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Botanical extracts with biostimulant potential for agriculture in Mexico: a review

Extractos botánicos con potencial bioestimulante para la agricultura en México: una revisión

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ABSTRACT

The main challenge facing the agricultural sector today is to ensure continued access to enough nutritious food with minimal environmental impact. Therefore, the search for sustainable and effective methods to stimulate crop yield and quality is a priority. Plants are emerging as an attractive local resource for biostimulant extracts due to their abundant biomass and high content of bioactive compounds. However, the factors that support the implementation of botanical extracts as alternative bioproducts to reduce dependence on agrochemicals are not yet fully understood. This review aimed to analyze the available information on the potential use of botanical extracts as biostimulants for Mexican agriculture. An updated overview of the definition and regulation of agricultural biostimulants worldwide and in Mexico is provided. In addition, research trends on the biostimulant properties of extracts derived from Mexican plants related to the species used as raw material, extraction methods, modes of application, and types of crops evaluated are analyzed. Finally, the effects and mechanisms of action of botanical extracts on crop plants are discussed, and future research needs to optimize their application in agricultural production.

Keywords: Sustainable agriculture, plant extracts, phenolic compounds, biostimulant regulation, abiotic stress tolerance.

RESUMEN

El reto principal del sector agrícola actualmente es garantizar el acceso constante a cantidades suficientes de alimentos nutritivos con un impacto ambiental mínimo. Por ende, la búsqueda de métodos sostenibles y eficaces para estimular el rendimiento y calidad de las cosechas representa una prioridad. Las plantas son recursos locales atractivos para la obtención de extractos bioestimulantes debido a su disponibilidad de biomasa y alto contenido en compuestos bioactivos. Sin embargo, la comprensión de los factores que sustentan la aplicación de los extractos botánicos como bioproductos alternativos para reducir la dependencia de los agroquímicos es aún limitada. El objetivo de esta revisión es analizar la información disponible sobre el uso potencial de los extractos botánicos como bioestimulantes para la agricultura mexicana. Se ofrece una visión general y actualizada de la definición y regulación de los bioestimulantes agrícolas en el mundo y en México. Además, se analizan las tendencias de la investigación sobre las propiedades bioestimulantes de los extractos derivados de plantas mexicanas relacionadas con las especies empleadas como materia prima, los métodos de extracción, las vías de aplicación y los tipos de cultivos evaluados. Finalmente, se discuten los efectos y mecanismos de acción de los extractos botánicos en los cultivos, así como las necesidades futuras de investigación para optimizar su implementación en la producción agrícola.

Palabras clave: Agricultura sostenible, extractos de plantas, compuestos fenólicos, regulación de bioestimulantes, tolerancia a estrés abiótico.

INTRODUCTION

The production and productivity of the Mexican agricultural sector depend, among other factors, on the use of agrochemicals such as fertilizers and pesticides (DataMéxico, 2025). According to the 2019 National Agricultural Survey (ENA) 67.4 % of the sector's production units used chemical fertilizers, and the remaining 24.4 % used natural fertilizers (INEGI, 2019). In Mexico, as in the rest of the world, the use of fertilizers is mainly concentrated in nitrogenous ones (65.3 %), followed by phosphorous fertilizers (16.1 %) and potassium fertilizers (18.6 %) (Banco de México, 2022).

For decades, the dependence on synthetic chemical inputs has significantly boosted food production. However, the inadequate application of these products has also generated harmful effects on human health and agroecosystems due to their high toxicity, rapid mobility in the soil, and high residuality in harvested products (Rouphael and Colla, 2020; Carvalho *et al.*, 2021). Therefore, one of the fundamental challenges currently facing the agricultural sector is the implementation of new and efficient strategies that promote the sustainability of agroproductive systems (Ali *et al.*, 2021; Martínez-Lorente *et al.*, 2024). Also, the accelerated growth of the population has promoted the need to enhance the development and yield of crops to meet consumer demands in terms of quantity and quality (Zulfiqar *et al.*, 2020; Akter *et al.*, 2024).

In the search for sustainable solutions in agriculture, natural products derived from renewable raw materials

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emerge as a promising alternative (Jasso de Rodríguez *et al*., 2020; Godlewska *et al*., 2021). Plant biostimulants comprise substances, compounds, and growth-promoting microorganisms that regulate or enhance plant-soil system processes (Calvo *et al*., 2014; Rouphael and Colla, 2020; Sanjuán *et al*., 2023). Nevertheless, despite the remarkable advances in biostimulant science in recent years, their definition, classification, and regulation continue to evolve. The importance of these bioproducts lies in their ability to enhance nutrient use efficiency and promote plant vigor, independently of their nutrient content and without directly affecting pest or disease control (Yakhin *et al*., 2017; Corsi *et al*., 2022). Therefore, the integration of biostimulant products into agricultural production systems has been demonstrated to promote desirable agronomic traits and help reduce the use of agrochemical inputs (Caradonia *et al*., 2019; Han *et al*., 2024).

However, to generalize the implementation of biostimulants in agriculture, it is important to consider the availability and accessibility of raw materials for processing (Xu and Geelen, 2018). In this context, there is growing interest in exploring the potential of botanical extracts as sources for novel biostimulant formulations. This interest stems from their high content of bioactive compounds, including polyphenols, phytohormones, organic acids, sterols, and terpenoids, as well as their availability from diverse and abundant raw materials, including weeds, invasive species, and agro-industrial byproducts (Godlewska *et al*., 2021; Han *et al*., 2024). Moreover, the cost-effectiveness of their production by the same manufacturer underscores their economic viability (Ahmad *et al*., 2022; Velasco-Ramírez *et al*., 2022b).

Due to the novelty of this topic, understanding the factors that underpin the use of botanical extracts as alternative bioproducts, to improve agricultural production and reduce dependence on agrochemicals, is still limited. The main objective of the current review was to analyze the available scientific information on the potential use of botanical extracts as biostimulants for Mexican agriculture. In particular, this review provides an updated overview of the definition and regulation of agricultural biostimulants worldwide and in Mexico. Furthermore, it examines current research trends on botanical extracts, focusing on the species used as raw materials, extraction methods, modes of application, and the types of crops evaluated. Finally, the effects and mechanisms of action of botanical extracts on plants are discussed, along with future research needs to optimize their application in agricultural production.

Global and Mexican context of plant biostimulants

Although interest in biostimulants in agriculture began in the second half of the twentieth century, in recent years it has experience exponential growth, consolidating itself as an innovative tool to improve the production and productivity of crops (Ahmad *et al*., 2022; Corsi *et al*., 2022; Han *et al*., 2024). Throughout history, plant biostimulants have received various names in the scientific literature, such as biogenic stimulants, plant strengtheners, metabolic enhancers, phytostimulants, positive plant growth regulators, growth promoters and biofertilizers, among others (Yakhin *et al*., 2017). However, there is no single, universally accepted definition of the term "plant biostimulant" (Sanjuán *et al*., 2023).

