Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Food Science

Design of sporopollenin-based functional ingredients for gastrointestinal tract targeted delivery

Pien JC Schouten[1], Diana Soto-Aguilar[1], Ali Aldalbahi[2], Tansir Ahamad[2], Shaykha Alzahly[2] and Vincenzo Fogliano[1,2]

The design of intelligent delivery systems is becoming popular in the food ingredient sector. The increasing consumer demand for natural ingredients prompted researchers to look for new green solutions and use the goods of nature. Sporopollenin exine capsules (SECs) are obtained from plant pollen by washing out the proteins and genetic material. SECs are empty hollow particles that can be easily filled with hydrophobic and hydrophilic small molecular compounds, but also with proteins, enzymes, and even living probiotic bacteria. The release of compounds from the SECs is regulated by passive diffusion and can be further modulated by coating the SECs with a layer of lipids, proteins, or polysaccharides. SECs can also be functionalized to function as an intelligent delivery system that targets the intestinal mucus or the receptors located on epithelial cell membranes.

Addresses
[1] Food Quality & Design Group, Wageningen University, Wageningen, The Netherlands

[2] Department of Chemistry, College of Science, King Saud University, Riyadh, 11451, Saudi Arabia

Corresponding author: Fogliano, Vincenzo (vincenzo.fogliano@wur.nl)

Current Opinion in Food Science 2022, **44**:xx–yy

This review comes from a themed issue on **Functional foods and nutrition**

Edited by **Amanda J Wright**

https://doi.org/10.1016/j.cofs.2022.100809

Introduction

Designing functional materials to protect bioactive compounds during processing and to deliver them to the site of action has received increased interest [1]. The pharmacological sector developed efficient systems already for many years, while in food, nutrition, as well as in cosmetic fields, the utilization of the delivery system approach is still in its infancy. Vitamins, colourants, flavours, and key tastant molecules need to be protected to ensure quality until the end of the food shelf-life period. Bioactives should also be protected against degradation during transit along the gastrointestinal tract to favour uptake and utilization by the human body. During processing and storage, heat, oxygen, light, and interactions with food-matrix components can lead to the compounds' degradation [2,3]. Along the gastrointestinal tract, compounds are susceptible to highly acidic conditions and protease activity in the stomach and to the action of bile salts and pancreatic enzymes in the small intestine [4•,5,6].

Smart delivery systems could vehicle food bioactives along the gastrointestinal tract to favour the absorption of some nutrients by the small intestine. They might also contribute to increase gut health by providing microbiota suitable substrates [7] and a new generation of prebiotics can be envisaged where both the carrier and encapsulated compound contribute to the growth of a healthy microbiota [4•].

Bioactive compounds that show potential for encapsulated target release include lipids, carbohydrates, proteins, peptides, and phytochemicals such as flavonoids and carotenoids [8,9]. Delivery systems can be classified based on the target organ as saliva responsive (mouth delivery), pH responsive (gastric delivery), enzyme responsive (duodenum delivery), target delivery (bind to specific receptors), mucus adhesive, or mucus penetrating (colon delivery) [10]. The available coating strategies for target delivery within the gastrointestinal tract can be distinguished according to the target zone: acid-labile polymers that are dissolved already in the stomach [11]; protein coatings that are mainly degraded by the proteolytic activities in the duodenum [11]; and lipids, emulsions, or liposomes that are opened in the ileum [12]. Alternatively, the chemical nature of the coating can be considered as starting point. Maltodextrin/starch-based coatings are probably the most applied, followed by lipid and protein-based capsules [8]. The size and morphology of the encapsulated particles and their embedding into different food matrixes also play a role in target delivery. Folic acid decorated capsules have been developed to specifically interact with intestinal epithelial cancer cells and thereby favour the absorption of phytochemicals inducing apoptosis of these cells [13]. Intestinal target delivery systems have been proposed: mucoadhesive (strongly interact with the layer of intestinal mucus) and mucodiffusive coatings (penetrate the layer of intestinal mucus) [14]. By adhering to the intestinal mucus, these capsules can enhance the sustained release of the encapsulated compound at the target location.

All the above-described coatings can be designed by ingredient manufacturers with various sophisticated techniques. However, the characteristics of the capsules is not uniform. The mounting interest in using natural sources, prompted researchers to consider the possibilities offered by plant pollen as delivery systems. Pollen grains are made up of one of the most resistant natural material as they were developed by mother nature as a transport unit for the plant genetic material. The pollen interior is protected against harsh environmental conditions by a double-layered wall. The exine, the outer part of the pollen wall, consists mainly of sporopollenin, a biopolymer resistant to chemical and enzymatic treatment [15]. Pollen can be used as encapsulation material by removing the intine, the inner part of the pollen wall, and the cellular material to create a cavity in which compounds can be loaded. Protein-free and nucleic acid-free sporopollenin exine capsules (SECs) with different sizes and morphology can be obtained from a variety of plants [16], offering many possibilities. By further adjusting the SECs features, the compound can be released in different places of the gastrointestinal tract. In the following paragraphs, we will explain how SECs can be used to protect unstable bioactive compounds both during food industrial processing as well as during the gastrointestinal passage. We aim to provide an insight into the possibilities to design sporopollenin-based functional ingredients for food applications and various biological targets.

