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Temporal Distribution of *Cladosporium* and *Alternaria* Spores in the Atmosphere of Gelibolu (Çanakkale), Turkey

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Abstract: This aeropalynological study aims to determine the inter-annual and seasonal variation of atmospheric spores belonging to *Cladosporium* Link and *Alternaria* Nees genera during the three years. *Cladosporium* and *Alternaria* spores are generally recorded as dominant in many atmospheric fungal spore studies and have high allergic effects on susceptible individuals. The species belonging to these two genera may also live as pathogens on the plants and cause low yield. This study was carried out for three years, between January 2018 to December 2020. Durham sampler, which is the device of the gravimetric method, was used in the research. A total of 17399 *Cladosporium* and *Alternaria* spores were recorded in the Gelibolu (Çanakkale) atmosphere for three years. The total numbers of *Cladosporium* spores were found about five times more than the total numbers of *Alternaria* spores. The maximum spore levels were recorded in June for both spore types. Summer months can be reported as a risky period in terms of *Alternaria* and *Cladosporium* pathogenicity and atmospheric fungal spore allergy in the region.

Keywords: Atmospheric fungal spores, Aeromycology, Allergy, Northwest Turkey.

Gelibolu (Çanakkale), Türkiye Atmosferindeki *Cladosporium* ve *Alternaria* Sporlarının Zamansal Dağılımı

Öz: Bu aeropalinolojik çalışma, *Cladosporium* Link (Havaküfü) ve *Alternaria* Nees (Arıküfü) cinslerine ait atmosferik sporların üç yıl süre zarfındaki yıllar arası ve mevsimsel değişimlerini belirlemeyi amaçlamaktadır. *Cladosporium* ve *Alternaria* Nees sporları birçok atmosferik mantar sporu çalışmasında dominant olarak kaydedilmiştir ve duyarlı bireyler üzerinde yüksek allerjik etkilere sahiptirler. Ayrıca bu iki cinse ait türlerin sporları, bitkiler üzerinde patojen olarak yaşayabilmekte ve bitkilerde verim kaybına da neden olmaktadılar. Bu çalışma Ocak 2018 – Aralık 2020 tarihleri arasında üç yıl süreyle gerçekleştirilmiş olup araştırmada gravimetrik yönteme dayalı Durham cihazı kullanılmıştır. Üç yıl boyunca, Gelibolu (Çanakkale) atmosferinde toplam 17399 adet *Cladosporium* ve *Alternaria* sporu kaydedilmiş ve *Cladosporium* sporlarının toplam sayısı, *Alternaria* sporlarının toplam sayısından yaklaşık beş kat daha fazla bulunmuştur. Her iki spor türü için de maksimum spor mikatarınna Haziran ayında ulaştığı kaydedilmiştir. Yaz ayları bölgede *Alternaria* ve *Cladosporium* patojenitesi ve atmosferik fungal spor alerjisi açısından riskli dönemler olarak bildirilebilir.

Anahtar Kelimeler: Atmosferik mantar sporları, Aeromikoloji, Allerji, Kuzeybatı Türkiye.

Introduction

Fungi are a large kingdom with a wide distribution on the earth, can live in a wide variety of habitats, and contain many species (Gregory, 1961; Blackwell, 2011). These organisms can produce large numbers of spores, and these spores are mostly dispersed into the atmosphere, generally with the help of wind (Money, 2015). The most abundant spores in the atmosphere



influence various aspects of human health and agriculture (Caretta, 1992; Anton et al., 2021).

Fungal spores, especially *Cladosporium* (Havaküfü) and *Alternaria* (Arıküfü) spores, cause many allergic reactions on susceptible individuals and damage plants and animals. Due to their presence in the air, we breathe, they can cause respiratory diseases such as asthma and allergic rhinitis in people and may cause skin diseases in animals and yield losses in plants (D'Amato et al., 1984; Buck and Levetin, 1985; Vjay et al., 1991; Angulo-Romero et al., 1999; Sesli et al., 2020).

