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Full Length Research Paper

Species richness and true diversity of cynipid galling-wasps community in oak forests of West-Azerbaijan Province (Iran)

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In this survey, the oak gall wasps (Hymenoptera: Cynipidae: Cynipini) were collected from oak forests of West-Azerbaijan Province in six sites, from April to October. Species richness, heterogeneity, evenness and true diversity were measured. Based on the result of this study, 37 of oak gall wasps species on oak trees (*Quercus infectoria* Oliv. and *Quercus brantii* Lindl.) were identified in West-Azerbaijan Province in 2009 and 2010. Most galls occurred on *Q. infectoria*. All the collected oak gall wasps belonged to seven genera: *Andricus*, *Cynips*, *Neuroterus*, *Chilaspis*, *Pseudoneuroterus*, *Biorhiza* and *Aphelonyx*. The expected number of oak gall wasps species on *Q. infectoria* and *Q. brantii* was estimated to be 27 and 10, respectively. The highest amount of gal wasps' species richness with rarefaction method was recorded in Pardanan, with 28 oak gall wasps species. Furthermore, the highest amount of Gini-Simpson and Shannon entropy index were recorded in Sardasht, while the highest evenness was recorded in Shalmash. Differences in the local distribution of oak species, especially their oak subspecies, and the climate of the locations should be considered as one main factor in species diversity and the distribution of oak gall wasps species in different locations.

Key words: Alpha diversity, distribution, rarefaction, evenness, forest.

INTRODUCTION

Oak gall wasps (Hymenoptera: Cynipidae: Cynipini) are obligate parasites of plants in the family Fagaceae, and have induced the most structurally complex and diverse galls known as gall-inducing group (Stone et al., 2002; Atkinson et al., 2003). Iran is located in the west of the Palaearctic region and its major plant covering is treeless steppe. Oak forests of Iran are located as high as over 2200 m above sea level. In the Zagros Mountains, especially in West-Azerbaijan province of Iran, oak forests are composed of drought-tolerant species including *Quercus libani* and *Quercus brantii* and *Quercus infectoria* (Azizkhani, 2006; Zargaran et al., 2008). Iran lies at the eastern limit of the Western Palaearctic, and recent surveys confirm that its cynipid fauna includes widespread Western Palaearctic species (such as *Andricus kollari* and *Cynips quercusfolii*) limited to the eastern part of this region, such as the insana form of *Andricus quercustozae*, and *Aphelonyx persica* (Melika et al., 2004; Zargaran et al., 2008).

Species diversity has two parts. Richness refers to the number of species found in a community and evenness refers to the relative abundance of each species (Magurran, 1988). The simplest measure of diversity is to count number of species in an area (species richness), but the major problem in species richness measurement is that species richness does not take into account species abundance and is highly influenced by sample size. Therefore, diversity indices that incorporate both abundance and richness are three methods: Dominance indexes, Information-statistic indexes and Evenness indexes. One of the most important dominance indexes is Simpson's index. Simpson's index of diversity calculates the probability that two organisms sampled from a community belongs to different species, and its values ranges from 0 to one (Magurran, 2004). The higher the value of D, the lower the diversity; therefore, the index is usually expressed as the reciprocal, 1/Ds. Moreover, information-statistic indices can take into account rare species and based on the rationale that diversity in a natural system can be measured in a way that is similar to the way information contained in a code or message measured. Evenness, the other information-statistic indices, is affected by both number of species and their equitability. The value of E is constrained between 0 and 1.0. Also, rank abundance diagrams are a more complete picture of the distribution of species abundance and can be drawn for the number of individuals, biomass, and ground area covered (Magurran, 1988; Schowalter, 1996).

