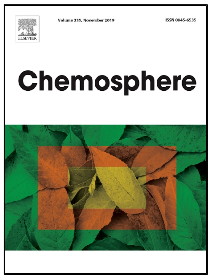
Journal Pre-proof



Analysis of Global Trends and Gaps for Studies about 2,4-D Herbicide Toxicity: A scientometric review

Natana Raquel Zuanazzi, Nédia de Castilhos Ghisi, Elton Celton de Oliveira

PII:	S0045-6535(19)32255-6
DOI:	https://doi.org/10.1016/j.chemosphere.2019.125016
Reference:	CHEM 125016
To appear in:	*Chemosphere*
Received Date:	06 July 2019
Accepted Date:	29 September 2019

Please cite this article as: Natana Raquel Zuanazzi, Nédia de Castilhos Ghisi, Elton Celton de Oliveira, Analysis of Global Trends and Gaps for Studies about 2,4-D Herbicide Toxicity: A scientometric review, *Chemosphere* (2019), https://doi.org/10.1016/j.chemosphere.2019.125016

Analysis of Global Trends and Gaps for Studies about 2,4-D Herbicide Toxicity: A scientometric review

Natana Raquel Zuanazzi[a], Nédia de Castilhos Ghisi[b*], Elton Celton de Oliveira[a]

[a]Postgraduate Program in Agroecosystems (PPGSIS), Federal University of Technology – Paraná, Campus Dois Vizinhos (UTFPR–DV), Estrada para Boa Esperança, km 4, 85660-000, Dois Vizinhos (PR) Brazil. natana_zuanazzi@hotmail.com, elton.c.oliveira2@gmail.com

[b]Postgraduate Program in Biotechnology (PPGBIOTEC), Federal University of Technology – Paraná – Paraná, Campus Dois Vizinhos (UTFPR–DV) Estrada para (Road to) Boa Esperança, km 4, 85660-000, Dois Vizinhos (PR) Brazil, nediaghisi@gmail.com

BULLET POINTS (3 to 5 bullet points - maximum 85 characters)

- Research on 2,4-D toxicology and mutagenicity has advanced rapidly.
- USA stands out in studies involving 2,4-D toxicity, followed by Canada and China
- Toxicology and Biochemical/Molecular Biology were the areas of greatest influence
- strongly related to the keywords glyphosate, atrazine, water and gene expression
- Focus on occupational risk, degradation, neurotoxicity and herbicide tolerance

*Corresponding author at: Nédia C. Ghisi, UTFPR (Universidade Tecnológica Federal do Paraná) – Campus Dois Vizinhos, Estrada para Boa Esperança, Km 04, Comunidade de São Cristóvão, ZIP CODE: 85660-000– Dois Vizinhos /PR – Brazil. Tel: +55 (46)3536-8900/Email: nediaghisi@gmail.com

ABSTRACT

2,4-dichlorophenoxyacetic acid (2,4-D) is a herbicide that is used worldwide in agricultural and urban activities to control pests, reaching natural environments directly or indirectly. The research on 2,4-D toxicology and mutagenicity has advanced rapidly, and for this reason, this review summarizes the available data in Web of Science (WoS) to provide insights into the specific characteristics of 2,4-D toxicity and mutagenicity. Contrary to traditional reviews, this study uses a new method to quantitatively visualize and summarize information about the development of this field. Among all countries, the USA was the most active contributor with the largest publication and centrality, followed by Canada and China. The WoS categories 'Toxicology' and 'Biochemical and Molecular Biology' were the areas of greatest influence. 2,4-D research was strongly related to the keywords glyphosate, atrazine, water and gene expression. The studies trended to be focused on occupational risk, neurotoxicity, resistance or tolerance to herbicides, and to non-target species (especially aquatic ones) and molecular imprinting. In general, the authors have worked collaboratively, with concentrated efforts, allowing important advances in this field. Future research on 2,4-D toxicology and mutagenicity should probably focus on molecular biology, especially gene expression, assessment of exposure in human or other vertebrate bioindicators, and pesticide degradation studies. In summary, this scientometric analysis allowed us to make inferences about global trends in 2,4-D toxicology and mutagenicity, in order to identify tendencies and gaps and thus contribute to future research efforts.

Keywords: Citespace, Degradation, Emerging trends, Mutation, Occupational exposure, pesticides mixture.

1. Introduction

2,4-dichlorophenoxyacetic acid (2,4-D) was developed during World War II (Suzuki, 2017) and introduced on the market in the 1940s (Islam et al., 2018) to control broad-leaved weeds (Aquino et al., 2007). Due to its efficiency, selectivity, low cost and broad spectrum in pest control, it has become a widely-used herbicide in agricultural and urban areas around the world (Dehnert et al., 2018).

According to Islam et al., (2018), 2,4-D has been registered as an ingredient in approximately 1500 agricultural and home use herbicides, as a sole active ingredient or in conjunction with other active ingredients. The compound mimics natural plant auxins and promotes division, differentiation and elongation of plant cells, acting as a regulator of plant growth (Islam et al., 2017; Song, 2014). However, even in low concentrations, 2,4-D has a herbicidal effect on dicotyledonous due to its ability to produce uncoordinated cell growth (Stackelberg, 2014), damage to conducting vessels and root, leaf and stem malformation (RED, 2006). Its action mode is effective through the increase in cell-wall plasticity, biosynthesis of proteins and production of ethylene.

The main markets for 2,4-D are the developed and developing countries with an agricultural vocation. In the US, for example, the estimated annual cost of marketing the product is close to 45 million pounds or 57 million dollars (RED, 2006). In China, 2,4-D production in 2010 reached the mark of 40,000 tons (Liu et al., 2013). Developing countries in South America, such as Argentina and Brazil, also have a large consumption of 2,4-D (Merini et al., 2007). Data from IBAMA (Brazilian Institute of the Environment) ranked 2,4-D as the second best-selling pesticide in the country in 2017 (behind glyphosate), with 57,389.35 tons (IBAMA, 2018).

A worldwide increase in 2,4-D use in soybean, corn and wheat has recently come to be expected, due to the development of new genetically modified crops resistant to this pesticide, since glyphosate-resistant technology is outdated (Freydier and Lundgren, 2016). It is important to highlight that 2,4-D resistant crops were registered by the US-EPA in 2014 (EPA Registration Number 62719-649; Decision Number 457755). This factor, and the characteristics of the chemical molecule and its kinetics in the environment, have led to concerns about the consequences of its interaction with different biological compartments (Freydier and Lundgren, 2016).

Once 2,4-D is applied directly on the soil or sprayed on the crops, it easily reaches water bodies (Chinalia and Killham, 2006). Being a polar molecule, 2,4-D and its ester and amines are quite mobile in aqueous systems because of its acidic carboxyl group (pKa = 2.8) and low soil adsorption that may be the reason for its widespread occurrence in the environment (Islam et al., 2018). In addition, it can reach the water bodies by surface runoff or through infiltration, leaching and soil percolation, becoming an environmental and human health problem (Gaultier et al., 2008; Kearns et al., 2014; Shareef and Shaw, 2008). It is also noteworthy that 2,4-D is a moderately persistent substance in the environment, with a half-life between 20 and 312 days, depending on environmental conditions (Walters, 2011).

Studies have demonstrated the bioaccumulation capacity of this herbicide in non-target organisms, exposed for a short period of time (da Fonseca et al., 2008; Kiljanek et al., 2016). Regarding the post-2,4-D exposure effects in different organisms, studies have shown endocrine disruption (Guerrero-Estévez and López-López, 2016), reproductive disorders (Pattanasupong et al., 2004), genetic alterations (Lajmanovich et al., 2015) and carcinogenic effects (Loomis et al., 2015). In humans, 2,4-D has been associated with the development of Parkinson's neurodegenerative disease (Tanner et al., 2009) and autism (Román, 2007).

Related and unrelated information on the 2,4-D toxicology is published daily in various journals around the world, making it difficult to effectively follow the progress of information in one or more areas of knowledge. Scientometric methods become adequate tools to summarize the complex and growing available information set, in order to show global trends and gaps in knowledge. It is possible to map journals, countries, authors, main research areas and the most active areas, and identify and visualize the main factors that contribute to the evolution of knowledge about a chosen theme (Chen, 2014, 2006a, 2004).Therefore, in recent years, there has been a growing interest in the scientific community to evaluate state-of-the-art research through the use of the scientometric methods (He et al., 2019; Konur, 2011).

Considerable attention has been given to the problems caused by 2,4-D and excellent narrative (Islam et al., 2018) and meta-analytical reviews (Goodman et al., 2015; Smith et al., 2017; Stackelberg, 2014) have been published recently. Nevertheless, up to the present, there has been no published scientometric revision that addresses this issue systematically. Moreover, no previous review in this area has defined the articulations or the working relationships between the research groups and their countries. In addition, there are no studies that include aspects such as clusters, co-citations or keywords.

The aim of this study is to comprehensively and systematically provide a scientometric review of the research on the toxicity and mutagenicity of 2,4-D. More specifically, our analysis mainly focuses on the network of cooperation between countries, co-authorship and co-occurring keywords analyzing publications from 1949 to 2018. Therefore, the knowledge domains, quantified research patterns, intellectual structure and emerging trends in this field can be explored, which is helpful in obtaining more accurate and complete information and offering insights into research topics and trend evaluation from different perspectives.

2. Materials and methods

Data was obtained from the ISI – Web of Science (WoS) database of Clarivate Analytics. The searched terms included TS (Topic Search) = (2,4-D OR "dichlorophenoxyacetic acid" AND mutation AND toxicity).

The search generated 10,735 references between 1949 and 2018. Later refining manually analyzed each publication through the reading of titles and abstracts to select only 2,4-D studies involving toxicity and mutation. A total of 9,319 publications were excluded (1,416 selected), since several publications dealt with the use of 2,4-D as a stimulation hormone for auxin production.

Document type, publication year, source and its impact factor, category, country and author were identified for each publication. The impact factor was obtained from Journal Citation Reports (JCR) (clarivate.com/products/web-of-science).

The results were analyzed using Microsoft Office Excel and CiteSpace software. The latter provides functions to facilitate understanding and interpretation of network patterns, including thematic area identification, finding specific points, and labeling selected terms. The software allows mapping authors, journals (sources) and countries, helping to identify and visualize the main factors contributing to the knowledge evolution in the search field under study (Chen, 2014, 2006a, 2004).

