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"DIRECT NOSE TO BRAIN DELIVERY OF NEUROTHERAPEUTICS- LOADED NANOEMULSION"

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Abstract:

The blood–brain barrier (BBB) plays a fundamental role in protecting the brain from toxic substances and therefore also controls and restricts the entry of therapeutic agents. The nasal administration of drugs using the nose-to-brain pathway allows direct drug targeting into the brain, avoiding the first-pass effect and bypassing the BBB. Through the nasal route, the drug can access the brain directly along the trigeminal and olfactory nerves, which are located in the upper part of the nasal cavity. Nanoemulsions are formulations belonging to the field of nanomedicine. They consist of emulsions (commonly oil in water) stabilized by one or more surfactants—and eventually co-surfactants—delivered in droplets of small dimensions (sizes of 100–300 nm or less) with a high surface area. A mucoadhesive polymer such as chitosan can be added to the formulation to impair rapid nasal clearance. Nanoemulsions represent promising formulations to deliver drugs directly into the brain through the intranasal route. Therefore, they can be used as a possible alternative to oral administration, avoiding problems such as low solubility in water, poor bioavailability, enzymatic degradation and slow onset of action. This review focuses the present situation in literature regarding the use of Nanoemulsions for nose-to-brain targeting, with particular attention to recent publications. Nasal Nanoemulsions appear to be effective, non-invasive and safe drug delivery systems to achieve brain targeting for the treatment of neurological diseases.

Keywords: *blood-brain barrier; brain targeting; Nanoemulsions; nose-to-brain delivery; nasal mucosa; Olfactory pathway.*

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INTRODUCTION:

The microvasculature of the central nervous system (CNS) is defined as a blood–brain barrier (BBB) because it isolates the brain from the remaining part of the body. CNS vessels, at the levels of arteriole–capillary–venules are non-fenestrated and continuous; they are able to control the exchange of molecules, ions and cells between the blood and the brain [1]. This strong barrier capacity allows the BBB to protect the CNS from pathogens, toxins and external agents. The barrier structure of BBB depends on the specific properties of the brain endothelial cells (BECs) that constitute the walls of the blood vessels. BECs are different from endothelial cells of non-neural tissues—they are polarized cells, held together by tight junctions that strongly limit them paracellular flux of solutes (molecules and ions) and restrict vesicle-mediated transcellular transports (Pinocytosis/transcytosis) .[1,2]. BECs are characterized by two kinds of systems of transport in both directions that include the expression of efflux transporters, whose function is the elimination of the lipophilic toxins able to passively diffuse through the cell membrane, and the expression of influx transporters, which are specific carriers that deliver the nutrients across the BBB into the brain .[1] Among efflux transporters, P-glycoprotein plays an important role, as its activity has been associated with diseases of the CNS such as drug-resistant tumors and epilepsy.[1] BBB is not only a cellular self-defense barrier but also an active interface that strictly controls the CNS microenvironment, allowing its normal neuronal functions. BBB cells are able to communicate with CNS cells and they adapt their behavior to the needs of the CNS, responding to pathological conditions and sometimes becoming a cause of the progression of the disease. [1]. Neurological diseases, such as neuroinfections, Parkinson's disease, Alzheimer's disease, multiple sclerosis, chronic age-related neurodegenerative diseases, cerebral ischemia and so on represent a group of severe pathologies with a broad spectrum of pathological conditions that result in alterations of neural functions. Healthcare statistics confirm that the incidence of CNS diseases is rapidly increasing around the world with higher healthcare costs. [5]. The nose is responsible both for respiration and for olfaction. The human olfactory region, where olfactory and trigeminal nerve terminations are present, occupies 2–12.5 cm2, representing approximately 1.25–10% of the total surface area of the nasal cavity, and it is about 60 _m thick [6]. Olfactory and trigeminal pathways are the only routes by which the brain is connected to the outside environment.[8,9] Thanks to the direct connection provided by the olfactory and trigeminal nerves present between the olfactory epithelium and the brain, drug targeting can be achieved with the administration of formulations onto nasal mucosa [10,12]. Therefore, particular attention must be given to studies of the blood–nerve barrier (BNB), which consists of endoneurial microvessels within the nerve fascicle and the investing perineurium [13, 14]. These microvessels are actively involved in the mechanisms that regulate the permeability of the perineurium and endoneurial capillaries, and surely they play an important role in the passage of substances from olfactory and trigeminal pathways into the CNS. Nose-to-brain drug delivery is a painless, non-invasive administration route that can be used to deliver therapeutic agents into the brain by bypassing the BBB.[10,14,17] These drug administration pathways are characterized by many advantages such as increased patient compliance, high safety, remarkable ease of administration and rapid onset of action, as well as minimized systemic exposure [7] . Furthermore, the use of nasal mucosa as a route of drug administration permits drugs to avoid hepatic first-pass metabolism. Consequently, nasal doses are often 2–10 times lower than oral doses. Direct transport of drug to brain through nasal administration is therefore more promising than oral or intravenous routes of administration [18]. However, despite its numerous advantages, nose-to-brain drug delivery can be limited by possible low bioavailability due to enzymatic degradations of sensitive drugs onto the mucosal surface, high clearance and restrictions determined by the anatomy of the nasal cavity (e.g., small volume, limited surface area of the olfactory mucosa, mucociliary clearance, etc.). These problems should be correctly addressed in designing suitable nose-to-brain formulations (Figure 1). Despite these limitations, examples of promising results are present in clinical trials [19, 20] According to the present literature, different kinds of nanocarriers are used to prepare nasal formulations able to target the brain, constituted by polymer-based and lipid-based nanoparticles [21–23].