Diversity indices like the Shannon entropy ("Shannon-Wiener index") and the Gini-Simpson index are not themselves diversities. The number of equally-common species required to produce a particular value of an index is called the "effective number of species". This is the true diversity of the community in question. For example, the true diversity associated with a Shannon-Wiener index of 4.5 is exp (4.5) = 90 effective species. Converting indices to true diversities (effective numbers of species) gives them a set of common behaviors and properties. After conversion, diversity is always measured in units of number of species. This enables us compare and interpret them easily, and develop formulas and techniques that do not depend on a specific index. It also helps to avoid the serious misinterpretations spawned by the nonlinearity of most diversity indices (Jost, 2006).

Although recent surveys have been conducted on the cynipids fauna in Iran (Azizkhani, 2006; Tavakoli et al., 2008; Zargaran et al., 2008), the oak cynipid gall wasps diversity is yet to be studied. Nazemi et al. (2008) reported species richness of oak gall wasps from Kurdistan, Ilam and Kermanshah provinces of Iran. Reducing the oak gall wasps diversity will be as an alarm for environmental health of oak forests. In this study, the oak gall wasps species diversity and their distribution in West-Azerbaijan Province of Iran were carried out in 2009 and 2010.

MATERIALS AND METHODS

Samplings were performed in west-southern regions of West-Azerbaijan province, Sardasht and Piranshahr, in areas such as: **Pardanan**, Mirabad, Nalas, Hamran, Sardasht and Shalmash (Table 1). The cynipids galls in these areas were gathered during these inspections made since the early growth season till the end of the season. The best number of samples was determined to be 40 trees base on Southwood and Henderson formula (2000) so that $N = (t \times s / D \times m)^2$, where *t* is student's *t-test* of standard statistical tables, *D* is predetermined confidence limit for the estimation of the mean expressed as a decimal, *m* is sampling mean and *s* is the standard deviation. In each tree, as unit of sampling, all of the occurred galls by oak gall wasps species on four branches (each branch length was 50 cm) in four cardinal directions were counted in the stations. In this study, 960 trees were counted in six sites (40 trees per each site) at four dates.

Measure of species diversity

Aspects of species diversity can be measured in a number of ways: measures of species richness count the number of species in a defined area; measures of species abundance sample the relative numbers among species; while measures of species diversity that simplify information on species richness and relative abundance into a single index are in extensive use (Magurran, 2004). The species richness of oak gall wasps in West-Azerbaijan province was estimated using:

1) The rarefaction quotient:

$$E(\hat{S}_n) = \sum_{i=1}^{s} \left[1 - \frac{\binom{N - N_i}{n}}{\binom{N}{n}} \right]$$

Where, *E(S n)* is the expected species number in a random sample of *n* individuals; S is the total number of species in the entire collection; Ni is the number of individuals in species *i;* N is the total number of individuals in collection = *I: Ni; n* is the value of sample size (number of individuals) chosen for standardization (n<N); and $\begin{bmatrix} N \\ n \end{bmatrix}$ is the number of combinations of *n* individuals that can be chosen from a set of N individuals =N! *In!* (N - *n)!*

2) Shannon-Weiner entropy indices: The computation of this diversity index uses this formula:

$$H_{SW} = -\sum_{i=1}^{N_o} [p_i * \log p_i],$$

Where, p_i is the proportion of the total number of individuals belonging to a morphotype, and N_o is the total number of individuals seen in this sample.

3) Gini-Simpson's indices: The diversity index is calculated using the following formula:

$$d = 1 - \sum_{i=1}^{N} \frac{n_i(n_i - 1)}{N(N - 1)}$$

Where, n_i is the number of individuals of a particular individuals and N is the total number seen in the sample.

Shannon evenness is measured by =H/Hmax

Conversion of common indices to true diversities

These measurements were conducted base on the formula of Jost (2006). Conversion of common indices to true diversities are:

Index x: Diversity in terms of x: Diversity in terms of p_i :

Shannon entropy x ≡ $-\sum_{i=1}^{S} p_i \ln p_i$ exp(x)

Table 1. Characteristics of study area with different habitats in West-Azerbaijan province.

