

# Conservation possibilities of *Isophya costata* (Orthoptera: Tettigoniidae: Phaneropterinae) based on frequency, population size, and habitats

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## Abstract

*Isophya costata* Brunner von Wattenwyl, 1878, commonly called the Keeled Plump Bush-cricket, is an endemic Natura 2000 species in the Carpathian Basin and is included in the IUCN Red List of Threatened species. We used extensive data collection from Hungary retrieved between 2004 and 2019 from 700 sampling sites spread over an area of 12,700 km<sup>2</sup> to examine the occurrence of the species in different regions in grasslands of similar structure but different origin, naturalness, and character. The results confirmed that *I. costata* currently occurs with the highest number of populations and highest density in regularly mowed, mesophilic hayfields rich in dicotyledonous plants (Arrhenatheretalia). The species also appears in smaller numbers in grasslands adjacent to hayfields, such as wetland meadows (*Molinion coeruleae*), marsh meadows (*Deschampsion caespitosae*, *Alopecurenion pratensis*), and edge habitats dominated by herbaceous plants. However, the results show that the extension of these habitats has a negatively significant correlation with both the occurrence of the species and its density. *Isophya costata* occurs in steppe meadows much less frequently than in mesophilic hayfields. The species is endemic to the Pannonian Steppe, and the key to their conservation is by maintaining stocks of hayfields in the species' area of distribution. According to this study, overseeding of mowed grasslands leads to the decline of the species. To preserve *I. costata*, it is necessary to eliminate trampling in its areas of occurrence (prohibition of grazing) and encourage late-season mowing adapted to the phenology of the species (not as early as mid-July) or, if this is not feasible, mosaic-type treatment leaving unmown patches (e.g., 1/3 of the plot).

## Keywords

Carpathian Basin, endemic species, grassland character, hayfields, mowing, overseeding

## Introduction

*Isophya costata* Brunner von Wattenwyl, 1878 is an endemic, postglacial-steppe relict species (Varga 1995) found in the Carpathian Basin. It is a highly protected species of community importance (Natura 2000) in Hungary, and as such, it is included in the IUCN Red List of Threatened species (Chobanov et al. 2016). The species has long been the focus of research because of its hab-

itat loss due to cultivation, abandonment of mowing, etc. (Nagy 1974) that has led to the Hungarian Red Book classifying it as an endangered species (Rakonczay 1989). *I. costata* is mostly found in Hungary (Nagy and Rácz 2014). Based on previous studies, *I. costata* may have been a characteristic species of loess meadows and closed-steppe meadows rich in dicotyledonous plants that later adapted to hayfields rich in dicotyledonous species, similar to their previous habitats but formed under anthropogenic influence. These hayfields have become the primary habitat of *I. costata* (Kenyeres et al. 2004, Bauer and Kenyeres 2006, Kenyeres et al. 2009).

Thanks to recent systematic research new occurrence data have been revealed – not only in Hungary, but also in Slovakia (Nuhlíčková et al. 2017), Serbia (Szövényi and Szekeres 2011, Ivković and Horvat 2020), Austria (Zuna-Kratky et al. 2017), and Romania (Iorgu et al. 2008).

In the present study, the data collection covers about a quarter of the territory of Hungary and took place between 2004 and 2019. We sought to answer the following questions: (1) What regional differences can be detected in the frequency of the occurrence of *I. costata* among the regions within the study area? (2) Are there any differences in terms of the frequency of occurrence of the species among its potential habitats with a similar structure but different species composition, origin, and landscape history? (3) Can the effects of the condition and use of the studied grasslands (regularly mowed, overseeded, shrubbing, humid, semi-dry, dry, etc.) on the occurrence of the species be revealed? (4) Does the density of the species correlate with the local surface cover of the studied habitat types?

## Methods

Between 2004 and 2019, we examined the presence/absence, density, and habitat requirements of *I. costata* (Figs 1, 2) at 700 sites (12,700 km<sup>2</sup>) in northwestern Hungary. The study area was divided into 12 regions (Fig. 3) according to landscape and biogeography. The CORINE Landcover habitat structure of the regions and the distribution of the studied grasslands among habitat types are shown in Fig. 4 and Table 1.





Fig. 1. Male (left) and female (right) specimens of *Isophya costata*.

