# AKADEMİK PERSPEKTİFTEN BİTKİ KORUMA

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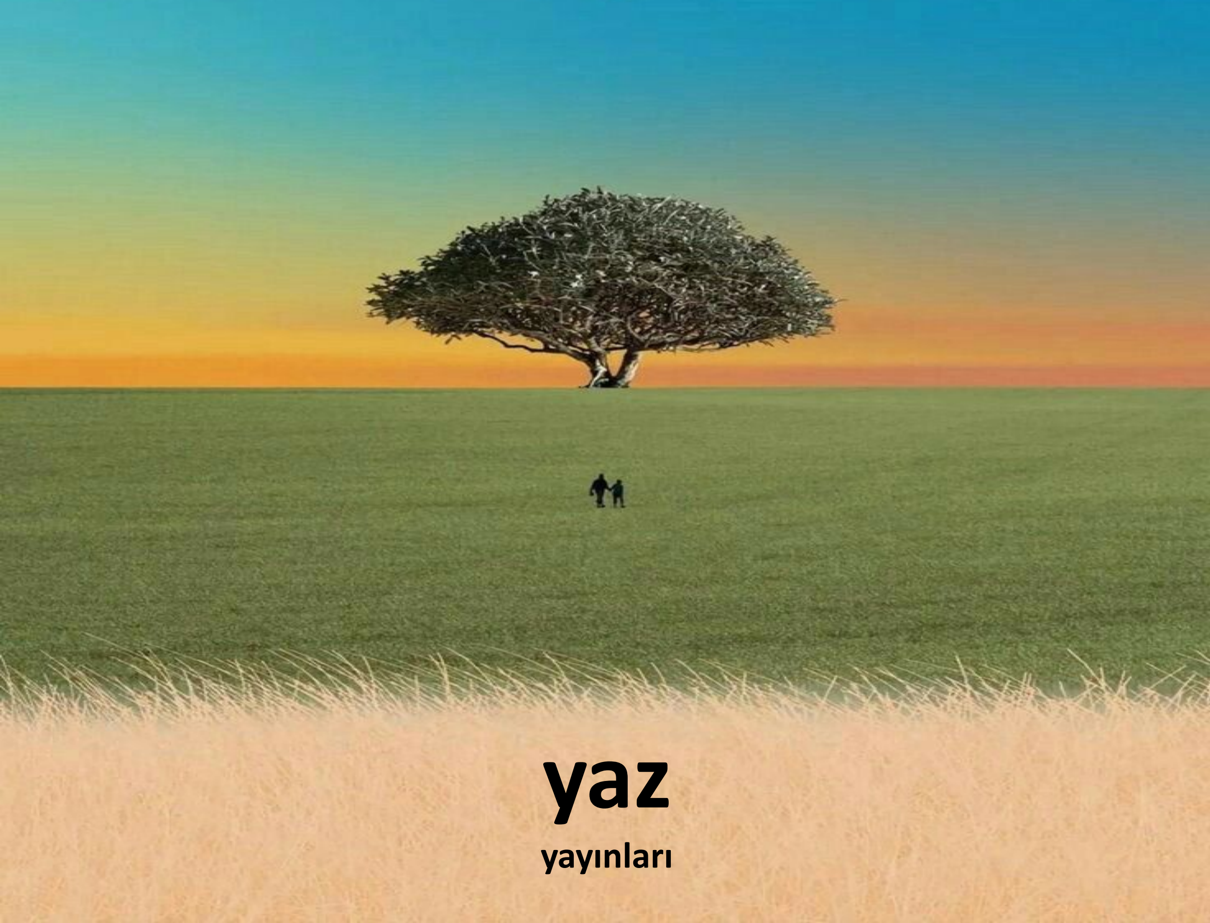

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# İÇİNDEKİLER

*"Bu kitapta yer alan bölümlerde kullanılan kaynakların, görüşlerin, bulguların, sonuçların, tablo, şekil, resim ve her türlü içeriğin sorumluluğu yazar veya yazarlarına ait olup ulusal ve uluslararası telif haklarına konu olabilecek mali ve hukuki sorumluluk da yazarlara aittir."*

# BIBLIOMETRIC ANALYSIS IN THE BIOLOGICAL CONTROL OF *ACIDOVORAX CITRULLI* AND DETERMINATION OF LOCAL ANTAGONISTS [1]

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**Mesude Figen DÖNMEZ YEŞİLDAĞ[3]**

## 1. INTRODUCTION

Cucurbit crops are widely cultivated throughout the world and are among the economically important vegetable species. Some diseases seen in plants belonging to this family, which includes species such as melon, watermelon, cucumber, can seriously threaten the quantity and quality of the product. One of these diseases is bacterial fruit spot disease caused by Acidovorax citrulli, which causes significant economic losses in cucurbit plants. The disease agent is an important risk factor for producers, especially because it spreads through contaminated seeds (Bahar et al., 2009) and is considered a potential source of disaster in all regions where cucurbit production is carried out (Hopkins and Thompson, 2002). This seed-borne pathogen can cause infection at any stage of plant development, but is most common during the seedling period or fruit ripening stage (Hopkins et al., 1993). However, incorrect practices in agricultural production (such as

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[1] Bu çalışma, yazarın Doç. Dr. Mesude Figen Dönmez Yeşildağ danışmanlığında Iğdır Üniversitesi Lisansüstü Eğitim Enstitüsünde yapmış olduğu doktora tezinin bir kısmından üretilmiştir.