This lack of consensus is due to multiple factors, among which the following stand out: (1) heterogeneity in their sources of production, elaboration processes, and composition; (2) the difficulty in elucidating its mode of action; (3) similarities with other categories of agricultural products regulated under existing regulations; (4) the variability in regulatory frameworks between countries and regions; (5) the commercial interests of large companies that dominate the agrochemical market and; (6) the progressive advancement of scientific knowledge about the nature of its effects on the different components of the plant-soil systems (Yakhin *et al*., 2017; Corsi *et al*., 2022; Sanjuán *et al*., 2024).

Despite this context, the academic community agrees that biostimulants are substances and microorganisms that, when applied at low concentrations to plants, seeds, or the rhizosphere, stimulate natural processes that improve or benefit nutrient absorption, nutritional efficiency, tolerance to abiotic stress, growth, productivity, and/or crop quality regardless of their nutrient content (du Jardin, 2015; Yakhin *et al*., 2017; Zulfiqar *et al*., 2019; Han *et al*., 2024; Sanjuán *et al*., 2024). By extension, products containing mixtures of such substances and/or microorganisms are also recognized as plant biostimulants (du Jardin, 2015).

One of the most supported premises is that the recognition of a product as a plant biostimulant should focus on its agricultural functions, rather than on the nature of its components or its modes of action, as these can originate from multiple sources (Ricci *et al*., 2019; Sanjuán *et al*., 2024). These include humic substances, animal and plant protein hydrolysates, algae and plant extracts, biochemicals, beneficial microorganisms, and agroindustrial residues (Yakhin *et al*., 2017). More recently, a trend has been to classify this wide diversity into microbial and non-microbial biostimulants (Corsi *et al*., 2022).

Commercial biostimulants may include compounds not naturally present in the feedstock (Yakhin *et al*., 2017; Sanjuán *et al*., 2024). Adding to the lack of a clear definition and consistent criteria that distinguish biostimulants, this situation generates confusion with other agricultural formulations such as fertilizers and growth regulators (Corsi *et al*., 2022). To address this problem, it is essential to establish a regulatory framework that guarantees the quality, efficacy, and safety of biostimulants at the national and regional levels (Yakhin *et al*., 2017; Ricci *et al*., 2019).

In this sense, the European Union established the "Regulation (EU) 2019/1009 of the European Parliament and the Council", which is the first legislative text that provides a legal framework for the marketing of biostimulant products (European Union, 2019). On the other hand, in the United States, despite significant progress in defining and proposing a bill with the backing of regulatory agencies and industry, there is still no federal or state regulation for these products

(GPO, 2023). There are no specific legal provisions for plant biostimulants in other countries such as India, South Africa, Canada, and Brazil. Instead, these products are indirectly regulated under national laws that address other agricultural inputs (Caradonia *et al*., 2019).

In Mexico, despite the wide availability of both national and imported biostimulants in the agricultural market, current legislation lacks specific regulations for their production, labeling, and commercialization. In the absence of a legal definition of "plant biostimulant", they are intended to be regulated under other categories recognized in the Norma Oficial Mexicana NOM-077-FITO-2000, which establishes the specifications, criteria, and procedures to regulate studies of the biological effectiveness of plant nutrition inputs for agricultural use (DOF, 2020). Biological effectiveness results from applying a plant nutrition input, which can be said to have a biostimulant effect if it improves nutritional uptake efficiency, abiotic stress tolerance, and/or crop quality (DOF, 2020). These inputs are highly diverse and include:

Inoculant: Plant nutrition input made from microorganisms that, when applied, favors the use of nutrients in association with the plant or its rhizosphere.

Organic soil improvers: plant nutrition inputs made from organic products or microorganisms that favorably modify the physical, chemical, or biological conditions of the soil for the best plant development.

Type 1 growth regulators: Growth regulators are based on substances found naturally in plant tissues, whether obtained by extraction, fermentation, synthesis, or other methods. These types of regulators are also known as phytohormones or plant hormones.

Type 2 growth regulators: Products based on substances obtained by synthesis and not found naturally in the plant.

However, these definitions do not fully capture the diverse and multifunctional nature of plant biostimulants. In addition, when analyzing these definitions, it can be observed that establishing an exclusive regulation for biostimulants would be complicated due to the overlaps in their composition and modes of action with some existing categories. For example, botanical or algae extracts are complex mixtures of different bioactive compounds, including plant hormones and their precursors or intermediates. This raises the question of whether they should be classified as type 1 growth regulators since they contain phytohormones, or if they should be considered biostimulants as they include substances other than phytohormones. Therefore, fostering a clear and accurate understanding of plant biostimulants is crucial to promote their adoption as sustainable alternatives to agrochemicals. Addressing this challenge requires coordinated efforts among academic institutions, industry stakeholders, and regulatory agencies to update and harmonize existing legislation.

Analysis of research on the use of botanical extracts as biostimulants in México

In México, just over 25,000 species of vascular plants have been documented, representing approximately 9.1 % of all known species worldwide. These figures position the country among the five nations with the most incredible wealth of vascular plants (SEMARNAT, 2012). Therefore, one of the promising strategies to promote economic and environmental sustainability in Mexican agriculture is using local plant species as a source of bioactive compounds with biostimulant activity.

Evolution of scientific production

From 2008 to date, 15 original articles by Mexican authors have been published evaluating the use of botanical extracts as plant biostimulants. Of the total scientific production analyzed, 73 % corresponds to the years 2022 (14 %), 2023 (36 %) and 2024 (27 %). In contrast, between 2008 and 2021, the number of publications on this topic remained constant (Figure 1). The increase observed in recent years reflects an increased visibility of research that uses extracts obtained from plants as a source of biologically active substances to enhance crop growth, development, and productivity.

This growing interest in the topic can be attributed to the remarkable global boom in plant biostimulants over the past decade, which have established themselves as innovative alternatives to synthetic chemical fertilizers to address the challenges of sustainable agriculture (Zulfiqar et *al*., 2019; Ahmad *et al*., 2022; Han *et al*., 2024). Mexico is among the 10 countries with the highest production of scientific articles regarding the benefits of the use of biostimulants in agricultural production systems (Corsi *et al*., 2022), with special emphasis on research related to botanical extracts (Carvalho *et al*., 2022).

Also, nine scientific journals were identified that published articles included in this review, predominantly those edited by Mexican institutions. Among the Mexican journals, *Biotecnia* and *Ecosistemas y Recursos Agropecuarios* stands out for concentrating 53 % of the scientific production on the potential of plant extracts as biostimulants in agriculture. Both journals are included in the Emerging Sources Citation Index (ESCI), which is part of the primary collection of the Web of Science, and the SCIELO Citation Index, another database integrated into this same scientific information platform.

Plants as raw material for biostimulant extracts

The research analyzed highlights the biostimulant properties of extracts from 22 plant species originating from diverse habitats, including semiarid zones, disturbed vegetation, halophytic grasslands, and freshwater ecosystems. These species represent approximately 0.1 % of the vascular plant diversity reported in Mexico (SEMARNAT, 2012). The analyzed species belong to 14 botanical families, with Asteraceae and Anacardaceae being the most represented, accounting for 23 % and 18 % of the total species, respectively (Figure 2A).

