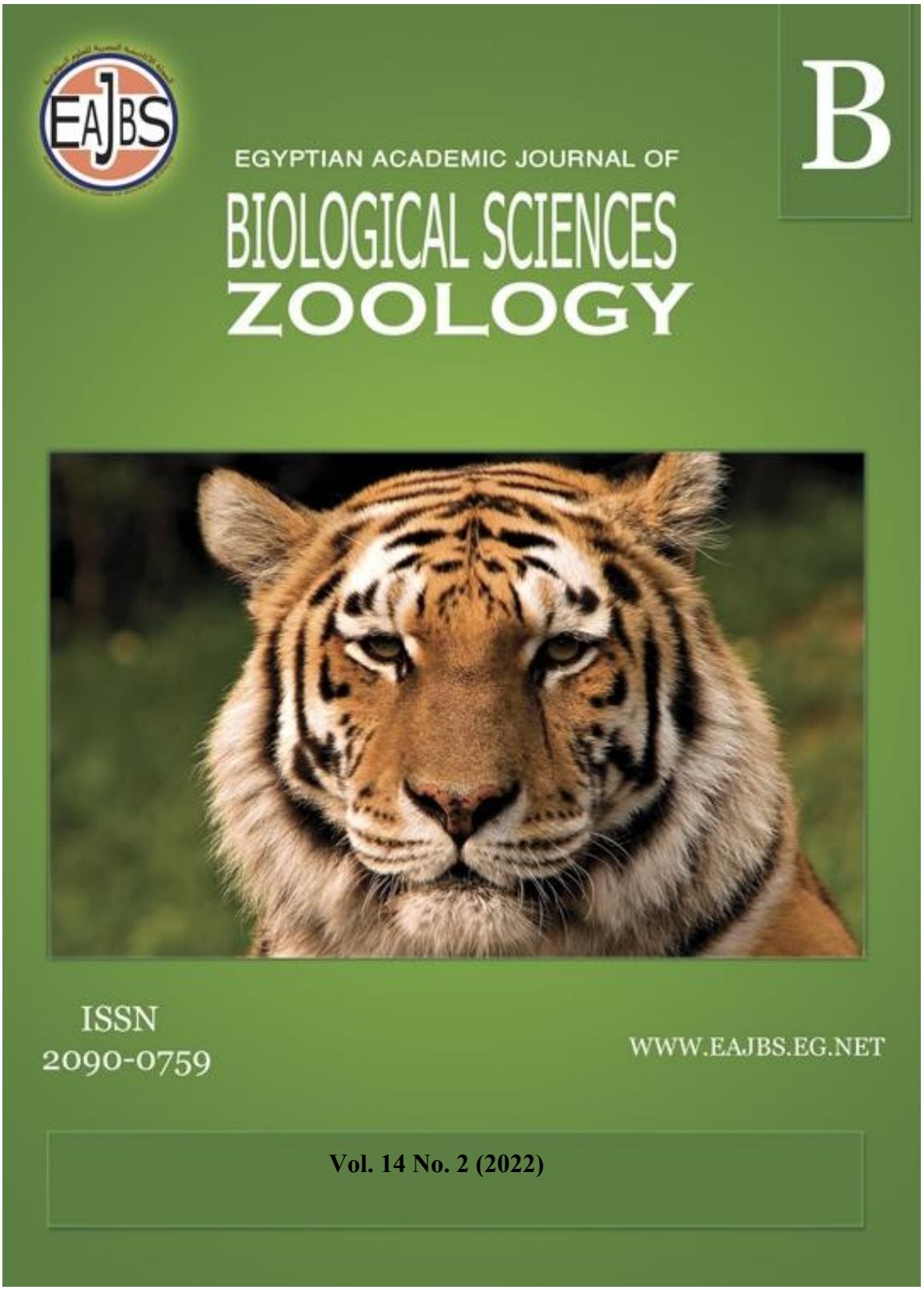

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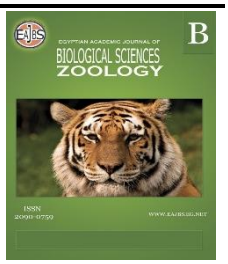

