RESEARCH ARTICLE

Therapeutic Effects of *Passiflora foetida* L. Fruit on Wound Healing in Streptozotocin-Induced diabetes

Budda Leena[*1], M. Sivakumar[2]

[1]*Bharath institute of higher education and research, Chromepet, Chennai- 600044*
[2] *Department of Pharmacognosy, Faculty of Pharmacy, Sree Balaji Medical College and Hospital, BIHER (DU), Chromepet, Chennai - 600044.*

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Abstract:
The present study investigates the antidiabetic and wound healing potential of *Passiflora foetida* fruit extracts. Administration of the fruit extracts to glucose-loaded normal rats, fasted for 18 hours, resulted in a significant decrease in blood glucose levels. In contrast, the diabetic control group showed a marked increase in blood glucose levels by the end of the 14-day experimental period. The treatment with *Passiflora foetida* extracts led to a significant reduction in fasting blood glucose levels and enhanced wound healing, as assessed by excision wound healing methods over a 21-day period. Ethanolic extract at doses of 400 mg/Kg and 200 mg/Kg exhibited a notable decrease in plasma glucose levels on days 0, 7, and 14. Flavonoids present in the ethyl acetate extract contributed to hypoglycemic activity, particularly at the higher doses of 400 mg/Kg and 200 mg/Kg. Regarding wound healing, the ethanolic and ethyl acetate extracts at the 400 mg/Kg dose demonstrated significant improvements in wound contraction, with percentages increasing from 25.52% to 97.37% and 21.34% to 96.87%, respectively, on the 15th day. Lower doses (100 mg/Kg and 200 mg/Kg) exhibited minimal improvements in wound contraction. Complete wound healing was observed by the 18th day for both extracts at the 400 mg/Kg dose. This study highlights the potential of Passiflora foetida fruit extracts, particularly at higher doses, as effective agents for managing diabetes and promoting wound healing.

Keywords: *Passiflora foetida* L, ethyl acetate extract, ethanol extract, Streptozotocin, antidiabetic activity
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INTRODUCTION

Diabetes mellitus is a chronic metabolic disorder characterized by hyperglycemia resulting from impaired insulin secretion, action, or both. It leads to a variety of complications, including delayed wound healing, which significantly affects the quality of life. One of the major challenges in managing diabetes is controlling blood glucose levels and improving wound healing, especially in diabetic patients who exhibit slow recovery from cuts and wounds. As a result, there is increasing interest in the therapeutic potential of natural products for both antidiabetic and wound healing properties. Passiflora foetida L. (Passion fruit) is a climbing plant native to tropical regions and is known for its diverse pharmacological effects. The fruit of Passiflora foetida contains a variety of bioactive compounds, including flavonoids, alkaloids, and saponins, which have been associated with several health benefits. Recent studies have shown that plant extracts, especially those rich in flavonoids, may have significant antidiabetic properties, which help in regulating blood glucose levels. Additionally, Passiflora foetida extracts have been reported to possess wound healing properties, potentially aiding in tissue regeneration and reducing recovery times.

The current study investigates the potential of Passiflora foetida fruit extracts in managing blood glucose levels and enhancing wound healing in experimental animal models. The aim is to assess the effects of ethanolic and ethyl acetate extracts of Passiflora foetida on blood glucose reduction and wound contraction in streptozotocin-induced diabetic rats. By evaluating both antidiabetic and wound healing effects, this study aims to establish the fruit extracts as potential therapeutic agents for managing diabetes and accelerating wound healing.

MATERIALS AND METHODS

Streptozotocin (Sigma–Aldrich Canada, Oakville, Ontario, Canada). All other chemicals and reagents used were of analytical grade.

Reagents

Buffer (pH 5): 50 g citric acid monohydrate, 12 ml glacial acetic acid, 120 g sodium acetate trihydrate, and 34 g sodium hydroxide added to distilled water up to 1000 ml.