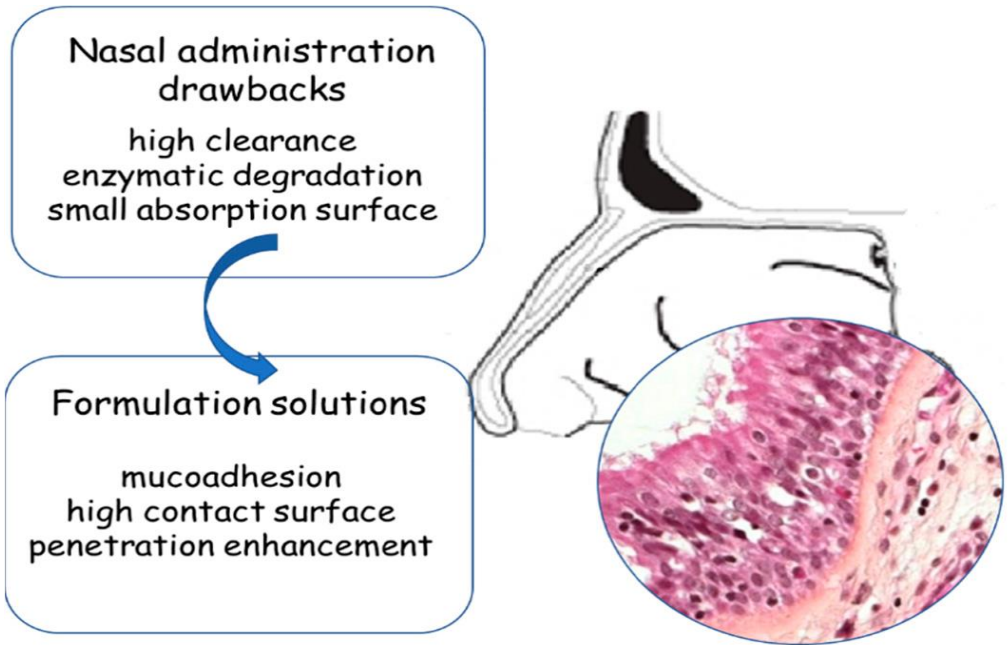

Figure 1. The nose as a route for the administration of drugs.

General Characteristics of NEs in Brief:

Nanoemulsions (NEs) are oil-in-water (O/W) or water-in-oil (W/O) dispersions of two immiscible liquids stabilized using appropriate surfactant(s)[24] , with a mean droplet diameter of about 100 nm ,[25] even if in literature upper size limits up to 300 nm[26]. have been reported. As the size of the droplets is significantly smaller than the wavelength of visible light, NEs have either a transparent or from transparent-to-milky-white appearance[25]. NEs can be formulated into different kinds of dosage forms like liquids, creams, gels, foams, sprays and so on, and can be administered by different routes including oral, parenteral and ocular, in addition to nasal [27] . NEs have droplets of a small size and therefore they are characterized by a higher surface area with respect to other formulations, and by a long-term physical stability, because the small droplet size impairs destabilization phenomena like coalescence, creaming and sedimentation. NEs can be used to solve problems of drug solubility and/or of drug stability (oxidation, pH, hydrolysis and enzymatic degradation at the mucosal level, under physiological conditions) . Hydrophobic drugs are expected to dissolve in the oily phase, and when the drug (dissolved in the oily phase) is released from the NE and comes in contact with the surrounding aqueous environment, a nanoprecipitation can occur. This determines the formation of particles with an enormously high surface and a remarkable improvement of drug dissolution rate, according to the Noyes–Whitney equation [29] . NEs can be used also to mask the bitter or unpleasant taste of drugs and to carry products of natural origin [30,31]. The preparation methods of NEs have been reviewed in exhaustive papers [24,32]. According to these authors, NEs can be prepared through different techniques that can be classified into two broad categories: high-energy methods and low-energy methods (Figure 2). In the case of high-energy methods, such as ultrasonication and high pressure homogenization, the constitution of the small droplets involves a mechanical device that generates disruptive forces breaking up oil and water phases to produce small oil droplets, a process that consumes significant energy. The devices used are microfluidic, ultrasound or high pressure homogenizers [33].

