Burn Wound Healing Effect and Hair Growth Promoting Activity of *Lawsonia inermis* L. and Honey in *Oryctolagus cuniculus* Rabbits

1,2Zouhir Djerrou, 1Imane Mokhbi, 1Khadidja Saci Hadef, 1Noudjoud Boutoba, 1Saïda Bouzeguine, 1Ilhem Brighet and 1Besma Khelfa

1Department of Sciences of Nature and Life, Faculty of Sciences, University of August 20th 1955, Skikda, Algeria
2Laboratory of Pharmacology and Toxicology, University of Mentouri Constantine, Algeria

Abstract: This study aimed to assess *Lawsonia inermis* L. and honey mixture effects on burn wounds and hair growth in rabbits. Nine male *Oryctolagus cuniculus* local rabbits were allocated randomly in 3 groups. After antisepsis and local anesthesia, four circular burns were created on the animal’s backs. Immediately after burning, the wounds were covered with honey in three rabbits (HON group), another group was treated first with honey and then *L. inermis* powder was added on honey (LI_HON group). Three untreated animals were used as control (CRL group). The treatment of rabbits was applied once daily (6/7 days) until 24th day post burns. The animals were observed for their general state, aspects of wounds and the healing times were also recorded. At 35th day post burns, the hair growth was investigated in the different groups. LI_HON group has recorded the best results compared to other groups; a short significant necrosis duration (LI_HON Vs CRL, p<0.001; LI_Hon Vs HON, p<0.05), precocious start of 1st crust detachment (LI_HON Vs CRL, p<0.05; LI_Hon Vs HON, p<0.001), a quick complete detachment of the first crust (LI_HON Vs CRL, p>0.05; LI_Hon Vs HON, p<0.05) and a significant reduction of healing time (LI_HON Vs CRL, p<0.05; LI_Hon Vs HON, p<0.01). LI-HON group has shown the best promoting activity of hair growth in term of recovered surface percentage (LI_HON Vs CRL, p = 9.13038E−7; LI_Hon Vs HON, P = 1.70745E−7) and hair length (LI_HON Vs CRL, p<0.01; LI_Hon Vs HON, p<0.001). The study concludes that the topic use of *L. inermis* powder and honey simultaneously accelerates burn wound healing process in rabbit’s model. Henna has also shown a remarkable hair growth promoting activity in term of hair recovered area and hair length.

Keywords: *Lawsonia inermis* L., Honey, Burns, Wound Healing, Hair Growth

Introduction

Wound healing is a natural and dynamic process; it occurs in three fundamental steps including: inflammation, proliferative and remodeling phases (Wulff and Wilgus, 2013). Inflammation includes coagulation and inflammatory cell recruitment. The fibrin clot formed contains fibrin molecules, fibronectin, vitronectin and thrombospondins; it hosts the active arriving cells and different cytokines and growth factors (Reviewed by Broughton et al., 2006). Macrophages are present throughout all stages of the healing process, producing a variety of substances that regulate healing including growth factors, prostaglandins and complement factors (Nathan, 1987). Numerous growth factors, cytokines and chemokines implicated in the execution and regulation of the wound healing stages were investigated. (Barrientos et al., 2008). The physiological processes of angiogenesis, granulation tissue formation, epithelialization, collagen synthesis and wound
contraction characterized the proliferative phase (Broughton et al., 2006). The wound healing cascade finishes by remodeling phase which occurs in three weeks to several years according to wound’s complexity (Clark, 1988). The apoptosis of cells in this step prevent the appearance of hypertrophic scar or keloid; in normal conditions a mature wound was formed and characterized as avascular and acellular (Broughton et al., 2006).

According to Khorasani et al. (2008), thermal burn injury represents a major cause of death and disability and may cause a high costs in health care. The conventional drugs used in the management of burn injuries were reported to exert unwanted side effects (Fan et al., 2015).

