



Relationship Between Ecological Species Groups and Environmental Factors in Fandoghlu Rangelands of Ardabil, Iran

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ABSTRACT

Aim This study was performed to investigate the relationship between ecological species groups and environmental factors in Fandoghlu rangelands of Ardabil province in the northwest of Iran, assuming that plant species distribution has been correlated with various of complex environmental gradients.

Materials & Methods The samples were digested by a combination of nitric acid and perchloric acid and the concentration of elements was measured by atomic absorption spectroscopy. Data were collected from 180 sampling plots (1m²) in an area of 3.27km² using the systematic-random method. In each plot, environmental factors (topography, climate, and soil variables) and the percent of vegetation cover for each species were recorded. TWINSpan method and Canonical Correspondence Analysis (CCA) were used to define ecological species groups and determine the relationship between ecological species groups and environmental factors, respectively.

Findings Results of using TWINSpan for 180 plots classified rangeland communities into three groups. The first group contained 110 plots, the second group 40 plots, and the third group 30 plots. Using ISA, vegetation species groups in the first group contained 8 species, the second group 11 species, and the third group 8 species. In each group, the name of the species with the highest index value was selected as the group's name. Thus using CCA, the group of *Trifolium pratense* had a relation with aspect, slope, organic matter, magnesium, temperature, and volumetric soil moisture. The *Leucanthemum vulgare* was related to phosphorus, elevation, lime, sand, clay, and potassium. The group of *Trifolium repense* had a relation with pH, electrical conductivity, calcium, diffusible clay, temperature, silt, and sodium. The factors of diffusible clay and electrical conductivity with the first CCA axis and potassium and phosphorus factors with the second CCA axis had the strongest correlations.

Conclusion Using the results of this study, we can evaluate the habitat conditions and vegetation quality of Fandoghlu rangelands. Moreover, used high-quality rangeland species such as *Trifolium* to counteract the spread of invasive species such as *Leucanthemum vulgare*.

Keywords Environmental Factors; TWINSpan; Indicator Species; CCA

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Introduction

Plants are a conspicuous and stationary habitat element that can be highly sensitive to biotic and abiotic factors, expressing localized changes through survival, growth, and reproduction [1-4]. Therefore, phytosociology's major applications are ecological assessment, vegetation mapping, monitoring environmental changes, and nature conservation inventory [2-5]. Some approaches can select several indicator plants, often a small subset of the total ground flora [6]. So indicator species groups are used instead of individual indicator species. Ecological species groups include plants that repeatedly occur together when certain combinations of site factors occur; they are distinguished by their species composition and abundance patterns among sampling plots. Identifying ecological species groups involves recognizing species that share similar environmental affinities and typically occupy the same sites across the landscape in predictable relative proportions [2, 7]. They help distinguish and map landscape ecosystems in the field by their presence or absence and by the relative coverage of plants in each group [8, 9]. Besides, the data of ecological species groups are used as sources of information on the spatial distribution of vegetation cover [10]. Well-defined species-environment relationships are important to understand vegetation patterns on rangelands [9].

Moreover, determining which factors control the presence, number, identify, and relative abundance of plant species remains a central goal in ecology. Quantitative analysis of ecological relationships between vegetation and the environment has become an essential means in modern vegetation ecology [3, 4, 11-13]. Quantitative analysis, especially quantitative classification methods and ordination techniques, has been widely used to illustrate the ecological relationships between vegetation cover and the environment [12, 14]. Zhang & Oxley [15] conclude using classification; one can evaluate the importance of the assignment of abundance coefficients to vegetation composition. Also, they had stated classification is the importance of cluster quadrats of high similarity. These clusters are key factors that are related to abiotic variables [12]. It is, therefore, possible to analyze floristic structures by a hierarchical classification [12]. One of the most popular hierarchical divisive clustering techniques in community ecology is the two-way

