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Biological Integrity of Azorean Native Forests Is Better Measured in Cold Season

Noelline Tsafack [1,2,3,*], Sébastien Lhoumeau [2], Alejandra Ros-Prieto [2], Loic Navarro [2], Timea Kocsis [2], Sónia Manso [3], Telma Figueiredo [3], Maria Teresa Ferreira [3,*] and Paulo A. V. Borges [2,4,5,*]

1 Institut Méditerranéen de Biodiversité et d'Ecologie, Aix Marseille Université, CNRS, IRD, Avignon Université, 84911 Avignon, France
2 cE3c-Centre for Ecology, Evolution and Environmental Changes, Azorean Biodiversity Group, CHANGE—Global Change and Sustainability Institute, School of Agricultural and Environmental Science, University of the Azores, Rua Capitão João d'Ávila, Pico da Urze, 9700-042 Angra do Heroísmo, Portugal; seb.lhoumeau@gmail.com (S.L.); alejandrarosprieto@gmail.com (A.R.-P.); loicnavarro@orange.fr (L.N.); tkocsis.kt@gmail.com (T.K.)
3 Regional Secretariat of Environment and Climate Change, Project LIFE-BEETLES (LIFE 18 NAT/PT/000864), Rua do Galo n118, 9700-040 Angra do Heroísmo, Portugal; sonia.mf.manso@azores.gov.pt (S.M.); telma.sr.figueiredo@gmail.com (T.F.)
4 IUCN SSC Mid-Atlantic Islands Invertebrate Specialist Group, 9700-042 Angra do Heroísmo, Portugal
5 IUCN SSC Species Monitoring Specialist Group, 9700-042 Angra do Heroísmo, Portugal
* Correspondence: noellinetsafack@gmail.com or noelline.tsafack@gmail.com or noelline.tsafack-menessong@univ-avignon.fr (N.T.); maria.tm.ferreira@azores.gov.pt (M.T.F.); paulo.av.borges@uac.pt (P.A.V.B.)

Abstract: The Azorean archipelago, recognized as one of the world's biodiversity hotspots, is home to a diverse and unique community of arthropod species, highlighting a notable degree of endemism. However, the native forests that support these species are facing significant degradation due to habitat loss and fragmentation. In this study, we aimed to determine the ideal season for measuring the biological integrity of forest sites using a biological integrity index (IBI) based on arthropod communities captured with Sea, Land, and Air Malaise (SLAM) traps. Drawing on more than thirty years of research experience in the Azorean forests, we selected twelve reference sites, six representing preserved native forest and six representing disturbed native forest, and compared how IBI values vary between seasons. IBI values exhibited consistent variations between seasons in disturbed sites, indicating that measuring the biological integrity in these areas can be conducted at any time of the year without a specific seasonal preference. In contrast, significant differences were observed in pristine forest sites, with the winter season and the combination of winter and spring data (cold semester) showing notably higher values compared to other seasons and semesters. This finding suggests that measuring the biological integrity of preserved sites is best optimized in the cold seasons, while the detection of exotic species impact is most effective in summer and autumn. Consequently, if resources are limited, monitoring efforts should be concentrated in the winter and summer seasons to obtain the maximum and minimum values of IBI, respectively. Additionally, our study suggests that the summer season is the optimal time to detect potentially invasive exotic species.

Keywords: integrity biological index; Azorean archipelago; native forests; endemic species; exotic species; disturbance

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1. Introduction

The Azorean archipelago, recognized as one of the world's biodiversity hotspots, is home to a diverse and unique community of species. These species exhibit a high level of endemicity [1–3]. This biodiversity is mainly explained by the diverse environmental features as well as smooth climate (high humidity and average temperatures) that characterize the Azorean islands. Vegetation and animal species communities are highly diverse. In the Azores, there are two major forest types: native and exotic forests. Native forests, which support numerous endemic tree species [4,5] such as the Azorean cedar *Juniperus brevifolia* (Hochst. ex Seub.) Antoine [6], the laurel *Laurus azorica* (Seub) Franco [7], *Ilex azorica* Gand., and *Erica azorica* Hochst. ex Seub, and endemic shrub species such as *Vaccinium cylindraceum* Sm. and *Myrsine retusa* Aiton. Bryophytes are present on a variety of substrates and are also major components of the Azorean native forests [8–10]. And exotic forests that are mostly represented by plantations of *Cryptomeria japonica* D. Don and *Eucalyptus* spp. used for forestry, and patches of the invasive *Pittosporum undulatum* Vent.

