Expert Opinion

informa
healthcare

Applications of ultrasonic skin permeation in transdermal drug delivery

Nadine Barrie Smith
The Pennsylvania State University, 21 Hallowell Building, University Park, PA 16802, USA

Transdermal ultrasound-mediated drug delivery has been studied as a method for needle-less, non-invasive drug administration. Potential obstacles include the *stratum corneum*, which is not sufficiently passively permeable to allow effective transfer of many medications into the blood-stream without active methods. A general review of the transdermal ultrasound drug delivery literature has shown that this technology offers promising potential for non-invasive drug administration. Included in this review are the reported acoustic parameters used for achieving delivery, along with the known intensities and exposure times. Ultrasound mechanisms are discussed as well as spatial field characteristics. Accurate and precise quantification of the acoustic field used in drug delivery experiments is essential to ensure safety versus efficacy and to avoid potentially harmful bioeffects.

Keywords: bioeffects, drug delivery, intensity, transdermal, ultrasound

Expert Opin. Drug Deliv. (2008) 5(10):1107-1120

1. Introduction

Studies over many years have shown that ultrasound-mediated transdermal drug delivery offers a promising method for non-invasive transdermal drug administration [1-3]. Ultrasound is one of several needle-less methods currently under investigation or on the market [4-6]. In addition to ultrasound, other transdermal drug delivery methods include active transport mechanisms such as iontophoresis [7], electroporation [8] and microneedles [9]. The use of ultrasound can be classified as a physical penetration enhancement approach, along with electrical methods such as iontophoresis and electroporation. In comparison, chemical enhancers include the use of liposomes for crossing the *stratum corneum* and researchers have investigated combining these effective strategies for synergistic results [10]. Of the numerous methods that exist, one advantage of ultrasound is the accepted safety of ultrasound, based on careful research on bioeffects at diagnostic imaging energy levels [11,12]. As an extension of this concept, ultrasound heating in sports or physical therapy is commonly used in injury treatment. To the general public, other methods such as laser radiation, magnetophoresis and skin stretching, for example, can have an aura of mystery as to the specifics regarding the technology. Given this responsibility, it is essential that the physical nature of ultrasound used in drug delivery be scientifically well understood and characterized so as to ensure both safe and effective delivery.

Although they are not explored here, ultrasound-based methods are also being used for gene delivery or organ-specific drug delivery. For example, there exist many therapeutic drugs that can potentially treat several neurological disorders if they could cross the blood–brain barrier. Ultrasound has recently been shown to be effective for non-invasive, transient, blood–brain barrier disruption in the delivery of chemotherapy agents such as liposomal Doxorubicin and Herceptin [13].

10.1517/17425240802399604 © 2008 Informa UK Ltd ISSN 1742-5247

These studies also show the safe but effective nature of ultrasound, since the blood–brain barrier heals within a few hours post-exposure. In the use of ultrasound for transdermal delivery, it is similarly important to deliver drugs across the dermal barriers for drug delivery, but with minimal disruption to the tissue. This follows the radiological principle of 'as low as reasonably achievable' (ALARA) to minimize the dose while accepting some risk to achieve efficacy.

Readers are also directed to several recent, comprehensive publications for further commentary on transdermal drug delivery, including ultrasound-based methods [3,4,6,14]. This review differs from previous publications [15] in the additional information given regarding the energy levels used for ultrasound drug delivery. To this end, a case is presented for the need for accurate and precise methods for reporting repeatable energy levels used in the scientific literature with respect to ultrasound transdermal drug delivery and the safety of these methods.

2. Mechanism of ultrasound

Ultrasound refers to mechanical waves with frequencies above human hearing (> 20 kHz), and clinical ultrasound imaging specifically uses frequencies in the range of 1 – 15 MHz. Unlike X-rays, mechanical sound propagation requires a medium to support transmission. The medium, such as a fluid or solid, can be described as atoms or molecules which are bound by internal elastic forces. Ultrasound is a sinusoidal pressure wave which causes the molecules to become displaced from their equilibrium positions. A one-dimensional representation of this interaction can be used to simplify this description. **Figure 1** shows one wavelength of a sinusoidal pressure wave propagation in the z-direction. The pressure oscillates between a maximum (compressional, P_c) and minimum (rarefactional, P_r) about the ambient pressure as it moves through the medium. Within the medium, molecules move closer together due to the compressional pressure and spread apart due to the rarefactional pressure. Wave propagation also depends on other parameters such as density, particle displacement, temperature, attenuation and other variables. While sound can travel through air, fluids and solids, this review will focus on fluids, since the body is mostly water-based tissue (> 70%).

Cavitation describes the oscillation or rapid expansion and collapse of gaseous bubbles in response to an alternating pressure field. Cavitation types can be broken down into two non-exclusive categories. The first is stable, or non-inertial cavitation, where the cavity (or bubble) oscillates about its equilibrium radius in response to relatively low acoustic pressures [11]. The second is transient, or inertial cavitation, whereby the equilibrium size varies greatly within very few acoustic cycles. During inertial cavitation, the rapid, violent collapse of bubbles is associated with high acoustic pressures and temperatures on the order of 1000 – 2000 K [16,17]. With inertial cavitation, short-lived fissures in the liquid are generated in response to high acoustic pressures and/or lower frequencies. The violent hydrodynamic forces due to a collapsing bubble can cause severe damage within biological media and free radicals can be produced by this phenomenon [18,19]. From previous research, the measured cavitation pressure amplitude in dog thigh muscle *in vivo* was found to depend linearly on frequency, with a slope of 5.3 MPa/MHz [20].

Compared to the kilohertz range, ultrasound in the megahertz range also produces cavitation, although much higher pressures are required to exceed the cavitation threshold. Above this threshold, cavitation has been shown to disrupt cells and damage tissue [21,22]. Mechanical bioeffects in tissues with gas bodies include lung hemorrhage in mice, rats, monkeys and pigs [23]. Cavitation also occurs at diagnostic ultrasound imaging power levels [24,25], which has motivated the introduction and continuous re-examination of the mechanical index to identify a threshold pressure for the onset of inertial cavitation [26,27]. Even in the absence of well-defined gas bodies, there exist non-thermal bioeffects due to ultrasound that are known to occur in the absence of excessive heating or evidence of cavitation bubbles. In this situation, the mechanism is in the form of radiation force, torque or acoustic streaming [28].

Determining the threshold and energy of a cavitation event is difficult in the best of circumstances [29,30]. Scientists try to follow three experimental rules with respect to cavitation: understand the liquid (including impurities), understand the sound field and know when a cavitation effect happens [29]. These cavitation study 'golden rules' are better stated as, 'know thy liquid, know thy sound field and know when something happens'. The first rule refers to the cavitation threshold, while the second rule relates to accurate measurements of the acoustic field. The third relates to observable cavitation events or secondary related information. There are various reliable and scientifically established methods for quantifying an acoustic field [31], including passive detection methods for cavitation events. By sensing acoustic emissions generated by the collapsing bubble, passive cavitation detection identifies cavitation events and has previously been used with lithotripters at low frequencies [32,33] and high intensity focused ultrasound at 1.5 MHz [34]. Other methods include laser scattering methods based on the detection of scattered light reflected during the bubble's lifetime [35], coupling the light of a laser diode into a light fiber to register the ultrasound-induced modification of the refractive index in a sample [36], or spectral analysis of cavitation noise from a heterodyne fiber-optic hydrophone [37].

In contrast to mechanical effects from an acoustic wave, thermal effects also occur in response to a wave propagating in tissue. Associated attenuation losses are due to absorption and scattering. The absorption mechanism consists of viscous

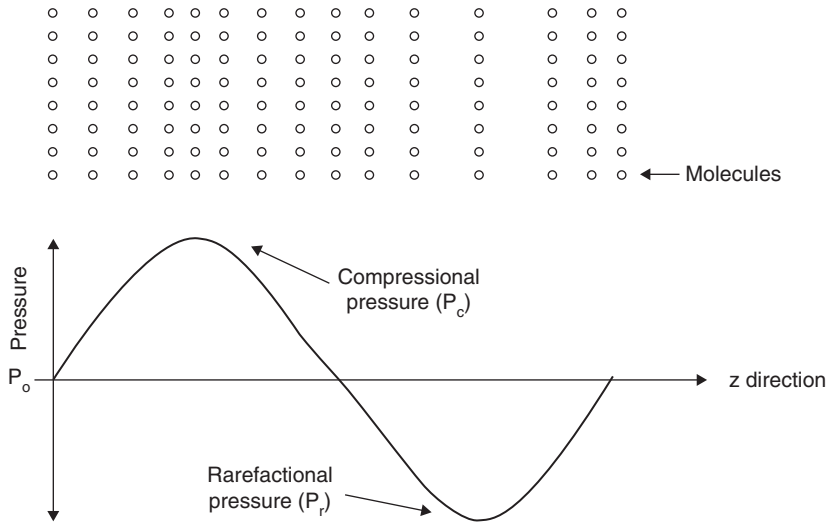

Figure 1. A linear, ultrasound wave is sinusoidal with an increased (compressional, P_c) and decreased (rarefactional, P_r) pressure field with respect to the ambient pressure (P_o). Molecules within the media are pushed closer or separated due to the compressional and rarefactional pressure.

