World Journal of Microbiology and Biotechnology (2023) 39:330
https://doi.org/10.1007/s11274-023-03760-8

REVIEW

The increasing importance of novel deep eutectic solvents as potential effective antimicrobials and other medicinal properties

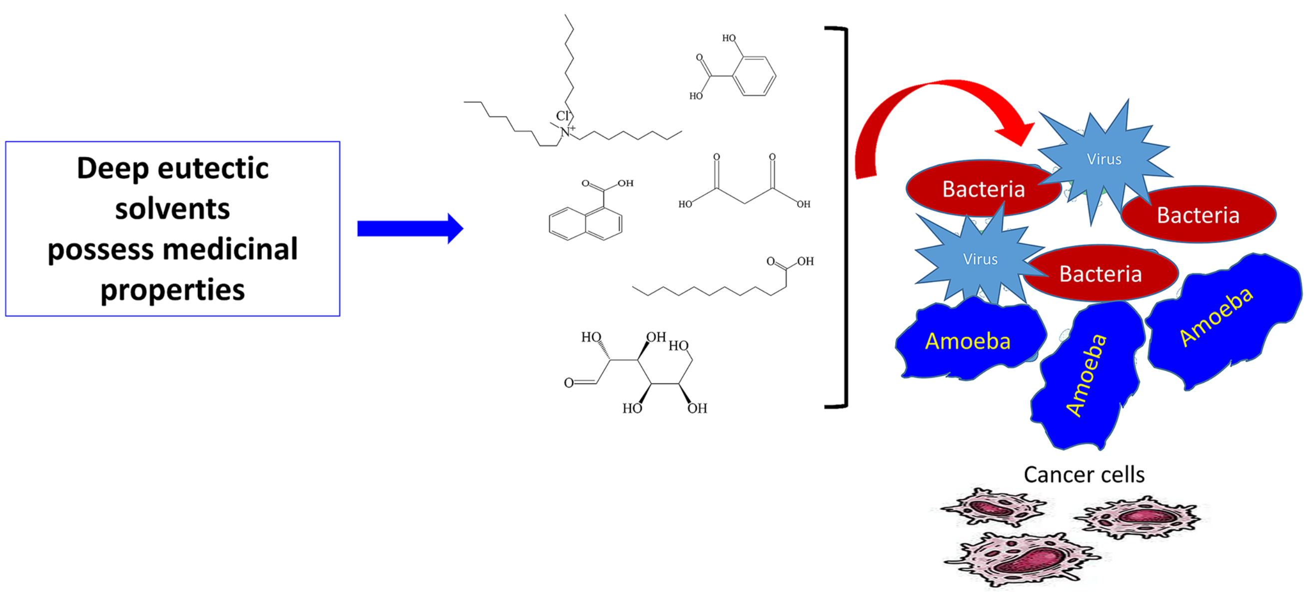
Ruqaiyyah Siddiqui[1,2] · Abdelhamid Khodja[2] · Taleb Ibrahim[3] · Mustafa Khamis[2] · Ayaz Anwar[4] · Naveed Ahmed Khan[1]

Received: 22 May 2023 / Accepted: 11 September 2023 / Published online: 4 October 2023

Abstract

With the rise of antibiotic resistance globally, coupled with evolving and emerging infectious diseases, there is an urgent need for the development of novel antimicrobials. Deep eutectic solvents (DES) are a new generation of eutectic mixtures that depict promising attributes with several biological implications. DES exhibit unique properties such as low toxicity, biodegradability, and high thermal stability. Herein, the antimicrobial properties of DES and their mechanisms of action against a range of microorganisms, including bacteria, amoebae, fungi, viruses, and anti-cancer properties are reviewed. Overall, DES represent a promising class of novel antimicrobial agents as well as possessing other important biological attributes, however, future studies on DES are needed to investigate their underlying antimicrobial mechanism, as well as their in vivo effects, for use in the clinic and public at large.

Graphical abstract



Keywords Deep eutectic solvents · Antimicrobial agents · Antibiotic resistance · Anti-virals · Anti-fungals · Anti-bacterials

Extended author information available on the last page of the article

Introduction

The emergence of antibiotic-resistant bacteria poses a significant threat to public health (Leonard et al. 2022). According to a recent report based on statistical models, approximately 4.95 million deaths in 2019 were linked to bacterial antimicrobial resistance, with 1.27 million deaths specifically attributed to bacterial antimicrobial resistance (Murray et al. 2022). Infectious diseases continue to evolve, and new pathogens or strains may emerge, as evidenced by the recent global pandemic of coronavirus disease 2019 (COVID-19), Ebola virus disease epidemic from 2013 to 2016, Zika virus disease epidemic in 2015, Middle East respiratory syndrome coronavirus in 2012, the swine flu pandemic in 2009, and severe acute respiratory syndrome coronavirus in 2003, to name a few (Baker et al. 2022; Mungroo et al. 2020). Incidentally, existing antimicrobials have limitations in their spectrum of activity, thus the development of novel antimicrobials is necessary to provide a broader range of coverage, targeting a wider variety of pathogens and ideally causing minimal toxicity to the host human. The development of novel antimicrobials is crucial to address the evolving challenges posed by antimicrobial resistance, emerging infections, and the need for more effective treatment options (Lai et al. 2022).

Deep eutectic solvents (DES) have recently secured attention from the scientific, pharmaceutical and industrial communities, as a potential new class of antimicrobial agents. Their growing popularity is attributed to a plethora of factors, such as their versatile applications, cost-effectiveness, and their ability to serve as alternatives to both organic solvents and ionic liquids (Hansen et al. 2020). In this review, various studies describing the antibacterial, anti-amoebic, anti-fungal, anti-viral activities as well as anti-cancer effects of DES are discussed. Moreover, DES mechanisms of action, and other attributes are deliberated upon. However, there is a need for future work to comprehend precise and detailed mechanism of action of the DES systems, improve their selectivity whilst minimizing toxicity, and importantly evaluation of their safety in vivo and in clinical trials. Such research efforts will play a crucial role in advancing the development and optimization of DES for eventual use in the clinic.

What are deep eutectic solvents (DES)?

