Microneedles for drug delivery: trends and progress

Citation: CHEUNG, K. and DAS, D.B., 2015. Microneedles for drug delivery: trends and progress. Drug Delivery, doi: 10.3109/10717544.2014.986309.

Microneedles for Drug Delivery: Trends and Progress

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ABSTRACT

In recent years there has been a surge in the research and development of microneedles, a transdermal delivery system that combines the technology of transdermal patches and hypodermic needles. The needles are in the hundreds of micron length range and therefore allow relatively little or no pain. For example, biodegradable microneedles have been researched in the literature and have several advantages compared to solid or hollow microneedles, as they produce non-sharp waste and can be designed to allow rapid or slow release of drugs. However they also pose a disadvantage as successful insertion into the stratum corneum layer of the skin relies on sufficient mechanical strength of the biodegradable material. This review looks at the various technologies developed in microneedle research and shows the rapidly growing numbers of research papers and patent publications since the first invention of microneedles (using time series statistical analysis). This provides the research and industry communities a valuable synopsis of the trends and progress being made in this field.

KEY WORDS: Microneedle, time series analysis, autoregressive integrated moving average (ARIMA)

1. INTRODUCTION

Drug delivery (DD) has historically been pivotal in ensuring drugs can be administered in a manner that leads to therapeutic efficacy. Methods of DD, such as oral ingestion or hypodermic injections are considered to be the most common forms of drug administration [1, 2]. However, they possess several limitations, e.g., pain associated with hypodermic injections due to the long needles piercing nerve endings and absorption and metabolism issues associated with oral administration that lead to variability in bioavailability and also side effects from metabolites [3]. These disadvantages have led to the development of alternative DD methods. An encouraging alternative to the traditional methods is DD directly through the skin surface, i.e., transdermal drug delivery (TDD). Commercially available transdermal patches (TDP) now exist

which provide controlled release of medicines to patients in a minimally invasive manner. However, large molecules cannot passively permeate through the stratum corneum (SC), thereby, limiting the number of such transdermal drug products available to patients. Problems can also arise from the TDP's peeling off due to the long periods for which they are required to be applied. Several approaches have been researched to overcome the problems with TDPs and allow delivery of compounds like proteins, DNA [4-9], so that they can permeate through the skin.

1.1 DD Routes

An overview of DD routes are described in this section briefly to provide some perspective on this important area of research, in particular, to provide an understanding of why there is a need to develop new technologies and where microneedles fit in to the picture. Numerous forms of DD routes are currently being used, which include, oral administration (gastric, colonic, enteric, etc.), hypodermic injections (e.g., intra-venous, intra-muscular, intra-cranial, sub-cutaneous injections, etc.), inhalation (pulmonary) and TDD (skin appendages) [10, 11]. Oral administration and hypodermic injections are the most common delivery methods with approximately 80% of drugs administered orally. However, several difficulties associated with oral dosage forms exist, e.g., pH changes within the body causing degradation to drugs, enzymatic activity, variable transit time, side effects and first pass metabolism [10]. The main disadvantages of using hypodermic injections is the resultant infection, pain caused during application, patient fear, anxiety and patient incompetence.

A good alternative that can overcome such problems is transdermal delivery of drugs (TDD), which can resolve issues such as by-passing first-pass metabolism (thus eliminating harmful metabolites whilst increasing bioavailability), and patient compliance. The method operates by transporting drug molecules from the surface of the skin into the body. It has been continuously developed ever since the first transdermal product was approved in 1979 in the United States to treat motion sickness [12]. Transdermal delivery includes the applications of gels, creams, ointments and more recently transdermal patches [13-15]. One transdermally delivered drug that is commonly used is nicotine [16], first developed in 1984 by Jarvik and Rose [17] to help smokers give up smoking. The drug was FDA approved in 1991 and has been on the market since 1992 [13].

However, there are disadvantages associated with TDPs, for example, the prevention of large molecules from bypassing the SC, the outermost layer of the skin, which is the rate-limiting barrier [18-21]. TDPs are applicable for molecules that are traditionally smaller than 500 Da [22]. Methods that have been employed to solve the short comings of both TDPs and hypodermic injections are ultrasound [20, 23, 24], MNs, iontophoresis [25], electroporation,

chemical enhancers, and others [23, 26]. These techniques can overcome the protective SC barrier as they allow the passage of large molecular compounds such as proteins and DNA [4, 5, 11, 27, 28]. MN technology in particular has grown over the past 15 years and can permit drugs to bypass the SC layer by the insertion of micron sized needles that create micro channels through the SC [29-31]. The MNs are small enough in length to avoid touching nerve endings of a patient, thereby, causing little or no pain [32]. Furthermore, MN technology has shown to be more advantageous in comparison to the other TDD techniques [33], such as the ability to deliver large molecular molecules that are larger than 500 Da [22] and the versatility in the application to allow solid or liquid formulations to be developed for disease specific applications.

There are numerous journal papers that have reviewed different types of drug delivery technologies [9, 34-41] and specific drug deliveries, such as insulin [42-48]. A number of review papers have also discussed specific technologies, e.g., various methods to fabricate MNs, or the devices that are currently being used or are likely to be used in clinical trials [49, 50]. Indeed one can safely assume that the most significant aspects of MNs research have been discussed in review or research papers. However, one issue that is obvious is that there is little attempt to quantify the trend in the progress of MN technology. In other words, it is not clear how slow or fast the rate of progress is the development the MNs based methods are. It is also not clear from the existing literature what method one could use to quantify the trends and, if the trend could be quantified reliably given that the MNs based research is still relatively new as compared to most other TDD methods. This review paper will look into assessing these gaps in the literature by using a time series analysis of the journal papers found in Scopus, using a time series analysis tool namely, ‘autoregressive integrated moving average (ARIMA) model (also known as univariate Box-Jenkins analysis) to look at the future trend patterns in the number of publications based on several key word searches. The information was gathered by conducting searches in the home page document search function only, as opposed to searching “microneedle” and then searching within the results the second key work (for example “solid”). Quantifying and predicting past and future trends are important to determine the market values of these products as there is a growing interest to produce MN’s to a commercial quality and scale. It is therefore important to view what is the current trend in literature, similar to how a start-up a business would require market research. We assume that the number of publications is the key indicator of how well the MN technology has developed and use the keywords “microneedle”, “solid microneedle”, “hollow microneedle” and “dissolvable microneedle” to determine the number of publications corresponding to each keywords (using Scopus). The program IBM SPSS Statistics version 21 was used to produce the time series analysis tool and analyse the data from Scopus. The ARIMA model was created by omitting 2014 data as it is

currently incomplete, therefore the analysis will give an indication of the trend till 2013 [51]. For the completeness of the paper, we discuss other relevant issues as well as follows.

1.2 Why microneedles?

TDPs have been delivering a variety of drugs since the early 1980's such as testosterone, nicotine, Selegiline and Clonidine [33]. Although there are several commercially available TDPs, they are limited by the SC which includes overcoming the mass transfer resistance [13]. TDPs administer drugs via passive diffusion as illustrated in Figure 1a [52]. There are two mechanisms for the storage of drugs on TDPs. A drug is either stored in a reservoir or incorporated into the transdermal patch fabric, whereby it is transported across the skin via a concentration gradient [13]. Although these methods have shown to be applicable for a variety of drug formulations, they are still limited by the drug molecules that can permeate through the SC barrier. As mentioned earlier, MNs have been proposed as an alternative delivery method to TDPs and hypodermic syringes as they have shown to overcome the various short comings previously outlined. Indeed, it has been illustrated by Benson and Namjoshi [4] and many other researchers [52-54] that MN research is a promising field of research to be pursued more extensively as it can be used to overcome the skin's natural defensive barrier, the SC in both adults and children. The rate of release of the drug depends upon the controlling membrane of the TDP [55] and therefore ensuring a consistent drug release profile can be uncertain.

MNs have been made possible to make due to the technological advances that have occurred in the last 20 years. Since the independent invention of the hypodermic needle in the mid 1850's [56] by Wood and Pravaz, hypodermic needles [57], or syringes, have been the most common form to administer biotherapeutics [10]. MN use has the advantage of simple patient administration of drugs with minimal invasiveness to the patients [58, 59]. The MNs would only permeate the SC and not the nerve receptors, consequently, the patients would feel little or no pain [32], though longer microneedles can also be used where deeper delivery of the drug is desired. Therefore, the development of MNs is important as it has the potential to overcome numerous disadvantages posed by the traditional DD systems [60].

Current applications of MNs include the delivery of macromolecules such as vaccines, proteins and peptides including insulin for diabetics. One such example for vaccines delivery is reported by Edmonston-Zagreb for measles vaccination [21, 61-68]. Vaccinations have the capacity to be delivered using MN patches which require the patient to have no specialised training or the need for cold/refrigerated storage; this in turn can reduce the spread of diseases [63, 69]. Therefore, the successful delivery of patch biotheraputics is a desirable drug administration method. Furthermore, there are initial challenges to overcome, such as the low permeability of skin which can limit the permeation of drugs. With MN, this challenge can be overcome.

1.3 Skin

We discuss skin and its properties briefly in this section because the skin barrier needs to be understood and overcome before MNs can be designed and applied successfully. Human skin is the largest organ in the human anatomy with an approximate surface area of 20,000 cm^2 on an average human male, which spans the entire external surface of the body [70], in which its main function is to contain all the internal organs, protect them against foreign organisms and bacteria and act as a passive barrier. Some of the skin's other functions include the regulation of internal body temperature, insulation, resistance against foreign bacteria and to provide protection to the internal organs [71]. The surface area of the skin constantly changes depending on age, weight loss and gain, [72, 73] height and sex of a human being [74]. It is also a receptive organ to thermal heat and pain. The skin comprises of several layers of which the main layers consist of the epidermis, dermis and subcutaneous tissue [71], see Figure 1. The figure illustrates the SC, a non-living layer, which provides the first barrier to foreign bodies and DD. Below the SC layer is the epidermis containing living tissue with no blood vessels. Below this is where drugs are taken up by capillaries in the dermis layer [52]. As evident in the figure, there are several pathways in which molecules can permeate through the skin which can be categorised into two routes, the appendageal and transepidermal routes. The appendageal route includes the movement through hair follicles and sweat glands (Figure 1b) and offers a high permeability to ions and large polar molecules [22]. The surface area is considerably small, and therefore, the exploration of passing drugs using this form of route is considered of little importance. The transepidermal route is a direct pathway through the SC (Figure 1c). This can occur via an intracellular (Figure 1a) or transcellular route (Figure 1c) [22].

With the varying skin surface areas there is also a dramatic difference in the skin thickness within the human body which is important in the absorption of therapeutics [37]. Therefore, analysis into skin thickness is important, as outlined in a study of skin thickness of Korean males and females conducted by Lee and Hwang [74]. In the study they found that the thickness of skin is vastly variable due to a number of factors. These include, the race, sex, and age of a person and also on different areas of the body. It has also been shown that aging and diabetes have an effect on skin thickness [72]. In particular it was found that subcutaneous fat thickness in different body regions was thinnner in aging skin and in diabetic patients. A reduction in the skin thickness of the hand was also observed for diabetic patients. Therefore, consideration of skin thickness would need to be examined when choosing an active site for TDD in general [37] and MNs in specific.

