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Review

A Bibliometric Analysis of the Scientific Literature on Biostimulants

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Abstract: A search of the term biostimulants on the most renowned scientific online databases such as Web of Science results in more than one thousand documents. Although some reviews have been previously published, there is no unified and comprehensive bibliometric review of the scientific literature related to biostimulants. This study examines the scientific literature on biostimulants from 2000 to February 2022 by conducting a bibliometric analysis of the literature published on the Web of Science database to deepen its evolution, trends, and macroareas to represent a quick reference guide for interdisciplinary researchers. We identify the most productive countries and journals, detect the major research streams and perspectives, and trace overall research development over the years. Furthermore, the results highlight aspects that have had little consideration in the current scientific literature, such as economic assessments of the use of biostimulants and more comprehensive explanations of the molecular mechanisms responsible for their positive effects.

Keywords: bibliometric analysis; biostimulants; seaweeds; protein hydrolysates; humic substances

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1. Introduction

After decades in which agricultural innovation and practices have been mainly focused on improving crop yields, efforts in the food systems are also oriented towards enhancing the quality of products and sustainability of food production [1]. Similarly, consumers are increasingly aware of food production's environmental and social repercussions, and the demand for sustainable products has grown. This growing interest in environment-friendly food coincided with the promotion in 2015 of the Sustainable Development Goals (SDG), which identify the linkages between human well-being and the health of natural systems: from the fight against hunger to the elimination of inequalities, from the protection of natural resources to the establishment of sustainable production and consumption patterns [2]. In this context, identifying sustainable inputs to address modern agricultural challenges is a current and highly relevant topic in the scientific and general debates on food systems' economic and environmental sustainability.

Biostimulants are innovative agronomic tools composed of organic substances and inorganic materials that include several substances and microorganisms [3]; this term was proposed for the first time in 1997 by Zhang and Schmidt [4] to indicate substances whose application in small quantities can favour plant growth and health [5]. Previously referred to as plant strengtheners, positive plant growth regulators, and metabolic enhancers, among others, biostimulants result from diverse starting materials and production processes, and their concept and definition are still evolving [6]. The definition most experts agree on is the one provided by Patrick Du Jardin, which defines biostimulants as "substances and materials, with the exception of nutrients and pesticides, which when applied to the plant, seeds or growing substrates in specific formulations, can modify physiological processes of plants in a way that provides potential benefits to growth development and/or stress response" [7,8]. Biostimulants differ from fertilisers in that they do not directly supply nutrients but promote in plants the improvement of nutrient uptake and assimilation efficiency and tolerance to abiotic stresses. They are defined for what they do more than what they are because they can be produced from multiple products: seaweeds, protein hydrolysates, humic substances, and microorganisms. Recently, biostimulants have been regulated and included among the fertilisers in the EU regulation 1009/2019, classified as either microbic or non-microbic. This recent regulation opened new opportunities for all industries involved in biostimulant production, leaving open the opportunity to exactly decipher their mode of action [9].

The lack of a recognised definition and criteria that identify biostimulants from the numerous types of formulations used in agriculture has meant that data show widely divergent statistics for the size and value of the biostimulants market. Indeed, some estimations include some families of biopesticides and liquid fertilisers among the biostimulants that technically do not fall into the category [10]. However, the global biostimulant market is estimated at approximately USD 3 billion in 2021 and is expected to reach more than USD 5.1 billion by 2027 [11]. Europe has the largest share at around 45%, North America and Asia have approximately 20% each, and Latin America has around 15% [12]. The value of the European biostimulant market ranged from 200 to 400 million in 2011 and grew to 500 million in 2013 [10,13], up to USD 1500.79 million by 2025. France, Italy, and Spain are the main European countries producing biostimulants [10]. The biostimulant market in North America was valued at USD 0.27 billion in 2011 [14]. Existing data on Latin America and the Asia-Pacific date back to 2013: in Latin America, the market was valued at USD 0.16 billion and for the Asia-Pacific at USD 0.25 billion [14]. The Australian biostimulants market was estimated at USD 233.8 million in 2015 and is expected to reach USD 451.8 million by 2022 [15].

In principle, the use of biostimulants could represent an economically viable option for farmers to meet the growing quality standards and consumers' expectations in terms of sustainability and environment protection, as it has been associated with reductions in production costs and increases in product quality [10,13]. Biostimulants can perform numerous agronomic functions, such as boosting the growth and development of plants during their entire life cycle [16]; increasing soil fertility, in particular by promoting the development of soil microorganisms [17,18]; increasing the resistance of plants to abiotic stresses, such as heat, cold or lack of water, [19] and biotic stresses, such as parasites including viruses, bacteria, and insects [20,21]; improving the use efficiency of nutrients by plants [5]; and improving crop quality [22] and yield [23,24]. With the agricultural industry experiencing an extraordinary increase in fertiliser prices [25], biostimulants could help reduce the use of agricultural inputs and, therefore, reduce production costs. In this respect, the use of biostimulants could be particularly strategic for organic farmers, who, not being able to use most fertilizers and pesticides, could find a tool to reduce the gap between organic and conventional yields in full compliance with the regulations. However, for the best results, biostimulants require tailor-made administration strategies at specific times and in optimal doses depending on the crop and variety [26].

It is crucial for nonmicrobial biostimulants to be produced from biomass available in large quantities and readily accessible for processing [27]. Materials to produce biostimulants should also have low collection and storage costs to maintain a low price and be competitive as substitutes for synthetic products. However, most natural products are unstable and undergo chemical modifications when exposed to heat, light, or oxygen, resulting in loss of bioactivity [28]. Various active ingredients found in industrial waste streams and byproducts of biological origins pose the perfect opportunity to implement a circular economy by extracting molecules that improve plant growth and resistance to pathogens [27]. However, some understanding of the intrinsic biochemical characteristics of raw materials is needed to preserve specific bioactive ingredients [29].