Within animal species communities, arthropods are the most diverse species group of the Azores pristine forests and the most investigated [8], which places Azorean arthropod communities among the most documented worldwide. Arthropods have been used as biological models in numerous studies in the Azores with a variety of aims. For instance, they have been used to investigate species–climate change scenarios [11–13], to investigate the biodiversity crisis [14], to reveal the influence of the abiotic environment [15], to understand the vertical distribution of spider species communities in native forests [16], to investigate species invasiveness, and to test macroecological patterns [17–19].

Despite the interest in Azorean native forests, which is also illustrated by the studies cited above, they are experiencing significant degradation of their biological integrity as a result of habitat loss and fragmentation. A request to guide the ongoing conservation programs concerns the development of efficient tools to measure the extent of degradation. In attempts to fulfill those requests, multi-metric arthropod-based indices were developed to assess the capability of native forest fragments in the Azorean islands [20,21] in supporting and maintaining stable species communities. This capability was called biological integrity, or biotic integrity, or bio-integrity by Karr et al. [22]. They defined it as the capability of an ecosystem to support and maintain a balanced, stable, and persistent species community (see also Cardoso et al. [21]). In a previous study [20], we developed a multi-metric index based on arthropods collected with diverse methods to measure native forest biological integrity called the Index of Biological Integrity (IBI). In the present study, we focused on an IBI based on arthropods collected with passive flight-intercept traps (called IBI-SLAM in the remaining text). Because arthropod species communities are sensitive to microclimates induced by seasons [23], we focused on identifying the best season during which arthropods should be monitored in order to obtain the most accurate IBI value.

Due to more than thirty years of research experience in Azorean forests, large arthropod databases are available [1]. From these long-term arthropod monitoring projects [1], we extracted arthropod abundance trapped in preserved and disturbed native forest sites. We calculated IBI values for the four seasons and for cold and warm semesters. Species colonization status was included in IBI calculations to fit the island context. Species were assigned to one of the three colonization statuses according to their distribution in the Azorean Archipelago [8,24]: endemic (species restricted to the Azores), native non-endemic (species that arrived naturally to the Archipelago, but are also present elsewhere), or introduced (species accidentally or deliberately introduced by man).

Therefore, the main aims of this study were: (i) to investigate IBI values across the four seasons and cold and warm semesters and detect which season and semester are suitable to measure IBI; (ii) to investigate the biogeographic colonization status of species in calculating IBI values, categorizing species into endemic, native non-endemic, and introduced. Overall, the study aimed at refining the bio-integrity index using arthropod data and identifying the optimal season for monitoring while considering the colonization status of species in the Azorean Archipelago.

2. Materials and Methods

2.1. Study Areas

The Azores is an oceanic archipelago of nine volcanic islands located on the North Atlantic Ocean between 37° and 40° North latitude and 25–31° West longitude [25]. The archipelago consists of nine islands: Corvo, Flores Faial, Pico, São Jorge, Graciosa, Terceira, São Miguel, and Santa Maria, distributed into three main groups: Occidental (Corvo and Flores); Central (Faial, Pico, São Jorge, Graciosa, and Terceira), and Oriental (São Miguel and Santa Maria). The Azores have a mild climate year-round, with an average annual temperature of 17 °C [6,12] and high levels of humidity, up to 95% in higher-altitude native forests.

The archipelago of the Azores is part of the Mediterranean biodiversity hotspots and, as such, is monitored through several projects, including those of our research team, the Azorean Biodiversity Group, on native and exotic forest arthropods. Under long-term monitoring studies (BALA I, II, III, SLAM projects) of up to 25 years, arthropods have been monitored using different types of techniques (SLAM traps, canopy beating, pitfall traps, handling, etc.). See team publications [26–29] to learn more about the different sampling techniques and arthropod identification. These projects, which have lasted nearly 30 years, have generated a massive amount of data that are made available to the public [1,27] in open sources. These databases were used to build the biotic integrity indices (IBI) [20,21] and are also used in the current study to investigate the best season during which IBI-SLAM values are the most accurate.