Program outcomes are shown as infographic forms, with a node indicating research items, and links between nodes describing citations or mutual references between these nodes. Each node is represented by a series of tree rings in different colors; a spectrum of colors serves to indicate the chronological order of occurrence of links and items (Xiang et al., 2017). Thus, blue represents older articles, while recent articles appear in orange, with an outer purple ring that indicates good centrality (or simply as yellow circles). Betweenness centrality is defined in the following Equation (1).

$$Centrality\ (node\ i) = \sum_{i \neq j \neq k} \frac{\rho_{jk}(i)}{\rho_{jk}} \quad \text{(Eq. 1)}$$

In Eq. (1), ρ_{jk} represents the number of shortest paths between node *j* and node *k*, and ρ_{jk} *(i)* is the number of those paths that pass through node *i*. At document level, the importance of each document in a co-citing network can be partially evaluated by the indicator betweenness centrality (Li et al., 2017; Ouyang et al., 2018).

In addition, some data was subjected to cluster analysis, which is an analysis in which similar objects are combined to determine related areas of study. The size of the set is the number of grouped objects. CiteSpace assigns ID zero (#0) to the largest cluster, ID one to the second largest, and so on (Chen, 2015). Statistica (StatSoft, 2007) was used to perform Pearson's correlation analysis.

3. Results and Discussion

3.1 Publication characteristics

An increase was observed in the number of yearly publications, from one in 1949 to 70 in 2018. The highest numbers were observed in 2016 and 2017 (81 and 82 articles, respectively). These publications were classified into 9 types of documents: article, procedure paper, abstract, review, letter, note, editorial material, book review and other items. Articles were predominant comprising 88.63% of the total. The predominant language was English (97.81%), regardless of the country of origin, since it is the preferred language of communication in the scientific community (Schuermans et al., 2010).

The articles in our refined dataset were cited 25,051 times, with an average of 17.69 citations per item and an H-index of 66. This suggests that this line of research has a relevant impact factor. The H-index was developed by Hirsch in 2005 and reveals the impact or relevance of a paper, author or field. The H-index is the maximum value of *h* for which an author or area has published *h* papers that have been cited at least *h* times. In summary, our dataset had 66 papers with at least 66 citations each (Hirsch, 2005). Thus, citation analysis provides important information about the scientific impact of 2,4-D herbicide toxicity and mutagenicity research.

Fig. 1 shows the total number of publications and citations per year. It also shows that since 1991 there has been a considerable and growing increase in the publications and citation numbers regarding 2,4-D toxicity and mutagenicity. In the whole period analyzed, a strong positive correlation was observed between citations and publications (Pearson's correlation: $r = 0.876$; $r^2 = 0.767$; $t = 14,725$; $p < 0.001$).

Insert Fig. 1.

3.2 Countries

Information analysis based on the origin of the authors of the publications is fundamental to understand the geographic distribution of the regions and countries with the highest publication indices about the toxicity and mutation of 2,4-D. This analysis can be seen in Fig. 2A. The publication leader was the USA, followed by Canada, China and India. In Fig. 2B, information on the number of citations and cooperation network between countries is summarized, showing the U.S. as the country that stands out in studies involving 2,4-D toxicity. This figure also revealed the lack of cooperation between some countries. Therefore, there is a need to improve the lines of communication between research and publication in these countries. It is worth noting that the letter font size in Fig. 2B is proportional to the volume of publication, while the thickness of the connecting lines between countries demonstrates the intensity of cooperation. The thicker the connection line the higher the frequency of cooperation between two or more countries.

Insert Fig. 2

The formation of clusters of countries separated by different colors can be seen in Fig. 2B. The numbers represent the citation counts of each country. The clusters are from the largest (#0) to the second largest (#1) and so on. The top ranked item by citation counts is the U.S. in Cluster #1, with citation counts of 399. The 2nd is Canada in Cluster #0 (100). The 3rd is China in Cluster #0 (78). The 4th is Argentina in Cluster #2 (62). The 5th is India in Cluster #1 (56). The 6th is Turkey in Cluster #2 (43). The 7th is Japan in Cluster #0 (43). The 8th is Germany in cluster #0 (38). The 9th is Brazil in Cluster #2 (37). The 10th is France in Cluster #2 (30).

Fig. 2B also presents the centrality of some countries (yellow circles). Centrality is an important measure that varies from 0 to 1 (Chen, 2015), representing the influence of a country. In this study, the USA has the greatest centrality (0.68), followed by Germany (0.29), Canada (0.13) and Spain (0.12).

Moreover, some countries presented citation bursts. A citation burst is an indicator of a most active region (in this case), which can last for multiple years as well as a single year. A citation burst provides evidence that a publication/country is associated with a surge of citations. In other words, the item has evidently attracted an extraordinary degree of attention from its scientific community (Chen, 2015). The top-ranked item in bursts is China (2000), with bursts of 13.23. The 2nd was Turkey (1999) (7.93). The 3rd is Argentina (1991) (7.39). The 4th is the U.S. (1973) (7.34). The 5th is Germany (1991) (6.92). The 6th is Canada (1974) (6.25). The 7th is Japan (1997) (6.20). Lastly, the 8th is England (1998) (4.54).

3.3 Category analysis

Based on the analysis of occurrence of a subject category, it is possible to describe which area is highlighted in toxicity and mutation studies of 2,4-D. In this study, the most recurring areas for analysis of the characteristics of categories listed by WoS were chosen. Subject category information has been analyzed using CiteSpace. Fig. 3A shows the co-occurrence network, where the node represents a subject category, while a line connecting the two nodes demonstrates the co-occurrence of two subject categories. In this figure, the three main categories in relation to the frequency of this theme are “Toxicology” (citation counts: 514), “Environmental Sciences & Ecology” (506) and “Environmental Sciences” (489), followed by “Public, Environmental & Occupational Health” (160) and “Chemistry” (119).

Centrality is represented in Fig. 3A as purple rings and are evident in the areas of “Biochemistry & Molecular Biology” and “Toxicology” and “Chemistry”. A node of high betweenness centrality is usually one that connects two or more groups of nodes with the node itself in-between, hence the term betweenness (Chen, 2014). Fig. 3B emphasizes the main points seen in Fig. 3A, where the centrality and frequencies are related to the 10 largest categories.

Insert Fig. 3

In Fig. 3B, “Biochemistry & Molecular Biology” and “Toxicology” had the largest centralities (0.38 for both), indicating greater influence as regards the publications about mutation and toxicity of 2,4-D. “Chemistry” and “Environmental Sciences & Ecology” also stand out for their centralities (0.24 and 0.23, respectively). Thus, the research field of this theme is multidisciplinary, covering a wide range of interests. “Toxicology”, “Environmental Sciences & Ecology” and “Environmental Sciences” had the highest frequencies in descending order, indicating that these categories have greater visibility in the scientific environment (Chen, 2014). Therefore, the category with the highest visibility will not always have the greatest influence. This is the case of the "Biochemistry and Molecular Biology" area, not so frequent, but highly influent.

3.4 Sources co-citation analysis

Studies on the mutagenicity and toxicity of 2,4-D were published in 419 journals. Table 1 shows the top 10 most-cited journals in this study. The best-classified journal by citation count was Chemosphere, with 314 records, thus being considered the best source for scientists to develop and disseminate papers on the subject. The second best-ranked journal in citations (Journal of Agricultural and Food Chemistry) also stands out in centrality. Undoubtedly, at the Impact Factor (IF) level, Science had the highest (41.037) and a profound influence on research. Moreover, it is remarkable that Science had a citation half-life of 36 years, followed by Environmental Science & Technology (31 years), Journal of Agricultural and Food Chemistry (31) and Chemosphere (27 years).

Table 1. Top 10 journals in terms of frequency and Impact Factor IF (JCR) in 2018.

Journal	Frequency	Centrality	Half-life	Burst	Impact Factor
Chemosphere	314	0.05	27		5.108
Journal of Agricultural and Food Chemistry	295	0.19	31	9.79	3.571
Bulletin of Environmental Contamination and Toxicology	223	0.09	24	13.13	1.650
Environmental Health Perspectives	216	0.04	19	5.03	8.049
Environmental Science & Technology	213	0.06	32		7.149
Toxicology and Applied Pharmacology	211	0.17	28	23.37	3.585
Toxicology	195	0.07	22		3.547
Environmental Toxicology and Chemistry	194	0.04	19		3.421
Archives of Environmental Contamination and Toxicology	183	0.08	17	5.32	2.135
Science	183	0.06	36	8.42	41.037

Fig. 4 shows the main journals and citation burst detection, which is an indicator of a more active area. It is possible to note that the journal with the largest explosion of citations, represented by red rings, was "Toxicology and Applied Pharmacology", followed by "Bulletin of Environmental Contamination and Toxicology ", "Journal of

Agricultural and Food Chemistry", "Toxicology" and "Environmental Health Perspectives". A citation burst provides evidence that a particular source is associated with a rapid increase in citations, indicating that the journal has attracted a great deal of attention from its peers in a short time (Chen, 2014).

Insert Fig. 4.

3.5 Authors co-citation analysis

Fig. 5 shows the collaboration network for productive authors according to CiteSpace. In this network, each node represents an author, expressed "author's name". This co-citation analysis refers to the co-citation relationship between two documents from the list of references of a third cited document. In the co-citation network, the location of clusters and their relationship may reveal an intellectual structure that gives the reader a holistic view of the subject in question (Sun et al., 2019). Thus, the authors work collaboratively, using the available literature, and making evident the high productivity and cooperation between them, while making great advances in this field of research. It is important to highlight that the connection between the nodes represents a common reference and the links between two nodes suggest a collaboration between individual authors (Chen et al., 2009; Rorissa and Yuan, 2012).

Insert Fig. 5.

As seen in Fig. 5, the top-ranked item in citation counts and centrality was CHARLES JM (1996), with 71 in Cluster #0. The important article of Charles et al., (1996) evaluated the chronic dietary toxicity and oncogenicity of 2,4-D in rodents. The general findings of these studies indicate low chronic toxicity of 2,4-D and the lack of oncogenic response to 2,4-D following chronic dietary 2,4-D exposure in rats and mice.

The following important references were also listed in descending order of citation counts: Grover et al., (1986) in cluster #1, Munro et al., (1992) in Cluster #2, and Sauerhoff et al., (1977) in Cluster #1. The USEPA has had several related publications, one standing out in #3, and Bradford (1976), with its citation peak in 1993, in cluster #0. The research of Grover et al. evaluated the amount of 2,4-D metabolite in the urine of farmers and was in the same Sauerhoff et al. (1977) cluster. The research of Sauerhoff was remarkable because it evaluated the elimination of 2,4-D from plasma in five male human volunteers who ingested 2,4-D in a single dose (5 $mg.kg^{-1}$). Several publications dealing with the genotoxicity of 2,4-D have appeared in the literature and were reviewed by Munro et al., (1992). On the other hand, the research of Bradford is globally important in toxicology and several other areas, because it presented a rapid and sensitive method for the quantitation of microgram quantities of protein using the principle of protein-dye binding.