Animals

Wistar albino rats, weighing between 150–200 g, were selected for the experiment. These rats were sourced from the disease-free small animal facility at Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar (Reg. No. 1669/GO/abc/12/CPCSEA Dated 08/04/2013). They

**Author for Correspondence: leenaangel27@gmail.com*

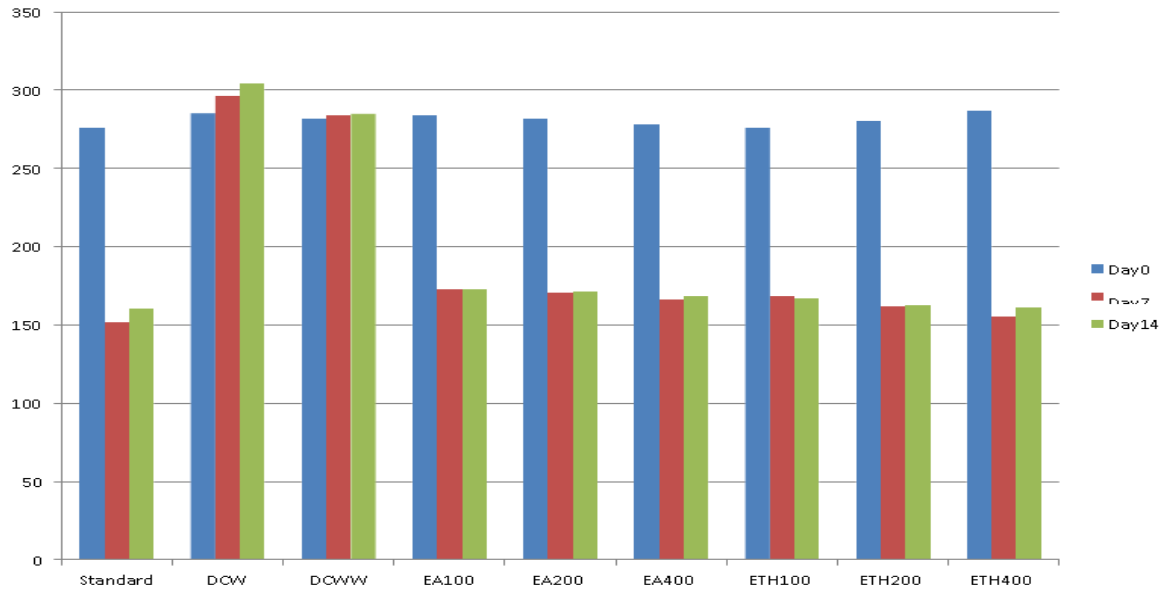

Fig 1: Effect of *Passiflora foetida* L. in streptozotocin-induced diabetes mellitus

Table 1: Antidiabetic activity of fruits of *Passiflora foetida* L. in streptozotocin-induced diabetes mellitus

S. No	Group	Plasma glucose level(mg/dl)		
		0 day	7^{th}day	14^{th}day
1	Standard (Metformin)	275.83±4.945	151.66±3.626*	160.33±2.21*
2	DiabeticControlwith wound	285.16±2.072	296.33±3.412	304.16±6.263
3	DiabeticControlwithout wound	281.66±5.420	284.16±4.490	285.00±5.721
4	Ethylacetateextract100mg/Kg	284.00±13.00	172.50±2.500*	173.00±6.000*
5	Ethylacetateextract200 mg/Kg	281.50±7.500	170.50±5.500*	171.50±4.500*
6	Ethylacetate extract400mg/Kg	278.00±14.000	166.00±10.000*	168.52±4.500*
7	Ethanolextract100 mg/Kg	276.00±13.000	168.50±12.500*	167.05±12.000*
8	Ethanolextract200 mg/Kg	280.50±12.500	162.00±7.000*	162.50±4.500*
9	Ethanolextract400 mg/Kg	286.50±7.500	155.50±2.500*	161.50±9.000*

Values are expressed as mean±SEM, n=6, $p<0.05$ versus diabetic control group (Dunnett'st-test after analysis of variances)

were maintained in a pathogen-free environment and housed in polypropylene cages under standard conditions (25±2°C, with a 12-hour light/dark cycle). The rats were provided with pelleted food (Purina) and had access to tap water ad libitum (Hedrich HH, 2006). Prior to the start of the study, they were acclimatized to the standard diet and water for two weeks. All experimental procedures followed international guidelines for the use and care of laboratory animals and were approved by the Institutional Animal Ethics Committee, Department of Pharmaceutical Sciences, M.D. University, Rohtak (1767/GO/Re/S/14/CPCSEA, 18/07/2014).

