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REVIEW

Artificial reproductive techniques in multiple sclerosis

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Abstract Fertility does not seem to be impaired to a larger extent in women with MS. However, infertility and MS might just come coincidentally together and, therefore, MS patients might undergo assisted reproductive treatment (ART). Thus, during the last years different studies evaluating the effects of ART on MS relapses have been published. All of them reported an increase in annualized relapse rate after ART. Furthermore, in a recent study clinical worsening was associated with an increase in MRI activity. Putative mechanisms involved in MS worsening after ART include: temporary interruption of disease modified therapies, stressful events associated with infertility, and immunological changes induced by hormones such as increase in pro-inflammatory cytokines and anti-MOG antibodies, as well as an increase in immune cell migration across the blood–brain-barrier. Overall, neurologists should be aware of this risk and discuss the pros and cons of the procedures with MS patients.

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1. Introduction

Multiple sclerosis (MS) is a common neurological disease in women of childbearing age. Although no systematical studies on fecundity exist, fertility does not seem to be impaired to a larger extent in women with multiple sclerosis [1]. Nevertheless, women with multiple sclerosis are more common childless, and according to a Finnish dataset undergo more common assisted reproductive techniques (ART) [2]. To which extent this is reflecting a true biological fact caused by MS is not clear yet. Several factors might contribute to a slightly impaired fertility in women with MS: hormonal alteration, including higher levels on prolactin, LH, FSH in combination with reduced estrogens is described [3,4]. Women with MS might have concomitant (autoimmune) thyroid disease (AIT) [5], which could interfere with fertility, although it is not evident if the prevalence of AIT is raised in MS patients [6]. Besides other hormonal alterations, MS itself may cause symptoms of sexual dysfunction including decreased libido, vaginal sensory disturbances and lubrication, which are known to occur also in patients in early states of the disease [7]. Psychosocial factors influencing the decision to become mother or not may be underestimated (reviewed in [8]). Finally, fertility is affected in MS patients treated with cyclophosphamide or high dose corticosteroids.

However, although the true incidence of infertility in MS patients is not known, in Western countries 10–20% of all couples suffer from infertility [8] and infertility and MS might just come coincidentally together.

After ART was introduced successfully with the delivery of Louise Brown in 1978, the methods of different hormonal approaches have grown extensively and are going to be presented shortly for a better understanding [9], as neurologists might not be familiar with these methods in detail.

2. ART and hormonal approaches in general

Different protocols, indicating the length of the protocol are generally applied. The length of the whole process is mainly dependent on the different types of downregulation.

Downregulation with gonadotropin releasing hormone (GnRH) agonist or antagonists are commonly applied to make the hypothalamic–pituitary gland axis extinct and to prevent an uncontrolled LH surge and therefore ovulation. GnRH agonists have an agonistic action on the receptor first, but block the receptor in the following permanently through a longer half-life time and lack of pulsatility of the natural hormone. Patients therefore develop hypo-estrogenic (“climacterium-like”) side-effects, flare-up and need longer down-regulation periods. This effect is observed 10 to 14 days after the use of GnRH.

The antagonists, in contrast, directly and rapidly inhibit gonadotropin release within several hours through competitive binding to pituitary GnRH receptors [10]. Interestingly,

according to a Cochrane review the use of antagonists compared with long GnRH agonist protocols seems to be associated with a large reduction in ovarian hyperstimulation syndrome (OHSS) and there was no evidence of a difference in live-birth rates.

In the following, controlled ovarian hyperstimulation is reached through different gonadotropins, mainly human menopausal gonadotrophins (HMG) and recombinant human follicle stimulating hormone (rFSH), or a combination. Clomifen, an antiestrogen is mainly used in insemination approaches. After successful stimulation controlled ovulation is induced with human choriongonadotropin (hCG), followed by progesterone to support the luteal phase [9,11]. Fertilization is mostly achieved by intrauterine insemination (IUI), in vitro fertilization (IVF), or intracytoplasmatic sperm injection (ICSI).