Regarding both authors and publications, Table 2, obtained from WoS, outlined the top 10 highly-cited papers in 2,4-D toxicity and mutagenicity studies. This table also shows the values of the total and annual citation counts of each article in addition to information about the authors and publication year. These documents are essential base readings for researchers seeking to work with 2,4-D toxicology. Several of these articles evaluated a mixture of pesticides, including 2,4-D. Several publications are about research with human beings.

Table 2 – top 10 most-cited publications about 2,4-D mutagenicity and toxicity.

Publication	Citation total	Average citation per year	Author/year
An ecologically effective water treatment technique using electrochemically generated hydroxyl radicals for *in situ* destruction of organic pollutants: Application to herbicide 2,4-D	284	14.05	Oturan, MA, 2000
Pesticide residues in urine of adults living in the United States: Reference range concentrations	215	8.60	Hill, RH; Head, SL; Baker, S; et al., 1995
A cocktail of contaminants: how mixtures of pesticides at low concentrations affect aquatic communities	214	19.45	Relyea, Rick A., 2009
Semen quality in relation to biomarkers of pesticide exposure	210	12.35	Swan, SH; Kruse, RL; Liu, F; et al., 2003
Aquatic phyto-toxicity of 23 pesticides applied at expected environmental concentrations	207	7.96	Peterson, Hg; Boutin, C; Martin, Pa; et al., 1994
A method for establishing acceptable toxicant limits for fish - malathion and butoxyethanol ester of 2,4-D	203	3.83	Mount, Di; Stephan, Ce, 1967
Aggregate exposures of nine preschool children to persistent organic pollutants at day care and at home	191	11.24	Wilson, NK; Chuang, JC; Lyu, C; et al., 2003
Non-Hodgkin's lymphoma and specific pesticide exposures in men: Cross-Canada study of pesticides and health	175	9.21	McDuffie, H H; Pahwa, P; McLaughlin, JR; et al., 2001
A review of 2,3,7,8-tetrachlorodibenzo-para-dioxin-induced changes in immunocompetence - 1991 update	175	6.03	Holsapple, Mp; Snyder, Nk; Wood, Sc; et al., 1991
Genotoxicity of select herbicides in *Rana catesbeiana* tadpoles using the alkaline single-cell gel DNA electrophoresis (comet) assay	166	7.22	Clements, C; Ralph, S; Petras, M., 1997

3.6 Keywords co-occurrence analysis

The keywords can be considered one of the most important points of a publication and co-words analysis can be used to detect research topics, analyze research hotspots, and monitor research frontier transitions from a certain knowledge domain (Yu et al., 2017).

In Fig. 6A, the main keywords were presented. The color represents temporality: blue for the first citations and red for the most recent. The words herbicide and pesticide appear because they are strongly linked to the subject under study. Words such as 2,4-D and dichlorophenoxyacetic acid were excluded from the results, since they are used to identify the pesticide.

Figure 6B shows the top 15 keywords with the frequency and centrality that appear in the studies. As mentioned above, the keyword that most frequently appears in the literature is herbicide and the most central word is pesticide, which means that this keyword has greater visibility in toxicology and mutagenicity connected to the 2,4-D area. There is a strong association of the subject in question with the keywords "water", "atrazine", and "glyphosate". Indeed, as a result of its intensive agricultural use, 2,4-D is recognized as one of the main pollutants of aquatic ecosystems (Atamaniuk et al., 2013). The estimated environmental concentrations of 2,4-D in freshwater bodies can range from 4 to 24 μg.L^{-1} and may locally reach concentrations up to 4,000 μg.L-1 (Borges et al., 2004).

Insert Fig. 6.

As regards freshwater bodies, different countries have different legislation determining the maximum quantity allowed. Table 3 shows the different levels of 2,4-D allowed in Brazil (Brasil., 2005), Argentina (Merini et al., 2007), the European Union (Nalcaci et al., 2006; Salman and Hameed, 2010), Canada (Health Canada, 2019), the

USA (USEPA, 2005) and by the WHO (World Health Organization, 2003). It should be noted that the lowest values are in Europe, where 0.1 µg L^{-1} is permitted in drinking water, while the sum of all molecules detected must not exceed 0.5 µg L^{-1}. The amount of 2,4-D permitted in drinking water is 1000 times greater in Canada and 700 times greater in the USA.

Table 3 –Maximum allowed 2,4-D limit around the world

Country	Maximum allowable quantity
European Union	0.1 µg L^{-1}
Argentina	4 µg L^{-1}
Brazil	30 µg L^{-1}
USA	70 µg L^{-1}
Canada	100 µg L^{-1}
World Health Organization (WHO)	30 µg L^{-1}

A word with strong centrality was *expression*, referring to genic expression. In this point, it is necessary to discuss a mechanist approach about 2,4-D analysis, its impact and remedial strategies. Several studies have evaluated the expression of genes of interest either in genetically modified plants or in non-target organisms, such as the research by Park and Kwak (2012), which evaluated the Metallothionein and Vitellogenin mRNA expressions in benthic *Chironomus riparius* larvae exposed to 2,4-D. We can cite also the study of Hattab et al., (2015) that investigated the transcriptional expression levels in the earthworm *Eisenia andrei* after exposure to 2,4-D. As outcome, they observed that, compared to expression in controls, the expression levels of the SOD, CAT, and GST genes increased in worms exposed to all three 2,4-D doses for 7 days, and after 14 days of exposure, only the expression of the GST gene remained higher than controls (Hattab et al., 2015). Studies in soil are important too, because the highest concentrations of 2,4-D were detected in soil, air and surface water surrounded by crop field (Islam et al., 2018). In the next topic it will be approached more about effects on individuals (plants and animal) and about remedial strategies.

3.7 Hot research topics on 2,4-D toxicity and mutagenicity: clusters

The "Keyword" node type in CiteSpace was selected, and the top 10 most frequently researched topics were obtained. Fig. 7 shows the main clusters labeled by the keywords indicated by the authors themselves. Table 4 shows the details of these clusters, where the size of the set is the number of terms included in each set. In summary, a cluster is a group of closely-coupled documents representing different directions of research. From the topics of this group of documents, representative terms were extracted and labeled by a log-likelihood ratio, expressed as "# + Cluster ID + representative term" (Chen et al., 2009; Rorissa and Yuan, 2012). To better understand the meaning and scope of each cluster, it is necessary to consider other factors in the cluster shown in Table 4, such as Silhouette, Size, and Year (mean), for example, in addition to their identification.

Insert Fig. 7.

Table 4. Summary of the 10 largest clusters, showing size, silhouette, and representative terms to characterize each cluster.

Cluster ID	Size	Silhouette	Mean (year)	Representatives terms
0	78	0.524	2007	Somatic Embryogenesis
1	74	0.734	2002	Occupational Exposure
2	68	0.596	2005	Degradation
3	65	0.657	2005	Transdermal
4	42	0.716	2000	Neurotoxicity
5	40	0.80	2013	Molecular Imprinting
6	34	0.843	2013	Equitoxic Mixtures
7	29	0.893	2013	Herbicide Resistance
8	24	0.908	2016	Herbicide Tolerance
9	12	0.958	2010	Goldfish
10	5	0.995	2017	Cross-resistance

The silhouettes in our clusters ranged from 0.524 to 0.995. The significant silhouette of scores is one of the most important metrics, impacting the overall structural properties of the network. The silhouette shows the homogeneity of a cluster ranging from -1 to 1 and the highest possible value 1 represents a perfect solution (Chen, 2006b). The higher the silhouette scores, the more consistent the cluster members are with each other (if the compared clusters are similar in size). A good range of cluster numbers would be about 7 ~ 10 major clusters with 10 or more members, with each of the clusters having high silhouette values (e.g. > 0.70) (Chen, 2015). All our 10 clusters had silhouettes above 0.5, and most were around 0.8. The mean publication year of a cluster indicates whether it is mostly made up of recent or old articles. This is a simple and useful indicator (Chen, 2015).

We can see that the keywords in the Fig. 6 and 7 and Table 4, also are represented in the highly cited literature (Table 2). The first most cited article studied methods of oxidative degradation of 2,4-D (Oturan, 2000). 'Degradation' is an important keyword in centrality, as well as 'pesticide'. Several studies investigate more than one pesticide and the 2,4-D is frequently associated, e.g., with 'glyphosate' and 'atrazine' – two important other keywords. It is corroborated by one of the most frequently cited studies entitled "A cocktail of contaminants: how mixtures of pesticides at low concentrations affect aquatic communities" (Relyea, 2009), and by Peterson et al. (1994) in "Aquatic phyto-toxicity of 23 pesticides applied at expected environmental concentrations". The keyword 'water' is important too, and well represented in the top cited literature. The study of Relyea (2009) evaluated aquatic comunities; Clements et al. (1997) evaluated the genotoxicity of selected herbicides in the tadpoles *Rana catesbeiana*; and Mount and Stephan (1967) evaluated a new method for establishing acceptable toxicant limits for Fish to 2,4-D and Malathion. *Genotoxicity* and *Fish* were keyword present in the figure 6 and 7. *Occupational exposure* was an highlighted trend, as seen in the 4th most cited Swan et al. (2003) and in the 8th (Mcduffie et al., 2001).

As can be seen in Fig. 7 and Table 4, the major objectives of the investigations involving 2,4-D toxicity and mutagenicity are related to the environment and studies on the toxic potential of this herbicide in non-target organisms. The representative term was "Somatic Embryogenesis" with the largest size, but with the lowest silhouette, showing a weak network structure. Despite the data refinement, it is impossible to deny that 2,4-D is classically known to induce somatic embryogenesis in routine protocols of seed cultures, alone or with other hormones (Raghavan, 2004). Other terms like Occupational Exposure, Transdermal (penetration), Neurotoxicity, Molecular Imprinting, Goldfish, Herbicide Resistance and Herbicide Tolerance attract attention.

Occupational exposure to pesticides is a major public health concern (Islam et al., 2018). Oxidative stress induced by this herbicide may be important in the pathogenesis of cancer and other chronic disease outcomes, especially in occupational exposure (Lerro et al., 2018). According to Stackelberg, (2014), epidemiologic studies suggest that exposure to 2,4-D may be associated with increased risk of non-Hodgkin's lymphoma, Hodgkin's disease, leukemia, and soft-tissue sarcoma, as well as Parkinson's disease (Tanner et al., 2009).

Neurotoxicity was an important keyword in the clusters view. A USEPA study (RED, 2006) demonstrated that clinical signs of neurotoxicity (ataxia, decreased motor activity, myotonia, prostration, lateral recumbency, impaired/loss of the righting reflex, and skin cold to the touch) were observed in pregnant rabbits following exposure to 2,4-D (in relatively high dose) and its amine salts and esters. Neuropathology (retinal degeneration) was observed following 2,4-D exposure in several studies of female rats (RED, 2006). A developmental neurotoxicity study of rats was required for 2,4-D (USEPA, 2,4-D RED Facts, 2005) and more recently Marty et al., (2013) assessed F1 rats for systemic toxicity, reproductive toxicity, developmental neurotoxicity (DNT), developmental immunotoxicity (DIT), and endocrine toxicity. In this study, the authors presented that 2,4-D did not cause reproductive toxicity or developmental neurotoxicity after oral exposure.

