases of treatment (BMT-ON/OFF, DBS-ON/OFF) and to estimate the possible usefulness of eye movement (EM) measurements as a biomarker of PD patients' response to surgical and pharmacological treatment.

Material and methods

Study concept

Patients enrolled to this study were clinically diagnosed as having idiopathic Parkinson's disease and fulfilled the UK Parkinson's Disease Society (UKPDS) Brain Bank criteria [31]. All of the study patients also met the CAPSIT-PD criteria [32] in order to have the qualification criteria for bilateral STN DBS implantation.

The patients were divided into three groups:

1) The BMT (Best Medical Therapy) group: 20 patients (mean age 56.7 years, 11 females, nine males) treated only with pharmacotherapy through the whole time of observation.
2) The DBS (Deep Brain Stimulation) group: 20 patients (mean age 51.1 years, eight females, 12 males) who underwent surgical procedure and pharmacotherapy.
3) The POP (Postoperative) group: 15 patients (mean age 51.4 years, seven females, eight males) who had been operated upon a median time of 30 months before the study began. This group was created in order to estimate any possible long-term motor effect of DBS.

The patients were examined during three visits (V1, V2, V3) made at intervals of 7-11 months. The UPDRS scale and reflexive saccades were evaluated twice during each visit in the BMT-group and preoperative assessment in the DBS-group (BMT-ON and BMT-OFF phase), and four times (Total-ON, DBS-ON/BMT-OFF, DBS-OFF/BMT-ON, Total-OFF) during postoperative evaluations (V2, V3) in the DBS group, and during all visits (V1, V2, V3) in the POP group.

The characteristics of the patients are set out in Table 1. All of the patients signed informed consents. This study was approved by the Ethics Committee of the Medical University of Warsaw. The experiments were conducted in accordance with the ethical standards of the Declaration of Helsinki.

UPDRS examination and saccadic assessment

The motor evaluation of the patients was performed by a neurologist experienced in movement disorders, using the UPDRS scale and saccadometry. The assessment was conducted in different phases of treatment: BMT-ON/DBS-ON when patients were on/off antiparkinsonian drugs and (in postoperative evaluation) with both stimulators switched on/off.

Saccadometry was evaluated using a head-mounted saccadometer (Ober Consulting, Poznan, Poland), which analyses binocular infra-red reflections from each eye. The saccadic step task paradigm was used with 20 calibration saccades followed by 50 random horizontal points projected in a random fore-period of 0.5–1.5s, which were always preceded by fixation central points. The parameters analysed were: saccadic latency [ms], saccades' amplitude [deg], and peak of velocity of saccades [deg/s]. The data was analysed using LatencyMeter software, version 6.9.

Surgical procedure

All of the patients who underwent surgery qualified for bilateral subthalamic nucleus deep brain stimulation

Table 1. Study population characteristics

	BMT group	DBS group	POP group
Gender	11 F, 9 M	8 F, 12 M	7 F, 8 M
Mean age	56.7 ± 15.4 years	51.1 ± 15.3 years	51.4 ± 8.7 years
Mean age at onset	46.3 ± 15.1 years	39.7 ± 13.3 years	40.9 ± 8.3 years
Mean symptoms' duration time	10.4 ± 4.9 years	11.3 ± 3.9 years	10.5 ± 3.5 years
Mean LEDD	1,254.0 ± 511.6 mg	1,379.5 ± 510.0 mg	1,273.2 ± 464.3 mg
Mean time of dyskinesia	1.8 ± 2.6 hours / day	4.9 ± 2.9 hours / day	5.9 ± 2.6 hours / day
Mean OFF time	2.7 ± 1.3 hours / day	4.6 ± 3.2 hours / day	4.4 ± 1.8 hours / day

(STN-DBS). The procedure was performed using microrecording and macrostimulation (Leadpoint®, Medtronic) and permanent electrodes (3389-28, Medtronic, Minneapolis, MN, USA) which were connected to internal pulse generators (Activa SC, Medtronic).

Data analysis and statistical assessment

The linear mixed model analysis was implemented by the use of LME4 (version 1.1) with intercepts for subjects included as random effects. Pairwise interactions between each fixed factor were included in the model. Tukey contrasts (from lsmeans package, version 2.25) were used to compare results between timepoints and treatments [33]. All calculations were performed in statistical computing software R (version 3.3) [34]. P values < 0.05 were considered significant.

Results

The mixed model analysis of RS showed a significant inter-phase latency difference ($p < 0.05$) with a visible relation in inter-visit changes in BMT-ON and DBS-ON phases ($p = 0.1$). The pharmacotherapy (levodopa and other dopaminergic treatment) did not significantly influence the saccades latency ($p > 0.05$). On the other hand, bilateral STN-DBS ON significantly improved RS latency (regardless of pharmacotherapy phase) in both (short-term and long-term) postoperative groups ($p < 0.05$). There was also statistically significant amplitude reduction in the first postoperative (ΔV2-V1) Total-OFF evaluation in DBS group ($p < 0.05$), not observed in other groups and other inter-visit assessments. The same results were also found in saccades' peak velocity: no statistically significant inter-phase or inter-visit changes in RS peak velocity, other than a definite reduction in peak velocity of saccades in the first postoperative (ΔV2-V1) examination of the DBS group in Total-OFF phase ($p < 0.05$) (Fig.1C–E).

The mixed model analysis of inter-phase UPDRS III score alterations was statistically significant ($p < 0.05$), as was the analysis of UPDRS III score alterations among visits in all three groups of patients ($p < 0.05$). The inter-phase analysis showed a significant improvement in UPDRS III score in Total-ON phase (compared to Total-OFF phase) in all three study groups ($p < 0.05$) (Fig. 1A). The improvement was observed in consecutive visits in all study groups ($p < 0.05$), with a more visible impact of the STN-DBS procedure. The mean inter-phase change was more evident in both postoperative groups in phases with stimulators switched ON (DBS and POP group) ($p < 0.05$) than in BMT-ON only phase (with stimulators switched off).

The analysis of inter-phase UPDRS TOTAL score was also statistically significant ($p < 0.05$) as were UPDRS TOTAL score changes among visits in the BMT, DBS and POP groups ($p < 0.05$). The improvement of UPDRS TOTAL score was observed in all three groups of patients, with the most evident effect of STN-DBS procedure in the short-term postoperative DBS group ($p < 0.05$), but was not observed in long-term postoperative assessment (POP-group, $p > 0.05$). The mean inter-phase alterations in the UPDRS TOTAL score were observed in phases with stimulators switched ON (DBS and POP group) rather than in phases with BMT-only ON ($p < 0.05$) (Fig. 1B).

The mixed model analyses between inter-phase and inter-visit UPDRS III and saccades' latency, amplitude and peak velocity in the DBS, BMT and POP groups were performed to compare the possible usefulness of both methods of assessment

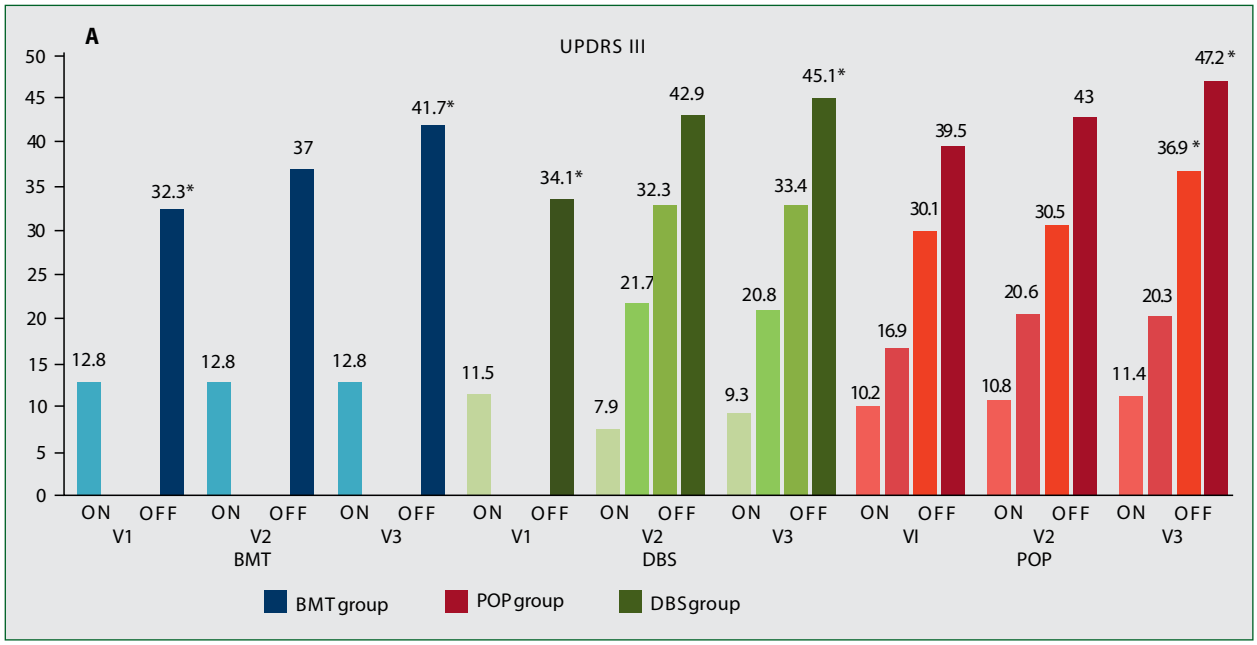

Figure 1. A. UPDRS III

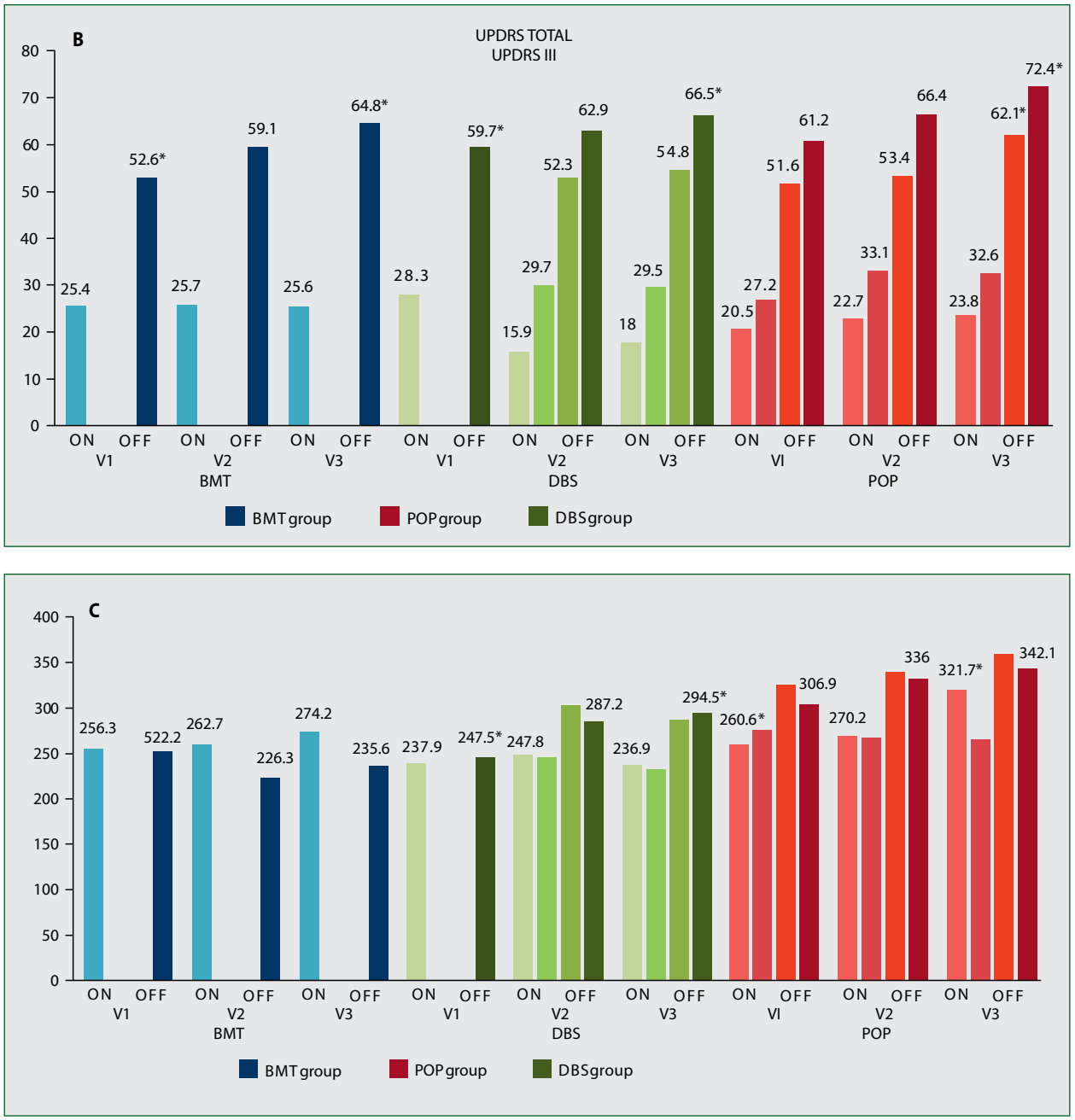

Figure 1. B. UPDRS TOTAL, **C.** mean RS latency

in operative and non-operative PD patients. The analyses revealed a statistically significant correlation between RS latency increase and UPDRS III score worsening in all study groups, with the most evident effect in the UPDRS III ON phase ($p < 0.05$). Such a clear correlation between UPDRS III and RS amplitude and / or RS peak velocity was not demonstrated ($p > 0.05$).

Inter-visit PDQ-39 and AIMS evaluations in all study groups were also carried out to compare the influence of surgical procedure and pharmacotherapy on the patients' quality of life and the level of dyskinesia intensity. The analyses showed a significant improvement of quality of life and a decrease of AIMS in the short-term postoperative DBS group ($p < 0.05$), which was not observed either in the BMT or the POP group ΔV3-V1 assessment ($p > 0.05$).

Discussion

The motor improvement of PD patients after STN-DBS has been previously proven in randomised trials [26–29], but to date there has been no clear evidence on UPDRS and RS application as biomarkers of STN-DBS treatment.

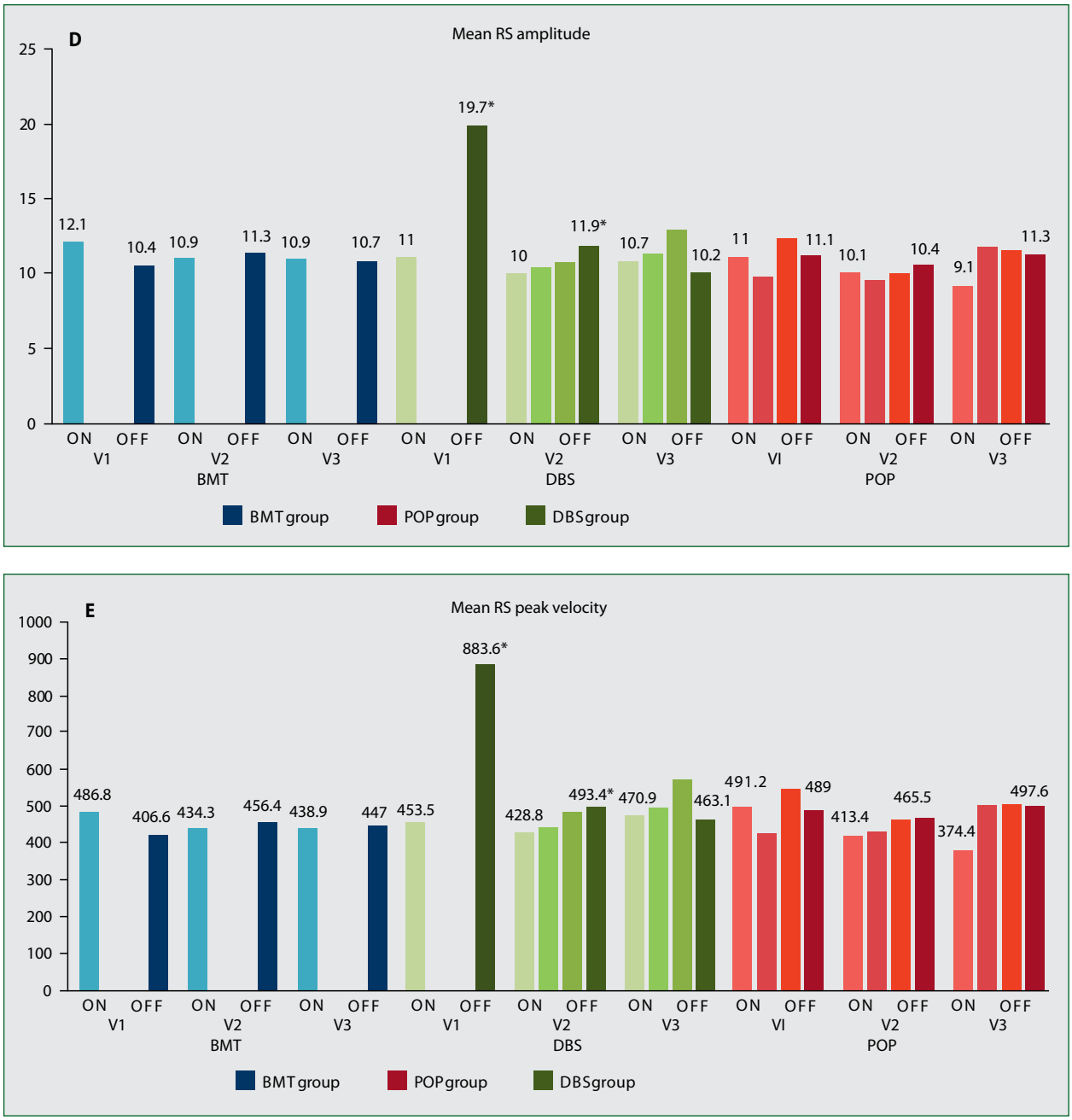

Figure 1. D. mean RS amplitude, **E.** mean RS peak velocity in BMT, DBS and POP group (V1, V2, V3). Consecutive phases: Total-ON, DBS-ON/BMT-OFF, DBS-OFF/BMT-ON, Total-OFF * $p < 0.05$

In all previous randomised studies, the authors proved a great improvement in UPDRS III DBS-ON BMT-OFF evaluation in patients after STN-DBS surgery, and a significant impact of STN-DBS procedure on quality of life in STN-DBS patients [26–29], but none of them compared total OFF phase in the DBS group in consecutive examinations.

In contrast, our study completed this comparison in all four treatment phases in order to establish the full impact of STN-DBS on UPDRS-III and saccadic alterations in BMT/STN-DBS PD patients.

Saccadometry was first described as a possibly useful clinical tool for the quantification of the motor effects of STN-DBS in PD in 2009 by Temel et al. [17], but there were also some other prior studies on monkeys and humans which showed a potential use of this method as a parametric tool in the assessment of motor changes in PD treatment [8, 11, 15, 16, 18–23]. Because of the fact that STN is a part of the saccadic system, various methods of treatment which influence its stimulation can result in alterations of saccadic movements. Therefore we assessed reflexive saccades' parameters in DBS

ON/OFF and BMT ON/OFF in order to estimate the possible correlation between motor improvement in UPDRS III scale and reflexive saccades' variations. Our results are consistent with previous studies, and show a greater improvement in RS latency in the DBS-ON phase (compared to the OFF-phase) rather than in the BMT-ON phase (compared to the BMT--OFF), which may also be an indicator of a more significant effect of STN-DBS, rather than pharmacotherapy, on saccadic system alterations. The other clinical problem of using saccadic movements' assessment as a parametric evaluation of motor deficits in PD patients under various methods of treatment is a lack of information concerning the rate of progression of saccadic alterations in PD. The only assessment has been performed by Antoniades et al. [13] who examined nine PD patients during four visits in order to observe the variations of saccades, which were anomalous: first postoperative assessment in OFF-phase revealed deterioration which was later improved in the following assessment [13], possibly due to astroglial neuroinflammatory reaction to stimulation confirmed in recent animal studies [35–37].

Conclusions

The definite improvement of RS latency with significant correlations to UPDRS III and UPDRS TOTAL score improvement in DBS-ON phase in both DBS (short and long term postoperative) groups with a co-existent non-significant improvement in RS latency (but with preserved significant UPDRS III and UPDRS TOTAL improvement) in the BMT group in the ON phase may suggest that BMT interferes mostly with the dopaminergic system, while STN-DBS may affect other systems as well.

Our results show that the application of RS measurement as a parametric tool (apart from UPDRS III assessment, which is a subjective scale) may be a good prognostic indicator of STN-DBS and pharmacological treatment effect on PD patients' motor outcome and quality of life. The limitations of our study, i.e. the study group sizes and the restricted duration of the study, may necessitate a prolonged assessment on larger populations in order to confirm the quality of these results.

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Abbreviations:

PD — Parkinson's disease
RS — reflexive saccades
DBS — deep brain stimulation
STN-DBS — subthalamic nucleus deep brain stimulation
BMT-group — Best Medical Treatment group
POP-group — Postoperative group
UPDRS — Unified Parkinson's Disease Rating Scale
LEDD — Levodopa equivalent daily dose

References

1. Krack P, Pollak P, Limousin P, et al. Stimulation of subthalamic nucleus alleviates tremor in Parkinson's disease. Lancet. 1997; 350(9092): 1675, doi: 10.1016/s0140-6736(97)24049-3, indexed in Pubmed: 9400514.
2. Krack P, Batir A, Van Blercom N, et al. Electrical stimulation of the subthalamic nucleus in advanced Parkinson's disease. N Engl J Med. 1998; 339(16): 1105–1111, doi: 10.1056/NEJM199810153391603, indexed in Pubmed: 9770557.
3. Limousin P, Pollak P, Benazzouz A, et al. Bilateral subthalamic nucleus stimulation for severe Parkinson's disease. Mov Disord. 1995; 10(5): 672–674, doi: 10.1002/mds.870100523, indexed in Pubmed: 8552123.
4. Krack P, Limousin P, Benabid AL, et al. Chronic stimulation of subthalamic nucleus improves levodopa-induced dyskinesias in Parkinson's disease. Lancet. 1997; 350(9092): 1676, doi: 10.1016/s0140-6736(05)64273-0, indexed in Pubmed: 9400515.
5. Fawcett AP, Dostrovsky JO, Lozano AM, et al. Eye movement-related responses of neurons in human subthalamic nucleus. Exp Brain Res. 2005; 162(3): 357–365, doi: 10.1007/s00221-004-2184-7, indexed in Pubmed: 15599721.
6. Fawcett AP, Cunic D, Hamani C, et al. Saccade-related potentials recorded from human subthalamic nucleus. Clin Neurophysiol. 2007; 118(1): 155–163, doi: 10.1016/j.clinph.2006.09.016, indexed in Pubmed: 17097341.
7. Sieger T, Bonnet C, Serranová T, et al. Basal ganglia neuronal activity during scanning eye movements in Parkinson's disease. PLoS One. 2013; 8(11): e78581, doi: 10.1371/journal.pone.0078581, indexed in Pubmed: 24223158.
8. Chan F, Armstrong IT, Pari G, et al. Deficits in saccadic eye-movement control in Parkinson's disease. Neuropsychologia. 2005; 43(5): 784–796, doi: 10.1016/j.neuropsychologia.2004.06.026, indexed in Pubmed: 15721191.
9. Choi SM, Lee SH, Choi KH, et al. Directional asymmetries of saccadic hypometria in patients with early Parkinson's disease and unilateral symptoms. Eur Neurol. 2011; 66(3): 170–174, doi: 10.1159/000330671, indexed in Pubmed: 21894020.
10. Pinkhardt EH, Jürgens R, Lulé D, et al. Eye movement impairments in Parkinson's disease: possible role of extradopaminergic mechanisms. BMC Neurol. 2012; 12: 5, doi: 10.1186/1471-2377-12-5, indexed in Pubmed: 22375860.
11. Rivaud-Péchoux S, Vermersch AI, Gaymard B, et al. Improvement of memory guided saccades in parkinsonian patients by high frequency subthalamic nucleus stimulation. J Neurol Neurosurg Psychiatry. 2000; 68(3): 381–384, doi: 10.1136/jnnp.68.3.381, indexed in Pubmed: 10675227.
12. Yugeta A, Terao Y, Fukuda H, et al. Effects of STN stimulation on the initiation and inhibition of saccade in Parkinson disease. Neurology. 2010; 74(9): 743–748, doi: 10.1212/WNL.0b013e3181d31e0b, indexed in Pubmed: 20194913.
13. Antoniades CA, Buttery P, FitzGerald JJ, et al. Deep brain stimulation: eye movements reveal anomalous effects of electrode placement and stimulation. PLoS One. 2012; 7(3): e32830, doi: 10.1371/journal.pone.0032830, indexed in Pubmed: 22427894.
14. Nilsson MH, Patel M, Rehncrona S, et al. Subthalamic deep brain stimulation improves smooth pursuit and saccade performance in patients with Parkinson's disease. J Neuroeng Rehabil. 2013; 10: 33, doi: 10.1186/1743-0003-10-33, indexed in Pubmed: 23551890.
15. Kumru H, Summerfield C, Valldeoriola F, et al. Effects of subthalamic nucleus stimulation on characteristics of EMG activity underlying rea-

ction time in Parkinson's disease. Mov Disord. 2004; 19(1): 94–100, doi: 10.1002/mds.10638, indexed in Pubmed: 14743367.
16. Sauleau P, Pollak P, Krack P, et al. Subthalamic stimulation improves orienting gaze movements in Parkinson's disease. Clin Neurophysiol. 2008; 119(8): 1857–1863, doi: 10.1016/j.clinph.2008.04.013, indexed in Pubmed: 18567536.
17. Temel Y, Visser-Vandewalle V, Carpenter RHS. Saccadometry: a novel clinical tool for quantification of the motor effects of subthalamic nucleus stimulation in Parkinson's disease. Exp Neurol. 2009; 216(2): 481–489, doi: 10.1016/j.expneurol.2009.01.007, indexed in Pubmed: 19320006.
18. Vermersch AI, Rivaud S, Vidailhet M, et al. Sequences of memory-guided saccades in Parkinson's disease. Ann Neurol. 1994; 35(4): 487–490, doi: 10.1002/ana.410350419, indexed in Pubmed: 8154878.
19. Rascol O, Clanet M, Montastruc JL, et al. Abnormal ocular movements in Parkinson's disease. Evidence for involvement of dopaminergic systems. Brain. 1989; 112 (Pt 5): 1193–1214, doi: 10.1093/brain/112.5.1193, indexed in Pubmed: 2804611.
20. Michell AW, Xu Z, Fritz D, et al. Saccadic latency distributions in Parkinson's disease and the effects of L-dopa. Exp Brain Res. 2006; 174(1): 7–18, doi: 10.1007/s00221-006-0412-z, indexed in Pubmed: 16544135.
21. Hood AJ, Amador SC, Cain AE, et al. Levodopa slows prosaccades and improves antisaccades: an eye movement study in Parkinson's disease. J Neurol Neurosurg Psychiatry. 2007; 78(6): 565–570, doi: 10.1136/jnnp.2006.099754, indexed in Pubmed: 17178817.
22. Yugeta A, Terao Y, Fukuda H, et al. Effects of levodopa on saccade performance in Parkinson's disease. Mov Disord. 2008; 23(Suppl. 1): S296.
23. Dec-Ćwiek M, Tutaj M, Gracies JM, et al. Opposite effects of l-dopa and DBS-STN on saccadic eye movements in advanced Parkinson's disease. Neurol Neurochir Pol. 2017; 51(5): 354–360, doi: 10.1016/j.pjnns.2017.06.002, indexed in Pubmed: 28669542.
24. Alexander GE, Crutcher MD. Functional architecture of basal ganglia circuits: neural substrates of parallel processing. Trends Neurosci. 1990; 13(7): 266–271, doi: 10.1016/0166-2236(90)90107-l, indexed in Pubmed: 1695401.
25. Terao Y, Fukuda H, Ugawa Y, et al. New perspectives on the pathophysiology of Parkinson's disease as assessed by saccade performance: a clinical review. Clin Neurophysiol. 2013; 124(8): 1491–1506, doi: 10.1016/j.clinph.2013.01.021, indexed in Pubmed: 23499161.
26. Deuschl G, Schade-Brittinger C, Krack P, et al. German Parkinson Study Group, Neurostimulation Section. A randomized trial of deep-brain stimulation for Parkinson's disease. N Engl J Med. 2006; 355(9): 896–908, doi: 10.1056/NEJMoa060281, indexed in Pubmed: 16943402.
27. Weaver FM, Follett K, Stern M, et al. CSP 468 Study Group. Bilateral deep brain stimulation vs best medical therapy for patients with advanced Parkinson disease: a randomized controlled trial. JAMA. 2009; 301(1): 63–73, doi: 10.1001/jama.2008.929, indexed in Pubmed: 19126811.
28. Williams A, Gill S, Varma T, et al. PD SURG Collaborative Group. Deep brain stimulation plus best medical therapy versus best medical therapy alone for advanced Parkinson's disease (PD SURG trial): a randomised, open-label trial. Lancet Neurol. 2010; 9(6): 581–591, doi: 10.1016/S1474-4422(10)70093-4, indexed in Pubmed: 20434403.
29. Schuepbach WMM, Rau J, Knudsen K, et al. EARLYSTIM Study Group. Neurostimulation for Parkinson's disease with early motor complications. N Engl J Med. 2013; 368(7): 610–622, doi: 10.1056/NEJMoa1205158, indexed in Pubmed: 23406026.
30. Movement Disorder Society Task Force on Rating Scales for Parkinson's Disease. The Unified Parkinson's Disease Rating Scale (UPDRS): status and recommendations. Mov Disord. 2003; 18(7): 738–750, doi: 10.1002/mds.10473, indexed in Pubmed: 12815652.
31. Gibb WR, Lees AJ. The relevance of the Lewy body to the pathogenesis of idiopathic Parkinson's disease. J Neurol Neurosurg Psychiatry. 1988; 51(6): 745–752, doi: 10.1136/jnnp.51.6.745, indexed in Pubmed: 2841426.
32. Defer GL, Widner H, Marié RM, et al. Core assessment program for surgical interventional therapies in Parkinson's disease (CAPSIT-PD). Mov Disord. 1999; 14(4): 572–584, indexed in Pubmed: 10435493.
33. Douglas Bates, Martin Maechler, Ben Bolker, Steve Walker . Fitting Linear Mixed-Effects Models Using lme4. Journal of Statistical Software. 2015; 67(1): 1–48.
34. Lenth R. Least-Squares Means: TheRPackagelsmeans. Journal of Statistical Software. 2016; 69(1), doi: 10.18637/jss.v069.i01.
35. Hirshler YK, Polat U, Biegon A. Intracranial electrode implantation produces regional neuroinflammation and memory deficits in rats. Exp Neurol. 2010; 222(1): 42–50, doi: 10.1016/j.expneurol.2009.12.006, indexed in Pubmed: 20026042.
36. Orlowski D, Michalis A, Glud AN, et al. Brain Tissue Reaction to Deep Brain Stimulation-A Longitudinal Study of DBS in the Goettingen Minipig. Neuromodulation. 2017; 20(5): 417–423, doi: 10.1111/ner.12576, indexed in Pubmed: 28220987.
37. Colangelo AM, Alberghina L, Papa M. Astrogliosis as a therapeutic target for neurodegenerative diseases. Neurosci Lett. 2014; 565: 59–64, doi: 10.1016/j.neulet.2014.01.014, indexed in Pubmed: 24457173.

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Anti-interferon-beta antibodies in Polish multiple sclerosis patients: prevalence and clinical significance in a long-term prospective study

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ABSTRACT

Aim of the study. To determine the prevalence of anti-interferon-β binding (BAb) and neutralising antibodies (NAb), and to investigate whether NAb measured by luciferase-based cell assay can predict treatment response in multiple sclerosis (MS) patients treated with interferon-β-1b (IFNβ-1b).

Clinical rationale for the study. A subgroup of IFNβ-treated MS patients develop NAb directed against the drug. The clinical significance remains controversial, which could be explained to some extent by technical difficulties in NAb detection and quantification. A simple, specific and reproducible test for NAb might help elucidate these uncertainties.