Encapsulation with sporopollenin: challenges and opportunities

As shown in Figure 1, pollen grains cover a large variety of shapes and sizes [17], but within single pollen species, size and morphology are uniform. This makes pollen capsules an interesting alternative to current encapsulation technologies that face the difficulty of ensuring particles homogeneity. Because of a large variety in pollen among the plant kingdom, the pollen with the right shape and grain size can be selected for different applications.

In Figure 2, a sketch of a pollen grain is shown. The outer layer of the pollen wall, the exine, consists mainly of sporopollenin [18••]. SECs can be obtained from pollen grains by removing the pollen's interior: the intine and the cellular material [19–21]. Removal of all interior and surface pollen constituents creates a large SEC cavity and eliminates the allergenicity of the pollen while keeping the native microstructure intact, which is a key factor for successive applications.

The pollen structure varies a lot between pollen species (Figure 1) and therefore requires different extraction methods. Extraction strategies include the use of organic solvents and alkaline or acid hydrolysis. These procedures are very laborious, as they need extensive washing and multiple drying steps, and can also result in SEC damage [22]. Therefore, new strategies are being developed to provide milder and easier extraction procedures [23,24••,25]. Multiple studies successfully kept the pollen's microstructure intact by using ionic liquids to remove cellular material and the intine of the pollen [23,24••,26]. Alternatively, SECs can also be obtained by exposing the pollen to a cocktail of enzymes, but this procedure is longer and more expensive [24••]. Thomasson *et al.* recently found a one-step extraction with 9 M HCl that resulted in *Lycopodium clavatum* SECs that left the antioxidant properties of encapsulated ω-3 oils unaffected [27•], thereby providing a cheap and easy alternative for industrial-scale application. However, the effect of this treatment on the whole exine microstructure has not yet been studied. For pine pollen SECs, hydrochloric acid alone was found to be insufficient to effectively remove the proteins, instead, 85% phosphoric acid could be used [28]. Improving the extraction methods, with a special focus on pollen species that are more prone to damage, is still a main need [29].

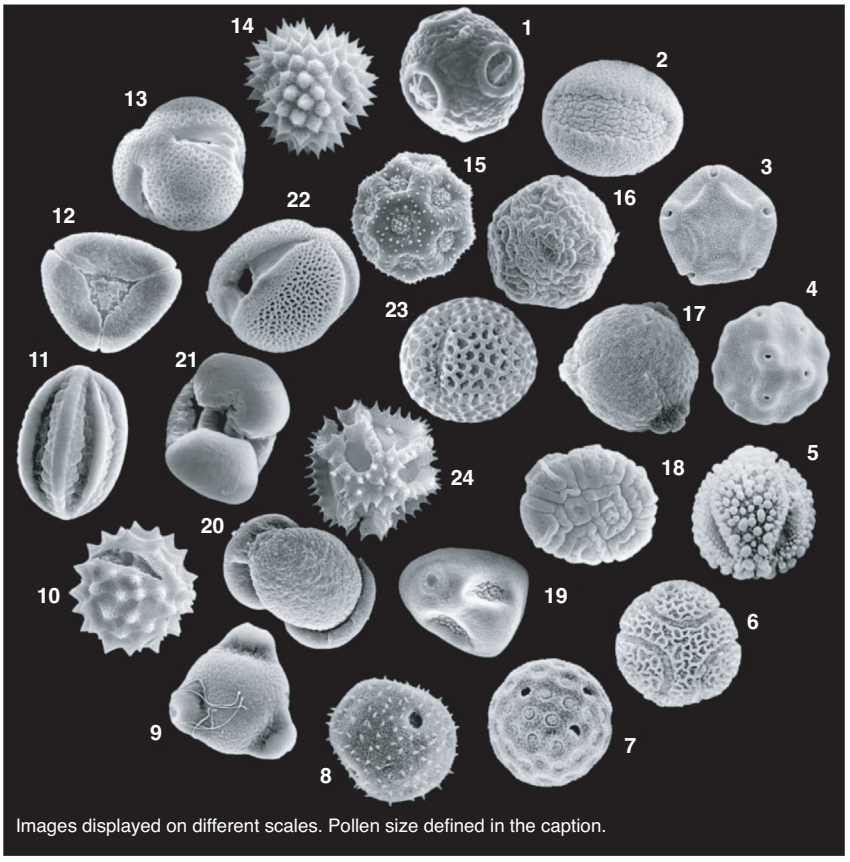
The empty SECs are a perfect material to be re-filled with the compound of interest. Bioactive loading can be achieved by physically adsorbing onto the surface of the SECs or by diffusing into the SECs' interior through the nanochannels of the exine membrane via vacuum-assisted diffusion [30,31]. Also bacteria can be loaded through a procedure named tableting: the SECs are subjected to pressure which opens up the aperture through which the bacterial cell can enter. Because of the SECs' elasticity, the aperture will close again [32].

