

# The First Record of *Gregarina typographi* Fuchs (Protista: Apicomplexa: Gregarinidae) from the European Spruce Bark Beetle, *Ips typographus* (Linnaeus) (Coleoptera: Curculionidae, Scolytinae) in Turkey

Türkiye’de *Ips typographus* (Linnaeus) (Coleoptera: Curculionidae, Scolytinae)’dan *Gregarina typographi* Fuchs (Sporozoa, Gregarinidae)’nin İlk Kaydı

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## ABSTRACT

**Objective:** Aim of this study is determining pathogens of *Ips typographus* from Eastern Black Sea Region in Turkey.

**Methods:** Samples collected from the field were taken to laboratuar as soon as possible. Microscopic examination was completed by dissection with Ringer’s solution.

**Results:** 780 *Ips typographus* beetles from Giresun, Rize and Artvin were examined. Gregarines were observed in the populations of *I. typographus* in the three regions. 27 of 780 beetles were found to be infected by the *Gregarina typographi*. Total rate of infection was 3.4% in three localities. During the study several life stages of the gregarine pathogen (trophozoite, gamont, Cysts and associative form) were observed. Gametocysts were spherical and from 77 to 85 µm in diameter. Total lengths of solitary gamonts were measured. from 90 to 155 µm. Measurements of gamonts and gametocysts of *G. typographi* were given and compared with other gregarines isolated from bark beetles. This pathogen is described as *Gregarina typographi*.

**Conclusion:** The gregarine pathogen of *Ips typographus* is reported from Turkey for the first time. (*Türkiye Parazitol Derg* 2010; 34: 179-82)

**Key Words:** Gregarine pathogen, *Ips typographus*, *Gregarina typographi*

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## ÖZET

**Amaç:** Doğu Karadeniz bölgesinde yayılış gösteren *Ips typographus* türündeki patojen varlığının belirlenmesi amaçlanmıştır.

**Yöntemler:** Araziden toplanan örnekler en kısa sürede laboratuvara getirilmiştir. Ringer solüsyonu ile diseksiyon yapılarak mikroskopik incelemeleri tamamlanmıştır.

**Bulgular:** Artvin, Rize ve Giresun illerinden toplam 780 adet *Ips typographus* örneği incelenmiştir. Her 3 ilde de gregarin patojeninin varlığı tespit edilmiştir. Disekte edilen 780 böcekten 27 tanesinde gregarin enfeksiyonuna rastlanılmıştır. Enfeksiyon yüzdesi % 3,4 tür. Çalışma boyunca patojene ait çeşitli hayat safhaları (trofozoit, gamont, kist ve şizigi) gözlenmiştir. Gametokistler küresel şekilli ve 77-85 µm çapında ölçülmüştür. Gamontların total uzunluğu min. 90 µm, max. 155 µm olarak ölçülmüştür. Patojenin vücut kısımlarına ait gerekli ölçümler karşılaştırılarak karakterizasyonu yapılmış ve *Gregarina typographi* olarak tanımlanmıştır.

**Sonuç:** Bu çalışmada, *Ips typographus*’tan *G. typographi* (Apicomplexa: Gregarinidae) patojeninin varlığı ülkemiz için ilk kez kaydedilmiştir. (*Türkiye Parazitol Derg* 2010; 34: 179-82)

**Anahtar Sözcükler:** Gregarin patojeni, *Ips typographus*, *Gregarina typographi*

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## INTRODUCTION

There are many economically harmful bark beetles in Turkey (1). The European spruce bark beetle *Ips typographus* is one. This pest is causing damage to *Picea orientalis* forests in the eastern Black Sea Region, leading to severe economic loss. In order to control this important pest, methods such as pheromone traps, chemical pesticides and mechanical control strategies have been used up to the present. These methods are inadequate because undulating land conditions make the application of these methods difficult and expensive. Apparently, chemicals have harmful effects on the environment, building up in the water, soil and living organisms. In addition, another disadvantage of using chemicals is that they destroy the natural enemies of target organisms. Natural enemies are very important in biological control. For this reason, many studies have been carried out on using natural enemies in biological control (2-5). In recent years, microorganisms have come in to prominence as biological control agents. The use of microorganisms as pathogens may very effective. Pathogens of *Ips typographus* are known from different parts of the world (6-12), but there is a lack of knowledge about the pathogens in Turkey. In the present paper, a gregarine pathogen of *I. typographus* from Turkey is reported for the first time.

## MATERIALS AND METHODS

Adult *I. typographus* specimens were collected from spruce (*Picea orientalis*) forests using pheromone traps in Giresun, Rize and Artvin (Turkey), from May to August in 2009. Each beetle was dissected in a Ringer solution and its intestine was examined microscopically at magnifications of 400x to 1000x. Observed pathogens were measured and photographed using an Olympus BX 51 microscope with a DP-25 digital camera and DP2-BSW Soft Imaging System. The following abbreviations, according to Lipa (13), Geus (14) and Clopton (15) are used to identify the gregarine pathogen in this paper: TL=total length; LP=length of protomerite; LD=length of deutomerite; WP=width of protomerite; WO=width of deutomerite; LP:TL=ratio of the length of protomerite to total length; WP:WD=ratio of the width of protomerite to the width of deutomerite; and WP:TL= ratio of the width of protomerite to total length. The systematic examination of the parasite was made by comparing the same or systematically close species given by Lipa (13) and Geus (14).

