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Emerging Trend of Thiolated Polymers/materials and nanomedicine in wound healing

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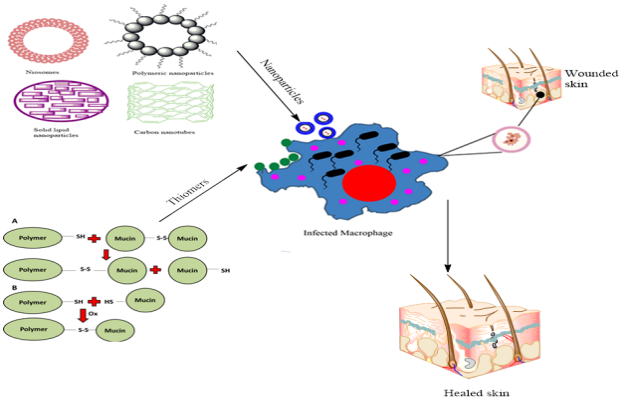
Abstract *Antimicrobial therapy failure against wound infections and arise of extended resistance highly encourages the discovery of novel mechanisms to overcome underlying issues. Major causative agent for causing wound infection is "Staphylococcus aureus" and recent medications lack antibacterial action, mucoadhesion and patient compliance. Thiolation proves itself as a novel technique for treatment of wound infections via topical application. Thiomers possesses features such as higher porosity, biodegradation, and swelling index. Thiolated products are resistant to the harsh environmental changes. Similarly, with the advancements in drug delivery, various smart drug delivery techniques lead to enhancement in drug delivery. Nanoparticles allow the drugs to penetrate the cells by diffusion or energy dependent process and degrade themselves at specified site of action by either attachment of recognition ligands or responsive stimuli. Overall, the Thiolated polymers and Nanomedicines depicted potential to cure wounds with increased severity meeting the required features of an ideal wound dressing.*

Key Words: Wounds, Thiolated Polymers, Microbes, Healing Cascade, Nanoparticles, Drug Resistance

Graphical Abstract



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Introduction

Skin appears to be largest organ of integumentary system consisting of the outermost tissue layer epidermis, that provides a protective barrier to underlying anatomical structures (Joshi & Patravale, 2008). while underlying to the epidermis is the dermis which is the source of nourishment.(Wokovich, Prodduturi, Doub, Hussain, & Buhse, 2006) The inner most layer is the hypodermis, which anchors the skin, provides support to the dermis and act as insulation due to adipose tissue. The skin functions as the first line of defense against microbes, which otherwise would have free access to internal organs resulting in fatal outcomes. Injury which results in breaking of skin is defined as a wound which disturbs defense mechanism by this physical, chemical, and biological trauma or injury.(Safferling et al., 2013)Wounds are classified in various ways. The most general way of classification is wounds without tissue loss and with tissue loss.(Dhivya, Padma, & Santhini, 2015)Wounds with tissue loss includes burn injuries (first and second-degree burns) caused by both thermal, electrical, chemical or radiation the sources and diabetic foot ulcers. Wounds without tissue loss include laceration and first-degree burns which have the superficial effect to epidermis only. Wounds are also classified the on the basis of their morphology as tidy wound and untidy wounds. Tidy wounds contain no damaged tissues and are caused by sharp instrument incisions. Untidy wounds are due to crushing, tearing and burns and results in vascular damage to the skin. (Li, Overend, Maitz, & Kennedy, 2012)In terms of duration, wounds are categorized as acute and chronic. Acute wounds (Minor surgeries or trauma) healing time is not more than one week. While chronic wounds (Pressure ulcers, venous leg ulcers) fail to heal in approximate time duration and often reoccur and healing may require months to years. On the basis of wound depth in skin tissues, wounds are categorized as superficial, partial thickness and full thickness wounds. In case of superficial wounds, the only epidermis is affected (abrasions and blisters). In terms of partial thickness wounds, epidermis and dermis are damaged and often bleeding occurs. In full thickness wounds the epidermis, dermis, subcutaneous, underlying fatty tissues, bones, muscles, and tendons are damaged.(Nichols & Florman, 2001)

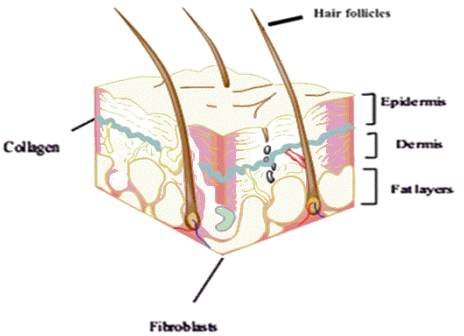

Figure 1: Anatomical structure of the skin

Pathogenesis of wound infection

Microbial infection at the site of wound complicate wound healing as shown in Figure 2.(Kumar et al., 2013) A well-established balance and commensalism between human and microorganism exist under normal physiological conditions. Due to this mutually

