Multifloral honey wound healing evaluation on Class IIB depth burns induced in rats of the Wistar strain

Abstract

Introduction: Burns are frequent. Can wound care after burn be done using honeys available in our context? This is why this study was conducted to evaluate the burn wound healing activity of three types local types of honey on rats.

Materials and methods: This was a comparative experimental study held at the Faculty of Medicine and Biomedical Sciences of the University of Yaoundé I. We used 3 varieties of honey designated by their cities of origin: Ngaoundal, Okuh and Yaounde. Brulex® (Zinc Oxide) was our reference medicine against burns. Male rats of the wistar strain have served as animal material on which the burns were performed. There were 36 divided into 4 groups of 9 rats per batch for treatment with reference medicine and 3 honey samples. We realized experimental burns unger general anesthesia by a heated mass. The surface areas were calculated using the AUTOCARD 2014 software. The photos were taken using an XTIGI V10 telephone. At the various dates selected, the wound surfaces calculated for each batch were expressed in the form of the mean ± standard deviation. For their comparison, we used Fisher's ANOVA test.

Results: In the evolution of the healing process (Table I and Figure 1), the group of rats treated with Brulex® showed the largest injured surfaces on day 0 (8.83 ± 2.34 cm2), the least extensive being those of the Okuh group (6.83 ± 0.66 cm^2). On the 24th day, the smallest areas were in order those of the Ngaoundal, Yaounde and Okuh lots. While the rats in the Brulex® and negative control lots were not yet completely cured.

Conclusion: The three honey types are effective in burn wound care

Key words : Burns – Wound healing - Honey

INTRODUCTION

According to the World Health Organisation (WHO), the International Society for Burn Injuries (ISI) and the WHO Emergency Medical Teams Technical Working Group on Burns (WHO TWGB), burns are a global public problem[1-3]. Burns are responsible for 180,000 deaths per year worldwide, the vast majority occurring in low and middle income countries [1]. In Cameroon, a study by the major burns center in Douala showed that domestic accidents are the most frequent causes (63.13%), followed by work accidents (22.50%) [4]. Different methods of burns wound are exist [5]. However, the use of many products is often limited because of their unavailability and their high price, especially in developing countries. An alternative is therefore the use of local natural products, which have proven their effectiveness in the treatment of burns, are available, safe and affordable. Honey is one of these natural products. It is of plant and animal origin, obtained by transformation of flower nectar or honeydew by the bee *Apis mellifera* [6]. Some studies have already been carried out on honey and its properties, worlwide and in Africa [7-22]. In relation to its climatic diversity, Cameroon presents a variety and a diversity of honeys for such scientific experiments. This is why it seemed important for us to evaluate the healing activities of three varieties of honey from Cameroon, harvested in three localities, different both by their climate and their vegetation. The general objective was to investigate the healing properties of these three varieties of Cameroonian honey on experimental burns in rats.

MATERIAL AND METHODS

Nature, type and location of the study

This was a comparative experimental study related to the therapeutic efficacy of varieties of honey in Cameroon. It was held at the Faculty of Medicine and Biomedical Sciences of the University of Yaoundé I (Anatomopathology, and Animal Care Laboratories) and at the National Polytechnic School of the same university, where the manufacture of a small mass was done in the Mechanical Engineering laboratory.

Equipment

For our study, we used 3 varieties of honey designated by their cities of origin: Ngaoundal, Okuh and Yaounde. Brulex® was our reference medicine against burns. 100g of this topical ointment contains: 5g of Zinc Oxide; 1g of Peruvian Balsam; 0.5g of Phenazone, 30mg of phenol, 3.6mg of sodium salicylate and excipients (glycerol, sodium carbonate decahydrate, sodium benzoate, levomenthol, wool fat and purified water, according to the 2020 version of the French Vidal reference drug book. Male rats have served as animal material on which the burns were performed.

