Alimentary Pharmacology & Therapeutics

WILEY

AP&T Alimentary Pharmacology & Therapeutics

ORIGINAL ARTICLE OPEN ACCESS

Evaluation of the Effectiveness and Safety of Mesenchymal Stem Cell Treatment in Fistulising Crohn's Disease: An International Real-Life Retrospective Multicentre Cohort Study

Péter Bacsur[1,2] | Daniel Shaham[3] | Zuzana Serclova[4,5] | Tamás Resál[1] | Bernadett Farkas[1] | Patrícia Sarlós[6] | Pál Miheller[7] | Nitsan Maharshak[3] | Meir Zemel[8] | Ariella Bar-Gil Shitrit[9] | Shlomo Yellinek[10] | Anita Bálint[1] | Anna Fábián[1] | Renáta Bor[1] | Zsófia Bősze[1] | Emese Iványi[1] | Zoltán Szepes[1] | Klaudia Farkas[1,2] | Illés Tóth[11] | György Lázár[11] | Katerina Vlkova[4] | Aneta Tremerova[4,5] | Petra Zuskova[4,5] | Szabolcs Ábrahám[11] | Tamás Molnár[1]

[1]Department of Medicine, Albert Szent-Györgyi Medical School, University of Szeged, Szeged, Hungary | [2]HCEMM-USZ Translational Colorectal Research Group, Szeged, Hungary | [3]IBD Unit, Tel Aviv Medical Center, Affiliated to the Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel | [4]Surgical Department, Clinical IBD Center ISCARE, Prague, Czech Republic | [5]Surgical Department, University Hospital Kralovske Vinohrady, Prague, Czech Republic | [6]Division of Gastroenterology, First Department of Medicine, Medical School, University of Pécs, Pécs, Hungary | [7]Department of Surgery, Transplantation and Gastroenterology, Faculty of Medicine, Semmelweis University, Budapest, Hungary | [8]Colorectal Unit, Surgical Division, Tel Aviv Medical Center, Affiliated to the Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel | [9]Digestive Diseases Institute–IBD-MOM Unit, Shaare Zedek Medical Center, Hebrew University, Jerusalem, Israel | [10]Department of General Surgery, Shaare Zedek Medical Center, The Hebrew University School of Medicine, Jerusalem, Israel | [11]Department of Surgery, Albert Szent-Györgyi Medical School, University of Szeged, Szeged, Hungary

Correspondence: Szabolcs Ábrahám (abraham.szabolcs@med.u-szeged.hu) | Tamás Molnár (molnar.tamas@med.u-szeged.hu)

Received: 23 June 2024 | **Revised:** 15 July 2024 | **Accepted:** 13 October 2024

Handling Editor: Cynthia Seow

Funding: This work was supported by research grants from the National Research, Development and Innovation Office (grant IDs: 125377 and 143549 to TM, 129266 to AB, 134863 to FK) and the New National Excellence Programme of the Ministry of Human Capacities (UNKP-23-3-SZTE-268 and EKÖP-368 to BP, UNKP-22-4 -SZTE-293 to BR, UNKP-23-3-SZTE-317 to RT, UNKP-22-4-SZTE-296 to FA) and by a Janos Bolyai Research Grant (BO/00598/19/5) and a Géza Hetényi Research Grant (to FK, RM and BA) from the Faculty of Medicine, University of Szeged. The project has received funding from the EU's Horizon 2020 research and innovation program under grant agreement No. 739593.

Keywords: Crohn's disease | darvadstrocel | mesenchymal stem cell treatment | perianal disease

ABSTRACT

Background: Perianal fistulas of Crohn's disease (CD) create a significant burden on patients' lives. However, the efficacy and safety of adipose-derived mesenchymal stem cell treatment are contradicting, and real-world evidence is lacking.

Aims: To examine the usability of darvadstrocel therapy in managing perianal CD.

Methods: We enrolled patients with CD and perianal fistulas in this retrospective multicenter study. The primary outcome was perianal clinical remission (defined as all treated fistulas closed) at weeks 26 and 52. Secondary outcomes were clinical response

Abbreviations: AE, adverse events; CD, Crohn's disease; CRP, C-reactive protein; IBD, inflammatory bowel disease; IQR, interquartile range; MSC, mesenchymal stem cell; PDAI, Perianal Disease Activity Index; PFCD, perianal fistulising Crohn's disease.

[Correction added on 20 November 2024, after first online publication: The author's name Ariella Shitrit has been corrected as Ariella Bar-Gil Shitrit and Co-author Shlomo Yellinek has been added to in this version.]

Péter Bacsur, Daniel Shaham Zuzana Serclova share first authorship.

Szabolcs Ábrahám and Tamás Molnár share senior co-authorship.

rates (≥1 fistulas closed), perianal activity (PDAI), patient satisfaction, and adverse events. Data were recorded at baseline and weeks 12, 26 and 52. Prediction of primary outcomes was performed by logistic regression.

Results: Overall, among 223 patients (male/female ratio: 0.48), perianal clinical remission was achieved in 78.2% and 62.3% until weeks 26 and 52. Baseline PDAI score (OR 0.75), number of fistulas (OR 0.28) and the number of weeks after preparation for surgery (OR 0.98) were associated with treatment failure. The clinical response rates were 84.8% and 79.8% at weeks 26 and 52. Improvement of subjective perianal symptoms was achieved in 77.8% and 78.4% of patients, respectively. Adverse events occurred in 13.5% of patients; perianal abscesses and proctalgia were the most frequently reported.

Conclusion: Effectiveness data were higher than in clinical trials. The safety profile was reassuring, and patients' satisfaction was high. Appropriate patient selection, fistula preparation and expertise may help to achieve treatment success.

1 | Introduction

Crohn's disease (CD) is a chronic, immune-mediated, idiopathic inflammatory condition of the gastrointestinal tract characterised by ulcerative lesions, strictures, penetrating complications and perianal fistulas. This is in addition to several extraintestinal manifestations, resulting in a high disease burden on patients' lives and healthcare services [1]. In particular, 10%–26% of CD patients exhibit perianal fistulising complications during the course of the disease (perianal fistulising Crohn's disease, PFCD) [2–4]. In this case, patients with perianal disease suffer from fistulas between the anal canal or rectal segment and the perianal skin (which are often associated with fluid collections or abscesses), resulting in an extremely difficult-to-treat disease [3, 5]. Meanwhile, inflammatory mechanisms of CD result in the upregulation of matrix-metalloproteases and the weakening of the epithelial cell layer, guided by inflammatory cytokines and leading to epithelial–mesenchymal transition and fistula formation [5].

PFCD has a high disease burden, creating a significant challenge for the healthcare system. Thus, the quality of life of CD patients is generally poor, based on their luminal complaints and disability due to perianal symptoms [5, 6].

The management of PFCD requires a multidisciplinary approach involving colorectal surgeons, gastroenterologists and radiologists. According to the European Crohn's and Colitis Organisation's guidelines, complete fistula closure may be achieved through anti-TNF drugs (infliximab and adalimumab), even though evidence supporting antibiotics or systemic immunosuppressants as monotherapy remains limited [7]. In regard to the ineffectiveness of pharmacotherapy, several surgical approaches have been suggested. In selected cases, fistula closure with advancement flaps, ligation of the intersphincteric fistula tracts, fibrin glue and fistula plugs may be considered, while seton placement has been suggested [7, 8]. Despite the high number of potential surgical techniques, the refractory disease rate and related complications remain high [9].

