CHLORHEXIDINE USE IN BURN PATIENTS: CYTOTOXICITY AND IMPLICATIONS IN WOUND HEALING

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ORIGIN OF THIS IN-DEPTH REPORT ON THE USE OF CHLORHEXIDINE FOR BURN WOUNDS

This evaluation on the current use of chlorhexidine has an origin from practical experience in the CHUV Burn Center. The chlorhexidine used is routinely prepared by the Hospital Pharmacy. Unfortunately, there was an absence of chlorhexidine in the correct concentration in the burn Operation Theater. Prof. Lee Ann Laurent-Applegate, director of the Cellular Therapy Unit of the CHUV and Burn Center, was called to seek immediate advice what to do in this case. The Hospital Pharmacy would not release magistral preparations directly so it was decided to make the correct dilution in the Operating Theatre to be able to continue with patient treatment. It was noted that there was a lack of specific guidelines in general for chlorhexidine as in the literature it can be found with many different concentrations and in different excipients for the same use. As there were reports that chlorhexidine could even create serious burn injury, it was decided that a complete review of the subject would be necessary to provide updated recommandations for specific use. Therefore, I chose this as my Masters project and divided the work-packages into three specific areas:

I. complete literature review;
II. collection of standard protocols within the University Hospital (CHUV);
III. development of a survey that was submitted to burn specialists and related centers all over the world.

The CHUV has been the first validated / accredited Burn Center by the European Burns Association (EBA) with the goal of having standards of quality in burn patient care. Therefore, the use of chlorhexidine is under evaluation in this report to help support the effective concentration and preparations of chlorhexidine for specific uses for best patient care with the intention of implemention in routine clinical pathways. With this Masters Project, I hope to contribute to the development of a Standardized Operating Protocol (SOP) for the Burn Center and perhaps also for the Institution as a whole.

To begin my report, a brief introduction of burns, burn management and infections will be presented. This will be followed by the methodology, the literature review and the survey development.
The literature review is organized firstly with case reports followed by an overview of the toxicity of chlorhexidine, effects of chlorhexidine on wounds and finally on burn wounds. The survey of chlorhexidine use in other centers is presented and a collection of Institutional protocols using chlorhexidine have been examined.

INTRODUCTION TO BURNS

SKIN AND BURN WOUNDS

The skin is the largest organ of the body and therefore is very important. Composed of stem cells, vessels, nerves glands and hairs the skin has three principal layers. Its role is the protection of the body against injury, infections, heat and light.

By definition, a wound is a traumatic opening in the skin and/or associated organs. It can be an open wound when there is a traumatic rupture of the integument or a contusion when the injury is closed.
Burn wounds are caused by contact with hot solid or liquid matter, from flame or scald, by chemicals, electricity and others.

Burn wounds are classified in three degrees which are important for wound management. In the first degree only the epidermis is destroyed. The second degree affects the dermis and is divided in two categories, superficial and deep. In the third degree, the three layers of the skin are completely destroyed. Because of no spontaneous healing possible and risks of infections, third degree burns have to be treated in specialized centers.

After the burn, an inflammation of the wound area occurs. Secondly, keratinocytes and fibroblasts migrate to revascularize and close the wound. A provisional matrix is produced. At the end, myofibroblasts end the closure of the wound and the fibroblasts produce the final matrix made of collagen and elastin.

Major burns also cause a systemic reaction involving cardiovascular, respiratory, metabolic and immunological reactions, and thus require specialized care in an intensive care unit.

This is a brief introduction of the skin and burn wounds. In order to understand burn wounds and their treatment, more detailed information is presented in APPENDIX I.

Since bacteria cover all the body, any wound will normally be contaminated except those of burn wounds at the very beginning, but most of the time the patient can resist invasive infection. Infection can occur, however, when the immune system is compromised or when there are more than 10^5 microorganisms per gram of tissue(11).
As seen before, when the skin protection of the body is compromised, it makes an easy entry for microorganisms. Major burns induce systemic changes such as an increase of metabolism and alteration of the immune system which can make patients more susceptible to infections(11).

An infection can inhibit wound healing by the invasion and dissemination of the microorganisms. This process can extend the inflammatory phase by having an increase of proinflammatory cytokines and proteases, by delaying the deposition of collagen and by degradation of granulation tissue and tissue growth factors.

Infectious agents can include virus, bacteria, fungi, protozoa, helmintes and prions. They can enter the human body through skin, lungs, blood, digestive tract and genitals and through cathetors during treatment. The major defense against infections are intact skin and mucous. They form a physical and chemical barrier by producing antimicrobial agents.
Microbes can injure or kill cells by releasing toxins which can cause hypercoagulative state, gastrointestinal symptoms, septic shock and acute respiratory syndrom(12).

Microorganisms that infect burn wounds differ depending on the anatomical site and time after injury. Most of the time, Staphylococcus and Pseudomonas are responsible for infections and sepsis in burn cases(11).
At any sign of rapid overall status of the patient, infection or sepsis should be investigated(11). An infection can be seen on the skin for exemple by a discoloration of the wound, sloughing of burned tissue, increased oedema and particularly odor.

Figure 1: Pathophysiology and Current Management of Burn Injury Photo Credit: Gregory Moran, M.D., from the Center for Disease Control and Prevention.

A wound can be colonized by endogenous micro-organisms present on the surface of the skin. This is the principal reason why wounds have to be desinfected in order to prevent an infection especially for a serious wound.
There are many types of desinfectants. They usually do not function on all micro-organisms and can have side effects.

By definition, an antiseptic is a substance that prevents, inhibits or reduces to a significant degree the microbial flora of skin or mucous membranes(13). Substances with antiseptic properties have been used for years by physicians and care professionals. For example, the antimicrobial effect of the coniferous resin was known by Egyptians which were using this for embalming mummies(14). Afterwards, the antiseptic effect of few substances were discovered but the true development of antiseptics date from the 19th century and consequent research has taken place principally during wars of the 20th century. A brief history of the development of antiseptics is presented to understand the problematic of antisepsis.

In ancient medical practices, the antiseptic effect of honey, vinegar and wine was well known(15).
In 1811, Bernard Courtois, discovered iodine(16) which is nowadays a commonly used antiseptic agent.
During the Civil War (1861-1865), sanitary practices had been developed and recognized as effective(17). Gangrene necrosis, by insufficient blood perfusion or infection, was a huge factor of mortality of war wounds. Bromine, carbolic acid and sodium hypochlorite were established to be effective in treating gangrene.

Then, in 1897, inspired by the work of Louis Pasteur on bacteria, Joseph Lister discovered the antiseptic properties of phenol (carbolic acid) in surgery(18).
During the First World War Alexis Carrel, a surgeon and Henry Dakin, a chemist, invented a technique of irrigation of wounds with Dakin's solution(17) which is composed of sodium hypochlorite.
The Vietnam war (1955-1975) permited numerous studies against infections. Mafenide acetate and silver sulfadiazine, topical antimicrobial agents still used today, were developed at that time(17).

Modern antiseptic agents have now less than 70 years of experience and their effects on wounds are not entirely understood. Chlorhexidine, used since 1954, has been studied for wound care in the last 20-30 years. Nowadays, mecanisms and actions of chlorhexidine on the wound are not entirely understood. Therefore, a study on the effects of chlorhexidine on burn wounds, which are specific wounds, is justified. The project to follow will be focused on chlorhexidine and its use.

