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Review

Bibliographic Analysis of Scientific Research on Downy Mildew (*Pseudoperonospora humuli*) in Hop (*Humulus lupulus* L.)

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Abstract: This study focused on downy mildew in hop caused by the pathogen *Pseudoperonospora humuli*. A systematic literature review was conducted using bibliometric analysis to explore trends in publishing, prominent research themes, and where research is being conducted on hop downy mildew. The databases Scopus, Web of Science, and ScienceDirect were used to identify publications spanning from 1928 to 2023. The analysis yielded 54 publications, with the most cited studies primarily focusing on disease management and host resistance. Additionally, these studies explored the genetic and pathogenic relationship between *P. cubensis* and *P. humuli*. A word co-occurrence map revealed that the main themes addressed in the publications included "hop", "disease", "downy", "*humuli*", "mildew", and "*Pseudoperonospora*". Notably, there was a particular emphasis on subtopics such as disease management, the disease reaction of hop cultivars, and the influence of weather factors on hop downy mildew. Notably, there was limited knowledge about the disease in regions with tropical climates. This study provides valuable information that can support and guide future research endeavors concerning downy mildew in hop cultivation.

Keywords: bibliographic analysis; disease management; fungal disease; overview; review

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1. Introduction

Hop (*Humulus lupulus* L.) is primarily cultivated for beer production [1]. The global production of hops is concentrated in temperature regions in the Northern Hemisphere, between latitudes 35° and 55°, with the United States and Germany being the largest producers [2]. Smaller production regions exist in the Southern Hemisphere, which now includes a developing industry in Brazil between the latitudes 0° and 23.5° in regions with a tropical climate.

The productivity of hop cultivation is closely linked to the health conditions of the plants, and one of the major challenges affecting productivity is crop diseases [3]. One of the most destructive diseases of cultivated hop is downy mildew, which is caused by *Pseudoperonospora humuli*, an obligate biotrophic oomycete that can lead to 100% crop loss in susceptible cultivars [4]. Hop downy mildew is favored by moderate temperatures (15 to 20 °C), high humidity (>90% relative humidity), and prolonged periods of wetness [5].

In general, disease management practices for hop downy mildew involve the use of fungicides, cultivars with varying levels of resistance to the disease, and cultural practices that reduce inoculum or modify the crop microclimate [6]. To effectively control the disease and develop sustainable disease management strategies, it is crucial to have a comprehensive understanding of pathogen biology and disease ecology. This knowledge enables one to identify and utilize new sources of resistance and to develop more sustainable approaches [7].

Researchers often face questions that drive them to plan and organize their studies in a more effective manner to meet the scientific requirements related to the subject. This may involve establishing research networks that complement existing research capacity [8]. Bibliographic analysis is useful for understanding trends in publishing, research themes, and authors and institutions conducting research. Bibliographic analysis typically involves textual data analysis. By treating textual data as numerical data, conducting analyses that integrate qualitative and quantitative elements becomes possible. This approach enables researchers to address qualitative questions using quantitative elements as a foundation [9].

The present study conducted a bibliographic analysis of the scientific literature related to hop downy mildew. This systematic literature review employed textual and content analysis techniques to summarize scientific publications from 1928 to 2023. We examined their geographical distribution and content, with particular emphasis on studies conducted in tropical climates.

2. Material and Methods

A bibliometric review of the literature was carried out using a quantitative approach to identify the major themes addressed in existing studies on hop downy mildew. The analysis was performed using the keywords "downy mildew", "*Pseudoperonospora humuli*", and "hop". These keywords were combined using the Boolean operator "AND" in a search string, as shown in Table 1. The studies were extracted from the databases of the scientific publications Scopus (Elsevier), Web of Science (Clarivate), and Science Direct (Elsevier), considering works published from 1928 to 2023.

Table 1. Parameters of the query performed in the Scopus, Web of Science, and ScienceDirect databases.

Database	Parameters
Scopus	(TITLE-ABS-KEY (hop) AND TITLE-ABS-KEY (downy AND mildew) AND TITLE-ABS-KEY (*Pseudoperonospora* AND *humuli*) AND PUBYEAR < 2023 AND PUBYEAR < 2023)
Web of Science	hop (Topic) and downy mildew (Topic) and *Pseudoperonospora humuli* (Topic)
ScienceDirect	hop AND downy AND mildew AND *Pseudoperonospora humuli*

Based on the research questions, inclusion and exclusion criteria were established, as were the data extraction and analysis methods. In the inclusion and exclusion criteria, filters such as language, year, document type, and area of knowledge were not applied to encompass the earliest studies on the subject. Articles that were available in full were included in the review. However, articles that did not address downy mildew in hop cultivation or focused on diseases other than downy mildew in hop were excluded. Duplicate articles were eliminated before classifying and organizing the articles according to the year of publication and theme.