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not removing diseased plants from the field or using contaminated seeds) cause the pathogen to spread to large areas and threaten the cultivation of species such as melon and watermelon on a global The use of pathogen-free seeds and seedlings is important in disease control. However, if the pathogen comes to the growing environment later, it is insufficient in the struggle. Although the use of resistant cultivars is an effective control strategy, the lack of a commercial melon cultivar resistant to *A. citrulli* (Sales Ju'nior et al. 2015) causes difficulties in controlling the disease. Disease control using pesticides raises serious concerns about food safety, environmental quality and resistance to pesticides, which determine the need for alternative disease and pest management techniques (Dordas, 2008). Therefore, in addition to the use of pesticides, it has become necessary to develop more sustainable and healthy alternatives in our food systems. This has led to the term sustainability being frequently encountered in many areas of our lives, especially in agriculture (Hanson et al., 2007). The increasing interest in sustainable agriculture in recent years has made this field one of the most important issues. At this point, it is important to implement alternative measures in the control of plant diseases that do not harm the environment, do not threaten living health, and at the same time increase yield and improve product quality (Batish et al., 2007; Camprubí et al., 2007). One of the most promising alternative methods to achieve this goal is biological control using antagonist bacteria (Vinale et al., 2007; Santoyo et al., 2016). This is one of the most economical and long-term effective strategies to control plant diseases and minimise crop loss (El-Saadony et al., 2022). Numerous studies have shown that antagonistic bacteria-plant interactions are important in reducing the severity of plant diseases, protecting plants from pathogens and increasing plant productivity (Jiang et al., 2015; Lemos et al., 2016; Adhikari et al., 2017; Glick and Gamalero 2021). The effective examination of all these studies requires a comprehensive analysis of the literature on the subject. In this context, bibliometric analyses contribute to the understanding of trends in the research field, author and country collaborations, prominent topics and scientific impact (Guleria and Kaur 2021; Arruda et al., 2022).

In this study, bibliometric analysis of published studies on the control of *Acidovorax citrulli* by biological methods was performed using the VOSviewer program. With this analysis, it is aimed to obtain comprehensive information about the research in this field both in the world and in Turkey, and to identify the prominent authors, countries and topics. In addition, it was aimed to determine potential antagonist bacterial strains isolated from local sources to be used in the fight against this pathogen detected in the region.

## 2. MATERIALS AND METHODS

### 2.1. Bibliometric analysis using VOSviewer software

For the bibliometric analysis of the studies on *Acidovorax citrulli*, which causes bacterial spot disease in cucurbits, the keyword "*Acidovorax citrulli*" was searched in the Web of Science database and bibliometric analysis was performed on the obtained studies using the VOSviewer (https://www.vosviewer.com/) program.

### 2.2. Bacterial stains tested against Acidovorax citrulli in the study

KVN 21, which was selected among 21 *A. citrulli* strains isolated and identified from melon fields in Iğdır by İnik (2019) and had the highest virulence, was used as pathogenic bacteria. Strains isolated from alternative forage plants (*Chepodium qinoa, Amaranthus paniculatus, Atriplex nitens* and *Salsola rutenica*, *Solanum lycopersicum*) grown in Iğdır and volcanic rocks present in the region by Temel (2023) and identified by biochemical tests, fatty acid methyl ester (FAME) and 16S rDNA sequence analysis were tested as candidate antagonist bacteria. The identification results of these strains are presented in Table 1.

**Table 1. Identification results of candidate antagonist bacteria**

| Strain No | FAME Identification Result | 16S rDNA Identification Result |
|---|---|---|
| **IT 14** | *Bacillus amyloliquefaciens* | *Bacillus amyloliquefaciens* |
| **IT 58** | *Variovorax paradoxus* | No record on NCBI Site |
| **IT 70** | *Pseudomonas fluerescens* | *Pseudomonas fluerescens* |
| **IT 115** | *Serratia rubidaea* | *Serratia rubidaea* |
| **IT 156** | *Bacillus subtilis* | *Bacillus subtilis* |
| **IT 159** | *Bacillus subtilis* | *Bacillus subtilis* |
| **IT 162** | *Bacillus subtilis* | *Bacillus subtilis* |
| **IT 188** | *Pseudomonas fluorescens* | *Pseudomonas fluorescens* |
| **IT 189** | *Bacillus atrophaeus* | *Bacillus atrophaeus* |
| **2/2** | *Bacillus atrophaeus* | *Bacillus atrophaeus* |
| **3/2** | *Bacillus thuringiensis* | *Bacillus thuringiensis* |
| **3/5** | *Bacillus subtilis* | *Pantoea agglomerans* |
| **3/9** | *Bacillus subtilis* | *Bacillus subtilis* |
| **4/2** | *Bacillus pumilus* | *Bacillus pumilus* |
| **6/4** | *Bacillus atrophaeus* | *Bacillus atrophaeus* |
| **6/5** | *Bacillus atrophaeus* | *Bacillus atrophaeus* |
| **8/3** | *Bacillus viscosus* | No record on NCBI Site |

FAME: Fatty acid methyl ester

### 2.3. Hypersensitivity in tobacco and pectolytic activity test of bacterial strains

In order to determine whether bacterial strains cause hypersensitivity reactions in tobacco, solutions of the strains at a concentration of $10^8$ cfu/ml were prepared and injected into the interveins of the lower leaf surface of tobacco (*Nicotiana tabacum* L. Samsun) plants using 3 ml plastic syringes. Necrosis (dead tissue formation) occurring in the inoculated areas after 48 hours was evaluated as HR positive (Lelliot and Stead, 1987). In order to determine the pectolytic activities of bacterial strains, potato tubers that were superficially disinfected and cut into 1 cm thick were placed in petri dishes containing sterile moist filter paper. Colonies taken from bacterial cultures using swab were inoculated onto the surface of potato slices and incubated for 48 hours. Softening in the area where the strains were inoculated was evaluated as positive, and absence of any change was evaluated as negative (Lelliot and Stead 1987).