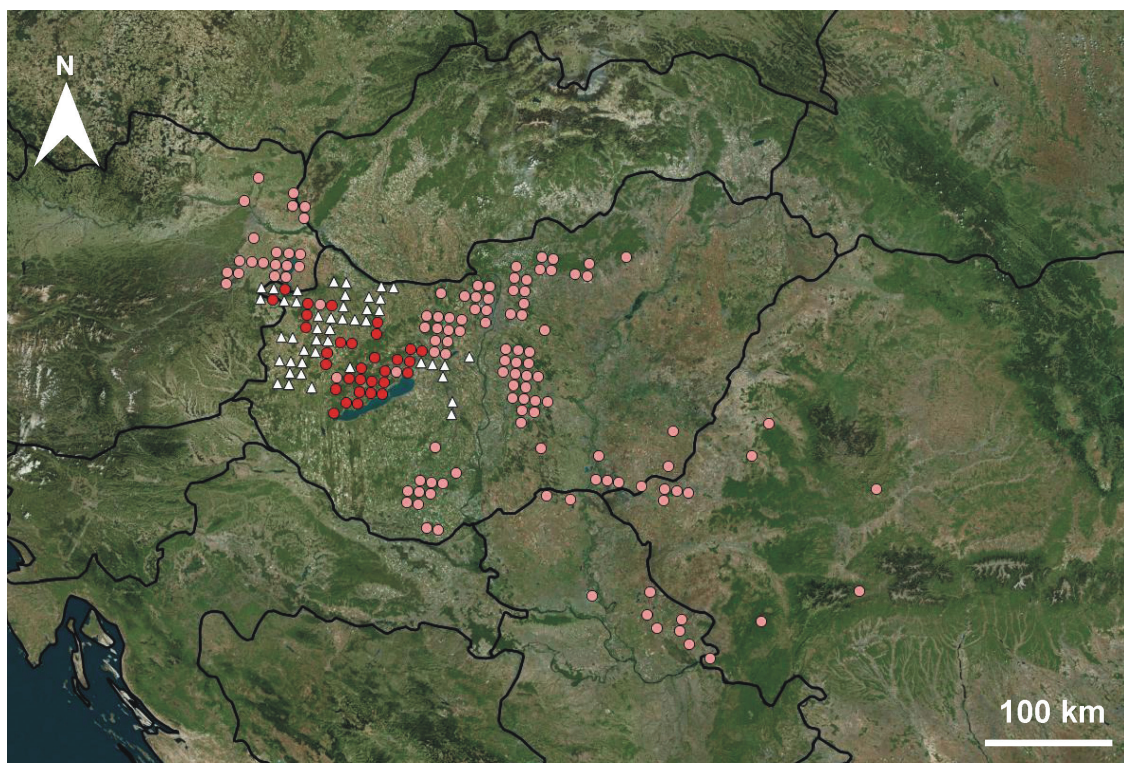
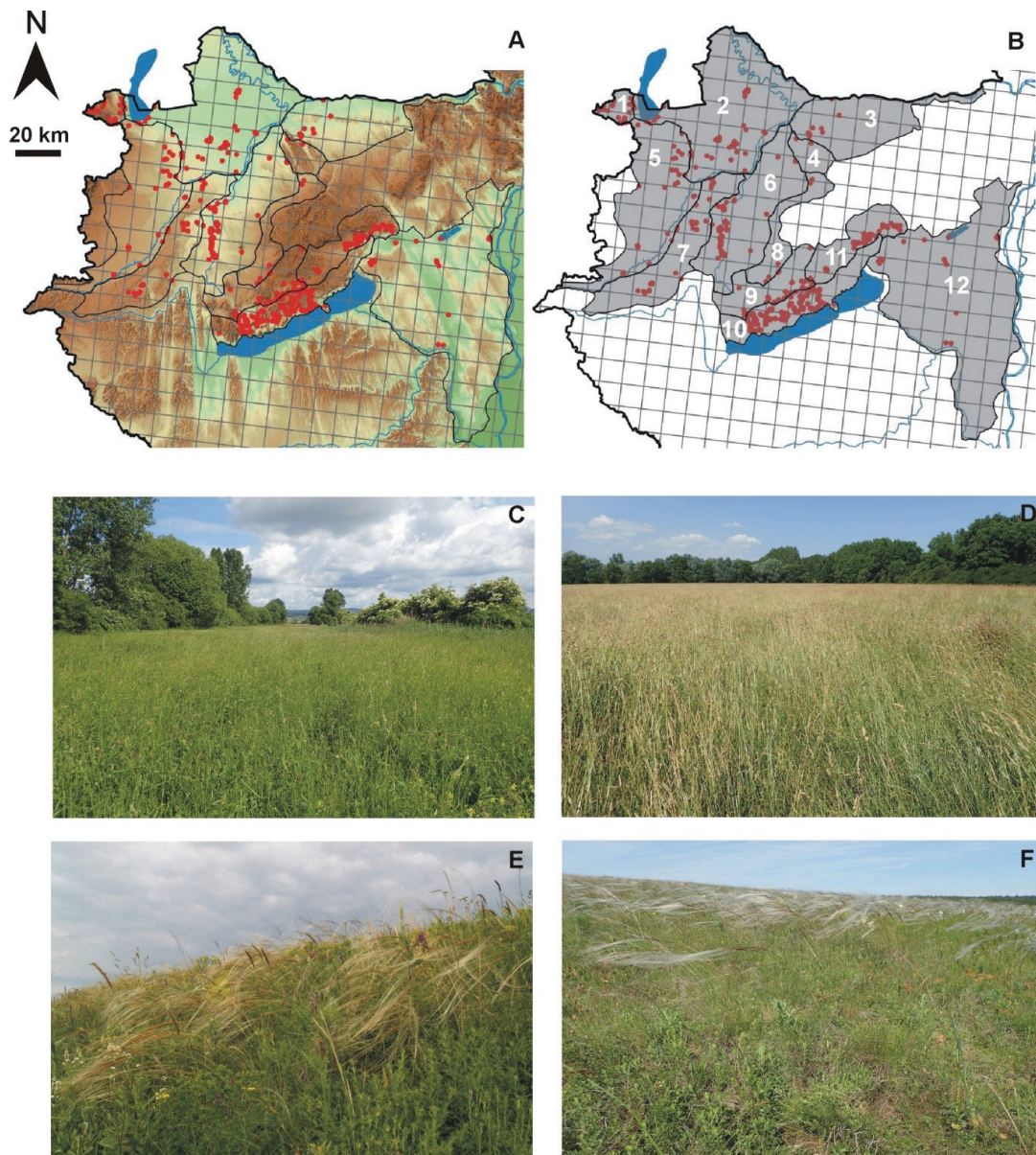


