

Next generation ion pairs for the delivery of diclofenac

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Overview

- Why?
 - Background
- How?
- The skin as a drug delivery route
 - What are ion pairs
 - Factors affecting the choice of counter ions
- Detection and quantification of API (DF)
 - Confirmation of non-interference of potential counter ions
- Partition coefficient experiments
- Application: permeation studies with mass balance
- Conclusion

Background

Diclofenac is one of the most prescribed and effective NSAIDs, used both orally and topically

As a topical product: one of the main treatment options for the condition osteoarthritis

Osteoarthritis affects 7% of global population > 500 million people: Global burden of disease study (2019)

DF in environment and detected in drinking water

Can we reduce the amount of DF used in a topical product without compromising the efficacy of the product “ (using an ion pairing as a technique)

The skin as a drug delivery route

- The skin and in particular the SC provide **very effective barriers to any exogenous substances**
- In general, only a small % of drug applied to the skin permeates, as the majority of pharmaceutical compounds are ionized at physiological pH.
- As the skin is a lipidic environment, it is not easy for ionized substances to pass the skin (SC) and reach the bloodstream
- Numerous strategies have evolved to increase the delivery of topically applied drugs, which can be divided into

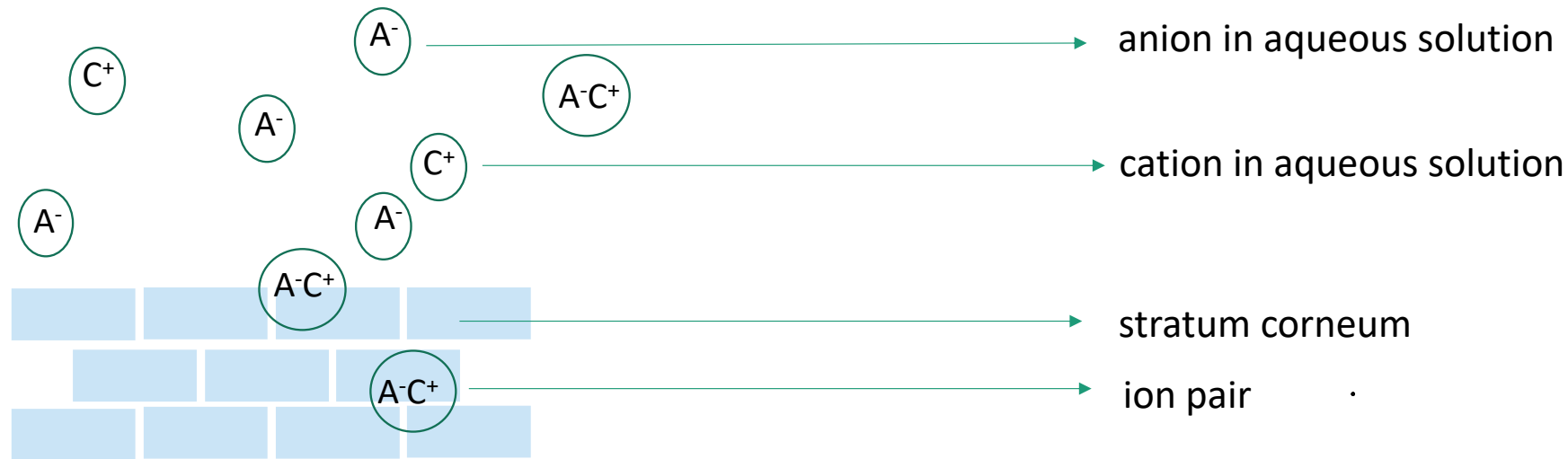
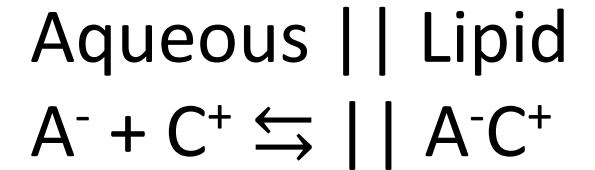
Active methods

- Using more physical means and sometimes equipment egs iontophoresis, phonophoresis and also microneedles

Passive methods

- Focus on the formulation (API and its excipients) egs: supersaturation of active, use of permeation enhancers and **ion pairing**

What are ion pairs?