Materials and methods. Sera from 101 consecutive MS patients initiating treatment with IFNβ-1b were collected at baseline and during the first two years, and assessed for BAbNAb with a novel luciferase-based cell assay. Median clinical follow-up lasted 5.1 years.

Results. BAb were present in 97% and NAb in 88% of the study cohort. Unexpectedly, 92% of patients tested positive for BAb and 12.5% for NAb at baseline, before drug exposure. Patients with baseline NAb positivity were more likely to remain free of disease activity in the first three years of treatment. When baseline-positive cases were grouped together with those who remained NAb-negative, and the resulting group was compared to those who became positive after drug exposure, NAb positivity was associated with a higher risk of disease activity during the entire follow-up. Direct comparison of BAb/Nab-positive and BAb/Nab-negative patients only revealed an association of BAb positivity with more active disease after four years of treatment, while NAb failed to predict the outcome.

Conclusions and clinical implications. Antibodies developed after treatment initiation are associated with a worse outcome. Naturally-occurring antibodies appear to predict more benign disease. Their prevalence and specificity require further investigation.

Key words: multiple sclerosis, interferon-beta, neutralising antibodies, treatment outcome

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Introduction

Interferon beta (IFNβ) continues to be commonly used as a first-line disease modifying drug (DMD) in relapsing-remitting multiple sclerosis (RRMS) patients, but its efficacy varies among individual cases.

In response to IFNβ treatment, a proportion of patients develop either binding (BAb) or neutralising anti-IFN antibodies (NAb). IFNβ-1b is thought to be more immunogenic than IFNβ-1a, and a subcutaneous route of administration more immunogenic than an intramuscular route [1]. BAb appears in up to 97% of patients, usually between the third and twelfth months of treatment [1]. NAb appears later (6-24 months) and less commonly (28–42%) [1–3]. For pegylated IFNβ-1A, NAb prevalence has been shown to be below 1% after two years of treatment [4].

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The significance of these findings remains controversial. There is evidence from clinical trials and open-label observational studies that NAb in high titres are persistent [5] and associated with a loss of IFN efficacy [2, 6–8]. In fact, NAb may be the most common cause for early treatment failure in IFNβ-1b-treated patients [9]. In many studies however, Nab's influence was most apparent in NAb-positive periods [3, 10, 11], and a considerable proportion of NAb-positive patients revert to a NAb-negative status over time [3, 10], with restoration of IFN activity [10]. On the other hand, some researchers [8, 12] have observed only sporadic conversion to NAb-negativity and worse outcomes in long term follow-ups.

At least in part, these uncertainties stem from the complexity of NAb measurement. All currently used assays are based on NAb-mediated inhibition of IFN's effects on IFN-responsive cells. The effect can be observed as protection from the cytopathic effect in a viral challenge, as in cytopathic effect assay, a gold standard since 1985 [13], or confirmed via MxA mRNA detection [14]. Attempts to design a non-cellular assay have thus far been unsuccessful.

A new generation of tests uses growth-arrested cells transfected with luciferase under a selective class-I IFN controlled promotor [15]. While this involves live cells, there is no need for continuous cell line culture. Currently, two luciferase-based tests have been validated and are commercially available [16, 17], but no long-term follow-up is available with regards to their prognostic value.

Clinical rationale for the study

We sought to determine the influence of IFNβ NAb, measured by luciferase-based cell assay, on disease activity in RRMS patients treated with IFNβ-1b in a long-term prospective observation, in the hope that their detection could predict drug response and so guide treatment decisions.

Materials and methods

Patients

A total of 101 RRMS patients were recruited consecutively for this study while initiating treatment with IFNβ-1b in the setting of the national MS treatment programme at the outpatient clinic at Heliodor Swiecicki University Hospital in Poznan, Poland, from 2008 to 2013. The study protocol was approved by the Ethics Board of Poznan University of Medical Sciences. Inclusion criteria were: age ≥ 18 years; diagnosis of RRMS according to the 2005 revision of the McDonald criteria [18] (all patients also met the 2010 criteria [19]); no prior disease modifying treatment; fulfillment of the national treatment programme eligibility criteria (Tab. 1); and written informed consent for study participation.

Treatment consisted of interferon beta-1b (Betaferon, n = 96, or Extavia, n = 5) 250μg (8 MIU) subcutaneously every other day.

Baseline clinical information was obtained: sex, age at first relapse, time to second relapse, time to treatment initiation, number of relapses, and EDSS score prior to treatment initiation.

Follow up

The follow up spanned the period from 2008 to 2018 and consisted of monthly neurological assessments.

In addition, 1.5 Tesla head magnetic resonance imaging (MRI, Siemens Avanto, Erlangen, Germany) with a 12-channel head coil, including T1, T2, FLAIR (Fluid Attenuated Inversion Recovery) and PD (Proton Density) sequences, with gadolinium contrast administration, was obtained at baseline and was repeated each year. For individual indications, spinal MRI was performed.

Each year, we recorded the number of relapses, EDSS change, and the presence of new T2/FLAIR or enhancing lesions in MRI.

Definition of NEDA

Consistently with previous works [20], NEDA was defined as no relapses, no disability progression and no MRI activity in a given time period. A relapse was defined as the appearance or worsening of symptoms applicable to multiple sclerosis, with focal neurological abnormality, lasting 24 hours and preceded by ≥ 30 days of neurological stability, in the absence of infection or fever. Progression was recognised when an increase in EDSS occurred and was confirmed after three months. The minimal required increase depended on baseline EDSS: ≥ 1.5 for a baseline score of 0 ≥ 1.0 for scores ranging from 1.0 to 5.0 and ≥ 0.5 for baseline EDSS of ≥ 5.5. New or enlarging lesions in T2/FLAIR or contrast enhancing lesions were considered as MRI activity.

Sampling

Serum samples were collected prior to treatment initiation and again after one, three, six, 12, 24 and 36 months of treatment, then stored at -70°C until further analysis.

The treating and sampling staff were unaware of the patients' antibody status.

Laboratory

Binding antibody detection

We employed a customised indirect ELISA developed at the Department of Neurochemistry and Neuropathology, Poznan University of Medical Sciences, Poznan, Poland [21].

ELISA plates (Nunc, Roskilde, Denmark) were coated with Betaferon (Bayer Pharma AG, Germany) diluted in 0.05M sodium bicarbonate (final concentration: 1 g/mL). The plates were incubated at room temperature for 12 hours, then washed with phosphate-buffered salt (PBS) with 0.05% (vol / vol) Tween 20. Nonspecific binding sites were blocked with 1% bovine albumin solution in PBS-0.05% Tween 20 and the plates were washed with PBS-0.05% Tween 20 again. Then, the

Table 1. National Multiple Sclerosis treatment programme eligibility criteria in Poland 2008–2013

Criterion	2008	2012
Diagnosis	2005 McDonald criteria and contrast-enhanced head MRI consistent with MS	
Disease activity	At least two relapses within the past two years	not specified
Required score	21 pt	15 pt
Scoring system	Age: 16–40 years — 6 pt 40–60 years — 3 pt over 60 years — 1 pt Disease duration: 0–3 years — 6 pt 3–6 years — 3 pt 6–10 years — 2 pt over 10 years — 1 pt RRMS with no neurological deficit — 5 pt. Number of relapses in the last year: 3–4 — 5 pt 1–2 — 4 pt 6–7 — 2 pt none (less than 1/year) — 1pt over 7 — 0 pt EDSS score: 0–2 — 6 pt 2.5–4 — 3 pt 4.5–5 — 2 pt over 5 — 1 pt	Disease duration: 0–3 years — 6 pt 3–6 years — 4 pt 6–10 years — 2 pt over 10 years — 1 pt RRMS with no neurological deficit — 5 pt Number of relapses in the last year: 3 and more — 5 pt 1–2 — 4 pt none — 1pt EDSS score: 0–2 — 6 pt 2.5–4 — 5 pt 4.5–5 — 2 pt over 5 — 1 pt
Exclusion criteria	1. hypersensitivity to IFNβ 2. primarily or secondarily progressive MS 3. pregnancy 4. decompensated liver disease (aminotranferase levels ≥ 2x upper reference limit) 5. thyroid disease (no euthyreosis) 6. intractable depressive mood disorder or history of suicidal ideation 7. epilepsy	

MRI — magnetic resonance imaging; MS — multiple sclerosis; RRMS —relapsing–remitting multiple sclerosis; pt — points; EDSS — Expanded Disability Status Score

standard and patient sera were added. As standard, we employed goat anti-human interferon antibodies (Sigma-Aldrich) in dilutions of 1:20, 1:50, 1:100, 1:200 and 1: 400. Patient sera were assessed at a dilution of 1:100. After incubation at room temperature, the plates were washed with PBS-0.05% Tween 20. Then, secondary antibodies were added (goat anti-rabbit IgG and rabbit anti-human IgG conjugated with alkaline phosphatase, Sigma-Aldrich). P-nitrophenyl phosphate was used as the alkaline phosphatase substrate (Sigma-Aldrich) and 1 M HCl as the inhibitor. Absorbance was measured at wavelength = 405 nm with ELx800 ELISA reader (Bio-TEK), as optical density, in arbitrary units (AU/ml), following the formula:

(10 × tested sample absorbance) / (cut-off values absorbance)

The cut-off was defined at the absorbance's 95th percentile. A standard curve, based on log-log regression (R2 = 0.978), was prepared for the correlation between absorbance and reciprocal standard dilution. Reciprocal serum dilutions were then read from the standard curve. A standard curve was also drawn for correlation between concentration of standard and absorbance.

A patient was considered BAb positive, BAb (+), if BAb were detected at any time, and BAb negative, BAb (-), if no sample tested positive and at least one was obtained after ≥ 6 months of treatment. A patient was considered persistently BAb (+) if he or she did not revert to BAb (-) in further samples, and transient BAb (+) otherwise.

Neutralising antibody detection

To detect NAb, we used a luciferase reporter gene assay (iLite® Type I IFN Assay Ready Cells, Euro-Diagnostica, Sweden) which involves live cells transfected with luciferase under a selective IFN type I-controlled promotor. When exposed to

Table 2. Study cohort baseline characteristics; P values for gender difference

	All	Female	Male	p
Age at first relapse, years, mean ± SD	29.9 ± 8.7	31.3 ± 9.2	25.7 ± 5.2	< 0.001
Time to second relapse, months, median (IQR)	10.0 (4.0 to 24.25)	10.0 (4.0 to 24.0)	11.5 (4.5 to 31.5)	0.401
Time to treatment initiation, months, median (IQR)	18.0 (12.0 to 35.0)	23.0 (12.0 to 35.0)	16.5 (7.5 to 34.5)	0.294
Relapses before treatment, median (IQR)	2 (2 to 3)	2 (2 to 3)	2 (2 to 3)	0.568
EDSS at baseline, median (IQR)	1.0 (0 to 1.5)	1.0 (0 to 1.5)	1.0 (0 to 1.25)	0.445
OCB in CSF, % positive	94%	95%	93%	1.000
Follow-up duration, years, median (IQR)	5.1 (2.9 to 7.15)	4.7 (2.9 to 6.7)	5.75 (2.9 to 8.25)	0.446

SD — standard deviation; IQR — interquartile range

IFNβ, the cells synthesise luciferase which generates bioluminescence. In the presence of IFN-neutralising antibodies, the intensity of luminescence decreases.

We used standard recombinant human IFNβ protein (Abcam) and standard positive and negative control sera (Euro-Diagnostica).

To inactivate native IFNβ, serum samples were incubated at 56°C for 30 minutes. The samples were then diluted with the diluent provided (Diluent D): 100 µL for a 60 µL sample. Pre-diluted samples and control sera were added to wells on a microplate, which were then closed with a lid, mixed by swirling, and left for 30 minutes at 37°C with 5% CO_2.

iLite Type I IFN Assay Ready Cells were thawed in a 15-minute 37°C water bath, then diluted according to the manufacturer's instructions and 50 µL of the cell suspension was added to each well. The microplate was mixed and incubated at 37°C with 5% CO_2 for 18 hours. Thirty minutes before the incubation completion, Bright-Glo™ Luciferase Assay System was thawed. The substrate was prepared as instructed by the manufacturer and added to each well in 50 µL portions. The microplate was mixed and incubated for two minutes in darkness at room temperature, then placed in a luminometer (FLx800, Bio-TEK). Luminescence intensity was recorded.

A calibration curve was prepared for the correlation between light intensity in relative light units (RLU) and standard concentration. A sample was considered NAb-positive if the ratio of luminescence intensity of the sample and standard mean was ≤ 1.0. If the ratio was >1.0 the sample was labelled negative.

A patient was NAb positive, NAb (+), if NAb were detected at any time during follow-up, and NAb negative, NAb (-), if all samples, including at least one collected at ≥ 24 months, were negative. If a NAb (+) patient did not test negative later, the patient was considered to be persistently NAb (+). Otherwise, he or she would be labelled transient NAb (+).

Statistical methods

Variables are presented as either mean ± standard deviation (SD) for normally distributed variables, or median with interquartile range (IQR) for variables without normal distribution.

Baseline characteristics were assessed for mutual correlations and compared in subgroups depending on BAb and NAb status.

Associations were considered between antibody status and the results of the follow-up (NEDA, relapse occurrence, disability progression, MRI activity) in each subsequent year, for each period up to a given year and after a given year, and for the entire follow-up. They were compared between the subgroups: BAb (+) and (-), persistent BAb (+) and (-), NAb (+) and (-), persistent NAb (+) and (-).

We used Fisher's exact test for nominal variables and Mann-Whitney *U* test for ordinal variables. For interval variables, normality of distribution and equality of variances was assessed with d'Agostino-Pearson and Levene's tests. For normally distributed variables, comparisons were made with the use of t-test. For non-normal data distribution, Mann-Whitney *U* test was employed instead.

P values of ≤ 0.05 were considered statistically significant.

Statistical analyses were performed using StatSoft STATISTICA version 13 [22] and MedCalc, version 15.8 [23].

Results

Baseline

A total of 101 RRMS patients, 77 females and 24 males, were recruited for the study. In Table 2, the baseline characteristics of the study cohort are presented. Women were older than men. There were no other significant gender-specific differences.

Of 101 patients included in the study, nine had compensated thyroid disease, including six with subclinical hypothyroidism, one with Hashimoto disease, one with non-toxic goitre treated with levothyroxine, and one on levothyroxine following thyroidectomy due to toxic goitre. Two subjects had asthma, one had a history of uveitis, and another one of hepatitis of unknown aetiology (both without treatment at the time of our study). Four patients had a history of anxiety and/or mood disorders, including one who was treated with mirtazapine. In two patients, arterial hypertension was present.

Table 3. Proportion of patients with positive and negative anti-IFNβ antibody status

	All	Female	Male
BAb: All tested	97	74	23
BAb (+) at any time	95 (98%)	72 (97%)	23 (100%)
transient BAb (+)	7 (7%)	5 (7%)	2 (9%)
persistent BAb (+)	88 (91%)	67 (91%)	21 (91%)
BAb (-)	2 (2%)	2 (3%)	0 (0%)
baseline BAb (+)	66 of 72 (92%)	47 of 53 (89%)	19 of 19 (100%)
NAb: All tested	77	58	19
NAb (+) at any time	68 (87%)	50 (84%)	18 (95%)
transient NAb (+)	1 (1%)	1 (2%)	0 (0%)
persistent NAb (+)	67 (86%)	49 (84%)	18 (95%)
NAb (-)	9 (12%)	8 (14%)	1 (5%)
baseline NAb (+)	9 of 72 (12.5%)	8 of 53 (15%)	1 of 19 (5%)

BAb — binding antibodies; NAb — neutralising antibodies
No difference in baseline characteristics was found between BAb (+) and BAb (-) or between NAb (+) and NAb (-) cases

Antibody prevalence

200 samples were included in the final analysis. This allowed us to determine BAb status in 97 and NAb status in 77 patients.

Most patients developed both binding and neutralising antibodies. Overall, 95 (97%) had at least one BAb (+) sample. Of these, seven reverted to BAb (-), making 91% persistently BAb (+). Three cases (2%) remained negative throughout the entire study. For NAb, 68 (88%) tested positive, and among them, one reverted to NAb (-), leaving 87% persistently NAb (+). Nine patients (12%) were NAb (-).

The overwhelming majority of patients tested positive for BAb at baseline (66 out of 72 sampled = 92%). The patients who developed BAb later tested positive between one and 24 months. Notably, many patients with baseline BAb had transient reversal to BAb (-) in the first two years, and became BAb (+) again at 24 months.

The prevalence of NAb positivity at baseline was lower: nine out of 72 (12.5%) tested positive. An additional three patients converted to NAb (+) within the first two years, the rest tested positive at 24 months. Of note, few samples were collected at baseline and 24 months.

The summary of antibody prevalence is shown in Tables 3 and 4.

Follow-up

The patients were followed up for a median of 60 months (range 4 to 110).

Between 70% and 80% of patients maintained NEDA criteria of no disease activity in each year from year 1 to year 6, 60% in year 7, 70% in year 8, and 60% in year 9 (Fig. 1). The cumulative count of patients who maintained NEDA-3 is shown in Figure 2.

Discontinuation statistics

A total of 66 patients stopped treatment during the study. Twenty-three patients (35%) had their treatment revoked after three years because the Polish MS treatment programme was limited to three years until 2012. Seven patients (11%) became pregnant or were considering doing so. Another seven patients resigned for personal reasons. Adverse effects were reasons for discontinuation in 14 patients (21%). In 15 cases (23%), the drug was switched due to lack of efficacy.

No associations between discontinuation for any reason and antibodies or baseline characteristics were found.

Correlations with baseline clinical characteristics

Patients with NEDA for the first seven years had fewer relapses prior to treatment ($p = 0.038$). Likewise, fewer pre-treatment relapses were reported in patients with no disease progression after the first and second years ($p = 0.011$ and 0.014), within the first six years ($p = 0.023$), and at any time during the observation ($p = 0.011$). Patients with progression after the second year were older at the time of their first relapse ($p = 0.025$). Most of the significant differences were in baseline EDSS score, which was higher in cases with progression within the first two, three, seven and eight years (p ranging from 0.017 to 0.041), after the first and the second year ($p = 0.017$ and 0.020), and at any time during the follow-up ($p = 0.017$).

Patients with MRI activity in the first seven years apparently had fewer relapses prior to treatment ($p = 0.041$).

Correlations with BAb and NAb status

For BAb, a significant correlation with NEDA was found: patients with persistent BAb were more likely to experience disease activity after year 4 [$p = 0.014$, RR 7.333 (95% CI 0.511 to 105.267), p for RR 0.143 and year 5 ($p = 0.049$, RR 6.75 (95% CI 0.468 to 97.264), p for RR 0.161]. There were no other associations between BAb status and NEDA, relapses, progression, MRI activity, number of relapses, or EDSS score at any time period.

Table 4. Anti–IFNβ antibody prevalence: by samples and cumulative (with assumption of no change from last known result for missing data)

				BAb			
Samples	**Baseline**	**1 month**	**3 months**	**6 months**	**12 months**	**24 months**	**36 months**
All	72	30	3	14	4	49	28
Positive	66	14	0	4	1	45	28
Negative	6	16	3	10	3	3	0
% positive	91.7%	46.7%	0.0%	28.6%	25.0%	91.8%	100.0%
% negative	8.3%	53.3%	100.0%	71.4%	75.0%	6.1%	0.0%
				NAb			
Samples	**Baseline**	**1 month**	**3 months**	**6 months**	**12 months**	**24 months**	**36 months**
All	72	30	3	14	4	49	28
Positive	9	3	0	3	0	45	22
Negative	63	27	3	11	4	3	6
% positive	12.5%	10.0%	0.0%	21.4%	0.0%	91.8%	78.6%
% negative	87.5%	90.0%	100.0%	78.6%	100.0%	6.1%	21.4%
Cases				**BAb**			
Cumulative	**Baseline**	**1 month**	**3 months**	**6 months**	**12 months**	**24 months**	**36 months**
Positive	66	67	65	65	64	73	87
Negative	31	30	32	32	33	24	10
% positive	68.0%	69.1%	67.0%	67.0%	66.0%	75.3%	89.7%
% negative	32.0%	30.9%	33.0%	33.0%	34.0%	24.7%	10.3%
Cases				**NAb**			
Cumulative	**Baseline**	**1 month**	**3 months**	**6 months**	**12 months**	**24 months**	**36 months**
Positive	9	11	11	13	12	55	67
Negative	68	66	66	64	65	22	10
% positive	11.7%	14.3%	14.3%	16.9%	15.6%	71.4%	87.0%
% negative	88.3%	85.7%	85.7%	83.1%	84.4%	28.6%	13.0%

BAb — binding antibodies; NAb — neutralising antibodies

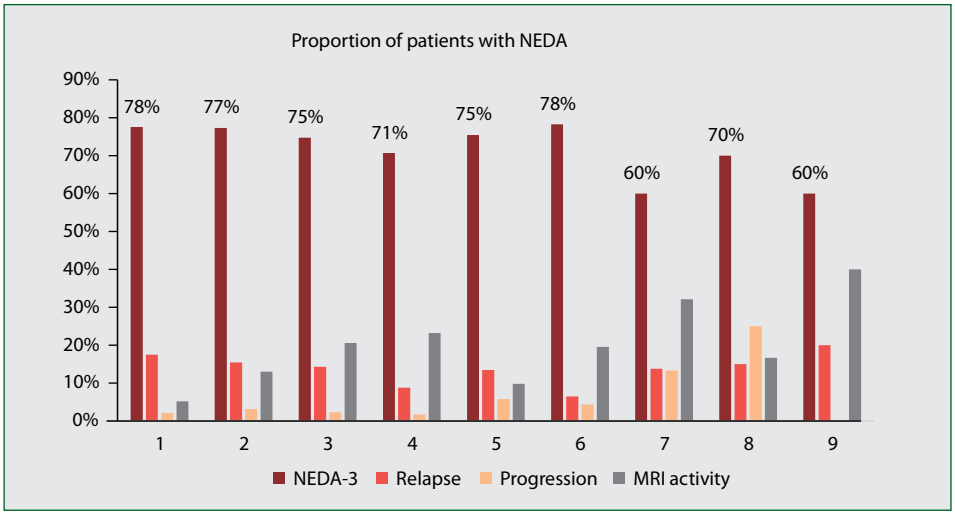

Figure 1. Proportion of patients with no disease activity (according to NEDA-3 criteria) and patients with relapses, progression and MRI activity in subsequent years

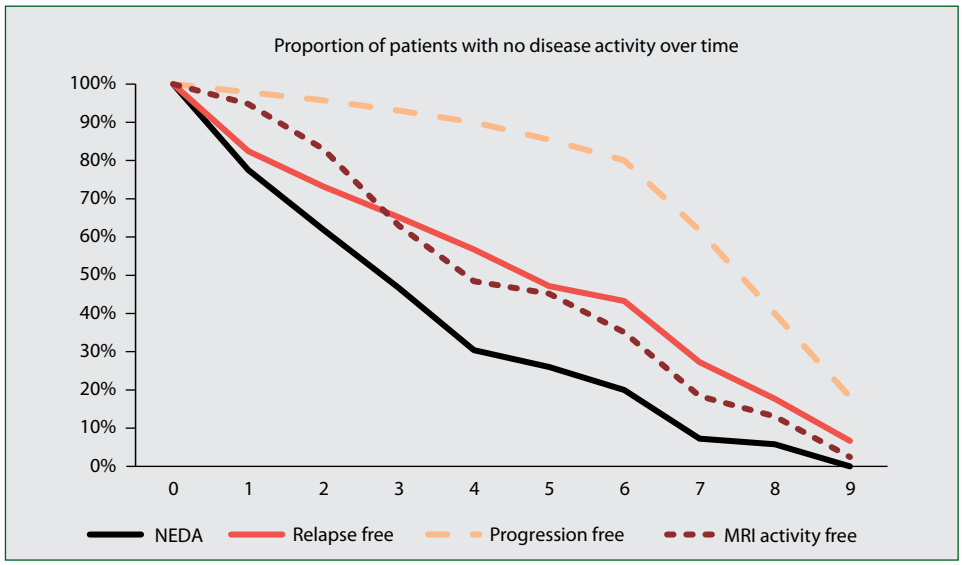

Figure 2. Proportion of patients with no disease activity (according to NEDA-3 criteria), relapses, progression and MRI activity over time

No association was found between NAb and any form of disease activity.

Excluding patients with baseline positivity and with unknown baseline status, attempting to distinguish between the antibodies present at baseline and those developed after drug exposure was impossible due to the small sample size [i.e. no BAb (-) cases were left, three baseline NAb (-) patients]. Therefore, only patients with known baseline positivity were excluded. A trend for higher disease activity was found for NAb (+) in the first year ($p = 0.098$), and for the first two years ($p = 0.069$). In addition, a trend for higher disease activity in BAb (+) cases after years 4 and 5 (both with $p = 0.061$) re-emerged.

After moving the baseline positive patients to antibody negative groups, NAb (+) was associated with a higher risk of disease activity in the first two [$p = 0.019$, RR 3.724 (95% CI 0.990 to 14.017), p for RR 0.052] and three years [$p = 0.041$, RR 2.210 (95% CI 0.928 to 5,260), p for RR 0.073], as well as in the entire follow up [$p = 0.044$, RR 1.575 (95% CI 0.930 to 2.668), p for RR 0.091]. It also correlated with MRI activity in the first three years [$p = 0.043$, RR 6.938 (95% CI 0.458 to 105.035), p for RR 0.162].

In logistic regression analysis, this modified NAb (+) status was a significant independent variable in enter method and was retained as the only significant variable in forward method in models for disease activity within the first two years of treatment.

When considering baseline BAb (+) as BAb (-), BAb (+) was associated with a higher risk of treatment discontinuation, of NEDA-3 loss after the first and the second year, a lower risk of relapse in year 6 although higher in year 4, a higher risk of disease progression in year 6, but again a lower risk after the first and the second year and in the follow-up as a whole. BAb (+) also showed less MRI activity after the first year and in the entire observation.

Comparing patients with and without baseline BAb and NAb [BAb0 (+) and BAb0 (-), NAb0 (+) and NAb0 (-)], regardless of subsequent antibody status, NAb0 (+) was associated with a lower risk of disease activity in the first three years ($p = 0.017$, RR = 0.2239, 95% CI: 0.03611 to 1.3890, p for RR = 0.1080). For baseline BAb, no significant associations were found in direct comparisons, but they remained the only significant variable in a forward model for relapse activity in the first year of treatment, where the absence of baseline BAb increased the likelihood of relapses (Tab. 5).

Logistic regression

For each time period and activity measure, logistic regression models were computed with combinations of BAb and NAb, persistent BAb and Nab, and baseline clinical characteristics.

Neither BAb nor NAb status, persistent or not, was retained as a significant independent contributor. Conversely, pre-treatment EDSS and relapses were abundantly included in models predicting progression, while several relapse and MRI activity models contained age, treatment delay, or time to second relapse. Only two models reached significant goodness of fit, one including NAb, the other persistent NAb (outcome: progression after third year, both with $p = 0.015$ and Hosmer-Lemeshow $p < 0.001$) (Tab. 6). For these variable sets, in forward, backward and stepwise method, only pre-treatment EDSS and relapses were retained.

Discussion

The abundance of NAb and BAb positivity before treatment is the most surprising finding of our study. Previous works have reported naturally occurring BAb and NAb in less than 1% of the healthy population and treatment-naive MS patients [1].

Table 5. Logistic regression analysis: relapses in the first year of treatment

Variable	Coefficient	Standard error	Wald's χ^2	p
Baseline Bab (—)	1.974	0.977	4.084	0.043
Constant	-1.569			

Table 6. Logistic regression analysis: progression after third year of treatment

Variable	Coefficient	Standard error	Wald's χ^2	p
NAb (+) / persistent NAb (+)*	17.226	2,881.824	< 0.001	0.995
Age at first relapse, years	-0.001	0.056	< 0.001	0.983
Gender	-0.743	1.182	0.395	0.530
Time to second relapse, months	0.041	0.063	0.424	0.515
Time to treatment start, months	-0.063	0.053	1.437	0.230
Baseline EDSS score	**2.481**	**1.146**	**4.691**	**0.030**
Number of relapses prior to treatment	**1.490**	**0.716**	**4.330**	**0.037**
Constant	-23.075			

*the models were otherwise identical

On the other hand, NAb developed in 11% of patients in the placebo group of a stage III clinical trial of IFNβ-1b [2]. It is understandable that positivity prevalence depends to a great extent on the assay's sensitivity and cut-off thresholds [1, 24]. Therefore these pre-treatment antibodies probably represent naturally occurring, nonspecific antibodies of very low affinity and variable IFN neutralising activity. Alternatively, this could potentially be caused by a methodological issue (i.e. sample contamination) or, less likely, a cross-reactivity with an antigen commonly encountered in the Polish population.

It is plausible that a different kind of anti-interferon antibodies develops in response to drug exposure and undergoes gradual maturation, eventually gaining IFN neutralising properties.

Removal of the baseline NAb-positive cases from the analysis brought a trend towards more disease activity in NAb (+) and, curiously, BAb (+) patients, supporting the hypothesis that the baseline and aquired antibodies represent two different phenomenons. An assignment switch involving moving baseline positive patients to the antibody negative groups revealed an association between early disease activity and NAb positivity, not unlike the previous evidence [2, 6–11]. An analogous grouping shift changed BAb correlations greatly, with contradictory associations with disease activity.

These inconsistent results suggest that the co-occurrence of unspecific and specific binding antibodies is probably common, and renders this analysis invalid.

Interestingly, pre-treatment antibodies predicted a more indolent course in the first three years of treatment. This resembles the observations from pivotal trials of IFNβ in MS [2, 25, 26], where patients who were to develop NAb in later observations had lower relapse rates in the first six or 12 months of treatment. This was thought to be caused by low-affinity anti-IFN antibodies that could initially prolong the drug's half-life, but would eventually develop into harmful, detectable NAb due to affinity maturation [24].

We are aware of our study's limitations. Due to the strict eligibility criteria for the Polish MS treatment programme, patients with low pre-treatment disease activity were preferentially included. This may explain why the study cohort experienced a relatively benign disease course. However, disease activity statistics were similar in other recently described populations [27]. Thirty-five percent of cases were lost after the third year of treatment due to the programme duration limit, regardless of response, while many continued treatment despite sustained disease activity (20–30% non-NEDA in each year), as second-line therapeutics were either not yet registered or out of reach due to programme criteria.

Because of poor patient co-operation and technical limitations, a significant number of scheduled samples were not collected.

Conclusions and future directions

In our RRMS cohort, we observed a high prevalence of naturally occurring anti-IFNβ BAb and NAb.

Exclusion of the NAb0 (+) cases brought results reminiscent of previous ones, with a trend towards more disease activity in NAb (+) and, notably, BAb (+) patients. An assignment switch (moving NAb0 (+) and BAb0 (+) patients to the NAb (-) and BAb (-) groups) revealed an association between

early disease activity and NAb, consistent with previously published research.

Patients with baseline positivity in BAb or NAb experienced less disease activity in the first one or three years of treatment.

We propose that two types of anti-interferon antibodies were detected by our assays: specific, drug-induced antibodies and also low-affinity, naturally occurring antibodies. The naturally occurring antibodies are beneficial or reflect an advantageous immune state. Drug-induced antibodies, once they reach a certain neutralising activity, inhibit IFNβ activity and cause loss of drug efficacy.

Our findings require confirmation in further studies. The prevalence and specificity of low-affinity antibodies should be determined. Also, a titre or anti-IFN activity threshold of predictive significance should be established if possible.