losses, heat conduction and relaxation processes, while scattering occurs when acoustic energy is deflected or redirected from its normal propagation direction. Absorption mechanisms can be divided into three categories: viscous losses, heat or thermal conduction losses and losses due to molecular exchanges of energy. To appreciate viscous losses it must be recalled that sound waves in a medium cause expansions and contractions (**Figure 1**). Fluids exhibit resistance to the distortion or viscosity. Thus, the relative motion between adjacent parts of the medium caused by expansions and compressions due to a sound wave leads to a viscous loss or frictional loss. Thermal losses result from conduction of thermal energy between higher temperature compressions and lower temperature rarefactions. Taking into account both viscous and thermal conductivity losses of the media gives rise to a classical absorption coefficient [38]. However, the classical absorption coefficient does not account in full for values of the absorption coefficient, which are experimentally measured in fluids. Much of the excess absorption beyond the classical absorption is accounted for by three processes known as structural, thermal and chemical relaxation.

Disturbance to the structure of a fluid due to a propagating wave changes the equilibrium molecular structure. The energy going into rearranging the molecular structure, thereby causing the structure to be more closely packed, causes energy to be removed from the wave. Another mechanism for acoustic absorption can be predicted by taking into account the internal structure of the molecules and the interaction between the molecules that lead to internal vibrations, rotations and translations. Absorption arising from perturbations in molecular equilibrium due to temperature alterations in the compressional wave is considered to be due to thermal relaxation mechanisms. Finally, the volume change which results from the perturbation of a chemical reaction from equilibrium by the pressure disturbance when an acoustic wave propagates through a liquid medium, is considered to be a chemical relaxation mechanism.

Understanding the acoustic field can be complicated since it depends on the frequency, shape and size of the transducer, along with the properties of the medium and a host of other variables that can lead to measurement variations [31]. A first order examination considers the acoustic field from a plane piston transducer with a radius a. The plane piston is very similar to the radiating source found on most ultrasound equipment used for tissue heating in sports therapy [39]. For a piston, certain on-axis pressure field characteristics are well known and can be used for characterizing acoustic fields. How quickly the wave travels though the material is given by $c = \lambda f$ where c is the speed of sound in the media (m/s), λ is the wavelength (m) and f is the frequency (s^{-1} or Hz). For water at 20°C, the speed of sound is 1481 m/s; thus for a 1 MHz ultrasound signal the wavelength would be 1.5 mm. Close to the face of the transducer the pressure field oscillates between a series of maximum values and nulls (**Figure 2A**). The final oscillation is known as the last axial maximum located at $\approx a^2/\lambda$. For a plane piston, this location also forms the boundary between the near field ('Fresnel zone') and the far field ('Fraunhofer zone') of the transducer [38]. Beyond the far field, the beam diverges at an angle of $\theta \cong \sin^{-1}(0.61\lambda/a)$. Similar to a radiating radio antenna, the off-axis field pattern also has a series of much smaller pressure field lobes and nulls (**Figure 2B**). The first null between the main lobe and first side lobe is at $\theta \cong \sin^{-1}(0.61\lambda/a)$. The exact radiation

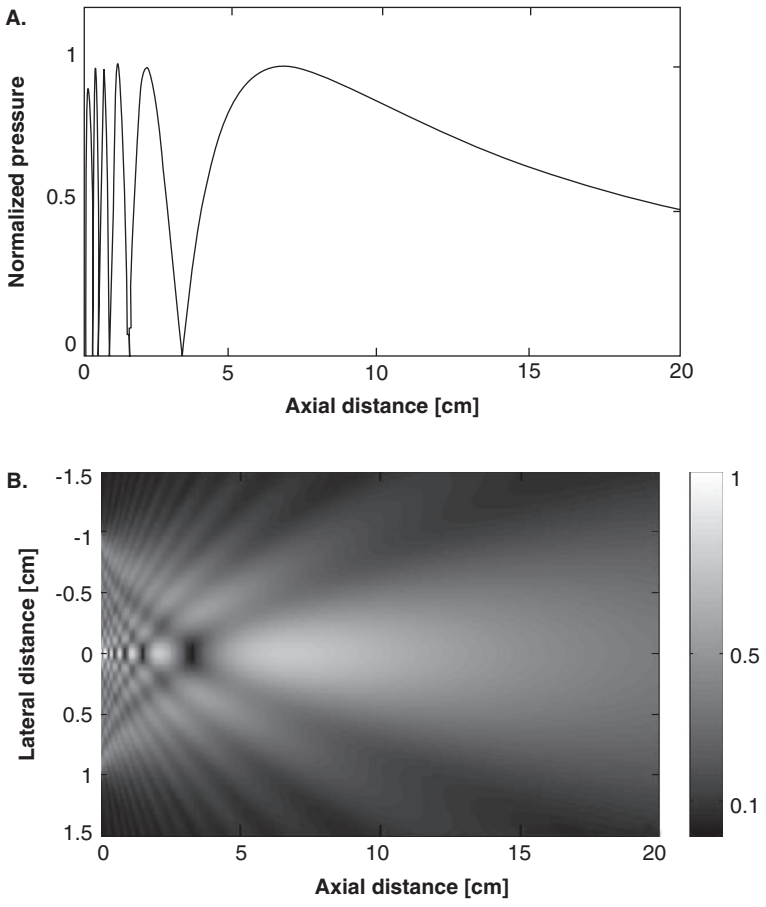

Figure 2. A. A plane piston transducer (located at an axial distance = 0 cm) operating at 1 MHz with a radius of 1 cm has a last axial maximum located at 6.7 cm from the face of the transducer. Close to the face of the transducer the pressure field oscillates between a series of absolute pressure maxima and nulls. **B.** For the same transducer, the calculated axial and lateral spatial pressure field is graphed showing the variable pressure field based on spatial location. The grayscale bar on the right indicates the normalized pressure.

or spatial intensity field from a circular piston is a complex integral requiring a computer to generate the three-dimensional pattern (**Figure 2B**).

Figure 2A and **B** show the respective on-axis and 2D normalized pressure fields for a plane piston 1 MHz transducer with a radius of 1 cm, which is similar to those on therapeutic ultrasound systems [39]. What both plots show is the high spatial variation of the acoustic field. At the most basic level, the intensity is the pressure squared divided by 2 times the characteristic impedance of the tissue. What has been recognized by the clinical diagnostics ultrasound community is that the intensity reported as such is not descriptive of the true acoustic field, given the spatial non-uniformity (**Figure 2B**) and temporal driving conditions (i.e., continuous wave versus pulsing).

For use in humans, diagnostic ultrasound devices are characterized by a range of descriptive intensity measures, including the spatial peak temporal peak (I_{sptp}), spatial peak temporal average (I_{spta}), spatial peak pulse average (I_{sppa}) and spatial average temporal average (I_{sata}). As one example, the spatial peak temporal peak intensity describes the location of the maximum of the spatial intensity field regardless of the pulsing of the transducer. However, the other intensities are just as descriptive of the operation of the system and these calculations with measurement guidelines can be found [40]. A level of careful understating of the acoustic field leads to proper use and reduced risk [12].

3. Current transdermal applications

For transdermal drug delivery, the *stratum corneum* forms a barrier to drug diffusion. This *stratum corneum* barrier varies in thickness (≈ 10 – 30 μm) depending on the location [41], with the palm having one of the thickest layers (> 150 μm). In passive drug delivery across the *stratum corneum*, molecules with a weight of less than 500 Da are at the upper end of the transport boundary [42,43]. For larger molecules, the low permeability is attributed to this outermost skin layer, which

consists of a compact and organized structure of cells named keratinocites surrounded by lipid bilayers. Once the drug has traversed the *stratum corneum*, the next layer is easier to cross and subsequently the drug can reach the capillary vessels to be absorbed [44].