## RESULTS

A gregarine pathogen was found in *Ips typographus*; *Gregarina typographi* FUCHS (Protista, Eugregarinida). Gregarines were observed in the populations of *I. typographus* collected from three localities (Giresun, Rize and Artvin). During the study, several life stages of the gregarine pathogen, such as trophozoite with epimerite (Figure 1A), gamont (Figure 1B), associative form previous to the cyst (Figure 1C), precyst (Figure 1D) and cyst (Figure 1E) were observed. Measurements (in  $\mu\text{m}$ ) of gregarine gamonts are given Table 1. Gametocysts were spherical and from 77 to 85  $\mu\text{m}$  in diameter (Figure 1E). Its gamonts were found in the mid-gut lumen. Gamonts were oval to elongated and solitary.

04 of 135 beetles examined from Giresun, 06 of 177 beetles from Rize, and 17 of 468 beetles from Artvin were found to be infected by the gregarine pathogen. Infection rates were found as 2.9% in Giresun, 3.3% in Rize and 3.6% in Artvin. Total rate of infection was 3.4% for the three localities.



**Figure 1.** Trophozoite (A), Gamont (B), Associative form, previous of cyst (C), Precyst (D), Cyst (E) of the *G. typographi*. (Bar 50  $\mu\text{m}$ )

## DISCUSSION

In the literature there are several reports on the gregarine pathogen of bark beetles (6, 7, 13, 16, 17). The first gregarine pathogen record from bark beetles in Turkey is *Gregarina typographi* from *Ips sexdentatus* (18). The gregarine presented in the present study is the second record from bark beetles in Turkey, but the first gregarine pathogen record from *Ips typographus* in Turkey. The pathogen shows similarities to *Gregarina typographi* Fuchs, recorded by Fuchs (6) from *Ips typographus* for the first time, in terms of its characteristics identified with the genus *Gregarina* (Table 2). The gregarine in the present study is apparently similar to *G. typographi* in its morphological characters. In the present study, total lengths of solitary gamonts were measured from 90 to 155  $\mu\text{m}$ . Lipa (13) reported that the total length of solitary gamonts measured from 78 to 118  $\mu\text{m}$  and Takov et al. (19) observed gamonts between 32.5-193.8  $\mu\text{m}$  for *G. typographi* from *Ips typographus*. Yaman (18) reported that solitary gamont total length measured from 80 to 275  $\mu\text{m}$ . Lipa (13), Purrini (17), Wegensteiner et al. (7) and Takov et al. (19) recorded this pathogen from the same host.

There are several reports in the literature about the occurrence of *G. typographi* from two *Ips* species (*I. sexdentatus* and *I. typographus*) which are the most economically important in Turkey. *I. sexdentatus* and *I. typographus* are from the family Curculionidae (Coleoptera) and they share the same terrestrial habitat, even the same tree (*Picea orientalis*) in the Eastern Black Sea Region, therefore they also share pathogens. According to these facts, *G. typographi* has two different hosts which are *I. sexdentatus* and *I. typographus* in Turkey. *G. typographi* was

**Table 1.** Measurements of gamonts of *Gregarina typographi* in *Ips typographus* (in  $\mu\text{m}$ ).

	TL	LP	LD	WP	WD	LP:TL	WP:WD	WP:LP
Gamonts	150.32	41.44	108.88	43.72	42.23	0.28	1.04	1.06
	91.25	24.76	66.49	39.15	45.38	0.27	0.86	1.58
	102.88	25.82	77.06	40	41.21	0.25	0.97	1.55
	109.78	30.9	78.88	30.33	35.96	0.28	0.84	0.98
	104.73	31.25	73.48	41.02	41.23	0.30	0.99	1.31
	138.44	31.67	106.77	39.54	39.55	0.23	1.00	1.25
	143.33	40.64	102.69	36.88	37.27	0.28	0.99	0.91
	155.56	36.25	119.31	49.53	66.5	0.23	0.74	1.37
	128.27	40.65	87.62	41.65	41.94	0.32	0.99	1.02
	90.73	28.93	61.8	36.35	44.35	0.32	0.82	1.26
	154.79	37.84	116.95	52.26	66.84	0.24	0.78	1.38
	95.69	24.43	71.26	26.67	32.77	0.26	0.81	1.09
	118.51	27.63	90.88	40.26	37.54	0.23	1.07	1.46
	95.94	23.66	72.28	33.55	38.01	0.25	0.88	1.42
	115.69	29.33	86.36	36.72	43.57	0.25	0.84	1.25
	146.61	45.77	100.84	41	38.01	0.31	1.08	0.90
	101.4	27.79	73.61	37.27	35.77	0.27	1.04	1.34
	98.13	20.62	77.51	32.49	35.87	0.21	0.91	1.58
	115.93	28.1	87.83	41.2	39.64	0.24	1.04	1.47
	118.67	26.06	92.61	25.78	34.84	0.22	0.74	0.99
128.87	26.53	102.34	41.24	40.41	0.21	1.02	1.55	
117.43	35.91	81.52	37.05	40.61	0.31	0.91	1.03	
95.71	20.61	75.1	33.65	37.04	0.22	0.91	1.63	
146.43	42.46	103.97	39.59	36.5	0.29	1.08	0.93	
Mean	119.37	31.21	88.16	38.20	41.37	0.26	0.93	1.26
Min.	90.73	20.61	61.8	25.78	32.77	0.21	0.74	0.90
Max.	155.56	45.77	119.31	52.26	66.84	0.32	1.08	1.63
SD.	21.64	7.23	16.28	6.07	8.40	0.03	0.11	0.24