On the other hand, *Rhus muelleri* Standl. and F.A. Barkley is positioned as the most researched species as a source of extracts with biostimulant properties in agriculture, representing 27 % of the articles analyzed (Figure 2B). Other prominent species include *Flourensia microphylla* S.F. Blake,

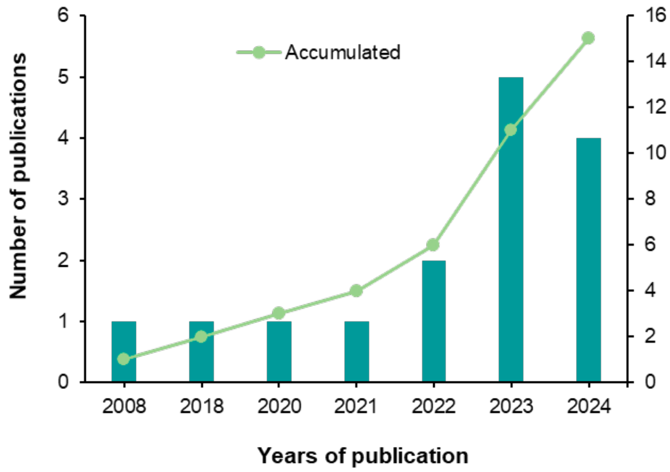


Figure 1. Articles published by Mexican authors on the use of botanical extracts as biostimulants for agriculture. Data for the first months of 2025 are not represented.
Figura 1. Artículos publicados por autores mexicanos sobre el uso de extractos botánicos como bioestimulantes en agricultura. Datos para los primeros meses de 2025 no están representados.

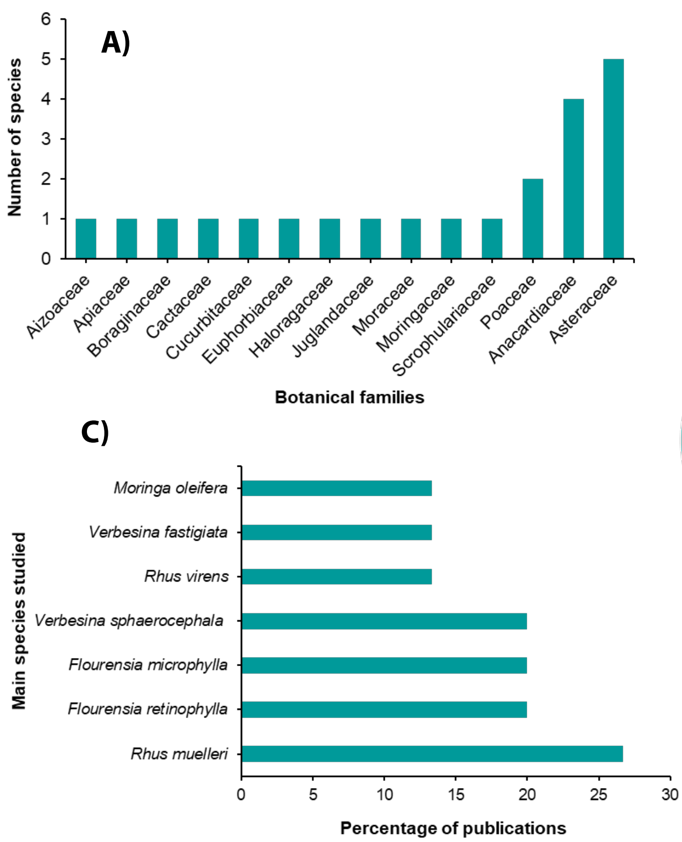
Flourensia retinophylla S.F. Blake and *Verbesina sphaerocephala* A. Gray ex S.Wats., which were the subject of study in 20 % of the studies (Figure 2B). *Rhus virens* Lindh. ex A. Gray and *Moringa oleifera* Lam. were represented in 13 % of the studies (Figure 2B). The remaining set of species assessed includes *Cenchrus echinatus* L., *Distichlis spicata* L., *Rhus trilobata* Nutt., *Amphipterygium glaucum* (Hemsl. & Rose) Hemsl. & Rose ex Standl., *Cucurbita foetidissima* Kunth, *Bacopa monnieri* (L.) Wettst, *Heliotropium curassavicum* L., *Flaveria trinervia* (Spreng.) C. Mohr, *Sesuvium verrucosum* Raf., *Maclura tinctoria* (L.) D. Don ex Steud, *Myriophyllum aquaticum* (Vell.) Verdc., *Jatropha dioica* Cerv., *Juglans mollis* Engelm., *Opuntia ficus-indica* (L.) Mill., and *Coriandrum sativum* L. Each species represents 5 % of the studies included in this work.

The biogeographical status of the plant species reveals that 59 % are native and 27 % are endemic to Mexico (DOF, 2010) (Figure 2C). The utilization of local flora as a source of bioactive extracts underscores the value of national resources, and opens new possibilities for discovery of compounds that stimulate plant growth (Godlewska *et al.*, 2021). In addition, using weeds as raw material for obtaining biostimulant extracts, as in the case of *C. echinatus*, could represent a promising strategy (Huez-Lopez *et al.*, 2008; Leopardi-Verde *et al.*, 2021).

In contrast, 14 % of the species evaluated are exotic (Figure 2C), that is, those not native to Mexico and have been introduced intentionally or accidentally. Among these, *M. aquaticum* is classified as an invasive species (Bonilla-Barbosa and Santamaría, 2013), whose benefits in horticultural production were documented by Viveros-Legorreta *et al.* (2018).

Methods of preparation

Botanical extracts are complex mixtures of primary and secondary metabolites whose biostimulant properties depend on various factors (Godlewska *et al.*, 2021). One of the key



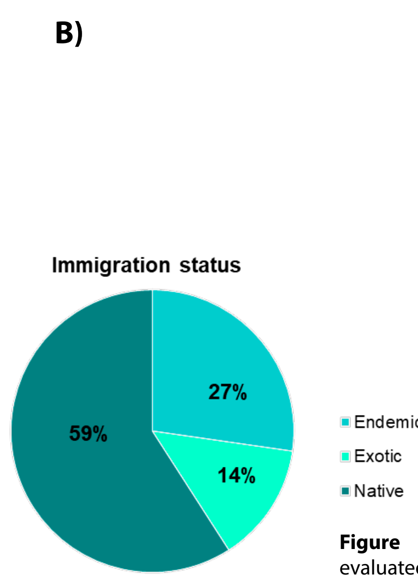


Figure 2. (A) Number of species evaluated by botanical family, (B) Main species evaluated as a source of biostimulant extracts, and (C) Percentage of the species studied according to their biogeographical status in México.
Figura 2. (A) Número de especies evaluadas por familia botánica, (B) Principales especies evaluadas como fuente de extractos bioestimulantes y (C) Porcentaje de especies estudiadas de acuerdo a su estado migratorio en México.

aspects is the selection of the plant biomass source used as a feedstock, which largely determines the chemical composition of the resulting extract (Zulfiqar *et al*., 2019; Ahmad *et al*., 2022). As shown in Table 1, the extracts evaluated in different studies as stimulants of crop growth and development are obtained from both whole plants and specific parts, including leaves, cladodes, or fruits. In addition, agricultural residues, often underutilized, represent valuable sources for obtaining plant-based biostimulants (Huez-López *et al*., 2008).