The long-term monitoring projects focus on the main forest ecosystems of the archipelago: native and exotic forests. Native forests are evergreen forests dominated by endemic tree species and shrubs [4,5] including *Juniperus brevifolia* (Hochst. ex Seub.) Antoine, *Laurus azorica* (Seub.) Franco, Ilex azorica Gand., and *Erica azorica* Hochst. ex Seub—trees and *Vaccinium cylindraceum* Sm.—shrub. The current dominant native forest occupies less than 5% of the original area, dominated by the *Juniperus–Ilex* forests and *Juniperus* woodlands and restricted to some patches at 500–700 m elevation [5]. These forests are subject to direct and indirect anthropogenic disturbance. Some spots are considered mixed forests because they are progressively invaded by invasive species such as *Pittosporum undulatum* Vent. (Pittosporaceae) and *Hedychium gardnerianum* Sheppard ex Ker—Gawl. A large proportion of land is currently dominated by intensively managed pastures and exotic forest fragments. Exotic forests are represented by plantations of *Cryptomeria japonica* D. Don (Cupressaceae) used for forestry and patches of the invasive *P. undulatum*.

2.2. Selecting Data Sets

We selected data from 12 sites placed on three islands (3 sites in Flores Island, 3 on Pico Island, and 6 on Terceira Island) (original data in [26–29]) (Table 1). Half of the sites were placed in high-elevation, well-preserved native forest fragments, and the other half were in either highly invaded native forests or exotic and mixed forest fragments, which are less preserved compared to native forest, and we named them disturbed sites in the rest of the text. The disturbance is therefore due to the fact that the soil of native forests is covered by the invasive plant *H. gardnerianum* or the site is now partly invaded by other trees like *Pittosporum undulatum* Vent.

Therefore, the selected datasets concern arthropod species communities trapped in native and mixed forests using passive flight-intercept Sea, Land, and Air Malaise (SLAM) trap; 110 × 110 × 110 cm (MegaView Science Co. Ltd., Taichung City, Taiwan) in a ten-year period extending from 2012 to 2022. Samples were collected one time per season around 15th March for the winter sample, 15th June for the spring sample, 15th September for the summer sample, and 15th December for the autumn sample. In the lab, samples were sorted as morphospecies and identified to the species level by the senior co-author (P.A.V.B.). Because some species remained unidentified at the species level, our investigation was based on morphospecies.

These datasets include the total abundance of the following target groups: Diplopoda (Chordeumatida, Julida), Chilopoda (Geophilomorpha, Lithobiomorpha, Scolopendromorpha), Arachnida (Araneae, Opiliones, Pseudoscorpiones), and Insecta (Archaeognatha, Blattaria, Coleoptera, Hemiptera, Microcoryphia, Neuroptera, Psocodea, Thysanoptera, Trichoptera, Hymenoptera-Formicidae). Furthermore, arthropods are classified according to their biogeographic origin: endemic (restricted to the Azores), native non-endemic (species that arrived naturally and have a distribution wider than the archipelago), and introduced (species with an original distribution that did not include the Azores before human settlement in the 15th century), based on the last checklist of Azorean arthropods [24]. Based on the trophic distinction, we also considered functional subgroups, including fungivores, predators, and herbivorous, generalist, and saprophagous species groups, which are necessary for the calculation of the IBI.

Table 1. Study sites, description, and location. Six sites were located in well-preserved forest fragments and 6 in disturbed forest fragments.