The selected publications had their geographic coordinates (latitude and longitude) extracted in CSV format. These coordinates were obtained either from the study sites mentioned in the articles or the location of the institutions conducting the research. The distribution of these locations was visualized on a map in a Geographic Information Systems (GIS) environment.

The obtained data were subjected to analysis of the most frequent topics using two open-source and free software programs: VOSviewer (version 1.6.17, Centre for Science and Technology Studies, Leiden University, NL) and IRAMUTEQ (version 0.7, alpha 2, Toulose, FR). While VOSviewer is standalone software, IRAMUTEQ is anchored in the statistical environment of R* software (version 3.2.3) [10] and utilizes Python language. These software tools were employed to perform topic analysis based on the data collected.

Using IRAMUTEQ, a textual data analysis was conducted to explore the correlation between the keywords found in the researched articles, as represented by the distance between them [11]. This analysis involved a similarity analysis based on graph theory, which helped identify co-occurrences between words and provided insights into their connections [11]. Additionally, a lexical analysis was performed using a word cloud, which visually grouped and organized keywords based on their frequency. In the word cloud, the size of each word represents its importance and frequency, with larger words indicating more prominent and frequent topics; the frequency of the topics is represented through clusters.

VOSviewer is widely used software in bibliometric analysis. By employing graph theory, VOSviewer facilitates the extraction of information from scientific collaborative networks and presents graphical representations of bibliometric maps in an intuitive manner [12]. One of the key features of VOSviewer is its ability to identify co-occurrences between words, which provides insights into the connections between them. This analysis helps to identify the main structures within a textual corpus, distinguishing commonalities and specificities based on the descriptive variables identified in the analysis [13].

In VOSviewer, a bibliometric map of the co-occurrence of keywords is generated. This map illustrates the frequency of each keyword used by the authors as well as the intensity of the connections, indicating the relationship of each term with other words. Additionally, VOSviewer allows for the visualization of clusters, which are groups of items (nodes) with greater affinity on the map. These clusters represent closely related topics or concepts [12].

3. Results and Discussion

The selection of scientific databases such as Scopus, Web of Science, and ScienceDirect was made due to their extensive collections of abstracts and citations, making them valuable sources of literature on the subject. These databases also provide bibliometric data tools for tracking, analysis, and research visualization.

In the initial search, a total of 194 publications were identified (Figure 1). However, to create the bibliographic portfolio, the articles were filtered based on their titles and abstracts. Using the exclusion criteria, 140 articles were excluded, resulting in a final selection of 54 articles for this review.