The loading capacity, expressed as the w/w percentage of compound versus carrier, is an important parameter to monitor the efficiency of the encapsulation process. The loading technique has a huge effect on the loading capacity [33]. For instance, looking at the 5-fluorouracil loading into club moss SECs, vacuum loading was more effective than either tableting or passive diffusion [33].

Other factors affecting loading capacity are the type [22] and size of the pollen [34]. Most pollen species have a similar wall thickness, therefore, the loading factor increases with a bigger exine diameter [34]. The type of compound to be encapsulated also determines the loading capacity [33,35], with low soluble compounds resulting in a lower loading efficiency. To increase loading, Paunov *et al.* [35] developed a method in which two high soluble compounds are filled into the SECs. Once in the SECs, the two compounds react, forming a compound with lower solubility. Moreover, the compound size influences the loading as well, as the compound needs to enter via either the nanochannels or the aperture of the pollen.

The same nanochannels through which the capsules are filled during vacuum-assisted diffusion can also release the encapsulated compound once the ingredient is in the

Figure 1



SEM microscopy picture of pollen from different plants. Picture retrieved from Suárez-Cervera *et al.* [17]. Pollen morphological diversity by SEM. (1). Fumariaceae, Fumaria capreolata (18.5 μm); (2). Arecaceae, Phoenix canariensis (30 × 17 μm); (3). Betulaceae, Alnus glutinosa (18 × 23 μm); (4). Juglandaceae, Juglans regia (40 × 35 μm); (5). Aquifoliaceae, Ilex aquifolium (25 × 21 μm); (6). Passifloraceae, Passiflora incarnata (49 μm); (7). Caryophyllaceae, Silene legionensis (33 μm); (8). Campanulaceae, Campanula herminii (29 μm); (9). Onagraceaae, Epilobium angustifolium (65 μm); (10). Asteraceae, Aster sedifolius (30 μm); (11). Ephedraceae, Ephedra sp. (32.5 × 20 μm); (12). Myrtaceae, Feijoa sellowiana (18.5 μm); (13). Euphorbiaceae, Neoguillauminia cleopatra (41 × 37 μm); (14). Asteraceae, Arnica montana (22.5 μm); (15). Ranunculaceae, Anemone sp. (21.7 μm); (16). Ulmaceae, Ulmus minor (33.5 μm); (17). Saxifragaceae, Saxifraga granulata (27 × 25 μm); (18). Mimosaceae, Acacia spinosa (32 × 27 μm); (19). Cyperaceae, Carex sp. (24 × 20 μm); (20). Pinaceae, Pinus radiata (72 μm); (21). Pinaceae, Pinus halepensis (65 μm); (22). Zygophyllaceae, Zygophyllum fabago (14 × 16 μm); (23). Oleaceae, Ligustrum vulgare (29 × 25 μm); (24). Asteraceae, Taraxacum officinalis (26630 μm).

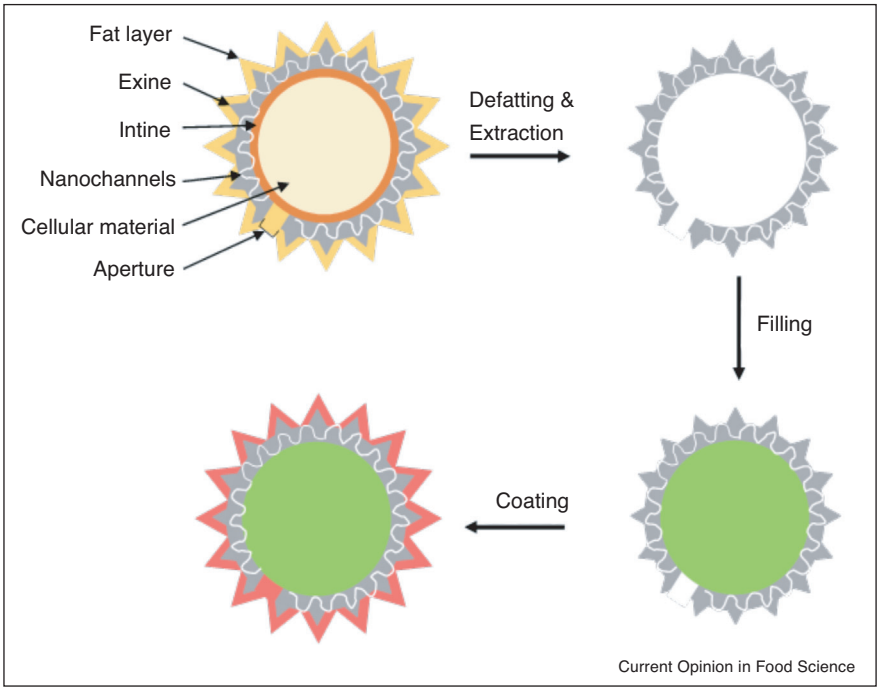
food or within the gastrointestinal tract. Depending on the pollen source, these nanochannels can range in size. Large pores such as in sunflower pollen lead to rapid leakage of the compound from the SECs [36]. When a slow sustained release is required, SECs can be coated to provide additional functionality with chitosan [37], zein/tannic acid [38•], or polymethacrylate [33].