beneficial relationship, endogenous bacteria significantly contribute to maintaining the normal flora and natural habitat. (Bowler, 2002)Natural habitat of bacteria especially, on host skin is advantageous for the host in a way that it prevents overgrowth and colonization of antibiotic-resistant bacteria. (Broughton 2nd, Janis, & Attinger, 2006) The loss of skin integrity due to any injury produce wounds leading to the disturbance in the normal flora of the skin. This eventually disturbs endogenous bacteria habitat and facilitate bacteria to colonize a new habitat and becomes pathogenic at the wound site (Kühne, Ullmann, & Kühne, 1985). Thus, the skin is exposed to microbial infections and wound care management becomes difficult. The process of wound infections results in increased wound exudates containing nutritional proteins for bacteria which further helps in their proliferation, delays healing and results in improper collagen deposition.(Oestem & Tscheme, 1984). The wound is sterile at the initial stage but rapidly becomes colonized with gram-positive bacteria majorly *Staphylococcus aureus* as well as *Staphylococcus aeruginosa* which impede the human immune system by degrading neutrophil extracellular traps (first defensive mechanism) and after reaching increased bacterial density wound gets infected and chances of sepsis becomes obvious. Infectious wounds impose serious complications leading to high morbidity and mortality. There are also some risk factors which obstruct and complicate the healing process(King, Cohn, Proctor, & Group, 2004). Diagnosis as well as management are controversial and varies among clinicians.

- Microbial related risk factors (*Staphylococcus aureus, Staphylococcus aeruginosa)*
- Host-related risk factors (obesity, co-morbidity, the severity of disease, aging, malnutrition. lack of protein-calorie diet, diabetes, hypertension, and systemic infection)
- Operation related risk factors (Hospitalization, traumatic injury of tissues)

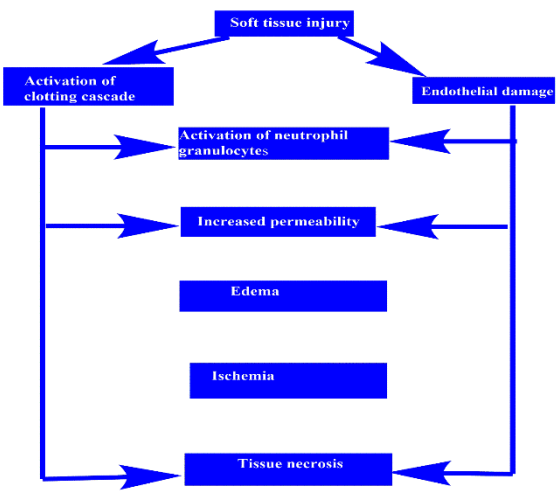

Figure 2: Pathogenesis of wound infection in different steps

Cell signaling and Response to wound Healing

Wound healing is designated as a complicated

biological process with factors influencing and possess potential to delay healing. Different cell signal such as growth factors as well as cytokines control the wound healing cascade. The growth factors control cell migration, matrix production, enzyme production, and cell differentiation. These factors are present in very small quantity performing their regular function, however, the presence of neutrophils at wound site release enzymes destroying these factors. These cytokines regulate the functions via endocrine, paracrine, and autocrine mechanisms as shown in Fig. The specific activity of a cytokine is induced by its attachment to a specific cellular receptor which initiates intracellular signals and producing a specific response. Excessive healing is characterized by stricture, fibrosis, adhesions, as well as contractures which show hypertrophic scars due to excessive tissue deposition and excessive contraction of tissues. Deficient healing is the other type which shows insufficient tissue and matrix deposition producing weakened tissue. Regeneration is the most sophisticated form of healing and regeneration in which the tissue is replaced with exactly what was lost in injury. Regeneration is found to a very limited capacity in human beings. Regeneration and tissue repair process comprise of a series of molecular and cellular events starting after the tissue lesion to restore tissue damage. Tissue repair is a simple linear process resulting due to dynamic changes involving the production of soluble mediators, blood cells, enzymes, and extracellular matrix. The process involves various blood cells, mediators, and extracellular matrixes. Wound healing follow the four mechanisms strictly i.e. hemostasis, inflammation, epithelialization as well as remodeling at injury site (Sudheesh Kumar et al., 2012) as shown in Figure 3.

Hemostasis

First stage of wound healing is hemostasis. Hemostasis begins after the wound formation or tissue injury due to the disruption of blood vessels and results in the release of clotting factors responsible for vasoconstriction and clot formation. Hemostasis mechanism is based on the following mechanisms as mentioned below:

1. Vascular constriction
2. Induction of the platelet plug
3. Blood clot formation
4. The closing of the wound hole with fibrous tissues permanently

Constriction of smooth muscles is followed by sealing with platelet plug. Platelets have cytoplasm which contains contractile actin and myosin molecules along with contractile protein thrombosthenin. Platelet's cytoplasm also has storage capacity for calcium ions as well as ATP and growth factors. After platelet plug formation, Blood clotting gets started in wounded vessels within seconds to minutes. Promoter substances from wounded vessels, from platelets and from blood proteins initiate blood clotting. Least step in hemostasis is an invasion of fibroblasts at the wound site and dissolve the preformed clot and this process requires 1-2 weeks for completion(Paul & Sharma, 2004)

Inflammation

As bleeding stops, proinflammatory cells initiates inflammation.(Ribeiro et al., 2009) Macrophages and neutrophils are responsible for eradicating wound exudates and invading microbes.(Renuka, Nishadh, Jigar, & Tejal, 2012) Hence macrophages ease the process of inflammation by inducing apoptotic cells as well as clearing them and changing the defective phenotype of various apoptotic cells into their repairing state. The apoptotic cells result in the production of fibroblasts as well as keratinocytes that initiates angiogenesis and tissue regeneration. (Yin, Fei, Cui, Tang, & Yin, 2007)