Sites Code	Island	Site Quality	Longitude	Latitude
FLO-NFFR-T-06	Flores	Disturbed	−31.2235	39.4074
FLO-NFFR-T-07	Flores	Disturbed	−31.2175	39.4032
FLO-NFMA-T-08	Flores	Preserved	−31.2094	39.4600
PIC-ML-400	Pico	Disturbed	−28.4311	38.5207
PIC-NFCA-T-08	Pico	Preserved	−28.2000	38.4408
PIC-NFMP-T-10	Pico	Disturbed	−28.2759	38.4630
TER-0M	Terceira	Disturbed	−27.3748	38.7666
TER-NFBF-T-01	Terceira	Preserved	−27.2193	38.7618
TER-NFPG-T-33	Terceira	Disturbed	−27.2271	38.7334
TER-NFSB-T164	Terceira	Preserved	−27.3074	38.7355
TER-NFTB-T-15	Terceira	Preserved	−27.2006	38.7364
TER-NFTB-T-18-O	Terceira	Preserved	−27.1980	38.7323

Analyses were conducted on the four seasons and on two semesters: cold semester assembling winter and spring seasons and warm semester assembling summer and autumn seasons. However, the number of seasons or semesters available for each site were not the same (Tables S1 and S2). Statistical methods were selected accordingly.

2.3. IBI Calculation and Data Analysis

The methodology employed involved the use of a Biotic Integrity Index (IBI), namely IBI-SLAM. Such indices were designed to evaluate the ecological health of Azorean native and disturbed forests [20,21]. The output is a score of bio-integrity based on the arthropod assemblage composition.

The groundwork for the design of these indices was laid by a prior study by Cardoso et al. [21] that investigated the biological integrity of Azorean native forests for epigean (ground-dwelling) arthropod communities. This earlier IBI (referred to as IBI-Pitfall) was founded on a multi-metric framework, incorporating seven taxonomical and ecological parameters chosen from a selection of candidate parameters based on their correlation with environmental disturbance (16 candidate parameters after a first screening). These parameters encompassed indicators such as biogeographic groups, including endemic, native non-endemic, introduced abundance, and species richness; functional groups such as predator, herbivore, fungivore, saprophagous, and generalist abundance and species richness; and more. Notably, these selected parameters demonstrated not only strong positive or negative ties to disturbance but also desirable scalability properties and relatively low correlation among themselves, rendering the IBI reliable and less susceptible to variations in sampling effort.

Following a similar methodology, we, Tsafack et al., in a previous study [20], entailed the creation of IBI-SLAM and IBI-Canopy, referring to IBI for intermediate strata species communities and canopy communities, respectively. To gather the requisite data for these indices, two distinct arthropod collection methods were employed: passive flight interception SLAM traps (referred to as SLAM-species community) and beating techniques designed to collect arthropod species dwelling in the canopy (referred to as Canopy-species community). This comprehensive approach allowed for the assessment of biological integrity across diverse arthropod communities inhabiting distinct forest strata.

Therefore, the IBI-SLAM is a multi-metric index based on arthropods collected with SLAM traps. The index accounts for the percentages of endemic species (%s End) and individuals (%n End); of native species (%s Nat) and individuals (%n Nat); of saprophytic species (%s Sap) and individuals (%n Sap); and introduced individuals (%n Int). Each metric takes one of the three discrete scores: 0, 1, or 2, according to the percentages observed in the species community structure. The IBI value is the sum of the scores assigned to each of the seven abovementioned components, so that the IBI-SLAM score can range from 0 (worst biotic integrity) to 14 (best biotic integrity) [20] (see Table 2).

Table 2. Quantitative values for each metric and scores of IBI-SLAM [20]. The parameters are all percentages.

Parameters	Score 0	Score 1	Score 2
sEnd	<18	18–30	>30
nEnd	<35	35–63	>63
sNat	<38	38–46	>46
nNat	>47	33–47	<33
sSap	>25	20–25	<20
nSap	>33	14–33	<14
nInt	>19	5–19	<5

The mathematical formula is: $IBI = \sum S$ with S the score of each of the seven parameters, for an overall IBI value ranging from 0 to 14.

Using the Wilcoxon test, we compared IBI values between disturbed and preserved forest sites to confirm their difference, which was first based on a priori knowledge (Figure S1). Kruskal–Wallis tests were run to compare seasonal IBI values within sites with the same quality. Then, Wilcoxon tests were used to perform pairwise comparison analyses between seasons and semesters. The Bonferonni correction was applied to adjust p-values. Statistical analyses were run using the R analysis language [30] and the rstatix R package version 0.7.2 [31].