Contribution to the Study of The Healing Effect of *Zingiber officinale* Extracts on an Animal Model

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ABSTRACT

Natural plant extracts contain a variety of biologically active molecules. In this context, and within the framework of the valorization of the extracts of the plant Zingiber officinale, our work was carried out on the study of the cicatrization of the cutaneous pleasures caused in the Wistar rats by comparing them with a batch control (without any treatment) and a batch positive (subjected to a treatment of references).The evaluation of the healing power of the different batches shows that our tested extract has a remarkable The evaluation of the healing power of the different batches shows that our tested extract has remarkable healing potential, which is justified by the values of wound contraction over a period of 15 days.

Finally, it should be noted that this work will open up targeted research horizons in the field of plants used in traditional medicine in arid zones.This work will open up new research horizons in the field of plants used in traditional medicine in arid zones, particularly in terms of highlighting their active principles and evaluating their biological activities.

By means of this study, our aspiration is to have provided a humble contribution towards promoting traditional medicine, with the aim of rendering efficacious and accessible herbal remedies to the populace.

INTRODUCTION

Natural products are increasingly playing an important role in the pharmaceutical and agronomic industries. Research strategies for active natural substances from plants are often based on ethnobotanical approaches, highlighting the use of these plants in traditional medicine by local populations (Sahli, R., 2017). Plants represent an inexhaustible source of traditional and effective remedies due to the active principles they contain, such as alkaloids, flavonoids, heterosides, saponins, quinones, vitamins, and essential oils (Ouis, N., 2015). These secondary metabolites play a decisive role in the adaptation of plants to their environment. They thus participate very effectively in the tolerance of plants to various stresses (pathogen and insect attacks, drought, UV light, etc.) (Hadj Salem, J., 2009). Plants are not always safe; they may appear innocuous but can be toxic or deadly to the body. Many medicinal plants and drugs are therapeutic at a certain dose and toxic at another (Elyebdri, N., Boumediou, A., & Addoun, S.2017). Phytochemicals are known to possess antioxidant, antibacterial, antifungal, antidiabetic, anti-inflammatory, anti-arthritic, and healing activities, and due to these properties, they are widely used for medicinal purposes (Gupta, A. *et al.*, 2012). Due to the side effects of antimicrobial chemicals and the resistance that pathogenic microorganisms establish against antibiotics, much attention has been paid to crude plant extracts, which are beginning to receive much interest as a potential source of bioactive natural molecules (Yakhlef, G. *et al.*, 2011, Gbogbo, K. A. *et al.*, 2013). The extraction of high-value active principles from plant material, notably the case of polyphenols, which currently arouse much interest thanks to their antioxidant power, is a very important step in both the isolation and identification of phenolic compounds (Mahmoudi, S. *et al.*, 2013). To isolate new substances from plants and thus find new bioactive natural molecules for the treatment of infectious diseases and wounds, and to make the isolation strategy as effective as possible, it is necessary to carefully select the plants to be studied. The purpose of this study is to enhance the phytochemical knowledge of ginger extracts.

MATERIALS AND METHODS

Preparation of Decoctions:

We brought the ginger back to the Saida region and thoroughly washed it to remove any impurities and unwanted traces so that it is clean and healthy (Fig. 1).

Fig. 1: Ginger

Preparation of Extracts:

The extracts of the spices under study were obtained by maceration and decoction. This type of extraction is simple and the separation is done by filtration.

Procedure:

The decoctions were prepared according to therapeutic dosages. 50 g of the plant (Ginger) was added to 500 ml of distilled water and boiled (100°C) in a beaker for 2 hours (Fig. 2**).**

The prepared extracts were placed in glass flasks and stored in a refrigerator at 4°C.

Fig. 2: Preparation of decoctions.

Animal Material

Rats:

The experiments were conducted at the Biology Laboratory of the University of Saida. We used male white Wistar rats (obtained from the Pasteur Institute of Algiers) that were 2 months old, with a body weight ranging from 85 to 145 g, for the evaluation of the studied effects.

The animals are kept at room temperature of 22°C (±4°C) in conventional cages with a feeder and a water bottle. They have free access to a standardized diet that corresponds to commercially available food products (Alf).

