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Alternative Approaches in the Control of Grape Bud Mite *Eriophyes vitis* (Pgst., 1859) (Acarina: Eriophyidae)

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Abstract
This study aimed to evaluate the effectiveness of three different compounds and one beneficial fungus as alternative control agents for Grape Bud mite *Eriophyes vitis* **(**Pgst., 1859), which causes significant damage in vineyards, under field conditions. In vineyards identified as infested, various doses of Kaligreen fungicide (82% Potassium Bicarbonate), the *Taloromyces funiculosus* st976 strain, Pascal+ Copper compound, and traditional wettable sulphur (WP) were applied. The trial was established following the Ministry of Agriculture and Forestry's Standard Pesticide Testing Method for *Eriophyes vitis* (Grape Leaf Blister Mite) Control. Pre-application leaf scale values for the pest were recorded, and statistical differences among applications were assessed on days 1, 7, 14, 21, 28, and 36 post-application. On day 7, Kaligreen and *Taloromyces funiculosus* st976 strain were found to be significantly more effective against the pest than Pascal applications. By day 14, the *T. funiculosus* st976 strain, Kaligreen, and traditional wettable sulphur treatments were significantly more effective than the Pascal+ Copper compound. On day 21, *T. funiculosus* st976 was significantly more effective than traditional sulphur on leaves. By day 28, *T. funiculosus* st976 and Kaligreen applications were significantly more effective than traditional wettable sulphur. By day 36, Kaligreen was significantly more effective than Pascal+ Copper on leaves. Overall, *Taloromyces funiculosus* st976 strain and Kaligreen outperformed traditional wettable sulphur and the Pascal +Copper compound in effectiveness. In terms of phytotoxicity, *T. funiculosus* st976 strain and traditional wettable sulphur did not cause toxicity on leaves, whereas Kaligreen and the Pascal+ Copper compound occasionally caused slight to moderate phytotoxicity. The results of this study contribute to the alternative approaches for integrated pest management (IPM) in vineyards for controlling grape leaf blister mite. Based on literature, this study is the first global record to establish the effectiveness of *Taloromyces funiculosus* ST976 strain against *Eriophyes vitis* (Grape Bud Mite).

Keywords: *Eriophyes vitis*, Kaligreen, *Taloromyces funiculosus* st 976 strain, effectiveness, vineyard

Introduction
The history of viticulture is quite ancient globally. Türkiye, situated in an ideal climate zone for viticulture, has a deeply rooted production tradition. Grapes belong to the order *Rhamnales*, family *Vitaceae*, and genus *Vitis*, known for their climbing growth habit. Based on global grape production figures from 1994-2016, Turkiye ranked 6th, following the United States and Spain, with a total production of 87,118,427 tons. According to 2016 FAO data, Turkiye ranked 6th in the world, with a production of 4,000,000 tons, while China led with 14,763,000 tons (Anonymous, 2020) [1]. Additionally, regions where grape cultivation is challenging have seen an increased demand for pickling vine leaves, a growing production model within viticulture due to its high yield per area, suitability for family farming, and relatively low maintenance and costs (Cangi *et al*., 2012) [3]. For pickled vine leaves, preferred characteristics include thin leaves with minimal hair and lobbing, as seen in Türkiye's Narince and Sultani Seedless grape varieties.

Among the main pests of vine leaves from Narince grapes is the grape leaf blister mite (*Colomerus vitis* Pagenstecher (Acarina: Eriophyidae). Recorded as early as 1680 in Italy, this pest has become a problem for vineyards in Europe, America, Africa and Australia. In Turkiye, *C. vitis* is commonly found in Central Anatolia, Southeast Anatolia, and the Aegean regions (Yazıcı and Yeşilayer, 2020) [4].

The primary area of damage by *C. vitis* is the leaves, where nymphs and adults settle on the undersides, feeding on the sap. After feeding, dimples form under the leaves and corresponding bumps above; these dimples eventually turn brown and develop a hairy tissue called *erineum*. High pest density negatively affects the quality of preferred vine leaves.
Sulphur applied to control powdery mildew often keeps this pest in check without additional pesticides, though chemical control is used if population density increases or resistance to sulphur occurs. In such cases, sulphur sprays may be supplemented with other pesticides in spring, depending on early damage signs. Overuse of systemic fungicides against the pest in recent years has led to increased pest populations, making alternative control strategies essential. Among these strategies, farmers frequently use sulphur-based compounds and copper-containing formulations in combination with traditional wettable sulfur, the potassium-based fungicide Kaligreen, and the fungus *Taloromyces funiculosus* st976 strain, which is biotechnologically important due to its enzyme production, pigment synthesis, and ability to mobilize phosphorus in soil. These substances were tested for their field effectiveness in combating *Eriophyes vitis* (Grape bude mite) (Türkölmez *et al*., 2022) [6].