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References

1. Ross C, Clemmesen KM, Svenson M, et al. Immunogenicity of interferon-beta in multiple sclerosis patients: influence of preparation, dosage, dose frequency, and route of administration. Danish Multiple Sclerosis Study Group. Ann Neurol. 2000; 48(5): 706–712, indexed in Pubmed: 11079533.
2. Neutralizing antibodies during treatment of multiple sclerosis with interferon beta-1b: experience during the first three years. The IFNB Multiple Sclerosis Study Group and the University of British Columbia MS/MRI Analysis Group. Neurology. 1996; 47(4): 889–894, doi: 10.1212/wnl.47.4.889, indexed in Pubmed: 8857714.
3. Sorensen PS, Koch-Henriksen N, Ross C, et al. Danish Multiple Sclerosis Study Group. Appearance and disappearance of neutralizing antibodies during interferon-beta therapy. Neurology. 2005; 65(1): 33–39, doi: 10.1212/01.WNL.0000166049.51502.6A, indexed in Pubmed: 15888603.
4. Cocco E, Marrosu MG. Profile of PEGylated interferon beta in the treatment of relapsing-remitting multiple sclerosis. Ther Clin Risk Manag. 2015; 11: 759–766, doi: 10.2147/TCRM.S69123, indexed in Pubmed: 26056458.
5. Gneiss C, Reindl M, Lutterotti A, et al. Interferon-beta: the neutralizing antibody (NAb) titre predicts reversion to NAb negativity. Mult Scler. 2004; 10(5): 507–510, doi: 10.1191/1352458504ms1074oa, indexed in Pubmed: 15471365.
6. Malucchi S, Sala A, Gilli F, et al. Neutralizing antibodies reduce the efficacy of betaIFN during treatment of multiple sclerosis. Neurology. 2004; 62(11): 2031–2037, doi: 10.1212/01.wnl.0000129265.73259.9e, indexed in Pubmed: 15184610.
7. Perini P, Calabrese M, Biasi G, et al. The clinical impact of interferon beta antibodies in relapsing-remitting MS. J Neurol. 2004; 251(3): 305–309, doi: 10.1007/s00415-004-0312-8, indexed in Pubmed: 15015010.
8. Tomassini V, Paolillo A, Russo P, et al. Predictors of long-term clinical response to interferon beta therapy in relapsing multiple sclerosis. J Neurol. 2006; 253(3): 287–293, doi: 10.1007/s00415-005-0979-5, indexed in Pubmed: 16151600.
9. Malucchi S, Gilli F, Caldano M, et al. One-year evaluation of factors affecting the biological activity of interferon beta in multiple sclerosis patients. J Neurol. 2011; 258(5): 895–903, doi: 10.1007/s00415-010-5844-5, indexed in Pubmed: 21153733.
10. Petkau AJ, White RA, Ebers GC, et al. IFNB Multiple Sclerosis Study Group. Longitudinal analyses of the effects of neutralizing antibodies on interferon beta-1b in relapsing-remitting multiple sclerosis. Mult Scler. 2004; 10(2): 126–138, doi: 10.1191/1352458504ms1004oa, indexed in Pubmed: 15124756.
11. Paolicelli D, D'Onghia M, Pellegrini F, et al. The impact of neutralizing antibodies on the risk of disease worsening in interferon β-treated relapsing multiple sclerosis: a 5 year post-marketing study. J Neurol. 2013; 260(6): 1562–1568, doi: 10.1007/s00415-012-6829-3, indexed in Pubmed: 23417273.
12. Paolicelli D, Manni A, Iaffaldano A, et al. The role of neutralizing antibodies to interferon-β as a biomarker of persistent MRI activity in multiple sclerosis: a 7-year observational study. Eur J Clin Pharmacol. 2016; 72(8): 1025–1029, doi: 10.1007/s00228-016-2073-6, indexed in Pubmed: 27251359.
13. WHO Expert Committee on Biological Standardization. Thirty-fifth report. World Health Organ Tech Rep Ser. 1985; 725: 1–140, indexed in Pubmed: 2417419.
14. Bertolotto A, Sala A, Caldano M, et al. Development and validation of a real time PCR-based bioassay for quantification of neutralizing antibodies against human interferon-beta. J Immunol Methods. 2007; 321(1-2): 19–31, doi: 10.1016/j.jim.2006.12.012, indexed in Pubmed: 17335844.
15. Farrell R, Kapoor R, Leary S, et al. Neutralizing anti-interferon beta antibodies are associated with reduced side effects and delayed impact on efficacy of Interferon-beta. Mult Scler. 2008; 14(2): 212–218, doi: 10.1177/1352458507082066, indexed in Pubmed: 17986510.
16. Lallemand C, Meritet JF, Erickson R, et al. Quantification of neutralizing antibodies to human type I interferons using division-arrested frozen cells carrying an interferon-regulated reporter-gene. J Interferon Cytokine Res. 2008; 28(6): 393–404, doi: 10.1089/jir.2007.0142, indexed in Pubmed: 18593334.
17. Lam R, Farrell R, Aziz T, et al. Validating parameters of a luciferase reporter gene assay to measure neutralizing antibodies to IFNbeta in multiple sclerosis patients. J Immunol Methods. 2008; 336(2): 113–118, doi: 10.1016/j.jim.2008.03.014, indexed in Pubmed: 18511063.
18. Polman CH, Reingold SC, Edan G, et al. Diagnostic criteria for multiple sclerosis: 2005 revisions to the „McDonald Criteria". Ann Neurol. 2005; 58(6): 840–846, doi: 10.1002/ana.20703, indexed in Pubmed: 16283615.

19. Polman CH, Reingold SC, Banwell B, et al. Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. Ann Neurol. 2011; 69(2): 292–302, doi: 10.1002/ana.22366, indexed in Pubmed: 21387374.
20. Kappos L, De Stefano N, Freedman MS, et al. Inclusion of brain volume loss in a revised measure of ‚no evidence of disease activity' (NEDA-4) in relapsing-remitting multiple sclerosis. Mult Scler. 2016; 22(10): 1297–1305, doi: 10.1177/1352458515616701, indexed in Pubmed: 26585439.
21. Wencel-Warot A, Michalak S, Warot M, et al. The cross-reactivity of binding antibodies with different interferon beta formulations used as disease-modifying drugs in multiple sclerosis patients. Medicine (Baltimore). 2016; 95(45): e5337, doi: 10.1097/MD.0000000000005337, indexed in Pubmed: 27828855.
22. TIBCO Software Inc. StatSoft STATISTICA. ; 2017.
23. MedCalc Software bvba. MedCalc statistical software. Ostend, Belgium 2015. https://www.medcalc.org/ (May 13, 2017).
24. Sorensen PS, Koch-Henriksen N, Bendtzen K. Are ex vivo neutralising antibodies against IFN-beta always detrimental to therapeutic efficacy in multiple sclerosis? Mult Scler. 2007; 13(5): 616–621, doi: 10.1177/1352458506072344, indexed in Pubmed: 17548440.
25. Rudick RA, Simonian NA, Alam JA, et al. Incidence and significance of neutralizing antibodies to interferon beta-1a in multiple sclerosis. Multiple Sclerosis Collaborative Research Group (MSCRG). Neurology. 1998; 50(5): 1266–1272, doi: 10.1212/wnl.50.5.1266, indexed in Pubmed: 9595973.
26. PRISMS Study Group and the University of British Columbia MS/MRI Analysis Group.. PRISMS-4: Long-term efficacy of interferon-beta-1a in relapsing MS. Neurology. 2001; 56(12): 1628–1636, doi: 10.1212/wnl.56.12.1628, indexed in Pubmed: 11425926.
27. Rotstein DL, Healy BC, Malik MT, et al. Evaluation of no evidence of disease activity in a 7-year longitudinal multiple sclerosis cohort. JAMA Neurol. 2015; 72(2): 152–158, doi: 10.1001/jamaneurol.2014.3537, indexed in Pubmed: 25531931.

RESEARCH PAPER

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Biomechanical evaluation of single- and multi-level anterior cervical discectomy and fusion with polyetheretherketone cages: radiological and clinical outcomes

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ABSTRACT

Objective. The aim of this study was to analyse the outcomes of single- and multi-level anterior cervical discectomy and fusion (ACDF) with standalone polyetheretherketone (PEEK) cages, with particular emphasis on the risk of secondary adjacent segment disease.

Materials and methods. This retrospective study included 30 patients with single- or multi-level cervical disc herniation. Before the ACDF, and one year thereafter, the patients underwent clinical and radiological evaluation including determination of cervical pain severity with a numerical rating scale (NRS), and a survey with a Polish adaptation of the neck disability index questionnaire (NDI-PL). Biomechanical parameters of the cervical spine were determined using the Cobb method.

Results. One year after ACDF, all patients had achieved complete fusions, and 97% showed a significant reduction of pain severity. Also, a significant decrease in all NDI-PL indices was observed. A significant decrease in overall cervical spine mobility coexisted with a significant increase in the mobility of the segment above the one operated upon and a non-significant decrease in the mobility of the segment below. No statistically significant change was found in the intervertebral disc space height (IVH) above and below the operated segment, and no evidence of degeneration within the segments adjacent to the operated one was documented.

Conclusion. One- and two-level ACDF with standalone PEEK cages provided high fusion rates. Surgical spondylosis contributed to a reduction of spinal mobility despite the hypermobility in adjacent spinal segments. No degeneration in adjacent spinal segments was documented within a year of ACDF, and the treatment seemed to improve patients' quality of life.

Key words: adjacent segment degeneration, adjacent segment disease, anterior cervical discectomy with fusion, cervical myelopathy, cervical radiculopathy, PEEK cages

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Introduction

Anterior cervical discectomy and fusion (ACDF) is a well-known form of surgical intervention in symptomatic cervical spondylosis. ACDF includes removal of the migrated disc (discectomy), decompression of neural structures within the spinal canal, and stabilisation of the interbody implant at the operated level. Adjacent segment disease (ASD) has become an important issue in patients with single- or multi-level cervical disc disease who undergo the ACDF procedure. ASD is defined as new degenerative changes in spinal segments adjacent to the previously operated one(s), associated with related symptoms [1].

Theoretically, the development of surgical techniques for interbody fusion with preservation of spinal mobility should contribute to a lower incidence of ASD. The ACDF-imposed alterations of spinal biomechanics may accelerate degenerative changes within adjacent segments of the cervical spine;

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however, the exact pathomechanism of this process is still not completely understood [2]. Patients after ACDF showed radiographic evidence of various degenerative changes within adjacent spinal segments, which may pose a substantial diagnostic and therapeutic challenge. Although ACDF results in the attenuation of clinical symptoms, it is also associated with decreased spinal mobility, greater mechanical overload, and accelerated degeneration of adjacent segments, which may produce new clinical symptoms such as radiculopathy, myelopathy, stenosis and instability [3, 4].

In this paper, we present radiological and clinical outcomes of single- and multi-level ACDFs with polyetheretherketone (PEEK) implants, with particular emphasis on the risk of secondary ASD.

Materials and methods

The retrospective study included 30 consecutive patients with single- or multi-level cervical disc herniation who between January and December 2013 underwent ACDF using a standard Cloward procedure with standalone PEEK cages at the Department of Neurosurgery, Medical University of Bialystok, Poland. Three patients were excluded from the analysis because of a history of systemic diseases (rheumatoid arthritis - one patient, osteoporosis – two patients).

The protocol of the study was approved by the Local Bioethics Committee at the Medical University of Bialystok (decision no. R-I-002/40/2013).

The study patients presented with single- or multi-level cervical spine disease (C3 to C7). Only those patients with cervical radiculopathy, myelopathy with neurological deficits or without, and spondylosis diagnosed based on MRI, were included in the analysis. Patients with physical deformities, infections, metabolic bone diseases or tumours were excluded.

Before the ACDF, and 12 months post-procedure, all participants of the study were subjected to a comprehensive clinical and radiological evaluation. The clinical evaluation consisted of history taking, physical and neurological examination, assessment of pain severity with a numerical rating scale (NRS) and a survey with a Polish adaptation of the neck disability index questionnaire (NDI-PL).

Spinal radiographs (anterior-posterior, lateral and flexion--extension) were obtained preoperatively and one year after the ACDF. Functional parameters of the cervical spine were determined based on Cobb angle measurement, a method used routinely to calculate the spinal curvature angles. Overall mobility of cervical spine and segmental mobility in the operated and adjacent segments determined prior to the ACDF were compared with respective parameters obtained 12 months post-procedure. The measurement methodology is set out in Figure 1.

Intervertebral disc space height (IVH) was measured for the operated segment and adjacent segments. Moreover, fusion (subsidence, antero-posterior implant displacement) and reconstruction of the intervertebral disc space height were evaluated. Fusion was defined as the lack of translucency around the PEEK cage, the presence of adhesion mass-fusion bone between the vertebral bodies, and the absence of motion between the spinous processes on flexion-extension radiographs. An interspinous distance ≥2 mm on functional radiographs was defined as non-fusion [5]. Cervical alignment was determined as the angle formed by the imaginary lines tangent to the posterior edges of C2 and C7. Subsidence was defined as the loss of height in the operated segment(s) on lateral radiographs, >3 mm and >5 mm for one- and two-level procedures, respectively [6, 7]. Intervertebral disc space height (IVH) was defined as the mean value of the anterior and posterior height of the disc, expressed in millimetres.

The follow-up radiographs were screened for ASD by two independent observers blinded to the clinical outcomes. ASD was defined by the presence of at least one of the following: calcification of the anterior longitudinal ligament; narrowing of the disc space with or without posterior osteophytes; and/or formation of a new anterior osteocyte or enlargement of a preexisting osteophyte [8].

Statistical analysis

The results were analysed with a Statistica 10.0 package from StatSoft. Statistical characteristics of the study variables were presented as means, their standard deviations, medians, minimum and maximum values. NDI-PL scores were compared using the Wilcoxon test, and mobility of the spinal segments with the non-parametric Kruskal-Wallis test. The significance of intragroup differences in overall and segmental spinal mobility was verified with the Wilcoxon test and the significance of intergroup differences in the spinal mobility with the non-parametric Mann-Whitney U-test. The threshold of statistical significance for all tests was set at $p < 0.05$.

Results

The study included 16 men (53.3%) and 14 women (46.7%) with a mean age of 56 years (range 27–67 years). All data was collected retrospectively. Mean duration of follow-up was 12 months. Four patients achieved fusion at C3/C4, another four at C4/C5, 19 at C5/C6, and 14 at C6/C7. No patients required additional surgery for recurrent symptoms.

Up to 97% of the patients who underwent ACDF reported a significant decrease in pain severity expressed on NRS. Furthermore, a statistically significant decrease in all NDI-PL indices (i.e. pain intensity, personal care, lifting, reading, headaches, concentration, work, driving, sleeping, and recreation) was documented, along with a significant improvement in daily functional activity. The first pre-op measurement amounted to c. 47% of disability, the last amounted to c. 15%, defining minimal disability.

Fusion rates for C2–C7 at three and 12 months post--procedure were 95.7% and 100%, respectively. No subsidence

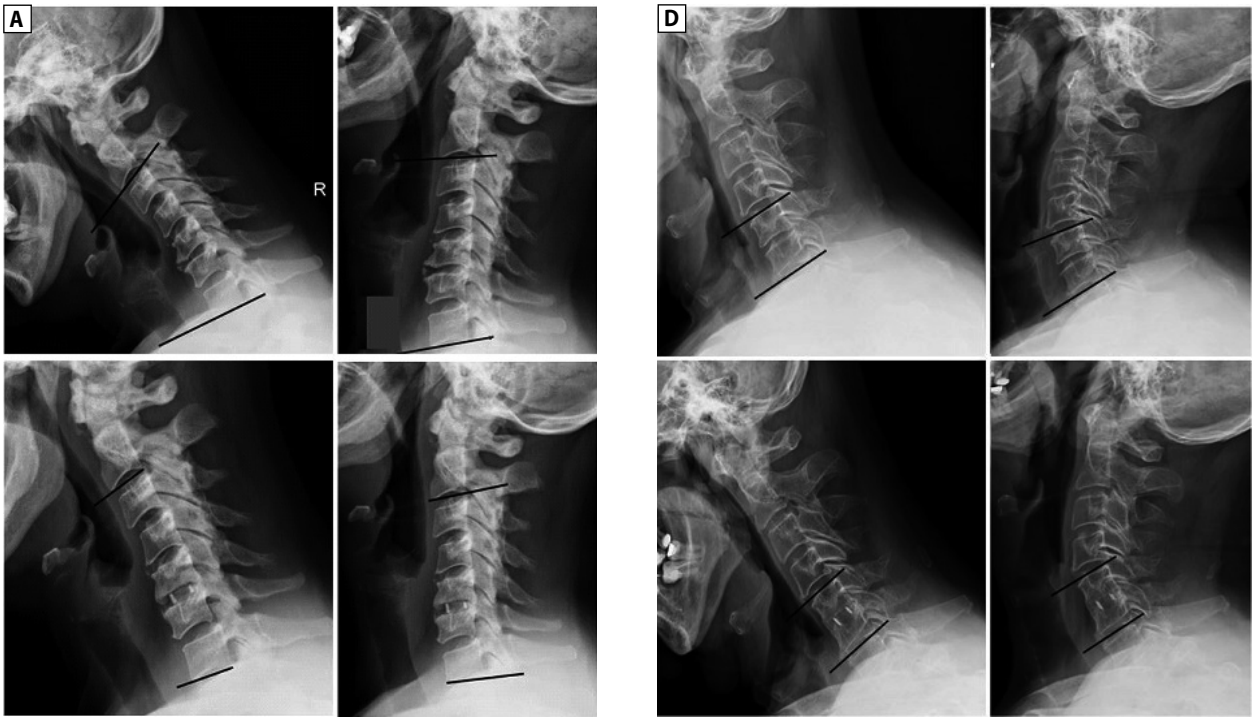

Figure 1. Measurement of spinal mobility: (**A**) ROM for C2–C7, (**B**) ROM for the segment located above the operated one, (**C**) ROM for the segment located below the operated one, (**D**) ROM for the operated segment. All ROM values determined based on Cobb angle measurement, on archival radiographs from the Department of Neurosurgery, Medical University of Bialystok

of vertebral bodies on the PEEK implant was observed at the operated level. At the end of the follow-up period, a statistically significant decrease in overall cervical spine mobility (C2–C7) was observed, along with a significant increase in the mobility of the segment located above the operated one, and a non-significant decrease in the mobility of the segment located below. No statistically significant change was found in the IVH above and below the operated segment. No evidence of degeneration within the segments adjacent to the operated one was documented during the follow-up period, and none of the patients required repeated ACDF for the adjacent segments 12 months after the primary procedure.

Discussion

Although ACDF is an effective method for the treatment of degenerative cervical disease, it can also result in overload and hypermobility of adjacent spinal segments [3]. Based on a systematic review of the literature, Sugawara [9] concluded that 2–36% of patients subjected to ACDF may develop symptomatic degenerative changes in adjacent spinal segments within 10 years of the procedure, which in 6–19% of cases may require further surgical treatment. In two studies with a minimum three-year follow-up [10], the incidence of radiological adjacent segment pathologies (RASP), such as heterotrophic calcification of the anterior longitudinal ligament and narrowing of the disc space with or without posterior osteophytes, exceeded 30%, and 10.5% of patients after cervical spine surgeries showed evidence of heterotrophic calcification. This can cause a secondary lack of flexibility within the previously operated segment of cervical spine. According to Mehren et al. and Heidecke et al., heterotrophic calcifications may be found in up to 29% of patients, whereas Tortolani et al. reported them as a rare and late complication of cervical surgeries [11–13].

In our present study, patients subjected to ACDF did not show any evidence of degenerative changes such as heterotrophic calcifications within one year of the procedure. The lack of symptomatic ASD in our series was probably associated with the fact that the patients were followed-up for a shorter period than the participants in previous, larger studies [14]. Our follow-up was short, limited to one year only. This represents a major limitation of our study. However, we continue to follow this cohort, and we hope that additional data will be available in the future.

The aetiology of ASD is complex, and no single cause or risk factor for this pathology have been identified thus far. ASD may be a consequence of a preexisting disease, physiological age-related degenerative process, hypermobility in adjacent spinal segments, changes in intradiscal pressure, anatomical anomalies or sagittal misalignment [15]. According to Lawrence et al. [16], the risk factors for ASD after cervical fusion surgery included age < 60 years, fusion adjacent to C5-C6 and/or C6-C7, preexisting disc herniation, and/or dural compression secondary to spinal stenosis. In turn, Katsuura et

al. [17] found a direct correlation between symptomatic ASD and the loss of lordotic curvature in the sagittal cervical spine. Published opinions vary about the contribution of surgical technique or operative management to the incidence of ASD. Song et al. [18] found no association between the incidence of clinical ASD and the number of fused segments. Hilibrand et al. [1] followed up 374 patients who underwent 409 cervical fusion procedures over a 20-year period. Approximately 25% of the patients developed symptomatic ASD within 10 years of the procedure, with an annual incidence rate of 2.9%. The incidence of ASD was shown to be higher after C5–C6 and C6–C7 fusions. According to some authors, the risk of ASD within an adjacent segment after a single-level fusion is higher in patients with CT-myelography or MRI evidence of preexisting degenerative changes [19].

Other key factors implicated in ASD are altered biomechanics and mobility of adjacent spinal segments, resulting from inappropriate cervical alignment. Biomechanical changes in adjacent segments after the fusion, such as altered range of motion or intradiscal pressure, have been reported by several authors [10, 18]. However, in our present study, the hypermobility of the adjacent spinal segment above the operated one did not exert an effect on the incidence of ASD within one year of ACDF, and we did not find statistically significant changes in IVH above and below the operated segment.

Also, the effect of soft tissue disruption on the biomechanics of adjacent spinal segments should be considered as a factor predisposing to ASD. Some previous studies found an association between the ossification of adjacent spinal segments in patients implanted with anterior cervical plates who eventually developed ASD. Park et al. [20] measured the distance between the intervertebral disc and the metal plate implanted during ACDF with plate fixation and found no association between this parameter and the risk of secondary ASD. We cannot comment on this observation as no plate fixation was used in our patients.

In previous studies, ACDF provided good or excellent outcomes in 70–90% of patients with cervical radiculopathy, primarily due to the decompression of the neural structures [21]. Bohlman et al. [22] found a correlation between the lack of fusion and the incidence of postoperative neck pain. In our present study, the lack of subsidence and the presence of stable intervertebral spondylosis observed in all patients during the final control visit turned out to be associated with a significant attenuation of pain. No patients developed symptomatic pseudarthrosis that required a secondary surgery.

These findings imply that the implantation of PEEK cages is a safe procedure resulting in high fusion rates and good clinical and radiological outcomes.

Published evidence suggests that the appropriate alignment of sagittal cervical spine might be associated with better quality of life after ACDF. According to some authors, the reconstruction of IVH is of lesser importance. However, worse cervical alignment has been shown to correlate with poorer outcomes with regards to neck and arm pain, as well as with a higher likelihood of repeated surgical procedure. This implies that preservation of cervical lordosis is an important determinant of ACDF outcomes. Lordotic alignment contributes to better mobility and functioning of cervical spine. In turn, sagittal misalignment has been demonstrated to be associated with cervical instability, pain, and even unfavourable functional outcomes [23]. Attenuation of pain is without doubt a key determinant of the quality of life. Similarly to previous studies, our experiment demonstrated that ACDF contributed to a considerable improvement in the quality of life determined with NRS scores and the NDI-PL questionnaire. Thus, the outcomes of the treatment were not impeded by either a decrease in spinal mobility or a hypermobility of the spinal segment located above the operated one.

In this study, we analysed spinal biomechanics with a functional X-ray, using a routine, albeit highly reliable, Cobb method. Dvorak et al. [24] analysed functional radiographs from 64 patients with disorders of cervical spine. The patients were divided into three groups, i.e. cervical degeneration, radiculopathy, and cervical trauma, which were then compared to healthy controls so as to identify pathological patterns of motion. Tęsiorowski et al. [25] compared cervical mobility at three, six and 12 months post-surgery with normal cervical mobility of people aged 40-50. The analysis of functional radiographs demonstrated an evident local kyphotic deformity on lateral views.

Conclusions

One- and two-level ACDF with standalone PEEK cages provided high fusion rates. Spondylosis at one or two levels contributed to a reduction of spinal mobility despite the hypermobility in adjacent spinal segments. No degenerative changes in adjacent spinal segments were documented within a year of ACDF. Restoring IVH at the operated level seems to be less important from the perspective of functional outcome. ACDF improved the quality of life in patients with single- or multi-level cervical disc herniation.

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References

1. Hilibrand AS, Robbins M. Adjacent segment degeneration and adjacent segment disease: the consequences of spinal fusion? Spine J. 2004; 4(6 Suppl): 190S–194S, doi: 10.1016/j.spinee.2004.07.007, indexed in Pubmed: 15541666.
2. Litrico S, Lonjon N, Riouallon G, et al. French Society of Spine Surgery (SFCR). Adjacent segment disease after anterior cervical interbody fusion: a multicenter retrospective study of 288 patients with long-term follow-up. Orthop Traumatol Surg Res. 2014; 100(6 Suppl): S305–S309, doi: 10.1016/j.otsr.2014.07.004, indexed in Pubmed: 25129704.

3. Carrier CS, Bono CM, Lebl DR. Evidence-based analysis of adjacent segment degeneration and disease after ACDF: a systematic review. Spine J. 2013; 13(10): 1370–1378, doi: 10.1016/j.spinee.2013.05.050, indexed in Pubmed: 23891293.
4. Matsumoto M, Okada E, Ichihara D, et al. Anterior cervical decompression and fusion accelerates adjacent segment degeneration: comparison with asymptomatic volunteers in a ten-year magnetic resonance imaging follow-up study. Spine (Phila Pa 1976). 2010; 35(1): 36–43, doi: 10.1097/BRS.0b013e3181b8a80d, indexed in Pubmed: 20023606.
5. Kaiser MG, Mummaneni PV, Matz PG, et al. Joint Section on Disorders of the Spine and Peripheral Nerves of the American Association of Neurological Surgeons and Congress of Neurological Surgeons. Radiographic assessment of cervical subaxial fusion. J Neurosurg Spine. 2009; 11(2): 221–227, doi: 10.3171/2009.3.SPINE08719, indexed in Pubmed: 19769501.
6. Gercek E, Arlet V, Delisle J, et al. Subsidence of stand-alone cervical cages in anterior interbody fusion: warning. Eur Spine J. 2003; 12(5): 513–516, doi: 10.1007/s00586-003-0539-6, indexed in Pubmed: 12827473.
7. Karikari IO, Jain D, Owens TR, et al. Impact of subsidence on clinical outcomes and radiographic fusion rates in anterior cervical discectomy and fusion: a systematic review. J Spinal Disord Tech. 2014; 27(1): 1–10, doi: 10.1097/BSD.0b013e31825bd26d, indexed in Pubmed: 24441059.
8. Li J, Li Y, Kong F, et al. Adjacent segment degeneration after single-level anterior cervical decompression and fusion: disc space distraction and its impact on clinical outcomes. J Clin Neurosci. 2015; 22(3): 566–569, doi: 10.1016/j.jocn.2014.08.019, indexed in Pubmed: 25487176.
9. Sugawara T. Anterior Cervical Spine Surgery for Degenerative Disease: A Review. Neurol Med Chir (Tokyo). 2015; 55(7): 540–546, doi: 10.2176/nmc.ra.2014-0403, indexed in Pubmed: 26119899.
10. Maldonado CV, Paz RDR, Martin CB. Adjacent-level degeneration after cervical disc arthroplasty versus fusion. Eur Spine J. 2011; 20 Suppl 3: 403–407, doi: 10.1007/s00586-011-1916-1, indexed in Pubmed: 21796395.
11. Heidecke V, Burkert W, Brucke M, et al. Intervertebral disc replacement for cervical degenerative disease--clinical results and functional outcome at two years in patients implanted with the Bryan cervical disc prosthesis. Acta Neurochir (Wien). 2008; 150(5): 453–9; discussion 459, doi: 10.1007/s00701-008-1552-7, indexed in Pubmed: 18421412.
12. Mehren C, Suchomel P, Grochulla F, et al. Heterotopic ossification in total cervical artificial disc replacement. Spine (Phila Pa 1976). 2006; 31(24): 2802–2806, doi: 10.1097/01.brs.0000245852.70594.d5, indexed in Pubmed: 17108833.
13. Tortolani PJ, Cunningham BW, Eng M, et al. Prevalence of heterotopic ossification following total disc replacement. A prospective, randomized study of two hundred and seventy-six patients. J Bone Joint Surg Am. 2007; 89(1): 82–88, doi: 10.2106/JBJS.F.00432, indexed in Pubmed: 17200314.
14. Bydon M, Xu R, De la Garza-Ramos R, et al. Adjacent segment disease after anterior cervical discectomy and fusion: Incidence and clinical outcomes of patients requiring anterior versus posterior repeat cervical fusion. Surg Neurol Int. 2014; 5(Suppl 3): S74–S78, doi: 10.4103/2152-7806.130676, indexed in Pubmed: 24843815.
15. Anakwenze OA, Auerbach JD, Milby AH, et al. Sagittal cervical alignment after cervical disc arthroplasty and anterior cervical discectomy and fusion: results of a prospective, randomized, controlled trial. Spine (Phila Pa 1976). 2009; 34(19): 2001–2007, doi: 10.1097/BRS.0b013e3181b03fe6, indexed in Pubmed: 19730207.
16. Lawrence BD, Hilibrand AS, Brodt ED, et al. Predicting the risk of adjacent segment pathology in the cervical spine: a systematic review. Spine (Phila Pa 1976). 2012; 37(22 Suppl): S52–S64, doi: 10.1097/BRS.0b013e31826d60fb, indexed in Pubmed: 22885828.
17. Katsuura A, Hukuda S, Saruhashi Y, et al. Kyphotic malalignment after anterior cervical fusion is one of the factors promoting the degenerative process in adjacent intervertebral levels. Eur Spine J. 2001; 10(4): 320–324, doi: 10.1007/s005860000243, indexed in Pubmed: 11563618.
18. Song KJ, Choi BW, Kim JK. Adjacent segment pathology following anterior decompression and fusion using cage and plate for the treatment of degenerative cervical spinal diseases. Asian Spine J. 2014; 8(6): 720–728, doi: 10.4184/asj.2014.8.6.720, indexed in Pubmed: 25558313.
19. Ishihara H, Kanamori M, Kawaguchi Y, et al. Adjacent segment disease after anterior cervical interbody fusion. Spine J. 2004; 4(6): 624–628, doi: 10.1016/j.spinee.2004.04.011, indexed in Pubmed: 15541693.
20. Park JB, Watthanaaphisit T, Riew KD. Timing of development of adjacent-level ossification after anterior cervical arthrodesis with plates. Spine J. 2007; 7(6): 633–636, doi: 10.1016/j.spinee.2006.10.021, indexed in Pubmed: 17998121.
21. Rao RD, Currier BL, Albert TJ, et al. Degenerative cervical spondylosis: clinical syndromes, pathogenesis, and management. Instr Course Lect. 2008; 57: 447–469, indexed in Pubmed: 18399602.
22. Bohlman HH, Emery SE, Goodfellow DB, et al. Robinson anterior cervical discectomy and arthrodesis for cervical radiculopathy. Long-term follow-up of one hundred and twenty-two patients. J Bone Joint Surg Am. 1993; 75(9): 1298–1307, doi: 10.2106/00004623-199309000-00005, indexed in Pubmed: 8408151.
23. Ferch RD, Shad A, Cadoux-Hudson TAD, et al. Anterior correction of cervical kyphotic deformity: effects on myelopathy, neck pain, and sagittal alignment. J Neurosurg. 2004; 100(1 Suppl Spine): 13–19, indexed in Pubmed: 14748568.
24. Dvorák J, Panjabi MM, Grob D, et al. Clinical validation of functional flexion/extension radiographs of the cervical spine. Spine (Phila Pa 1976). 1993; 18(1): 120–127, doi: 10.1097/00007632-199301000-00018, indexed in Pubmed: 8434312.
25. Tęsiorowski M, Lipik E, Zarzycki D, et al. Wyniki leczenia jedno- i wielopoziomowej szyjnej choroby dyskowej z zastosowaniem sztucznego dysku szyjnego. J Orthop Trauma Surg Rel Res. ; 3(15): 55–61.

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Association of trans-myocardial repolarisation parameters with size of the diffusion limitation area in acute ischaemic stroke

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ABSTRACT

Objectives. This study aimed to evaluate the relationship between transmyocardial repolarisation parameters and the size of the diffusion limitation area measured using diffusion weighted magnetic resonance imaging (DWMRI) in patients diagnosed with ischaemic stroke without known cardiac diseases.

