ZHITTYA
REGENERATIVE MEDICINE INC.

Healing Venous Ulcers with FGF-1: Positive Data from a U.S. Phase II Clinical Trial

Summary: Venous stasis wounds occur as a result of a poorly functioning venous system in the legs. Venous valves, which prevent the backflow of blood into the legs, are often damaged in this disorder, and blood pools in the veins. This results in edema, poor tissue perfusion, and eventually the development of painful wounds which often remain open. There is currently no effective treatment to heal this type of open wound.

It has recently been announced that the human growth factor FGF-1 (fibroblast growth factor) is a potent accelerator in closing venous ulcers. This data was obtained from a U.S. FDA-authorized Phase II clinical study which enrolled a total of 57 patients with venous ulcers. A summary of the results of this trial is as follows:

- No significant safety or adverse events attributable to the drug were reported with topically-applied FGF-1.
- A total of 57 patients with venous stasis wounds were treated in a double-blinded, placebo-controlled Phase IIa trial.
- Subjects treated with FGF-1 had a wound healing rate over twice that of subjects in the placebo group.
- As early as 10 days after treatment, there was a statistically significant difference ($p=0.02$) between placebo and the FGF-1 treated wounds.

According to a recent report (MedMarket Diligence, LLC; Report #S247), there are 12.5 million venous stasis wounds globally and the incidence is growing at approximately 10% annually. The U.S. market for a viable product that can heal venous ulcers (VUs) has been estimated by Shire to be ***$3 billion.***

Venous Ulcers, An Unmet Medical Need: The treatment and healing of chronic wounds (diabetic foot ulcers, bedsores, etc.) is difficult and this is especially true for venous ulcers (VUs), also known as venous stasis ulcers. The healing rates for VUs are typically poor with up to 50 percent of venous ulcers open and unhealed for nine months or longer. Venous ulcer recurrence rates are also troubling with up to one-third of treated patients experiencing four or more episodes of recurrence.[1]
Venous ulcers are costly in terms of both economics and quality of life. Economic considerations include direct medical costs as well as indirect costs from reduced productivity. An estimated 5% to 8% of the world's population is afflicted with venous disease, 1% suffers from VUs of the lower extremities, 3.5% of people over age 65 have VUs and these numbers are rising as the population ages.[2]

There are approximately 12.5 million venous ulcers in the world requiring treatment.[3] It is estimated that venous ulcers affect 500,000-600,000 people in the US every year and it is therefore the most common type of leg ulcer. The total U.S. cost for treating venous ulcers is estimated at $5 billion, with the charge per case ranging from $2,000 to $10,000.[4]

The incidence rate of venous ulcers in people 65 and older within Europe is estimated to be 980,000 while the overall cost is estimated at an annual $8.9 billion.[5]

Pathophysiology of the Venus Ulcer: The return of blood from the peripheral lower limbs to the heart occurs through the venous system. The movement of blood to the heart is assisted by contraction of skeletal muscles that pump the returning blood through a series of one-way valves within the veins to maintain the forward blood flow rate and to prevent the buildup of hydrostatic pressure and backflow into smaller veins and capillaries.

Chronic venous disease can develop when the valves within the veins are damaged resulting in venous hypertension. In patients with damaged valves blood flow rates are diminished facilitating migration of inflammatory white cells into the venous valves, which might exacerbate the valve damage.[6] In addition, the increase in venous pressure causes leakage of fluid from the small thin-walled veins and capillaries in the leg muscles and skin. The accumulation of extravascular immune white cells and fluid in the tissues results in inflammation and edema in the lower leg. The edematous fluid accumulating in the dermis contains not only abundant proteases that can degrade skin connective tissue and inactivate endogenous growth factors but also angiogenesis inhibitors[7], all of which can lead to the formation and persistence of dermal venous ulcers. Cells from these ulcers have diminished proliferative responses that can be reversed with potent mitogenic growth factors such as FGFs.[8] Once a dermal venous ulcer forms, the edematous fluid that accumulates within the tissue leaks out of the ulcer surface as wound exudate, which is characteristic of these ulcers.

The initial cause of the valve damage is not certain but risk factors include heredity, number of previous pregnancies, occupation and age. Heredity determines the number and position of the

valves in the veins. Individuals with fewer valves appear to be at higher risk for chronic venous disease. Pregnancy is also considered a source of stress on the venous system, and females are three times more likely to develop venous leg ulcers. Professions that involve standing or sitting still for extended periods have been correlated with a higher incidence of venous complications and the aging process appears to contribute to valve failure, perhaps as a consequence of co-morbidities and the diminished capacity to repair tissue damage.

Healing Venous Ulcers: For many years, the standard approach to venous ulcers has been compression bandages of various types, with dressings applied to help moisturize the wounds and to kill bacteria. In addition, wounds often have to be debrided surgically, as needed. The standard of care in all treatment regimens has always included compression bandages, with off-loading and elevation designed to alleviate edema.

To heal VUs, or any wound, requires the regeneration of new skin. Human fibroblast growth factor (FGF-1) is the only growth factor demonstrated to directly promote the growth of vascular endothelial cells, fibroblasts and keratinocytes, the three major cell types present in skin. Microvascular endothelial cells are required for the development of the microvasculature that supports the growth and maintenance of regenerating skin, fibroblasts produce collagen and other connective tissue components that give skin its strength and flexibility, and keratinocytes generate the epidermal barrier to dehydration and infection. Therefore, FGF-1 as a single factor can support the regeneration of the principal cellular components needed to heal venous ulcers.

In a double-blinded, placebo-controlled Phase II clinical trial, the effect of topically applied FGF-1 was examined for its ability to accelerate the healing of venous wounds on the legs of patients. The data shown here has been collected from publicly available documents (publications, press releases, investor solicitations) that have been put into the public domain by CardioVascular Bio-Therapeutics, a Las Vegas-based biotechnology company. Venous ulcers were treated 3 times per week for 3 weeks with either topical FGF-1 or the corresponding vehicle placebo, and the rate of ulcer closure monitored as a function of time. As calculated from healing distances shown in the Figure below, wounds treated with FGF-1 healed ***over twice as fast*** ($p = 0.02$) versus placebo at day 10. At day 20, the increased healing rate was still two-fold greater with FGF-1 treatment ($p = 0.07$). Repeated measured ANOVA analysis of healing through day 20 indicated a strong positive trend toward acceleration ($p = 0.06$).

Figure 1: Acceleration of Wound Healing Rate with FGF-1 in Venous Ulcers: *Total healing distance is shown as a function of time in per protocol evaluable subjects with venous stasis ulcers treated with FGF-1 or placebo in a Phase IIa trial. The number of subjects in each group is in parenthes*