Breeding of Rats:

The rats are raised in cages lined with bedding that is renewed three times a week.

Group Allocation:

The rats were mated one week after their arrival, three females and one male per cage (Fig.3).

Fig. 3: Rat cages

Gel Preparation:

The gel was composed of carbopol-934 (carboxy polymethylene with a molecular weight of 3*106) 2.5%, propylene glycol (25.75%), propylparaben (0.25%), triethanolamine (0.8 ml), and distilled water in sufficient quantity to prepare 100 g of gel In the case of the white gel, 2.5 and 5 g of extract were respectively incorporated for 100 g of total gel during sample preparation.The required water for these formulations was divided into volume parts. In one part, the exact amount of extract (10 g) was dissolved, and in the other part, carbopol was soaked overnight, and propylene glycol and propylparaben were added to this solution. These two solutions were mixed in a beaker, and triethanolamine was added dropwise to the mixture to obtain the appropriate gel consistency. The gel prepared in this method was vacuum-sealed for 2 hours to eliminate trapped air. The pH of the gel preparation in this manner was neutral. This gel was placed in colla-cibles tubes and stored in a cool, dry place during the studies. Similarly, the white gel was prepared without incorporating the extract. -Evaluation des activités Cicatrisants

Biological Activity:

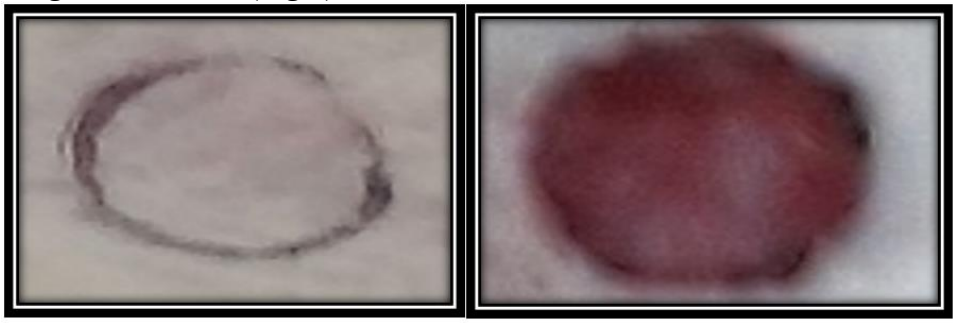
This test involves creating wounds on animals and then treating them with the tested extract preparation.The animals used were male white Wistar rats, weighing between 85 and 145 g, selected for laboratory use; so that they are treated separately in cages.

Analgesia:

-Local anesthetics are used for minor interventions or to block sensations in a limited region.

- It can be avoided by adhering to the recommended maximum dosages. It is also important to exercise caution not to introduce it into the circulatory system.

-Performing skin excision.(Fig.4)



-Before- -After-

Fig. 4: Surgical wound creation.

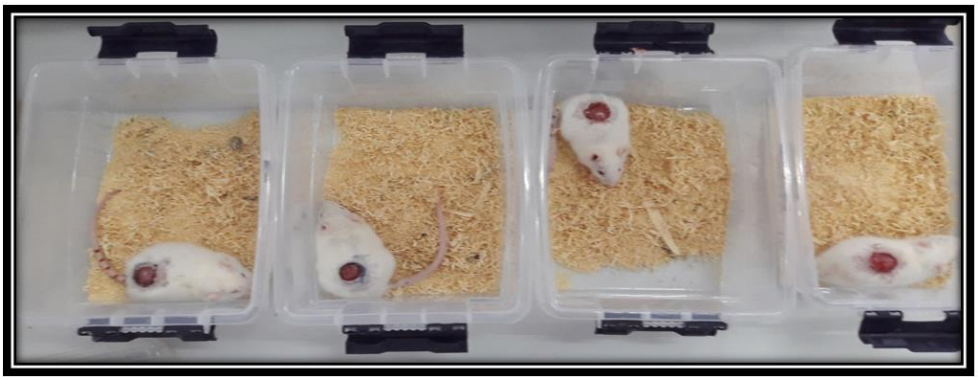


Fig.5: Individual isolation cage.

