Application Note

Pharma/BioPharma

Agilent

Trusted Answers

Dissolution of a PLGA Nanoparticle Formulation Using the Agilent NanoDis System

Authors

Emre Türeli
MyBiotech
Ueberherrn, Germany

Karen Krauel-Göllner
Agilent Technologies, Inc.

Abstract

Nanocarriers and nanoformulations have been investigated in pharmaceutical research over many years. These nanomedicines offer multiple solutions from protecting the active pharmaceutical ingredient from biological environments to enhancing the bioavailability and stability of the drug at the site of action. Although nanomedical research is in the spotlight and more commercialized nanoformulations are on the market, there are no standardized procedures or regulations for the assessment of the quality and safety profiles of such products. In this study, the release profile of poly(lactic co-glycolic) (PLGA) nanocapsules loaded with all-*trans* retinoic acid (RA) was characterized using an innovative sample and separation setup, the Agilent NanoDis system. The results were compared to the release profile measured with a dialysis technique.

Introduction

Nanocarriers and nanoformulations have been investigated in pharmaceutical research over many years. These nanomedicines offer multiple solutions from protecting the active pharmaceutical ingredient from the biological environments to enhancing the bioavailability and stability of the drug at the site of action. Although nanomedical research is in the spotlight and more commercialized nanoformulations are on the market, there are no standardized procedures or regulations for the assessment of the quality and safety profiles of such products. Among these assessments, the accurate estimation of the *in vitro* release profiles of the drug delivery systems should be considered. This estimation can provide a better understanding and prediction of the behavior of the nanoformulations in the *in vivo* environment and to ease the translation of nanomedicines from the bench to the bedside. These assessments can evaluate the safety and quality profiles of such systems. To address this effort, traditional and commercialized techniques such as dialysis, sample and separation techniques, *in situ* techniques, and combined methods have been developed. These techniques have been applied for the assessment of the *in vitro* release characteristics of the nanocarriers. Moreover, various commercialized *in vitro* dissolution testing systems are on the market for the automation of the processes. Although there has been a tremendous effort to understand the *in vitro* release characteristics of such systems, efficient separation of the drug from the nanocarrier remains a challenge for most of the techniques. In this study, the release profile of poly(lactic co-glycolic) (PLGA) nanocapsules loaded with all-*trans* retinoic acid (RA) was characterized using an innovative sample and separation setup, the Agilent NanoDis system, and compared to the release profile measured with a dialysis technique. See reference 1 for more information on the overall functionality of the NanoDis system.

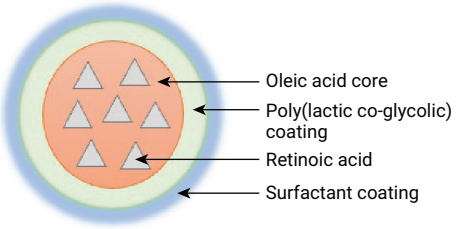

Figure 1. Retinoic acid-encapsulated PLGA particles.

Experimental

Dissolution experiments were conducted with an Agilent 708-DS dissolution apparatus coupled with a NanoDis module and Agilent 850-DS sampling station (see Table 1).

Table 1. Parameters of dissolution experiments.

Dissolution Apparatus	USP II
Volume	900 mL
Rotation speed	50 rpm
Membrane	500 kD mPES Spectrum MicroKros hollow fiber filters
Buffer	Phosphate buffer saline with 0.5% polysorbate 80 (PBS-PS80) or SDS (PBS-SDS)

The nanocapsules were diluted to reach 5 µg/mL of RA in the vessel and reach sink conditions in 900 mL of PBS-PS80 or PBS-SDS. At predetermined time points, the samples were pulled from the dissolution vessel, filtered by the NanoDis module, and collected by the 850-DS sampling station (Table 2). The whole workflow was controlled with Dissolution Workstation software.

Table 2. Parameters of the NanoDis module.

Operation	Setting
Pretest	
Peristaltic Flow Through Duration	240 s
Syringe Purge Volume	4 mL
Peristaltic Air Purge Duration	60 s
Pre Time Point Filter Conditioning	
Peristaltic Flow Through Duration	60 s
Syringe Purge Volume	2 mL
Time Point Sampling Properties	
Sample Volume	5 mL
Filter Outer Cylinder Rinse Volume	4 mL
Peristaltic Pump Sample Duration	45 s
Peristaltic Syringe Overlap	0 s

For the dialysis method, the nanocapsules were diluted to reach 20 µg/mL of RA in release medium, either PBS-PS80 or PBS-SDS, to ensure sink conditions. A 10 mL sample of the nanoparticle suspension was placed inside a 10 mL, 300 kD cellulose ester Float-A-Lyzer from Repligen.