indicator species analysis (TWINSpan) [12, 15]. TWINSpan simultaneously classifies species and samples [9, 16]. Ordination is a collective term for multivariate techniques that adapt a multidimensional group of data points so that when it is projected onto a two-dimensional space, any intrinsic pattern the data may possess becomes apparent upon visual inspection [12, 17, 18]. Canonical Correspondence Analysis (CCA) is a ranking method based on correspondence analysis [2, 12, 18]. The method, which is also called "multivariate direct gradient analysis", combines correspondence analysis with multiple regression analysis [2]. Each step of the calculation is regressed with environmental factors. The basic ideas behind CCA are that the iterative process of the correspondence analysis sorts the quadrat's coordinate values, which are not always available. The data is then subject to multivariate linear regression with the environmental factors [2, 9, 18].

Assuming that plant species distribution is associated with various complex environmental gradients, this study was performed to investigate the relationship between environmental and ecological species groups in the Fandoghlu rangelands of Ardabil province in northwest Iran. Vegetation groups were defined using the TWINSpan technique, and determining relationships between environmental factors with ecological species groups to identify the main factors that affect vegetation types' distribution was conducted using the CCA technique.

Materials and Methods

Area of study

Fandoghlu rangelands are located in the northwest of Iran in 24km northeast of Ardabil, the capital city of Ardabil province, and in 9km southeast of Namin county along with the mountain ranges of Alborz (Talesh) between 38°23'55" N to 38°24'55" N and 48°33'05" E to 48°34'16" E (Figure 1). The minimum altitude in the study area is about 1400, and the maximum is 1600m. The area's average rainfall using the nearest station (Namin synoptic station with a height of 1345m above sea level) is 272.21 mm, and the average temperature is 10.9°C. The study region has moderate summers and cold winters (Diagram 1). For 3 to 4 months of the year, snow and ice are covered, and most rainfall occurs in autumn and winter and early spring [19, 20]. The study area in the classification of Iran's

ecosystems is the ecotone between the two regions of Europe-Siberian and Iran-Turonian regions [21, 22]. These rangelands have changed from forest ecosystems and present a physiognomic structure of vegetation is meadow, which is considered one of Iran's best rangelands [19, 22]. The main part of the forage of these rangelands harvested in the spring and summer and used for winter forage of livestock. Each year, these rangelands by the local livestock (cattle, sheep, and goats) from mid-May to mid-November are grazed for almost seven months. This region is one of the main recreational areas of Ardabil province.

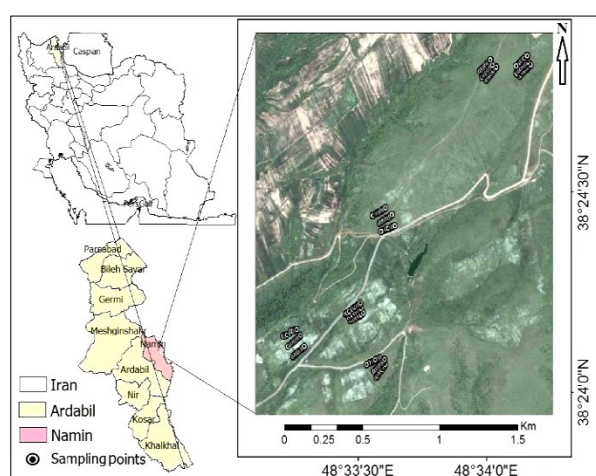


Figure 1) Location of the study area and sampling points in Namin county, Ardabil province, and Iran