### 2.4. Determination of antibacterial activity of bacterial strains against Acidovorax citrulli

In order to determine the effect of strains on pathogen development, a pathogen suspension prepared at a concentration of 108 cfu/ml was sprayed onto petri dishes containing Nutrient Agar (NA) and inoculated. 1 hour after application, antagonist bacteria were seeded in 3 spots on the medium at equal distances from each other using the dot seeding method and incubated at 27°C for 48 hours. At the end of the incubation period, the inhibition zones formed around the antagonist bacteria were measured in mm and recorded (Fan et al. 2017).

### 2.5. Statistical analysis

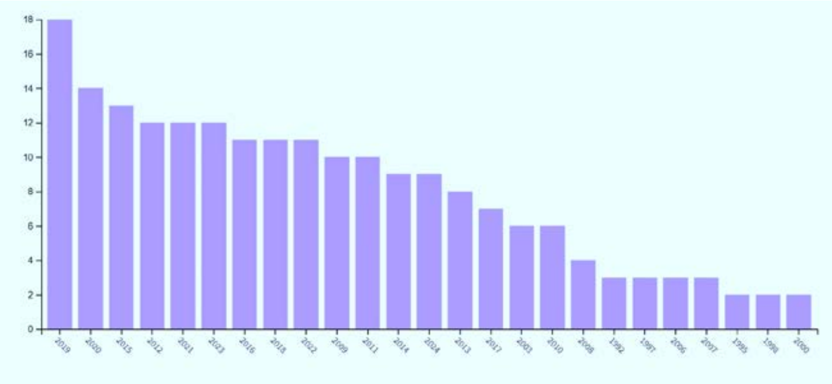
The data obtained from the trials conducted in the study were subjected to Variance Analysis in the SPSS (Version 17.0) statistical program and the differences between the applications were compared with the Duncan Multiple Comparison test ($P \leq 0.01$).

## 3. RESULTS

As a result of the search made with the terms "*Acidovorax citrulli*", "*Acidovorax avenae* subsp. *citrulli*" and "Bacterial fruit blotch" (ALL FIELDS) in the Web of Science database, 398 publications were obtained. Later; When the limitation was made with the criteria (EXACTKEYWORD, "Plant Pathology"), 212 publications were reached. When these studies were limited with the term "Biological Control", 21 publications were obtained. It was observed that the studies on the biological control of

*Acidovorax citrulli* started to increase in 2009 and the most publications were made in 2019 (Figure 1).

**Figure 1. Distribution of publications by years**




From the studies obtained, it was determined that the countries with the most publications on the subject were the United States and China, while there were 9 publications from Turkey (Figures 2,3). The keywords analyzed in the search for biological control of *Acidovorax citrulli* are given in Table 2. When the keywords obtained on the subject were examined, it was seen that the terms *Acidovorax citrulli* and biological control, which were frequently used in the studies, were at the center (Figure 4).

**Figure 2. Distribution of publications on biological control by country**

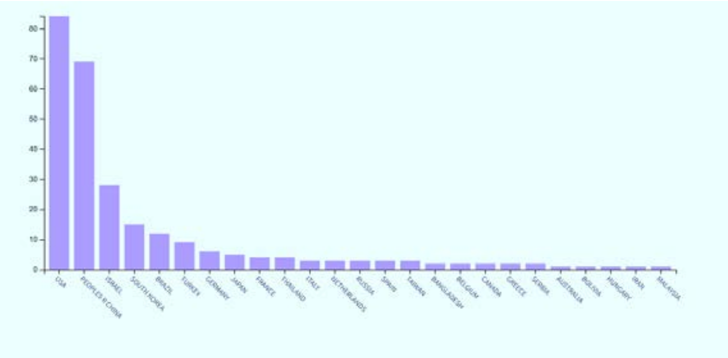


**Figure 3. Correlation analysis of countries regarding publications on biological control**

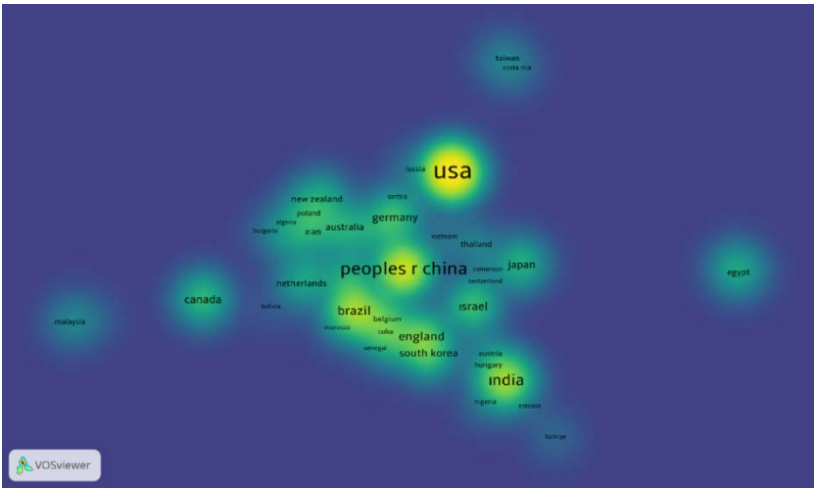
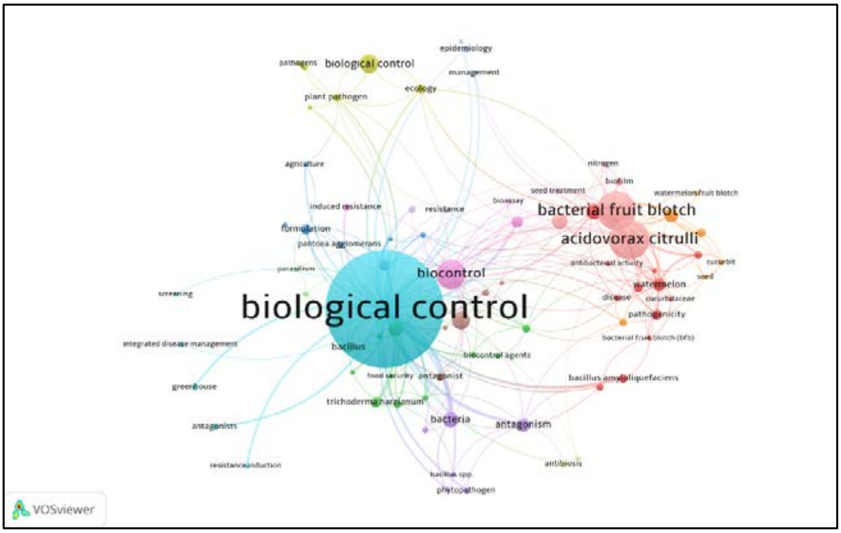