Location	Latitude/Longitude	Climate	Oak species
Pardanan	36°28□′ N 45°18′ W	Very humid and cold	*Q. infectoria, Q. brantii* and *Q. libani*
Mirabad	36°15□′ N 45°22′ W	Very humid and cold	*Q. infectoria, Q. brantii*
Hamran	36°14□′ N 45°33′ W	Humid, mild cold	*Q. infectoria, Q. brantii*
Nalas	36°16□′ N 45°28′ W	Very humid Mediterranean	*Q. infectoria, Q. brantii*
Sardasht	36°11□′ N 45°24′ W	Humid, mild cold	*Q. infectoria, Q. brantii* and *Q. libani*
Shalmash	36°07□′ N 45°30′ W	Very humid Mediterranean	*Q. infectoria, Q. brantii* and *Q. libani*

$$\exp\left(-\sum_{i=1}^{S} p_i \ln p_i\right)$$

Gini-Simpson index $x \equiv 1-\sum_{i=1}^{S} p_i^2$ $1/(1-x)$ $1/\sum_{i=1}^{S} p_i^2$

RESULTS

In this research, six sites were studied in the spring, summer and fall seasons. The identified gall wasps were (37 species) classified in seven genera: *Andricus* (25 species), *Cynips* (three species), *Neuroterus* (four species), *Pseudoneuroterus* (one species), *Chilaspis* (one species), *Biorhiza* (one species) and *Aphelonyx* (two species) (Table 2). A number of seven and 30 galls was collected in spring and summer-fall, respectively. The highest and the least number of gall wasp species were collected from Pardanan station (28 species), and Nalas and Mirabad station (12 species), respectively. The genus *Andricus* had the highest number of species and the genera *Chilaspis*, *Pseudoneuroterus*, and *Biorhiza* were only found with a single species in all sites (Table 2). In all sites studied, seven gall wasp species were collected in spring, four of them were caught from catkins, two from leaves, and only a single species was found to induce galls on shoots. Most spring gall wasp species were found on *Q. brantii* and as this oak species was dominant in Mirabad area vegetation; the highest number of galls was counted in this region. The species *Pseudoneuroterus macropterus* and *Chilaspis israeli* were only collected on *Q. brantii* trees in Hamran station (Figure 1).

The oak species, *Q. infectoria*, *Q. brantii* and *Q. libani*, grow in West-Azerbaijan. These three species are of uneven distribution in the stations under study, while all three oak species are found in Pardanan and Sardasht stations, only two species (*Q. infectoria* and *Q. brantii*) grow in Hamran and Nalas. These oak species are of several sub-species which are under identification in and lowest numbers of spring gall wasps were res-pectively recorded in Mirabad and Sardasht stations (Figure 1). The highest number of gall wasps inducing summer and fall galls was found in Pardanan and Sardasht stations, while Mirabad station had the lowest number of gall wasps in the summer and fall (Table 3). This is in obvious contrast to the highest number of spring gall wasps (spring galls) found in Mirabad region. The highest number of gall wasps species was collected on *Q. infectoria*. Lists of oak gall wasps and their host plant as well as location of galls on oak trees, number of larval chamber and types of generation are shown in Table 2. Our findings show that two asexual generations of cynipid wasps were related to *Q. infectoria*. Four and one gall wasps species were collected on catkins and leaves of *Q. brantii*, respectively (Table 2). Our results show that there are no galls in spring and summer-fall on *Q. libani*. Species diversity in Sardasht, with only one species, was not measured. The highest and the least value of Gini-Simpson indices in the spring's and the fall's galls were recorded from Mirabad and Shalmash regions and from Sardasht and Mirabad regions, respectively (Table 3). Also, the highest and the least value of Shannon's entropy indices of spring and the fall galls were recorded from Mirabad and Shalmash regions and from Sardasht and Mirabad regions, respectively (Table 3).