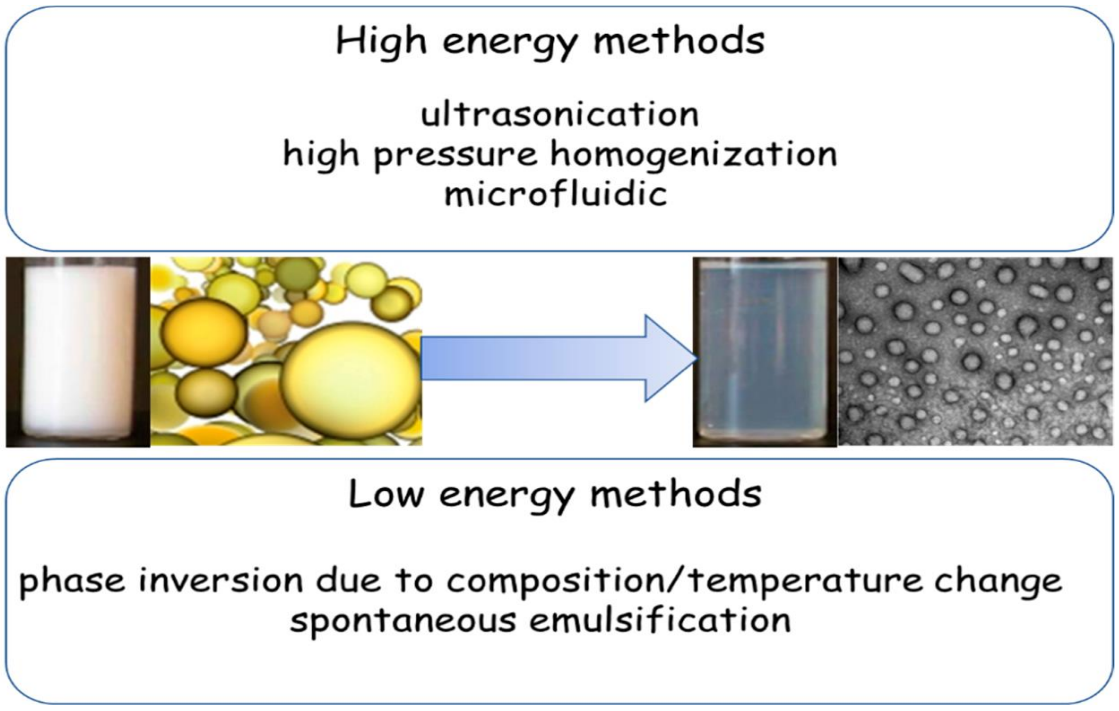

Figure 2. Preparation methods of nanoemulsions (NEs).

The low-energy methods involve specific physico-chemical processes such as phase inversion temperature and emulsion inversion points to make small droplets without consuming significant energy. In the low-energy methods, the droplets are constituted when the system undergoes an inversion of phase in response to changes, such as composition or temperature, and consequently passes through a low interfacial tension state [32]. The quantity of lipidic component(s) used in NEs is dependent on the kind of emulsion (generally they contain 5–20% lipidic droplets in cases of O/W emulsion) and on solubility of the drug to loaded in the system [29] . The solubilizing capacity of the oil phase is a factor that plays an important role in the oil selection because the oil phase must dissolve and maintain dissolved the drug. Furthermore, the highest possible solubility is required to decrease the amount of oil used, reducing the required amounts of surfactants that might determine toxicity problems [34]. Usually the lipids used for the preparation of NEs, alone or in mixture, are fractions of oils of natural origin such as sesame oil, cottonseed oil, soybean oil, coconut oil, and so on that can be classified according to chain lengths as long-chain, medium-chain and short-chain triglycerides [29] . The kind of oil components used in the formulation process of NEs influences (sometimes strongly) the bioavailability of the drug. Many studies have been performed to clarify this aspect for oral NE delivery. For example, it is reported that the bioavailability of curcumin is maximized in NEs made with medium-chain and long-chain triglycerides [35] . Furthermore, it has been reported that several NEs are characterized by a direct lymphatic absorption (avoiding first-pass metabolism) [29] and it is likely that this behavior strongly depends on lipidic phase composition. No data seem available at the momentregarding the relevance of oil choice for nasal and in particular for nose-to-brain administration [37]. Emulsifiers are needed to facilitate NE formation and to ensure its kinetic stability during storage, even if NEs typically require less surfactant than other colloidal dispersions. The emulsifier used for the preparation of NEs is very often a surfactant, but proteins and lipids have also been used. Lecithin (phosphatidylcholine) [36] , sodium deoxycholate (bile salt) [37] , polyoxyethlene sorbitan monolaurate (Tween 20, 40, 60, 80)[39] and sorbitan monolaurate (Span 20, 40, 60, 80) [38]are commonly employed in the preparation of NEs. Other surfactants proposed are poloxamers ,[40] sodium dodecyl sulfate[41] , amphiphilic proteins like casein ,[42] polysaccharides (starch derivatives, gums) and poly-ethylene-glycol (PEG)-containing block copolymers .[44] Co-surfactants, such as polyethylene glycol, ethylene glycol, propylene glycol, ethanol and glycerine can be used for the stabilization of NEs . All of these can be used alone or in combination.

The selection of the surfactants is critical, as they must be safe at the concentrations used. The choice of the surfactant or of the surfactant blend influences both the size of the droplets and the stability of the NE. However, it must be taken into account that this choice can influence pharmacokinetics and toxicity; for example, poloxamer 188 has renal toxicity at a concentration higher than 0.5% in parenteral NEs [29].

