Treatment of Diabetic Foot Ulcers and Peripheral Artery Disease with Placenta-Derived Adherent Cells

Steven Fischkoff, MD
Celgene Cellular Therapeutics
Warren, NJ, USA
Celgene Cellular Therapeutics
Placental Cells & Biomaterials

Placenta

Adherent Cells

PDAC

PDA-001/2

AMDAC

Tissue Processing

Amniotic Membrane

Placental Collagen

Chorionic Matrix

Extracellular Matrix

Suspension Cells

PSC-100

PiNK

RBC

PDAC, active ingredient; PDA-001, IV formulation; PDA-002, IM formulation
PDA-002: Mesenchymal-like Stromal Cells Derived From Normal Full Term Human Placenta

- Mesenchymal stromal cells (MSCs) have been shown to modulate inflammation and secrete factors to promote angiogenesis.
- Therapeutic angiogenesis with PDA-002 offers a promising treatment option for patients who have DFU with PAD.
PDA-002 Process Overview

Procurement, Donor Screening

Clinical Grade Placenta

Dissection, Enzymatic Digestion

Establishment Culture:
- Cell Line

Product:
- Frozen vial of cells
- 20 million / vial

- Thaw
- Formulate
- Inject

Master Cell Stock

Working Cell Stock

Clinical Dose Lot

Expansion Culture in Bioreactors
PDA-002 Product Overview

- Cryo-preserved cells in a 6 ml sterile, low endotoxin vial
- Derived from non-maternal portions of placenta and cord
- No HLA matching required
- LN2 storage (-120°C to -196°C)
- ≥80% viability post-thaw
- Identity/purity markers: CD10+/CD200+/CD105+/ CD34-
Manufacturing at Scale

One placenta > 100,000 doses
Shift to High Capacity Manufacturing Technologies

- Reduced labor/COGS
- Greater capacity
- Higher quality products
**Diabetic Foot Ulcer in Patients With PAD**

- Over 29 million people with diabetes in the US\(^1\)
  - 25% will get at least one DFU during their lifetime\(^2\)
  - 30% of patients with DFU will go on to have some level of amputation
- Patients with DFU and limb ischemia due to PAD have a higher risk of amputation and prolonged wound healing\(^3\)
- No approved products for the treatment of patients who have DFU with PAD

---

**Patients with Ischemic, Neuroischemic, and Neuropathic DFU 5-Year Amputation Rates**

- Ischemic: 29%
- Neuroischemic: 25%
- Neuropathic: 11%


**Probability of Wound Healing Based on TcPO\(_2\), Toe Pressure, and Ankle Pressure**

- TcPO\(_2\)
- Toe pressure
- Ankle pressure


---

DFU, diabetic foot ulcer; PAD, peripheral arterial disease; TcPO\(_2\), transcutaneous oxygen pressure; US, United States.

PDA-002 Demonstrated Improved Blood Flow and Angioscore in Rodent Hind Limb Ischemia Models


- A long term healing effect was observed with IM injection of PDA-002 (multiple HLI models in rat and mice)
- Animals administered PDA-002 demonstrated enhanced blood flow and increased blood volume
- No therapeutic efficacy observed for human fibroblasts, or dead PDA-002

HLI, hind limb ischemia; IM, intramuscular.
Angiogenesis Assessment: Histochemical Assessment

- Indoxyl-tetrazolium method for alkaline phosphatase to was used to identify newly synthesized vessels
- The two lose doses of PDA-002 demonstrated enhanced angiogenesis compared with vehicle
PDA-002 in Diabetic Foot Ulcer with Peripheral Artery Disease – MoA Hypothesis

PDA-002: MESENCHYMAL STROMAL CELL-LIKE POPULATION FROM FULL-TERM HUMAN PLACENTA

ANGIOGENESIS/ VASCULOGENESIS

Vascular Growth Factors
- PDGF
- FGF
- VEGF, HGF

IMMUNE MODULATION
Local, Regional, Systemic

Cytokine Modulation
- TNF-α
- IL-17
- IL-23
- IL-10
- IL-6
- MCP-1

Immune Cell Modulation
- Dendritic cells
- Monocytes
- Macrophages

IMPROVED PERIPHERAL CIRCULATION AND TISSUE OXYGENATION

Tissue Homeostasis
- Endothelial cell survival
- Epithelial cell proliferation/migration
- Myofibroblast differentiation