Ultrasound-enhanced transdermal drug delivery offers advantages over traditional injection drug delivery methods, which are invasive and painful. From a clinical application viewpoint, there are relatively few drugs, proteins or peptides that are commonly administered transdermally using ultrasound technology, but there are many companies that are working to change this. From a research viewpoint, in contrast, the list of compounds that have been shown to transdermally cross skin via ultrasound is increasing. **Table 1** presents an overview of the transdermal ultrasound-mediated drug delivery literature from the past several years with some commentary on notable contributions. This review endeavors to be different from previous reviews on this topic with the inclusion of the ultrasound intensity and exposure time used for the transdermal drug delivery. Additionally, the table lists the delivered compound or drug, its respective molecular weight (Daltons) and the experimental sample (animal) preparation, including whether the experiment was performed under *in vivo* or *in vitro* conditions. While the table endeavors to present accurate information regarding the experiments, some information was missing or difficult to interpret. Readers are therefore directed to the original publications to determine the exact experimental conditions.

Of major interest is the ability to translate this research to clinical practice and therefore the reported ultrasound frequency and device are listed. Ultrasound devices can be classified into different categories. Commercial-made include sonicators (≈ 20 kHz), ultrasound heating devices intended for therapy (> 1 MHz) and transducers, which are commercially purchased or fabricated in-house. Noteworthy differences between high (1 – 3 MHz) and low (≈ 20 kHz) frequency ultrasound appear to be that low frequency ultrasound enhances transdermal drug transport ≈ 1000 times more than high frequency ultrasound [45]. Additionally, a direct relationship between enhancement of transdermal transport and ultrasound frequency has been shown [46,47]. As shown in the table, effective delivery depends on acoustic parameters such as frequency, intensity and exposure time. Other operational conditions, for example continuous wave or pulsed wave with duty cycle (pulse duration/pulse repetition period) information, were not always presented. If specific intensity parameters were known (for example spatial peak temporal peak intensity, I_{sptp}), then this was listed.

While a comment on each drug could be made, insulin deserves particular focus given the public impact. This is due to the ever-increasing number of people with diabetes in the USA and the need to develop needle-less methods for insulin delivery [48]. In the USA alone, approximately 24 million people or 8% of the population suffer from diabetes. From a human and economic perspective, diabetes is one of the most costly diseases. Management of diabetes often requires painful repetitive blood glucose tests and insulin injections up to three or four times each day. Between injections, blood sugar levels can fluctuate and remain out of balance until the next test or injection, increasing the risk of tissue or organ damage. Aversion to needles remains one of the most difficult challenges to maintaining tight glucose control.

Early research using ultrasound technology includes effective *in vivo* transport of insulin at 48 kHz using an ultrasonic bath [1], and at 105 kHz [49] using a commercially obtained transducer. Many early experiments were performed using either an ultrasound sonicator, ultrasonic bath or commercial transducer (**Table 1**). A major drawback so far in exploiting ultrasound for non-invasive drug delivery is the large size and poor mobility of the ultrasound devices. In general, commercial sonicators are large, heavy, table-top devices specifically designed for lysis of cells, catalyzing reactions, creating emulsions or cleaning. Yet, until a cure is found, needle-less alternatives to syringes and pumps is a medical need and a major portion of the healthcare market. In addition to ultrasound methods for insulin, other transdermal methods include iontophoresis [50], electroporation [51] and microneedles [52].

4. Consideration of bioeffects with transdermal delivery

A bioeffect is 'any biological change occurring as the result of an exposure, whether physiological or harmful' [53]. In response to ultrasound exposure, harm is 'an adverse outcome' and safety is 'freedom from unacceptable risk'. Using these definitions, the resulting outcome from the use of ultrasound for drug delivery (**Table 1**) is technically a bioeffect. Whether or not the resulting effect on skin can be classified as safe or harmful requires a closer examination of the experimental conditions. For example, subtle, non-obvious mechanical bioeffects from sonoporation have been shown in the literature [27]; these include free radical generation, erythrocyte agglutination, DNA strand breaks and sister chromatid exchange.

As many researchers are interested in effective transdermal drug delivery, many are also interested in the possible harmful bioeffects that can occur. Using 20 kHz ultrasound with hairless rats, erythema (redness due to capillary dilatation) was shown at an intensity exposure level of 2.5 W/cm^2. Further inspection of the skin *in vivo* at 24 h post-exposure showed deep dermal and muscle necrosis [54]. Others have reported noticeable skin damage from ultrasound transdermal drug delivery experiments [55]. One group has examined the morphological changes induced with *in vitro* hairless mouse skin and human skin after ultrasound exposure from a transdermal drug delivery system. The skins

Table 1. Broad list of compounds that have been delivered transdermally, and the corresponding frequency, exposure time and intensity of the ultrasound used.

Compound and Ref.	MW	Preparation	Frequency	Time	Intensity	Device
Aldosterone [65]	832	*In vitro* human	20 kHz	24 h	125 mW/cm^2	Sonicator[1]
Benzene [44]	78	*In vitro* human	1 – 3 MHz	4 h	0 – 2 W/cm^2	Therapeutic US[5]
Bicarbonate [66]	136	*In vivo* rat	20 kHz	15/5 min[1a]	1 W/cm^2	Sonicator[1]
Butanol [44]	74	*In vitro* human	1 – 3 MHz	24 h	125 mW/cm^2	Therapeutic US[5]
Butanol [65]	74	*In vitro* human	20 kHz	4 h	0 – 2 W/cm^2	Sonicator[1]
Caffeine [44]	194	*In vitro* human	1 – 3 MHz	4 h	0 – 2 W/cm^2	Therapeutic US[5]
Caffeine [67]	194	*In vitro* human	20 kHz	10 min CW 1 h pulsed	2.5 W/cm^2	Sonicator[7]
Calcein [68]	623	*In vitro* rat	41, 158, 445 kHz	30 min	60 – 240 mW/cm^2	US transducer[13]
Calcein [69]	623	*In vitro* cell membrane	20, 57, 76, 93 kHz	0 – 180 s	0 – 3 W/cm^2	US transducer[17]
Calcein [70]	623	*In vitro* pig	20 kHz	2 h	15 W/cm^2	US transducer[21]
Calcein [71]	623	*In vitro* pig	20 kHz	[§]	7.2 W/cm^2	Sonicator[1]
Calcein [72]	623	*In vitro* rat	41 kHz	[§]	60 – 300 mW/cm^2	US transducer[9]
Calcium [66]	40	*In vivo* rat	20 kHz	15/5 min[1a]	1 W/cm^2	Sonicator[1]
Corticosterone [65]	346	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic US[2]
Corticosterone [44]	346	*In vitro* human	1 – 3 MHz	24 h	125 mW/cm^2	Therapeutic US[5]
Corticosterone [65]	346	*In vitro* human	20 kHz	4 h	0 – 2 W/cm^2	Sonicator[1]
Dexamethasone [65]	392	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic US[2]
Dexamethasone [73]	392	*In vivo* human	1 MHz	10 min	1.0 W/cm^2	Therapeutic US[10]
Dextran [66][‡]	2000	*In vivo* rat	20 kHz	15/5 min[1a]	1 W/cm^2	Sonicator[1]

NB: Apologies are offered for any omitted or misinterpreted information. Readers are directed to the original publication for the most comprehensive source regarding the experimental methods.

Compound legend

*Tracer.

[‡]Extraction experiment; Fluorescein isothiocyanate (FITC, derivative of fluorescein)-labeled dextrans.

[§]Details unclear or not indicated.

Time, intensity and device legend

1. VCX 400, Sonics and Materials, Newtown, CT, USA; 1a. 15 min after 5 min; 1b. 15 min after 5 min; 2. Sonopuls 463, Henley International, Sugar Land, TX, USA; 3. Precision Acoustic Devices, Fremont, CA, USA and Panametrics, Waltham, MA, USA; 4. Leader Electronics Corporation, Yokohama, Japan; 5. Sonopuls 474, Henley International, Sugar Land, TX, USA; 6. W-385, Heat Systems Ultrasonics, Inc., Farmingdale, NY, USA; 7. Brand not indicated; 8. Cole Palmer Instrument, Chicago, IL, USA; 9. Transducer company not indicated; 10. Omnisound 3000, Accelerated Care Plus-Physio Technology, Topeka, KS, USA; 11. Sonics & Materials, Newtown, CT, USA; 12. ITO, Tokyo, Japan; 13. Dai-ichi High Frequency, Tokyo, Japan; 14. Model XL2020, Misonix, Farmingdale, NY, USA; 15. Pro Seven 977 to 2000 model, Quark Productos Médicos, Piracicaba, São Paulo, Brazil; 16. Noblelife™, Duplogen, Suwon, Korea; 17. Piezo Systems, Cambridge, MA, USA; 18. Cymbal, low profile ultrasound transducer, Penn State University, University Park, PA, USA; 19. Model S-110, Branson Instruments, Standford, CT, USA; 20. Sofranel, Zurich, Switzerland; 21. VCX 400, Sonics and Materials, Danbury, CT, USA; 22. Sonoplus 590, Enraf-Nonius BV, AV Delft, The Netherlands; 23. Peterson® 250 Ultrasound Equipment Petas, Turkey; 24. Pro Seven 977 to 2000 model, Quark Produtos Médicos, Piracicaba, São Paulo, Brazil; 25. SonoPrep, Sontra Medical, Cambridge, MA, USA.