A eutectic system refers to a combination of two or more components in a certain ratio, which have a melting point that is lower than that of the individual components (Zainal-Abidin et al. 2019). DES are commonly described as mixtures of compounds that primarily associate through hydrogen bonding. When these compounds are combined in specific proportions, a eutectic mixture is formed. The term "eutectic" originates from the Ancient Greek word eútēktos, meaning easily melted. A eutectic point represents the specific composition and temperature at which a mixture of two solids completely melts at the lowest temperature compared to either individual compound (Paiva et al. 2014; El Achkar et al. 2021). These systems have various widespread applications, including pharmaceutical, biotechnological, and industrial fields, and are commonly utilized in drug delivery, as a medium for enzymatic reactions, and for solubilizing drugs (Mbous et al. 2017; Abbott et al. 2001; Stott et al. 1998; Cherukuvada and Nangia 2014). DES are a new generation of eutectic mixtures made from a combination of Lewis or Brønsted–Lowry acids and bases with anionic and/or cationic species (Smith et al. 2014). The lower melting points of DES mixtures are attributed to charge delocalization caused by hydrogen bonding between the hydrogen bond donor and the halide anion (Carriazo et al. 2012; Abbott et al. 2004; Smith et al. 2014). Recently, DES have garnered widespread scientific and industrial interest and are being explored as novel antimicrobial agents. This is evidenced by the literature, which reveals a remarkable surge in publications over the past two decade or thereabouts, in comparison to the 1990s whereby there were only a few reports on DES describing eutectic mixtures as substrates for enzymatic reactions. This widespread attention is due to a multitude of reasons, namely, numerous applications, low cost, and alternatives to organic solvents and ionic liquids(Zainal-Abidin et al. 2019). This review is concerned mainly in exploring the applications of DES as novel antimicrobial agents as well as other biological activities such as anti-cancer effects. Previous studies have demonstrated the antimicrobial activity of a variety of DES systems (Zainal-Abidin et al. 2019). DES have several unique and advantageous properties that contribute to their antimicrobial activity, including low volatility, low toxicity, thermal stability, biodegradability, and the ability to form complexes with certain biomolecules (Mbous et al. 2017; Juneidi et al. 2015; van Osch et al. et al. 2015; Abo-Hamad et al. 2015; Zdanowicz et al. 2018; Mannu et al. 2021; Ling and Hadinoto 2022; Curreri et al. 2021; Li 2022).

Natural deep eutectic solvents (NADES) are a new category of green DES prepared from complexations of naturally occurring primary metabolites, amino acids, sugar alcohols, sugars, organic acids, amines, carboxyl groups, or amino groups (Dai et al. 2013, 2016; Liu et al. 2018; Hayyan et al. 2016; Mukhopadhyay et al. 2016; Choi et al. 2011). NADES have enabled the solubilization and production of semi-polar substances that cannot be dissolved in water or a lipid phase (Liu et al. 2018; Dai et al. 2013; Markham et al.

2000; Pisano et al. 2018; Wikene et al. 2017). They have the potential to replace toxic industrial organic solvents used in the synthesis, extraction, and purification of bioactive macromolecules (i.e., proteins, polysaccharides, polyphenols) (Zainal-Abidin et al. 2017; García et al. 2016; Wei et al. 2015; Zhang and Wang 2017; Li and Lee 2016; Liu et al. 2017). Furthermore, NADES can act as stabilizing agents for drugs with poor water solubility and are being explored as delivery vehicles for pharmaceutical ingredients such as antibiotics (Cao et al. 2020; Olivares et al. 2018). Beta-lactam antibiotics, such as penicillin, are unstable in water-based solutions and lose their antimicrobial activity. NADES may be a useful alternative as non-toxic, non-aqueous solvents for clinical use (Olivares et al. 2018). Betaine and urea (BU) are being explored as an interesting alternative solvent, but further testing is needed to confirm their biological compatibility in formulations (Olivares et al. 2018).

Antibacterial properties of DES

The overuse and misuse of antibiotics has led to the generation of resistant bacteria, making many drugs ineffective against bacterial infections. To exacerbate the issue further, the development of new antibiotics has relied mainly on synthetic modification of natural compounds (Ghosh et al. 2019; Akbar et al. 2023). However, this approach has been slow and with the pipeline for new drugs dwindling the need for new methods for the development and enhancement of antibacterial agents is a pressing issue (McKinney and Pruden 2012; Bassetti et al. 2015). DES hold promising potential in this regard as novel antimicrobial agents and a variety of DES mixtures are being explored for their antimicrobial properties (Zainal-Abidin et al. 2019; Olivares et al. 2018; Silva et al. 2019; Zhou et al. 2022; Siddiqui et al. 2022, 2023).

Initial reports documenting the antibacterial activity of DES were described with cholinium-based and phosphonium-based DES against several Gram-positive and Gram-negative bacteria (Hayyan et al. 2013). Both DES systems demonstrated antibacterial properties; however, the overall phosphonium-based DES demonstrated significantly higher antibacterial activity towards *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Escherichia coli*. It is thought that the type of hydrogen bond donor utilized in the preparation of the cholinium-based and phosphonium-based DES may play a critical role in the antibacterial activity of the DES (Zainal-Abidin et al. 2019).

The antibacterial activity of fatty acids and their synergistic effects in combination have been well documented and can be attributed to their non-specific action by destabilizing the cellular membrane. This is beneficial as it enables fatty acids to have broad-spectrum action with a reduced chance of resistance spread (Kitahara et al. 2004; Batovska et al. 2009; Nakatsuji et al. 2009; Lee and Jo 2016; Kabara Jon et al. 1972; Ouattara et al. 1997; Zheng et al. 2005; Desbois and Smith 2010). Recently, the antimicrobial efficacy of saturated fatty acid-based DES systems was investigated. The antimicrobial potential of saturated fatty acid-based DES was evaluated against a broad range of microorganisms, and data revealed that the DES retained the antimicrobial activity of the individual fatty acids and possibly had synergistic effects between components, especially in the capric acid-myristic acid formulation (Silva et al. 2019). DES showed significant antimicrobial activity against *Candida albicans*, *S. aureus*, Methicillin-Resistant *S. aureus* (MRSA), Methicillin-Resistant *Staphylococcus epidermidis* (MRSE), with the capric acid–lauric acid formulation having the greatest overall bactericidal activity (Silva et al. 2019). However, when tested against *E. coli* and *P. aeruginosa*, all the fatty acid-based DES systems demonstrated minimal activity (Silva et al. 2019). The capric acid–lauric acid system also had the ability to promote membrane detachment/removal in all of the microorganisms tested, however, the mechanisms are not clearly understood, and further research is required to elucidate these (Silva et al. 2019). It was found that the overall antimicrobial activity of fatty acid-based DES increased with a lower fatty acid chain length (Silva et al. 2019). Other studies concur and a chain length between 10 and 12 carbon atoms displays the most promising antimicrobial activity due to hydrophobicity, bioavailability, and microbial membrane permeability (Silva et al. 2019; Batovska et al. 2009; Kitahara et al. 2004; Nakatsuji et al. 2009; Huang et al. 2014). Data suggests that saturated fatty acid-based DES may have potential as novel broad-spectrum antimicrobial agents and may be useful alternatives to antibiotics, which could facilitate the dissemination of antibiotic resistance (Silva et al. 2019).

Another comprehensive study assessing the broad-spectrum antimicrobial activity of DES utilized a choline bicarbonate and geranic acid (CAGE) DES mixture (Zakrewsky et al. 2016). These mixtures were tested against a wide variety of bacterial strains, and a plethora of multi drug resistant strains (Zakrewsky et al. 2016). This study also reported that CAGE exhibited broad-spectrum antimicrobial activity against several of the tested strains. Specifically, the CAGE system used was able to effectively target *Mycobacterium tuberculosis, S. aureus*, and *C. albicans*. Furthermore, experimentation conducted on human keratinocytes and mice demonstrated that CAGE DES system exhibited minimal toxicity at both local and systemic levels (Zakrewsky et al. 2016). Furthermore, this system provided a significantly improved efficacy-toxicity ratio over the currently used antiseptic agents (Zakrewsky et al. 2016). Additionally, in vivo experiments revealed that CAGE DES had the ability to penetrate deeply into the dermis and effectively treat infections caused by *Propionibacterium acnes*, which are located in the deeper layers of the skin (Zakrewsky et al. 2016). This CAGE system should be further evaluated in clinical trials.