2. Materials and Methods

This study made use, as an experimental material, a vineyard of the Öküzgözü grape variety which was affected by the grape leaf blister mite as shown from (Figure 1). The vineyard consisted of vines of the same age and variety. The randomized block design with four blocks was used for the experiment. Each plot consisted of at least 4x4=16 vines. Data was collected on four vines located at the centre of each plot. A 10-litre battery-powered automatic sprayer pump containing four different compounds was used to administer treatments.
Care was taken to thoroughly spray all green parts of the vines. Uniform coverage was ensured across the test area and specific regions (e.g., branches, trunks) as required. The compounds used in the study included Kaligreen (potassium salt), the fungus *Taloromyces funiculosus* strain, a formulation containing Pascal/Copper, and traditional sulphur dust. Traditional sulphur dust served as the control group. Kaligreen was applied with the concentration of 9 g/10L of water, the *T. funiculosus* st976 solution was applied at 45 mL/10L of water. The traditional wettable sulphur powder containing 73% sulphur was mixed at 50 g/10L of water. Pascal 20 SP containing 20% Acetamiprid was used at a concentration of 2 g/10L of water along with Copper oxychloride, equivalent to 50% metallic copper, with a concentration of 30 g/10L of water.

Fig 1: Vineyard Area Where the Study Was Conducted, Application Process, and Infected Leaf

Data collection was carried out on a total of 100 leaves, with 25 randomly selected leaves from the third leaf above the base of the shoots around each vine, across four vines (Figure 2). The evaluation scale for leaf damage caused by grapevine mite is provided in Table 1. All leaves were initially rated at a scale of 2, and statistical analyses were performed based on changes to the leaves over time following application of treatments with reference to the damage scale.

Fig 2: Field counting and evaluation for grapevine bud mite *Eriophyes vitis* Pgst.

Table 1: Evaluation Scale for Grapevine Bud Mite Damage on Leaves

Scale Value	Damage Description
0	No blister mite symptoms on the leaf
1	Blister mite symptoms in 1-2 spots on the leaf
2	Blister mite symptoms in 3-10 spots on the leaf
3	Blister mite symptoms in more than 10 spots on the leaf

To determine the phytotoxic effects of the compounds used on leaves, samples were collected post-application on days 1, 7, 14, and 21. A total of 10 leaves per application were brought to the laboratory and examined for any phytotoxic effects on the leaf surfaces using an Olympus SZX 7 stereomicroscope. In total, 10 leaves were examined each day, amounting to 40 leaves per application on counting days. Additionally, a modified scale based on the powdery mildew scale as shown in Table 2 was used for the phytotoxicity assessment (Balıç, 2005) [2].

Table 2: Phytotoxicity Evaluation Scale

Scale Value	Phytotoxicity Description
0	No spots on the leaf
1	1-2 spots present on the leaf
2	3-10 spots present on the leaf
3	More than 10 spots present on the leaf

2.1. Statistical Applications: The Shapiro-Wilk Normality Test and the Kolmogorov-Smirnov Test were used to test normality of data collected. Since the normality assumption was not met, the Kruskal-Wallis Analysis was employed for comparisons between independent groups, and the Friedman Analysis was used for comparing dependent measurements over time. Descriptive statistics such as mean, median, and standard deviation were used to describe the data in this study. Statistically significance differences levels were recorded at $p<0.05$. Statistical analyses were performed using SPSS (Statistical Package for Social Sciences; SPSS Inc., Chicago, IL) version 22.