Generally speaking, these formulations appear safe because, to our knowledge, no mutagenic effects are reported in literature, and only safe pharmaceutical excipients have been used for their preparation. In the present literature there is some confusion regarding the classification of NEs because they are sometimes confused with microemulsions, which are thermodynamically stable systems that form spontaneously [24,46] . For this reason, macroemulsions and NEs can be prepared with high- and low-energy methods, while microemulsions are prepared with low-energy methods only. Important differences can be found between classical macroemulsions (classical emulsions), NEs and microemulsions in droplet size range and stability characteristics [32,47].

General Overview of NEs for Nose-to-Brain Delivery:

The easiest classification of NEs designed for nose-to-brain delivery is based on the drug loaded and the therapeutic purpose. As the final target is the brain through the nose, the pharmacological actions regard pathologies of the CNS. In one case, a probe is loaded in the NE to obtain brain imaging. The formulations used in the intranasal administrations of drugs are always, in our knowledge, O/W emulsions. A general overview of the present literature about NEs for nose-to-brain targeting shows clearly that intranasal use is often an alternative to the oral therapy. In fact, if the drug is administered orally to reach the brain, this kind of administration can present problems for some drugs, which are summarized in Table 1. CNS delivery through the nasal mucosa sometimes performs better than parenteral administration as well, as shown by in vivo experiments. One of the first examples in the literature of the use of NEs to reach the brain through the administration onto nasal mucosa is a paper by Kumar et al. in which NEs were utilized to carry risperidone, an antipsychotic drug belonging to the group of benzisoxazole derivatives [48]. According to the authors' opinion, these positive results were related to the enhancing of the nasal retention time due to the presence of chitosan, thus confirming the importance of this polymer as mucoadhesive agent in nasal formulations [50].

Table 1. Characteristics that make the central nervous system (CNS) drug a candidate for nasal NE, as an alternative to oral administrations.

Low capacity to cross the Blood-Brain-Barrier
Low solubility in water (and consequently poor bioavailability)
Low/irregular absorption (and consequently poor bioavailability)
Active substrate of intestinal P-glycoprotein
Problems of stability (pH, hydrolysis, oxidation, under gastro-intestinal conditions)
Intestinal metabolism (enzymatic degradation)
First-pass metabolism (enzymatic degradation)
Need to reduce dosage (i.e., in case of chronic therapy and/or to avoid side effects connected to the high oral
dosage used for overcoming the Blood-Brain-Barrier)
Slow onset of action
Bitter/unpleasant taste of the drug

An important application of nose-to-brain delivery with NEs was that described by Mahajan et al., who used NEs to carry anti-HIV drugs [28] . It is known that after the initial infection, CNS is the region in which HIV viruses constitute a sort of "anatomic reservoir", and from which they can reactivate the infection. Furthermore, the brain infection from HIV can determine neuro-AIDS, a form of dementia and cognitive impairment. It is clear that improved drug delivery to the CNS will reduce the possibility of underlying persisting infections. Saquinavir mesylate is a protease inhibitor with activity against HIV-Type 1 (HIV-1). However, its bioavailability is low, owing to its low solubility in water. Furthermore, saquinavir permeability through the BBB is poor and is a P-glycoprotein and cytochrome P450 substrate. For all

these reasons, nasal O/W NEs containing saquinavir mesylate were prepared by the spontaneous emulsification technique using Capmul MCM, a mono-diglyceride of medium-chain fatty acids (mainly caprylic and caproic). NEs were characterized in terms of drug content, droplet size and zeta potential. Ex vivo permeation studies were carried out using excised fresh sheep nasal mucosa. NEs showed an increase in drug permeation compared to plain drug suspension. Cilia toxicity was low. In vivo biodistribution studies, carried out after nasal administration of 99mTc formulations, showed higher drug concentration in the brain after nasal administration of NE with respect to intravenous administration. Gamma scintigraphy imaging of a rat brain demonstrated increased drug transport to the CNS after NE nasal administration. One of the most recent papers published on the use of NEs for nose-to-brain delivery is by Haider et al., who prepared rivastigmine NEs for enhanced brain delivery through nasal administration .[53] Rivastigmine hydrochloride is a drug that has been proposed for the therapy of Alzheimer's disease, as it helps in prevention of acetylcholine hydrolysis through the enzymes acetylcholinesterase and butyrylcholinesterase and thereby increases central cholinergic function and availability of acetylcholine. Box–Behnken design was used to statistically optimize the formulation parameters for the preparation of NEs. Ex vivo diffusion studies were carried out using Franz diffusion cells through freshly excised goat nasal mucosa. In vivo studies were carried out on Wistar rats. For the preparation of NEs, the selection of the oils led to the choice of Capmul MCM, whereas among surfactants Tween 80 and among co-surfactants Transcutol-P were chosen. Ex vivo diffusion studies done on goat nasal mucosa showed that the cumulative amount of drug permeation through nasal mucosa for NE was higher than that of the drug solution used as a comparison. In vivo studies showed that the brain drug concentration after nasal administration of NEs was significantly higher than that achieved after nasal administration of solutions and intravenous administration of NEs. NEs loaded with chloramphenicol, based on palm kernel oil esters and suitable for meningitis treatment have been described [54,55] , even if no specific indication about the possible nasal use of these formulations has been given. NEs containing amiloride have been prepared and administered intranasally in mice. Oleic acid, Tween 20 and Carbitol were employed as oil, surfactant and co-surfactant, respectively. For the emulsion preparation, a high-energy ultra-sonication method was used. An enhanced drug brain bioavailability was found as compared to intravenous administration [56]. Lastly, a paper by El-Zaafarany et al. describes lipid-based nanovectors defined as emulsomes and containing oxcarbazepine [57] . These dispersed systems can be considered the borderline between NEs and Solid Lipid Nanoparticles (SLNs). In fact, according to the authors, in the emulsomes the dispersed phase is constituted by "particles" comprising lipid cores stabilized by phospholipid layer(s) that surrounds the lipid core at the aqueous interface. The lipidic core of these particles is in a solid phase, rather than oil in a fluid phase. Oxcarbazepine is a drug used for the treatment of epilepsy. The aim of this work was the preparation of this lipidic nanocarrier aimed at a direct nose-to-brain targeting. Ex vivo studies carried out using rat nasal mucosa to check possible changes induced by Emulsomes showed no histopathological alteration. Pharmacokinetic studies carried out in rats showed that oxcarbazepine-loaded emulsomes slowed drug elimination, and a high drug concentration was achieved in the blood stream for a longer time periods, as compared to the free drug administrations. In Table 2 the schematic classification based on drug loaded in NEs is reported.