Market research companies and agricultural organisations are investing significantly in research to meet the needs of this rapidly expanding market; this aspect is also clearly highlighted by publications: although the interest in biostimulants had begun already in 1975, only in recent years has it increased exponentially. Today, a search of the term biostimulants on the most important scientific online databases, such as Web of Science, leads to more than 1500 results. However, although some reviews have been previously published, there is no unified and comprehensive bibliometric review of the scientific literature related to biostimulants. Therefore, to analyse the scientific literature regarding biostimulants, we carried out a quantitative study using bibliometric methodologies and tools; this allowed us to detect the most productive countries and journals, identify the major research streams and topics, and trace the overall development of research over the years. This study uses bibliometric methodologies to explore the current and previous research on biostimulants to deepen its evolution, trends, and macro areas to represent a quick reference guide for interdisciplinary researchers. Furthermore, this analysis highlights some aspects that have received little or no consideration in scientific literature.

2. Materials and Methods

This study examines the scientific literature on biostimulants from 2000 to February 2022 by conducting a bibliometric analysis of the literature published on the Web of Science database, including almost one thousand articles written by more than 2500 authors. The approach adopted in this paper is rooted in bibliometrics, a field of information and library science used to measure and analyse published scientific research that applies mathematical and statistical techniques to analyse the distribution models of publications and explore the impact within scientific communities [30–32]. Bibliometrics develops on two main types of analysis: performance analysis and scientific mapping [33–35]. The first type evaluates players (e.g., authors, institutions, journals, and countries) based on bibliographic data, while scientific mapping graphically represents the different bibliometric networks to explore the interrelation between disciplines, fields, institutions, authors and papers [36]. In bibliometrics, a citation is assumed to generally reflect the resonance of a paper in the academic community in an objective and measurable way. Several bibliometric techniques have been developed to build a scientific map; the most used is co-citation network analysis [37].

Co-citation analysis is a bibliometric technique proposed by Small [32,38,39] that allows mapping the structure of a research field through the analysis of the most cited documents in a specific body of literature [37]. Two documents are said to be co-cited when they appear in a third document's reference list; the more articles that cite them simultaneously, the stronger their association. This technique provides a comprehensive view of the intellectual structure of a specific research field, as the co-cited documents tend to share some common themes and are considered to represent the core knowledge base of a research area: the key concepts, methods or experiments that researchers build on [40]. Furthermore, cluster analysis can further inspect the heterogeneity within the co-cited documents. Co-citation analysis allows the exploration of the dynamics of scientific development and conceptual shifts of a specific subject [32]. The main drawback of co-citation analysis is that it places a greater emphasis on "the past" of an academic field, as it is more likely to capture older contributions and well-established scholars rather than the forefront of the research [41,42].

The bibliographic data gathered in this study were collected from Web of Science, a scientific citation indexing service produced by the Institute for Scientific Information (ISI), now managed by Clarivate Analytics. The term "biostimulant" was used as a keyword in the field "Topic"; doing so reported 1498 documents by searching for the term in titles, abstracts, and keywords. We have selected only English language articles from 2000 to February 2022, reducing the database to 1168 documents. Consistency with the topic discussed was verified by reading the titles, the abstracts and, in some cases, the articles.

This process removed almost one hundred documents from the initial database, leading to a final data set of 1088 documents. The skimming of documents, the extraction of the cited literature, and the creation of the final database are computed through the software Bibexcel; the software VOSviewer [43] and Citespace [44] are used for mapping and clustering.

3. Results

3.1. The Characteristics Analysis of Biostimulant Research

Although the first research dates back to the 1980s, research on biostimulants has developed significantly only in recent years, as it has become an affirmed research topic since 2007 and even more so since 2015. Table 1 and Figure 1 show the increase in the number of publications and researchers interested in the topic. This growing interest in the topic of biostimulants by research has occurred in conjunction with an increase in the number of industries involved in their production; the different prototypes need to be developed and then tested in different environments and crops before commercialisation, and collaborations with academic institutions are strategic for demonstrating their stimulatory properties and their patent registration [45,46]. The increase in publications also coincides with the program for sustainable development, Agenda 2030, denoting an increased interest in understanding the modes of action of biostimulants and how their use can increase agricultural sustainability by promoting the growth and development of plants, their nutrient use efficiency, and stress tolerance [47].

Table 1. Quantitative analysis of the trend of research on biostimulants.

Year	No. of Articles	No. of Authors	No. Authors/No. Docs	Times Cited	Average Times Cited Count	Average Reference Count
2000	1	3	3.0	7	7.0	10.0
2002	1	3	3.0	44	44.0	11.0
2003	2	6	3.0	72	36.0	18.0
2004	1	4	4.0	32	32.0	36.0
2005	2	3	1.5	33	16.5	34.0
2006	1	6	6.0	24	24.0	36.0
2007	7	25	3.6	305	43.6	29.1
2008	6	26	4.3	57	9.5	19.2
2009	4	15	3.8	217	54.3	42.0
2010	7	32	4.6	178	25.4	40.6
2011	6	25	4.2	246	41.0	36.5
2012	8	42	5.3	116	14.5	31.9
2013	12	66	5.5	431	35.9	43.4
2014	18	93	5.2	848	47.1	62.8
2015	29	149	5.1	1695	58.4	54.5
2016	51	237	4.6	1140	22.4	44.1
2017	69	353	5.1	1496	21.7	54.0
2018	132	647	4.9	2211	16.8	50.8
2019	164	899	5.5	1691	10.3	56.3
2020	247	1391	5.6	1267	5.1	57.4
2021	298	1735	5.8	417	1.4	65.0
2022	22	122	5.5	4	0.2	57.3