3. Results

Overall, the selected dataset assembles a total of 90,492 individuals belonging to 351 morphospecies. A total of 1011 individuals belonging to 51 morphospecies and representing about 1% of the total abundance were removed from the analysis because of uncertainty about their colonization status.

Native non-endemic species dominated overall samples, each season samples, and the two semester samples, and they represented about 56% of the total abundance. On the other hand, exotic species represented 7% and endemic species 36% of the total abundance (Table 3). About 64% of total abundance was collected in the warm semester (summer and autumn samples) (Table S3). Native non-endemic species also dominated warm and cold semester samples, whereas exotic species were the least abundant group, representing about 2% and 5% of total abundance in cold and warm semesters, respectively (Table 3).

Most species were exotic species in cold or warm semester samples and in each season sample. In contrast, endemic species showed the lowest number of species overall in cold or warm semester samples and in each season sample (Table 3).

After IBI calculation, we found that the average index of bio-integrity (IBI) value was 6.26 in the cold semester and 5.09 in the warm semester, whatever the type of forest. And the average IBI value was 3.38 in disturbed forest sites and 7.79 in preserved sites (Table 4).

In all disturbed sites, no difference in IBI values between seasons was observed (Figures 1 and S2, Table 5), whereas significant differences between seasons were observed in preserved sites (Figure 2, Table S3). Pairwise analysis showed that IBI values measured in winter and spring were significantly higher than IBI values in summer or autumn (Figure 2, Table S3), suggesting a difference between cold and warm semesters that is confirmed by the Wilcoxon test (Figure S3, Table 4 with $p < 0.0001$).

Table 3. Number of individuals and number of morphospecies for endemic (END), native non-endemic (NAT), and non-native introduced (INT) arthropods samples collected in cold (winter and spring) and warm (summer and autumn) semesters. Differences were assessed using a Chi-square test. Chi-square test estimates and significance are indicated.

		END	**NAT**	**INT**	χ^2	p
		Number of individuals				
Cold semester	Winter	3233	4131	463	2802.3	<0.001
	Spring	10,863	12,179	1131	9039.1	<0.001
Warm semester	Summer	13,737	25,846	2608	19,210	<0.001
	Autumn	5155	8250	1885	3975.5	<0.001
	Total	32,988	50,406	6087	33,429	<0.001
		Number of morpho (species)				
Cold semester	Winter	32	43	58	7.6842	0.021
	Spring	50	59	76	5.6541	0.059
Warm semester	Summer	48	72	85	10.312	<0.01
	Autumn	41	62	82	13.632	<0.01
	Total	171	236	301	35.805	<0.001

Table 4. Mean and range values of bio-integrity index values in disturbed and preserved reference sites for the two semesters. U and p are, respectively, the statistic and probability values of the Wilcoxon test comparing the two semesters.

Islands	**Site Quality**	**Cold (Winter/Spring)**		**Warm (Summer/Autumn)**		**U**	p
		Mean ± SD	**Range**	**Mean ± SD**	**Range**		
Flores	Disturbed	4.3 ± 1.03	[3–6]	3.6 ±1.06	[1–4]	5.04	0.283
Flores	Disturbed	4.0 ± 1.41	[3–5]	1.7 ± 0.58	[1–2]	4.00	0.261
Pico	Disturbed	1.13 ± 1.46	[0–4]	0.9 ± 1.36	[0–4]	0.68	0.878
Pico	Disturbed	8.5 ± 0.71	[8–9]	6.7 ± 0.58	[6–7]	4.00	0.261
Terceira	Disturbed	2.2 ± 1.09	[1–5]	1.8 ± 1.23	[1–4]	8.83	0.032
Terceira	Disturbed	5.7 ± 1.64	[4–8]	4.1 ± 1.81	[1–7]	9.82	0.133
Flores	Preserved	8.5 ± 0.71	[8–9]	7.0 ± 1	[6–8]	2.33	0.506
Pico	Preserved	6.5 ± 2.12	[5–8]	8.7 ± 0.58	[8–9]	2.33	0.311
Terceira	Preserved	8.5 ± 0.53	[8–9]	6.3 ± 1.29	[4–8]	14.55	0.013
Terceira	Preserved	9.6 ± 0.53	[9–10]	7.9 ± 0.74	[7–9]	12.93	$p < 0.01$
Terceira	Preserved	8.5 ± 0.53	[8–9]	7.2 ± 0.75	[6–8]	11.52	$p < 0.01$
Terceira	Preserved	8.4 ± 0.79	[8–10]	7.0 ± 1.12	[5–8]	6.53	0.258
All Disturbed sites		3.7 ± 2.5	[0–9]	2.8 ± 2.1	[0–7]	648	0.112
All Preserved sites		8.6 ± 0.9	[5–10]	7.2 ± 1.1	[4–9]	289	$p < 0.001$
Total		6.3 ± 3.0	[0–10]	5.1 ± 2.7	[0–9]	2471.5	$p < 0.001$