Re-epithelialization and Remodeling

In epithelialization, fibroblasts and endothelial cells play an important role thus helping in the

growth of capillaries, collagen and granulation tissues thus promoting angiogenesis and proliferation which activates healing and growth (Roselli, Finamore, Garaguso, Britti, & Mengheri, 2003)After epithelialization and proliferative phase, wound undergoes neo-vascularization, physical contraction. Neovascularization results in the synthesis of various new capillaries and blood vessels, while the physical contraction in wound occurs due to the release of contractile fibroblasts and collagen release at this stage. All these factors are responsible for remodeling and regain in the skin texture.(Dohmen et al., 2011)

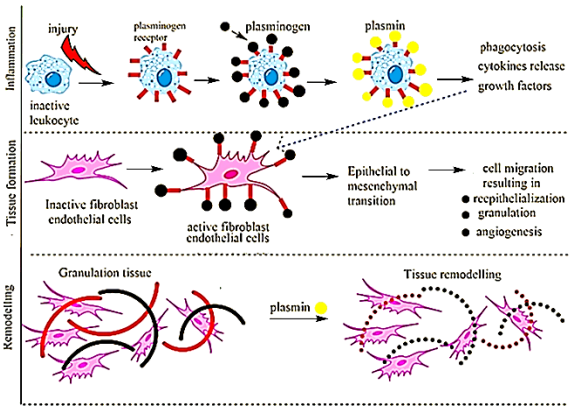

Figure 3: Mechanistic Representation of four stages of wound Healing

Treatment Goals

Wound healing a complex process that can be compromised by both exogenous (microorganisms) and endogenous (pathophysiological) factors. Microbial colonization of both acute and chronic wound is inevitable and in most cases, endogenous microbes predominate. The risk of infection increases as local conditions favor bacterial the growth rather than host defense.

There is high recognition of the concept of critical colonization when wound healing may be delayed in the absence of typical clinical features of infection. The progression from wound colonization to infection depends on various factors like the bacterial count, species, host immune response, virulence, and synergistic interaction present between different organisms. It's been suggested that bacteria within chronic wounds live within biofilm communities, where they are protected and develop resistance against antibiotics. Consequently, the prime objective of wound management is to restore the host-bacterial balance by ensuring that wound is cleared from cell debris and microbes. First line therapy for wound infections includes antibiotics, among which cephalosporin are the first drug of choice. But due to cephalosporin's allergic reaction issues, drug line switches to vancomycin, metronidazole, clindamycin, and aminoglycosides. Other available medications are protective dressings (gauze, moist cotton gauze, and antiseptic solutions), antibacterial dressings (iodine dressings, silver-based dressings), absorbent dressings (foams, hydrogels, and hydrofibrils) and autolysis debridement's (films, hydrogels). (Dumville, Gray, Walter, Sharp, & Page, 2014)Nanoparticles based dressings (bandages). But the main disadvantage of oral

antibiotics is non-compliance, economic burden, and antimicrobial resistance while dressings lack mucoadhesion and antimicrobial action. They also have solubility related issues.(Kandavilli, Nair, & Panchagnula, 2002) Chronic wounds are difficult to heal because of heavy bleeding and increased microbial load and this alarming situation highly evoke the need for ideal topical bandage/dressing. An ideal dressing has the capacity to resolve the solubility issues. Minimize the toxicity associated with non-degradable polymers by utilizing biocompatible and biodegradable natural polymers. Increase hemostatic potential, mucoadhesion, antimicrobial activity, degradation, swelling, porosity, wound healing and control drug release. (Rangari, Kalyankar, Puranik, & Chaudhari, 2012)

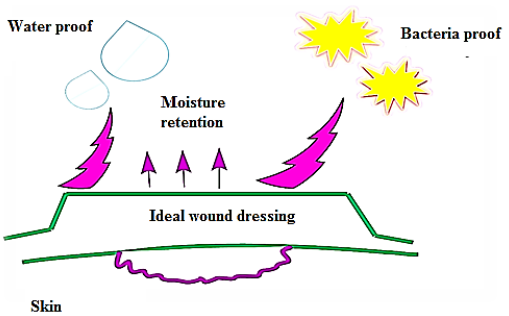

Figure 4: Characteristics of the ideal wound dressing

Thiomers (Thiolated Polymers) and their Mechanistics

Thiolation is the process of conjugating thiol groups (thioglycolic acid or N acetyl cysteine) to any moiety *via* different chemical reaction. Mostly, biocompatible and biodegradable polymers (chitosan, polyacrylates) are conjugated with thiol group resulting thiolated polymer or thiomer having superior properties compared to unmodified polymers. Many research groups have reported the superiority of thiolated polymers for their applications in various dimensions of drug delivery and therapeutics. Lacrimera, a thiomer (Chitosan-N acetylcysteine) based first commercial product is available in the European market. These terminal thiol groups, once in the body, develop disulfide linkage by exchange or simple oxidation-reduction reactions as shown in Figure 5 with cystine-rich subunits present in the mucosa, glycoproteins or any other place resulting in stronger mucoadhesion(Bernkop-Schnurch, 2005), (Bernkop-Schnurch, Hornof, & Guggi, 2004).Amide or amidine bond formation is the usual mechanism for thiomer synthesis between thiol-bearing moiety and polymer backbone. The reactivity of thiomer is strongly dependent on pka of thiol-bearing ligand attached to the polymer. The low the pka is, the more thiolated anions are available at physiological pH showing more reactive sulfhydryl groups for developing disulfide bond e.g. N acetylcysteine (pka=8.2) is more reactive than cysteine (pka = 8.4) or thioglycolic acid (pka= 10.3). These thiomers have are explored and reported in 4 major areas, mucoadhesion, in situ gelations, permeation enhancement, and efflux pump inhibition. Mechanism of thiolation is based on the "immobilization" process. Immobilization is a promising process of making a polymer inert, confined, localized and reusable thus ensuring it advance safety, therapeutic efficacy and stability thus rendering it resistant to adverse changes. Immobilization can be achieved by physical entrapment, microencapsulation, adsorption and covalent