Many of today’s modern drugs have their origin in traditional plant medicine (Blanks et al., 1998). The use of plants for the healing purposes cited around 3000 B.C, in China, Egypt and subcontinent (Morse, 1934).

Henna is a tall shrub or small flowering tree. The scientific name of this plant is Lawsonia inermis L., which belongs to Lythraceae family. It is a shrub that grows in North Africa and Middle East (Gibbons et al., 2005). Henna is native to a number of tropical regions in Asia, Northern Africa and Australia. It is naturalized and cultivated in the tropics of America, Egypt, India and parts of the Middle East (Muhammad and Muhammad, 2005). The different parts of henna have been reported to exert antioxidant, hypoglycemic, antidiabetic, hepatoprotective, antibacterial, antifungal, anticancer and wound healing properties (Majtan et al., 2013; Hadagali and Chua, 2014; Rajashri and Sachin, 2014; El Bergadi et al., 2015; Devasvaran and Yong, 2016; Mohamed et al., 2016).

Honey is a natural product of Apis mellifera, it is prescribed in external applications and in the management of wound healing since the ancient times, from ancient Egypt, Chinese, Indian, to Greek civilizations (Siedentopp, 2009). Several studies have focused the pharmacological properties of honey including antibacterial activities (Molan, 1992; Khoo et al., 2010; Al-Nahari et al., 2015), antifungal (Irish et al., 2006; Boukraa et al., 2008), anti-inflammatory (Beretta et al., 2010), antioxidant (Al-Mamary et al., 2002; Gheldof and Engeseth, 2002), hypoglycemic and hypolipidemic (Al-Waili, 2004), healing activities (Khoo et al., 2010), cardioactive, vasoactive effects (Rakha et al., 2008) and anticancer activity (Fernandez-Cabezudo et al., 2013; Erejuwa et al., 2014).

The aim of the present study is to evaluate the effect of a combination of honey bee and Lawsonia inermis on burn wound healing and to investigate the possible hair growth promoting activity in rabbit’s model.

Materials and Methods

Drugs

200 g of commercial Lawsonia inermis L. powder was purchased from local market. It is an imported product from Pakistan under the name “Special mehndi”, fabrication date: October, 2013 and expiration date: September, 2018. Honey was obtained from a local bee keeper from Collo, East of Algeria.

Animals and Housing Conditions

The study was conducted on 9 healthy male, adult, Oryctolagus cuniculus local rabbits weighing 1500 to 1800 g at the beginning of experiment. The animals were kept in normal conditions of temperature and lighting (12 h light: 12 h dark) and given laboratory food and water, ad libitum during the study. The experimental protocol was approved by the scientific committee of the department of sciences of nature and life, faculty of sciences, University of August, 20th 1955, Skikda, Algeria.

Fig. 1. Localisation of cranial and caudal burns
**Experimental Design**

Three days before burning, the animal backs were clipped and a depilatory cream was used to remove all the hairs from the rabbit’s skin. On day zero, a local anesthesia was practiced for all rabbits using xylocain 2% (s/c) after disinfectant by surgical alcohol 70%. Then, four burns of identical size (22 mm in diameter) were created on the back of each animal, two cranially (left and right) and two caudally (left and right) (Fig. 1), by a cylinder metal weighing 200 g immersed in prior in boiled water for 3 min and maintained on animal skin 15 sec (Djerrou et al., 2010).

**Treatment and Assessment of Healing Process**

The animals were randomly allocated into three groups, the first untreated was used as control (CRL group), in the second 3 rabbits (HON group), the burns were immediately covered by a thin layer of honey (≈1 mm), the third group was treated as HON group and then honey was covered by 0.5 g of *Lawsonia inermis* L. powder. All these drugs were applied topically slowly and they were repeated once daily until 24th day post burns for non healed burns. The clinical aspects of wounds and the different healing times were noted. Photographs were taken from the wounds on days 0, 8, 12 and 20.

**Hair Growth Assessment**

At 35 day post burns, the hair growth was compared in the different rabbits; the percentages of recovered areas were calculated and the lengths of back’s new hair were compared with the lengths of hairs in the left and right normal sides of animals.