The antibacterial activity of choline chloride (ChCl)-based DES were investigated against *Listeria monocytogenes, S. aureus*, *E. coli* and *Salmonella enteritidis* (Zhao et al. 2015). Inhibition of bacterial growth was documented for several of these DES namely: ChCl/oxalic acid, ChCl/p-toluenesulfonic acid, ChCl/malonic acid, ChCl/malic acid, ChCl/levulinic acid, ChCl/citric acid, and ChCl/tartaric acid. The efficacy of ChCl-based DES was also compared to sugar-based, alcohol-based, and amine-based DES mixtures; and ChCl-based DES outperformed the other preparations in terms of antimicrobial activity (Zhao et al. 2015). Moreover, the findings indicated that DES based on choline chloride were effective solvents characterized by minimal toxicity and favourable biodegradability (Zhao et al. 2015).

Recently, DES that were based on hydrogen donors and acceptors were tested for their bactericidal properties against several multi drug resistant bacteria (Akbar et al. 2023). It was reported that specific DES systems prepared from methyl-trioctylammonium chloride and fructose, exerted the highest bactericidal activity against multidrug resistant bacteria (Akbar et al. 2023). Importantly, these DES showed limited toxicity towards human cells, making them promising candidates for further research and potential applications in antimicrobial therapies. Previous studies have also reported the antibacterial properties of choline chloride-based DES, medicinal plants' extracts in amalgamation with glycerol-based choline chloride with citric acid-1,2-propanediol, natural DES extracted from phenolic compounds, and phosphonium-based deep eutectic solvents against various bacteria (Jurić et al. 2021; Grozdanova et al. 2020; Zhao et al. 2015). The hydrophobicity of DES systems may play a role in their antibacterial activity as the hydrophobic phase that forms can interact with bacteria, thus eliminating them (Syed et al. 2022). Although there is an abundance of studies examining effects of DES on a range of bacteria in vitro, currently there are limited in vivo studies being accomplished (Lomba et al. 2021). Thus, further research should be encouraged in various animal models of bacterial infectious diseases as well as determination of the efficacy and safety of more DES systems, before progressing to clinical trials.

Anti-fungal properties of DES

With the rise in drug-resistant fungal infections there is a demand for novel, efficient, and safe antifungal agents (Shao et al. 2007). Recently, several studies have shown the promising anti-fungal properties of DES (Abdullah et al. 2021; Grozdanova et al. 2020; Juneidi et al. 2015, 2016). The anti-fungal properties of DES may depend on their composition, including the type and concentration of individual components and the type of DES formed. DES have also been demonstrated as an alternative green solvent to toxic organic solvents used in the extraction of plant biocompounds for anti-fungal application (Grozdanova et al. 2020; Abdullah et al. 2021). In a recent study the in vitro anti-fungal activity of the medicinal plant *Premna odorata* Blanco extract against two pathogenic fungi *C. albicans* and *Monilinia* spp. was assessed (Abdullah et al. 2021). DES extracts of *P. odorata* Blanco demonstrated significant antifungal activity against *C. albicans* and *Monilinia* spp.. Whereas, the choline chloride:laevulinic acid extract had the lowest inhibitions against fungi (Abdullah et al. 2021).

In another study NADES were utilized as an alternative to toxic organic solvents for the extraction of bioactive compounds, phenols and flavonoids, from medicinal plants *Sideritis scardica*, and *Plantago major* (Grozdanova et al. 2020). Four NADES were evaluated for their ability to extract phenolic and flavonoid compounds from the plants and their antimicrobial and genotoxicity profiles. Choline chloride-glucose NADES had the highest extraction efficiency, but their plant extracts demonstrated no antimicrobial or anti-fungal activity against all tested microorganisms including *Candida albicans*. However, when combined with NADES, plant extracts exhibited significant antimicrobial activity against all the microorganisms. Specifically, the citric acid-1,2-propanediol (CAPD) and choline chloride-glycerol (XXGly) systems exhibited the highest activity against *S. pyogenes*, *E. coli*, *S. aureus*, and *C. albicans*. In addition to the NADES extracts tested, the antimicrobial properties of the NADES was higher than that of traditional ethanol extracts (Grozdanova et al. 2020). In another study, the addition of excess water to NADES extracts showed that it could potentially destroy the supramolecular complex; however, the individual constituents may still contribute to the overall antimicrobial effect (Gutiérrez et al. 2009). Furthermore, all four NADES revealed negligible genotoxicity and minimal cytotoxicity profiles determined on the normal mouse fibroblast cell lines followed by genotoxicity assays, suggesting that these could be evaluated further for use in vivo and clinical trials (Grozdanova et al. 2020).

In a previous study, the anti-fungal properties of choline chloride DES and N,N-diethylethanolammonium chloride (EAC)-based DES were compared (Juneidi et al. 2015). The results revealed that the EAC-based DES exhibited higher anti-fungal activity compared to choline chloride-based DES when tested against *Aspergillus niger*. Namely, the EAC:zinc chloride system exhibited the highest activity, followed by the EAC:zinc nitrate hexahydrate and EAC:malonic acid. In this study a DES classification system was utilized as proposed previously (Smith et al. 2014): DES Type I (organic salts and metal salts), DES Type II (organic salt and hydrate metal salt), DES Type III (organic salts and hydrogen bond donors), and DES Type IV (metal salt with hydrogen bond donor). Type I, II, and IV DES systems comprised metal salts that are capable of ionic interactions and have higher anti-fungal properties than a type III system. The study highlighted that the type I system exhibited the highest level of toxicity whereas type III exhibited the lowest toxicity (Juneidi et al. 2015). Of note, the proposed mechanism of action explaining the anti-fungal activity of DES is through a dehydrating effect, however this needs to be confirmed (Cardellini et al. 2014, 2015).

The anti-fungal properties of cholinium-based DES versus various fungi such as *Candida cylindracea, Lentinus tigrinus*, *Aspergillus niger* and *Phanerochaete chrysosporium* were elucidated(Juneidi et al. 2016). The individual components of the DES exhibited lower anti-fungal activities than did their conjugated DES systems which is consistent with previous work on bacterial and fungal species (Wen et al. 2015; Juneidi et al. 2015). It is believed that the observed differences in anti-fungal activity between DES and their individual starting materials are likely a result of the synergistic effects resulting from DES formation through the hydrogen bonding between the salt anion and hydrogen bond donor which results in changing their properties through charge delocalization (Hayyan et al. 2013). This delocalization of charges is thought to develop through hydrogen bonding and is expected to enhance DES toxicity (Hayyan et al. 2015; Modica-Napolitano and Aprille 2001).

In another recent study the ecotoxicological profile of ammonium-based DES with glycerol, ethylene glycol, and diethylene glycol as hydrogen bond donors was investigated (Inayat et al. 2023). Various biological systems, including fungi, bacteria, fish, and human fibroblast cell lines, were evaluated for their interaction with these DES. The results revealed that tetrabutylammonium bromide: ethylene glycol exhibited the most potent antimicrobial activity, while tetrabutylammonium bromide: diethylene glycol had greater toxicity against *Cyprinus carpio* fish (Inayat et al. 2023). Nonetheless, although studies investigating the impact of DES on various fungi are present, again there is a lack of in vivo research on this topic. Therefore, it is important to encourage further investigations using animal models of fungal infections to assess the effectiveness and safety of different DES systems.