Converting the species diversity indices to effective number of species showed that the difference is in fact huge; for example, the oak gall wasps community in Sardasht with a Gini-Simpson index of 0.9556 has the same diversity as a community with 22.52 equally-common species, while oak gall wasps community in Mirabad with a Gini-Simpson index of 0.8120 has the same diversity as a community with 5.32 equally-com-mon species. The second community is therefore only about 1/4 as diverse as the first community, and so on. Compared to other areas, spring galls were of the lowest rate of diversity in Shalmash region. The highest even-ness of species of spring and the fall galls were recorded from Shalmash and Mirabad regions, respectively (Table 3). The highest and the least expected number of oak gall wasps species (for the spring gall) in the sample with 100 individuals was the term of a comprehensive research plan. The highest estimated to be 4.86 species and 2.72 species in Mirabad and Shalmash regions. In this study, species richness of oak gall wasps on *Q. brantii* was the

Table 2. Gall wasps species associated with oak trees in West-Azerbaijan Province.

No.	Oak gall wasps species	Location of gall formation	Number of larvae chamber	Sexual or asexual generation	*Quercus* Host
1	*Andricus aries* (Giraud, 1859)	Shoot	One	Asexual	*Q. infectoria*
2	*Andricus askewi* (Melika and Stone, 2001)	Shoot	One	Asexual	*Q. infectoria*
3	*Andricus burgundus* (Giraud, 1859)	Catkin	One	Sexual	*Q. brantii*
4	*Andricus caputmedusae* (Hartig, 1843)	Shoot	Many	Asexual	*Q. infectoria*
5	*Andricus cecconii* (Kieffer, 1901)	Catkin	One	Sexual	*Q. brantii*
6	*Andricus conglomeratus* (Giraud, 1859)	Shoot	One	Asexual	*Q. infectoria*
7	*Andricus coriarius* (Hartig, 1843)	Shoot	Many	Asexual	*Q. infectoria*
8	*Andricus curvator* (Hartig, 1840)	Leaf	One	Sexual	*Q. infectoria*
9	*Andricus galeatus* (Giraud, 1859)	Shoot	One	Asexual	*Q. infectoria*
10	*Andricus grossulariae* (Giraud, 1859)	Catkin	One	Sexual	*Q. brantii*
11	*Andricus hystrix* (Trotter, 1899)	Shoot	One	Asexual	*Q. infectoria*
12	*Andricus kollari* (Hartig, 1843)	Shoot	One	Asexual	*Q. infectoria*
13	*Andricus lucidus* (Hartig, 1843)	Shoot	Many	Asexual	*Q. infectoria*
14	*Andricus mediterraneae* (Trotter, 1901)	Shoot	One	Asexual	*Q. infectoria*
15	*Andricus megalucidus* (Melika et al., 2003)	Shoot	Many	Asexual	*Q. infectoria*
16	*Andricus multiplicatus* (Giraud, 1859)	Leaf	Many	Sexual	*Q. brantii*
17	*Andricus panteli* (Kieffer, 1901)	Branch	Many	Asexual	*Q. infectoria*
18	*Andricus polycerus* (Giraud, 1859)	Branch	One	Asexual	*Q. infectoria*
19	*Andricus quercuscalicis* (Borgsdorf, 1783)	Fruit	One	Asexual	*Q. infectoria*
20	*Andricus quercustozae* (Bosc, 1792)	Shoot	One	Asexual	*Q. infectoria*
21	*Andricus seckendorffi* (Wachtl, 1879)	Shoot	Many	Asexual	*Q. infectoria*
22	*Andricus sternlichtii* (Bellido and Melika, 2003)	Shoot	One	Asexual	*Q. infectoria*
23	*Andricus theophrastea* (Trotter, 1901)	Shoot	One	Asexual	*Q. infectoria*
24	*Andricus tomentosus* (Trotter, 1901)	Shoot	One	Asexual	*Q. infectoria*
25	*Andricus megatruncicolus* Giraud	Shoot	One	Asexual	*Q. infectoria*
26	*Aphelonyx cerricola* (Giraud, 1859)	Shoot	Many	Asexual	*Q. brantii*
27	*Aphelonyx persica* (Melika et al., 2004)	Shoot	One	Asexual	*Q. brantii*
28	*Biorhiza pallida* (Olivier, 1791)	Shoot	Many	Sexual	*Q. infectoria*
29	*Chilaspis israeli* (Sternlicht, 1968)	Catkin	One	Sexual	*Q. brantii*
30	(Hartig, 1843) *Cynips cornifex*	Leaf	One	Asexual	*Q. infectoria*
31	*Cylindrocopturus quercus* (Fourcroy, 1785)	Leaf	One	Asexual	*Q. infectoria*
32	*C. quercusfolii* (Linnaeus, 1758)	Leaf	One	Asexual	*Q. infectoria*
33	*Neuroterus saliens* (Kollar, 1857)	Leaf	Many	Asexual	*Q. brantii*
34	*Neuroterus lanoginosus* (Giraud, 1859)	Leaf	One	Asexual	*Q. brantii*
35	*Neuroterus numismalis* (Geoffroy, 1785)	Leaf	One	Asexual	*Q. infectoria*
36	*Neuroterus quercusbaccarum* (Linnaeus, 1758)	Leaf	One	Asexual	*Q. infectoria*
37	*Pseudoneuroterus macropterus* (Hartig,1843)	Shoot	Many	Asexual	*Q. brantii*