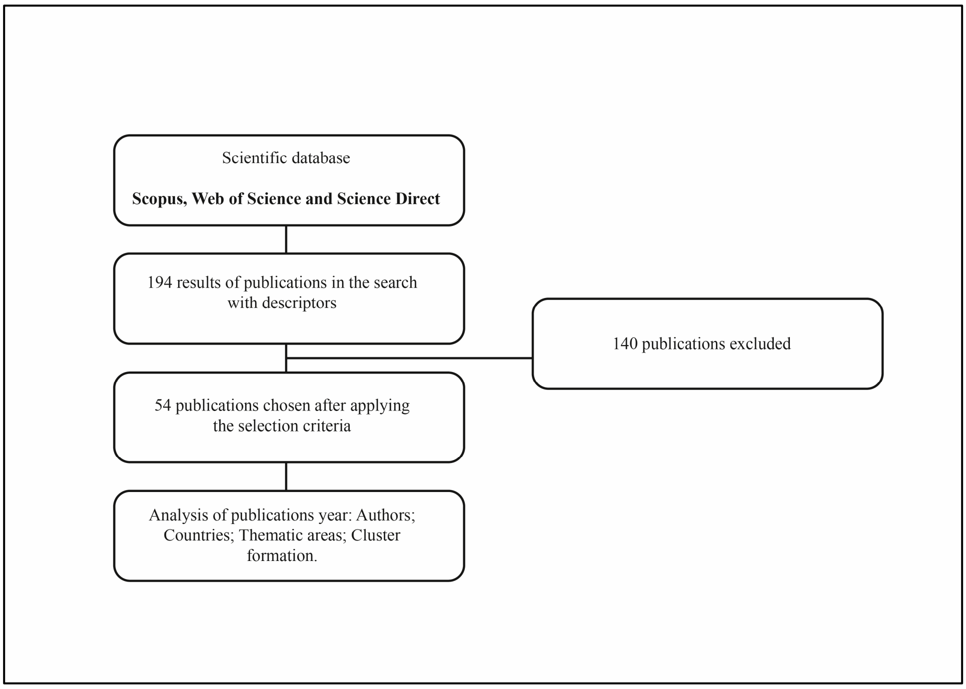

Figure 1. Process of selection and analysis of scientific literature on hop downy mildew.

After applying the exclusion criteria, only complete and unique articles were selected for further analysis. The Scopus database yielded 50 articles that met the criteria, while the Web of Science database provided 3 articles, and ScienceDirect contributed 1 article (Table 2). The analysis of the selected articles indicates that there is still a relatively limited number of studies focusing on hop downy mildew in generally, and a particular dearth of information globally in tropical climates. This suggests that further research in this area is needed to expand the current knowledge base.

Table 2. Hop downy mildew found in Scopus, Web of Science, ScienceDirect repositories.

	Author	Title of the Article	Journal
		Web of Science	
1	[14]	Reevaluation of Host Specificity of the Closely Related Species *Pseudoperonospora humuli* and *P. cubensis*	Plant disease
2	[15]	A Multiplex TaqMan qPCR Assay for Detection and Quantification of Clade 1 and Clade 2 Isolates of *Pseudoperonospora cubensis* and *Pseudoperonospora humuli*	
3	[16]	Detection of Airborne Sporangia of *Pseudoperonospora cubensis* and *P. humuli* in Michigan Using Burkard Spore Traps Coupled to Quantitative PCR	
		ScienceDirect	
1	[17]	Effects of light during infection on the incidence of downy mildew (*Pseudoperonospora cubensis*) on cucumbers	Physiological Plant Pathology
		Scopus	
1	[18]	Inoculation experiments with the downy mildews of the Hop and Nettle (*Pseudoperonospora humuli* (Miy. et Taka.) Wils. and P. Urticare (Lib.) Salmon et Ware)	Annals of Botany
2	[19]	Experiments on the Production of Diseased Shoots by the Hop Downy Mildew, *Pseudoperonospora humuli* (Miy. et Takah.), Wils.	
3	[20]	The downy mildew of the hop in 1930.	
4	[21]	Overwintering of hop downy mildew *Pseudoperonospora humuli* (Miy. and Tak.) Wilson	Annals of Applied Biology
5	[22]	Persistence and identification of downy mildew *Pseudoperonospora humuli* (Miy. and Tak.) Wilson in hop rootstocks	
6	[23]	Infection of hop rootstocks by downy mildew *Pseudoperonospora humuli* (Miy. & Tak.) Wilson and its control by early-season dusts	
7	[24]	Early -season control of hop downy mildew, *Pseudoperonospora humuli* (Miy. and Tak.) Wilson, with streptomycin and protectant fungicides in severely infected plantings	
8	[25]	Infection periods in relation to the natural development of hop downy mildew (*Pseudoperonospora humuli*)	
9	[26]	The influence of stomatal opening on the infection of hop leaves by *Pseudoperonospora humuli*	Physiological Plant Pathology
10	[27]	Quantitative relationships between infection by the hop downy mildew pathogen, *Pseudoperonospora humuli*, and weather and inoculum factors	Annals of Applied Biology
11	[28]	Factors affecting zoospore responses towards stomata in hop downy mildew (*Pseudoperonospora humuli*) including some comparisons with grapevine downy mildew (*Plasmopara viticola*)	Physiological Plant Pathology
12	[29]	Epidemic related decision model for control of downy mildew in hop (*Pseudoperonospora humuli* Miy. et Tak.), based on critical amount of spores	Invasive Species Compendium
13	[30]	Ultrastructure of the Host-Parasite Relationships of *Pseudoperonospora humuli* on Hops	Australian Journal of botany
14	[31]	Marker-assisted hop (*Humulus lupulus* L.) breeding	Monatsschrift fur Brauwissenschaft
15	[32]	Forecasting climate suitability of Australian hop-growing regions for establishment of hop powdery and downy mildews	Australasian Plant Pathology