For this study, we used 26 male Wistar rats that were of adult age. with body weights ranging from 85 to 145 grams, obtained from the central animal facility of the

university. The rats were acclimated to our animal facility for a period of 2 weeks before the experiment.

From the first day of the experiment, the rats were transferred to individual cages (Fig.5). They had free access to their food and drink. (06) batches of 05 rats each were constituted (Table 2).

Table: Different groups based on different types of treatment.

LOT1	Operated and Untreated Rats
LOT2	Operated Rats + Treated with Madécassol (15 mg/kg)
LOT3	Operated Rats + Ginger Extract 01 cc (Maceration)

Preparation of Animals:

General Anesthesia:

The animals receive general anesthesia with chloroform. This anesthesia is supplemented by a local administration at the site of choice of Xylocaine 2% (10 mg/kg) for complete numbness of the animal.

Operative Procedure:

 The rat is placed under general anesthesia (chloroform); Place the animal in a clean and quiet place.

 The rats were shaved with scissors and a razor blade.

 Using a scalpel blade, a circular surgical wound of 1cm diameter is created.

 The skin is removed and the wound is left for a few minutes without any treatment, and the product is applied directly (then they take their usual diet in the same environmental conditions as before the intervention).

 Separate the rats into individual cages in groups, each group containing (5) rats:

The first group: It is used as control.

The second group: Treatment of the wound by a mixture of gel (1cc) + extract of the plant (Ginger) (1cc).

To prepare the Mixture:

Take 1cc of gel and 1cc of plant extract using a syringe and transfer them to a dry tube.Vortex the mixture to homogenize it.

The third group: Application of a reference healing ointment (Madécassine) on the wound. The ointment is applied using a spatula for 15 days.

During the 15-day treatment period, the temperature of all rats in each group is measured daily. The diameter of the wounds is also measured daily using transparent paper and a millimeter paper ruler.

Surgical Excision of Skin Flaps:

A circular area of approximately 1cm in length and 1cm in width is delimited by four points on the skin of the dorsolombar zone and then excised.

The excision wounds are immediately treated after the surgical procedure, and the animals are placed in individual cages with clean bedding.

Treatment of Wounds:

The wounds are treated once a day with a localized to using an insulin syringe with one of the following preparations: aqueous extract of the plant (Ginger) 1cc + hydrophilic gel (2%), or Madécassol (ointment), which is used as a reference healing agent (Lawrence, 1967; Poizot and Dumez, 1978; Pointel et al., 1987).

Evaluation of Parameters (temperature and healing):

- The evolution of the body temperature of the animals is measured during the 2-week period from J0 to J15.
- Temperature readings are taken using a rectal thermometer with a precision of 1/100 C°.

Evaluation of the Healing Process (Photography, Wound planimetry):

After being photographed, the dimensions (length, width) of the excision wounds are measured daily during the trial period (15 days) using a digital caliper and transparent paper with a metric scale for accuracy. The percentage of wound contraction is calculated using the following formula (Lodhi *et al.*, 2006):

Percentage of wound contraction (%) = [Healed wound surface area/Initial wound surface area] x 100

Mode and Route Of Administration:

The method of administration is cutaneous (topical application on the skin).

Treatment Duration:

The duration of the treatment is two weeks (15 days).

RESULTS

Study of the Healing Activity:

The healing activity is expressed in terms of the degree of wound contraction.

Evaluation of parameters (temperature and healing)

Body Temperature (°C). (Figs. 6 and 7)

The temperature is evaluated daily rectally. According to the results obtained, there is stability in the recorded values (ranging between 37° and 38°) throughout the experiment. Regardless of the nature of the animals (operated and intact) and/or the type of treatment, the body temperature remains statistically identical and close to normal or physiological values, with values ranging between 36.9 and 37.5**.**