Oppositely charged ions are attracted to each other



If electrostatic energy of attraction > mean thermal energy, it draws them closer together



If they achieve a critical distance, charges are masked = ion pair



ion pair can partition into a lipid environment



What factors affect the choice of counter ions?

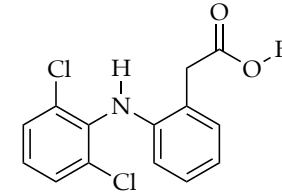
Factors affecting choice of counter ions

Considerations

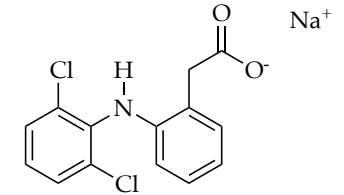
- Size
- Oppositely charged at physiological pH (DF negatively charged)
- Non - toxic

Potential ion pairs: L-arginine, L-histidine, L-lysine & salts

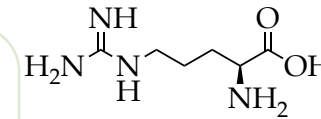
- Size: all smaller than DF / DNa
- Oppositely charged at physiological pH (isoelectric points i.e. points where the AAs have no net charge, exceed physiological pH)
- Toxicity: GRAS by FDA
- Bacterial fermentation production method, low environmental impact, sustainable



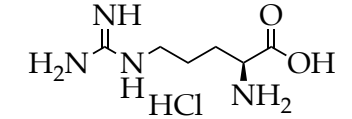
Diclofenac



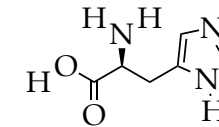
Diclofenac Sodium



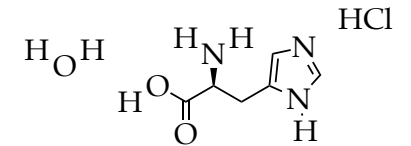
L-Arginine



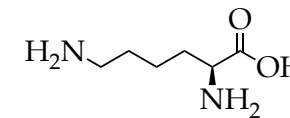
L-Arginine Monohydrochloride



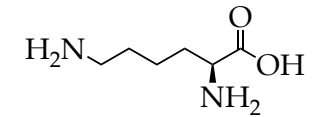
L-Histidine



L-Histidine Hydrochloride Monohydrate



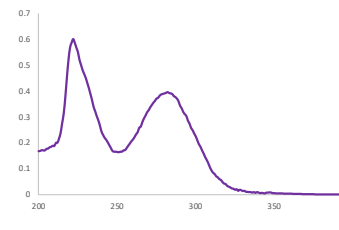
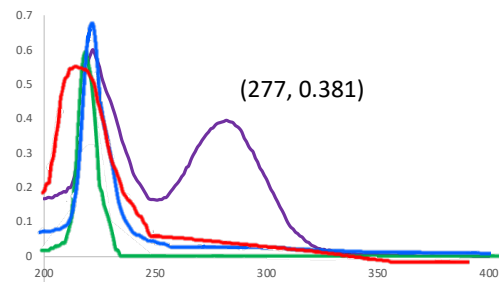
L-Lysine