Table 2. *Cont.*

	Author	Title of the Article	Journal
16	[33]	A re-consideration of *Pseudoperonospora cubensis* and *P. humuli* based on molecular and morphological data	The British Mycological Society
17	[34]	Hop (*Humulus lupulus* L.) Transformation with Stilbene Synthase for Increasing Resistance against Fungal Pathogens	Acta Horticulturae
18	[35]	Population Biology of *Pseudoperonospora humuli* in Oregon and Washington	The American Phytopathological Society
19	[36]	Persistence of Phenylamide Insensitivity in *Pseudoperonospora humuli*	Plant Disease
20	[37]	Predicting Infection Risk of Hop by *Pseudoperonspora humuli*	The American Phytopathological Society
21	[38]	PCR detection of *Pseudoperonospora humuli* in air samples from hop yards	Plant Pathology
22	[39]	Forecasting and Management of Hop Downy Mildew	Plant Disease
23	[40]	Genetic and Pathogenic Relatedness of *Pseudoperonospora cubensis* and *P. humuli*	The American Phytopathological Society
24	[41]	Registration of 'Dana'—A Bittering Hop Cultivar with a Pleasant Hoppy Aroma	Journal of Plant Registrations
25	[42]	Spatial analysis and incidence–density relationships for downy mildew on hop	Plant Pathology
26	[43]	Association of Spring Pruning Practices with Severity of Powdery Mildew and Downy Mildew on Hop	The American Phytopathological Society
27	[44]	Pre-and postinfection activity of fungicides in control of hop downy mildew	Plant Disease
28	[45]	Precision QTL mapping of downy mildew resistance in hop (*Humulus lupulus* L.)	Euphytica
29	[46]	*Pseudoperonospora cubensis* and *P. humuli* detection using species-specific probes and high-definition melt curve analysis	Canadian Journal of Plant Pathology
30	[47]	Genotyping-by-sequencing of a bi-parental mapping population segregating for downy mildew resistance in hop (*Humulus lupulus* L.)	Euphytica
31	[48]	Susceptibility of Hop Cultivars to Downy Mildew: Associations with Chemical Characteristics and Region of Origin	Plant Health Progress
32	[49]	Homothallism in *Pseudoperonospora humuli*	Plant Pathology
33	[50]	Genotyping-by-Sequencing Reveals Fine-Scale Differentiation in Populations of *Pseudoperonospora humuli*	Phytopathology
34	[51]	Investigating Phenylamide Insensitivity in Wisconsin Populations of *Pseudoperonospora humuli*	Plant Health Progress
35	[52]	Genome sequencing and transcriptome analysis of the hop downy mildew pathogen *Pseudoperonospora humuli* reveal species-specific genes for molecular detection	Phytopathology
36	[53]	Post-harvest recognition of various fungicide treatments for downy mildew of hops using comprehensive pesticide residue monitoring	International Journal of Pest Management
37	[54]	High Levels of Insensitivity to Phosphonate Fungicides in *Pseudoperonospora humuli*	Plant Disease
38	[55]	The Effector Repertoire of the Hop Downy Mildew Pathogen *Pseudoperonospora humuli*	Frontiers Genetic
39	[4]	Hop Downy Mildew Caused by *Pseudoperonospora humuli*: A Diagnostic Guide	Plant Health Progress
40	[56]	Downy mildew resistance is genetically mediated by prophylactic production of phenylpropanoids in hop	Plant, Cell & Environment
41	[57]	Susceptibility of Hop Cultivars and Rootstock to Downy Mildew Caused by *Pseudoperonospora humuli*	HortScience
42	[58]	*Pseudoperonospora humuli* might be an introduced species in Central Europe with low genetic diversity but high distribution potential	Jornal Plant Pathology
43	[59]	"Jumping Jack": Genomic Microsatellites Underscore the Distinctiveness of Closely Related *Pseudoperonospora cubensis* and *Pseudoperonospora humuli* and provide new insights into their evolutionary past	Frontiers Microbiology
44	[7]	The hop downy mildew pathogen *Pseudoperonospora humuli*	Wiley Molecular Plant Pathology
45	[60]	Results from Hop Cultivar Trials in Mid-Atlantic United States	HortTechnology