Antiamoebic properties of DES

Amoebae such as *Acanthamoeba* are prevalent in diverse environments globally, with over 80% of the population possessing IgG antibodies to these species (Ozpinar et al. 2021; Haston et al. 2023). Free-living amoebae *Naegleria fowleri*, *Acanthamoeba castellanii*, *Balamuthia mandrillaris* and *Vermamoeba vermiformis* thrive in water, soil and other environments and thus pose a threat to human health, especially in developing countries, particularly for those communities that reliant on water storage tanks as a result of limited water supplies (Siddiqui et al. 2021; Mungroo et al. 2022; Gharpure et al. 2021). Unfortunately, these tanks often harbour a variety of microbial organisms and serve as potential sources for the transmission of amoebic infections. Rise in temperatures due to global warming, may further exacerbate the situation, as amoebae are thermophilic (Maciver et al. 2020). Currently, the treatments for these infections are ineffective and consist of a cocktail of antimicrobial drugs which can cause nephrotoxicity and thus the rate of mortality is more than 90% (Debnath 2021; Tillery et al. 2021).

While the antiviral, antibacterial, and antifungal properties of DES have been documented previously, only recently have the antiamoebic properties of DES been explored. The antiamoebic properties of DES were first documented recently (Siddiqui et al. 2022, 2023). As indicated earlier, many studies have reported the antibacterial activity of salicylic acid and malonic acid-based DES (Bandara et al. 2016; Radošević et al. 2018; Sadaf et al. 2018). Using similar salicylic acid and malonic acid-based DES, a recent study investigated the antiamoebic effects of five DES on *Acanthamoeba castellanii* (Siddiqui et al. 2022). The DES showed significant antiamoebic activity by suppressing excystment and encystment, and cell-mediated cytopathogenicity of *A. castellanii*. Specifically, salicylic acid-trihexylamine DES (DES2) and salicylic acid-trioctylamine (DES3) demonstrated low cytotoxicity to human cells at micromolar concentrations. The individual components of the DES did not display any significant activity which was consistent with previous work on bacterial and fungal species (Wen et al. 2015). However, the mechanism of action of the salicylic acid and malonic acid-based DES was not determined, although the observed amoebicidal effects of the DES are likely mediated through hydrogen bond donor-acceptor interactions that disrupt the amoebic membranes. The study also found that the hydrogen bond donor: salicylic acid is superior to the weaker acid: malonic acid, contributing to the efficacy of DES2 and DES3. Additional in vivo studies are required to assess and confirm their safety and efficacy (Siddiqui et al. 2022). Nevertheless, the results of this study suggested that salicylic acid-based DES have potential as novel antimicrobial agents against *A. castellanii* and may represent a new avenue for the development of therapeutic strategies against amoebic infections.

Another study demonstrated that methyltrioctylammonium chloride (Aliquat 336)-based DES exhibit potent anti-amoebic properties at micromolar doses (Siddiqui et al. 2023). The investigation revealed that Aliquat 336-glucose (DES-B), Aliquat 336-ethylene glycol (DES-C), Aliquat 336-citric acid (DES-D), and Aliquat 336-fructose (DES-E) displayed potent antiamoebic properties at micromolar concentrations versus *Acanthamoeb*a. This was demonstrated through various amoebicidal, encystment, excystment, cytotoxicity, and cytopathogenicity assays (Siddiqui et al. 2023). Specifically, only DES-D showed elevated cytotoxicity, which may be due to the toxicity of its shared hydrogen bond acceptor, methyltrioctylammonium chloride. Of interest, the potency of DES versus the cyst form of pathogenic *Acanthamoeba* was noted. This is encouraging, as current treatment of this infection is problematic, and often there is reoccurrence of infection due to the resistant nature of the cyst form. It is speculated that this maybe likely due to DES disrupting hydrogen bonding in the cellulose inner wall of *A. castellanii* cysts.

The underlying mechanism accounting for DES action in amoebae remains unclear, and further mechanistic studies are necessary. The study suggests that DES could be a valuable ingredient in the development of contact lens cleaning solutions and other antiamoebic treatments given the inefficacy of most currently solutions and treatments. Future research should evaluate the efficacy of DES against other free-living amoebae. Specifically, against *Naegleria fowleri*, *Balamuthia mandrillaris*, and *Vermamoeba vermiformis*. Furthermore, comprehension of various doses, incubation periods, use of different bases, as well as their efficacy and safety in corneal epithelial cell lines and in animal models of *Acanthamoeba keratitis* as well as other amoebae are necessitated.

Anti-viral properties of DES

Given the connection of the ecosystem and the ongoing interaction between viruses and their hosts, it is inevitable that zoonotic outbreaks, epidemics, and pandemics in the future will occur (Holmes 2022). Despite a plethora of studies versus bacteria, studies examining anti-viral effects of DES have been limited, and should be explored given the potency of some DES against other microorganisms. A study evaluating the anti-viral activities of DES was accomplished (Zakrewsky et al. 2016). It was found that a choline and geranate-based DES (CAGE DES) exhibited anti-viral activity against herpes simplex virus type-1 (HSV-1) and herpes simplex virus type-2 (HSV-2) through the mechanism of neutralization (Zakrewsky et al. 2016).However, the mechanism of the neutralization process is still unknown and requires further investigations (Zakrewsky et al. 2016; Zainal-Abidin et al. 2019).

In another recent study, the andrographolide content and antiviral activity of methanol extract and extracts of DES from *Andrographis paniculata* were evaluated against human coronavirus OC43 (HcoV-OC43) (Komaikul et al. 2023). Eleven DES extracts were prepared using choline chloride and proline as hydrogen bond acceptors, and malic acid, glycerol, dextrose, and citric acid as hydrogen bond donors. The results showed that the methanol extract exhibited the highest andrographolide content and displayed the most significant antiviral activity compared to the ethanolic extract. For this reason, the authors compared the methanol extract with the *A. paniculata* DES extract to assess anti-HcoV-OC43 activity. DES-8 and DES-4 had the highest andrographolide content. The in vitro anti-HcoV-OC43 activities of the *A. paniculata* methanolic extract, eight DES extracts, and andrographolide were evaluated using Remdesivir as a positive control (Komaikul et al. 2023). The acidic DES comprising citric acid and malic acid without *A. paniculata* extract showed no antiviral activity at non-cytotoxic doses. The antiviral tests demonstrated that 5 acidic DES extracts displayed higher anti-HcoV-OC43 activity than the methanol extract with the highest andrographolide concentration. Eleven acidic DES, five DES extracts demonstrated greater anti-viral efficacy against HcoV-OC43 when compared to the methanol extract containing the highest andrographolide concentration. Moreover, the stability test of andrographolide in DES extracts showed that the extracts remained stable at 4 °C for at least one month. The study highlighted the potential of DES as an alternative and efficient method for extracting andrographolide from *A. paniculata*, with promising antiviral activity against HcoV-OC43. Further studies are needed to fully comprehend the mechanism of action of DES and DES extracts of *A. paniculata* against HcoV-OC43.