Material and methods. The study was a prospective, observational clinical study. Patients without cardiac disease with acute ischaemic stroke were included in the study. Electrocardiography (ECG) was received from the patients. P, QT, QTc and Tp-e dispersions were calculated. All the patients had computerised brain tomography (CT) and then DWMRI carried out so as to calculate infarct areas.

Results. Seventy ischaemic stroke patients and 30 control patients were included in the study. All parameters except for QTc dispersion ($p = 0.88$) were higher in the stroke group than in the control group ($p < 0.05$ for all values). The infarct area calculated with DWMRI was divided into four groups according to quartiles, and QT, QTc, P, and Tp-e dispersions of patients were evaluated. Patients were found to have a prolonged dispersion as the infarct area expanded, and this difference was statistically significant ($p < 0.05$ for all values).

Conclusions. When we compared the patients with ischaemic stroke who had no known cardiac disease to those in the control group we found an increase in transmyocardial repolarisation parameters. As diffusion limitation areas grew larger, QT, QTc, P, and Tp-e dispersions increased. Physicians should be aware of dysrhythmias and sudden cardiac death in acute stroke and should observe these patients, especially those with larger stroke lesions.

Key words: stroke, diffusion-limitation area, trans-myocardial repolarisation parameters, electrocardiography

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Introduction

Strokes are among the most frequent and most important of all neurological disorders encountered during adulthood [1]. In the acute phase of a stroke, repolarisation abnormalities or ischaemic-like electrocardiographic changes can be observed [2]. Electrocardiographic abnormalities in patients with ischaemic and haemorrhagic strokes are a well-known problem which leads to diagnostic difficulties. In stroke patients, electrocardiographic changes have been observed without cardiac pathology [3]. The mechanism behind the electrocardiographic changes during an acute stroke has aroused great interest for years. It is thought that these changes could either be related to an underlying cardiac disorder, or indicate myocardial damage developing under acute stress. They might also result from neuro-hormonal interaction

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between the central nervous system and the cardiovascular system, which has been understood better in recent years [4, 5]. These abnormalities include ST segment changes (elevation or depression), changes in T waves, QT interval prolongation, and supraventricular or ventricular arrhythmias [2]. It has been reported that an increase in QT dispersion, which is regarded as an indicator of regional heterogeneity in myocardial repolarisation, leads to severe ventricular arrhythmias and sudden cardiac death [6, 7]. Even so, an increase in the Tpeak–Tend (Tp-e) interval from the peak of the T wave to the end, considered a new arithmogenic marker in ECG in recent times, has been shown to be related to life-threatening ventricular arrhythmias [8–10]. It has been demonstrated in previous studies that the above mentioned transmyocardial repolarisation parameters were extended in patients with a history of stroke [11–13].

In this study, we have evaluated the relationship between transmyocardial repolarisation parameters and the size of the diffusion limitation area measured using diffusion weighted magnetic resonance imaging (DWMRI) in patients diagnosed with ischaemic stroke without known cardiac diseases.

Material and methods

This study was a prospective and observational clinical study whose protocol was approved by the local ethics committee. The patients, who were aged over 18, were successively included in the study between 31 January and 1 June 2016 after they had received the diagnosis of an ischaemic stroke based on the clinical physical examination and the findings of imaging when they arrived at the emergency department within six hours of the first appearance of stroke symptoms. The exclusion criteria are set out in Table 1. The control group consisted of healthy subjects who were matched for age and sex, and had no malignancies, active infections, or coronary arterial disease.

Table 1. Exclusion criteria

Patients under 18 years of age
Patients with an accompanying acute coronary syndrome at admission
Those with a history of coronary arterial disease (stable / unstable angina, myocardial infarct)
Those undergoing a bypass operation
Those with transient ischaemic attack or lacunary infarct, and those who underwent haemorrhagic stroke
Pregnant women
Those with a history of cardiac medication (beta blocker, nitrate, calcium antagonist, digoxin)
Those who suffered cardiac embolism-related stroke
Patients with electrolyte disorders
Patients who had a branch block, pathological Q wave or atrial fibrillation, the criteria of left ventricle hypertrophy
Patients who refused to join in the study
Severe mitral or aortic valve disorder (stenosis or insufficiency)

All the patients were given detailed physical and neurological examinations, electrocardiography, imaging and echocardiographic imaging. Patients' neurological examinations were done according to the National Institutes of Health Stroke Scale (NIHSS).

The methods of imaging and their analyses

All the patients had computerised brain tomography (CT) and then DWMRI. Patients' CTs were obtained with a Toshiba (Japan) Activion 16 Detector tomography device, and DWMRI with a General Electric (USA) 1.5 Tesla closed MRI. All CTs and DWMRIs were evaluated by the same radiologist.

Measuring diffusion limitation area

Using a 1.5-T General Electric MR device, images with diffusion sensitivities of b 0 and b 1,000 s / mm^2 and 20 slice diffusion weighted images with a cross-sectional thickness of 5 mm were obtained. In cases with multiple acute infarct areas, the largest lesion was measured. On the iMAC medical image-processing system, the volumetric volume in cm^2 was automatically calculated from the images created with 1,000 s / mm^2 using the region of interest (ROI) from the widest outer edge of the lesion.

ECG

During the first hour, 12-channel ECGs were taken with a Nihon Kohden CardioFax GEN® (Japan) ECG device at 10 mm / mV and 25 mm / sec. ECG images were enlarged to 600-dpi resolution and read in Adobe Photoshop CS3 (USA). ECGs were assessed by two experts who were blinded to each other and the groups.

The QT interval was measured from the beginning of the QRS to the end of the T-wave in all derivations. QTc was calculated using the Bazzett formula ($QTc = QT / \sqrt{RR}$).

The Tp-e interval was determined from the T wave peak to the junction of the T wave with the isoelectric line junction. Tp-e was measured using the tangent method. P wave duration was measured from all leads. All dispersions were defined as the difference between the maximum and minimum duration in the measurements.

Statistical analysis

Analysis of the data was performed using the SPSS package program (version 15, Chicago, USA). The Shapiro-Wilk test was used to determine whether the distribution of continuous variables was normal. Descriptive statistics were expressed as mean ± standard deviation or median (quartiles) for continuous variables, and the number of cases (n) and (%) as categorical variables. Categorical variables were assessed by Chi-square test. Two independent sample t-tests were used to determine whether there was a statistically significant

Table 2. Demographic data of the patients

	Stroke patients	Control subjects	p value
Age, median (IQR 25–75%)	74 (68–82)	71 (68–80)	0.5
Gender, n (%)			
Female	25 (35.7)	13 (43.3)	0.5
Male	45 (64.3)	17 (56.7)	
Comorbidity, (%)			
Diabetes mellitus	7 (23.3)	28 (40)	0.16
Hypertension	14 (46.7)	42 (60)	0.15
COPD	4 (13.3)	4 (5.7)	0.23
Prior stroke, n (%)	13 (18.6)	—	
NIHSS score at admission, median (IQR 25–75%)	5 (0–31)	—	
Localisation, n (%)			
Right	41 (58.6)	—	
Left	23 (32.9)		
Bilateral	6 (8.6)		
Area, (cm^2)	1.5 (0.70–4.03)	—	

COPD — chronic obstructive lung disease; NHISS — national institutes of health stroke scale; IQR — inter-quartile range

Table 3. Characteristics of ECG parameters associated with trans-myocardial repolarisation in control subjects and stroke patients

	Stroke patients	Control subjects	p value
QT-min	386 (370–402)	365 (348–381)	< 0.05
QT-max	426 (398–458)	393 (370–409)	< 0.05
QT dispersion	39 (22–57)	25 (20–30)	< 0.05
QTc-min	441 (408–473)	398 (378–398)	< 0.05
QTc-max	490 (445–529)	441 (424–456)	< 0.05
QTc dispersion	44 (25–67)	42 (34–49)	0.88
P-min	67 (60–76)	65 (55–65)	< 0.05
P-max	90 (80–104)	76 (72–80)	< 0.05
P dispersion	21 (12–36)	15 (12–20)	0.017
Tp-e min	70 (62–76)	59 (52–69)	< 0.05
Tp-e max	90 (80–103)	73 (68–80)	< 0.05
Tp-e dispersion	20 (14–29)	13 (10–18)	< 0.05

ECG — electrocardiography

change in the mean values of the patient and control groups. Whether there was a statistically significant change in the median values and in the non-normal distribution data was examined using the Mann-Whitney U test. In the patient and control groups, Spearman correlation test was used to determine whether there was a statistically significant correlation between ECG measurements and infarct size. The Kruskal Wallis test was used to examine the significance of median values among the groups.

For $p < 0.05$, the results were considered statistically significant. However, Bonferroni correction was performed to check for Type I error in all possible multiple comparisons.

Results

Seventy ischaemic stroke patients, and 30 control patients with similar features, were included in the study. The mean age of stroke patients was 72 (68–80) years while that of the control group was 74 (68–82) years. 18.6% of the stroke patients had a stroke history. Patients' demographic data and characteristics are set out in Table 2.

When the transmyocardial parameters were evaluated in ECGs of patients and of the control group, all parameters except for QTc dispersion ($p = 0.88$) were higher in the stroke group than in the control group, and this difference was statistically significant ($p < 0.05$ for all values) (Tab. 3).

Table 4. Relationship between infarct areas and transmyocardial repolarisation parameters

	Correlation coefficient	p-value	Number of patients (n)
QT-min	0.543	< 0.05	70
QT-max	0.710	< 0.05	70
QT dispersion	0.608	< 0.05	70
QTc-min	0.442	< 0.05	70
QTc-max	0.651	< 0.05	70
QTc dispersion	0.636	< 0.05	70
P-min	0.083	0.49	70
P-max	0.486	< 0.05	70
P dispersion	0.517	< 0.05	70
Tp-e min	0.472	< 0.05	70
Tp-e max	0.689	< 0.05	70
Tp-e dispersion	0.663	< 0.05	70

Table 5. Parameters of transmyocardial repolarisation according to DWMRI-calculated infarct area quartiles

	DWMRI calculated infarct area quartiles (cm^2)				
	1 (< 0.70)	2 (0.71–1.50)	3 (1.51– 4.03)	4 (> 4.04)	p-value
Parameters	(n = 20)	(n = 19)	(n = 14)	(n = 17)	
QT dispersion	24 (20–38)	32 (20–40)	39 (24–47)	68 (52–76)	< 0.001
QTc dispersion	25.7 (22–42)	34 (24–58)	45 (33–49)	79 (65–86)	< 0.001
P dispersion	12 (10–20)	18 (12–28)	20 (7–27)	40 (36–44)	< 0.001
Tp-e dispersion	14 (10–20)	16 (14–24)	24 (20–24)	38 (34–50)	< 0.001

DWMRI — diffusion weighted magnetic resonance imaging

When the relationship between diffusion limitation areas and transmyocardial repolarisation parameters in the stroke patients was evaluated, a statistically significant similarity between the transmyocardial repolarisation parameters and diffusion limitation areas was found (correlation coefficients and p values are shown in Table 4) except for P-min (r = 0.083 and p = 0.49).

The diffusion limitation area calculated using DWMRI was divided into four groups according to quartiles and the QT, QTc, P, and Tp-e dispersions of patients were evaluated. Patients were found to have a prolonged dispersion as the diffusion limitation area expanded, and this difference was statistically significant ($p < 0.05$ for all values) (Tab. 5).

Discussion

According to the results of our study, transmyocardial repolarisation parameters were higher in patients with acute ischaemic stroke than in the control group. The transmyocardial repolarisation parameters in ischaemic stroke patients and the diffusion limitation areas measured with DWMRI were positively related. QT, QTc, P, and Tp-e dispersions were prolonged as the infarct areas expanded.

Ischaemic stroke is the most common of all stroke types. Markers and tests that can be used to determine prognosis, especially arrhythmia and the possibility of sudden death, in patients with frequent and high mortality strokes are helpful in the management of patients. Many studies have shown that cardiovascular abnormalities are caused by cerebral infarction, depending on its localisation and size [2, 14, 15]. Prolongation of the QT interval and enlargement of the QRS complex are electrical instability of the ventricular myocardium; ST-T changes, which are ischaemia-like changes, are the abnormalities most commonly observed on ECG [2]. Parameters such as QT, QTc, P, and Tp-e dispersions which can be used in the prediction of sudden death and arrhythmia and obtained only from a standard 12-lead ECG become increasingly attractive, due to their low cost and usefulness [6]. Measurements of these parameters in ECG reveal heterogeneity of cardiac repolarisation and useful parameters used in the definition of risky patients [6–8, 16, 17]. T wave peak-to-end point interval (Tp-e) measured in the ECG has recently been introduced into the literature, and is a parameter used to evaluate ventricular arrhythmogenity in many diseases [8, 9]. Studies comparing QT, QTc, and QTd for relatively long periods of time have shown that these studies yield reliable results at least as accurate as these measurements without signifying ventricular repolarisation [9].

It is thought that a stroke leads to ECG changes especially due to its effect on the autonomic nervous system, its capacity to cause haemodynamic changes, and to trigger catecholamine

release [18, 19]. For these reasons, cardiac involvement in cerebral lesions that have sustained certain areas of the brain for a long time has been the subject of studies. Since the hypothalamus and insulin have effects on the autonomic nervous system, various cardiac effects have been observed with the stimulation of these [18, 19]. We have already carried out a study in our own department. This, and other studies in the literature, have shown a prolonged range of transmyocardial repolarisation parameters that have been proven to be associated with ventricular arrhythmias and sudden cardiac death in stroke patients [13, 20, 21]. In addition, these parameters were evaluated not only for stroke, but also for stroke severity, type, localisation, and prognostic value. These parameters have been shown to be longer than stroke severity, ischaemic stroke in haemorrhagic stroke, and in patients with insula or brainstem involvement [13, 21-24]. The prognostic effect of QTc prolongation has been investigated in several studies. Most of these studies have shown that QTc prolongation adversely affects prognosis [11, 12].

There have been a number of studies evaluating the relationship between infarct area and QT dispersion in the literature; Pd, Tp-e interval, and Tp-e dispersion have not been identified in studies [22, 24]. In the study conducted by Chugh on patients with ischaemic and haemorrhagic stroke but without a cardiovascular disease history, patients with a large lesion on DWMRI were shown to have longer QTds on ECGs taken in the first 24 hours [22]. Also Avsar found the same result in a similar group of patients [24]. In our study, we showed that as the diffusion limitation area of the patients expanded, the myocardial repolarisation parameters were prolonged. Previous studies reported that Tp-e interval and Tp-e dispersion were longer in patients with long- and short-QT syndrome, Brugada syndrome, and myocardial infarction. [8, 25, 26]. Tp-e and Tp-e dispersions were validated in various cardiac conditions that led to sudden cardiac death [10].

We showed that trans-myocardial repolarisation parameters, including Pd, Tp-e and Tp-e dispersion, are longer in stroke patients, and especially in patients with a large diffusion limitation area.

We think that early attention should be paid to malignant ventricular arrhythmias and sudden cardiac death in these patients.

Limitations

Our study has some limitations. Firstly, our study comprised a relatively small sample size. Therefore, we believe that our results cannot be generalised to all populations. Secondly, none of the patients included in our study had arrhythmia during their entire hospital stay. This could have been due to the exclusion of patients with a known cardiac disease, cardiac drug use, and cardioembolic stroke in the aetiology.

Another limitation of our study was the absence of long-term follow-up of our patients. We have not evaluated the patients over a long time in terms of mortality, neurological status or cardiac intervention / implantation of pacemaker.

Finally, patients with other stroke types, including intracranial haemorrhages with fewer cases, were not included in the study. Patients were followed only during their hospitalisation period, and so could not be followed for long-term rhythm disturbances or mortality.

Conclusion

No studies in the literature have investigated the relationship between the diffusion limitation volume and other transmyocardial repolarisation parameters, including Pd, Tp-e interval and Tp-e dispersion in patients with ischaemic stroke. When we compared the patients with ischaemic stroke who had no known cardiac disease to those in the control group, we found an increase in these parameters. As diffusion limitation areas grew larger, QT, QTc, P, and Tp-e dispersions increased. We think that physicians should be aware of dysrhythmias and sudden cardiac death in ischaemic stroke patients and should observe these patients, especially those with larger stroke lesions.

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References

1. Aygencel G, Karamercan A, Akinci E, et al. Metabolic syndrome and its association with ischemic cerebrovascular disease. Adv Ther. 2006; 23(3): 495–501, indexed in Pubmed: 16912032.
2. Khechinashvili G, Asplund K. Electrocardiographic changes in patients with acute stroke: a systematic review. Cerebrovasc Dis. 2002; 14(2): 67–76, doi: 10.1159/000064733, indexed in Pubmed: 12187009.
3. Jensen JK, Bak S, Flemming Høilund-Carlsen P, et al. Prevalence of electrocardiographic ST-T changes during acute ischemic stroke in patients without known ischemic heart disease. Int J Cardiol. 2008; 128(1): 137–138, doi: 10.1016/j.ijcard.2007.05.055, indexed in Pubmed: 17683810.
4. Tokgözoglu SL, Batur MK, Topçuoglu MA, et al. Effects of stroke localization on cardiac autonomic balance and sudden death. Stroke. 1999; 30(7): 1307–1311, doi: 10.1161/01.str.30.7.1307, indexed in Pubmed: 10390300.
5. Oppenheimer SM, Cechetto DF, Hachinski VC. Cerebrogenic cardiac arrhythmias. Cerebral electrocardiographic influences and their role in sudden death. Arch Neurol. 1990; 47(5): 513–519, doi: 10.1001/archneur.1990.00530050029008, indexed in Pubmed: 2185720.
6. Kautzner J, Malik M. QT interval dispersion and its clinical utility. Pacing Clin Electrophysiol. 1997; 20(10 Pt 2): 2625–2640, doi: 10.1111/j.1540-8159.1997.tb06112.x, indexed in Pubmed: 9358510.
7. Aitchison JD, Campbell RW, Higham PD, et al. QT dispersion. Br Heart J. 1994; 71(6): 508–510, doi: 10.1136/hrt.71.6.508, indexed in Pubmed: 8043327.

8. Castro Hevia J, Antzelevitch C, Tornés Bárzaga F, et al. Tpeak-Tend and Tpeak-Tend dispersion as risk factors for ventricular tachycardia/ventricular fibrillation in patients with the Brugada syndrome. J Am Coll Cardiol. 2006; 47(9): 1828–1834, doi: 10.1016/j.jacc.2005.12.049, indexed in Pubmed: 16682308.
9. Morin DP, Saad MN, Shams OF, et al. Relationships between the T-peak to T-end interval, ventricular tachyarrhythmia, and death in left ventricular systolic dysfunction. Europace. 2012; 14(8): 1172–1179, doi: 10.1093/europace/eur426, indexed in Pubmed: 22277646.
10. Panikkath R, Reinier K, Uy-Evanado A, et al. Prolonged Tpeak-to-tend interval on the resting ECG is associated with increased risk of sudden cardiac death. Circ Arrhythm Electrophysiol. 2011; 4(4): 441–447, doi: 10.1161/CIRCEP.110.960658, indexed in Pubmed: 21593198.
11. Stead LG, Gilmore RM, Bellolio MF, et al. Prolonged QTc as a predictor of mortality in acute ischemic stroke. J Stroke Cerebrovasc Dis. 2009; 18(6): 469–474, doi: 10.1016/j.jstrokecerebrovasdis.2009.02.006, indexed in Pubmed: 19900651.
12. Hjalmarsson C, Bokemark L, Fredriksson S, et al. Can prolonged QTc and cTNT level predict the acute and long-term prognosis of stroke? Int J Cardiol. 2012; 155(3): 414–417, doi: 10.1016/j.ijcard.2010.10.042, indexed in Pubmed: 21093074.
13. Lederman YS, Balucani C, Lazar J, et al. Relationship between QT interval dispersion in acute stroke and stroke prognosis: a systematic review. J Stroke Cerebrovasc Dis. 2014; 23(10): 2467–2478, doi: 10.1016/j.jstrokecerebrovasdis.2014.06.004, indexed in Pubmed: 25282188.
14. Goldstein DS. The electrocardiogram in stroke: relationship to pathophysiological type and comparison with prior tracings. Stroke. 1979; 10(3): 253–259, doi: 10.1161/01.str.10.3.253, indexed in Pubmed: 462510.
15. Golan S, Livneh A. [ECG changes during stroke]. Harefuah. 2008; 147(6): 568–9, 572, indexed in Pubmed: 18693637.
16. Okutucu S, Aytemir K, Oto A. P-wave dispersion: What we know till now? JRSM Cardiovasc Dis. 2016; 5: 2048004016639443, doi: 10.1177/2048004016639443, indexed in Pubmed: 27081484.
17. Gialafos J, Dilaveris P, Gialafos E, et al. P Wave Dispersion: A Valuable Electrocardiographic Marker for the Prediction of Paroxysmal Lone Atrial Fibrillation. Annals of Noninvasive Electrocardiology. 1999; 4(1): 39–45, doi: 10.1111/j.1542-474x.1999.tb00363.x.
18. Oppenheimer S. Cerebrogenic cardiac arrhythmias: cortical lateralization and clinical significance. Clin Auton Res. 2006; 16(1): 6–11, doi: 10.1007/s10286-006-0276-0, indexed in Pubmed: 16477489.
19. Purushothaman S, Salmani D, Prarthana KG, et al. Study of ECG changes and its relation to mortality in cases of cerebrovascular accidents. J Nat Sci Biol Med. 2014; 5(2): 434–436, doi: 10.4103/0976-9668.136225, indexed in Pubmed: 25097430.
20. Emektar E, Çorbacıoğlu ŞK, Korucu O, et al. The evaluation of a new marker of transmyocardial repolarization parameters in ischemic stroke patients; T -T (T), T /QT. Acta Neurol Belg. 2017; 117(2): 461–467, doi: 10.1007/s13760-017-0744-4, indexed in Pubmed: 28110482.
21. Lazar J, Busch D, Wirkowski E, et al. Changes in QT dispersion after thrombolysis for stroke. Int J Cardiol. 2008; 125(2): 258–262, doi: 10.1016/j.ijcard.2007.03.114, indexed in Pubmed: 17509702.
22. - Ch, Garg A, Yadav A, et al. QT- dispersion in patients with stroke without known cardiac disease. JIACM. 2011; 12: 102–105.
23. Lazar J, Manzella S, Moonjelly J, et al. The prognostic value of QT dispersion in patients presenting with acute neurological events. J Invasive Cardiol. 2003; 15(1): 31–35, indexed in Pubmed: 12499526.
24. Afsar N, Fak AS, Metzger JT, et al. Acute stroke increases QT dispersion in patients without known cardiac diseases. Arch Neurol. 2003; 60(3): 346–350, doi: 10.1001/archneur.60.3.346, indexed in Pubmed: 12633145.
25. Letsas KP, Weber R, Astheimer K, et al. Tpeak-Tend interval and Tpeak--Tend/QT ratio as markers of ventricular tachycardia inducibility in subjects with Brugada ECG phenotype. Europace. 2010; 12(2): 271–274, doi: 10.1093/europace/eup357, indexed in Pubmed: 19897501.
26. Lubinski A, Kornacewicz-Jach Z, Wnuk-Wojnar AM, et al. The terminal portion of the T wave: a new electrocardiographic marker of risk of ventricular arrhythmias. Pacing Clin Electrophysiol. 2000; 23(11 Pt 2): 1957–1959, doi: 10.1111/j.1540-8159.2000.tb07061.x, indexed in Pubmed: 11139966.

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POLR3B-associated leukodystrophy: clinical, neuroimaging and molecular-genetic analyses in four patients: clinical heterogeneity and novel mutations in *POLR3B* gene

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ABSTRACT

Introduction and aim of the study. White matter disorders represent a spectrum of neurological diseases frequently associated with an unfavourable prognosis and a delay in diagnostics. We report the broad phenotypic spectrum of a rare hypomyelinating leukodystrophy and three novel mutations. Further, we aim to explore the role of the combined clinical and neuroimaging diagnostic approach in the era of whole exome sequencing.

Materials and methods. We present a clinical, neuroimaging and molecular-genetic characterisation of four patients from three families suffering from a rare genetic leukoencephalopathy. Two severely affected siblings (P1, P2) manifested a profound developmental delay, cerebellar symptomatology, microcephaly, failure to thrive, short stature and delayed teeth eruption with oligodontia. The other two patients (P3, P4), on the contrary, suffer from substantially less serious impairment with mild to moderate developmental delay and cerebellar symptomatology, delayed teeth eruption, or well-manageable epilepsy. In all four patients, magnetic resonance revealed cerebellar atrophy and supratentorial hypomyelination with T2-weight hypointensities in the areas of the ventrolateral thalamic nuclei, corticospinal tract and the dentate nuclei.

Results. Using whole-exome sequencing in P1, P2 and P3, and targeted sequencing in P4, pathogenic variants were disclosed in *POLR3B*, a gene encoding one of 17 subunits of DNA-dependent RNA polymerase III — all patients were compound heterozygotes for point mutations. Three novel mutations c.727A>G (p.Met243Val) and c.2669G>A (p.Arg890His) (P1, P2), and c.1495G>A (p.Met499Val) (P3) were found. Magnetic resonance revealed the characteristic radiological pattern of POLR3-leukodystrophies in our patients.

Conclusion and clinical implications. The diagnosis of POLR3-associated leukodystrophies can be significantly accelerated using the combined clinical and neuroradiological recognition pattern. Therefore, it is of crucial importance to raise the awareness of this rare disorder among clinicians. Molecular-genetic analyses are indispensable for a swift diagnosis confirmation in cases of clear clinical suspicion, and for diagnostic search in patients with less pronounced symptomatology. They represent an invaluable tool for unravelling the complex genetic background of heritable white matter disorders.

Key words: POLR3B, DNA-dependent RNA polymerase III, leukodystrophy, hypomyelination, hypodontia

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Introduction

Leukodystrophies represent a heterogenous group of heritable neurological disorders affecting the white matter of the central nervous system (CNS) with or without peripheral nervous system involvement [1]. They can be differentiated into hypomyelinating and demyelinating leukodystrophies using CNS magnetic resonance imaging (MRI) recognition patterns. CNS MRI in demyelinating leukodystrophies shows prominent hyperintensity of the white matter in the T2-weighted (T2-W) sequence and hypointensity in the T1-weighted (T1-W) sequence, as opposed to hypomyelinating leukodystrophies in which mild white matter hyperintensity on T2-W sequence and mild hypo-, iso- or hyperintensity on T1-W sequence can be observed [1, 2, 3]. The radiological criteria for hypomyelination further require an unchanged pattern of deficient myelination on two successive MRI scans at least six months apart, and at least one of them performed after the age of one year, separating thus delayed myelination and hypomyelination [2]. At least 91 heritable disorders affecting the white matter are known, 30 of which are the classic leukodystrophies with primary myelin infliction, the rest being formed by genetic leukoencephalopathies in which myelin homeostasis disruption is part of a systemic metabolic disorder [1].

The DNA-dependent RNA polymerase III-associated (POLR3-associated) leukodystrophies comprise a set of clinically, radiologically and genetically defined, yet heterogeneous, hypomyelinating leukodystrophies, for which a common aetiopathogenesis has been disclosed recently – mutations in the *POLR3A, POLR3B,* and *POLR1C* genes encoding subunits of the DNA-dependent RNA polymerase III. The prominent clinical symptoms of POLR3-associated leukodystrophies include progressive motor dysfunction or regression with hypotonia and marked cerebellar and pyramidal or extrapyramidal signs with a variable degree of intellectual impairment. Dental abnormalities constitute a distinct clinical sign. The severity of impairment may vary greatly among individual patients, and the disease may present in several distinct phenotypic forms. A specific radiological pattern exists offering a highly sensitive and specific diagnostic tool. Currently, only supportive symptomatic treatment exists [4–9].

Here, we present a thorough clinical, neuroimaging and molecular-genetic characterisation of four patients from three unrelated families. The sibling patients (P1, P2) presented severe both neurological and non-neurological phenotypes with early onset and rapid disease progression with profound developmental delay, various intriguing clinical features, and hypomyelination on CNS MRI. The other two patients (P3, P4) developed similar MRI findings, although they were much less dramatically affected with mild to moderate intellectual disability and other somatic symptoms that were fewer and less distinctly pronounced. Compound heterozygosity for mutations in *POLR3B* gene was found in all the patients, and three previously unreported mutations were revealed.

Case reports

Two affected siblings (P1, P2) and two other unrelated patients (P3, P4) born to healthy non-consanguineous parents are presented (Tab. 1).

P1, P2: The older sister (P1) was born prematurely in the 34th week of gestation with adequate birth weight of 2,300 g and length of 44 cm. The imminent postnatal adaptation was uneventful with an Apgar score of 9-10-10. Due to an early postnatal infection, the girl had had to be temporarily treated with antibiotics since her second day of life; however, recovery occurred within days, and discharge from hospital in a good clinical condition followed. Psychomotor milestones achievement was satisfactory until the age of six months, when a developmental deceleration and arrest were observed. Subsequently, the girl's clinical state started to deteriorate markedly. During regular clinical evaluations, she began to present with profound psychomotor retardation, central hypotonic syndrome, and a progressive cerebellar symptomatology. The prominent evolving features were dysmetria, dyskinetic choreatic movements, lower extremities spasticity, failure to thrive (17 kg, < 1st percentile at 10 years), short stature (114 cm, < 1st percentile at 10 years), microcephaly (45 cm, < 1st percentile at 10 years), convergent strabismus, central nystagmus, optic nerve atrophy, hypertelorism, inverted mammils and oligodontia with atypical teeth shape, and delayed eruption of both deciduous and permanent teeth (Fig. 1). Her clinical state is now still slowly progressive, predominantly concerning the cerebellar symptomatology. At the age of 11, she is neither able to sit nor to walk, and her cognitive development corresponds to the 3rd trimenone.

The second sister (P2) was born at term with a birth weight of 3,220 g and birth length of 49 cm. The clinical course and disease progression were very similar to her sister. At the age of seven months, developmental slowdown and practical arrest occurred. Progressively, substantial psychomotor retardation, hypotonic syndrome and cerebellar symptomatology evolved, with tremor, acral dyskinesias, failure to thrive (12.5 kg, < 1st percentile at six years), short stature (100 cm, < 1st percentile at 5.5 years), microcephaly (45 cm, < 1st percentile at six years), craniofacial dysmorphia (hypertelorism, enlarged auricles, mild macroglossia), strabismus, hypermetropia, inverted mammils and abnormal dental findings with oligodontia and delayed deciduous and permanent teeth eruption (Fig. 1). At the age of seven years, showing mild intrafamilial difference, she is able to sit unassisted and recently began to stand up and walk around the furniture, milestones her older sister never achieved . Her cognitive development is approximately at the level of eight months. Similarly to her sister, the cerebellar symptomatology is currently slowly progressive.

In both sisters, biochemical work-up did not reveal any abnormalities. Genetic analyses including micro-array hybridisation assays (aCGH) and targeted gene (*MECP2, SMN1*) analyses were negative. Thorough metabolic evaluation was also normal.

