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Forest Health Assessment in Four Jordanian Reserves Located in Semi-Arid Environments

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Abstract: Healthy forests are essential to human life because they provide food, energy, and other benefits including carbon sequestration. The objective of this study was to assess the forests health status in Mediterranean ecosystems, specifically, arid to semi-arid. Four forest reserves directed by Royal Society for the Conservation of Nature, Jordan were evaluated. Plant health indicators [(gas exchange (photosynthesis, stomatal conductance, transpiration), chlorophyll, middy stem water potential (Ψsmd), relative water content], regeneration, lichens, plant disease, as well as soil variables (respiration CO_2-C, electrical conductivity (EC), pH, microorganisms' abundance) were measured. The Ψsmd values in those semi-arid/arid ecosystems were within the normal ranges (−0.3 to −1.3 MPa) in spring but was under extreme water stress (−1.6 to −5.3 MPa) in summer in three reserves. Similarly, gas exchange variables reduced by 25%–90% in summer (compared to spring) across the studied forests. Although the regeneration (seedling per 1000 m^2) was higher than 100 in two forest (Ajloun and Dibbeen), the number of seedlings in hiking sites was extremely low in both forests. Soil health indicators reveled that soil respiration CO_2-C were higher than 25 mg kg^{-1} in two forests [Ajloun, Dibbeen, (except hiking zone)]. The mean soil saprophytes (number g^{-1}) ranged from 86 to 377 across the forests reserves. In addition, the mean arbuscular mycorrhizal fungi (spores $100g^{-1}$ soil) was between 350 and 877. Soil EC was consistently optimal (less than 0.5 dS m^{-1}) and pH was slightly basic (7.5–8.3) across the reserves. The results revealed that the fluctuation of rainfall and anthropogenic pressures (grazing, hiking) led to partial forest degradation. When forests (Dana Biosphere Reserve) received 81 mm annual precipitation, Ψsmd values in *Juniperus phoenicea* at summer ranged from −4.4 to −5.3 MPa, regeneration and lichens were less than 20 per 1000 m^2, and several trees were dead after infected with soil and air borne pathogens including wilt diseases and die back. Intensive hiking activities (Dibbeen forests, tourism area) and heavy grazing (Yarmouk frosts) reduced regeneration, lichens and soil respiration. Interestingly, the native species had better water relations (RWC, Ψsmd) and gas exchange performance than the introduced species. Overall, it is better to grow native species, and exclude anthropogenic pressure on the territory of introduced species. The conservation programs must persist to sustain several native historical forest trees including *Juniperus phoenicea* (>600 year old), *Quercus ithaburensis* (>500 year old), and *Pinus halepensis* (>100 year old) at Mediterranean semi-arid forests.

Keywords: soil health; forest tree diseases; climate change; physiological status; semi-arid ecosystems

Citation: Alananbeh, K.M.; Othman, Y.A.; Tahat, M.M.; Al-Dakil, H.; Yahya, A.A.; Ayasrah, B.; Al-Share, T.; Alkhatatbeh, S.; Al-Zoubi, R.; Alnaanah, M.; et al. Forest Health Assessment in Four Jordanian Reserves Located in Semi-Arid Environments. *Forests* **2023**, *14*, 918. https://doi.org/10.3390/f14050918

Academic Editor: Minaco Adachi

Received: 3 March 2023
Revised: 25 April 2023
Accepted: 27 April 2023
Published: 28 April 2023

1. Introduction

Human depend on healthy forests to supply food and energy and to provide services such as carbon sequestration, hosting biodiversity (flora and fauna), and regulating the changing climate [1]. The term "Forest Health" is generally used to describe the state of a forest in terms of natural and human processes. Forest health was described by O'Laughlin et al. [2] as "a condition of forest ecosystems that sustains their complexity while meeting human demands". At individual scale, health can be defined as the absence of disease but at larger scale (entire forest stands), indicators of forest health become more difficult to assess [1]. According to Fairweather [3], the management goals of a site can be linked to the ecosystem health of the forest by utilizing certain indicators to measure the performance over an extended period. For example, growth, diversity, damage, crown, and site quality are health indicators used in forest health assessment and each has a weighting score [4]. Forest health assessment is critical for a number of reasons including the following: (1) forests are invaluable ecological, economic and aesthetic resource, (2) management plans of any forest should be carried out based on the knowledge of its health condition because the forests are sensitive to biotic and abiotic stresses and regular monitoring would allow any problem to be solved; and (3) there is a growing awareness that the changing climate (e.g., temperature, rainfall) interact with pests and pathogens leading to more serious threat to forest ecosystems [5]. Vegetation variables (physiological indicators, regeneration, tree pathogens, mortality, etc.), soil indicators (soil biota abundance and activity, soil respiration, pathogens, pH, electrical conductivity (EC), etc.), lichen abundance and diversity have been used as key indicators of forest status or health [6,7].

In the forest ecosystem, the healthy soil have higher levels of active carbon recycle, eliminate greenhouse gases from the environment, release more mineral nutrient for plants, and functions as a dynamic living system for plants and microbes [8]. Chlorophyll pigments provide the required reaction energy for the photosynthesis process by absorbing energy from the light [9]. Photosynthesis is a critical component of plant growth and development, and it is greatly affected by management practices and climatic conditions such as rainfall [9,10]. The U.S. Forest Service and other government agencies have designated lichen communities as a forest health indicator because they provide critical information about ecosystem contaminants and biodiversity [11]. Lichens play a variety of roles in nutrient cycling and as components of food webs. Because lichens are epiphytic, they are completely dependent on woody plants, making them vulnerable to forest management practices [11]. As a result, high levels of pollution or inappropriate forest management practices will reduce the abundance of epiphytic lichens. Overall, understanding and measuring soil and tree-level indicators is essential to evaluate the forest health.

Jordan is Mediterranean country with diverse climatic zones and a geographic location at the meeting point of three continents. Jordan's major biogeographic regions are the Mediterranean, Irano-Turanian, Saharo-Arabian, and Sudanian (sub-tropical) [12]. This distinct region, however, has long hot summers, and relatively short cold winters. In fact, approximately 80% of the Jordanian lands receive less than 200 mm precipitation per year. Furthermore, Jordanian forestlands are limited, diverse and fragmented [7]. Jordan total forest's area is less than 1% (dense forests 398 km^2, spares forest, 394 km^2) [13]. More than 60% of these forests are found in the northern regions (Jerash, Ajloun, and Irbid). At the national level, four types of woody species have been identified: Aleppo pine (*Pinus halepensis*), deciduous oak (*Quercus coccifera*), Phoenicean Juniper (*Juniperus phoenicea*), and evergreen oak (*Quercus coccifera*) [12]. Although Jordanian native forests in northern (e.g., Ajloun, Dibbeen, Yarmouk) and southern regions (Dana) are adapted to minor degree of disturbance, all forests now encounter unusual stresses in the form of climate change, fires, illegal logging, and disease. Identifying how amplification of these novel stresses will affect the trajectory of forests in the future is a major scientific challenge [1]. In fact, determining thresholds for rapid forest deterioration is critical because it take several decades for forests to restore the services they provide, particularly in arid environments [1,7]. For example, ground measurements of trees height in severely burned areas of Aleppo pine (*Pinus halepensis*) forest in Northern Jordan in 2003 revealed that, after 20 years of regeneration, trees in severely burned areas were significantly shorter than those from the nearby unburned areas [7]. The exponential rise in global temperatures caused by climate change, as well as the frequent drought periods, have had a negative impact on forestlands. The impact of these harsh conditions on Jordanian forests, however, is not fully understood. In fact, no study that we aware of has assessed the health of Jordanian forests. Sustainable forest management and conservation efforts necessitate a clear understanding of the ecological status (health) of forests. As a result, assessing forest health by measuring soil and vegetation health indicators is critical to improving our understanding of how arid ecosystems interact and function as well as the impact of changing climate on those ecosystems. The objective of this study was to assess forests health status in Mediterranean ecosystems, specifically, arid to semi-arid by evaluating forest tree physiology and pathology, regeneration, lichens as well as soil health components (respiration, and microbial abundance).