Institutes such as WHO (World Health Organization) defend that "all people, whatever their stage of development and their social and economic conditions, have the right to have access to an adequate supply of safe drinking water" (World Health Organization, 2003). Thus, to ensure water safety, a trend in the search for new technologies for analytical detection of pesticides is molecular imprinting (Cluster #5). Jia et al., (2017), for example, described a novel molecular imprinting fluorescence sensor construction by anchoring mesoporous structured imprinting microspheres on the surfaces of quantum dots (QDs) for the selective and sensitive detection of 2,4-D.

There is no doubt that modern agricultural techniques are based in biotechnologies and agrochemicals. With the cultivation of 2,4-D GM-resistant crops, the 2,4-D use will be inevitably increased in the next years. In this sense, intending to reduce the environmental impacts and seeking remedial strategies, the most recent papers about 2,4-D are researching more efficient and complete degradation methods for this pesticide. It can be cited Cai et al., (2020) that investigate the influencing factors and mechanism in degradation of 2,4-D by anodic oxidation and electro-Fenton using BDD anode. Other recent article investigated the degradation of 2,4-D by photoelectrochemical methods using electrochemically self-doped Blue TiO_2 nanotube arrays with formic acid as electrolyte (Xu et al., 2020). Likewise, Li et al., (2019), used CuO-Co_3O_4@CeO_2 nanoparticles and get 100% of 2,4-D (20 mg/L) decomposed. It is promising and very important line of research.

When non-target organisms are exposed to 2,4-D during or after application, they may trigger stress responses (Liu et al., 2011; Subrahmaniyan et al., 2008). The 2,4-D concentrations in the environment may be well above the lethal concentration for numerous aquatic species (e.g. fish), especially when these applications are made directly (Browne and Moore, 2014). Some of these investigations were done with goldfish *Carassius auratus* L. (Clusters #9) (ex. Kubrak et al., 2013; Matviishyn et al., 2014). Various fish species also stand out as bioindicators of the effects of exposure to 2,4-D (e.g. rainbow trout, bluegill, striped bass, banded killifish, white perch, eel, and carp) (RED, 2006). The fathead minnows (*Pimephales promelas*) (Dehnert et al., 2018) are included in this group.

In this sense of evaluation of the impacts on non-target organisms, there are articles that investigate the substances that can increase the transdermal absorption of the 2,4-D (Clusters #3). Researches investigated the mechanisms by which substances as alcohol, topical sunscreen application and moisturizing lotions can increase transdermal absorption (Brand et al., 2007a, 2007b, 2007c). Brand et al., (2007c) compared the 2,4-D transdermal absorption after exposure to both ethanol and sunscreen and demonstrates that these two treatments enhance additively at the higher doses tested. As the organisms are exposed to a mixture of xenobiotics, studies as theses are important to show the synergic effect the pollutants.

Lastly, the words resistance and tolerance were highlighted in three keywords that labeled the clusters: Herbicide Resistance, Herbicide Tolerance, Cross-resistance. The rapid evolution of weeds resistant to the herbicide glyphosate required the development of GMO technologies resistant to another herbicide, in this case, 2,4-D. The commercialization of these technologies led to a greater consumption of this herbicide, which, since 2001 in the USA, has been the most commonly used herbicide in the residential market, and went from the seventh to the fifth most commonly used herbicide in the agricultural market from 2007 to 2012 (https://www.epa.gov/pesticides/pesticides-industry-sales-and-usage-2008-2012-market-estimates) (Stackelberg, 2014; USEPA, 2012). Continued and increasing use of 2,4-D in conventional agriculture is inevitable, and research efforts have therefore been made to evaluate the potential toxicological effects of this substance on human health and ecosystems (Egan and Mortensen, 2012).

4. CONCLUSION

This study provides a comprehensive review of the mutagenicity and toxicity of 2,4-D by means of scientometric methods, which clarified the emerging trends. Research on 2,4-D toxicology and mutagenicity has advanced rapidly and the United States has maintained the leading research position with the largest production, number of citations and centrality. On the other hand, the Chinese citation burst was remarkable. “Toxicology”, “Environmental Sciences & Ecology” and “Environmental Sciences” are three main frequency categories. “Biochemistry & Molecular Biology” had the same centrality as “Toxicology”, indicating that they are the areas with the largest influence. Chemosphere is the most published source, probably the best source for scientists to disseminate papers on the studied subject. In relation to the principal articles, several of them are evaluations of human beings. The others also evaluated a mixture of pesticides, including 2,4-D. Studies of rats, fish or amphibians had great prominence as well. The keywords co-occurrence analysis and the cluster view point in the same direction. 2,4-D toxicity research is remarkably associated with words such as glyphosate and atrazine (other pesticides), degradation, molecular imprinting, water, goldfish, rat, neurotoxicity, occupational exposure (in humans), genetic expression, herbicide resistance/tolerance. Therefore, future research on 2,4-D toxicology and mutagenicity should probably focus on molecular biology, especially gene expression, assessment of exposure in human or other vertebrate bioindicators, and pesticide degradation studies. In summary, this scientometric analysis allowed us to make inferences about global trends, in order to identify tendencies and gaps in 2,4-D toxicology and mutagenicity and thus contribute to future research efforts.

References

Aquino, A.J.A., Tunega, D., Haberhauer, G., Gerzabek, M.H., Lischka, H., 2007. Interaction of the 2,4-dichlorophenoxyacetic acid herbicide with soil organic matter moieties: A theoretical study. Eur. J. Soil Sci. 58, 889–899. doi:10.1111/j.1365-2389.2007.00928.x

Atamaniuk, T.M., Kubrak, O.I., Storey, K.B., Lushchak, V.I., 2013. Oxidative stress as a mechanism for toxicity of 2,4-dichlorophenoxyacetic acid (2,4-D): Studies with goldfish gills. Ecotoxicology 22, 1498–1508. doi:10.1007/s10646-013-1136-z

Borges, S., Dzubow, C., Orrick, G., Stavola, A., 2004. 2,4-Dichlorophenoxyacetic Acid Analysis of Risks to Endangered and Threatened Salmon and Steelhead.

Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem. 72, 248–254. doi:10.1016/0003-2697(76)90527-3

Brand, R.M., Charron, A.R., Sandler, V.L., Jendrzejewski, J.L., 2007a. MOISTURIZING LOTIONS CAN INCREASE TRANSDERMAL ABSORPTION OF THE HERBICIDE 2 , 4-DICHLOROPHENOXACETIC ACID ACROSS HAIRLESS MOUSE SKIN. Cutan. Ocul. Toxicol. 15–23. doi:10.1080/15569520601182791

Brand, R.M., Jendrzejewski, J.L., Charron, A.R., 2007b. Potential mechanisms by which a single drink of alcohol can increase transdermal absorption of topically applied chemicals. Toxicology 235, 141–149. doi:10.1016/j.tox.2007.03.008

Brand, R.M., Mcmahon, L., Jendrzejewski, J.L., Charron, A.R., 2007c. Transdermal absorption of the herbicide 2 , 4-dichlorophenoxyacetic acid is enhanced by both ethanol consumption and sunscreen application 45, 93–97. doi:10.1016/j.fct.2006.08.005

Brasil., 2005. Ministério do Meio Ambiente. Resolução n. 357. 23p.

Browne, A.M., Moore, P.A., 2014. The effects of sublethal levels of 2,4-dichlorophenoxyacetic acid herbicide (2,4-D) on feeding behaviors of the crayfish O. rusticus. Arch. Environ. Contam. Toxicol. 67, 234–244. doi:10.1007/s00244-014-0032-8

Cai, J., Zhou, M., Pan, Y., Lu, X., 2020. Degradation of 2,4-dichlorophenoxyacetic acid by anodic oxidation and electro-Fenton using BDD anode: influencing factors and mechanism. Sep. Purif. Technol. in proof, 115867. doi:10.1016/j.seppur.2019.115867

Charles, J.M., Bond, D.M., Jeffries, T.K., Yano, B., Stott, W.T., Johnson, K.A., Cunny, H.C., Wilson, R.D., S.Busb, J., 1996. Chronic Dietary Toxicity / Oncogenicity Studies on 2 , 4-Dichlorophenoxyacetic Acid in Rodents. Fundam. Appl. Toxicol. 33, 166–172. doi:https://doi.org/10.1006/faat.1996.0154

Chen, C., 2015. The CiteSpace Manual, http://cluster.ischool.drexel.edu/~cchen/citespace/CiteSpaceManual.pdf. doi:10.1007/s11192-015-1576-8

Chen, C., 2014. The CiteSpace Manual, College of Computing and Informatics Drexel University. doi:10.1007/s11192-015-1576-8

Chen, C., 2006a. CiteSpace II: Detecting and Visualizing Emerging Trends and Transient Patterns in Scientific Literature. J. Am. Soc. Inf. Sci. Technol. 3, 359–377. doi:10.1002/asi

Journal Pre-proof

Chen, C., 2006b. CiteSpace II : Detecting and Visualizing Emerging Trends. J. Am. Soc. Inf. Sci. Technol. 57, 359–377. doi:10.1002/asi

Chen, C., 2004. Searching for intellectual turning points: Progressive knowledge domain visualization. Proc. Natl. Acad. Sci. 101, 5303–5310. doi:10.1073/pnas.0307513100

Chen, C., Chen, Y., Horowitz, M., Hou, H., Liu, Z., Pellegrino, D., 2009. Towards an explanatory and computational theory of scientific discovery. J. Informetr. 3, 191–209. doi:10.1016/j.joi.2009.03.004

Chinalia, F.A., Killham, K.S., 2006. 2,4-Dichlorophenoxyacetic acid (2,4-D) biodegradation in river sediments of Northeast-Scotland and its effect on the microbial communities (PLFA and DGGE). Chemosphere 64, 1675–1683. doi:10.1016/j.chemosphere.2006.01.022

Clements, C., Ralph, S., Petras, M., 1997. Genotoxicity of select herbicides in Rana catesbeiana tadpoles using the alkaline single-cell gel DNA electrophoresis (Comet) assay. Environ. Mol. Mutagen. 29, 277–288. doi:10.1002/(SICI)1098-2280(1997)29:3<277::AID-EM8>3.0.CO;2-9

da Fonseca, M.B., Glusczak, L., Silveira Moraes, B., de Menezes, C.C., Pretto, A., Tierno, M.A., Zanella, R., Gonçalves, F.F., Lúcia Loro, V., 2008. The 2,4-D herbicide effects on acetylcholinesterase activity and metabolic parameters of piava freshwater fish (Leporinus obtusidens). Ecotoxicol. Environ. Saf. 69, 416–420. doi:10.1016/j.ecoenv.2007.08.006