Induction and assessment of the burn

Quantitatively, the sampling selected albino rats of the wistar strain, from the animal facility of the Faculty of Medicine and Biomedical Sciences. There were 60 of them in the test phase. While 12 rats were included in the batch of untreated burns and 12 in that of unburned specimens the 36 remaining were divided into 9 rats per batch for treatment with Brulex and the 3 honey samples. Before this distribution, a pre-test was carried out with five batches of two 02 rats of the wistar strain. The realization of experimental burns, according to the method of Cai and al., was performed under general anaesthesia by intravenous administration of ketamine® and diazepam® for 15 to 20 minutes. It used a mass of of 100g and a round contact surface of 22mm in diameter. The mass was heated in boiling water (100 ° C) until thermal equilibrium (reached at 5 min). After heating, it is removed from the water, quickly wiped off, and then applied without pressure for 20 seconds at the place of election. The photos were taken using an XTIGI V10 telephone; the surface areas were calculated using the AUTOCARD 2014 software. The evaluation of healing by digital planimetry proceeded from the calculation and the percentage of retraction, using the equation developed by Gopinath and al. in 2004. We then calculated the average of the areas of the six wounds of the same batch at time t, in comparison to the area of the initial burn.

Statistical analysis and bioethical considerations

At the various dates selected, the wound surfaces calculated for each batch were expressed in the form of the mean ± standard deviation. For their comparison, we used Fisher's ANOVA test. The data was saved as a Microsoft Office Excel® 2013 file and then analyzed using SPSS version 20.0 software. From a bioethical point of view, the experimental protocol used in this study complies with the recommendations for animal care used in research and teaching of the NRC of 1996. In addition, this work has been validated by the institutional research ethics committee of the Faculty of Medicine and Biomedical Sciences, located in Yaounde.

RESULTS

Morphological parameters and healing of burns

In the evolution of the healing process (Table I and Figure 1), the group of rats treated with Brulex® showed the largest injured surfaces on day 0 (8.83 ± 2.34 cm2), the least extensive being those of the Okuh group (6.83 ± 0.66 cm^2). On the 24th day, the smallest areas were respectively and crescendo of the Ngaoundal, Yaounde and Okuh lots. While the rats in the Brulex® and negative control lots were not yet completely cured. Figure 1 represents the variations in the rate of contraction of the surfaces of the wounds of the Okuh, Brulex® and negative control groups. We observe that the rats of the Okuh batch have a faster surface contraction than that of the other two batches. Similarly, the same evolutionary pattern is observed in the comparison of two other batches with the Yaounde and Ngaoundal batches. For a comparison of the 3 therapeutic trials, it is observed that the rats of the Okuh batch were those which had a slower healing rate than that of the other two batches. But all of those rats were cured around the 22nd day. Figure 1 shows the macroscopic characteristics observed on the burns on days 0, 10 and 22. The wounds treated with honey had scabs at the start of the experiment, which ended up falling off after. Also in the negative control group, the wounds tended to dry quickly and crust, but with very slow healing.

DISCUSSION

We observed a real healing effect that was rapid and variable depending on the origin of the honey, but also in the three cases significantly better than that of Brulex®. This property of honey clinically constitutes proof of its addition as an important excipent, usable in cosmetology and pharmaceutical products for dermatological use. The efficacy of honey for the treatment of burns, already mentioned other authors [7-22], is currently valid in our local context. Because it is biocompatible and associated with established antimicrobial and antibacterial activity, the honey dressing of burns, wounds and loss of skin material is being suggested as a future solution to burn wound dressing.

CONCLUSION

We can conclude that the topical treatment of degree IIB burns was effective with the varieties of honey tested. In the evaluation of the scar process, the healing time is shorter, with a higher rate of contraction when batches of rats are treated with honey, compared to those treated with the reference drug.