Recently, the efficacy and safety of adipose-derived allogenic mesenchymal stem cell (MSC) treatment (darvadstrocel) for PFCD have been raised by the ADMIRE-CD placebo-controlled clinical trial [10]. Darvadstrocel is administered as an intralesional injection at the site of the perianal fistula after a preparation including rigorous curettage and seton placement. During the procedure, 120 million cells (5 million cells/mL) should be administered. The exact mechanism of action is unclear; however, the immunomodulatory and anti-inflammatory effects are presumed by inhibition of the proliferation and activation of lymphocytes via anti-inflammatory cytokines. This regulation results in mucosal healing and repairing of the fistula tract [10, 11]. In ADMIRE-CD trial, 51.5% of the treated patients showed combined clinical and radiological remission, compared to 35.6% in the placebo group (only surgical intervention) at week 24. Meanwhile, the 1-year outcome was even better (80% vs. 47%). Although the safety profile was reassuring, anal abscesses and proctalgia appeared in high proportions in both groups [10–12]. Moreover, the preliminary results of the ADMIRE CD-II randomised-controlled trial did not show superiority of darvadstrocel over the placebo [13]. As for the meta-analyses of the randomised trials, they suggest the efficacy and safety of MSC treatment for CD patients [9, 14]. However, the few real-world studies published included a low number of treated patients [15–17].

Therefore, the overall objective of this study was to assess the long-term effectiveness and safety of adipose-derived MSC treatment for PFCD in a real-world, multicenter setting.

2 | Materials And Methods

2.1 | Study Design and Participants

This retrospective multicenter cohort study was conducted in six tertiary inflammatory bowel disease (IBD) centres in three countries, including Israel, the Czech Republic, and Hungary. A sample of adult (age ≥ 18 years) CD patients who underwent MSC treatment (darvadstrocel) between January 1, 2019 and September 30, 2023, due to PFCD (high transsphincteric or suprasphincteric localisation, ≤ 2 internal and ≤ 3 external openings) refractory to traditional PFCD treatment (ineffectiveness of surgical fistula treatment and pharmacotherapy with antibiotics or immunomodulators or anti-TNFs within 6 weeks before the baseline) and with stable pharmacotherapy at least 3 months before inclusion were enrolled and examined. In this case, the baseline was defined as the day of the application of the MSC treatment, while follow-ups were conducted after 6–12 months. Patients with moderate to severe luminal activity (CDAI ≥ 220 [18]), active CD of the rectum, > 2 cm intra-abdominal abscess, concomitant rectovaginal fistula or deviating stoma, rectal or anal stricture and surgical intervention (except for surgical/seton drainage regarding perianal fistulas) were excluded from further analysis. Patients with multiple MSC treatments during the follow-up were also excluded. The reporting of this study conforms to the STROBE statement [19]. All patients provided their written informed consent to regular care.

2.2 | Administration of Darvadstrocel

Darvadstrocel was administered as an intralesional injection at the site of the perianal fistula after a preparation including rigorous curettage and seton placement according to the manufacturer's label. After the closure of the internal openings, approximately 120 million cells (5 million cells/mL) were administered during the procedure. All the participating centres were involved in the ADMIRE study and surgeons have treated subjects guided by ADMIRE experience. Adherence to the ADMIRE protocol was not a condition for participation in the study.

2.3 | Data Collection

Data were collected into a uniform database at each centre (based on the medical history available in the medical record systems) and anonymously transferred to the coordinating centre (the University of Szeged). The data regarding the preparation for surgery, including curettage of the fistula tracts and seton placement/revision before the baseline was obtained. Baseline demographic data (age, sex, date of birth, age at diagnosis), clinical data (disease localisation by Montreal classification [20], previous medications, current medications, previous surgery), and MSC surgery data were recorded at the baseline, while details of perianal clinical activity were measured at the baseline and weeks 12, 26 and 52 (if available). Due to the study design, data collection was allowed to be performed within 1 week of the reference timepoint.

Moreover, Parks's fistula classification was used [21], while luminal clinical activity was measured by the Crohn's Disease Activity Index (CDAI) [18], endoscopic activity was measured by the Simple Endoscopic Score for Crohn's Disease (SES-CD) [22], and perianal complaints were objectivised by the Perianal Disease Activity Index (PDAI) [23]. The C-reactive protein (CRP) level of the patients and modified Van Assche index [24] (if MRI scans were performed) were recorded, while the patients' satisfaction levels and improvement of perianal symptoms were obtained at weeks 12, 26 and 52 by using a 3-point Likert scale (i.e., improved, not altered, worsened) [25]. Time-dependent variables, such as adverse events (AEs), the reopening of a treated and closed fistula, and the occurrence of anal stenosis after treatment, were also recorded. Disease activity characteristics were recorded using prospectively followed patient records. Data collection was performed between February 1 and April 30, 2024.

2.4 | Outcome Measurements and Definitions

The primary co-outcomes were the perianal clinical remission at weeks 26 and 52, defined by the closure of all external openings of the treated fistulas that were draining at baseline and assessed by a gastroenterologist/colorectal surgeon at regular visits. The secondary co-outcomes were the clinical response rates at weeks 26 and 52 (defined by the closure of at least one external opening of the treated fistulas and assessed by a gastroenterologist/colorectal surgeon at regular visits), whereas treatment failure was defined if no treated fistula tract was closed. The tertiary co-outcomes were the perianal clinical activity rates at weeks 12, 26, and 52 (defined by the PDAI score of >4) [26], the patients' satisfaction levels, and AEs requiring hospitalisation (presented as incidence rates as frequency/100 patient-years). A sub-group analysis evaluating treatment experience was also conducted on the primary outcomes. Patients were stratified according to the overall experience of the treating centre and >100 treatments with darvadstrocel was used as cut-off (high experience and low experience).

2.5 | Statistical Considerations

In this study, a power analysis was not performed, since it analysed all eligible patients, and there was no control group, making a sample size calculation unfeasible. The descriptive statistics included the mean and standard deviation or the median and interquartile range (IQR) of the continuous variables, and the numbers and percentages of the categorical variables. Normality was tested by using visual interpretations (histograms and quantile–quantile plots). After checking the assumptions, the groups described with the categorical variables were compared by using a chi-square test or Fisher's exact test, while the continuous variables were compared with independent samples *t*-tests. Potential covariates and confounders associated with the primary and secondary effectiveness outcomes were analysed via univariable and multivariable logistic regression models. Specifically, the variables in the univariable analysis with a $p<0.15$ were fitted and included in the multivariable analysis. Final multivariable models were obtained by forward selection using likelihood ratios. Moreover, the goodness of fit was analysed by the Hosmer–Lemeshow test and the Nagelkerke R^2 was presented. Kaplan–Meier survival curves with a log-rank test were used to determine the characteristics of AEs over time. Cox proportional hazard models were also created to investigate the covariates associated with AEs. We performed a complete case analysis to reduce bias and to achieve the most accurate description of the cohort. Bonferroni correction was used to reduce multiple comparisons bias. A two-sided $p<0.05$ indicated a statistically significant difference, while 95% confidence intervals (95% CI) were communicated. Statistical analysis was performed by using IBM SPSS software (Windows, Version 29.0, IBM Corp., Armonk, NY).