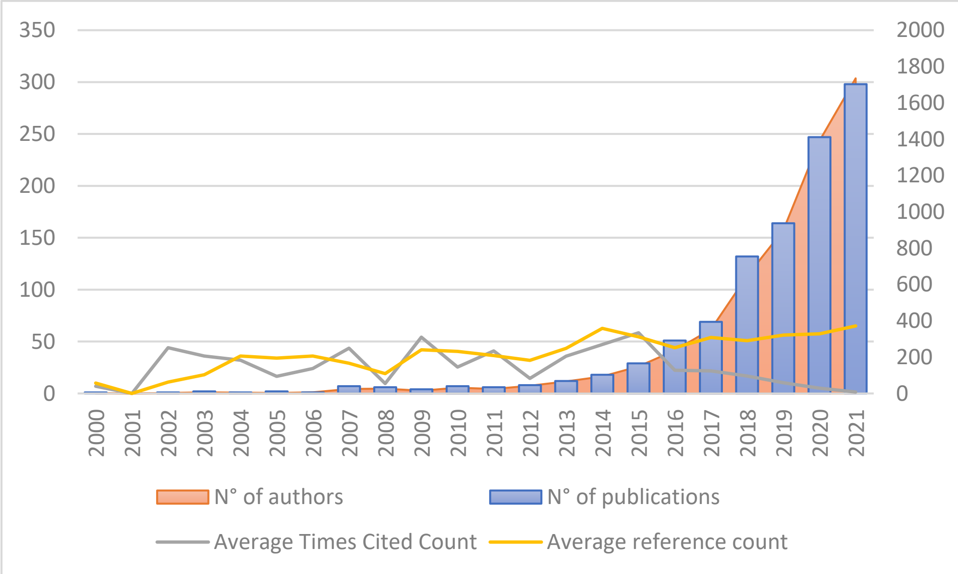

Figure 1. Number of authors and publications, average citations and the reference count of biostimulant publications between 2000 and 2021. Data for the first two months of 2022 are not represented in the figure.

Table 2 shows the top 10 journals that published studies related to biostimulants from 2000 to 2022. A large portion of the publications can be found in crossdisciplinary journals with experimental and theoretical research on food and cropping systems, horticulture and plant science, sustainable development in agriculture, biodiversity, and ecosystem services of the food system. Of the journals included in the WoS core search database, the journals "Agronomy", "Frontiers in Plant Science", and "Scientia Horticulturae" published the most biostimulant papers in the study period. Horticultural crops seem to be of particular interest for the research on biostimulants, probably due to their short growth cycle and high input needs, for which the applications of biostimulants could provide beneficial effects from the economic and environmental points of view. The research papers report the effectiveness of biostimulants in increasing yield, quality, and tolerance to abiotic stresses. Moreover, journals in Table 2 aim to disseminate forefront multidisciplinary research in plant biology and physiology, while others deal with food quality and phycology. Overall, these results show that biostimulants in agriculture have relevance in a wide range of disciplines related to life science.

Table 2. Biostimulant publications' top 10 most productive journals.

Journal	No. of Biostimulants Publications	No. of Total Publications	No. of Citations
Agronomy Basel	95	5948	671
Frontiers in Plant Science	78	16,359	1783
Scientia Horticulturae	45	9981	1782
Plants Basel	43	5409	222
Journal of Applied Phycology	39	4613	1016
Agriculture Basel	18	2620	66
Molecules	15	33,917	199
Sustainability	15	42,222	245
Hortscience	13	26,595	191
Journal of the Science of Food and Agriculture	13	12,380	196

As shown in Table 3, the subject categories reflect the multidisciplinarity and complementarity of the different disciplines that have approached biostimulants observed in the journals in Table 2. Research on biostimulants involves several disciplines: from agriculture to environmental and ecological sciences to chemistry and food science. The most popular subject categories are agriculture and plant sciences, which are then addressed by specific branches such as chemistry, biotechnology, and microbiology. Interestingly, the categories related to business and economics are not among the most frequent subject categories and are generally missing in the entire dataset, as only one document of the 968 falls into the business and economics category. This lack of assessments in the economic field is crucial considering that the profitability of biostimulants could depend on optimising the costs requested for the preparation/isolation/stabilisation/conservation of the products and the economic valorisation of the raw materials. In this context, the increasing competition in the biostimulants market reinforces the importance of economic analysis.

Table 3. Scientific macroareas in which research on biostimulants is concentrated.

Subject Categories	Frequency	%	Cumulative %
Agriculture	504	46%	31.48
Plant Sciences	348	32%	53.22
Environmental Sciences & Ecology	128	12%	61.22
Chemistry	94	9%	67.09
Biotechnology & Applied Microbiology	89	8%	72.65
Science & Technology-Other Topics	54	5%	76.02
Food Science & Technology	52	5%	79.27
Microbiology	50	5%	82.39
Engineering	49	5%	85.45
Marine & Freshwater Biology	49	5%	88.51

3.2. Contribution of Countries

From 2000 to 2022, universities and research institutes from 40 countries have contributed to the research on biostimulants: Italy has a dominant role with 18% of the total publications, followed by Poland (16%), Brazil (13%), Spain (10%), the USA (10%), and India (6%). The 15 most productive countries are shown in Table 4. For this statistic, only the corresponding author's affiliation for each document has been considered.

Table 4. Top 15 most productive countries in biostimulant research.

Country	Frequency	%
Italy	197	18%
Poland	144	13%
Brazil	84	8%
Spain	76	7%
USA	64	6%
India	44	4%
Egypt	41	4%
People's Republic of China	40	4%
Mexico	27	2%
South Africa	25	2%
Iran	21	2%
France	19	2%
Canada	17	2%
Portugal	16	1%
Morocco	14	1%

Figure 2 shows in which period researchers from different countries have addressed this topic. Italy occupies a leading position in the literature on biostimulants; Italian institutions occupy four positions among the top five institutions publishing on the topic, Poland being the second-most productive country. The high research contribution from Italy might be due to the high number of industries involved in biostimulants located in Italy. In fact, the leading companies in biostimulant production are Italian, and several research activities are performed in collaboration with Italian research institutions. European countries are the major producers and users of biostimulants; therefore, studies and research are essential to understanding the effectiveness, methods, doses, and application periods of biostimulants. Furthermore, research has been crucial to demonstrating the real benefits of using these materials for plants, soil, and the environment and to improving farmers' profitability [13].