Diabetes Induction

Streptozotocin-induced diabetes mellitus

After an overnight fast, streptozotocin (STZ; 50 mg/Kg, i.p.) (Sigma-Aldrich Canada, Oakville, Ontario, Canada) was administered to the rats, dissolved in a citrate buffer (0.1M, pH 4.5), to induce diabetes (Junod A et al., 1969). Fasting blood glucose levels were measured 24 hours post-injection using a Glucometer (Accu-Chek® Extra Care, Roche Diabetes Care India Pvt. Ltd., 601B, Silver Utopia, Chakala Road, Andheri (East), Mumbai, Maharashtra), with glucose oxidase reagent strips, after blood was drawn from the retro-orbital plexus. Rats with blood glucose levels exceeding 250 mg/dl were selected for the study, which commenced 7 days following the streptozotocin injection.

Diabetic Excision Model for Wound Healing Activity

Surgical Procedures and Treatment

On the 7th day after inducing diabetes, excision wounds were created for biochemical parameter analysis and to assess the rate of wound contraction. The animals were anesthetized with thiopentone sodium (40 mg/Kg i.p.), and the right side of each rat was shaved. The shaved area was disinfected with 70% ethanol. Excision wounds of 4 cm^2 (2 cm x 2 cm) were made along the dorsal midline. For 21 days, the rats were orally administered ethanol and ethyl acetate extracts at concentrations of 100 mg/Kg, 200 mg/Kg, and 400 mg/Kg. The control group received an equivalent amount of the vehicle (citrate buffer).

Excision Wound Procedure

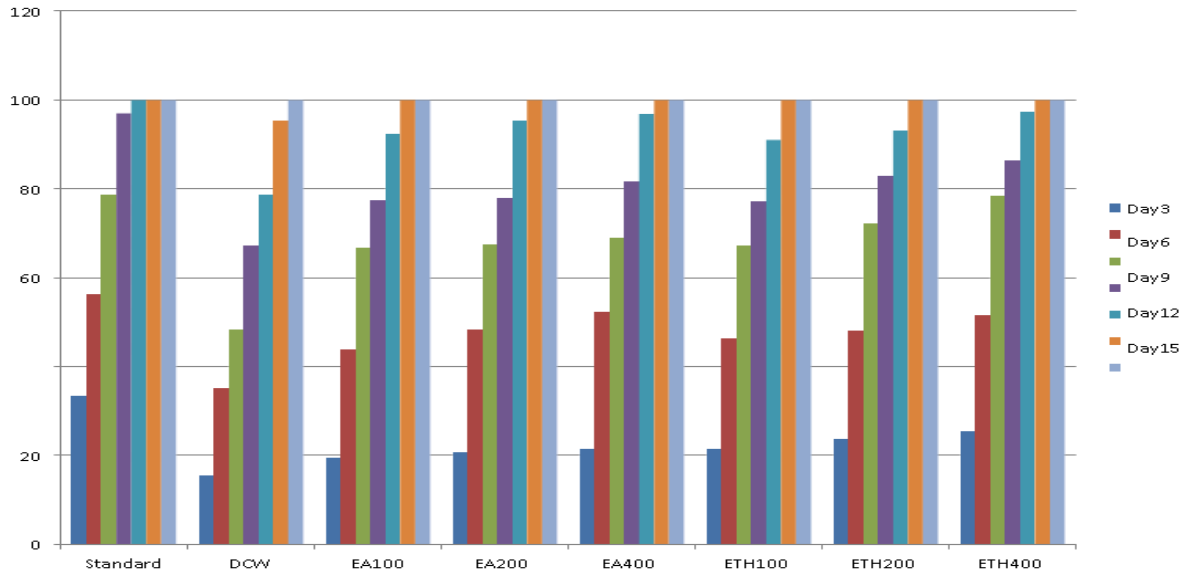

Fig 2: Wound healing activity of *Passiflora foetida* L. in diabetic excision model.

The epithelialization time was recorded (Villegas LF et al., 1997) when no raw wound remained and the scar had fallen off. The excision wounds were traced on transparent paper with a millimeter scale to determine the rate of wound contraction. The percentage of wound healing was calculated based on the change in wound size. The epithelialization period was noted as the time taken for complete healing when no raw wound remained.