Table 1. Clinical symptoms in two severely (P1, P2) and two moderately (P3, P4) impaired patients with three novel variants in *POLR3B* gene and their comparison to literature

Clinical feature	Patient 1	Patient 2	Patient 3	Patient 4	Wolf et al., 2014 n = 103
Age of onset	6 months	7 months	3 years	2 years	< 10 years (90%)
Age at referral	10 years	6 years			n.d.
Developmental delay	+	+	+	+	most patients
Hypotonia	+	+	-	+	n.d.
Ataxia	+	+	+	+	almost all patients
Tremor	-	+	-	+	many patients
Dyskinesias	-	+	+	+	a few patients
Acral spasticity	+	-	+	+	+
Epilepsy	-	-	+	-	19%
Dental abnormalities	+	+	-	+	87%
Delayed dentition	+	+	-	+	71%
Hypo- /oligodontia	+	+	-	+	72%
Abnormal teeth shape	+	+	-	+	+
Microcephaly	+	+	-	-	n.d.
Short stature	+	+	-	n.d.	51 %
Failure to thrive	+	+	-	-	n.d.
Facial dysmorphy	+	+	-	-	n.d.
Inverted mammils	+	+	-	-	n.d.
Strabismus	+	+	-	-	n.d.
Nystagmus	+	+	-	-	most patients
Optic atrophy	+	-	+	-	+
Myopia	-	-	+	+	87%
Hypermetropia	-	+	-	-	n.d.

n.d. — not determined

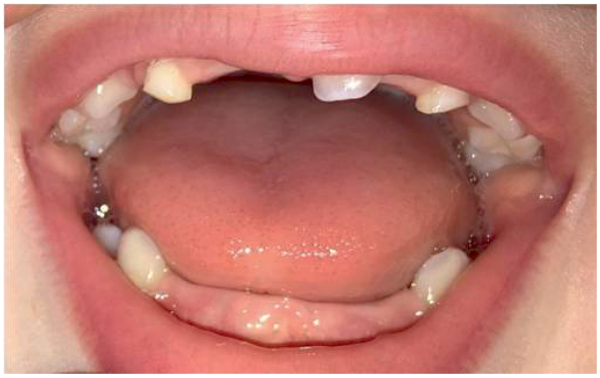

Figure 1. Oligodontia of P2 at the age of 6 years due to delayed eruption of both the deciduous and permanent teeth with atypically shaped teeth and enamel hypoplasia

P3: The perinatal data of the currently 28 year-old male P3 document delayed imminent postnatal adaptation necessitating resuscitation. Nevertheless, further development was unremarkable and appropriate until three years of age when slow retardation of psychomotor milestones achievement started to become apparent. Subsequently, apart from in the long term almost stationary mild psychomotor delay, clumsiness and behavioural problems began to develop. The clinical state then remained for a long time without any substantial progress. The patient attended a regular elementary school initially. Later on, however, he had to be assigned to a special educational programme. At the age of 11 years, the first epileptic seizure of a generalised tonic-clonic nature appeared. Antiepileptic therapy was initiated with a satisfactory effect, and epilepsy has since then been well-managed. In subsequent years, however, the patient's communication skills deteriorated mildly, and mood oscillations were noted. Furthermore, cerebellar and extrapyramidal symptomatology developed during the second decade of life and started to dominate the clinical picture, including ataxia, dyskinetic movements of the extremities and the neck, and hyperreflexia with hyp-/paresthesias of the lower limbs. Also, the patient's behavioural disorder then progressed with sexually inappropriate behaviour being noted. Psychological examination objectified a below average performance in the majority of the areas tested.

Marked myopia required ophthalmological correction. Due to the development of gynaecomastia, the patient underwent a thorough endocrinological examination, which yielded no abnormal findings. Neither dental abnormalities, nor growth impairment, failure to thrive, nor any other conspicuous symptoms were observed.

For the time being, the patient's clinical state seems to remain stationary, except for mild worsening of his cognitive functions.

Complex biochemical and metabolic workup were inconclusive, as were genetic analyses including the common microdeletion syndromes and X-linked mental retardations panel.

P4: Patient 4 is a 19 year-old woman born without any perinatal burden and with physiological birth anthropometric data. The early developmental milestones achievement remained within broader limits until two years of age, when mild psychomotor impairment started to become apparent. Teeth eruption was already delayed at that time. Simultaneously, growth velocity slowdown occurred and insufficient growth hormone production was confirmed. Substitution treatment was therefore initialised, with a satisfactory response. At around three years of age, apart from the developmental delay, intentional tremor with clumsiness and ataxia with frequent falls came to the forefront of the phenotypic picture and set off a broad diagnostic process. The patient's clinical condition subsequently slowly progressed, dominantly in the cerebellar component; choreiform dyskinesias and hyperreflexia together with pyramidal signs were noted further on. The absence of any signs of puberty initiation provoked endocrinology examinations disclosing borderline levels of both central and peripheral sex-hormones. Thus, hormonal substitution therapy was commenced. However, psychiatric problems including depression, food refusal and pseudohallucinations occurred thereafter. These were judged however to be a possible adverse consequence of the hormonal therapy. Another CNS MR at 16 years of age revealed mild progression of the cerebellar atrophy; yet the neurological findings of the cerebellar symptomatology seem to be long-term stationary. In summary, the current clinical state of P4 is non-progressive and dominated by mild intellectual disability, cerebellar symptomatology, optic atrophy and markedly disrupted dental eruption resulting in oligodontia with abnormally shaped teeth.

As with the other patients, biochemical and metabolic examinations remained unremarkable. Genetic testing, aiming at e.g. various spinocerebellar ataxias, yielded no results.

Material and methods

Exome sequencing of the trio (proband(s) and unaffected parents; P1, P2, P3) was performed using an Illumina HiSeq 2000 system (Illumina, USA) and SeqCap EZ Exome Enrichment kit v3.0 (Roche NimbleGen, USA) [10]. Sanger sequencing of all exons and exon–intron boundaries of *POLR3B* gene (NG_031837.1, NM_018082.5) was used in P4. *POLR3B* mutations identified by exome sequencing were confirmed by Sanger sequencing in probands and their parents.

Ethics

This study was approved by the ethics committee of the General University Hospital in Prague and was conducted in agreement with institutional guidelines. Written informed consent was obtained from parents for genetic analysis.

Results and Discussion

Mutations affecting POLR3 subunits, in particular the two largest subunits POLR3A (DNA-dependent RNA-polymerase III, subunit A) and POLR3B (DNA-dependent RNA-polymerase III, subunit B), are causative of a continuous spectrum of allelic and clinically, radiologically and genetically defined group of hypomyelinating leukodystrophic disorders categorised as POLR3-associated leukodystrophies. This group includes five disorders initially described as individual entities: Hypomyelination with Oligodontia (HO), Tremor-Ataxia with Central Hypomyelination (TACH), Hypomyelination with Cerebellar Atrophy and Hypoplasia of the Corpus Callosum (HCAHCC), Ataxia, Delayed Dentition and Hypomyelination (ADDH) and Hypomyelination, Hypodontia, Hypogonadotropic Hypogonadism (4H). With the expansion of molecular genetic testing, these disorders have been revealed to have a similar genetic basis: mutations in genes encoding for POLR3 subunits. Therefore, rather than separate diseases, they represent a broad phenotypic spectrum with distinct phenotypes of the so-called POLR3-associated leukodystrophies [11, 12, 13]. They represent an extremely rare disorder with single case reports reported worldwide. However, as there is no database gathering these patients, neither the precise number of patients suffering from POLR3-associated leukodystrophies nor other epidemiological data are currently available. No more than a few hundred patients have been reported in the literature, the largest cohort amounting to 105 patients [11]. Genetic testing in our patient cohort revealed that all patients were compound heterozygotes for point mutations in *POLR3B* gene, a gene encoding one of the 17 subunits of the DNA-dependent RNA polymerase III.

In P1 and P2, with the severe phenotype, two novel heterozygous mutations — maternally inherited c.727A>G (p.Met243Val) and paternally inherited c.2669G>A (p.Arg890His) — were found. Using exome sequencing, P3 was found to bear a novel variant c.1495G>A (p.Met499Val) inherited from his mother and a previously characterised pathogenic variant c.2084-6A>G leading to a frameshift and a premature stop codon (p.Gly695Valfs*5) [5]. The novel variants change evolutionally conserved amino acids. The impact of these three missense mutations on *POLR3B* was assessed with MutationTaster [14], PolyPhen2 [15] and SIFT [16] programmes and mutations were predicted to be pathogenic. Sequence analysis

Table 2. MRI findings in two severely (P1, P2) and two moderately (P3, P4) impaired patients with three novel variants in *POLR3B* gene

MRI feature	Patient 1	Patient 2	Patient 3	Patient 4
Age at MRI examination (years)	10	5	15	16
Diffuse supratentorial hypomyelination	+	+	+	+
T2-weight hypointensities				
ventrolateral thalamus	+	+	+	+
globus pallidus	-	-	+	+
corticospinal tracts (internal capsule)	+	+	+	+
nucleus dentatus	+	+	+	+
optic radiation	-	-	-	-
Cerebellar atrophy	+	+	+	+
Cortical dysplasia	+	+	-	-
Arachnoideal cysts	-	+	-	-

of the *POLR3B* in P4 revealed compound heterozygosity for two reported mutations, a frameshift variant c.2570+1G>A leading to a premature truncated protein (p.Gly818fs), and a common missense mutation c.1568T>A (p.Val523Glu) found in patients from a European background, homozygosity of which causes a milder phenotype. Apart from this exception, patients with compound heterozygous mutations show no difference in disease progression from those bearing homozygous mutations [17].

POLR3 is one of the three DNA-dependent RNA polymerases found in eukaryotic cell nuclei, each of them being responsible for the transcription of a specific set of genes [5, 18]. POLR3 transcribes a number of various non-coding RNAs involved in essential cellular processes such as translation, RNA processing or transcription regulation [19]. POLR3 is composed of 17 subunits, the two largest of which, POLR3A and POLR3B, form the catalytic centre of the enzyme and are responsible for the vast majority of POLR3-associated leukodystrophies [11, 18]. A mutation in POLR1C – a POLR1 (DNA-dependent RNA polymerase I) and POLR3 shared subunit previously associated with the autosomal recessive Treacher Collins syndrome, has recently been proved to also be causative of a minority of cases of POLR3-associated leukodystrophies [20].

The pathophysiological pathway underlying POLR3-leukodystrophies remains elusive. However, it is presumed that either defective POLR3 assembly, stability or nuclear import or decreased enzymatic activity could affect the levels of various RNAs, which in turn are indispensable for embryonic CNS myelin formation and subsequent myelin homeostasis [5]. POLR3 function is cell-type and cell-cycle dependent. With certain transcripts being brain-specific, this might be the mechanism partially explanatory of the profound CNS impairment and the relative sparing of other tissues and organs [21]. However, the cause of the common dental impairment still remains unclear, as does the endocrine infliction frequently seen in POLR3 patients.

The CNS MRI findings of POLR3-associated leukodystrophies comprise a set of features forming together a highly sensitive (84.6%) and specific (92.9%) recognition pattern [9]. The characteristic POLR3-leukodystrophy CNS MRI features include diffuse supratentorial hypomyelination accompanied by relative T2-W hypointensities in the areas of the globus pallidus, ventrolateral nuclei of the thalamus, corticospinal tract (at the level of the posterior limb of the internal capsule) and in the dentate nucleus and the optic radiation. Cerebellar atrophy and thinning of the corpus callosum complete the neuroradiological pattern characteristic of POLR3-leukodystrophies (Tab. 2). Patients not bearing this MRI pattern are unlikely to suffer from POLR3-associated leukodystrophy [3, 9, 22].

Following standard procedures, CNS MRI was performed at the age of 32 months and at seven and 10 years in P1, at 15 months, 28 months and five years in P2, at 15 years at P3, and at eight and 16 years in P4. All our patients meet the majority of the criteria of the POLR3-associated leukodystrophy recognition pattern, with diffuse non-progressive symmetrical supratentorial hypomyelination and cerebellar atrophy (Fig. 2), affecting both vermis and the hemispheres. Cerebellar atrophy is slightly more common in POLR3B cases compared to POLR3A patients [11]. The degree of white matter hypomyelination reached approximately similar levels among all the patients. Cerebellar atrophy seems to be less prominent in P3 (Fig. 2F), arguably corresponding to a milder cerebellar symptomatology in this patient. The typical T2-W hypointensities in the regions of the ventrolateral thalami (Fig. 2E, H), the dentate nuclei (Fig. 2I) and the corticospinal tract at the level of the posterior limb of the internal capsule (Fig. 2E, H, Fig. 3B) were observed in all our patients. Hypointensities in the globi pallidi (Fig. 2H, K) were only seen in P3 and P4 (Tab. 2). Optic radiation involvement, also often accompanying

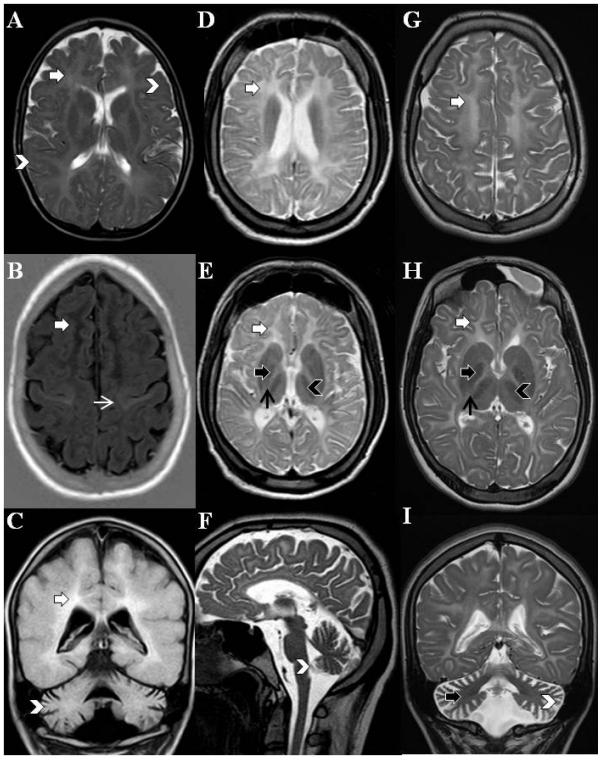

Figure 2. CNS MRI of P1, P2, P3 and P4. Diffuse hypomyelination (white arrows) visible as white matter T2-weight or FLAIR (A, C, D, G, E, H) hyperintensity and T1-weight hypointensity (B) is documented in P1 (B), P2 (A, C), P3 (D, E, F) and P4 (G, H, I). Mildly asymmetric gyrification compatible with the diagnosis of nonlissencephalic cortical dysplasia with polymicrogyria (A; white arrowheads) is depicted in P2. Cerebellar atrophy, affecting both vermis and the hemispheres, can be observed in FLAIR (C; white arrowheads) and T2-weight sequences (F, I; white arrowheads). T2-weight hypointensity in the area of nucleus dentatus (I; black arrow) can be observed in P4. T2-weight hypointensities in globi pallidi (E, H; black arrows), ventrolateral thalami (E, H; black arrowheads) and the corticospinal tract at the level of the posterior limb of the internal capsule (E, H; black thin arrows) are demonstrated in axial images of P3 and P4. Preserved myelination of gyrus praecentralis and of the corticospinal tract upstream of internal capsule (B; white thin arrow) corresponding to T1-weight hyperintensity is observed in P1

POLR3-leukodystrophies, was not present in either of these patients [9]. Thin hypoplastic corpus callosum, described in approximately 50% of children younger than 10 years and more frequently associated with *POLR3B* mutations, was found in all four patients (Fig. 3A). Interestingly, both P1 and P2 expressed additional findings of dysmyelination of the white matter with asymmetric gyrification compatible with a diagnosis of nonlissencephalic cortical dysplasia in both sisters - cortical dysplasia with pachygyria in P1, and cortical dysplasia and polymicrogyria in P2 (Fig. 2B). The aetiology of these findings was not clarified; nevertheless, cortical dysplasia has never been described in any POLR3 patient before. Given its simultaneous occurrence in both siblings, the presence of another underlying genetic cause cannot be excluded, despite negative findings in the whole exome sequencing. P2 was also noted to possess a temporobasal enlargement of the external cerebrospinal fluid spaces due to an arachnoid cyst (30 x 25 x 22 mm). However, as arachnoid cysts are not an especially rare finding even in healthy asymptomatic populations, they cannot be related to POLR3-leukodystrophies. In a subset of patients, a small cyst within the splenium has been observed [11].

The onset of POLR3-associated leukodystrophies ranges from early childhood to adolescence, with only a minority (10%) of patients presenting after 10 years of age. The prominent clinical features include progressive motor dysfunction or regression with hypotonia and marked cerebellar and pyramidal or extrapyramidal signs, presenting as gait abnormalities, ataxia, tremor, dysmetria, abnormal eye smooth pursuit, nystagmus or other gaze limitations, dystonia, diskinetic movements, spasticity or hyperreflexia and others [11] (Tab. 1).

In spite of the few exceptions documented, POLR3A-associated disease has been found to be associated with a more rapid disease progression, more severe disease course, and shorter life expectancy, despite having a slightly later disease onset compared to patients bearing mutations in *POLR3B*. Patients from a European background are more likely to have mutations in *POLR3B* [11, 23].

The clinical presentation in both our severely (P1, P2) and moderately (P3, P4) impaired patients is in accordance with the hitherto described broad phenotypic spectrum of POLR3-associated leukodystrophies.

As with the majority of patients, both siblings manifested early and similarly with developmental delay, cerebellar and pyramidal symptomatology and a corresponding dental abnormality pattern, which to a lesser extent was also the clinical presentation of P4. On the other hand, P3 manifested with unprogressive mild to moderate intellectual disability with behavioural problems and unspecified clumsiness in early childhood, with a subsequent onset of epilepsy at the beginning of his second decade and further deterioration of cognitive skills and the slow progression of other symptoms. No dental abnormalities have been detected in P3 so far, although they manifest in around three quarters of patients [11] (Tab. 1). Seizures occurred neither in the sibling patients (P1, P2), nor in P4. Because epilepsy is present in only approximately 20% of patients, these patients may remain without seizures. However, if epilepsy eventually evolves, it is usually well-controllable by pharmacotherapy [11]. Cognitive impairment of POLR3 patients can be of variable severity. Our sibling patients manifested a profound intellectual disability, while most described patients have developed only a mild to moderate impairment, just like P3 and P4. Some patients may even have normal cognitive abilities, or only variably expressed learning difficulties [11]. Intercurrent infections have been noted to worsen the disease course, with not all patients

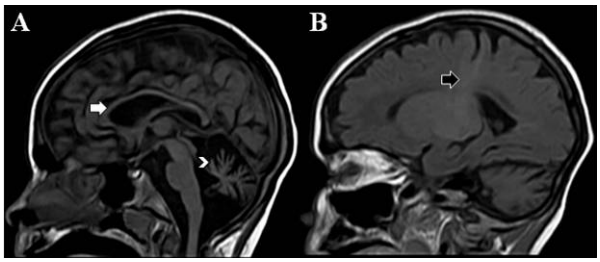

Figure 3. CNS MRI in P1, T1-weight sequence. Atrophy of the corpus callosum (A; white arrow) and of the cerebellum (A; white arrowhead) is demonstrated, together with preserved myelinisation of the corticospinal tract and its proximity upstream of the internal capsule (B; black arrow). Similar findings have been found in P2, the sister of P1

regaining their previous level [11], although this phenomenon has not so far been observed in our patients.

The non-neurological symptoms are dominated by dental abnormalities, with an abnormal or delayed deciduous and definitive teeth eruption, and hypo- /oligodontia and dysmorphic teeth, as seen in P1, P2 and P4. Other non-neurological symptoms, as observed in P4, may include endocrine dysfunctions such as prolactin or growth hormone deficiencies or hypogonadotropic hypogonadism, which leads to delayed puberty in three quarters of POLR3B patients [11, 24, 25]. Short stature, affecting about 50% of patients, was a dominant feature in our siblings, as well as microcephaly and failure to thrive. Worthy of note concerning the POLR3-leukodystrophies is the substantial diversity in disease onset and clinical course severity and progression, including even intrafamilial heterogeneity [11]. Progressive myopia seems to be a frequent part of the syndrome, as observed in our moderately affected P3 and P4; cataracts can sometimes develop. Interestingly, convergent strabismus has not been mentioned yet, while optic atrophy, as seen in P1 and P3, has only been described occasionally and mostly in older patients [23].

The precise pathophysiological mechanism leading to the substantial difference in the clinical phenotype severity between P1, P2 with two novel mutations and the other two patients, P3 and P4, remains currently elusive and cannot be correlated purely to CNS MRI findings, which are of approximately similar severity. Additional analyses need to be performed. Since *POLR3B* possesses a high degree of mutational heterogeneity with mutational sites distributed throughout the whole gene, further studies on more patients are necessary in order to determine a precise genotype-phenotype correlation, something which cannot be drawn for the time being [5].

Clinical implications and conclusion

Leukodystrophies are a growing group of heritable white matter disorders, for which comprehensive diagnostic algorithms have been created to assist in reaching the proper diagnosis as early as possible [1].

In certain cases, the clinical and neuroimaging signs and symptoms may assist in narrowing the differential diagnosis and focusing the diagnostic process, including the molecular-genetic analyses, as witnessed by POLR3-associated leukodystrophies [3, 9].

Therefore, awareness of POLR3-associated leukodystrophies ought to be increased among clinicians, as this particular entity offers highly specific and sensitive clinical and neuroimaging recognition patterns, making thereby targeted sequencing a method of choice, rather than whole exome sequencing. This may be of substantial significance not only for clinicians, but also for patients and the general healthcare system.

However, there still are a number of mono-/oligosymptomatic phenotypes, some of them especially with the non-neurological presenting symptoms, which may pose a more challenging diagnostic task and thus require broader molecular-genetic analyses, such as whole exome sequencing [26].

Such sequencing, whether genome, exome or targeted panel sequencing, offers a valuable diagnostic tool if properly used. Concomitantly, it offers an opportunity to further unravel the underlying genetic and pathophysiological background of these disorders, and shift today's mostly symptomatic care towards a search for disease-specific targeted therapies and novel therapeutic modalities (for an overview of novel therapies see Helman et al. [27]).

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References

1. Ashrafi MR, Tavasoli AR. Childhood leukodystrophies: A literature review of updates on new definitions, classification, diagnostic approach and management. Brain Dev. 2017; 39(5): 369–385, doi: 10.1016/j.braindev.2017.01.001, indexed in Pubmed: 28117190.
2. Schiffmann R, van der Knaap MS. Invited article: an MRI-based approach to the diagnosis of white matter disorders. Neurology. 2009; 72(8): 750–759, doi: 10.1212/01.wnl.0000343049.00540.c8, indexed in Pubmed: 19237705.
3. Steenweg ME, Vanderver A, Blaser S, et al. Magnetic resonance imaging pattern recognition in hypomyelinating disorders. Brain. 2010; 133(10): 2971–2982, doi: 10.1093/brain/awq257, indexed in Pubmed: 20881161.

4. Bernard G, Thiffault I, Tetreault M, et al. Tremor-ataxia with central hypomyelination (TACH) leukodystrophy maps to chromosome 10q22.3--10q23.31. Neurogenetics. 2010; 11(4): 457–464, doi: 10.1007/s10048-010-0251-8, indexed in Pubmed: 20640464.
5. Daoud H, Tétreault M, Gibson W, et al. Mutations in POLR3A and POLR3B are a major cause of hypomyelinating leukodystrophies with or without dental abnormalities and/or hypogonadotropic hypogonadism. J Med Genet. 2013; 50(3): 194–197, doi: 10.1136/jmedgenet-2012-101357, indexed in Pubmed: 23355746.
6. Saitsu H, Osaka H, Sasaki M, et al. Mutations in POLR3A and POLR3B encoding RNA Polymerase III subunits cause an autosomal-recessive hypomyelinating leukoencephalopathy. Am J Hum Genet. 2011; 89(5): 644–651, doi: 10.1016/j.ajhg.2011.10.003, indexed in Pubmed: 22036171.
7. Sasaki M, Takanashi Ji, Tada H, et al. Diffuse cerebral hypomyelination with cerebellar atrophy and hypoplasia of the corpus callosum. Brain Dev. 2009; 31(8): 582–587, doi: 10.1016/j.braindev.2008.09.003, indexed in Pubmed: 18851904.
8. Timmons M, Tsokos M, Asab MA, et al. Peripheral and central hypomyelination with hypogonadotropic hypogonadism and hypodontia. Neurology. 2006; 67(11): 2066–2069, doi: 10.1212/01.wnl.0000247666.28904.35, indexed in Pubmed: 17159124.
9. La Piana R, Tonduti D, Gordish Dressman H, et al. Brain magnetic resonance imaging (MRI) pattern recognition in Pol III-related leukodystrophies. J Child Neurol. 2014; 29(2): 214–220, doi: 10.1177/0883073813503902, indexed in Pubmed: 24105487.
10. Mazurova S, Magner M, Kucerova-Vidrova V, et al. Thymidine kinase 2 and alanyl-tRNA synthetase 2 deficiencies cause lethal mitochondrial cardiomyopathy: case reports and review of the literature. Cardiol Young. 2017; 27(5): 936–944, doi: 10.1017/S1047951116001876, indexed in Pubmed: 27839525.
11. Wolf NI, Vanderver A, van Spaendonk RML, et al. 4H Research Group. Clinical spectrum of 4H leukodystrophy caused by POLR3A and POLR3B mutations. Neurology. 2014; 83(21): 1898–1905, doi: 10.1212/WNL.0000000000001002, indexed in Pubmed: 25339210.
12. Bernard G, Vanderver A. POLIII-Related Leukodystrophies. In: GeneReviews® [Internet] Pagon RA, Adam MP, Ardinger HH, Wallace SE, Amemiya A, Bean LJH, Bird TD, Ledbetter N, Mefford HC, Smith RJH, Stephens K, editors. University of Washington, Seattle. 2012: 1993–2017.
13. Tétreault M, Choquet K, Orcesi S, et al. Recessive mutations in POLR3B, encoding the second largest subunit of Pol III, cause a rare hypomyelinating leukodystrophy. Am J Hum Genet. 2011; 89(5): 652–655, doi: 10.1016/j.ajhg.2011.10.006, indexed in Pubmed: 22036172.
14. Schwarz JM, Cooper DN, Schuelke M, et al. MutationTaster2: mutation prediction for the deep-sequencing age. Nat Methods. 2014; 11(4): 361–362, doi: 10.1038/nmeth.2890, indexed in Pubmed: 24681721.
15. Adzhubei I, Jordan DM, Sunyaev SR. Predicting functional effect of human missense mutations using PolyPhen-2. Curr Protoc Hum Genet. Chapter 7: Unit 7. 20 1-7. 2013; 20: 41.
16. Kumar P, Henikoff S, Ng PC. Predicting the effects of coding non-synonymous variants on protein function using the SIFT algorithm. Nat Protoc. 2009; 4(7): 1073–1081, doi: 10.1038/nprot.2009.86, indexed in Pubmed: 19561590.
17. Gutierrez M, Thiffault I, Guerrero K, et al. Large exonic deletions in POLR3B gene cause POLR3-related leukodystrophy. Orphanet J Rare Dis. 2015; 10: 69, doi: 10.1186/s13023-015-0279-9, indexed in Pubmed: 26045207.
18. Hu P, Wu Si, Sun Y, et al. Characterization of human RNA polymerase III identifies orthologues for Saccharomyces cerevisiae RNA polymerase III subunits. Mol Cell Biol. 2002; 22(22): 8044–8055, doi: 10.1128/mcb.22.22.8044-8055.2002, indexed in Pubmed: 12391170.
19. Bernard G, Chouery E, Putorti ML, et al. Mutations of POLR3A encoding a catalytic subunit of RNA polymerase Pol III cause a recessive hypomyelinating leukodystrophy. Am J Hum Genet. 2011; 89(3): 415–423, doi: 10.1016/j.ajhg.2011.07.014, indexed in Pubmed: 21855841.
20. Thiffault I, Wolf NI, Forget D, et al. Recessive mutations in POLR1C cause a leukodystrophy by impairing biogenesis of RNA polymerase III. Nat Commun. 2015; 6: 7623, doi: 10.1038/ncomms8623, indexed in Pubmed: 26151409.
21. Haurie V, Durrieu-Gaillard S, Dumay-Odelot H, et al. Two isoforms of human RNA polymerase III with specific functions in cell growth and transformation. Proc Natl Acad Sci U S A. 2010; 107(9): 4176–4181, doi: 10.1073/pnas.0914980107, indexed in Pubmed: 20154270.
22. La Piana R, Cayami FK, Tran LT, et al. Diffuse hypomyelination is not obligate for POLR3-related disorders. Neurology. 2016; 86(17): 1622–1626, doi: 10.1212/WNL.0000000000002612, indexed in Pubmed: 27029625.
23. Jurkiewicz E, Dunin-Wąsowicz D, Gieruszczak-Białek D, et al. Recessive Mutations in POLR3B Encoding RNA Polymerase III Subunit Causing Diffuse Hypomyelination in Patients with 4H Leukodystrophy with Polymicrogyria and Cataracts. Clin Neuroradiol. 2017; 27(2): 213–220, doi: 10.1007/s00062-015-0472-1, indexed in Pubmed: 26478204.
24. Billington E, Bernard G, Gibson W, et al. Endocrine Aspects of 4H Leukodystrophy: A Case Report and Review of the Literature. Case Rep Endocrinol. 2015; 2015: 314594, doi: 10.1155/2015/314594, indexed in Pubmed: 26113998.
25. Potic A, Popovic V, Ostojic J, et al. Neurogenic bladder and neuroendocrine abnormalities in Pol III-related leukodystrophy. BMC Neurol. 2015; 15: 22, doi: 10.1186/s12883-015-0283-7, indexed in Pubmed: 25868523.
26. Richards MR, Plummer L, Chan YM, et al. Phenotypic spectrum of POLR3B mutations: isolated hypogonadotropic hypogonadism without neurological or dental anomalies. J Med Genet. 2017; 54(1): 19–25, doi: 10.1136/jmedgenet-2016-104064, indexed in Pubmed: 27512013.
27. Helman G, Van Haren K, Bonkowsky JL, et al. GLIA Consortium. Disease specific therapies in leukodystrophies and leukoencephalopathies. Mol Genet Metab. 2015; 114(4): 527–536, doi: 10.1016/j.ymgme.2015.01.014, indexed in Pubmed: 25684057.

SHORT COMMUNICATION

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Migraine headache facilitators in a population of Polish women and their association with migraine occurrence — preliminary results

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ABSTRACT

Aim of the study. The occurrence of migraine is linked with some common lifestyle activities and conditions preceding the attack. Our study presents known and presumptive lifestyle factors and activities related to migraine, and compares them to the frequency of headache attacks.

Material and methods. 40 female patients of the Headache Outpatient Clinic in Warsaw, Poland, diagnosed with migraine, mean age 44.6 years, and 40 female participants from the control group, mean age 39.5 years, were included in the study. The study employed questionnaires reporting the presence of lifestyle factors and socioeconomic predispositions as well as the Migraine Disability Assessment Test (MIDAS) as data collection methods.

Results. Correlations between some of the lifestyle factors and the frequency of migraines occurred statistically significantly.

Conclusions. Some factors and lifestyle activities such as stress, relaxation, specific dietary products, fasting, fatigue, bright light, noise, weather changes or menstruation may have an influence on migraine frequency and severity in female patients, which can have an impact on migraine prevention.

Key words: migraine, trigger factors, lifestyle, headaches

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Introduction

Migraine is a common health problem occurring in up to 12% of the Caucasian population. According to epidemiological studies in Poland, the cause of about 49% of chronic daily headaches (CDH) is chronic migraine [1]. The prevalence of migraine is up to three times more frequent among women depending on populations, and varies by age, with its peak between the ages of 35 and 45, regardless of sex [2, 3]. Stępień et al. determined the prevalence of migraine in the Polish population to be about 10%. Based on their study, 75% of Polish migraineurs were female and migraine was more common in patients over 40 years old (72%) [4]. Low household income and lower educational level can further increase migraine prevalence, significantly among women [5, 6]. Migraine can have a major impact on the patient's quality of life, affecting it directly and causing complications such as functional disability or depression [2, 7, 8].

Peroutka tried to identify the top ten migraine factors based on an analysis of 25 publications, of which stress was predominant [9]. Between 50% and 80% of migraine patients associated stress with headaches [9, 10]. Other frequently mentioned factors include sleep disturbance and dietary factors like hunger, skipping meals, alcohol intake (especially red wine and beer), caffeine, chocolate and cheese, fatigue and weather changes [9, 11, 12, 13]. Physical activity can further worsen the severity of headaches induced by alcohol intake [14]. Sleep and stress can play a greater role for patients suffering from migraine with aura, as opposed to environmental factors which may be more crucial to migraine patients without aura

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[10]. Another Polish study focused on the potential impact of body mass index (BMI) and serum lipid levels on increasing the risk of migraine [15].

Determining headache trigger factors among migraine patients could possibly help improve prevention in this group. Due to insufficient data in the Polish population, our present study was conducted to analyse the association between migraine and factors which are known to trigger, or are presumed to trigger, migraine headaches such as marital status, educational level, alcohol intake, sleep deprivation, dietary problems, stress, weather changes, exposure to flashing lights or a high noise level, staying at a high altitude, drug administration, menstruation, or working out.