In another study, NADES were utilized as a solvent to extract bioactive compounds from spices in cinnamon, curcumin, and pepper (Długosz et al. 2022). The resulting spice extracts were then used to synthesize selenium sulfide nanoparticles. These selenium sulfide nanoparticles stabilized in the spice extracts from NADES were tested for their antimicrobial and antiviral activity, rather than testing NADES directly against microbes. The selenium sulphide nanoparticle suspensions displayed potent antimicrobial and antiviral activity against the various bacteria and fungi, as well as Human influenza virus A/H1N1, and Betacoronavirus 1 (Human coronavirus HcoV-OC43) (Długosz et al. 2022). Although research on viruses can be problematic, given their potent antimicrobial activities, future studies evaluating DES on other viruses of concern such as HIV, dengue, monkeypox, coronaviruses should be accomplished.

Concluding remarks and prospective studies

A multitude of studies show that DES demonstrate promising and potent antimicrobial properties against a plethora of pathogens, including multidrug resistant bacteria, fungi, protists and some viruses. Additional reports indicate the potential of DES as novel, eco-friendly, and cost-effective antimicrobial agents. Studies have demonstrated that DES can disrupt the cell membrane, alter cellular morphology and interfere with vital metabolic processes in cancer cells

(Zainal-Abidin et al. 2019). Albeit, these data are encouraging, in some cases the mechanisms of action of DES on pathogens remains poorly understood, thus research is needed to elucidate the underlying mechanisms of DES.

Importantly, investigations are warranted to explore potential synergistic effects of various combinations of DES in combination with other known clinically approved antimicrobial agents. Furthermore, current research utilizing DES as antimicrobial agents is mostly at the exploratory stage and most studies have been conducted in vitro. Prospective translational research is needed in relevant animal models of infectious diseases, before progression to clinical trials. Research on anti-viral effects of DES is still in its infancy. Furthermore, DES reveal potential potent antimicrobial properties against a wide range of pathogens, nonetheless there is ambiguity regarding their toxicity, their chemical inertness, and their non-requirement for purification, which also needs to be clarified in vivo studies. Perhaps DES can be combined with nanotechnology to further reduce any toxicity and augment targeted delivery to pathogens, to mitigate potential toxicity issues. In addition, DES systems could be attached to membranes and utilized for water filtering. Against pathogens, which will be particularly useful for developing countries or countries that utilize water tanks for storage due to water shortages.

Author contributions RS and NAK conceptualized the study amid critical discussions with TI and MK. AK reviewed the literature under supervision of RS and NAK. AK and RS prepared the first draft that was corrected by NAK, AA, TI and MK. All authors approved the final manuscript and will act as guarantors.

Funding Ruqaiyyah Siddiqui and Naveed Ahmed Khan are supported by the Air Force Office of Scientific Research (AFOSR).

Data availability No new data were created or analyzed in this study.

Declarations

Conflict of interest The authors have not disclosed any conflict of interest.

References

Abbott AP, Capper G, Davies DL, Munro HL, Rasheed RK, Tambyrajah V (2001) Preparation of novel, moisture-stable, Lewis-acidic ionic liquids containing quaternary ammonium salts with functional side chains. Chem Commun 19:2010–2011. https://doi.org/10.1039/B106357J

Abbott AP, Boothby D, Capper G, Davies DL, Rasheed RK (2004) Deep eutectic solvents formed between choline chloride and carboxylic acids: versatile alternatives to ionic liquids. J Am Chem Soc 126(29):9142–9147. https://doi.org/10.1021/ja048266j

Abdullah AK, Dheyab AS, Saleh RO (2021) Evaluation of antifungal activity derivative from *Premna odorata* blanco extract by deep eutectic solvents. Plant Sci Today. https://doi.org/10.14719/pst.2021.8.4.1381

Abo-Hamad A, Hayyan M, AlSaadi MA, Hashim MA (2015) Potential applications of deep eutectic solvents in nanotechnology. J Chem Eng 273:551–567. https://doi.org/10.1016/j.cej.2015.03.091

Akbar N, Khan NA, Ibrahim T, Khamis M, Khan AS, Alharbi AM, Alfahemi H, Siddiqui R (2023) Antimicrobial activity of novel deep eutectic solvents. Sci Pharm. https://doi.org/10.3390/scipharm91010009

Baker RE, Mahmud AS, Miller IF, Rajeev M, Rasambainarivo F, Rice BL, Takahashi S, Tatem AJ, Wagner CE, Wang LF, Wesolowski A (2022) Infectious disease in an era of global change. Nat Rev Microbiol 20(4):193–205. https://doi.org/10.1038/s41579-021-00639-z

Bandara M, Sankaridurg P, Zhu H, Hume E, Willcox M (2016) Effect of salicylic acid on the membrane proteome and virulence of *Pseudomonas aeruginosa*. Investig Ophthalmol Vis Sci 57(3):1213–1220. https://doi.org/10.1167/iovs.15-18990

Bassetti M, De Waele JJ, Eggimann P, Garnacho-Montero J, Kahlmeter G, Menichetti F, Nicolau DP, Paiva JA, Tumbarello M, Welte T, Wilcox M, Zahar JR, Poulakou G (2015) Preventive and therapeutic strategies in critically ill patients with highly resistant bacteria. J Intensive Care Med 41(5):776–795. https://doi.org/10.1007/s00134-015-3719-z

Batovska DI, Todorova IT, Tsvetkova IV, Najdenski HM (2009) Antibacterial study of the medium chain fatty acids and their 1-monoglycerides: individual effects and synergistic relationships. Pol J Microbiol 58(1):43–47. https://doi.org/

Cao J, Cao J, Wang H, Chen L, Cao F, Su E (2020) Solubility improvement of phytochemicals using (natural) deep eutectic solvents and their bioactivity evaluation. J Mol Liq 318:113997. https://doi.org/10.1016/j.molliq.2020.113997

Cardellini F, Tiecco M, Germani R, Cardinali G, Corte L, Roscini L, Spreti N (2014) Novel zwitterionic deep eutectic solvents from trimethylglycine and carboxylic acids: characterization of their properties and their toxicity. RSC Adv 4(99):55990–56002. https://doi.org/10.1039/c4ra10628h

Cardellini F, Germani R, Cardinali G, Corte L, Roscini L, Spreti N, Tiecco M (2015) Room temperature deep eutectic solvents of (1S)-(+)-10-camphorsulfonic acid and sulfobetaines: hydrogen bond-based mixtures with low ionicity and structure-dependent toxicity. RSC Adv 5(40):31772–31786. https://doi.org/10.1039/C5RA03932K

Carriazo D, Serrano MC, Gutiérrez MC, Ferrer ML, del Monte F (2012) Deep-eutectic solvents playing multiple roles in the synthesis of polymers and related materials. Chem Soc Rev 41(14):4996–5014. https://doi.org/10.1039/C2CS15353J

Cherukuvada S, Nangia A (2014) Eutectics as improved pharmaceutical materials: design, properties and characterization. Chem Commun 50(8):906–923. https://doi.org/10.1039/C3CC47521B

Choi YH, van Spronsen J, Dai Y, Verberne M, Hollmann F, Arends IWCE, Witkamp G, Verpoorte R (2011) Are natural deep eutectic solvents the missing link in understanding cellular metabolism and physiology? Plant Physiol 156(4):1701–1705. https://doi.org/10.1104/pp.111.178426