Table 2. Nanoemulsions designed for nose-to-brain delivery, and classification based on the drug.

Drug/Probe	**Therapy of**
Risperidone	Schizophrenia
Olanzapine	Schizophrenia
Ergoloid mesylate	Antiaging
Amiloride	Antiepileptic
Ziprasidone hydrochloride	Antipshychotic
Saquinavir mesylate	HIV infections
Nimodipine	Cerebrovascular spasm and senile dementia
Curcumin	Neurodegenerative diseases
Resveratrol	Parkinson's disease
Curcumin/Resveratrol	Age-related neurodegenerative diseases

Cyclosporine-	A Neuro-protective
anti-TNF_	RNA Neuro-inflammations
Riluzole Amyotrophic	lateral sclerosis (ALS)
Selegiline	Parkinson's disease
Thymoquinone	Cerebral ischemia
Paroxetine	Depression and anxiety
Quetiapine	Schizophrenia
Zolmitriptan	Migraines
Aggregation-	caused quenching (ACQ) probes Labeling action
Kaempferol	Gliomas
Rivastigmine	Alzheimer's disease
Chloramphenicol	Bacterial meningitis
Amiloride	Epilepsy
Oxcarbazepine	Epilepsy

CONCLUSIONS:

Nanoemulsions are formulations that are more and more important in the field of nanomedicine. Their characteristics (nanodroplets with a high surface) make them suitable for nose-to-brain delivery. Mucoadhesive polymers can be added in their composition to slow down nasal clearance. The presence of chitosan as an additional excipient plays a double role, because it is mucoadhesive and has penetration enhancing properties on nasal mucosa. As shown by Table 2, there are many examples in the literature of recent years of nanoemulsionloaded drugs with different therapeutic goals in brain diseases. The pathologies are all important and serious; many of these diseases, if not treated effectively, can reduce the quality of life or even lead to death. The nose-to-brain delivery is often an alternative to oral therapies for the CNS that can present problems, usually related to the characteristics of the drug. There are many reasons a CNS drug can be a good candidate for intranasal nanoemulsion, as an alternative to the oral administration (Table 1). However, as demonstrated by the reported literature, intranasal administrations of NEs often lead to better results, also with respect to intravenous administrations. These good results can be explained by mechanisms of transcytosis/endocytosis of the nanodroplets by the brain endothelial cells. Moreover, the surfactant(s) present in the nanoemulsions could have a fluidizing effect on endothelial cell membranes, determining an enhanced drug permeability and favoring by this mechanism the olfactory and trigeminal pathways. Nanoemulsions for nasal administration represent a promising strategy for nose-to-brain drug delivery and to achieve CNS targeting for the treatment of neurodiseases. However, clinical studies of these formulations are still needed to demonstrate their appropriateness in clinical practice. Many efforts must be made to further improve the performances of nanoemulsions. Future perspectives could consider the use of additional excipients. For instance, it is known that cyclodextrins can be used for the preparation of formulations able to cross the BBB, administered through nasal or oral/parenteral routes [16,23,56,57] . New penetration enhancers and modulators of cytochrome P450 Could be proposed to improve the selectivity and entity of penetration through the BBB .