cross-linking (Roldo, Hornof, Caliceti, & Bernkop-Schnürch, 2004). Various anionic and cationic polymers are conjugated with thioglycolic acid to enhance their mucoadhesive, healing and advanced therapeutic properties. Cationic polymers most commonly included chitosan and its derivatives and it can be conjugated with thioglycolic acid, as a result, thiol groups are immobilized by chitosan amino group at position-2 of glucosamine subunits. Similarly, an anionic polymer like alginate is conjugated with cysteine to achieve immobilization of thiol groups(Naz et al., 2016) as shown in Figure 6. Thiolation can be achieved by the covalent crosslinking method. In this way, thiolated polymers are stable and overcome the nanoparticles stability issues in the wound healing medications as well as mucoadhesive because of high retaining capability at the wound surface.(Carvalho, Bruschi, Evangelista, & Gremião, 2010; Roldo et al., 2004; Sohail et al., 2016)

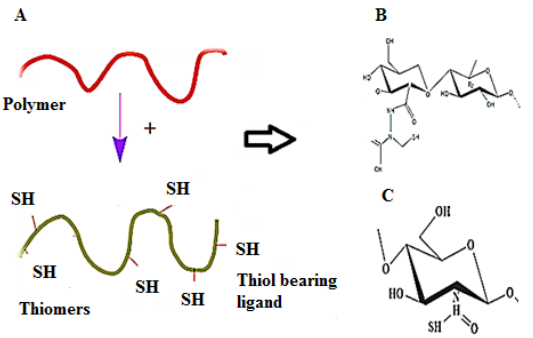

Figure 5: Attachment of thiol-bearing ligand to the polymer for Thiolation. (A) Thiolated alginate (anionic thiolated polymer) B and Thiolated chitosan (cationic thiolated polymer) C

Role of Thiolation in wound Healing

Proper healing is ensured in an ideal healing environment. Ideal healing can be achieved with the medication which can be accomplished with stability, safety, efficacy, porosity, swelling, degradation, hemostasis, drug release, and mucoadhesion.(Carvalho et al., 2010) Thiolated polymers accelerate wound healing by aggrandizing solubility, porosity, swelling, mucoadhesion , permeation and inhibition of P-gp. Polymers present in the dressings have different solubility issues and may degrade at wound pH and shows abrupt drug release.(Daunton, Kothari, Smith, & Steele, 2012) Such solubility issues can be resolved by thiolation of the polymer because thiolation renders the polymer in situ gelling properties at the wound site. In situ gelling property of the thiolated polymer is achieved due to the strong cross-linking and inter/ intra molecular interactions (Carbinatto, De Castro, Evangelista, & Cury, 2014). Moreover thiolated polymer is instilled with safety, stability, and efficacy due to its basic mechanism of immobilization. Porosity is dependent on the polymeric solution and cross-linking method. Thiolated polymers based dressings will be highly porous due to stronger cross-linking and slow freeze-drying. The porosity of the wound dressing accelerates the absorption capacity of the infectious wound exudates as well as the rate of colonization, cellular organization and angiogenesis of the skin (Barrett, Joyner, & Halenda, 1951) Thiolated polymer induce swelling because of its increased porosity and strong adhesion capabilities. Ideal swelling is responsible for proliferation and distribution of

cells as well as the transfer of oxygen and nutrients through polymers of the dressing at the wound site. (Begam, Nagpal, & Singhal, 2003)A most important feature of thiolated polymers is their enhanced mucoadhesion. Mucoadhesion is the most important and promising factor responsible for proper dosing, patient compliance, and cost-effectiveness.(Heggset, 2012) Mucoadhesion is achieved because of immobilization of thiol groups due to thiolation and secondly due to the medium molecular mass of both cationic and anionic polymers. Thiolated polymers have increased polymeric strength and are robust enough to resist any mechanical pressure, so they promote the rate of dry mass loss and biodegradation. Thiolated polymers are highly hemostatic because of strong intermolecular interaction between any cationic polymer (positively charged) and blood cells (negatively charged).(Kandavilli et al., 2002; King et al., 2004; Takeuchi, Matsui, Yamamoto, & Kawashima, 2003) Hence, thiolation embellishes and facilitates the overall process of wound healing due to increased mucoadhesion, absorption and antibacterial properties that help tissue repair and angiogenesis bringing normal skin back into position.(Quirynen, Bollen, Eyssen, & Van Steenberghe, 1994)Tyrosine subunits are responsible for the opening of tight junctions thus allowing permeation. But these subunits are prone to dephosphorylation of tyrosine phosphatase. Permeation is responsible for inhibiting tyrosine phosphatase due to glutathione (GSH) phosphorylating capacity. But GSH is readily oxidized by GSSG thus limiting permeation. Thiolated polymers help in the balancing of GSH/GSSG shift, encouraging GSH production resulting in reversible tight junction openings. P-glycoprotein (P-gp) is an important protein of the cell membrane dependent on ATP efflux pump and responsible for leaking foreign substances outside the cell. Thiomers are helpful in sustaining and retaining of the drug by inhibiting P-gp by transforming the fluidity of the cell membrane. This inhibition results in the blockage of P-gp ATPase and allosteric membrane transporter site which are important for moving of drug outside the cell membrane.