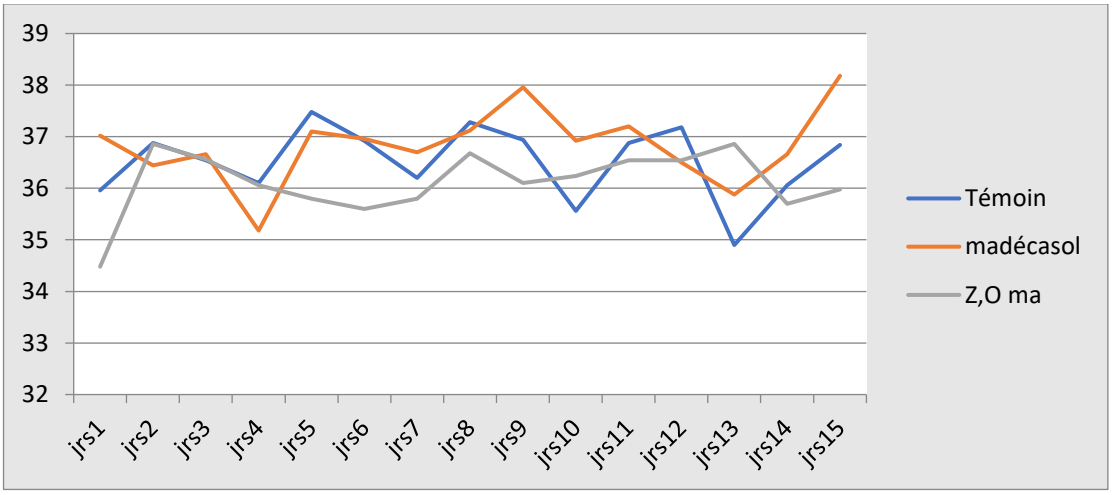


Fig. 6: Evolution of body temperature (°C) during the healing period according to different treatments.

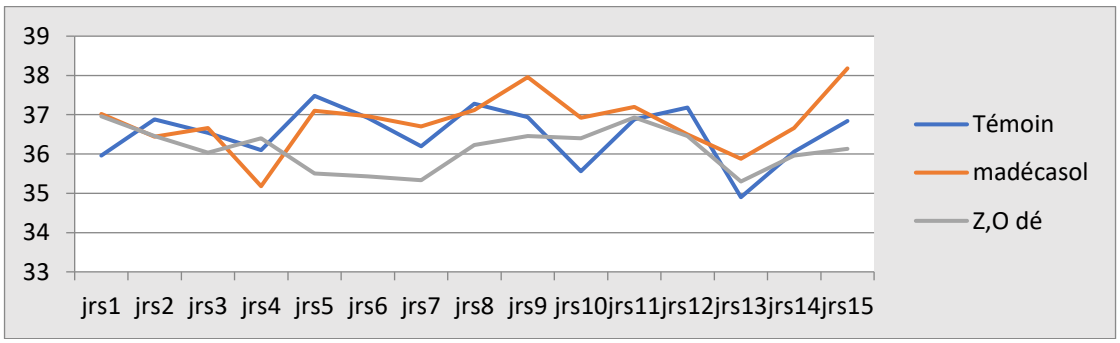


Fig. 7: Evolution of body temperature (°C) during the healing period according to different treatments.

Evolution of the Wound Healing Process (evaluation of excision wounds):

The process of wound healing occurred in several phases: a progressive disappearance of inflammation (wounds became less red on the first day), a contraction phase (the wounds became hard and covered with slightly blackish crusts on the 5th day), and the treatment allowed for complete healing of the wounds (on the 15th day).

Our results show that the ginger gel extract (decoction) accelerates the healing process compared to the controls; indeed, we notice that the contraction percentages of the wounds in different groups of rats (Madécassol and Z.O extract controls) indicate an increase every day. However, the ginger extract shows the highest percentage of wound contraction on the 11th day **(**Fig .8**).** Other studies have shown that pre-treatment of animals with Catharanthus roseus extract also induces an acceleration of the healing process (Nayak and Pintopereira, 2006).