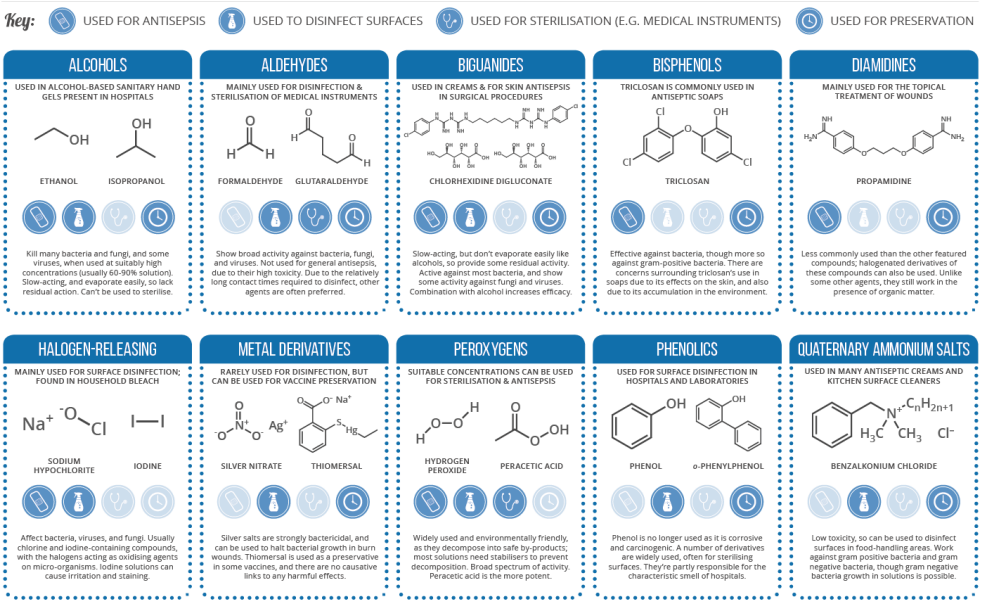


***Figure 2**: Brief summary of disinfectants and antiseptics*
(https://lh3.googleusercontent.com/7tVpWO_HcjU/VyDp3FazR4I/AAAAAAAA_2s/De0dWxNbPpwH5PudFXsln--h7IhGq8qZQ/w1488-h1052/Guide-to-Disinfectants-Antiseptics.png)

INTRODUCTION TO CHLORHEXIDINE

This part of the report will present the routinely used antiseptic, chlorhexidine. In order to understand the effects of this agent on wounds and specifically on burns, the structure of this chapter follow these points:

- Brief history of chlorhexidine
- Mechanisms of action of chlorhexidine
- Main forms used in Switzerland
- Problems occuring during the use of chlorhexidine

HISTORY OF CHLORHEXIDINE

Imperial Chemical Industries in Manchester discovered chlorhexidine in the 50's when they were researching an agent against malaria. As a very promising antiseptic agent, chlorhexidine gluconate was commercialized in the UK in 1954. In the 70's, the USA commercially introduced chlorhexidine. It was then developed into hand washing soap, oral agents, alcoholic solution and also for impregnating catheters or needles. Chlorhexidine is used in different forms as diacetate, dihydrochloride, gluconate (which is the most used form), digluconate and phosphanilate.

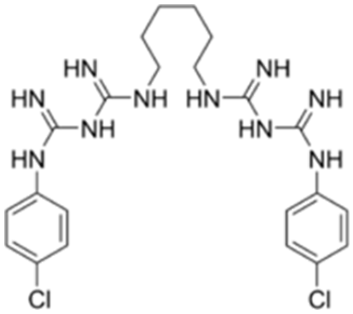


Figure 3 : Chlorhexidine formula (http://chlorhexidinefacts.com/history-of-chlorhexidine.html

In gels, creams, wound dressings, and other solutions, chlorhexidine can be formulated into in a wide variety of products for both medical and non-medical use and is widely used; for the desinfection of skin before and after a surgery or for hand hygiene, to disinfect surfaces, oral care, disinfection of urologic and vascular catheters and needles. It is also a component of deodorants, antiperspirants, creams, toothpastes and used in pharmaceutical products as acting as conservation agents.

Chlorhexidine is applied on wounds in different manners. It can be directly poured on the skin, applied in gel or cream on the wound, or covered by an impregnated dressing. The wound can be cleansed or irrigated (delivering a flow of chlorhexidine with pressure in order to clean and remove debris of the wound) with chlorhexidine.

MECHANISM OF TOXICITY OF CHLORHEXIDINE

Chlorhexidine's positive charges at physiologic pH bind to the negatively-charged bacteria's membrane producing a disbalance of bacteria osmotic equilibrium. This engenders a loss of potassium and phosphorus causing death of the bacteria at a high enough concentration by precipitation of the bacteria cytoplasmic content(19).

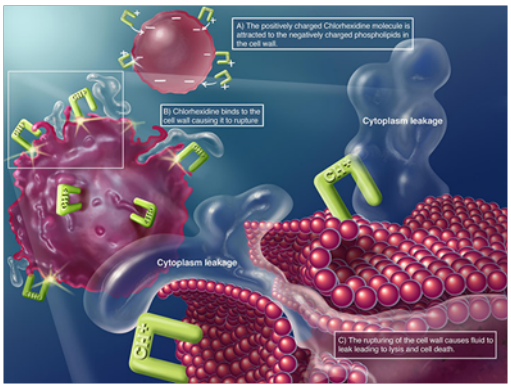


Figure 4 *: Chlorhexidine mechanism (http://chlorhexidinefacts.com/history-of-chlorhexidine.html*

Mecanisms of chlorhexidine's toxicity on eucaryotic cells has not been well understood to date. It seems to be related to the binding of charges on the cell's membrane causing an increase of permeability to calcium and inhibition of Na-K atpase pump. Other mechanisms such as alteration of protein synthesis or alteration of proliferation have been reported(19).

Compared to other antiseptics chlorhexidine can be bacteriostatic or active against bacteria, virus, mycobateria and fungi depending on the final concentration.

	Gram +	Gram -	Virus	Fungus	Side effects
Alcohol	+	+	-	-	Skin irritation
Providone iodine	+	+	+	-	Resorption, skin irritation
Chlorhexidine	+	+	+/-	+/-	Ototoxicity
Aldehyde	+	+	+	+	Allergic risk
Sulfadiazine	+	+	-	-	Allergic risk, resorption, skin irritation

Table 1 *: Effect of different antiseptics on microorganisms and their main side effects*
Adapted from http://www.cclinparisnord.org/Guides/guide_desinfectant.pdf and Conseils pratiques pour le traitement aigü des plaies, Roche, 3M, Flawa

DOSAGE FORM OF CHLORHEXIDINE

Solutions containing chlorhexidine can be found in different concentrations and with different excipients. The main forms are listed in Table 2 but other products containing chlorhexidine are available in Switzerland. See APPENDIX III (Médicaments contenant de la chlorhexidine autorisés en Suisse)

As seen in Table 3, chlorhexidine is mainly in a 70% alcohol solution and concentrations range from 0.5% to 4% for a topical use and 0.12% for oral use.

Form	Name	Concentration	Excipient
Liquid, External	Betasept Surgical Scrub	4%	isopropyl alcohol
	Hibiclens	4%	isopropyl alcohol
Miscellaneous, External	Hibistat	0.5%	isopropyl alcohol
	Tegaderm	2%	water, polymers
Solution, External	ChloraPrep One Step	2%	isopropyl alcohol
	Dyna-Hex 2	2%	isopropyl alcohol
Solution, Mouth/Throat	Paroex	0.12%	propylene glycol
	Peridex	0.12%	alcohol, saccharin sodium
	Periogard	0.12%	water, 11.6% alcohol

Table 2: *Different solutions containing chlorhexidine(adapted from uptodate drug information)*

PROBLEMS RELATED TO CHLORHEXIDINE

Daily use in surgery would indicate that it appears to be relatively safe with low problems related to wound healing. However, studies have shown that chlorhexidine is cytotoxic for human cells and can inhibit wound healing. Furthermore, it can even induce burn wounds or anaphylactic shock in certain patients.

As toxicity of chlorhexidine is concentration-dependent there are no studies published and therefore no overall recommendations on which concentration and for which use chlorhexidine is the most effective. Recommendations in the literature can even be contradictory. Chlorhexidine has been shown to inhibit or delay the healing process of a wound. In practice, chlorhexidine is used at different concentrations and in different formulations for the same use and this seems to be the same all over the world.

The main problems related in the literature are inhibition of wound healing, skin irritation, burn induction or anaphylactic shock.

The aim of this report is to look for reasons of the existing problems seen and hopefully to try to avoid a maximum of side effects on patients. Reasons that should be explored are the association of chlorhexidine with 70% alcohol, the percentage of chlorhexidine used in concentrations compared to the concentration studied in vitro and chlorhexidine effects on fragile skin as in burn cases and premature infants.