Since the sporulation times are different for each fungus, the periods and the number of spores of these fungi also differ in the atmosphere. The types and densities of atmospheric spores may vary according to geographical, ecological, meteorological factors and the floristic structure of the region. For this reason, it is necessary to find out together with spore calendars as a result of long-term studies by comparing the atmospheric spore types and their distribution during the year with meteorological factors in regions with topography and especially climate difference.

The first aeromycological study of Turkey was conducted by Özkaragöz in 1969 with the open Petri method to determine atmospheric fungal spores in Ankara. Atmospheric fungal spore studies have been continuing since 1969 in the different cities in Turkey and in the world (Özkaragöz, 1969; Gioulekas et al., 2004; Oliveira et al.. 2010; Mallo et al., 2011; Grinn-Gofron et al., 2016; Kasprzyk et al., 2016; Akgul et al., 2016; Ding et al., 2016; Saatcioglu et al., 2016; Yılmazkaya et al. 2019; Kilic et al., 2020; Bekil et al., 2021; Sevindik et al. 2021).

The aims of this study can be summarized as follows: (i) to contribute to taking the necessary measures to prevent yield losses in the economically important plants growing in the region; (ii) to benefit the people of the region and allergologists in the treatment process and in taking precautions to prevent allergies in risky periods; (iii) to investigate the annual variation of atmospheric *Cladosporium* and *Alternaria* spores and the effects of meteorological factors (Mean temperature, relative humidity, total rainfall, and wind speed) on them in the Gelibolu (Çanakkale) atmosphere.

Material and Method

The description of the Study area, Climate, and the Floristic Structure

Gelibolu is derived from its old name "Gallipoli", which the ancient Greeks gave, and it means good/beautiful city. Its history dates back to before Christ. The Gelibolu is a peninsula, stretches between the Çanakkale Boğazı "Dardanelles Strait" and the Saroz Körfezi "Saros Gulf", expanding to the south. Located in the Northwest of Turkey, the peninsula is also the last piece of land in the South-East of the European continent. The district of Gelibolu is situated on the North-Eastern coast of the peninsula with the same name, at the point where the Dardanelles Strait opens to the Sea of Marmara (40° 27' 44.38" N–26° 37' 54.30" E). The district has an area of 825 km², and the altitude is at sea level (Figure 1).

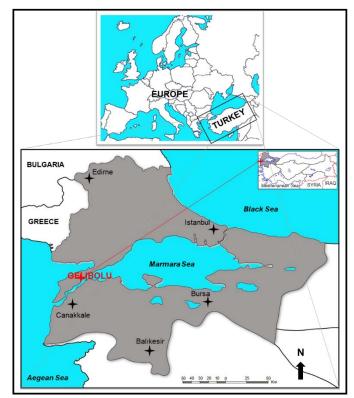


Figure 1. Location of study area (Gelibolu).

According to meteorological data provided by the Turkish State Meteorological Service, for three studied years (2018-2020), the annual average temperature was 16.8 °C (min. temperature 6.01°C - in January, max. temperature 25.95°C - in August). The total amount of precipitation was 1655.8 mm; the annual average relative humidity is 76.7%, and the annual average wind speed is 4.4 m/s. Since the district is located on the coast of the Dardanelles strait, it is under the influence of continuous air currents in all four seasons of the year.

The region is under the influence of the Mediterranean climate. *Pinus brutia* Ten. is the dominant species in the forest formation in the region, and there are *Pinus nigra* Arn., *Fagus* L., *Carpinus* L. pure or mixed forests in the higher parts of the region, and maquis



