Enhancing the topical delivery of N-acetyl-D-glucosamine: screening emulsifying systems

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Introduction

N-acetyl-D-glucosamine (GlcNAc) is a hydrophilic compound (log P = –2.1) and a versatile topical active. A monomer of hyaluronan (HA), it shares many of its physiological effects: it improves skin hydration, promotes the production of collagen and HA by fibroblasts and, as a tyrosinase inhibitor, also reduces hyperpigmentation [1–3]. However, contrary to HA, GlcNAc has a low molecular weight (221.21 g.mol^{-1}), which makes it a suitable compound to permeate the skin. Amphiphilic emulsifiers are used to stabilise emulsions, yet there is limited information about their influence on skin delivery [4]. **This study explored the influence of different emulsifiers in optimising the topical delivery of GlcNAc.**

Methods

- 5 O/W emulsions (Table 1) were made with different emulsifying systems:
  - F1: Polyglyceryl-3 Stearate;
  - F2: Steareth-2 / Steareth-21;
  - F3: Cetearyl Glucoside;
  - F4: Sorbitan Oleate / Polysorbate 80;
  - F5: Oleth-5 / Oleth-20.
- Additional emulsions with Cetearyl Alcohol (2.5, 5.0, 7.5 and 10.0%) were formulated under the same conditions.

Table 1. Emulsion samples

Phase	INCI	% (w/w)
Water	Aqua	73.0 – 63.0
	N-Acetyl-D-Glucosamine	2.0
Oil	Emulsifying System (details on the left)	5.0
	Cetyl Palmitate	10.0
	Isopropyl Myristate	10.0
	Cetearyl Alcohol	2.5 – 10.0 (where appropriate)

- All emulsions were characterised: pH, conductivity and apparent viscosity.
- The viscosity curves of emulsions F1 and those with Cetearyl Alcohol were obtained with a HAAKE™ RotoVisco 1 cone/plate rheometer (Thermo Fisher Scientific, UK);
- *In vitro* release studies were performed with vertical Franz diffusion cells and HT Tuffryn® membranes (Pall Corporation®, USA); the receptor phase was a phosphate-buffered solution (pH 7.4) maintained at 32 °C and stirred at 600 rpm. Samples were collected after 0.5, 1, 2, 3, 4 and 6 h.
- GlcNAc was quantified by a DNS-colourimetric method, based on the oxidation of 3,5-dinitrosalicylic acid, DNS, under alkaline conditions, followed by the formation of 3-amino-5-nitrosalicylic acid, which was detected at 545 nm [5].

Results and Discussion

The results showed similar pH and conductivity values for all emulsions. However, apparent viscosity (T = 22 °C, 5 rpm) values were different:

F1 – 33,200 mPa.s > F2 – 7,080 mPa.s > F3 – 2,500 mPa.s > F4 – 1,610 > F5 – 80 mPa.s

Figure 1:

- All formulations showed similar release patterns, but different release concentrations;
  - F1 – higher viscosity – had the lowest GlcNAc release after 6 h;
  - F2 and F3 released similar amounts of GlcNAc after 6 h;
  - F4 and F5 – lower viscosity – had the highest GlcNAc release after 6 h.

Viscosity was confirmed to be the main variable responsible for the differences in GlcNAc release, in line with previous studies [6,7].

To evaluate the effect of viscosity further:

- F4 had the highest GlcNAc release and low viscosity – it was reformulated with the addition of Cetearyl Alcohol in a range of concentrations: 2.5, 5.0, 7.5 and 10.0%.

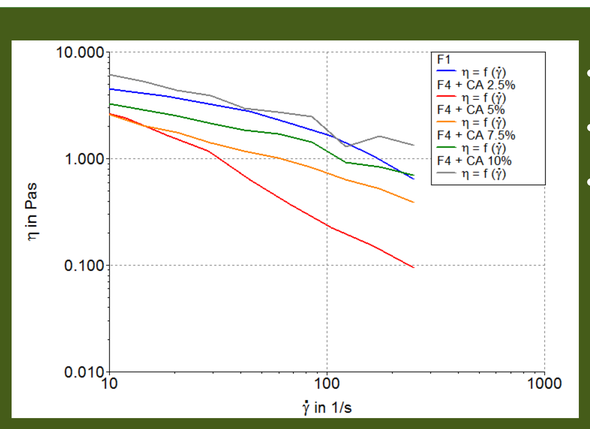

Figure 2. Viscosity curves of F1 and the emulsions with Cetearyl Alcohol.

- ↑ Cetearyl Alcohol (CA) = ↑ viscosity;
- Shear thinning behaviour;
- F1 has shown a slightly different rheological profile from the CA-containing emulsions, indicating a different internal structure.

Figure 3:

- Similar release profile for all samples with Cetearyl Alcohol;
- Increasing viscosity lowered the amount of GlcNAc released.