Table 2. *Cont.*

	Author	Title of the Article	Journal
46	[61]	Fungicide efficacy against *Pseudoperonospora humuli* and point-mutations linked to carboxylic acid amide (CAA) resistance in Michigan	Plant Disease
47	[62]	Optimizing Molecular Detection for the Hop Downy Mildew Pathogen *Pseudoperonospora humuli* in Plant Tissue	Phytopathology
48	[63]	Use of botanicals to protect early stage growth of hop plants against *Pseudoperonospora humuli*	Crop Protection
49	[64]	Genetic characterization of downy mildew resistance from the hop (*Humulus lupulus* L.) line USDA 64035M	Crop Science
50	[65]	Diversity in genetic and downy mildew resistance among wild and mutagenized hops as revealed by single nucleotide polymorphisms and disease rating	Canadian Journal of Plant Science

Based on the research results, it is evident that the number of articles published on hop downy mildew has shown a slow evolution across all databases. From 1928 to 2006, a span of 78 years, only 19 articles were published, resulting in an average of 1 publication every 4.1 years. However, starting from 2007, there has been a notable increase in the number of published articles. Over the period of 2007 to 2023, which spans 16 years, a total of 35 publications were made available, averaging 2.1 publications per year. This indicates that in recent years, the scientific production of hop downy mildew has been more actively explored (Figure 2).

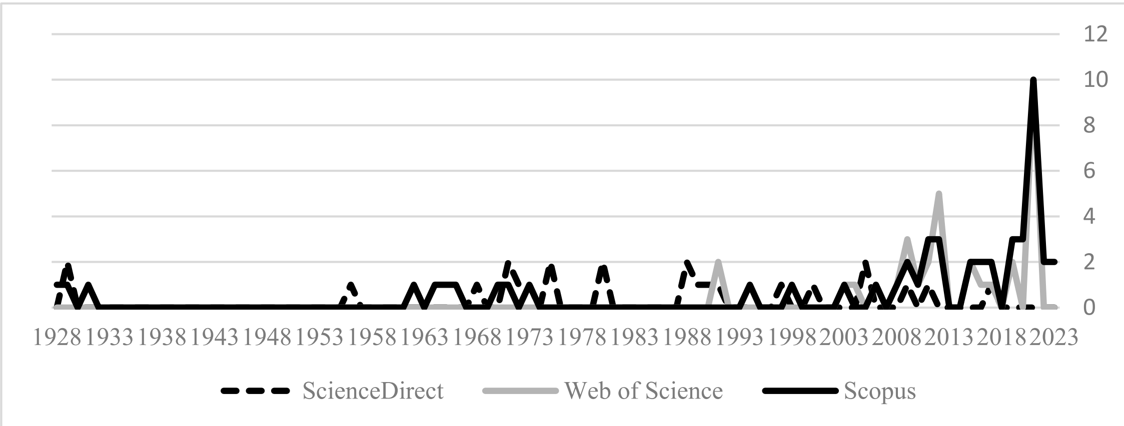

Figure 2. Comparison of the number of articles published on hop downy mildew in the Scopus, Web of Science, and ScienceDirect databases from 1928 to 2023.

Figure 3 illustrates the distribution of publications on hop downy mildew among different countries. The United States emerges as the country with the highest number of publications, with 29 articles, followed by the United Kingdom, with 12 articles. Together, these two countries contribute to approximately 76% of the total number of publications. This concentration of scientific production aligns with the fact that the United States is one of the largest producers of hops and with the long-standing hop industry in the United States and the United Kingdom [2]. The fact that the geographical location of these countries lies within the latitudinal range of 35° to 55°, where hops are traditionally cultivated, may also contribute to their significant scientific contributions in this area, as well as the long-term institutional support for research.

It is worth noting that no studies have been conducted on hop downy mildew in South America. However, the issues related to hop adaptability have been gaining attention in Brazil, as highlighted by [66–68]. While research on hop cultivation is accumulating in Brazil, there is still a lack of knowledge about downy mildew specifically. The absence of studies on this topic in regions with tropical climates underscores the need for further research and understanding in these areas.