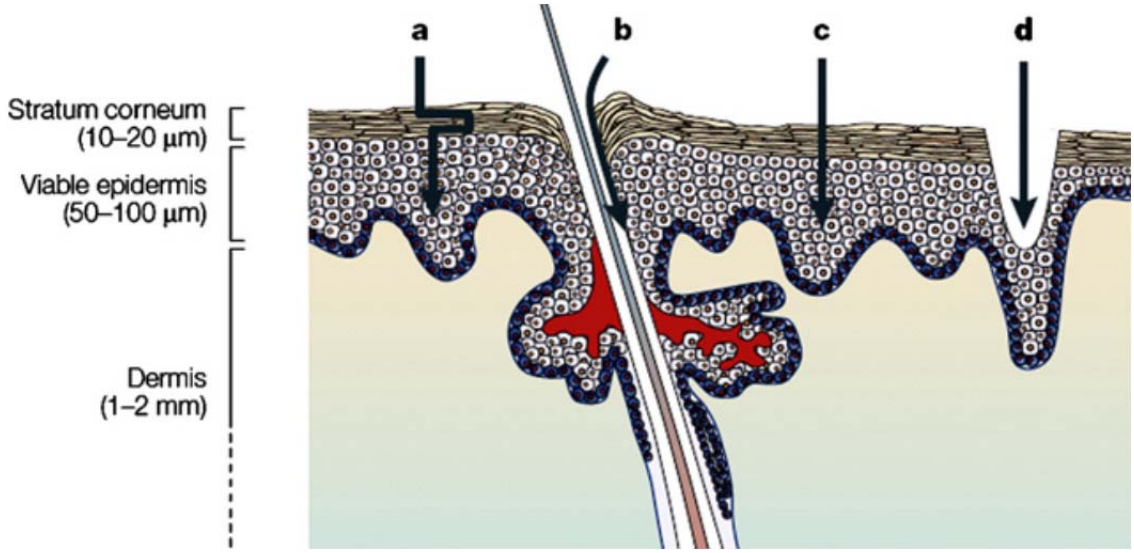


Figure 1 Cross section through human skin a: intracellular, b: hair follicles and sweat glands, c: direct pathway through the SC and, d: depicts the micron sized holes that can be created by MNs upon the skin (modified from [52])

1.3.1 Mechanical properties of skin

Understanding the mechanical properties of skin and detailed knowledge on various skin layers [76] are important as the insertion of needles into the skin would alter the skins mechanical response to all skin layers [77]. Knowledge of the properties of skin would help better understand the effect MNs have and therefore help determine what needs to be overcome when designing or inserting a MN. Skin is a viscoelastic material in which research has been conducted to ascertain what factors can influence skin properties. These include the site at which a material can be located, the age of the patient, the thickness of the skin, orientation, etc. [76, 78-80]. It is a complex organ that continuously changes as we age [79]. A property of skin that has been studied is the skin's ability to fold upon itself, such as wrinkling [81]. Smalls *et al*. [76] investigated the body's biomechanical skin properties which showed a significant difference in elastisity, stiffness and laxity for the right side in comparison to the left side of the body. This was possibly the result of 90% of subjects being right handed, which illustrated that an increase in muscle tone can also have an effect on the biomechanical skin properties. Cua *et al.* [79]. concluded that the biomechanical properties of skin decreased with increasing age, which may be due to natural degeneration as we get older. SC thickness on human forearm, palm, cheek and lower leg were also studied to determine by using two non-invasive measuring techneques namely, confocal Raman spectroscopy and confocal laser scanning microscopy, to measure SC thickness which were then compared to the thickness in literature data. It was found that it was possible to accurately measure the SC thickness with both techniques [82].

1.3.2 Increasing Permeability of Skin

The use of MN as a drug delivery system is an important development as the potential to allow a wider scope of molecules to be transdermally delivered through the skin is greatly increased

[83-86]. The amount of drug that is delivered can also be increased. However, an understanding of the permeability of skin would need to be established in order to determine how to increase drug content [69, 87, 88]. It is important to look at skin thickness when investigating increasing permeability of skin, as increasing skin permeability is important for transdermal drug delivery (TDD). The invention of the MN can overcome this factor as the needles bypass the SC layer, which is the rate dependent layer and can allow large molecular weight proteins to pass into the blood stream [89]. A well-known method to quantify the drug release through skin is the use of Franz diffusion cells, and therefore have been used in literature extensively to calculate the permeability of skin [32, 90, 91].

There have been multiple papers outlining various methods conducted to analyse different techniques to increase the permeability of skin. They can be categorised into chemical and physical enhancing techniques, some of which include, thermal ablation, sonophoresis and electroporation [3, 5, 19, 20, 30, 92]. Sinha and Kaur [19] stated that an individual enhancement technique cannot possess all the desired properties to facilitate the transport of drugs transdermally. However, the data published by Prausnitz *et al*. [33] illustrate that the use of MNs is a promising technique as it possessed many of the required properties for the delivery of drug therapeutics. Figure 1d depicts the micron sized holes that can be created by MNs upon the skin.

There are several physical methods that have been used in conjunction with MN technology to increase skin permeation [93-95]. One such example is the use of sonophoresis with MNs. It is a technique which allows molecules to permeate through the barrier of the skin more readily as ultrasonic waves create micro-vibrations on the skin [92]. Monomeric insulin analogues were studied to investigate the rate of iontophoresis transport on mice skin [25]. The study showed clinically relevant results for insulin regulation.

Several review papers have been published on different types of enhancement techniques [15, 22, 96-103] which outline various uses of chemical enhancers like N-methyl-2-pyrrolidone, a pyrrolidone [19] used in the application of insulin, ibuprofen and flurbiprofen. The applications of these techniques for increasing skin permeability would be useful when delivering drugs transdermally. Table 1 outlines some examples of enhanced protein/peptide delivery systems across the skin and the outcome of the experiments.

2. Trends in Microneedle (MN) Drug Delivery Method

MNs can be considered to be a micron scale hybrid between TDPs and hypodermic syringes, to overcome limitations that are associated with the individual application. They are small arrays of needles that are generally less than 1 mm in length [104-107]. There are multiple MN designs that have been created over the last decade. They can be categorised into two types,

solid or hollow MNs [108]. The materials that have been used to fabricate them range from metal [109], glass, silicon [110] and biodegradable polymers (polydimethylsiloxane) [111, 112] and silk fibroin [113]. More recently the use of fish scales have also been investigated [114]. Ideally the materials used would be pharmacologically inert, non-toxic, compatible with pharmaceutical ingredients, etc. [19]. Metals traditionally used for MN fabrication consist of stainless steel, nickel coated in gold, titanium, platinum, palladium [47, 115, 116]. Although numerous journal papers have been published on the use of silicon as a primary substituent of MN formulation, the material itself has yet to be FDA approved [90].

MNs provide a direct pathway for drugs to access the viable dermis, allowing for a painless DD that by-passes the SC [4]. They also differ in shape, ranging from square, circular, flat tipped, sharp tipped etc., [32, 90, 116-121]. There has been a lot of research conducted on MNs for the delivery and monitoring of various drugs such as glucose control for diabetics [61, 46, 122-125], Alzheimer's disease [110], anti-cancer [126] and other conditions [127]. Vaccines have also been a prominent research field with numerous studies developed to allow dose sparing effects [63, 128, 129] . There have been multiple studies conducted to optimise the delivery of drugs using MNs with numerous methods to fabricate them.

Table 1 Examples of enhanced protein/peptide delivery to and across the skin

Protein/Peptide	Delivery Method	Main Outcome	Reference
Bovine serum albumin	Polymeric microneedles	All drug released in 6 hours for in vitro permeation studies	[130]
Bovine serum albumin	Microneedles, ultrasound	BSA permeability of 1 μm/s is achieved with a 1.5 mm height microneedle and 15 W ultrasound output	[20]
Bovine insulin and bovine serum albumin	Microneedles coupled with iontophoresis	MN design containing 361 MNs/cm^2 of drug permeation in 6 hours on neonatal porcine skin in vitro	[6]
Botulinum toxin A	Stainless steel microneedles	Efficient diffusion through the dermis	[118]
Hepatitis B Surface Antigen	Elastic liposomes	Systemic and mucosal antibody response elicited in mice	[4]
Hepatitis B Virus DNA Vaccine	Jet propulsion (Powderject)	Application to healthy human volunteers resulted in both humoral and cell-mediated immune responses	[4]
Insulin	Insulin loaded dissolvable microneedle made of starch and gelatin	Dissolved in 5 minutes in rats to relative pharmacological availability and relative bioavailability of 92%	[21]
Influenza subunit vaccine	Microneedles Delivery	Application to intramuscular injection in guinea pigs. Hemagglutinin concentrations as high as 20.	[117]

		mg/ml	
Measles	Microneedles Delivery	Measles was able to be coated and dried onto MN. Vaccine delivery into rats was comparable to using hypodermic needles which gave similar antibody tilters	[63]

Park *et al*. [90] used biodegradable polymer MNs with sharp tips, to overcome problems associated with safety and disposal. They found that the use of biodegradable MNs increased skin permeability by three fold which increase the delivery of drugs transdermally. Gupta et al. [119] used hollow MNs for bolus deilvery of lispro insulin in comparison to catheter infusion. They found that MNs inserted 1 mm into human skin showed rapid insulin absortption with no pain observed from the volunteers in comparison to catheter infusion [131].

A method to fabricate MNs is important, as desirable attributes would be to have a simplistic fabrication method that is low cost and reproducible. This would be advantageous when mass producing MNs for industrial application [132]. However, the delivery of drugs using MNs can be conducted in four different ways. These can be known as “poke and patch”, “coat and poke”, formulating the API (active pharmaceutical ingredient) with a biodegradable polymeric MN [133, 134] or by channelling the drug through the channels of a hollow MN [89]. Multiple MN designs have been fabricated that facilitate the piercing of the SC allowing the permeation of drugs [110].

The rate of the development in MN research has increased significantly since the first papers have been published. A time series analysis forecast based on ARIMA model has been applied to the 1228 papers that have been found using the database Scopus with the search term “microneedle”. ARIMA is used to extrapolate future trends in data based on previous data sets. The forecast is shown in Figure 2. Trends will also be determined for specific microneedle types in the forthcoming sections in the paper.

A “single series” of ARIMA forecast is based only on the past values of the variables being forecasted, the variables in this case are the number of journal publications (NOJP). It tells us how an observation on the NOJP is statistically related to past observation on the same variable. All forecasts are extrapolations and therefore produce projections based on past patterns or relationships into the future. The model is used for short term forecasting in this paper which relies upon the data collected from recent pasts as opposed to distant past.

Figure 2 represents the time series analysis in graphical form. The observed and fitted data show a good match which give the confidence that the ARIMA model is reliable in predicting the trend. The trend shows that there are some fluctuations in the number of publications in various years. However, overall, there is a considerable increase in the number of MN publications in

the past 10 years. The ARIMA fit shows a good prediction of the trend (beyond 2015) when the total number of future MN publications are forecasted. The figure shows the yearly observations of the number of publications published since 1982-2013 (1228 total publications) and forecasted to 2016. The predicted NOJP for 2016 are shown to be approximately 300 which is a significant increase from the 218 papers found in Scopus for year 2013.

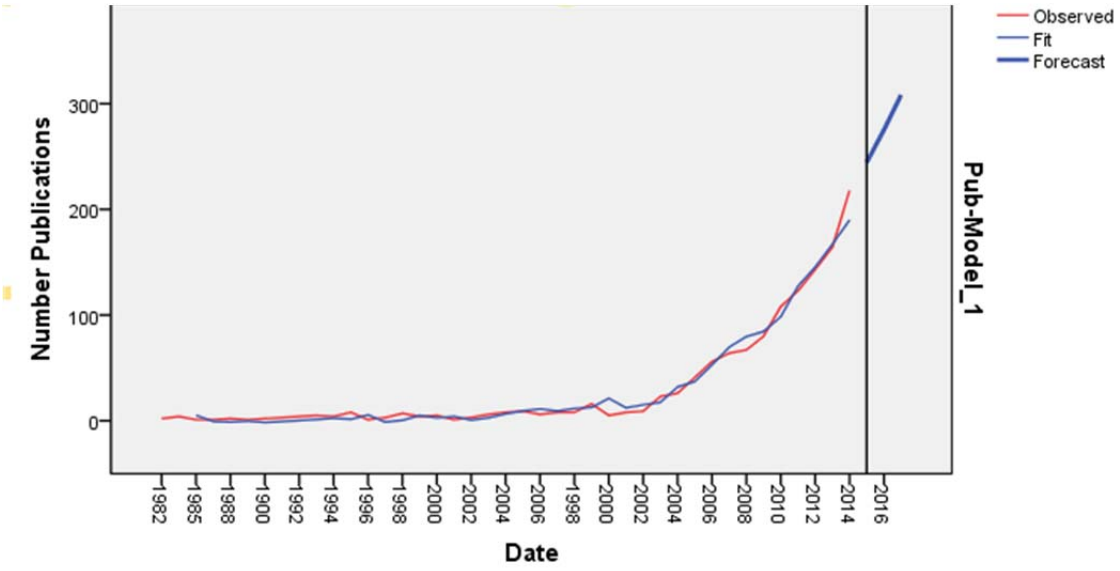


Figure 2. Forecasted, fitted and observed results on the trend of publications on microneedles using the keyword "microneedle"

2.1 Hollow MNs

Hollow MNs are traditionally used to allow liquid formulations through the SC and act like micron scale syringes. They have an added advantage as they can permit the administration of a larger drug dose compared to solid MNs [4]. They have also been shown to allow an increase in drug infusion rate due the fact that pressure can be applied across the length of the MN with administration. Ahmad *et al*. [135] have shown the in-situ assembly of hollow liquid filled polymeric microneedles for drug delivery Hollow metal MN arrays have been fabricated to allow the continuous administration of drugs. Micromachining methods have been used to make machine moulds from polyethylene terephthalate using UV lasers [77, 136, 137]. However, hollow MNs are still considered to be mechanically weaker involving complicated manufacturing processes and more complex to use than solid MNs as solid MNs are considered to be more robust [1]. There have been other advancements in recent research to allow manufacture of any height, pitch and lumen-lumen spacing of the MNs [116, 138]. Griss and Stemme [137] fabricated out-of-plane hollow MNs to overcome the short comings of blockages caused by hollow MNs. The design consists of an opening on the shaft of the MN as opposed to the tip for the delivery of drugs using a microfluidic liquid transfer. These were made by a deep reactive-ion etching (DRIE) method.