Once the plant material has been selected, several biomass pretreatment steps must be carried out before extracting the bioactive compounds from the plant tissues (Godlewska *et al*., 2021). The first step is to wash the collected material several times with tap water to remove surface impurities (Ahmad *et al*., 2022). The biomass intended for extraction can then be used in its fresh state or subjected to drying processes (Table 1). These processes can be carried out at room temperature (~26 °C) in the laboratory (Velasco-Ramírez *et al*., 2021; 2022a) or at low temperatures (-80 °C) by freeze-drying to preserve the properties of the compounds present in the extract (e.g., polyphenols and flavonoids) (Garza-García *et al*., 2023). The dried plant material is then crushed and screened to obtain a homogeneous sample and increase the surface-to-volume ratio (Michalak and Chojnacka, 2014). In most studies, the crushing stage is not described in detail, although it is usually done with the help of an electric mill or mixer.

Several methods exist to obtain biostimulant extracts from plant biomass (Godlewska *et al*., 2021; Han *et al*., 2024). However, the method and solvent selection are essential to ensure a high yield of the bioactive compounds of interest and preserve their integrity and properties (Michalak and Chojnacka, 2014; Ahmad *et al*., 2022). In addition, identifying an economical, simple, effective, and environmentally friendly method for preparing biostimulants based on plant extracts is crucial to scaling up the process (Michalak and Chojnacka, 2014). The research analyzed in this study describes methods for extracting plant biomass that use various approaches (Table 1).

The 54 % of the studies used physical methods for the extraction of bioactive compounds, such as maceration, autoclaving, or ultrasound, which facilitates the release of these from plant tissue (Velasco-Ramírez *et al*., 2022a; Garza-García *et al*., 2023; Moreno-Guerrero *et al*., 2024; Velazquez-Gamboa *et al*., 2024). The choice of solvent is crucial for extraction efficiency (Michalak and Chojnacka, 2014). The 38 % of the research used organic solvents such as methanol, ethanol, and acetone, which selectively dissolve the compounds of interest (Jasso de Rodríguez *et al*., 2020; Brazales-Cevallos *et al*., 2022). The evaporation of these solvents ensures that the final extract is safe for crops application (Godlewska *et al*., 2021; Ahmad *et al*., 2022).

In addition to the methods previously described, 15 % of the studies included in this work employed biological processes such as fermentation to obtain plant extracts with biostimulant activity. This method consists in inoculating fresh plant material with natural sources of microorganisms, such as manure, which are responsible for carrying out the fermentation process under anaerobic conditions (Huez-López *et al*., 2008; Alcalán-López *et al*., 2023). Microbial activity facilitates the release of bioactive compounds in the plant matrix and improves its chemical and functional properties, increasing its effectiveness in agricultural applications (Pérez-Alva *et al*., 2022).

In general, water-based extraction methods are the most common to produce botanical extracts for use in agriculture (Table 1). These extractions are simple and relatively inexpensive and dispense with the use of organic solvents, reducing toxicity and residue generation while improving crop yield and quality (Godlewska *et al*., 2021; Velasco-Ramírez *et al*., 2021). However, using water as a solvent for specific compound extractions has disadvantages, as its low selectivity and extraction efficiency reduce the yield per unit of biomass used as feedstock (Michalak and Chojnacka, 2014).

Application into plants and seeds

The application method of botanical extracts determines their effectiveness as agricultural biostimulants (Han *et al*.,

Table 1. Summary of the methods for obtaining plant extracts as biostimulants.
Tabla 1. Resumen de métodos para la obtención de extractos de plantas como bioestimulantes.

Type of extraction	Method	Solvent	Plant material	Biomass pretreatment	References
Chemistry	Organic solvent extraction	Methanol	Leaves and fruits	Drying and crushing	Jasso de Rodríguez *et al*. (2020; 2023; 2024a; 2024b)
		Ethanol/ Acetone	Leaves	Fresh	Brazales-Cevallos *et al*. (2022)
Physics		Water	Young leaves	Drying and crushing	Velasco-Ramírez *et al.* (2021; 2022; 2023)
	Mechanical extraction with blender	Water	Leaves	Fresh	Brazales-Cevallos *et al*. (2022)
	Maceration	Water	Leaves	Drying and crushing	Garza-García *et al*. (2023)
		Methanol	Leaves and cladodes	Fresh	Moreno-Guerrero *et al.* (2024)
	Ultrasound-assisted extraction	Water	Leaves	Fresh	Velázquez-Gamboa *et al*. (2024)
Biological	Anaerobic fermentation	Water	Waste	Fresh	Huez-Lopez *et al*. (2008)
		Water	The whole plant	Fresh and segmented	Alcalán-López *et al*. (2023)

2024). In this study, four methods for applying plant extracts in crops were identified and evaluated under various experimental conditions. These methods include seed treatment, direct application to plants or soil, and a combination of the latter approaches (Figure 3).

The 47 % of the studies explored the biostimulant effects of seed treatment by hydration with different concentrations of extracts. Of these, 27 % were performed in petri dishes under laboratory conditions, 7 % in germination trays or pots under greenhouse conditions, and 13 % in both experimental environments (Figure 3). Also, direct application to plants by foliar spray accounted for 47 % of the research analyzed in this study, all of which were conducted under greenhouse conditions (Figure 3).

These results highlight the potentialities of using botanical extracts by both application methods to promote desirable agronomic traits in crops. For example, seed priming with botanical extracts is consolidated as an effective preconditioning strategy, which optimizes germination capacity and promotes adequate seedling establishment (Alcalán-López *et al*., 2023; Velasco-Ramírez *et al*., 2022a; Carvalho *et al*., 2024; Jasso de Rodríguez *et al*., 2024b). On the other hand, foliar spraying allows an immediate interaction of the bioactive substances present in plant extracts with plant tissues, thanks to their rapid absorption through stomata and cuticles, which induces a direct response in crops (Youssef *et al*., 2022; Han *et al*., 2024). This application method is most effective when it is performed in the morning, between 8:00 and 10:00 a.m., when the stomata remain open (Jasso de Rodríguez *et al*., 2023). Likewise, the complementary use of agricultural adjuvants contributes to optimizing the absorption of extracts through plant foliage (Jasso de Rodríguez *et al*., 2020).

The applications of botanical extracts by irrigation to the soil or to the growing substrate were evaluated in 34 % of the studies, 27 % in the greenhouse, and 7 % in the laboratory (Figure 3). This method delivers plant extracts to the base of the crop, promoting root development and microbial activity in the rhizosphere (Youssef *et al*., 2022). However, bioactive substances can be adsorbed by soil particles or lost through leaching, limiting their bioavailability (Mattner *et al*., 2013; Han *et al*., 2024). The combined application of botanical extracts to both plants and soil has been demonstrated to enhance their effectiveness (Velasco-Ramírez *et al*., 2022b). Only 7 % of studies have investigated this approach under greenhouse conditions (Figure 3), underscoring the need for further research in this sense.