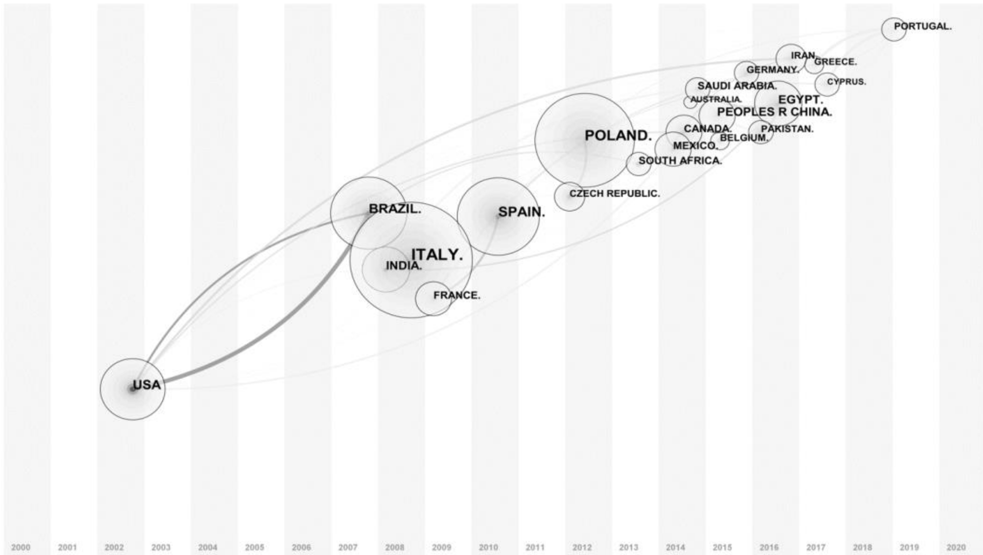

Figure 2. Contribution of countries/territories on biostimulant research during 2000–2020.

3.3. High Frequency Keywords over Time

The distribution and evolution over time of research hotspots can intuitively demonstrate research themes, perspectives, and method changes throughout the years. Analysing the evolution of the main keywords used in biostimulant research can help us understand the literature's focus and core issues and the hotspots and topics of interest during a specific period. The time zone view represented in Figure 3 represents different research hotspots. Figure 3 allows us to deduce the objectives on which the research focused over the years. In the early years, the focus was on plant growth, increasing yields, and specific categories, such as humic acids, and seaweed extracts; from 2013, the central themes have shifted towards topics related to quality and resistance to different types of stress with the appearance of other biostimulant categories such as amino acids and protein hydrolysates. This trend seems to reflect the changes occurring in the main aims of the agricultural sector, which joined high yields in crop production with growing attention toward sustainability and food quality. It also seems to be accompanied by an increasing focus on stress and climate change. Interestingly, some of the most active countries producing biostimulants, such as Italy, are among those most exposed to the negative effects of climate change in the future [48]. Over the years, authors have used several keywords connected to the term biostimulant, but the inclusion of this term in the list of keywords suggests that it was fully recognised by the scientific community around 2017. There is an evolution of the materials used; this progress is attributable to the fact that more and more efforts are being focused on reusing materials to save new natural resources, raw materials, and energy and protect the environment. For these reasons, the production of biostimulants can be part of bioeconomy strategies and consistent with the Green Deal of future European strategies.

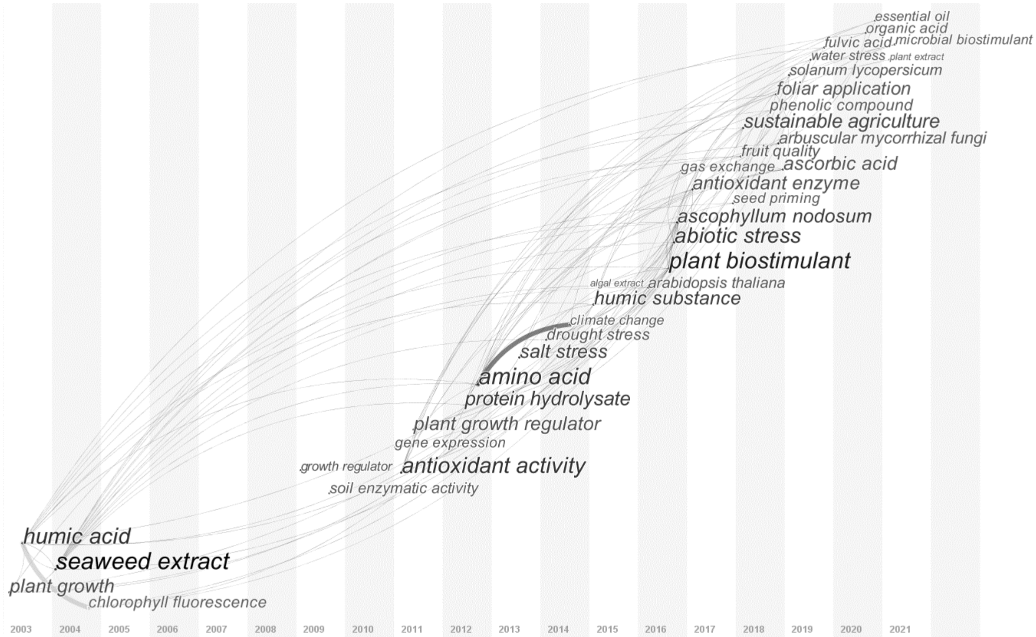

Figure 3. Time zone view of WoS biostimulant hotspots keywords from 2000 to 2021; the size of the keywords is proportional to their frequency.

The analysis of the graph reflects the evolution over time of the different types of matrices used for the production of biostimulants, starting from bacteria and seaweeds that have been traditionally used, either directly or following composting, as soil fertilisers to enhance crop productivity [49,50]. Given the large availability of algae in the coastal regions of the oceans—about 15 million metric tons of seaweed—based products are produced annually;the benefits they can bring to plants and the possibility of exploiting them as biostimulants have resulted in significant interest from researchers for many years [51]. Since 2014, arbuscular mycorrhizal fungi (AMF) and beneficial bacteria (PGPR) have gradually received more scientific attention; however, the research was limited to a limited number of strains and species. To date, different mechanisms of action of PGRP have been proposed [52].

Due to the early studies on humic acids, the stimulating properties of humic substances were recognised very early on. Since the early 1960s, it has been known that they are natural substances resulting from the transformation of organic matter in the soil through the metabolic activity of the microbes that, using these substrates, exert a direct positive influence on plant growth and nutrition [53]. However, due to their action mechanisms, they have been recently reconsidered as biostimulants [7].