Table I : Evolution of the burn surface size during experimentation for each group (in cm^2)

Day	Negative control	Brulex®	Ngaoundal	Okuh	Yaounde
0	7,69 ± 0,89	8,83 ± 2,34	7,76 ± 1,3	6,83 ± 0,66	7,80 ± 0,73
4	6,99 ± 1,515	8,14 ± 1,549	6,38 ± 0,483	6,16 ± 0,483	6,34 ± 0,906
8	5,95 ± 0,097	7,61 ± 2,130	5,56 ± 0,471	5,04 ± 0,602	4,93 ± 0,416
12	5,22 ± 0,964	6,14 ± 0,850	3,34 ± 0,103	3,62 ± 1,805	3,54 ± 1,091
16	3,49 ± 0,082	3,98 ± 0,205	1,21 ± 0,131	1,95 ± 0,710	1,02 ± 0,143
20	1,67 ± 0,777	2,51 ± 0,026	0,42 ± 0,204	0,79 ± 0,024	0,42 ± 0,101
24	0,80 ± 0,062	0,74 ± 0,247	0,0 ± 0,000	0,0 ± 0,000	0,00 ± 0,000

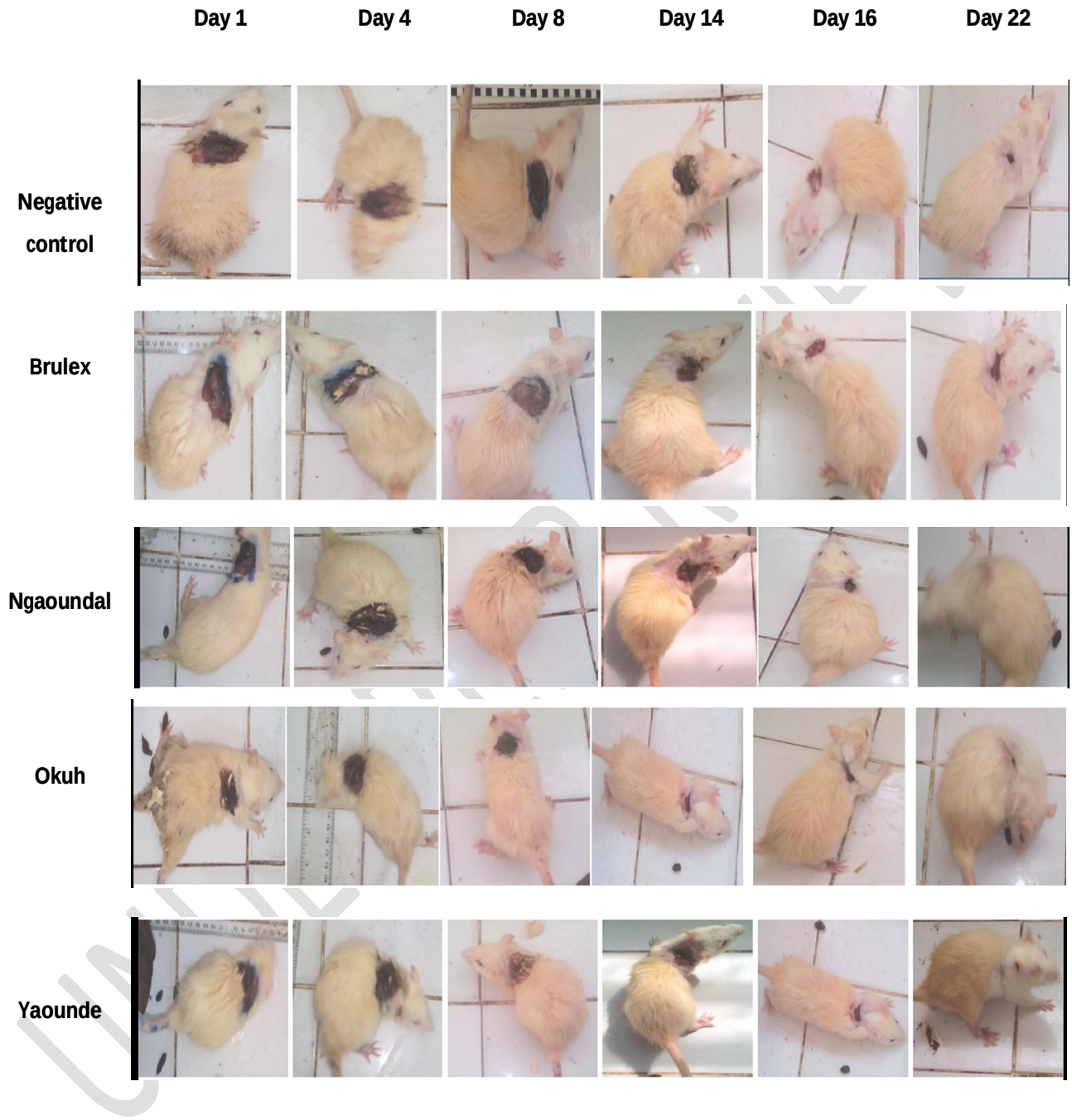

Figure 1 : General clinical wound evolution for each group

REFERENCES

1. World health organisation website on burns page. Consulted on the 15 January 2023 https://www.who.int/news-room/fact-sheets/detail/burns.