Table 5. Pairwise comparison of bio-integrity index values between seasons and semesters in disturbed and preserved sites. The number n of data available for comparison are provided in brackets after season as well as Wilcoxon static U and the adjusted p-values p. No differences were observed in disturbed sites. In preserved sites, IBI values in the cold season were significantly higher than measures in the warm season, but no difference was observed in seasons of the same semester.

Site Quality	**Season 1 (n)**	**Season 2 (n)**	**H**	p
Disturbed	Winter (29)	Spring (35)	385.5	1
Disturbed	Winter (29)	Summer (41)	560	1
Disturbed	Winter (29)	Autumn (35)	521.5	1
Disturbed	Spring (35)	Summer (41)	899	0.9
Disturbed	Spring (35)	Autumn (35)	779.5	0.7
Disturbed	Summer (41)	Autumn (35)	778.5	1
Preserved	Winter (31)	Spring (34)	465.5	1

Table 5. *Cont.*

Site Quality	Season 1 (n)	Season 2 (n)	H	*p*
Preserved	Winter (31)	Summer (44)	1030	0.001
Preserved	Winter (31)	Autumn (42)	1062.5	$p < 0.001$
Preserved	Spring (34)	Summer (44)	1165	$p < 0.001$
Preserved	Spring (34)	Autumn (42)	1185.5	$p < 0.001$
Preserved	Summer (44)	Autumn (42)	1079.5	1
Disturbed	Winter/Spring (37)	Summer/Autumn (44)	980	1
Preserved	Winter/Spring (40)	Summer/Autumn (47)	1591	$p < 0.001$

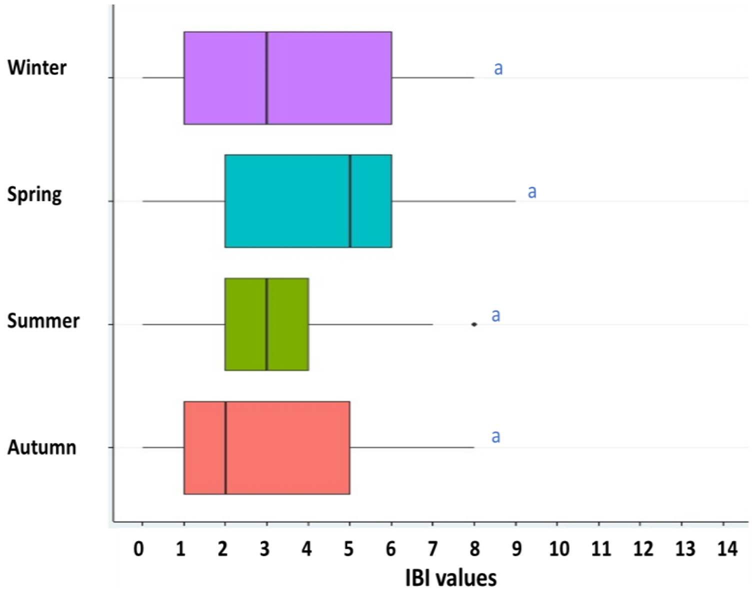

Figure 1. Boxplots of total IBI values of the six sites representing disturbed native forest per season: winter (n = 29), spring (n = 35), summer (n = 41), autumn (n= 35). Same letter indicates non-significant differences according to Kruskal–Wallis test (H = 5.48, p = 0.14).