METHODOLOGY

SEARCH METHODS FOR IDENTIFICATION OF PUBLISHED STUDIES

Databases Medline/Pubmed, Cochrane and Uptodate were used with the following criteria:

- chlorhexidine
- side effects / case report / burn / infection / toxicity anaphylaxis related with chlorhexidine
- management of wound / burn wound
- topical agent on burn, infection management
- some others were found on articles read

No limits were established for year of publication and references in English and French were considered.

LITERATURE DATA COLLECTION AND ANALYSIS

All data involving mechanism, toxicity, case reports, side effects of chlorhexidine and it's use on wounds, burn wounds, infections, or in surgery were analysed. In total, 88 articles and web pages have been analysed to form the bibliography, references and writing of this report.

Data were analysed in this order:

1. Definition, general use, indications and mechanism of chlorhexidine
2. Case reports involving chlorhexidine to see if it was frequent, well documented, the consequences of side effects
3. Toxicity of the chlorhexidine in vitro and in vivo
4. Effect on wound healing
5. Effect on burn wounds

SURVEY

A survey was sent to 213 people from mailing lists accumulated from burn meeting attendance lists, associations and to burn centers. The answers came from professionals of burn care, mostly European plastic surgeons and nurses working in burn care centers. More details of the survey are found on page 25. The aim was to have a worldwide overview of the use of chlorhexidine in actual burn care. With these data, we could analyse the tendencies of solutions and their associated concentrations of agents containing chlorhexidine in burn wound management. A comparison of other medical units within the CHUV and other burn centers for their use of chlorhexidine was also part of the aim of this study.

The survey was sent at two separate times in the calander year at an interval of 5 months (May and September) to achieve the maximum number of responses.

DATA OF CHUV

In order to compare the CHUV to other centers and the literature, internal protocols of patient care involving chlorhexidine in the CHUV were consulted.

The data for the use of chlorhexidine in different solutions and concentrations in the whole of the CHUV were requested at the CHUV Pharmacy (protocols are included in Appendix IV).

RESULTS OF LITERATURE SURVEY

The effects that chlorhexidine can have on humans have been reviewed in the literature following the structure of this report's reflexion. The first step was to search if serious side effects were associated with chlorhexidine in the literature. Secondly, chlorhexidine toxicity has to be understood to know why it can induce lesions. Finally, the main point that this report is interested in is how chlorhexidine affects wounds and particularly burns.

The overview of all articles involving the use of chlorhexidine have been arranged in a Table format (Appendix VI) and separated into several categories for ease of analysis which include:

- Case reports of serious side effects
- *In vitro* and *in vivo* toxicity
- Effect on wound healing
  - Inhibition of healing process
  - Neutral effect on healing process
  - Promoting the healing process
- Use of chlorhexidine in burn care

REVIEW ARTICLES AND CASE REPORTS INVOLVING CHLORHEXIDINE SENSITIVITY

All active substances can have side effects which are other manifestations than the one desired. They are more or less frequent depending on the dose and the patient. They can have no incidence or be life threatening. This part of the report presents case reports with serious side effects induced by chlorhexidine found in the literature.

Article	Population	Type of chx	Side effects
Lashkari HP et al.	Extremely low birth weight	Aqueous 2%	Burn
Kutsch J, Ottinger D.	Extremely low birth weight		Burn
Sivathasan N et al.,	All population	Alcohol	Skin irritation, burn.
Stables GI, Turner WH, Prescott S, Wilkinson SM.	67 year-old man	0.5%	Allergic reaction. Urticaria.
Sharp G, Green S, Rose M.	All population	4%	Anaphylaxis
Beaudouin E, Kanny G, Morisset M, Renaudin JM, Mertes M, Laxenaire MC, et al.	All population		Anaphylaxis.
Wittczak T, Dudek W, Walusiak-Skorupa J, Świerczyńska-Machura D, Pałczyński C.	All population		Chlorhexidine as an occupational allergen
Sanders TH, Hawken SM.	Adults		Burn
Palmanovich E, Brin YS, Laver L, Nyska M, Kish B.	55 year old woman		Burn
http://www.theguardian.com/society/2014/jun/30/premature-babies-burned-antiseptic-mhra-	All population		Burn Death
Siddique H.	Extremely low birth weight	0.5% in alcohol	Burn
Bringué Espuny X, Soria X, Solé E, Garcia J, Marco JJ, Ortega J, et al.	Extremely low birth weight	0.5% in alcohol	Burn

Table 3: *Case reports and side effects of chlorhexidine in the literature.*

Side effects of solutions containing chlorhexidine can come from two sources; either from chlorhexidine itself or from excipients which are all substances in the solution aiming to stabilize, color, change the properties or dilute the active drug. The major excipient that can have side effects in chlorhexidine solution is alcohol.

Chlorhexidine can induce skin irritation, burn wounds and allergic reactions. Chlorhexidine can still be considered as an underestimated allergic hazard for health care professionals. The nocive effects can be in clinical situations involving skin, airway or systemic reactions and anaphylaxis/allergy and all of these side effects can affect all ages.

The majority of burn wound cases reported were with premature newborns and for specific skin affections(20)(21)(22)(23)(24)(25)(26). Extremely low birth weight populations seems to be very susceptible to chlorhexidine adverse skin reactions represented mostly by burns(20).

In one case report(21), authors mentioned that it is the use of alcohol in the preparation on premature infant skin that causes irritations. Alcohol is also well known for its antibacterial properties and is probably enhancing the disinfection when associated with chlorhexidine. Furthermore, many case reports that were studied were presenting cases with the use of alcoholic chlorhexidine(22)(23)(24)25)(27)(28). In our research, we did not find any other article having this point-of-view but alcohol could be a factor of irritation or burn induction in skin for premature infants. More research on this question would be necessary to draw a complete conclusion.

There could be multiple reasons why most of the burns by chlorhexidine were on premature infants. Perhaps they have a different skin type or could be more sensitive to chlorhexidine. In the case where it is known that the skin is more sensitive and that chlorhexidine is known to cause burn on premature infants, why would physicians still use chlorhexidine. Other antiseptic agents that do not provoke burns in this population would be an interesting question that could also be studied.
The cause of burns could also come from the use of alcohol for the excipient as *in vitro* studies show that chlorhexidine is as cytotoxic as normal saline water(29). Alcohol solutions and providone-iodine with alcohol preparations are also known to induce burns(22)(23)(24)(25)(30)(31)(32). Alcohol is definitely responsible for burns in extremely low birthweight infants and should absolutely be removed in routine care of extremely low birthweight infants.
However, one case reported that chlorhexidine was indeed in aqueous solution(26) which would indicate that the alcohol is probably not the only responsible agent for burns. The role of chlorhexidine and providone-iodine in burns should be seriously investigated.

Cases reported for adults showing nocive effects of chlorhexidine occured when the surface of the body was not completely dry and in situations where pressure zones were hidden and solutions had pooled(27)(28).

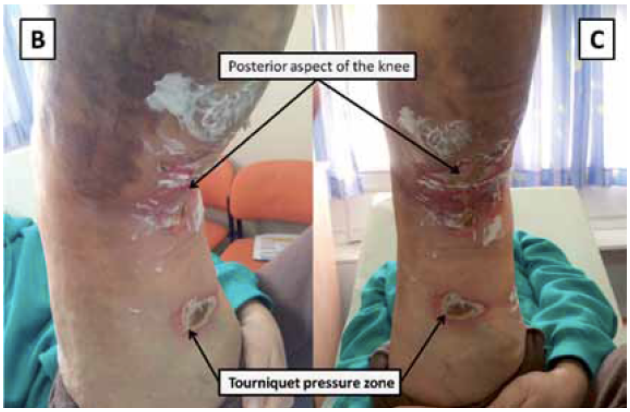


Figure 5: Zone of pressure burns (Third-degree chemical burns from chlorhexidine local antisepsis)

Cases of anaphylaxis induced by chlorhexidine are increasingly known. In one article(33), the authors have related up to fifty cases of anaphylaxis induced by chlorhexidine all over the world in the last 10 years and only fifteen of the fifty occurred in surgery situations.

Overall, anaphylaxis is present in adult elective surgery mostly in urology. This article also mentioned that chlorhexidine is in the top-five causes of perioperative anaphylaxis. The authors have also noted the lack of allergic testing of chlorhexidine. They stress the lack of recognition of chlorhexidine as a potential source of anaphylaxis.