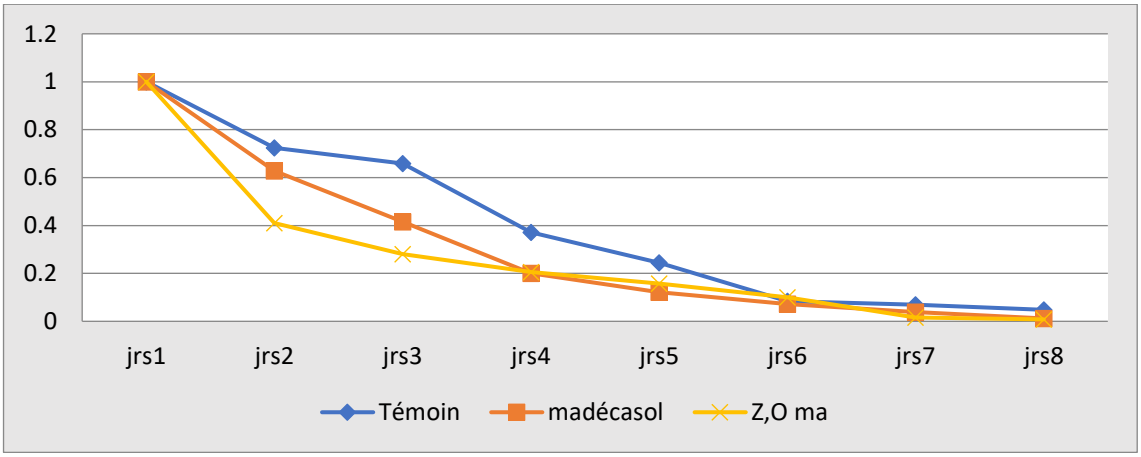


Fig. 8: Graphical representation of the evolution of wound healing over 15 days.