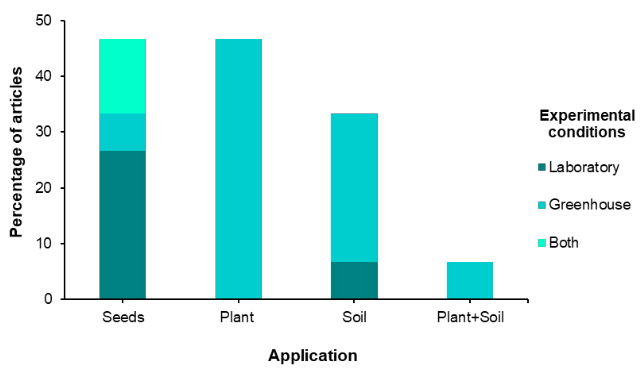


Figure 3. Main applications of plant extracts in agriculture, and the experimental conditions evaluated in the articles published by Mexican authors during the study period.

Figura 3. Principales aplicaciones de los extractos vegetales en agricultura, y las condiciones experimentales evaluadas en publicaciones por autores mexicanos durante el periodo de estudio.

On the other hand, bioactive compounds in botanical extracts tend to oxidize or degrade over time, limiting the duration of their beneficial effects on plants (Ali *et al*., 2021). Therefore, applications should be every 7 to 14 days to obtain an optimal response in crops (Brazales-Cevallos *et al*., 2022; Velasco-Ramírez *et al*., 2022a; Garza-García *et al*., 2023; Jasso de Rodríguez *et al*., 2024a; Moreno-Guerrero *et al*., 2024). In addition, the efficacy of the extracts also depends on the developmental stage of the crop, with transplanting, flowering, and fruiting being key moments for their application (Jasso de Rodríguez *et al*., 2020; 2023).

Crops evaluated with botanical extracts

The analyzed studies used 12 crop species as experimental models to evaluate the biostimulant properties of botanical extracts. Among these, horticultural crops were the most extensively studied, accounting for 67 % of the research included in this review. Cucumber (*Cucumis sativus* L.) was the most frequently studied crop, appearing in 27 % of the publications. Other key vegetables, such as tomato (*Solanum lycopersicum* L.) and lettuce (*Lactuca sativa* L.), were evaluated in 20 % of the studies. Bell pepper (*Capsicum annuum* L.) was addressed in 13 % of the publications, while carrot (*Daucus carota* L.) and radish (*Raphanus sativus* L.) were each featured in only 7 %.

This preference for leaf, root, or fruit vegetables is probably attributed to their short growth cycle, which facilitates the observation of rapid treatment responses. This group of crops has been recommended in NOM-077-FITO-2000 to carry out biological effectiveness studies of plant nutrition inputs (DOF, 2020). In addition, vegetables represent a fundamental part of agricultural production in Mexico, where approximately 20.3 % of arable areas are used for this type of crop (SIAP, 2024).

On the other hand, 7 % of the studies investigated the biostimulant effects of botanical extracts on fruit (*Fragaria ananassa* Duch.), ornamental (*Catharanthus roseus* L.), and medicinal (*Calendula officinalis* L.) crops. The remaining 20 % of the published articles included crops such as *Salicornia bigelovii* Torr., *Stevia rebaudiana* Bertoni, and *Arabidopsis thaliana* L., species with various agroindustrial and biological applications.

Biostimulant effects of botanical extracts

Several studies in recent years have demonstrated the positive effects of crude botanical extracts or their organic fractions on seed germination, vegetative growth, flowering, and fruiting of crops of commercial interest (Jasso de Rodríguez *et al*., 2020; Velasco-Ramírez *et al*., 2021; Jasso de Rodríguez *et al*., 2023; Velasco-Ramírez *et al*., 2022b; Alcalán-López *et al*., 2023; Jasso de Rodríguez *et al*., 2024a; Moreno-Guerrero *et al*., 2024; Velázquez-Gamboa *et al*., 2024; Godlewska *et al*., 2021; Han *et al*., 2024). These processes are critical to agricultural production, directly impacting crop yield and quality (Martínez-Lorente *et al*., 2024). Table 2 summarizes the biostimulant effects of Mexican plant extracts and the metabolites responsible for their biological activities.

Seed germination

Seed germination is a key stage for agricultural production because it marks the beginning of plant growth and largely determines crop yield (Carvalho *et al*., 2022). Therefore, it is important to implement natural strategies that ensure successful and uniform germination to obtain vigorous plants resistant to various environmental conditions (Gupta *et al*., 2022). Seed treatment (i.e., imbibition or soaking) with botanical extracts is a simple and effective technique to improve germination and stimulate seedling growth (Martínez-Lorente *et al*., 2024). However, the concentration of the extracts evaluated is a determining factor in their effectiveness as biostimulants (Han *et al*., 2024).

For example, liquid extracts obtained from different halophytes at 5 % increased the relative germination percentage, relative radicle growth, seedling length, and germination index of crops such as carrot (*D. carota*), lettuce (*L. sativa*), radish (*R. sativus*), and cucumber (*C. sativus*). High concentrations (20 %) significantly reduced these parameters (Alcalán-López *et al*., 2023). In another study, aqueous extracts of *V. sphaerocephala* and *V. fastigiata* at 0.5 % and 1.0 % adversely affected the germination percentage, mean germination time, and vigor index of tomato and cucumber seeds. However, they promoted the growth of leaves and seedling roots of these potted vegetables (Velasco-Ramírez *et al*., 2023). These authors suggested that *Verbesina* extracts contain allelopathic compounds that inhibit germination. On the other hand, Jasso de Rodríguez *et al.* (2024b) reported the highest values of total plumule and radicle length, as well as the fresh weight of seedlings of two tomato varieties, at

Table 2. Biostimulant effects of extracts obtained from plants distributed in Mexico, and responsible metabolites identified in the study * or suggested from the literature **.
Tabla 2. Efectos bioestimulantes de extractos obtenidos de plantas distribuidas en México, y metabolitos responsables identificados en el estudio * o sugeridos en la literatura **.