3. Results and Discussion

Table 1: Comparison of Applications via Kruskal-Wallis Analysis

	Groups	N	Mean	SS	Rank Mean	H	P Value	Pairwise Comparisons
	Kaligreen	100	2.00	0.00	200.50	0.000	1.000	
	Pascal	100	2.00	0.00	200.50			
	Traditional Sulphur Powder	100	2.00	0.00	200.50			
	T. funiculosus st976 strain	100	2.00	0.00	200.50			
Day 1	Kaligreen	100	1.98	0.14	199.50	2.025	0.567	
	Pascal + Copper	100	1.98	0.14	199.50			
	Traditional Sulphur Powder	100	1.98	0.14	199.50			
	T. funiculosus st976 strain	100	2.00	0.00	203.50			
Day 7	Kaligreen	100	0.69	0.46	178.91	31.218	<0.001	*T. funiculosus* < Pascal, Kaligreen < Pascal
	Pascal + Copper	100	1.05	0.63	238.81			
	Traditional Sulphur Powder	100	0.85	0.36	209.15			
	T. funiculosus st976	100	0.67	0.47	175.13			
Day 14	Kaligreen	100	0.53	0.50	188.21	66.126	<0.001	*T. funiculosus* < Pascal, Kaligreen < Pascal, G. Sulphur < Pascal
	Pascal + Copper	100	0.93	0.36	265.18			
	Traditional Sulphur Powder	100	0.56	0.50	194.16			
	T. funiculosus st976 strain	100	0.36	0.48	154.46			
Day 21	Kaligreen	100	0.36	0.48	193.50	10.226	0.017	*T. funiculosus* < G. Sulphur
	Pascal + Copper	100	0.43	0.50	207.50			
	Traditional Sulphur Powder	100	0.50	0.50	221.50			
	T. funiculosus st976 strain	100	0.29	0.46	179.50			
Day 28	Kaligreen	100	0.08	0.27	180.50	21.761	<0.001	*T. funiculosus* < G. Sulphur, Kaligreen < G. Sulphur
	Pascal + Copper	100	0.19	0.39	202.50			
	Traditional Sulphur Powder	100	0.32	0.47	228.50			
	T. funiculosus st976 strain	100	0.13	0.34	190.50			
Day 36	Kaligreen	100	0.06	0.24	186.50	11.466	0.009	Kaligreen < Pascal
	Pascal + Copper	100	0.20	0.40	214.50			
	Traditional Sulphur Powder	100	0.17	0.38	208.50			
	T. funiculosus st976 strain	100	0.09	0.29	192.50			

From Table 1, statistically significant differences among the applications can be observed on the 7th, 14th, 21st, 28th, and 36th days ($p<0.05$). On the 7th day, the applications of Kaligreen and the *T. funiculosus* st976 strain were found to be significantly more effective on the leaves compared to the Pascal application. By the 14th day, *T. funiculosus* st976 strain, Kaligreen, and traditional wettable sulphur applications also showed significantly greater effectiveness on the leaves than the Pascal treatment (see Figure 3). On the 21st day, the *T. funiculosus* application was significantly more effective than the traditional sulphur application. On the 28th day, both *T. funiculosus* st976 strain and Kaligreen were significantly more effective on the leaves compared to the traditional wettable sulphur application. Finally, on the 36th day, the Kaligreen application was again found to be significantly more effective compared to the Pascal treatment.

Fig 3: The effectiveness of the *T. funiculosus* st976 strain application on the 14th day (Inactivity of the pest/renewal of the damaged leaf).

Table 2: Comparison of measurement times using the dependent samples Friedman Test

	Groups	N	Mean	SS	Rank Mean	Chi-Square	P Value
Kaligreen	Before Application	100	2.00	.00	6,51	490.376	<0.001
	1. day	100	1.98	.14	6,48		
	7. day	100	.69	.46	3,87		
	14. day	100	.53	.50	3,47		
	21. day	100	.36	.48	3,05		
	28. day	100	.08	.27	2,34		
	36. day	100	.06	.24	2,29		
Pascal+ Sulphur	Before Application	100	2.00	.00	6,39	477.460	<0.001
	1. day	100	1.98	.14	6,35		
	7. day	100	1.05	.63	4,22		
	14. day	100	.93	.36	3,93		
	21. day	100	.43	.50	2,75		
	28. day	100	.19	.39	2,19		
	36. day	100	.20	.40	2,19		
Traditional Sulphur Powder	Before Application	100	2.00	.00	6,51	476.974	<0.001
	1. day	100	1.98	.14	6,47		
	7. day	100	.85	.36	3,93		
	14. day	100	.56	.50	3,21		
	21. day	100	.50	.50	3,05		
	28. day	100	.32	.47	2,61		
	36. day	100	.17	.38	2,23		
T. funiculosus st 976 strain	Before Application	100	2.00	.00	6,50	488.112	<0.001
	1. day	100	2.00	.00	6,50		
	7. day	100	.67	.47	3,91		
	14. day	100	.36	.48	3,13		
	21. day	100	.29	.46	2,96		
	28. day	100	.13	.34	2,56		
	36. day	100	.09	.29	2,46		