highest in the spring galls (sexual generation) (Figure 2). Also, the highest and the least expected number of cynipid species in a sample with 250 individuals were estimated to be 22.85 and 6.9 species in the summer galls (asexual generation of gall wasps) in Pardanan and Mirabad regions (Figure 3).

DISCUSSION

Overall, 37 oak gall wasps species were collected in this research, and the 25 species belong to the genus *Andricus.* Cynipid-genera can be large; about 1000 species are distributed in 41 genera, with a mean of 24

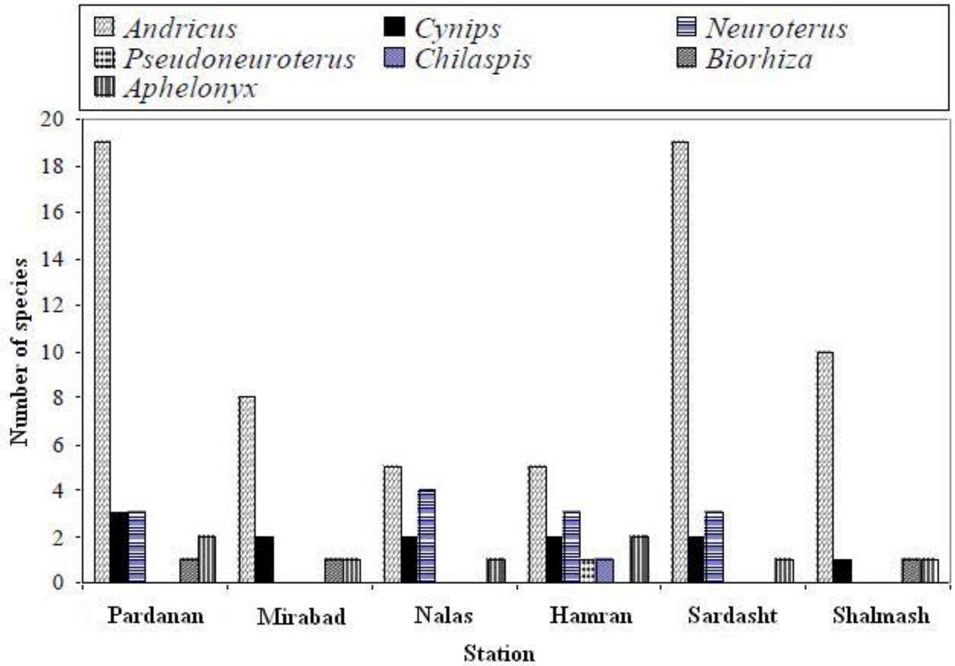

Figure 1. Distribution of oak gall wasps species in West-Azerbaijan province, 2009-2010