Plant source for extract	Responsible metabolites	Biostimulant effect	Plant evaluated	References
Grasses	Humic substances**	Improved seed germination and radicle growth under salt stress	Chili pepper	(Huez-López *et al*., 2008)
Myriophyllum aquaticum	Phenolic compounds*	Increased root elongation	Lettuce	(Viveros-Legorreta *et al*., 2018).
Rhus trilobata, *Rhus muelleri*, *Flourensia microphylla*, *Flourensia retinophylla*, *Cucurbita foetidissima*	Phenolic compounds**	Increased vegetative growth, crop yield, and fruit quality	Tomato	(Jasso de Rodríguez *et al*., 2020)
Verbesina sphaerocephala	Phenolic compounds*	Stimulated vegetative growth and flowering	Cucumber	(Velasco-Ramírez *et al*., 2021)
Moringa oleifera	Flavonoids, Amino Acids**	Induce tolerance to salinity	Arabidopsis	(Brazales-Cevallos *et al*., 2022)
Verbesina sphaerocephala *Verbesina fastigiata*	Phenolic compounds**	Promoted seedling growth	Tomato, cucumber	(Velasco-Ramírez *et al*., 2022a)
Verbesina sphaerocephala, *Verbesina fastigiata*	Phenolic compounds**	Increased vegetative growth, flowering, and fruit production	Strawberry	(Velasco-Ramírez *et al*., 2022b)
Bacopa monieri, *Distichlis spicata*, *Heliotropium curasavicun*, *Flaveria trinervia*, *Sesuvium verrucosum*	Not mentioned	Increased germination parameters	Carrot, lettuce, radish, and cucumber	(Alcalán López *et al*., 2023)
Maclura tinctoria	Phenolic compounds*	Stimulation of plant growth, height, and flowering	*Salicornia bigelovii*	(Corona *et al*., 2023)
Amphipterygium glaucum	Flavonoids, polyphenols, reducing sugars*	Improved vegetative development, flower production, and photosynthetic pigment content	Vinca, Calendula	(Garza-García *et al*., 2023)
R. muelleri, *Rhus virens*, *F. microphylla*, *F. retinophylla*	Phenolic compounds**	Promotion of growth, yield, and fruit quality	Bell pepper	(Jasso de Rodríguez *et al*., 2023)
R. muelleri, *F. microphylla F. retinophylla*	Phenolic compounds**	Increased growth, yield, and fruit quality	Cucumber	(Jasso de Rodríguez *et al*., 2024a)
R. muelleri, *R. virens*, *Jatropha dioica*, *Juglans mollis*	Phenolic compounds**	Stimulated root, stem, and leaf growth	Tomato	(Jasso de Rodríguez *et al*., 2024b)
Opuntia ficus-indica, *Coriandrum sativum*	Phenolic compounds, phytohormones**	Increased mineral nutrient content	Lettuce	(Moreno-Guerrero *et al*., 2024)
Moringa oleifera	Not mentioned	Increased phenolic compounds and glycosides content in leaves	*Stevia rebaudiana*	(Velazquez-Gamboa *et al*., 2024)

the lowest (75 mg mL^{-1}) and highest (200 mg mL^{-1}) concentrations of semi-desert plant extracts.

Also, the application of botanical extracts has been shown to improve the germination percentage and growth of bell pepper (*C. annuum*) and *Arabidopsis thaliana* seedlings under salinity stress conditions (Huez-Lopez *et al*., 2008; Brazales-Cevallos *et al*., 2022). In general, the variability in observed effects across studies can be attributed to differences in extract type, the plant species or cultivar evaluated, and the experimental conditions (Carvalho *et al*., 2022).

Crop growth and productivity

Enhancing crop morphology and physiology during the growth phase is associated with increased productivity, which is the primary objective of agricultural production (Zulfiqar *et al*., 2020). Appropriate treatment with botanical extracts at different phenological stages of crops has positive effects on growth and productivity parameters (Godlewska *et al*., 2021; Han *et al*., 2024).

In a study, extracts of *R. muelleri* and *J. dioica* stimulated root and aerial growth of tomato seedlings under greenhouse conditions, more than commercial rooting products (Jasso de Rodríguez *et al*., 2024b). Similarly, another study reported that the application of botanical extracts increased stem length and diameter, as well as leaf dry weight, in tomato plants. Moreover, the extract of *R. muelleri* showed greater efficacy in increasing the number, weight, and yield of tomatoes, reaching values similar to those obtained with the bioregulator 6-benzylaminopurine (Jasso de Rodríguez *et al*., 2020). Likewise, applying *F. retinophylla* extract to bell pepper plants has stimulated longitudinal stem growth and enhanced total dry matter accumulation and root volume. This, in turn, increased fresh weight, volume, and yield of pepper fruit (Jasso de Rodríguez *et al*., 2023).

The biostimulant effect of botanical extracts, whether applied individually or in combination, has also been demonstrated to improve growth parameters (e.g., plant height, leaf number, chlorophyll index, and biomass accumulation) and enhance the productivity of cucumber plants (Velasco-Ramírez *et al*., 2021; Jasso de Rodríguez *et al*., 2024a).

A study by Velasco-Ramírez *et al*. (2022b) found that extracts of *V. sphaerocephala* and *V. fastigiata* improved the growth (leaf width and length, crown number, and root length) and reproductive development (number of buds, flowers, and fruits) of strawberry plants grown in pots. On the other hand, the foliar application of nanoparticles synthesized from the methanolic extract of *A. glaucum* stimulated better vegetative development and increased the content of photosynthetic pigments in vinca (*C. roseus*) and calendula (*C. officinalis*) plants (Garza-García *et al*., 2023). In contrast, nopal (*O. ficus-indica*) and coriander (*C. sativum)* extracts did not positively affect the growth of lettuce plants (Moreno-Guerrero *et al*., 2024).

Tolerance of abiotic stress

In recent years, there has been a notable increase in the prevalence of abiotic stress conditions in agriculture, largely due to the effects of climate change (Ali *et al*., 2021; Martínez-Lorente *et al*., 2024). These situations induce physiological stress that can significantly reduce crop yields (Ahmad *et al*., 2022). The use of plant-derived biostimulants is emerging as a cost-effective and sustainable strategy to mitigate the effects of adverse environmental conditions on crops (Godlewska *et al*., 2021; Ahmad *et al*., 2022; Martínez-Lorente *et al*., 2024). However, research on the potential of Mexican plant extracts to enhance abiotic stress tolerance is still in its early stages.

The study by Huez-Lopez *et al*. (2008) reported that *C. echinatus* extract, obtained by fermentation, improved seed germination percentage and the length of the radicle and hypocotyl of chili pepper (*C. annuum*) seedlings under salinity conditions. Likewise, Brazales-Cevallos *et al*. (2022) observed that *M. oleifera* leaf extracts induce tolerance to saline stress on *A. thaliana* seeds. This study offers significant insights into the mechanisms at the transcriptomic level underlying the protective effects of plant-based biostimulants during the initial stages of plant development.

Harvest quality

Ensuring access to sufficient, safe, and high-quality nutritious food to meet dietary needs represents one of the major challenges facing agriculture in the quest for food security. The quality of agricultural products is a key factor influencing their commercial value, consumer satisfaction, and the reduction of post-harvest losses (Akter *et al*., 2024). The concept of quality encompasses the physical appearance of the product (i.e., color, size, shape, firmness) as well as the organoleptic, nutritional and nutraceutical properties that determine its acceptability and health benefits (Reis *et al*., 2022). A recently recognized strategy for enhancing crop quality is the application of plant biostimulants (Calvo *et al*., 2014; Rouphael and Colla, 2020; Han *et al*., 2024). In this regard, several studies included in this review highlight the potential of botanical extracts to improve various quality attributes, demonstrating their capacity to positively influence the characteristics of the final product.

The foliar application of extracts from plants native to the semiarid regions of northeastern Mexico resulted in tomato fruits (*S. lycopersicum*) with polar length, equatorial diameter, firmness, pH, total soluble solids (TSS), lycopene, and vitamin C content comparable to those obtained with the synthetic cytokinin 6-BAP (Jasso de Rodríguez *et al*., 2020). Similarly, foliar spraying of semi-desert plant extracts enhanced several quality attributes of bell peppers, including longitudinal and equatorial diameters, volume, TSS, anthocyanin, and vitamin C content. The values obtained were comparable to or exceeded, those obtained with commercial bioregulators (Jasso de Rodríguez *et al*., 2023). In another study, combining various botanical extracts improved several of the aforementioned quality parameters in cucumber fruits (Jasso de Rodríguez *et al*., 2024a). On the other hand, *S. rebaudiana* plants treated with nanoparticles synthesized from the aqueous extract of *M. oleifera* showed a higher leaf content of phenolic compounds (chlorogenic acid and caffeic acid) and glycosides (Velázquez-Gamboa *et al*., 2024). Moreover, foliar application of nopal and coriander extracts increased the concentration of macro and micronutrients in the lettuce leaf tissue (Moreno-Guerrero *et al*., 2024).