**Statistical Analysis**

The results were expressed as mean with their variance. One-way Analysis Of Variance (ANOVA) was used to compare the different group means. A value of p<0.05 was considered statistically significant.

**Results**

Generally, there was a favorable trend towards healing in different rabbits either treated or untreated. Body weight of the animals has recorded a significant decrease during the first 15 days after burns in different groups; noting a return to the original state that was obtained within 2 weeks. One rabbit from LI-HON group showed a dermal reaction following application of the depilatory and that has healed at the 6th day post burns (Fig. 2), the same rabbit also expressed a skin thickening 15 days after the beginning of the application of honey and henna mixture.

**Healing Process**

The results of healing process recorded in Table 1 and Fig. 3 showed that application of *Lawsonia inermis* and honey simultaneously stimulated significantly the healing process in its different stages compared to honey applied only or untreated animals. HON group has enregisted a significant reduction (p<0.005) of necrosis duration compared to CRL group but no significant differences were observed between these two groups in the processes of crust detachment or the total healing time. LI_HON group has recorded the best results compared to other groups; a short significant necrosis duration (LI_HON Vs CRL, p<0.001; LI_Hon Vs HON, p<0.05), precocious start of 1st crust detachment (LI_HON Vs CRL, p<0.05; LI_Hon Vs HON, p<0.001), a quick complete detachment of the first crust (LI_HON Vs CRL, p>0.05; LI_Hon Vs HON, p<0.01) and a significant reduction of healing time (LI_HON Vs CRL, p<0.05; LI_Hon vs HON, p<0.01).

**Hair Growth**

The results relative to hair growth registred in Table 2 and illustrated by Fig. 4 showed a real promoting activity of hair growth in LI-HON group in term of recovered surface percentage (LI_HON Vs CRL, p = 9.13038E⁻⁷; LI_Hon Vs HON, P = 1.70745E⁻⁷), the untreated rabbits have recorded a significant hair growth compared to HON group (p<0.01), however, there was no significant difference between these two groups in term of new hair length (p>0.05). In LI_HON group, the new hair length has reached a very significant difference compared to other groups (LI_HON Vs CRL, p<0.01; LI_Hon Vs HON, p≤0.001).
Fig. 3. Photographs of some skin wounds of control group, honey group and *Lawsonia inermis* + honey group at 0, 8, 12 and 20 days after burns. D: Day, CRL: Control (untreated animals), HON: Honey group, LI_HON: *Lawsonia inermis* + honey treated rabbits

Fig. 4. Photographs of hair growth in the different groups at 27th day post burn

Table 1. Evolution of healing process in control and treated groups

|                  | CRL       | HON       | LI_HON     |
|------------------|-----------|-----------|------------|
| Necrosis         | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     |
|                  | 10.75     | 0.916     | 8          | 0.666     | 6.25        | 0.916     |
| Start of 1st crust detachment | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     |
|                  | 12.5      | 1.66      | 13.5       | 1.66      | 10.5        | 0.333     |
| Complete detachment of 1st crust | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     |
|                  | 21,375    | 17,696    | 19,75      | 1,583     | 17,25       | 1,583     |
| Healing time     | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     | Mean (n = 12) | Var.     |
|                  | 24.5      | 12        | 25         | 6         | 19.25       | 1,583     |

Statistical data (P value)

- HON Vs CRL: 0.004
- LI_HON Vs CRL: 5.599 E^-4
- LI_Hon Vs HON: 0.031

CRL: Control (untreated animals), HON: Honey group, LI_HON: *Lawsonia inermis* + honey treated rabbits
Discussion