Table 1. Thiolated Polymers as a source of Mucoadhesive TDDS

S. No	Polymer	Thiolation	Dosage Form	Conclusion	References
1	Carboxymethyl-Hyaluronic-Acid	Thiolated Carboxymethyl-Hyaluronic-Acid	Cross-linked gels and films	Minimized wound closure time and reduced risk of the infection and pain as compared to non-thiolated polymer	(Yang,Prestwich, & Mann, 2012)
2	Chitosan	Chitosan-TGA	Multifunctional hydrogels for chronic wound treatment	Increased mucoadhesiveness and inhibitory activity against Staphylococcus aureus than simple un-modified chitosan	(Stefanov, Hinojosa-Caballero, Maspoch, Hoyo, & Tzanov, 2018)

S. No	Polymer	Thiolation	Dosage Form	Conclusion	References
3	Alginate	Alginate cysteine	itu thiolated alginate hydrogel	Increased wound healing and proficient hemostasis	(Xu et al., 2016)

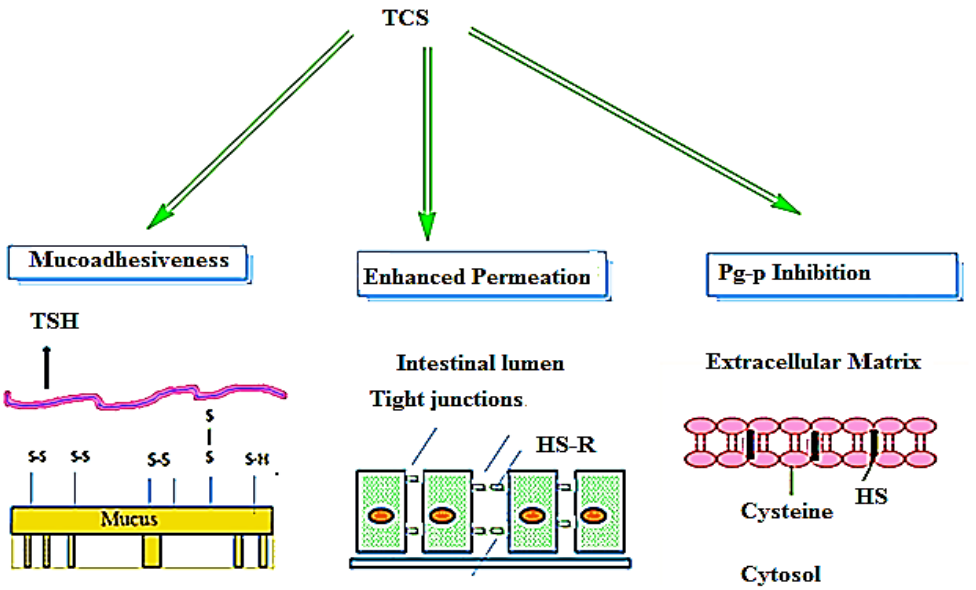

Figure 6: Enhanced features of thiomers i.e., Mucoadhesiveness, permeation and Pg-p inhibition

Advantages of Nanomedicine over Traditional Drugs

Traditional medicines are mostly of plant and animal origin and have been widely used to treat several pathogenic as well as hormonal disorders. Now days due to acquired resistance of pathogens as well as drug resistant food are contributing towards the failure of traditional medicinal therapy. Moreover multi drug resistance has also been increased from 1983 to 2012, due to decreased number of USA approved antibiotics (Spellberg, 2009) . Main problem with immensely used traditional medicines i.e, antibacterial and antifungals is that these medicines are non- site specific. Traditional antibiotics circulate in the whole body, only minimal quantity reaching the site of action thus rendering minimal efficacy and maximum adverse effects. Moreover, all of the traditional medicines followed immediate drug release, low bioavailability and shorter half- life leading to inability to cross biological barriers. With the advancements in drug delivery various smart drug delivery techniques and novel nanomedicines lead to enhancement in drug delivery. Nanoparticles bypass lack of traditional medicinal therapy by preventing their degradation by directly penetrating the cells by diffusion or energy dependent process, rather they get entrapped in endosomes and degrade themselves at specified site of action by either attachment of recognition ligands or responsive stimuli. Advantages of nanomedicine and their endocytic pathways are discussed in detail in site-specific targeting.

Site specific Targeting

Site specific targeting is designated as smart

drug delivery to attain maximum and effective treatment goals (Mishra et al., 2016). For achieving successful site specific drug delivery action, the drug must retain at the site of action followed prolonged release and bypass degradation (Ladaviere & Gref, 2015).