Table 3. Species diversity indices and true diversity in West Azerbaijan province

Station	Index						
	D	Species richness	Gini-Simpson	True diversity	Shannon's H'	True diversity	Shannon's evenness
April							
Pardanan	0.3243	4	0.6757	3.08	1.2419	3.46	0.8958
Mirabad	0.24	5	0.76	4.16	1.5159	4.55	0.9419
Nalas	0.3948	3	0.6052	2.53	1.0081	2.74	0.9176
Hamran	0.3851	3	0.6149	1.71	1.0104	2.75	0.9197
Sardasht	1	1	0	1	0	1	0
Shalmash	0.5131	2	0.4869	1.94	0.6773	1.97	0.9771
May							
Pardanan	0.3142	4	0.6858	3.18	1.2496	3.49	0.9014
Mirabad	0.2361	5	0.7684	4.31	1.5248	4.59	0.9474
Nalas	0.3629	3	0.6371	2.76	1.0534	2.87	0.9588
Hamran	0.3788	3	0.6212	2.64	1.0195	2.77	0.928
Sardasht	1	1	0	1	0	1	0
Shalmash	0.5121	2	0.4879	1.95	0.6765	1.96	0.976
September							
Pardanan	0.0454	24	0.9546	22.02	2.5562	12.88	0.9022
Mirabad	0.188	7	0.812	5.32	1.7929	6.07	0.9214
Nalas	0.1248	9	0.8752	8.01	1.9291	6.88	0.8779
Hamran	0.1044	11	0.8956	9.58	2.1916	8.95	0.914

Table 3. Contd.

Sardasht	0.0444	24	0.9556	22.52	2.6895	14.02	0.8978
Shalmash	0.1229	11	0.8771	8.13	2.1701	8.76	0.905
October							
Pardanan	0.0419	24	0.9581	23.87	2.5638	12.91	0.9049
Mirabad	0.186	7	0.814	5.38	1.7926	6	0.9212
Nalas	0.1189	9	0.8811	8.41	1.9671	7.15	0.8953
Hamran	0.1029	11	0.8971	9.72	2.1735	8.79	0.9064
Sardasht	0.0427	24	0.9573	23.42	2.634	13.93	0.8793
Shalmash	0.1035	11	0.8965	9.67	2.1586	8.66	0.9002

D, Predetermined confidence limit for the estimation of the mean expressed as a decimal.