Material and methods

We retrospectively evaluated the clinical data of 40 randomly selected female patients with a mean age (± SD) of 44.62 ± 10.64 years (range 21 to 66) diagnosed with episodic or chronic migraine with or without aura, based on the criteria of the International Headache Society [14]. Our control group consisted of 40 women with chronic tension type headaches (TTH) diagnosed according to the International Classification of Headache Disorders 3rd edition (ICHD-3) [14], with a mean age (± SD) of 39.5 ± 15.59 years (range 23 to 76 years). This group was adjusted to the age and gender of the study group. All individuals in our study groups (the control-TTH group and the migraine group) were Caucasians. The mean age for the study groups was not significantly different (Wald-Wolfowitz runs test – $Z = -1.350$; $p = 0.177$). The consecutively incoming migraine patients were examined in the Headache Outpatient Clinic from November 2015 to February 2017. A basic neurological and general examination was performed during planned appointments in the Clinic. Depending on individual needs, the patients were receiving either prophylactic or acute treatment. During our study, there were only a few male patients, so we excluded them from the study to avoid selection bias,deciding to pay attention only to the unified female patients group and the control group. Our study group did not omit individuals in migraine preventive treatment or those receiving medication for chronic migraine. We also did not select patients for the study on the basis of the onset of migraine or its duration.

All the patients completed a questionnaire that was created for the researchers' own use. Each set of questions was asked directly by the researchers, not over the telephone, after the confirmation of the migraine diagnosis and the examination of physical condition. All the patients agreed to their data included in the questionnaire being used for our study.

The questionnaire was divided into three parts. In the first, socioeconomic, part we assessed educational level, type of work (physical or mental), marital status and number of children. In the second part, each patient was asked about the concomitance of headaches and the following aspects of life: a stressful situation (e.g. exams, work interviews), relaxation after stress, the intake of dietary products such as chocolate, dairy, cocoa, seasonings, citrus, as well as alcohol intake, fasting, sleep deficiency or excess, workout/fatigue, exposure to bright or pulsing light, high levels of noise, staying at high altitude (e.g. in the mountains), sudden weather changes, medicine intake, and menstruation. We also asked patients if there were any other factors that they had linked with their headaches. In the final part, the Migraine Disability Assessment Test (MIDAS) [16] was used as a means to estimate patients' migraine severity and daily activity impairment. We compared triggers of headache in the migraine and in the TTH group. The complete questionnaire is set out in Table 1.

Statistical analysis

Non-parametric Wald-Wolfowitz runs test was performed for the group comparison. Null hypothesis was that the groups were independent and identically distributed, which was proved correct (p-value 0.177).

The presumptive headache trigger factors were presented as numerical variables and were analysed with chi-square tests such as Pearson's Chi-square test, Yates Chi-square test, and V-square test. The null hypothesis in this case, and for all the subsequent tests, was that a statistical correlation between the headache occurrence in the patients' group and the trigger factors existed, while the alternative hypothesis was that there was no correlation. Pearson correlation coefficients were calculated to determine if a correlation existed between the frequency of headaches and the trigger factors we had taken into account in our study, in both the migraine and the control group.

The level of statistical significance was set at a p-value < 0.05.

Results

According to the MIDAS, we categorised patients due to their migraine severity as Grade I, II, III or IV, which amounted respectively to 10%, 10%, 22.5% and 57.5% of the patients. 34 patients (85%) were diagnosed as patients with migraine without aura, and only two patients (5%) had attacks of migraine with aura. Four patients (10%) had chronic migraine. The demographic and clinical data of patients is set out in Table 2 and Table 3 respectively.

There were non-uniform sample sizes in terms of educational level, type of work, number of children, aura and no-aura migraine, marital status and the MIDAS test groups, which made it impossible to obtain reliable data.

Some of the patients from both groups were post-menopausal and so could not associate menstruation with headaches. For this reason, we excluded such patients from the study and the control group, and for this specific factor the number of females in the compared groups has changed.

The results of the tests revealed a weak connection between the occurrence of headaches and trigger factors such as alcohol

Table 1. Patients' questionnaire

Please answer the following questions:		
First name:	……..	
Last name:	……..	
Age:	……..	
Education:	Primary / Secondary / Higher	
Type of work:	Physical / Mental	
Marital status:	……..	
Number of children:	……..	
Do you associate the occurrence of headaches with the following situations? Please circle the correct answer (YES or NO).		
After a stressful situation?	YES	NO
After relaxation, holidays or other forms of rest?	YES	NO
After eating products like chocolate, dairy, cocoa, seasonings or citruses? If yes, please state which products exactly ……	YES	NO
After alcohol consumption?	YES	NO
After fasting?	YES	NO
After sleep deficiency?	YES	NO
After excessive sleeping?	YES	NO
After workout/fatigue?	YES	NO
After exposure to bright or pulsing light?	YES	NO
After exposure to noise?	YES	NO
After staying at a high altitude (e.g. in the mountains)?	YES	NO
After sudden weather changes?	YES	NO
After medicine intake? If yes, please state which medicines exactly …….	YES	NO
After menstruation?	YES	NO
Do you find any other situations that trigger headaches? If yes, please describe them. ……..	YES	NO
Please answer the following questions:		
On how many days in the last 3 months did you miss work or school because of your headaches?	……..	
How many days in the last 3 months was your productivity at work or school reduced by half or more because of your headaches?	……..	
On how many days in the last 3 months did you not do household work because of your headaches?	……..	
How many days in the last 3 months was your productivity in household work reduced by half of more because of your headaches?	……..	
On how many days in the last 3 months did you miss family, social or leisure activities because of your headaches?	……..	
On how many days in the last 3 months did you have a headache?	……..	
On a scale of 0–10, on average how painful were these headaches? (where 0 = no pain at all, and 10 = pain as bad as it can be)	……..	

intake, sleep deficiency, excessive sleeping, staying at high altitudes, or medicine intake.

On the other hand, in the following situations: stress exposure, relaxation after stress, intake of dietary products, fasting, workout/fatigue, exposure to bright or pulsing light, high noise level, sudden weather changes and menstruation, the results of the chi-square tests showed statistically significant differences, and the null hypothesis cannot be rejected.

The results are set out in Table 4.

Discussion

Our study shows that we can identify headache-triggering factors in migraineurs which have been many times reported as acknowledged migraine trigger factors in previous research studies [9–13, 17]. Kelman retrospectively evaluated 1,750 patients and showed that acute migraine attacks were triggered in around 75% of patients, with different frequencies, and no triggers were seen in around 25%. The most common triggers included stress (79.7%), hormones in women (65.1%), fasting (57.3%), weather (53.2%), sleep disturbance (49.8%), perfume or odour (43.7%), neck pain (38.4%), light(s) (38.1%), alcohol (37.8%), smoking (35.7%), sleeping late (32.0%), heat (30.3%), food (26.9%), and exercise (22.1%) [17].

Stress (82.5%) was the most frequent migraine headache-triggering factor in our study, which corresponds to the results of other researchers [9, 10, 12, 17, 18]. Stress is said to trigger headaches in 50-80% of migraineurs [9, 10, 17]. Moreover,

Table 2. Demographic data

		Migraine group (n = 40)	TTH group (n = 40)
Age range		21–66	23–76
Age (years)	Mean	44.62	39.5
	SD[1]	10.64	15.59
Gender	Female	n = 40	n = 40
	Male	n = 0	n = 0
Type of work	Physical	5	0
	Mental	35	40
Education	Primary	0	0
	Secondary	13	15
	Higher	27	25
Marital status	Single	12	21
	Married	25	14
	Divorced	1	2
	Widowed	2	3
Number of children	0	12	22
	1	9	10
	2	17	7
	3+	2	1

[1]standard deviation

chronic migraine can increase perceived stress and further lower patients' quality of life [12]. Comparing this trigger in other populations, the frequency of stress as a trigger was similar in Spanish and Pakistani populations (46.3% and 44%), and slightly higher in Brazilian migraineurs (73%) [19]. Similar results have been noted in Caucasians (87% recognised stress as a trigger) and African-Americans (84%) in a questionnaire study focused on identifying ethnic disparities, involving 131 migraine patients [20].

Sudden weather changes (75%) — our next most frequent trigger, consist of many variables, such as barometric pressure

Table 3. Clinical data

		Number of migraine patients (n = 40)	
		n	%
MIDAS grade	I	4	10
	II	4	10
	III	9	22.5
	IV	22	57.5
Migraine without aura		34	85
Migraine with aura		2	5
Chronic migraine		4	10

Table 4. Patients with headaches triggered by the researched factors

	Migraine group n = 40		Control group n = 40		P value[2]	Type of chi-square test
	n	%	n	%		Pearson Chi-square
Stress	33	82.5	15	37.5	**0.00004**	
Relaxation after stress	17	42.5	5	12.5	**0.003**	
Alcohol intake	20	50.0	16	40.0	0.369	
Fasting	26	65.0	15	37.5	**0.014**	
Sleep deficiency	25	62.5	25	62.5	1.000	
Excessive sleeping	24	60.0	17	42.5	0.117	
Workout/fatigue	21	52.5	6	15.0	**0.0004**	
Exposure to bright or pulsing light	25	62.5	6	15.0	**0.00001**	
High noise level	23	57.5	12	30.0	**0.013**	
Sudden weather changes	30	75.0	20	50.0	**0.021**	
Medicine intake	3	7.5	4	10.0	1.000	
Menstruation[1]	26 (n = 39)	66.67	5 (n = 26)	19.2	**0.0002**	
Dietary products intake	15	37.5	3	7.5	**0.001**	**V-square**
Staying at high altitudes	11	27.5	5	12.5	0.096	
Medicine intake	3	7.5	4	10.0	1.000	**Yates Chi-square**

[1]numbers in brackets state the total number of patients in groups due to excluding females after the menopause
[2]p-values in bold are statistically significant < 0.05

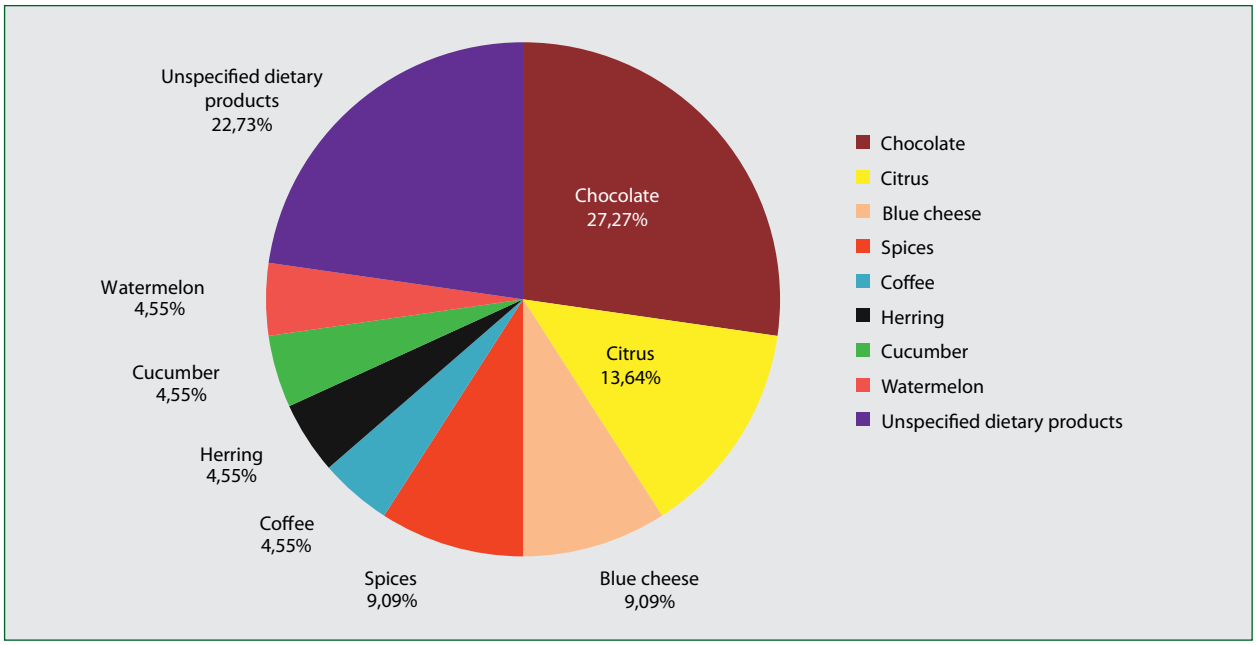

Figure 1. Percentage frequency distribution of dietary products associated with headaches in migraine patients

fluctuations, humidity, temperature, or weather pattern. Prince et al., in a study of 77 patients with migraine, found 39 (50.6%) to be sensitive to weather changes [21]. Regional weather conditions, like the presence of Chinook winds in Canada, may affect migraine occurrence. Prince et al. wrote: "For the patient group (75) as a whole, headache probability was significantly increased on pre-chinook days, and also on chinook days with high-velocity wind (top quartile of chinook days)" [22]. Other researchers have observed that different air mass types in central North Carolina have an influence on migraine attacks as well: "We found statistically significant differences in ED migraine visits between air mass types. Tropical (i.e. warmer) air masses generally resulted in the highest frequency of ED visits, while polar (i.e. colder) air masses resulted in the lowest frequency" [23].

Fasting (65%) is among the best studied and the most reliable natural migraine triggers. Out of all the dietary triggers of migraine attack, fasting is very frequently reported, with percentages ranging from 39% to 66% depending on the study [24]. Our questionnaire did not define the duration of fasting nor did it include questions about how much food a patient ate before or during fasting or what kind of food it was. Objective research studies tend to concentrate on specifically defined periods of fasting such as that encouraged by religions. In one study focused on the month of Ramadan, patients identified 9.4 ± 4.3 migraine days on average (range 3–20) as compared to 3.7 ± 2.1 migraine days on average (range 1-10) during the control month ($p < 0.001$) [25]. Researchers investigating the phenomenon of Yom Kippur headache reported that in a population including 211 who fasted, 39% of the fasters developed headaches, compared to only 7% of non-fasters ($p < 0.000001$). Also, the number of headache sufferers increased in direct correlation to the duration of the fast [26].

Chocolate was the most common dietary product associated with headaches. Food triggers frequency is similar to many others studies [27, 28]. Tai et al. in their study involving 319 (46.6%) Malaysian migraine patients, pointed out that the most common dietary trigger factors were coffee (136 patients, 19.9%), followed by chocolate (51 patients, 7.5%) [29]. The comparison of the migraine and the TTH group in Tai's study showed a statistical significance of their findings — chocolate (OR 2.16, 95% CI 1.06–4.41, $p = 0.035$) and coffee (OR 1.73, 95% CI 1.12–2.68, $p = 0.014$). Peatfield et al. found that 19.2% of 490 migraine patients reported sensitivity to chocolate, 18.2% to cheese, and 11.1% to citrus fruit [30]. The percentage frequency distribution of different dietary products associated with migraine headaches of our patients is presented in Figure 1.

Our study has some limitations. The study group was relatively small, and all the patients came from a single headache clinic. This study was conducted according to patients' perceived headache triggering factors and observations. The questionnaire did not implement objective scales for said trigger factors. We did not measure noise level, altitudes, or weather parameters such as air pressure or temperature. Furthermore, some patients were on preventive medication, which could have affected the analysed data. All these factors could have an impact on the study's results.

Conclusion

Although our study presented statistical significance and stated specific trigger factors, which are consistent with studies

from other countries such as Canada, USA, Turkey, Spain and Korea, we cannot generalise the results to the whole Polish population, due to the study group being too small [10, 13, 17, 29, 31]. In our opinion, this study could prove useful for clinical practice in non-pharmacological preventive treatment in migraine patients, although it should be verified by other studies evaluating the Polish population. Future studies should include larger study groups that would allow the gathering of cross-sectional population data, possibly including additional factors.

Conflict of interest: *All authors have no conflict of interest to disclose.*

Abbreviations:
MIDAS — Migraine Disability Assessment Test
TTH — Tension type headache
ICHD-3 — The International Classification of Headache Disorders 3rd edition
CDH — Chronic daily headache
BMI — Body mass index

References

1. Karbowniczek A, Domitrz I. Frequency and clinical characteristics of chronic daily headache in an outpatient clinic setting. Neurol Neurochir Pol. 2011; 45(1): 11–17, indexed in Pubmed: 21384288.
2. Younger DS. Epidemiology of Migraine. Neurol Clin. 2016; 34(4): 849–861, doi: 10.1016/j.ncl.2016.06.011, indexed in Pubmed: 27719997.
3. Vetvik K, MacGregor E. Sex differences in the epidemiology, clinical features, and pathophysiology of migraine. The Lancet Neurology. 2017; 16(1): 76–87, doi: 10.1016/s1474-4422(16)30293-9.
4. Stępień A, Prusiński A. Wybrane dane epidemiologiczne wystepowania migreny w Polsce, Ból : kwartalnik Polskiego Towarzystwa Badania Bólu. 2003, 1640-324X. T. 4, nr 3 s. : 9–11.
5. Chu MK, Kim DW, Kim BK, et al. Gender-specific influence of socioeconomic status on the prevalence of migraine and tension-type headache: the results from the Korean Headache Survey. J Headache Pain. 2013; 14: 82, doi: 10.1186/1129-2377-14-82, indexed in Pubmed: 24093215.
6. Stewart WF, Roy J, Lipton RB. Migraine prevalence, socioeconomic status, and social causation. Neurology. 2013; 81(11): 948–955, doi: 10.1212/WNL.0b013e3182a43b32, indexed in Pubmed: 23990405.
7. Kim SY, Park SP. The role of headache chronicity among predictors contributing to quality of life in patients with migraine: a hospital-based study. J Headache Pain. 2014; 15: 68, doi: 10.1186/1129-2377-15-68, indexed in Pubmed: 25278151.
8. Bera SC, Khandelwal SK, Sood M, et al. A comparative study of psychiatric comorbidity, quality of life and disability in patients with migraine and tension type headache. Neurol India. 2014; 62(5): 516–520, doi: 10.4103/0028-3886.144445, indexed in Pubmed: 25387621.
9. Peroutka SJ. What turns on a migraine? A systematic review of migraine precipitating factors. Curr Pain Headache Rep. 2014; 18(10): 454, doi: 10.1007/s11916-014-0454-z, indexed in Pubmed: 25160711.
10. Mollaoğlu M. Trigger factors in migraine patients. J Health Psychol. 2013; 18(7): 984–994, doi: 10.1177/1359105312446773, indexed in Pubmed: 23104993.
11. Fernández-de-Las-Peñas C, Fernández-Muñoz JJ, Palacios-Ceña M, et al. Sleep disturbances in tension-type headache and migraine. Ther Adv Neurol Disord. 2018; 11: 1756285617745444, doi: 10.1177/1756285617745444, indexed in Pubmed: 29399051.
12. Moon HJ, Seo JG, Park SP. Perceived stress in patients with migraine: a case-control study. J Headache Pain. 2017; 18(1): 73, doi: 10.1186/s10194-017-0780-8, indexed in Pubmed: 28733942.
13. Zaeem Z, Zhou L, Dilli E. Headaches: a Review of the Role of Dietary Factors. Curr Neurol Neurosci Rep. 2016; 16(11): 101, doi: 10.1007/s11910-016-0702-1, indexed in Pubmed: 27714637.
14. Headache Classification Committee of the International Headache Society (IHS) The International Classification of Headache Disorders, 3rd edition. Cephalalgia. 2018; 38(1): 1–211, doi: 10.1177/0333102417738202, indexed in Pubmed: 29368949.
15. Chorążka K, Janoska M, Swić P, Domitrz I, Body mass index and serum lipid levels in effect on the incidence and course of migraine, Neurol Neurochir Pol. 2013; 47(6): 572–6.
16. Stewart WF, Lipton RB, Kolodner K, et al. Reliability of the migraine disability assessment score in a population-based sample of headache sufferers. Cephalalgia. 1999; 19(2): 107–14; discussion 74, doi: 10.1046/j.1468-2982.1999.019002107.x, indexed in Pubmed: 10214536.
17. Kelman L. The triggers or precipitants of the acute migraine attack. Cephalalgia. 2007; 27(5): 394–402, doi: 10.1111/j.1468-2982.2007.01303.x, indexed in Pubmed: 17403039.
18. Yalinay Dikmen P, Yavuz BG, Aydinlar EI. The relationships between migraine, depression, anxiety, stress, and sleep disturbances. Acta Neurol Belg. 2015; 115(2): 117–122, doi: 10.1007/s13760-014-0312-0, indexed in Pubmed: 24889393.
19. Carod-Artal FJ, Ezpeleta D, Martín-Barriga ML, et al. Triggers, symptoms, and treatment in two populations of migraneurs in Brazil and Spain. A cross-cultural study. J Neurol Sci. 2011; 304(1-2): 25–28, doi: 10.1016/j.jns.2011.02.027, indexed in Pubmed: 21402387.
20. Nicholson RA, Rooney M, Vo K, et al. Migraine care among different ethnicities: do disparities exist? Headache. 2006; 46(5): 754–765, doi: 10.1111/j.1526-4610.2006.00453.x, indexed in Pubmed: 16643578.
21. Prince PB, Rapoport AM, Sheftell FD, et al. The effect of weather on headache. Headache. 2004; 44(6): 596–602, doi: 10.1111/j.1526-4610.2004.446008.x, indexed in Pubmed: 15186304.
22. Cooke LJ, Rose MS, Becker WJ. Chinook winds and migraine headache. Neurology. 2000; 54(2): 302–307, doi: 10.1212/wnl.54.2.302, indexed in Pubmed: 10668687.
23. Elcik C, Fuhrmann CM, Mercer AE, et al. Relationship between air mass type and emergency department visits for migraine headache across the Triangle region of North Carolina. Int J Biometeorol. 2017; 61(12): 2245–2254, doi: 10.1007/s00484-017-1432-z, indexed in Pubmed: 28900742.
24. AA S, CAC d, RP SN. Headaches and Food Abstinence: A Review. Journal of Clinical Case Studies. 2018; 3(2), doi: 10.16966/2471-4925.163.
25. Abu-Salameh I, Plakht Y, Ifergane G. Migraine exacerbation during Ramadan fasting. J Headache Pain. 2010; 11(6): 513–517, doi: 10.1007/s10194-010-0242-z, indexed in Pubmed: 20652352.
26. Mosek A, Korczyn AD. Yom Kippur headache. Neurology. 1995; 45(11): 1953–1955, doi: 10.1212/wnl.45.11.1953, indexed in Pubmed: 7501139.
27. Fukui PT, Gonçalves TR, Strabelli CG, et al. Trigger factors in migraine patients. Arq Neuropsiquiatr. 2008; 66(3A): 494–499,

doi: 10.1590/s0004-282x2008000400011, indexed in Pubmed: 18813707.

28. Wöber C, Wöber-Bingöl C. Triggers of migraine and tension-type headache. Handb Clin Neurol. 2010; 97: 161–172, doi: 10.1016/S0072-9752(10)97012-7, indexed in Pubmed: 20816418.
29. Tai MLS, Yap JF, Goh CB. Dietary trigger factors of migraine and tension-type headache in a South East Asian country. J Pain Res. 2018; 11: 1255–1261, doi: 10.2147/JPR.S158151, indexed in Pubmed: 29988763.
30. Peatfield RC, Glover V, Littlewood JT, et al. The prevalence of diet-induced migraine. Cephalalgia. 1984; 4(3): 179–183, doi: 10.1046/j.1468-2982.1984.0403179.x, indexed in Pubmed: 6498931.
31. Park JW, Chu MK, Kim JM, et al. Analysis of Trigger Factors in Episodic Migraineurs Using a Smartphone Headache Diary Applications. PLoS One. 2016; 11(2): e0149577, doi: 10.1371/journal.pone.0149577, indexed in Pubmed: 26901341.

SHORT COMMUNICATION

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Direct oral anticoagulants in the treatment of cerebral venous sinus thrombosis: a single institution's experience

DOACs in treatment of cerebral venous sinus thrombosis: case series

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ABSTRACT

Aim of the study. Oral anticoagulants, preferentially vitamin K antagonists (VKA), are recommended for 3–12 months in patients with cerebral venous sinus thrombosis (CVST). We present a series of patients with CVST treated with direct oral anticoagulants (DOAC).

Materials and methods. We prospectively recruited 36 patients with CVST (aged 40.3 ± 9.2 years, 58.3% female) treated with DOAC based on the physician's or patient's preferences. Functional outcome was assessed with modified Rankin Scale. Recanalisation was assessed on imaging at 3–6 months post the event. Patients were followed for a median of 30 [interquartile range (IQR) 25–37] months.

Results. After use of heparin (median: 6 days; IQR 5–8.75), patients received dabigatran (150 mg bid, n = 16 or 110 mg bid, n = 2), rivaroxaban (20 mg qd, n = 10) or apixaban (5 mg bid, n = 8) for a median of 8.5 months (IQR 6.25–12). Complete or partial recanalisation was observed in 34 cases (94.4%). Three patients (8.3%) experienced major bleeding: menorrhagia on rivaroxaban (n = 2) and gastrointestinal bleeding on dabigatran (n = 1). A favourable functional outcome was observed in 24 (66.7%) patients, without any fatality. CSVT recurred in two patients (5.6%) and two venous thromboses developed in two other patients with inherited thrombophilia after anticoagulation withdrawal.

Conclusions and clinical implications. DOACs could be an alternative to VKA in CVST patients.

Key words: anticoagulation, cerebral venous sinus thrombosis, direct oral anticoagulants, bleeding, venous thromboembolism

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Introduction

Cerebral venous sinus thrombosis (CVST) is rare, with an annual incidence estimated at 3 to 4 cases per million [1] and with a female to male ratio of 3:1 [2]. CVST is associated with genetic or acquired thrombophilias, malignancy, infection or head trauma, pregnancy, and oral contraceptives [3]. Prothrombotic risk factors are identifiable among 85% of CVST patients [3].

European Stroke Organisation guidelines recommend using low-molecular-weight heparin (LMWH) in the treatment of the acute phase [4], followed by vitamin K antagonists (VKAs) for 3–12 months. The guidelines of the American Heart Association/American Stroke Association are similar [5].

Direct oral anticoagulants (DOACs) are at least as effective as VKAs in the treatment and prevention of the recurrence of venous thromboembolism (VTE) [6]. DOACs might be a promising alternative to VKAs in the long-term treatment of

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CVST, especially among patients for whom VKAs are contraindicated or unsuitable [7].There have been few observational studies on DOAC in CVST. Mendonça et al. [8] reported full recanalisation in 27% of 15 dabigatran-treated CVST patients (mostly 150 mg bid) and excellent long-term functional outcomes in 87% of patients, without any CVST recurrence or major bleeding during follow-ups for a median of 19 months. Geisbüsch et al. [9] reported on seven patients with CVST on rivaroxaban 20 mg qd. During follow-up (median: 7 months), no thrombotic events and two minor bleeds occurred. Complete recanalisation was observed in four patients.

Several experts have advised using DOAC in this disease [6]. To the best of our knowledge, there have been no reports on Polish patients with CVST treated with DOACs. This single-centre case series study was aimed to assess the efficacy and safety of DOACs in CVST.

Materials and methods

We enrolled 36 consecutive patients with a documented episode of CVST referred for further diagnostic work-up to an outpatient clinic at John Paul II Hospital in Krakow, Poland, between December 2013 and March 2018. On the first visit, all eligible subjects were on dabigatran, rivaroxaban or apixaban and provided medical records including imaging data. They were enrolled 3–6 months since a diagnosis of CVST. The exclusion criteria were as follows: indications for long-term anticoagulant therapy other than VTE, diagnosed malignancy, pregnancy, breastfeeding, and advanced kidney disease (stage 4–5). Written consent was obtained to participate in this observational study.

Clinical diagnosis of CVST was made according to the international criteria [5] by brain imaging — computed tomography (CT) with CT venography. Following the use of LMWH at therapeutic doses or unfractionated heparin, the DOAC therapy was initiated. To assess the vessel recanalisation status, CT angiography was performed 3–6 months after CVST.

We collected demographic characteristics, clinical data on CVST, VTE risk factors, comorbidities and current treatment using a standardised questionnaire. Unprovoked CVST was established if there was no history of malignancy, major surgery, trauma, immobilisation, pregnancy or childbirth at least three months before CVST diagnosis and no use of oestrogen. Family history of VTE was defined as VTE in first-degree relatives. Obesity was recognised when Body Mass Index was ≥ 30 kg/m^2.

Follow-up included the time of DOAC use and the time since its withdrawal. Clinical data was collected every six months via a visit to the outpatient clinic or telephone contact. The decision to cease DOAC use was left to the discretion of the attending physician based on the patient's preferences. Thrombophilia screening was performed (antiphospholipid syndrome [APS], Factor V Leiden [FVL] or prothrombin G20210A mutations and deficiencies in protein C, protein S or antithrombin, as described previously [10]). Functional outcome was assessed using modified Rankin Scale (mRS) before initiation of the DOAC therapy and after 6–12 months of follow-up. A favourable functional outcome was defined as 0-1 point in mRS. The occurrence of VTE (including CVST) and major bleeding (according to the definition by Schulman et al. [11]) were recorded.

The local ethical committee issued approval for the study according to the Declaration of Helsinki.

Statistical analysis

Categorical variables were reported as numbers and percentages. Continuous variables were presented as means (standard deviation) or median (IQR, interquartile range). Normality was assessed by the Shapiro-Wilk test. The chi-squared test was used to compare categorical variables. The ANOVA or Kruskal-Wallis tests for continuous variables were conducted to assess differences between the groups using different DOACs. Analyses were performed using SPSS Software (IBM, USA). A P-value below 0.05 was considered statistically significant.

Results

A total of 36 patients with CVST (aged 40.3 ± 9.2 years, 58.3% female) were analysed (Table 1). Unprovoked CVST was found in 26 (72.2%). Thrombophilia was observed in 18 (50%) patients, including eight patients (22.2%) with FVL.

The most common single location of CVST was the transverse sinus. Following the use of heparin (median, 6 days; IQR 5–8.75), DOAC was initiated and continued for a median of 8.5 months (IQR 6.25–12). There were 18 patients (50%) on dabigatran (n = 16, 150 mg bid and n = 2, 110 mg bid). Ten subjects (27.8%) received rivaroxaban (20 mg daily) and eight patients (22.2%) were on apixaban (5 mg bid). Apart from older age in the dabigatran users (44.3 ± 8.3 years *vs.* rivaroxaban 35.7 ± 9.1 and apixaban 37.1 ± 8.4 years, p = 0.029), the patients on the three DOACs were similar with regard to demographic and clinical characteristics (data not shown).

A repeat brain imaging after 3–6 months showed at least partial vessel recanalisation in 34 patients (94.4%). Complete recanalisation was observed in 20 individuals (55.6%), comprising 10 on dabigatran (55.6%), six on rivaroxaban (60.0%) and four on apixaban (50.0%).

On anticoagulant therapy, three patients (8.3%), two on rivaroxaban (20 mg qd) and one on dabigatran (110 mg bid), experienced major bleeding, including two heavy menstrual bleedings (HMB) in a 25-year-old woman and a 46-year-old woman both with previously abundant menses (haemoglobin, 8 g/dL and 9 g/dL, respectively) and an upper gastrointestinal bleeding in a 56-year-old woman who had reported dysphagia in previous weeks despite pantoprazole use.

