

Chitosan as Coating for Biocontrol

Subjects: Biology

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This entry focused on the scientific production, trends, and characteristics of a knowledge domain of high worldwide importance, namely, the use of chitosan as a coating for postharvest disease biocontrol in fruits and vegetables, which are generated mainly by fungi and bacteria such as *Aspergillus niger*, *Rhizopus stolonifera*, and *Botrytis cinerea*. For this, the analysis of 875 published documents in the Scopus database was performed for the years 2011 to 2021. The information of the keywords' co-occurrence was visualized and studied using the free access VOSviewer software to show the trend of the topic in general.

Keywords: chitosan ; coating ; biocontrol ; postharvest ; fruit ; vegetables

1. Introduction

The most effective and used edible coatings for the protection of fruits and vegetables are made up one or more natural polymers such as cellulose [1], alginate [2], gellan [3], pectin, starch and its derivatives [4], methylcellulose, carboxymethylcellulose [5], Arabic gum [6], whey protein concentrate [7], and chitosan or chitosan nanoemulsion [8][9]. Chitosan is a deacetylated form of chitin, (poly β -(1 \rightarrow 4) N-acetyl-d-glucosamine), and is the second most abundant biopolymer found in nature after cellulose, with prominent film-forming properties, non-toxicity, biodegradable and biocompatible properties, high mechanical strength, and excellent antimicrobial activity [10], and it has been used as a coating in various foods [11]. Furthermore, chitosan has been approved by the United States Food and Drug Administration (USFDA) as a food additive and listed as generally recognized as safe (GRAS) in the USA and Japan

In the last years, chitosan has gained more attention from researchers due to its broad-spectrum activity and high destruction rate against Gram-positive and Gram-negative bacteria [12] and filamentous fungi [13][14][15]. However, its quality, chemical and biological properties, and therefore its applications are closely related to numerous intrinsic and extrinsic factors such as the degree of deacetylation (DD). Chitosan is a polysaccharide mainly obtained from invertebrates; insect cuticles; fungal cell walls; green algae; yeast; crustacean shells such as those of cockles, shrimp, crabs, etc.; by chemical (alkaline hydrolysis with NaOH solutions) and other less-used methods such as alkaline treatment at high temperature and high pressure [16]; or a process that uses ultrasound [17][18][19] and sometimes, less frequently, enzymatic deacetylation [20]. Moreover, it is important to mention that the method used increases or decreases the deacetylation degree (DD) and determines the content of free amino groups and the cationic character [21].

On the other hand, the DD grade of chitosan indicates the number of acetyl groups removed from chitin, which corresponds to the release of the amino groups from the N-acetylglucosamine monomers. In this regard, He et al. [22] mentioned the chitosan classification according to its DD as being low between 55 and 70%, medium between 70 and 85%, high between 85 and 95%, and ultra-high between 95 and 100% [22][23]. [24] found that the chitosan microspheres with a 63.6% degree of deacetylation (DD) exhibited the highest antibacterial activity concerning the microspheres made with chitosan at 83.7% of DD, which exerted at least some antibacterial activity.

Furthermore, although the antifungal or antimicrobial mechanism of action of chitosan is still being studied, some hypotheses have already been proposed, e.g., Yang [25] mentioned that this polymer permeates and perforates the fungus nuclei, the protein cell membranes, and intracellular constituents, inactivating bacterial metabolism due to the presence of an amino group that has a positive charge with a pH lower than 6.3 and interacts with negative charges of the cell wall of microorganisms, generating the rupture or lysis of these structures, causing the loss of protein compounds and other intracellular constituents [26]. However, because there are still many factors that need to be analyzed during the coating process such as the coating suspensions properties, the fruit surface microstructure and wettability [1], and the synergy between biocontrol agents and natural bioactive compounds, it is necessary that more studies be conducted [27].

On the other hand, due to the vast amount of existing information, an “intoxication” problem can be generated, as reported by Flórez-Martínez et al. (2021) [28]. Consequently, in this paper, we carried out a bibliometric analysis to show and quantify the evolution of the research, perspectives, challenges, and prospects, not only a cross-sectional study, as this provides limited information.

2. Bibliometric Analysis

For this, a Boolean search string was used. First, the search was realized for those coming from the relevant keyword fields (e.g., chitosan, postharvest, biocontrol, fungi, and phytopathogens). Subsequently, the Scopus service was used, with the option to combine searches, using the “Combine queries” field, where the syntax applied is the # symbol with the “OR” and “AND” operators. AND (EXCLUDE (EXACTKEYWORD, “Biomedical Applications”)); in order to limit the topic additional phrases, (AND (LIMIT-TO (EXACTKEYWORD, “Fruits”))) was added into the query string, which resulted in 92 articles [29][30].