Roxhed *et al*. [77] developed a method of fabricating MNs that combines a controlled flow of drugs using out of plane MNs. Nordquist *et al*. [61] conducted a study using hollow MNs fabricated from metal which improved the Griss and Stemme design. MNs of length 400 µm and pitch 500 µm were produced.

ARIMA model has been applied to the 167 journal papers that have been found in the journal data base Scopus with the search term “hollow microneedle” (Figure 3). It is obvious that the fitted and the observed data do not match well which is due to the small number of publications per year. This suggests that while the total numbers of publications relating to hollow microneedles are increasing, the trend (e.g., the number of publications in different years) cannot be reliably predicted. Nevertheless, the ARIMA model has been applied to predict the future trend in the journal papers. It shows that the number of publications on hollow MNs would decline, which seems to support a hypothesis that research groups are potentially leading away from hollow MNs use due to the complications in manufacture and their brittle nature.

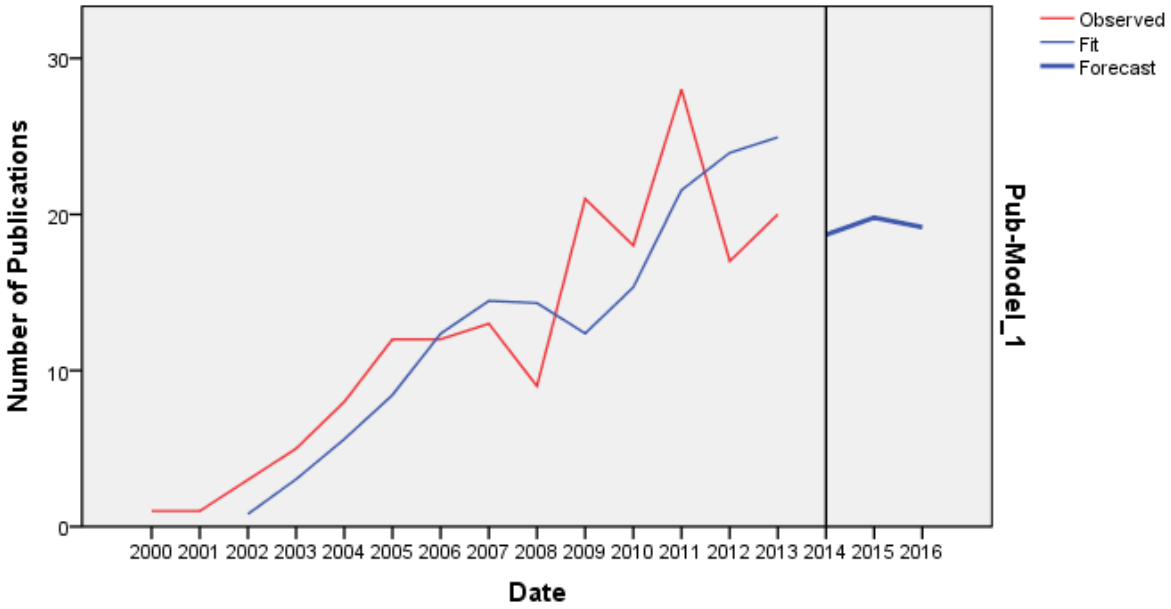


Figure 3 Forecasted, fitted and observed results on the trend of publications on hollow microneedle using the keyword "hollow microneedle"

2.2 Solid MNs

Solid MNs are more robust than hollow MNs and have a stronger mechanical strength [1]. MNs can produce micro pores in skin, which bypasses the SC layer, and allow drugs to permeate to the viable epidermis [110]. There have been a number of methods and materials used to fabricate solid MNs [139] [140], some of which are represented in Table 2.

The first reported case of solid MNs in literature was in the study of gene therapy [141]. However, Henry *et al*. [32] was the first to demonstrate the feasibility of delivering drugs

transdermally. They designed conical shaped MNs, 150 μm in length and <1 μm tip diameter (Figure 4A). This allowed easy piercing of the MN into skin and produced a 4 order of magnitude increase in skin permeability. In this study, a deep reactive ion etching microfabrication technique was used. Although the fabrication method yielded reproducible fabrication of MNs, the insertion of MNs into human cadaver skin left a small proportion of the needles bent at the tip 5-10 μm. Khan *et al.* [140] prepared coated MNs utilising a process named 'electrohydrodynamic atomisation (EHDA)' to produce pharmaceutical coatings on a single MN patch for the purpose of applying personalised medicine. Martanto *et al*. [142] fabricated solid metal MNs by cutting metal sheets with an infrared laser. The MNs were then manually bent into the 90° angle. The MNs were in an array containing 105 MNs, 1000 μm in length and a cross section at base of 50 μm x 200 μm. The MN was tapered to a sharp tip (angle 20°) (Illustrated in Figure 4B). This method of fabrication was shown to be laborious and used a number of strong acids which could pose problems when disposing of such chemicals. The application of this MN into hairless rats also required an external high velocity applicator to successfully insert the MN into the skin. Although the MNs used to elucidate the transdermal delivery of insulin produced successful data in reducing glucose levels by up to 80%, the fabrication method would not be considered a simple process.

The fabrication of biodegradable and biocompatible polymers was proposed by Park *et al.* [90] to address the issues associated with cost effective fabrication materials and problems associated with MN fabrication materials, such as metal, a sharp hazardous waste, and silicon which has yet to be FDA approved [90]. The material was considered to be a safer alternative as it is mechanically strong and relativly inexpensive.

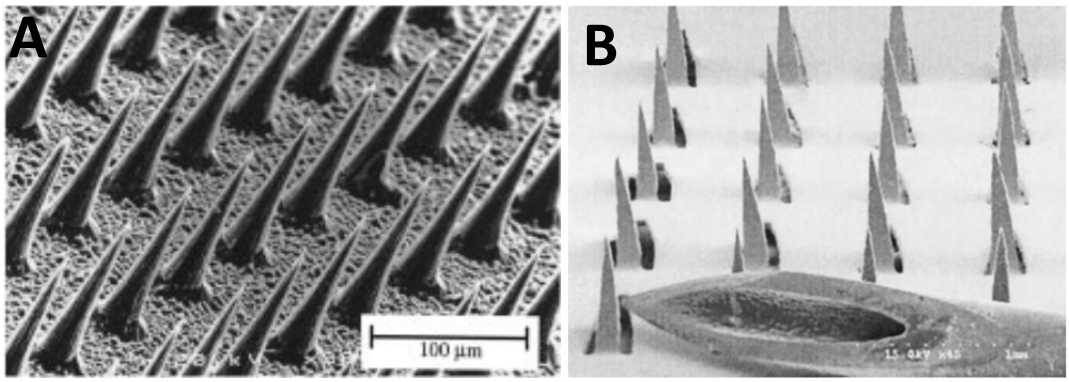


Figure 4 SEM images of various microneedles: (A) solid conical shaped microneedle [32], (B) solid microneedle next to a 27-gauge syringe [47]

As shown in Table 2, silicon has been widely researched when developing MNs. Polydimethylsiloxane was used by Chu and Prausnitz [111] to show the material combined the mechanical strength that metal MNs can provide and the useful properties of silicon drug based arrow head (Figure 6). They showed that the production of a blunt metal shaft with a detachable

drug encapsulated arrow head on the end could provide drug to the viable epidermis within seconds to porcine and human cadaver skin.

There have been several types of polymer MNs that have been created to overcome the non-biocompatible and non-biodegradable properties of metal and silicon MNs [90, 143-145]. The biodegradable MNs were fabricated using a lithographic approach. Dissolving MNs were fabricated by Ito *et al.* [46] in which insulin was mixed at room temperature to dextrin and deionised water and dried in a desiccator after thread was attached. Three different mixtures of insulin were made and tested on rats which illustrated the uses of dissolving MNs to successfully deliver the drug precutaneously. Table 2 illustrates a general overview of the transdermal MN methods used to administer drugs and vaccines. For example, the table shows that dissolving insulin based MNs were made with starch and gelatin [21] for a 5 minute dissolution time when inserted into the skin. It was shown that 600 μm height and 300 μm base MN retained pharmalogical activity in the starch/gelatin matrix.

ARIMA model has been applied to the 112 journal papers that have been found using the journal data base Scopus with the search term “solid microneedle”. Although the trend has declined in the observed data for 2013-2014, the results show that there would be an increase in the amount of solid MN papers in the future.

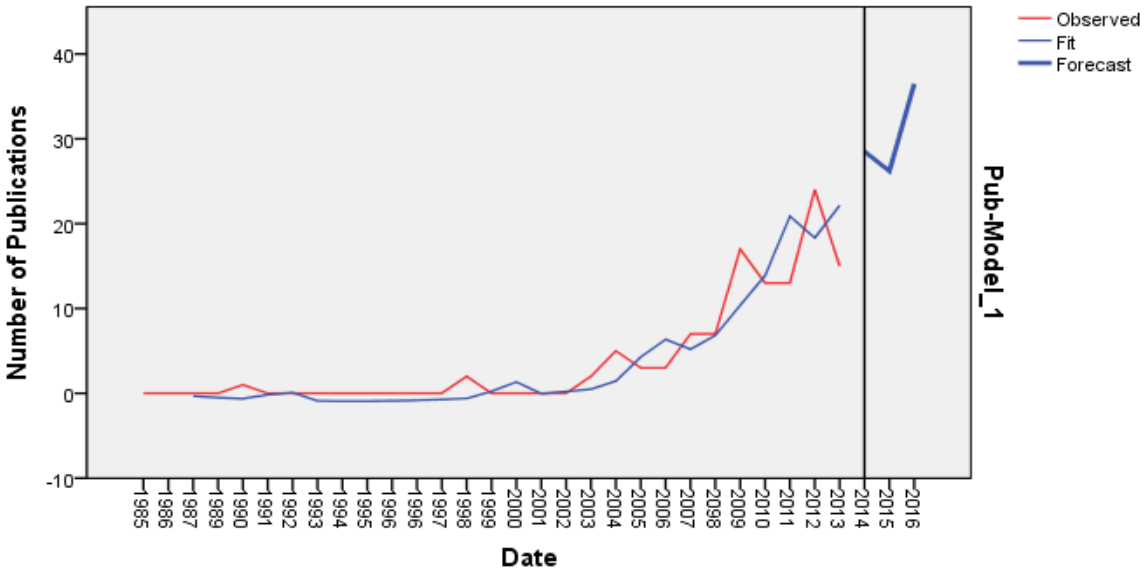
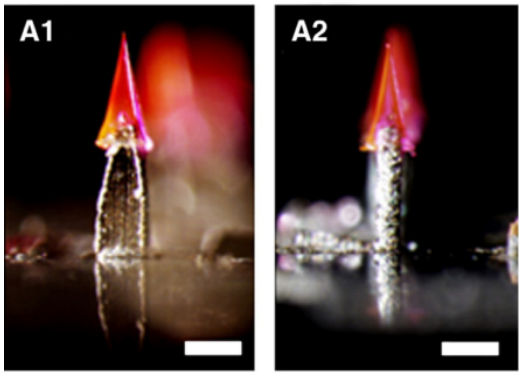


Figure 5 Forecasted, fitted and observed result on the trend of publications on solid microneedle using the keyword "solid microneedles"



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387 Figure 6 Images of an arrow head microneedle [111]