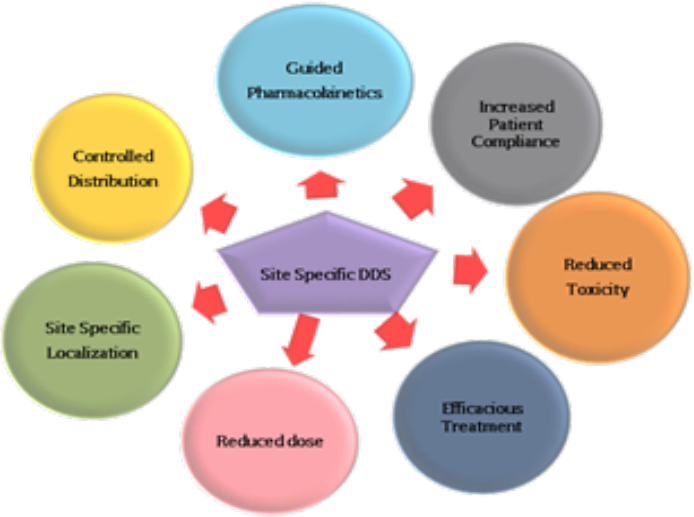

Figure 7: Advantages of site-specific drug delivery system as compared to the Traditional Medicines.

Endocytic Pathways for Site Specific Delivery of Nanomedicine

Nanoparticles undergo site specific targeting via bypassing the first pass effect by enduring themselves with specific endocytic pathways which allow them for site specific drug delivery. Endocytic pathways can be classified as

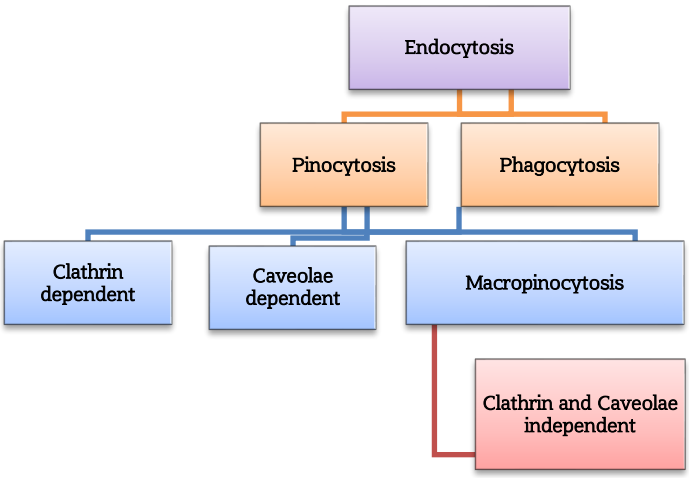

Figure 5: Classification of endocytosis pathway of nanoparticulate system

Phagocytosis

Phagocytosis is a vital step in endocytosis. Opsonins recognize nanoparticles and then bind with receptors followed by formation of phagolysosome on combining with lysosomes to release entrapped drug.

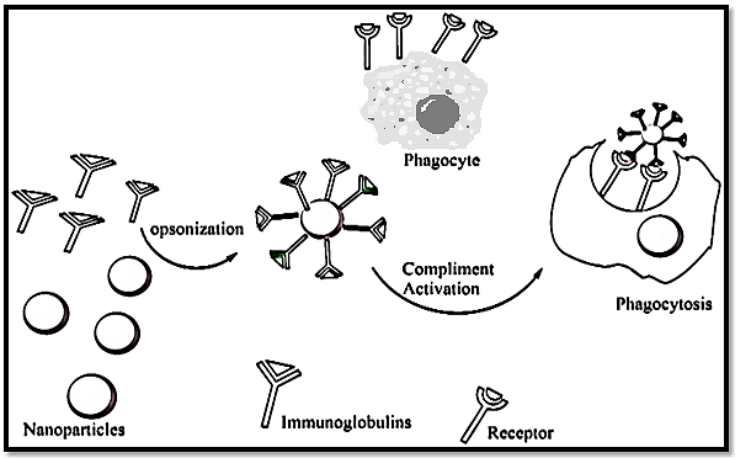

Figure 6: Nanoparticles uptake by Phagocytes and lysosomal Degradation

Pinocytosis

Pinocytosis leads to the functionalization of all other types of endocytosis like clathrin dependent endocytosis, caveola dependent endocytosis, macropinocytosis and clathrin and caveola dependent endocytosis. Clathrin-dependent endocytosis is present in all mammalian cells, occupying an important part in cellar entry. After the interaction of nanomedicines with the receptors present on the cell membrane, clathrin -1 protein is formed. Nanomedicines get transported and engulfed in the clathrin protein, and this protein has ability to wrap them inside the vesicle. Vesicle is then released by the activity of dynamin. GTPase resulting in the formation of clathrin coated vesicles (CCV). CCV can locomote inside the cytoskeleton with the help of energy provided by actin. In the end, coated clathrin sheds of in the cytosol. Macropinocytosis transport is present in the brain and is clathrin and caveola independent endocytosis.

