Supramolecular Metal–Phenolic Gels for the Crystallization of Active Pharmaceutical Ingredients

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The use of supramolecular gel media for the crystallization of active pharmaceutical ingredients (APIs) is of interest for controlling crystal size, morphology, and polymorphism, as these features determine the performance of pharmaceutical formulations. In contrast to supramolecular systems prepared from synthetic gelators, herein, supramolecular metallogels based on a natural polyphenol (tannic acid) are used for the crystallization of APIs. The gel-grown API crystals exhibit considerable differences in size, morphology, and polymorphism when compared with those formed in solutions. These physical features can also be tailored by varying the gel composition and additives, suggesting an influence of the gel medium on the crystallization outcomes. Furthermore, these gel–API crystal composites can be used for sustained drug release, indicating their potential as drug delivery systems. The facile preparation of these supramolecular gels and the use of naturally abundant components in their synthesis provide a generic platform for studying gel-mediated crystallization of diverse APIs.

1. Introduction

The crystallization of active pharmaceutical ingredients (APIs) plays a key role in addressing different formulation issues in the pharmaceutical industry, as the crystal size, morphology, and polymorphism significantly influence the bioavailability, solubility, and processing of APIs.\(^1\)–\(^3\) In this context, the use of supramolecular gel media to control the crystallization of APIs has emerged as a versatile strategy.\(^4\)–\(^7\) Foster et al.\(^8\) in their seminal work, used supramolecular gel systems based on low-molecular weight gelators (typically, \(M_w < 2000\) Da)\(^9\) for API crystallization. These gelators were synthesized to leverage the gelation of multiple solvents under specific crystallization conditions, thus enabling crystallization of the APIs mediated by such gel systems.\(^6\)–\(^8\)\(^,\)\(^10\) However, the supramolecular gel systems developed to date are mostly limited to the use of synthetic gelators. Furthermore, such supramolecular gels systems depend exclusively on a heat–cool-triggered gelation process, which mostly affords the preparation of mono-component gels, thereby limiting the possible variables used during API crystallization.\(^11\)–\(^14\) Therefore, supramolecular gel systems, which are assembled directly from naturally occurring organic gelators and whose preparation is independent
of a heat–cool-triggered gelation process (to allow for the preparation of bi- and multi-component gels),\textsuperscript{[15–17]} are an attractive alternative for the gel-mediated crystallization of APIs.

Recently, we demonstrated the gelation of tannic acid (TA, $M_w = 1701.2$, Figure S1, Supporting Information), a ubiquitous natural polyphenol, with titanium(IV) (Ti\textsuperscript{IV}) ions via coordination-driven supramolecular assembly; this gelation was observed to be specific to group IV transition metals.\textsuperscript{[18]} Contrary to the conventional heat–cool-triggered gelation,\textsuperscript{[6,8,9]} the TA–Ti\textsuperscript{IV} metallogels could be formed by simple mixing at ambient conditions in various solvents over a large concentration range and metal–ligand stoichiometries. Main features of this system are its robust and adaptive nature, which enable in situ co-gelation of a range of additives and control over other concurrent assembly processes such as crystallization of metal–organic frameworks.\textsuperscript{[18]} Based on these features, we hypothesized that the TA–Ti\textsuperscript{IV} system could act as a facile supramolecular gel medium for API crystallization.

Herein, we report the crystallization of APIs in a TA–Ti\textsuperscript{IV} metallogel medium. To our knowledge, this is the first metallogel system, prepared from a natural organic gelator without the need of a heat–cool trigger, which is used as a medium for API crystallization. As model APIs (Figure S2, Supporting Information), we demonstrate the gel-mediated crystallization of caffeine (a central nervous system stimulant), carbamazepine (an anti-convulsant, CBZ), and piroxicam (an anti-inflammatory drug, PX).\textsuperscript{[8]} The size, morphology, and polymorphism of the gel-grown API crystals show striking differences when compared with those of the corresponding API crystals grown in solution. Furthermore, the size and morphology of the gel-grown crystals can be modulated by varying the crystallization parameters and gel composition, including metal–ligand stoichiometry and additive type (i.e., multi-component environment). Thus, the crystallization outcomes can be controlled by the gel medium. Additionally, we use the gel–API crystal composites as carriers for sustained drug release, where, the gel–API crystal composites show a significantly delayed release of APIs from the gel matrix when compared with the gel–API (molecular) composites.
2. Results and Discussion