In recent years, research has focused on protein hydrolysates that have been used in the production of biostimulants; protein hydrolysates are complex organic materials obtained through chemical and/or enzymatic hydrolysis of agroindustrial byproducts from animal or plant origins or dedicated biomass crops [54,55]. The use of protein hydrolysates can increase the supply of agricultural products while reducing the impact of agriculture on the environment and human health [19,24]. The aspects that make protein hydrolysates very suitable products are that they can be derived from sewage sludge, wastewater treatment plants, waste from the agro-food industry, manure, and compost extracts from urban waste fermentation tank residue [7]. Despite being materials that were considered waste until a few years ago, today, they not only have a "second life", but are also able to bring several benefits to agricultural production.

3.4. Document Co-Citation Analysis Results

The most cited documents by the researchers of biostimulants are shown in Table 5. Although the number of citations does not necessarily indicate the quality of an article, it is a reliable indicator of its impact or visibility, and to an even greater extent, of the extent to which the specific document represented a point of reference for subsequent research. The top five publications with multiple citations in biostimulant research are five reviews, namely Du Jardin [8], Calvo [3], Khan [49], Yakhin [5], and Battacharyya [56]. These documents explain in depth what biostimulants are, their main types and categories, the materials that can be used for their production, and other information, providing a comprehensive overview and a fundamental starting point for future learning and research. Du Jardin [8] and then Yakhin et al. [5] review the different concepts of biostimulants and their legal and regulatory status in the EU and the US (before 2019); moreover, Du Jardin's definition of biostimulants is the most commonly accepted used today. Calvo's research [3] points out common plant responses to different biostimulants, such as increased root growth, enhanced nutrient uptake, and stress tolerance. Furthermore, the authors hypothesised that the combination of more than one category of biostimulants could improve their beneficial effects. Khan's paper focuses mainly on the use of macroalgae in agriculture, reporting the effects on the growth and development of plants [49]. Battacharyya et al. [56] provide a review of the literature on the chemical constituents of brown seaweed extracts and their physiological effects on plants, with particular reference to horticultural crops

Table 5. Top 15 most cited publications in the biostimulants research.

Cited Reference	Citations from Biostimulant Studies	Total Citations
Du Jardin, P. Plant biostimulants: Definition, concept, main categories and regulation. Sci. Hortic. 2015, 196, 3–14. https://doi.org/10.1016/J.SCIENTA.2015.09.021 [8]	334	713
Calvo, P.; Nelson, L.; Kloepper, J.W. Agricultural uses of plant biostimulants. Plant Soil 2014, 383, 3–41. https://doi.org/10.1007/S11104-014-2131-8/TABLES/1 [3]	261	763
Khan, W.; et al. Seaweed Extracts as Biostimulants of Plant Growth and Development Tropical Phyconomy Coalition Development #1—The eucheumatoids View project PGPR and plant growth View project Seaweed Extracts as Biostimulants of Plant Growth and Development. Artic. J. Plant Growth Regul. 2009, 28, 386–399. https://doi.org/10.1007/s00344-009-9103-x [49]	165	646
Yakhin, O.I.; Lubyanov, A.A.; Yakhin, I.A.; Brown, P.H. Biostimulants in plant science: A global perspective. Front Plant Sci. 2017, 7, 2049. https://doi.org/10.3389/FPLS.2016.02049/BIBTEX [5]	151	362
Battacharyya, D.; Babgohari, M.Z.; Rathor, P.; Prithiviraj, B. Seaweed extracts as biostimulants in horticulture. Sci. Hortic. 2015, 196, 39–48. https://doi.org/10.1016/J.SCIENTA.2015.09.012 [56]	132	293
Colla, G.; Nardi, S.; Cardarelli, M.; Ertani, A.; Lucini, L.; Canaguier, R.; Rouphael, Y. Protein hydrolysates as biostimulants in horticulture. Sci. Hortic. (Amsterdam). 2015, 196, 28–38, https://doi.org/10.1016/J.SCIENTA.2015.08.037 [57]	121	212
Craigie, J.S. Seaweed extract stimuli in plant science and agriculture. J. Appl. Phycol. 2011, 23, 371–393, https://doi.org/10.1007/S10811-010-9560-4 [50]	119	436
Sharma, H.S.S.; Fleming, C.; Selby, C.; Rao, J.R.; Martin, T. Plant biostimulants: a review on the processing of macroalgae and use of extracts for crop management to reduce abiotic and biotic stresses. J. Appl. Phycol. 2013 261 2013, 26, 465–490, https://doi.org/10.1007/S10811-013-0101-9. [58]	119	238
Van Oosten, M.J.; Pepe, O.; De Pascale, S.; Silletti, S.; Maggio, A. The role of biostimulants and bioeffectors as alleviators of abiotic stress in crop plants. Chem. Biol. Technol. Agric. 2017, 4, 1–12, https://doi.org/10.1186/S40538-017-0089-5 [59]	100	240

Table 5. *Cont.*

Cited Reference	Citations from Biostimulant Studies	Total Citations
Bulgari, R.; Cocetta, G.; Trivellini, A.; Vernieri, P.; Ferrante, A. Biostimulants and crop responses: A review. Biol. Agric. Horticult. 2015, 31, 1–17. https://doi.org/10.1080/01448765.2014.964649 [6]	96	199
Colla, G.; Rouphael, Y.; Canaguier, R.; Svecova, E.; Cardarelli, M. Biostimulant action of a plant-derived protein hydrolysate produced through enzymatic hydrolysis. Front Plant Sci. 2014, 5, 448. https://doi.org/10.3389/FPLS.2014.00448/BIBTEX [54]	93	171
Colla, G.; Hoagland, L.; Ruzzi, M.; Cardarelli, M.; Bonini, P.; Canaguier, R.; Rouphael, Y. Biostimulant action of protein hydrolysates: Unraveling their effects on plant physiology and microbiome. Front. Plant Sci. 2017, 8, 2202, https://doi.org/10.3389/FPLS.2017.02202/BIBTEX [60]	83	147
Bradford, M.M. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem. 1976, 72, 248–254, https://doi.org/10.1016/0003-2697(76)90527-3 [61]	82	237,327
Canellas, L.P.; Olivares, F.L.; Aguiar, N.O.; Jones, D.L.; Nebbioso, A.; Mazzei, P.; Piccolo, A. Humic and fulvic acids as biostimulants in horticulture. Sci. Hortic. (Amsterdam). 2015, 196, 15–27 [16]	82	322
Ertani, A.; Cavani, L.; Pizzeghello, D.; Brandellero, E.; Altissimo, A.; Ciavatta, C.; Nardi, S. Biostimulant activity of two protein hydrolyzates in the growth and nitrogen metabolism of maize seedlings. J. Plant Nutr. Soil Sci. 2009, 172, 237–244, https://doi.org/10.1002/JPLN.200800174 [55]	77	143