Table 1. Characteristics of the study participants

VARIABLE	TOTAL (n = 36)
Age, years	40.3 ± 9.2
Sex female, n (%)	21 (58.3%)
BMI [kg/m^2]	26.8 ± 4.2
Cigarette smoking, n (%)	8 (22.2%)
OC/HRT, n/females (%)	10/21 (47.6%)
Family history of VTE, n (%)	8 (22.2%)
Unprovoked CVST, n (%)	26 (72.2%)
Obesity, n (%)	10 (27.8%)
Site of CVST	
Transverse sinus, n (%)	5 (13.9%)
Cavernous sinus, n (%)	3 (8.3%)
Straight sinus, n (%)	1 (2.8%)
Combined, n (%)	26 (72.2%)
Other locations, n (%)	1 (2.8%)
Concomitant VTE, n (%)	4 (11.1%)
Heparin regimen	
LMWH, n (%)	33 (91.7%)
UFH, n (%)	3 (8.3%)
Duration of heparin use [days]	6 (5–8.75)
Aspirin use, n (%)	8 (22.2%)
Type of DOAC	
Dabigatran, n (%)	18 (50.0%)
Rivaroxaban, n (%)	10 (27.8%)
Apixaban, n (%)	8 (22.2%)
Full dose of DOAC, n (%)	34 (94.4%)
Duration of DOAC use [months]	8.5 (6.25–12)
mRS score at the start of DOAC	
Favourable outcome (mRS 0–1), n (%)	19 (52.8%)
Independent (mRS 2), n (%)	13 (36.1%)
Significant disability (mRS 3–5), n (%)	4 (11.1%)
Follow-up [months]	30 (25–37)
Recanalisation 3-6 months after CVST	
Complete, n (%)	20 (55.6%)
Partial, n (%)	14 (38.9%)
No recanalisation, n (%)	2 (5.6%)
mRS score 6-12 months after CVST	
Favourable outcome (mRS 0–1), n (%)	24 (66.7%)
Independent (mRS 2), n (%)	10 (27.8%)
Significant disability (mRS 3–5) , n (%)	2 (5.6%)
Recurrent CSVT, n (%)	2 (5.6%)
New DVT, n (%)	2 (5.6%)
Major bleeding, n (%)	3 (8.3%)
Thrombophilia testing	
Factor V Leiden, n (%)	8 (22.2%)
Prothrombin G20210A mutation, n (%)	3 (8.3%)
Protein C deficiency, n (%)	1 (2.8%)
Protein S deficiency, n (%)	2 (5.6%)
Antithrombin deficiency, n (%)	1 (2.8%)
Antiphospholipid syndrome, n (%)	4 (11.1%)

Data reported as median (interquartile range), mean ± standard deviation or number (percentage)
BMI — Body Mass Index; CVST — cerebral venous sinus thrombosis; DVT — deep vein thrombosis; LMWH — low-molecular-weight heparin; mRS — modified Rankin Scale; DOAC — direct oral anticoagulant; OC/HRT — oral contraceptives/hormonal replacement therapy; UFH — unfractionated heparin; VTE — venous thromboembolism

After withdrawal of anticoagulant therapy, we followed patients for a median of 30 months (IQR 25–37). Neurological evaluation at 6–12 months showed that 66.7% of CVST patients (n = 24) had a favourable functional outcome. During follow-up, two patients (5.6%) had recurrent CVST. One episode occurred in a 37-year-old man, free of thrombophilia but with a positive VTE family history, five months after rivaroxaban withdrawal. The other was diagnosed in a 52-year-old woman, diagnosed with single-positive APS, following hospitalisation for pneumonia, 20 months after the first CSVT, while on aspirin. There were also two episodes of deep-vein thrombosis (DVT). The first DVT event, provoked by a leg injury and oral contraception, occurred in a 29-year-old woman heterozygous for prothrombin G20210A mutation with previous VTE, 18 months after apixaban therapy was ended. The other unprovoked DVT was observed in a 46-year-old woman heterozygous for FVL.

Discussion

To the best of our knowledge, this is the first Polish case study and the largest worldwide report presenting treatment outcomes among DOAC-treated patients with CVST.

We noted complete cerebral vessel recanalisation in more than 55% of rivaroxaban users, which is similar to the previous study [9], in which such an outcome was observed in 57.1% of cases. Among dabigatran-treated patients, the complete recanalisation rate was 60% (n = 6), which is much higher than in a Portuguese study [8] where full recanalisation was noted in 26.7% of patients. The favourable functional outcomes during follow-up were similar to previous studies [8–9] while a favourable functional outcome was observed in 58–89% of CVST patients on warfarin [12–13].

Since among VKA-treated patients with CVST, the major bleeding rate is estimated at 0.21%/patient-month [14], the present rate with 8.3% of patients (n = 3) with major bleeding over a median of 8.5 months of DOAC therapy appears to be higher. However, there were no life-threatening episodes and the 50% lower risk of intracranial bleeds on DOAC is of vital importance. We confirmed an increased HMB risk on this drug [15–16], which supports the suggestion that in women of reproductive age and previously abundant menses, rivaroxaban should be avoided. Previous small studies [8–9] did not report any major haemorrhages, which disagrees with randomised VTE studies and registries [17–18].

Two recurrent episodes of CVST while off anticoagulation are consistent with the rates reported in previous studies, i.e. 2–4.4% over a median 16–40 months of follow-up (0.5–1.5 per 100 person-years) [3, 12]. Regarding non-cerebral VTE, the current two episodes among 36 CVST patients correspond to the rate for VKA-treated CVST patients (6.5% over a median follow-up of 40 months) [12].

Clinical implications/future directions

While we await the results of a randomised trial with dabigatran in CVST patients [19], based on growing evidence from observational studies, DOACs do appear to be an attractive alternative to VKA due to similar efficacy and safety without even taking into account advantages including no need for laboratory monitoring, no dietary interactions, and no interference with most medications [20]. Randomised trials with all DOACs should be performed to prove the benefits available from DOACs in this disease.

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References

1. Stam J. Thrombosis of the cerebral veins and sinuses. N Engl J Med. 2005; 352(17): 1791–1798, doi: 10.1056/NEJMra042354, indexed in Pubmed: 15858188.
2. Coutinho JM, Ferro JM, Canhão P, et al. Cerebral venous and sinus thrombosis in women. Stroke. 2009; 40(7): 2356–2361, doi: 10.1161/STROKEAHA.108.543884, indexed in Pubmed: 19478226.
3. Ferro JM, Canhão P, Stam J, et al. ISCVT Investigators. Prognosis of cerebral vein and dural sinus thrombosis: results of the International Study on Cerebral Vein and Dural Sinus Thrombosis (ISCVT). Stroke. 2004; 35(3): 664–670, doi: 10.1161/01.STR.0000117571.76197.26, indexed in Pubmed: 14976332.
4. Ferro JM, Bousser MG, Canhão P, et al. European Stroke Organization. European Stroke Organization guideline for the diagnosis and treatment of cerebral venous thrombosis - endorsed by the European Academy of Neurology. Eur J Neurol. 2017; 24(10): 1203–1213, doi: 10.1111/ene.13381, indexed in Pubmed: 28833980.
5. Saposnik G, Barinagarrementeria F, Brown RD, et al. American Heart Association Stroke Council and the Council on Epidemiology and Prevention. Diagnosis and management of cerebral venous thrombosis: a statement for healthcare professionals from the American Heart Association/American Stroke Association. Stroke. 2011; 42(4): 1158–1192, doi: 10.1161/STR.0b013e31820a8364, indexed in Pubmed: 21293023.
6. Mimier MK, Janczak DT, McBane RD, et al. Thrombosis of atypical location: how to treat patients in the era of direct oral anticoagulants? Pol Arch Intern Med. 2018; 128(10): 604–608, doi: 10.20452/pamw.4333, indexed in Pubmed: 30233080.
7. Behrouzi R, Punter M. Diagnosis and management of cerebral venous thrombosis. Clin Med (Lond). 2018; 18(1): 75–79, doi: 10.7861/clinmedicine.18-1-75, indexed in Pubmed: 29436443.
8. Mendonça MD, Barbosa R, Cruz-e-Silva V, et al. Oral direct thrombin inhibitor as an alternative in the management of cerebral venous thrombosis: a series of 15 patients. Int J Stroke. 2015; 10(7): 1115–1118, doi: 10.1111/ijs.12462, indexed in Pubmed: 25708372.
9. Geisbüsch C, Richter D, Herweh C, et al. Novel factor Xa inhibitor for the treatment of cerebral venous and sinus thrombosis: first experience in 7 patients. Stroke. 2014; 45(8): 2469–2471, doi: 10.1161/STROKEAHA.114.006167, indexed in Pubmed: 25070963.
10. Siudut J, Świat M, Undas A. Altered Fibrin Clot Properties in Patients With Cerebral Venous Sinus Thrombosis: Association With the Risk of Recurrence. Stroke. 2015; 46(9): 2665–2668, doi: 10.1161/STROKEAHA.115.009528, indexed in Pubmed: 26173730.
11. Schulman S, Kearon C. Subcommittee on Control of Anticoagulation of the Scientific and Standardization Committee of the International Society on Thrombosis and Haemostasis. Definition of major bleeding in clinical investigations of antihemostatic medicinal products in non-surgical patients. J Thromb Haemost. 2005; 3(4): 692–694, doi: 10.1111/j.1538-7836.2005.01204.x, indexed in Pubmed: 15842354.
12. Dentali F, Poli D, Scoditti U, et al. CErebral VEin Thrombosis International Study Investigators. Long-term outcomes of patients with cerebral vein thrombosis: a multicenter study. J Thromb Haemost. 2012; 10(7): 1297–1302, doi: 10.1111/j.1538-7836.2012.04774.x, indexed in Pubmed: 22578023.
13. Herweh C, Griebe M, Geisbüsch C, et al. Frequency and temporal profile of recanalization after cerebral vein and sinus thrombosis. Eur J Neurol. 2016; 23(4): 681–687, doi: 10.1111/ene.12901, indexed in Pubmed: 26667584.
14. Cundiff DK. Anticoagulants for cerebral venous thrombosis: harmful to patients? Stroke. 2014; 45(1): 298–304, doi: 10.1161/STROKEAHA.113.003519, indexed in Pubmed: 24232450.
15. Bryk AH, Piróg M, Plens K, et al. Heavy menstrual bleeding in women treated with rivaroxaban and vitamin K antagonists and the risk of recurrent venous thromboembolism. Vascul Pharmacol. 2016; 87: 242–247, doi: 10.1016/j.vph.2016.11.003, indexed in Pubmed: 27865826.
16. Beyer-Westendorf J, Michalski F, Tittl L, et al. Management and outcomes of vaginal bleeding and heavy menstrual bleeding in women of reproductive age on direct oral anti-factor Xa inhibitor therapy: a case series. Lancet Haematol. 2016; 3(10): e480–e488, doi: 10.1016/S2352-3026(16)30111-9, indexed in Pubmed: 27692306.
17. Agnelli G, Buller HR, Cohen A, et al. AMPLIFY Investigators. Oral apixaban for the treatment of acute venous thromboembolism. N Engl J Med. 2013; 369(9): 799–808, doi: 10.1056/NEJMoa1302507, indexed in Pubmed: 23808982.
18. Schulman S, Kakkar A, Goldhaber S, et al. Treatment of Acute Venous Thromboembolism With Dabigatran or Warfarin and Pooled Analysis. Circulation. 2014; 129(7): 764–772, doi: 10.1161/circulationaha.113.004450.
19. Ferro JM, Dentali F, Coutinho JM, et al. Rationale, design, and protocol of a randomized controlled trial of the safety and efficacy of dabigatran etexilate versus dose-adjusted warfarin in patients with cerebral venous thrombosis. Int J Stroke. 2018; 13(7): 766–770, doi: 10.1177/1747493018778125, indexed in Pubmed: 29775170.
20. Tripodi A, Braham S, Scimeca B, et al. How and when to measure anticoagulant effects of direct oral anticoagulants? Practical issues. Pol Arch Intern Med. 2018; 128(6): 379–385, doi: 10.20452/pamw.4287, indexed in Pubmed: 29968697.

LETTER TO THE EDITOR

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Is vitamin D deficiency a reliable risk factor for multiple sclerosis development?

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Key words: vitamin 25(0H)D, multiple sclerosis

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We greatly appreciate, and are impressed by, the article written by Halina Bartosik-Psujek and Marek Psujek who reviewed the state of current knowledge regarding vitamin D [25(0H)D] as an immune modulator in multiple sclerosis (MS), especially regarding 25(0H)D's effect on immune cells subsets in relation to experimental and clinical studies [1].

25(0H)D maintains calcium-phosphate homeostasis and modulates an immune response. 25(0H)D inhibits the maturation of antigen-presenting dendritic cells (DCs), the proliferation of T and B cells, the Th1 and Th17 response, the expression of MHC class II, CD40, CD80, CD86, and the production of IgG, IgM and pro-inflammatory IL-1, IL-6, IL-12, TNFα. 25(0H)D increases the differentiation of T regulatory cells (Treg), the proliferation of macrophages, and the Th2 response and production of anti-inflammatory IL-10 [1]. The main route for obtaining 25(0H)D is sunlight exposure, because dietary intake accounts only for 30% of the total amount of 25(0H)D [2].

MS is a chronic inflammatory and neurodegenerative disorder of the central nervous system (CNS) leading to demyelination, axonal loss and damage of oligodendrocytes. 25(0H)D is one of the environmental factors which seem to play an important role in the aetiopathogenesis of MS and influences the course of the disease.

However, the impact of 25(0H)D concentration on MS development and the clinical and radiological activity of the disease is still inconclusive [1]. In many studies, higher concentrations of 25(0H)D have been associated with a reduced risk for MS development and with reduced clinical activity of MS, in terms of a low rate of disease relapse, slow disability progression, and low disease activity measured on brain MRI [3, 4]. 25(0H)D supplementation alone, or as an add-on to a disease-modifying therapy (DMT) in patients with MS, has been shown to significantly reduce new T2-hyperintense and gadolinium-enhanced lesions on brain MRI [5], although other studies have found no influence on disease activity on brain MRI [6, 7].

Referring to the article, we would like to present the results of a study comparing the concentrations of 25(0H)D in serum, assessed using an ELISA Kit (ImmunDiagnostik AG, Bensheim, Austria) with a microplate reader μQuant (Biotek Instruments Inc.,Winooski, Vermont, USA) in patients with relapsing-remitting multiple sclerosis (RRMS) and in healthy subjects from a control group (CG) (Tab. 1). None of the individuals was taking a 25(0H)D supplement, and none had symptoms of acute inflammation (their C reactive protein concentration was within the normal range i.e. < 10 mg/L) (Tab. 1). The study protocol was approved by the Medical University of Bialystok Ethics Committee for Research on Humans and Animals (R-I-002/171/2018). The MS patients (mean time from diagnosis 8 ± 0.5 years) were treated with interferon-β 1b in the Department of Neurology, Medical University of Bialystok. All patients with MS within the last 12 months had had a brain MRI and none had been treated with corticosteroids within the past six months. In patients with MS we assessed correlations between 25(0H)D concentrations and the season of the year in which they were born, the number of disease relapses, and

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Table 1. Clinical characteristics of patients with relapsing-remitting multiple sclerosis (RRMS) and the control group (CG)

Diagnosis	Number of patients (women)	Age (years)	EDSS	CRP (mg/L)
RRMS	57 (40)	43.7 ± 9.5	1.9 ± 1.2	1.0 ± 1.5
CG	19 (17)	41.7 ± 2.5	—	0.9 ± 0.5

CRP — C reactive protein; EDSS — Expanded Disability Status Scale

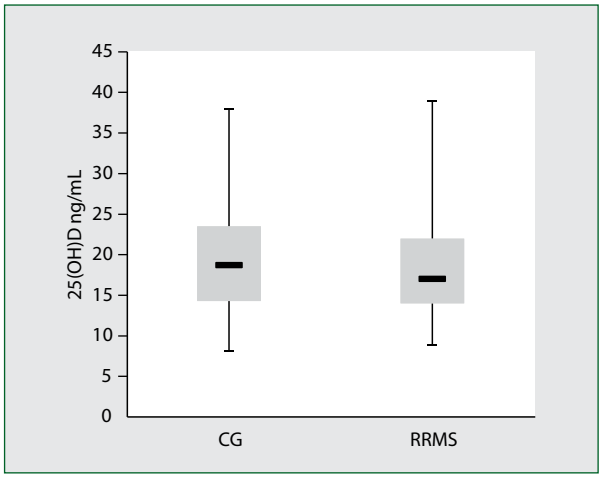

Figure 1. 25(0H)D concentrations in serum of patients with relapsing-remitting multiple sclerosis (RRMS) and in the control group (CG); Kruskal-Wallis test with post hoc Dwass-Steele-Critchlow-Fligner test ($p < 0.897$)

the new T2-hyperintense and gadolinium-enhanced lesions on brain MRI and the EDSS score.

We found no statistically significant differences between the concentrations of 25(0H)D in the serum of patients with RRMS (18.90 ± 6.96 ng/mL) and the control group (19.47 ± 7.78 ng/mL) ($p < 0,897$) (Fig. 1). 25(0H)D concentrations were below laboratory standards (30–40 ng/mL) in 52 patients (91.2%) with RRMS and in 18 subjects in the control group (94.7%). In patients with RRMS we observed a statistically significant relationship between concentrations of 25(0H)D and the number of disease relapses ($p < 0.032$). There was no statistically significant relationship between concentrations of 25(0H)D and the number of new T2-hyperintense lesions and the new gadolinium-enhanced lesions on brain MRI, the season of the year of birth, or the EDSS score.

In relation to the article written by Halina Bartosik-Psujek and Marek Psujek, our results cast doubt as to whether low serum 25(0H)D concentration is a reliable risk factor for developing MS. This is because almost all patients suffering from MS, as well as healthy controls, had low serum levels, which may result from insufficient sun exposure, the use of sun block, limited time spent in outdoor activity, staying indoors, and / or low 25(0H)D dietary intake [8].

Therefore, 25(0H)D should be supplemented in the whole population, especially in MS patients, according to the proven relationship between its concentration and the number of disease relapses.

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Ethics. *The work described in this article has been carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans; Uniform Requirements for Manuscripts submitted to Biomedical Journals.*

References

1. Bartosik-Psujek H, Psujek M. Vitamin D as an immune modulator in multiple sclerosis. Neurol Neurochir Pol. 2019; 53(2): 113–122, doi: 10.5603/PJNNS.a2019.0015, indexed in Pubmed: 30916776.
2. Holick MF. Vitamin D: importance in the prevention of cancers, type 1 diabetes, heart disease, and osteoporosis. Am J Clin Nutr. 2004; 79(3): 362–371, doi: 10.1093/ajcn/79.3.362, indexed in Pubmed: 14985208.
3. Fitzgerald KC, Munger KL, Köchert K, et al. Association of Vitamin D Levels With Multiple Sclerosis Activity and Progression in Patients Receiving Interferon Beta-1b. JAMA Neurol. 2015; 72(12): 1458–1465, doi: 10.1001/jamaneurol.2015.2742, indexed in Pubmed: 26458124.
4. Sintzel MB, Rametta M, Reder AT. Vitamin D and Multiple Sclerosis: A Comprehensive Review. Neurol Ther. 2018; 7(1): 59–85, doi: 10.1007/s40120-017-0086-4, indexed in Pubmed: 29243029.
5. Smolders J, Hupperts R, Barkhof F, et al. SOLAR study group. Efficacy of vitamin D3 as add-on therapy in patients with relapsing-remitting multiple sclerosis receiving subcutaneous interferon β-1a: a Phase II, multicenter, double-blind, randomized, placebo-controlled trial. J Neurol Sci. 2011; 311(1-2): 44–49, doi: 10.1016/j.jns.2011.04.013, indexed in Pubmed: 21620416.
6. Golan D, Halhal B, Glass-Marmor L, et al. Vitamin D supplementation for patients with multiple sclerosis treated with interferon-beta: a randomized controlled trial assessing the effect on flu-like symptoms and immunomodulatory properties. BMC Neurol. 2013; 13: 60, doi: 10.1186/1471-2377-13-60, indexed in Pubmed: 23767916.
7. Zheng C, He L, Liu L, et al. The efficacy of vitamin D in multiple sclerosis: A meta-analysis. Mult Scler Relat Disord. 2018; 23: 56–61, doi: 10.1016/j.msard.2018.05.008, indexed in Pubmed: 29778041.
8. Płudowski P, Ducki C, Konstantynowicz J, et al. Vitamin D status in Poland. Pol Arch Med Wewn. 2016; 126(7-8): 530–539, doi: 10.20452/pamw.3479, indexed in Pubmed: 27509842.

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'Falling off' the dopamine wagon

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Key words: Parkinson's disease, fracture, levodopa, neurodegenerative disease

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A 70 year-old man with Parkinson's disease (PD) of nearly 20 years' duration decided to see an acupuncturist for another opinion on the treatment of his PD. He was prescribed a series of herbal teas as well as 12 weeks of Chinese acupuncture. Following this treatment he felt better, not in terms of his motor symptoms, but because the teas relieved his chronic constipation. He was so delighted with the removal of this symptom that he questioned the need for PD medications, which amounted to total daily doses of 750 mg levodopa, 9 mg ropinirol, and 300 mg amantadine. Without consulting his neurologist, he abruptly decreased each of the prescribed doses by two-thirds. Shortly afterwards, he stumbled and fell on a tile floor, resulting in a sharp pain in the right groin radiating to the medial thigh. Radiographic evaluation of the right hip revealed an impacted right garden II femoral neck fracture, for which he underwent percutaneous pinning (Fig. 1).

Multiple factors contribute to the risk of falls for persons with parkinsonism. Hallmark symptoms of bradykinesia, rigidity, and postural instability may apply to all forms of parkinsonism. Neurogenic orthostatic hypotension is a manifestation of autonomic dysfunction that may lead to falls and is more common with parkinsonism due to synuclein pathology, such as PD or multiple system atrophy [1, 2]. Some persons with parkinsonism may have extraocular eye movement abnormalities, further increasing their risk of falling. This occurs most often in progressive supranuclear palsy and is thought to play a role in the 'early falls' often reported by patients at the time of diagnosis. Our patient had no evidence of orthostatic intolerance or eye movement abnormalities surrounding the time of his fall. He did however suffer from frequent freezing of gait (FOG) in addition to his tremor, bradykinesia, and rigidity, which led to his high intensity dopamine supplementation.

While persons with PD are at risk of falling, those with less mobility/core strength and declining bone density (which may follow reductions in dopaminergic therapy), are at increased risk for not only falls, but also significant injury and mortality stemming from subsequent surgery and complications [3, 4]. Furthermore, there is an increased risk of fractures among patients with PD and dementia compared to those without dementia; however, this risk is not solely explained by the presence of dementia [5]. Vitamin D deficiency and perhaps bone loss may also contribute to this risk. It has even been proposed that low vitamin D levels may be related to the pathomechanism underlying PD [6]. Our patient had no cognitive impairment at the time of his fall, but was diagnosed with osteopenia shortly thereafter.

The cornerstone of treatment for PD is dopamine supplementation via levodopa, which improves motor symptoms of resting tremor, bradykinesia, and rigidity. Other symptoms associated with more advanced disease states, such as postural instability and FOG, are recalcitrant to dopaminergic therapies. However, there is secondary improvement in ambulation probably due to decreased bradykinesia and rigidity.

In advanced stages of PD, patients may begin to think that their medication has lost its effectiveness due to an accumulation of 'dopa-resistant' symptoms. It can be tempting for patients, with or without the guidance of their neurologist, to seek a simplified medication regimen and decrease dosage or frequency of dopaminergic drug therapy. In the case of our patient, this decision was catalyzed by the assumption that the improvement of non-motor symptoms indicated a concomitant improvement of motor symptoms. Our patient's experience vividly illustrates how abruptly altering dopaminergic medications can have disastrous consequences.

In addition to emphasising the critical role of dopamine supplementation in PD, this case teaches us an important lesson that non-motor symptoms of PD deserve attention. Such symptoms, including constipation, affect up to 90% of PD patients [7]. In his *Essay on the Shaking Palsy*, James Parkinson wrote: "The bowels, which had been all along torpid,

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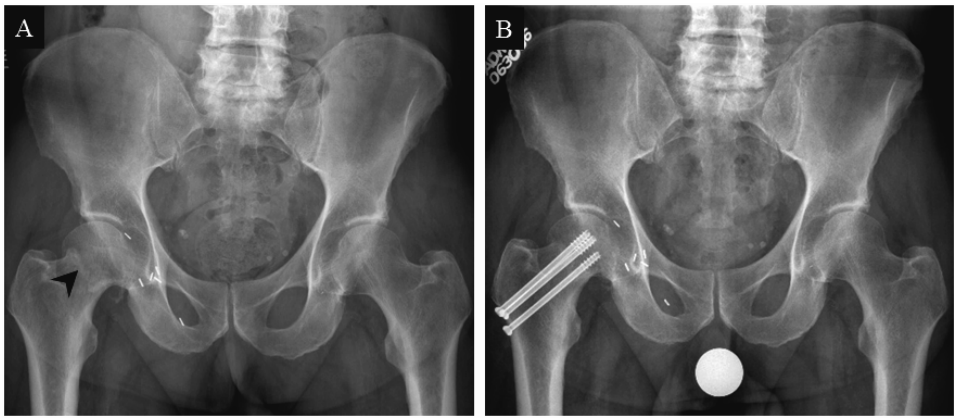

Figure 1. Pelvic X-ray. **A.** Pelvic X-ray of a 70 year-old man showing right garden II femoral neck fracture (black arrowhead); **B.** Follow-up pelvic X-ray after surgical repair with pinning

now, in most cases, demand stimulating medicines of very considerable power" [8]. Treating such non-motor symptoms may not only improve quality of life, but avoid catastrophe.

Because PD is classically thought of as a disorder of movement, patients may be less likely to mention non-motor symptoms in the setting of a neurological follow up visit, unless explicitly questioned on the topic by their neurologist. Due to the complexity of PD, this can be a challenge to accomplish within the confines of a short office visit. However, its importance cannot be overstated because many non-motor symptoms such as mood disorders, cognitive impairment, and autonomic dysfunction have a negative impact on quality of life [9].

This case underscores the importance of a strong therapeutic alliance in which patients are comfortable discussing these issues and therapy options they may be considering. Our patient pursued a non-traditional approach with Eastern medicine, which he credited with improving his constipation, and that led him to reduce his dopaminergic medications. Discussing the option of supplementing rather than replacing his medications might have prevented his injury.

Fortunately, our patient's fall resulted in morbidity that was repairable. It is advisable to stress to patients and caregivers not to decrease dopaminergic therapy, even in advanced PD, unless serious adverse effects develop. In such instances, considering alternative forms of treatment (i.e. medications and surgery) may be appropriate.

References

1. Nojszewska M, Potulska-Chromik A, Jamrozik Z, et al. Electrophysiological and clinical assessment of dysautonomia in multiple system atrophy (MSA) and progressive supranuclear palsy (PSP): a comparative study. Neurol Neurochir Pol. 2019; 53(1): 26–33, doi: 10.5603/PJNNS.a2019.0005, indexed in Pubmed: 30620042.
2. van Gerpen JA, Al-Shaikh RH, Tipton PW, et al. Progressive supranuclear palsy is not associated with neurogenic orthostatic hypotension. Neurology. 2019; 93(14): e1339–e1347, doi: 10.1212/WNL.0000000000008197, indexed in Pubmed: 31484717.
3. Johnell O, Melton LJ, Atkinson EJ, et al. Fracture risk in patients with parkinsonism: a population-based study in Olmsted County, Minnesota. Age Ageing. 1992; 21(1): 32–38, doi: 10.1093/ageing/21.1.32, indexed in Pubmed: 1553857.
4. Genever RW, Downes TW, Medcalf P. Fracture rates in Parkinson's disease compared with age- and gender-matched controls: a retrospective cohort study. Age Ageing. 2005; 34(1): 21–24, doi: 10.1093/ageing/afh203, indexed in Pubmed: 15591480.
5. Melton LJ, Leibson CL, Achenbach SJ, et al. Fracture risk after the diagnosis of Parkinson's disease: Influence of concomitant dementia. Mov Disord. 2006; 21(9): 1361–1367, doi: 10.1002/mds.20946, indexed in Pubmed: 16703587.
6. Newmark HL, Newmark J. Vitamin D and Parkinson's disease--a hypothesis. Mov Disord. 2007; 22(4): 461–468, doi: 10.1002/mds.21317, indexed in Pubmed: 17230473.
7. Palma JA, Kaufmann H. Treatment of autonomic dysfunction in Parkinson disease and other synucleinopathies. Mov Disord. 2018; 33(3): 372–390, doi: 10.1002/mds.27344, indexed in Pubmed: 29508455.
8. Parkinson J. An essay on the shaking palsy. 1817. J Neuropsychiatry Clin Neurosci. 2002; 14(2): 223–36; discussion 222, doi: 10.1176/jnp.14.2.223, indexed in Pubmed: 11983801.
9. Rahman S, Griffin HJ, Quinn NP, et al. Quality of life in Parkinson's disease: the relative importance of the symptoms. Mov Disord. 2008; 23(10): 1428–1434, doi: 10.1002/mds.21667, indexed in Pubmed: 18543333.

LETTER TO THE EDITOR

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Neurosurgery residency burnout: what can prevent this?

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ABSTRACT

Burnout is an occupational phenomenon indicating that the work and the workplace are responsible. We here discuss how a supportive resident-mentor relationship, and a positive working environment, could help to prevent resident burnout. A positive resident-mentor relationship can be achieved by understanding the mentor, the mentee, and the generational differences of each individual. A positive working environment depends on a healthy work-life balance and the atmosphere in the department. The benefits of preventing burnout include not only happier physicians but also fewer medical errors and better medical care. The universal reminders and proven suggestions in our paper could help address the burnout problem among working physicians worldwide.

Key words: burnout, mentorship, neurosurgery residency, work-life balance

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Neurosurgery is regarded as one of the most prestigious specialities in the medical world, and so it is understandable that the profession requires the input of an enormous amount of time and effort. During the marathon that is a neurosurgery residency, one can become exhausted. Burnout is more common among physicians compared to the average United States (USA) population [1] and residents are particularly at an increased risk, with a lower quality of life than attending physicians [2]. Older age, female sex, and junior residents all independently carry an increased rate of attrition, particularly in neurosurgery [3]. Suicide is still one of the leading causes of death among residents [4]. Fortunately, despite the rigours of residency, neurosurgery residents actually have a lower prevalence of burnout than other medical specialities [5]. Burnout is a diagnosis included in Revision 11 of the International Classification of Diseases [6] as a strictly occupational phenomenon, indicating that the work and workplace are responsible. Given this sobering background, what preventative interventions can well-meaning residency programmes take? Is there a way to support residents within the bounds of an 80-hour working week? In our paper, we suggest how an encouraging resident-mentor relationship and a positive working environment can help eliminate resident burnout.

A meaningful mentorship

Having a mentor results in greater work satisfaction, higher academic scores, and more publications [7]. A study conducted among American neurosurgery residents proved that neurosurgery residents can handle a tremendous workload and still be satisfied in their work so long as they have a meaningful mentorship [8]. A positive resident-mentor relationship is especially necessary since surgeon-to-surgeon training is needed to acquire advanced neurosurgical skills. Moreover, a fruitful mentorship not only benefits the mentor and the mentee, but also the host institution which benefits from higher physician retention and higher productivity [9].

Suggestions for mentors

Some attending physicians might find themselves having to fulfill the role of a mentor by chance. This is challenging, as mentoring is a tremendous responsibility that requires not only the ability to teach neurosurgical skills but also the social intelligence to guide, counsel and orient mentees in a new environment. Despite its challenges, mentoring can offer some people a sense of personal fulfillment, development of leadership skills, and even a renewed interest in neurosurgery [9].

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Active leadership has been proven to increase work satisfaction and ameliorate against burnout [10]. Therefore, we propose that mentors encourage residents to seize leadership opportunities. These might include designing their own research projects, running for a local council or serving as a delegate for interim meetings. Moreover, it is the feeling of accomplishment that is most related to happiness — not the number of hours a week a physician works [11]. We propose that mentors advocate specific, measurable and attainable goals for residents, since unrealistic expectations will wear down a resident. Finally, we urge mentors to instill the core values of neurosurgery at career-entry so that the mentee can navigate academic culture and establish helpful contacts [12]. The end goal is to provide support and guidance so that a motivated resident can prosper, rather than become overwhelmed with additional responsibilities. Therefore, we suggest that a mentor engages in these activities *jointly* with the mentee.

Suggestions for residents

We advise residents to consider the resident-mentor relationship as empowering and to be open to suggestions and feedback. With a mentor, the steep learning curve of neurosurgery can be curtailed, productivity may be advanced, and new networking opportunities may appear. Mentors can also impart informal knowledge that might not be found in a textbook.

Junior residents must regularly deal with bureaucratic burdens and copious documentation because they are necessary obligations involved in patient care. In consequence, they may spend less time in the operating theatre [13].