TL: total length; LP: length of protomerite; LD: length of deutomerite; WP: width of protomerite; WD: width of deutomerite; LP:TL: ratio of the length of protomerite to total length; WP:WD: ratio of the width of protomerite to the width of deutomerite. WP:LP: ratio of the width of protomerite to the length of protomerite; Mean: Arithmetic mean; SD: standard deviation; Min and Max: extreme values, n: the number of the gamonts measured (n=24)

**Table 2.** The comparative characters of gregarines observed in beetles (Coleoptera) compared to *Gregarina typographi* Fuchs (mm).

Characters	<i>G. typographi</i>	<i>G. typographi</i> Fuchs	<i>G. typographi</i>	<i>G. typographi</i>	<i>G. typographi</i>
TL <sup>a</sup>	80-275	78-118	32.5-193.8	55-237.5	90-155
LP:TL <sup>a</sup>	1: 2.7-5.3	1 : 2.2-5	1: 0.14-0.49	1: 0.09-0.38	1: 0.21-0.32
WP:WD <sup>a</sup>	1: 1.1-1.3	1 : 1.1-2	1: 0.63-1.13	1: 0.22-1.8	1: 0.74-1.08
Gametocyst	Spherical 90 to 100	.....	.....	.....	Spherical 77 to 85
Host	<i>Ips sexdentatus</i>	<i>Ips typographus</i>	<i>Ips typographus</i>	<i>Ips sexdentatus</i>	<i>Ips typographus</i>
Reference	18	13	19	19	Present study

<sup>a</sup>; minimal and maximal: TL; total length; LP:TL: ratio of the length of protomerite to total length; WP:WD: ratio of the width of protomerite to the width of deutomerite.

reported from *I. sexdentatus* by several authors (16-19). The occurrence of *G. typographi* from *I. sexdentatus* was reported for the first time from Turkey by Yaman (18) and *G. typographi* from *I. typographus* is reported for the first time from Turkey in the present study.

*G. typographi* was found from *I. typographus*, *I. sexdentatus* and *I. acuminatus* in Bulgaria, infection rates were found as 10.1% for *I. typographus*, 27.4% for *I. sexdentatus* and 11.8% for *I. acuminatus* (19). Händel et al. (2) found 0.8-59.4% infection for *G. typographi* in *I. typographus* and Wegensteiner & Weiser (20) reported 6.3% infection in the same host in Austria. Wegensteiner et al. (21) reported 16.7-45.5% infection rates in *I. sexdentatus* in Austria. Wegensteiner & Weiser (22) reported that this pathogen from *I. typographus* in Germany and its infection rate was 11%. Holusa et al. (11) reported it from *I. typographus* and *I. duplicatus* in Czech Republic with the infection rates, 5% and ≤5% respectively. Kereselidze & Wegensteiner (23), Kereselidze et al., (10) and Burjanadze & Goginashvili (12) reported *G. typographi* from *I. typographus* in Georgia, the pathogen infection rates were 23.2-48.6% for 2005, 24% for 2007 and 7.5-49.5% for 2009 from *I. typographus*. Takov et al. (9) reported the same pathogen from *I. typographus* in Bulgaria, max total infection rate was 45.2% and min total infection rate was 5.3% from four different areas in this report. Yaman (18) reported that this pathogen causes 6% infection in *I. sexdentatus* in Turkey. Ünal et al. (24) found 25.8% infection in the same host. All results show that *G. typographi* is present in neighbor countries, Bulgaria, Georgia and Turkey, indicating distribution of this pathogen. In addition to this, Germany, Austria and Czech Republic are neighbor countries too. Although, infection rate of *G. typographi* is high in Georgia and Bulgaria, it is low in Turkey.

*Ips typographus* is important pest in spruce forests for Turkey. These insects cause great economic loss in oriental spruce forest in Black Sea Region. Tanada & Kaya (25) reported that the pathological effects of gregarines are generally weak. However the gregarine pathogen may play an important role in the biological control of bark beetles by reducing the life span and reproductive potential of the host (18). So, we need much more studies in this subject.

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#### Conflict of Interest

No conflict of interest was declared by the authors.

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