There are few cases of life threatening side effects documented in the literature. However, most of the medical professional caregivers are not aware of chlorhexidine-induced reactions and to the extent of potential injury(34). Swissmedic mentions that case reports of anaphylaxis with agents containing chlorhexidine are probably underreported. From 2005 to 2013, 18 cases of anaphylaxis induced by chlorhexidine have been reported to Swissmedic. (https://www.swissmedic.ch/marktueberwachung/00135/00157/00285/index.html?lang=fr)

In order to prevent such cases, the literature would support that articles above recommend to:

- Increase the awareness of chlorhexidine-induced reactions in the medical profession
- Recognise effects of chlorhexidine-induced reactions
- Recognise products containing chlorhexidine
- Wipe with normal saline solution and dry the excess of chlorhexidine on the body
- Use chlorhexidine at the lowest concentration available, 0.05%
- Test allergic reaction systematically to chlorhexidine and not use it in this case

LITERATURE INVOLVING TOXICITY EFFECTS OF CHLORHEXIDINE *IN VITRO* AND *IN VIVO*

Chlorhexidine is an antiseptic which can kill microorganisms. Therefore, it has a toxicity against living organisms and thus could be harmful to human cells. Research has been done on the toxicity of chlorhexidine *in vitro* and *in vivo*. Different concentrations have been tested on different animal cell types. This part of the report will present the different results of research on the toxicity of chlorhexidine.

Toxicity of chlorhexidine has been shown by *in vitro* studies. It can kill microorganisms as well as human cells. The mechanism of action is that chlorhexidine, by a non-specific binding on the phospholipid membrane of the cell, induces an alteration of the osmotic equilibrium. At high concentrations, the content of the cytoplasm precipitates and leads to cell death(19).
In vitro studies show that human cells involved in wound healing such as fibroblasts, keratinocytes and macrophages are sensitive to chlorhexidine toxicity. At a concentration of 0.05%, chlorhexidine has been tested for cultured graft skin. Results are that this agent can prevent infections but at the same time can inhibit the growth of dermal fibroblasts and keratinocytes(35). Another *in vitro* study(19), states that chlorhexidine inhibits the growth of fibroblasts even at a concentration lower than that used clinically. It also has been reported that it could also have a negative effect on macrophages and thereby induce an immunosuppression(36).

On the other hand, *in vivo* studies show that chlorhexidine cytotoxicity is comparable to physiological saline water(29)(37). It can also be safely used to preserve contact lenses even at higher doses than commercially available. Chlorhexidine remaining on the contact lenses was reported to cause no damage to the cornea(38).

There are several questions which still remain unclear. Why is there a significant difference in the results of toxicity of chlorhexidine between *in vitro* and *in vivo* studies? What are the mechanisms underlying these differences? Why are products containing chlorhexidine for clinical disinfection at a concentration well above effective concentrations against microorganisms and proven to be cytotoxic *in vitro*? Why, if the toxicity of chlorhexidine is dose-dependent, cases reported are at a concentration varying from 0.5% to 4%, regardless of the gravity of the injury?

Overall, chlorhexidine toxicity *in vitro* is well proven at the concentrations tested but *in vivo* studies differ. It would suggest that overall safety is indeed existent but more extensive studies conducted *in vivo* on wounds would be required to attest that chlorhexidine is or is not harmful for the healing process.

Article	In vivo / in vitro	Toxicity
Boyce ST, Warden GD, Holder IA.	In vitro	Chlorhexidine 0.05% was uniformly toxic to both human cell and microorganisms.
Brennan SS, Foster ME, Leaper DJ.	In vivo	Toxicity of chlorhexidine does not differ from saline water.
Hidalgo E, Dominguez C.	In vitro	Significant adverse effects on dermal fibroblast growth.
Severyns AM, Lejeune A, Rocoux G, Lejeune G.	In vivo	Low toxicity which was comparable to physiological saline.
Bonacorsi C, Raddi MSG, Carlos IZ.	In vivo	May have an immunosuppressive effect on exposed macrophages.
Ar G, Y I.	In vivo	Chlorhexidine is safe for use on contact lenses.

Table 4: *Toxicity of chlorhexidine in literature in vitro and in vivo.*

LITERATURE ON EFFECTS OF WOUND HEALING WITH CHLORHEXIDINE

As seen in the two previous sections, chlorhexidine interacts with human cells and can be toxic and can have side effects. Whether the interaction with the wound healing process is beneficial or not is what we are interested in this part of the report.

With respect to all of the literature cited, chlorhexidine may be cytotoxic and could therefore interfere with the wound healing process. Chlorhexidine's effects were studied at a concentration of 0.05% to 4%. The interference is expressed in a delay of the formation of the granulation tissue and its decrease of thickness.
Numerous studies discourage the use of chlorhexidine on wounds because it can inhibit the healing process(39)(40)(41)(42). With a concentration of chlorhexidine at 0.05%, investigators have demonstrated that the average number of days for healing was doubled compared to physiological saline(43). In an animal study(44), chlorhexidine was also found to inhibit the healing process compared to saline water. At a concentration of 4%, the effect was more pronounced than at lower concentration.

Drosou *et al.* and Tatnall *et al.* have stated that *in vitro* studies did not corroborate *in vivo* studies(45)(46). They suggested the reason was that the different toxicities shown were due to the differences of concentration and the period of exposure to the agent. In spite of that, they recommended to use it to disinfect intact skin or dirty traumatized wounds but not on clean healing wounds. Instead of using chlorhexidine, they recommended to irrigate the wound with physiological saline abundantly.

Article	Type of article	Effects on the healing process
Salami AA, Imosemi IO, Owoeye OO, SALAMI A, IMOSEMI I, OWOEYE O.	Article review	Inhibitory effect
Morgan ED, Bledsoe SC, Barker J.	Article review	Inhibitory effect
Saatman RA, Carlton WW, Hubben K, Streett CS, Tuckosh JR, DeBaecke PJ.	Article review	Inhibitory effect
Uptodate : Emergency care of moderate and severe thermal burns in children	Uptodate	Inhibitory effect
Paunio KU, Knuttila M, Mielitynen H.	Journal review	Inhibitory effect
Niedner R, Schöpf E.	Journal review	Inhibitory effect

Table 5: *Inhibitory effect of chlorhexidine.*

Neutral effects on wound healing have also been stated. Some authors have noted that chlorhexidine does not interfere with the reepithelialization of the wound(47)(48). No difference for the toxicity of chlorhexidine on wound healing(29), no effect on the number of days in hospital(49) or no delay in the wound healing process have been reported(50)(51).

Brennan SS, Foster ME, Leaper DJ.	Journal review	Neutral effect
Dai T, Huang Y-Y, Sharma SK, Hashmi JT, Kurup DB, Hamblin MR.	Article review	Neutral effect
Crosfill M, Hall R, London D.	Journal review	Neutral effect
Fumal I, Braham C, Paquet P, Piérard-Franchimont C, Piérard GE.	Article review	Neutral effect
Popp JA, Layon AJ, Nappo R, Richards WT, Mozingo DW.	Article review	Neutral effect
Uptodate	Uptodate	Neutral effect

Table 6: *Neutral effect of chlorhexidine.*

On the other hand, some studies present chlorhexidine as being beneficial to wound healing(45)(52) and recommended chlorhexidine on open wounds but also state that data are insufficient to clearly know the effect on wound healing. In another report, authors have studied 0.05% chlorhexidine on wounds and concluded that chlorhexidine was more beneficial than normal saline solution(52). They have also stated that the concentration of chlorhexidine which is cytotoxic *in vitro* is not cytotoxic *in vivo*.

Sanchez IR, Swaim SF, Nusbaum KE, et al.	Journal review	Helping effect
Drosou A, Falabella A, Kirsner RS.	Article review	Helping effect

Table 7: Helping effect of chlorhexidine.

In conclusion, the effect of the chlorhexidine on wound healing processes has been studied by many authors but the results are contradictory and even confusing. No fixed recommendations have been cited up to now. In order to be sure of the overall effect, more studies on human wound healing are required and with different galenic formulations.