Figure 2 vs Figure 3:

- The internal of emulsions, as opposed to viscosity alone, affects GlcNAc release;
- Fatty alcohols + emulsifiers = lamellar gel phases in O/W emulsions [8];

The results indicate that lamellar gel phases hindered GlcNAc release

Lamellar gel phases have been described to enhance dermal delivery of hydrophobic compounds [4]

The microstructure of the emulsions must be assessed and related to rheological and release profiles

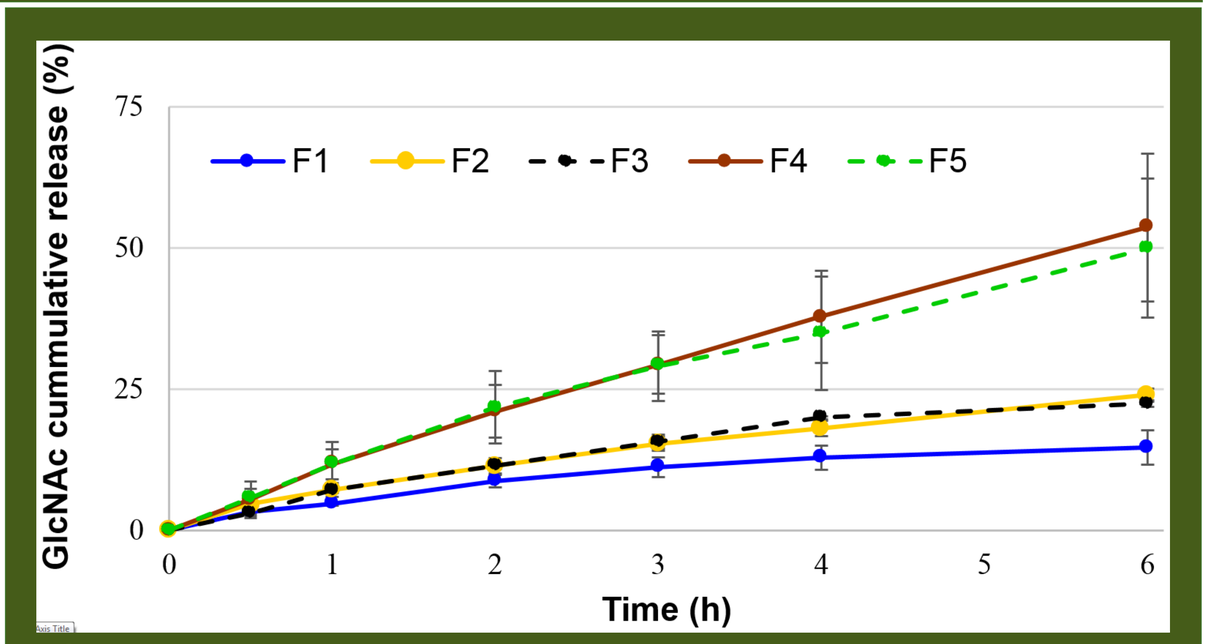

Figure 1. *In vitro* release profiles of GlcNAc from emulsions F1 – F5.

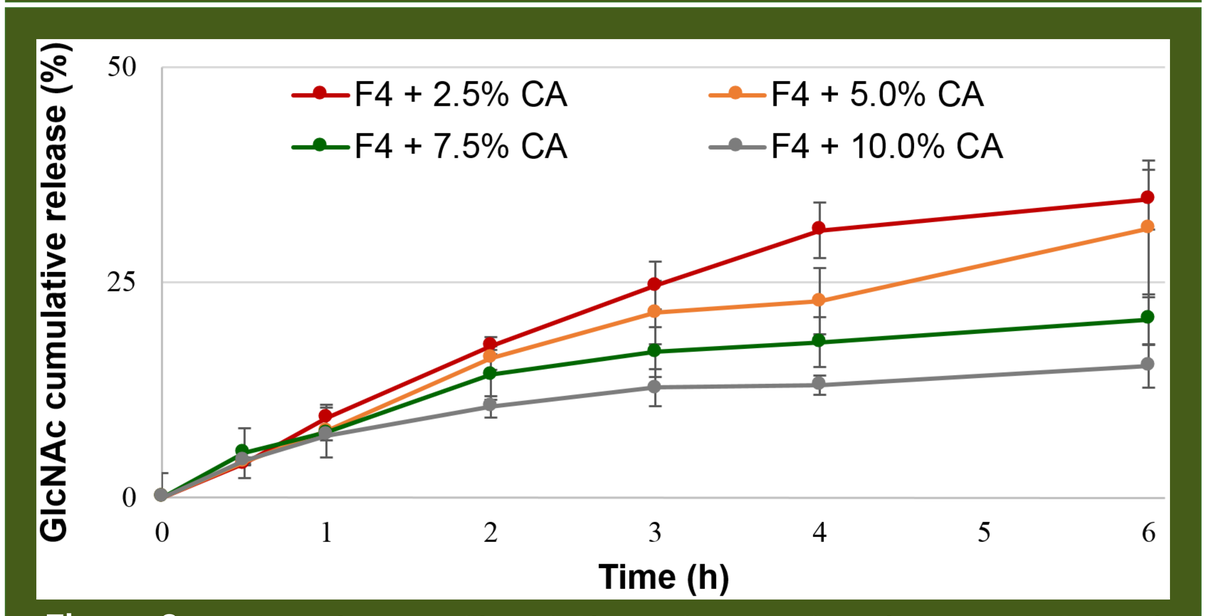

Figure 3. *In vitro* release profiles of GlcNAc from the formulations with Cetearyl Alcohol: F4 + CA 2.5%, F4 + CA 5%, F4 + CA 7.5% and F4 + CA 10.0%.

Conclusion

- The *in vitro* release of GlcNAc from O/W emulsions is affected by viscosity. Thus, viscosity should be controlled in studies to optimise the topical delivery of GlcNAc.
- To understand the role of emulsifiers, further analysis of the internal structure of emulsions is required.

Although viscosity has been described to influence release from emulsions, it may not affect skin permeation [4]. Therefore, *in vitro* permeation studies using skin-similar membranes must be performed to understand the influence of the emulsifying system on GlcNAc delivery, followed by *in vivo* efficacy tests.

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