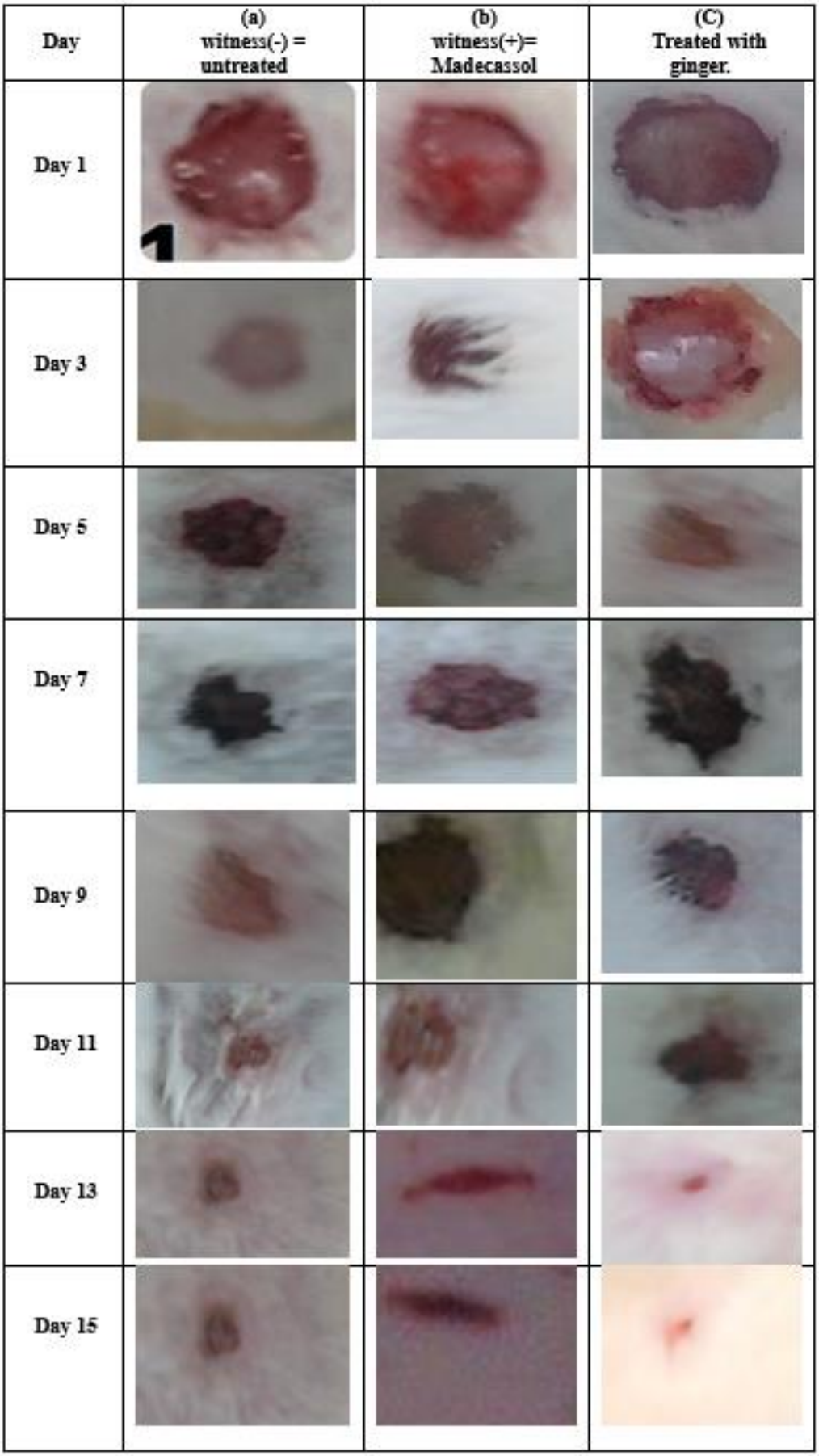


Fig. 9: Timeline of wound contraction after treatment.

The results of the evaluation of the wound healing properties show that the evolution of the percentage of contraction of the wounds treated with MADECASSOL is better compared to those of untreated wounds. We also notice that the evaluation of the percentage of contraction of wounds treated with ginger cream is much faster (more effective) compared to untreated ones. We can conclude that the evaluation of the percentage of contraction of wounds treated with the cream is more effective than those treated with MADECASSOL **(**Fig .9**).**

DISCUSSION

Several factors can affect the evolution of different phases of wound healing: the degree of trauma (Lee et al., 1989), fluid accumulation (Fowler, 1989), infection and the bacterial species (Diss, 2005; Dudleey, 1990), the pH of the local environment (Turner, 1978), trimming (systematic removal of any foreign body), antisepsis, and wound protection (Diss, 2005; Swain *et al.,* 1997; Bensegueni, 2007).

Local Wound Treatment:

The treatment of wounds depends on their size and severity, They may require general treatment in addition to local treatment.

Commonly Used Conventional Products: Products used in wound care include:

Antiseptics: These are chosen according to their spectrum of action against pathogens. (bacteria, fungi, and viruses) and the pH of the surrounding environment. Among these products iodinated products (iodised alcohol), chlorinated products (DAKIN),(Lozier *et al.,* 1992; Bensegueni, 2007).

Antibiotics: chlorotetracycline (aureomycin), combination (bacitracin, nalidixic acid). Polymixin B and Neomycin), etc.

Ethnic Medicine And Healing Products:

Among the products used in the treatment of wounds, honey, thanks to its antibacterial activity,antibacterial activity (Moore *et al.,* 2001; Williams, 1999), and propolis is often used to seal cracks and for its bacterial activity (Farstvedt *et al.,* 2004; Bensegueni, 2007).

Plants and Herbal Products:

Among the phototherapy products used, some have been the subject of experimental trials in an attempt to try to demonstrate their healing potential.*Centella asiatica* (asiaticoside) stimulates granulation tissue and angiogenesis (Rosen et coll, 1967).

Conclusion:

The use of medicinal plants from traditional African pharmacopoeia in the treatment of various diseases has been known for a long time; the positive effects of this phytotherapy are well established. However, empiricism is the basis of these practices. The extract of the Ginger Officinale plant has analgesic, wound healing, and antipyretic properties, which justify its traditional use. These properties are probably related to the presence of polyphenols, flavonoids, and saponins identified by phytochemical investigations. The in vivo tests used can be useful in predicting these biological activities:

- The results obtained by the analgesic study showed that the extracts have significant analgesic activity.
- The tests evaluating the activity of wound healing showed that the ginger extract has a significant effect.
- Given the results obtained from the antipyretic test, the ginger extract was more or less effective in reducing hyperthermia induced by brewer's yeast.

Therefore, the GINGER extract would be an advantageous source of improved traditional medicine that is very accessible and cheaper for populations**.**

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