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Note on the *Medicago rigidula* (L.) All. in Iran

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ABSTRACT

A taxonomic status of the *Medicago rigidula* was reviewed in Iran. Morphological characteristics and pollen grains were studied in 72 populations of this species collected from natural habitats were. The results showed that the populations of *M. rigidula* have three-pore or four-pore pollen grains. Three-pores populations had less coils of pod, long and hooked spine that correspond with the *M. rigidula*, but four-pores populations had more coils without spine or with tubercles or straight, short spine tip that correspond with the *M. rigiduloides*. Thus, both species *M. rigidula* (L.) All. and *M. rigiduloides* E. Small were distributed across Iran.

Keywords: Iran, *Medicago rigidula*, *M. rigiduloides*, Pollen grains.

INTRODUCTION

Medicago rigidula (L.) All. is currently interpreted as a Mediterranean species, indigenous to North Africa, Europe and Asia, and spontaneous in the new world and elsewhere [1]. The most intensive analysis of intraspecific variation of *M. rigidula* is that of Heyn [2, 3], who concluded her account by stating the great variability as well as the two recorded chromosome numbers ($2n=14, 16$) suggest that further research might prove the need for subdividing the species. Lesins and Lesins [4] in their monograph on *Medicago*, recognized only the chromosome count of $2n=14$ for *M. rigidula* and accepted the four varieties recognized by Heyn [3], while noting that she admits that it is impossible to define boundaries between intraspecific entities with precision, and she lists various characters, which vary greatly, and occur in different combinations. The name *Medicago rigidula* (L.) All. has been applied to two groups that merit separate species status [1]. Almost all of the plants of Europe and North Africa are preferable to one of these species, and almost all of the plants of Asia are preferable to the other. *Medicago rigidula* recognized as European plants and *M. rigiduloides* recognized as Asian plants and he made the *M. rigidula* var. *submissa* (Boiss.) Heyn, as a synonym of *M. rigiduloides*[5].

Here we report the pollen morphology of populations of *Medicago rigidula* and its significance in their taxonomic relationships in Iran.

MATERIALS AND METHODS

In this study, several locations in Iran have been visited and specimens were collected and reserved in the National Plant Gene Bank of Iran (NPGBI) which showed by TN. Therefore, herbarium specimens of 72 *M. rigidula* populations in NPGBI have been studied. The studied populations and their localities are listed in (Table 1). Morphological characters and pollen morphology were studied in five plants from each population. The pollens were acetolysed in the standard way [6,7]. For light microscopy (LM), slides were prepared by mounting the pollen in glycerol jelly. For scanning electron microscopy (SEM), pollen grains were coated as dry specimens with gold.

Table1. Voucher specimen, pollen pores, number of coils and spine type in studied populations.