**Table 2. Keywords that stand out as a result of the analysis**

| Keywords | Occurrences* | Total link strength |
|---|---|---|
| Biological control | 264 | 359 |
| *Acidovorax citrulli* | 57 | 81 |
| Bakteriyel fruit blotch | 55 | 77 |
| Biocontrol | 37 | 50 |
| Bacteria | 34 | 53 |
| Biological control agents | 32 | 35 |
| *Cucumis melo* | 15 | 32 |
| Virulence | 15 | 24 |
| Antagonizm | 12 | 27 |
| Watermelon | 11 | 25 |
| *Bacillus subtilis* | 10 | 19 |
| *Pseudomonas fluorescens* | 9 | 25 |
| Formulasyon | 8 | 14 |
| *Acidovorax avenae* subsp. *citrulli* | 7 | 14 |
| *Bacillus amyloliquefaciens* | 7 | 10 |
| Sustainable agriculture | 6 | 15 |
| Antibacterial activity | 4 | 9 |
| Seed treatment | 3 | 5 |
| *Cucurbita pepo* | 2 | 5 |

*Occurrences: Number of terms in the text, Total Link Strength: Total relationship strength

**Figure 4. Keyword analysis of publications on biological control of *Acidovorax citrulli***




When the researchers were evaluated according to their productivity, it was seen that Walcot R.R. had the most publications and that he and Burdman played a leading role in the biological control of *Acidovorax citrulli* (Figure 5). When the obtained data were examined, it was determined that Burdman S. had the highest number of citations (703) on this subject. As a result of the collaboration analysis with other authors, it was determined that 10 different clusters were formed and that Burdman was the author with the most collaborations. It was seen that Walcot, who published the most, did not have many studies on the subject in recent years (Figure 6).

**Figure 5. Distribution of publications according to author productivity**

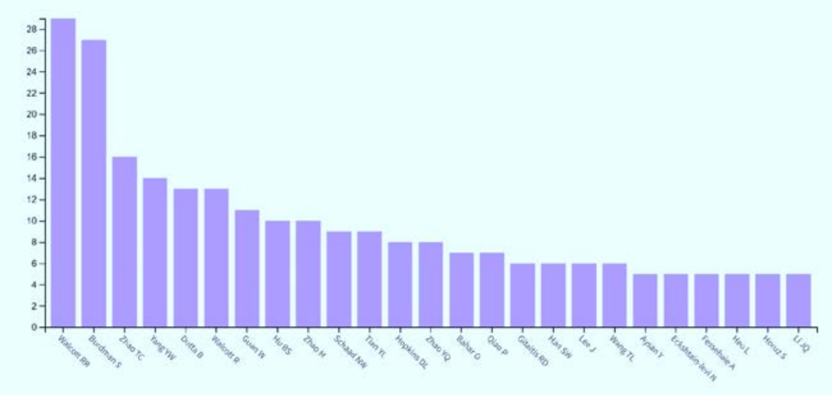
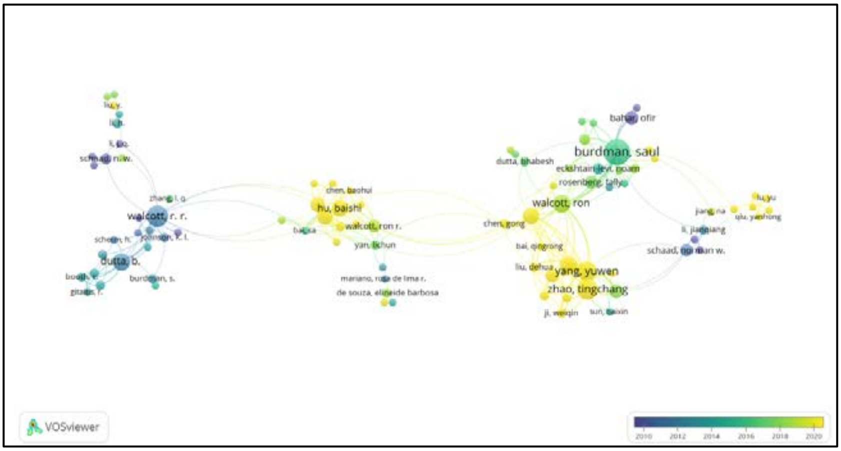
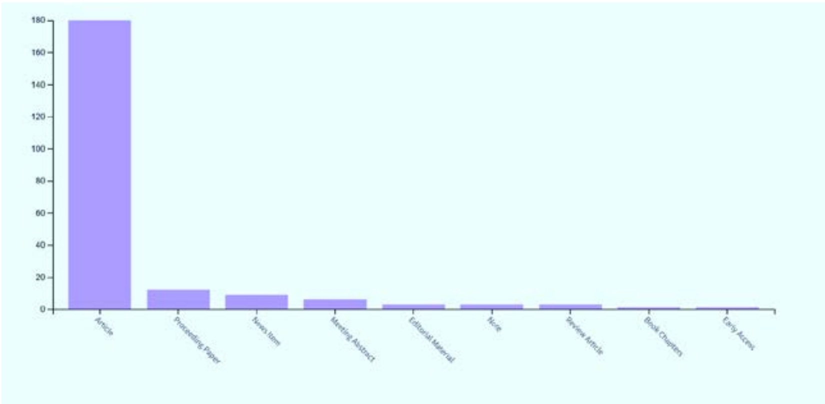