3 | Results

3.1 | Patient Characteristics

Overall, the data of 223 CD patients (male/female ratio: 0.48) was analysed, with a median follow-up duration of (51.9 weeks [IQR 48–53]). Among the patients, 69% (154/223), 22% (48/223) and 9% (21/223) were from the Czech Republic, Israel and Hungary, respectively. At inclusion, the median age was 39 years (IQR 33–49), and patients suffered from CD with a median duration of 13 years (IQR 7–21). Approximately three-quarters (75.7%) of the patients did not smoke at inclusion. In total, 69.1% of the patients were treated in high-volume tertiary centres. A flowchart of the enrolled patients is presented in Figure 1.

The most common localisation was ileocolonic (43.5%), while 40.7% of the patients had undergone prior bowel resection. The

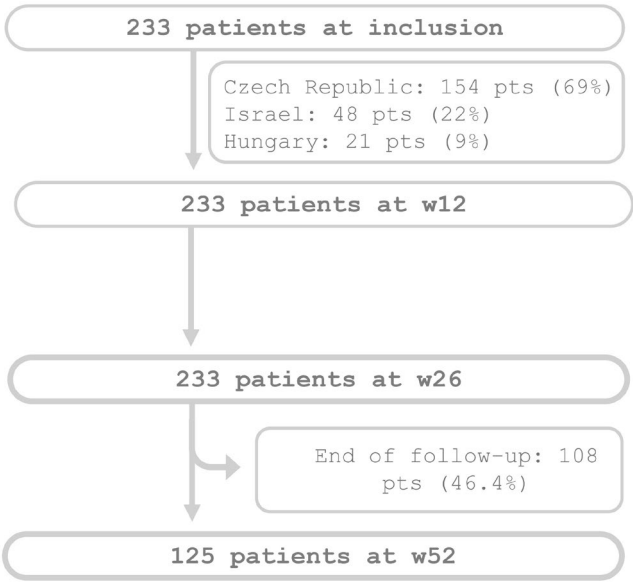

FIGURE 1 | Flowchart of the patients' enrollment and follow-ups. Pts, patients.

majority of the cohort (91.5%) were in clinical remission at the baseline. In addition, an immunomodulator was applied in 41.7% of the patients, with more than three quarters on anti-TNF treatment. Meanwhile, 57.4% of the patients had only one fistula tract at enrollment, with the majority transsphincteric fistulas. However, branching tracts were registered in 13.9% of the cases. Finally, more than half of the patients showed a PDAI score of >4 (57.8%). Further details of the baseline clinical and demographic characteristics are presented in Table 1.

3.2 | Effectiveness Assessment

In total, the data of 100% and 82.1% of the patients were available at weeks 26 and 52, respectively. Perianal clinical remission was achieved in 72.2% (161/223) and 62.3% (114/183) of the patients by weeks 26 and 52, respectively. The clinical response rates were 84.8% (189/223) and 79.8% (146/183) at weeks 26 and 52. Neither the clinical remission rates ($X^2=0.143$; $p=0.71$) nor the clinical response rates ($X^2=1.722$; $p=0.19$) changed until week 52, respectively. Figure 2 presents the data regarding the proportion of patients achieving perianal clinical remission and response at weeks 26 and 52.

At the baseline, 57.8% (129/223) of the patients had perianal disease with PDAI score > 4. This rate decreased to 20.6% (41/199, $X^2=60.638$, $p<0.001$), 17.2% (34/198, $X^2=32.596$, $p<0.001$) and 19.6% (32/163, $X^2=56.566$, $p<0.001$) at weeks 12, 26 and 52, respectively. Among week 26 responder patients, a similar decrease was found. However, the rate of perianal disease with PDAI score > 4 did not decrease among the patients with treatment failure at week 26. Figure 3 shows the proportion of patients with perianal disease with PDAI score > 4 at the baseline and weeks 12, 26 and 52, according to the treatment response at week 26.

It is important to note that 81.2% (164/202), 77.8% (161/207) and 78.4% (131/167) of the patients experienced improvement in subjective symptoms, whereas 16.8% (34/202), 7.7% (16/207) and 7.2% (12/167) did not experience any change until weeks 12, 26 and 52, respectively. Figure 4 presents the patients' experiences.

3.3 | Prediction of Treatment Efficacy

Week 52 clinical remission was negatively associated with antibiotic usage at MSC injection (OR 0.159, 95% CI 0.066–0.382), the level of the baseline PDAI score (OR 0.753, 95% CI 0.659–0.859), and the presence of branching fistulas (OR 0.417, 95% CI 0.172–1.011), while the week 26 remission outcome was coupled with the shorter period between preparation and transplantation surgery (OR 0.976, 95% CI 0.965–0.986) and the low number of fistulas at the baseline (OR 0.278, 95% CI 0.127–0.609). Achieving primary outcomes was observed more frequently in the centres with high-volume treatments in week 26 (82.5% vs. 49.3%, $X^2=26.153$; $p<0.001$) and week 52 (72.0% vs. 41.4%, $X^2=15.815$; $p<0.001$).

Clinical response rates at week 26 were associated with ileocolonic disease localisation (OR 3.482, 95% CI 1.228–9.879), bowel resection history (OR 2.998, 95% CI 1.123–8.006), a high number of external fistula openings (OR 7.997, 95% CI 2.223–28.767), and prior usage of advanced treatments (OR 0.140, 95% CI 0.052–0.378).

Treatment failure at week 52 was associated with female sex (OR 0.392, 95% CI 0.157–0.980) and with longer periods between preparation and transplantation surgery (OR 0.992, 95% CI 0.984–0.999). Table 2 presents the results of the multivariable regression after model selection, while Tables S1–S4 detail the univariable and multivariable regression models on each outcome.

3.4 | Safety Analysis

Overall, 204 patient-years were recorded, of which 13.5% (14.7/100 PY) suffered from AEs requiring hospitalisation during the follow-ups. Adverse events not requiring hospitalisation were not observed. The median time until occurrence of an AE was 51.1 weeks (IQR 27.9–52.9) after treatment. Meanwhile, the characteristics of AEs were similar between the groups, stratified by achieving perianal clinical remission at week 26 ($X^2=0.522$, $p=0.47$). The occurrence of AEs was also associated with a higher baseline PDAI score (HR 1.148, 95% CI 1.021–1.290), while active smoking (HR 0.227, 0.054–0.957) was coupled with decreased risk. Table 3 presents further details of the AEs, while Figure 5 displays a survival curve of the time to AEs, according to perianal clinical remission at week 26. Additionally, Table S5 lists the descriptions of the Cox models.

4 | Discussion

In this study, the effectiveness and safety of MSC treatment in fistulising CD was evaluated in a large multicenter cohort study, including 223 CD patients from tertiary IBD centres in a retrospective setting. The perianal clinical remission rates at weeks 26 and 52 were 72.2% and 62.3%, respectively, and were negatively associated with antibiotic usage, the baseline PDAI

TABLE 1 | Baseline demographic and clinical characteristics of enrolled patients.