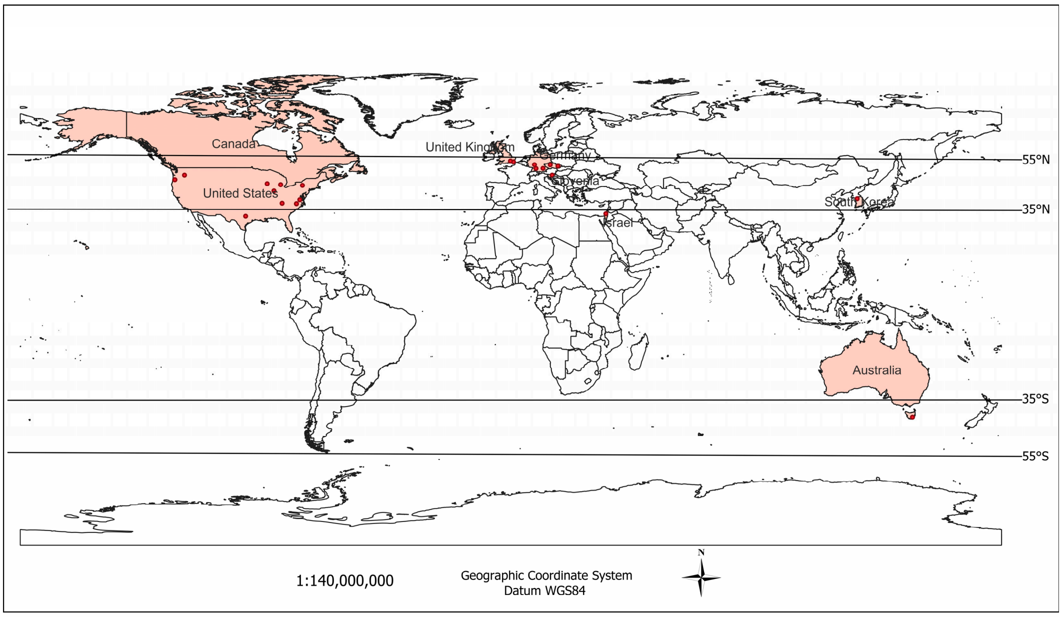

Figure 3. Spatial distribution of the selected articles on hop downy mildew.

The limited knowledge of downy mildew in hop cultivation in regions with tropical climates poses challenges for hop growers and researchers in these regions. Future research efforts should aim to address this gap in knowledge and management strategies for downy mildew under tropical conditions.

Among the published articles, several studies have addressed various aspects related to downy mildew in hop cultivation. Early studies focused on understanding the pathogen lifecycle, with specific emphasis on overwintering mechanisms and conditions that favor secondary infection [17,22,23,41]. More recently, other studies have explored the reproductive behavior and spatial patterns of downy mildew [42,43,49,50]. Additionally, the relationship between different *Pseudoperonospora* species, morphological comparisons, and environmental conditions necessary for infection have been investigated [14,25–27].

Agronomic factors, such as the interactions of pests and diseases, climate influence, and hop genetics, have also been studied concerning downy mildew [37,47,60]. These studies provide insights into the factors that can affect yield and disease resistance in hops.

Recent studies have focused on exploring the hop response to the pathogen at the biochemical, molecular, and genetic levels. They have identified resistance-associated markers, analyzed genetic diversity, and developed detection methods for *Pseudoperonospora* species [15,16,35,36,52,53,56,57,59,64,66,67]. These molecular approaches are crucial for the effective management and control of pathogens, facilitating early detection and decision making regarding crop protection.

Furthermore, studies have focused on genomics, the timing of fungicide application, and resistance factors related to downy mildew [4,16,47,50,55]. The impact of climate on downy mildew development has also been investigated over long-term periods [32]. Evaluating hop cultivars and understanding the disease symptoms, life cycle, virulence factors, and management strategies have been the focus of other studies [7,60].

These studies have collectively contributed to a better understanding of downy mildew in hops, including its molecular characteristics, epidemiology, management strategies, and factors influencing disease development. They have played a crucial role in guiding disease control practices, improving crop resilience, and optimizing hop production.

3.1. Graphic Analysis of Keywords Using VOSviewer Software

The co-occurrence analysis of keywords revealed three distinct clusters on the map. The first cluster, represented in red, is centered around keywords such as "*Pseudoperonospora humuli*", "*Peronosporaceae*", and "*Humulus*", which are related to the biological classification of downy mildew and hop. This cluster likely includes studies that specifically focus on the taxonomy, classification, and characteristics of *Pseudoperonospora humuli* and its relationship with the Peronosporaceae family and *Humulus* genus.