Fig. 2. Known distribution of *Isophya costata* in the Carpathian Basin at the level of ETRS quadrates (circle: presence data of the species; red circle: presence data of this study; triangle: absence data of this study).





**Fig. 3.** Sub-areas of the study area (A, B.) (1: Alpokalja, 2: Győr Basin, 3: Komárom-Esztergom Plain, 4: Pannónhalma Region, 5: Sopron-Vas Plain, 6: Marcal Basin, 7: Kemeneshát, 8: Bakonyalja, 9: Southern Bakony, 10: Balaton Uplands, 11: Eastern Bakony, 12: Mezőföld), sampling sites (red points), and the most typical studied habitat-types: C. Typical hayfield rich in dicotyledonous plant species; D. Overseeded hayfield poor in dicotyledonous plant species; E. Loess steppe; F. Slope steppe.

**Table 1.** Extent (mean $\pm$ SE) and habitat types of the grasslands containing the study quadrates (N=700).

Habitat type	Range in hectare (mean $\pm$ SE)	N
Red beds	3.64 $\pm$ 0.64	3
Rich fens	14.32 $\pm$ 3.52	12
Mesotrophic wet meadows	6.74 $\pm$ 0.67	107
Hayfields	6.40 $\pm$ 0.97	190
Overseeded hayfields	13.20 $\pm$ 3.41	88
Calcareous rocky steppes	4.03 $\pm$ 0.45	116
Slope steppes on stony soils	3.59 $\pm$ 0.62	39
Closed forest steppe meadows	2.29 $\pm$ 0.47	32
Closed steppes on loess	0.68 $\pm$ 0.27	12
Uncharacteristic mesic grasslands	2.86 $\pm$ 0.42	23
Uncharacteristic dry and semi-dry grasslands	5.20 $\pm$ 0.86	78