2. Materials and Methods

2.1. Study Sites

The study was conducted in four natural Jordanian reserves (Figure 1), Dibbeen (32°14′31.47″ N, 35°49′45.97″ E), Ajloun (32°22′55.37″ N, 35°45′51.26″ E), Yarmouk (32°39′36.40″ N, 35°41′40.53″ E), and Dana (30°41′16.89″ N, 35°34′17.23″ E). These forests located within the following bio-geographic zones, Mediterranean Irano-Turanian, and Sudanian (sub-tropical). The mean temperatures and precipitation for the reserves are shown in Figure 2. Each reservation was divided into different zones based on the density and type of tree cover (pure, mixed), elevation, and land usage (open to tourism or no). Dibbeen Forest Reserve is in Jerash, Northern Jordan. This ecological zone (Jerash) contains the last remaining stand of old pine forest in Jordan, as well as approximately 190 km^2 of the world southernmost native Aleppo pine (*Pinus halepensis*) forest [14]. The reserve is divided into three main habitats: the Aleppo pine area, where the native *Pinus halepensis* is dominant (zones A, D, and E), the mixed area (zones B, F, and G) where *Pinus halepensis*, *Quercus coccifera* and *Arbutus andrachne* are grown and the dominant evergreen oak (*Quercus coccifera*) region (Zone C). The reserve's total area is approximately 8.5 km^2. Zone A has an elevation of about 680 m, a dense canopy and is the only site in this reserve that is regularly open for tourism activities. Zones B (840 m), C (970 m), D (710 m), and E (750 m, orchids location) have dense canopies, whereas zone G (673 m) has a scattered to moderate canopy (25%–50%). Zone F (840 m) experienced multiple fires over the past ten years and has moderate canopy density (40%–60%).

Ajloun Forest Reserve covers approximately 13 km^2 and has an altitude ranging from 700–1100 m (Figure 1). The soil is fertile red Terra-Rosa type, and the climate is typical Mediterranean (cold rainy winters and hot, dry summers) [12]. The reserve is dominated by the evergreen oak *Quercus coccifera* [12]. Furthermore, a few Palestinian pistachio (*Pistacia palaestina*), Strawberry Tree (*Arbutus andrachne*), Hawthorn (*Crataegus aronia*), Buckthorn (*Rhamnus palaestinus*), and deciduous oak trees (*Quercus infectoria*) can be found scattered among the dominant evergreen oak. In this study, the reserve was divided into four zones: A, B, C, and D. Zone A has an elevation of 1010 m, a low cover density (<50%) and is regularly open for tourism. Zones B (970 m), and C (960 m) have dense oak cover density (more than 80%), whereas Zone D (890 m) has low to moderate (25–50%) oak cover density.

The Yarmouk Forest Reserve is located on Jordan's Northern border and covers an area of approximately 20.5 km^2 (Figure 1). The reserve consists mainly of three habitat zones, A, B, C. Across the reserve, the canopy density of the trees is less than 60%. Mountainous zone (zone A, 400 m) is dominated by sporadic deciduous oak trees (*Quercus ithaburensis*). Zone B (219 m) is a mixture of scattered oak (*Quercus ithaburensis*) and carob (*Ceratonia siliqua*), while zone C (155 m) is dominated by the introduced pine (*Pinus halepensis*).

Jordan's natural Phoenician juniper (*Juniperus phoenicea*) grows in the country's south. Even though human activity has destroyed a large portion of the juniper forests, several stand formations are protected in Dana Biosphere Reserve. The total area of Dana Biosphere Reserve is 292 km^2 (Figure 1). Dana Biosphere Reserve includes three different bio-geographical zones: Mediterranean, Irano-Turanian, and Sudanian penetration [15]. It is also home to the southernmost remaining forest community of *Cupressus simpervirens* [15]. In this study, the reserve was divided into four zones: A, B, C, and D (Figure 1). Zone A (1310 m) are a mixture of scattered *Juniperus phoenicea* and *Cupressus sempervirens* trees. The tourism camp area (zone B, 1175 m) is a mixture of scattered *Juniperus phoenicea* and *Quercus coccifera*. Zone C (1300 m) is dominated by *Juniperus phoenicea* with a few *Quercus coccifera* and *Pinus halepensis* trees. Zone D (1220 m) is open for tourism activities and is dominated by native *Juniperus phoenicea*.

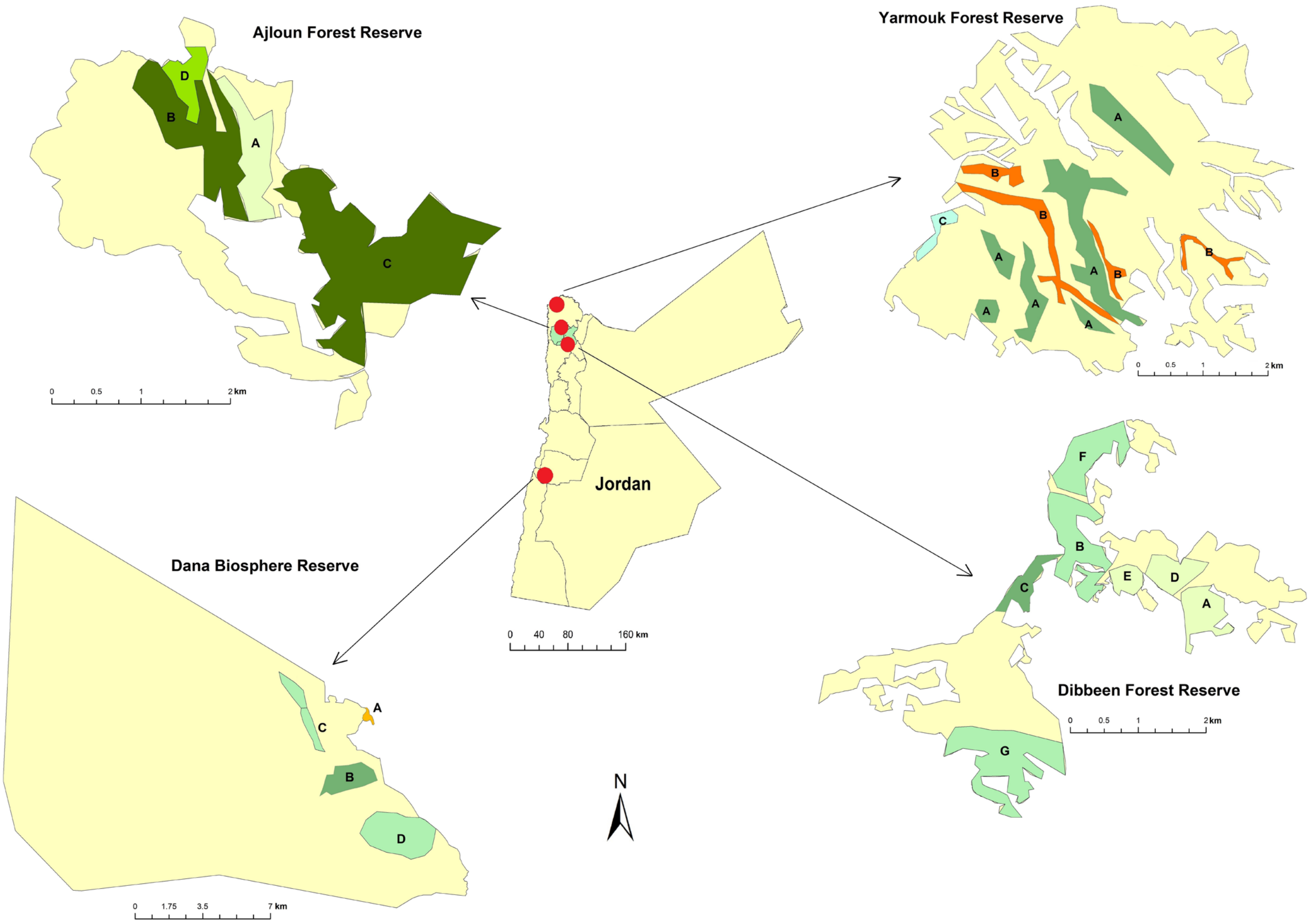

Figure 1. Ajloun, Yarmouk, Dibbeen and Dana Forest reserves, Jordan. A, B, C, D, E, F, and G are the location of sampling sites which represent different vegetation type/and or cover density across the reserves.