LITERATURE SUPPORTING EFFECT ON INFECTIONS WITH CHLORHEXIDINE USE

Infections are the invasion of an organism by germs. In the case of a wound, the skin can not fill its role of protection anymore and an infection can occur. Antiseptics such as chlorhexidine can be useful to kill germs in order to prevent or treat infections. This part of the review is focused on the impact of chlorhexidine on infections.

Many studies on the effects of chlorhexidine regarding infections have been described. This disinfectant is largely used in many forms for the known bactericidal activity or to help wound healing. Chlorhexidine is commonly used to treat odontogenic infections(53) and to reduce clinical symptoms(54).

Article	Type of article	Effect on infections
Uptodate : Complications, diagnosis, and treatment of odontogenic infections	Uptodate	Chlorhexidine 0.12 percent oral rinse can be used in most cases for odontogenic infections.
Bhate D, Jain S, Kale R, Muglikar S.	Journal review	0.12% CHX mouth rinse effectively reduced the clinical symptoms of plaque-induced gingivitis

Table 8: *Effect of chlorhexidine on odontogenic infections.*

Chlorhexidine is well known to be effective for the prevention and treatment against a large range of microorganisms(55). In a comparative study, authors have stated that chlorhexidine has an excellent activity against microorganisms present in burn wounds at a concentration up to 0.5% *in vitro*(43). It has also been proven that cleansing a wound with chlorhexidine reduces wound colonization(56)(57). Antisepsis such as chlorhexidine is recommended to be used on burn wounds to prevent and treat infections(58). On the other hand, another study mentions that chlorhexidine has no effect on contaminated surgical wounds(49).
The efficiency of chlorhexidine against Hospital acquired infection is also reported. Some studies even recommend to use it particulary against MRSA(59), VRE(56) and patients in an intensive care unit(60).
Bathing patients with chlorhexidine can prevent(59), decrease(61) or even eradicate drug-resistant bacteria colonization and Hospital acquired infections(51) which is particulary important for intensive care and particularly for burn patients(59). Even though bathing is widespread, modern recommendations are to not apply chlorhexidine on deep wounds and not to repeatedly clean large surfaces of skin except from necessary conditions which have been adapted(60).

Gunjan K, Shobha C, Sheetal C, Nanda H, Vikrant C, Chitnis DS	Journal review	Excellent antibacterial activity up to 0.5% concentration. The effect was marginally reduced for Pseudomonas at 0.25% concentration
Snelling CF, Inman RJ, Germann E, Boyle JC, Foley B, Kester DA, et al.	Journal review	Reduced wound colonization with chlorhexidine when compared to nonantibacterial soap.
Sanchez IR, Swaim SF, Nusbaum KE, Hale AS, Henderson RA, McGuire JA.	Article review	More bactericidal activity of chlorhexidine than saline and providone-iodine and more beneficial to wound healing.
Rubin C, Louthan RB, Wessels E, McGowan M-B, Downer S, Maiden J.	Article review	Greater reduction of Hospital acquired infections compared to bathing with soap and water.
Vernon MO, Hayden MK, Trick WE, Hayes RA, Blom DW, Weinstein RA.	Article review	Reduced VRE contamination of patients' skin
Uptodate : Epidemiology and prevention of infections and antimicrobial resistance in the intensive care unit	Uptodate	Bathing patients decreases both hospital-acquired infections and colonization with drug-resistant organisms among patients in the ICU.

Popp JA, Layon AJ, Nappo R, Richards WT, Mozingo DW.	Journal review	Decrease of HAI rate in an intensive care unit to zero with use twice a day of chlorhexidine 0.9% for bathing. No delay of wound healing.
D'Avignon LC, Saffle JR, Chung KK, Cancio LC.	Journal review	A broad-spectrum surgical detergent such as chlorhexidine gluconate should be used for burns in the combat casualty.
Lee I, Agarwal RK, Lee BY, Fishman NO, Umscheid CA.	Web page	Preoperative skin antisepsis with chlorhexidine is more effective than with iodine for preventing surgical site infection and results in cost savings.
Climo MW, Sepkowitz KA, Zuccotti G, Fraser VJ, Warren DK, Perl TM, et al.	Article review	Daily chlorhexidine bathing among ICU patients may reduce the acquisition of MRSA and VRE.
Uptodate : Chlorhexidine gluconate/ Drug information	Uptodate	Chlorhexidine has activity against gram-positive and gram-negative organisms, facultative anaerobes, aerobes, and yeast; it is both bacteriostatic and bactericidal, depending on its concentration.
Uptodate : Basic principles of wound management	Uptodate	For wound irrigation, the addition of dilute iodine or other antiseptic solutions (eg, chlorhexidine and hydrogen peroxide) is generally unnecessary.
Dai T, Huang Y-Y, Sharma SK, Hashmi JT, Kurup DB, Hamblin MR.	Article review	Majority of bacteria and yeast with the exception of mycobacteria, are eradicated by chlorhexidine.

Table 9: Beneficial effect of chlorhexidine on infections.

Chlorhexidine has also been proven to considerably reduce surgical site infections and is even less expensive compared to other antiseptics(62). In one study, the authors emphasize the fact that the prevention of infections after surgery must be globally approached(63).

Hannan MM, O'Sullivan KE, Higgins AM, Murphy A-M, McCarthy J, Ryan E, et al.	Article review	Significant lower SSI infection rates in cardiothoracic surgeries use of chlorhexidine as pre-operative antiseptic in a risk-adjusted cohort with education of the surgical team with when compared with API.
Eiselt D.	Article review	Surgical site infection rates reduced by 50% with 2% chlorhexidine cloth before surgery

Table 10: *Beneficial effect of chlorhexidine on surgical site infections.*

However, Crossfill *et al.* have shown that treatment of surgical wounds with chlorhexidine has no better effect on the sepsis rate when compared with saline water or no treatment at all(49). The addition of chlorhexidine in wound irrigation would therefore not be necessary according to their study.
The lack of clear clinical benefits for using chlorhexidine in the management of skin microbial burden is also mentioned(64). The authors also say that the reduction of the burden could be a surrogate outcome which would not necessary mean a better clinical result.
Furthermore, a recent pragmatic trial on 9,340 patients in the United States concluded that daily chlorhexidine 2% bathing did not decrease the number of hospital acquired infections(65). They do not recommend daily bathing for patients in intensive care units but did not describe the particular use for severe burn patients.

Noto MJ, Domenico HJ, Byrne DW, Talbot T, Rice TW, Bernard GR, et al.	Article review	In this pragmatic trial on 9,340 patients, daily bathing with chlorhexidine did not reduce the incidence of healthcare-associated infections.
Alawadi ZM, Kao LS.	Web page	Although there is clear biological rationale for chlorhexidine gluconate in terms of reduction of skin microbial burden, clinical evidence of benefit has been lacking.
Crossfill M, Hall R, London D.	Journal review	No effect of chlorhexidine in contaminated surgical wounds. No better effect on the sepsis rate when compared with saline water or no treatment at all

Table 10: *No beneficial effect demonstrated for chlorhexidine on infections.*

In conclusion, chlorhexidine has its place for treating and preventing infections in wound care. Discussion is still open concerning the benefit of using chlorhexidine if no decrease of infection and side effects are obvious. There should still be inquiries into which concentration, type of solution and kind of use for chlorhexidine should be studied. Caring for infections of burn wounds is specific and also requires focused studies. This specificity will be reviewed in the next section.

LITERATURE SUPPORTING USE OF CHLORHEXIDINE IN BURN CARE

Burn wounds are particularly subject to infections. As seen in the pathophysiology of burns, a major burn destroys the vessels and therefore the immune system is not able to fight against microorganisms present on the wound. Furthermore, major burns induce an immunodepressive state making the maintainence of sterility of the wound more difficult. An overview of the literature on the use of chlorhexidine in burn management will be presented in this section.

Just after the burn injury, the wound is sterile except for the presence of some microorganisms deep in the sebaceous glands and hair follicles(4). Therefore, it is necessary to keep the wound sterile and to prevent colonization from present microorganisms and from new ones existing on exterior dressings with antibacterial treatments(4).