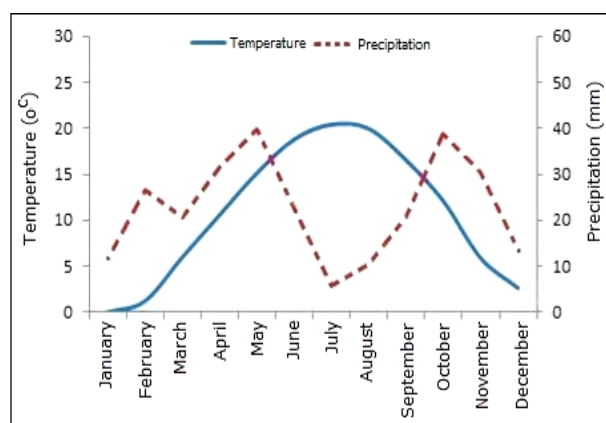


Diagram 1) Embrotermic curve of Namin synoptic station

Site selection and sampling

Using the land-use map [19] and field observations of Fandoghlu rangelands, in an area of 3.27km², 6 sites were selected [20] by considering-vegetation structure (dominant species are forb and grass) and previous studies

[22-24]. Three sampling transect [25, 26] with a length of 200m [27] and a distance of 100m from each other were established in each site. 10 plots (1m²) with a distance of 20m were established [20]. The location of plots was recorded with a global position system (GPS) [20]. In each plot, the canopy cover percentage of each species was recorded. Each species in the sampling plots was collected as a herbarium specimen and transferred to the herbarium of the University of Mohaghegh Ardabili and used valid references [28-30]. Soil samples were collected from the beginning, middle, and end of each transect and mixed. These samples were taken from the depth of the rootstock of species [31]. Soil attributes such as pH (using pH meter in saturated extracts), electrical conductivity (using EC meter in saturated extracts), calcium and magnesium (using the titrimetric method), potassium and sodium (using flame photometer), phosphorus (using Olsen method with a spectrophotometer), lime (using the titrimetric method), organic matter (by calculating organic carbon using the modified Walkley-Black wet oxidation procedure), soil texture (using two hydrometer readings method), diffusible clay (using double hydrometer method) and volumetric soil moisture (using measuring the weight difference between wet and dry soil in the oven) were measured in the laboratory [32]. Topographic data (elevation, slope, and aspect) were derived from a 20m digital elevation model, and climate data (temperature) were derived using a gradient equation [20, 22, 24, 33].

Data analysis

Kolmogorov-Smirnov test was used to test all parameters' normality, and Levene's test was used to examine the homogeneity of variances. A two-way indicator species analysis (TWINSPAN) was used to classify the 180 plots with similar species abundance patterns. The cut-off level of 'pseudo-species' followed the PC-ORD Software's default. Indicator species analysis (ISA) was used to extract those significantly associated with each group. Therefore, the indicator species and species groups were distinguished. TWINSPAN and ISA were performed by PC-ORD 5.10 [34]. To determine whether to use linear or unimodal based numerical methods, Detrended Correspondence Analysis (DCA) [16] for preliminary analysis of indicator species data and to determine lengths of the gradients was

used [35] in R software, version 3.6. The gradient was sufficiently long to justify to use of CCA, which assumes species have a unimodal response to the environmental gradients [36]. Thus after classification of the plots and species, relationships between environmental factors (topography, climate, and soil variables) and indicator species were assessed using CCA [37] in R software. CCA is used with a forward selection of explanatory variables to provide an estimate of the best set of variables for predicting species-environment relation and to provide a ranking of the relative importance of the individual explanatory variables [38]. Statistically significant environmental factors (elevation, slope, aspect, temperature, pH, electrical conductivity, calcium, magnesium, potassium, sodium, phosphorus, lime, organic matter, soil texture (clay, silt, sand), diffusible clay, and volumetric soil moisture) were examined using the Monte Carlo permutation test [38]. Permutation tests were run with 1000 permutations.