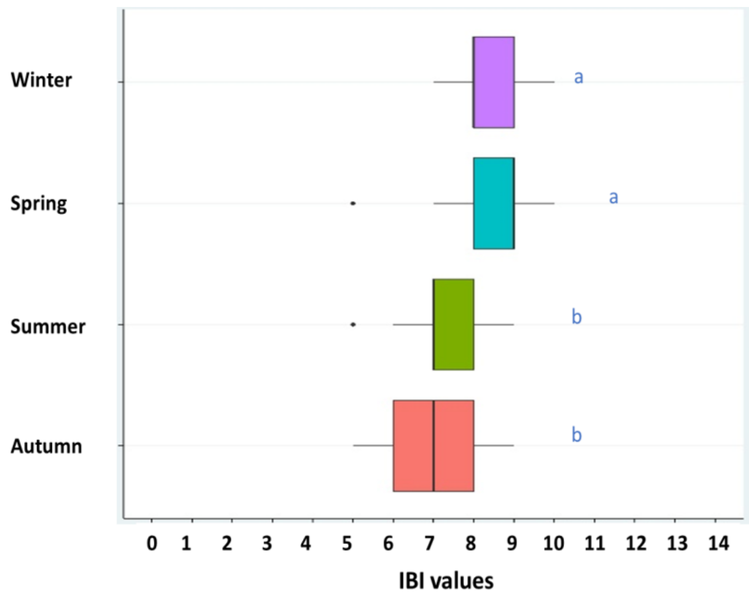

Figure 2. Boxplots of total IBI values of the six sites representing preserved native forest per season: winter (n = 31), spring (n = 34), summer (n = 44), autumn (n = 42). Same letter indicates non-significant differences according to Kruskal–Wallis test (H = 43, $p < 0.001$).

A total of 1011 individuals belonging to 51 morphospecies were removed from the analysis because of uncertainty regarding their biogeographic colonization status.

4. Discussion

The data set used in this study is exceptionally large and is likely one of the most complete datasets on arthropods on islands. The dataset assembles arthropod species communities collected at every season in twelve forest sites over a period of ten years (see also Lhoumeau and Borges [32] for a recent review of ecological trends).

Our primary objective was to optimize the use of the bio-integrity index (IBI) to measure how well a forest site could support stable species communities using arthropods as a biological model. We investigated to detect the best season in which IBI was optimal. First, our results confirmed that IBI decreases with disturbance. This has been shown in the past with a dwelling arthropod-based IBI in Azorean forests [21], with a bird-based IBI in south-eastern Brazilian Atlantic forest patches [33], and also using phytoplankton to assess the quality of lakes [34,35]. For all these studies, disturbance was characterized by a number of environmental factors that were expected to impact species. In our study, disturbance was characterized by differences between native, well-preserved forest fragments and less-preserved exotic forest fragments. The main difference that we observed on sites was the fact that the soil of some native forest fragments started to be covered by the invasive plant *H. gardnerianum*, or that some sites are now partly invaded by other trees like *P. undulatum*.

Furthermore, we found that the variation of seasonal or semi-annual IBI values also depends on the disturbance at the forest site. The more the site was disturbed, the less important was the time of the year in which IBI was measured. In fact, in disturbed sites, IBI values were consistent, without significant differences between seasons or even between semesters. Despite the natural yearly dynamic of arthropod species communities due to abiotic factors (climate features), species composition did not change enough to influence IBI in disturbed forests. Studies that have investigated the temporal variation of biotic integrity of sites are rare. To our knowledge, this is the first study that has attempted to understand how biotic integrity of a system can change across time.

However, for more pristine forests, we found that IBI values were significantly higher in the cold semester, including winter and spring seasons, than in the warm semester (summer and autumn). This result suggests that Azorean high-elevation pristine forest sites present better biological integrity in cold seasons than in warm seasons. One main difference between cold and warm semester samples is the number of exotic introduced species individuals, which is about three times higher in warm seasons. Exotic introduced species are usually characterized by a high level of flexibility to face environmental changes. The increase of introduced species during summer in native forest fragments explains the decline in biological integrity of the more pristine forest sites. Unfortunately, studies reported an increasing trend of introduced species in native forests with clear filters in the advantage of small-sized dominant species [32].