Use of chlorhexidine is recommended by some authors for the treatment of burn wounds. There are contradictory results regarding application. Some articles advise to use it only on large burns to prevent sepsis(9) and others only for superficial burns as a disinfectant(58)(66)(67)(48) or within dressings(4)(48). Chlorhexidine surgical scrub brushes can also be required for strong debridements(66)(48).
As burn impedes the innate immune system of the skin, vulnerability to bacterial infections is greater and burn wounds also take several weeks for complete healing. Therefore, several dressing changes are required with the same antibacterial cream to prevent microorganism resistance(68).

Article	Type of article	Effect on burn wounds
Tiwari VK.	Journal review	Dressings of burn wound by antimicrobial agent helps in killing of organisms and keeps the wound sterile for a longer period.
Waitzman AA, Neligan PC.	Article review	"Topical chemoprophylaxis should not be used routinely on small burns but is useful for large burns to reduce the incidence of burn wound sepsis"
D'Avignon LC, Saffle JR, Chung KK, Cancio LC.	Journal review	A broad-spectrum surgical detergent such as chlorhexidine gluconate should be used.
Uptodate : Topical antimicrobial agents for the management of superficial partial thickness burns	Uptodate	Only for superficial burns. Does not interfere with reepithelialization.
http://emedicine.medscape.com/article/213595-treatment	Web page	A broad-spectrum surgical antimicrobial topical scrub such as chlorhexidine gluconate should be used for burn wounds infections.
DeSanti L.):323–32; quiz 332–4.	Article review	Open areas are gently cleansed daily with a dilute chlorhexidine solution to remove crust and surface exudate

Table 10: *Articles that recommend chlorhexidine for the treatment of burn wounds.*

In one article(69), *P. aeruginosa* is the most prevalent bacteria in burns. In this study, silver sulfadiazine is said to be the most potent agent against *P. aeruginosa* but requires daily change of the dressing. In order to reduce unnecessary dressing changes, alternative agents such as chlorhexidine should be studied(70). This antiseptic is presented as a very potent agent for the treatment of burns since uptake is rapid and antibacterial action can last up to 6 hours(69).
A Cochrane review mentions that chlorhexidine dressings do not reduce the time of burn wound healing compared to hydrocolloid dressings(71). On the other hand, avoidance of disinfectants such as chlorhexidine has been advised due to inhibitory effects on wound healing(42). Other authors suggest to use it only for isolated cases(72). As studies are contradictory, we can see that the effect on wound healing and especially for burn is not clearly known up to now.

Acar A, Uygur F, Diktaş H, Evinç R, Ulkür E, Oncül O, et al.	Article review	Silver sulfadiazine is the most potent antipseudomonal agent in this study.
McManus AT, Denton CL Mason AD, Jr.	Article review	Data suggesting that chlorhexidine diphosphanilate should be evaluated in a clinical trial for use as an alternative topical therapy for the burn wound.
Wasiak J, Cleland H, Campbell F.	Cochrane review	There was no significant difference in mean time to wound healing between hydrocolloid dressing and chlorhexidine impregnated paraffin gauze dressing.

Table 11: *Chlorhexidine in dressings for the treatment of burn wouds.*

One of the complications of burn management is the non-capacity of action of the antiseptic against a class of micro-organisms and the emergence of drug-resistant bacteria. Silver sulfadiazine is one of the most used and recommended in Europe(70) but development of sulfonamide-resistant gram-negative organisms has been signalled. Chlorhexidine is as active as sulfadiazine silver for treatment of burn wound sepsis(47) and there is no evidence of cross resistance with sulfonamide-resistant strains observed(70).
Even if increased resistance against chlorhexidine with bacteria such as Proteus has been reported(73), this antiseptic appears to be a very good alternative to silver sulfadiazine for the treatment of burn wounds. This is mainly because few chlorhexidine-resitant bacteria exist to date and because it may broaden sulfadiazine antimicrobial spectrum such as for *Staphylococcus species*.

Furthermore, chlorhexidine dressings, currently used for burns in Australia(73), do not interfere with wound reepithelialization in contrast to silver sulfadiazine(71)(74).

In conclusion, the role of chlorhexidine in burn care is still under discussion. As a topical solution, within dressings or brushes and with different solutions could prevent or treat infections of burn wounds. Effects on wound healing and reepithalisation are still contradictory.
These results indeed seem to be chaotic. On the other hand, its large spectrum of action and its role in the treatment of bacterial-resistant infections is well established. The reasons for these different results can be relative to the concentration of chlorhexidine, the frequency and lasting effects of the application, the amount of chlorhexidine poured or applied on the wound, the alcohol as excipient and others that I certainly have not taken into account.

Based on the literature, simple changes can diminish adverse reactions. Recommendations are therefore:

- Inform the medical workers that chlorhexidine can be harmful if not used properly
- Use chlorhexidine in an aqueous solution at a low concentration (0.05%)- especially on fragile skin
- Different packaging design and color between aqueous and alcoholic solutions should be implemented
- Wiping with saline water after disinfection with chlorhexidine could be included in burn SOP's
- Testing allergic reaction before using intravenous chlorhexidine

All of these defined summarized elements have been extracted from this extensive literature search, survey and SOP retrieval from the internal protocols. Therefore, all of the evidence established should now help to make recommendations for an updated SOP for the CHUV burn center. It is interesting to note that the CHUV has many SOPs on disinfection and use of chlorhexidine. This is an excellent model for other burn care centers.

OVERALL USE OF CHLORHEXIDINE IN CHUV PRACTICES

Chlorhexidine is an antisepsis largely used in the CHUV for cleansing or for disinfection of hands, skin, wounds or materials. Concentrations vary from 0.02% to 4%. Chlorhexidine can be found to be used in alcoholic or aqueous solution. Table 4 summarizes the use of chlorhexidine in the different protocols of the CHUV.

Percentage of Chlorhexidine	Clinical use
0.02%	Vesical wash. Disinfection of wounds on ear-nose-throat area.
0.05%	Disinfection of the external urethral orifice before placing a urinary catheter or vulval cleansing if prescribed. Cleansing and dressing of burned patient.
0.5%	Disinfection of surgery field or intact skin before an invasive surgery, blood sample or injection. Disinfection of central venous catheter and venous catheter site by oncologic patients. Disinfection of the skin before introduction of a pacemaker. Disinfection of intravenous connections, gloves, taps.
2%	Replacement of one-way valve of venous catheter. Desinfection of parenteral feeding field. Impregnated gloves for cleansing. Disinfection of skin before connection on hemodialysis fistula. Hand wash. Impregnated dressings for intensive care of burn patients.
4%	Hand wash. Preoperative patient wash whether or not colonized by multidrug-resistant germs. Entire cleansing of neutropenic patient.

***Table 12**: Use of chlorhexidine in CHUV (from CHUV protocols)*

Product	Quantity (units)
Chlorhexidine Alcoolique Colorée Braun sol 2 % 1 flac 100 mL	632, 200.00
Chlorhexidine Alcoolique Colorée Braun sol 2 % 1 flac 500 mL	3, 589, 000.00
Chlorhexidine Alcoolique Incolore sol 2 % 1 flac 250 mL	7, 519, 750.00
Chlorhexidine Aqueuse Bichsel sol 0.100 % 1 flac 100 mL	465, 700.00
Chlorhexidine Aqueuse Braun sol 0.500 % 1 flac 100 mL	133, 200.00
Chlorhexidine Aqueuse CHUV sol 0.050 % 1 flac 500 mL	787, 500.00
Chlorhexidine Aqueuse CHUV sol 2 % 1 flac 100 mL	69, 900.00
Chlorhexidine Incolore Braun teinture 0.500 % 1 flac 500 mL	10, 000.00
Hibidil Stérile sol 0.050 % 5 flac 15 mL	1, 157, 475.00
Hibiscrub sol 4 % 1 flac 250 mL	1, 250.00
Lifo-Scrub sol 4 % 1 flac 100 mL	1, 168, 400.00

***Table 13**: Solutions for disinfection containing chlorhexidine in the CHUV (CHUV Pharmacy)*

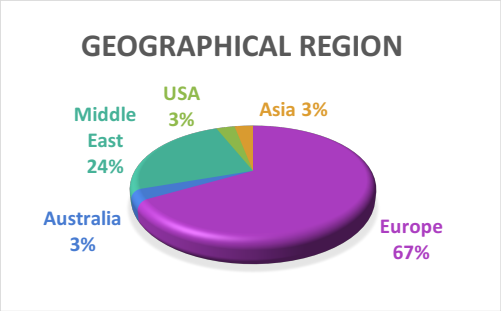
From the different protocols used in the CHUV, what evolves as being important is that this antiseptic is mainly present for disinfection of skin before surgery and for hand wash procedures.
From 2009 to 2015, about 11 million units of chlorhexidine in alcoholic solution were used which represents nearly 2 million liters of chlorhexidine solution. By comparison, about 2.5 million units were in aqueous solution. The alcoholic solution is therefore dominant (data extracted from CHUV Pharmacy).