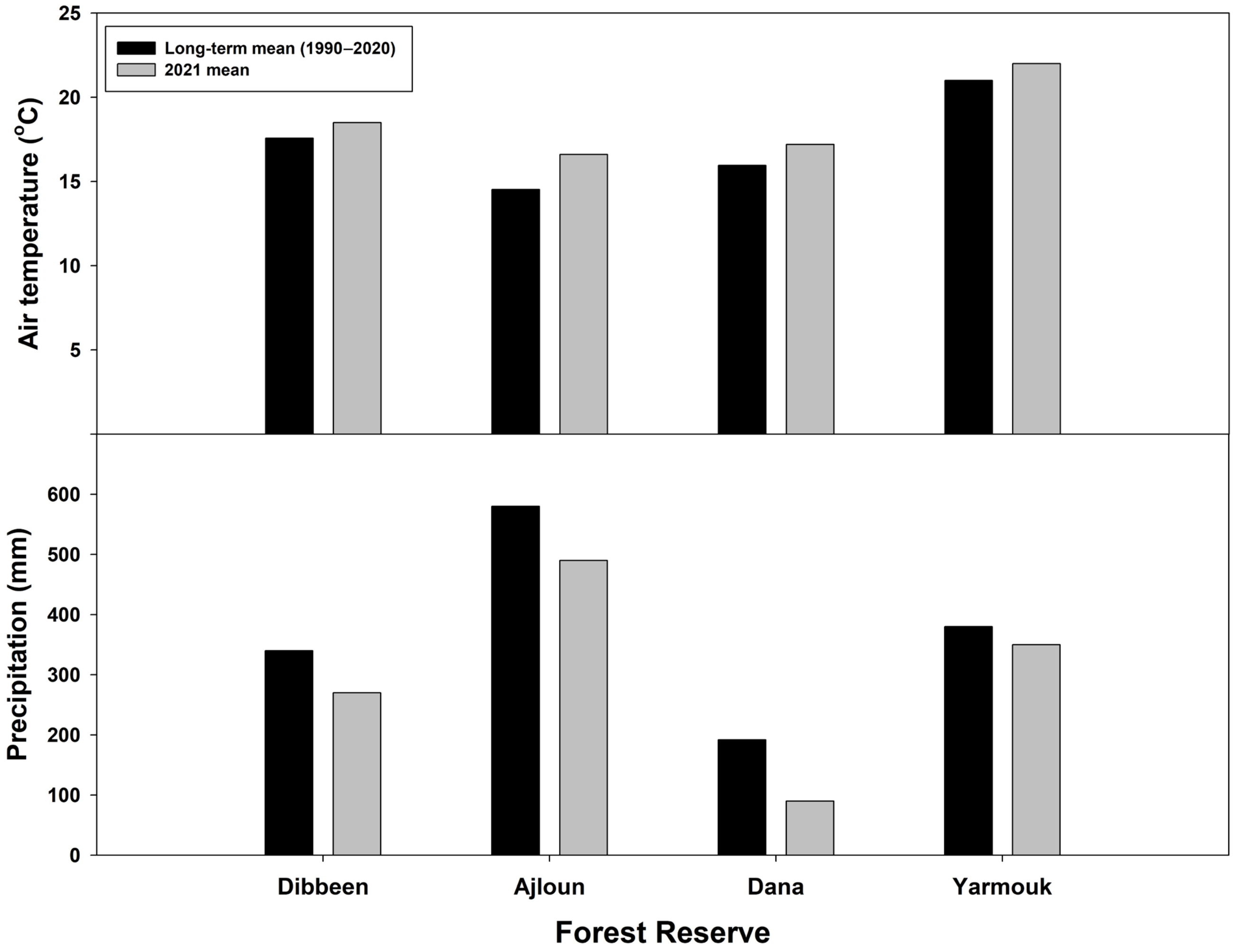

Figure 2. Long-term (1990–2020) annual temperature and precipitation for Dibbeen, Ajloun, Dana and Yarmouk reserves, Jordan.

2.2. Measurements

Tree and soil measurements were conducted in April and September 2021. Plant health indicators, gas exchange [photosynthesis (*Pn*), stomatal conductance (*gs*), and transpiration (*E*)], midday stem water potential (Ψsmd), canopy temperature and chlorophyll content index (SPAD) were determined (10 samples per zone) between 11:00 a.m. and 1:00 p.m. Gas exchange component was measured using a portable photosynthesis system (LI-6400XT; LI-COR, Lincoln, NE) as described in Leskovar and Othman [16]. Reference CO_2 was set to 400 µmol, light intensity was set to track ambient photosynthetically active radiation, area of chamber head to 6 cm^2 and flow rate to 500 µmol s^{-1}. Midday stem water potential was measured on fully equilibrated leaves following the procedure of Othman et al. [17]. Leaf from the lower part of the tree was enclosed with aluminum foil for two hours to equilibrate the leaf moisture with stem and then Ψsmd was measured using a pressure chamber (PMS instrument, Corvallis, OR, USA). The chlorophyll content index (SPAD) was determined on two mature leaves per tree (middle part of the tree) using a chlorophyll meter (CCM-200 plus; Opti-Science, NH, USA). In addition, shoot samples were collected from infected trees (leaves, branches, bark, and fruits) for pathogenic analysis. For, regeneration and lichens, a 100 m × 10 m block was in each zoon across the reserves and then the total number of regeneration tree seedlings were determined. In addition, the total number of trees that have lichens on its branches were determined.

Soil samples were collected to determine the baseline soil health. To ensure consistency in soil sampling horizons and periods, soil cores (4 cores per zone, from 0–30 cm depth) were collected, sieved in a 2 mm sieve, and transferred in ice box into laboratory from the four reserves. Soil respiration was measured using the Solvita Soil Respiration System (Solvita) [18]. The test was determined on 40 g air dried soil. Soil CO_2-C released was measured using a digital-color reader (Solvita-DCR, Woods End Laboratories, Inc., Mt. Vernon, ME, USA). Soil pH, and EC was determined in a 1:1 (water:soil) extract. Soil samples (sieved with 2-mm sieves) and root parts were also collected from the same horizons (0–30 cm) and locations used for soil respiration test, for microorganisms (fungi and nematodes) quantification and culturing [19]. For leaves, at least 10 random leaves from each tree were randomly collected and packed in paper bags, dried, and stored until further processing [20]. For roots and shoot parts, samples were cut into small pieces with a diameter of 0.5 mm, sterilized with 5% hypochlorite for three minutes, washed three times with sterile distilled water, and then cultured on potato dextrose agar (PDA) medium and incubated at 25 °C for 7–10 days. Colonies were hyphal tipped and pure cultures from fungi were obtained. For obligate fungi, leaves were inspected using the microscope and the fungal genera were identified [20]. The serial dilution technique was also used to isolate fungi from the soil by spreading one milliliter of 10^{-3} dilution over 9-cm petri's of PDA with the same previous conditions. Nematodes and mycorrhizal fungi in the soil were determined using soaking method for 24 h. A compound microscope was then used to identify fungal morphological characteristics from the pure cultures.

3. Results

3.1. Dibbeen Forest Reserve

Tree physiology (*Pn*, *gs*, *E*, Ψsmd, RWC, canopy temperature) and soil EC, pH, respiration (CO_2-C) and surface temperature for Dibbeen Forest Reserve are presented in Table 1. In April, the canopy temperature was between 12–19 °C and the soil temperature was 24–31 °C; in September the canopy temperatures was 32–30 °C and the soil temperature was 33.5–45 °C. The water status of the trees is represented by the midday stem water potential and RWC. More negative Ψsmd values indicate that the tree was under water deficit stress. In this study, Ψsmd values for Dibbeen Forest Reserve in April ranged between −0.3 and −1.07 MPa (good conditions). On the other hand, Ψsmd exceeded the −1.5 MPa threshold value across all zones in September. The highest Ψsmd values (more negative) was noticed for *Arbutus andrachne* trees. RWC was also high in April (79.6%–85.1%) and low in September (64.7%–81.5%). Gas exchange component (*Pn*, *gs*, *E*) are critical for tree health assessment. These indicators (*Pn*, *gs*, *E*) are highly associated with the tree water status (e.g., Ψsmd) and chlorophyll content. In spring (April), *Pn* ranged from 27 to 37 µmol m^{-2} s^{-1}, *gs* 0.1–0.2 mol m^{-2} s^{-1} and *E* 1.5–3.0 mmol m^{-2} s^{-1}. However, during the summer (September), the gas exchange of trees is significantly reduced by more than 30% across all tree species and zones. In April, the chlorophyll content index (SPAD) ranged between 19 and 31, and in September, it ranged between 18.5 and 25.