Dehnert, G.K., Freitas, M.B., Dequattro, Z.A., Barry, T., Karasov, W.H., 2018. Effects of Low , Subchronic Exposure of 2 , 4- Formulations on Early Life Stages of Fathead Minnows (Pimephales promelas) 37, 2550–2559. doi:10.1002/etc.4209

Egan, J.F., Mortensen, D.A., 2012. Quantifying vapor drift of dicamba herbicides applied to soybean. Environ. Toxicol. Chem. 31, 1023–1031. doi:10.1002/etc.1778

Freydier, L., Lundgren, J.G., 2016. Unintended effects of the herbicides 2,4-D and dicamba on lady beetles. Ecotoxicology 25, 1270–1277. doi:10.1007/s10646-016-1680-4

Gaultier, J., Farenhorst, A., Cathcart, J., Goddard, T., 2008. Degradation of [carboxyl-14C] 2,4-D and [ring-U-14C] 2,4-D in 114 agricultural soils as affected by soil organic carbon content. Soil Biol. Biochem. 40, 217–227. doi:10.1016/j.soilbio.2007.08.003

Goodman, J.E., Loftus, C.T., Zu, K., 2015. 2,4-Dichlorophenoxyacetic acid and non-Hodgkin's lymphoma, gastric cancer, and prostate cancer: Meta-analyses of the published literature. Ann. Epidemiol. 25, 626-636.e4. doi:10.1016/j.annepidem.2015.04.002

Grover, R., Franklin, C.A., Muir, N.I., Cessna, A.J., Riedel, D., 1986. Dermal exposure and urinary metabolite excretion in farmers repeatedly exposed to 2,4-D amine. Toxicol. Lett. 33, 73–83.

Guerrero-Estévez, S.M., López-López, E., 2016. Effects of endocrine disruptors on reproduction in viviparous teleosts with intraluminal gestation. Rev. Fish Biol. Fish. 26, 563–587. doi:10.1007/s11160-016-9443-0

Hattab, S., Boughattas, I., Boussetta, H., Viarengo, A., Banni, M., Sforzini, S., 2015. Transcriptional expression levels and biochemical markers of oxidative stress in the earthworm Eisenia andrei after exposure to 2,4-dichlorophenoxyacetic acid (2,4-D). Ecotoxicol. Environ. Saf. 122, 76–82. doi:10.1016/j.ecoenv.2015.07.014

He, K., Zhang, J., Zeng, Y., 2019. Knowledge domain and emerging trends of agricultural waste management in the fi eld of social science : A scientometric review. Sci. Total Environ. 670, 236–244. doi:10.1016/j.scitotenv.2019.03.184

Health Canada, 2019. Guidelines for Canadian Drinking Water Quality—Summary Table. Water and Air Quality Bureau, Healthy Environments and Consumer Safety Branch. Ottawa, Ontario.

Hirsch, J.E., 2005. An index to quantify an individual ' s scientific research output 102, 16569–16572.

IBAMA, I.B. do M.A., 2018. Os 10 ingredientes ativos mais vendidos - 2017 - Consolidação de dados fornecidos pelas empresas registrantes de produtos técnicos, agrotóxicos e afins, conforme art. 41 do Decreto n° 4.074/2002.

Islam, F., Farooq, M.A., Gill, R.A., Wang, J., Yang, C., Ali, B., Wang, G.X., Zhou, W., 2017. 2,4-D attenuates salinity-induced toxicity by mediating anatomical changes, antioxidant capacity and cation transporters in the roots of rice cultivars. Sci. Rep. 7, 1–23. doi:10.1038/s41598-017-09708-x

Islam, F., Wang, J., Farooq, M.A., Khan, M.S.S., Xu, L., Zhu, J., Zhao, M., Muños, S., Li, Q.X., Zhou, W., 2018. Potential impact of the herbicide 2,4-dichlorophenoxyacetic acid on human and ecosystems. Environ. Int. 111, 332–351. doi:10.1016/j.envint.2017.10.020

Jia, M., Zhang, Z., Li, J., Yang, X., Chen, L., 2017. A molecular imprinting fluorescence sensor based on quantum dots and a mesoporous structure for selective and sensitive detection of 2,4-dichlorophenoxyacetic acid. Sensors Actuators, B Chem. 252, 934–943. doi:10.1016/j.snb.2017.06.090

Kearns, J.P., Wellborn, L.S., Summers, R.S., Knappe, D.R.U., 2014. 2,4-D adsorption to biochars: Effect of preparation conditions on equilibrium adsorption capacity and comparison with commercial activated carbon literature data. Water Res. 62, 20–28. doi:10.1016/j.watres.2014.05.023

Kiljanek, T., Niewiadowska, A., Semeniuk, S., Gaweł, M., Borzęcka, M., Posyniak, A.,

2016. Multi-residue method for the determination of pesticides and pesticide metabolites in honeybees by liquid and gas chromatography coupled with tandem mass spectrometry—Honeybee poisoning incidents. J. Chromatogr. A 1435, 100–114. doi:10.1016/j.chroma.2016.01.045

Konur, O., 2011. The scientometric evaluation of the research on the biofuels. J. Orthop. Surg. 7, 890–898.

Kubrak, O.I., Atamaniuk, T.M., Husak, V. V, Lushchak, V.I., 2013. on some metabolic and free radical processes in goldfish white muscle. Food Chem. Toxicol. 59, 356–361. doi:10.1016/j.fct.2013.06.023

Lajmanovich, R.C., Attademo, A.M., Simoniello, M.F., Poletta, G.L., Junges, C.M., Peltzer, P.M., Grenón, P., Cabagna-Zenklusen, M.C., 2015. Harmful Effects of the Dermal Intake of Commercial Formulations Containing Chlorpyrifos, 2,4-D, and Glyphosate on the Common Toad Rhinella arenarum (Anura: Bufonidae). Water. Air. Soil Pollut. 226. doi:10.1007/s11270-015-2695-9

Lerro, C.C., Freeman, L.E.B., Portengen, L., Kang, D., Lee, K., Blair, A., Lynch, C.F., Bakke, B., Roos, A.J. De, Vermeulen, R.C.H., 2018. oxidative stress markers among Iowa corn farmers 58, 30–38. doi:10.1002/em.22069.A

Li, M., Porter, A.L., Lin, Z., 2017. Nano Energy Evolutionary trend analysis of nanogenerator research based on a novel perspective of phased bibliographic coupling. Nano Energy 34, 93–102. doi:10.1016/j.nanoen.2017.02.020

Li, W., Li, Y., Zhang, D., Lan, Y., Guo, J., 2019. CuO-Co3O4@CeO2 as a heterogeneous catalyst for efficient degradation of 2,4-dichlorophenoxyacetic acid by peroxymonosulfate. J. Hazard. Mater. 121209. doi:10.1016/j.jhazmat.2019.121209

Liu, F., Jin, Z.L., Naeem, M.S., Tian, T., Zhang, F., He, Y., Fang, H., Ye, Q.F., Zhou, W.J., 2011. Applying Near-Infrared Spectroscopy and Chemometrics to Determine Total Amino Acids in Herbicide-Stressed Oilseed Rape Leaves. Food Bioprocess Technol. 4, 1314–1321. doi:10.1007/s11947-010-0445-y

Liu, W., Li, H., Tao, F., Li, S., Tian, Z., Xie, H., 2013. Formation and contamination of PCDD/Fs, PCBs, PeCBz, HxCBz and polychlorophenols in the production of 2,4-D products. Chemosphere 92, 304–308. doi:10.1016/j.chemosphere.2013.03.031

Loomis, D., Guyton, K., Grosse, Y., El Ghissasi, F., Bouvard, V., Benbrahim-Tallaa, L., Guha, N., Mattock, H., Straif, K., 2015. Carcinogenicity of lindane, DDT, and 2,4-dichlorophenoxyacetic acid. Lancet. Oncol. 16, 891–892. doi:10.1016/S1470-2045(15)00081-9

Marty, M.S., Neal, B.H., Zablotny, C.L., Yano, B.L., Andrus, A.K., Woolhiser, M.R., Boverhof, D.R., Saghir, S.A., Perala, A.W., Passage, J.K., Lawson, M.A., Bus, J.S., 2013. An F1-Extended One-Generation Reproductive Toxicity Study in Crl : CD (SD) An F 1 -Extended One-Generation Reproductive Toxicity Study in Crl : CD (SD) Rats With 2 , 4-Dichlorophenoxyacetic Acid. Toxicol. Sci. 136, 527–547. doi:10.1093/toxsci/kft213

Matviishyn, T.M., Kubrak, O.I., Husak, V. V., Storey, K.B., Lushchak, V.I., 2014. Tissue-specific induction of oxidative stress in goldfish by 2,4-dichlorophenoxyacetic acid: Mild in brain and moderate in liver and kidney. Environ. Toxicol. Pharmacol. 37, 861–869. doi:10.1016/j.etap.2014.02.007

Mcduffie, H.H., Pahwa, P., Mclaughlin, J.R., Spinelli, J.J., Fincham, S., Dosman, J.A., Robson, D., Skinnider, L.F., Choi, N.W., 2001. Non-Hodgkin ' s Lymphoma and Specific Pesticide Exposures in Men : Cross-Canada Study of Pesticides and Health 1. Cancer Epidemiol. Biomarkers Prev. 10, 1155–1163.

Merini, L.J., Cuadrado, V., Flocco, C.G., Giulietti, A.M., 2007. Dissipation of 2,4-D in soils of the Humid Pampa region, Argentina: A microcosm study. Chemosphere 68, 259–265. doi:10.1016/j.chemosphere.2007.01.012

Mount, D.I., Stephan, C.E., 1967. A Method for Establishing Acceptable Toxicant Limits for Fish — Malathion and the Butoxyethanol Ester of 2 , 4-D. Trans. Am. Fish. Soc. 96, 186–193. doi:10.1577/1548-8659(1967)96

Munro, I.C., Carlo, G.L., Orr, J.C., Sund, K.G., Wilson, R.M., Kennepohl, E., Lynch, B.S., Jablinske, M., 1992. A Comprehensive, Integrated Review and Evaluation of the Scientific Evidence Relating to the Safety of the Herbicide 2,4-D. J. Am. Coll. Toxicol. 11, 559–664. doi:10.3109/10915819209141893

Nalcaci, O.O., Sirin, S., Ovez, B., 2006. Toxicity determination of various phenoxyalkanoic acid herbicides using cress seed in phosphate contaminated aqueous media. WIT Trans. Ecol. Environ. 95, 301–307. doi:10.2495/WP060311

Oturan, M.A., 2000. An ecologically effective water treatment technique using electrochemically generated hydroxyl radicals for in situ destruction of organic pollutants : Application to herbicide 2 , 4- ... An ecologically e □ ective water treatment technique using electroc. J. Appl. Electrochem. 475±482. doi:10.1023/A

Ouyang, W., Wang, Y., Lin, C., He, M., Hao, F., Liu, H., Zhu, W., 2018. Heavy metal loss from agricultural watershed to aquatic system : A scientometrics review. Sci. Total Environ. 637–638, 208–220. doi:10.1016/j.scitotenv.2018.04.434

Park, K., Kwak, I., 2012. Assessment of Potential Biomarkers , Metallothionein and Vitellogenin mRNA Expressions in Various Chemically Exposed Benthic Chironomus riparius Larvae. Ocean Sci. J. 47, 435–444.