**Figure 6. Analysis of the collaboration of authors publishing on the biological control of *Acidovorax citrulli***




When the studies on biological control of *Acidovorax citrulli* were evaluated, it was seen that most documents were published as research articles and approximately 85% of the 212 publications were research articles (Figure 7).

**Figure 7. Distribution of publications on the subject according to document types**




Hypersensitivity tests were performed on tobacco for all 189 bacterial strains used as candidate antagonist bacteria and it was determined that none of the tested bacterial strains caused hypersensitivity reactions. No pectolytic activity was observed in any of the tested bacterial strains. Antibacterial activities of these strains against *Acidovorax citrulli* were determined by in vitro dual culture tests. It was determined that 17 of the strains showed antibacterial activity against the pathogen and the effect of the application on pathogen development was found to be statistically significant. The statistical analysis results of the zone values formed by the bacterial strains against the pathogen are presented in Table 3.

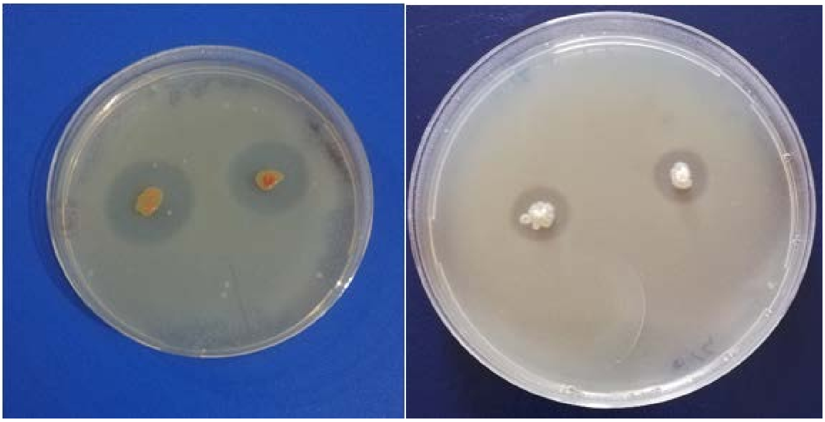
**Table 3. Effect of antagonist bacterial strains on pathogen development *in vitro***

| Strain No | FAME Identification Result | Zone value (mm) |
|---|---|---|
| **IT 14** | *Bacillus amyloliquefaciens* | 28,26±0,14[a] |
| **IT 58** | *Variovorax paradoxus* | 10,00±0,28[ıj] |
| **IT 70** | *Pseudomonas fluerescens* | 14,76±0,14[de] |
| **IT 115** | *Serratia rubidaea* | 13,26±0,14[ef] |
| **IT 156** | *Bacillus subtilis* | 21,26±0,43[c] |
| **IT 159** | *Bacillus subtilis* | 24,76±0,14[b] |
| **IT 162** | *Bacillus subtilis* | 15,26±0,43[d] |
| **IT 188** | *Pseudomonas fluorescens* | 23,76±0,72[b] |
| **IT 189** | *Bacillus atrophaeus* | 12,0±0,28[f-h] |
| **2/2** | *Bacillus atrophaeus* | 14,26±0,14[de] |
| **3/2** | *Bacillus thuringiensis* | 10,5±0,57[hı] |
| **3/5** | *Bacillus subtilis* | 8,76±0,14[j] |
| **3/9** | *Bacillus subtilis* | 12,0±0,86[fgh] |
| **4/2** | *Bacillus pumilus* | 14,26±0,14[de] |
| **6/4** | *Bacillus atrophaeus* | 12,5±0,00[fg] |
| **6/5** | *Bacillus atrophaeus* | 10,5±0,28[hı] |
| **8/3** | *Bacillus viscosus* | 11,26±0,14[g-ı] |
| **F Değeri** | | 236,47 |

Data are the average of three replications and there is no statistical difference between values indicated with the same letter in the same column ($P < 0.01$).

When the data in Table 3 are examined, it is seen that the strains prevent pathogen development at varying rates, and *Bacillus amyloliquefaciens* strain IT 14 significantly inhibits pathogen development. Following this strain, the largest inhibition zone was measured as a result of the application of *Bacillus subtilis* strain IT 159. The lowest antibacterial activity against the pathogen among the strains was observed in the application of *Bacillus subtilis* strain 3/5 (Photo 1).