The total regeneration number of Dibbeen Forest Reserve zones in April 2021 ranged from 25 to 230 seedlings per plot (Table 1). Zone A (tourism area) had two *Pinus halepensis* and 23 *Quercus coccifera* seedlings (for a total of 25), zone B (mixed forest) had 54 *Pinus halepensis*, 7 *Arbutus andrachne* and 127 *Quercus coccifera* seedlings (for a total of 188), zone C (pure oak) had 109 *Quercus coccifera* seedlings, zone D had 120 *Pinus halepensis* seedlings, zone F had 30 *Pinus halepensis*, 6 *Arbutus andrachne* and 82 *Quercus coccifera* seedlings (a total of 118), zone G had 174 *Pinus halepensis*, and 55 *Quercus coccifera* seedlings (a total of 229). Lichens number per plot were 11 for zone A, 146 for zone B, 160 for zone C, 150 for zone D, 95 for zone F and 71 for zone E.

Table 1. Tree physiology gas exchange [photosynthesis (*Pn*), stomatal conductance (*gs*), and transpiration (*E*)], midday stem water potential (Ψsmd), relative water content (RWC), leaf temperature and chlorophyll content index (SPAD), soil pH, electrical conductivity (EC), respiration (CO_2-C), temperature, seedling total regeneration and lichens number of Dibbeen Forest Reserve zones in April and September 2021. Zone A elevation, 680 m; B, 840 m; C, 970 m; D, 710 m; E, 750 m; F, 840 m and G 973 m. Plot size for seedling regeneration and lichens number evaluation was 100 m × 10 m.

		Tree Leaves							Soil					
Zone/Time	**Tree Species**	**Temp. (°C)**	**RWC (%)**	**Ψsmd (MPa)**	***Pn*** **(µmol m^{-2} s^{-1})**	***gs*** **(mol m^{-2} s^{-1})**	***E*** **(mmol m^{-2} s^{-1})**	**SPAD**	**pH**	**EC (dS m^{-1})**	**CO_2-C (mg kg^{-1})**	**Temp. (°C)**	**Regeneration (No. $Plot^{-1}$)**	**Lichenes (No. Plo^{-1})**
April														
A	*Pinus halepensis*	14.2 bc	82.7 cd	−0.75 bcd	27.6 e	0.07 c	1.50 ef	- *	8.1 a	0.24 a	16.3 de	24.0 e	25 **	11
	Arbutus andrachne	10.4 f	80.0 d	−0.87 ab	31.9 bc	0.19 a	2.97 a	31.1 a	7.9 a	0.22 a	16.8 de	28.0 bcd	-	-
	Quercus coccifera	12.7 ef	80.8 d	−0.80 bc	28.2 e	0.11 bc	1.59 ef	27.7 ab	7.8 a	0.23 a	6.67 e	28.0 bcd	-	-
B	*Pinus halepensis*	15.7 abc	81.3 cd	−0.77 bcd	32.5 b	0.15 ab	2.99 a	-	8.0 a	0.24 a	48.0 abc	26.6 cd	188	146
	Arbutus andrachne	14.0 bc	85.1 bc	−0.60 cd	31.6 bcd	0.18 a	2.85 ab	26.6 ab	7.9 a	0.22a	71.3 ab	27.0 cd	-	-
	Quercus coccifera	14.0 bc	87.7 ab	−0.53 cde	28.8 e	0.09 bc	1.71 c-f	21.8 b	7.8 a	0.24 a	53.3 abc	27.0 cd	-	-
C	*Quercus coccifera*	12.0 ef	89.8 a	−0.35 e	37.5 a	0.15 ab	3.01 a	19.0 c	7.9 a	0.33 a	53.3 abc	26.3 d	109	160
D	*Pinus halepensis*	14.7 bc	80.0 d	−0.80 bcd	28.2 e	0.10 bc	1.75 c-f	-	7.9 a	0.25 a	69.7 ab	29.0 abc	120	150
E	*Pinus halepensis*	19.0 a	81.6 cd	−0.73 bcd	28.0 e	0.15 ab	1.53 ef	-	7.9 a	0.33 a	74.0 a	27.0 cd	130	140
F	*Pinus halepensis*	18.0 ab	79.6 d	−0.77 bcd	28.3 e	0.13 abc	1.97 b-e	-	7.8 a	0.24 a	63.3 abc	27.6 cd	118	92
	Arbutus andrachne	19.0 a	81.3 cd	−1.07 a	32.0 bc	0.18 a	2.39 abc	27.7 ab	7.7 a	0.25 a	56.7 abc	31.0 a	-	-
	Quercus coccifera	17.0 abc	82.6 cd	−0.57 cde	27.2 e	0.13 abc	1.80 c-f	31.0 a	7.9 a	0.26 a	58.3 abc	27.0 cd	-	-
G	*Quercus coccifera*	16.0 abc	80.6 d	−0.57 cde	29.0 de	0.15 ab	2.50 abc	27.3 ab	8.2 a	0.25 a	40.8 cd	29.6 ab	229	71
	Pinus halepensis	16.7 abc	82.0 cd	−0.70 cd	29.6 cde	0.11	2.17 a-d	-	7.9 a	0.24 a	45.0 bcd	28.0 abc	-	-
p-value		<0.0001	0.001	0.001	0.02	0.01	0.002	0.04	0.06	0.1	0.0003	0.002	-	-
September														
A	*Pinus halepensis*	25.7 de	79.3 abc	−1.87 ef	19.3 c	0.05 c	1.20 e	-	8.0 a	0.30 a	33.3 efg	41.1 bcd	-	-
	Arbutus andrachne	24.3 ef	70.2 de	−3.67 a	15.0 d	0.05 c	2.38 a	22.2 a	7.9 a	0.25 a	20.7 fg	38.9 de	-	-
	Quercus coccifera	22.9 g	64.7 e	−2.97 b	15.4 d	0.08 abc	1.27 de	23.3 a	7.9 a	0.22 a	14.7 g	36.6 ef	-	-
B	*Pinus halepensis*	29.3 abc	81.5 a	−1.62 f	22.7 b	0.12 a	2.39 a	-	7.8 a	0.27 a	133 b	41.0 bcd	-	-
	Arbutus andrachne	24.0 ef	75.1 a-d	−3.53 a	12.6 d	0.05 c	2.28 ab	24.9 a	8.0 a	0.23 a	130 b	33.5 g	–	-
	Quercus coccifera	25.7 de	65.6 e	−2.40 c	20.2 bc	0.07 abc	1.37 cde	19.9 a	7.6 a	0.20 a	136 b	36.0 fg	-	-
C	*Quercus coccifera*	30.5 a	77.1 abc	−2.70 b	26.2 a	0.11 ab	2.41 a	20.0 a	7.9 a	0.28 a	193 a	39.6 cd	-	-
D	*Pinus halepensis*	28.4 bc	78.3 abd	−1.77 ef	19.7 c	0.08 abc	1.40 cde	-	8.0 a	0.27 a	89.0 bcd	39.8 bcd	-	-
E	*Pinus halepensis*	30.0 ab	80.0 ab	−1.60 f	19.6 d	0.12 a	1.23 de	-	7.9 a	0.31 a	70.0 def	42.0 bc	-	-
F	*Pinus halepensis*	29.2 abc	77.8 abc	−2.10 d	19.8 c	0.10 ab	1.57 b-e	-	7.8 a	0.22 a	121 bc	40.9 bcd	-	-
	Arbutus andrachne	30.4 a	73.6 bcd	−3.40 a	15.0 d	0.07 b	1.91 a-d	20.7 a	7.9 a	0.22 a	60.3 c-f	42.6 ab	-	-
	Quercus coccifera	28.2 bc	72.6 cd	−2.70 b	19.1 c	0.10 ab	1.44 cde	18.5 a	8.0 a	0.25 a	75.7 cde	39.5 cd	-	-
G	*Quercus coccifera*	30.0 ab	77.9 abc	−2.40 c	20.3 bc	0.12 a	2.00 abc	20.5 a	8.1 a	0.40 a	56.7 c-f	45.0 a	-	-
	Pinus halepensis	27.6 cd	77.9 abc	−2.03 de	20.7 bc	0.08 abc	1.74 b-e	-	8.0 a	0.25 a	61.3 c-f	41.3 bcd	-	-
p-value		0.0001	0.002	<0.0001	<0.0001	0.03	0.02	0.25	0.8	0.2	<0.0001	<0.0001	-	-

Means in columns followed by different letters are significantly different at $p < 0.05$. * SPAD was not available for *Pinus halepensis* because the leaf is needle-like and cannot be measured used chlorophyll meter. ** Only one block per zone was collected for regeneration and lichens number in April 2021 and therefore no statistical analysis can be carried out.