Pattanasupong, A., Nagase, H., Inoue, M., Hirata, K., Tani, K., Nasu, M., Miyamoto, K., Science, E., 2004. Ability of a microbial consortium to remove pesticide , carbendazim and. J. Microbiol. 517–522.

Peterson, H.G., Boutin, C., Martin, P.A., Freemark, K.E., Ruecker, N.J., Moody, M.J., 1994. Aquatic phyto-toxicity of 23 pesticides applied at expected environmental concentrations. Aquat. Toxicol. 28, 275–292.

Raghavan, V., 2004. ROLE OF 2,4-DICHLOROPHENOXYACETIC ACID (2,4-D) IN SOMATIC EMBRYOGENESIS ON CULTURED ZYGOTIC EMBRYOS OF ARABIDOPSIS: CELL EXPANSION, CELL CYCLING, AND MORPHOGENESIS DURING CONTINUOUS EXPOSURE OF EMBRYOS TO 2,4-D. Am. J. Bot. 91, 1743–1756. doi:10.3732/ajb.91.11.1743

RED, 2006. Reregistration Eligibility Decision (RED) 2,4-D. United States Environ. Prot. Agency. doi:EPA738-R-98-010

Relyea, R.A., 2009. A cocktail of contaminants : how mixtures of pesticides at low concentrations a V ect aquatic communities. Oecologia 159, 363–376. doi:10.1007/s00442-008-1213-9

Román, G.C., 2007. Autism: Transient in utero hypothyroxinemia related to maternal flavonoid ingestion during pregnancy and to other environmental antithyroid agents. J. Neurol. Sci. 262, 15–26. doi:10.1016/j.jns.2007.06.023

Rorissa, A., Yuan, X., 2012. Visualizing and mapping the intellectual structure of information retrieval. Inf. Process. Manag. 48, 120–135. doi:10.1016/j.ipm.2011.03.004

Salman, J.M., Hameed, B.H., 2010. Adsorption of 2,4-dichlorophenoxyacetic acid and carbofuran pesticides onto granular activated carbon. Desalination 256, 129–135. doi:10.1016/j.desal.2010.02.002

Sauerhoff, M.W., Braun, W.H., Blau, G.E., Gehring, P.J., 1977. The Fate of 2,4-Dichlorophenoxyacetic Acid (2,4-D) Following Oral Administration to Man. Toxicology 8, 3–11.

Schuermans, N., Meeus, B., De Maesschalck, F., 2010. Is there a world beyond the Web of Science? Publication practices outside the heartland of academic geography. Area 42, 417–424. doi:10.1111/j.1475-4762.2010.00938.x

Shareef, K., Shaw, G., 2008. Sorption kinetics of 2,4-D and carbaryl in selected agricultural soils of northern Iraq: Application of a dual-rate model. Chemosphere 72, 8–15. doi:10.1016/j.chemosphere.2008.02.056

Smith, A.M., Smith, M.T., La Merrill, M.A., Liaw, J., Steinmaus, C., 2017. 2,4-dichlorophenoxyacetic acid (2,4-D) and risk of non-Hodgkin lymphoma: a meta-analysis accounting for exposure levels. Ann. Epidemiol. 27, 281-289.e4. doi:10.1016/j.annepidem.2017.03.003

Song, Y., 2014. Insight into the mode of action of 2,4-dichlorophenoxyacetic acid (2,4-D) as an herbicide. J. Integr. Plant Biol. 56, 106–113. doi:10.1111/jipb.12131

Stackelberg, K. Von, 2014. A Systematic Review of Carcinogenic Outcomes and Potential Mechanisms from Exposure to 2 , 4-D and MCPA in the Environment. J. Toxicol. doi:10.1155/2013/371610

StatSoft, I., 2007. STATISTICA (data analysis software system), version 8.0.

Subrahmaniyan, K., Kalaiselvan, P., Balasubramanian, T.N., Zhou, W., 2008. Soil properties and yield of groundnut associated with herbicides, plant geometry, and plastic mulch. Commun. Soil Sci. Plant Anal. 39, 1206–1234. doi:10.1080/00103620801925919

Sun, Y., Wu, S., Gong, G., 2019. Trends in Food Science & Technology Trends of research on polycyclic aromatic hydrocarbons in food : A 20-year perspective from 1997 to 2017. Trends Food Sci. Technol. 83, 86–98. doi:10.1016/j.tifs.2018.11.015

Suzuki, J.B., 2017. ROTULAGEM DE TRANSGÊNICOS NO BRASIL : O RETROCESSO DO PL N ° 95–123.

Swan, S.H., Kruse, R.L., Liu, F., Barr, D.B., Drobnis, E.Z., Redmon, J.B., Wang, C., Brazil, C., Overstreet, J.W., 2003. Semen Quality in Relation to Biomarkers of Pesticide Exposure. Environ. Health Perspect. 1478–1484. doi:10.1289/ehp.6417

Tanner, C.M., Ross, G.W., Jewell, S.A., Hauser, R.A., Jankovic, J., Factor, S.A., Bressman, S., Deligtisch, A., Marras, C., Lyons, K.E., Bhudhikanok, G.S., Roucoux, D.F., Meng, C., Abbott, R.D., Langston, J.W., 2009. Occupation and Risk of Parkinsonism. Arch. Neurol. 66, 1106–1113. doi:10.1001/archneurol.2009.195

USEPA, U.S.E.P.A., 2012. Pesticides Industry Sales and Usage.

USEPA, U.S.E.P.A., 2005. Reregistration Eligibility Decision for 2,4-D.

Walters, J., 2011. Environmental Fate of 2 , 4-Dichlorophenoxyacetic Acid. Environ. Monit. Pest Manag. 3510, 1–18.

World Health Organization, 2003. 2,4-D in Drinking-water:Background document for development of WHO Guidelines for Drinking-water Quality. 2,4-D Drink. doi:(WHO/SDE/WSH/03.04/24)

Xiang, C., Wang, Y., Liu, H., 2017. A scientometrics review on nonpoint source pollution research. Ecol. Eng. 99, 400–408.

Xu, X., Cai, J., Zhou, M., Du, X., Zhang, Y., 2020. Photoelectrochemical degradation of 2 , 4-dichlorophenoxyacetic acid using electrochemically self-doped Blue TiO 2 nanotube arrays with formic acid as electrolyte. J. Hazard. Mater. 382, 121096. doi:10.1016/j.jhazmat.2019.121096

Yu, D., Xu, Z., Pedrycz, W., Wang, W., 2017. Information sciences 1968–2016: A retrospective analysis with text mining and bibliometric. Inf. Sci. (Ny). 418–419, 619–634. doi:10.1016/j.ins.2017.08.031

Figure Captions

Fig. 1. Publication *vs.* Citation output performance between 1949 and 2018.

Fig. 2. A. Geolocalization of scientific papers published in peer-reviewed journals about toxicity and mutagenicity of 2,4-D. The geographic distribution of the reviewed papers is related to the color of each country, proportional to the publication number. B. The cooperation network of the productive countries. Different colors mean different clusters, and the numbers represent the citation counts. Yellow circles represent the proportional centrality.

Fig. 3. A. Subject category network between 1949 and 2018, with a threshold of 20 citations. The purple rings represent centrality and the different colored lines mean different clusters. B. Top 10 categories in terms of frequency and centrality.

Fig. 4. The network organized by the cited journal. Note: Red circles represent the citation burst.

Fig. 5. The co-authorship analysis.

Fig. 6. A. The network organized by the cited keyword. B.Top 10 keywords in terms of frequency and centrality.

Fig. 7. Main clusters labeled by keyword in the field 2,4-D toxicity and mutagenicity.

Ministério da Educação
Universidade Tecnológica Federal do Paraná
Câmpus Dois Vizinhos