3. ART and multiple sclerosis existing studies

In total 5 studies on the effects of ART in MS have been published [12–16], which are summarized in the Table 1.

3.1. German experience

For our first publication [12] we followed 6 patients with relapsing remitting MS (RRMS) who underwent 14 cycles of ART retrospectively. We recruited patients nationwide.

Patients were not informed about the concrete aim of the study (possible increase of relapses after ART), to avoid reporting bias. Women were interviewed with a standardized questionnaire, which evaluated the number of MS relapses before, during and after ART over an interval of 15 months in addition to their neurological and gynecological history including the various hormonal approaches. In nine stimulation cycles they were treated with GnRH agonists and in five with GnRH antagonists (five stimulation cycles) in combination with gonadotropins, HCG followed by progesterone. We included only fertilization with IVF (five treatments/two patients) or ICSI (nine treatments/four patients). Two patients had spontaneous miscarriages during the first trimester, two patients gave birth to a healthy baby. No patient received an immunomodulatory drug during the treatment cycles of ART. The annualized relapse rate (ARR) increased significantly after ART, but was not dependent on the different hormonal approaches of downregulation.

In our second publication [13] we were able to collect data from 23 women, of whom 14 were followed prospectively, undergoing 78 cycles of ART. Again we searched participants by advertising on the website of the national German MS society (www.dmsg.de) and other MS organizations, on ART websites (www.wunschkind.de, www.kleinpuetz.de) and by informing gynecological colleagues of every clinic or practice specialized in ART in Germany to assign eligible patients to the study. Into this analysis we included patients who underwent IUI (n=32), IVF (n=15)

Table 1 Effects of assisted reproductive treatment on multiple sclerosis.

Study	Design	Sample size (n)/ ART cycles (n)	Treatment	Main results	Reference
Laplaud et al.	Retrospective case series	6/10	GnRH agonists (n=6) GnRH antagonists (n=4)	Increase in relapse rate 3 months following ART in the group treated with GnRH agonists. No evidence of increase in relapse rate in patients treated with GnRH antagonists	[15]
Michel et al.	Retrospective case series	32/70	GnRH agonists (n=48); GnRH antagonists (n=19) 3 cases lack of data	Increase in relapse rate 3 months following ART in the group treated with GnRH agonists. No evidence of increase in relapse rate in patients treated with GnRH antagonist	[16]
Hellwig et al.	Retrospective case series	6/14	GnRH agonists(n=9); GnRH antagonists (n=5)	Increase in relapse rate 3 months following ART, independent of treatment.	[13]
Hellwig et al.	Retrospective and prospective case series	23/78	GnRH agonists (n=33); GnRH antagonists (n=11)	Increase in relapse rate 3 months following ART, independent of treatment, and time interval between repetitive stimulation, in the whole cohort. Trend in prospectively followed subgroup (p=0.05).	[14]
Correale et al.	Prospective	16/26	GnRH agonists in all cases	Increase in relapse rate 3 months following ART (relative risk=7) Increase in MRI activity (relative risk=9)	[17]

ART: assisted reproductive treatment; GnRH: gonadotrophin releasing hormone.

and ICSI ($n=31$) and again observed a significant increase of the ARR in the 3 months following ART, again independent in the different hormonal approaches of downregulation ($n=39$) with GnRH agonists or antagonists. Interestingly IUI showed the strongest association of significance of the increased RR. This time we also included the different gonadotropin regimens into the analysis, but did not observe any significant difference between different gonadotropin use and the RR, neither did we detect a significant difference for the different time interval between stimulations. 14 women became pregnant, with 2 early miscarriages during the first trimester and 12 healthy babies were born. None of the women who became pregnant had a relapse. Four patients were treated with intravenous immunoglobulins and 1 with glatiramer acetate during ART. The remaining 18 patients did not receive any immunomodulatory drug during the approximately 4- to 6-week-lasting ART treatment cycles.