Considering the possibility of adding encapsulated bioactives in a food product, it is of interest to investigate how the physical characteristics of the food matrix could influence the release of the bioactives from the SECs. The food matrix might behave as an external coating and limit the bioactive diffusion. Another interesting field for future study is the functionalization of SECs outer surface taking advantage of the reactivity of the hydroxyl groups on the sporopollenin polymer [26,39]. In this way, target delivery could be achieved by decorating the SECs with molecules that bind to specific areas of the gastrointestinal tract [13].

Effect of sporopollenin encapsulation on stability to digestion and gut microbiota delivery

In this case, the goal of encapsulating bioactive compounds is to protect them during the gastric phase and

Figure 2



Sketch of a (sunflower) pollen grain that is subjected to defatting and extraction to obtain a sporopollenin exine capsule (SEC). This is followed by filling the SEC with a compound (green) and subsequent coating (red) of the filled SEC.

favour a slow release from the SECs into the intestines and/or to cross the intestinal epithelial barrier. Small molecular weight bioactives (hydrophilic or hydrophobic), such as enzymes, antibiotics, drugs, and vitamins, have been successfully incorporated in the SECs following the procedures described above [34]. An overview of articles that studied the release of a compound from coated or uncoated SECs in different digestion models is reported in Table 1.

As discussed in the previous section, the release of compounds from the SECs depends on the size of the nanochannels and by the presence of an additional coating on the SECs' surface. Multiple studies found that uncoated sunflower SECs resulted in a rapid release of compounds during digestion [21,38•,40]. Most likely this is because the sunflower SECs have large channels or an exposed aperture through which the compound can easily be released. Therefore, to sustain the release, sunflower SECs are usually covered by a layer of polysaccharides [40,41] or proteins [38•]. The target release of the SECs can be improved by applying pH sensitive or enzyme degradable coatings.

Club moss SECs have been largely investigated: some studies found minimal release of compounds during the gastric phase [42,43], whereas others found a burst release [31,33]. This is likely due to the fact the SECs were produced by different extraction procedures. Even though club moss might indeed be capable of providing sustained release in the gastric phase, gastric fluids may enter the SECs and modify the release fluxes. Therefore, also for club moss, the use of additional coating is advised to provide sustained release of compounds and to protect the compounds within the SECs.

A sustained release of bioactives from the SECs is desired to achieve a slow and continuous absorption over time. Adhesion of the SECs to the mucosa is a very efficient way to prolong exposure to the bioactives and in this respect, the selection of an appropriate coating is key. SECs themselves have good mucoadhesive properties [42,44] but they could be further enhanced by a chitosan [45] or zein [46] coating which can adhere to the colonocyte mucus, thereby prolonging the contact time of the compound with the epithelial layer [47].

Interestingly, it was observed that in some cases the entire SEC can cross the intestinal epithelial barrier and reach the bloodstream [48,49]. This happens thanks to a phenomenon called 'persorption', which allows the passage from the intestine to the bloodstream of

Table 1

Overview of studies that show the effect of sporopollenin encapsulation as a delivery system on the bioavailability of bioactive compounds. SGF = simulated gastric fluid; SIF = simulated intestinal fluid; SLIF = simulated large intestine fluid; PBS = physiological buffer solution