Sweep-netting, direct observation, and acoustic detection were performed at the studied 10 $\times$ 10 m quadrates for the duration (30 minutes). All detected individuals were recorded. Based on the number of individuals registered in the study quadrates, we determined the presence/absence data and the density of the species per square meter related to the habitat patch concerned. During sampling, we recorded the type of the habitat according to the Hungarian General National Habitat Classification System (Á-NÉR): red bed, rich fen, mesotrophic wet meadow, hayfield/overseeded hayfield, calcareous rocky steppe, slope steppe on stony soils, closed forest steppe meadow, closed steppe on loess, uncharacteristic mesic, and dry and semi-dry grassland. We also recorded the naturalness (natural, pseudo-natural: anthropogenic hayfields with good naturalness, shrubby, disturbed, and overseeded) and microclimatic conditions (humid: directly affected by water and



Fig. 4. Relative frequency of CORINE habitat types in the studied sub-areas.

characterized by water supply for at least a part of the year; mesophilic: closed, semi-dry grasslands; dry: xerophilous grasslands with open soil surfaces).

We determined the size of the habitat type corresponding to the quadrates studied based on data collected in the field by handheld computer (Trimble Juno3B) with the use of QGIS 2.16 software (QGIS Development Team 2016).

A basic database containing the records of all sites ( $N = 700$ ) was used for analyses, but a database containing the relative frequency (rel. freq.) of the recorded variables at the ETRS quadratic level was also generated ( $N = 78$ ). Furthermore, we determined the sub-areal (12 regions) indicators for the presence/absence of the species ( $N_s$ : number of sampling sites with presence/absence of *I. costata*, and  $R_f$ : relative frequency of presence of *I. costata*). The relationships between the presence/absence of the species and the presence/absence of the habitat types and habitat characteristics studied were examined by Generalized Linear Models (GLM) with binomial distribution. Division of the habitats with the presence of *I. costata* among habitat characteristics recorded (habitat types, naturalness, structure, and microclimate) were described by bar charts. The paired t-test was used to evaluate statistical differences among the variables. In order to analyse the relationship between the density of *I. costata* and environmental data at the European Terrestrial Reference System (ETRS) quadrate scale (in which the dependent variable was the mean density of *I. costata*; the habitat variables were mean of the patch size; and the relative frequencies (rel. freq.) of the recorded habitat types were categorized according to in the Habitat Classification System (Á-NÉR), to their naturalness and to their microclimatic conditions; see above), single tests of GLM (Poisson distribution) were used. Overdispersion was not detected in the variables. Statistical procedures were performed using PAST 2.16 (Hammer et al. 2001) and CANOCO 4.5 (Braak and Smilauer 2002) software packages.

## Results

*Regional differences.*—Of the 700 study sites, the presence of the species was detected at 280 locations.

The frequency of the species showed significant regional differences (Fig. 5). *I. costata* proved to be particularly frequent (rel. freq. of positive cases: 0.63) in the Balaton Uplands sub-area (267 study sites), presumably because of the richness of potential habitats in the region. Among the studied quadrates, the proportion of hayfield habitat type was high (rel. freq.: 0.42), but slope steppes on stony soils (rel. freq.: 0.15), rich fens (rel. freq.: 0.15), and uncharacteristic dry and semi-dry grasslands (rel. freq.: 0.15) habitat types were also significantly present. Of the grasslands studied, the proportion of pseudo-natural (rel. freq.: 0.55) and mesophilic (rel. freq.: 0.59) patches was very high.

Roughly a third of the 112 study sites showing the presence of the species were in the Eastern Bakony region (rel. freq.: 0.29). In this region, most of the grasslands studied were calcareous rocky steppes habitat type (rel. freq.: 0.80) with a significant share of uncharacteristic dry and semi-dry grassland habitat types (rel. freq.: 0.15). Most sites included in the study were edaphic dry grasslands (rel. freq.: 0.76) with a dry microclimatic character (rel. freq.: 0.98).

Also, we found the presence of the species in about a third of the 72 study sites in the Marcal Basin region (rel. freq.: 0.32). The majority of the examined patches (rel. freq.: 0.51) belonged to the rich fens habitat type, with a significant number of hayfields and overseeded hayfields (rel. freq.: 0.32 and 0.10). In terms of character, the grasslands studied were pseudo-natural (rel. freq.: 0.81), humid (rel. freq.: 0.58), or semi-dry (rel. freq.: 0.32).