2. Rajeev B. Ahuja, Nicole Gibran, David Greenhalgh, James Jeng, David Mackie, Amr Moghazy, Naiem Moiemen, Tina Palmieri, Michael Peck, Michael Serghiou, Stuart Watson, Yvonne Wilson, Ariel Miranda Altamirano, Bechara Atieh, Alberto Bolgiani, Gretchen Carrougher, Dale Edgar, Linda Guerrero, Marella Hanumadass, Lisa Hasibuan, Helma Hofland, Ivette Icaza, Leo Klein, Hajime Matsumura, Richard Nnabuko, Arash Pirat, Vinita Puri, Nyoman Putu Riasa, Fiona Wood, Jun Wu, Xia Zhao-Fan, Paul van Zuijlen International Society for Burn Injuries (ISBI) Practice Guidelines Committee, *Burns*, Volume 42, Issue 5, 2016, Pages 953-1021, ISSN 0305-4179 https://doi.org/10.1016/j.burns.2016.05.013. (https://www.sciencedirect.com/science/article/pii/S0305417916301449)
3. Amy Hughes, Stian Kreken Almeland, Thomas Leclerc, Takayuki Ogura, Minoru Hayashi, Jody-Ann Mills, Ian Norton, Tom Potokar. Recommendations for burns care in mass casualty incidents: WHO Emergency Medical Teams Technical Working Group on Burns (WHO TWGB) 2017-2020, *Burns*, Volume 47, Issue 2, 2021, Pages 349-370, ISSN 0305-4179. https://doi.org/10.1016/j.burns.2020.07.001. (https://www.sciencedirect.com/science/article/pii/S0305417920304599)
4. Beyiha G., Binam F.,Batamack J.F., Sosso M.A. Traitement et pronostic de la brulure grave au centre des grands brules de douala, cameroun. *Annals of Burns and Fire Disasters* - vol. XIII - n. 3 - September 2000 P1-10.
5. Norman_G, Christie_J, Liu_Z, Westby_MJ, Je!eries_JM, Hudson_T, Edwards_J, Mohapatra_DP, Hassan_IA, Dumville_JC. Antiseptics for burns. *Cochrane Database of Systematic Reviews* 2017, Issue 7. Art. No.: CD011821. DOI: 10.1002/14651858.CD011821.pub2.
6. Giulia Papa, Roberto Maier, Alessandra Durazzo, Massimo Lucarini, Ioannis K. Karabagias, Manuela Plutino, Elisa Bianchetto, Rita Aromolo, Giuseppe Pignatti, Andrea Ambrogio, Marco Pellecchia and Ilaria Negri. The Honey Bee Apis mellifera: An Insect at the Interface between Human and Ecosystem Health. *Biology* 2022, 11(2), 33. https://doi.org/10.3390/biology11020233
7. Nicolaas J Basson and Sias R Grobler. Antimicrobial activity of two South African honeys produced from indigenous Leucospermum cordifolium and Erica species on selected micro-organisms. *BMC Complementary and Alternative Medicine* 2008, 8:41 doi:10.1186/1472-6882-8-41 http://www.biomedcentral.com/1472-6882/8/41.