**Photo 1. Effect of antagonist bacteria on pathogen development *in vitro***



## 4. CONCLUSION

evaluated, it was seen that the studies on the determination of local potential biological control elements for use in the biological control of *Acidovorax citrulli*, the bacterial fruit spot agent that causes significant yield losses in Turkey, were insufficient. Therefore, this study is important in terms of preparing the ground for future studies on the determination of effective control elements against pathogens isolated from existing sources in Turkey. When the study was evaluated based on the results of in vitro dual culture tests, it was shown that 17 strains showed antibacterial activity against the pathogen, and among the strains, *Bacillus amyloliquefaciens* strain IT 14, *Bacillus subtilis* strain IT 156, Bacillus subtilis strain IT 159 and *Pseudomonas fluorescens* strain IT 188 have significant potential as biological control agents.

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# PHEROMONES IN ACARI AND USAGE POSSIBILITIES[1]

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**Ramazan AKTÜRK[3]**

**Sibel YORULMAZ[4]**

## 1. INTRODUCTION

Pheromones are specific compounds that enable chemical communication between individuals within the same species and trigger specific behavioral or physiological responses (Wyatt, 2014). First identified by Karlson and Lüscher (1959), these chemical signals play fundamental roles in a wide variety of biological processes in organisms, such as reproduction, defense, feeding, and social organization. Although the existence and roles of pheromones in insects have long been investigated, pheromone-based communication systems in other arthropod groups, such as mites (Acari), are a relatively new area of research (Basu et al., 2021).

The subclass Acari is a member of the class Arachnida and represents an extremely diverse group with approximately 55,000 described species (Walter and Proctor, 1999). Mites with

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different ecological roles such as agricultural pests [*Tetranychus urticae* (Acari: Tetranychidae)], storage pests [*Rhizoglyphus robini* (Acari: Acaridae)], human and animal parasites [*Dermanyssus gallinae* (Acari: Dermanyssidae)] and beneficial species used in biological control [*Phytoseiulus persimilis* (Acari: Phytoseiidae)] are included in this group. Pheromones stand out as a critical communication tool in the life cycle of these species and in their interactions with the environment and each other (Walter and Proctor, 1999; Zeeman et al., 2022).

Among mites, pheromones play a role in many processes such as regulating sexual behavior (Zeeman et al., 2022), providing social organization (Benoit et al., 2004), organizing the defense of the colony against dangers (Basu et al., 2021) and sharing food resources (Kuwahara, 2004). The fact that the microhabitats where mites live are generally dark, humid and have limited visual signals has made chemical communication even more important (Walter and Proctor, 1999; Basu et al., 2021).

## 2. CLASSIFICATION AND GENERAL PROPERTIES OF PHEROMONES IN ACARI SPECIES

In Acari, pheromones are generally divided into six basic groups according to their mechanism of action: alarm, sexual, aggregation, trail, epideictic and dispersing pheromones (Wyatt, 2014).

### 2.1. Alarm Pheromones

Alarm pheromones are secreted when individuals are threatened and usually trigger defensive behaviors such as escape, attack or hiding in colony members, and sometimes cause dispersal (Amrine et al., 2003; Basu et al., 2021). As in some social insects, alarm pheromones play important roles in warning the colony in mite species.

### 2.2. Sex Pheromones

Sex pheromones are generally secreted by females and trigger male mating behavior. Since these pheromones are species specific, they play an important role in providing intraspecific communication (Kuwahara, 2004). For example, in *R. robini*, the α-acaridial compound secreted by females initiates mating behavior in males, and some males even produce this pheromone by imitating females, indicating alternative reproductive strategies (Zeeman et al., 2022).

### 2.3. Aggregation Pheromones

These pheromones encourage individuals to gather in a certain area and can attract both males and females. Aggregation behavior can facilitate social organization, feeding or mating (Vaníčková et al., 2017). Both inter- and intra-sexual aggregation signals have been detected in some mite species (Kuwahara, 2004). For example, the compound neryl formate, which attracts both males and females in the species *Dermatophagoides pteronyssinus* (Acari: Pyroglyphidae), shows a strong aggregation effect (Skelton et al., 2010). Additionally, the presence of different aggregation pheromones has been detected in species such as *Lardoglyphus konoi* (Acarina: Acaridae) and *Chortoglyphus arcuatus* (Sarcoptiformes: Chotoglyphidae) (Akyazı and Altunç, 2018).

### 2.4. Trail Pheromones

These pheromones, which are generally seen in social species, allow individuals to leave chemical traces in the areas they move. They are especially used to mark paths leading to food sources (Kuwahara, 2004). In creatures that are small and have limited mobility, such as mites, such pheromones can facilitate navigation within the habitat. For example, in studies conducted on the species *Phytoseiulus macropilis* (Acari: Phytoseiidae) and *Neoseiulus fallacis* (Acari: Phytoseiidae), it was observed that the chemical traces left by these individuals during their prey search behavior were perceived and followed by other individuals of the same species. This situation reveals that these species use trail pheromones for social guidance (Hislop and Prokopy, 1981, Akyazı and Altunç, 2018).

### 2.5. Epideictic Pheromones

These pheromones are generally used to mark spawning areas and prevent excessive density. These signals left by females prevent other females from laying eggs in the same area (Wyatt, 2014). Such pheromones can reduce resource competition in areas where mites are densely populated. For example, in studies conducted on the species *Tyrophagus putrescentiae* (Acari: Acaridae), it was observed that the chemical traces left by females in the environment after spawning prevented other individuals from laying eggs in the same area. This situation reveals that epideictic pheromones function as a signal that regulates space and food sharing (Kuwahara et al., 1980).