The total number of searches resulted in 875 publications (from January 2011 to 14 April 2021). The raw data obtained (CSV Format) from Scopus was analyzed using the VOSviewer software (www.vosviewer.com, accessed on 25 March 2021; Van Eck and Waltman, 2009–2020, version 1.6.15, Leiden University, Leiden, The Netherlands) for the construction and bibliometric visualization of networks of institutions, countries, keywords, and citations per article.

San Jose, CA, USA) was used to design and produce the graph shown in Figure 1; then, the “Dynamic fit wizard” plugin was applied for curve fitting, using a linear regression model. The obtained equation was Equation (1). $(1)f=y_0+a*x$ where Y = response (number of published papers);

3. Results

Figure 1 shows the number of scientific publications per year, limiting this research to a period from January 2011 to 14 April 2021. It can be observed that the temporal evolution between the number of articles versus years, in this field of research, has had linear growth in the last decade. From 2011 to 2013, few documents per year were published on the subject (11, 34, and 41, respectively), but these have increased. In order to perform the data analysis and have it be explained by a linear regression model using the best fit ($R^2 = 0.9859$) of the results, we did not consider this year and only included studies until the year 2020, as shown in Figure 1.

In all published documents, the central focus of an article is highlighted using keywords, which are essential and facilitate mapping for readers [29] so that their analysis is necessary. One method is by word cloud (Figure 2), which provides a first view of the dataset, allowing us to explore and visually analyze, as well as to size and create the first classification for our data. The size of the words “chitosan” and “coatings” suggests that in most research, these words have been the most persistent theme. However, this technique only provides qualitative information, and therefore it is necessary to execute a more in-depth analysis.

A more accurate method is the co-keyword cluster mapping (Figure 3), obtained from author keywords. Here, the software (VOSviewer) analyzed the 84 most frequently terms, and each one of them was repeated at least six times. It is imperative to mention that each circle represents a keyword, and its size indicates its appearance frequency in the articles. Data analysis generated nine clusters marked with different colors, e.g., the first cluster (in red) contained 19 terms, with “chitosan” being the most frequent term, with a greater node keyword, closely associated with the 81 terms belonging at the nine clusters, but mainly with largest nodes such as “edible coating, coating, shelf life, antibacterial activity”.

It is important to note that the node’s color also determined when the term or keyword was introduced for the first time in the network [14]. These results could be significant for the scientific development research that involves the topic of food waste that generates so many economic losses. For example, the generation of coatings involved added nanoparticles for protection and longer shelf life of different foods. However, much research is still needed to establish the existing interactions between food matrices and these coatings.

On the other hand, as shown in Figure 4, the keyword “chitosan”, highlighted with the larger circle in blue, also determined a central position, indicating its importance and direct connection with other smaller nodes such as “fruit coating”, “useful life”, “strawberry”, “mango”, “guava”, “tomato”, and “papaya”. Terms that gain importance as will be seen later.

On the other hand, Table 1 shows the top 20 most-cited articles, extracted from the search of 875 documents. Obtained data such as the year of publication, authorship, journal title, publication count, and citation count were analyzed. Due to the high quantity of published papers, our analysis focused on the most highly cited papers and those related to the

keyword “fruits”, which generated 92 documents, analyzed as described in Table 2, Table 3 and Table 4.

The top of 20 most cited authors and documents between 2011 and 2021.

This study was published in Scientific Reports and intended to explore the interaction pattern role of the iron oxide nanoparticle (IONP)–bacteria interface that enhances the antimicrobial activity of IONP using positively charged chitosan. In analyzing the rest of the authors and the most cited scientific papers in the domain under study, we noted the importance of the use of chitosan and its multiple applications as well as their effect as a coating in various fruits. In this sense, the second most cited document [31] reported the use of chitosan as an effective control in reducing weight loss, maintaining firmness, delayed changes in the peel color, and soluble solids in papaya (*Carica papaya* L.), which is one of the most important fruit crops in the world and has a short post-harvest life. However, it did not study the damage by opportunistic plant pathogens capable of producing diseases or loss of crops, which has led to countless studies, as shown in Table 2, Table 3 and Table 4.

It is worth noting that the emerging interdisciplinary field of nanotechnology has been a recurring phenomenon in recent studies, as shown in Table 1, with the highest cited document or the documents published by various authors [32][33][34][35].