Tecnológica há mais de 100 anos

HIGHLIGHTS

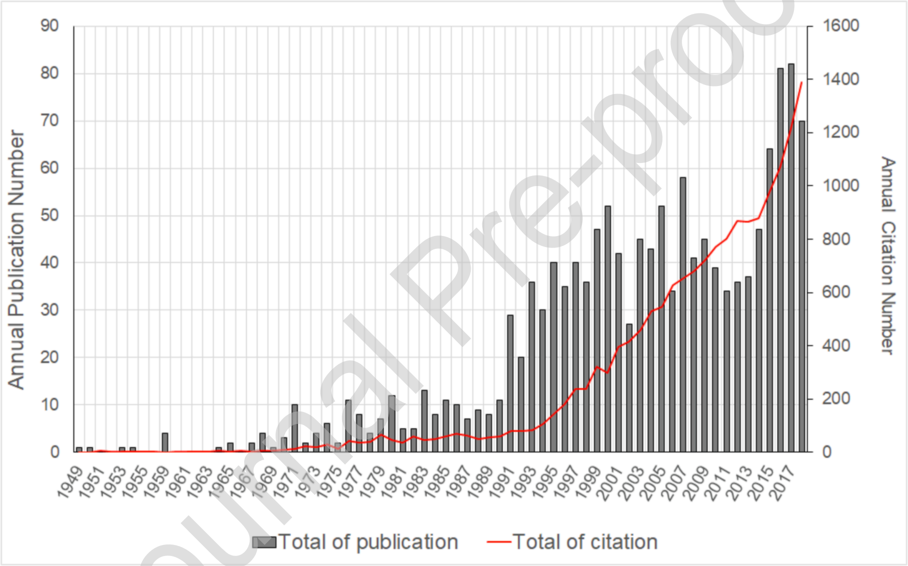
Dois Vizinhos 2019, September

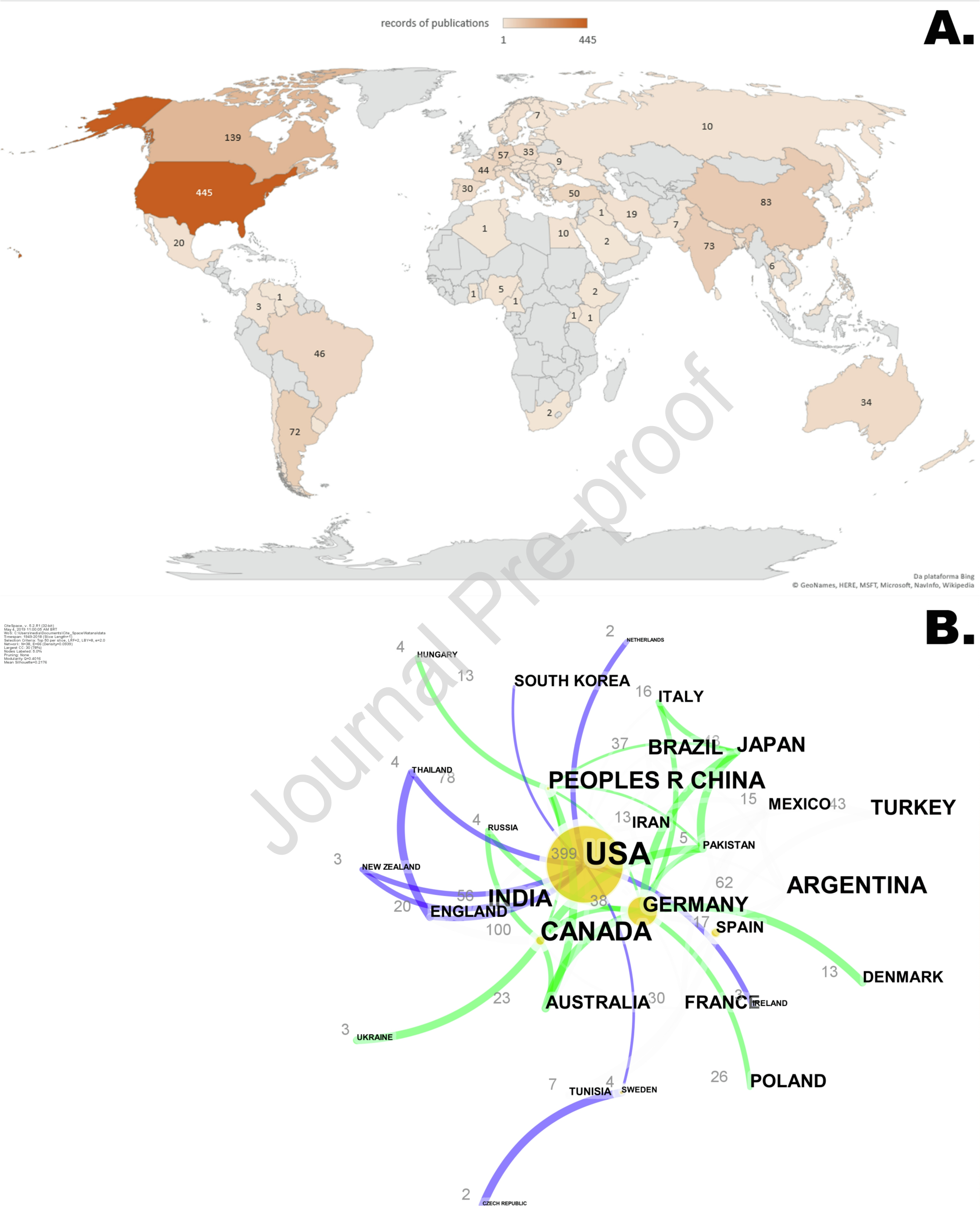
Manuscript **"Analysis of Global Trends and Gaps for Studies about 2,4-D Herbicide Toxicity: A scientometric review"**
Authors: Natana Raquel Zuanazzi, Nédia de Castilhos Ghisi, Elton Celton de Oliveira

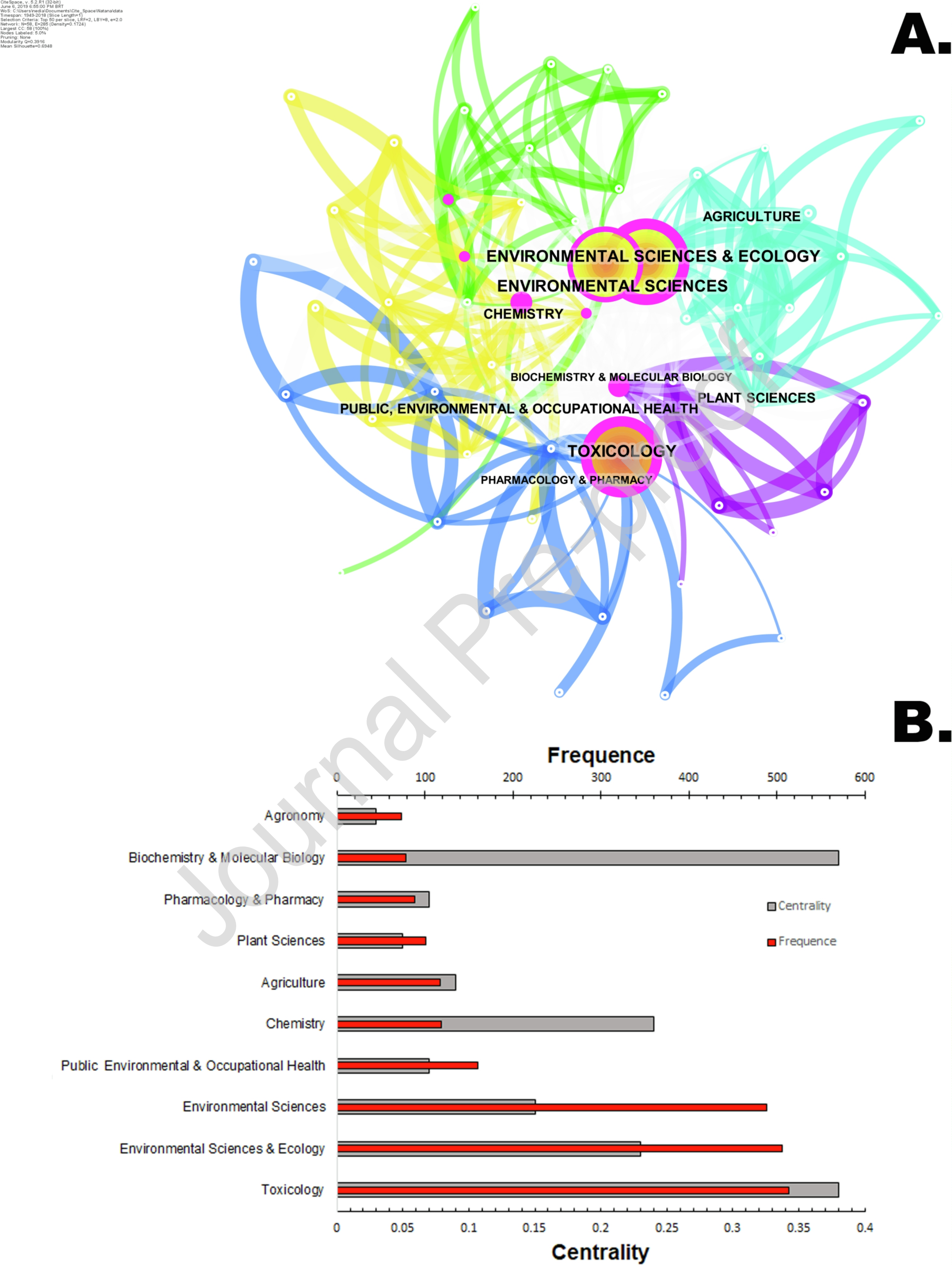
BULLET POINTS (3 to 5 bullet points - maximum 85 characters)

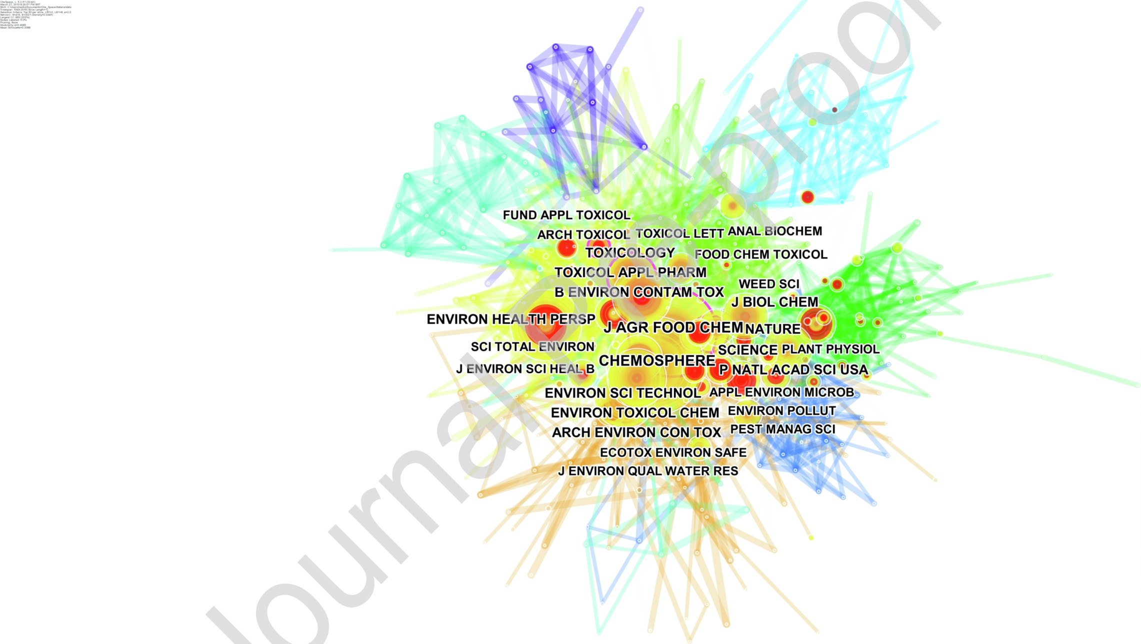
- Research on 2,4-D toxicology and mutagenicity has advanced rapidly.
- USA stands out in studies involving 2,4-D toxicity, followed by Canada and China
- Toxicology and Biochemical/Molecular Biology were the areas of greatest influence
- strongly related to the keywords glyphosate, atrazine, water and gene expression
- Focus on occupational risk, degradation, neurotoxicity and herbicide tolerance

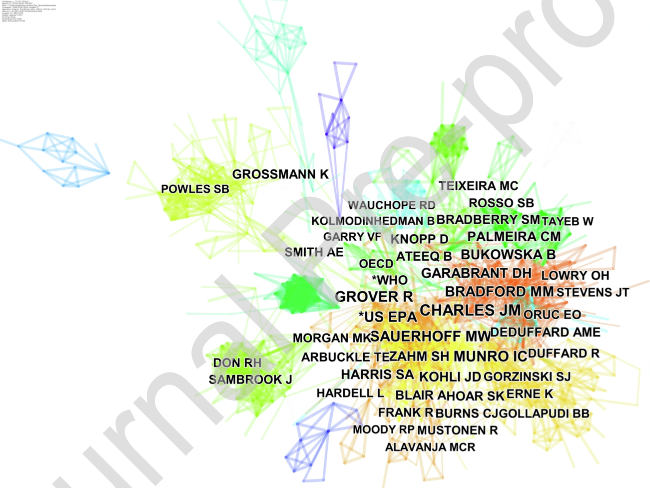
Nédia de Castilhos Ghisi
Corresponding Author
Federal University of Technology - Paraná