From the use of chlorhexidine in the CHUV evolves the following questions:

- Is chlorhexidine in aqueous solution as efficient as in the alcoholic solution in practice?
- Is the alcoholic solution appropriate since we believe from the literature that the alcohol contained within can be responsible for negative side effects?
- Is the alcoholic solution really indicated on fragile skin as burn wounds and preterm infants or should it be replaced by aqueous solution because of the effect of alcohol reported in the literature?
- Are the high concentrations of chlorhexidine still useful if it is known that chlorhexidine is effective at lower concentrations and furthermore when side effects are concentration-dependent?
- Is it economically wise to purchase such high quantities of solution compared to making the dilution by the CHUV pharmacy, especially if the concentration of chlorhexidine is low?
- If the price is lower, is it safe enough to make the chlorhexidine in the Pharmacy knowing that a rupture of stock can occur?
- In case of a rupture of stock of chlorhexidine solution at a specific concentration, is it possible to make the dilution in the operation theater?

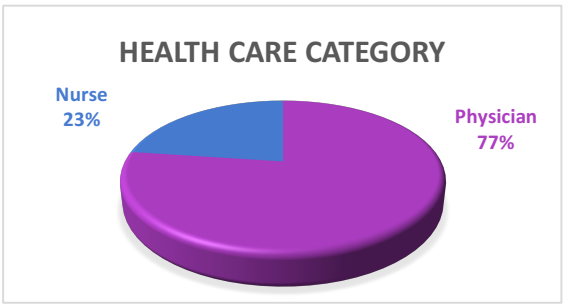
From my entire work on chlorhexidine, few responses can be that aqueous chlorhexidine in low concentration seems to be satisfactory to treat infections. It also seems to cause less side effects than in an alcohol preparation. It may be less expensive to order high amounts of aqueous chlorhexidine as no alcohol is needed and a less amount of chlorhexidine is present in preparations.

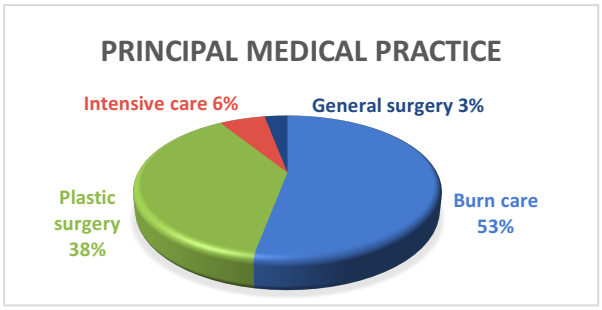
In order to determine how chlorhexidine is used in other burn care centers, I have sent a survey to 213 healthcare workers in burn care at two different intervals of time with one in the Spring and one in the Fall (May and September) to maximize potential responses. We had 36 participants providing 17% of responses. Overall, this is very similar to general surveys for health care professionals.



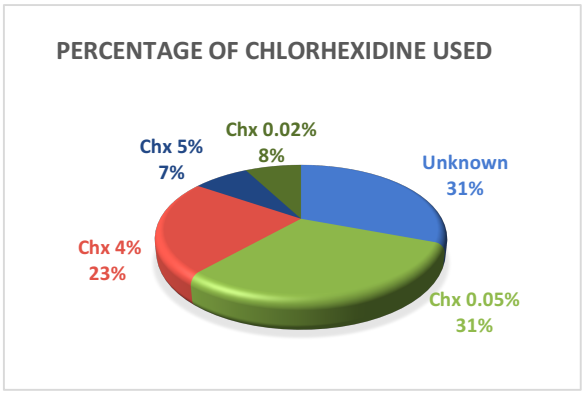
The health care category, medical practise and geographical region of the people who completed the questionnaire are presented in the following graphs.
67% of the responses came from Europe, 24% from the Middle East, 3% from USA, 3% from Asia and 3% from Australia.

77% were physicians and 23% were nurses. Their medical fields were Burn Care 53%, Plastic Surgery 38%, Intensive Care 6% and General Surgery 3%.



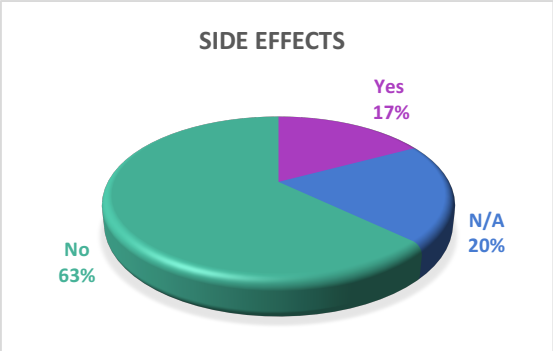


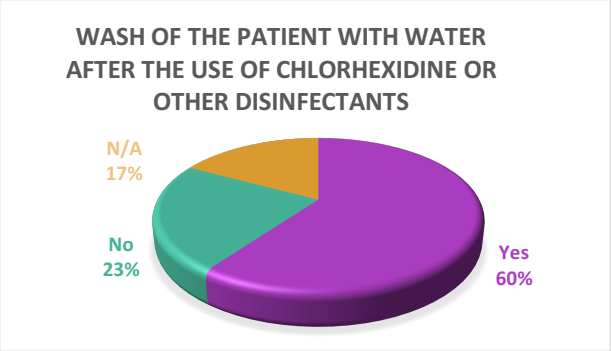
Chlorhexidine is used on burn wounds by 54% of the survey participants and the percentage of chlorhexidine varied from 0.02% to 4%. For approximately 31%, the percentage used was not known (no specific response).
The most used percentages were found to be 0.05% for 31% of the users and 4% for 23% of the users, a difference of 100 times in concentration. Other concentrations are 5% for 7% of the users, and 0.02% for 8% of the users.



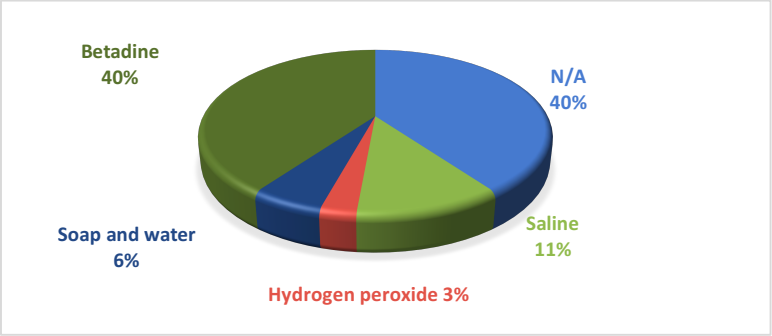
Although we can imagine that this difference may have effects on wound healing and side effects, washing the patient with water after the disinfection (what the majority of the survey particpants do) may reduce the possibility of side effects.

Side effects reported were skin irritation, dryness, inhibition of wound healing and pseudomonas contamination.



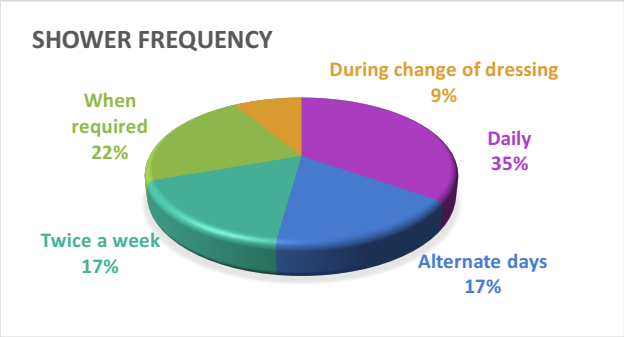


Substances used for disinfection of burn wounds instead of chlorhexidine were Betadine® for the majority (40%), saline solution (11%), soap and water (6%) and hydrogen peroxide (3%). Overall, 40% of survey participants did not answer this specific question.



