Preservation of Oral Nanosuspensions with Food Preservatives

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ABSTRACT SUMMARY
An alternative dosage form to nanocrystal tablets are suspensions of nanocrystals. Nanocrystal suspensions are also of interest to deliver nutraceuticals in health/wellbeing drinks. In this study, food preservatives which do not impair nanosuspension stability and are suitable for use in oral nanosuspensions were screened. The most suitable one was glycerol at a concentration of 20% (w/w).

INTRODUCTION
Nanocrystals for the oral delivery of poorly soluble drugs are on the market since the year 2000 (e.g. Rapamune), mainly tablets and capsules, Megace ES being the only oral nanosuspension. Meanwhile, nanocrystals are also used for dermal delivery, cosmetic products are on the market since 2007 (e.g. platinum rare) [1]. Additionally, there has been also interest to add nanocrystals to beverages, e.g. poorly soluble anti-oxidants – especially to higher priced health/wellbeing drinks. In focus, are compounds such as resveratrol and rutin.

Of course, the oral use requires often preservation or at least preservation of the nanocrystal concentrates prior to their use in drink production. Preservatives established for dermal use (e.g. Euxyl PE9010) are not allowed as food additive. Therefore, in this study, a screening was performed using food preservatives allowed for use in beverages.

Preservatives can destabilize suspensions, e.g. electrolyte type ones by reduction of the zeta potential and non-electrolytes ones by dehydration of the stabilizer layer. Both types were used in this study and rutin nanocrystals were used as nutraceutical.

EXPERIMENTAL METHODS
Rutin nanocrystals were produced by dispersing rutin powder (10% w/w) in Tween® 80 solution (1% w/w) and processing by wet bead milling using a PML-2 (Bühler AG, Switzerland) with 0.4-0.6 mm yttria stabilized zirconium oxide beads. Subsequently, 2 cycles of high pressure homogenization (HPH) (LAB 40, APV, Germany) at 300 bar were applied.

Particle size analysis of the rutin nanosuspension was performed by photon correlation spectroscopy (PCS) (Zetasizer Nano ZS, Malvern Instruments, UK). Destabilization of the nanocrystals after incorporation of different preservatives was monitored using light microscopy and turbidimetry. The absorbance was measured at wavelengths between 340 and 800 nm using a PharmaSpec UV-1700 photometer (Shimadzu Corporation, Japan). Storage was performed at room temperature (20°C) for 2 months.

Zeta potential (ZP) was analyzed in water (pH 5.5-6, 50µS/cm) using a Zetasizer Nano ZS (Malvern Instruments, UK).

Preservatives added were glycerol at 5, 10, 15 and 20% (w/w), potassium sorbate (PS) and sodium benzoate (SB) at concentrations of 0.02, 0.05, 0.10 and 0.15% (w/w).

RESULTS AND DISCUSSION
Nanocrystals can either be added to the health drink or alternatively, a nanocrystal concentrate can be admixed to the drink prior to consumption. Placing the nanocrystals and other ingredients in a concentrate opens the option to provide optimized storage conditions for the ingredients, e.g. optimized pH to ensure chemical stability of vitamins or physical stability of nanocrystals.

The unpreserved rutin nanosuspension had a mean PCS diameter of 860 nm, and a zeta
potential in water of -21 mV, indicating that the good stability of the nanosuspension itself (stability data of this formulation available for over 2 years) is due to not only electrostatic, but also steric stabilization.

For ease of detection and to avoid interference of concentrate ingredients with PCS, physical stability was monitored by light microscopy and turbidimetry. Figure 1 shows the absorbance of the non-preserved nanocrystals in the concentrate at different wavelengths, with a peak at 377 nm.

![Absorbance spectrum of non-preserved nanocrystals in the concentrate.](image1)

Based on this, wavelengths on the interval between 340 and 540 were chosen to observe the absorbance pattern of the concentrates containing different preservatives. Figure 2 shows the change in the absorbance pattern, exemplarily for the destabilizing preservatives potassium sorbate and sodium benzoate, and the most stable preservative, glycerol. It is noted that as the nanosuspension destabilizes, the absorbance tends to decrease. First at minor aggregation the absorbance increases, large aggregates (= reduced number of particles) have then a lower absorption.

After 2 months, samples preserved with glycerol at all concentrations proved to be stable, even for the highest concentration, 20%, while the samples preserved with the ionic molecules aggregated at all concentrations, except for 0.02% (for both SB and PS), which is not ideal, since it is the lower preservation concentration for sodium benzoate and below the lowest preservation concentration for potassium sorbate. The combination of both ionic preservatives also showed destabilization effect.

![Absorbance spectrum of preserved nanocrystals concentrates after 2 months storage.](image2)

Destabilization effects detected by turbidimetry measurements were in good agreement with light microscopy, by which aggregation could be observed in case of destabilization.

CONCLUSIONS

Preservation with glycerol at 20% was established, not impairing the physical stability of the nanocrystals. This supports the use of nanocrystal in health beverages.

REFERENCES


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