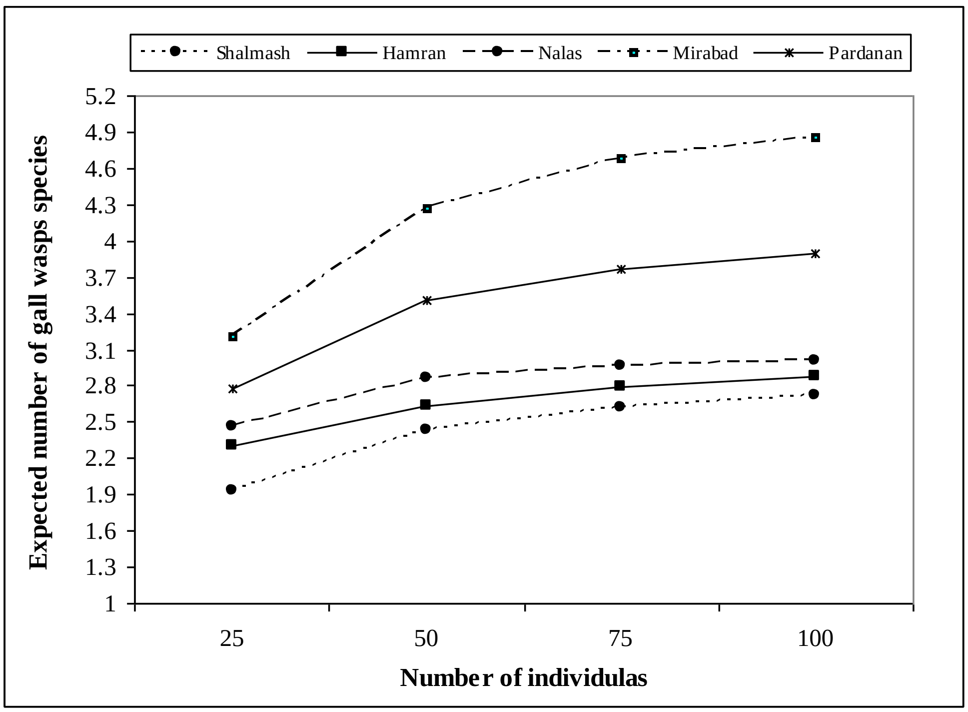

Figure 2. Oak gall wasps species richness base on Rarefaction curve in West-Azerbaijan in April-May (spring galls)

species per genus. Some genera are very large, with 300 species in *Andricus*, and 150 species in *Callirhytis* (Price, 2005). The highest number of oak gall wasps species was collected in this study during summer and fall seasons. Considering the dominance of *Q. infectoria* in all regions studied, the most abundant number of galls was found on this oak species. There are specific relationships between gall-inducing wasp species and the host oak tree species, and host plant distribution has a determinative impact on the distribution of a particular gall wasp species. The most amount of diversity in the number as well as the abundance of gall wasps was recorded in Pardanan, where *Q. infectoria* had formed about 90% of the local vegetation. Some species of gall wasps were collected from only a single area. While factors like altitude (based on the vertical distance from the surface of free seas), and regional climate are similar in some areas under comparison. The differences in the geographical distribution of oak subspecies may be attributed to the difference in the presence of the regions. This implies to the important role of the host plant sub-species distribution in the determination of gall wasp distribution. Therefore, the diversity in the local population of oak trees at the level of species and inferior