Healing Process

The effect of honey applied alone was significant compared to control in the first 10 days of healing process with no infection of wounds. This period correspond to hemostasis and inflammatory phases (through 4 to 6 days post burns) and a part of the proliferative phase (which prolonged until 14th day) (Broughton et al., 2006). This result could be a consequence of honey pharmacological properties demonstrated in several studies. According to Namias (2003), honey has anti-inflammatory effect, promotes granulation tissue formation and exert antibacterial activity. The anti-inflammatory property may be associated with the antioxidant content of honey (Tanaka et al., 1995). The high osmolarity of honey has been considered a valuable tool in the management of sloughy and septic wounds; it produces a cleansing effect and naturally debrides non-viable tissue. A reduction but not significantly of time of first crusts detachment was recorded in honey group compared to control; this later has recorded a better healing time but not significantly than that of honey. Several studies have explained the effects of honey on granulation tissue formation and epithelialization by the generation of hydrogen peroxide (stimulation of angiogenesis), the growth of fibroblasts and the wound acidification. The nutrient content of honey may also stimulate growth because it has a wide range of amino acids, vitamins and trace elements (Reviewed by Molan, 1999). The addition of L. inermis on honey in the third group has resulted in very significant results in the different stages of wound healing. Phytochemical screening of henna plant has revealed the presence of numerous chemicals including alkaloids, tannins, flavonoids, steroids, glycosides, saponins etc. (Ibrahim et al., 2008; Arun et al., 2010). According to Muhammad and Muhammad (2005), the plant constituent are made up of mnnite, tannic acid, mucilage and gallic acid, but the main constituent is 2-hydroxynaphthoquinone (lawsonie), known to be the major bioactive constituent of this plant. In an in vitro study, the henna leaves extracts were able to inhibit the growth pattern of Aspergillus niger and Fusarium oxysporum, Streptococcus sp. and Staphylococcus aureus which are the primary invaders of burnt wounds. Inhibition of these microorganisms’ growth suggests that henna may be valuable in the management of burnt wound infections (Muhammad and Muhammad, 2005). Chloroform extract of leaves of L. inermis had shown a highest antioxidant activity compared to α-tocopherol (Endrini et al., 2007). In addition, L. inermis L. extract exhibited absolute toxicity and showed broad fungitoxic spectrum when tested against 13 ring worm fungi (Singh and Pandey, 1989). A significant analgesic and antipyretic activities were also shown with leaves extract of this plant (Molsin et al., 1989). In another study, isopombagin and lawsaritol, isolated from stem bark and root of L. inermis L. showed anti-inflammatory activity against Carrageenan induced paw edema in rats (Gupta et al., 1993). In a study of Nayak et al. (2007), the healing activity of L. inermis extract was compared with the control and reference standard animals; the results showed a positive effects on term of wound contraction, epithelialization period, skin breaking strength, granulation tissue weight and hydroxyproline content in this plant group. The authors have conducted a histological study which has showed increased and well organized bands of collagen, more fibroblasts and few inflammatory cells in henna group.

Hair Growth

Honey group has recorded a significant delay in hair growth, compared to control in term of percentage of recovered area, but no statistical significance was observed in comparing new hair length to normal hair length; this later may be due to individual characteristics and/or due to stress because these animals were manipulated more than the untreated animals. The real hair growth promoting activity recorded in LI_HON group is due to Lawsonia inermis powder that spreads, after it’s sprinkling on the honey, in the whole surface of the shaved back. L. inermis has been cited as a growth accelerator and was used in an ancient Egyptian formula to cure the loss of hair. Henna has also been recognized to act as a very good conditioner to the hair.
participated in laboratory experiments. Ahmadian, S. and M.A. Fakhree, 2009. Henna material. The corresponding author confirms that all of above described mechanisms of influence of the different hair-cycle stages on skin wound healing may be implicated in the interpretation of the present study results.

Conclusion

The mixture of Lawsonia inermis L. powder and honey stimulates burn wound healing process in its different stages. In addition to this, henna promotes hair growth. Further studies, including immunohistochemical investigations are required to confirm and elucidate the mechanism behind this promoting hair growth activity.

Acknowledgement

We would like to thank the staff of Department of Sciences of Nature and Life, University of August 20th 1955 Skikda, for help in achieving the experiment.