communities took place in areas where forests were destroyed (Turrill, 1924). Populus L. and Quercus L. species are primarily found in the valleys and Cupressus L. species in graveyards. On the side of the peninsula facing the Dardanelles strait, at an altitude of 150-200, species belonging to the genus Quercus, Pinus L., Juniperus L., Arbutus L., Myrtus L. on the steep slopes. A vegetation type consisting of grasses and scrub bushes covers the soil-poor and stony slopes of the peninsula, such as Quercus L., Cistus L., Erica L., Astragalus L., Tymelaea Mill. (Turrill, 1924; Şahin and Kartepe, 2020). The main species of the urban flora are Pinus brutia, Pinus pinea L., Pinus pinaster Ait., Pinus nigra, Carpinus orientalis Mill., Quercus coccifera L., Quercus ithaburensis subsp. macrolepis (Kotschy) Hedge & Yalt., Juniperus oxycedrus L., Arbutus andrachne L., Arbutus unedo L., Olea europea L., Erica arborea L., Myrtus communis L., Pistacia terebinthus L. and Laurus nobilis L. (Bağcı et al., 2004). The field crops grown around are; Helianthus L., Triticum L., Hordeum L., Avena L. Sesamum L., Nicotiana L. Phaseolus L. species.

Aeromycological study

This study was conducted between January 2018 to December 2020. Durham sampler, the device of the gravimetric method used in the research, was placed at the top roof of a building, 9 meters above ground level.



Figure 2. Percentage distribution of *Alternaria* and *Cladosporium* spores detected in Gelibolu atmosphere (three-year average of 2018-2020).

Before being placed on the device, slides were covered with glycerin-jelly (Charpin et al., 1974) and were changed weekly. Counting was conducted on a 24 x 24 mm area of the slide extrapolated to 1 cm² later; all spore numbers were given for per cm².

Results

In studied years, a total of 17399 spores were recorded, 14533 spores (83.47%) belong to the

Cladosporium genus, and 2876 spores (16.53%) belong to the *Alternaria* genus (Figure 2).

4086 spores (79.69% *Cladosporium* – 20.31% *Alternaria*); 4843 spores (81.88% *Cladosporium* –18.12% *Alternaria*); 8509 spores (86.19% *Cladosporium* –13.81% *Alternaria*) (Table 1,2) counted in order between 2018 - 2020. The total amounts of *Cladosporium* spores have been found higher than the amounts of *Alternaria* spores in each of the studied years.

Considering the average values of three years, the months that reached the highest spore levels were May, June and July. The months with the lowest amount of spores are December, January and February. Both *Cladosporium* and *Alternaria* spores have reached their maximum level in June every three years (Figure 3). Two peaks occurred in the total number of *Cladosporium* and *Alternaria* spores each year. The first peak occurred in June in all three years. The second peak occurred in October in 2018 and 2020, and November in 2019. These three years' the first peaks have been found larger than the second peaks. Both peaks have been higher in 2020 particularly the second peak showed a greater difference (Figure 3).

The atmospheric amount of *Cladosporium* has reached the lowest level in November in 2018 and 2020, and in January in 2019. The lowest amount of *Alternaria* spores have been found in May in 2018, in February in 2019, and in January in 2020 (Figure 4, Table 2).

In May, June and July, which the months that reached the highest spore levels, the temperature has been rising, and humidity has fallen every three years. In December, January, and February, which have the lowest amount of spores, the temperature has decreased, and the rainfall and humidity have increased in all these three years. In June, the temperature has increased but has not reached its maximum level and humidity has decreased, but not to its minimum level (Figure 5).

In this study, when the total number of *Cladosporium* and *Alternaria* spores in 3 years is examined seasonally, summer was recorded as the season with the most spores, and winter was recorded as the season with the fewer spores. 47.69% of *Cladosporium* spores and 10.17% of *Alternaria* spores were recorded in the summer and 2.97% of *Cladosporium* spores and 0.43% of *Alternaria* spores were recorded in the winter (Figure 6). On the other hand, the lowest amount of humidity and the highest amount of