Variables	CD ($n = 223$)
Follow-up duration, weeks, median (IQR)	51.9 (48–53)
Sex, male (%)	107 (48)
Age at inclusion, years, median (IQR)	39.0 (33–49)
Smoking habits at inclusion, *n* (%)	
Never smoking	137 (61.4)
Active smoker	49 (22.0)
Former smoker	32 (14.3)
Missing data	5 (2.2)
CD disease duration at inclusion, years, median (IQR)	13 (7–21)
Prior bowel resection, *n* (%)	
Small bowel segment	5 (2.2)
Large bowel segment	17 (7.6)
Ileocoecal	69 (30.9)
Disease localisation, *n* (%)[a]	
Ileal	71 (31.8)
Colonic	55 (24.7)
Ileocolonic	97 (43.5)
Upper GI involvement	11 (4.9)
Extraintestinal manifestations, *n* (%)	
Arthropathy	41 (18.4)
Skin disease	4 (1.8)
Eye disease	3 (1.3)
Hepatic disease	0 (0)
Disease activity	
CDAI, mean (±SD)	51.1 (53.1)
PDAI, > 4, *n* (%)	129 (57.8)
CRP, mg/L, mean (±SD)	14.7 (9.5)
SES-CD, mean (±SD)	0.5 (1.1)
Prior advanced treatment, *n* (%)	
1 agent	63 (28.3)
2 agents	38 (17.0)
≥ 3 agents	15 (6.7)
Anti-TNF	95 (42.6)
Conventional treatment at baseline, *n* (%)	
Oral 5-ASA	23 (10.3)
Topical 5-ASA	3 (1.3)
Budesonide	1 (0.4)
Azathioprine	93 (41.7)
Systemic corticosteroid	10 (4.5)
Advanced treatment at baseline, *n* (%)	
Adalimumab	57 (25.6)
Infliximab	108 (48.4)
Golimumab	1 (0.4)
Certolizumab	1 (0.4)
Vedolizumab	12 (5.4)
Natalizumab	2 (9.0)
Ustekinumab	35 (15.7)
Guselkumab	1 (0.4)
Upadacitinib	0 (0)
Ozanimod	1 (0.4)
Description of fistulas, *n* (%)	
Number of fistulas	
1	128 (57.4)
2	81 (36.3)
3	14 (6.3)
Number of external openings	
1	127 (57.0)
2	79 (35.4)
3	17 (7.6)
Number of internal openings	
1	177 (79.4)
2	46 (20.6)
Branching fistula present	31 (13.9)
Classification of fistulas, *n* (%)[b]	
Intersphincteric is present	33 (14.8)
Transsphincteric is present	177 (79.4)
Extrasphincteric is present	13 (5.8)
Suprasphincteric is present	18 (8.1)
MRI was performed during FU	24 (10.8)

[a]Montreal classification.
[b]Parks classification.
Abbreviations: CD, Crohn's disease; CDAI, Crohn's Disease Activity Index; CRP, C-reactive protein; FU, follow-up; IQR, interquartile range; MRI, magnetic resonance imaging; *n*, number of patients; PDAI, Perianal Disease Activity Index; SD, standard deviation of mean; SES-CD, Simple Endoscopic Severity of Crohn's Disease; TNF, tumour necrosis factor-alpha.

score, the interval between preparation and surgery, and with branching of the fistulas. AEs occurred in 13.5% of the patients, including perianal abscesses and pain. Instances of AEs were coupled with elevated baseline PDAI scores.

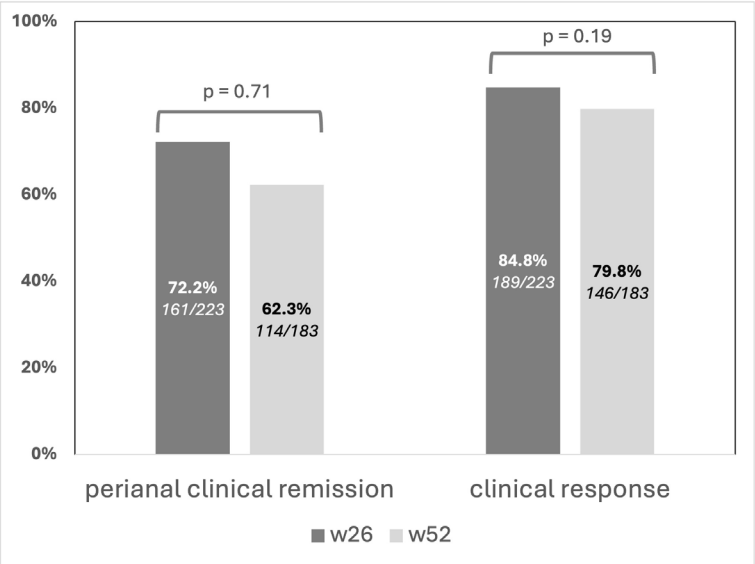

FIGURE 2 | The proportion of patients achieving perianal clinical remission and response at weeks 26 and 52. No significant difference was observed between the timepoints and endpoints. P, significance level.

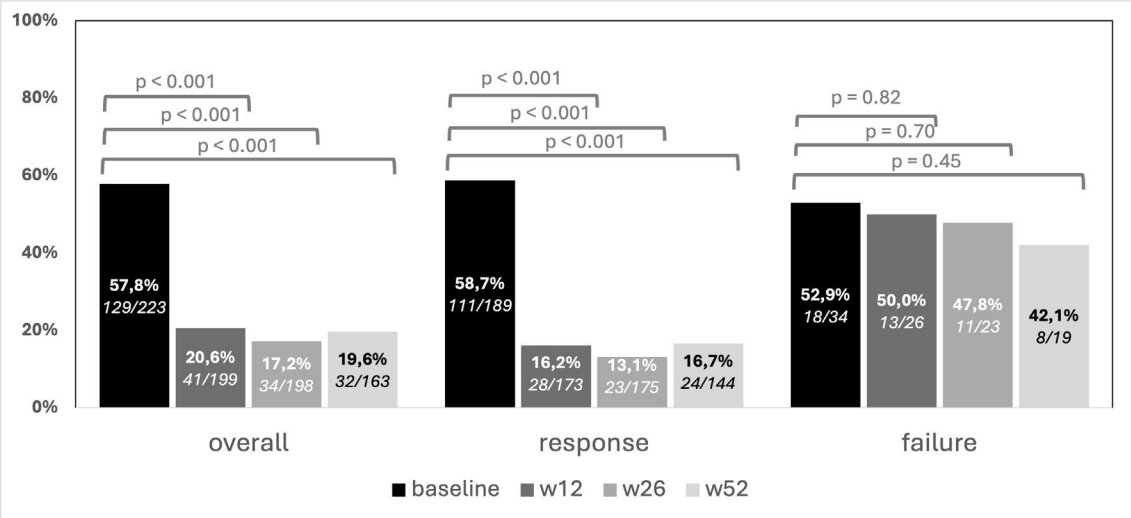

FIGURE 3 | The proportion of patients with >4 PDAI score for perianal disease during the follow-ups, stratified by achieving perianal clinical remission at week 26. Among the patients, perianal disease immediately decreased after treatment. P, significance level.

Several clinical trials were conducted to evaluate the efficacy and safety of MSC treatment among the CD patients with perianal fistulas. The ADMIRE CD placebo-controlled pivotal clinical trial included 212 CD patients. Among the 107 patients who were treated with darvadstrocel, a higher proportion (55%) achieved clinical remission (closure of all treated external openings), compared to the placebo (43%) at week 24 [10]. Interestingly, a long-term extension study of the ADMIRE CD trial reported an 80% clinical remission rate at week 52 [12] in the treatment group.