Author’s Contributions

Zouhir Djerrou: The first author designed and supervised the study and assisted in data analysis and manuscript preparation.

Imane Mokhbi: Sample collection and laboratory experiments.

Khadija Saci Hadeff: Sample collection and participated in laboratory experiments.

Noudjoud Boutebza, Saïda Bouzeguine, Ilhem Bright and Besma Khelfa: Participated in laboratory experiments.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues involved.

References

Ahmadian, S. and M.A. Fakhree, 2009. Henna (Lawsonia inermis) might be used to prevent mycotic infection. Med. Hypothesis, 73: 629-630. DOI: 10.1016/j.mehy.2009.06.001

Al-Mamary, M., A. Al-Meeiri and M. Al-Habori, 2002. Antioxidant activities and total phenolics of different types of honey. Nutr. Res., 22: 1041-1047. DOI: 10.1016/S0271-5317(02)00406-2

Al-Nahari, A.A.M., S.B. Almasaudi, ES.M.A. El-Ghany, E. Barbour and S.K. Al Jaouni et al., 2015. Antimicrobial activities of Saudi honey against Pseudomonas aeruginosa. Saudi J. Biol. Sci., 22: 521-525. DOI: 10.1016/j.sjbs.2015.04.006

Al-Waili, N.S., 2004. Natural honey lowers plasma glucose, C-reactive protein, homocysteine and blood lipids in healthy, diabetic and hyperlipidemic subjects: Comparison with dextrose and sucrose. J. Med. Food, 7: 100-107. DOI: 10.1089/109662004322984789

Aln-Agha, T., A. Aldeya, K. Alarifi, M. Al-Mamary, M. Al-Meeiri and M. Al-Habori, 2002. Antioxidant activities and total phenolics of different types of honey. Nutr. Res., 22: 1041-1047. DOI: 10.1016/S0271-5317(02)00406-2

Ansell, D.M., J.E. Kloepper, H.A. Thomason, R. Paus and M.J. Hardman, 2011. Exploring the “hair growth-wound healing connection”: Anagen phase promotes wound re-epithelialization. J. Invest. Dermatol., 131: 518-528. DOI: 10.1038/jid.2010.291

Arun, P., K.G. Purushotham, J. Jayarani and V. Kumari, 2010. In vitro antibacterial activity and flavonoid contents of Lawsonia inermis (Henna). Int. J. Pharmaceutical Technol. Res., 2: 1178-1181.

Barrientos, S., O. Stojadinovic, M.S. Golinko, H. Brem and M. Tomic-Canic, 2008. Growth factors and cytokines in wound healing. Wound Repair Regen., 16: 585-601. DOI: 10.1111/j.1524-475X.2008.00410.x

Beretta, G., F. Gelmini, V. Loddi, A. Piazzalunga and R. Maffei Facino, 2010. Profile of Nitric Oxide (NO) metabolites (nitrate, nitrite and N-nitroso groups) in honeys of different botanical origins: Nitrate accumulation as index of origin, quality and of therapeutic opportunities. J. Pharm. Biomed. Anal., 53: 343-349. DOI: 10.1016/j.jpba.2010.04.010

Blanks, T., S. Brown, B. Cosgrave, J. Woody and V. Bentley et al., 1998. The Body Shop Book of Wellbeing: Mind, Body and Soul. 1st Edn., Ebury, London, ISBN-10: 0091868173, pp: 256.

Boukraa, L., H. Benbarek and M. Ahmed, 2008. Synergistic action of starch and honey against Candida albicans in correlation with diastase number. Braz. J. Microbiol., 39: 40-43. DOI: 10.1590/S1517-83822008000100010

Broughton, G., J.E. Janis and C.E. Attinger, 2006. Wound healing: An overview. Plastic Reconstr. Surgery, 117: 1e-S-32e-S. PMID: 16801750

Chen, P., M. Cescon and P. Bonaldo, 2015. Lack of collagen VI promotes wound-induced hair growth. J. Invest. Dermatol., 135: 2358-2367. DOI: 10.1038/jid.2015.187

Clark, R.A.F., 1988. Overview and General Considerations of Wound Repair. In: The Molecular and Cellular Biology of Wound Repair, Clarke, R.A.F. and P.M. Henson (Eds.), Plenum, New York, pp: 3-33.
Devasvaran, K. and Y.K. Yong, 2016. Anti-inflammatory and wound healing properties of Malaysia Tualang honey. Curr. Sci., 110: 47-51.