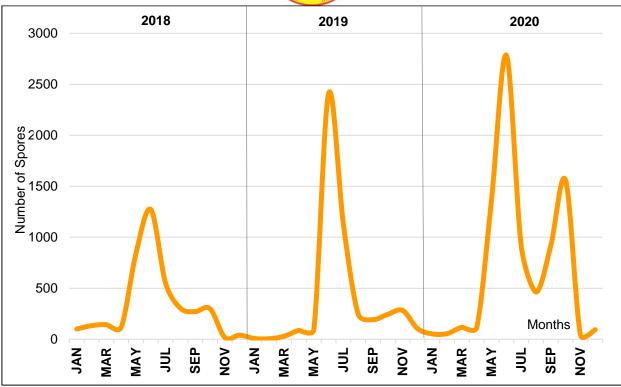


Figure 3. Monthly variation of total spore amounts of *Alternaria* and *Cladosporium* during 2018-2020.

Table 1: Total amounts of *Cladosporium* and *Alternaria* spores in Gelibolu atmosphere in years 2018-2020 (mean and percentage values).

	2018	%	2019	%	2020	%	MEAN	TOTAL	%
Cladosporium	3256	79.69	3950	81.88	7317	86.19	4841.00	14523	83.47
Alternaria	830	20.31	874	18.12	1172	13.81	958.67	2876	16.53
TOTAL	4086	100	4824	100	8489	100	5799.67	17399	100.00

temperature was recorded in the summer and the opposite was observed in the winter (Figure 4).

When the total number of *Cladosporium* and *Alternaria* spores were examined weekly, the week with the highest amount of spores was recorded at 24th week in 2018, at 26th week in 2019, and 41st week in 2020. The lowest amount of spores was recorded at 17th and 44th weeks in 2018, at 9th and 12th weeks in 2019, and at 2nd week in 2020 (Figure 7).

Discussions

In Turkey and worldwide spores of *Cladosporium* and *Alternaria* are often recorded as the dominant

airborne spore types many researchers relate the occurrence of respiratory allergy symptoms with the presence of these spores in the ambient air (Kauffman et al., 1995; Downs et al., 2001).

In this study, the total number of *Cladosporium* spores was recorded much more than *Alternaria* spores (Figure 2, Table 1). *Cladosporium* is the dominant airborne fungal spore type observed in many countries (Katial et al., 1997; Bustos et al., 2001; Henriguez et al., 2001; Al-Subai, 2002; Pepeljnjak and Segvi, 2003; Gioulekas et al., 2004, Mitakakis et al., 2005; Asan, 2015). This could be due to many factors; as seen in many fungal spore studies, *Cladosporium* forms more



colonies than *Alternaria*, and the number of species belonging to *Cladosporium* genus is higher than that of *Alternaria* as observed in many systematic studies (Şen and Asan, 2001; Dugan et al., 2004; Asan,2015; Kireçci and Alagöz, 2019).

The annual total number of spores in 2020 (8489 spores) has been nearly doubled compared to 2019 (4824 spores) and 2018 (4086 spores), and this is why related to meteorological factors (Figure 3). In many studies, it has been determined that the weather conditions have the most important and greatest effect on airborne spore concentration (Hjelmroos, 1993; lanovici, 2016; Grinn-Gofron et al., 2016; Olsen et al., 2020). Meteorological factors need to be examined to explain the difference in spore amounts between 2020, 2019, and 2018 (Figure 3, Table1-2). The total amount of rainfall in 2020 was lower than in 2018 and 2019 (Figure 5). The amount of precipitation is one factor influencing the concentration of spores in the atmosphere. It's known that annual variation in climate, particularly rainfall, can determine the fungal spore concentration in the atmosphere, and in dryer years, higher fungal concentration was observed in different studies (Kendrik 2001; Pakpour et al., 2014). Also, precipitation may clean the air by forcing fungal propagules back to the ground or onto other surfaces (Katial et al., 1997; Polymenakou, 2012).

One of the other important factors affecting spore concentration is relative humidity, and many researchers reported a negative correlation between atmospheric fungal spore concentrations and relative humidity (Kurkela, 1997; Stepalska and Wolek, 2005; Oliveira et al., 2007). In Gelibolu, relative humidity was recorded in 2020 (74.74%) is lower than in 2019 (77.28%) and 2018 (78.05%) and there was not much difference between mean temperature and wind speed for these three years (Figure 5).