Standard Operating Procedures (SOP) for cleaning burn wounds varies between the different hospitals. Some reported to not have any SOP in place and some transfer directly to specialized centers. However, for the majority, SOP's are in routine practice. The presentation of the different SOP's is presented below in order of frequency.

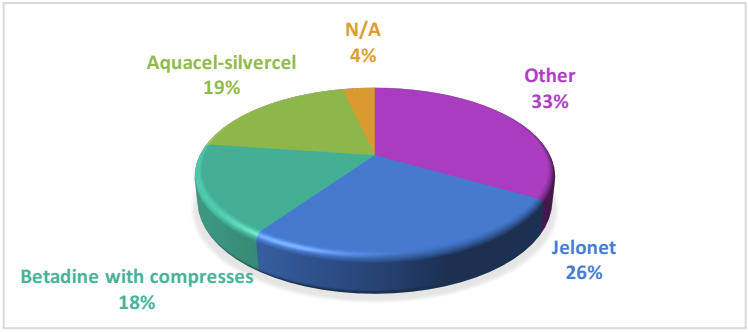
- Disinfection with Betadine® only or Betadine® wiped afterward with saline solution. In case of allergy of Betadine®, washed only with saline water.
- Disinfection with Chlorhexidine at concentration 0.015% and 4%. Some of them use only Chlorhexidine or after a first wash with Betadine® and normal saline solution. Others use Chlorhexidine at first and then wash with soapy water, normal saline water or Betadine®.
- Application of MEBO® ointment on the burn. Then wipe the old ointment and apply the new layer three times per day.
- Application of Flamazine® directly on the wound. Also used to remove the eschar and then clean with normal saline solution.
- Bathe or wash with soapy water and wipe with saline water.



Management of burn wounds can include showering of the burn. In our results we found different shower systems and frequencies. Only three hospitals do not use shower systems. One hospital transfers severe burned patients to specialized centers.
Shower frequencies are different depending on the hospitals. It varies from daily or every two days for the majority and only when required.

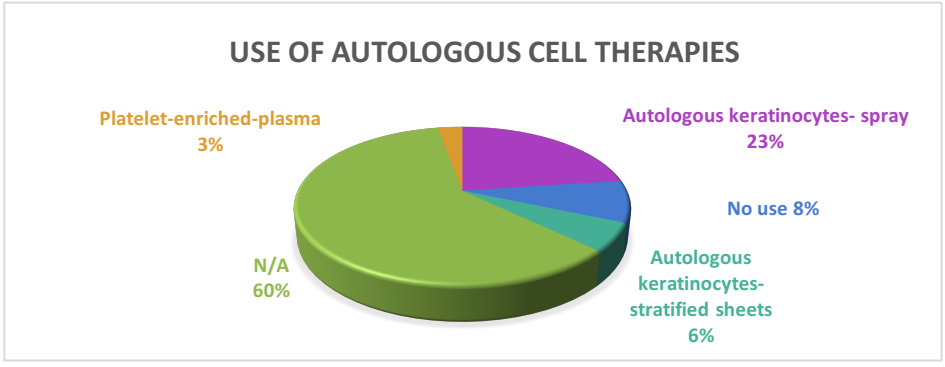
Usual procedures in the CHUV requires applying wound cover after shower or cleansing of the burn wound. In our survey, physicians and nurses use Jelonet in 26% of cases, Aquacel-sivercel in 19% of cases and Betadine® with compresses in 18% of cases. Other wound covers are also employed by 33% of the users such as Mebo ointment®, Flamazine®, Flaminal®, Silver sulfadiazine, hypochlorite solution for some infected wounds, dressings Mepitel®, Mepiplex®, Acticoat® or Urogotul SSD®, Polyfax® and Biobrane®.

Reasons answered for applying wound cover were for anti-bacterial effect and to help the wound healing depending on the presence of infection, size and depth of wound, age of patient and stage of healing.



Cellular therapies, which consist of applying living cells onto wounded patient skin can also be used in burn wound management. 60% of survey participants did not answer if they use these techniques. 8% answered that they do not. Autologous keratinocyte spray (23%), Platelet-enriched-plasma (3%) and Autologous keratinocyte stratified sheets (6%) are the cell therapies utilized by hospital centers within our survey results.

Procedures reported before the application of any cell type or graft procedure are debridement and disinfection. Debridments are typically done with scalpels, versajet or with the Watson knife. Wounds are disinfected with Betadine® or chlorhexidine.



DISCUSSION OF SURVEY RESULTS

Briefly summarized, the answers of the distributed survey came from the majority of European physicians specialized in treatment of burn wounds as the email addresses came from a database of several burn congresses. More than half of the responders were using chlorhexidine for burn care in heterogenous concentrations varying from 0.05% to 4%.
Approximately 20% of side effects were seen when using chlorhexidine and were reported. Surprisingly, 60% of the professionnals wash the patient after the use of a disinfectant. This is not what is usually recommended in the literature. Therefore, we may think that the interpretation is that chlorhexidine is seen as possibly harmful and wiping may reduce frequence and importance of side effects. This procedure should be investigated associated with the efficiency with or without post-disinfection wash.
Betadine® and saline water are the most employed disinfectants instead of chlorhexidine. Most of the participants disinfect the wound with Betadine® or chlorhexidine and they wipe after with normal saline.
Nearly all hospitals shower burn wounds and the frequencies vary from daily to three time a week.
Wound coverings are very often used and cellular therapies do not seem to be well known.

The survey was aimed to see if side effects were current and which concentrations and solutions were most used. Results showed that side effects, 20%, were more frequent than thought and concentrations are extremely varied. No trend of dose-dependent side effects was noted. The survey was not able to detect if the alcoholic solutions were more susceptible to cause adverse skin reactions. The strength of this study was that we are able to show frequent side effects at different concentrations, the lack of knowledge on the concentration used, the heterogeneous manner of application and the fear about using chlorhexidine seen by the usual post-wiping.

At the very end of the research, a very interesting article was found. It is an analysis from 2010 of a national survey on the use of chlorhexidine in Neonatal Intensive Care Units in Baltimore. Authors aimed to assess the current practice and the safety of chlorhexidine gluconate on neonates less than two months old.
It has been shown that effects of this antisepsis are not entirely known in specialized fields. Adverse effects on fragile skin can be serious and the safety of chlorhexidine in those fields has not been totally approved. It would further show that more studies have then to be made for correct use.

This very unique research was made only on neonatal population and is therefore not comparable to our research. But what comes out is that it shows the same results of the effects of chlorhexidine seen on adults. Then, similar effects with neonatal population may be found on fragile skin as burn wounds. This confirms the fact that more studies on chlorhexidine effects on burns has to be initiated.

OVERALL DISCUSSION

This search of literature shows that chlorhexidine seems to be a very potent antiseptic with a clear beneficial role in burn care.
In summary, case reports found in the literature mention skin reactions, burns and allergic reactions mainly on extremely low birth weight infants but also can occur on adults.
Dose dependent toxicity of chlorhexidine is well demonstrated in vitro but in vivo results are rare and seem contradictory. Due to the toxicity of chlorhexidine, its effects on wound healing and specifically on burn wound healing should appear to be harmful but are not really known. Opinions of authors are heterogeneous in the literature.
However, the role of chlorhexidine in the prevention and treatment of infections have been clearly demonstrated.

What remains unclear is if side effects of chlorhexidine are more related to "how to use it" than about effects of the substance itself. Indeed, the alcohol contained in chlorhexidine solution can induce burns and mostly if the solution has not had the time to dry which therefore can induce a maceration of the skin in alcohol. Mistaken of identity between alcoholic and non-alcoholic solutions has been made owing to the same packaging design of the products. High concentrations of chlorhexidine often used are not proven to be more effective than lower concentrations and its toxicity is dose dependent. Therefore, there is no reason to use it at higher concentrations especially on fragile skin such as on burn wounds. Furthermore, another advantage that can be imagined may be the lower price of lower concentrations in aqueous solutions.
Finally, further studies are required on the effects of chlorhexidine on burns and adult populations.

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