Among the regions with less than 10% of the total sample, *I. costata* was found in about a third of the study sites. This



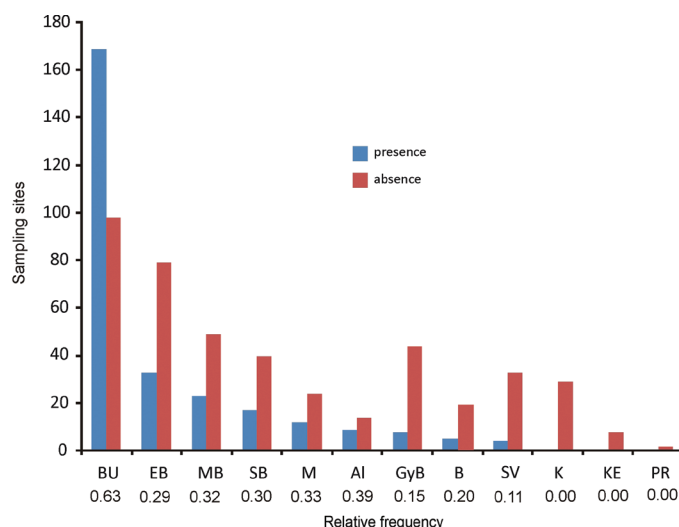


Fig. 5. Presence or absence and relative frequency of presence of *Isophya costata* in the studied regions (BU: Balaton Uplands; EB: Eastern Bakony; MB: Marcal Basin; SB: Southern Bakony; M: Mezőföld; Al: Alpokalja; GyB: Győr Basin; B: Bakonyalja; SV: Sopron-Vas Plain; K: Kemeneshát; KE: Komárom-Esztergom Plain; PR: Pannonhalma Region).

includes Southern Bakony (no. of samples: 57, rel. freq.: 0.30), Mezőföld (no. of samples: 36, rel. freq.: 0.33), and in the Alpokalja region (no. of samples: 23, rel. freq.: 0.39). From a microclimatic point of view, the grasslands under review were dry (rel. freq.: 0.68) and semi-dry (rel. freq.: 0.30) in all three regions. In the sampling quadrates of the Southern Bakony, uncharacteristic dry and semi-dry grasslands (rel. freq.: 0.28), hayfields (rel. freq.: 0.26), rich fens (rel. freq.: 0.28), and slope steppes on stony soil (rel. freq.: 0.26) habitats occurred in a roughly balanced proportion. Among the grasslands examined, pseudo-natural patches (rel. freq.: 0.40) were dominant, but the proportion of disturbed (rel. freq.: 0.30) and shrubby (rel. freq.: 0.25) habitats were also high. Closed forest-steppe meadows (rel. freq.: 0.31) and closed steppes on loess habitat types (rel. freq.: 0.31) were dominant in the studied grasslands of the Mezőföld region, but hayfields (rel. freq.: 0.26) also occurred (rel. freq.: 0.17). Among the grasslands examined, shrubby (rel. freq.: 0.33), pseudo-natural (rel. freq.: 0.22), disturbed (rel. freq.: 0.22), and edaphic patches (rel. freq.: 0.22) were present in a balanced proportion. In the Alpokalja region, in addition to the dominant hayfields (rel. freq.: 0.26), rich fens (rel. freq.: 0.13), and overseeded hayfields (rel. freq.: 0.13) habitat types also occurred. Of the grasslands studied, pseudo-natural patches (rel. freq.: 0.61) were dominant, but the proportion of overseeded (rel. freq.: 0.13) habitats was also high.

In the other sub-areas, either the species was not detected or the frequency of its occurrence was very low. Of these, the Bakonyalja should be highlighted, where most of the grasslands examined belonged to the pseudo-natural, but rather dry, microclimate occurrences of the hayfield (rel. freq.: 0.80) habitat type. However, the number of samples (corresponding to the occurrence of potential *I. costata* habitats) was low in this region. The other sub-areas can be divided into two groups. The examined patches of the Győr Basin (N = 52), Kemeneshát (N = 29) and Sopron-Vas Plain (N = 37) predominantly belonged to overseeded hayfield habitat type (rel. freq.: 0.55–0.68), the character of which was determined by overseeding (rel. freq.: 0.55–0.70) and dry stock climate (rel. freq.: 0.59–0.83). The dominant habitat types in the Komárom-Eszter-