Djerrou, Z., Z. Maamuri, Y. Hamdi-Pacha, M. Serakta and F. Riacho et al., 2010. Effect of virgin fatty oil of Pistacia lentiscus on experimental burn wound's healing in rabbits. Afr. J. Tradit. Complement., 7: 258-263. PMID: 21461154

El Bergadi, F., F. Laachari, M. Sadiki, A. Megzari and H.M. El Abed Iraqui et al., 2015. Antifungal effect of Moroccan Lawsonia inermis leaf extracts on the growth of filamentous fungi isolated from historical wood. Int. J. Curr. Res., 7: 14237-14240.

Endrini, S., A. Rahmat, P. Ismail and Y.H. Taufiq-Yap, 2014. The anti-inflammatory and wound healing properties of honey. Eur. J. Med. Sci., 7: 1098-1102. DOI: 10.3923/jms.2007.1098.1102

Erejuwa, O.O., S.A. Sulaiman and M.S. Ab Wahab, 2007. Comparing of the cytotoxicity properties and mechanism of Lawsonia inermis and Sambucus crpispus extract against several cancer cell lines. J. Med. Sci., 7: 1098-1102. DOI: 10.3923/jms.2007.1098.1102

Fan, Z.W., Y.X. Pang, K. Wang, F.L. Yu and D. Wang et al., 2015. Blumea balsamifera oil for the acceleration of healing of burn injuries. Molecules, 20: 17166-17179. DOI: 10.3390/molecules20091766

Fernandez-Cabezudo, M.J., R. El-Kharrag, F. Torab, G. Bashir and J.A. George et al., 2013. Intravenous administration of manuka honey inhibits tumor growth and improves host survival when used in combination with chemotherapy in a melanoma mouse model. PLoS One, 8: e55993-e55993. DOI: 10.1371/journal.pone.0055993

Ghelfdof, N. and N.J. Engeseth, 2002. Antioxidant capacity of honeys from various floral sources based on the determination of oxygen radical absorbance capacity and inhibition of in vitro lipidprotein oxidation in human serum samples. J. Agric. Food Chem., 50: 3050-3055. DOI: 10.1021/jf0114637

Gibbons, L.G., R.H. Gopalla, A. Hunter, K. Kerr and P.M. Mulrey, 2004. Plants Encyclopedia. 1st Edn., US, pp: 232.

Gupta, S., M. Ali, K.K. Pillai and M.S. Alam, 1993. Evaluation of antiinflammatory activity of some constituents of Lawsonia inermis. Fitoterapia, 64: 365-366.

Hadagali, M.D. and L.S. Chua, 2014. The anti-inflammatory and wound healing properties of honey. Eur. Food Res. Technol., 239: 1003-1014. DOI: 10.1007/s00217-014-2297-6

Ibrahim, M., A.J. Hameed and A. Jalbout, 2008. Molecular spectroscopic study of river nile sediment in the greater Cairo region. Applied Spectroscop., 62: 306-311. DOI: 10.1366/00037020878359795

Irish, J., D.A. Carter, T. Shokohi and E.S. Blair, 2006. Honey has an antifungal effect against Candida species. Med. Mycol., 44: 289-291. DOI: 10.1080/13693780500417037

Khoo, Y.T., A.S. Halim, K.K.B. Singh and N.A. Mohamad, 2010. Wound contraction effects and antibacterial properties of Tualang honey on full-thickness burn wounds in rats in comparison to hydrofibre. BMC Complement. Altern. Med., 10: 48-48. DOI: 10.1186/1472-6882-10-48