3.2. French experience

A first case series describing a significant increase in relapses in MS patients following in vitro fertilization (IVF) was reported in 2006 by Laplaud and co-workers [15]. By matching MS and IVF databases from the University Hospital of Nantes authors found four patients who had undergone six IVF cycles. Two additional patients who underwent four IVF cycles were also reported by the referring neurologist. In all treated patients disease modifying therapies were stopped at least one year before IVF. The treatments for IVF consisted of either gonadotrophin releasing hormone (GnRH) agonists (six cases) or GnRH antagonists (four cases), both associated with recombinant FSH. In this first report, none of the patients have relapses during the 3 months preceding IVF, while the annualized relapse rate (ARR) for the following 3 months was 2.4 ± 2.8 . A significant difference was also observed in the ARR after IVF (2.4 ± 2.8) as compared to the ARR 1 year before IVF (0.5 ± 1.2) and 1 year after IVF (0.4 ± 1.2). The increase in the ARR after IVF was observed particularly in those patients using GnRH agonists.

A second larger French systematic study was performed to confirm the preliminary results [16]. In this more recent investigation MS and IVF data were extracted from 13 French University Hospital databases or from referring neurologists. No significant differences in clinical and demographic characteristics were observed between these two groups of patients, and therefore they were pooled for further analysis. During this study 32 women with MS were undergoing 70 IVF treatments. Eighteen patients have never received any immunomodulatory or immunosuppressive drugs before their first IVF. At the time of their first IVF four patients had continued on immunoactive treatment, and 10 had stopped the treatment 19 ± 5.0 months before. In this study, in the 3 month period following IVF the ARR significantly increased to 1.60 ± 2.40 , compared with both the 3 month period before IVF (0.80 ± 1.61), and the control period 1 year before (0.68 ± 1.51).

This study demonstrated that in patients for whom the IVF procedures failed the ARR in 3 months post-IVF was significantly higher than in patients in which IVF led to pregnancy. 48 IVF procedures were performed using GnRH agonists and 19 with GnRH antagonists. The pregnancy rate

was lower in the group treated with GnRH antagonists compared with those treated with agonists (10% vs. 40%). A significant increase in ARR was observed in the 3-month period following IVF in the group treated with GnRH agonists (1.60 ± 2.29), compared with the 3 months preceding IVF (0.75 ± 1.58). In contrast, IVF performed with GnRH antagonists did not lead to significant differences in ARR.

3.3. Argentinean experience

In a recent investigation from Argentina 16 relapsing remitting MS patients who underwent 26 cycles of hormonal treatment as assisted reproductive technology (ART) were studied prospectively [17]. Neurological examinations, brain MRI and immunological testing were conducted every 3 months. Patients were eligible after 12 months of naturally seeking pregnancy without success. Disease modifying therapies were discontinued in all of them for at least 15 months. Participants received GnRH agonist, recombinant FSH for 7–10 days and vaginal progesterone as luteal phase support. Twelve of 16 patients (75%) experienced exacerbations following ART treatment. Relapses were noted in 15 of 26 cycles (58%) in the 3-month period following ART procedures. No patients had suffered relapses or shown MRI activity 9 months preceding ART. ARR was 3.28 during the 3 months following ART versus 0.42 during the non risk periods (12 months prior to the first cycle of ART, and 9 months after the final ART cycle). Thus receiving an ART cycle was associated with a 7-fold increase in risk of relapse.

Radiological activity showed on average 1.99 ± 0.3 new or enlarging T2 lesions and 1.27 ± 0.2 gadolinium (Gd) enhancing lesions at 3 months post ART, compared with 0.36 ± 0.2 T2 lesions and 0.23 ± 0.1 Gd-enhancing lesions during the remaining observation period. Thus, ART was linked to a 9-fold increase in risk of increased MRI activity.

The prospective design of the study overcomes some limitations (small sample size, none of the patients continued under disease modified therapies during ART, and all of them were treated with GnRH agonists) of the study.