Excision Wound Model

The excision wound model was carried out as described above, with parameters such as wound contraction percentage, epithelialization period, and granulated tissue scar area being evaluated (Nayak BS et al., 2007). Photographs were taken every third day, and wound boundaries were traced on transparent paper to measure the wound area in all groups.

Monitored Parameters

Rate of Wound Contraction:

Before treatment (day 0) and after wounding on days 3, 6, 9, 12, 15, and 18, excision wounds were traced on transparent paper with a millimeter scale. The percentage of wound healing was calculated as the change in wound size. The wound contraction percentage was determined using the formula:

% wound contraction = $(A_0 – A_t) / A_0 * 100$

Where (A_0) is the original wound area and (A_t) is the area of the wound at a specific time period after wounding (Yates CC et al., 2007; Rashed AN et al., 2003).

Epithelialization Period:

The epithelialization period refers to the number of days required for the scar to fall off with no raw wound remaining. It was expressed as the time taken for complete epithelialization (Dinesh M et al., 2010).

Animal Grouping

The rats were divided into nine groups, with six animals in each group. They were administered extracts for 21 days. Ethyl acetate and ethanol extracts were selected for pharmacological activity studies, as these extracts from various plant species contained the highest number of potent chemical constituents, determined by qualitative phytochemical analysis and chromatographic profiles. Based on previous studies indicating their safety at doses up to 2000 mg/Kg, the doses of 100 mg/Kg (1/20th of the safe dose), 200 mg/Kg (1/10th), and 400 mg/Kg (1/5th) were chosen for further investigation (Vikram PK et al., 2012; Bhide NK, 1962).

Group I: Standard (Metformin 5 mg/Kg)

Group II: Diabetic rats with wound without treatment (normal control group)

Group III: Diabetic rats without wound (for diabetes only)

Group IV: Diabetic rats with wound treated with ethyl acetate extract by oral route at a dose of 100 mg/Kg

Group V: Diabetic rats with wound treated with ethyl acetate extract by oral route at a dose of 200 mg/Kg

Group VI: Diabetic rats with wound treated with ethyl acetate extract by oral route at a dose of 400 mg/Kg

Group VII: Diabetic rats with wound treated with ethanol extract by oral route at a dose of 100 mg/Kg

Group VIII: Diabetic rats with wound treated with ethanol extract by oral route at a dose of 200 mg/Kg

Group IX: Diabetic rats with wound treated with ethanol extract by oral route at a dose of 400 mg/Kg

Statistical Analysis

Wound area was measured as the percentage contraction in wound size. Analysis of data was performed using Dunnett's t-test with GraphPad Prism 7.0. When $P < 0.05$ compared with control, the data is considered significant.

Results and discussion

Antidiabetic activity of Fruits of *Passiflora foetida* L. in streptozotocin-induced diabetes mellitus

There is a decrease in blood glucose level when *Passiflora foetida fruit* L. extracts were administered to glucose loaded normal rats fasted for 18 h. There is a significant increase in blood glucose level in the diabetic control group at the end of 14th day experimental period and increase in wound healing by excision wound healing method observed for 21 days. There is a significant

Table 2: Wound healing activity of stem of *Passiflora foetida* L. in diabetic excision model

S. No.	Group	Percentage contraction in wound area							Epithelia lization period (in days)
		3^{rd}day	6^{th}day	9^{th}day	12^{th}day	15^{th}day	18^{th}day	21^{st}day	
1	Standard (Metformin)	33.30±0.304	56.34±0.432*	78.71±0.354*	96.96±0.692*	100	100	100	14.86±0.307*
2	Control with wound	15.54±0.164	35.06±0.284	48.37±0.189	67.19±0.276	78.62±0.392	95.45±0.761	100	20.50±0.365
3	Ethylacetate extract100 mg/Kg	19.43±0.245	43.77±0.160*	66.84±0.180*	77.46±0.190*	92.48±0.065*	100	100	17.50±0.500
4	Ethylacetate extract200 mg/Kg	20.71±0.340	48.27±0.150*	67.36±0.005*	77.98±0.090*	95.22±0.495*	100	100	17.00±0.00
5	Ethyl acetate extract400 mg/Kg	21.34±0.185	52.36±0.160*	69.00±0.225*	81.76±0.285*	96.87±0.050*	100	100	16.50±0.50
6	Ethanolextract 100 mg/Kg	21.31±0.040	46.30±0.035*	67.10±0.445*	77.10±0.020*	91.05±0.430*	100	100	18.00±0.00
7	Ethanolextract 200 mg/Kg	23.65±0.075	48.08±0.115*	72.26±0.040*	82.94±0.125*	93.12±0.305*	100	100	17.50±0.50
8	Ethanol extract 400 mg/Kg	25.52±0.005	51.57±0.015*	78.42±0.300*	86.31±0.385*	97.37±0.500*	100	100	16.50±0.50