388 **Table 2 Methods of fabricating solid MNs**

Methods	Materials	Dimensions (μm)	Advantages	Disadvantages	Reference
Separable dissolving Arrow Heads	• (polydimethylsiloxane (PDMS) Sylgard 184,), • Metal shaft • Water soluble excipients-PVP, sucrose • PLGA	600 μm-long PVP/PVA arrowhead capped onto a metal shaft with an exposed length of 600 μm and a 100 μm overlap.	• Rapid (1-5 seconds) /painless drug/ vaccines delivery, • PDMS: Inert/Non-toxic/ Non flammable Rapid (1-5 seconds) /painless drug/ vaccines delivery, convenient/safe/potential self-administration, can allow controlled release of active ingredient • Advantage over coated needles-elimination of bio-hazardous waste, allow self-administration • Does not require extended patch wearing even for long duration of drug releases. • PDMS: Inert • PDMS: Non-toxic • PDMS: Non flammable	• Difficult insertion into skin as requires wider needle geometry (non-blunt shaft) • Non reusable • fibrotic reaction	[111, 146]
Dissolving	• Mixture Insulin, water, dextrin	basal diameter: 3.24±0.16 and 0.55±0.03 mm	• Bio-compatible	• Produce bio hazardous sharp waste • Large tipped	[46]
Pyramidal dissolving Polymer	• polyvinyl alcohol (PVA), Polyvinylpyrrolidone (PVP) • A = Pyramidal MN • B= Extended pyramidal MNs • C = Pedestal MNs	base width x base depth x needle height: A: 300 μm x 300 μm x 600 μm. B: 300 μm x300 μm x 900 μm. C: 340 μm x 340 μm x 900 μm) 10x10 array	• Ability to increase drug capacity and localise to the MN tip • Allowed a deeper insertion • Higher increase of drug to be dissolved	• Produce bio hazardous sharp waste	[147]
Deep reactive Ion etching	• Chromium • Silicon wafers	120 μm length, <1 μm tip diameter	• Mechanically strong	• Reusability questionably • Top 5- 10 μm of MN damaged for a few samples	[32]
Dissolving. Fabricated by a	• Maltose	1200 μm length 60 μm tip diameter	• Requires no moulds, therefore no sharp waste	• Complicated process	[148]

drawing technique to create a sharp tip					
Cutting metal using infrared laser, manually bending the MN structure, electropolished	• Metal	1000 µm length, 50 µm x200 µm cross section at base, tapered to a sharp tip (angle 20°), 106 array	• A short MN insertion time was better than a longer insertion time to facilitate drug permeation	• Insertion of large array was difficult by hand without external device • Strong acids used	[47]
Silicon master mould to make PDMS inverse mould	• Sugar glass MN: • Trehalose/mannitol (50:50 w/w) • Trehalose dehydrate/sucrose (75:25w/w) • Trehalose/sucrose (75:25 w/w) • Trehalose/sucrose (50:50 w/w) • 2% (w.w) methylene blue	200 µm base 20 µm tip 300 µm height	• Fast dissolution of drug into skin • Use of simple sugars to create biodegradable MNs	• MN patch needles to be left on the skin for potentially over 20 minutes	[149]
Stainless steel MN produced by chemical etching	• Stainless steel	750 µm height 200 µm x 50 µm at the base. Single row of 5 MNs	• Dose of Measles vaccine is small, therefore the dimensions of the MN are sufficient to allow coating and delivery of the vaccine • Needles long enough for rat dorsal thickness (700 µm-1000 µm) penetration • Cost effective manufacturing	• Produce bio hazardous sharp waste	[63]
Dissolving MN	• Starch/gelatin (1:1 ratio) • PDMS mould	600 µm height 300 µm base 5 µm tip	• Rapid dissolution of 5 minutes was achieved • Pharmalogical activity retain in the starch/gelatin	• Time to reach minimum plasma glucose level slightly longer for MN than for subcutaneous injection of insulin	[21]

It has been reported in the literature that breakage of MNs is considered to be minimal as long as the insertion of the MNs is gentle. It was also stated that metal MNs are more robust than other materials used to manufacture MNs and that biodegradable needles are safer [4].

Although the use of silicon as a primary material in some MN fabrication methods is favoured, it is hindered by the fact that the material is currently not approved by the Food and Drug Administration (FDA) [90]. Research has been conducted on the use of large sharp MNs but little has been carried out on the effect of short blunt MNs on increasing the permeability of skin. What little research has been conducted shows increasing permeability to longer sharper MNs. This shows that there is a potential for developing a new MN which is blunt and short in length.

Solid MNs have been shown in the literature to have a typical length of 150 μm-350 μm. However, studies into the fabrication of super short MNs with a length of 70-80 μm were conducted by Wei-Ze *et al*. who have shown that super-short MNs are capable of successfully delivering galanthamine (GAL), a drug used for Alzheimer's. The study compared the use of sharp super-short MNs against blunt super-short MNs and longer sharp needles of 1500 μm as shown in Figure 7. The super short MNs were made using wet etching of silicon using acupuncture needles backed onto the minipore of a basement. The study found that as more pressure was applied to insert the MNs, the permeation of the drug increased [110]. There is currently little literature regarding the feasibility of blunt short MNs against sharp long MNs. Based on the findings of Wei-Ze *et al*. there could potentially be a gap in the literature to pursue further the implications of using the is method of DD on various drugs and see the effect the MN has on skin permeability. This shows a promising fabrication method for the delivery of drugs transdermally. Table 3 shows the main parameters of super short MNs in the study conducted by [110]. However, considering the various fabrication methods, there seems to be a need for a simpler robust technique that requires minimal cost, as the process is economically viable and relatively robust.

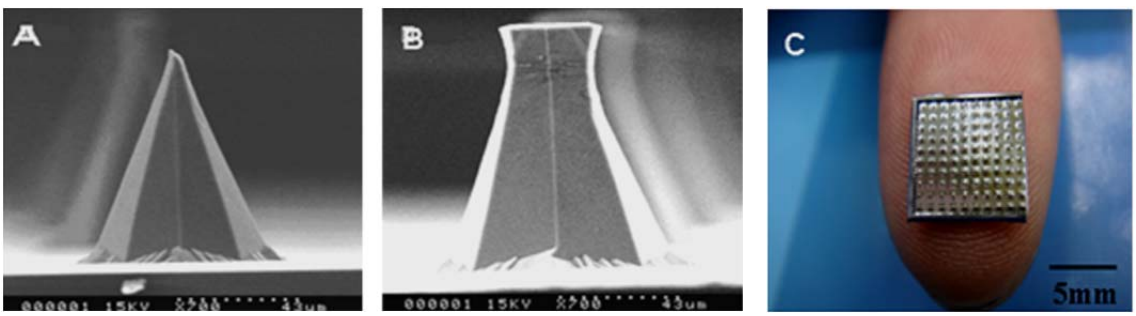

Figure 7 Super-short microneedles used in this study, SEM images of A: a single sharp tipped super-short microneedle, B: a single flat tipped super-short microneedle and C: an angled view of a flat tipped super-short microneedle array [110]

ARIMA model has been applied to the 16 journal papers that have been published in the journal data base Scopus with the search term “dissolvable microneedle”.

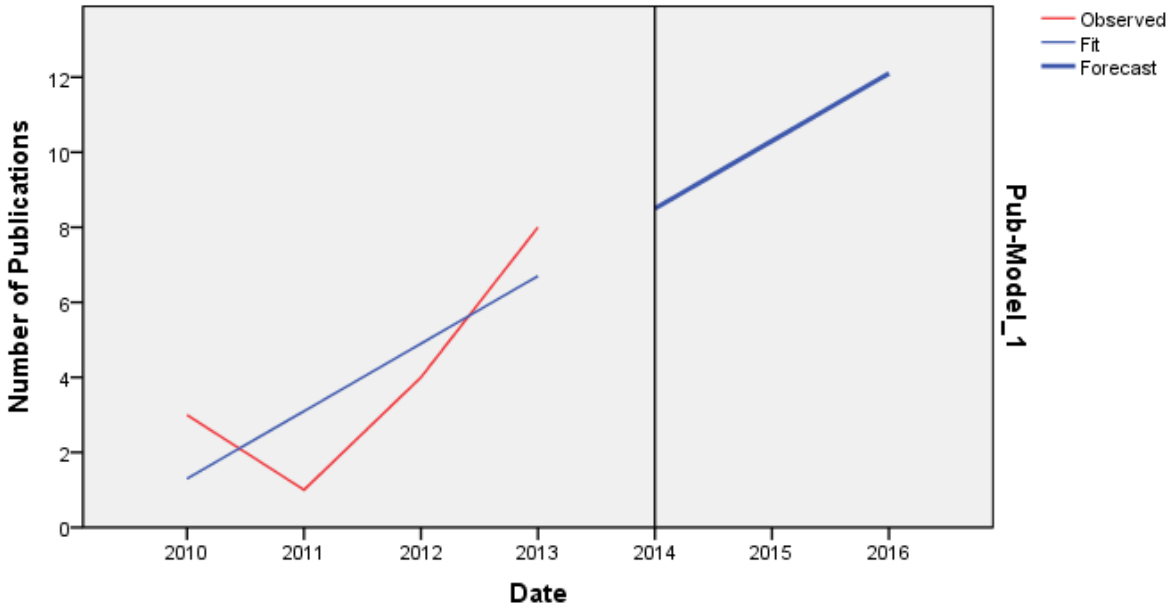

Figure 8 Forecasted, fitted and observed result on the trend of publications on dissolvable microneedle using the keywords "dissolvable microneedle"

There is little data on the publication of dissolvable MNs. However there is a growing trend in the use of dissolvable MNs, which is apparent due to the benefits of incorporating active drug directly to the manufacture of MNs. Although the ARIMA forecast in Figure 8 seems to fit this trend due to the lack of observations the forecast would not be a useful estimate of the mean as there would essentially only be one observation per mean. Therefore this forecast would not be an ideal assumption of the predicted NOJP for 2016.

430 Table 3 Parameters of super short MNs [110]

Method	**Materials**	**Dimensions (μm)**	**Advantages**	**Disadvantages**
Sharp tipped Super Short microneedles	Microneedle: Silicon with 30% potassium hydroxide Patch: • Backing: polyethylene • Adhesive: polyisobutylene	75	• Strong material which does not break with insertion forces exceeding 8 N • Reusable	• Permeability of drug is lower compared to flat tipped microneedle • Skin folding shown upon insertion of needle compared to flat tipped microneedle
Flat tipped Super Short microneedles	Microneedle: Silicon with 30% potassium hydroxide Patch: • Backing: polyethylene • Adhesive: polyisobutylene	80	• Increase permeability in skin compared to sharp tipped microneedle • Decreases skin barrier function • No skin folding shown upon insertion of needle compared to sharp tipped microneedle • Strong material which does not break with insertion forces exceeding 8 N • Reusable	N/A

3. MNs Patents

MNs have become more prominent within the last decade. The use of MNs as a method for the transdermal delivery of drugs has become a more appealing technique as it overcomes many disadvantages such as discomfort and pain that can be caused by hypodermic needles or the non-bioavailability that oral dosage forms provide. Therefore it is important to look at patents that have been filed within the last decade to correlate the trends of MNs [150-152]. Table 4 shows a summary of patents that have been taken out on materials to fabricate MNs. Table 5 illustrates some example patents on the methods of MN fabrication. Table 6 shows patents on chemicals used for MN transdermal delivery system. Figure 9 illustrates the number of patents taken for various MN designs. It can be seen that there is a considerable amount of MN patens taken out on hollow MNs. This may be due to the fact that hollow MNs have the advantage of delivering a higher dosage in comparison to other methods [108].

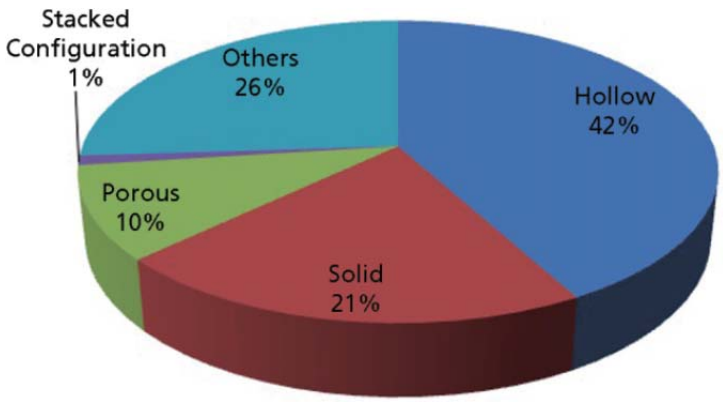

Figure 9 Division of patents filed based on type of MNs [108]

Transdermal delivery of drugs is a continually growing field with an abundant of journal papers being published each year on MN technology and an increasing number of patents filed. Therefore an increase in development for more commercially viable MN products would need to be conducted.