However, we ask residents to recognise that even the most menial tasks play a vital role in a hospital's ecosystem, and that even the most renowned neurosurgeons of today faced the same paperwork in the past.

Generational obstacles

A resident-mentor relationship may be hindered by a generational gap; baby boomers perceive the independent-minded Millennial generation to be distrustful and reluctant to establish a resident-mentor relationship. Baby boomers generally take pride in the patience and discipline required for mundane work, while Millennials consider it a hindrance. You could say that in general baby boomers 'live to work' while Millennials 'work to live'. [14]. Some authors feel that while duty hour regulations have improved the conditions of residency, they have also lessened empathy in the workplace [14]. We recommend that the mentorship style should maintain some flexibility to take account of generational differences. For example, the resident-mentor relationship can take place face to face, over an online chat (e.g. Skype), or even in a group setting. "Personal inadequacies and relationship problems" are quoted as the most common cause of dysfunctional mentoring. Therefore, we believe that effective communication — in whatever medium and capacity — is paramount [9, 15]. No matter what style is applied, shared values and effective communication have proven to be the pillars of an effective mentorship [16].

A positive working environment

While a mentorship is crucial to reducing physician burnout, studies show that facilitating the proper environment at an institution is necessary for such a relationship to prosper [15]. A positive and encouraging work environment results in more productive and happier residents. This has been proven true when considering the deleterious effects of rudeness among physicians on patient care and safety [17, 18]. In the words of Juha Hernesniemi — a Finn who is one of the world's leading neurosurgeons: "The atmosphere in the department should be open and supportive of good work, and the employees should be proud of their clinic... Express your appreciation of your hardworking colleagues" [19]. Furthermore, a resident under Hernesniemi's department recalled that when serious complications are encountered, "the colleagues are very supportive, and from their own experience understand that there is no room for accusations and cynicism, but constructive re-evaluation of the case and circumstances" [19]. We feel that by understanding the resident, and by establishing a sustainable work-life balance, a positive work environment can be brought about.

Understanding the resident

In the USA, neurosurgical residents have already proven their competency, work ethic and dedication when they match into one of the most selective medical residency programmes. They require some of the highest United States Medical Licensing Examination scores, and illustrate their dedication to neurosurgery with numerous scientific publications. On average, international medical graduates have more than 47 research projects (including abstracts, presentations, and publications) [20]. Compare this to only 12 research projects for orthopaedic surgery [20]. With these rigorous entry standards, few individuals can match into residency. We maintain that it is important for students to recognise residents as being competent individuals even though they might not have a chance to exhibit it with their surgical skills during residency.

During residency a resident might question their specialisation and career choice. We encourage such a resident to discuss these issues with family, friends, mentors or even a psychiatrist to reflect on their achievements and establish reasonable expectations. Psychological support can also come in the form of yoga, mindfulness–based cognitive therapy, and cognitive behavioural therapy as they have all proved to increase health-related quality of life [21].

Work-life balance

While neurosurgery offers exciting new opportunities for research, intellectual development, social prestige, and wealth, it consumes time otherwise spent on one's family, friends, hobbies, spiritual and other goals. The work–life balance is perhaps complicated when a spouse is not a physician and does not always appreciate the long hours spent away from home. Research shows that a lack of work–life balance can be detrimental to a trainee's learning and well-being; this has been seen especially in women with children [22]. Work-life balance can be understood as rungs on a ladder, where both aspects are dependent one on another i.e. a better attitude at home results in a better attitude at work [23]. Therefore, we assert that to maintain a positive work environment the academic institution and resident should work together to try to establish a sustainable work-life balance early.

A preliminary study in Europe showed that the 2003 Working Time Directive — which attempted to promote health and safety by cutting excessive working hours — resulted in a steep decline in surgical cases over time and less surgical exposure in residents graduating [24]. Thus, we emphasise that perhaps it is not long hours that are to blame for physician burnout, but rather the invalidating environment that needs to be addressed. So, even with the same *quantity* of hours, the *quality* of practice can improve to cultivate a resident to his or her most productive potential.

In summary, we encourage physicians to establish a positive and supportive environment in the department and strengthen the resident-mentor relationship despite generational obstacles. The problem of burnout is the responsibility of a department's entire team. Its benefits include not only happier physicians but fewer medical errors [25] and better medical care at an institution. We invite neurosurgeons to serve as leaders and set an example for other medical specialities. The universal reminders and suggestions in our paper can help address the burnout problem among working physicians worldwide.

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References

1. Dyrbye LN, West CP, Satele D, et al. Burnout among U.S. medical students, residents, and early career physicians relative to the general U.S. population. Acad Med. 2014; 89(3): 443–451, doi: 10.1097/ACM.0000000000000134, indexed in Pubmed: 24448053.
2. Pulcrano M, Evans SRT, Sosin M. Quality of Life and Burnout Rates Across Surgical Specialties: A Systematic Review. JAMA Surg. 2016; 151(10): 970–978, doi: 10.1001/jamasurg.2016.1647, indexed in Pubmed: 27410167.
3. Agarwal N, White MD, Pannullo SC, et al. Analysis of national trends in neurosurgical resident attrition. J Neurosurg. 2018 [Epub ahead of print]: 1–6, doi: 10.3171/2018.5.JNS18519, indexed in Pubmed: 30497159.
4. Yaghmour NA, Brigham TP, Richter T, et al. Causes of Death of Residents in ACGME-Accredited Programs 2000 Through 2014: Implications for the Learning Environment. Acad Med. 2017; 92(7): 976–983, doi: 10.1097/ACM.0000000000001736, indexed in Pubmed: 28514230.
5. Shakir HJ, McPheeters MJ, Shallwani H, et al. The Prevalence of Burnout Among US Neurosurgery Residents. Neurosurgery. 2018; 83(3): 582–590, doi: 10.1093/neuros/nyx494, indexed in Pubmed: 29088408.
6. Burn-out an „occupational phenomenon": International Classification of Diseases. WHO 2019. https://www.who.int/mental_health/evidence/burn-out/en/ (June 7, 2019).
7. Feldman MD, Arean PA, Marshall SJ, et al. Does mentoring matter: results from a survey of faculty mentees at a large health sciences university. Med Educ Online. 2010; 15, doi: 10.3402/meo.v15i0.5063, indexed in Pubmed: 20431710.
8. Attenello FJ, Buchanan IA, Wen T, et al. Factors associated with burnout among US neurosurgery residents: a nationwide survey. J Neurosurg. 2018; 129(5): 1349–1363, doi: 10.3171/2017.9.JNS17996, indexed in Pubmed: 29424650.
9. Burgess A, van Diggele C, Mellis C. Mentorship in the health professions: a review. Clin Teach. 2018; 15(3): 197–202, doi: 10.1111/tct.12756, indexed in Pubmed: 29318730.
10. Hamade YJ, Aoun RJ, Zimmerman RS, et al. The Modern Neurosurgical Leader as a Cure for Team Burnout. Neurosurgery. 2015; 77(2): N13, doi: 10.1227/01.neu.0000467292.26010.c3, indexed in Pubmed: 26181785.
11. Eckleberry-Hunt J, Kirkpatrick H, Taku K, et al. Relation Between Physicians' Work Lives and Happiness. South Med J. 2016; 109(4): 207–212, doi: 10.14423/SMJ.0000000000000437, indexed in Pubmed: 27043800.
12. Nick JM, Delahoyde TM, Del Prato D, et al. Best practices in academic mentoring: a model for excellence. Nurs Res Pract. 2012; 2012: 937906, doi: 10.1155/2012/937906, indexed in Pubmed: 22685645.
13. Spiotta AM, Kalhorn SP, Patel SJ. Letter: How to Combat the Burnout Crisis in Neurosurgery? Cathedrals and Mentorship. Neurosurgery. 2019; 84(4): E257–E258, doi: 10.1093/neuros/nyy611, indexed in Pubmed: 30566685.
14. Spiotta AM, Patel S. Generational tensions are a distraction from addressing the burnout crisis in Neurosurgery. Neurol India. 2018; 66(6): 1572–1574, doi: 10.4103/0028-3886.246283, indexed in Pubmed: 30504542.
15. Sambunjak D, Straus SE, Marusic A. A systematic review of qualitative research on the meaning and characteristics of mentoring in academic medicine. J Gen Intern Med. 2010; 25(1): 72–78, doi: 10.1007/s11606-009-1165-8, indexed in Pubmed: 19924490.
16. Straus SE, Johnson MO, Marquez C, et al. Characteristics of successful and failed mentoring relationships: a qualitative study across two academic health centers. Acad Med. 2013; 88(1): 82–89, doi: 10.1097/ACM.0b013e31827647a0, indexed in Pubmed: 23165266.
17. Riskin A, Erez A, Foulk TA, et al. The Impact of Rudeness on Medical Team Performance: A Randomized Trial. Pediatrics. 2015; 136(3): 487–495, doi: 10.1542/peds.2015-1385, indexed in Pubmed: 26260718.

18. Riskin A, Erez A, Foulk T, et al. Rudeness and Medical Team Performance. Pediatrics. 2017; 139(2): e20162305, doi: 10.1542/peds.2016-2305.
19. Lehecka M, Laakso A, Hernesniemi J. Helsinki Microneurosurgery Basics and Tricks. Helsinki: Druckerei Hohl GmbH & Co. KG / Germany; 2011.
20. Charting Outcomes in the Match: International Medical Graduates; 2018.
21. Grensman A, Acharya BD, Wändell P, et al. Effect of traditional yoga, mindfulness-based cognitive therapy, and cognitive behavioral therapy, on health related quality of life: a randomized controlled trial on patients on sick leave because of burnout. BMC Complement Altern Med. 2018; 18(1): 80, doi: 10.1186/s12906-018-2141-9, indexed in Pubmed: 29510704.
22. Rich A, Viney R, Needleman S, et al. ‚You can't be a person and a doctor': the work-life balance of doctors in training-a qualitative study. BMJ Open. 2016; 6(12): e013897, doi: 10.1136/bmjopen-2016-013897, indexed in Pubmed: 27913563.
23. George N, Kiran PR, Sulekha T, et al. Work-life Balance among Karnataka State Road Transport Corporation (KSRTC) Workers in Anekal Town, South India. Indian J Occup Environ Med. 2018; 22(2): 82–85, doi: 10.4103/ijoem.IJOEM_25_18, indexed in Pubmed: 30319228.
24. Stienen MN, Bartek J, Czabanka MA, et al. EANS Young Neurosurgeons and EANS Training Committee. Neurosurgical procedures performed during residency in Europe-preliminary numbers and time trends. Acta Neurochir (Wien). 2019; 161(5): 843–853, doi: 10.1007/s00701-019-03888-3, indexed in Pubmed: 30927157.
25. Shanafelt TD, Balch CM, Bechamps G, et al. Burnout and medical errors among American surgeons. Ann Surg. 2010; 251(6): 995–1000, doi: 10.1097/SLA.0b013e3181bfdab3, indexed in Pubmed: 19934755.

AVONEX (interferon beta 1a) 30 mikrogramów/0,5 ml roztwór do wstrzykiwań. **Skład:** Każdy wstrzykiwacz półautomatyczny napełniony jednorazowego użytku zawiera 30 mikrogramów (6 milionów j.m.) interferonu beta-1a w roztworze o objętości 0,5 ml. Stężenie wynosi 30 mikrogramów na 0,5 ml. 30 mikrogramów produktu AVONEX zawiera 6 milionów j.m. (jednostek międzynarodowych) aktywności przeciwwirusowej według Międzynarodowego Standardu dla Interferonu Światowej Organizacji Zdrowia (WHO). Nie jest znana aktywność według innych standardów. **Postać farmaceutyczna:** Roztwór do wstrzykiwań we wstrzykiwaczu. Klarowny i bezbarwny roztwór. **Wskazania do stosowania:** Produkt AVONEX jest wskazany w leczeniu: 1) pacjentów ze zdiagnozowaną nawracającą postacią stwardnienia rozsianego (SR) określonego w badaniach klinicznych jako dwa lub więcej zaostrzeń choroby (nawrotów) w czasie ostatnich trzech lat bez oznak postępu choroby między nawrotami; AVONEX spowalnia postęp niesprawności i zmniejsza częstość nawrotów, 2) pacjentów, u których wystąpił pojedynczy przypadek demielinizacji z czynnym procesem zapalnym, którego ciężkość kwalifikuje do leczenia podawanymi dożylnie kortykosteroidami, jeśli alternatywna diagnoza została wykluczona, i istnieje duże ryzyko rozwoju klinicznie zdefiniowanego stwardnienia rozsianego (patrz punkt: Właściwości farmakodynamiczne w Charakterystyce Produktu Leczniczego (ChPL)). Produkt AVONEX należy odstawić u pacjentów, u których rozwinie się postępujące SR. **Dawkowanie i sposób podawania:** Leczenie powinno być rozpoczęte pod kontrolą lekarza prowadzącego, który ma doświadczenie w leczeniu tej choroby. Dawkowanie: Dorośli: W leczeniu nawrotowego stwardnienia rozsianego (SR) zalecana dawka wynosi 30 mikrogramów (0,5 ml roztworu) we wstrzyknięciu domięśniowym (im.) podawana raz w tygodniu (patrz punkt: Specjalne środki ostrożności dotyczące usuwania i przygotowania produktu leczniczego do stosowania w ChPL). Nie wykazano dodatkowych korzyści po stosowaniu większej dawki (60 mikrogramów) raz na tydzień. Dostosowywanie dawki: Aby zmniejszyć częstość występowania i nasilenie objawów grypopodobnych (patrz punkt: Działania niepożądane), leczenie można rozpocząć od dostosowywania dawki. Jeżeli stosuje się zestaw ampułko-strzykawkę, leczenie zaczynamy od ¼ pełnej dawki, zwiększając ją w odstępach tygodniowych do osiągnięcia pełnej dawki (30 mikrogramów na tydzień) w czwartym tygodniu. Innym sposobem dostosowywania dawki jest rozpoczęcie leczenia produktem AVONEX od dawki odpowiadającej w przybliżeniu ½ pełnej dawki podawanej raz w tygodniu przed zwiększeniem jej do pełnej dawki. Aby uzyskać odpowiednią skuteczność, po początkowym okresie dostosowywania należy osiągnąć i utrzymywać dawkę 30 mikrogramów, podawaną raz w tygodniu. Po osiągnięciu pełnej dawki, pacjenci mogą zacząć stosować wstrzykiwacz AVONEX PEN. Przed wstrzyknięciem oraz przez 24 godziny po każdym wstrzyknięciu zaleca się podawanie leku przeciwbólowego o działaniu przeciwgorączkowym, aby złagodzić objawy grypopodobne związane ze stosowaniem produktu AVONEX. Te objawy zwykle występują przez pierwsze kilka miesięcy leczenia. Dzieci i młodzież: Nie określono dotychczas bezpieczeństwa stosowania ani skuteczności produktu leczniczego AVONEX w grupie nastolatków w wieku od 12 do 16 lat. Aktualne dane przedstawiono w punktach: Działania niepożądane i Właściwości farmakodynamiczne w ChPL, ale brak zaleceń dotyczących dawkowania. Nie ustalono jak dotąd bezpieczeństwa ani skuteczności produktu AVONEX u dzieci poniżej 12. roku życia. Brak dostępnych danych. Osoby w podeszłym wieku: Badania kliniczne nie obejmowały dostatecznej liczby pacjentów w 65. roku życia i powyżej, aby ustalić, czy odpowiedź na lek jest w tej grupie wiekowej odmienna niż u młodszych pacjentów. Jednakże na podstawie klirensu substancji czynnej nie istnieje teoretyczne uzasadnienie, aby ustalać jakiekolwiek wymaganie w zakresie dostosowywania dawki u osób starszych. Sposób podawania: Obecnie nie ustalono, jak długo pacjenci powinni być leczeni. Należy dokonać klinicznej oceny stanu pacjenta po dwóch latach leczenia. Decyzję o dłuższym leczeniu podejmuje lekarz w zależności od indywidualnego stanu pacjenta. Należy przerwać leczenie, jeśli u pacjenta rozwinie się postępujący proces przewlekłego SR. AVONEX PEN jest to wstrzykiwacz półautomatyczny napełniony, przeznaczony do jednorazowego użytku, który należy stosować jedynie po odpowiednim przeszkoleniu. Zalecanym miejscem do wykonania domięśniowego wstrzyknięcia za pomocą wstrzykiwacza AVONEX PEN jest górna, zewnętrzna część mięśni uda. Miejsce wstrzyknięcia należy zmieniać co tydzień. Podając produkt AVONEX za pomocą wstrzykiwacza AVONEX PEN, należy postępować zgodnie z instrukcją podaną w ulotce dołączonej do opakowania. Przeciwwskazania: - Leczenie pacjentów, u których stwierdzono w wywiadzie nadwrażliwość na naturalny lub rekombinowany interferon beta lub którąkolwiek substancję pomocniczą. - Leczenie pacjentów z istniejącą ciężką depresją i (lub) myślami samobójczymi (patrz punkty: Specjalne ostrzeżenia i środki ostrożności dotyczące stosowania i Działania niepożądane). **Specjalne ostrzeżenia i środki ostrożności dotyczące stosowania:** Identyfikowalność: W celu poprawienia identyfikowalności biologicznych produktów leczniczych należy czytelnie zapisać nazwę i numer serii podawanego produktu. Produkt AVONEX należy podawać z dużą ostrożnością pacjentom z występującymi w przeszłości lub obecnie istniejącymi zaburzeniami depresyjnymi, zwłaszcza pacjentom, u których przed rozpoczęciem leczenia występują myśli samobójcze. Wiadomo, że depresja i myśli samobójcze występują ze zwiększoną częstością w populacji ze stwardnieniem rozsianym oraz podczas stosowania interferonu. Pacjentów należy poinformować o konieczności natychmiastowego zgłoszenia lekarzowi objawów depresji i (lub) myśli samobójczych. Pacjentów wykazujących objawy depresji należy ściśle monitorować podczas terapii i odpowiednio leczyć. Należy rozważyć przerwanie leczenia produktem AVONEX. Produkt AVONEX należy podawać z zachowaniem ostrożności pacjentom z napadami drgawkowymi w przeszłości, pacjentom, którym podaje się leki przeciwpadaczkowe, zwłaszcza jeśli padaczka nie jest właściwie kontrolowana przez stosowanie leków przeciwpadaczkowych. Należy zachować szczególną ostrożność i rozważyć przeprowadzanie częstych kontroli lekarskich, podając produkt AVONEX pacjentom z ciężką niewydolnością nerek i wątroby oraz pacjentom z ciężkim zahamowaniem czynności szpiku. Mikroangiopatia zakrzepowa (ang. thrombotic microangiopathy, TMA): podczas leczenia interferonem beta zgłaszano przypadki TMA (w tym przypadki śmiertelne) występującej pod postacią zakrzepowej plamicy małopłytkowej (TTP) lub hemolitycznego zespołu mocznicowego (HUS). Zdarzenia zgłaszano w różnych okresach leczenia i mogą one występować po kilku tygodniach, a nawet kilku latach, od rozpoczęcia leczenia interferonem beta. Wczesne objawy kliniczne obejmują trombocytopenię, nowo rozpoznane nadciśnienie, gorączkę, objawy ze strony ośrodkowego układu nerwowego (np. splątanie i niedowład) i zaburzenie czynności nerek. Do wyników badań laboratoryjnych wskazujących na TMA należą: zmniejszona liczba płytek, podwyższone stężenie dehydrogenazy mleczanowej (LDH) w surowicy wskutek hemolizy oraz obecność schistocytów (fragmentów erytrocytów) w rozmazie krwi. Dlatego w razie stwierdzenia klinicznych objawów TMA zaleca się wykonanie dodatkowych badań poziomu płytek, LDH w surowicy, rozmazu krwi i czynności nerek. W razie rozpoznania TMA konieczne jest bezzwłoczne wdrożenie leczenia (w tym rozważenie wymiany osocza) i zalecane jest natychmiastowe odstawienie produktu AVONEX. Zespół nerczycowy: podczas leczenia produktami zawierającymi interferon beta zgłaszano przypadki zespołu nerczycowego wywołanego przez różne rodzaje nefropatii, w tym ogniskowe segmentalne stwardnienie kłębuszków nerkowych z zapadnięciem pętli włośniczkowych (ang. collapsing FSGS), zmianę minimalną (ang. MCD), błoniasto-rozplemowe kłębuszkowe zapalenie nerek (ang. MPGN) i mezangialne kłębuszkowe zapalenie nerek (ang. MGN). Zdarzenia te zgłaszano w różnych okresach w trakcie leczenia i mogą one występować po kilku latach stosowania interferonu beta. Zaleca się okresowe monitorowanie wczesnych objawów podmiotowych lub przedmiotowych, takich jak obrzęki, białkomocz czy zaburzona czynność nerek, zwłaszcza u pacjentów z wysokim ryzykiem wystąpienia choroby nerek. Konieczne jest szybkie podjęcie leczenia zespołu nerczycowego i rozważenie przerwania leczenia produktem AVONEX. Podczas leczenia interferonem beta, w badaniach po wprowadzeniu leku na rynek, opisywano uszkodzenie wątroby, w tym zwiększoną aktywność enzymów wątrobowych, zapalenie wątroby, autoimmunologiczne zapalenie wątroby i niewydolność wątroby. W niektórych przypadkach reakcje te występowały w obecności innych produktów leczniczych uszkadzających wątrobę. Nie określono addytywnego działania podczas jednoczesnego stosowania kilku produktów leczniczych lub innych czynników hepatotoksycznych (np. alkoholu). Należy obserwować pacjentów pod kątem wystąpienia objawów uszkodzenia wątroby i zachować ostrożność w przypadku jednoczesnego stosowania interferonu z innymi produktami leczniczymi uszkadzającymi wątrobę. Pacjentów z chorobami serca, takimi jak dławica piersiowa, zastoinowa niewydolność serca lub arytmia, należy uważnie obserwować ze względu na możliwość pogorszenia się ich stanu klinicznego podczas leczenia produktem AVONEX. Objawy grypopodobne związane z leczeniem produktem AVONEX mogą pogorszyć stan zdrowia pacjentów z chorobami serca. Z leczeniem interferonem są związane nieprawidłowe wyniki testów laboratoryjnych. Podczas leczenia produktem AVONEX oprócz rutynowych badań wymaganych do monitorowania pacjentów ze SR, zalecane jest również wykonanie pełnego obrazu białokrwinkowego krwi obwodowej, liczby płytek i badań biochemicznych krwi, w tym badań czynności wątroby. Pacjenci z zahamowaniem czynności szpiku mogą wymagać wzmożonej kontroli pełnej krwi obwodowej z różnicowym rozpoznaniem płytek krwi. U leczonych pacjentów mogą powstać przeciwciała przeciw produktowi AVONEX. U niektórych pacjentów przeciwciała te (przeciwciała neutralizujące) zmniejszają in vitro aktywność interferonu beta-1a. Przeciwciała neutralizujące są związane ze zmniejszeniem in vivo biologicznego działania produktu AVONEX i mogą być związane z obniżeniem skuteczności klinicznej. Ocenia się, że częstość wytwarzania przeciwciał neutralizujących osiąga stały poziom po 12 miesiącach leczenia. Dane zebrane u pacjentów leczonych produktem AVONEX do trzech lat, wskazują, że u około 5% do 8% pacjentów zostają wytworzone przeciwciała neutralizujące. Stosowanie różnych metod oznaczania w surowicy przeciwciał neutralizujących interferony ogranicza możliwość porównania antygenowości różnych produktów. **Działania niepożądane:** Najczęstszym występującym działaniem niepożądanym związanym ze stosowaniem produktu AVONEX są objawy grypopodobne. Najczęściej opisywane objawy grypopodobne to: bóle mięśniowe, gorączka, dreszcze, pocenie się, osłabienie, bóle głowy i nudności. Dostosowywanie dawki produktu AVONEX na początku leczenia, pozwala zmniejszyć nasilenie i częstość występowania objawów grypopodobnych. Objawy grypopodobne są szczególnie nasilone w początkowej fazie leczenia, a częstość ich występowania zmniejsza się podczas kontynuowania terapii. Po wstrzyknięciu leku mogą wystąpić przejściowe objawy neurologiczne przypominające objawy nasilenia stwardnienia rozsianego (SR). W każdym momencie leczenia może wystąpić przejściowe wzmożenie napięcia mięśniowego i (lub) ciężkie osłabienie mięśniowe uniemożliwiające wykonywanie ruchów dowolnych. Objawy te występują w ograniczonym czasie trwania w zależności od kolejnych wstrzyknięć i mogą powtarzać się po kolejnych wstrzyknięciach. W niektórych przypadkach objawy te są związane z występowaniem objawów grypopodobnych. Częstość występowania działań niepożądanych jest wyrażona w pacjentolatach według następujących kategorii: Bardzo często (≥1/10 pacjentolat); Często (≥1/100 do <1/10 pacjentolat); Niezbyt często (≥1/1 000 do <1/100 pacjentolat); Rzadko (≥1/10 000 do <1/1 000 pacjentolat); Bardzo rzadko (<1/10 000 pacjentolat); Nieznana (częstość nie może być określona na podstawie dostępnych danych). Pacjentoczas jest sumą indywidualnych jednostek czasu, w których pacjent uczestniczący w badaniu był poddany działaniu produktu AVONEX przed wystąpieniem działania niepożądanego. Na przykład 100 osobolat można obserwować u 100 pacjentów leczonych przez okres jednego roku lub u 200 pacjentów leczonych przez okres pół roku. Działania niepożądane zaobserwowane w trakcie badań(badania kliniczne i badania obserwacyjne z okresem obserwacji od dwóch do sześciu lat) i inne działania niepożądane o nieznanej częstości zgłoszone w spontanicznych raportach z rynku: bardzo często - ból głowy, objawy grypopodobne, gorączka, dreszcze, pocenie się; często - zmniejszenie liczby limfocytów, zmniejszenie liczby białych krwinek, zmniejszenie liczby granulocytów obojętnochłonnych, zmniejszenie hematokrytu, zwiększenie stężenia potasu we krwi, zwiększenie stężenia azotu mocznikowego we krwi, spastyczność mięśni, niedoczulica, wyciek wodnisty z nosa, wymioty, biegunka, nudności, wysypka, nasilone pocenie się, stłuczenia, kurcz mięśni, ból karku, bóle mięśni, bóle stawów, bóle kończyn, bóle pleców, sztywność mięśni, sztywność mięśniowo szkieletowa, brak łaknienia, nagłe zaczerwienienie, ból w miejscu wstrzyknięcia, rumień w miejscu wstrzyknięcia, siniak w miejscu wstrzyknięcia, osłabienie, ból, zmęczenie, złe samopoczucie, nocne pocenie się, depresja, bezsenność; niezbyt często - zmniejszenie liczby płytek krwi, łysienie, uczucie pieczenia w miejscu wstrzyknięcia, krwotok maciczny, krwotok miesiączkowy; rzadko - mikroangiopatia zakrzepowa, w tym zakrzepowa plamica małopłytkowa (TTP) lub hemolityczny zespół mocznicowy (HUS) (Dotyczy klasy produktów zawierających interferon beta, patrz punkt: Specjalne ostrzeżenia i środki ostrożności dotyczące stosowania), duszność, zespół nerczycowy, stwardnienie kłębuszków nerkowych; częstość nieznana - zmniejszenie masy ciała, zwiększenie masy ciała, nieprawidłowe wyniki testów czynności wątroby, kardiomiopatia, zastoinowa niewydolność serca, kołatanie serca, arytmia, tachykardia, pancytopenia, trombocytopenia, objawy neurologiczne, omdlenie, wzmożone napięcie, zawroty głowy, parestezje, drgawki, migrena, tętnicze nadciśnienie płucne (dotyczy klasy produktów zawierających interferon, patrz poniżej Tętnicze nadciśnienie płucne.), obrzęk naczynioruchowy, świąd, wysypka pęcherzykowa, pokrzywka, nasilenie łuszczycy, układowy toczeń rumieniowaty, osłabienie mięśni, zapalenie stawów, niedoczynność tarczycy, nadczynność tarczycy, ropień w miejscu wstrzyknięcia, rozszerzenie naczyń, odczyn w miejscu wstrzyknięcia, zapalenie w miejscu wstrzyknięcia, zapalenie tkanki łącznej w miejscu wstrzyknięcia, martwica w miejscu wstrzyknięcia, krwawienie w miejscu wstrzyknięcia, bóle w klatce piersiowej, reakcja anafilaktyczna, wstrząs anafilaktyczny, reakcje nadwrażliwości (obrzęk naczynioruchowy, duszność, pokrzywka, wysypka, wysypka ze świądem), niewydolność wątroby, zapalenie wątroby, autoimmunologiczne zapalenie wątroby, próby samobójcze, psychozy, niepokój, splątanie, niestabilność emocjonalna. Tętnicze nadciśnienie płucne W związku ze stosowaniem produktów zawierających interferon beta zgłaszano przypadki tętniczego nadciśnienia płucnego (ang. pulmonary arterial hypertension, PAH). Zdarzenia zgłaszano w różnych momentach trwania leczenia interferonem beta, w tym kilka lat po jego rozpoczęciu. Dzieci i młodzież: Ograniczone opublikowane dane sugerują, że profil bezpieczeństwa u nastolatków w wieku od 12 do 16 lat, przyjmujących produkt AVONEX w dawce 30 mikrogramów domięśniowo raz w tygodniu jest zbliżony do obserwowanego u dorosłych. **Podmiot odpowiedzialny:** Biogen Netherlands B.V., Prins Mauritslaan 13, 1171 LP Badhoevedorp, Holandia **Numer pozwolenia na dopuszczenie do obrotu:** EU/1/97/033/005-6. **Data zatwierdzenia lub częściowej zmiany tekstu ChPL:** 09/2019. **Kategoria dostępności:** Produkt leczniczy wydawany z przepisu lekarza - Rp. **Odpłatność dla pacjenta:** produkt bezpłatny, refundowany w ramach programu lekowego B.29: „Leczenie stwardnienia rozsianego". Urzędowa cena zbytu - brutto 3402,00 PLN. Obwieszczenie Ministra Zdrowia z dnia 30 sierpnia 2019 r. w sprawie wykazu refundowanych leków, środków spożywczych specjalnego przeznaczenia żywieniowego oraz wyrobów medycznych na 1 września 2019 r. (DZ. URZ. Min. Zdr. 2019.65). **Przedstawiciel podmiotu odpowiedzialnego:** Biogen Poland sp. z o.o. tel.: + 48 22 116 86 94 **Informacja medyczna:** Biogen Poland Sp. z o. o., ul. Salsy 2, 02-823 Warszawa, tel.: 48 22 351 51 00, e-mail: informacja.medyczna@biogen.com **Zgłaszanie działań niepożądanych:** e-mail: safetypl@biogen.com, tel. 22 351 51 31. Szczegółowe informacje o tym produkcie leczniczym są dostępne na stronie internetowej Europejskiej Agencji Leków http://www.ema.europa.eu.

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Piśmiennictwo:

1. Charakterystyka Produktu Leczniczego AVONEX. Październik 2019. www.ema.europa.eu

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*p=0.02; † p=0.002; ‡ p=0.05.

ZMIANY W ZAPISACH AVONEX CHPL:

„Jeśli jest to klinicznie uzasadnione, można rozważyć stosowanie produktu Avonex w okresie ciąży.”

„Produkt Avonex może być stosowany podczas karmienia piersią.[1]”

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Dane z badań rejestracyjnych oraz zgłoszenia po wprowadzeniu do obrotu obejmujące dużą liczbę (ponad 1000 kobiet w ciąży) zastosowań produktu wskazują na brak zwiększonego ryzyka poważnych wad wrodzonych po ekspozycji na interferon beta przed zapłodnieniem oraz w pierwszym trymestrze ciąży. Niemniej jednak nie można było dokładnie określić czasu trwania ekspozycji w pierwszym trymestrze, ponieważ dane były gromadzone w okresie, gdy stosowanie interferonu beta w czasie ciąży było przeciwwskazane i leczenie prawdopodobnie było przerywane po stwierdzeniu i/lub potwierdzeniu ciąży. Istnieją bardzo ograniczone dane dotyczące ekspozycji w drugim i trzecim trymestrze ciąży.

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