The second cluster, represented in green, encompasses keywords like "Peronospora", "microbiology", "genetics", "Plant Disease", and "oomycetes". These terms indicate a focus on studies related to the genus *Peronospora*, including aspects of microbiology and genetics. The inclusion of "Plant Disease" suggests that these studies might be published in the journal *Plant Disease*, which is a prominent journal in the field of phytopathology. This cluster likely represents research that explores microbiological and genetic aspects of *Peronospora* species, including phylogenetic and taxonomic studies.

The third cluster, depicted in blue, includes keywords such as "hop", "*Pseudoperonospora cubensis*", and "downy mildew". These terms are closely associated with the genus of hop downy mildew and suggest studies spanning the sister species, *P. cubensis*. cucurbit downy mildew, caused by *P. cubensis*, is a re-emergent disease (CITATIONS). Multiple studies have been conducted to understand the genetic and pathogenic relatedness of *P. cubensis* and *P. humuli* (CITATIONS). The presence of "*Pseudoperonospora cubensis*" indicates research investigating the genetic and pathogenic similarities between *P. cubensis* and *P. humuli*, two species of *Pseudoperonospora* associated with downy mildew in hops. This cluster likely encompasses studies exploring the genetic diversity, pathogenicity, and management of downy mildew specifically in hops (Figure 4).

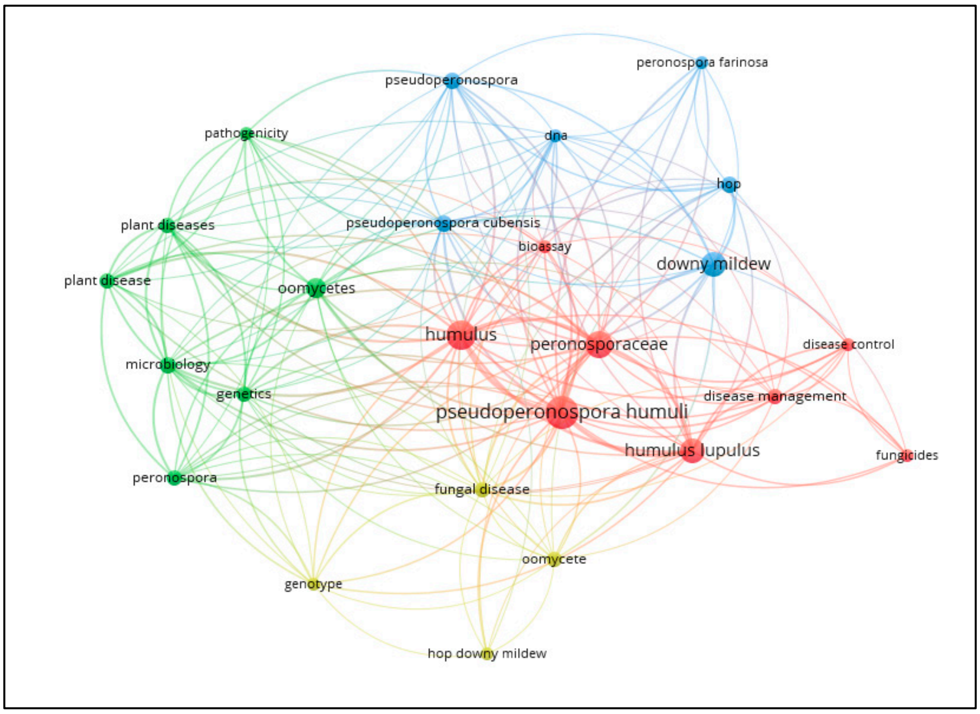

Figure 4. Bibliometric map of co-occurrence networks of keywords used in this review.

3.2. Graphic Analysis of Keywords Using IRAMUTEQ Software

IRAMUTEQ's graphical representation of the keywords with more co-occurrences provides a simpler depiction of the relationships between subjects and their depths compared to VOSviewer. The similarity chart generated by IRAMUTEQ highlights the main topics addressed in the 54 objects of study, namely, "hop", "disease", "downy", "*humuli*", "mildew", and "*Pseudoperonospora*" (Figure 5A).