In a Phase II study, fibrin glue was applied to close the fistula tracts after MSC treatment. Although superiority was reported (compared to fibrin glue alone), the retrospective evaluation indicated that a high proportion of patients did not achieve sustained closure of the tracts during the follow-ups (11/21 vs. 10/13). However, the population was not restricted to CD patients [27, 28]. Additionally, meta-analyses and systematic reviews evaluated the overall efficacy of the darvadstrocel treatment, which showed superiority in fistula healing, compared to the placebo [14, 29, 30]. Overall, 72.2% of the patients were in clinical remission at week 26, while this proportion decreased to 62.3% at week 52. Although the clinical response rate was 84.8% at week 26, which was higher than that in the ADMIRE population (69% vs. 55%), the outcome was stricter (closure of at least 50% of the treated tracts). It should be highlighted, that the similarity of outcome results in our study are presumably due to the similarity of outcome definitions.

Nevertheless, the ADMIRE-II randomised-controlled trial displayed the nonsuperiority of MSC treatment, compared to the placebo (combined remission rate at week 24: 48.8% vs. 46.3%). Although this questions the usefulness of the procedure, a full publication is not yet available. Meanwhile, even though the clinical remission rates were lower in the ADMIRE-II trial, higher effectiveness was observed in a

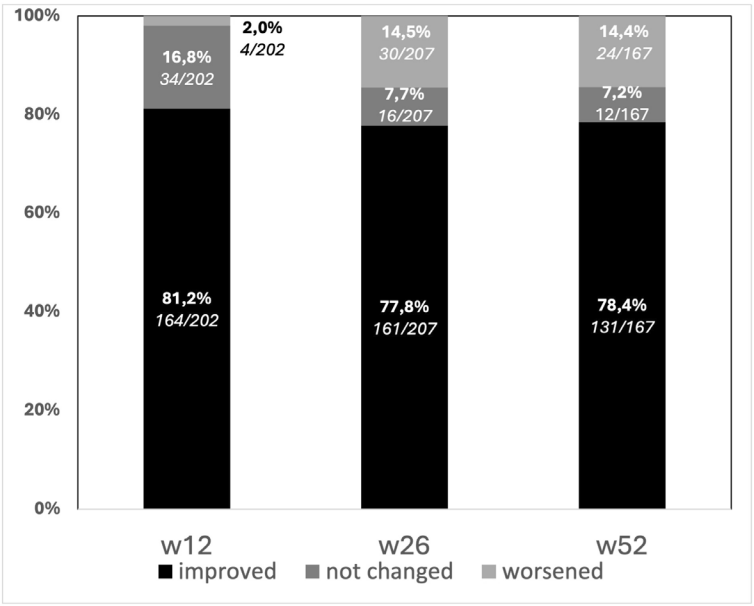

FIGURE 4 | The change in patients' symptoms during the follow-ups, measured on a 3-point Likert scale. The majority of the treated patients experienced an improvement in perianal symptoms. P, significance level.

TABLE 2 | Multivariable logistic regression model to predict treatment effectiveness.

Outcomes	Variables	Sig.	OR	95% CI	
Clinical remission at w26	Antibiotic use at injection	< 0.001	0.159	0.066	0.382
	PDAI	< 0.001	0.753	0.659	0.859
	Branching fistula	0.053	0.417	0.172	1.011
Clinical remission at w52	Time between preparation and injection (week)	< 0.001	0.976	0.965	0.986
	Number of fistulas, 1	< 0.001			
	2	0.001	0.278	0.127	0.609
	3	< 0.001	0.062	0.014	0.281
Clinical response at w26	Localisation[a], L1	0.027			
	L2	0.936	0.957	0.329	2.785
	L3	0.019	3.482	1.228	9.879
	Prior resection	0.028	2.998	1.123	8.006
	Prior advanced treatment	< 0.001	0.140	0.052	0.378
	Number of external openings	0.001	7.997	2.223	28.767
Clinical response at w52	Sex (female)	0.045	0.392	0.157	0.980
	Time between preparation and injection (week)	0.030	0.992	0.984	0.999

[a]Montreal classification.
Abbreviations: 95% CI, 95% confidence interval; OR, odds ratio (exp(B)); PDAI, Perianal Disease Activity Index; Sig, significance level.

real-world clinical setting. This is in line with the postmarketing INSPIRE registry and retrospective analysis of ADMIRE CD patients (INSPECT), indicating 76.6% and 67.4% clinical remission rates in 1 year [31, 32]. However, since the INSPIRE data have yet to be published, real-world data remain limited, with only a few publications reporting experiences with < 100 patients per report.

Both ADMIRE reports assessed the efficacy with combined clinical and radiologic remission (absence of fluid collections > 2 cm) as primary endpoints, which made it impossible to adequately compare them with our results. Moreover, MRI scans were only performed in 10.8% (24/223) of the patients, and a modified VanAssche score was only available in 7.2% of the patients (16/223). Thus, a statistical analysis, including radiological

TABLE 3 | Number and details of adverse events requiring hospitalisation during the follow-up.

	Event (patient number = 223)	Incidence rate per 100 PY (total PY = 204.2)
Total adverse event	30	14.7
Perianal abscess	14	6.9
Perianal pain	9	3.9
Perianal haematoma	2	0.49
Increased perianal discharge	1	0.49
Pulmonary embolism	1	0.49
Invasive ductal carcinoma	1	0.49
Urinary tract infection	1	0.49
Spontaneous abortion	1	0.49

Abbreviation: PY, patient-year.

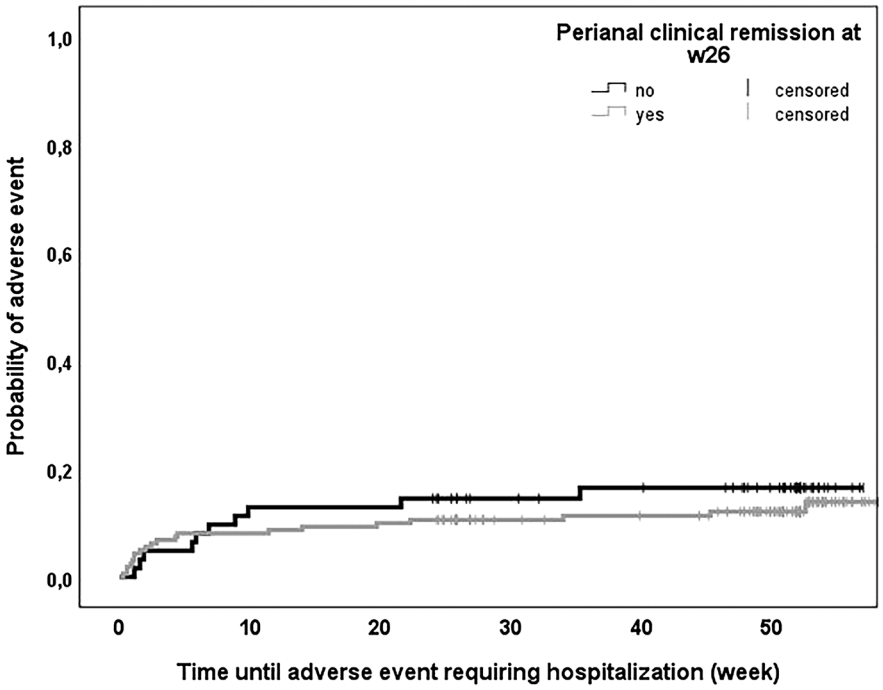

FIGURE 5 | Survival characteristics of adverse events requiring hospitalisation during the follow-ups. The patients were stratified by achieving perianal clinical remission at week 26. Differences were not observed regarding outcome ($X^2 = 0.522$, $p = 0.47$).

characteristics, was not performed. The lack of MRI data in a real-world setting may be explained by the costs, poor availability of the procedure, and the obligatory need for experts for interpretation of the findings which was reported in an experts' opinion including Southeast Europe and Israel [33].