Results and discussion

In the first set of experiments, the dissolution of dissolved RA was compared with RA contained in PLGA nanocapsules using the dialysis method (Figure 2).

Release profiles depended on the dissolution medium, and there was no significant difference between the release profiles of RA encapsulated in PLGA and RA dissolved alone in the donor compartment. These data show that the dissolution kinetic is solely controlled by the membrane permeation and that the membrane permeation kinetics of RA is slower than the actual dissolution rate. Studying the drug release of PLGA nanocapsules over 40 hours via the dialysis method could easily lead to an overestimation if it was not compared to the control of the permeation kinetics of the free drug.

In the second part of the experimental design, PLGA nanoparticle drug release was quantified using the NanoDis system (Figure 3).

The burst release of RA from PLGA nanocapsules could be observed clearly using the NanoDis system since the release kinetic was not limited by the permeation kinetic of the dialysis membrane. The burst release was followed by a slower controlled release from the nanocapsules. When using the dialysis method, the burst release of the nanocapsules was severely underestimated, particularly for the early time points.

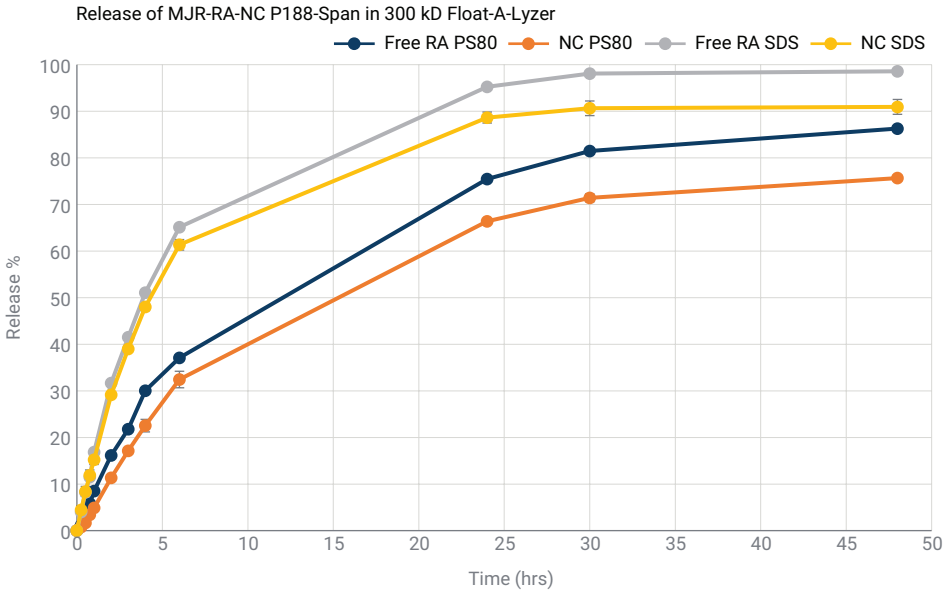

Figure 2. Release profile of nanocapsules (NC) and dissolved retinoic acid (RA) in PBS-PS80 and PBS-SDS by dialysis.

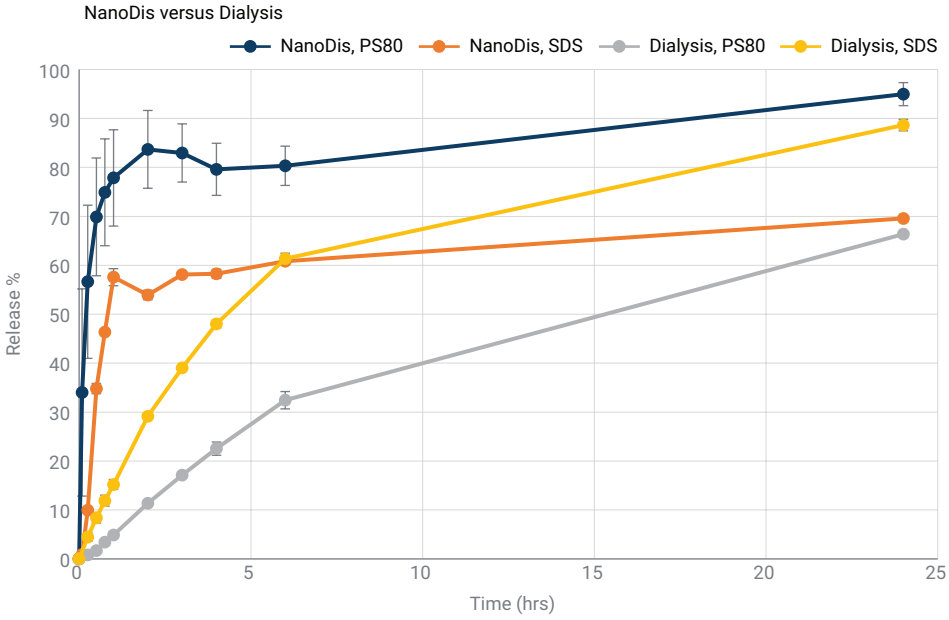

Figure 3. Release profiles of RA loaded PLGA nanocapsules in PBS-PS80 and PBS-SDS, determined by dialysis and cross flow filtration with an Agilent NanoDis system.