Figure 4 shows the top 15 references with the strongest citation burst. Citation burst is an indicator of the most active area of research and detects a burst event, which can last for multiple years or only for a single year [44]. This parameter indicates that a publication has attracted an extraordinary degree of attention from the scientific community over a certain time. The authors with the strongest citation burst are Parrado [62], Garcia-Martinez [63], and Ertani [55]. Parrado [62] has a high burst on the topic of biostimulants as it describes a biological process that converts carob germ, a proteinic vegetable byproduct, into a water-soluble enzymatic hydrolysate extract. This extract has significant beneficial impacts, most notably regarding plant height and the number of flowers and fruits per plant. In Garcia-Martinez's [63] paper, the effects of protein hydrolysates obtained from wheat-condensed distiller solubles on soil biochemical parameters and microbial community are described. The treated soils showed higher enzyme activity without modifying the soil microbiome. It is interesting to note that the literature dealing with the effects of biostimulants on soil proprieties is still very scarce. Ertani [55] showed that the protein hydrolysates of alfalfa or meat flour could be of practical interest because they can stimulate plant growth and the assimilatory nitrate pathway in maize. Consequently, these products could potentially help reduce the amounts of nitrogen fertiliser inputs and the environmental impact of agricultural production.

Figure 5 shows the network of co-citation of the database. The size of the labels is larger when the number of citations within the series of publications is greater; the references that are most likely to be mentioned together are closer to one another. Co-citation analysis highlighted four main clusters that correspond well to the most investigated topics in the research field on biostimulants.

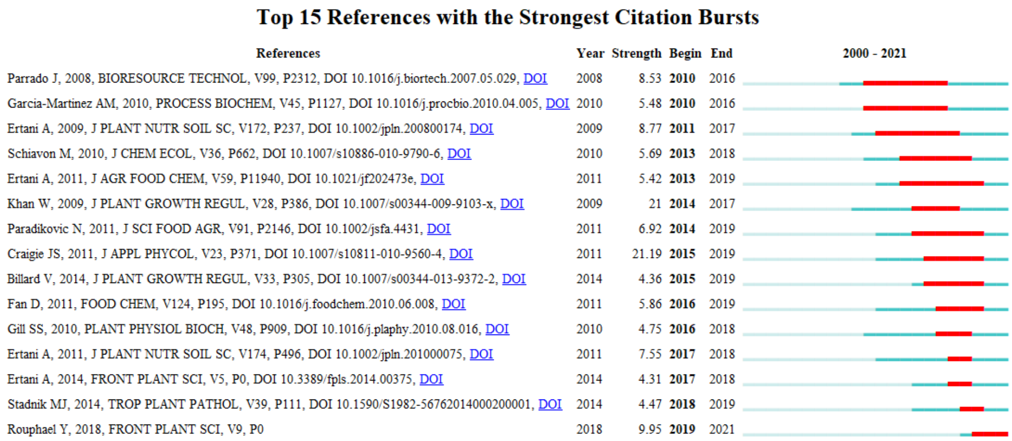

Figure 4. Articles with the strongest citation burst.

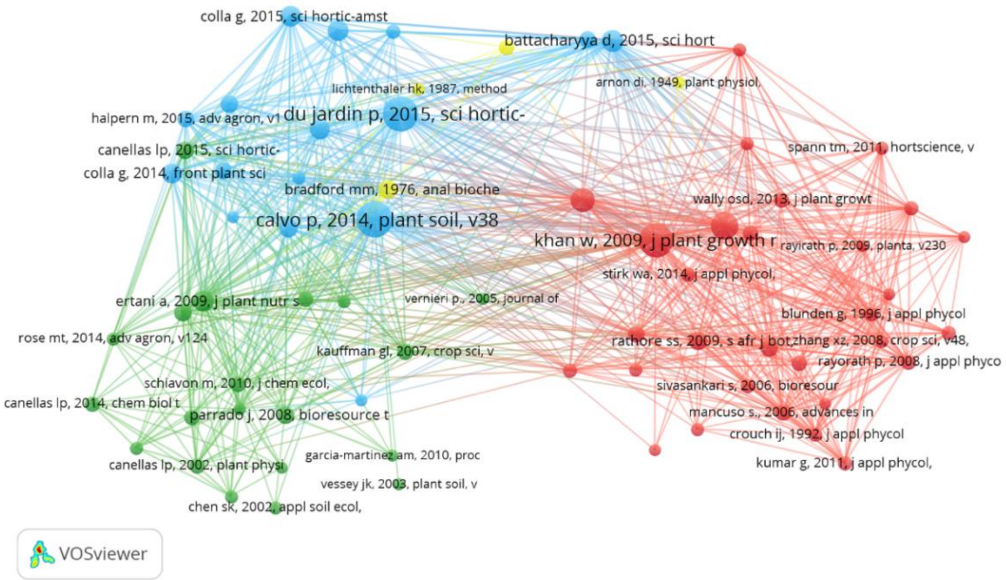

Figure 5. Co-citation analysis network.

Cluster 1 (red)—use of seaweeds as biostimulant to increase crop productivity

Documents of the first cluster mainly focus on the use of seaweeds as biostimulants, as they are biodegradable and nonpolluting for the environment [64] and therefore have become popular as tools to increase production yields and improve plant growth.