Row	Voucher specimen (TN)	species	Pollen pores	Number of coils	Spine curvature	Row	Voucher specimen (TN)	species	Pollen pores	Number of coils	Spine curvature
1	1845	<i>M.rigiduloides</i>	Four-pores	5	1	37	1552	<i>M.rigiduloides</i>	Four-pores	6	1
2	1884	<i>M.rigiduloides</i>	Four-pores	6	2	38	1037	<i>M.rigidula</i>	Three-pores	4	4
3	1854	<i>M.rigiduloides</i>	Four-pores	6	1	39	1617	<i>M.rigidula</i>	Three-pores	5	3
4	1861	<i>M.rigiduloides</i>	Four-pores	6	1	40	1554	<i>M.rigiduloides</i>	Four-pores	6	1
5	1842	<i>M.rigiduloides</i>	Four-pores	6	1	41	1557	<i>M.rigiduloides</i>	Four-pores	5	0
6	1853	<i>M.rigiduloides</i>	Four-pores	6	1	42	1616	<i>M.rigiduloides</i>	Four-pores	5	0
7	1955	<i>M.rigiduloides</i>	Four-pores	5	0	43	1643	<i>M.rigiduloides</i>	Four-pores	7	1
8	1957	<i>M.rigidula</i>	Three-pores	4	4	44	1682	<i>M.rigiduloides</i>	Four-pores	5	1
9	1964	<i>M.rigidula</i>	Three-pores	5	4	45	1666	<i>M.rigiduloides</i>	Four-pores	6	1
10	2290	<i>M.rigiduloides</i>	Four-pores	5	1	46	1112	<i>M.rigiduloides</i>	Four-pores	5	0
11	2213	<i>M.rigiduloides</i>	Four-pores	7	0	47	1860	<i>M.rigiduloides</i>	Four-pores	6	1
12	2047	<i>M.rigidula</i>	Three-pores	3	4	48	1848	<i>M.rigidula</i>	Three-pores	4	3
13	2285	<i>M.rigidula</i>	Three-pores	4	4	49	1847	<i>M.rigiduloides</i>	Four-pores	5	1
14	2211	<i>M.rigiduloides</i>	Four-pores	6	0	50	1888	<i>M.rigidula</i>	Three-pores	5	4
15	2212	<i>M.rigiduloides</i>	Four-pores	6	0	51	2264	<i>M.rigiduloides</i>	Four-pores	6	1
16	2269	<i>M.rigiduloides</i>	Four-pores	6	2	52	2137	<i>M.rigiduloides</i>	Four-pores	6	2
17	2206	<i>M.rigiduloides</i>	Four-pores	6	1	53	2153	<i>M.rigiduloides</i>	Four-pores	5	0
18	2209	<i>M.rigiduloides</i>	Four-pores	5	0	54	2161	<i>M.rigiduloides</i>	Four-pores	7	1
19	2268	<i>M.rigiduloides</i>	Four-pores	6	1	55	2219	<i>M.rigiduloides</i>	Four-pores	5	0
20	2292	<i>M.rigiduloides</i>	Four-pores	6	1	56	2203	<i>M.rigiduloides</i>	Four-pores	6	0
21	2220	<i>M.rigidula</i>	Three-pores	5	4	57	2081	<i>M.rigiduloides</i>	Four-pores	5	2
22	2138	<i>M.rigiduloides</i>	Four-pores	5	1	58	2207	<i>M.rigiduloides</i>	Four-pores	6	1
23	2294	<i>M.rigiduloides</i>	Four-pores	6	1	59	2296	<i>M.rigiduloides</i>	Four-pores	6	1
24	2325	<i>M.rigidula</i>	Three-pores	4	3	60	2205	<i>M.rigiduloides</i>	Four-pores	6	1
25	2324	<i>M.rigiduloides</i>	Four-pores	5	1	61	2218	<i>M.rigiduloides</i>	Four-pores	6	0
26	2322	<i>M.rigiduloides</i>	Four-pores	5	0	62	2216	<i>M.rigiduloides</i>	Four-pores	5	1
27	2327	<i>M.rigiduloides</i>	Four-pores	5	1	63	2210	<i>M.rigiduloides</i>	Four-pores	6	1
28	2307	<i>M.rigiduloides</i>	Four-pores	6	1	64	2215	<i>M.rigiduloides</i>	Four-pores	6	0
29	2305	<i>M.rigiduloides</i>	Four-pores	5	1	65	2214	<i>M.rigiduloides</i>	Four-pores	6	1
30	945	<i>M.rigidula</i>	Three-pores	4	2	66	2223	<i>M.rigiduloides</i>	Four-pores	6	0
31	927	<i>M.rigidula</i>	Three-pores	4	4	67	2284	<i>M.rigidula</i>	Three-pores	5	3
32	653	<i>M.rigiduloides</i>	Four-pores	5	0	68	214	<i>M.rigiduloides</i>	Four-pores	5	1
33	899	<i>M.rigiduloides</i>	Four-pores	5	1	69	62	<i>M.rigiduloides</i>	Four-pores	5	0
34	999	<i>M.rigiduloides</i>	Four-pores	5	1	70	206	<i>M.rigiduloides</i>	Four-pores	6	1
35	837	<i>M.rigiduloides</i>	Four-pores	5	0	71	2226	<i>M.rigiduloides</i>	Four-pores	5	1
36	1652	<i>M.rigiduloides</i>	Four-pores	6	1	72	892	<i>M.rigiduloides</i>	Four-pores	5	1

TN: Number of genotypes in national plant gene bank of Iran
 Spine curvature: 0= none; 1= uncurved; 2, 3, 4= highly curved

RESULTS AND DISCUSSION

Results showed that based on number of fruit coils and curvature of fruit spines it is possible to identify two kinds of plants. Figure 1 shows how these two characters can be used rapidly for differentiating two groups of populations. Pods in some populations had less coils (≤ 5) and fruit spines usually more hooked tip and longer than others. The others had more coils (≥ 5) with straight tip spines or spineless, or with tubercles.