When the total number of spores in these three years for both *Cladosporium* and *Alternaria* is examined, the maximum number of spores belong to summer, and the minimum number of spores belong to the winter (Figure 6). *Cladosporium* and *Alternaria* were reported as the most prevalent fungal spore type during summer (lanovici 2017; Oliveira et al., 2005). They are classified as dry-air spores, too, and for this reason, they are found in the greatest abundance in the atmosphere with high temperatures. Low humidity and these conditions are most visible in the summer (Peternel et al., 2004). In each of these three years, it was observed that the temperature has increased and humidity has decreased in summer (June, July, August) and the temperature has decreased,

and humidity and precipitation have increased in winter (December, January, February) (Figure 5).

Temperature is stated as one of the most influential and important factors on fungal spore concentration in the atmosphere and has a positive effect on spore concentration in the air (Hjelmroos, 1993). The positive impact of the high temperature on the spore amounts was observed in the summer in Gelibolu (Figure 5). Similar results have been obtained in other atmospheric fungal spore studies with similar climatic conditions (Şakıyan and İnceoğlu, 2003; Kizilpinar and Dogan, 2011; Gioulekas et al., 2004). As mentioned before many studies were reported a negative correlation between atmospheric fungal spores and relative humidity. Another reason why the summer has the maximum number of spores is the low relative humidity in the summer. In addition to low temperature and high humidity in winter, both of which have a negative effect on the atmospheric amount of spores in the air, the amount of rainfall is another effective factor. Some researchers have considered that the rain removes ambient fungal spores by both rain-out and wash-out effects (Magyar et al., 2009; Artac et al., 2014).

Two peaks occurred in the total number of Alternaria and Cladosporium spores in each of these three years (Figure 3). The first peak has occurred in June, same this in Timisoara (Romania) maximum spore amounts (first peak) were determined in June too (lanovici, 2016). The second peak occurred in October in 2020 and 2018 and in November in 2019 (Figure 3). The first peak was more significant than the second peak in all three years and had a more significant number of spores; this is related to the conditions in June (Figure 3, 5). High temperature and low humidity in June can be seen as an optimum sporulation conditions for *Cladosporium* and Alternaria. Although it has been mentioned before that precipitation has a negative effect on the spore concentration, much rainfall was recorded in June (Figure 5); this is probably because of different rainfall types, which have different effects on atmospheric spore amounts. Heavy rain such as rain storms and thunderstorms may increase the spore concentration in the atmosphere because the spore of fungi such as Cladosporium and Alternaria, which can live on plants or in the soil, can be separated from the conidium by force and dispersed into the atmosphere but the light rain decrease spore concentration in the air (Rich and Waggoner, 1962). Besides, rainfall may cause a more accessible release of fungal spores to the atmosphere by splash and tap-and-puff mechanisms (Ho et al., 2005).



In July, the spores of Cladosporium and Alternaria spores started to decrease rapidly, which may be due to the extreme temperature in August (Figure 4, 5). Several authors found a negative correlation between fungal spores and temperature (Aira et al., 2003; Calderon et al., 1997). Common fungal spore only develops under a certain temperature threshold; therefore, with extremely high or low temperature, the spore concentration decreases (Oliveira et al., 2005). In this study was observed that the high temperature of August and the cold weather of January correspond to a decrease in the spore amounts. Same this was observed in another study in Porto city (Oliveira et al., 2005) was reported the exceedance of the higher temperature limit for growth might kill fungi. In contrast, temperatures below the lower limit are less lethal (Eduard, 2009). It is known that, for each fungal species, there is an optimal temperature range for growth to occur; outside of that optimal temperature range, more water is necessary for growth (Ianovici, 2016). Another reason for decreasing the number of *Cladosporium* spores sharply in July may be the excessive humidity reduction. It is well known that fungi typically require moisture to grow and sporulate, many spores eventually become airborne (Kendrik, 2001, Tilak, 2009) and low relative humidity and extreme temperature inhibit growth and spore germination (Talley et al., 2002), as seen in our study.