CW: Continuous wave; I_{sptp}: Spatial peak temporal peak intensity; I_{spta}: Spatial peak temporal average intensity; I_{sapa}: Spatial peak pulse average intensity; I_{sata}: Spatial average temporal average intensity; US: Ultrasound.

Table 1. Broad list of compounds that have been delivered transdermally, and the corresponding frequency, exposure time and intensity of the ultrasound used (continued).

Compound and Ref.	MW	Preparation	Frequency	Time	Intensity	Device
Dextran [74]	70,000	*In vitro* pig	58 kHz	1 – 25 h	1.08 W/cm^2	US transducer[17]
Diclofenac [75]	296	*In vivo* human	1 MHz	5 min	0.5 W/cm^2 (I_{sata})	Therapeutic US[24]
Diclofenac [76]	296	*In vivo* rat	1 MHz	5 min	0.5 W/cm^2 (I_{sata})	Therapeutic US[12]
Erythropoeitin [58]	48,000	*In vitro* human, *in vivo* rat	20 kHz	0.5, 1, 2 h	12.5 ~ 225 mW/cm^2	Sonicator[1]
Estradiol [65]	272	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic US[2]
Estradiol [44]	272	*In vitro* human	1 – 3 MHz	4 h	0 – 2 W/cm^2	Therapeutic US[5]
Estradiol [45]	272	*In vitro* human	20 kHz	5 h	125 mW/cm^2	Sonicator[1]
FD-4 [72]¶	4400	*In vitro* rat	41 kHz	§	60 – 300 mW/cm^2	US transducer[9]
FD-40 [72]¶	38,000	*In vitro* rat	41 kHz	§	60 – 300 mW/cm^2	US transducer[9]
Fentanyl [67]	336	*In vitro* rat and human	20 kHz	10 min CW – 1 h pulsed	2.5 W/cm^2	§
Fluorescein [77]	389	*In vitro* human	20 kHz	1.5 – 30 min	8 – 24 mW/cm^2 (I_{spta})	Sonicator[14]
Fluorescein probes Nile red [70]	535	*In vitro* pig	20 kHz	2 h	15 W/cm^2	US transducer[21]
Glucose [66]	182	*In vivo* rat	20 kHz	15/5 min[1a] min	1 W/cm^2	Sonicator[1]
Glucose [78]‡	182	*In vitro* human	20 kHz	15/1 min[1b]	1 W/cm^2	Sonicator[1]
Glucose [47]	182	*in vitro* pig	10 MHz	2 h	0.2 – 2 W/cm^2	US transducer[20]
Glucose [47]	182	*in vitro* pig	20 kHz	2 h	10 – 70% of max	Sonicator[21]
Hyaluronan [79]	1000	*In vivo* rabbit	1 MHz	10 min	400 mW/cm^2	Therapeutic[16]
Hydrocortisone [80]	362	*In vivo* rat	1 MHz	5 min	0.5 W/cm^2 (I_{sata})	§
Ibuprofen [81]	206	*In vivo* human	1 MHz	5 min	1 W/cm^2	US transducer[23]

NB: Apologies are offered for any omitted or misinterpreted information. Readers are directed to the original publication for the most comprehensive source regarding the experimental methods.

Compound legend

*Tracer.

‡Extraction experiment; Fluorescein isothiocyanate (FITC, derivative of fluorescein)-labeled dextrans.

§Details unclear or not indicated.

Time, intensity and device legend

1. VCX 400, Sonics and Materials, Newtown, CT, USA; 1a. 15 min after 5 min; 1b. 15 min after 5 min; 2. Sonopuls 463, Henley International, Sugar Land, TX, USA; 3. Precision Acoustic Devices, Fremont, CA, USA and Panametrics, Waltham, MA, USA; 4. Leader Electronics Corporation, Yokohama, Japan; 5. Sonopuls 474, Henley International, Sugar Land, TX, USA; 6. W-385, Heat Systems Ultrasonics, Inc., Farmingdale, NY, USA; 7. Brand not indicated; 8. Cole Palmer Instrument, Chicago, IL, USA; 9. Transducer company not indicated; 10. Omnisound 3000, Accelerated Care Plus-Physio Technology, Topeka, KS, USA; 11. Sonics & Materials, Newtown, CT, USA; 12. ITO, Tokyo, Japan; 13. Dai-ichi High Frequency, Tokyo, Japan; 14. Model XL2020, Misonix, Farmingdale, NY, USA; 15. Pro Seven 977 to 2000 model, Quark Productos Médicos, Piracicaba, São Paulo, Brazil; 16. Noblelife™, Duplogen, Suwon, Korea; 17. Piezo Systems, Cambridge, MA, USA; 18. Cymbal, low profile ultrasound transducer, Penn State University, University Park, PA, USA; 19. Model S-110, Branson Instruments, Standford, CT, USA; 20. Sofranel, Zurich, Switzerland; 21. VCX 400, Sonics and Materials, Danbury, CT, USA; 22. Sonoplus 590, Enraf-Nonius BV, AV Delft, The Netherlands; 23. Peterson® 250 Ultrasound Equipment Petas, Turkey; 24. Pro Seven 977 to 2000 model, Quark Produtos Médicos, Piracicaba, São Paulo, Brazil; 25. SonoPrep, Sontra Medical, Cambridge, MA, USA.

CW: Continuous wave; I_{sptp}: Spatial peak temporal peak intensity; I_{spta}: Spatial peak temporal average intensity; I_{sapa}: Spatial peak pulse average intensity; I_{sata}: Spatial average temporal average intensity; US: Ultrasound.

Table 1. Broad list of compounds that have been delivered transdermally, and the corresponding frequency, exposure time and intensity of the ultrasound used (continued).

Compound and Ref.	MW	Preparation	Frequency	Time	Intensity	Device
Insulin [58]	5807	*In vitro* human, *in vivo* rat	20 kHz	0.5, 1, 2 h	12.5 ~ 225 mW/cm^2	Sonicator[1]
Insulin [82]	5807	*In vivo* rat	20 kHz	15, 60 min	2.5, 5, 10 W/cm^2	Sonicator[1]
Insulin [1]	5807	*In vivo* rat	48 kHz	90 min	0.61 – 4.32 mW/cm^2	LAG-26[4]
Insulin [49]	5807	*In vivo* rabbit	105 kHz	90 min	1.69 mW/cm^2	LAG-26[4]
Insulin [83]	5807	*In vitro* human	20 kHz	2, 5 h	0.1 ~ 1 W/cm^2	Sonicator[6]
Insulin [84]	5807	*In vivo* rat	20 kHz	15, 60 min	2.5, 5, 10 W/cm^2	Sonicator[1]
Insulin (Humulin® and Humalog®) [85]	5807	*In vitro* human	20 kHz	1 h	173 mW/cm^2 (I_{sptp})	US transducer[18]
Insulin-Humulin® [86]	5807	*In vivo* rabbit	20 kHz	1 h	100 mW/cm^2 (I_{sptp})	US transducer[18]
Insulin-Humulin® [86]	5807	*In vivo* rabbit	20 kHz	1 h	100 mW/cm^2 (I_{sptp})	US transducer[18]
Insulin-Humulin® [62]	5807	*In vivo* pig	20 kHz	1 h	100 mW/cm^2 (I_{sptp})	US transducer[18]
Insulin-Humulin® [87]	5807	*In vivo* rabbit	20 kHz	1 h	50 mW/cm^2 (I_{sptp})	US transducer[18]
Insulin [88]	5000	*In vivo* rat	20 kHz	2 min	7 W/cm^2 (I_{sapa})	Sonicator[1]
Insulin [74]	5000	*In vitro* pig	58 kHz	1 min	1.6 – 14 W/cm^2	US transducer[17]
Ketoprofen [89]	254	*In vivo* human	1 MHz	5 min	1.5 W/cm^2	Sonicator[22]
Ketorolac-tromethamine [90]	376	*In vitro* rat	1 MHz	30 min	3 W/cm^2	Sonicator[7]
Lanthanum hydroxide [91]*	189	*In vivo* guinea-pigs	2, 10, 16 MHz	5, 10 min	0.2 W/cm^2	Panametrics[3]
Lidocaine [65]	234	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic US[2]
Lidocaine and prilocaine mix [92]	234/220	*In vivo* human	55 kHz	10 sec	§	SonoPrep[25]

NB: Apologies are offered for any omitted or misinterpreted information. Readers are directed to the original publication for the most comprehensive source regarding the experimental methods.