Additionally, our study reveals that summer season is the optimum time to detect potentially invasive exotic species in the Azores. The dynamics of arthropod invasive species in native forests can vary depending on several factors, including the characteristics of invasive species communities, the ecological conditions of the forest, and the interactions with native species. Moreover, the success of invasive species establishment can depend on factors such as the availability of suitable habitat, availability of resources, and absence of natural enemies or competitors. However, the assessment of the colonization status of arthropod species based on expert knowledge is not free of errors. A recent study conducted in the Canary Islands, based on DNA analysis, has shown that some species previously classified as indigenous to the region belong to the non-indigenous group [36]. If this applies to our data, this implies that the higher IBI values obtained in the colder seasons may be overestimated.

The diversity of exotic species in the Azores is increasing [14], but their impacts are still unknown. However, Cardoso et al. [37] suggested impacts of invasive species on the extinction of Azorean endemic spiders. In some cases, invasive species may become naturalized and persist in the ecosystem, leading to long-term changes in community composition and dynamics. In a recent study, Pozsgai et al. [15] showed that in Azorean forests, the proportion of negative links as indicators of competition did not increase with the increase of introduced species in the habitats, implying that introduced species were occupying empty niches when they colonized the islands. But this should be taken with caution because we observed in a previous study [20] that an arthropod species community can be stratified with particularities along forest vertical strata, and introduced exotic species seem to be less observed at an intermediate stratum than at the ground or in the canopy. The most frequent and abundant exotic species at our sites include the known invasive species *Dysdera crocata* C.L. Koch, 1838 (Araneae, Dysderidae) and *Ommatoiulus moreleti* (Lucas, 1860) (Diplopoda, Julida), which are common on the ground of the forest, but also many pasture-dwelling exotic ballooning spider species can be found in the canopy of the forest (see Costa et al. [16]).

It is important to note that the dynamics of potentially invasive, introduced arthropod species can be complex and context-dependent. Different invasive species can have different impacts, and the response of native ecosystems can vary based on their resilience and ability to adapt to new ecological pressures. Effective long-term monitoring programs of island forests are critical [1]. Additionally, early detection and rapid response measures are crucial for managing invasive arthropod species in native forests and minimizing their negative impacts on biodiversity and ecosystem functioning.

5. Conclusions

The measure of biological integrity in disturbed forest sites can be conducted at any time of the year without a specific seasonal preference, contrary to pristine forest sites, where cold seasons (winter and spring) should be preferentially monitored. Additionally, our study suggests that the summer season is the optimum time to detect potentially invasive exotic species. The results of this study can serve as inspiration for other systems, but a precaution should be taken to adapt the IBI to the given system, for which standardized endemic, native non-endemic, and introduced exotic species richness and abundance should be obtained.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/d15121189/s1, Figure S1: Boxplots of total IBI values all preserved sites and disturbed sites. IBI values are significantly higher in preserved sites with low variance compared to disturbed sites. Letters indicate significant difference according to Wilcoxon test (U = 3279.5, $p < 0.0001$); Figure S2: Boxplots of total IBI values of the six sites representing disturbed native forest per semester Cold semester—Winter/Spring ($n = 37$), Warm semester—Summer/Autumn ($n = 44$), Same letter indicates non-significant differences according to Wilcoxon test (U = 980, $p = 1$); Figure S3: Boxplots of total IBI values of the six sites representing preserved native forest per semester Cold semester—Winter/Spring ($n = 40$), Warm semester—Summer/Autumn ($n = 47$), Same letter indicates non-significant differences according to Wilcoxon test (U = 1591, $p < 0.0001$); Table S1: Number of seasons data available in disturbed and preserved reference sites; Table S2: Number of semesters data available in disturbed and preserved reference sites. Seasons winter and spring are merged into Cold semester. Seasons summer and autumn are merged into warm semester; Table S3: Mean and range values of biointegrity index in disturbed and preserved reference sites for the four seasons. H and P are respectively the statistic and Probability values of the Kruskal-Wallis test comparing the four seasons. – Indicates situations where data are insufficient to run a Kruskal-Wallis test.

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