The process of API crystallization in the TA–Ti IV metallogel is presented in Scheme 1, and detailed characterization of the TA–Ti IV system can be found elsewhere. For the synthesis, the APIs were dissolved in their respective solvents and mixed with the TA/Ti IV sol—the solvents of which were chosen to act as anti-solvents for the APIs (e.g., water for CBZ, see details in Supporting Information). The conditions were maintained so that gelation (typically <30 min) preceded crystallization (2–24 h depending on the API, Scheme 1b and Figure S3 (Supporting Information)) and crystallization during mixing could be avoided. For simplicity, we denote the TA/Ti IV system according to its composition as TA–Ti IV–XYZ, where X is the concentration of TA in % (w/v) and YZ is the TA:Ti IV stoichiometry. For example, a gel with a composition of TA = 5% (w/v) and TA:Ti IV = 1:5 is denoted as TA–Ti IV–515. The API concentration is also presented in % (w/v).

We first investigated the crystallization of caffeine in the TA–Ti IV–515 system with the caffeine concentration varying from 1.6 to 5.4%. Initially, the crystallization was monitored at room temperature (~23 °C) for 24 h. For caffeine concentrations greater than 3.0%, crystallization was completed within 2 h (Figure 1a–d) and for concentrations less than 3.0%, no crystals were observed.

**Scheme 1.** Illustration of the a) crystallization of the API in the TA–Ti IV gel system and b) sol–gel transition and subsequent crystallization of the API in the gels.
even after 24 h. Note that, the transparency of the TA–Ti IV system allowed clear visualization (in-gel observation) of the crystals formed in the gels by optical microscopy.[18] Differential interference contrast (DIC) microscopy images of the needle-like caffeine crystals formed in the gels with caffeine concentrations of 3.1, 4.1, and 5.4% showed a more uniform distribution of shorter needles (Figure 1b,c) when compared with the size of the needles obtained (>10 mm) through crystallization in solution in the absence of gelators (specimens obtained in the absence of gelators are referred to as controls, Figure 1a and S4). Specifically, at caffeine concentrations of 3.1 and 4.1%, the gel-grown crystals were similar in length (500–700 µm) and particle number (40–50 mm−2). In contrast, at a caffeine concentration of 5.4%, the gel-grown crystals were considerably shorter (200–300 µm) with a higher particle number (160–180 mm−2). Scanning electron microscopy (SEM) images of the crystal needles formed in the control system with a caffeine concentration of 5.4% revealed the relatively smooth surface of the needles (Figure S5, Supporting Information). In contrast, the gel-grown caffeine crystals revealed mesh-like microscopic domains (Figure S6, Supporting Information). X-ray diffraction (XRD) analyses confirmed the crystallinity of the caffeine crystals formed in the control system and the gels (Figure S7, Supporting Information).
As the solubility of caffeine in water decreases with decreasing temperature,\textsuperscript{[19]} we also examined the crystallization of caffeine with a concentration of 2.5\% in the gel at 4 °C for 24 h, as shown in Figure 1f–h. The caffeine crystals formed in the gels and control system under this condition were stable at ~23 °C (i.e., crystallization was irreversible). The gel-grown crystals showed remarkably different morphologies when compared with those grown in the control system (Figure 1e), which produced long needles similar to those observed at ~23 °C. The crystals formed in TA–Ti\textsuperscript{IV}-515 featured hair-like structures on rod-like cores whose length varied from 200 to 250 µm (Figure 1f). Increasing the TA:Ti\textsuperscript{IV} stoichiometry (while maintaining a caffeine concentration of 2.5\%), that is, in the TA–Ti\textsuperscript{IV}-517 and TA–Ti\textsuperscript{IV}-519 systems, resulted in the formation of caffeine crystals with a leaf-like (length, 60–80 µm) and blade-like (length, 100–120 µm; width, 50–70 µm) morphologies (Figure 1g,h), respectively. In contrast, increasing the gel concentration, that is, in the TA–Ti\textsuperscript{IV}-615 and TA–Ti\textsuperscript{IV}-715 systems, resulted in caffeine crystals of comparable sizes but with a
more complex morphology (bamboo shoot-like) (Figure S8, Supporting Information). These results demonstrate the influence of different parameters of the TA–Ti$^{IV}$ gel medium that control the size and morphology of the caffeine crystals.