Compound legend

*Tracer.

‡Extraction experiment; Fluorescein isothiocyanate (FITC, derivative of fluorescein)-labeled dextrans.

§Details unclear or not indicated.

Time, intensity and device legend

1. VCX 400, Sonics and Materials, Newtown, CT, USA; 1a. 15 min after 5 min; 1b. 15 min after 5 min; 2. Sonopuls 463, Henley International, Sugar Land, TX, USA; 3. Precision Acoustic Devices, Fremont, CA, USA and Panametrics, Waltham, MA, USA; 4. Leader Electronics Corporation, Yokohama, Japan; 5. Sonopuls 474, Henley International, Sugar Land, TX, USA; 6. W-385, Heat Systems Ultrasonics, Inc., Farmingdale, NY, USA; 7. Brand not indicated; 8. Cole Palmer Instrument, Chicago, IL, USA; 9. Transducer company not indicated; 10. Omnisound 3000, Accelerated Care Plus-Physio Technology, Topeka, KS, USA; 11. Sonics & Materials, Newtown, CT, USA; 12. ITO, Tokyo, Japan; 13. Dai-ichi High Frequency, Tokyo, Japan; 14. Model XL2020, Misonix, Farmingdale, NY, USA; 15. Pro Seven 977 to 2000 model, Quark Productos Médicos, Piracicaba, São Paulo, Brazil; 16. Noblelife™, Duplogen, Suwon, Korea; 17. Piezo Systems, Cambridge, MA, USA; 18. Cymbal, low profile ultrasound transducer, Penn State University, University Park, PA, USA; 19. Model S-110, Branson Instruments, Standford, CT, USA; 20. Sofranel, Zurich, Switzerland; 21. VCX 400, Sonics and Materials, Danbury, CT, USA; 22. Sonoplus 590, Enraf-Nonius BV, AV Delft, The Netherlands; 23. Peterson® 250 Ultrasound Equipment Petas, Turkey; 24. Pro Seven 977 to 2000 model, Quark Produtos Médicos, Piracicaba, São Paulo, Brazil; 25. SonoPrep, Sontra Medical, Cambridge, MA, USA.

CW: Continuous wave; I_{sptp}: Spatial peak temporal peak intensity; I_{spta}: Spatial peak temporal average intensity; I_{sapa}: Spatial peak pulse average intensity; I_{sata}: Spatial average temporal average intensity; US: Ultrasound.

Table 1. Broad list of compounds that have been delivered transdermally, and the corresponding frequency, exposure time and intensity of the ultrasound used (continued).

Compound and Ref.	MW	Preparation	Frequency	Time	Intensity	Device
Linoleic acid [65]	280	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic US[2]
Luteinizing hormone release hormone [74]	1311	*In vitro* pig	58 kHz	1 – 25 h	1.08 W/cm^2	US transducer[17]
Mannitol [88]	183	*In vivo* rat	20 kHz	2 min	7 W/cm^2 (I_{sapa})	Sonicator[1]
Mannitol [93]	183	*In vitro* pig	20 kHz	15 – 60 min	1.6 ~ 14 W/cm^2	Sonicator[1]
Mannitol [94]	183	*In vitro* pig, *in vitro* human	20 kHz	Pig/human 20 h human 9 h	1.6 W/cm^2	Sonicator[1]
Mannitol [74]	182	*In vitro* pig	58 kHz	1 min	1.6 – 14 W/cm^2	US transducer[17]
Mannitol [47]	183	*In vitro* pig	10 MHz	2 h	0.2 – 2 W/cm^2	US transducer[20]
Mannitol [47]	183	*In vitro* pig	20 kHz	2 h	10 – 70% of max	Sonicator[21]
Methylpredni-solone/ciclosporin [95]	374	*In vivo* human	25 kHz	10 min	50 – 100 mW/cm^2	Sonicator[7]
Octa l-lysine-fluorescein isothiocyanate [96]	2500	*In vitro* human	20 kHz	10 min	2 – 50 W/cm^2 (I_{sapa})	Sonifier[19]
Poly l-lysine-fluorescein isothiocyanate [96]	51,000	*In vitro* human	20 kHz	15 min	19 W/cm^2 (I_{sapa})	Sonifier[19]
Progesterone [44]	274	*In vitro* human	1 – 3 MHz	4 h	0 – 2 W/cm^2	Therapeutic US[5]
Salicylic acid [65]	138	*In vitro* human	20 kHz	24 h	125 mW/cm^2	Sonicator[1]
Sodium lauryl sulfate [46]	288	*In vitro* pig	19.6, 36.9, 58.9, 76.6, 93.4 kHz	10 – 15 min	0.2 – 2.7 W/cm^2	US transducer[17]
Sodium lauryl sulfate [97]	288	*In vitro* pig	20 kHz	3 min	2.4 W/cm^2	Sonicator[11]
Sucrose [65]	342	*In vitro* human	20 kHz	24 h	125 mW/cm^2	Sonicator[1]

NB: Apologies are offered for any omitted or misinterpreted information. Readers are directed to the original publication for the most comprehensive source regarding the experimental methods.

Compound legend

*Tracer.

‡Extraction experiment; Fluorescein isothiocyanate (FITC, derivative of fluorescein)-labeled dextrans.

§Details unclear or not indicated.

Time, intensity and device legend

1. VCX 400, Sonics and Materials, Newtown, CT, USA; 1a. 15 min after 5 min; 1b. 15 min after 5 min; 2. Sonopuls 463, Henley International, Sugar Land, TX, USA; 3. Precision Acoustic Devices, Fremont, CA, USA and Panametrics, Waltham, MA, USA; 4. Leader Electronics Corporation, Yokohama, Japan; 5. Sonopuls 474, Henley International, Sugar Land, TX, USA; 6. W-385, Heat Systems Ultrasonics, Inc., Farmingdale, NY, USA; 7. Brand not indicated; 8. Cole Palmer Instrument, Chicago, IL, USA; 9. Transducer company not indicated; 10. Omnisound 3000, Accelerated Care Plus-Physio Technology, Topeka, KS, USA; 11. Sonics & Materials, Newtown, CT, USA; 12. ITO, Tokyo, Japan; 13. Dai-ichi High Frequency, Tokyo, Japan; 14. Model XL2020, Misonix, Farmingdale, NY, USA; 15. Pro Seven 977 to 2000 model, Quark Productos Médicos, Piracicaba, São Paulo, Brazil; 16. Noblelife™, Duplogen, Suwon, Korea; 17. Piezo Systems, Cambridge, MA, USA; 18. Cymbal, low profile ultrasound transducer, Penn State University, University Park, PA, USA; 19. Model S-110, Branson Instruments, Standford, CT, USA; 20. Sofranel, Zurich, Switzerland; 21. VCX 400, Sonics and Materials, Danbury, CT, USA; 22. Sonoplus 590, Enraf-Nonius BV, AV Delft, The Netherlands; 23. Peterson® 250 Ultrasound Equipment Petas, Turkey; 24. Pro Seven 977 to 2000 model, Quark Produtos Médicos, Piracicaba, São Paulo, Brazil; 25. SonoPrep, Sontra Medical, Cambridge, MA, USA.

CW: Continuous wave; I_{sptp}: Spatial peak temporal peak intensity; I_{spta}: Spatial peak temporal average intensity; I_{sapa}: Spatial peak pulse average intensity; I_{sata}: Spatial average temporal average intensity; US: Ultrasound.

Table 1. Broad list of compounds that have been delivered transdermally, and the corresponding frequency, exposure time and intensity of the ultrasound used (continued).

Compound and Ref.	MW	Preparation	Frequency	Time	Intensity	Device
Sucrose [94]	342	*In vitro* human *in vitro* pig	20 kHz	Pig/human 20 h human 9 h	1.6 W/cm^2	US transducer[1]
Testosterone [65]	288	*In vitro* human	1 MHz	24 h	1.4 W/cm^2	Therapeutic[2]
Tetanus toxoid (TTx vaccine) [98]	150,000	*In vivo* mice	20 kHz	40 s increments	2.4 W/cm^2	600 W Sonicator[11]
Triamcinolone-Acetonide [99]	434	*In vitro* mice	1, 3 MHz	10 h	1, 2.5 W/cm^2	UStransducer[9]
Urea [100]	60	*In vivo* rat	20 kHz	15 min after 5 min	1 W/cm^2	Sonicator[1]
Vasopressin [83]	1056	*In vitro* human	20 kHz	2 or 5 h	0.1 – 1 W/cm^2	Sonicator[6]
Water [65]	18	*In vitro* human	20 kHz	24 h	125 mW/cm^2	Sonicator[1]

NB: Apologies are offered for any omitted or misinterpreted information. Readers are directed to the original publication for the most comprehensive source regarding the experimental methods.