### 2.6. Dispersal Pheromones

These pheromones, which are often seen as a sub-function of alarm pheromones, enable individuals to disperse rapidly (Shimizu et al., 2001). They are important in the rapid response of the colony to danger (Basu et al., 2021). For example, in the *T. putrescentiae* species, it has been observed that the pheromones secreted by individuals under stress trigger dispersal behavior in colony individuals. One of these components, neryl formate, acts as a dispersal pheromone by causing the colony to disperse in a short time when a threat perception occurs in the environment (Kuwahara et al., 1980, Akyazı and Altunç, 2018).

## 3. PHEROMONE COMPOUNDS DETERMINED IN ACARI SPECIES

In Acari species, pheromones are generally secreted through opisthonotal glands, and these glands can produce both sexual and alarm and aggregation pheromones simultaneously (Kuwahara, 2004). As a result of chemical analyses, many unique pheromone compounds belonging to different species have been identified (Table 1).

**Table 1. Pheromone Compounds in Important Acari Species.**

| **Pheromon Compound** | **Species** | **Pheromone Type** | **References** |
|---|---|---|---|
| α-acaridial | *Rhizoglyphus robini* (Acari: Acaridae) | Sex | Zeeman et al. (2022) |
| β-acaridial | *Tyrophagus longior* (Acari: Acaridae) | Alarm/ Sex | Kuwahara et al. (1980) |
| γ-acaridial | *Rhizoglyphus sp.* (Astigmata: Acaridae) | Sex | Murakami et al. (2006) |
| Neral | *Carpoglyphus lactis* (Astigmata, Carpoglyphidae), *Oulenzia sp.* (Astigmata: Winterschmitiidae), | Alarm | Kuwahara et al. (1980); Shimizu et al. (2004) |
| Geranial | *Histiostoma laboratorium* (Astigmata: Histiostomidae) | Alarm | Kuwahara et al. (1992) |
| 2-Hydroxy-6-methylbenzaldehyde | *Acarus immobilis* (Astigmata: Acaridae), *Aleuroglyphus ovatus* (Astigmata: Acaridiae) | Sex | Kuwahara et al. (1992) |
| S-(+)-Isorobinal | *Rhizoglyphus setosus* (Astigmata: Acaridae) | Sex | Mizoguchi et al. (2005) |
| Rosefuran | *Caloglyphus sp.* (Astigmata: Acaridae) | Sex | Mori et al. (1998) |
| Neryl formate | *Schwiebea sp.* (Acari: Acaridae), *Dermatophagoides pteronyssinus*(Acari: Pyroglyphidae) | Aggregation /Alarm | Skelton et al. (2010) |
| (2R,3R)-Epoxyneral | *Caloglyphus sp.* (Astigmata: Acaridae) | Sex | Mori and Kuwahara (1995) |
| S(+)-Isopiperitenone | *Tyrophagus similis* (Astigmata: Acaridae) | Alarm | Kuwahara et al. (1987) |

## 4. PHEROMONE SECRETION REGIONS IN ACARI

**Production Sites: Opisthonotal Glands:**

In most mites, pheromone production is carried out by opisthonotal glands or similar special secretory structures located in the opisthonotum region. These glands are activated during the mating period, especially in astigmatid mite species, and secrete pheromone (Kuwahara, 2004). For example, in *R. robini*, the α-acaridial compound is synthesized intensively in the opisthonotal gland and is released into the external environment to attract the attention of individuals of the opposite sex (Zeeman et al., 2022). Similarly, in species such as *Caloglyphus sp*., pheromone synthesis and secretion are directly related to the gland systems in the posterior part of the body (Mori et al., 1998). Opisthonotal glands generally work with enzymatic mechanisms that can volatilize compounds in liquid form. This feature allows the effective spread of chemical signals in the microscopic environment where the mite lives.

## 5. PHEROMONE DETECTION MECHANISMS IN ACARI

### 5.1. Sensory Structures and Sensilla

Chemoreceptive sensilla located on the tarsus, palpus and body surfaces of mites play a role in the recognition of volatile compounds. Pores located within these structures take in volatile chemicals from the environment and initiate signal transmission. Especially in species living in microhabitats such as astigmatid mites, these structures have evolved to be extremely sensitive (Kuwahara, 2004).

In some mite species, the single sensillum on the palpaltibiotarsal segment may play a role in the perception of sex pheromones. For example, in the species *R. robini*, it has been shown that male individuals are directed towards α-acaridial-containing traces left by females (Zeeman et al., 2022).

### 5.2. Molecular Sensors: OBP and OR Proteins

After the chemical signals of pheromones are perceived, they are processed through various odorant-binding proteins (OBP) and odorant receptors (odorantreceptors – OR) before being transmitted to the nervous system. This system, which is common in insects (Zhang et al., 2014), is also increasingly found to exist at the molecular level in mite species (Basu et al., 2021).

### 5.3. Neural Transmission and Behavioral Response

The electrical signal resulting from pheromone perception is transmitted from peripheral sensory neurons to the central nervous system. Various motor responses are generated here: escape, mating search, gathering, etc. (Basu et al., 2021). Especially in alarm pheromones, the signal compound released in high concentrations in a short time is detected via sensilla and escape or defensive behaviors are initiated in individuals. This rapid perception-response relationship is an evolutionary advantage that mites have developed against threats.

Although mites do not have antennae that directly detect pheromones, they can give extremely sensitive responses to chemical signals thanks to specialized tarsal and palpal sensilla, OBP/OR proteins, and fast neural transmission mechanisms.

The pheromone perception mechanism in Acari consists of 6 stages:

1. Pheromone Secretion → Pheromone compound is released into the environment (e.g. α-acaridial, neral).
2. Sensory Reception → Pheromone is taken in through pores on tarsal or palpal sensilla.