451 **Table 4 Example patents on microneedles technology**

Patent Number	Date of Filing	Applicant	Key Invention	Country	Reference
US 2010042050	16th April 2007	Nemaura Pharma Ltd, USA	Applicator for microneedles	USA, European, Japan, China, India	[153]
WO 2011016230	4th Aug 2010	Medrx Co., Ltd., Japan	Provided is a microneedle device which protects microneedles, has an easily portable shape, is free from such problems as breakage of fine needles in the step of puncturing the skin with the microneedles, and ensures appropriate skin puncture to administer a drug.	Japan	[154]
WO 2011084951, US 20110172645	4th Jan 2011, 8th Jan 2010	Ratio, Inc., USA	Microneedle configured to facilitate delivery of the drug to the subject. The microneedle includes a tip portion and is moveable from an inactive position to an activated position	USA	[155]
WO 2011014514	27th Jul 2010	3M Innovative Properties Company, USA	The present disclosure relates to apparatus, assemblies, combinations, and methods for infusing fluids by hollow microneedles.	USA	[156]
CA 2696810 JP 2011078618 WO 2011043086 AU 2010201434 KR 2011067009 EP 2343102 A1	15th Jan 2010	Bioserentach Co. Ltd., Japan	Microneedle sheets are produced by injecting a needle raw material into a stamper formed with a concavity in a base material.	Japan	[157]
WO 2013096026	12th Dec 2012	3M Innovative Properties Company, USA	Assembly of a microneedle adhesive patch. The assembly can include a backing, and an adhesive and a matrix coupled to the backing	USA	[158]
WO 2013066262	2nd Nov 2011	Singapore	Invention relates to plastic microneedle strips that are used in TDD for increasing the DD rate through the skin	Spain	[159]

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453 **Table 5 Example patents on methods of microneedle fabrication**

Patent Number	Date of Filing	Applicant	Key Invention	Country	Reference
EP 2289843	31st Aug 2009	University College Cork-National University of Ireland, Cork, Ireland.	The invention relates to a method of fabricating a microneedle device of the type comprising an array microneedles on a flexible polymer support layer.	European	[160]
JP 2011083387	14th Oct 2009	Kyushu Institute of Technology, Japan; Nichiba n Co., Ltd.	Manufactured. by (1) performing Bosch process to a Si wafer to form a Si microneedle having a tapered tip and a columnar part with the same diameteror a decreasing diameter in the longitudinal direction, etching the tip with an etching solution and/or a reactive radical.	Japan	[161]
CN 102000020	17th Nov 2010	Beijing Pharmaceutical Research Institute, Henan Lingrui Pharmaceutical Co., Ltd., People Republic of China; Beijing Lingrui Hi-tech Co., Ltd.	The title polymer (molecular weight: . 1000-1000,000) is selected from poly(p-dioxanone) or p-dioxanone containing copolymer (containg p-dioxanone 10-100 wt.%), such as poly(p-dioxanone-lactide), poly(p-dioxanone-glycolide), etc.	China	[162]
CN 103263727	22nd May 2013	Tsinghua University, People Republic of China.	A metallic microneedle array, including : substrate; and a metal sheet fixed on the surface of the substrate.	China	[163]
US 20130030374 A1	11 Oct 2012	Toppan Printing Co., Ltd.	Microneedle including forming a plurality of first linear grooves on a substrate in parallel to one another along a first direction using grinding and forming a plurality of second linear grooves on the substrate in parallel to one another in a second direction intersecting the first direction using grinding.	USA	[164]
CN 103181887	30 Dec 2011	Shanghai No.7 People's Hospital, People Republic of China.	invention relates to triamcinolone acetonide biodegradable maltose microneedle array which contains (1) triamcinolone acetonide or its pharmaceutically acceptable salt and (2) maltose or its hydrate, wherein the length of microneedle is 800-1500 μm, the diam. of the microneedle is 100-300 μm and array d. is 9-100 needles/cm^2.	China	[165]

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456 **Table 6 Patents on chemicals used for microneedle transdermal delivery system**

Patent Number	Date of Filing	Applicant	Key Invention	Country	Reference
GB2472778A	17th Aug 2009	PANGAEA LAB LTD	Microneedle Roller.	Great Britain	[166]
WO 2011026144 or S 20110052694	31st Aug 2009	AllTranz Inc., USA	DD system for pharma active ingredients (e.g., cannabidiol and prodrugs of cannabidiol.	USA	[167]
US 20110118560, US 20110118656, US 20110117150, US 20110118652, US 20110118696, US 20110118653, US 20110118697, US 20110118698, US 20110118652 US 20110118656	13th Nov 2009, 19th Feb 2010, 9th Mar 2010	Searete LLC, USA	Provides one or more medicines to mammalian subject.	USA	[168]
CN 101991846	17th Nov 2010	Chifeng Boen Pharmaceutical Co., Ltd., People Republic of China	The invention relates to inactivated vaccine for immunoprophylaxis of bovine mastitis caused by Escherichia coli and its preparation method.	China	[169]
US 20130171722	3rd Jan 2012	City University of Hong Kong, Hong Kong	Injection of a substance into a subject including elongate non-hollow micro-nanoneedles for delivering bioactive substance including drug and gene molecules such as plasmid DNA, siRNA, miRNA, shRNA,	USA	[170]

4. Conclusion

Oral administration and hypodermic syringes are the most commonly used delivery methods in today's society. However, they pose several disadvantages such as painful side effects of using hypodermic syringes or the problem associated with oral administration such as drug bioequivalence [3]. TDPs pose numerous advantages as an alternative method as they provide controlled release of medicine to the patient in a minimally invasive manner. However, they cannot permeate large molecules to pass the SC (the top layer of skin), thereby limiting the medical application to patients. MNs have been proposed to overcome this limitation and provide the transdermal delivery of large molecular weight proteins such as shown by Benson and Namjoshi [4]. Various MN designs and fabrication methods have been explored in the literature ranging from the fabrication of, hollow, solid, desolvable, sharp and short MNs. MNs can be made from polymers, metals, glass and silk. There is a gap in the literature that has not been explored – the commercial viability of MNs – for an industrially viable product to be manufactured on an industrial scale. Various techniques discussed in this literature review have shown laborious techniques and involve the use of non FDA approved excipients such as silicon. There is a growing trend in the amount of publications surrounding MN technology. With the increase in the use of MN for commercial scale products, the upward trend in number of publications is unlikely to change in the next decade, due to continuous advances in technology.

The use of time series analysis allows the extrapolation of trends in data to predict the number of journal papers published. This can occur provided the number of data sets (observations) is sufficient to produce a good estimate. In the case of microneedles, solid and hollow, this was possible. However this was not sufficient for dissolvable microneedles.

5. Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

6. Funding

This work was partly supported by Loughborough University, UK and EPSRC, UK.

487 **7. References**

488

[1] J. J. Escobar-Chávez, D. Bonilla-Martínez, M. A. Villegas-González, E. Molina-Trinidad, N. Casas-Alancaster and A. L. Revilla-Vázquez, "Microneedles: a valuable physical enhancer to increase transdermal drug delivery," *Journal of clinical pharmacology,* vol. 51, no. 7, pp. 964-977, 2011.

[2] S. P. Davis, B. J. Landis, Z. H. Adams, M. G. Allen and M. R. Prausnitz, "Insertion of microneedles into skin: measurement and prediction of insertion force and needle fracture force," *Journal of Biomechanics,* vol. 37, no. 8, pp. 1155-1163, 2004.

[3] R. H. Guy, "Current Status and Future Prospects of Trandermal Drug Delivery," *Pharmaceautical Research,* vol. 13, no. 12, pp. 1765-1769, 1996.

[4] H. A. E. Benson and S. Namjoshi, "Proteins and Peptides: Strategies for Delivery to and Across the Skin," *Journal of Pharmaceutical Sciences,* vol. 97, no. 9, 2008.

[5] L. Daugimont, N. Baron, G. Vandermeulen, N. Pavselj, D. Miklavcic, M.-C. Jullien, G. Cabodevila, L. M. Mir and V. Pre´at, "Hollow Microneedle Arrays for Intradermal Drug Delivery and DNA Electroporation," *Journal of Membrane Biology,* vol. 236, p. 117–125, 2010.

[6] M. J. Garland, E. Caffarel–Salvador, K. Migalska, A. D. Woolfson and R. F. Donnelly, "Dissolving polymeric microneedle arrays for electrically assisted transdermal," *Journal of Controlled Release,* vol. 159, no. 1, p. 52–59, 2012.

[7] H. Kalluri and A. K. Banga, "Transdermal delivery of proteins," *American Association of Pharmaceutical Scientists: PharmSciTech,* vol. 12, no. 1, pp. 431-441, 2011.

[8] H. Katsumi, Y. S. Quan, F. Kamiyam, T. Sakane and A. Yamamoto, "Development of a novel transdermal delivery system of peptide and protein drugs using microneedle arrays," *Yakugaku Zasshi,* vol. 134, no. 1, pp. 63-67, 2014.

[9] S. Chandrasekhar, L. K. Iyer, J. P. Panchal, E. M. Topp, J. B. Cannon and V. V. Ranade, "Microarrays and microneedle arrays for delivery of peptides, proteins, vaccines and other applications," *Expert Opinion on Drug Delivery,* vol. 10, no. 8, pp. 1155-1170, 2013.

[10] S. P. Sullivan, N. Murthy and M. R. Prausnitz, "Minimally Invasive Protein Delivery with Rapidly Dissolving Polymer Microneedles," *Advanced Materials,* vol. 20, pp. 993-938, 2008.

[11] H. Li, Y. Yu, D. S. Faraji, B. Li, C.-Y. Lee and L. Kang, “Novel engineered systems for oral, mucosal and transdermal drug delivery,” *Journal of Drug Targeting,* vol. 21, no. 7, pp. 611-629, 2013.

[12] M. R. Prausnitz and R. Langer, “Transdermal drug delivery,” *Nature Biotechnology,* vol. 26, no. 11, p. 1261–1268, 2008.

[13] L. Margetts and R. Sawyer, “Transdermal drug delivery: principles and opioid therapy,” *Continuing Education in Anaesthesia, Critical Care and Pain,* vol. 7, no. 5, pp. 171-176, 2007.

[14] S. R. Behin, I. S. Punitha and F. Saju, “Development of matrix dispersion transdermal therapeutic system containing glipizide,” *Der Pharmacia Lettre,* vol. 5, no. 3, pp. 278-286, 2013.

[15] M. Nalesniak, k. Iwaniak, R. Kasperek and E. Poleszak, “A review of TTS-development, types and preparations,” *Current Issues in Pharmacy and Medical Sciences,* vol. 26, no. 1, pp. 88-93, 2013.

[16] S. Ravi, P. K. Sharma and M. Bansal, “A review: Transdermal drug delivery of nicotine,” *International Journal of Drug Development and Research,* vol. 3, no. 2, pp. 1-8, 2011.

[17] J. E. Rose, M. E. Jarvik and K. D. Rose, “Transdermal administration of nicotine,” *Drug and Alcohol Dependence,* vol. 13, no. 3, pp. 209-213, 1984.

[18] M. J. Garland, K. Migalska, T. Mazlelaa, T. Mahmood, T. R. R. Singh, D. A. Woolfson and R. F. Donnelly, “Microneedle arrays as medical devices for enhanced transdermal drug delivery,” *Expert Review of Medical Devices,* vol. 8, no. 4, pp. 459-482, 2011.

[19] V. R. Sinha and M. P. Kaur, “Permeation Enhancers for Transdermal Drug Delivery,” *Drug Development and Industrial Pharmacy,* vol. 26, no. 11, pp. 1131-1140, 2000.

[20] T. Han and D. B. Das, “Permeability Enhancement for Transdermal Delivery of Large Molecule Using Low-Frequency Sonophoresis Combined with Microneedles,” *Journal of Pharmaceutical Sciences,* pp. 1-9, 2013.