REFERENCES:

1. Daneman, R.; Prat, A. The blood-brain barrier. Cold Spring Harb. Perspect. Biol. **2015**, 7, a020412.
2. Gloor, S.M.; Wachtel, M.; Bolliger, M.F.; Ishihara, H.; Landmann, R.; Frei, K. Molecular and cellular permeability control at the blood-brain barrier. Brain Res. Brain Res. Rev. **2001**, 36, 258–264.
3. Zlokovic, B.V. The blood-brain barrier in health and chronic neurodegenerative disorders. Neuron **2008**, 57, 178–201.
4. Banks,W.A. From blood-brain barrier to blood-brain interface: New opportunities for CNS drug delivery. Nat. Rev. Drug Discov. **2016**, 15, 275–292.
5. Kaushik, A.; Jayant, R.D.; Bhardwaj, V.; Nair,M. Personalized nanomedicine for CNS diseases. Drug Discov. Today **2018**, 23, 1007–1015.
6. Dhuria, S.V.; Hanson, L.R.; Frey, W.H., 2nd. Intranasal delivery to the central nervous system: Mechanisms and experimental considerations. J. Pharm. Sci. **2010**, 99, 1654–1673.
7. Bourganis, V.; Kammona, O.; Alexopoulos, A.; Kiparissides, C. Recent advances in carrier mediated nose-to-brain delivery of pharmaceutics. Eur. J. Pharm. Biopharm. **2018**, 128, 337–362.

8. Thorne, R.G.; Pronk, G.J.; Padmanabhan, V.; Frey,W.H. Delivery of insulin-like growth factor-I to the rat brain and spinal cord along olfactory and trigeminal pathways following intranasal administration. Neuroscience **2004**, 127, 481–496.
9. Mistry, A.; Stolnik, S.; Illum, L. Nanoparticles for direct nose-to-brain delivery of drugs. Int. J. Pharm. **2009**, 379, 146–157.
10. Alam, M.I.; Beg, S.; Samad, A.; Baboota, S.; Kohli, K.; Ali, J.; Ahuja, A.; Akbar, M. Strategy for effective brain drug delivery. Eur. J. Pharm. Sci. **2010**, 40, 385–403.
11. Pardeshi, C.V.; Belgamwar, V.S. Direct nose to brain drug delivery via integrated nerve pathways bypassing the blood-brain barrier: An excellent platform for brain targeting. Expert Opin. Drug Deliv. **2013**, 10, 957–972.
12. Dalpiaz, A.; Fogagnolo, M.; Ferraro, L.; Capuzzo, A.; Pavan, B.; Rassu, G.; Salis, A.; Giunchedi, P.; Gavini, E. Nasal chitosan microparticles target a zidovudine prodrug to brain HIV sanctuaries. Antiviral Res. **2015**, 123, 146–157.
13. Weerasuriya, A.;Mizisin, A.P. The blood-nerve barrier: Structure and functional significance.MethodsMol. Biol. **2011**, 686, 149–173.
14. Gavini, E.; Rassu, G.; Ferraro, L.; Beggiato, S.; Alhalaweh, A.; Velaga, S.; Marchetti, N.; Bandiera, P.; Giunchedi, P.; Dalpiaz, A. Influence of polymeric microcarriers on the in vivo intranasal uptake of an anti-migraine drug for brain targeting. Eur. J. Pharm. Biopharm. **2013**, 83, 174–183.
15. Sharma, D.; Sharma, R.K.; Sharma, N.; Gabrani, R.; Sharma, S.K.; Ali, J.; Dang, S. Nose-To-Brain Delivery of PLGA-Diazepam Nanoparticles. AAPS PharmSciTech. **2015**, 16, 1108–1121.
16. Yalcin, A.; Soddu, E.; Turunc Bayrakdar, E.; Uyanikgil, Y.; Kanit, L.; Armagan, G.; Rassu, G.; Gavini, E.; Giunchedi, P. Neuroprotective Effects of Engineered Polymeric Nasal Microspheres Containing Hydroxypropyl-_-cyclodextrin on _-Amyloid (1-42)-Induced Toxicity. J. Pharm. Sci. **2016**, 105, 2372–2380.
17. Rassu, G.; Porcu, E.P.; Fancello, S.; Obinu, A.; Senes, N.; Galleri, G.;Migheli, R.; Gavini, E.; Giunchedi, P. Intranasal Delivery ofGenistein-LoadedNanoparticles as a Potential Preventive System against NeurodegenerativeDisorders. Pharmaceutics **2019**, 111, 8.
18. Ugwoke, M.I.; Verbeke, N.; Kinget, R. The biopharmaceutical aspects of nasal mucoadhesive drug delivery. J. Pharm. Pharmacol. **2001**, 53, 3–21.
19. Quintana, D.S.; Westlye, L.T.; Rustan, Ø.G.; Tesli, N.; Poppy, C.L.; Smevik, H.; Tesli, M.; Røine, M.; Mahmoud, R.A.; Smerud, K.T.; et al. Low-dose oxytocin delivered intranasally with Breath Powered device affects social-cognitive behavior: A randomized four-way crossover trial with nasal cavity dimension assessment. Transl. Psychiatry **2015**, 5, e602.
20. Stützle, M.; Flamm, J.; Carle, S.; Schindowski, K. Nose-to-brain delivery of insulin for Alzheimer's disease. Admet Dmpk. **2015**, 3, 190–202.
21. Rassu, G.; Soddu, E.; Posadino, A.M.; Pintus, G.; Sarmento, B.; Giunchedi, P.; Gavini, E. Nose-to-brain delivery of BACE1 siRNA loaded in solid lipid nanoparticles for Alzheimer's therapy. Colloids Surf. B Biointerfaces **2017**, 152, 296–301.
22. Sonvico, F.; Clementino, A.; Buttini, F.; Colombo, G.; Pescina, S.; Stanisçuaski Guterres, S.; Raffin Pohlmann, A.; Nicoli, S. Surface-Modified Nanocarriers for Nose-to-Brain Delivery: From Bioadhesion to Targeting. Pharmaceutics **2018**, 10, 34.
23. Rassu, G.; Gavini, E.; Carta, A.; Obinu, A.; Porcu, E.P.; Giunchedi, P. Hydroxypropyl-_-Cyclodextrin Formulated in Nasal Chitosan Microspheres as Candidate Therapeutic Agent in Alzheimer's Disease.Curr. Drug Deliv. **2018**, 15, 746–748.
24. Comfort, C.; Garrastazu, G.; Pozzoli, M.; Sonvico, F. Opportunities and challenges for the nasal administration of nanoemulsions. Curr. Top. Med. Chem. **2015**, 15, 356–368.
25. Rodrigues, R.F.; Costa, I.C.; Almeida, F.B.; Cruz, A.S.; Ferreira, A.M.; Vilhena, J.C.E.; Florentino, A.C.; Carvalho, J.C.T.; Fernandes, C.P. Development and characterization of evening primrose (Oenothera biennis) oil nanoemulsions. Revista Brasileira de Farmacognosia **2015**, 25, 422–425.
26. Anton, N.; Benoit, J.P.; Saulnier, P. Design and production of nanoparticles formulated from nano-emulsion templates-A review. J. Control. Release **2008**, 128, 185–199.
27. Sarker, D.K. Engineering of nanoemulsions for drug delivery. Curr. Drug Deliv. **2005**, 2, 297–310.
28. Mahajan, H.S.; Mahajan, M.S.; Nerkar, P.P.; Agrawal, A. Nanoemulsion-based intranasal drug delivery system of saquinavir mesylate for brain targeting. Drug Deliv. **2014**, 21, 148–154.
29. Singh, Y.; Meher, J.G.; Raval, K.; Khan, F.A.; Chaurasia, M.; Jain, N.K.; Chourasia, M.K.