In addition, results showed that based on palynology, pollen grains divided into two groups, triangular on polar view with three-pores and the other group was rectangular on polar view with four-pores [8]. The pollen grain of three-pores and four-pores populations are illustrated in Figure 2 and important data of them are presented in Table 1. Having three-pores or four-pores in this species had been noted by Small *et al.* 1989. The pod of three-pores populations had less coils with hooked tip and long spine that correspond with the Small's *M. rigidula*, but four-pores populations had more coils with straight tip spines that correspond with the Small's *M. rigiduloides*. Mehregan [9] said the study of Small and their colloquies [1] represents a cline variation in *M. rigidula* from Europe toward Asia and this kind of variation is a common aspect of most species with spiny pods, and regarding this distinction of Asian populations of *M. rigidula* and placing them in *M. rigiduloides* did not accepted. Although this species has variation in Iran and is closely allied to *M. rigiduloides* but the last one it can be distinguished specially by the substantial presence of 4-pored pollen (mostly 3-pored in *M. rigidula*), and also by fruits with more coils and straighter tip spines and with fruits spineless or with tubercles more than *M. rigidula* [1].

This study demonstrated that both species were distributed across Iran against Small's *et al.*, opinion but *M. rigiduloides* populations were more distributed than *M. rigidula* (Fig.3). In addition, 13 populations of 72 studied populations were belonging to *M. rigidula* and the others were *M. rigiduloides*.

Fig.1. variation in pods shape and spines in *Medicago* populations (magnifications 10X)

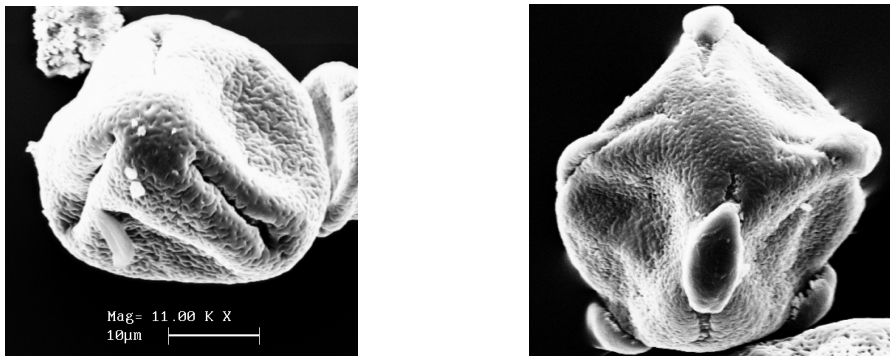
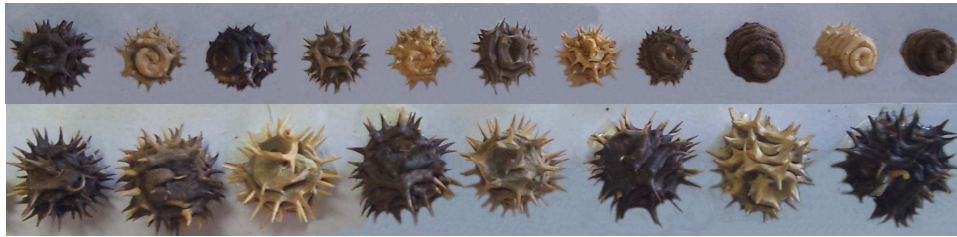


Fig. 2. Pollen grains in *Medicago* populations, three-pores (left) and four-pores (right) (Magnifications 1100X)

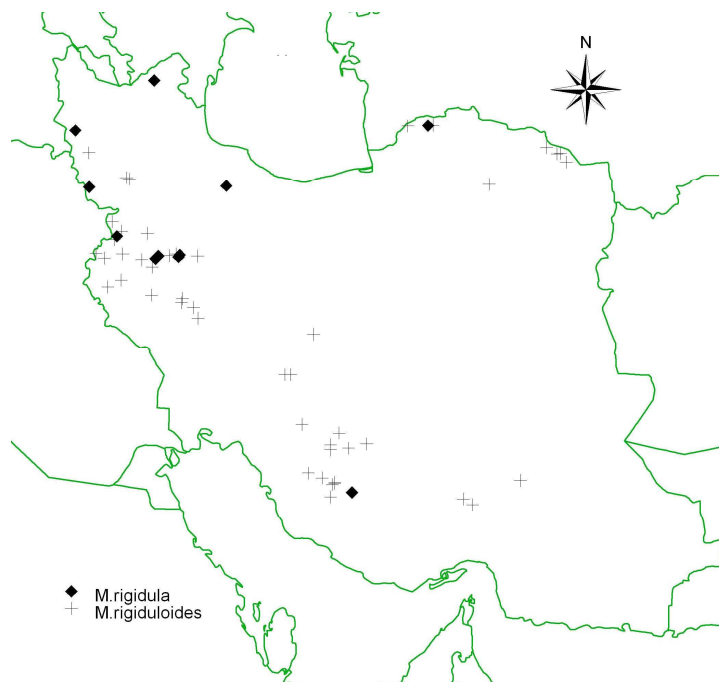


Fig 3. Distribution of *M. rigidula* and *M. rigiduloides* species in Iran.

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