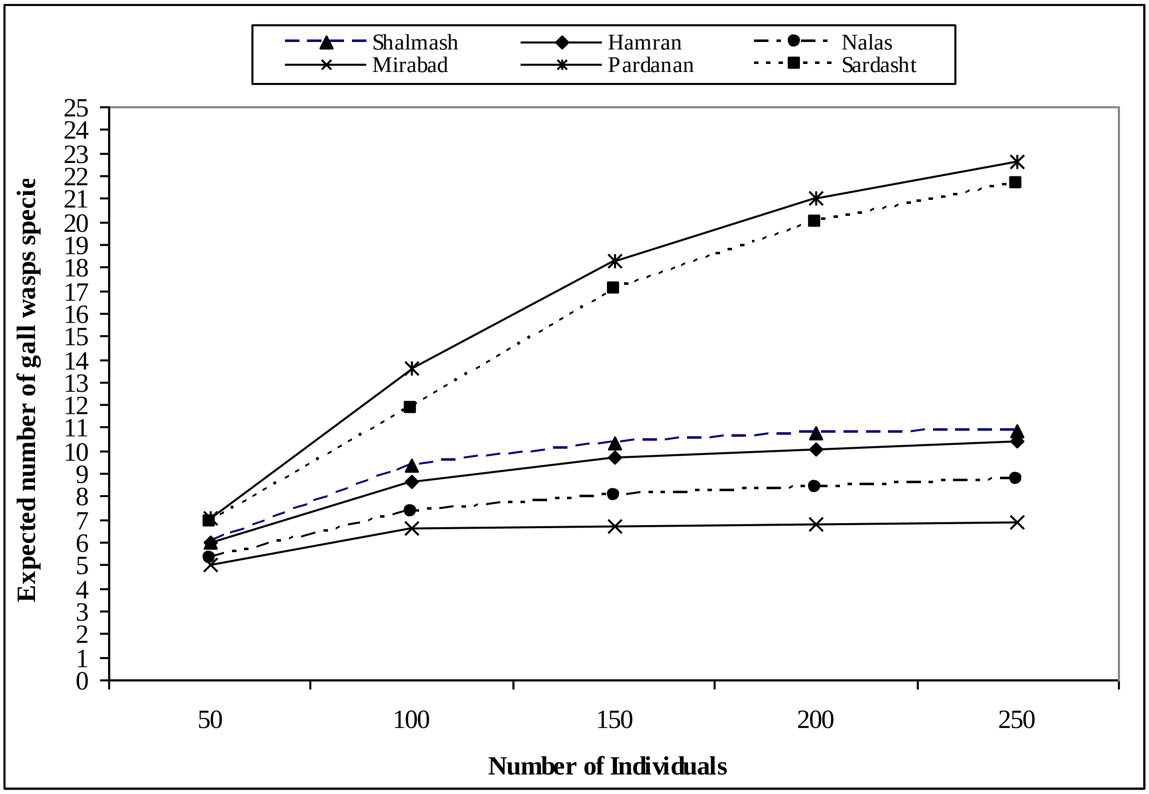

Figure 3. Oak gall wasps species richness base on Rarefaction curve in West-Azerbaijan in September-October (summer-fall galls)

can be considered as one main factor involved in the distribution of gall wasp species besides other factors such as regional climate, annual temperatures, and vegetation.

Some studies indicate that the species richness gall-inducing insect increases as environments become hotter and drier, while others suggest that these factors have no effect. The role of plant species richness in determining species richness of galling insects is also controversial (Blanche, 2000). Local climate was not so effective on some gall wasp species such as *Andricus grossulariae*, whose galls are formed during spring and encountered in all three types of climate. It is expectable that vegetation and the kind of oak subspecies are involved in this case. Pardanan and Mirabad stations are of similar climate, however, the specific diversity of gall wasps was found to be very different. The difference must be sought in vegetation discrepancies of two regions. The Pardanan and Sardasht areas despite of their different climates were of similar vegetations, a fact that had ended to a similar rate of specific diversity in both districts (28 species in Pardanan versus 25 species in Sardasht). Local climate affects gall wasp diversity and impacts on the distribution of some gall wasp species. All three oak species were present in Shalmash as in Pardanan (Table 1), but because of the difference in climate, the latter region was of less diversity of gall wasp species. With change in latitude and presence of different oak species in the sites, species richness of oak gall wasps was very different. The most important point is that the role of the oak sub-species and region climate in the distribution of oak gall wasps should not be ignored. Higher species diversity is generally thought to indicate a more complex and healthier community because a greater variety of species allows for more species interactions, hence greater system stability, and indicates good environmental conditions. Oak gall wasps species richness might be expected to increase with the richness of host plant species (Starzomski et al., 2008). Also, abiotic factors, such as water stress, and other biotic factors (plant age and natural enemies) may also affect tropical gall wasps species richness at different scales (Veldtman and Mcgeoch, 2003; Price, 2005).

The results obtained herein therefore add to the few data available on species richness of oak gall wasps on *Q. infectoria* and *Q. brantii* in Iran. Previously, Chodjai (1980) reported 36 oak gall wasps related to *Q. infectoria* from Iran. Azizkhani (2006) also reported 24 gall wasps on *Q. infectoria* and *Q. brantii*. In addition, it has been observed that gall wasps exhibit a high degree of host plant specificity and chemistry that may be crucial for the success of these insects (Cook et al., 2002; Rokas et al., 2003). Results have also shown that plant chemistry may play an appreciable role in the selection of plant species by gall wasps (Abrahamson et al., 2003).

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