Pollen type	Coating	Compound	Digestion model	Results	Reference
Clubmoss	–	Erythromycin (EM) and bacitracin	*In vitro* in dialysis bag with PBS and *in vivo* with adult male albino rats	*In vitro* the initial burst release was followed by a sustained release. *In vivo* the total drug exposure over time in plasma was higher for encapsulated EM compared to the control.	[44]
Clubmoss	–	Ibuprofen	*In vitro* SGF and PBS	Maximum release of 12% was found in a SGF and a burst release was found in a PBS.	[42]
Clubmoss	–	Folic acid	*In vitro* SGF and SIF in dialysis bag	A sustained release in SGF and a faster but sustained release in SIF. After 10 hour up to 45.5% and 94.4% was released in SGF and SIF, respectively.	[43]
Clubmoss	–	*Lactobacillus casei*	*In vitro* fasted SGF, SIF and SLIF. *In vitro* fed SGF, SIF and SLIF	The viability of *L. casei* was higher in a fed state compared to a fasted state. The protective effect of encapsulation was most pronounced in the fed state.	[32]
Clubmoss	Carboxymethyl-pachymaran	β-Galactosidase	*In vitro* SGF and SIF	A burst release in SGF without coating and a sustained release in SIF with increased coating concentration.	[31]
Clubmoss	Eudragit RS100 (2.5–10%)	5-fluoroucil	*In vitro* SGF and SIF	A burst release in SGF without coating and a sustained release in SIF with increased coating concentration.	[33]
Date palm	Chitosan	Ibuprofen	*In vitro* SGF and PBS	A cumulative release with coating in SGF (42.40%) and in PBS (94.20%).	[37]
Sunflower	Alginate	Bovine serum albumin	*In vitro* SGF and PBS	A burst release in SGF and PBS without coating. With coating there was a more sustained release in SGF compared to PBS.	[21]
Sunflower	Alginate	Nobiletin	*In vitro* SGF and SIF	~20% released in SGF without coating and 2% with coating. A slow release in SIF without coating and a sustained release up to 100 hour with coating.	[54]
Sunflower	Carboxymethyl-pachymaran (CMP) and CMP/metal ion	β-Galactosidase	*In vitro* SGF and SIF	A burst release in SGF without coating. With CMP there was a sustained release in SGF and SIF. The addition of metal ions ($CaCl_2$ and $AlCl_3$) led to a more long-term release and decreased the required CMP dosage.	[40]
Sunflower	Zein and zein/tannic acid	β-Galactosidase	*In vitro* SGF and SIF	A burst release in SGF without a coating or with a zein coating. A sustained release in SIF with a zein/tannic acid coating.	[38•]

micrometric particles. Persorption is still poorly studied in human physiology [50]: evidence is still weak for persorption of particles larger than 2 μm [51] and even weaker for particles bigger than 20 μm. However, a recent article showed that club moss SECs, with a size of 25 μm, were able to cross the epithelial surface and enter the intestinal wall of mice [50] also showing the potential of persorption in applications seeking for intact delivery to the bloodstream from oral administration. Several older studies also found that some macromolecules and particles crossed the intestinal wall intact [52] and into the bloodstream [51]. Diego-Taboada *et al.* found that SECs will break down in the bloodstream and release their content thereby opening interesting drug delivery possibilities [34]. Up to now, it is unclear to what extent the SECs penetrate the intestinal wall and Fasano [53] stated that persorption is unlikely to be an appropriate delivery mechanism for a therapeutic dose of drugs. Persorption mechanisms deserve further investigations as they can provide sporopollenin encapsulation with additional benefits.

Sensory opportunities of sporopollenin encapsulated ingredients

Besides compound protection during processing and along the gastrointestinal tract, SECs also offer opportunities to improve the food sensory experience. Examples are taste masking, colour masking, and protection of key taste compounds during shelf life.

It has been shown that SECs possess good taste-masking properties, a beneficial trait to incorporate compounds with a strong undesirable taste in food products. To illustrate, the bitter-tasting ibuprofen [42] and fish oils with strong unpalatable flavours [41] were successfully masked by encapsulating it within SECs. This also poses opportunities for fortifying functional foods with phenolic compounds that would otherwise be disliked by the consumer due to astringency and bitterness. Similarly, unwanted colour compounds can be masked by SECs encapsulation. This can be interesting when green or blue microalgae-based ingredients are used in fortified bakery or dairy products. In these cases, the strong chlorophyll

green colour, which also turns brownish during processing and shelf life, is usually not desired. On the other hand, SECs might also be able to protect colourants and tastants during shelf life. Once dissolved in a liquid, these compounds presumably escape the uncoated SECs, which can be especially interesting for powdery products that need to be dissolved before usage. Up to date, research on the potential ability of colour masking or protection by SECs encapsulation is lacking. It should be considered that SECs >30 μm can start to feel gritty in the mouth [34].

Conclusions

Sporopollenin is a natural polymer that is still poorly utilized in the food sector. The availability of effective systems to obtain an allergen-free versatile tool for encapsulation and delivery of a large variety of food bioactives make this resource valuable for research and practical applications.

In comparison with other commonly used delivery systems in food ingredients, SECs have some advantages, such as: the high loading factor; the possibility to incorporate both hydrophobic and hydrophilic compounds in a wide range of molecular weight; and the facile modulation of the kinetic release by varying the cavity diameter as well as the physical-chemical characteristics of the coating. While pollen is a General Recognized as Safe material, SECs as an ingredient should undergo the novel food procedure to be used in commercial products. From the researchers' point of view, SECs represent a very interesting opportunity to investigate the behaviour of encapsulated ingredients in several foods, using *in vitro* cell studies and artificial gut systems.