However, due to the pragmatical design, the control group was not able to be used to handle confounders. Nevertheless, to resolve the discrepancies between the efficacy of the clinical trials and the real-world data, several covariates and possible confounders were evaluated via univariable and multivariable regression analyses in this study to predict treatment success. First, a higher baseline PDAI score, antibiotic usage at treatment, a higher number of fistulas and branching fistula tracts, a delay of treatment after preparation surgery, female sex and prior advanced treatment were associated with worse clinical outcomes. Second, although a higher number of external openings seemed to predict treatment failure at week 26, this was not definite, due to the skewed distribution of the variables (three tracts in 7.6% of the patients). Third, prior usage of any advanced treatment can potentially select patients with more aggressive disease courses. However, the possible effect of pharmacological treatment before MSC treatment should not be overlooked. Based on these results, considering the baseline perianal clinical activity and the number of fistulas and branching tracts immediately after preparation for surgery may help to select patients with potentially better treatment outcomes. The impact of antibiotic use during MSC application on treatment outcomes may be questionable and controversial, since the exact pathological connection is lacking, however, the interaction of antibiotics

with MSC have been raised [34]. Extensive usage of systemic antimicrobial medications may be due to fear about infections by the surgical team, and preventive antibiotic usage should be avoided. Another possible reason for higher effectiveness may be the similar population descriptive features in our cohort, compared to the ADMIRE CD trials. The relatively low proportion of branching fistulas and high proportion of patients with mild perianal symptoms further reinforces the importance of population characteristics when comparing available data. Since a high proportion of the patients were on anti-TNF biologics, the clinical and biochemical activity parameters were low and fistula characteristics were comparable. Nevertheless, an accurate comparison with the population description of the overall ADMIRE study is necessary. Although, many of our data were based on a prospective data collection, real-world controlled research in prospective viewpoint considering different disease characteristics (perianal disease, multiple treatments and deviating stoma) is needed to confirm our results.

In this study, our population encompassed 69.1% of the patients treated in a high-volume tertiary centre (where >150 patients were treated with darvadstrocel). In this subcohort, the effectiveness was higher, compared to the rest of the cohort. Although not significant, this raises the question of the experience of the colorectal surgeons, gastroenterologists and radiologists when selecting the ideal number of patients treated in a centre.

As for the correlation between the clinical course of perianal fistulas and symptoms, they have been investigated in previous studies, in which a PDAI score of <4 was considered an inactive disease [26]. In the clinical trial, the PDAI score decreased after treatment. However, a decrease of >1 was observed in the placebo trial (ADMIRE CD week 24: −2.4 vs. −1.2; week 52: −2.3 vs. −1.4). In our study, the proportion of the patients with a PDAI score of <4 increased after treatment and remained until week 52 (57.8%–19.6%), while no significant improvement was detected in regard to the non-responders.

In both ADMIRE reports, a relatively high proportion of patients achieved clinical remission in the control groups (>40%) without darvadstrocel treatment. This result may be explained by the effect of the rigorous curettage of the fistula tracts performed before treatment. Hence, the importance of preparation for surgery should be emphasised in light of the high effectiveness rate of our cohort.

In the literature, assessments of patient-reported symptoms and quality of life are extremely limited. The exception was a French pilot study of 27 CD patients with perianal fistulas who showed significant improvement in their quality of life (CAF-QoL) after the treatment. This is in line with our study, in which 77.8%–81.2% of the patients experienced improvement of clinical symptoms, with a high level of treatment satisfaction [17]. Meanwhile, the ADMIRE CD trials showed an improvement in the IBD quality of life (IBDQ) score after treatment at week 26 (mean difference: 3.8). However, a decrease in this score was also observed in the placebo trial (mean difference: 4.0) [10].

Moreover, in our cohort, the safety profile was reassuring, with an overall AE incidence rate of 14.7/100 PY. Abscesses and proctalgia were reported in most of the cases, which is in line with the pivotal ADMIRE CD trial. In a Swiss cohort, testicular carcinoma and a cytomegalovirus (CMV) infection were reported [15], while in our study, invasive ductal carcinoma of the breast and a urinary tract infection were observed. However, the exact association with darvadstrocel treatment remains a question. Meanwhile, pulmonary embolism was registered in one case, which may be associated with the surgical procedure and hospital admission. These findings suggest that MSC treatment is well tolerated following its administration.

Finally, the present study includes several strengths. First, this was the largest real-world study on the efficacy and safety of darvadstrocel in fistulising CD. Second, regression analyses were applied to assess potential covariates and confounders, select possible predictors, and reduce type I errors. Third, a relatively long follow-up period and strict patient selection were applied to obtain reliable data, while pragmatic endpoints helped to draw real-life conclusions.

It should be noted that the main limitation of the darvadstrocel treatment is its cost. Unfortunately, data regarding absolute cost are not available and only a few costs effectiveness analyses have been published yet. According to an Irish pharmacoeconomic analysis based on ADMIRE CD results, darvadstrocel was associated with an additional 0.898 QALYs for an additional cost of 53.544€ compared to control [35]. Based on a Norwegian analysis, darvadstrocel strategy was associated with a total cost increase of NOK 744095.57 and a QALY gain of 0.416 compared to placebo strategy [36]. To be highlighted, it seems that the overall treatment cost of CD patients with PFCD is higher than patients without PFCD [37]. Therefore, high cost and restrictive reimbursement policies make it impossible to access treatment in high-income countries as well with high prevalence of IBD such as Canada or Denmark.

Nevertheless, some limitations should be noted. First, despite the high number of enrolled patients, the number of involved centres was relatively low and the treatment volume differed. Second, the retrospective design did not allow us to draw exact conclusions, but only potential explanations. Third, it was not possible to specifically analyse the differences between the experiences of each centre, which clearly impacted the treatment effectiveness and safety issues. Fourth, due to the pragmatic design, a control group was not used to independently handle the covariates associated with treatment such as spontaneous fistula closings. Fifth, due to the lack of sufficient amount of results from cross-sectional imaging, analysis including MRI outcomes was not performed. Despite these limitations, our study provides robust real-world evidence of using darvadstrocel in fistulising CD.

Based on our results, the data regarding the effectiveness of the darvadstrocel treatment was higher than the data from the ADMIRE trials. However, the spontaneous closure of fistulas in the placebo-treated patients remains high in the literature, presumably due to the rigorous curettage of the tracts. Moreover, the safety profile was reassuring, while the patients' satisfaction levels were high. Although precise conclusions may not be drawn, due to the retrospective setting and the lack of control group, the roles of several aspects of the preliminary

assessment of treatment success and patient selection may be considered. In this regard, the patients with lower perianal clinical activity at the baseline and a lower number of fistulas may achieve better outcomes, while tract preparation immediately before transplantation and appropriate use of antibiotics should be prioritised. In sum, multidisciplinary medical and surgical treatment approaches are necessary for managing CD patients with perianal disease and further real-world data are needed.