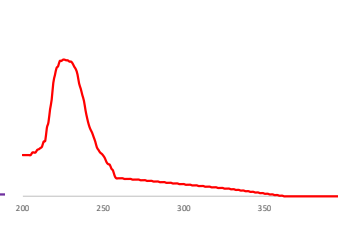
L-Lysine Monohydrochloride

Detection and quantification of DF

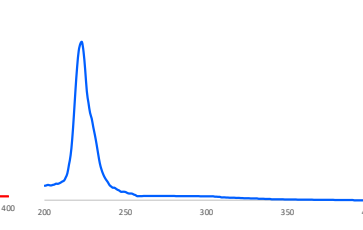
HPLC details	Agilent 1100 series including:
	G1379A Degasser
	G1311A Quaternary Pump
	G1313A Autosampler
	G1316 Column Compartment
Column	Shiseido
	C18, 250 x 4.6 mm
Mobile phase (v/v)	Acetonitrile 70%
	0.1% Trifluoroacetic acid in water (H ₂ O) 30%
Temperature ° C	25
Flow rate (mL/ min)	1 mL / min
Wavelength (nm)	277
Run time (min)	6
Injection volume (μL)	10



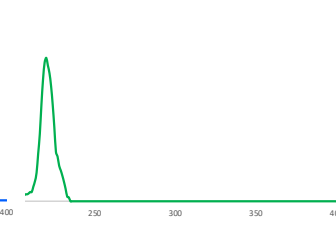
Diclofenac sodium



L-Arginine



L-Histidine



L-Lysine

Primary aim:
Detection and
quantification of
DF



Considerations:
± 0.1 μg - 400 μg
range
Large number of
samples
No interference
from counter ions



HPLC was chosen
as the analytical
device: deal with
large number of
samples,
detection of DF at
a range of
concentrations



Non-interference:
absorption
spectra for DF and
the AA counter
ions



This was tested
making up
solutions of DF
with and without
the CI at both 1:1
and 1:50 molar
ratios.

Partition coefficient studies

PC studies were used to select final candidates for permeation studies.

If successful: apparently neutral species partitions from aqueous into organic layer

DF, a weak acid, was combined with basic amino acids, while Diclofenac sodium (DNa) was combined with their salts at different ratios (n=3).

DNa combined with L-histidine salt (L-HSS) was able to increase the PC of DF > 8 times

FREE ACID PLUS FREE BASE

DFA: L - Arginine				DFA: L - Histidine				DFA: L - Lysine			
	Ratio	PC Mean	SD		Ratio	PC Mean	SD		Ratio	PC Mean	SD
D:A	1:0	14.36	0.27	D:H	1:0	14.21	0.36	D:L	1:0	15.24	0.33
D:A	1:0.5	12.85	0.07	D:H	1:0.5	14.60	0.07	D:L	1:0.5	14.96	0.07
D:A	1:1	12.97	0.26	D:H	1:1	14.42	0.18	D:L	1:1	14.44	0.15
D:A	1:5	8.58	0.09	D:H	1:5	14.14	0.23	D:L	1:5	10.33	0.10
D:A	1:10	7.36	0.06	D:H	1:10	14.41	0.14	D:L	1:10	8.71	0.15
D:A	1:50	6.88	0.24	D:H	1:50	13.32	0.21	D:L	1:50	7.61	0.20

SALTS

DNa: L - Arginine hydrochloride				DNa: L - Histidine monochloride monohydrate				DNa: L - Lysine monochloride			
	Ratio	PC Mean	SD		Ratio	PC Mean	SD		Ratio	PC Mean	SD
D	1:0	15.33	0.27	D:H	1:0	14.79	0.31	D:L	1:0	17.97	0.24
D:A	1:0.5	15.34	0.41	D:H	1:0.5	16.51	0.14	D:L	1:0.5	17.98	0.20
D:A	1:1	15.38	0.26	D:H	1:1	17.27	0.18	D:L	1:1	17.28	0.20
D:A	1:5	15.78	0.13	D:H	1:5	28.72	0.40	D:L	1:5	18.09	0.09
D:A	1:10	15.85	0.33	D:H	1:10	45.24	2.76	D:L	1:10	18.18	0.18
D:A	1:50	17.00	0.45	D:H	1:50	121.43	14.56	D:L	1:50	19.92	0.26