The articles of the first cluster provide an overview of the potential positive and beneficial effects of marine algae [49], both microalgae and macroalgae [50], which could allow reducing the use of phytochemicals [65]. Khan provides a comprehensive review of the effects of various algae species and algae products on plant growth and development, emphasising the use of this renewable biological source in sustainable agricultural systems [49].

In this cluster, there are most of the documents related to the effects of seaweed extracts on different types of crops such as tomatoes [66,67], carrots [68], spinach seedlings [69], and vines [70].

Cluster 2 (green)—applications and effects of humic acids, fulvic acids, and hydrolysed proteins on different crops

The green cluster highlights the biostimulant activity of products based on humic and fulvic acids and hydrolysed proteins. Canellas et al. [71] conducted experimental research analysing how humic acids obtained from vermicompost stimulate lateral root development and plasma membrane H+-ATPase activity in maise. In 2015, Canellas et al. summarised the literature describing the effects of humic and fulvic acids [16]. Among other things, it emerges how they affect plant primary and secondary metabolism and enhance root growth, nutrient uptake, and crop tolerance to environmental stresses. Several authors in this cluster report examples of how the action of humic acids can increase plant growth [72] and act on their metabolism [73–75]. Documents that analyse the biostimulant activity of products based on hydrolysed proteins are also included in this cluster [20,55]. Ertani's research, the most cited in this cluster, presents robust data on the positive effects of protein hydrolysed in increasing the growth of roots and leaves and inducing morphological changes in the architecture of the roots [55].

Cluster 3 (blue)—reviews on biostimulants: what they are, categories, effects, and benefits

This cluster reports reviews and articles on biostimulants [5] and their application in agriculture [3,76]. In addition, research addressing the definition and classification of biostimulants is also included, among which is the review by Du Jardin [8].

It is worth noting that the most co-cited documents belong to the studies that define and explain the type of products, the actions they can perform and—above all—the benefits they can bring to crops [5]. For example, Calvo [3] reviews the emerging definitions of biostimulants and the relevant literature on five categories of biostimulants: microbial inoculants, humic acids, fulvic acids, protein hydrolysates, and seaweed extracts.

Other studies in the cluster inspect different effects of biostimulants on crops, evaluating their agronomic efficiency and how their application influences in plants some key biochemical activities [6,54,56,77,78]. Another theme among the documents of the third group is the chemical characterisation of two biostimulants: hydrolysed proteins derived from alfalfa plants and an extract from red grapes [79].

Cluster 4 (yellow)—Methodologies for the quantification of plant metabolites affected by biostimulant treatment

The last cluster groups quite old documents mainly focusing on the methodologies to quantify some fundamental plant molecules, such as chlorophyll [80], carotenoids [81], proline [82], and proteins [61]. These documents are often cited in biostimulant research because the levels of these compounds can be associated with their efficiency. Protein content in plant tissues is indeed strictly related to nitrogen metabolism, and chlorophylls and carotenoids are fundamental in photosynthesis. For example, carotenoids are involved in protecting the photosynthetic apparatus against abiotic stresses and increasing the nutraceutical value of fruits and vegetables [83], and their concentration in plants increases following treatments with biostimulants.

4. Discussion

This research reveals that the scientific literature on biostimulants has exponentially grown over the last years, with a particularly evident increase in the last five, possibly due to the increasing importance of environmental protection and sustainability of food systems in scientific and public debates.

Several authors from different disciplines have published scientific research on biostimulants, resulting in a vast scientific production that is heterogeneous in terms of approaches, methodologies, and objectives. Within this heterogeneity, the co-citation analysis identified four main lines of research that have been covered up to now. As biostimulants derive from different materials, the formation of the clusters partly resembles the source from which they are produced. The largest cluster is composed of publications focusing on biostimulants deriving from seaweeds. There is a long tradition of using algae for agricultural purposes due to the numerous advantages it can bring to crops, particularly as soil amendments [49]. On the other hand, at the beginning of the 1950s, a process to produce liquid extracts of seaweed was already being described, opening the way for the production of several algae-extract-based products that are still used in agriculture and horticulture as biostimulants [49,50]. This could justify the abundance of studies linked to algae as biostimulants in literature. Recently, there has been an interest in cultivating microalgae and their use in the biostimulant production industries [84]. Seaweed extracts are mainly used to improve plants' photosynthesis and promote growth and nutrient use efficiency. The main effect is on crops' primary metabolism, and there is much evidence of the ability of seaweed extracts to counteract abiotic stresses.

The second cluster groups publications dealing with fulvic acids, humic acids, and hydrolysed proteins. The effects of humic substances extracted from soils on plants' growth and physiological processes have been known for a long time—decades before they were defined as biostimulants [85]. The recent literature confirms these properties and highlights the potentialities of these biostimulants to alleviate symptoms due to abiotic stress conditions with particular reference to plant nutrition [16,53,86,87]. On the other hand, although one possible target of their stimulatory action could be the plasma membrane H+-ATPase [88], the biochemical and molecular bases of the modes of action of these molecules are still under debate [53,87,89]. As far as hydrolysed proteins are concerned, it is interesting to observe that fifty years ago, a foliar "organic fertiliser" based on free amino acids and peptides was launched on the Italian agrochemical market [87]. Several pieces of evidence suggest that treatment with this product stimulates the activity of some enzymes and causes exhibition of a hormone-like activity, allowing scholarship to define the product as an "organic biostimulant" [90]. In recent years, the effects of hydrolysed proteins have been studied at molecular levels [91–94]. These "omics" studies give a first picture of the changes in transcriptome, proteome, and metabolome in response to hydrolysed protein, confirming their action in promoting root growth and nutrient uptake and increased resistance to abiotic stresses. Papers in the third cluster focus on classifications, application modalities, and characterisation of biostimulants. Some papers propose few modes of action but simultaneously declaim the need to broaden scientific research to understand these biological mechanisms better. Studies have shown that the efficacy and applicability of biostimulants are deeply influenced by the raw materials used and industrial preparation. Given the compositional complexity of biostimulants, the chemical analyses hardly detect all product components. Hence, to date, the characterisation of biostimulants must be obtained from feedback analysis reporting the effects on plant physiology and biochemistry [76]. As mentioned, "Omics" approaches appear promising to obtain a more comprehensive characterisation of biostimulant effects on plants [95]. However, it is essential to recall that there are no examples of the exact deciphering of the mode of action in identifying the mechanism of interaction between a biostimulant component and plant cell (e.g., receptors and signalling transduction). It is important to link the transcriptional changes with proteins and metabolic changes that allow crops to adapt to stressful conditions or improve their nutrient use efficiency.