When the total number of Cladosporium and Alternaria spores was examined weekly, in the week with the highest number of spores in each of the three years, the relative humidity and temperature were recorded at moderate levels, neither maximum nor minimum (Figure 7). And this is because as has been mentioned common fungal spore only develops under a certain temperature threshold; therefore, with extremely high or low temperature, the spore concentration decreases (Oliveira et al., 2005). In all three years in the weeks when the lowest number of spores was recorded, the total precipitation was also low (Figure 5). The low amount of precipitation negatively affected the number of weekly spores. Rainfall may cause a more accessible release of fungal spores to the atmosphere by splash and tap-andpuff mechanisms (Ho et al., 2005).

The formation of the peaks in our study may have many reasons, as Dugan et al. stated earlier (2004); related meteorological factors may have been influential for sporulation and sporulation times of different species belonging to both genera. Also, the second peak in the fall can be related to decaying leaves from the plants, which constitute a substrate where fungal spores can grow, as shown by Oliveira et al. (2005). Also, one factor that increases Cladosporium and Alternaria spores in summer and autumn can be related to agricultural activities. Harvest time corresponds to the June-October term depending on the plants that Cladosporium and Alternaria can live on as pathogens and that can be released and increase the spore concentration in the atmosphere (Landecker, 1996). The seasonal pattern of Cladosporium generally follows the life cycle of the local vegetation (Mitakakis et al., 1997). Many factors affect the concentration of Cladosporium and Alternaria spores in the atmosphere, including geobotanical characteristics of the region, vegetation, climate, the types of fungal spores, proximity and abundance of the source of fungal spore and sporulation periods of them (Hjelmroos, 1993; Şakıyan and İnceoğlu, 2003; lanovici, 2016; Sindt et al., 2016; Grinn-Gofron et al., 2016; Olsen et al., 2020; Damialis and Gioulekas, 2006). And many authors indicate that airborne fungal spore levels are dependent on crop production and proximity of grassland areas (Solomon, 1978; Mitakasis et al., 2001; Corden et al., 2003; Pepelinjak and Segvic, 2003; Damialis and Gioulekas, 2006). Among these, the weather condition has the most important and greatest effect on spore concentration (Hjelmroos, 1993).

The atmospheric concentrations of *Cladosporium* and *Alternaria* are very important for many reasons: *Cladosporium* and *Alternaria* spores have been in the atmosphere for a long time, and they can be a factor in the emergence of disease symptoms such as asthma, allergic rhinitis, and conjunction by entering the body through ways such as eye conjunctiva, skin, respiratory and nasal mucosa (Chakrabarti et al., 2012). And also have pathogenic effects on many plants of economic importance. *Cladosporium* and *Alternaria* spores may cause skin diseases in animals and yield losses in plants (D'Amato et al. 1984, Buck and Levetin 1985, Vjay et al. 1991, Angulo-Romero et al. 1999).

Conclusions

In this study, the atmospheric amounts of *Cladosporium* and *Alternaria* spores and the effect of meteorological factors were investigated in the Gelibolu atmosphere for three consecutive years (2018-2020). In the Gelibolu atmosphere, the temperature was positive,



Table 2. Monthly variation of Cladosporium and Alternaria spores in Gelibolu atmosphere during the years 2018-2020