Values are expressed as mean ± SEM, n=6,p<0.05 versus diabetic control group (Dunnett'st-test after analysis of variances)

decrease in fasting blood glucose level and percentage wound area contraction after administration of extracts.

A significant decrease in plasma glucose level in the ethanolic extract at the dose level of 400mg/Kg and 200 mg/Kg was observed for 0 days, 7^{th} day and 14^{th} day as illustrated in Table 6.5. Various chemical constituents present in the extract are responsible for the antidiabetic activity. Flavonoids present in ethyl acetate extract showed the hypoglycemic activity at the dose level of 400 mg/Kg and 200 mg/Kg on 0 days, 7^{th} day and 14^{th} day. There is an increase in the percentage area of wound contraction from 25.52% to 97.37% and 21.34%to 96.87%respectivelyon 15^{th} day in ethanolic and ethyl acetate extract at the dose of 400 mg/Kg. There is not much increase in the percentage contraction in the wound area in the lower doses (100 mg/Kg and 200mg/Kg) in ethyl acetate and ethanol extract. Complete wound healing is shown by ethyl acetate and ethanol extract at the dose of 400 mg/Kg on 18^{th} day.

CONCLUSION

In conclusion, the administration of *Passiflora foetida* fruit extracts resulted in a significant decrease in blood glucose levels in glucoseloaded normal rats that were fasted for 18 hours. In diabetic rats, there was a notable increase in blood glucose levels in the diabetic control group by the 14th day of the experimental period. However, after the administration of the extracts, there was a marked decrease in fasting blood glucose levels. Additionally, the percentage of wound area contraction showed a significant improvement after 21 days of treatment with the extracts, especially at higher doses.

The ethanolic extract at doses of 400 mg/Kg and 200 mg/Kg demonstrated a significant reduction in plasma glucose levels on days 0, 7, and 14. Flavonoids in the ethyl acetate extract contributed to hypoglycemic activity, particularly at the doses of 400 mg/Kg and 200 mg/Kg. The wound contraction rate increased significantly from 25.52% to 97.37% and from 21.34% to 96.87% on the 15th day for the ethanolic and ethyl acetate extracts at the 400 mg/Kg dose, respectively. Lower doses (100 mg/Kg and 200 mg/Kg) of both extracts showed less pronounced improvements in wound contraction. Complete wound healing was observed by the 18th day in the 400 mg/Kg dose of both extracts.

Overall, Passiflora foetida fruit extracts, particularly at higher doses, demonstrate promising antidiabetic and wound healing properties, supporting their potential as therapeutic agents in managing diabetes and promoting wound healing.

REFERENCES

1. Agarwal PK, Singh A, Gaurav K, Goel S, Khanna HD, Goel RK. Evaluation of wound healing activity of extracts of Plantain Banana (*Musa sapientum* var. *paradisiaca*) in rats. Indian J Exp Biol. 2009; 47: 32-40.
2. Ahmad N, Mukhtar H. Green tea polyphenols and cancer: biologic mechanism and practical implications. Nutr Rev. 1999; 57: 78–83.
3. Akhtar J, Jamil S, Azhar MU. Diabetes mellitus prevention and management. Nat Prod Rad. 2005; 4(5): 413-415.
4. BhideNK.Pharmacologicalstudyandfractionationof*Passiflora foetida*extract. Brit.J.Pharmacol.1962;18:7-18.
5. Biswas TK, Maitry LN, Mukherjee B. Wound healing potential of *Pterocarpus santalinus* Linn: a pharmacological evaluation. Int J Low Extrem Wounds. 2004; 3: 143-150.
6. Biswas TK, MukherjeeB. Plantmedicines of Indian origin for wound healing: a review. IntJLowExtremWounds. 2003; 2(1):25-39.