Conflict of interest statement

Nothing declared.

Acknowledgement

The authors acknowledge the King Saud University, Riyadh, Saudi Arabia, for funding this work through Research Supporting (Project No. RSP-2021/30).

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Đorđević V, Balanč B, Belščak-Cvitanović A, Lević S, Trifković K, Kalušević A, Kostić I, Komes D, Bugarski B, Nedović V: **Trends in encapsulation technologies for delivery of food bioactive compounds**. *Food Eng Rev* 2015, **7**:452-490.

2. Liu Y, Yang R, Liu J, Meng D, Zhou Z, Zhang Y, Blanchard C: **Fabrication, structure, and function evaluation of the ferritin based nano-carrier for food bioactive compounds**. *Food Chem* 2019, **299**:125097.

3. Nahum V, Domb AJ: **Recent developments in solid lipid microparticles for food ingredients delivery**. *Foods* 2021, **10**:1-25.

4. • Nowak E, Livney YD, Niu Z, Singh H: **Delivery of bioactives in food for optimal efficacy: what inspirations and insights can be gained from pharmaceutics?** *Trends Food Sci Technol* 2019, **91**:557-573

This recent review paper provides an overview of the take home message from pharmaceutical studies that can be implemented in the design of intelligent delivery systems for functional foods.

5. Bao C, Jiang P, Chai J, Jiang Y, Li D, Bao W, Liu B, Liu B, Norde W, Li Y: **The delivery of sensitive food bioactive ingredients: absorption mechanisms, influencing factors, encapsulation techniques and evaluation models**. *Food Res Int* 2019, **120**:130-140.

6. Zhang Z, Zhang R, McClements DJ: **Lactase (β-galactosidase) encapsulation in hydrogel beads with controlled internal pH microenvironments: impact of bead characteristics on enzyme activity**. *Food Hydrocoll* 2017, **67**:85-93.

7. Ercolini D, Fogliano V: **Food design to feed the human gut microbiota**. *J Agric Food Chem* 2018, **66**:3754-3758.

8. McClements DJ: *Nanoparticle-and Microparticle-based Delivery Systems: Encapsulation, Protection and Release of Active Compounds*. CRC Press; 2019.

9. Giaconia MA, dos P Ramos S, Pereira CF, Lemes AC, De Rosso VV, Braga ARC: **Overcoming restrictions of bioactive compounds biological effects in food using nanometer-sized structures**. *Food Hydrocoll* 2020, **107**:105939.

10. Chai J, Jiang P, Wang P, Jiang Y, Li D, Bao W, Liu B, Liu B, Zhao L, Norde W, Yuan Q, Ren F, Li Y: **The intelligent delivery systems for bioactive compounds in foods: physicochemical and physiological conditions, absorption mechanisms, obstacles and responsive strategies**. *Trends Food Sci Technol* 2018, **78**:144-154.

11. Salem A, Ramadan AR, Shoeib T: **Entrapment of β-carotene and zinc in whey protein nanoparticles using the pH cycle method: evidence of sustained release delivery in intestinal and gastric fluids**. *Food Biosci* 2018, **26**:161-168.

12. Berton-Carabin C, Schroën K: **Towards new food emulsions: designing the interface and beyond**. *Curr Opin Food Sci* 2019, **27**:74-81.

13. Su C, Li H, Shi Y, Wang G, Liu L, Zhao L, Su R: **Carboxymethyl-β-cyclodextrin conjugated nanoparticles facilitate therapy for folate receptor-positive tumor with the mediation of folic acid**. *Int J Pharm* 2014, **474**:202-211.

14. Semenova M, Antipova A, Martirosova E, Zelikina D, Palmina N, Chebotarev S: **Essential contributions of food hydrocolloids and phospholipid liposomes to the formation of carriers for controlled delivery of biologically active substances via the gastrointestinal tract**. *Food Hydrocoll* 2021, **120**:106890.

15. Shaw G: **The chemistry of sporopollenin**. In *Sporopollenin.* Edited by Brooks J, Grant PR, Muir M, Gijzel PV, Shaw G. Academic Press Inc.; 1971:305-334.

16. Mackenzie G, Boa AN, Diego-Taboada A, Atkin SL, Sathyapalan T: **Sporopollenin, the least known yet toughest natural biopolymer**. *Front Mater* 2015, **2**:66.

17. Suárez-Cervera M, Vega-Maray A, Castells T, Rodríguez-Rajo FJ, Asturias JA, Le Thomas A, Seoane-Camba JA: **An approach to the knowledge of pollen and allergen diversity through lipid transfer protein localisation in taxonomically distant pollen grains**. *Grana* 2008, **47**:272-284.