[21] M. H. Ling and M. C. Chen, “Dissolving polymer microneedle patches for rapid and efficient transdermal delivery of insulin to diabetic rats,” *Acta Biomaterialia,* 2013.

[22] H. A. Benson, “Transdermal drug delivery: penetration enhancement techniques,” *Current drug delivery,* vol. 2, no. 1, pp. 23-33, 2005.

[23] A. Yamashita, Y. Hirai and . K. Tojo, “Effect of Ultrasound on Rate of Drug Absorption through Skin,” *Journal of Chemical Engineering Japan,* vol. 29, no. 5, pp. 812-816, 1996.

[24] A. Nayak, H. Babla, T. Han and D. B. Das, “Lidocaine carboxymethylcellulose with gelatine co-polymer hydrogel delivery by combined microneedle and ultrasound,” *Drug Delivery,* no. DOI: 10.3109/10717544.2014.935985 (in press).

[25] L. Langkjær, J. Brange, G. M. Grodsky and R. H. Guy, “Iontophoresis of monomeric insulin analogues in vitro: effects of insulin charge and skin pretreatment,” *Journal of Controlled Release,* vol. 51, no. 1, pp. 47-56, 1998.

[26] Y. Yang, H. Kalluri and A. K. Banga, “Effects of Chemical and Physical Enhancement Techniques on Transdermal Delivery of Cyanocobalamin (Vitamin B12) In Vitro,” *Pharmaceutics,* vol. 3, no. 3, pp. 474-484, 2011.

[27] D. Zhang, D. B. Das and C. D. Rielly, “Potential of microneedle-assisted micro-particle delivery by gene guns: a review,” *Chemical Engineering Science,* no. DOI: 10.1016/j.ces.2014.06.031 (in press), 2014.

[28] D. Zhang, D. B. Das and C. D. Rielly, “Microneedle assisted micro-particle delivery by gene guns: Mathematical model formulation and experimental verification,” *Drug Delivery,* no. DOI: 10.3109/10717544.2013.864345 (in press), 2014.

[29] H. S. McAllister, D. V. Allen, M. G. Allen and M. R. Prausnitz, “Microfabricated Microneedles: A Novel Approach to Transdermal Drug Delivery,” *Journal of Pharmaceautical Sciences,* vol. 87, no. 8, pp. 922-925, 1998.

[30] S. Mitragotri, “Devices for overcoming biological barriers: The use of physical forces to disrupt the barriers,” *Advanced Drug Delivery Reviews,* vol. 65, no. 1, pp. 100-103, 2013.

[31] M. B. R. Pierre and F. C. Rossetti, “Microneedle-based drug delivery systems for transdermal route,” *Current Drug Targets,* vol. 15, no. 3, pp. 281-291, 2014.

[32] S. Henry, D. V. McAllister, M. G. Allen and M. R. Prausnitz, “Microfabricated Microneedles: A Novel Approach to Transdermal Drug Delivery,” *Journal of Pharmaceutical Sciences,* vol. 87, p. 922–925, 1998.

[33] M. R. Prausnitz, S. Mitragoti and R. Langer, “Current status and future potential of transdermal drug delivery,” *Nature Reviews Drug Discovery,* vol. 3, pp. 115-124, 2004.

[34] H. M. Wolff, “Future of transdermal drug delivery systems (TDDS),” *American*

Pharmaceutical Review, vol. 17, no. 4, 2014.

[35] K. B. Ita, "Transdermal drug delivery: Progress and challenges," *Journal of Drug Delivery Science and Technology,* vol. 24, no. 3, pp. 245-250, 2014.

[36] H. S. Nalwa, "A special issue on reviews in nanomedicine, drug delivery and vaccine development," *Journal of Biomedical Nanotechnology,* vol. 10, no. 9, pp. 1635-1640, 2014.

[37] S. Kamboj, V. Jhawat, V. Saini and S. Bala, "Recent advances in permeation enhancement techniques for Transdermal drug delivery systems: A review," *Current Drug Therapy,* vol. 8, no. 3, pp. 181-188, 2013.

[38] J. Zhang, Q. Zhu and L. Zhang, "Research advances in applications of microneedles in transdermal drug delivery," *Pharmaceutical Care and Research,* vol. 13, no. 5, pp. 326-331, 2013.

[39] M. Fazil, A. Ali, S. Baboota, J. K. Sahni and J. Ali, "Exploring drug delivery systems for treating osteoporosis," *Expert Opinion on Drug Delivery,* vol. 10, no. 8, pp. 1123-1136, 2013.

[40] T.-M. Tuan-Mahmood, M. C. McCrudden, B. M. Torrisi, E. McAlister, M. J. Garland, T. R. Singh and R. F. Donnelly, "Microneedles for intradermal and transdermal drug delivery," *European Journal of Pharmaceutical Sciences,* vol. 50, no. 5, pp. 623-637, 2013.

[41] F. Wu, S. Yang, W. Yuan and T. Jin, "Challenges and strategies in developing microneedle patches for transdermal delivery of protein and peptide therapeutics," *Current Pharmaceutical Biotechnology,* vol. 13, no. 7, pp. 1292-1298, 2012.

[42] R. J. Natayan, "Transdermal delivery of insulin via microneedles," *Journal of Biomedical Nanotechnology,* vol. 10, no. 9, pp. 2244-2260, 2014.

[43] C. Bronger, "Insulin pen needles and adherance," *Australian Journal of Pharmacy,* vol. 95, no. 1128, pp. 68-71, 2014.

[44] M. J. Taylor and T. S. Sahota, "New technologies in insulin delivery," *Practical Diabetes,* vol. 30, no. 1, pp. 21-26, 2013.

[45] J. D. Zahn, Y. C. Hsieh and M. Yang, "Components of an integrated microfluidic device for continuous glucose monitoring with responsive insulin delivery," *Diabetes Technology and Therapeutics,* vol. 7, no. 3, pp. 536-545, 2005.

[46] Y. Ito, E. Hagiwara, A. Saeki, N. Sugioka and K. Takada, “Feasibility of microneedles for percutaneous absorption of insulin,” *European Journal Of Pharmaceutical Sciences,* vol. 29, pp. 82-88, 2006.

[47] W. Martant, ,. S. P. Davis, N. R. Holiday, J. Wang, H. S. Gill and M. R. Prausnitz, “Transdermal Delivery of Insulin Using Microneedles in Vivo,” vol. 21, no. 6, 2004.

[48] P. Khanna, J. A. Strom and S. Bhansali, “Microneedle-based automated therapy for diabetes mellitus,” *Journal of Diabetes Science and Technology,* vol. 2, no. 6, pp. 1122-1129, 2008.

[49] M. S. Giri Nandagopal, R. Antony, S. Rangabhashiyam and N. Sreekumar, “Overview of microneedle system: A third generation transdermal drug delivery approach,” *Microsystem Technologies,* vol. 20, no. 7, pp. 1249-1272, 2014.

[50] A. Jain, P. Jain, J. Kurmi, D. Jain, R. Jain, S. Chandel, A. Sahu, N. Mody, S. Upadhaya and A. Jain, “Novel strategies for effective transdermal drug delivery: A review,” *Critical Reviews in Therapeutic Drug Carrier Systems,* vol. 31, no. 3, pp. 219-272, 2014.

[51] A. Pankratz, Forecasting with Univariate Box-Jenkins Models: Concepts and Cases, John Wiley & Sons, 1983.

[52] U. U. Shah, M. Roberts, M. O. Gul, C. Tuleu and M. W. Beresford, “Needle-free and microneedle drug delivery in children: A case for disease-modifying antirheumatic drugs (DMARDs),” *International Journal of Pharmaceutics,* vol. 416, no. 1, pp. 1-11, 2011.

[53] H. Liebl and L. C. Kloth, “Skin cell proliferation stimulated by microneedles,” *Journal of the American College of Clinical Wound Specialists,* vol. 4, no. 1, pp. 2-6, 2012.

[54] N. Li, L.-H. Peng, X. Chen, S. Nakagawa and J.-Q. Goa, “Transcutaneous vaccines: Novel advances in technology and delivery for overcoming the barriers,” *Vaccine,* vol. 29, no. 37, pp. 6179-6190, 2011.

[55] J. L. Osborne, I. C. Sanchez and D. R. Paul, “An asymptotic analysis of drug delivery from transdermal patches,” *Journal of Membrane Science,* vol. 442, no. 1, p. 27–30, 2013.

[56] K. M. Krapp and J. L. Longe, How Products are made, Gale Research: Detroit, 1998.

[57] G. Lawrence, “The hypodermic syringe,” *The Lancet,* vol. 359, no. 9311, p. 1074, 2002.

[58] R. F. Donnelly and A. D. Woolfson, “Patient safety and beyond: What should we expect

from microneedle arrays in the transdermal delivery arena?," *Therapeutic Delivery,* vol. 5, no. 6, pp. 653-662, 2014.

[59] R. F. Donnelly, K. Mooney, E. Caffarel-Salvador, B. M. Torrisi, E. Eltayib and J. C. McElnay, "Microneedle-mediated minimally invasive patient monitoring," *Therapeutic Drug Monitoring,* vol. 36, no. 1, pp. 10-17, 2014.

[60] T. Gratieri, I. Alberti, M. Lapteva and Y. N. Kalia, "Next generation intra- and transdermal therapeutic systems: Using non- and minimally-invasive technologies to increase drug delivery into and across the skin," *European Journal of Pharmaceutical Sciences (Article in Press),* 2013.

[61] L. Nordquist, N. Roxhed, P. Griss and G. Stemm, "Novel Microneedle Patches for Active Insulin Delivery are Efficient in Maintaining Glycaemic Control: An Initial Comparison with Subcutaneous Administration," *Pharmaceutical Research,* vol. 24, no. 7, pp. 1381-1388, 2007.

[62] E. E. Kis, G. Winter and J. Myschik, "Devices for intradermal vaccination," *Vaccine,* vol. 30, no. 3, p. 523– 538, 2012.

[63] C. Edens, M. L. Collins, J. Ayers, P. A. Rota and M. R. Prausnitz, "Measles vaccination using a microneedle patch," *Vaccine,* vol. 31, no. 34, p. 3403–3409, 2013.

[64] Y. Cai, M. Xu, M. Yuan, Z. Lui and W. Yuan, "Developments in human growth hormone preparations: Sustained-release, prolonged half-life, novel injection devices, and alternative delivery routes," *International Journal of Nanomedicine,* vol. 9, no. 1, pp. 3527-3538, 2014.

[65] H. S. Gill, S.-M. Kang, F.-S. Quan and R. W. Compans, "Cutaneous immunization: An evolving paradigm in influenza vaccines," *Expert Opinion on Drug Delivery,* vol. 11, no. 4, pp. 615-627, 2014.

[66] D. G. Koutsonanos, R. W. Compans and L. Skountzou, "Targeting the skin for microneedle delivery of influenza vaccine," *Advances in Experimental Medicine and Biology,* no. Doi: 10.1007/978-1-4614-6217-0_13, pp. 121-132, 2013.

[67] A. Herwadkar and A. K. Banga, "Peptide and protein transdermal drug delivery," *Drug Discovery Today: Technologies,* vol. 9, no. 2, pp. 147-154, 2012.

[68] N. Singh, H. Kalluri, A. Herwadkar, A. Badkar and A. K. Banga, "Transcending the skin barrier to deliver peptides and proteins using active technologies," *Critical Reviews in Therapeutic Drug Carrier Systems,* vol. 29, no. 4, pp. 265-298, 2012.

[69] M. R. Prausnitz, J. A. Mikszta, M. Cormier and A. K. Andrianov, “Microneedle-based vaccines,” *Current Topics in Microbiology and Immunology,* vol. 333, p. 369–393, 2009.

[70] H. Gray, Gray's Anatomy, 39th ed., S. Standring, Ed., Spain: Elsevier, 2005.

[71] H. Gray, Gray's Anatomy, 40th ed., S. Standring, Ed., Spain: Elsevier Limited, 2008.

[72] J. S. Petrofsky, M. Prowse and E. Lohman, “The Influence of Ageing and Diabetes on Skin and Subcutaneous Fat Thickness in Different Regions of the Body,” *The Journal of Applied Research,* vol. 8, no. 1, pp. 55-61, 2008.