3. OBP Binding → Odorant Binding Protein (OBP) carries the molecule to the receptor.
4. OR Activation → Odorant Receptor (OR) recognizes the pheromone and produces a signal.
5. Neural Transmission → Signal is processed through the nervous system.
6. Behavioral Response → Responses such as escape, mating, and aggregation occur (Walter and Proctor, 1999; Basu et al., 2021; Zeeman et al., 2022).

## 6. BIOASSAY METHODS BASED ON PHEROMONES IN ACARI

One of the most common methods for determining the biological effects of pheromones and evaluating their practical usability is bioassay applications. These methods aim to observe and measure the behavioral or physiological response of the target organism to a specific chemical (Borges and Aldrich, 1992). Although difficult to apply on small and fast-moving organisms such as mites, bioassay systems designed under the right conditions can clearly reveal the responses of these organisms to chemical signals (Wyatt, 2014).

### 6.1. Laboratory Bioassays

Bioassays performed under laboratory conditions are used to determine whether pheromone compounds are attractive, repulsive or ineffective under controlled conditions. Some techniques specially adapted for mites are as follows:

#### 6.1.1. Petri Dish Choice Tests

Mites are placed in a petri dish to choose between a pheromone-treated surface and a control surface. The direction and duration of movement of individuals are observed.

#### 6.1.2. Y-Tube Olfactometer

Mites are placed in a Y-shaped glass tube between two different currents, one of which is the air current containing pheromone. The direction they head is recorded (Noge et al., 2005; Çakmak et al., 2006).

#### 6.1.3. Video-Assisted Behavioral Observation Systems

Under the influence of pheromone, the movement speed, orientation degree and timing behaviors of individuals are analyzed by monitoring with high-resolution cameras (Zeeman et al., 2022).

## 7. THE PLACE OF ACARI PHEROMONES IN INTEGRATED PEST MANAGEMENT

Today, pheromones have gained an important place in biotechnical and biological control methods developed as an alternative to chemical control. While insect pheromones are widely used in agricultural production (El-Sayed et al., 2006; Ahmad and Kamarudin, 2011), studies on mite pheromones are still limited but promising (Benelli et al., 2019).

### 7.1. Monitoring and Density Determination

One of the most important potential areas of use of mite pheromones is the early detection and monitoring of population density. For example, by using α-acaridial-based traps in storage pests such as *R. robini*, it may be possible to take precautions before the population increases (Zeeman et al., 2022). This approach can make pesticide application decisions more accurate.

### 7.2. Attractant-Trapping Systems

Synthetic derivatives of sex or aggregation pheromones can be used in traps to attract pests and suppress their populations. Especially:

- β-acaridial in storage mites such as *T. longior*,
- neryl formate in *D. pteronyssinus*,
- *Caloglyphus sp*. species stand out as suitable targets for behavioral manipulation (Kuwahara, 2004; Skelton et al., 2010).

### 7.3. Mating Disruption

Preventing males from finding females by spreading sex pheromones into the environment is known as the "mating disruption" technique. This technique has been successfully applied in some insect species. Although there is no commercialized example for mites yet, laboratory studies are promising (Vaníčková et al., 2017). Artificial dispersion of the compound S-(+)-isorobinal, especially used in species such as *R. setosus*, can affect male behavior and reduce mating rates (Mizoguchi et al., 2005).

## 8. CONCLUSION AND FUTURE PERSPECTIVES

Acari, in terms of their ecological roles and economic importance, include agricultural pests, biological control agents, and even species that affect human health (Carr and Roe, 2016). This diversity makes it critical to understand complex chemical communication systems such as pheromones at both evolutionary and applied levels (Walter and Proctor, 1999). Recent studies have shown that sex, alarm, aggregation, and trail pheromones are common among mites, and these pheromones are mostly secreted through opisthonotal glands (Kuwahara, 2004). The identified compounds are not limited to sexual communication, but are involved in many behavioral processes such as ensuring social organization, sharing threat perception, and regulating intraspecific competition (Basu et al., 2021; Zeeman et al., 2022). The role of chemical signals in interactions between species (e.g. kairomone effect) reveals that these compounds are decisive not only for one species but for the entire habitat. Therefore, pheromones have a strategic importance both in resolving ecological interactions and in developing biotechnical applications.

### 8.1. Future Perspectives

- Pheromone Discovery in New Species: There may be many chemical signals that have not yet been fully identified in agriculturally important families such as Tetranychidae and Phytoseiidae. Molecular and metabolic studies in this area should be increased.
- Synthesizable and Stabilizable Analogs: Artificial production of natural pheromones and their application to the environment in stable forms may form the basis of commercial biotechnical solutions.
- Pheromone-Predator Mite Pairings: Mixed pheromone mixtures (e.g. sex + alarm) that direct natural enemies to target pests may increase the effectiveness of biological control.
- Use as a pesticide alternative: Pheromone-based monitoring and guidance systems may offer an environmentally friendly and sustainable solution in integrated control strategies by reducing pesticide use.

Acari pheromones offer both a deep field for basic scientific research and pave the way for innovative solutions in the fields of agriculture, storage and public health. This powerful tool offered by chemical ecology plays a key role in understanding not only mites but also the microsystems in which they live. In conclusion, the pheromone compounds identified in Acari species are biological tools that can be evaluated not only in the regulation of basic behaviors but also in sustainable pest management practices. The central role played by opisthonotal glands in this production lies at the intersection of both evolutionary adaptation and applied entomology.

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# AKADEMİK PERSPEKTİFTEN

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