Upon examining Table 2, statistically significant differences between the measurement times in the applications of Kaligreen, Pascal, Traditional Powder Sulphur, and *T. funiculosus* st976 strain are noted ($p<0.05$).

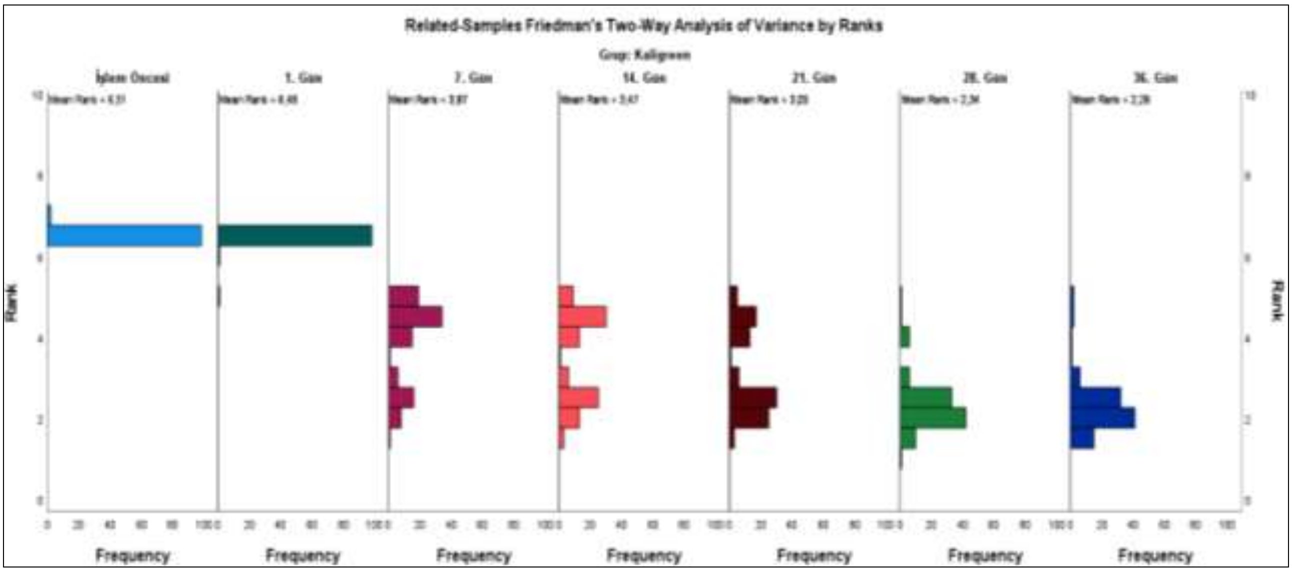

Fig 3: Pairwise Comparisons of Kaligreen Applications by Day

The pairwise comparisons for Kaligreen applications on the 36th day shows that leaf damages were significantly reduced more effectively compared to the pre-treatment, as well as observations on days 1, 7, and 14. Similarly, on the 28th day, leaf damages were significantly reduced more effectively than those taken before treatment, as well as on days 1, 7, and 14. On the 21st day, the leaf damages were significantly reduced more effectively compared to pre-treatment and the 1st day measurements. Additionally, the measurements on the 14th day were found to be significantly more effective compared to pre-treatment and the 1st day measurements. On the 7th day, the leaf damage measurements were significantly more effective than those taken before treatment, as well as on the 1st and 7th day measurements ($p<0.05$) (Figure 3).