The last cluster groups the different analytical methods used to quantify plant responses to biostimulant treatments. The methodologies highlighted in this cluster suggest that plants' protein, proline, sugars, and chlorophyll contents are primarily used as key biochemical indicators to evaluate biostimulants' beneficial effects on crops.

Despite the economic relevance of the global biostimulants market, our investigation found that few studies analysed the economic profitability of biostimulants. This is also despite the fact that it has been proved that these products can help farmers obtain more abundant, vigorous and sustainable food production while increasing the efficiency of fertilisers and reducing the number of treatments.

Furthermore, in light of the increase in fertiliser prices in recent years, which constantly threatens to undermine the entire agricultural sector, the use of products to improve the uptake and efficiency of nutrients and plants' tolerance to stress can contribute to the reduction of the use of fertilisers and therefore be strategic both for achieving greater environmental and economic sustainability. Moreover, consumers have access to healthy, abundant, affordable, and lower-environmental-impact food products.

Biostimulants have become central themes in some European countries, such as Italy, Poland, and Spain, which have been the major contributors to the evolution of research. Since the 1990s, environmental issues related to food production have played a central role in the debate on food systems in the EU and were already included in the Maastricht Treaty signed in 1992. In addition, the European Common Agricultural Policy has included environmental protection as one of its main objectives. Europe, therefore, aims at growth taking into account the "Europe 2020" strategy and the objectives of the circular economy; attention is paid to the careful management of natural resources such as water, soil and air; attempts are made to limit the use of antibiotics and to encourage the sustainable use of pesticides, to recycle and use renewable resources and to create jobs. In this scenario, biostimulants could significantly contribute to such objectives.

This study also identified the most productive actors and the most cited publications within biostimulant literature. The most cited articles focus on the characterisation of biostimulants, their impact on agriculture, their mechanisms of action, classifications, and the potential benefits for cultivation systems. These papers provide notions on biostimulants, highlighting their benefits and effects. This concept is also evident in the analysis of the keywords; those most used are related to the materials that can be used to produce biostimulants, the effects these products have on plants, and the goals to be achieved through their use. Initially, the production of biostimulants for agriculture made use of the most available materials: algae, fungi, and humic substances. However, over the years, the need has arisen to shift towards implementing production methods to be inserted in a circular economy scenario, with practices to be developed to ensure the growing demand for sustainable food, feed, and energy sources. In this regard, the most revolutionary aspect lies in using byproducts of different origins, e.g., recycled materials, which can bring the same benefits while also reducing the environmental impact of farming practices. In this regard, the study reveals that interest in the use of hydrolysed proteins has increased since 2014.

The recent Green Deal Europe guidelines will probably increase the research and innovation in the biostimulants sector. Biostimulants can be obtained from wastes from agroindustries such as fresh-cut industries, the olive oil extraction industry, the fish industry, and the meat industry, boosting the circular economy and the bioeconomy. The latest EU regulation precisely defined what can be considered a biostimulant, but from a research perspective, biostimulant proprieties should be studied in different materials not currently included in the biostimulant categories. The reduction of agrochemicals can be achieved by coupling biostimulant use and appropriate agronomic management strategies.

5. Conclusions and Limitations

The use of biostimulants in agriculture and horticulture can improve production in different ways through a sustainable and circular economy approach: without providing significant quantities of nutrients, they can improve plant growth and nutrient use efficiency, thus enabling a decrease in the use of synthetic fertilisers and improving plant resiliency to stress, starting from waste materials.

The analysis of biostimulant literature through bibliometric methods has highlighted how the scientific interest in this field has dramatically grown in recent years—in an era in which the agricultural sector is facing the challenges of the development of sustainable and environmentally friendly systems and meeting the nutritional needs of the growing world population. The use of biostimulants can contribute to achieving more sustainable food production systems, as it can reduce energy consumption and enhance the use of resources while improving crop quality, especially in unfavourable growing conditions. This would result in higher profits for farmers, who would have the opportunity to obtain higher crop yields with acceptable quality and nutritional values for increasingly aware consumers.

The interest for biostimulants has only increased in recent years, and for this reason, more comprehensive explanations of the molecular mechanisms responsible for the positive effects of these preparations on the crops analysed are needed. In this regard, further studies must be carried out at an observation level using -omic approaches, such as transcriptomics, proteomics, and metabolomics, and identifying the molecular actors involved in plant–biostimulant interaction. These data will allow a deeper knowledge of the effects and functions of the components of biostimulant products and can be useful in selecting new starting materials and classifying and evaluating the effectiveness of new commercial formulations [6]. In addition, the promotion of research will also help define regulations regarding the use of biostimulants becoming a tool used by everyone for cultivation.

Bibliographical methods are not without limitations. First, as mentioned in the introduction, until fairly recently, biostimulants were referred to as plant strengtheners, positive plant growth regulators, metabolic enhancers, and other names. Nevertheless, the present research used the term biostimulants as the search keyword, which may have reduced the presence of older relevant publications. Second, we limited the search to English-written articles, and we are aware that broadening the search to publications in other languages could yield more complete results, although the number of non-English documents is relatively small. Third, other possible biases could come from the tendency of authors to cite their own previous articles, which could lead to a greater emphasis on specific documents and authors. However, this distortion should be negligible when the number of authors is sufficiently high compared to the number of published articles. Moreover, there are several reasons why a document may or may not be cited, and the number of citations does not imply quality. Indeed, bibliometrics does not measure quality, and it is essential to combine bibliometric and qualitative analysis to evaluate the works' quality and contributions.

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