Tissue Repair Ulcer Healing

REDUCED INFLAMMATION

Antioxidants and Others
- CAT
- SOD
- HMOX1
- MMPs
- IDO
- PGE2

*Based on the available in vitro and in vivo non-clinical data for PDA-002.
Phase 1 Study Design

Multicenter, Open-label, Dose-escalation
Patient Population

- Patients with diabetic foot ulcer and peripheral arterial disease
- Grade 1 (full thickness only) or Grade 2 on the Wagner Grading Scale
- Ulcer of > 1 month duration which has not adequately responded to conventional ulcer therapy
- Peripheral arterial disease with ankle-brachial index > 0.5 and ≤ 0.9 or toe-brachial index > 0.35 and ≤ 0.7
The patient population is defined as those with diabetes, PAD and a DFU.

The initial trials will target patients with Type 1 and 2 diabetes, an ABI between 0.9 and 0.5 and a DFU of at least 1 month duration not responding to standard therapy.

The ABI range that is generally considered normal is 1.0–1.3.
> 1.30 Noncompressible
1.0 – 1.29 Normal
0.91 – 0.99 Borderline (equivocal)
0.41 – 0.90 Mild to moderate PAD
0.00 – 0.40 Severe PAD
### Baseline Patient Demographics and Disease Status

<table>
<thead>
<tr>
<th></th>
<th>Cohort 1</th>
<th>Cohort 2</th>
<th>Cohort 3</th>
<th>Cohort 4</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>3 × 10⁶ cells</td>
<td>10 × 10⁶ cells</td>
<td>30 × 10⁶ cells</td>
<td>100 × 10⁶ cells</td>
</tr>
<tr>
<td>n</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>6</td>
</tr>
<tr>
<td>Age, mean (y)</td>
<td>72</td>
<td>67</td>
<td>65</td>
<td>71</td>
</tr>
<tr>
<td>Gender, male (%)</td>
<td>67</td>
<td>100</td>
<td>33</td>
<td>83</td>
</tr>
<tr>
<td>Ulcer size (cm²)</td>
<td>2.1 ± 2.1</td>
<td>4.6 ± 4.3</td>
<td>2.9 ± 3.6</td>
<td>4.1 ± 4.6</td>
</tr>
<tr>
<td>Ulcer duration (weeks)</td>
<td>52 ± 22</td>
<td>62 ± 60</td>
<td>82 ± 123</td>
<td>27 ± 15</td>
</tr>
<tr>
<td>ABI</td>
<td>1 ± 0.2</td>
<td>0.96 ± 0.2</td>
<td>0.95 ± 0.14</td>
<td>0.81 ± 0.2</td>
</tr>
<tr>
<td>Ulcer Grade</td>
<td>Grade 1</td>
<td>Grade 1</td>
<td>Grade 1; n = 2</td>
<td>Grade 1; n = 5</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Grade 2; n = 1</td>
<td>Grade 2; n = 1</td>
</tr>
<tr>
<td>Rutherford Score</td>
<td>0, 1, 2</td>
<td>0, 2, 5</td>
<td>0, 0, 2</td>
<td>0, 0, 2, 5, 5, 5</td>
</tr>
</tbody>
</table>

- 15 patients were sequentially enrolled in Cohorts 1-4\(^a\)
- Coronary artery disease: 47%
- Prior amputation: 47%

\(^a\) N = 3 enrolled in Cohorts 1-4 since no patients met dose limiting toxicity; an additional 3 were enrolled in Cohort 4 to establish the maximum tolerated dose. ABI, ankle-brachial index.
Safety Results

- No patients met the criteria for study stopping rules\(^a\) or for a dose limiting toxicity\(^b\)
- No treatment-related SAEs
- Observed SAEs were consistent with what would be expected in a population that has DFU with PAD
  - Hypoglycemia, cellulitis, osteomyelitis
- One non-treatment related death
  - Acute myocardial infarction and cardiogenic shock

\(^a\) Study stopping rules: ≥ Grade 2 allergic reactions that were suspected to be related to the IP; experiencing an unexpected, treatment-related SAE or DLT within 14 days following the initial dose of the IP.