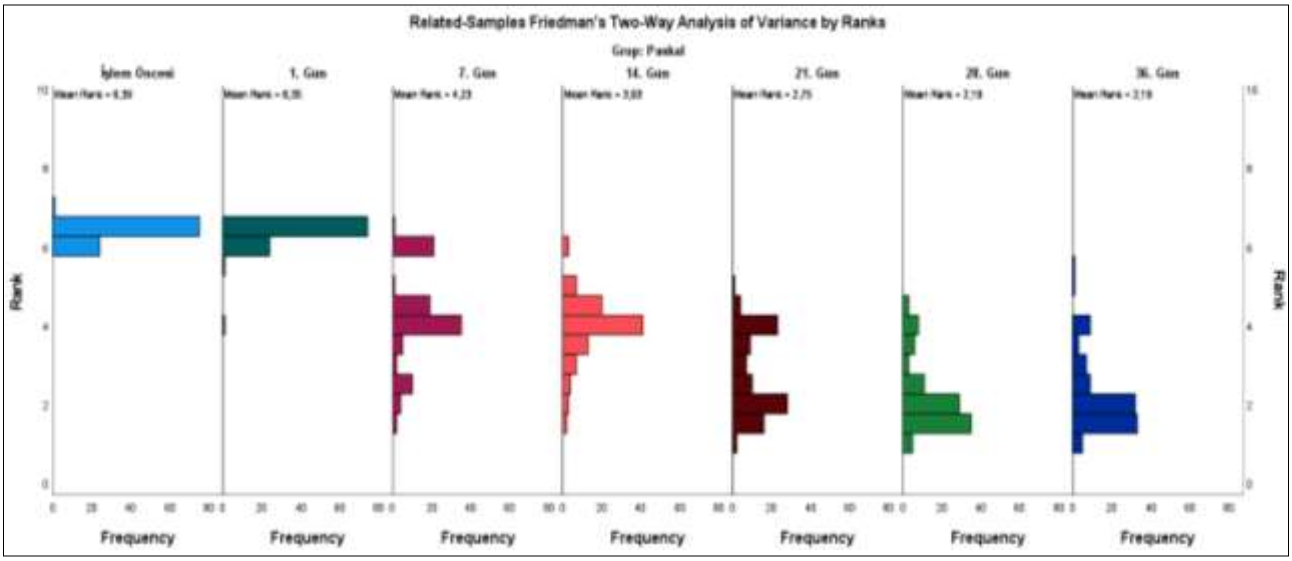

Fig 4: Pairwise Comparisons of Pascal+ Copper Applications by Days

The pairwise comparisons for Pascal+ Copper applications on the 36th day shows that leaf damage measurements were significantly more effectively reduced compared to pre-treatment, as well as the measurements taken on days 1, 7, and 14. On the 28th day. Similarly, on the 21st day, the leaf damage measurements were significantly more effectively reduced than those taken before treatment, as well as on days 1, 7, and 14. On the 14th day, the leaf damage measurements were significantly more effective compared to the measurements taken on the 1st day. Finally, on the 7th day, the leaf damage measurements were significantly more effective compared to pre-treatment and the 1st day measurements ($p<0.05$) as can be observed on (Figure 4).