gom Plain (N = 8) and Pannonhalma Hills (N = 2) regions can be classified as diverse: rich fens (rel. freq.: 0.38–0.50), overseeded hayfields (rel. freq.: 0.00–0.38), uncharacteristic dry and semi-dry grasslands (rel. freq.: 0.00–0.25), and closed steppes on loess (rel. freq.: 0.00–0.50). In terms of character, both humid (rel. freq.: 0.38–0.50) and dry (rel. freq.: 0.50–0.63) grasslands were determined to be in high proportion in these regions, and also in terms of naturalness, several types occurred: pseudo-natural (rel. freq.: 0.13–0.50), shrubbing (rel. freq.: 0.00–0.50), disturbed (rel. freq.: 0.00–0.50) and overseeded (rel. freq.: 0.00–0.70).

**Habitat requirements.**—Based on the data as a whole, more than half (51.1%) of the sampled quadrates with the presence of the species were hayfields rich in dicotyledonous plant species (Fig. 6). Additional occurrences were distributed among nine different habitat types, of which the proportion of marsh meadows (12.9%) and calcareous rocky steppes (12.5%) were significant.

In terms of habitat character (Fig. 6), most occurrences were found in regularly mowed hayfields (64.6%) showing good naturalness or edaphic steppes (14.6%). The proportion of disturbed or shrubby grasslands among positive samples was 12.1% and 7.14%, respectively, and the presence of *I. costata* was rarely observed on overseeded hayfields (1.4%). In terms of microclimate, 66.4% of the grasslands where the species occurred were semi-arid, 27.9% were dry, and only 5.7% were humid.

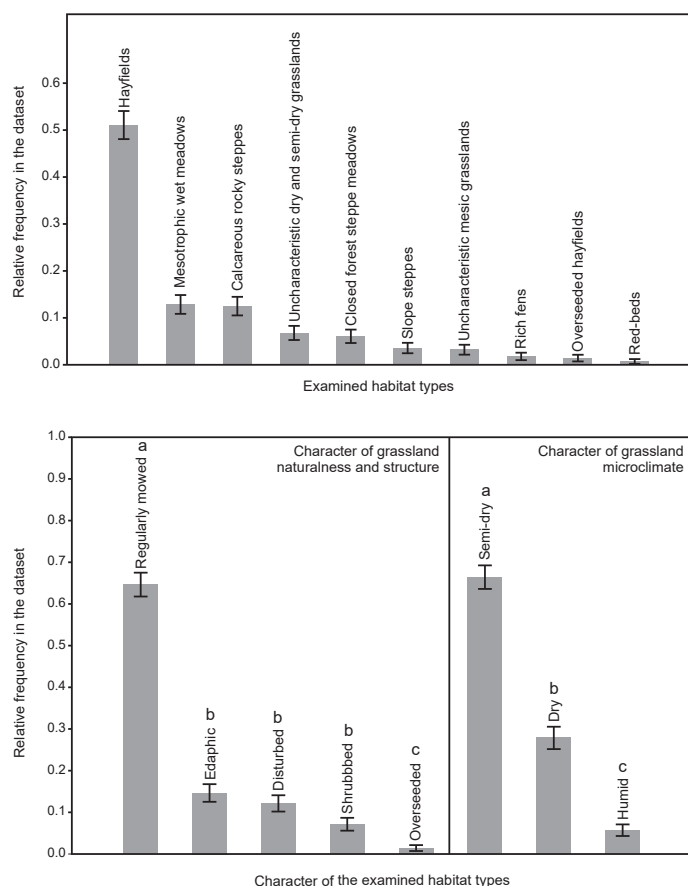
GLM (Table 2) of the data on all study quadrates showed a significant positive correlation between the presence of *I. costata* and the presence of hayfields rich in dicotyledonous plant species. Significant negative relationships were seen between the occurrence of *I. costata* and the presence of overseeded hayfields, mesotrophic wet meadows, and uncharacteristic dry and semi-dry grasslands.

**Density.**—The density of *I. costata* in the study quadrates was 0.24 individuals/m<sup>2</sup> (mean  $\pm$  0.11). There was no significant correlation between species density and size of the habitat patch ( $p=0.609$ ,  $R^2=0.00037$ ).