7. Day C. Traditional plant treatments for diabetes mellitus: Pharmaceutical foods. Br J Nutr. 1998; 80: 5-6.
8. Dinesh M, Manjunath C, Joshi VG, Karigar AA, Sikarwar MS. Wound healing property ofethanolicextractof*Crataevanurvala*rootbark.J.Pharm. Res.2010;3(11): 2788-9.
9. Eming SA, Werner S, Bugnon P, WickSiewe L, Utermohlen O, Davidson JM, Krieg T, RoersA.Acceleratedwoundclosureinmicedeficientforinterleukin-10. Am J Pathol. 2007; 170: 170-202.
10. International Diabetes Federation. IDF Diabetes Atlas, 8th edition, 2017 updated (www.idf.org/diabetesatlas). Accessed on 25 Nov. 2017.
11. Khan A, Safdar M. Role of diet, nutrients, spices and natural products in diabetes mellitus. Pak J Nutr. 2003; 2: 1–12.
12. Khan MTH. Diabetes mellitus treatment and complications. Hamdard Med. 2000; 43(1- 4): 77-83.
13. MemonBA,BhattiK.Diabetesmellitusachronicmetabolicdisorder.HamdardMed.
2002;45(1-4):57-67.
14. MukherjeePK,MukherjeeK,PalM,SahaBP.Woundhealingpotentialof*Nelumbo nucifera* (Nymphaceae) rhizome extract. Phytomedicine. 2000; 7: 66-73.
15. NayakBS,AndersonM,PereiraPLM.Evaluationofwound-healingpotentialof
*Catharanthusroseus*leafextract
inrats.Fitoterapia.2007;78:540-44.
16. Nayak BS, Krishma M. Influence of ethanolic extract of *Jasminum grandiflorum* Linn. flowers on wound healing activity in rats. Indian J Physiol Pharmacol. 2007; 51:189-194.
17. PierceGF,MustoeTA.Pharmacologicenhancementofwoundhealing.AnnuRevMed. 1995; 46: 467-481.
18. Prabhakar PK, Doble M. Mechanism of action of natural products used in the treatment of diabetes mellitus. Chin J Integr Med. 2011; 17(8): 563-574.
19. Vikram PK, Malvi R, Jain DK. Evaluation of analgesic and anti-inflammatory potential of *Passiflora foetida fruit*Linn. Int J Curr Pharm Res. 2012; 4(4): 47-50.
20. PurnaSK,BabuM.Collagenbaseddressings -a review.Burns.2000;26:54-62.
21. Rai N, Tripathi K, Sharma D, Shukla VK. Apoptosis: a basic physiological process in wound healing.Int J Low Extrem Wounds. 2005; 4: 138-144.
22. Rashed AN, Afifia FU, Disi AM. Simple evalution of the wound healing activity of a crude extract of *Portulaca oleracea* L. (growing in Jordan) in Mus musculus JVI-1. J. Ethnopharmacol. 2003; 88: 131-6.
23. Rosenthal SP. Acceleration of primary wound healing by insulin. Arch Surg. 1968; 96: 53–55.
24. Stipcevic T, Piljac A, Piljac G. Enhanced healing of full-thickness burn wounds usingdi-rhamnolipid. Burns. 2006; 32: 24-34.
25. Vasco C, Ruales J, Eldin A. Total phenolic compounds and antioxidant capacity ofmajor fruits from Ecuador. Food Chem. 2008; 111: 816–823.
26. Villegas LF, Fernandez ID, Maldonado H, Torres R, Zavaleta A, Vaisberg AJ, Hammond GB. Evaluation of the wound-healing activity of selectedtraditional medicinal plants from Peru. J. Ethnopharmacol. 1997; 55:193–200.
27. World Health Organization. 10 Facts About Diabetes, Updated April 2016. (http://www.who.int/features/factfiles/diabetes/en/index.html). Accessed on25 Nov 2017.
28. Yates CC, Whaley D, Babu R, Zhang J, Krishna P, Beckman E, Pasculle AW, Wells A. The effect of multifunctional polymer-based gels on wound healing in full thickness bacteria contaminated mouse models. Biomaterials. 2007; 28:3977– 3986.