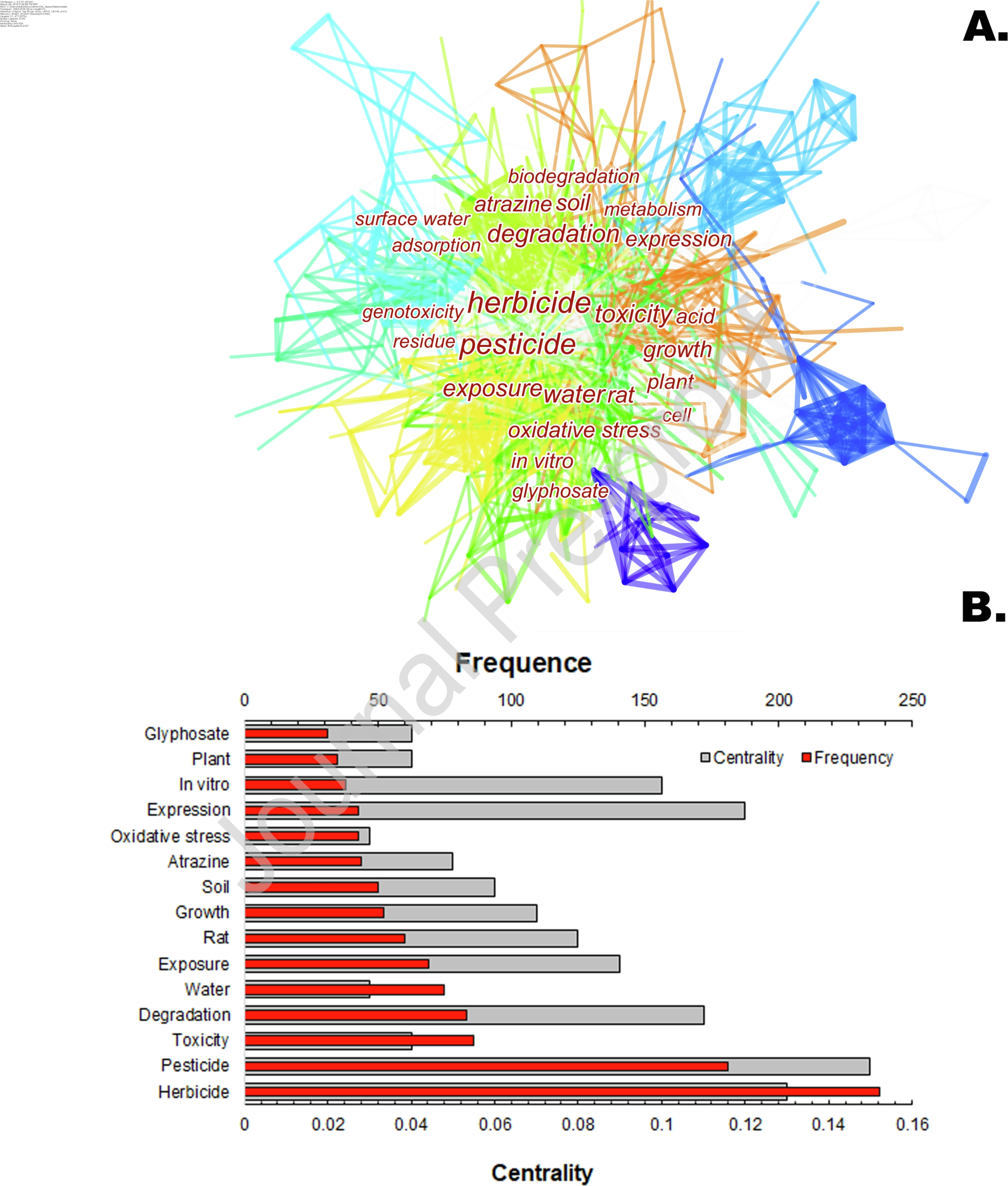

Annual Publication Number
90
80
70
60
50
40
30
20
10
0
Annual Citation Number
1600
1400
1200
1000
800
600
400
200
0
1949
1951
1953
1955
1959
1961
1963
1965
1967
1969
1971
1973
1975
1977
1979
1981
1983
1985
1987
1989
1991
1993
1995
1997
1999
2001
2003
2005
2007
2009
2011
2013
2015
2017
Total of publication
Total of citation

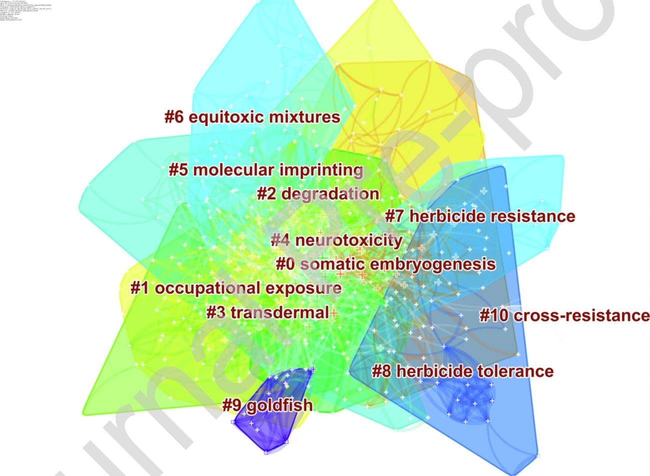

A.
records of publications
1
445
139
445
20
7
57
33
9
44
30
50
10
1
19
7
83
1
10
2
73
6
1
5
1
2
1
1
3
1
46
2
34
72
Da plataforma Bing
© GeoNames, HERE, MSFT, Microsoft, NavInfo, Wikipedia
B.
2
NETHERLANDS
4
HUNGARY
13
SOUTH KOREA
16
ITALY
37
BRAZIL
JAPAN
4
THAILAND
78
PEOPLES R CHINA
15
MEXICO
43
TURKEY
4
RUSSIA
13
IRAN
5
PAKISTAN
3
NEW ZEALAND
399
USA
62
ARGENTINA
56
INDIA
20
ENGLAND
38
GERMANY
17
SPAIN
100
CANADA
13
DENMARK
23
AUSTRALIA
30
FRANCE
IRELAND
3
UKRAINE
26
POLAND
7
TUNISIA
4
SWEDEN
2
CZECH REPUBLIC


CiteSpace, v. 5.2.R1 (32-bit)
June 6, 2019 6:55:00 PM BRT
WoS: C:\Users\nedia\Documents\Cite_Space\Natana\data
Timespan: 1949-2018 (Slice Length=1)
Selection Criteria: Top 50 per slice, LRF=2, LBY=8, e=2.0
Network: N=58, E=285 (Density=0.1724)
Largest CC: 58 (100%)
Nodes Labeled: 5.0%
Pruning: None
Modularity Q=0.3916
Mean Silhouette=0.6948
A.
AGRICULTURE
ENVIRONMENTAL SCIENCES & ECOLOGY
ENVIRONMENTAL SCIENCES
CHEMISTRY
BIOCHEMISTRY & MOLECULAR BIOLOGY
PLANT SCIENCES
PUBLIC, ENVIRONMENTAL & OCCUPATIONAL HEALTH
TOXICOLOGY
PHARMACOLOGY & PHARMACY
B.
Frequence
0
100
200
300
400
500
600
Agronomy
Biochemistry & Molecular Biology
Pharmacology & Pharmacy
Plant Sciences
Agriculture
Chemistry
Public Environmental & Occupational Health
Environmental Sciences
Environmental Sciences & Ecology
Toxicology
Centrality
Frequence
0
0.05
0.1
0.15
0.2
0.25
0.3
0.35
0.4
Centrality


FUND APPL TOXICOL
ARCH TOXICOL
TOXICOL LETT
ANAL BIOCHEM
TOXICOLOGY
FOOD CHEM TOXICOL
TOXICOL APPL PHARM
WEED SCI
B ENVIRON CONTAM TOX
J BIOL CHEM
ENVIRON HEALTH PERSP
J AGR FOOD CHEM
NATURE
SCI TOTAL ENVIRON
SCIENCE
PLANT PHYSIOL
CHEMOSPHERE
J ENVIRON SCI HEAL B
P NATL ACAD SCI USA
ENVIRON SCI TECHNOL
APPL ENVIRON MICROB
ENVIRON TOXICOL CHEM
ENVIRON POLLUT
ARCH ENVIRON CON TOX
PEST MANAG SCI
ECOTOX ENVIRON SAFE
J ENVIRON QUAL
WATER RES


GROSSMANN K
POWLES SB
TEIXEIRA MC
WAUCHOPE RD
ROSSO SB
KOLMODINHEDMAN B
BRADBERRY SM
TAYEB W
GARRY VF
KNOPP D
PALMEIRA CM
SMITH AE
ATEEQ B
BUKOWSKA B
OECD
GARABRANT DH
LOWRY OH
*WHO
BRADFORD MM
STEVENS JT
GROVER R
CHARLES JM
ORUC EO
*US EPA
SAUERHOFF MW
DEDUFFARD AME
MORGAN MK
ARBUCKLE TE
ZAHM SH
MUNRO IC
DUFFARD R
DON RH
HARRIS SA
KOHLI JD
GORZINSKI SJ
SAMBROOK J
HARDELL L
BLAIR A
HOAR SK
ERNE K
FRANK R
BURNS CJ
GOLLAPUDI BB
MOODY RP
MUSTONEN R
ALAVANJA MCR


A.
biodegradation
atrazine
soil
metabolism
surface water
degradation
expression
adsorption
genotoxicity
herbicide
toxicity
acid
residue
pesticide
growth
plant
exposure
water
rat
cell
oxidative stress
in vitro
glyphosate
B.
Frequence
0
50
100
150
200
250
Glyphosate
Plant
In vitro
Expression
Oxidative stress
Atrazine
Soil
Growth
Rat
Exposure
Water
Degradation
Toxicity
Pesticide
Herbicide
Centrality
Frequency
0
0.02
0.04
0.06
0.08
0.1
0.12
0.14
0.16
Centrality


#6 equitoxic mixtures
#5 molecular imprinting
#2 degradation
#7 herbicide resistance
#4 neurotoxicity
#0 somatic embryogenesis
#1 occupational exposure
#3 transdermal
#10 cross-resistance
#8 herbicide tolerance
#9 goldfish