Soil health indicators are important in determining forest health. In this study, soil chemical properties (EC and pH), soil microorganisms and total soil respiration CO_2-C were all considered to assess soil health (Table 1). Soil EC of and pH values were similar across Dibbeen Forest Reserve zones and were about 0.3 ± 0.1 dS m^{-1} and 7.9 ± 0.5, respectively. Although soil respiration was similar across species (*Pinus halepensis*, *Quercus coccifera*, *Arbutus andrachne*) the recreation zone (zone A) had lower values in both spring (April) and Summer (September) and across forest trees species, less than 25 mg kg^{-1} soil.

Soil microorganisms included a variety of saprotrophic organisms that decompose organic matter (beneficial microorganisms) such as *Alternaria*, *Aspergillus*, *Penicillium*, *Mucor*, etc. The average number of saprophytic and parasitic fungi that were isolated from the soil ranged from 89–156 and 4–9, respectively. The average number of soil free-living and parasitic nematodes ranged from 400 to 813 individuals 100 g^{-1} soil. For mycorrhizal spore numbers, the second reading that was conducted in September 2021 was at least three-folds than that of April 2021. The average number of mycorrhizae ranged from 88 to 138 spores 100 g^{-1} soil in April 2021 and 450–1100 spores 100 g^{-1} soil in September 2021. *Mycosphaerella*, *Diplodia*, and *Phomopsis* were the main genera responsible for the leaf spot diseases in the Dibbeen reserve (Table 2). Fusarium spp.-related infections that cause wilt diseases were noticed in zones C, D, and E.

Table 2. Soil biota and plant pathogens characterized from *Pinus halepensis* of Dibben reservation in April 2021.

Zone	Soil Microorganisms					Plant Fungal Pathogens	
	Average Number of Fungi/1 g Soil [1]		Dominant Nematodes	Mycorrhizae (Spores/100 g Soil)		Dominant Pathogens in the Forest	
	Saprophytic	Parasitic	Free and Parasitic	1st Reading	2nd Reading	Spots and Blight-Causing	Wilt-Causing
A	89 b	9 a	400 c	125 a	800 b	*Diplodia*, *Phomopsis* *Mycosphaerella*	-
B	96 b	6 a	813 a	88 a	830 b	*Mycosphaerella*, *Diplodia*, *Phomopsis*	-
C	156 a	9 a	550 b	138 a	450 d	*Mycosphaerella*, *Diplodia*, *Phomopsis*	*Fusarium* spp.
D	140 a	4 a	400 c	100 a	900 b	*Mycosphaerella*, *Diplodia*, *Phomopsis*	*Fusarium* spp.
E	-	-	800 a	-	450 d	*Mycosphaerella*	*Fusarium* spp.
F	-	-	785 ab	-	1100 a	-	-
G	-	-	600 b	-	700 c	-	-
p-value	0.04	0.07	0.02	0.08	0.03		

[1] Four samples were analyzed from each zone. Means in columns followed by different letters are significantly different at $p < 0.05$.

3.2. Ajloun Forest Reserve

Table 3 shows the tree, soil, regeneration, and lichens assessment at spring (April) and summer (September). At spring (April 2021), canopy temperature, RWC, gs, SPAD, soil EC were similar across studied zones (A, B, C, D). However, Ψsmd at recreation area (zone A) had more negative values and lower soil respiration compared to zones B, C, and D. During the summer, tree physiology of dominant species (*Quercus coccifera*) potentially changed over the reserve. Mean canopy temperatures increased by 89% (15.7 vs. 29.75 °C), RWC reduced by 20% (85% vs. 68%), Ψsmd increased (more negative) by 528% (−0.39 vs. −2.45 MPa), *Pn* reduced by 93% (31 vs. 16 µmol m^{-2} s^{-1}), *gs* by 75% (0.17 vs. 0.045 mol m^{-2} s^{-1}), E by 27% (2.3 vs. 1.7 mmol m^{-2} s^{-1}) and SPAD by 19% (23 vs. 18.6). Interestingly, soil respiration increased by 118% (67 vs. 118 mg kg^{-1}).

Table 3. Tree physiology gas exchange [photosynthesis (*Pn*), stomatal conductance (*gs*), and transpiration (*E*)], midday stem water potential (Ψsmd), relative water content (RWC), leaf temperature and chlorophyll content index (SPAD), soil pH, electrical conductivity (EC), respiration (CO_2-C), temperature, seedling total regeneration and lichens number of Ajloun Forest Reserve zones in April and September 2021. Zone A (recreation zone) elevation, 1010 m; B, 970 m; C, 960 m and D, 890 m. Plot size for seedling regeneration and lichens number evaluation was 100 m × 10 m. All measurements across zones (A, B, C, D) were conducted on *Quercus coccifera* species.

Zone/Time	Tree Leaves							Soil				Regeneration (No. Plot^{-1})	Lichenes (No. Plot^{-1})
	Temp. (°C)	RWC%	Ψsmd (MPa)	*Pn* (μmol m^{-2} s^{-1})	*gs* (mol m^{-2} s^{-1})	*E* (mmol m^{-2} s^{-1})	SPAD	pH	EC (dS m^{-1})	CO_2-C (mg kg^{-1})	Temp. (°C)		
April													
A	14.5 a	85.3 a	−0.63 c	31.6 ab	0.16 a	2.15 ab	24.5 a	7.15 ab	0.28 a	29.3 d	17.4 a	59 *	98
B	14.6 a	84.4 a	−0.30 ab	28.7 b	0.19 a	1.97 b	24.5 a	7.45 a	0.24 a	87.0 a	20.0 a	89	218
C	16.3 a	86.2 a	−0.24 a	33.6 a	0.18 a	2.90 a	23.7 a	6.79 b	0.24 a	66.0 b	20.9 a	63	144
D	17.4 a	84.1 a	−0.39 b	31.1 ab	0.18 a	2.24 ab	19.5 a	7.06 ab	0.30 a	48.0 c	18.5 a	37	71
p-value	0.11	0.78	0.005	0.02	0.81	0.05	0.44	0.04	0.17	<0.0001	0.12		
September													
A	33.0 a	48.5 b	−2.70 c	14.5 b	0.02 b	0.98 b	20.2 a	7.15 ab	0.26 a	91.3 b	48.0 a	-	-
B	30.0 b	80.0 a	−1.90 a	13.3 b	0.05 b	1.55 b	20.0 a	7.43 a	0.24 a	101 b	45.0 b	-	-
C	28.0 c	76.5 a	−2.50 b	16.5 ab	0.09 a	2.81 a	16.9 a	6.88 b	0.22 a	315 a	42.0 c	-	-
D	28.0 c	68.5 a	−2.70 c	20.4 a	0.02 b	1.37 b	17.4 a	7.08 ab	0.29 a	77.0 b	45.7 ab	-	-
p-value	<0.0001	0.005	<0.0001	0.01	0.004	0.017	0.11	0.05	0.28	<0.0001	0.002	-	-

Means in columns followed by different letters are significantly different at $p < 0.05$. * Only one block per zone was collected for regeneration and lichens number in April 2021 and therefore no statistical analysis can be carried out.

The total regeneration number of Ajloun Forest Reserve zones during the study period was between 37 and 89 seedlings per plot (100 m × 10 m). Zone A (tourism area, scattered oak) plot had 54 *Quercus coccifera* and 5 *Pistacia palaestina* seedlings (for a total of 59), zone B (dense oak forest) had 87 *Quercus coccifera* and 2 *Pistacia palaestina* seedlings (for a total of 89), zone C (dense oak forest) had 50 *Quercus coccifera*, 5 *Pistacia palaestina*, 2 *Arbutus andrachne*, 2 *Crataegus azarolus*, and 4 *Pyrus syriaca* seedlings (for a total of 63), zone D (scattered oak) had 29 *Quercus coccifera* and 8 *Pistacia palaestina* seedlings (for a total of 37). Lichens number per plot (100 m × 10 m) were 98 for zone A, 218 for zone B, 144 for zone C, and 71 for zone D.