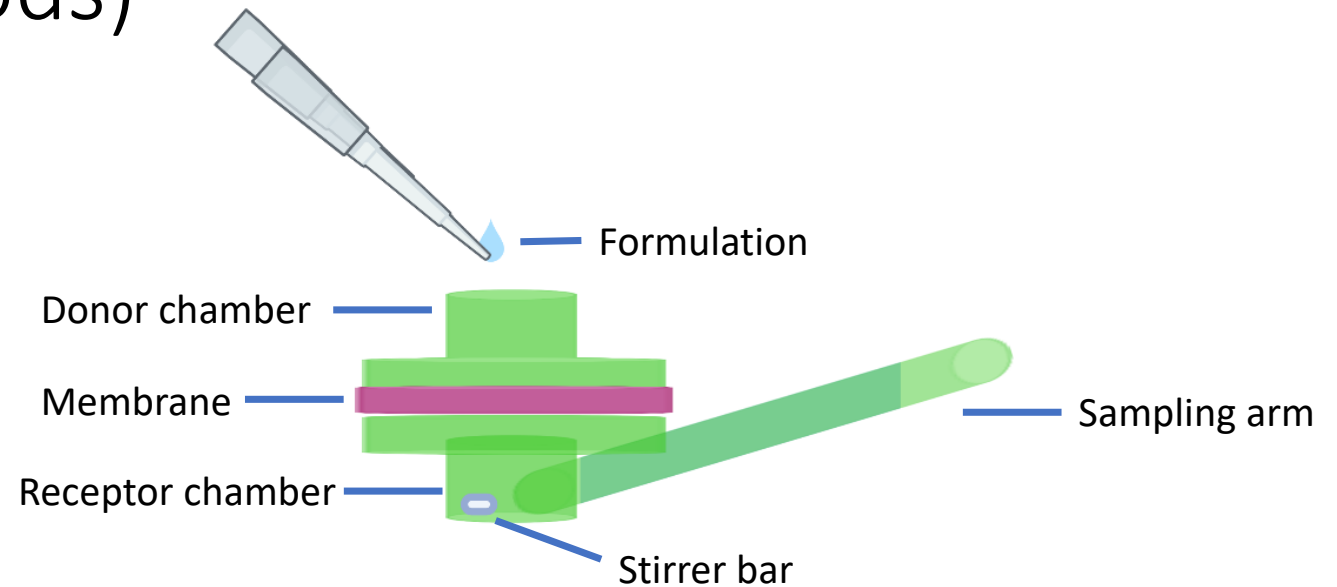
Permeation studies using porcine skin (experimental methods)

Permeation is based on passive diffusion can be described using Fick's laws of diffusion

Use Franz diffusion cells

Membrane: full thickness porcine skin

Mass balance studies



$$J_{ss} = \frac{DK\Delta c}{h}$$

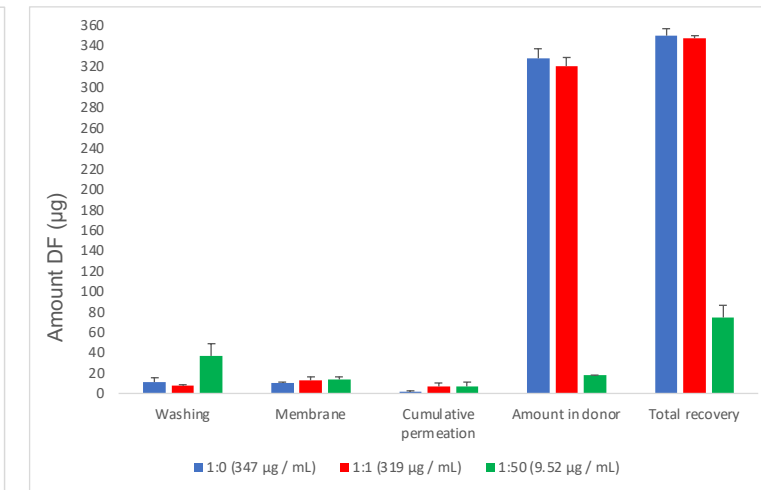
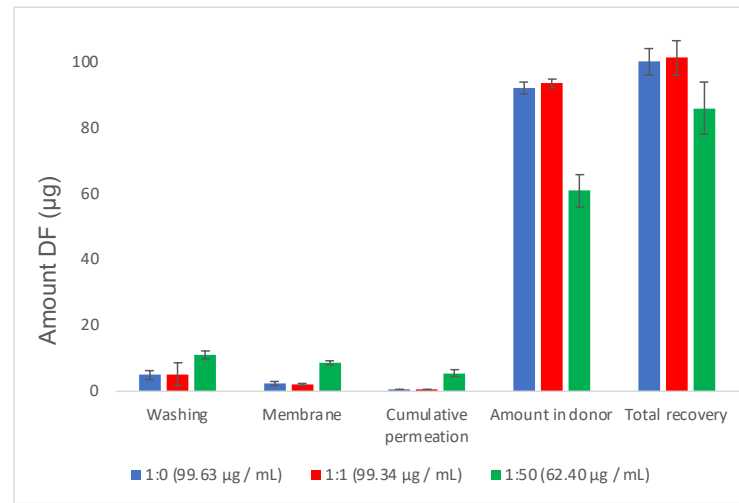
Fick's first law of diffusion