In the next set of dissolution experiments, the different release kinetics of RA from different PLGA formulations were compared using the NanoDis setup. The nanocapsules were coated either with chitosan to decrease the burst release, or the polymer and oil content of the particles were increased to achieve a longer diffusion path for RA (Figure 4).

Figure 4 shows the release profiles of RA form the various nanocapsule formulations, e.g. high and low chitosan coating and different amounts of PLGA and oil content. Oil content was varied between 0.9 and 1.5% between the high and low formulations. Furthermore, chitosan was added in two different concentrations as a surface coating. Another excipient polysorbate 80 was also included in the oil phase of the formualtions. With the NanoDis, it was possible to differentiate the release from the various formulations and also show the initial burst release. In general, the formulations with a chitosan coating show an incomplete release of RA and a slower burst release. These results are in comparison to the formulations without a chitosan coating and with a variation in PLGA and oil content.

Conclusion

Burst release differences in RA containing PLGA formulations were characterized using the Agilent NanoDis system for formulation development and to select the most promising approach to decrease the burst release.

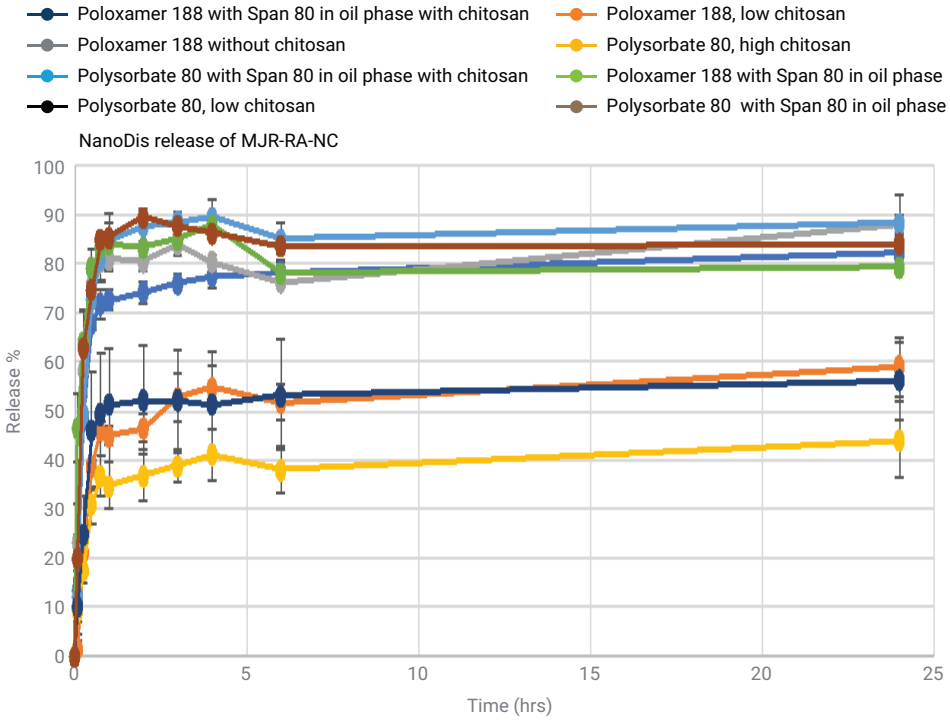

Figure 4. Release profile of different RA containing PLGA formulations in PBS 0.5% Tween 80 buffer using an Agilent NanoDis system.

It was not possible to observe the burst release using the dialysis method due to the slow permeation kinetics of RA through the membrane, which was the rate-limiting step. The NanoDis system showed its advantage in the formulation development process of PLGA nanocapsules. The system enabled differentiation between various formulations, showing their differences in the burst release, as the principle of crossflow filtration is able to show the real release profiles, without delay by a dialysis membrane.

Reference

1. Agilent NanoDis System Method Development Guide. *Agilent Technologies white paper*, publication number 5994-2347EN, **2020**.

www.agilent.com

DE37616851

This information is subject to change without notice.

© Agilent Technologies, Inc. 2022

Printed in the USA, December 9, 2022
5994-5536EN

Agilent
Trusted Answers