[73] S. Seidenari, A. Pagnoni, A. Di Nardo and A. Giannetti, “Echographic Evaluation with Image Analysis of Normal Skin: Variations according to Age and Sex,” *Skin Pharmacol,* vol. 7, pp. 201-209, 1994.

[74] Y. Lee and K. Hwang, “Skin thickness of Korean adults,” *Surgical and Radiologic Anatomy,* vol. 24, pp. 183-189, 2002.

[75] J. J. van de Sandt, J. J. van Burgsteden, S. Cage, P. L. Carmichael, I. Dick, S. Kenyon, G. Korinth, F. Larese, J. C. Limasset, W. J. Maas, L. Montomoli, J. B. Nielsen, J. P. Payan, E. Robinson, P. Sartorelli, K. H. Schaller, S. C. Wilkinson and F. M. Williams, “In vitro predictions of skin absorption of caffeine, testosterone, and benzoic acid: a multi-centre comparison study,” *Regulatory Toxicology and Pharmacology,* vol. 39, p. 271–281, 2004.

[76] L. K. Smalls, R. R. Wickett and M. O. Visscher, “Effect of dermal thickness, tissue composition, and body site on skin biomechanical properties,” *Skin Research and Technology ,* vol. 12, no. 1, pp. 43-49, 2006.

[77] N. Roxhed, T. C. Gasser, P. Griss, G. A. Holzapfel and G. Stemme, “Penetration-Enhanced Ultrasharp Microneedles and Prediction on Skin Interaction for Efficient Transdermal Drug Delivery,” *Journal of Microelectromechanical Systems,* vol. 16, no. 6, pp. 1429-1440, 2007.

[78] P. K. Karande, A. Jain and S. Mitragotri, Relationships between skin’s electrical impedance and permeability in the presence of chemical enhancers, vol. 110, Santa Barbara,: Journal of Controlled Release, 2006, pp. 307-313.

[79] A. B. Cua, K. P. Wilhelm and H. I. Maibach, “Elastic properties of human skin: relation to age, sex, and anatomical region,” *Archives of Dermatological Research ,* vol. 282, no. 5, pp. 283-288, 1990.

[80] B. Boettcher, K. J. Stahlhofer, V. Mattle, V. Seeber, C. Brezinka and L. Wildt,

"Ultrasonographic assessment of skin thickness in patients with PCOS - a case-control study," *Gynecological endocrinolog,* vol. 29, no. 4, pp. 380-383, 2013.

[81] N. Magnenat-Thalmann, P. Kalra, J. L. Lévêque, R. Bazin, D. Batisse and B. Querleux, "A Computational Skin Model: Fold and Wrinkle Formation," *IEEE Transactions on Information Technology in Biomedicine,* vol. 6, no. 4, pp. 317-323, 2002.

[82] A. Bohling, S. Bielfeldt, A. Himmelmann, M. Keskin and K. P. Wilhelm , "Comparison of the stratum corneum thickness measured in vivo with confocal Raman spectroscopy and confocal reflectance microscopy," *Skin Research and Technology,* vol. 0, no. 1, pp. 1-8, 2013.

[83] D. L. Dhamech, V. B. Rajendra, V.B., Rathi, A.A., Ghadl, A. A. Rathi, S. V. Ghadlinge, M. Saifee and M. G. Dehghan, "Physical approaches to penetration enhancement," *International Journal of Health Research,* vol. 3, no. 2, pp. 57-70, 2010.

[84] T. R. Singh, M. J. Garland, C. M. Cassidy, K. Migalska, Y. K. Demir, S. Abdelghany, E. Ryan, D. Woolfson and R. F. Donnelly, "Microporation techniques for enhanced delivery of therapeutic agents," *Recent Patents on Drug Delivery and Formulation,* vol. 4, no. 1, pp. 1-17, 2010.

[85] M. Milewski, N. K. Brogden and A. L. Stinchcomb, "Current aspects of formulation efforts and pore lifetime related to microneedle treatment of skin," *Expert Opinion on Drug Delivery,* vol. 7, no. 5, pp. 617-629, 2010.

[86] S. D. Gittard, A. Ovsianikov, B. N. Chichkov, A. Doraiswamy and R. J. Narayan, "Two-photon polymerization of microneedles for transdermal drug delivery," *Expert Opinion on Drug Delivery,* vol. 7, no. 4, pp. 513-533, 2010.

[87] R. K. Subedi, S. Y. Oh, M.-K. Chun and H.-K. Choi, "Recent advances in transdermal drug delivery," *Archives of Pharmacal Research,* vol. 33, no. 3, pp. 339-351, 2010.

[88] S. S. Lanke, J. G. Strom and A. K. Banga, "Enhancement of transdermal delivery of heparin by various physical and chemical enhancement techniques," *Critical Reviews in Therapeutic Drug Carrier Systems,* vol. 26, no. 6, pp. 581-606, 2009.

[89] M. R. Prausnitz, "Microneedles for transdermal drug delivery," *Advanced Drug Delivery Reviews,* vol. 56, p. 581– 587, 2004.

[90] J. H. Park, M. G. Allen and M. R. Prausnitz, "Biodegradable polymer microneedles: Fabrication, mechanics and transdermal drug delivery," *Journal of Controlled Release,*

vol. 104, pp. 51-66, 2005.

[91] N. D. Singh and A. K. Banga , “Controlled delivery of ropinirole hydrochloride through skin using modulated iontophoresis and microneedles,” *Journal of Drug Targeting,* vol. 21, no. 4, pp. 354-366, 2013.

[92] S. Mitragotri and J. Kost, “Low-frequency sonophoresis A review,” *Advanced Drug Delivery Reviews,* vol. 56, p. 589– 601, 2004.

[93] M. G. Nava-Arzaluz, L. Calderón-Lojero, D. Quintanar-Guerrero, R. Villalobos-García and A. Ganem-Quintanar, “Microneedles as transdermal delivery systems: Combination with other enhancing strategies,” *Current Drug Delivery,* vol. 9, no. 1, pp. 57-73, 2012.

[94] H. Trommer and R. H. Neubert, “Overcoming the stratum corneum: The modulation of skin penetration. A review,” *Skin Pharmacology and Physiology,* vol. 19, no. 2, pp. 106-121, 2006.

[95] J. Z. Hilt and N. A. Peppas, “Microfabricated drug delivery devices,” *International Journal of Pharmaceutics,* vol. 206, no. 1-2, pp. 15-23, 2005.

[96] S. M. Al-Saidan, “Transdermal self-permeation enhancement of ibuprofen,” *Journal of Controlled Release,* vol. 100, no. 1, pp. 199-209, 2004.

[97] T. Tanner and R. Marks, “Delivering Drugs by the Transdermal Route: Review and Comment,” *Skin Research and Technology,* vol. 14, no. 3, pp. 249-260, 2008.

[98] A. K. Banga, “Microporation applications for enhancing drug delivery,” *Expert Opinion on Drug Delivery,* vol. 6, no. 4, pp. 343-354, 2009.

[99] A. Arora, M. R. Prausnitz and S. Mitragotri, “Micro-scale devices for transdermal drug delivery,” *International Journal of Pharmaceutics,* vol. 364, no. 2, pp. 227-236, 2008.

[100] M. Milewski and A. Mitra, “Recent developments in microneedle technology for transdermal drug delivery & vaccination,” *Drug Development and Delivery,* vol. 12, no. 6, pp. 30-35, 2012.

[101] S. Mitragotri, “Mechanical disruption of skin barrier for vaccine delivery,” *Drug Delivery System,* vol. 27, no. 3, pp. 202-212, 2012.

[102] D. Ukawala Ravikumar and J. Vasava, “Physical enhancement techniques for transdermal delivery system: A review,” *International Journal of Pharmacy and Technology,* vol. 4, no. 3, pp. 2174-2200, 2012.

[103] J. D. Yadav, K. A. Vaidya, P. R. Kulkarni and R. Raut, “Microneedles: Promising technique for transdermal drug delivery,” *International Journal of Pharma and Bio Sciences,* vol. 2, no. 1, pp. 684-708, 2011.

[104] M. Shikida, “Fabrication of microneedles by MEMS technologies,” *Drug Delivery System,* vol. 27, no. 3, pp. 176-183, 2012.

[105] R. S. Desale, K. S. Wagh, A. M. Akarte, D. T. Baviskar and D. K. Jain, “Microneedle technology for advanced drug delivery: A review,” *International Journal of PharmTech Research,* vol. 4, no. 1, pp. 181-189, 2012.

[106] G. Aggarwal, A. Garg and S. Dhawan, “Transdermal drug delivery: Evolving technologies and expanding opportunities,” *Indian Journal of Pharmaceutical Education and Research,* vol. 43, no. 3, pp. 251-259, 2009.

[107] S. H. Bariya, M. C. Gohel, T. A. Mehta and O. P. Sharma, “Microneedles: An emerging transdermal drug delivery system,” *Journal of Pharmacy and Pharmacology,* vol. 64, no. 1, pp. 11-29, 2012.

[108] S. H. Bariya, M. C. Gohel, T. A. Mehta and O. P. Sharma, “Microneedles: an emerging transdermal drug delivery system,” *Journal of Pharmacy and Pharmacology,* vol. 64, no. 1, pp. 11-29, 2011.

[109] H. J. Choi, B. J. Bondy, D. G. Yoo, R. W. Compans, S. M. Kang and M. R. Prausnitz, “Stability of whole inactivated influenza virus vaccine during coating onto metal microneedles,” *Journal of Controlled Release,* vol. 166, no. 2, pp. 159-171, 2013.

[110] L. Wei-Ze, H. Mei-Rong, Z. Jian-Ping, Z. Yong-Qiang, H. Bao-Hua, L. T. Ting and Z. Y. Yong, “Super-short solid silicon microneedles for transdermal drug delivery applications,” *International Journal of Pharmaceutics,* vol. 389, pp. 122-129, 2010.

[111] L. Y. Chu and M. R. Prausnitz, “Separable arrowhead microneedles,” *Journal of Controlled Release,* vol. 149, p. 242–249, 2011.

[112] B. Bediz, E. Korkmaz, R. Khilwani, C. Donahue, G. Erdos, J. L. Falo and O. B. Ozdoganlar, “Dissolvable Microneedle Arrays for Intradermal Delivery of Biologics: Fabrication and Application,” *Pharmaceutical Research,* pp. 1-19, 2013.

[113] K. Tsioris, W. K. Raja, E. M. Pritchard, B. Panilaitis, D. L. Kaplan and F. G. Omenetto, “Fabrication of Silk Microneedles for Controlled-Release Drug Delivery,” *Advanced Functional Materials,* vol. 22, no. 2, pp. 330-335, 2012.

[114] O. Olatunji, C. C. Igwe, A. S. Ahmed, D. O. Alhassan, G. O. Asieba and D. B. Das,

"Microneedles from fish scale biopolymer," *Apoplied Polymer Science,* vol. 131, no. 12, pp. 1-10, 2014.

[115] X. Wang, X. Chen, Z. Yu and L. wang, "A Novel Fabrication Approach for Microneedles Using Silicon Micromaching Technology," in *1st IEEE International Conference on Nano/Micro Engineered and Molecular Systems*, Zhuhai, 2006.

[116] I. Mansoor, Y. Liu, U. O. Häfeli and B. Stoeber, "Arrays of hollow out-of-plane microneedles made by metal electrodeposition onto solvent cast conductive polymer structures," *Journal of Micromechanics and Microengineering,* vol. 23, no. 8, pp. 1-10, 2013.

[117] S. Kommareddy, B. C. Baudner, A. Bonificio, S. Gallorini, S. Palladino, A. S. Determan, D. M. Dohmeier, K. D. Kroells, J. R. Sternjohn, M. Singh, P. R. Dormitzer, K. J. Hansen and D. T. O'Hagan, "Influenza subunit vaccine coated microneedle patches elicit comparable immune responses to intramuscular injection in guinea pigs," *Vaccine,* vol. 31, no. 34, pp. 3435-3441, 2013.

[118] B. M. Torrisi, V. Zarnitsyn, M. R. Prausnitz, A. Anstey, C. Gateley, J. C. Birchall a and S. A. Coulman, "Pocketed microneedles for rapid delivery of a liquid-state botulinum toxin A," *Journal of Controlled Release,* vol. 165, no. 2, p. Pages 146–152, 2013.