Nanoemulsion: Concepts, development and applications in drug delivery. J. Control. Release **2017**, 252, 28–49.
30. Bonferoni, M.C.; Sandri, G.; Rossi, S.; Usai, D.; Liakos, I.; Garzoni, A.; Fiamma, M.; Zanetti, S.; Athanassiou, A.; Caramella, C.; Ferrari, F. A novel ionic amphiphilic chitosan derivative as a stabilizer of nanoemulsions: Improvement of antimicrobial activity of Cymbopogon citratus essential oil. Colloids Surf. B Biointerfaces **2017**, 152, 385–392.
31. Bonferoni, M.C.; Riva, F.; Invernizzi, A.; Dellera, E.; Sandri, G.; Rossi, S.; Marrubini, G.; Bruni, G.; Vigani, B.; Caramella, C.; et al. Alpha tocopherol loaded chitosan oleate nanoemulsions for wound healing. Evaluation on cell lines and ex vivo human biopsies, and stabilization in spray dried Trojan microparticles. Eur. J. Pharm. Biopharm. **2018**, 123, 31–41.
32. Gupta, A.; Eral, H.B.; Hatton, T.A.; Doyle, P.S. Nanoemulsions: Formation, properties and applications. Soft Matter. **2016**, 12, 2826–2841.
33. Prakash, R.T.; Thiagarajan, P. Nanoemulsion for drug delivery through different routes. Res. Biotechnology. **2011**, 2, 1–13.
34. Mahmoud, H.; Al-Suwayeh, S.; Elkadi, S. Design and optimization of self nanoemulsifying drug delivery systems of simvastatin aiming dissolution enhancement. Afr. J. Pharm. Pharmacol. **2013**, 7, 1482–1500.
35. McClements, D.J.; Xiao, H. Potential biological fate of ingested nanoemulsions: Influence of particle characteristics. Food Funct. **2012**, 3, 202–220.
36. Klang, V.; Valenta, C. Lecithin-based nanoemulsions. J. Drug Deliv. Sci. Technol. **2011**, 21, 55–76.
37. Vyas, T.K.; Shahiwala, A.; Amiji, M.M. Improved oral bioavailability and brain transport of saquinavir upon administration in novel nanoemulsion formulations, Int. J. Pharm. **2008**, 347, 93–101
38. Azeem, A.; Rizwan, M.; Ahmad, F.J.; Iqbal, Z.; Khar, R.K.; Aqil, M.; Talegaonkar, S. Nanoemulsion components screening and selection: A technical note. AAPS PharmSciTech **2009**, 10, 69–76.
39. Jadhav, C.; Kate, V.; Payghan, S.A. Investigation of effect of non-ionic surfactant on preparation of griseofulvin non-aqueous nanoemulsion. J. Nanostruct. Chem. **2015**, 5, 107–113.
40. Brüsewitz, C.; Schendler, A.; Funke, A.; Wagner, T.; Lipp, R. Novel poloxamer-based nanoemulsions to enhance the intestinal absorption of active compounds. Int. J. Pharm. **2007**, 329, 173–181.
41. Erramreddy, V.V.; Ghosh, S. Influence of emulsifier concentration on nanoemulsion gelation. Langmuir **2014**, 30, 11062–11074.
42. Maher, P.G.; Fenelon, M.A.; Zhou, Y.; Kamrul Haque, M.; Roos, Y.H. Optimization of _-casein stabilized nanoemulsions using experimental mixture design. J. Food Sci. **2011**, 76, C1108–C1117.
43. Combrinck, J.; Otto, A.; du Plessis, J. Whey protein/polysaccharide-stabilized emulsions: Effect of polymer type and pH on release and topical delivery of salicylic acid. AAPS PharmSciTech **2014**, 15, 588–600.
44. Rapoport, N.; Christensen, D.A.; Kennedy, A.M.; Nam, K.H. Cavitation properties of block copolymer stabilized phase-shift nanoemulsions used as drug carriers. Ultrasound Med. Biol. **2010**, 36, 419–429.
45. Zhang, Y.; Shang, Z.; Gao, C.; Du, M.; Xu, S.; Song, H.; Liu, T. Nanoemulsion for solubilization, stabilization, and in vitro release of pterostilbene for oral delivery. AAPS PharmSciTech **2014**, 15, 1000–1008.
46. Langasco, R.; Tanrıverdi, S.T.; Özer, Ö.; Roldo, M.; Cossu, M.; Rassu, G.; Giunchedi, P.; Gavini, E. Prolonged skin retention of clobetasol propionate by bio-based microemulsions: A potential tool for scalp psoriasis treatment. Drug Dev. Ind. Pharm. **2018**, 44, 398–406.
47. Abdou, E.M.; Kandil, S.M.; Miniawy, H.M.F.E. Brain targeting efficiency of antimigrain drug loaded mucoadhesive intranasal nanoemulsion. Int. J. Pharm. **2017**, 529, 667–677.
48. Kumar, M.; Misra, A.; Babbar, A.K.; Mishra, A.K.; Mishra, P.; Pathak, K. Intranasal nanoemulsion based brain targeting drug delivery system of risperidone. Int. J. Pharm. **2008**, 358, 285–291.
49. Kumar, M.; Misra, A.; Mishra, A.K.; Mishra, P.; Pathak, K. Mucoadhesive nanoemulsion-based intranasal drug delivery system of olanzapine for brain targeting. J. Drug Target. **2008**, 16, 806–814.
50. Rassu, G.; Soddu, E.; Cossu, M.; Gavini, E.; Giunchedi, P.; Dalpiaz, A. Particulate formulations based on chitosan for nose-to-brain delivery of drugs. A review. J. Drug Del. Sci. Technol. **2016**, 32, 77–87.
51. Yu, C.; Meng, J.; Chen, J.; Tang, X. Preparation of ergoloid mesylate submicron emulsions for enhancing nasal absorption and reducing nasal ciliotoxicity. Int. J. Pharm. **2009**, 375, 16–21.
52. Jain, N.; Akhter, S.; Jain, G.K.; Khan, Z.I.; Khar, R.K.; Ahmad, F.J. Antiepileptic intranasal