Curreri AM, Mitragotri S, Tanner EEL (2021) Recent advances in ionic liquids in biomedicine. Adv Sci 8(17):e2004819. https://doi.org/10.1002/advs.202004819

Dai Y, van Spronsen J, Witkamp G, Verpoorte R, Choi YH (2013) Natural deep eutectic solvents as new potential media for green technology. Anal Chim Acta 766:61–68. https://doi.org/10.1016/j.aca.2012.12.019

Dai Y, Rozema E, Verpoorte R, Choi YH (2016) Application of natural deep eutectic solvents to the extraction of anthocyanins from *Catharanthus roseus* with high extractability and stability replacing conventional organic solvents. J Chromatogr A 1434:50–56. https://doi.org/10.1016/j.chroma.2016.01.037

Debnath A (2021) Drug discovery for primary amebic meningoencephalitis: from screen to identification of leads. Expert Rev Anti Infect Ther 19(9):1099–1106. https://doi.org/10.1080/14787210.2021.1882302

Desbois AP, Smith VJ (2010) Antibacterial free fatty acids: activities, mechanisms of action and biotechnological potential. Appl Microbiol Biotechnol 85(6):1629–1642. https://doi.org/10.1007/s00253-009-2355-3

Długosz O, Ochnik M, Sochocka M, Franz D, Orzechowska B, Anna C, Agata D, Banach M (2022) Antimicrobial and antiviral activity of selenium sulphide nanoparticles synthesised in extracts from spices in natural deep eutectic solvents (NDES). Sustain Mat Technol 32:e00433. https://doi.org/10.1016/j.susmat.2022.e00433

El Achkar T, Greige-Gerges H, Fourmentin S (2021) Basics and properties of deep eutectic solvents: a review. Environ Chem Lett 19:3397–3408. https://doi.org/10.1007/s10311-021-01225-8

García A, Rodríguez-Juan E, Rodríguez-Gutiérrez G, Rios JJ, Fernández-Bolaños J (2016) Extraction of phenolic compounds from virgin olive oil by deep eutectic solvents (DESs). Food Chem 197:554–561. https://doi.org/10.1016/j.foodchem.2015.10.131

Gharpure R, Gleason M, Salah Z, Blackstock AJ, Hess-Homeier D, Yoder JS, Ali IKM, Collier SA, Cope JR (2021) Geographic range of recreational water-associated primary amebic Meningoencephalitis, United States, 1978–2018. Emerg Infect Dis 27(1):271–274. https://doi.org/10.3201/eid2701.202119

Ghosh C, Sarkar P, Issa R, Haldar J (2019) Alternatives to conventional antibiotics in the era of antimicrobial resistance. Trends Microbiol 27(4):323–338. https://doi.org/10.1016/j.tim.2018.12.010

Grozdanova T, Trusheva B, Alipieva K, Popova M, Dimitrova L, Najdenski H, Zaharieva MM, Ilieva Y, Vasileva B, Miloshev G, Georgieva M, Bankova V (2020) Extracts of medicinal plants with natural deep eutectic solvents: enhanced antimicrobial activity and low genotoxicity. BMC Chem 14(1):73. https://doi.org/10.1186/s13065-020-00726-x

Gutiérrez MC, Ferrer ML, Mateo CR, del Monte F (2009) Freeze-drying of aqueous solutions of deep eutectic solvents: a suitable Approach to deep eutectic suspensions of self-assembled structures. Langmuir 25(10):5509–5515. https://doi.org/10.1021/la900552b

Hansen BB, Spittle S, Chen B, Poe D, Zhang Y, Klein JM, Horton A, Adhikari L, Zelovich T, Doherty BW, Gurkan B (2020) Deep eutectic solvents: a review of fundamentals and applications. Chem Rev 121(3):1232–1285. https://doi.org/10.1021/acs.chemrev.0c00385

Haston JC, O'Laughlin K, Matteson K, Roy S, Qvarnstrom Y, Ali IK, Cope JR (2023) The epidemiology and clinical features of non-keratitis *Acanthamoeba* infections in the United States, 1956–2020. Open Forum Infect Dis 10(1):ofac682. https://doi.org/10.1093/ofid/ofac682

Hayyan M, Hashim MA, Hayyan A, Al-Saadi MA, AlNashef IM, Mirghani MES, Saheed OK (2013) Are deep eutectic solvents benign or toxic? Chemosphere 90(7):2193–2195. https://doi.org/10.1016/j.chemosphere.2012.11.004

Hayyan M, Looi CY, Hayyan A, Wong WF, Hashim MA (2015) In vitro and in vivo toxicity profiling of ammonium-based deep eutectic solvents. PLoS One 10(2):e0117934. https://doi.org/10.1371/journal.pone.0117934

Hayyan M, Mbous YP, Looi CY, Wong WF, Hayyan A, Salleh Z, Mohd-Ali O (2016) Natural deep eutectic solvents: cytotoxic profile. SpringerPlus 5(1):913. https://doi.org/10.1186/s40064-016-2575-9

Holmes EC (2022) The ecology of viral emergence. Annual Rev Virol 9:173–192. https://doi.org/10.1146/annurev-virology-100120-015057

Huang W, Tsai T, Chuang L, Li Y, Zouboulis CC, Tsai P (2014) Anti-bacterial and anti-inflammatory properties of capric acid against *Propionibacterium acnes*: a comparative study with lauric acid. J Dermatol Sci 73(3):232–240. https://doi.org/10.1016/j.jdermsci.2013.10.010

Inayat S, Ahmad SR, Awan SJ, Nawshad M, Ali Q (2023) In vivo and in vitro toxicity profile of tetrabutylammonium bromide and alcohol-based deep eutectic solvents. Sci Rep 13(1):1777. https://doi.org/10.1038/s41598-023-28928-y

Juneidi I, Hayyan M, Hashim MA (2015) Evaluation of toxicity and biodegradability for cholinium-based deep eutectic solvents. RSC Adv 5(102):83636–83647. https://doi.org/10.1039/C5RA12425E

Juneidi I, Hayyan M, Mohd Ali O (2016) Toxicity profile of choline chloride-based deep eutectic solvents for fungi and *Cyprinus carpio* fish. Environ Sci Pollut Res 23(8):7648–7659. https://doi.org/10.1007/s11356-015-6003-4

Jurić T, Mićić N, Potkonjak A, Milanov D, Dodić J, Trivunović Z, Popović BM (2021) The evaluation of phenolic content, in vitro antioxidant and antibacterial activity of *Mentha piperita* extracts obtained by natural deep eutectic solvents. Food Chem 362:130226. https://doi.org/10.1016/j.foodchem.2021.130226

Kabara Jon J, Swieczkowski Dennis M, Conley Anthony J, Truant Joseph P (1972) Fatty acids and derivatives as antimicrobial agents. Antimicro Agents Chemother 2(1):23–28. https://doi.org/10.1128/AAC.2.1.23

Kitahara T, Koyama N, Matsuda J, Aoyama Y, Hirakata Y, Kamihira S, Kohno S, Nakashima M, Sasaki H (2004) Antimicrobial activity of saturated fatty acids and fatty amines against methicillin-resistant *Staphylococcus aureus*. Biol Pharm Bull 27(9):1321–1326. https://doi.org/10.1248/bpb.27.1321