Lipid Based nano Formulations for wound HealingSolid lipid nanoparticles

In solid lipid nanoparticles, liquid lipid gets replaced with solid lipid. Some effective SLNs formulations for wound healing include SLN based silver sulfadiazine wound dressings, in which scaffolds are composed of Chitosan glutamate or HPMC. Whereas Sodium hyaluronate and chondroitin sulphate are bioactive polymers for initiating angiogenesis of the skin and they form the basics of SLNs. AgSD encapsulated in the SLNs in the presence of platelet lysate (PL) was loaded in aforesaid scaffold to enhance the spongy effect of wound scaffold and results in maximum softness and elasticity to provide comfort to the patients. Results also confirm that SLNs based AgSD are highly efficacious in treating difficult wounds and skin lesions alongwith eradication of wound colonization and antibacterial resistance.(Sandri et al., 2013) Similarly essential oil (eucalyptus and rosemary) loaded SLNs (Natural lipids, cocoa butter) were synthesized by shear homogenization following

application of ultrasonics for enhancing wound healing of skin.(Saporito et al., 2017) Advanced SLNs for treating wounds with highest efficacy include morphine loaded SLNs. Morphine loaded SLNs were synthesized (around 180 nm), and wound closure was noticed in human mimicked 3D wound model. The results were highly promising as they show aggrandize and maximum wound closure rates, angiogenesis, minimized cytotoxicity and no hypersensitivity reactions at all. Moreover another novel approach involves encapsulating morphine in the SLNs, which depicted prolonged drug release, thus promoting an advanced step towards accelerated wound healing.(Küchler et al., 2010) To get maximum activity against notorious pathogenic bacteria responsible for worsening wounds, burns and surgical incisions can be achieved in a best way by encapsulating antimicrobial peptides into SLNs.(Caldon, 2013)

Ethosomes

Ethosomes open new era for nano targeted drug delivery deep inside the skin and systemic circulation. Ethosomes bypass various other drug delivery systems because of their safety, stability, efficacy, and simplicity in preparation (Touitou, Dayan, Bergelson, Godin, & Eliaz, 2000). Ethosomes have the ability to encapsulate various lipophilic drugs. Ethosomes possessed increased bioavailability and penetration ability due to its composition specialty i.e. based on lipids and ethanol. While curcumin is well reputed for its wound healing properties and tissue repair, but it has low penetration capability and low bioavailability which renders it weak candidate for treating severe wounds. To overcome its low bioavailability issues, it was formulated with enhanced bioavailability ethosomes to overcome biological barriers and to achieve prolonged and sustain drug levels. Results concluded that ethosomal curcumin formulations produced pronounced advanced wound repair and anti-inflammatory as well as antimicrobial action on murine model.(Partoazar et al., 2016) Moreover, transdermal ethosomal gel was also prepared to deliver 5-fluorouracil in mice wounded scar model. Results deduced that ehthosomal gel combination with 5-fluorouracil reduced its toxicity and endured it with biocompatibilty and healing features (Wo et al., 2014). Skin wounds after worsening, get inhabited by staphylococcus bacteria and these pathogenic bacteria cause various dermal and subdermal infections. Now a days, such infections are being treated with invasive antibiotics which have various side effects along with, i.e. toxicity, poor bioavailability, drug resistance, and patient non-compliance. To alleviate all side effects of invasive antibiotics novel transdermal ethosomal carrier of antibiotic (erythromycin) was formulated and tested on murine model by developing severe wounds infected with *Staphylococcus aureus* and promising results regarding eradication of *Staphylococcus aureus* and healing were obtained (Godin, Touitou, Rubinstein, Athamna, & Athamna, 2005)

Transferosomes

Transferosomes are sophisticated nanoformulations which possess the ability to endure stress with high adaptability. Structurally they are very soft, elastic and deformed vesicular formulations having an inner aqueous core and outer complex lipid bilayer. Owing to the ultra-elastic nature of transferosomes they are able to squeeze through the skin pores of very small size. Tocopherol loaded transferosomes were reported to be very efficient in wound healing. Different polysorbates (Tween 20, 40, 60, 80) were loaded with Tocopherol acetate to develop tocopherol loaded transferosomes.

Developed Transferosomes were identified as efficient candidates for delivering tocopherol topically into the skin and showing biocompatibility, improved healing, protection from oxidative damage and promotion of cell proliferation and migration. (Balogh, Hagnauer, Tomalia, & McManus, 2001)

Liposomes

Liposomes appear to be spherical vesicle comprising of lipid layers with framework of phospholipids (Williams & Barry, 2012). Liposomes are best for encapsulating different drugs and it is an effective vehicle for various formulations. Growth factors exist in many forms and insulin growth factors are most common among them. Insulin growth factors are involved in skin growth, angiogenesis and reepithelialization. But insulin growth factor abruptly releases in the body and results in either hyperglycemia or hypoglycemia. Research was therefore conducted in 1997 to regulate the insulin growth factor in a proper and controlled way by combining them in the liposomal formulation. Insulin like growth factors in liposomal formulation were efficient in reepithelialization of burn wounds without causing any side effects of abrupt release.(Pierre et al., 1997) Another remarkable research was conducted on liposomal hydrogel with povidine-iodine to efficiently heal the wounds by reducing inflammation and oxidative stress caused by reacting oxygen species. Invitro studies confirmed that liposomal povidine-iodine hydrogel was highly efficient in effectively minimizing the inflammatory cascade and oxidative stress damage, thus resulting in efficient wound healing(Reimer et al., 2000). Similarly epidermal growth factor loaded with liposomes was incorporated into chitosan gel for efficiently and actively treating second degree burns by inducing reepithelialization and promoting angiogenesis.(Değim et al., 2011)