\(^b\) Dose limiting toxicity: Grade 2 toxicity not resolving within 14 days suspected to be related to the IP; any toxicity ≥ Grade 3 suspected to be related to the IP. DLT, dose limiting toxicity; IP, investigational product; SAE, serious adverse events.
Efficacy Results: Wound Healing

- 7/15 patients had evidence of ulcer healing within 3 months
  - 5 had ulcer closure
  - 2 had partial (approximately 50%) wound healing
- No association between dose and ulcer healing

**Patient With Complete Healing**

**Patient With Partial Healing**
Efficacy Results: Change in ABI

- There was a trend for an increase in ABI in patients following dosing in the overall population
  - $\Delta$ ABI from screening to prior to dosing: 0.003
  - $\Delta$ ABI at 3 months following dosing: 0.16
- There was an association between dose and change in ABI
- An increase in ABI was observed in patients whose DFU healed compared with patients whose DFU did not heal

Note: data only available for 14 patients.

Median Change in ABI at 3 Months

- Median change in ABI from baseline
- Healed
- Didn’t Heal
- Overall

(n = 5)
(n = 9)
(n = 14)
A trend of decrease in circulating endothelial cells, a biomarker of endothelial injury, correlated with ulcer healing in the DFU-001 study.
Phase I Study Conclusions

• PDA-002 was safe and well tolerated in patients who have DFU with PAD

• 5/15 patients with chronic DFU and PAD experienced closure of their ulcers within 3 months after treatment with PDA-002

• There was a trend for an increase in ABI in patients who healed their ulcer after 3 months following administration of PDA-002

• There was no association between dose and wound healing and change in ABI, but the sample size was limited

A placebo-controlled phase 2 study has been initiated to evaluate PDA-002 in patients who have DFU with PAD (NCT02264288)
**TREAD PDA-002 Phase 2 Study Design**

**Screening/Baseline**
(Day -28 to 0)

- Screening eligibility & informed consent document signature

**Treatment**
(Days 1 and 8)

- Randomized, double-blind, ~30 per arm (10 at highest dose)
  1: 3 \( \times 10^6 \) cells
  2: 10 \( \times 10^6 \) cells
  3: 30 \( \times 10^6 \) cells
  4: Placebo

**Follow-up**
(24 months)

- Primary Efficacy Endpoint – Month 3, (6??)
- Final Analysis Month 6

**Follow-up visits**

- Study Day 15
- Months 1, 2 & 3
- Months 6, 9, 12, & 24

**Baseline Assessments**

- Wound assessments, Basic hemodynamics, Sample collection

**Safety, Duration of Effect**

- Follow-up visits

First Patient Screened 10/23/14

Area = 0.2 cm$^2$
Transcutaneous Oxygen Measurement
Key Phase 2 Efficacy and Outcome Endpoints

The primary endpoint is complete wound closure of the index ulcer within 3 months and retaining closure for a subsequent 4 weeks in subjects who have DFU with PAD.

- **Clinical Endpoints**
  - Time to ulcer closure and complete wound closure of the index ulcer up to 6 months.
  - The number, size of all ulcers and 50% closure of the index ulcer
  - Wagner Grading Scale over time
  - Rutherford Criteria over time

- **Vascular Endpoints**
  - ABI/TBI over time
  - Transcutaneous oxygen measurements (baseline, 3 months, 6 months)

- **Health Outcomes**
  - SF-36
  - DFS-SF
  - EuroQOL-5D
  - Survival, Hospitalization, Amputation

- **Neuropathy**
  - Pain VAS
  - Patient Global Impression of change in neuropathy

- **Diabetes**
  - HbA1C
  - Urinary Albumin / Creatinine
PDA-002 DFU-003 Mechanism of Action Study Design