18. •• Halbritter H, Ulrich S, Grímsson F *et al.*: **Pollen morphology and ultrastructure**. *Illus Pollen Terminol* 2018:37-65 http://dx.doi.org/10.1007/978-3-319-71365-6_3. Published online

A complete picture of pollen morphology and ultrastructure is given in this book. It illustrates the large variety of pollen in nature, in this way inspiring ideas about the potential application in functional ingredient design.

19. Mundargi RC, Potroz MG, Park JH, Seo J, Lee JH, Cho NJ: **Extraction of sporopollenin exine capsules from sunflower pollen grains**. *RSC Adv* 2016, **6**:16533-16539.

20. Quilichini TD, Grienenberger E, Douglas CJ: **The biosynthesis, composition and assembly of the outer pollen wall: a tough case to crack**. *Phytochemistry* 2015, **113**:170-182.

21. Mundargi RC, Potroz MG, Park S, Shirahama H, Lee JH, Seo J, Cho NJ: **Natural sunflower pollen as a drug delivery vehicle**. *Small* 2016, **12**:1167-1173.

22. Fan T, Park JH, Pham QA, Tan EL, Mundargi RC, Potroz MG, Jung H, Cho NJ: **Extraction of cage-like sporopollenin exine capsules from dandelion pollen grains**. *Sci Rep* 2018, **8**:1-11.

23. Chiappe C, Demontis GC, Di Bussolo V, Rodriguez Douton MJ, Rossella F, Pomelli CS, Sartini S, Caporali S: **From pollen grains to functionalized microcapsules: a facile chemical route using ionic liquids**. *Green Chem* 2017, **19**:1028-1033.

24. •• Hegedüs K, Fehér C, Jalsovszky I, Kristóf Z, Rohonczy J, Vass E, Farkas A, Csizmadia T, Friedbacher G, Hantz P: **Facile isolation and analysis of sporopollenin exine from bee pollen**. *Sci Rep* 2021, **11**:1-16

This paper provides an excellent example of a facile extraction process of protein and genetic material to produce the SEC, allowing to overcome one of the main bottlenecks related to their utilization.

25. Corliss MK, Bok CK, Gillissen J, Potroz MG, Jung H, Tan EL, Mundargi RC, Cho NJ: **Preserving the inflated structure of lyophilized sporopollenin exine capsules with polyethylene glycol osmolyte**. *J Ind Eng Chem* 2018, **61**:255-264.

26. Palazzo I, Mezzetta A, Guazzelli L, Sartini S, Pomelli CS, Parker WO, Chiappe C: **Chiral ionic liquids supported on natural sporopollenin microcapsules**. *RSC Adv* 2018, **8**:21174-21183.

27. • Thomasson MJ, Diego-Taboada A, Barrier S, Martin-Guyout J, Amedjou E, Atkin SL, Queneau Y, Boa AN, Mackenzie G: **Sporopollenin exine capsules (SpECs) derived from *Lycopodium clavatum* provide practical antioxidant properties by retarding rancidification of an ω-3 oil**. *Ind Crops Prod* 2020, **154**:112714

This paper shows the applications of SECs for taste masking by preventing polyunsaturated lipid oxidation.

28. Prabhakar AK, Lai HY, Potroz MG, Corliss MK, Park JH, Mundargi RC, Cho D, Bang SI, Cho NJ: **Chemical processing strategies to obtain sporopollenin exine capsules from multi-compartmental pine pollen**. *J Ind Eng Chem* 2017, **53**:375-385.

29. Park JH, Seo J, Jackman JA, Cho NJ: **Inflated sporopollenin exine capsules obtained from thin-walled pollen**. *Sci Rep* 2016, **6**:1-10.

30. Wu D, Liang Y, Huang K, Jing X, Li B, Liang H: **Leveraging plant exine capsules as pH-responsive delivery vehicles for hydrophobic nutraceutical encapsulation**. *Food Funct* 2018, **9**:5436-5442.

31. Deng Z, Pei Y, Wang S, Zhou B, Li J, Hou X, Li J, Li B, Liang H: **Carboxymethylpachymaran entrapped plant-based hollow microcapsules for delivery and stabilization of β-galactosidase**. *Food Funct* 2019, **10**:4782-4791.

32. Stamatopoulos K, Kafourou V, Batchelor HK, Konteles SJ: **Sporopollenin exine microcapsules as potential intestinal delivery system of probiotics**. *Small* 2021, **17**:1-12.

33. Mundargi RC, Tan EL, Seo J, Cho NJ: **Encapsulation and controlled release formulations of 5-fluorouracil from natural *Lycopodium clavatum* spores**. *J Ind Eng Chem* 2016, **36**:102-108.

34. Diego-Taboada A, Barrier S, Thomasson M, Atkin S, Mackenzie G: **Pollen: a novel encapsulation vehicle for drug delivery**. *Innov Pharm Technol* 2007, **24**:63-68.