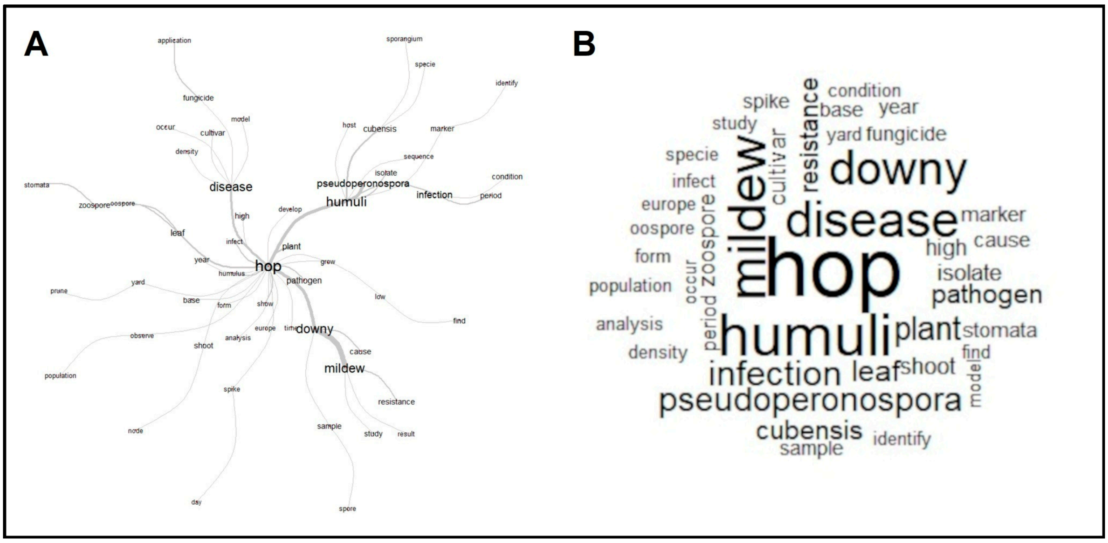

Figure 5. Similitude graph (**A**) and point cloud (**B**) of the 50 articles published on downy mildew in hops.

The prominence of these keywords can be attributed to the increasing demand for hop-derived products and the consequent emphasis on improving productivity and efficiency in hop cultivation. The scientific production on this subject has evolved in parallel with the agricultural industry's need for effective disease management strategies and a deeper understanding of the biology and control of downy mildew in hops.

The word cloud (Figure 5B) highlights the most frequent keywords present in the studies, including terms such as "hop", "disease", "*humuli*", "mildew", "downy", "infection", "resistance", and "*Pseudoperonospora*". By analyzing these keywords in connection, it becomes apparent that a significant focus of the studies is on the *P. humuli* species and its potential socioeconomic consequences in hop cultivation.

According to [33], downy mildew caused by *P. humuli* leads to substantial economic losses in hop plantations. [44] further emphasizes that inadequate disease management can result in significant crop losses. Therefore, managing downy mildew is a major challenge in hop cultivation, particularly in regions with high humidity, as highlighted by [56].

Although the term "cultivate" is less represented among the 54 publications, it deserves special attention as it appears more frequently in the keywords of recent articles. This can be attributed to the increasing global market demand for hops and their by-products, as noted by [60]. The concern with diseases and pests, including downy mildew, is a central theme in current studies, reflecting the need to develop strategies for maintaining hop crop health and productivity.

4. Final Considerations

In summary, this analysis of the scientific research on hop downy mildew highlights significant progress in recent decades, reflected in the notable increase in the number of articles published. The main topics covered in these 54 articles focus on "hops", "*humuli*", "disease", and "downy mildew", underlining the relevance of downy mildew in hop production. These studies focus on disease management, the susceptibility of hop varieties to downy mildew, the influence of climate on the development of the disease, and methods of detection and control. Even the older articles retain their relevance, as they discuss challenges in managing the disease, forecasts based on infection periods and meteorological factors, and the potential use of geotechnologies to address these challenges.

The expansion of hop cultivation to new regions, including Brazil, demonstrates the interest in exploring the adaptability of this crop in different climates. However, it is essential to point out that Brazil lacks more-in-depth research on hop downy mildew, highlighting the need to encourage studies and publications in this area, especially in regions with a tropical climate.

Ultimately, encouraging additional research and interdisciplinary collaborations could result in significant advances in the understanding, management, and control of hop downy mildew, benefiting both the hop industry and global beer production, as well as supporting sustainable hop cultivation practices.

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