8. Yan-Teng Khoo, Ahmad Sukari Halim, Kirnpal-Kaur B Singh, Noor-Ayunie Mohamad. Wound contraction effects and antibacterial properties of Tualang honey on full-thickness burn wounds in rats in comparison to hydrofibre. *BMC Complementary and Alternative Medicine* 2010, 10:48. http://www.biomedcentral.com/1472-6882/10/48

9. A.Werner et O. Laccourreye. Le miel en oto-rhino-laryngologie : quand, pourquoi et comment ? *Annales françaises d'Oto-rhino-laryngologie et de Pathologie Cervico-faciale.* Volume 128, Issue 3, June 2011, Pages 153-157 https://doi.org/10.1016/j.aforl.2010.12.013Get rights and content

10. Molan PC. The evidence and the rationale for the use of honey as a wound dressing. *Wound Practice and Research.* Volume 19 Number 4 – December 2011 Pages 204-220

11. Vallianou NG, Gounari P, Skourtis A, Panagos J, Kazazis C (2014) Honey and its Anti-Inflammatory, Anti-Bacterial and Anti-Oxidant Properties. *Gen Med (Los Angel)* 2: 132. doi: 10.4172/2327-5146.1000132

12. Tolga Eser, Cemil Kavalci, Cem Aydogan and Afsin Emre Kayipmaz. Epidemiological and cost analysis of burn injuries admitted to the emergency department of a tertiary burn center. Eser et al. *SpringerPlus* (2016) 5:1411Pages 1-6. DOI 10.1186/s40064-016-3107-3

13. MG Miguel1, MD Antunes1,2 and ML Faleiro3. Honey as a Complementary Medicine*. Integrative Medicine Insights*. 2017. Volume 12: 1–15. DOI: 10.1177/1178633717702869

14. Eyarefe, OD; Oguntoye, CO. Honey, an unexplored topical wound dressing agent in Nigerian veterinary practice. *Sokoto Journal of Veterinary Sciences* ISSN : 1595-093X. 2018 Volume: 14 Issue: 3 Page: 149922 - 149922

15. Galit H. Frydman, David Olaleye, Damodaran Annamalai, Kim Layne, Illina Yang, Haytham M. A. Kaafarani & James G. Fox. Manuka honey microneedles for enhanced wound healing and the prevention and/or treatment of Methicillin□resistant Staphylococcus aureus (MRSA) surgical site infection. *Scientific Reports* (2020) 10:13229. https://doi.org/10.1038/s41598-020-70186-9

16. Paulus H. S. Kwakman and Sebastian A. J. Zaat. Critical Review. Antibacterial Components of Honey. *IUBMB Life*, 64(1): 48–55, January 2012. ISSN 1521-6543 print/ISSN 1521-6551 online DOI: 10.1002/iub.578

17. Manisha Deb Mandal, Shyamapada Mandal. Honey: its medicinal property and antibacterial activity. *Asian Pac J Trop Biomed* 2011; 1(2): 154-160. doi:10.1016/S2221-1691(11)60016-6.

18. Iraj Ahmadi, Hossein Foruozandeh and Fatemeh Yekke . Evaluation the Healing Potential of Oleuropein on Second-Degree BurnWounds in a Rat Model. *Jundishapur J Nat Pharm Prod*. 2022 February; 17(1):e114568. Published online 2021 September 10. doi: 10.5812/jjnpp.114568

19. Scepankova, H.; Combarros-Fuertes, P.; Fresno, J.M.; Tornadijo, M.E.; Dias, M.S.; Pinto, C.A.; Saraiva, J.A.; Estevinho, L.M. Role of Honey in AdvancedWound Care. *Molecules* 2021, 26, 4784. https://doi.org/10.3390/molecules26164784

20. Hanaa Tashkandi. Honey in wound healing: An updated review. *Open Life Sciences* 2021; 16: 1091–1100.

21. Esa, N.E.F.; Ansari, M.N.M.; Razak, S.I.A.; Ismail, N.I.; Jusoh, N.; Zawawi, N.A.; Jamaludin, M.I.; Sagadevan, S.; Nayan, N.H.M. A Review on Recent Progress of Stingless Bee Honey and Its Hydrogel-Based Compound for Wound Care Management. *Molecules*. 2022, 27, 3080. https://doi.org/10.3390/molecules27103080.

22. Xiaoxuan Deng | Maree Gould | M. Azam Ali. A review of current advancements for wound healing: Biomaterial applications and medical devices. *J Biomed Mater Res.* 2022;110:2542–2573.