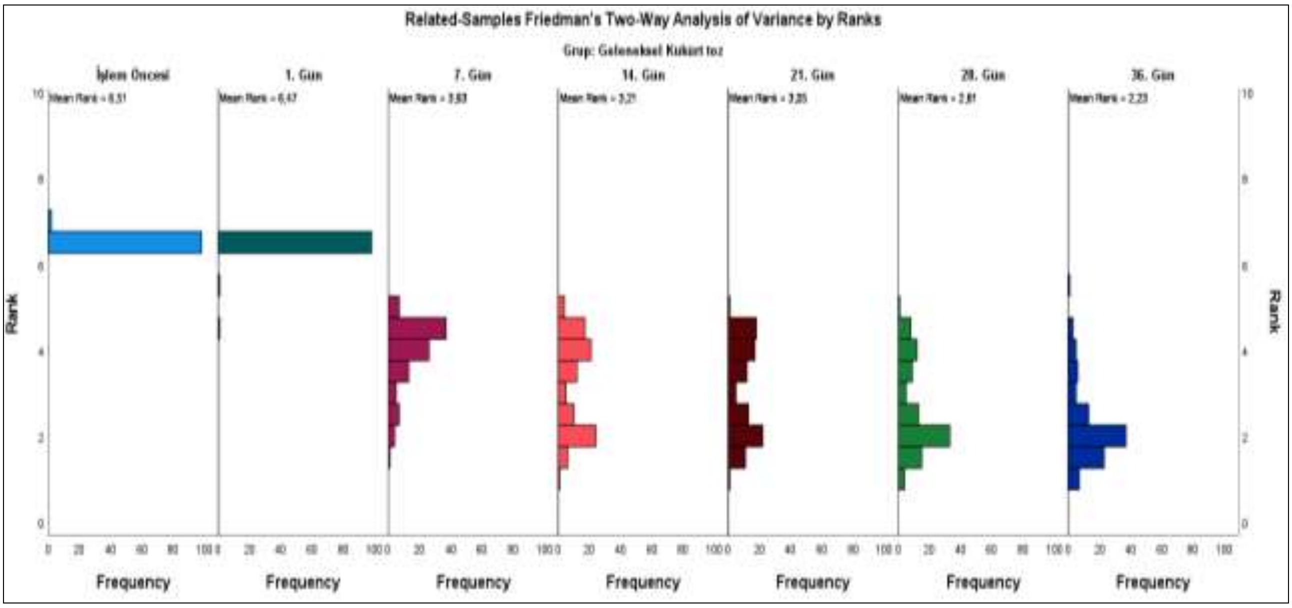

Fig 5: Pairwise Comparisons of Traditional Wettable Powder Sulphur Applications by Days

When the pairwise comparisons of traditional wettable powder sulphur applications was examined on the 36th day, the leaf damage measurements were significantly more effectively reduced compared to pre-treatment, as well as the measurements taken on days 1, 7, and 14 days. On the 28th day, the leaf damage measurements were significantly more effectively reduced compared to those taken before treatment and on days 1 and 7. Similarly, on the 21st day, the leaf damage measurements were significantly more effectively reduced than those taken before treatment, as well as on days 1, 7, and 14. On the 14th day, the leaf damage measurements were significantly reduced compared to the measurements taken on the 1st day. Finally, on the 7th day, the leaf damage measurements were significantly more effective compared to pre-treatment and the 1st day measurements ($p<0.05$) as can be observed on (Figure 5).

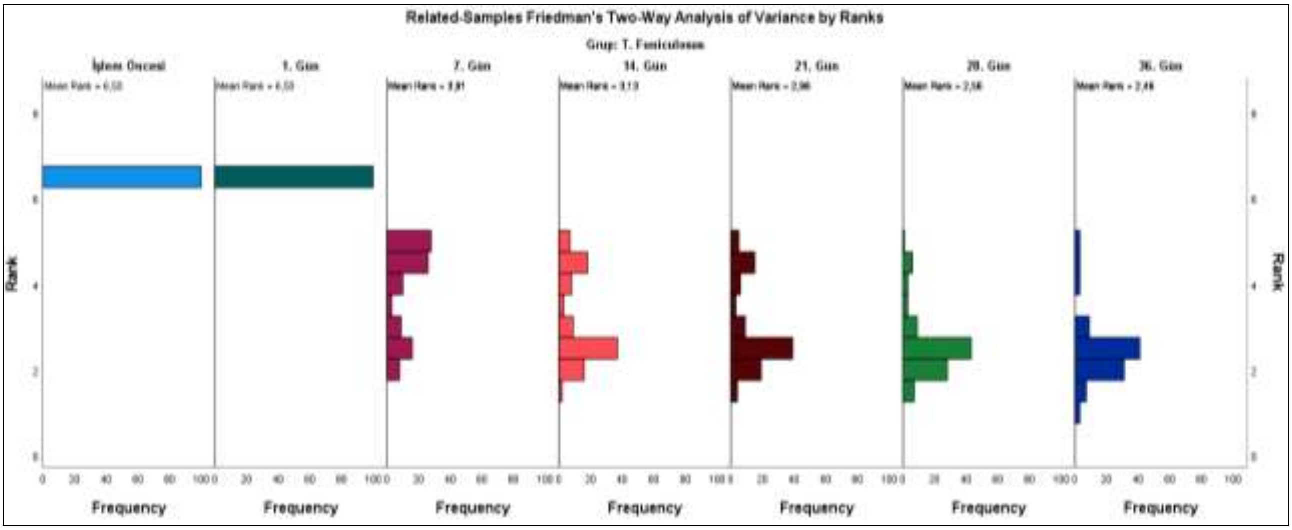

Fig 6: Pairwise Comparisons of *T. funiculosus* st976 Strain Applications by Days