GLM showed (a) a significant positive correlation ( $p=0.004$ ,  $R^2=0.301$ ) between the mean density of *I. costata* at the ETRS quadrate level and the frequency of hayfields rich in dicotyledonous plant species (Fig. 7) and (b) a significant negative correlation ( $p<0.001$ ,  $R^2=0.462$ ) between the mean density of *I. costata* at the ETRS quadrate level and the frequency of grasslands characterized by humid microclimate (Fig. 7).

## Discussion

Our results show that, although the Keeled Plump Bush-cricket can be found in several areas on steppe meadows, loess grasslands, and other grassland types (weedy humid grasslands, marsh meadows, etc.), their highest density populations live on regularly mowed hayfields rich in dicotyledonous plant species, confirming our earlier statement (Bauer and Kenyeres 2006). According to this study, the species rarely occurs in overseeded hayfields or, if it does, only in very small numbers. This may be related to the fact that overseeding is usually done with grass species, leading to the decline of dicotyledonous plant species cover (Blackmore and Goulson 2014). Also, the soil surface is regularly damaged by overseeding (e.g., harrowing, ploughing) (Cardarelli et al. 2020), which is fatal in terms of the reproduction of *I. costata*, which lays its eggs in the upper-most layers of the soil (Nagy and Rácz 2014). Intensive mowing of the whole plot is also pervasive, which can lead to a further reduction of the cover of dicotyledonous plant



**Fig. 6.** Bar charts ( $\pm$ SE) of the studied habitat types with a presence of *Isophya costata* and bar charts ( $\pm$ SE) of the studied habitats (characterized by different naturalness, structure, and microclimate) with a presence of *Isophya costata*.

species to a level that is no longer able to meet the dicotyledonous nutritional requirements of *I. costata* (Orci et al. 2007); in this situation, the grasslands become unsuitable for maintaining the species' populations (Kenyeres et al. 2004).

The presence of *I. costata* in hayfields rich in dicotyledonous plant species and the absence of the species in overseeded mowed grasslands was also reflected in the sub-areas of the study. In the Balaton Uplands sub-area, which showed the highest frequency of the species, the prevalence of large hayfields rich in dicotyledonous plant species was substantial. The Balaton Uplands sub-area is also characterized by the occurrence of steppe grasslands, which is a priori favourable for the species (Varga 1995). In addition to the above, the extent of potential habitats for *I. costata* in the Balaton Uplands has increased due to the impact of humans, such as through deforestation and the establishment of grasslands maintained by mowing. Typically, extensive mowing creates large anthropogenic hayfields rich in dicotyledonous plant species with a physiognomy very similar to steppes. However, according to our results, *I. costata* occurs with a much lower frequency in steppe grasslands, and in sub-areas dominated by that habitat type (e.g., Eastern Bakony), than in hayfields. These grasslands have a similar structure to hayfields but are less advantageous to *I. costata* in terms of protection from both predators (Kruess and Tschamtkke 2002, Gardiner and Haines 2008) and weather challenges, as these grasslands have a different structure and shorter grass (Kenyeres et al. 2018).

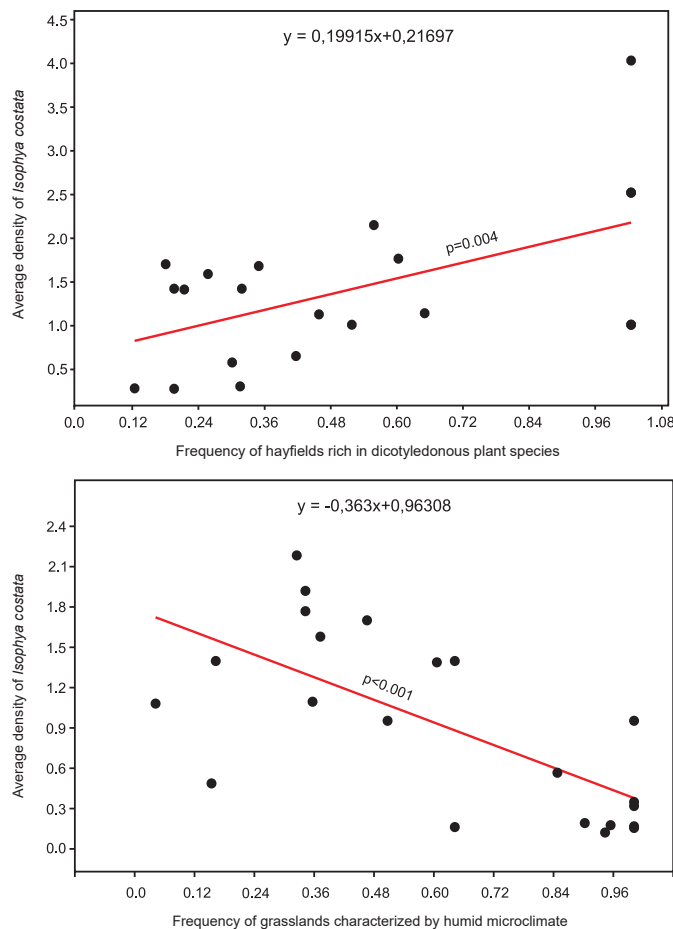
**Table 2.** Results of GLM with binomial distribution of presence/absence data of *Isophya costata* and the studied habitat types, with indication of p-, R-values and estimates (grassland types examined using more than five quadrates and with a mean larger than 1 hectare were used).

