Mesenchymal Stem Cells in Treatment of Perianal and Rectovaginal Fistulas

Abstract

Perianal fistulas are still a major challenge in everyday clinical practice. Particularly difficult fistulas, like those in Crohn’s Disease, often require multiple aggressive surgical interventions, with high risk of sphincter damage and incontinence. Intensive trials have been conducted for over 10 years on the treatment of difficult fistulas with mesenchymal stem cells (MSC). This paper presents a short review of basic mechanisms of mesenchymal stem cells in healing of damaged tissues and results of major clinical trials. MSC in perianal fistulas, regardless of research protocol has a 70% healing rate in recurrent fistulas and in fistulas in Crohn’s Disease. Further research is needed to precisely define the indications for MSC-based treatment of tissue damage, and particularly perianal and recto-vaginal fistulas.

Keywords: Fistula-in-ano; Recurrent fistula; Complex fistula; Crohn disease; Adipose-derived mesenchymal stem cells; Clinical trials

Introduction

Perianal and rectovaginal fistulas are pathological connections between the rectum and adjacent structures in the rectal region. About 80% of perianal fistulas are of cryptoglandular origin the remaining 20% develop in Crohn’s Disease, after trauma, or have iatrogenic etiology, etc.

Standard in perianal fistula treatment are cutting (fistulotomy, fistsectomy) or non-cutting (seton) surgical procedures, to remove granulation tissue, necrosis and translocated mucosal cells. The success rate of primary surgical treatment of perianal fistulas reaches about 70% [1]. In the remaining cases, there is no healing of the fistula after the procedure, or the fistula recurs after initial healing. Fistula recurrence rate in Crohn’s Disease is relatively high (34%) [2] which is due to constant inflammation of the intestines and soft tissues. Therefore surgical approach is preceded by immunosuppressive treatment, including anti-TNFα, which is necessary for proper soft tissues healing.

Regardless of the fistula etiology, the disease greatly reduces the patients’ quality of life both due to recurrence and incontinence [3]. The metaanalysis by Blumetti et al. [4] shows, that aggressive surgical approach (eg. fistulotomy) has a greater chance of successful healing, at the same time increasing the risk of fecal incontinence. Therefore, there is a constant search for better alternatives for fistula treatment.

In 2003 Prof. Garcia-Olmo made the first attempt to treat a rectovaginal fistula in a 33-year old female patient with Crohn’s Disease, using the mesenchymal stem cells derived from the fatty tissue of the patient (Adipose-derived mesenchymal Stem Cells) [5]. By implantation of about 9mln of autologous stem cells he achieved complete closure and lack of recurrence of the fistula. From that time, many approaches to soft tissue treatment with MSC were made. This paper presents a short review of clinical research on the MSC treatment of perianal and rectovaginal fistulas so far.

Discussion

Mechanism of action

Mesenchymal stem cells exist in the perivascular niche of almost all tissues [6], however, the most useful source of these cells are bone marrow, adipose tissue and umbilical cord [7]. In classic approach, MSC are pluripotent cells that support the natural healing process by increasing the overall pool of cells (self-renew) and differentiation in the direction of mesenchymal cells (fibroblasts producing collagen, adipocytes, myocytes, osteocytes, etc.) [8]. In addition, MSC modulate the healing process by stimulating dermal fibroblasts to divide, migrate toward the injury, and the expression of collagen [9]. In hypoxic conditions which occur on the edges of tissue damage, MSC secrete growth factors, such as vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), hepatocyte growth factor (HGF), insulin-like growth factor, and other [10] therefore inducing revascularisation [11]. Mesenchymal stem cells also possess immunosuppressive properties [12,13]. In a mouse model of inflammatory bowel disease MSC suppress Th1 lymphocytes, which leads to a reduction in clinical and histological symptoms of inflammatory bowel disease [13]. Multifactorial mechanism of action of MSC responds to the need for proper healing of soft tissue, but we still do not know which of the effects described by basic science have clinical significance. What’s more, there are no
direct studies on the characteristic of the microenvironment of the damaged tissue, and in particular its “weak spots” in relation to the properties of the MSC, which could allow the development of better treatment strategies.

**Clinical outputs**

Since 2005 at least 9 clinical trials with MSC treatment of the perianal fistula were conducted [14-24]. The summary of prospective studies are shown in Table 1. In all studies, 225 cell-treated patients were completed the protocol, 107 were healed in 8-24 weeks of follow-up. In all trial administration of the cells was combined with surgical closure of the fistula (one-stage procedure). Patients received 9 to 139 million cells suspended in fibrin glue [15,16,23] or in saline solution [15,19-22]. The cells were administered into the walls of the fistula and into its lumen.