4. Putative mechanisms involved in MS worsening after ART

Evidence from existing studies is arising, that hormonal stimulation during ART increases the relapse risk after unsuccessful (not pregnant) stimulation [4,9,12,15,16]. The exact reasons for this increase is not fully understood, but at least some of the studies suggest that the downregulation with GnRH agonists might account for the relapse risk [14,15]. It is well known, that hormonal levels have an influence on the disease activity of MS; this was shown in animal studies [17] and in humans as well (reviewed in [18,19]). Thus, estrogens are the most putative candidates, to decrease the disease activity being discussed, with a remarkable decrease of disease activity during pregnancy and an increase of that after birth in women with MS [20,21]. However, if changing estrogen levels alone or other hormonal factors as applied in ART are responsible for the relapse increase remains to be investigated in the future.

Beside the rapidly changing hormonal levels during ART several other factors might contribute to an increased relapse

risk. Most of the women undergoing ART stopped their disease modifying therapies before stimulation [9,12,14,16,17]. Also ART might for the affected couples represent an extremely stressful life event, normally preceded by the unfulfilled wish to become pregnant by several years. Therefore the essential hope to finally get pregnant is often linked to the beginning of ART. Although elusive, stressful life events may induce increasing disease activity in MS patients and decreased hypothalamic–pituitary–adrenal (HPA) function may play a role in the increased susceptibility on disease activity [22,23]. Interestingly while pre ART stress does not predict IVF failure, especially failure of IVF is associated with distress in the affected women [24].

4.1. Immunological mechanisms

Epidemiological and immunological studies have shown that reproductive and sex hormones play an important role in the etiology and course of different autoimmune diseases [25–27]. Thus, increases in ARR and MRI activity after ART treatment have been associated with 3 different possible mechanisms [17]: 1) GnRH effects: both GnRH and its receptor are not only expressed in the hypothalamus and pituitary gland, but also in extra pituitary tissues, including the immune system [28]. Notably, the GnRH receptor is upregulated by GnRH suggesting an autocrine function in immune cells [29]. GnRH increased proliferation of ex vivo isolated CD4+ T cells, as well as of myelin autoreactive T cells. Furthermore, the hormone increases the number of IL-8, IL-12, IFN- γ , and TGF- β secreting cells, as well as the production of endothelial growth factor (VEGF), and circulating levels of CXCL-12. 2) Estrogens effects: ART treatment was associated with a significant increase in estrogens and progesterone levels. However these levels are lower than those observed during normal pregnancy, a period during which considerable improvement of MS symptoms is reported [21]. ART treatment significantly increases the number of anti-MOG antibody-secreting cells, an effect mediated by estrogens. Moreover, purified CD19+ B cells isolated from ART-treated MS patients produced significantly higher amounts of the B-cell survival factor BAFF, and the anti-apoptotic Bcl-2 protein. 3) ART facilitates immune cell transmigration across the blood–brain-barrier by inducing 3 different molecules: IL-8, VEGF, and CXCL-12. Overall, ART involves complex and dynamic interactions between hormonal and immune factors, which could affect the course of autoimmune disease, explaining increased disease activity.

5. Concluding remarks

Overall, these data demonstrate a significant disease activity in MS patients after ART. Different approaches can be used. According to a Cochrane review the use of antagonists compared with long GnRH agonist protocols seem associated with a large reduction in OHSS and there was no evidence of a difference in live-birth rates [29]. Therefore, if possible, as more studies suggest that the treatment with GnRH agonists is harmful for women with MS, antagonists should be favored. However, the final choice of the best hormonal treatment depends on ART specialists' recommendations. Moreover, to what extent concomitant disease modified therapies showed to be administered during ART is not clear yet. Nevertheless,

neurologists should be aware of this risk, and discuss pros and cons of the procedure with MS patients.

Conflict of interest statement

The author(s) declare that there are no conflicts of interest.

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