Findings

Classification

Results of using TWINSpan for 180 plots classified rangeland communities into three groups (Diagram 2). The first group contained 110 plots, the second group 40 plots, and the third group 30 plots. Indicator species analysis (ISA) was used to extract those significantly associated with each group. Using ISA, from 80 species, 27 species were selected as dominant or indicator species of the region and formed 3 vegetation species groups. In the first group, vegetation species groups contained 8 species, the second group 11 species, and the third group 8 species. In the first group, the indicator species were *Trifolium pratense*, *Trifolium compositum*, *Trisetum flavescens*, *Stachys byzantine*, *Dactylis glomerata*, *Cynosurus echinatus*, *Ranunculus millefolius*, *Poa bulbosa*. Moreover, *Trifolium pratense* has the highest indicator value, and this group was named as *Trifolium pratense*. In the second group, the indicator species were *Leucanthemum vulgare*, *Luzula multiflora*, *Hieracium matrense*, *Hieracium pilosella*, *Lotus corniculatus*, *Polygala anatolica*, *Hypericum perforatum*, *Veronica gentianoides*, *Vulpia myuros*, *Cirsium* sp., *Leontodon hispidus*. Moreover, *Leucanthemum vulgare* has the highest indicator value, and this species named this group. In the third group, the indicator

species were *Trifolium repense*, *Trifolium micranthum*, *Poa pratensis*, *Bromus briziformis*, *Bromus scoparius*, *Taraxacum syriacum*, *Barbarea minor*, *Hypericum linarioides*, and the *Trifolium repense* has the highest indicator value. Thus this group was named by this species.

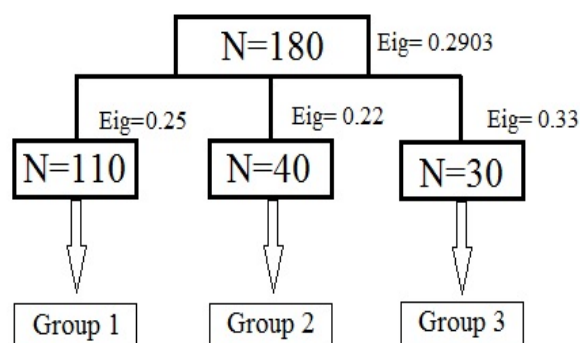


Diagram 2) Diagram of TWINSpan analysis on 180 sample plots in the study area

Ordination

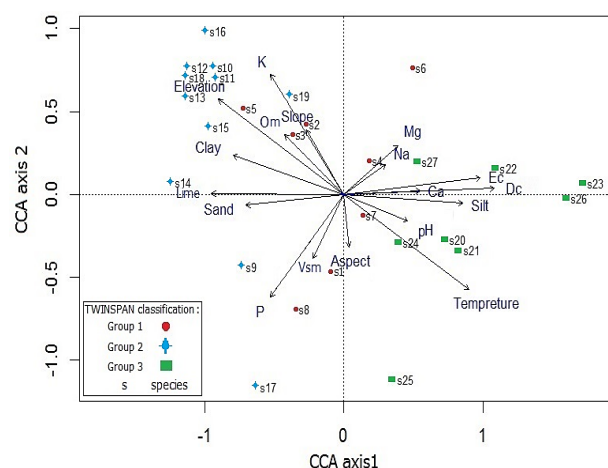
By considering the gradient's length, which was 3.18, thus CCA was selected as the ordination method. According to Table 1, first axis (eigenvalue=0.498) accounted for 35.33% variation of environmental factors and the second axis (eigenvalue=0.264) explained 18.71% variation. The species-environment correlations calculated for the first two axes of CCA were: 0.964 and 0.887 (Table 1). The Monte Carlo permutation test showed that the species-environment relationships revealed by axis 1 and 2 was significant ($p = 0.01$; Table 1). Correlations between the first two CCA axes and environmental factors showed the strongest correlations with the first CCA axis for diffusible clay, electrical conductivity, lime, elevation, temperature, and silt. The second CCA axis was highly correlated with K and P. These factors are significant (Table 2). The results of the CCA ordination is shown in Diagram 3.