Initial permeation studies

- Initial permeation studies (IVPS) using 1 mL PBS solutions of DNA alone and combination with L-HSS in 1:1 and 1:50 molar ratios confirmed that amino acids could be used as counter ions to increase the partition and permeation of DF.
- Main issues related to solubility: as the amounts of L-HSS increased, the amount of DNA and L-HSS in solution decreased.
- Despite this, cumulative permeation results with L-HSS were 4 to 10-fold greater than using DNA alone

Permeation study	Ratio DNA:L-HSS	Amount of DF in solution at time of application ($\mu\text{g/mL}$) $\pm$ SD	Permeation ($\mu\text{g/cm}^2$) $\pm$ SD at 24 h	Amount in membrane (μg) $\pm$ SD at 24 h	Total amount permeated and in membrane (μg) $\pm$ SD at 24 h
IVPS-100	1:0	99.63 $\pm$ 0.12	0.50 $\pm$ 0.20	2.44 $\pm$ 0.59	2.95 $\pm$ 0.75
	1:1	99.34 $\pm$ 0.38	0.54 $\pm$ 0.14	2.10 $\pm$ 0.23	2.64 $\pm$ 0.25
	1:50	62.40 $\pm$ 0.41	5.39 $\pm$ 1.00	8.61 $\pm$ 0.66	14.04 $\pm$ 1.67
IVPS-350	1:0	347.52 $\pm$ 0.18	1.65 $\pm$ 0.57	10.01 $\pm$ 0.72	11.66 $\pm$ 0.38
	1:1	318.94 $\pm$ 0.64	6.93 $\pm$ 2.97	12.74 $\pm$ 3.32	19.67 $\pm$ 5.19
	1:50	9.52 $\pm$ 0.22	6.91 $\pm$ 3.61	13.46 $\pm$ 2.83	20.37 $\pm$ 2.49

IVPS-100 and IVPS-350, n=4, mean $\pm$ SD



Recovery of DF (μg) for the 3 formulations of DF:L-His (salts) in the ratios 1:0, 1:1 and 1:50 following mass balance studies in porcine skin. Unfiltered formulations (1 mL) were applied. (n=4; mean $\pm$ SD)

Conclusion of the first steps of DF: amino acid ion pair investigation:

L-HSS appears to increase the partition and permeation of DF at different concentrations of DNa

We on our way to achieving our goal: to reduce the amount of DNa required in the formulation to achieve at least the same expected outcomes

Furthermore, the use of amino acids comply with increasing demands to turn to more green manufacturing processes and the use of more sustainable inputs.

This concludes the first steps of our challenge

Thank you

- to my supervisors Dr Bruno Sil dos Santos, Dr Majella Lane and Prof. Jonathan Hadgraft
- to my colleagues past and present at the UCL Skin Research Group and London Metropolitan University
- to all of you for listening

Questions?