We then examined the crystallization of CBZ in TA–Ti$^{IV}$-515 (Figure S9, Supporting Information) at ~23 °C for 24 h and increasing CBZ concentrations. In this case, the gel matrix exerted a stronger influence on the crystal morphology. The control system resulted in needle-like crystals only (Figure 2a and S10) with a slight increase in the needle length with increasing CBZ concentrations. In contrast, the CBZ crystals formed in the gels were of a near-spherical shape, as shown in Figure 2b–d. As the CBZ concentration increased, more spherical crystals formed; however, they became smaller as the CBZ concentration increased from 2.5% (500–600 µm) to 3.7% (250–300 µm). Changes in the surface morphology were also evident—the crystals initially featured a globular surface texture with a plate-like pattern that changed to a relatively smooth surface structure as the concentration increased. A similar surface smoothening was observed upon increasing the gel concentration (TA–Ti$^{IV}$-615, TA–Ti$^{IV}$-715; Figure 2f,g) at a CBZ concentration of 3.0%. However, the variation in the crystal size was negligible.
Figure 2. DIC images of the CBZ crystals formed a) in the control system (with 2.5% CBZ) and b–d) TA–Ti\textsuperscript{IV}–515 with increasing CBZ concentrations (2.5, 3.0, and 3.7%, respectively). DIC images of the CBZ crystals e) in the control system and f,g) TA–Ti\textsuperscript{IV}–615 and TA–Ti\textsuperscript{IV}–715, respectively (with 3.0% CBZ). h) Photograph showing the extraction process of the CBZ crystals (with 3.7% CBZ) after gel disassembly. i–k) SEM images of the control and extracted CBZ crystals from the gels (with 3.7% CBZ). l) XRD pattern of the extracted CBZ crystals from the gels (with 3.7% CBZ).

To extract the gel-grown CBZ crystals, we developed a competing ligand-mediated gel disassembly method. A solid mass of pyrocatechol (PC) was added on top of a gel sample containing crystals to initiate the process (see Supporting Information for details), and the crystals were collected when the gel disassembled (Figure 2h). The dissolved PC in the gel competed with the galloyl groups of TA (i.e., cross-linking sites) for coordination with Ti\textsuperscript{IV}, resulting in gel disassembly. SEM images
of the extracted CBZ crystals from the gels (TA–TiIV-515, 3.7% CBZ) revealed the spherical shape of the crystals (as consistent with the DIC images), with a densely stacked plate-like surface texture (Figure 2j,k). An SEM image of the control sample (3.7% CBZ) is also shown for comparison (Figure 2i). The crystallinity of the extracted and control samples (both with 3.7% CBZ) was confirmed by XRD (Figure 2l and S11). Furthermore, the XRD patterns indicated the presence of two polymorphs of CBZ (mixture of dihydrate and form III) in both the samples; the dihydrate form (CDCC-CSD: FEFNOT09) was the dominant phase in the extracted crystals, whereas form III (CDCC-CSD: CBMZPN01) was the dominant phase in the control sample. This result suggests that the TA–TiIV gel matrix can also influence the crystal polymorphism.

The crystallization of PX performed in TA–TiIV-515 (Figure S12, Supporting Information) with increasing PX concentrations from 1.1 to 2.0% (at ~23 °C for 24 h) also yielded crystals with remarkably different sizes and morphologies when compared with those obtained in the control system (Figure 3). The crystals obtained in the control system formed globular aggregates (Figure 3a and S13). In contrast, block-type crystals formed in TA–TiIV-515 (Figure 3b–d). With increasing PX concentrations from 1.1 to 1.5 and 2.0%, the size of the crystals increased from 15–30 to 40–60 and 70–100 µm, respectively, with the corresponding particle number decreasing from 600–800 to 200–300 and 70–90 mm–² (Figure 3b–d). This trend is opposite to that observed for the caffeine and CBZ crystals formed in TA–TiIV-515 gel (with increasing caffeine and CBZ concentrations). The different trends obtained can be attributed to the structural differences of the APIs and their interactions with the gel environments. SEM images of the control sample (with 1.5% PX) revealed that the globular aggregates composed of clustered thin needles (Figure 3e), and that of the corresponding crystals extracted from the gel (extraction process was identical to that of CBZ, Figure S14, Supporting Information) confirmed the irregular block-shaped morphology (Figure 3f). From the XRD analyses, the crystalline phase of the extracted crystal sample was identified to be a mixture of PX polymorphs of form I (CDCC-CSD: BIYSEH01) and form B (subclass of form II, ICDD PDF No. 40-1708), where form B was the dominant phase (Figure 3g). In contrast, the control sample showed a pure

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phase of form I (Figure S15). Furthermore, to provide an example of API crystallization in a multi-component gel environment, we performed the crystallization of PX (1.5%) in this system with iron(III) as an additive, that is, in TA–TiIV–515–FeIII gel. The preliminary result is shown in Figure 3h. The block-type PX crystals formed in this system were larger (>500 µm) but lower in number (<10 per 400 µL of gel) when compared with those formed in the TA–TiIV–515 gel (Figure 3c). This is possibly caused by the higher degree of crosslinking of the TA–TiIV–515–FeIII gel, which reduces the number of nuclei formed and retards the molecular convection compared with the TA–TiIV–515 gel.