Khorasani, G., S.J. Hosseinimehr, P. Zamani, M. Ghasemi and A. Ahmadi, 2008. The Effect of Saffron (Crocus Sativus) extract for healing of second-degree burn wounds in rats. Keio J. Med., 57: 190-195. DOI: 10.2302/kjm.57.190

Majtan, J., J. Bohova, R. Garcia-Villalba, F. Tomas-Barberan and Z. Madakova et al., 2013. Fir honeydew honey flavonoids inhibit TNF-α-induced MMP-9 expression in human keratinocytes: A new action of honey in wound healing. Arch. Dermatol. Res., 365: 619-627. DOI: 10.1007/s00403-013-1385-y

Mohamed, M.A., I.M. Taj Eldin, A.E.H. Mohammed and H.M. Hassan, 2016. Effects of Lawsonia inermis (L. (Henna) leaves’ methanolic extract on carbon tetrachloride-induced hepatotoxicity in rats. J. Intercult. Ethnopharmacol., 5: 22-26. DOI: 10.5455/jice.20151123043218

Mohsin, A., A.H. Shah, M.A. Al-Yahya, M. Tariq and M.O.M. Tanira et al., 1989. Analgesic, antipyretic activity and phytochemical screening of some plants used in traditional Arab system of medicine. Fitoterapia, 60: 174-177.

Molan, P.C., 1992. The antibacterial activity of honey: 1. The nature of the antibacterial activity. Bee World, 73: 5-28. DOI: 10.1080/0005772X.1992.11099109

Molan, P.C., 1999. The role of honey in the management of wounds. J. Wound Care, 8: 415-418. DOI: 10.12968/jowc.1999.8.8.25904

Morse, W.R., 1934. Chinese Medicine (Clio Medica). New York.

Muhammad, H.S. and S. Muhammad, 2005. The use of Lawsonia inermis linn. (henna) in the management of burn wound infections. Afr. J. Biotechnol., 4: 934-937.

Namias, N., 2003. Honey in the management of infections. Surg. Inf., 4: 219-26. DOI: 10.1089/109629603766957022

Nathan, C.F., 1987. Secretory products of macrophages. J. Clin. Invest., 79: 319-326. DOI: 10.1172/JCI112815
Nayak, B.S., G. Isitor, E.M. Davis and G.K. Pillai, 2007. The evidence based wound healing activity of *Lawsonia inermis* Linn. Phytotherapy Res., 21: 827-831. DOI: 10.1002/ptr.2181

Rajashri, R.T. and A.N. Sachin, 2014. Antifungal activity of *Nigella sativa* and *Lawsonia inermis* and its comparative study. World. J. Pharm. Res., 3: 1468-1472.

Rakha, M.K., Z.I. Nabil and A.A. Hussein, 2008. Cardioactive and vasoactive effects of natural wild honey against cardiac malperformance induced by hyperadrenergic activity. J. Med. Food, 11: 91-98. DOI: 10.1089/jmf.2006.172

Siedentopp, W., 2009. Honey: Effective against inflammation, cough and hoarseness. Deutsche Zeitschrift fuer Akkupunktur.

Singh, V.K. and D.K. Pandey, 1989. Fungitoxic studies on bark extract of *Lawsonia inermis* against ringworm fungi. Hindusthan Antibiot. Bull., 31: 32-35.

Tanaka, H., M. Hanumadass, H. Matsuda, S. Shimazaki and R.J. Walter *et al.*, 1995. Hemodynamic effects of delayed initiation of antioxidant therapy (beginning two hours after burn) in extensive third-degree burns. J. Burn Care Rehabil., 16: 610-615. DOI: 10.1097/00004630-199511000-00010

Wulff, B.C. and T.A. Wilgus, 2013. Mast cell activity in the healing wound: More than meets the eye? Exp. Dermatol., 22: 507-510. DOI: 10.1111/exd.12169