Author Contributions

Péter Bacsur: conceptualization, data curation, formal analysis, methodology, writing – original draft, investigation. **Daniel Shaham:** data curation, writing – review and editing. **Zuzana Serclova:** data curation, writing – review and editing. **Tamás Resál:** data curation, formal analysis, methodology, investigation, writing – review and editing. **Bernadett Farkas:** data curation, writing – review and editing. **Patrícia Sarlós:** data curation, writing – review and editing. **Pál Miheller:** data curation, writing – review and editing. **Nitsan Maharshak:** data curation, writing – review and editing. **Meir Zemel:** data curation, writing – review and editing. **Ariella Bar-Gil Shitrit:** data curation, writing – review and editing. **Shlomo Yellinek:** data curation, writing – review and editing. **Anita Bálint:** data curation, writing – review and editing. **Anna Fábián:** data curation, writing – review and editing. **Renáta Bor:** data curation, writing – review and editing. **Zsófia Bősze:** data curation, writing – review and editing. **Emese Iványi:** data curation, writing – review and editing. **Zoltán Szepes:** data curation, writing – review and editing. **Klaudia Farkas:** data curation, writing – review and editing. **Illés Tóth:** data curation, writing – review and editing. **György Lázár:** data curation, writing – review and editing. **Katerina Vlkova:** data curation, writing – review and editing. **Aneta Tremerova:** data curation, writing – review and editing. **Petra Zuskova:** data curation, writing – review and editing. **Szabolcs Ábrahám:** resources, writing – review and editing, conceptualization, formal analysis, methodology. **Tamás Molnár:** conceptualization, methodology, writing – review and editing, formal analysis, resources.

Acknowledgements

Writing support was provided by Enago and funded by Klebelsberg Library, University of Szeged. The authors are grateful to Csilla Tóth-Káli, Brigitta Csorba, Krisztina Ördög and Stefánia Csák for study support (Department of Medicine, University of Szeged).

Ethics Statement

This study was approved by the National Institute of Public Health and Pharmacy based on the proposal of the Scientific Research Ethics Committee of the Hungarian Medical Research Council (Registration No. NNGYK/GYSZ/12796-4/2024). This study was conducted according to the principles of the Declaration of Helsinki (1975 Declaration of Helsinki, 6th revision, 2008), while the protocol was approved by the local ethics committee of the participating centres. All the patients gave their written informed consent for regular health care.

Conflicts of Interest

T.M. has received speaker's honoraria from MSD, AbbVie, Egis, Goodwill Pharma, Takeda, Pfizer, Janssen, Sandoz, MundiPharma, Phytotec, Roche, Fresenius and Teva. K.F. has received speaker's honoraria from AbbVie, Janssen, Ferring, Takeda and Goodwill Pharma. N.M. has received speaking and/ or consulting fees from Pfizer, Abbvie, Lilly, Takeda, Janssen, Ferring, BiomX, BMS, Nestle, Trobix Innovation and Teva and grant support from Takeda, Janssen, Abbott, Abbvie, Pfizer, BMS and Nestle. P.B., D.S., Z.S., T.R., B.F., P.S., P.M., M.Z., A.B.-G.S., S.Y., A.B., A.F., R.B., Z.S.B., E.I., Z.S.Z., I.T., G.L., K.V., A.T., P.Z. and S.Á. have declare no conflicts of interest.

Data Availability Statement

TM is the guarantor of the article. All authors have read and agreed to the submitted version of the manuscript. Data are available upon request. The data cannot be shared publicly for the privacy of individuals that participated in the study. The data underlying this article will be shared on reasonable request to the corresponding author. The current manuscript, including related data and figures, has not been previously published and is not under consideration elsewhere.

References

1. J. D. Feuerstein and A. S. Cheifetz, "Crohn Disease: Epidemiology, Diagnosis, and Management," *Mayo Clinic Proceedings* 92, no. 7 (2017): 1088–1103.

2. J. Burisch, G. Kiudelis, L. Kupcinskas, et al., "Natural Disease Course of Crohn's Disease During the First 5 Years After Diagnosis in a European Population-Based Inception Cohort: An Epi-IBD Study," *Gut* 68, no. 3 (2019): 423–433.

3. G. Hellers, O. Bergstrand, S. Ewerth, and B. Holmström, "Occurrence and Outcome After Primary Treatment of Anal Fistulae in Crohn's Disease," *Gut* 21, no. 6 (1980): 525–527.

4. M. J. Solomon, "Fistulae and Abscesses in Symptomatic Perianal Crohn's Disease," *International Journal of Colorectal Disease* 11, no. 5 (1996): 222–226.

5. J. Panés and J. Rimola, "Perianal Fistulizing Crohn's Disease: Pathogenesis, Diagnosis and Therapy," *Nature Reviews. Gastroenterology & Hepatology* 14, no. 11 (2017): 652–664.

6. American Gastroenterological Association Clinical Practice Committee, "American Gastroenterological Association Medical Position Statement: Perianal Crohn's Disease," *Gastroenterology* 125, no. 5 (2003): 1503–1507.

7. J. Torres, S. Bonovas, G. Doherty, et al., "ECCO Guidelines on Therapeutics in Crohn's Disease: Medical Treatment," *Journal of Crohn's & Colitis* 14, no. 1 (2020): 4–22.

8. M. Adamina, S. Bonovas, T. Raine, et al., "ECCO Guidelines on Therapeutics in Crohn's Disease: Surgical Treatment," *Journal of Crohn's & Colitis* 14, no. 2 (2020): 155–168.

9. A. L. Lightner, Z. Wang, A. C. Zubair, and E. J. Dozois, "A Systematic Review and Meta-Analysis of Mesenchymal Stem Cell Injections for the Treatment of Perianal Crohn's Disease: Progress Made and Future Directions," *Diseases of the Colon and Rectum* 61, no. 5 (2018): 629–640.

10. J. Panés, D. García-Olmo, G. Van Assche, et al., "Expanded Allogeneic Adipose-Derived Mesenchymal Stem Cells (Cx601) for Complex Perianal Fistulas in Crohn's Disease: A Phase 3 Randomised, Double-Blind Controlled Trial," *Lancet* 388, no. 10051 (2016): 1281–1290.

11. J. Panés, D. García-Olmo, G. Van Assche, et al., "Long-Term Efficacy and Safety of Stem Cell Therapy (Cx601) for Complex Perianal Fistulas in Patients With Crohn's Disease," *Gastroenterology* 154, no. 5 (2018): 1334–1342.e4.

12. D. Garcia-Olmo, I. Gilaberte, M. Binek, et al., "Follow-Up Study to Evaluate the Long-Term Safety and Efficacy of Darvadstrocel (Mesenchymal Stem Cell Treatment) in Patients With Perianal Fistulizing Crohn's Disease: ADMIRE-CD Phase 3 Randomized Controlled Trial," *Diseases of the Colon and Rectum* 65, no. 5 (2022): 713–720.