Table 1) CCA for environmental data and Monte Carlo test for species-environment correlations

Parameters	Axis 1	Axis 2
Eigenvalue	0.4989	0.2641
Proportion Explained (%)	35.33	18.71
Cumulative Proportion (%)	35.33	54.04
Species-environment correlation	0.964	0.887
P (Monte Carlo)	0.01	0.01

Table 2) Intra-set correlations of environmental factors with the first two CCA axes

Environmental factors	Axis 1	Axis 2
Diffusible clay (%)	0.7311	0.0217
Electrical conductivity (ds/m)	0.6628	0.0747
Lime (%)	-0.6445	0.0106
Elevation (m)	-0.5934	0.4614
Temperature (°C)	0.5910	-0.4603
Silt (%)	0.5790	-0.0484
Potassium (meq/l)	-0.3378	0.5566
Phosphorus (meq/l)	-0.3346	-0.4755
Slope (%)	-0.1734	0.2971
Aspect (modified)	0.0252	-0.2363
pH	0.3079	-0.1341
Calcium (meq/l)	0.3716	0.0120
Magnesium (meq/l)	0.2514	0.2313
Sodium (meq/l)	0.1975	0.1426
Organic matter (%)	-0.2797	0.2852
Clay (%)	-0.5358	0.1955
Sand (%)	-0.4763	-0.0477
Volumetric soil moisture (%)	-0.1375	-0.2926

**Diagram 3)** CCA-ordination diagram of the environmental data

Each group reflects the ecological relationships between vegetation species groups and their environmental factors. The first CCA axis represents most of the factors, namely, the diffusible clay, electrical conductivity, temperature, and silt have increased from left to right, and elevation and lime have increased from right to left along the first axis. The second axis also shows two factors: potassium, which has declined from top to bottom, and phosphorus has declined from the bottom to top along the second axis. Considering each variable's angle relative to the main axes and the length of each variable, the ecological species groups in TWINSpan and ISA analyses were in accordance with the CCA analysis. The group of *Trifolium pratense* (group 1) was located in the center of the axis and showed a significant

relation with aspect, slope, organic matter, magnesium, temperature, and volumetric soil moisture. In other words, these factors are the indicators of sites of species group 1. The *Leucanthemum vulgare* (group 2) was mostly placed on the left of axis 1 and top of axis 2 and had a relation with phosphorus, elevation, lime, sand, clay, and potassium. Thus these factors are the indicators of sites of species group 2. The *Trifolium repense* (group 3) was mostly placed on the right of axis 1 and bottom of axis 2 and related to pH, electrical conductivity, calcium, diffusible clay, temperature, silt, and sodium (Diagram 3).

Discussion

Using TWINSpan, ISA, and CCA methods, this study presents quantitative information on the relationship between vegetation and environmental factors in the Fandoghlu rangelands of Ardabil in Iran. 180 plots were classified into three groups by TWINSpan, and 27 species were selected as indicator species of the region by ISA and formed three vegetation species groups. The distribution of plant species groups could demonstrate the effect of environmental factors. CCA ordination was used because the length of the gradient was 3.18. Classification and CCA ordination results were compared. TWINSpan groups could be recognized in the CCA graph.

Results showed that the first axis represented most of the factors, and it was positively correlated to diffusible clay, electrical conductivity, temperature, silt, and was negatively correlated with elevation and lime.

Table 2 shows that diffusible clay has the highest line length and has the lowest angle compared to the first axis. Thus diffusible clay has been one of the main factors controlling vegetation distribution and isolation of the ecological species grouping. Soil erosion has been directly linked to water dispersible clay's rate and soil [39]. When soils are submerged in water, diffusible clay affects a lot of soil physical and chemical properties [40]. Electrical conductivity has been the second leading factor controlling vegetation distribution and isolation of the ecological species grouping. This factor ranks second in terms of line length and angle with the first axis. EC is an index of salt concentration and an indicator of electrolyte concentration of the solution. EC of the nutrient solution is related to the number of ions available to plants in the root

zone [41]. Tavili & Jafari [42] stated that EC with other factors were the most important factors that correlated strongly with the distribution of ecological groups in the north of Iran.