**Screening/Baseline**
- Day -49 to 0

**Run-in Period**
- 3 weeks

**Treatment**
- IP Day 1
- VA+IP Month 1
- VA+IP Month 2
- VA Month 3
- VA Month 6

**Follow-up**
- Month 6 - Month 12

**VA**
- Vascular assessment

**IP**
- Administration of Investigational Product

**Screening Eligibility & Informed Consent Document signature**

**3 x 10^6 cells**

**30 x 10^6 cells**

**Placebo**

24 patients total, randomized 8 to each arm

VA=Vascular assessment
IP=Administration of Investigational Product
GCSF Mobilized PBMN for CLI and DFU MRA Results

Figure 2—Change in digital subtraction angiography of lower limbs in three patients (N5, N9, and N12) from the transplant group before and after cell transplantation, showing a significantly increased formation of new collateral vessels. Arrows direct to the same place of vessel or bone before and after transplantation.

Huang et.al., Diabetes Care 2005
Near Infrared Spectroscopy

**Figure 3.** Light absorption of hemoglobin (Hb) related to its state of oxygenation. In the near-infrared range, absorption of light is different in deoxygenated hemoglobin when compared with oxygenated hemoglobin. These differences can be measured and used to differentiate the adequacy of hemoglobin oxygenation in blood.
Figure 2. Shown on the left are four cross-sectional computed tomography images of the mid-thigh. These CT images are coregistered with the PET transmission images of the same locations and are then used as anatomical templates for placing regions of interest within skeletal muscle for assessment of [18F] FDG tissue activity. Shown to the right of the CT images are multiple PET images. These multiple PET images are taken from one anatomical location of mid-thigh, but they represent multiple time points of imaging following administration of [18F] FDG. The multiple PET images were obtained over 90 min, and began with the administration of the tracer bolus. By utilizing the same set of regions of interest on each dynamic PET image, data for the formulation of a tissue activity curve can be generated. In practice, this acquisition of tissue data is performed on multiple planes (anatomic locations) within the area of muscle being imaged.
Diabetic Neuropathy Study

Overall Study Design

**Screening Period**
Day -28 to Day -1

**Treatment Period**
Randomized double-blind, placebo-controlled
Treatment days 1, 29, 57
1: $3 \times 10^6$ cells N=8
2: $30 \times 10^6$ cells N=8
3: Placebo N=8

**Follow-up Period**
Day 187 to Day 425

Follow-up visits months 8,10,12,14
Patient Population
- Patients with abnormal symptoms of DPN
  - NTSS ≥ 6 or ≥ 2 for 1 or more Sx
- Patients with abnormal neurologic signs
  - UENS 2-24 and/or NIS-LL 2-10

Efficacy Assessments
- Primary: Histology of Peripheral Nerves
  - ENFD at 6 months
- Peripheral limb circulation
  - Hyperspectral Imaging
  - Vascular reactivity in the lower limb
- Signs and symptoms of DPN
- Nerve function
  - Nerve Conduction
  - Autonomic testing
- HRQoL

Epidermal Nerve Fiber Density (ENFD)

Hyperspectral Imaging

Nerve Conduction Velocity Test
Contributors

• **Phase 1 Trial**
  - S. Wu,¹ R. Pollak,² R. Frykberg,³ J. Caporusso,⁴ P. Lawrence,⁵ W. Zhou,⁶
    ¹Rosalind Franklin University, North Chicago, Illinois, USA; ²Endeavor Clinical Trials PA, San Antonio, Texas, USA; ³Phoenix VA Health Care System, Phoenix, Arizona, USA; ⁴Complete Family Foot Care, McAllen, Texas, USA; ⁵Ronald Reagan UCLA Medical Center, Los Angeles, California, USA; ⁶VA Palo Alto Health Care System, Palo Alto, California, USA

• **Celgene Cellular Therapeutics**
  - D. Chitkara, S. Fischkoff, A. Franck, R. Hariri, V. Hernandez, U. Herzberg, W. Hofgartner, V. Jankovic, M. Karnoub, B. Murphy, M. Rudinski, G. Russotti