Dendrimers

Morphology of dendrimers is comprised of various polymeric components such as core, dendrons and surface active groups. Dendrimers possess such unique features that the drug can be incorporated in their internal structure or physically linked on their surface. There is invasive novel research on wound healing activity enhancement of the dendrimers. Dendrimers lead to hallmark in wound healing after the revolutionary patented discovery (US62247=898B1) of antimicrobial dendrimers nanoparticles. These dendrimers were synthesized by incorporating metallic silver nanoparticles as a strong antimicrobial agent in the dendritic polymer. This formulation proved to be less toxic and more beneficial in case of wound healing. (Balogh et al., 2001) Similarly, in past polyvalent dendrimers after conjugating with glucosamine subunits resulted in prevention of scar formation , thus leading towards cosmetic effects.(Shaunak et al., 2004) Moreover, cationic dendrimers have been used to promote angiogenesis, growth and healing in diabetic patients by conjugating with plasmid DNA (encodes VGEF). For in vivo testing, these cationic conjugated dendrimers were injected subcutaneously in the mice skin. This highly efficacious and novel formulation lead to the new horizons for wound healing as results obtained were very promising in case of proliferation , wound healing and angiogenesis. (Kwon et al., 2012) Another novel approach involved electrospunning of silver acetate onto the constructs of nano fibers after the conjugation of star shaped and highly branched dendrimers (PAMAM) with the gelatin via EDC. This formulation was considered to be most effective in eradicating *Staphylococcus aureus* and *Staphylococcus aeruginosa* bacteria, which

are responsible for worsening skin wounds.(Dongargaonkar, Bowlin, & Yang, 2013)

Metallic Nanoparticles

They impart enhanced antibacterial action due to reduced particle size as well as by generating reacting oxygen species (ROS). ROS damage bacterial cell wall via localized action, internalization, and trafficking of nanoparticles because of loss of proton motive force as well as uptake of toxic ions. This results in inhibition of bacterial cell growth and necrosis. A research was conduct on noble metals platinum and palladium to reveal their antioxidant property by preventing the skin from atrophy.(Shibuya et al., 2014)

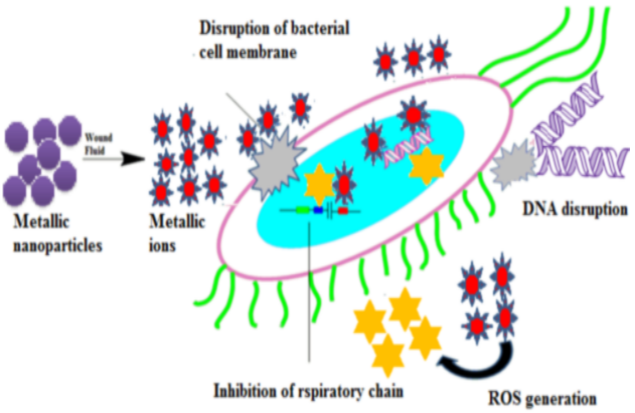

Figure 7: Interaction of metallic nanoparticles with the bacteria present in the wound fluid and causing the disruption of that bacteria.

Surfactant Based Niosomes

Niosomes consist of non-ionic surfactant and cholesterol. Since past, Niosomes possess increased bioavailability and reduced toxicity. For wound healing purpose, a research group identified a novel technology of incorporating puniglacin (obtained from the source of Punica granatum) into niosomes and resulting formulation was niosomal gel of puniglacin. Punica granatum (pomegranate) is innately featured with antimicrobial, antioxidant and anti-inflammatory capabilities, therefore it is considered as Chinese traditional medicine from many years. But it is entitled with certain deficiencies like low bioavailability, poor half-life etc Results revealed that puniglacin niosomal gel was efficient in wound healing in appropriate way(Chidambara Murthy, Reddy, Veigas, & Murthy, 2004). Similarly, phenytoin sodium loaded niosomal gel formulation via evaporation film hydration methodology for wound healing was founded remarkable in closure of the skin injuries by enhancing the penetration capacity of phenytoin the skin(Ali, Sarhan, & Magdy, 2014) Moreover, for eradicating microbial contamination of *Pseudomonas aeruginosa* efficiently and effectively , novel moxifloxacin loaded niosomal gel was synthesized.(Sohrabi, Haeri, Mahboubi, Mortazavi, & Dadashzadeh, 2016)

Conclusion

Wound healing is a dynamic biological. Among Bioadhesive drug delivery system thiolated polymer exhibit unique position because disulphide bonds between thiolated polymers (thiomers) and cysteine-rich subdomains of mucus glycoprotein provide enhanced mucoadhesive properties. The foremost advantage of thiomers is its innate property of wound healing due to its characteristic porosity, swelling ability, biocompatibility and biodegradation. Thiomers withstand high pressure and pH due to its cross linking effects and provide permanent immobilization characteristic to dressing. While nanoparticles overcome the resistance and low bioavailability of antimicrobial drugs and pass all the hurdles of skin barriers and with control release effect and act as antioxidant to prevent oxidative stress caused by microorganism. Inspite of all, thiomers and nanomedicines depicts potential as advanced therapeutic agent for treating wounds of all severities with cost-effectiveness effeciently and effectively.

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