Upon examining the pairwise comparisons of *T. funiculosus* applications, it was observed that on the 36th day, leaf damages were significantly reduced compared to pre-treatment, as well as the measurements taken on days 1 and 7. On the 28th day, the leaf damages were significantly reduced compared to those taken before treatment and on days 1 and 7. Similarly, on the 21st day, the leaf damages were significantly reduced compared to those measured before treatment and on days 1 and 7. On the 14th day, the leaf damage measurements were significantly lower compared to the measurements taken on the 1st day. Finally, on the 7th day, the leaf damage measurements were significantly lower compared to pre-treatment and the 1st day measurements ($p<0.05$) (Figure 4). The evaluation of the phytotoxicity of the compounds used is presented in Tables 3, 4, 5, and 6.

Table 3: Phytotoxicity Scales of Kaligreen Applications on Different Days

Kaligreen	Day 1	Day 7	Day 14	Day 21
Leaf 1	1	0	0	0
Leaf 2	0	0	0	0
Leaf 3	1	1	1	0
Leaf 4	1	1	0	0
Leaf 5	0	1	1	0
Leaf 6	0	0	0	0
Leaf 7	1	1	0	0
Leaf 8	0	0	0	1
Leaf 9	1	1	1	0
Leaf 10	0	0	0	1

When Table 3 is examined, it can be seen that the application of the Kaligreen product exhibited either no spots (0) or 1-2 spots (1).

Table 4: Phytotoxicity Scales of Paskal+ Copper Applications on Different Days

Paskal +Copper	Day 1	Day 7	Day 14	Day 21
Leaf 1	1	0	0	2
Leaf 2	0	0	2	0
Leaf 3	1	1	1	0
Leaf 4	2	1	0	0
Leaf 5	0	1	2	0
Leaf 6	0	2	2	0
Leaf 7	2	1	0	1
Leaf 8	0	2	0	1
Leaf 9	1	1	1	0
Leaf 10	0	0	0	1

In Table 4, it is observed that the application of the Pascal +Copper product exhibited either no spots (0), 1-2 spots (1), or 3-10 spots (2).

Table 5: Phytotoxicity Scales of Traditional Wettable Powder Sulphur Applications on Different Days

Traditional Wettable Powder Sulphur	Day 1	Day 7	Day 14	Day 21
Leaf 1	0	0	0	0
Leaf 2	0	0	0	0
Leaf 3	0	0	0	0
Leaf 4	0	0	0	0
Leaf 5	0	0	0	0
Leaf 6	0	0	0	0
Leaf 7	0	0	0	0
Leaf 8	0	0	0	0
Leaf 9	0	0	0	0
Leaf 10	0	0	0	0

When Table 5 is examined, it can be seen that the application of the traditional wettable powder sulphur exhibited no spots (0).

Table 6: Phytotoxicity Scales of *T. funiculosus* st 976 Applications on Different Days

T. funiculosus st976	Day 1	Day 7	Day 14	Day 21
Leaf 1	0	0	0	0
Leaf 2	0	0	0	0
Leaf 3	0	0	0	0
Leaf 4	0	0	0	0
Leaf 5	0	0	0	0
Leaf 6	0	0	0	0
Leaf 7	0	0	0	0
Leaf 8	0	0	0	0
Leaf 9	0	0	0	0
Leaf 10	0	0	0	0

When Table 6 is examined, it can be seen that the application of the *T. funiculosus* product resulted in a phytotoxicity scale where the leaves exhibited no spots (0).

In conclusion, the evaluation of the compounds used, particularly the *T. funiculosus* st976 strain, is significant for both its efficacy and its zero phytotoxicity. This highlights the potential for research into its use in pest control within viticulture. Although there are existing studies on the use of this fungus species as a phosphorus solubilizers, in biological control, and as a natural dye, the identification of its effectiveness against pests for the first time is promising (Oiuphisittraiwat *et al*., 2024) [7]. Furthermore, the pairwise comparisons of all applications over the days indicated increasing effectiveness against pests over time. This suggests that *T. funiculosus* st 976 and Kaligreen could be utilized in pest management. However, more detailed studies on dosage and the impact of *T. funiculosus* st 976 strain on plant nutrition and yield parameters within natural ecosystems need to be carried out.

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