Mechanisms of biostimulatory activity

According to Yakhin *et al*. (2017), the mode of action refers to the "specific effect on a discrete biochemical or regulatory process", while the mechanism of action involves the "impact on general biochemical or molecular pathways or physiological processes". The inherent chemical complexity of botanical extracts has thus far precluded the requisite level of characterization necessary to elucidate their specific modes of action. Therefore, their functions in plants are mainly inferred from the underlying mechanisms of action (Martínez-Lorente *et al*., 2024).

In this regard, the overall positive effects of the botanical extracts on plant growth and productivity are due to the modulation of various physiological and biochemical processes, such as (1) nutrient and water uptake, (2) photosynthetic activity, (3) modulation of phytohormones, (4) specialized and oxidative metabolism, (5) gene expression related to abiotic stress tolerance, and (6) plant architecture and phenology (Zulfiqar *et al*., 2020; Godlewska *et al*., 2021; Ahmad *et al*., 2022; Martínez-Lorente *et al*., 2024). However, it should be clarified that the biostimulatory properties of botanical extracts on plant performance are not the result of a single bioactive compound, but of the coordinated and synergistic action of predominant compounds, such as polyphenols, phytohormones, amino acids, sterols, terpenoids, and vitamins (Ahmad e*t al*., 2022; Han *et al*., 2024; Jasso de Rodríguez *et al*., 2024a). Although some studies have reported the presence of micro- and macronutrients in plants-derived extracts (Huez-Lopez *et al*., 2008; Velasco-Ramírez *et al*., 2022b; Alcalán-López *et al*., 2023), their concentrations are often insufficient to induce physiological responses in plants at the doses commonly employed (Han *et al*., 2024). This observation suggests that organic constituents, rather than mineral elements, are primarily responsible for growth-stimulating activity.

Most of the studies analyzed in this review attribute the beneficial effects of extracts from Mexican plants to their phenolic compound content. These compounds represent one of the most abundant families of secondary plant metabolites and include phenolic acids, flavonoids, tannins, and others (Michalak *et al*., 2006; Martínez-Lorente *et al*., 2024). Recently, interest in phenolics has increased considerably due to their potential as biostimulants in agriculture (Rouphael and Colla, 2020). Phenolics act as potent antioxidants because they can scavenge reactive oxygen species, thus helping to maintain redox homeostasis and protect cells from oxidative damage under normal and stressful conditions (Michalak *et al*., 2006). Furthermore, they modulate the metabolism and transport of auxins and ethylene, thereby indirectly influencing the processes regulated by these phytohormones (Santner *et al*., 2009). For example, auxins are primarily associated with growth and development, while ethylene regulates maturation, senescence, and stress response (Ali *et al*., 2021). By the above, several studies on plant methanolic extracts in various horticultural crops have reported biostimulatory activity comparable to that of synthetic bioregulators, such as indoleacetic acid, gibberellic acid, and 6-benzylaminopurine (Jasso de Rodríguez *et al*., 2020; Jasso de Rodríguez *et al*., 2024a; Jasso de Rodríguez *et al*., 2024b). In another study, moringa leaf extracts induced tolerance to salinity stress in *A. thaliana* seeds through the transcriptional activation of abscisic acid, salicylic acid, auxin, and ethylene-related signaling pathways (Brazales-Cevallos *et al*., 2022).

Additionally, phenolic compounds can stimulate photosynthesis, enhance CO_2 fixation, and regulate the stomatal opening/closing mechanism, promoting better plant adaptation to environmental conditions (Ertani *et al*., 2016; Agati *et al*., 2012; Martínez-Alonso *et al*., 2022). On the other hand, when applied to the soil, flavonoids can function as chelating agents for metal ions, enhancing their availability for plant nutrition (Cesco *et al*., 2012). Moreover, it has been proposed that flavonoids released into the rhizosphere stimulate the solubilization of inorganic phosphorus from the soil, allowing its plant uptake (Tomasi *et al*., 2008). These phenolic compounds have also been found to increase the expression of aquaporins in biological membranes, improving water uptake efficiency (Martinez-Alonso *et al*., 2022).

Another important role of phenolics is their participation in plant-microorganism interactions (Badri *et al*., 2009). It has been described that *nod* genes, which are activated in legumes during symbiosis with nitrogen-fixing bacteria, are stimulated by flavonoids acting as chemical signals that trigger their activation (Cooper, 2007). Furthermore, both flavonoids and some phenolic acids have been shown to interfere with quorum sensing, a microbial communication system used in symbiosis, defense, and other plant-microorganism interactions (Zhuang *et al*., 2005).

Finally, based on the scientific reports included in this work, we suggest that the bioactive compounds in botanical extracts can promote plant growth and increase plant productivity (yield and quality) through direct and indirect mechanisms of action. When absorbed through the leaves or roots, some compounds interact directly with plant metabolism and physiology. In contrast, others act indirectly by improving soil properties, enhancing the rhizospheric environment, and stimulating beneficial soil microbiota.

FUNDAMENTAL STAGES OF PLANTS BIOPROSPECTING FOR THE PRODUCTION OF BIOSTIMULANTS

Biostimulants derived from native plant extracts are currently experiencing a global surge in interest, aiming to valorize local natural resources to enhance agriculture and promote food sovereignty (Godlewska *et al*., 2021; Han *et al*., 2024).

However, the studies analyzed in this review show that only a limited number of plant species in Mexico have been evaluated for this purpose. In this context, the systematic search for natural compounds and whole organisms with potential commercial value—known as bioprospecting—is a valuable tool for increasing our understanding of species with known biostimulant properties (Torres and Velho, 2009). In the context of this publication, a bioprospecting was focused on the identification and development of novel plant-derived bioactive compounds for use as agricultural biostimulants, and a methodology in two-phases was proposed. To achieve this, the integration of knowledge, technologies and processes is needed through schemes with systematic approaches and phases such as the one presented in Figure 4.