Compound legend

*Tracer.

‡Extraction experiment; Fluorescein isothiocyanate (FITC, derivative of fluorescein)-labeled dextrans.

§Details unclear or not indicated.

Time, intensity and device legend

1. VCX 400, Sonics and Materials, Newtown, CT, USA; 1a. 15 min after 5 min; 1b. 15 min after 5 min; 2. Sonopuls 463, Henley International, Sugar Land, TX, USA; 3. Precision Acoustic Devices, Fremont, CA, USA and Panametrics, Waltham, MA, USA; 4. Leader Electronics Corporation, Yokohama, Japan; 5. Sonopuls 474, Henley International, Sugar Land, TX, USA; 6. W-385, Heat Systems Ultrasonics, Inc., Farmingdale, NY, USA; 7. Brand not indicated; 8. Cole Palmer Instrument, Chicago, IL, USA; 9. Transducer company not indicated; 10. Omnisound 3000, Accelerated Care Plus-Physio Technology, Topeka, KS, USA; 11. Sonics & Materials, Newtown, CT, USA; 12. ITO, Tokyo, Japan; 13. Dai-ichi High Frequency, Tokyo, Japan; 14. Model XL2020, Misonix, Farmingdale, NY, USA; 15. Pro Seven 977 to 2000 model, Quark Productos Médicos, Piracicaba, São Paulo, Brazil; 16. Noblelife™, Duplogen, Suwon, Korea; 17. Piezo Systems, Cambridge, MA, USA; 18. Cymbal, low profile ultrasound transducer, Penn State University, University Park, PA, USA; 19. Model S-110, Branson Instruments, Standford, CT, USA; 20. Sofranel, Zurich, Switzerland; 21. VCX 400, Sonics and Materials, Danbury, CT, USA; 22. Sonoplus 590, Enraf-Nonius BV, AV Delft, The Netherlands; 23. Peterson® 250 Ultrasound Equipment Petas, Turkey; 24. Pro Seven 977 to 2000 model, Quark Produtos Médicos, Piracicaba, São Paulo, Brazil; 25. SonoPrep, Sontra Medical, Cambridge, MA, USA.

CW: Continuous wave; I_{sptp}: Spatial peak temporal peak intensity; I_{spta}: Spatial peak temporal average intensity; I_{sapa}: Spatial peak pulse average intensity; I_{sata}: Spatial average temporal average intensity; US: Ultrasound.

were immersed in a commercial ultrasound water tank at 48 kHz and an intensity of 0.5 W/cm^2. Skins were compared to control skins under a scanning electron microscope and it was found that the cells of the *stratum corneum* of the mouse skin surface were almost completely removed. Also, in the mouse skin, large craterlike pores with a diameter of 100 microns were formed sporadically in some of the skin samples. However, in human skin the surface of skin exposed to ultrasound showed only slight removal of keratinocytes around the hair follicles [56]. While much of the drug delivery research looks at efficacy, bioeffects from the ultrasound deserve the same attention.

5. Conclusion

Research into ultrasound-based technology for transdermal drug delivery is growing. The most immediate need is probably for diabetes in either insulin delivery or glucose monitoring. While ultrasound-mediated insulin delivery and glucose sensing has been shown (**Table 1**), there is no guarantee that a sustainable market exists. Needle-less insulin was initially made available via an inhalable form using EXUBERA® (Pfizer, New York, NY, USA) in late 2006. However, side effects included decreased lung function and the delivery device was described as large and inconvenient. These are probably the main reasons as to why the device has been taken off the market [57]. Research is ongoing to develop a smaller inhaler device. Overall, the ultrasound drug delivery technology should look closely at this example regarding safety and device size.

Insofar as ultrasound is known to increase transdermal protein delivery [58], the mechanisms of this enhanced transport have not yet been fully characterized. One effect of ultrasound is to increase permeability of the outer skin layer (*stratum corneum*), which is considered to be a primary barrier to protein diffusion. However, depending on the distance from the transducer to the skin and the penetration depth, consequences of ultrasound administration could extend from the *stratum corneum* several millimeters into the underlying tissue.

The consequences of prolonged ultrasound administration to tissue below the *stratum corneum* can be viewed as either potentially beneficial or potentially harmful. With respect to the latter, many researchers suggest that further investigation is required, despite early preliminary indications that short-term, low-frequency ultrasound causes no histological abnormalities in the skin or underlying muscle. More researchers in the field are recognizing the need to understand the mechanism of ultrasound and the need for safety in its use for drug delivery [2,59].

6. Expert opinion on transdermal ultrasound drug delivery

Beyond the diagnostic imaging, researchers have harnessed the unique capabilities of ultrasound for controlled tissue destruction in the form of high intensity focused ultrasound. Through careful research, clinical application of focused ultrasound has been used, for example, in the treatment of uterine fibroids with the ExAblate® 2000 System (GE Healthcare, Milwaukee, WI, USA and InSightec, Tirat Carmel, Israel). In conjunction with focused ultrasound, magnetic resonance imaging derived thermometry provides an accurate and non-invasive method to destroy diseased tissue while leaving healthy tissue intact.

In order to achieve the same utility as diagnostic ultrasound imaging and focused ultrasound surgery, the use of ultrasound for transdermal drug delivery must follow the same careful scientific methods to become an accepted clinical practice. However, for a number of the transdermal drug delivery experiments in **Table 1**, it is difficult to duplicate the acoustic conditions based on the information given. Although many of the papers report an ultrasound intensity, the drawback is that a value alone without any spatial or temporal information does not enable repeatable experimental procedures. Low frequency experiments confirm the acoustic spatial variability for effective delivery [60]. This spatial effect can also be seen in the computed pressure field in **Figure 2B**. A mere 2.75 cm lateral of the last axial maximum would reduce the pressure field by half or -3 dB. This might explain the wide range in reported intensities, which varies by orders of magnitude in **Table 1** for the same drug. Additionally, while research often considers the intensity within a few millimeters from the transducer face or source, there is substantial acoustic energy transmitting several centimeters into the tissue, which can also contribute to bioeffects. Therefore, a limited understanding of the acoustic field can lead to a flawed description of the actual intensity. While the skin is permeabilized for drug delivery, there might also be deeper tissue damage or heating at a bone/muscle interface.

A previous opinion stated that 'small-sized low-frequency transducers need to be developed so that patients can wear them' [3]. While some researchers are working on these small, flat transducers, the technology is still in the research stage and is not yet a commercial product for drug delivery [61,62]. Some companies have developed larger ultrasound devices for effective pre-treatment and drug delivery such as the SonoPrep® (Sontra Medical, Cambridge, MA, USA). While the literature shows that ultrasound can deliver drugs like insulin across the skin, this is not yet sufficient to replace insulin needles, inject pens or pumps. Some diabetics require a large, quick dose (bolus) to counteract a glucose spike, which current research does not explore in detail. Raising the intensity to increase the dose might not be the best approach, given the potential for damage. One possibility might be to maintain a low intensity but increase the area of skin exposed to ultrasound for a larger dose delivery [63]. There is also the need for more longitudinal studies on animals, given the need for daily drug administration in humans.

Those who use ultrasound equipment for drug delivery should be concerned about bioeffects. Journals that publish research in this area should recruit reviewers with a detailed knowledge of acoustics and ultrasound. While the future for non-invasive drug delivery is encouraging, exploiting transdermal ultrasound drug delivery beyond the feasibility stage will require the cooperation of physicians and engineers. Physicians are needed to define dose guidelines (how often, how much) and engineers are required so that the technology is both safe and clinically practical. The diagnostic ultrasound community routinely assesses the risk–benefit of the current technology [12]. This does not represent a call for intensity limits (as with imaging ultrasound), but cautions that those who use ultrasound equipment should be concerned about bioeffects. This technology has the potential to be either as successful as the nicotine patch [4,64] or fall by the wayside like EXUBERA® [57]. As with diagnostic ultrasound, the bioeffects and safety of each device need to be carefully evaluated, since it will not matter how much of any drug can be transported if the skin is irreversibly damaged.

Acknowledgement

This work was supported by the Department of Defense Technologies for Metabolic Monitoring Award Number W81XWH-05-1-0617.

Declaration of interest

The author states no conflict of interest and has received no payment in preparation of this manuscript.