[119] J. Gupta, E. I. Felner and M. R. Prausnitz, "Minimally Invasive Insulin Delivery in Subjects with Type 1 Diabetes Using Hollow Microneedles," *Diabetes technology and Therapeutics,* vol. 11, no. 6, pp. 329-337, 2009.

[120] M. R. Pierre and F. C. Rossetti, "Microneedle-based drug delivery systems for transdermal route," *Current Drug Targets,* vol. 3, no. 2014, p. 15, 281-291.

[121] S. Swain, S. Beg, A. Singh, C. N. Patro and M. B. Rao, "Advanced techniques for penetration enhancement in transdermal drug delivery system," *Current Drug Delivery,* vol. 8, no. 4, pp. 456-473, 2011.

[122] A. El-Laboudi, N. S. Oliver, A. Cass and D. Johnston, "Use of microneedle array devices for continuous glucose monitoring: A review," *Diabetes Technology and Therapeutics,* vol. 15, no. 1, pp. 101-115, 2013.

[123] M. J. Taylor and T. S. Sahota, "New technologies in insulin delivery," *Practical Diabetes,* vol. 30, no. 1, pp. 21-26, 2013.

[124] K. B. Ita, "Transdermal drug delivery: Progress and challenges," *Journal of Drug Delivery Science and Technology,* vol. 24, no. 3, pp. 245-250, 2014.

[125] K. M. Ainslie and T. A. Desai, “Microfabricated implants for applications in therapeutic delivery, tissue engineering, and biosensing,” *Lab on a Chip - Miniaturisation for Chemistry and Biology,* vol. 8, no. 11, pp. 1864-1878, 2008.

[126] J. Y. Fang, P. F. Liu and C. M. Huang, “Decreasing systemic toxicity via transdermal delivery of anticancer drugs,” *Current Drug Metabolism,* vol. 9, no. 7, pp. 592-597, 2008.

[127] E. Ezan, “Pharmacokinetic studies of protein drugs: Past, present and future,” *Advanced Drug Delivery Reviews,* vol. 65, no. 8, pp. 1065-1073, 2013.

[128] J. J. Norman, J. M. Arya, M. A. McClain, P. M. Frew, M. I. Meltzer and M. R. Prausnitz, “Microneedle patches: Usability and acceptability for self-vaccination against influenza,” *Vaccine,* vol. 32, no. 16, pp. 1856-1862, 2014.

[129] K. van der Maaden , S. J. Trietsch, H. Kraan, E. M. Varypataki, S. Romeijn, R. Zwier, H. J. Van Der Linden, G. Kersten, T. Hankemeier, W. Jiskoot and J. Bouwstra, “Novel hollow microneedle technology for depth-controlled microinjection-mediated dermal vaccination: A study with polio vaccine in rats,” *Pharmaceutical Research,* vol. 31, no. 7, pp. 1845-1854, 2014.

[130] J. S. Kochhar, S. Zou, S. Y. Chan and L. Kang, “Protein encapsulation in polymeric microneedles by photolithography,” *International Journal of Nanomedicine,* vol. 7, no. 1, pp. 3143-3154, 2012.

[131] J. Gupta, E. I. Felner and M. R. Prausnitz, “Minimally Invasive Insulin Delivery in Subjects with Type 1 Diabetes Using Hollow Microneedles,” *DIABETES TECHNOLOGY & THERAPEUTICS,* vol. 11, no. 6, pp. 329-337, 2009.

[132] S. Indermun, R. Luttge, Y. E. Choonara, P. Kumar, L. C. Du Toit, G. Modi and V. Pillay, “Current advances in the fabrication of microneedles for transdermal delivery,” *Journal of Controlled Release,* vol. 185, no. 1, pp. 130-138, 2014.

[133] A. Nayak, D. B. Das and G. V. Vladisavljevic, “Microneedle assisted permeation of lidocaine carboxymethylcellulose with gelatine co-polymer hydrogel,” *Pharmaceutical Research,* vol. 31, no. 5, pp. 1170-1184, 2014.

[134] K. Cheung, T. Han and D. B. Das, “Effect of Force of Microneedle Insertion on the Permeability of Insulin in Skin,” *Journal of Diabetes Science and Technology,* vol. 8, no. 3, pp. 444-452, 2014.

[135] Z. Ahmad, E. Stride and M. Edirisinghe, “Novel preparation of transdermal drug-delivery patches and functional wound healing materials,” Journal of Drug Targeting,

vol. 17, no. 9, p. 724–729, 2009.

[136] S. P. Davis, W. Martanto, M. G. Allen and M. R. Prausnitz, “Hollow Metal Microneedles for Insulin Delivery to Diabetic Rats,” *Transactions on Biomedical Engineering,* vol. 52, no. 5, pp. 909-915, 2005.

[137] P. Griss and G. Stemme, “Side-Opened Out-of-Plane Microneedles for Microfluidic Transdermal Liquid Transfer,” *IEEE Journal of Microelectromechanical Systems,,* vol. 12, no. 3, pp. 296-301, 2003.

[138] K. Lee and H. Jung, “Drawing lithography for microneedles: A review of fundamentals and biomedical applications,” *Biomaterials,* vol. 33, no. 30, pp. 7309-7326, 2012.

[139] D. Jain, V. Jain and R. Singh, “Novel antigen delivery technologies: A review,” *Drug Delivery and Translational Research,* vol. 1, no. 2, pp. 103-112, 2011.

[140] H. Khan, P. Mehta, H. Msallam, D. Armitage and Z. Ahmad, “Smart microneedle coatings for controlled delivery and biomedical analysis,” Journal of Drug Targeting, vol. 22, no. 9, pp. 790-795, 2014.

[141] S. Hashmi, P. Ling, G. Hashmi, R. Gaugler and W. Trimmer, “Genetic transformation of nematodes using arrays of micromechanical piercing structures,” *Biotechniques,* vol. 19, no. 5, pp. 766-770, 1995.

[142] W. Martanto, S. P. Davis, N. R. Holiday, J. Wang, H. S. Gill and M. R. Prausnitz, “Transdermal Delivery of Insulin Using Microneedles in Vivo,” *Pharmaceutical Research,* vol. 21, no. 6, pp. 947-952, 2004.

[143] X. Hong, L. Wei, F. Wu, Z. Wu, L. Chen, Z. Liu and W. Yuan, “Dissolving and biodegradable microneedle technologies for transdermal sustained delivery of drug and vaccine,” *Drug Design, Development and Therapy,* vol. 7, no. 1, pp. 945-952, 2013.

[144] A. Nayak and D. B. Das, “Potential of biodegradable microneedles as a transdermal delivery vehicle for lidocaine,” *Biotechnology Letters,* vol. 35, no. 9, pp. 1351-1363, 2013.

[145] J. W. Lee, M. R. Han and J. H. Park, “Polymer microneedles for transdermal drug delivery,” *Journal of Drug Targeting,* vol. 21, no. 3, pp. 211-223, 2013.

[146] A. Backovic, D. Wolfram, B. Del-Frari, H. Piza, L. A. Huber and G. Wick, “Simultaneous analysis of multiple serum protiens adhering to the surface of medical grade polydimethylsiloxane elastomers,” *Journal of Immunological Methods,* vol. 328, p. 118–127, 2007.

[147] L. Y. Chu, S.-O. Choi and M. R. Prausnitz, “Fabrication of Dissolving Polymer Microneedles for Controlled,” *Journal of Pharmaceautical Science,* vol. 99, no. 10, pp. 4228-4238, 2010.

[148] K. Lee, C. Y. Lee and H. Jung, “Dissolving microneedles for transdermal drug administration prepared by stepwise controlled drawing of maltose,” *Biomaterials,* vol. 32, no. 11, p. 3134e3140, 2011.

[149] C. J. Martin, C. J. Allender, K. R. Brian, A. Morrissey and J. C. Birchall, “Low Temperature fabrication of Biodegradable sugar glass microneedles for transdermal drug delivery applications,” *Journal of controlled Release,* pp. 93-101, 2012.

[150] T. R. Singh, N. J. Dunne, E. Cunningham and R. F. Donnelly, “Review of patents on micro needle applicators,” *Recent Patents on Drug Delivery and Formulation,* vol. 5, no. 1, pp. 11-23, 2011.

[151] V. Sachdeva and A. K. Banga, “Microneedles and their applications,” *Recent Patents on Drug Delivery and Formulation,* vol. 5, no. 2, pp. 95-132, 2011.

[152] M. Rizwan, M. Aqil, S. Talegaonkar, A. Azeem, Y. Sultana and A. Ali, “Enhanced transdermal drug delivery techniques: An extensive review of patents,” *Recent Patents on Drug Delivery and Formulation,* vol. 3, no. 2, pp. 105-124, 2009.

[153] D. F. H. Chowdhury, “Microneedle Transdermal Delivery Device”. Loughborough Patent US 2010042050, 18 February 2010.

[154] K. Kobayashi and H. Hamamoto, “Applicator device of pinholder type microneedle”. Kagawa Patent WO 2011016230, 10 February 2011.

[155] B. J. Moga, K. B. Chase and G. D. Smith, “DD device”. Madison Patent WO 2011084951, US 20110172645, 14 July 2011.

[156] B. A. Gonzalez, S. A. Burton, J. Hu, C.-Y. Ng, R. P. Simmers, T. J. Gilbert, S. M. Burke, R. A. Harkins and L. A. Schleif, “Hollow microneedle arrays”. St Paul Patent WO 2011014514, 3 February 2011.

[157] K. Honda, M. Motoi and K. Takada, “Method for producing stamper for microneedle”. Kyoto Patent CA 2696810, JP 2011078618, WO 2011043086, AU 2010201434, KR 2011067009, EP 2343102 A1, 10 February 2011.

[158] A. S. Cantor and A. J. Stockholm, “Transdermal adhesive patch assembly with removable microneedle array and method of using same”. St Paul Patent WO 2013096026, 27 June 2013.

[159] C. Y. Lim, “The plastic microneedle strip”. Singapore Patent WO 2013066262, 10 May 2013.

[160] T. designation of the inventor has not yet been file, “A microneedle device and method for the fabrication thereof”. Cork Patent EP 2289843 , 2 March 2011.

[161] B. Akiyoshi, “Biocompatible microneedle, manufacture thereof, and sheet having the microneedle”. Kitakyushu-shi Patent JP 2011083387, 18 April 2012.

[162] Z. Huizhen, W. Huibin, X. WZhang and J. Eizheng, “Degradable polymer-based microneedle patch and its preparation method”. Beijing Patent CN 102000020, 6 April 2011.

[163] R. Yue and Y. Wang, “Metal microneedle array, transdermal patch, micro needle roller and micro needle electrode array”. Beijing Patent CN 103263727, 28 August 2013.

[164] H. Sugimura, G. Suzuki, M. Ueno, Y. Kodama and T. Tomono, “Microneedle and method of manufacturing microneedle”. Tokyo Patent US 20130030374 A1, 31 January 2013.

[165] L. Zhang and Z. Wu, “riamcinolone acetonide biodegradable maltose microneedle array for treating scar and its preparation method”. Shanghai Patent CN 103181887, 3 July 2013.

[166] E. Saacs and T. Cobbledick, “A microneedle roller”. Borehamwood Patent GB2472778A, 23 February 2011.

[167] A. L. Stinchcomb, S. L. Banks, M. J. Golinski, J. L. Howard and D. C. Hammell, “Cannabidiol prodrugs in topical and transdermal administration with microneedles”. Lexington Patent WO 2011026144, S 20110052694, 03 March 2011.

[168] P. A. Eckhoff, R. A. Hyde, M. Y. Ishikawa, J. T. Kare, E. C. Leuthardt, D. J. Rivet, E. A. Sweeney, L. L. J. Wood and V. Y. Wood, “Device, system, and method for targeted delivery of anti-inflammatory medicaments to a mammalian subject”. Bellevue Patent US 20110118560, US 20110118656, US 20110117150, US 20110118652, US 20110118696, US 20110118653, US 20110118697, US 20110118698, US 20110118652, US 20110118656, 19 May 2011.

[169] M. Li, Y. Liu, C. Wang, X. Yang, D. Yang, Y. Sun, G. Hao and X. Gao, “Inactivated vaccine for immunoprophylaxis of bovine mastitis caused by Escherichia coli and its preparation method”. Raleigh Patent CN 101991846, 30 March 2011.

[170] X. Chen and W. Zhang, “Method and apparatus for delivery of bioactive molecules to

cells”. Hong Kong Patent US 20130171722, 4 July 2013.

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