TAXA/MONTHS		JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	ост	NOV	DEC	TOTAL
Cladosporium	2018	90	116	141	108	704	1045	459	253	59	235	9	37	3256
	%	2.20	2.84	3.45	2.64	17.23	25.58	11.23	6.19	1.44	5.75	0.22	0.91	79.69
	2019	2	5	28	83	81	2004	887	181	123	211	254	91	3950
	%	0.04	0.10	0.58	1.72	1.68	41.54	18.39	3.75	2.55	4.37	5.27	1.89	81.88
	2020	46	48	108	103	1328	2450	657	363	699	1413	21	81	7317
	%	0.54	0.57	1.27	1.21	15.64	28.86	7.74	4.28	8.23	16.65	0.25	0.95	86.19
	Mean	46.00	56.33	92.33	98.00	704.33	1833.00	667.67	265.67	293.67	619.67	94.67	69.67	4841.00
	%	0.93	1.17	1.77	1.86	11.52	31.99	12.45	4.74	4.08	8.92	1.91	1.25	82.59
Alternaria	2018	10	17	2	9	138	227	89	51	212	64	8	3	830
	%	0.24	0.42	0.05	0.22	3.38	5.56	2.18	1.25	5.19	1.57	0.20	0.07	20.31
	2019	5	0	1	4	12	405	238	65	69	33	28	14	874
	%	0.10	0.00	0.02	0.08	0.25	8.40	4.93	1.35	1.43	0.68	0.58	0.29	18.12
	2020	6	7	9	13	46	334	261	100	220	143	20	13	1172
	%	0.07	0.08	0.11	0.15	0.54	3.93	3.07	1.18	2.59	1.68	0.24	0.15	13.81
	Mean	7.00	8.00	4.00	8.67	65.33	322.00	196.00	72.00	167.00	80.00	18.67	10.00	958.67
	%	0.14	0.17	0.06	0.15	1.39	5.96	3.40	1.26	3.07	1.31	0.34	0.17	17.41
TOTAL	2018	100	133	143	117	842	1272	548	304	271	299	17	40	4086
	%	2.45	3.26	3.50	2.86	20.61	31.13	13.41	7.44	6.63	7.32	0.42	0.98	100.00
	2019	7	5	29	87	93	2409	1125	246	192	244	282	105	4824
	%	0.15	0.10	0.60	1.80	1.93	49.94	23.32	5.10	3.98	5.06	5.85	2.18	100.00
	2020	52	55	117	116	1374	2784	918	463	919	1556	41	94	8489
	%	0.61	0.65	1.38	1.37	16.19	32.80	10.81	5.45	10.83	18.33	0.48	1.11	100.00
	Mean	53.00	64.33	96.33	106.67	769.67	2155.00	863.67	337.67	460.67	699.67	113.33	79.67	5799.67
	%	1.07	1.34	1.83	2.01	12.91	37.95	15.85	6.00	7.15	10.24	2.25	1.42	100.00



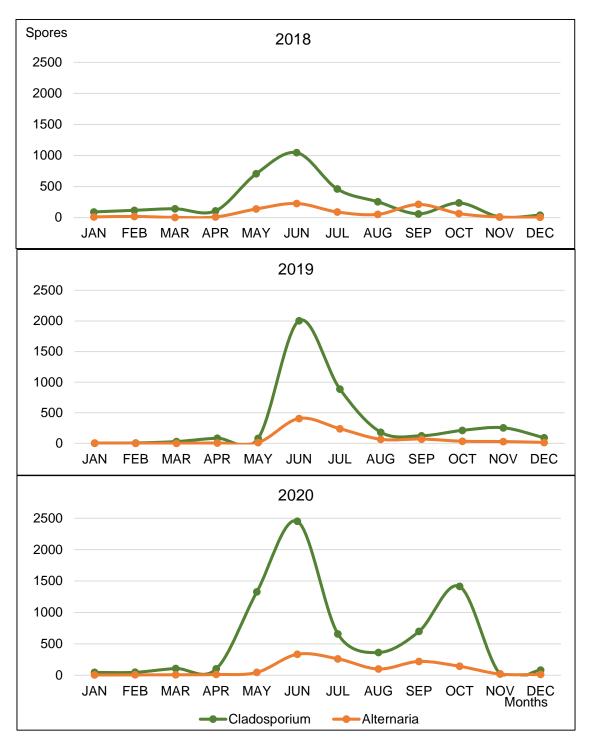


Figure 4. Monthly amounts of *Cladosporium* and *Alternaria* spores in Gelibolu atmosphere in the studied years (2018-2020).