Bibliography

1. Tachibana K, Tachibana S. Transdermal delivery of insulin by ultrasonic vibration. J Pharm Pharmacol 1991;43:270-1
2. Lavon I, Kost J. Ultrasound and transdermal drug delivery. Drug Discov Today 2004;9:670-6
3. Pitt WG, Husseini GA, Staples BJ. Ultrasonic drug delivery – a general review. Expert Opin Drug Deliv 2004;1:37-56
4. Prausnitz MR, Mitragotri S, Langer R. Current status and future potential of transdermal drug delivery. Nat Rev Drug Discov 2004;3:115-24
5. Nanda A, Nanda S, Ghilzai NM. Current developments using emerging transdermal technologies in physical enhancement methods. Curr Drug Deliv 2006;3:233-42
6. Brown MB, Traynor MJ, Martin GP, Akomeah FK. Transdermal drug delivery systems: skin perturbation devices. Methods Mol Biol 2008;437:119-39
7. Kumar MG, Lin S. Transdermal iontophoresis: impact on skin integrity as evaluated by various methods. Crit Rev Ther Drug Carrier Syst 2008;25:381-401
8. Hui SW. Overview of drug delivery and alternative methods to electroporation. Methods Mol Biol 2008;423:91-107
9. Vandervoort J, Ludwig A. Microneedles for transdermal drug delivery: a minireview. Front Biosci 2008;13:1711-5
10. Smith EI, Maibach HI. Percutaneous penetration enhancers. Boca Raton, FL: CRC Press; 1995
11. Dalecki D. Mechanical bioeffects of ultrasound. Ann Rev Biomed Eng 2004;6:229-48
12. Fowlkes JB. American Institute of Ultrasound in Medicine Consensus Report on Potential Bioeffects of Diagnostic Ultrasound: Executive Summary. J Ultrasound Med 2008;27:503-15
13. Hynynen K. Ultrasound for drug and gene delivery to the brain. Adv Drug Deliv Rev 2008;60:1209-17
14. Mitragotri S, Kost J. Low-frequency sonophoresis: a review. Adv Drug Deliv Rev 2004;56:589-601
15. Smith NB. Perspectives on transdermal ultrasound mediated drug delivery. Int J Nanomed 2007;2:585-94
16. Apfel RE. Acoustic cavitation: a possible consequence of biomedical use of ultrasound. Br J Cancer 1982;45:140-6
17. Wu J, Nyborg WL. Ultrasound, cavitation bubbles and their interaction with cells. Adv Drug Deliv Rev 2008;60:1103-16
18. Edmonds PD, Sancier KM. Evidence for free radical production by ultrasonic cavitation in biological media. Ultrasound Med Biol 1983;9:635-9
19. Mason TJ, Lorimer JP. Sonochemistry: theory, applications and uses of ultrasound in chemistry. West Sussex, UK: Ellis Horwood Limited; 1988
20. Hynynen KH. The threshold for thermally significant cavitation in dog's thigh muscle in vivo. Ultrasound Med Biol 1991;17:157-69
21. Dalecki D, Raeman CH, Child SZ, Carstensen EL. A test for cavitation as a mechanism for intestinal hemorrhage in mice exposed to a piezoelectric lithotripter. Ultrasound Med Biol 1996;22:493-6
22. Miller MW, Miller DL, Brayman AA. A review of in vitro bioeffects of intertial ultrasonic cavitation from a mechanistic perspective. 22nd edition. 1996. p. 1131-54
23. AIUM. Mechanical bioeffects from diagnostic ultrasound: AIUM consensus statements. J Ultrasound Med 2000;19:68-168
24. Apfel RE. Possibility of microcavitation from diagnostic ultrasound. IEEE Trans Ultrason Ferroelectr Freq Contr 1986;33:139-42
25. Roy RA, Madanshetty SI, Apfel RE. An acoustic backscattering technique for the detection of cavitation produced by microsecond pulses of ultrasound. J Acoust Soc Am 1990;87:2451-8
26. Laurel MD. AIUM-NEMA. Standard for real-time display of thermal and mechanical acoustic output indices on diagnostic ultrasound equipment: American Institute of Ultrasound in Medicine National Electrical Manufacturers Association; 1996
27. Stratmeyer ME, Greenleaf JF, Dalecki D, Salvesen KA. Fetal ultrasound: mechanical effects. J Ultrasound Med 2008;27:597-605
28. Beyer RT. Nonlinear acoustics. Sewickley, PA: Acoustical Society of America Publications; 1997
29. Apfel RE. Acoustic Cavitation. In: Edmonds PD, editor, Methods of experimental physics. New York: Academic Press; 1981. p. 356-411
30. Hamilton M, Blackstock D. Nonlinear acoustics. San Diego, CA: Academic Press; 1998

31. Lewin P, Ziskin M. Ultrasonic exposimetry. Boca Raton, FL: CRC Press; 1992
32. Everbach EC, Makin IRS, Azadniv M, Meltzer RS. Correlation of ultrasound-induced hemolysis with cavitation detector output in vitro. Ultrasound Med Biol 1997;23:619-24
33. Cleveland RO, Sapozhnikov OA, Crum LA. A dual passive cavitation detector for localized detection of lithotripsy-induced cavitation in vitro. J Acoust Soc Am 2000;107:1745-58
34. Hynynen KH. The threshold for thermally significant cavitation in dog's thigh muscle in vivo. Ultrasound Med Biol 1990;17:157-69
35. Jochle K, Debus J, Lorenz WJ, Huber P. A new method of quantitative cavitation assessment in the field of a lithotripter. Ultrasound Med Biol 1996;22:329-38
36. Huber P, Debus J, Peschke P, et al. In vivo detection of ultrasonically induced cavitation by a fiber-optic technique. Ultrasound Med Biol 1994;20:811-25
37. Takahashi S, Kikuchi T, Hasegawa A, Murakami Y. Cavitation Noise Measurement Using a Fiber-optic Hydrophone. J Acoust Soc Am 1990;87:2489-92
38. Kinsler LE, Frey AR, Coppens AB, Sanders JV. Fundamentals of acoustics. 3rd edition. New York: John Wiley and Sons; 1982
39. Johns LD, Straub SJ, Howard SM. Variability in effective radiating area and output power of new ultrasound transducers at 3 MHz. J Athl Train 2007;42:22-8
40. IEEE. Guide for medical ultrasound field parameter measurements. New York: Institute of Electrical and Electronics Engineers, Inc.; 1990
41. Egawa M, Hirao T, Takahashi M. In vivo estimation of stratum corneum thickness from water concentration profiles obtained with Raman spectroscopy. Acta Derm Venereol 2007;87:4-8
42. Boucaud A. Trends in the use of ultrasound-mediated transdermal drug delivery. Drug Discov Today 2004;9:827-8
43. Brown MB, Martin GP, Jones SA, Akomeah FK. Dermal and transdermal drug delivery systems: current and future prospects. Drug Deliv 2006;13:175-87
44. Mitragotri S, Edwards DA, Blankschtein D, Langer R. A mechanistic study of ultrasonically-enhanced transdermal drug delivery. J Pharm Sci 1995;84:697-706
45. Mitragotri S, Blankschtein D, Langer R. Transdermal drug delivery using low-frequency sonophoresis. Pharm Res 1996;13:411-20
46. Tezel A, Sens A, Tuchscherer J, Mitragotri S. Frequency dependence of sonophoresis. Pharm Res 2001;18:1694-700
47. Merino G, Kalia YN, Gado Charro MB, et al. Frequency and thermal effects on the enhancement of transdermal transport by sonophoresis. J Control Release 2003;88:85-94
48. Centers for Disease Control and Prevention. 2007 National Diabetes Fact Sheet. Phoenix: CDC Division of Diabetes Translation Public Inquiries/Publications; 2008
49. Tachibana K. Transdermal delivery of insulin to alloxan-diabetic rabbits by ultrasound exposure. Pharm Res 1992;9:952-4
50. Dixit N, Bali V, Baboota S, et al. Iontophoresis – an approach for controlled drug delivery: a review. Curr Drug Deliv 2007;4:1-10
51. Murthy SN, Zhao YL, Marlan K, et al. Lipid and electroosmosis enhanced transdermal delivery of insulin by electroporation. J Pharm Sci 2006;95:2041-50
52. Roxhed N, Samel B, Nordquist L, et al. Painless drug delivery through microneedle-based transdermal patches featuring active infusion. IEEE Trans Biomed Eng 2008;55:1063-71
53. Abramowicz JS, Barnett SB, Duck FA, et al. Fetal thermal effects of diagnostic ultrasound. J Ultrasound Med 2008;27:541-59
54. Boucaud A, Montharu J, Machet L, et al. Clinical, histologic, and electron microscopy study of skin exposed to low-frequency ultrasound. Anat Rec 2001;264:114-9
55. Wu J, Chappelow J, Yang J, Weimann L. Defects generated in human stratum corneum specimens by ultrasound. Ultrasound Med Biol 1998;24:705-10
56. Yamashita N, Tachibana K, Ogawa K, et al. Scanning electron microscopic evaluation of the skin surface after ultrasound exposure. Anat Rec 1997;247:455-61
57. Mack GS. Pfizer dumps Exubera. Nat Biotechnol 2007;25:1331-2
58. Mitragotri S, Blankschtein D, Langer R. Ultrasound-mediated transdermal protein delivery. Science 1995;269:850-3
59. Tachibana K, Feril LB Jr, Ikeda-Dantsuji Y. Sonodynamic therapy. Ultrasonics 2008;48(4):253-9
60. Terahara T, Mitragotri S, Kost J, Langer R. Dependence of low-frequency sonophoresis on ultrasound parameters; distance of the horn and intensity. Int J Pharm 2002;235:35-42
61. Newnham RE, Dogan A. Metal-electroactive ceramic composite transducer.5,729,077. 17 Mar 1998
62. Park EJ, Werner J, Smith NB. Ultrasound mediated transdermal insulin delivery in pigs using a lightweight transducer. Pharm Res 2007;24:1396-401
63. Luis J, Smith NB, Meyer RJ. Rectangular cymbal arrays for improved ultrasonic transdermal insulin delivery. J Acoust Soc Am 2007;122:2022-30
64. Stead LF, Perera R, Bullen C, et al. Nicotine replacement therapy for smoking cessation. Cochrane Database Syst Rev 2008;CD000146
65. Johnson ME, Mitragotri S, Patel A, et al. Synergistic effects of chemical enhancers and therapeutic ultrasound on transdermal drug delivery. J Pharm Sci 1996;85:670-9
66. Mitragotri S, Coleman M, Kost J, Langer R. Transdermal extraction of analytes using low-frequency ultrasound. Pharm Res 2000;17:466-70
67. Boucaud A, Machet L, Arbeille B, et al. In vitro study of low-frequency ultrasound-enhanced transdermal transport of fentanyl and caffeine across human and hairless rat skin. Int J Pharm 2001;228:69-77
68. Mutoh M, Ueda H, Nakamura Y, et al. Characterization of transdermal solute transport induced by low-frequency ultrasound in the hairless rat skin. J Control Release 2003;92:137-46
69. Sundaram J, Mellein BR, Mitragotri S. An experimental and theoretical analysis of ultrasound-induced permeabilization of cell membranes. Biophys J 2003;84:3087-101
70. Varez Roman R, Merino G, Kalia YN, et al. Skin permeability enhancement by low frequency sonophoresis: lipid extraction and transport pathways. J Pharm Sci 2003;92:1138-46
71. Kushner J, Blankschtein D, Langer R. Experimental demonstration of the