Komaikul J, Ruangdachsuwan S, Wanlayaporn D, Palabodeewat S, Punyahathaikul S, Churod T, Choonong R, Kitisripanya T (2023) Effect of andrographolide and deep eutectic solvent extracts of *Andrographis paniculata* on human coronavirus organ culture 43 (HcoV-OC43). Phytomedicine 112:154708. https://doi.org/10.1016/j.phymed.2023.154708

Lai CK, Ng RW, Leung SS, Hui M, Ip M (2022) Overcoming the rising incidence and evolving mechanisms of antibiotic resistance by novel drug delivery approaches–an overview. Adv Drug Deliv Rev 181:114078. https://doi.org/10.1016/j.addr.2021.114078

Lee J, Jo YW (2016) Antimicrobial effect of a lauric acid on *Streptococcus Mutans* biofilm. Ann Int Med Dent Res. https://doi.org/10.21276/aimdr.2016.2.4.21

Leonard AF, Morris D, Schmitt H, Gaze WH (2022) Natural recreational waters and the risk that exposure to antibiotic resistant bacteria poses to human health. Curr Opin Microbiol 65:40–46. https://doi.org/10.1016/j.mib.2021.10.004

Li D (2022) Natural deep eutectic solvents in phytonutrient extraction and other applications. Front Plant Sci 13:1004332. https://doi.org/10.3389/fpls.2022.1004332

Ling JK, Hadinoto K (2022) Deep eutectic solvent as green solvent in extraction of biological macromolecules: a review. Int J Mol Sci. https://doi.org/10.3390/ijms23063381

Liu R, Yu P, Ge X, Bai X, Li X, Fu Q (2017) Establishment of an aqueous PEG 200-based deep eutectic solvent extraction and enrichment method for pumpkin (*Cucurbita moschata*) seed protein. Food Anal Methods 10(6):1669–1680. https://doi.org/10.1007/s12161-016-0732-y

Liu Y, Zhang Y, Chen S, Friesen JB, Nikolić D, Choules MP, McAlpine JB, Lankin DC, Gemeinhart RA, Pauli GF (2018) The influence of natural deep eutectic solvents on bioactive natural products: studying interactions between a hydrogel model and *Schisandra chinensis* metabolites. Fitoterapia 127:212–219. https://doi.org/10.1016/j.fitote.2018.02.024

Lomba L, Ribate M, Sangüesa E, Concha J, Garralaga M, Errazquin D, García CB, Giner B (2021) Deep eutectic solvents: are they safe? Appl Sci 11(21):10061. https://doi.org/10.3390/app112110061

Maciver SK, Piñero JE, Lorenzo-Morales J (2020) Is *Naegleria fowleri* an emerging parasite? Trends Parasitol 36(1):19–28. https://doi.org/10.1016/j.pt.2019.10.008

Mannu A, Blangetti M, Baldino S, Prandi C (2021) Promising technological and industrial applications of deep eutectic systems. Materials. https://doi.org/10.3390/ma14102494

Markham KR, Gould KS, Winefield CS, Mitchell KA, Bloor SJ, Boase MR (2000) Anthocyanic vacuolar inclusions—their nature and significance in flower colouration. Phytochemistry 55(4):327–336. https://doi.org/10.1016/S0031-9422(00)00246-6

Mbous YP, Hayyan M, Wong WF, Looi CY, Hashim MA (2017) Unraveling the cytotoxicity and metabolic pathways of binary natural deep eutectic solvent systems. Sci Rep 7(1):41257. https://doi.org/10.1038/srep41257

McKinney CW, Pruden A (2012) Ultraviolet disinfection of antibiotic resistant bacteria and their antibiotic resistance genes in water and wastewater. Environ Sci Technol 46(24):13393–13400. https://doi.org/10.1021/es303652q

Modica-Napolitano JS, Aprille JR (2001) Delocalized lipophilic cations selectively target the mitochondria of carcinoma cells. Adv Drug Deliv Rev 49(1):63–70. https://doi.org/10.1016/S0169-409X(01)00125-9

Mukhopadhyay S, Mukherjee S, Adnan NF, Hayyan A, Hayyan M, Hashim MA, Sen Gupta B (2016) Ammonium-based deep eutectic solvents as novel soil washing agent for lead removal. Chem Eng J 294:316–322. https://doi.org/10.1016/j.cej.2016.02.030

Mungroo MR, Khan NA, Siddiqui R (2020) Novel coronavirus: current understanding of clinical features, diagnosis, pathogenesis, and treatment options. Pathogens 9(4):297. https://doi.org/10.3390/pathogens9040297

Mungroo MR, Khan NA, Maciver S, Siddiqui R (2022) Opportunistic free-living amoebal pathogens. Pathog Glob Health 116(2):70–84. https://doi.org/10.1080/20477724.2021.1985892

Murray CJ, Ikuta KS, Sharara F, Swetschinski L, Aguilar GR, Gray A, Han C, Bisignano C, Rao P, Wool E, Johnson SC (2022) Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. Lancet 399(10325):629–655. https://doi.org/10.1016/S0140-6736(21)02724-0

Nakatsuji T, Kao MC, Fang J, Zouboulis CC, Zhang L, Gallo RL, Huang C (2009) Antimicrobial property of lauric acid against *Propionibacterium acnes*: its therapeutic potential for inflammatory acne vulgaris. J Invest Dermatol 129(10):2480–2488. https://doi.org/10.1038/jid.2009.93

Olivares B, Martínez F, Rivas L, Calderón C, Munita JM, Campodonico PR (2018) A natural deep eutectic solvent formulated to stabilize β-lactam antibiotics. Sci Rep 8(1):14900. https://doi.org/10.1038/s41598-018-33148-w

Ouattara B, Simard RE, Holley RA, Piette GJ, Bégin A (1997) Antibacterial activity of selected fatty acids and essential oils against six meat spoilage organisms. Int J Food Microbiol 37(2):155–162. https://doi.org/10.1016/S0168-1605(97)00070-6

Ozpinar N, Culha G, Kaya T, Yucel H (2021) The amoebicidal activity of three substances derived from benzothiazole on *Acanthamoeba castellanii* cysts and trophozoites and its cytotoxic potentials. Acta Trop 220:105981. https://doi.org/10.1016/j.actatropica.2021.105981

Paiva A, Craveiro R, Aroso I, Martins M, Reis RL, Duarte ARC (2014) Natural deep eutectic solvents–solvents for the 21st century. ACS Sustain Chem Eng 2(5):1063–1071. https://doi.org/10.1021/sc500096j

Pisano PL, Espino M, Fernández MdlÁ, Silva MF, Olivieri AC (2018) Structural analysis of natural deep eutectic solvents. theoretical and experimental study. Microchem J 143:252–258. https://doi.org/10.1016/j.microc.2018.08.016

Radošević K, Čanak I, Panić M, Markov K, Bubalo MC, Frece J, Srček VG, Redovniković IR (2018) Antimicrobial, cytotoxic and antioxidative evaluation of natural deep eutectic solvents. Environ Sci Pollut Res 25(14):14188–14196. https://doi.org/10.1007/s11356-018-1669-z