13. Z. Serclova, D. Garcia-Olmo, S. T. Chen, et al., "OP18 Efficacy and Safety of Darvadstrocel Treatment in Patients With Complex Perianal Fistulas and Crohn's Disease: Results From the Global ADMIRE-CD II

Phase 3 Study," *Journal of Crohn's & Colitis* 18, no. Supplement_1 (2024): i34–i35.

14. H. Wang, H. Y. Jiang, Y. X. Zhang, H. Y. Jin, B. Y. Fei, and J. L. Jiang, "Mesenchymal Stem Cells Transplantation for Perianal Fistulas: A Systematic Review and Meta-Analysis of Clinical Trials," *Stem Cell Research & Therapy* 14, no. 1 (2023): 103.

15. D. Cabalzar-Wondberg, M. Turina, L. Biedermann, G. Rogler, and P. Schreiner, "Allogeneic Expanded Adipose-Derived Mesenchymal Stem Cell Therapy for Perianal Fistulas in Crohn's Disease: A Case Series," *Colorectal Disease* 23, no. 6 (2021): 1444–1450.

16. O. Schwandner, "Stem Cell Injection for Complex Anal Fistula in Crohn's Disease: A Single-Center Experience," *World Journal of Gastroenterology* 27, no. 24 (2021): 3643–3653.

17. N. Fathallah, M. Akaffou, M. A. Haouari, et al., "Deep Remission Improves the Quality of Life of Patients With Crohn's Disease and Anoperineal Fistula Treated With Darvadstrocel: Results of a French Pilot Study," *Techniques in Coloproctology* 27, no. 12 (2023): 1201–1210.

18. W. R. Best, J. M. Becktel, J. W. Singleton, and F. Kern, Jr., "Development of a Crohn's Disease Activity Index," *National Cooperative Crohn's Disease Study. Gastroenterology* 70, no. 3 (1976): 439–444.

19. E. von Elm, D. G. Altman, M. Egger, S. J. Pocock, P. C. Gøtzsche, and J. P. Vandenbroucke, "The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) Statement: Guidelines for Reporting Observational Studies," *Journal of Clinical Epidemiology* 61, no. 4 (2008): 344–349.

20. M. S. Silverberg, J. Satsangi, T. Ahmad, et al., "Toward an Integrated Clinical, Molecular and Serological Classification of Inflammatory Bowel Disease: Report of a Working Party of the 2005 Montreal World Congress of Gastroenterology," *Canadian Journal of Gastroenterology* 19 (2005): 5A–36A.

21. A. G. Parks, P. H. Gordon, and J. D. Hardcastle, "A Classification of Fistula-In-Ano," *British Journal of Surgery* 63 (1976): 1–12.

22. E. Koutroumpakis and K. H. Katsanos, "Implementation of the Simple Endoscopic Activity Score in crohn's Disease," *Saudi Journal of Gastroenterology* 22, no. 3 (2016): 183–191.

23. E. J. Irvine, "Usual Therapy Improves Perianal Crohn's Disease as Measured by a New Disease Activity Index. McMaster IBD Study Group," *Journal of Clinical Gastroenterology* 20, no. 1 (1995): 27–32.

24. K. L. van Rijn, C. A. Lansdorp, J. A. W. Tielbeek, et al., "Evaluation of the Modified Van Assche Index for Assessing Response to Anti-TNF Therapy With MRI in Perianal Fistulizing Crohn's Disease," *Clinical Imaging* 59, no. 2 (2020): 179–187.

25. A. T. Jebb, V. Ng, and L. Tay, "A Review of Key Likert Scale Development Advances: 1995-2019," *Frontiers in Psychology* 12 (2021): 637547.

26. A. Losco, C. Viganò, D. Conte, B. M. Cesana, and G. Basilisco, "Assessing the Activity of Perianal Crohn's Disease: Comparison of Clinical Indices and Computer-Assisted Anal Ultrasound," *Inflammatory Bowel Diseases* 15, no. 5 (2009): 742–749.

27. D. Garcia-Olmo, D. Herreros, I. Pascual, et al., "Expanded Adipose-Derived Stem Cells for the Treatment of Complex Perianal Fistula: A Phase II Clinical Trial," *Diseases of the Colon and Rectum* 52, no. 1 (2009): 79–86, https://doi.org/10.1007/DCR.0b013e3181973487.

28. H. Guadalajara, D. Herreros, P. De-La-Quintana, J. Trebol, M. Garcia-Arranz, and D. Garcia-Olmo, "Long-Term Follow-Up of Patients Undergoing Adipose-Derived Adult Stem Cell Administration to Treat Complex Perianal Fistulas," *International Journal of Colorectal Disease* 27, no. 5 (2012): 595–600, https://doi.org/10.1007/s00384-011-1350-1.

29. Y. Cao, Q. Su, B. Zhang, F. Shen, and S. Li, "Efficacy of Stem Cells Therapy for Crohn's Fistula: A Meta-Analysis and Systematic Review," *Stem Cell Research & Therapy* 12, no. 1 (2021): 32.

30. F. S. Fousekis, K. Mpakogiannis, G. D. Lianos, et al., "Effectiveness and Safety of Darvadstrocel in Patients With Complex Perianal Fistulizing Crohn's Disease: A Systematic Review," *Annals of Gastroenterology* 37, no. 1 (2024): 46–53.

31. N. Fathallah, O. Zmora, D. C. Baumgart, et al., "INSPIRE: Preliminary Data From an Observational Post-Marketing Registry on the Effectiveness and Safety of Darvadstrocel in Patients With Crohn's Disease and Complex Perianal Fistulas," *Journal of Crohn's & Colitis* Supplement_1 (2024): i191–i193.

32. J. Panés, G. Bouma, M. Ferrante, et al., "INSPECT: A Retrospective Study to Evaluate Long-Term Effectiveness and Safety of Darvadstrocel in Patients With Perianal Fistulizing Crohn's Disease Treated in the ADMIRE-CD Trial," *Inflammatory Bowel Diseases* 28, no. 11 (2022): 1737–1745.

33. G. Norčič, N. Smrekar, S. Marković, et al., "Insights Into Treatment of Complex Crohn's Perianal Fistulas," *BMC Proceedings* 18, no. Suppl 7 (2024): 7.

34. V. Johnson, T. Webb, A. Norman, et al., "Activated Mesenchymal Stem Cells Interact With Antibiotics and Host Innate Immune Responses to Control Chronic Bacterial Infections," *Scientific Reports* 7, no. 1 (2017): 9575.

35. "Cost-Effectiveness of Darvadstrocel (Alofisel®) for Treatment of Complex Perianal Fistulae About the National Centre for Pharmacoeconomics," 2019, https://www.ncpe.ie/wp-content/uploads/2019/09/Summary-Darvadstrocel-23.09.2019.pdf.

36. J. J. Jiang and J. J. Jiang, "Cost-Effectiveness of Treatment of Darvadstrocel in Combination With Standard of Care Compared to Placebo Added to Standard of Care for Refractory Complex Perianal Fistulas in Patients With Crohn's Disease in Norway."

37. G. Chen, V. Pedarla, K. D. Null, S. E. Cazzetta, Q. R. Khan, and D. A. Schwartz, "Health Care Costs and Resource Utilization Among Patients With Crohn's Disease With and Without Perianal Fistula," *Inflammatory Bowel Diseases* 28, no. 6 (2022): 870–877.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.