existence of highly permeable localized transport regions in low-frequency sonophoresis. J Pharm Sci 2004;93:2733-45

72. Morimoto Y, Mutoh M, Ueda H, et al. Elucidation of the transport pathway in hairless rat skin enhanced by low-frequency sonophoresis based on the solute-water transport relationship and confocal microscopy. J Control Release 2005;103:587-97
73. Darrow H, Schulthies S, Draper D, et al. Serum Dexamethasone Levels After Decadron Phonophoresis. J Athl Train 1999;34:338-41
74. Tezel A, Sens A, Mitragotri S. Description of transdermal transport of hydrophilic solutes during low-frequency sonophoresis based on a modified porous pathway model. J Pharm Sci 2003;92:381-93
75. Rosim GC, Barbieri CH, Lancas FM, Mazzer N. Diclofenac phonophoresis in human volunteers. Ultrasound Med Biol 2005;31:337-43
76. Hsieh YL. Effects of ultrasound and diclofenac phonophoresis on inflammatory pain relief: suppression of inducible nitric oxide synthase in arthritic rats. Phys Ther 2006;86:39-49
77. Cancel LM, Tarbell JM, Ben Jebria A. Fluorescein permeability and electrical resistance of human skin during low frequency ultrasound application. J Pharm Pharmacol 2004;56:1109-18
78. Kost J, Pliquett U, Mitragotri S, et al. Synergistic effect of electric field and ultrasound on transdermal transport. Pharm Res 1996;13:633-8
79. Park SR, Jang KW, Park SH, et al. The effect of sonication on simulated osteoarthritis. Part I. Effects of 1 MHz ultrasound on uptake of hyaluronan into the rabbit synovium. Ultrasound Med Biol 2005;31:1551-8
80. Koeke PU, Parizotto NA, Carrinho PM, Salate AC. Comparative study of the efficacy of the topical application of hydrocortisone, therapeutic ultrasound and phonophoresis on the tissue repair process in rat tendons. Ultrasound Med Biol 2005;31:345-50
81. Kozanoglu E, Basaran S, Guzel R, Guler Uysal F. Short term efficacy of ibuprofen phonophoresis versus continuous ultrasound therapy in knee osteoarthritis. Swiss Med Wkly 2003;133:333-8
82. Boucaud A, Tessier L, Machet L, et al. Transdermal delivery of insulin using low frequency ultrasound. Proceedings of the IEEE 2000 Ultrasonics Symposium; November 2000; San Juan Porto Rico; 2000. p. 1453-6
83. Zhang I, Shung KK, Edwards DA. Hydrogels with enhanced mass transfer for transdermal drug delivery. J Pharm Sci 1996;85:1312-6
84. Boucaud A, Garrigue MA, Machet L, et al. Effect of sonication parameters on transdermal delivery of insulin to hairless rats. J Control Release 2002;81:113-9
85. Smith NB, Lee S, Maione E, et al. Ultrasound mediated transdermal transport of insulin through in vitro human skin using novel transducer designs. Ultrasound Med Biol 2003;29:311-7
86. Lee S, Snyder B, Newnham RE, Smith NB. Non-invasive ultrasonic transdermal insulin delivery in rabbits using the light-weight cymbal array. Diabetes Technol Ther 2004;6:808-15
87. Luis J, Smith NB, Meyer RJ. Rectangular cymbal arrays for improved ultrasonic transdermal insulin delivery. J Acoust Soc Am 2007;122:2022-30
88. Mitragotri S, Kost J. Low-frequency sonophoresis: a non-invasive method of drug delivery and diagnostics. Biotechnol Prog 2000;16:488-92
89. Cagnie B, Vinck E, Rimbaut S, Vanderstraeten G. Phonophoresis versus topical application of ketoprofen: comparison between tissue and plasma levels. Phys Ther 2003;83:707-12
90. Tiwari SB, Pai RM, Udupa N. Influence of ultrasound on the percutaneous absorption of ketorolac tromethamine in vitro across rat skin. Drug Deliv 2004;11:47-51
91. Bommannan D, Menon GK, Okuyama H, et al. Sonophoresis. II. Examination of the mechanism(s) of ultrasound-enhanced transdermal drug delivery. Pharm Res 1992;9:1043-7
92. Katz NP, Shapiro DE, Herrmann TE, et al. Rapid onset of cutaneous anesthesia with EMLA cream after pretreatment with a new ultrasound-emitting device. Anesth Analg 2004;98:371-6, table
93. Mitragotri S, Farrell J, Tang T, et al. Determination of the threshold energy dose for ultrasound-induced transdermal drug transport. J Control Release 2000;63:41-52
94. Tang H, Mitragotri S, Blankschtein D, Langer R. Theoretical description of transdermal transport of hydrophilic permeants: application to low-frequency sonophoresis. J Pharm Sci 2001;90:545-68
95. Santoianni P, Nino M, Calabro G. Intradermal drug delivery by low-frequency sonophoresis (25 kHz). Dermatol Online J 2004;10:24
96. Weimann LJ, Wu J. Transdermal delivery of poly-l-lysine by sonomacroporation. Ultrasound Med Biol 2002;28:1173-80
97. Paliwal S, Menon GK, Mitragotri S. Low-frequency sonophoresis: ultrastructural basis for stratum corneum permeability assessed using quantum dots. J Invest Dermatol 2006;126:1095-101
98. Tezel A, Paliwal S, Shen Z, Mitragotri S. Low-frequency ultrasound as a transcutaneous immunization adjuvant. Vaccine 2005;23:3800-7
99. Yang JH, Kim DK, Yun MY, et al. Transdermal delivery system of triamcinolone acetonide from a gel using phonophoresis. Arch Pharm Res 2006;29:412-7
100. Mitragotri S, Coleman M, Kost J, Langer R. Analysis of ultrasonically extracted interstitial fluid as a predictor of blood glucose levels. J Appl Physiol 2000;89:961-6

Affiliation

Nadine Barrie Smith PhD
Graduate Program in Acoustics,
The Pennsylvania State University
21 Hallowell Building,
University Park, PA 16802, USA
Tel: +1 814 865 8087; Fax: +1 814 863 0490;
E-mail: nbs@engr.psu.edu