Soil microorganisms at Ajloun Forest Reserve included a variety of saprotrophic organisms that decompose organic matter like those found in Dibben Forest Reserve. The average number of saprophytic and parasitic fungi that were isolated from the soil ranged from 65–109 and 3–7, respectively. The average number of soil free-living and parasitic nematodes ranged from 350 to 600 individuals 100 g^{-1} soil. For mycorrhizal spore numbers, the second reading that was conducted in September 2021 was significantly higher than that of April 2021. The average number of mycorrhizae ranged from 56 to 169 spores 100 g^{-1} soil in April 2021 and 1000–2000 spores 100 g^{-1} soil in September 2021. *Taphrina* sp. And *Hypoxylon* sp. were the main genera responsible for the spotted and blight diseases in the Ajloun Forest reserve (Table 4). No wilt-disease-causing pathogens were found in this reserve.

Table 4. Soil biota and plant pathogens characterized from *Quercus coccifera* of Ajloun Forest Reserve in April 2021.

Zone	Soil Microorganisms					Plant Fungal Pathogens	
	Average Number of Fungi/1 g Soil [1]		Dominant Nematodes	Mycorrhizae (Spores/100 g Soil)		Dominant Pathogens in the Forest	
	Saprophytic	Parasitic	Free and Parasitic	1st Reading	2nd Reading	Spots and Blight-Causing	Wilt-Causing
A	65 b	7 a	-	56 c	1500 b	*Hypoxylon*, *Taphrina*	-
B	109 a	6 a	350 b	115 b	1300 b	*Hypoxylon*, *Taphrina*	-
C	83 ab	3 b	-	169 a	1000 c	*Hypoxylon*, *Taphrina*	-
D	-	-	600 a	-	2000 a	-	-
p-value	0.01	0.04	0.01	0.008	0.03		

[1] Four samples were analyzed from each section. Means in columns followed by different letters are significantly different at $p < 0.05$.

3.3. Yarmouk Forest Reserve

The assessment of tree and soil health variables of Yarmouk Forest Reserve are presented in Tables 5 and 6. Zone C which grows with introduced species (*Pinus halepensis*) had higher canopy temperatures, and Ψsmd (more negative) and lower gas exchange (*Pn*, *gs* and *E*) than zones A and B which have the native species *Ceratonia silique* and *Quercus ithaburensis* (Table 5). Interestingly, Ψsmd of *Quercus ithaburensis* species were never exceeded −0.8 MPa in spring and summer conditions. In addition, SPAD values of *Ceratonia silique* was extremely higher than *Quercus ithaburensis* over the study period (April and September 2021). Soil EC were about 0.4 ±0.1 dS m^{-1} and pH was about 7.3 ± 5.0 across the reserve zones. However, soil respiration CO_2-C was extremely low (less than 11 mg kg^{-1} dry soil) in April and increased (41 ± 2.0 mg kg^{-1}) in September 2021. In addition, the regeneration in Yarmouk Forest Reserve zones were zero (zones A, B and C) and the total number of lichens for zone A, B, C were 5, 6 and zero, respectively.

Table 5. Tree physiology gas exchange [photosynthesis (*Pn*), stomatal conductance (*gs*), and transpiration (*E*)], midday stem water potential (Ψsmd), relative water content (RWC), leaf temperature and chlorophyll content index (SPAD), soil pH, electrical conductivity (EC), respiration (CO_2-C), temperature, seedling total regeneration and lichens number of Yarmouk Forest Reserve zones in April and September 2021. Zone A elevation is 400 m; B, 219 m and C, 155 m. Plot size for seedling regeneration and lichens number evaluation was 100 m × 10 m.

Zone/Time	Tree Species	Tree Leaves							Soil				Regeneration (No. Plot^{-1})	Lichenes (No. Plot^{-1})
		Temp. (°C)	RWC%	Ψsmd (MPa)	*Pn* (µmol m^{-2} s^{-1})	*gs* (mol m^{-2} s^{-1})	*E* (mmol m^{-2} s^{-1})	SPAD	pH	EC (dS m^{-1})	CO_2-C (mg kg^{-1})	Temp. (°C)		
April														
A	*Q. ithaburensis*	22.1 b	81.0 ab	−0.63 b	41.3 b	0.26 a	4.96 a	31.7 b	7.51 a	0.41 a	10.7 a	34.0 a	0 **	5
B	*Q. ithaburensis*	20.0 c	85.3 a	−0.43 a	45.6 a	0.25 a	4.97 a	30.6 b	7.49 a	0.38 a	7.42 a	39.6 a	0	6
B	*C. siliqua*	18.4 c	85.7 a	−0.62 b	41.8 b	0.18 ab	4.91 a	64.2 a	7.30 a	0.33 a	5.00 a	34.0 a	-	-
C	*P. halepensis*	25.1 a	77.6 b	−0.67 b	33.4 c	0.05 b	1.72 b	- *	7.08 a	0.44 a	8.33 a	34.3 a	0.0	0.0
p-value		0.002	0.04	0.01	0.002	0.04	0.02	0.001	0.17	0.16	0.52	0.11		
September														
A	*Q. ithaburensis*	31.3 a	63.7 c	−0.73 b	28.5 a	0.28 a	7.21 a	33.8 b	7.5 a	0.32 a	21.0 a	43.7 a	-	-
B	*Q. ithaburensis*	31.7 a	80.9 a	−0.51 a	27.3 ab	0.22 b	7.11 a	35.9 b	7.4 a	0.41 a	24.0 a	41.7 a	-	-
B	*C. siliqua*	31.1 a	85.7 a	−1.30 c	24.2 b	0.10 c	3.96 b	61.6 a	7.2 a	0.36 a	30.7 a	42.3 a	-	-
C	*P. halepensis*	32.5 a	70.1 b	−2.30 d	16.9 c	0.03 c	1.15 c	-	6.8 b	0.43 a	17.7 a	41.2 a	-	-
p-value		0.44	<0.0001	<0.0001	0.004	0.0005	<0.0001	0.002	0.01	0.21	0.22	0.35	-	-

Means in columns followed by different letters are significantly different at $p < 0.05$. * (not available), SPAD was not available for *P. halepensis* because the leaf is needle-like and cannot be measured used chlorphyll meter. ** Only one block per zone for regeneration and lichens number in April 2021 and therefore no statistical analysis can be carried out.

Table 6. Soil biota and plant pathogens characterized from *Quercus ithaburensis* of Yarmouk Forest Reserve in April 2021.

Zone	Soil Microorganisms					Plant Fungal Pathogens	
	Average Number of Fungi/1 g Soil [1]		Dominant Nematodes	Mycorrhizae (Spores/100 g Soil)		Dominant Pathogens in the Forest	
	Saprophytic	Parasitic	Free and Parasitic	1st Reading	2nd Reading	Spots and Blight-Causing	Wilt-Causing
A	185 b	5 a	1000 a	175 b	550 b	*Taphrina caerulescens*, *Gnomonia*	*Ceratocystis* sp.
B	160 b	5 a	666 b	138 b	966 a	*Taphrina caerulescens*, *Gnomonia*	*Ceratocystis* sp.
C	267 a	5 a	200 c	275 a	900 a	*Taphrina caerulescens*, *Gnomonia*	*Ceratocystis* sp.
p-value	0.043	1.0	0.009	0.04	0.03		

[1] Four samples were analyzed from each zone. Means in columns followed by different letters are significantly different at $p < 0.05$.

The main genera responsible for spots illnesses in the Yarmouk reserve trees were *Taphrina caerulescens* and *Gnomonia* sp. Wilt-causing pathogens were also notices and belonged to the *Ceratocystis* sp. in the three studied zones. A variety of saprotrophic organisms that decompose organic matter (beneficial microorganisms) such as *Alternaria*, *Aspergillus*, *Penicillium*, etc. were morphologically identified (Table 6). The number of parasitic fungi was very low. An average of 160–167 parasitic and 5 saprophytic fungi were isolated from the soil (Table 6). The average number of parasitic and soil-free nematodes per 100g of soil ranged from 200 to 1000 individuals. In April 2021, mycorrhizal count was between 138 to 275 spores per 100 g of soil while in September the spore's number were between 550 to 900 spores.