**Table 1:** Summary of clinical trials and compassionate-use programmes with mesenchymal stem cells therapy in treatment of fistulas.

| Published Clinical Trial | Etiology of Fistula | Type and Amount of Cells | N / n | H / h |
|--------------------------|---------------------|--------------------------|-------|------|
| 1 Garcia-Olmo et al. [14]| Crohn               | Autogenic ASC, 3-30 mln  | 9     | 6    |
| 2 Garcia-Olmo et al. [23]| both                | Autogenic ASC, 20 mln    | 17/7* | 12/5*|
| 3 Ciccocioppo et al. [18]| Crohn               | Autogenic MSC, 54-139 mln| 10    | 7    |
| 4 de la Portilla et al. [20]| Crohn             | Allogenic ASC, 20-40 mln | 16    | 9    |
| 5 Herreros et al. [15]  | non-Crohn           | Autogenic ASC, 20 mln    | 124   | 51   |

Results in multicenter trial excluded a pioneering center

Results in the pioneering center (Garcia-Olmo)

|                |                |                |
|----------------|----------------|----------------|
|                |                |                |

N: number of cell-treated patients who completed the protocol, H – number of cell-treated patients with healed fistula in 8-24 weeks follow-up; * - Number of non-Crohn/Crohn patients; ** - Compassionate-use programme.

It is difficult to determine the efficacy of MSC / ASC as each of these studies had a slightly different protocol. For this reason, the total numbers of patients were counted - patients who received mesenchymal stem cells (isolated from bone marrow and adipose tissue, auto and allogeneic, alone or with fibrin glue) in clinical trials or registered treatment programs (compassionate use). Only prospective studies were taken into account, and the endpoint (ie. "healed") was considered as clinical healing in the period from 8 to 24 weeks after administration of the cells (the status of "partially healed" was classified as "not healed" unless the healing was achieved up to 12 months after the cells were administered).

Among 225 (or 154) patients who received the MSC or ASC status of "healed" were reported in 138 (54%) (or 103 (67%) - excluding non-pioneering center form the largest clinical trial conducted by Herreros et al. [15]) cases. The healing rate in non-Crohn fistulas were 45% (n=148 cell-treated patients) and (or 68% (n=47) excluding non-pioneering centers from Herreros et al.). We decided to separately present a results from Herreros et al. because of only in the pioneering center outcomes of cell-treatment patients were significantly better than in the rest centers (the healing rate in 19 centers was 50% (n=124) but in pioneering center reached 70% (n=23)). This fact suggest major influence of experience in cell-based products management on the final results.

In the case of Crohn’s Disease overall healing rate was 66% (n=107) which include Lee et al. [16] where reported healing rates even up to 82%. There are several different strategies reported in the literature (use of auto or allogeneic cells, the administration of from 9 to 139 million cells, the administration of cells in fibrin glue, or alone, administration of cells at once or sequentially until healed, and various ways to provide the fistula before administration of the cells). Therefore it is very difficult to compare results between studies. However, achieved results show, that there is a positive effect of the MSC on soft tissue healing.

In the presented studies some of AE (adverse effects) were reported but only in Herreros et al. [15] there were reporting in systematic condition. The most frequent adverse effects [15] were a proctalgia (43%) (that was also reported by our team (data not shown)) and abscess drainage (22.4%). Pain, perianal abscesses, pyrexia, swelling and pruritus were also reported in around 10%
or less patients who were included in protocol [15]. Frequency of these effects were no statistically differences within groups which received the cells or fibrin glue. In longer observation the most probability AE, directly associated with cell transplantation, uncontrolled cellular growth were no reported in Herreros et al. and another publication as well.

Lack of differences between groups in occurring of the AE and lack of the uncontrolled cellular growth suggest that reported adverse events were more related to the act of administration an ascetical products risky perianal location than the products were a mesenchymal stem cells. It must be stressed here that cells cannot be sterilized as typical pharmaceutics (ex. gamma sterilization)

In clinical trials so far, the healing rates of MSC fistula treatment was about 70%. Pleiotropic effect of action of the stem cells seems to be particularly beneficial in the process of perianal fistula healing, but we still don’t know whether the cells are permanently built into the tissue, and what clinical value have the properties of the cells we described. From that reason there is still grate necessity to explore a mechanism of soft tissue healing driven by autogenic as well as allogenic MSC. Developing of spin-labeling technics and magnetic resonance imaging seems to be useful tools in investigation of “fate of cells” after implantation and can be directly implemented in clinical trials protocols. The explanation of these aspects will help to develop better strategies to introduce the cells and thus contribute to the definition of precise indications for such treatment.

The cell-based therapies that are still in investigational phase require from all scientific and medical society to treat the patients only in restricted conditions of registered clinical trials or compassionate use programs to decrease chance of serious adverse effects. In over 10-year long research on safety and effectiveness of the mesenchymal stem cells in treatment of soft tissues give hope for a new, safer therapeutic option, especially useful in difficult fistula cases but still need more effort to better understand a nature and consequences of cell-based products.

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