Presence of	<i>Isophya costata</i>		
	p	R	est.
Rich fens	n.s.	-0.053	-57.81
Mesotrophic wet meadows	0.043	-0.471	-251.44
Hayfields	<0.001	2.358	-270.28
Overseeded hayfields	<0.001	-2.775	-184.61
Calcareous rocky steppes	n.s.	-0.464	-241.94
Slope steppes on stony soils	n.s.	-0.623	-97.71
Closed forest steppe meadows	n.s.	0.172	-75.84
Uncharacteristic mesic grasslands	n.s.	-3.319	-75.69
Uncharacteristic dry and semi-dry grasslands	<0.001	-1.331	-170.63

The other sub-areas characterized by typical, but fewer, occurrences of the species (Southern Bakony, Mezőföld, Bakonyalja, Marcal Basin) have a similar landscape to the Balaton Uplands sub-area. *I. costata* can also be considered frequent in the Alpok-alja sub-area (Vienna Basin), where loess grasslands and steppe meadows rich in dicotyledonous plant species (assumed to be the original habitat of the species) were present before the landscape was changed by human activities (Bauer and Kenyeres 2006, Kenyeres et al. 2009). Based on this extensive study, *I. costata* is also extremely rare in the intermediate, lowland areas (Győr Basin, Sopron-Vas Plain, Kemeneshát, and Komárom-Esztergom Plain). The potential habitats of *I. costata* in these areas, dominated by arable lands, have drastically changed, and most of the remaining grasslands in these areas are located in depressions and, thus, are dominated not by semi-dry hayfields, but by marsh meadows with a humid microclimate that are less suitable for the ecological requirements of *I. costata*. According to the results of previous (Kenyeres et al. 2017, 2018) and the present study, grasslands with humid climates negatively correlate with the occurrence of the species and its density.

**Conservation possibilities.**—From the perspective of species protection, it is essential to highlight that trampling, fires, and mowing in spring and early summer are a threat to *I. costata* due to its phenological characteristics and low mobility. The species places its eggs 1–2 cm deep in the soil, making them easily destroyed by passing fires. Among grassland management procedures, interventions involving soil damage seriously endanger a population's survival; ploughing and overseeding are lethal, but harrowing, with only a few centimeters of soil damage, can also significantly endanger the success of the species.

In areas where *I. costata* occurs, the prohibition of grazing and tourism that cause trampling is justified; in hayfields, the application of late mowing (beginning of July at the earliest) or, if this is not feasible, mosaic-type treatment leaving unmown patches (e.g., 1/3 of the plot) is recommended (Kenyeres et al. 2017, 2018). The latter is justified by the fact that from the second half of May, which is the typical mowing period of these meadows, the species is in its adult state when reproduction and egg-laying take place. Also, in the distribution area of *I. costata*, mowed grasslands are often located between intensively used arable lands, and it is important for their conservation to reduce the use of pesticides in this area (Ivković and Horvat 2020).



**Fig. 7.** Results of GLM testing of the effect of the presence of hayfields rich in dicotyledonous plant species and of the presence of grasslands characterized by a humid microclimate on the density of *Isophya costata* (based on data in ETRS quadrat, N=78).

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