Soil microorganisms at Yarmouk Forest Reserve included a variety of saprotrophic organisms that decompose organic matter like those found in Dibben and Ajloun Forest Reserve. The average number of saprophytic and parasitic fungi that were isolated from the soil ranged from 160–267 and 5, respectively. The average number of soil free-living and parasitic nematodes ranged from 200 to 1000 individuals $100g^{-1}$ soil. For mycorrhizal spore numbers, the second reading that was conducted in September 2021 was significantly higher than that of April 2021. The average number of mycorrhizae ranged from 138 to 275 spores $100g^{-1}$ soil in April 2021 and 550–966 spores $100g^{-1}$ soil in September 2021. *Taphrina* sp. and *Gnomonia* sp. were the main genera responsible for the spotted and blight diseases in the Yarmouk Forest reserve (Table 4). *Ceratocystis* sp. was the wilt-disease-causing pathogen in the three zones of the reserve.

3.4. Dana Biosphere Reserve

Although the canopy temperature at Dana Biosphere Reserve in April was low (12–15 °C), Ψsmd values for *Cupressus sempervirens*, *Juniperus phoenicea* and *Pinus halepensis* were higher (more negative) than −1.2 MPa (Table 7). Furthermore, *gs* ranged between 0.01 and 0.1 mol m^{-2} s^{-1}, *E* 0.32–1.38 mmol m^{-2} s^{-1}, and soil respiration was less than 30 mg kg^{-1} across all studied zones. In September, RWC, Ψsmd, gas exchange (*gs*, *E*) values were extremely low particularly for, *Juniperus phoenicea* trees. Across zones, Ψsmd values for that species ranged from −4.4 and −5.3 MPa (Table 7). *Pn* values, on the other hand, were never lower than 14µmol m^{-2} s^{-1} across all tree's species, zones and dates (April, September). In terms of soil, the EC of Dana Biosphere Reserve zones was approximately 0.3 ±0.1 dS m^{-1} and the pH were approximately 7.9 ± 0.5 across the reserve zones. Furthermore, soil respiration increased by about one-fold in September compared to April (mean of soil CO_2-C across zones 19 vs. 39 mg kg^{-1}).

Table 7. Tree physiology gas exchange [photosynthesis (*Pn*), stomatal conductance (*gs*), and transpiration (E)], midday stem water potential (Ψsmd), relative water content (RWC), leaf temperature and chlorophyll content index (SPAD), soil pH, electrical conductivity (EC), respiration (CO2-C), temperature, seedling total regeneration and lichens number of Dana Biosphere Reserve zones in April and September 2021. Zone A elevation is 1310 m; B, 1175 m, C, 1300 m; D, 1220 m. Plot size for seedling regeneration and lichens number evaluation was 100 m × 10 m.

Zone/Time	Tree Species	Tree Leaves							Soil				Regeneration (No. Plot^{-1})	Lichenes (No. Plot^{-1})
		Temp. (°C)	RWC%	Ψsmd (MPa)	*Pn* (µmol m^{-2} s^{-1})	*gs* (mol m^{-2} s^{-1})	*E* (mmol m^{-2} s^{-1})	SPAD	pH	EC (dS m^{-1})	CO_2-C (mg kg^{-1})	Temp. (°C)		
April														
A	*C. sempervirens*	12.2 a	69.4 a	−1.30 b	29.9 d	0.06 abc	0.57 bc	- *	8.2 a	0.26 a	21.3 ab	27.2 a	11 **	0
	J. phoenicea	12.1 a	68.1 a	−1.87 de	32.5 cd	0.07 ab	0.55 bc	-	8.0 a	0.35 a	13.6 b	29.7 a	-	-
B	*Q. coccifera*	13.1 a	78.6 a	−0.80 a	39.5 a	0.10 a	1.38 a	30.7 a	7.6 a	0.36 a	18.8 ab	30.0 a	5	9
	J. phoenicea	15.0 a	69.4 a	−2.41 f	39.6 a	0.07 ab	0.89 b	-	7.7 a	0.35 a	12.3 b	31.4 a	-	-
C	*P. halepensis*	12.7 a	73.8 a	−1.70 cde	35.5 bc	0.01 d	0.38 c	-	8.0 a	0.35 a	23.6 ab	28.6 a	0	10
	J. phoenicea	14.8 a	66.3 a	−1.97 e	40.4 a	0.01 d	0.32 c	-	7.6 a	0.40 a	30.3 a	30.2 a	-	-
	Q. coccifera	15.6 a	80.6 a	−1.40 bc	38.9 ab	0.05 c	0.71 bc	26.0 a	7.9 a	0.34 a	19.7 ab	31.6 a	-	-
D	*J. phoenicea*	14.2 a	72.6 a	−1.60 bcd	35.7 bc	0.06 abc	0.61 bc	-	7.8 a	0.31 a	12.0 b	26.2 a	8	0
p-value		0.27	0.21	<0.0001	0.001	0.001	0.007	0.12	0.23	0.16	0.05	0.11	-	-
September														
A	*C. sempervirens*	30.0 a	69.4 bc	−2.63 b	14.4 d	0.02 b	0.78 c	-	8.1 a	0.30 a	56.7 a	37.0 c	-	-
	J. phoenicea	30.0 a	64.6 d	−5.00 d	17.9 c	0.02 b	0.92 c	-	7.9 a	0.33 a	48.7 a	38.0 c	-	-
B	*Q. coccifera*	25.0 b	63.9 d	−2.37 ab	24.5 a	0.04 a	2.51 a	31.2 a	7.8 a	0.38 a	35.0 a	39.3 abc	-	-
	J. phoenicea	24.0 b	67.5 cd	−5.33 d	20.6 bc	0.02 b	0.99 c	-	7.4 a	0.38 a	30.0 a	43.3 a	-	-
C	*P. halepensis*	20.0 c	74.6 a	−2.20 a	19.5 bc	0.02 b	0.71 c	-	7.9 a	0.32 a	36.7 a	41.0 abc	-	-
	J. phoenicea	25.0 b	66.7 cd	−4.40 c	21.6 ab	0.01 b	0.68 c	-	7.8 a	0.35 a	34.0 a	40.0 abc	-	-
	Q. coccifera	21.0 c	74.2 ab	−2.27 ab	22.3 ab	0.04 a	1.73 b	29.8 a	7.7 a	0.38 a	28.0 a	39.0 abc	-	-
D	*J. phoenicea*	25.7 b	64.1 d	−5.17 d	19.7 bc	0.02 b	0.96 c	-	7.8 a	0.38 a	46.7 a	42.3 ab	-	-
p-value		0.01	0.01	0.001	0.002	0.003	0.007	0.27	0.22	0.47	0.56	0.05	-	-

Means in columns followed by different letters are significantly different at $p < 0.05$. * (not available), SPAD was not available for *Pinus halepensis* because the leaf is needle-like and cannot be measured used chlorophyll meter. ** Only one block per zone was collected for regeneration and lichens number in April 2021 and therefore no statistical analysis can be carried out.

In April, the total number of regeneration and lichens in Dana Biosphere Reserve ranged between 0 to 11 across the study zones (Table 7). For regeneration, Zone A plot had 6 *Cupressus sempervirens* and 5 *Juniperus phoenicea* seedlings (for a total of 11), zone B (camp area) had two *Quercus coccifera*, one *Pistacia atlantica* and two *Juniperus phoenicea* seedlings (for a total of 5), zones C had no regeneration and zone D had had one *Quercus coccifera* and 7 *Juniperus phoenicea* (for a total of 8). For lichens, the total number per plot (100 m $\times$ 10 m) for zone A was 0.0, zone B was 9, zone C was 10, and zone D was 0.0.