Amiloride loaded mucoadhesive nanoemulsion: Development and safety assessment. J. Biomed. Nanotechnol. **2011**, 7, 142–143.
53. Haider, F.; Khan, S.; Gaba, B.; Alam, T.; Baboota, S.; Ali, J.; Ali, A. Optimization of rivastigmine nanoemulsion for enhanced brain delivery: In-vivo and toxicity evaluation. J. Mol. Liquids **2018**, 255, 384–396.
54. Musa, S.H.; Basri, M.; Masoumi, H.R.; Karjiban, R.A.; Malek, E.A.; Basri, H.; Shamsuddin, A.F. Formulation optimization of palm kernel oil esters nanoemulsion-loaded with chloramphenicol suitable for meningitis treatment. Colloids Surf. B Biointerfaces **2013**, 112, 113–119.
55. Pécoul, B.; Varaine, F.; Keita, M.; Soga, G.; Djibo, A.; Soula, G.; Abdou, A.; Etienne, J.; Rey, M. Long-acting chloramphenicol versus intravenous ampicillin for treatment of bacterial meningitis. Lancet **1991**, 338, 862–866.
56. Ahmad, N.; Ahmad, R.; Alam, M.A.; Ahmad, F.J.; Amir, M. Impact of ultrasonication techniques on the preparation of novel Amiloride-nanoemulsion used for intranasal delivery in the treatment of epilepsy. Artif. Cells Nanomed. Biotechnol. **2018**, 23, 1–16.
57. El-Zaafarany, G.M.; Soliman, M.E.; Mansour, S.; Awad, G.A. Identifying lipidic emulsomes for improved oxcarbazepine brain targeting: In vitro and rat in vivo studies. Int. J. Pharm. **2016**, 503, 127–140.