Moreover, Zare Chahuki & Zare [43] expressed that electrical conductivity is the most effecting factors on the presence of *Tamarix ramosissima* in Garizat rangelands of Yazd province, which confirms the results of this study. The local climate constrains plant species distribution. The temperature in terms of line length and angle with the first axis is the most important factor in the separation of ecological species groups, and it plays an important role in the start of growing season, growth period, and type of vegetation. Borna *et al.* [44] reported that the average temperature of the wet season and average temperature during the dry season were the most important environmental factors influencing the distribution of *Astragalus gossypinus* in summer rangelands of Baladeh Nour, that it confirms the results of this study. Soil texture greatly influences on the formation of plant communities. Soil texture is effective in plant distribution due to humidity, soil water holding capacity, the cycle of nutrients, aeration, and depth of roots [45]. Silt has been one of the soil factors affecting the separation of plant communities in the study area. This factor has a longer line length and a slight angle with the first axis's positive part. The results confirm the findings of Zolfaghari *et al.* [46]. They reported that silt is the most important and effective factor in classifying plant communities in Plour in Amol, Iran. The existence of lime and appropriate size plays a role in generating good building and modifying soil acidity and, consequently, food absorption. Lime is the first factor in the left axis, with the first axis having a very slight angle and it has been one of the most important factors affecting the segregation of plant communities and the results of this study, which confirm the results of Zare Chahouki *et al.* [47]. They are noted that there is a relationship between plant communities' distribution and environmental factors and the parameters like lime, texture, nitrogen, and potassium, which have the greatest impact on the distribution of plant communities among the study factors. Elevation controls the vegetation patterns and isolation of the ecological species grouping in the Fandoghlu rangelands. Vittoz *et al.* [48] showed that altitude is an important factor because the elevation changes are associated

with humidity, temperature, soil type, and other factors that influence plant communities. Aghaei *et al.* [49] conclude that elevation is effective in the separation and distribution of ecological groups, which consistent with the results of this study. The second axis represented soil's chemical properties and was positively correlated to potassium and was negatively correlated with phosphorus. These factors have a longer line length and have an angle with the second axis. Thus, potassium and phosphorus have been important factors in separating ecological species groups in the study area. Potassium plays an important role in regulating photosynthesis, carbohydrate transport, protein synthesis, and other important physiological processes. In addition, the existence of potassium in the soil makes it easy to transform the water and nutrients in the soil, and then potassium can be taken into account as a fertile material of the soil [2]. Phosphorus plays a fundamental role in plant nutrition, and its concentration and availability determine, to a large extent, soil fertility and site productivity as this element is required in relatively large amounts by plants [2]. Amorin & Batalha [50], Eshaghi Rad & Banj Shafiei. [51], Naqinezhad *et al.* [52] and Adel *et al.* [2] have reported on the role of potassium and phosphorus in the distribution of plant species and delimiting ecological species groups, which confirms the results of this study.

Conclusion

We believe that studies of ecological species groups provide a better understanding of vegetation-environment relationships. This study showed that topographic, climate, and soil variables are useful in separating three ecological species groups. The results supply an ecological basis for rangeland management and developing strategies for rangeland conservation in the study area.

Using the results of this study, we can evaluate the habitat conditions and vegetation quality of Fandoghlu rangelands. Moreover, used high-quality rangeland species such as *Trifolium* to counteract the spread of invasive species such as *Leucanthemum vulgare*.

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Original researcher/Discussion author (34%); Ghorbani A. (Second author), Introduction author/Methodologist/Original/Statistical analyst/Discussion author (33%); Moameri M. (Third author), Methodologist/ Assistant researcher/ Statistical analyst (33%)

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