Phase 1 focuses on the initial exploration and selection of species of interest with agricultural potential. The process starts with a judicious assessment of potential plant species, weighing their favourable attributes (such as abundance in the ecosystem, ease of access and their ability to thrive in challenging environmental conditions) against potential constraints, such as biomass yield or the stability of their compounds. Once a species has been pre-selected, the research is aimed at locating the metabolites of interest within the plant, commonly concentrated in the leaves. Priority is given to aqueous extraction methods due to their simplicity and applicability both in the laboratory and in the field, also incorporating a sustainability perspective when considering the use of residual biomass. Preliminary biological assays, such as germination tests or rapid growth models, are then performed to evaluate the biostimulant potential of the extracts obtained. This stage requires previously defining the key variables that will allow the effect to be measured, such as germination growth, and vigor parameters of the plants. It is important that bioprospecting studies consider the specifications of NOM-077-FITO-2000, which are mainly applicable in phase 1 of this strategy (Figure 4). Once the exploratory phase is over, Phase 2 delves into scientific research and product development. It begins with an exhaustive chemical characterization of those extracts that showed promising activity, in order to identify the molecules responsible for the biostimulant effect. Accurate quantification of these active molecules, both in the extracts and in the original plant, is another crucial step in standardizing and understanding the dose-response. At the same time, it seeks to elucidate the underlying mechanisms of action, that is, how these molecules interact at a biochemical, physiological or genetic level in the plant. Subsequently, the research addresses the ways for the formulation of the product, will delve into the methods used to create or develop a product. At this point the research will explore the specific techniques, processes, and approaches involved in taking an idea and turning it into a tangible product up to sustainable and scalable production

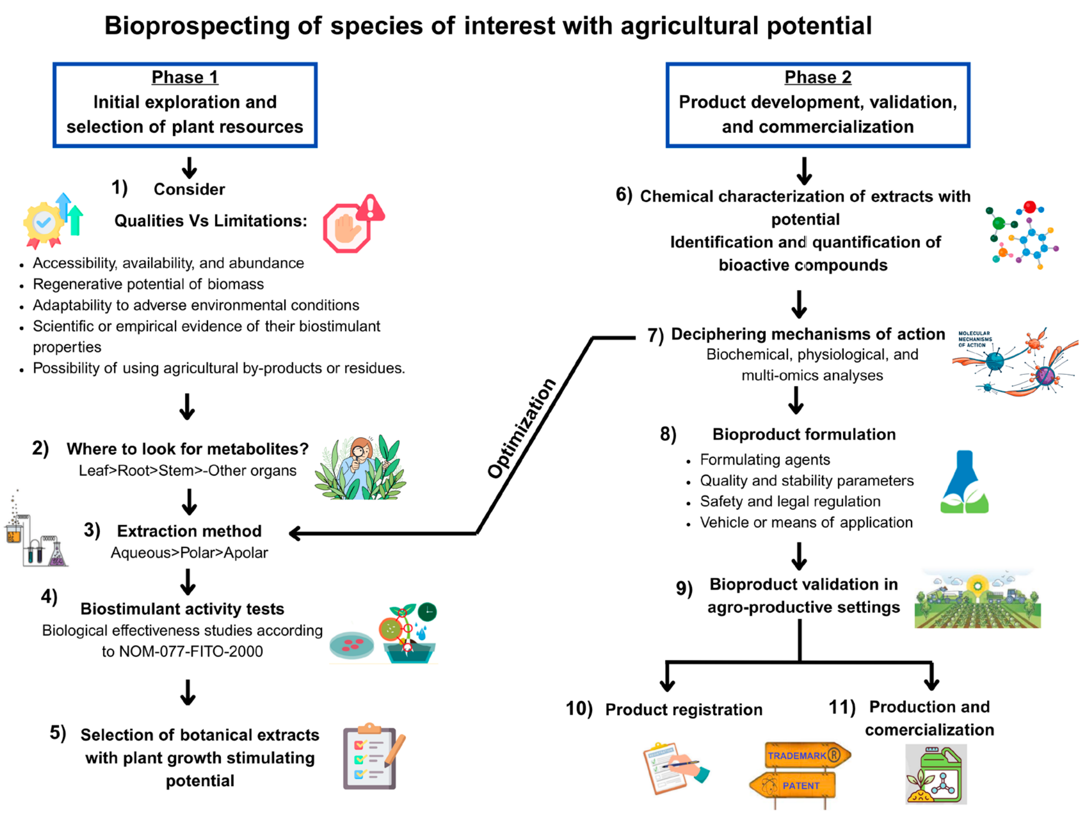


Figure 4. Phases of bioprospecting of plant species for the production biostimulants.
Figura 4. Fases de bioprospección de especies vegetales para la producción de bioestimulantes.

of the bioinput, exploring options such as the cultivation of the source species, the culture of plant tissues or even the organic synthesis of the identified molecules. Prior to final implementation, a rigorous cost-benefit analysis is performed to determine the economic viability of the technology. Finally, the process of registering intellectual property is contemplated to protect the innovation developed.

Concluding remarks and research perspectives

Interest in plant biostimulants has increased significantly in recent years, driven by the need to adopt more sustainable and efficient agricultural practices. However, there are still several issues to be addressed, mainly related to their definition and regulation. In Mexico, the production, commercialization, and use of biostimulants in agriculture remain limited, partly due to the absence of a specific regulatory framework. Currently, these products attempt to be regulated indirectly through the Norma Oficial Mexicana NOM-077-FITO-2000, which focuses on plant nutrition inputs, such as organic soil improvers, inoculants, and type 1 and 2 growth regulators. Therefore, a coordinated effort between academia, government regulatory agencies, and industry is needed to harmonize current regulations and enhance farmers' comprehension of the functions and benefits of biostimulants.

Among the diversity of raw materials recognized for biostimulant production, plants stand out for their accessibility, wide availability, and richness in primary and secondary metabolites with multiple biological functions. In this sense, the studies analyzed in this review support the biostimulant potential of extracts from plant species distributed in Mexico. However, despite the remarkable floristic richness of the country, few species have been studied for this purpose. In particular, the Asteraceae family concentrates the largest number of species evaluated in bioprospecting studies.

The application of botanical extracts to seeds, plants, or soil has been demonstrated to have positive effects on key processes such as germination, seedling vigor, vegetative growth, reproductive development, abiotic stress tolerance, as well as on crop yield and nutritional quality. Horticultural crops are among the most used experimental models in studies evaluating the biological effectiveness of botanical extracts, both under laboratory and greenhouse conditions. Most studies attribute the observed benefits to phenolic compounds, including phenolic acids and flavonoids. However, the efficacy of these extracts can be influenced by several factors, including the characteristics of the raw material (species, phenological stage, part of the plant), the extraction method (extraction technique and solvent), and the conditions of application to the plant (concentration, dose, frequency, and timing of application).

Although advances in the understanding of Mexican plant extracts highlight their potential as a promising strategy for sustainable crop management aimed at enhancing production and productivity, further research is still required. The exploration of new species with biostimulant properties, especially those native or endemic species with the ability to adapt to specific environmental conditions, would allow the identification of novel biological activities and bioactive compounds. Moreover, research on the mechanism of action requires the elucidation of the chemical compounds responsible for the effects observed in botanical extracts already studied, as well as a further understanding of the physiological, metabolic, and genomic modifications induced in plants. Likewise, to optimize the use of these bioproducts in agriculture, crop-specific research is needed to study in depth the duration of the effect after the application of the extract, the most appropriate phenological stages for its application and maximization of benefits, as well as the possibility of obtaining synergistic effects by combining extracts from different plant species or concentrations. Additional trials also should be conducted under field conditions to demonstrate the efficacy of these botanical formulations in natural settings, including abiotic stress situations. Finally, using advanced biotechnological tools, such as nanotechnology, can enhance the biostimulant properties of botanical extracts, improving their stability, bioavailability, and competitiveness against synthetic agro-inputs.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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