Exceptionally, the co-occurrence between keywords allows for the generation of knowledge in search of a common goal. Over the past decade, researchers around the world have developed many different methods to minimize postharvest fruit loss because they have the highest waste rates of any food product (45% waste [36]), which is a global problem. A novel method is the use of chitosan coatings as well as their different combinations with other polymers or with essential oils or nanoparticles, among others, as shown in Table 2. This allows for the storage period to be increased in order to postpone the deterioration of fruits and vegetables and preventing the growth of microorganisms transmitted by food on the surfaces of the products.

Fungi that affect postharvest fruit quality: analysis from 2011 to 2021.

The letters correspond to the fungi worked by each author: Letter A, B, and C correspond to [13]; letter B corresponds to [37], and letter C corresponds to [1].

It was observed that there is plenty of research involving published studies concerning the antimicrobial and antifungal activity of chitosan as well as a combination with other polymers or the application of different essential oils against foodborne pathogens. However, of the total reports (875 documents published), only 93 documents with the keyword “fruits” were analyzed due to the importance of this kind of food. The findings mentioned below and those in Table 2, Table 3, Table 4 and Table 5 correspond to these documents.

Published results of bacterial contamination by different microorganisms in fruits: from 2011 to 2021.

Published results from bacterial and fungal contamination by different microorganisms in fruits: from 2011 to 2021.

Recently, the impact of preharvest foliar spraying with chitosan and postharvest aloe vera gel coating (AVG) on the quality of table grapes during storage was evaluated, thereby extending the shelf life of the fruit up to 15 days by significantly reducing the decomposition index [38]. Another recent finding is the production of edible coating films based on Pickering emulsions, which showed a smaller droplet size, narrower size distribution, and improved stability. These could inhibit the growth of typical spoilage organisms such as *S. aureus* and *E. coli* in order to preserve fruits and vegetables [39]. [40] applied this method by adding oleic acid and cellulose nanocrystal in “Bartlett” pears (*Pyrus communis* L.) for delaying ripening and superficial scald during the long-term cold storage.

Table 2, Table 3 and Table 4 show studies concerning the application of chitosan as an antimicrobial and antifungal to maintain fruit and vegetable quality at the postharvest stage. It is highlighted that several studies have focused on reducing the antifungal activity of *Aspergillus niger*, *Rhizopus stolonifera*, *Botrytis cinerea*, *P. expansum*, *Alternaria alternate*, *Colletotrichum gloeosporioides*, etc. In some studies, the antifungal activity of chitosan depends on the extraction procedure, the deacetylation percentage, molecular weight, or the microstructure of the fruit and the interaction of the coating material.

Some fruits such as strawberries [1][13][41][42][37][43][44][45][46][47][48], mango (*Mangifera indica* L.) , tomato [49][50][51][52][53][54][55][56][57], guava [58][59][60], banana [61][62], apples [63][64][65][66], or fresh-cut apple slices [67][68] are especially perishable and therefore there is a larger number of documents focused on decreasing mechanical injury, desiccation, decay, and physiological disorders during storage [41], as observed in Table 2, Table 3, Table 4 and Table 5. In this sense, the most cited document (166 citations) [41] mentioned that the use of chitosan functionalized by acylation with palmitoyl chloride

increase its hydrophobicity in order to ensure a controlled release and improve its stability and adherence in strawberries. Notwithstanding, research in this area is still incipient, and therefore is necessary to carry out future research about the topic with other fruits or in different matrixes of food.

For this purpose, mixtures of chitosan and some other materials have also been used, as shown in Table 5; these results indicated that the coatings could reduce the damage in different fruits or vegetables.

Moreover, other studies have addressed that the chitosan coating applied in pummelo fruit mitigates the development of juice sac granulation and delays postharvest senescence in the same fruit during room temperature storage [69], and in eggplant cultivars (purple long, purple round, and white long), chitosan was effective in minimizing weight loss, maintained quality, and prolonged storability with good appearance and overall acceptability [70]. However, it is necessary to conduct more research focused on combinations of adequate techniques and different coating materials that consider the intrinsic and extrinsic factors that affect food, as well as allowing for enhancement of shelf life and decreases in the amount of waste.

Coating materials mixture with chitosan applied to extend the shelf-life and improve the quality of fruits.

4. Conclusions

This bibliometric review analyzed the evolutionary process over the past decade of topics about chitosan as coating and their fruits and vegetables' antifungal or antimicrobial effects. VOSviewer software is a useful and versatile tool that allows for easy visualization and analysis of bibliometric networks. In this paper, 875 documents reported that coatings made of chitosan only or chitosan in combination with other biopolymers are a natural and safe post-harvest biocontrol strategy to decrease microbial spoilage mainly by pre- Finally, this work can provide a useful perspective for future research in the studied field since it demonstrates the existence of an emerging area of study that is intended to reduce a global problem caused by the generation of agro-industrial waste due to the loss of post-harvest damaged crops.

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