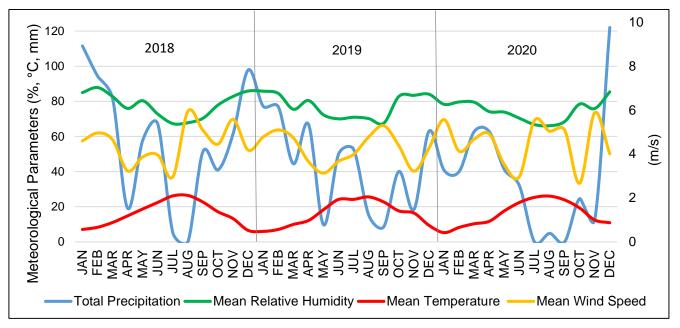


Figure 5. Monthly variation of meteorological parameters in Gelibolu during the years 2018-2020.

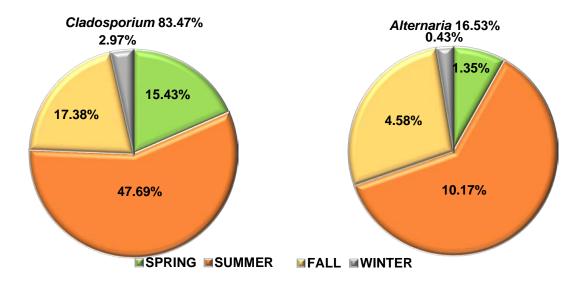


Figure 6. Seasonal dispersion of Cladosporium and Alternaria spores in Gelibolu atmosphere.



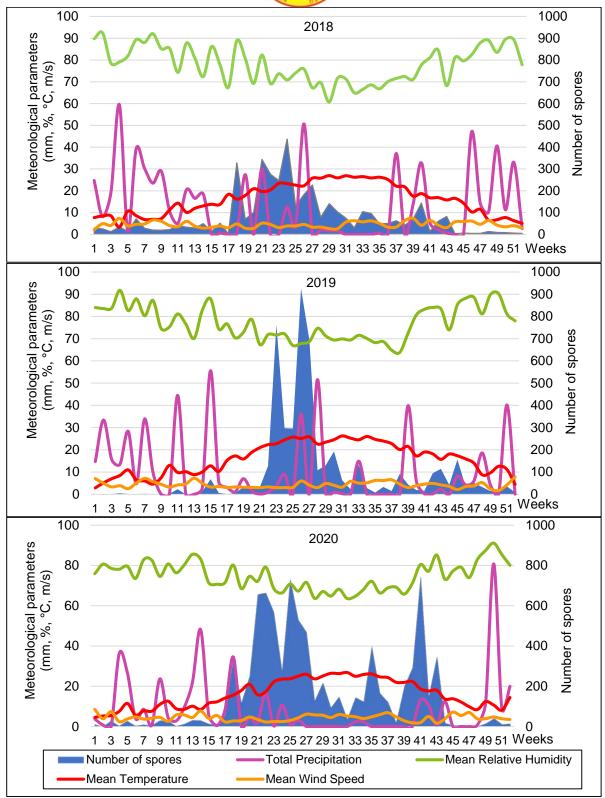


Figure 7. Weekly variation of meteorological parameters and number of spores in Gelibolu during the years 2018-2020.



and relative humidity and rainfall negatively affected spore amounts. Still, there could not be found any relation between wind speed and spore amounts. In all years, the highest spore amounts were recorded in the summer, and the total number of spores was the highest, especially in June. June had provided optimal conditions (high temperature and low humidity) for both *Cladosporium* and *Alternaria* sporulation. Therefore, June can be very risky for susceptible individuals, and conducting such studies can be helpful to doctors in the diagnosis, identification, and treatment of allergic patients.

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