Figure 3. DIC images of the PX crystals formed a) in the control system (with 1.5% PX) and b–d) TA–TiIV–515 with increasing PX concentrations (1.1, 1.5, and 2.0%, respectively). e,f) SEM images of the control and extracted PX crystals from the gel (with 1.5% PX). g) XRD pattern of the extracted PX crystals from the gel (with 1.5% PX). h) PX crystals formed in TA–TiIV–515–FeIII system (with 1.5% PX).

As previously shown,[8] the crystallization of these APIs in synthetic urea-based gels resulted in crystals with different sizes, morphologies, and polymorphism (e.g., the gel-grown PX crystals
composed of mostly needles with diverse sizes of form I and II polymorphs) when compared with our present results. Such differences are expected as every gel system obtained under a given condition would provide a unique physico-chemical environment for the crystallization process.\textsuperscript{[4,6]} However, such a process is governed by a complex interplay of factors, including API–solvent/gel interactions, nucleation, and molecular convection in the gel, which render the mechanistic details of a specific system or inter-system comparison difficult and thus require further studies.\textsuperscript{[5–7,10]}

In previous studies,\textsuperscript{[18,20]} we demonstrated the TA–Ti\textsuperscript{IV} gel system to be non-cytotoxic based on a 3-(4,5-dimethylthiazolyl-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. This leads us to envision that gel–API crystal composites can potentially be used as bi-modal release systems for drug delivery, where the confinement effect of the gel matrix and crystal dissolution can be combined for sustained drug release. This idea is schematically presented in Figure 4a. As a proof of concept, TA–Ti\textsuperscript{IV}-515 gel particles (prepared by an emulsion method, see Supporting Information for details) containing caffeine crystals (Figure 4b and S16) were examined. The caffeine release (dissolution) kinetics from the gel–API crystal composite was compared with the release kinetics of molecular caffeine-containing gel particles. Caffeine release was monitored at its characteristic wavelength of 273 nm by UV-visible absorption spectroscopy (Figure 4b).\textsuperscript{[21]} Note that at this condition, the gel particles were stable\textsuperscript{[18]} and no TA release was observed, which could potentially interfere with the absorption spectrum of caffeine (Figure S17). As shown in Figure 4c, the gel–crystal composites exhibited a significantly delayed release profile when compared with the gel–caffeine (molecular) composites (~240 vs ~60 min for over 95% dissolution), which can be attributed to a combination of the factors (i.e., the gel confinement effect and crystal dissolution) mentioned above.
3. Conclusion

In summary, we presented a natural polyphenol-based supramolecular metallogel system for API crystallization. The gel-mediated crystallization resulted in API crystals with different sizes, morphologies, and polymorphs when compared with those formed in solutions. The intra-system modifications in sizes and morphologies of these crystals were also achieved by altering the crystallization parameters for example, gel composition. The inherent advantages of the present system, including ease of preparation, use of cheap and abundant components, and ability to incorporate diverse additives, can provide a generic platform from which an infinite number of subsystems can be derived to study the crystallization of a range of APIs. We are currently exploring multi-component gel environments using a range of additives for the crystallization of diverse sets of hydrophilic and hydrophobic APIs. Furthermore, the use of gel-API crystal composites as carriers for sustained drug release (as demonstrated) is a promising approach to be explored further.
Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

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Competing financial interests

The authors declare no competing financial interest.

References


Simple yet elegant—the controlled crystallization of active pharmaceutical ingredients (APIs) in supramolecular metallogel media using a natural polyphenol, tannic acid, is reported. As opposed to synthetic gelator-based media, the present system possesses inherent advantages, including ease of preparation, and the use of inexpensive components and ability to incorporate various additives, thus providing a generic platform to study the crystallization of diverse APIs.

**Keywords:** active pharmaceutical ingredients, crystallization, supramolecular gels, morphology control, composites

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