Sadaf A, Kumari A, Khare SK (2018) Potential of ionic liquids for inhibiting the growth and β-lactamase production by *Bacillus cereus* EMB20. Int J Biol Macromol 107:1915–1921. https://doi.org/10.1016/j.ijbiomac.2017.10.053

Shao P, Huang L, Hsueh P (2007) Recent advances and challenges in the treatment of invasive fungal infections. Int J Antimicrob Agents 30(6):487–495. https://doi.org/10.1016/j.ijantimicag.2007.07.019

Siddiqui R, Makhlouf Z, Khan NA (2021) The increasing importance of *Vermamoeba vermiformis*. J Eukaryot Microbiol 68(5):e12857. https://doi.org/10.1111/jeu.12857

Siddiqui R, Makhlouf Z, Akbar N, Khamis M, Ibrahim T, Khan AS, Khan NA (2022) Antiamoebic properties of salicylic acid-based deep eutectic solvents for the development of contact lens disinfecting solutions against *Acanthamoeba*. Mol Biochem Parasitol 250:111493. https://doi.org/10.1016/j.molbiopara.2022.111493

Siddiqui R, Makhlouf Z, Akbar N, Khamis M, Ibrahim T, Khan AS, Khan NA (2023) Antiamoebic properties of methyltrioctylammonium chloride based deep eutectic solvents. Cont Lens Anterior Eye 46(2):101758. https://doi.org/10.1016/j.clae.2022.101758

Silva JM, Silva E, Reis RL, Duarte ARC (2019) A closer look in the antimicrobial properties of deep eutectic solvents based on fatty acids. Sustain Chem Pharm 14:100192. https://doi.org/10.1016/j.scp.2019.100192

Smith EL, Abbott AP, Ryder KS (2014) Deep eutectic solvents (DESs) and their applications. Chem Rev 114(21):11060–11082. https://doi.org/10.1021/cr300162p

Stott PW, Williams AC, Barry BW (1998) Transdermal delivery from eutectic systems: enhanced permeation of a model drug, ibuprofen. J Control Release 50(1):297–308. https://doi.org/10.1016/S0168-3659(97)00153-3

Syed UT, Leonardo IC, Mendoza G, Gaspar FB, Gámez E, Huertas RM, Crespo MTB, Arruebo M, Crespo JG, Sebastian V, Brazinha C (2022) On the role of components of therapeutic hydrophobic deep eutectic solvent-based nanoemulsions sustainably produced by membrane-assisted nanoemulsification for enhanced antimicrobial activity. Sep Purif Technol 285:120319. https://doi.org/10.1016/j.seppur.2021.120319

Tillery L, Barrett K, Goldstein J, Lassner JW, Osterhout B, Tran NL, Xu L, Young RM, Craig J, Chun I, Dranow DM (2021) *Naegleria fowleri*: protein structures to facilitate drug discovery for the deadly, pathogenic free-living amoeba. PLoS One 16(3):e0241738. https://doi.org/10.1371/journal.pone.0241738

van Osch Dannie JGP, Zubeir LF, van den Bruinhorst A, Rocha MAA, Kroon MC (2015) Hydrophobic deep eutectic solvents as water-immiscible extractants. Green Chem 17(9):4518–4521. https://doi.org/10.1039/C5GC01451D

Wei Z, Wang X, Peng X, Wang W, Zhao C, Zu Y, Fu Y (2015) Fast and green extraction and separation of main bioactive flavonoids from *Radix Scutellariae*. Ind Crops Prod 63:175–181. https://doi.org/10.1016/j.indcrop.2014.10.013

Wen Q, Chen J, Tang Y, Wang J, Yang Z (2015) Assessing the toxicity and biodegradability of deep eutectic solvents. Chemosphere 132:63–69. https://doi.org/10.1016/j.chemosphere.2015.02.061

Wikene KO, Rukke HV, Bruzell E, Tønnesen HH (2017) Investigation of the antimicrobial effect of natural deep eutectic solvents (NADES) as solvents in antimicrobial photodynamic therapy. J Photochem Photobiol B Biol 171:27–33. https://doi.org/10.1016/j.jphotobiol.2017.04.030

Zainal-Abidin MH, Hayyan M, Hayyan A, Jayakumar NS (2017) New horizons in the extraction of bioactive compounds using deep eutectic solvents: a review. Anal Chim Acta 979:1–23. https://doi.org/10.1016/j.aca.2017.05.012

Zainal-Abidin MH, Hayyan M, Ngoh GC, Wong WF, Looi CY (2019) Emerging frontiers of deep eutectic solvents in drug discovery and drug delivery systems. J Control Release 316:168–195. https://doi.org/10.1016/j.jconrel.2019.09.019

Zakrewsky M, Banerjee A, Apte S, Kern TL, Jones MR, Sesto RED, Koppisch AT, Fox DT, Mitragotri S (2016) Choline and Geranate deep eutectic solvent as a broad-spectrum antiseptic agent for preventive and therapeutic applications. Adv Healthc Mater 5(11):1282–1289. https://doi.org/10.1002/adhm.201600086

Zdanowicz M, Wilpiszewska K, Spychaj T (2018) Deep eutectic solvents for polysaccharides processing. a review. Carbohydr Polym 200:361–380. https://doi.org/10.1016/j.carbpol.2018.07.078

Zhang L, Wang M (2017) Optimization of deep eutectic solvent-based ultrasound-assisted extraction of polysaccharides from *Dioscorea opposita Thunb*. Int J Biol Macromol 95:675–681. https://doi.org/10.1016/j.ijbiomac.2016.11.096

Zhao B, Xu P, Yang F, Wu H, Zong M, Lou W (2015) Biocompatible deep eutectic solvents based on choline chloride: characterization and application to the extraction of Rutin from *Sophora japonica*. ACS Sustain Chem Eng 3(11):2746–2755. https://doi.org/10.1021/acssuschemeng.5b00619

Zheng CJ, Yoo J, Lee T, Cho H, Kim Y, Kim W (2005) Fatty acid synthesis is a target for antibacterial activity of unsaturated fatty acids. FEBS Lett 579(23):5157–5162. https://doi.org/10.1016/j.febslet.2005.08.028

Zhou P, Tang D, Zou J, Wang X (2022) An alternative strategy for enhancing stability and antimicrobial activity of catechins by natural deep eutectic solvents. LWT–Food Sci Technol 153:112558. https://doi.org/10.1016/j.lwt.2021.112558

Authors and Affiliations

Ruqaiyyah Siddiqui[1,2] · Abdelhamid Khodja[2] · Taleb Ibrahim[3] · Mustafa Khamis[2] · Ayaz Anwar[4] · Naveed Ahmed Khan[1]

✉ Naveed Ahmed Khan
naveed5438@gmail.com

1 Microbiota Research Center, Istinye University, 34010 Istanbul, Turkey

2 Department of Biology, Chemistry and Environmental Sciences, College of Arts and Sciences, American University of Sharjah, Sharjah, United Arab Emirates

3 Department of Chemical and Biological Engineering, College of Engineering, American University of Sharjah, Sharjah, United Arab Emirates

4 Department of Biological Sciences, School of Medical and Life Sciences, Sunway University, 47500 Petaling Jaya, Selangor, Malaysia