Design of polymeric-based nanoparticles for treatment of skin conditions: atopic dermatitis and acne vulgaris.

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INTRODUCTION AND OBJECTIVE

Skin diseases need long-term treatment and have a great impact on patients' life and a social stigma raised by the presence of visible skin marks. Conventional treatment options, based on topical corticosteroids for AD, and antimicrobial/ anti-inflammatory drugs for AV - as well as non-pharmaceutical therapies (*e.g.* emollients and ointments) - help to mitigate the symptoms; however, they are not highly effective, need frequent applications and, in the case of the topical corticosteroids, are associated with several local and systemic side effects (Ference 2009; Jih 2007). Advances in nanotechnology and drug delivery outstood the use of nano-scale particles as vehicles and revolutionized topical treatments (Prow, 2011). Particularly, our aim is to design innovative nanoparticles, based on polymers such as poly(lactic-co-glycolic acid) (PLGA) and poly (Ɛ-caprolactone) (PCL) to treat patients' with problematic skin conditions. With these systems we expect to improve drug residence in the skin and a targeting delivery to specific cell populations in the skin. Hydrocortisone acetate (HCA) and azelaic acid (AzA) were chosen as our models drugs to conduct these studies.

MATERIALS AND METHODS

HCA-loaded PCL nanoparticles were prepared by a modified solvent displacement method previously described (Alvarez-Román 2004) and AzA-loaded PLGA nanoparticles were prepared by a modified-spontaneous emulsification solvent diffusion method (Reis 2013). All systems were characterized in terms of mean particle size, polydispersity index (PI) and zeta potential using a Coulter Nanosize Delsa Nano™C (Fullerton, CA, USA). The morphological assay of the HCA-loaded nanoparticles was conducted by TEM (CM12, Philips, the Netherlands) and SEM (JEOL 5200LV, Tokyo, Japan) for AzA-loaded nanoparticles. After recovery, HCA and AzA encapsulation efficiency (EE) were determined by HPLC (Hájková, 2003; Ibrahierm 2002). *In vitro* drug release and permeation studies with Franz Cells, DSC (Q20, TA Instruments, New Castle, DE, USA), FTIR (Bruker Spectrometer IFS-66V, Karlsruche, Germany) were also conducted in our studies.

In vitro toxicity studies were performed with *Saccharomyces cerevisiae*; *in vitro* efficiency studies with *Staphylococcus epidermidis* and *Propionibacterium acnes* were also performed for the AzA-loaded nanoparticles. All experiments were made in triplicate. Finally, HCA-loaded nanoparticles and AzA-loaded nanoparticles were incorporated into topical formulations (hydrophobic ointment and Carbopol 940 gel, respectively) and occlusive patch tests for excipients' safety were conducted in human volunteers for 48h, according to the Declaration of Helsinki and after approval of the local Ethics Committee. Statistical analysis was conducted with Graph Pad Prism 5 Software (San Diego, CA, USA); $p < 0.05$ was considered statistically significant and the confidence level used was 95%.

RESULTS AND DISCUSSION

Table 1: Main results for HCA-loaded and AzA-loaded nanoparticles.

	HCA-loaded nanoparticles	**AzA-loaded nanoparticles**
Mean Particle Size (nm) [PI]	258.4 ± 24.5 [0.084]	378.6 ± 60.8 [0.094]
Zeta potential (mV)	-4.39 ± 0.62	-7.82 ± 9.01
EE (%)	36%	76%
Morphology	Spherical	Spherical
FTIR	Possible drug-polymer hydrogen bonding	-
DSC	-	Possible strong drug-polymer interactions
In vitro drug release studies	~ 7% in 24h (PBS pH 7.4)	~ 80% in 24h (PBS pH 5.5)
In vitro permeation studies	Small flux (5.35 ± 0.37 µg/cm^2/h) and drug retention	-
In vitro efficacy studies (*P.acnes* and *S. epidermidis*)	-	Similar activity to free drug
In vitro toxicity studies (*S. cerevisiae*)	Concentration-time dependent	Concentration-time dependent
In vivo excipients safety studies	No clinical reactions (n=10)	No clinical reactions (n=12)

Results of the two main nanosystems studied are briefly presented in table 1. The polymeric nanoparticles showed intrinsic differences allied with the polymers' nature and the drugs' properties. Both nanoparticles demonstrated to be stable and the slightly negative zeta potential, which may interact with the described positive charge (+ 23 mV) of the skin (Morykwas 1987). Chemical analysis may indicate that both systems are stabilized by electrostatic interactions and possible stronger interactions between drug and polymer. Still, each one is tailored for a different approach: HCA-loaded nanoparticles are developed for a highly controlled release and retention at the skin epidermis and dermis, to reduce the severe side effects and absorption, while AzA-loaded nanoparticles are suitable for a targeting delivery and retention inside the pilosebaceous follicles to conduct an effective antimicrobial and anti-inflammatory activity.

CONCLUSIONS

Overall conclusions demonstrate that PLGA and PCL nanoparticles are promising and safe systems to deliver corticosteroids and antimicrobials into the skin. Further studies should assess the effects of the long-term application of the described therapeutics.

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