35. Paunov VN, MacKenzie G, Stoyanov SD: **Sporopollenin micro-reactors for in-situ preparation, encapsulation and targeted delivery of active components**. *J Mater Chem* 2007, **17**:609-612.

36. Potroz MG, Mundargi RC, Gillissen JJ, Tan EL, Meker S, Park JH, Jung H, Park S, Cho D, Bang SI, Cho NJ: **Plant-based hollow microcapsules for oral delivery applications: toward optimized loading and controlled release**. *Adv Funct Mater* 2017, **27**:1-12.

37. Alshehri SM, Al-Lohedan HA, Chaudhary AA, Al-Farraj E, Alhokbany N, Issa Z, Alhousine S, Ahamad T: **Delivery of ibuprofen by natural macroporous sporopollenin exine capsules extracted from Phoenix dactylifera L**. *Eur J Pharm Sci* 2016, **88**:158-165.

38. • Deng Z, Wang S, Pei Y, Zhou B, Li J, Hou X, Li B, Liang H: **Tuning of molecular interactions between zein and tannic acid to modify sunflower sporopollenin exine capsules: enhanced stability and targeted delivery of bioactive macromolecules**. *ACS Appl Bio Mater* 2021, **4**:2686-2695

This paper opens the possibility of further coating and decorating SECs surface to obtain a sustained release during digestion or to add smart delivery functionalities.

39. Pellegrini N, Vittadini E, Fogliano V: **Designing food structure to slow down digestion in starch-rich products**. *Curr Opin Food Sci* 2020, **32**:50-57.

40. Deng Z, Pei Y, Wang S, Zhou B, Hou X, Li J, Li B, Liang H: **Designable carboxymethylpachymaran/metal ion architecture on sunflower sporopollenin exine capsules as delivery vehicles for bioactive macromolecules**. *J Agric Food Chem* 2020, **68**:13990-14000.

41. Barrier S, Rigby AS, Diego-Taboada A, Thomasson MJ, Mackenzie G, Atkin SL: **Sporopollenin exines: a novel natural taste masking material**. *LWT Food Sci Technol* 2010, **43**:73-76.

42. Diego-Taboada A, Maillet L, Banoub JH, Lorch M, Rigby AS, Boa AN, Atkin SL, Mackenzie G: **Protein free microcapsules obtained from plant spores as a model for drug delivery: ibuprofen encapsulation, release and taste masking**. *J Mater Chem B* 2013, **1**:707-713.

43. Mohammed ASY, Dyab AKF, Taha F, Abd El-Mageed AIA: **Encapsulation of folic acid (vitamin B9) into sporopollenin microcapsules: physico-chemical characterisation, in vitro controlled release and photoprotection study**. *Mater Sci Eng C* 2021, **128**:112271.

44. Dyab AKF, Mohamed MA, Meligi NM, Mohamed SK: **Encapsulation of erythromycin and bacitracin antibiotics into natural sporopollenin microcapsules: antibacterial, cytotoxicity, in vitro and in vivo release studies for enhanced bioavailability**. *RSC Adv* 2018, **8**:33432-33444.

45. Chuah LH, Roberts CJ, Billa N, Abdullah S, Rosli R: **Cellular uptake and anticancer effects of mucoadhesive curcumin-containing chitosan nanoparticles**. *Colloids Surf B Biointerfaces* 2014, **116**:228-236.

46. Deng Z, Wang S, Zhou B, Li J, Zhou P, Li B, Liang H: **Carboxymethylpachymaran-zein coated plant microcapsules-based β-galactosidase encapsulation system for long-term effective delivery**. *Food Res Int* 2020, **128**:108867.

47. Diego-Taboada A, Beckett ST, Atkin SL, Mackenzie G: **Hollow pollen shells to enhance drug delivery**. *Pharmaceutics* 2014, **6**:80-96.

48. Barrier S: *Physical and Chemical Properties of Sporopollenin Exine Particles. PhD Thesis*. Hull: University of Hull; 2008.

49. Blackwell LJ: *Sporopollenin Exines as a Novel Drug Delivery System. PhD Thesis*. Hull: University of Hull; 2007.

50. Atwe SU, Ma Y, Gill HS: **Pollen grains for oral vaccination**. *J Control Release* 2014, **194**:45-52.

51. Weiner ML: **Intestinal transport of some macromolecules in food**. *Food Chem Toxicol* 1988, **26**:867-880.

52. Volkheimer G: **Passage of particles through the wall of the gastrointestinal tract**. *Environ Health Perspect* 1974, **9**:215-225.

53. Fasano A: **Novel approaches for oral delivery of macromolecules**. *J Pharm Sci* 1998, **87**:1351-1356.

54. Wu D, Liang Y, Pei Y, Li B, Liang H: **Plant exine capsules based encapsulation strategy: a high loading and long-term effective delivery system for nobiletin**. *Food Res Int* 2020, **127**:108691.