Soil microorganisms at Dana Forest Reserve included a variety of saprotrophic organisms that decompose organic matter like those found in the other reserves. The average number of saprophytic and parasitic fungi that were isolated from the soil ranged from 270–451 and 3–6, respectively (Table 8). The average number of soil free-living and parasitic nematodes ranged from 200 to 800 individuals 100 g^{-1} soil. For mycorrhizal spore numbers, the average number ranged from 138 to 250 spores 100 g^{-1} soil in April 2021 and 300–750 spores 100 g^{-1} soil in September 2021. *Phomopsis*, *Cercospora*, and *Kabatina* sp. were the main genera responsible for the spotted and blight diseases in the Dana Forest reserve (Table 8). *Diplodia* and *Fusarium* sp. were the wilt-disease-causing pathogen in section C of the reserve.

Table 8. Soil biota and plant pathogens of Dana Biosphere Reserve in April 2021.

Zone	Tree Species	Soil Microorganisms					Plant Fungal Pathogens	
		Average Number of Fungi/1 g Soil [1]		Dominant Nematodes	Mycorrhizae (Spores/100 g Soil)		Dominant Pathogens in the Forest	
		Saprophytic	Parasitic	Free and Parasitic	1st Reading	2nd Reading	Spots and Blight-Causing	Wilt-Causing
A	*P. halepensis*	375 ab	6 a	583 b	200 ab	366 bc	*Phomopsis, Cercospora, Kabatina*	-
B	*J. phoenicea*	451 a	5 a	600 b	250 a	300 c	*Phomopsis, Cercospora, Kabatina*	-
C	*J. phoenicea*	270 b	3 b	800 a	138 b	750 a	*Phomopsis, Cercospora, Kabatina*	*Diplodia, Fusarium*
D	*J. phoenicea*	413 a	6 a	200 c	250 a	550 b	*Cercospora*	-
p-value		0.05	0.03	0.01	0.04	0.02		

[1] Four samples were analyzed from each section. Means in columns followed by different letters are significantly different at $p < 0.05$.

4. Discussion

4.1. Forest Tree Physiology

Forest health is defined as the ability of forest ecosystems to maintain their complexity while meeting human needs [2]. Steps to developing a sustainable forest health plan are (1) select a representative set of indicators for a specific ecosystem (2) develop baseline data (3) establish standards against which to compare current forest status; and (4) launch a monitoring program to evaluate current conditions and amend baseline data in the future [2]. The Forest Inventory and Analysis program (FIA) which collects, analyzes, and reports data on the status of American forests, used a variety of indicators to assess forest health including crown condition, tree damage mortality, lichen abundance and communities, down woody materials, vegetation profile, soil health, nonnative invasive plants, regeneration fragmentation and landscape context [6].

Several forest health indicators including water relations (RWC, Ψsmd), gas exchange, chlorophyll, diseases, regeneration, lichens abundance and soil components were measured in this study. Midday stem water potential Ψsmd has been proposed as a precise plant water status indicator [17]. For trees, the suggested Ψsmd values for optimal growth is between −0.3 and −1.0 MPa while stressed trees normally exceed −1.5 MPa and become more negative [17]; thus, normal ranges should be less than −1.5 MPa. In this study, Ψsmd values were within the normal ranges (−0.3 to −1.3 MPa) across the reserves and tree species in spring (April 2021) except for *Juniperus phoenicea* at Dana Biosphere Reserve (Tables 1, 3, 5 and 7). This can partially attributed in part to the low precipitation during that season (81 mm vs. long term mean 192 mm). However, when temperatures rose in summer (September 2021), Ψsmd values exceeded the −1.5 MPa thresholds (become more negative) at Dibbeen, Ajloun, and Dana reserves in all tree species. The high Ψsmd values were for *Arbutus andrachne* (−3.4 to −3.7 MPa) in Dibbeen and *Juniperus phoenicea* (−4.4 to −5.3 MPa) in Dana Reserve. Interestingly, while Ψsmd values in *Pinus halepensis* (introduced species) at Yarmouk Forest Reserve exceeded −1.5 MPa, native species *Quercus ithaburensis* and *Ceratonia silique* were able to maintain good water levels in their shoots (Ψsmd, −0.5 to −1.3), emphasizing the importance of selecting the proper tree species for afforestation, particularly the native ones.

The gas exchange component (*Pn*, *gs*, *E*) are required for tree growth and development [9,10,17]. Furthermore, leaf chlorophyll is an important component for carbon assimilation because these pigments are absorbing sunlight energy and supply the required reaction energy for the photosynthesis process [9]. The gas exchange component and canopy temperature strongly related to the tree's water status [10]. High RWC and low Ψsmd (less negative) reduce leaf and canopy temperature and increase carbon assimilation (photosynthesis) in the forest. However, high Ψsmd (more negative than −1.5 MPa) resulted in significant decrease in gas exchange and an increase in canopy temperatures [17]. In this study, gas exchange was within the normal range in spring (April) but potentially reduced in summer (September) across reserves, Dibbeen, Ajloun and Dana. This could be attributed in part to lower water levels (Ψsmd) in the trees.

4.2. Forest Regeneration and Lichens

Stressors such as grazing animals, invasive plants, and climate variability (e.g., unpredictable rainfall) have negative impact on young tree seedlings and overall regeneration of trees [10]. Forest regeneration typically establishes the long-term trajectory of the forest's composition and function of the forest [6]. In this study, the regeneration levels were higher than 100 seedlings per 1000 m^2 for Ajloun and Dibbeen reserve, except for the tourism area (Zone A) in Dibbeen Forest Reserve. Furthermore, when compared to the dense oak zones (B and C) at Ajloun, the scattered oak (Zone D) and tourism region (Zone A) had the lowest regeneration. In addition, regeneration in Dana Biosphere Reserve was extremely low (1–10 seedlings) and did not exist at Yarmouk Forest Reserve studied zones. The lack of regeneration at Yarmouk is due to reserve's high grazing rate, which browsed the groundcover layer including the regenerated seedlings. Although the grazing is s restricted at Dana, these areas experienced an extended drought period in the last decade (annual precipitation values were below 100 mm) resulting in decreased tree health and regeneration. Lichen is well-known ecological indicator of forest health. Lichens lack roots and rely entirely on the atmosphere for water and nutrition. This distinctive feature makes them highly sensitive to air pollutants, microclimate, nutrient availability and consequently, the changing climate [6]. During the study period, lichen numbers (per 1000 m^2) were less than 20 at tourism zone of Dibbeen and across all sites at Dana and Yarmouk reserves. These results highlight the impact of open recreation, grazing and changing climate (especially rainfall) on trees health as well as regeneration and lichens abundance.

4.3. Soil Health Indicators

Soil health is the list of intrinsic characteristics of soil that define its vigor [8]. Soil health has been defined in the literature as "the capacity of a soil to function as a vital living system within ecosystem and land use boundaries to sustain plant and animal production, maintain or enhance water and air quality, and promote plant and animal health" [21]. Soil biota are linked to pathogenic organism suppression, nutrient cycling, and water storage and detoxification [22,23]. Plant residues are mineralized by soil microorganisms into nutrients that are easily absorbed by plants [24]. Soil biota such as, bacteria and fungi can transform N between inorganic and organic forms and consequently, influences the uptake and composition of minerals by the tree [25]. In fact, microbial communities play critical roles in fundamental processes that ensure stability and productivity for ecosystems [26]. Root-associated soil microorganisms promote ecosystem stability by positively influencing how plants respond to environmental fluctuations and improving their adaptation to extreme stresses (e.g., drought and high temperatures) [27]. Soil microbial components such as abundance, diversity, activity, and stability are important indicators of soil health [21,28]. The abundance of soil biota such as Arbuscular mycorrhizal fungi (AMF), active bacteria, and beneficial nematode is significantly related to soil water storage, and nutrient cycling, soil fertility and plant performance (health) [21,28,29]. Arbuscular mycorrhizal fungi play a fundamental function in the establishment and maintenance of plant communities in forest soil rhizosphere, ecosystem, and biodiversity [30,31]. Several studies have concluded that AMF can be used as a biological and ecological indicator of forest health and quality [32]. Mycorrhizal fungi—associated with plant roots can benefit the host by improving minerals and water uptake. In contrast, carbohydrates are provided by the plant to mycorrhizal fungi [33]. Mycorrhizal association colonization has a bio-protective and control effect against soil inhibitors. Overall, soil indicators are critical for determining the health and composition of a forest over time.

In this study, soil EC was consistently optimal (less than 0.5 dS m^{-1}) across the zones and over the studied reserves. In addition, soil pH was slightly basic (7.5–8.3) across the reserves. However, soil respiration and biota abundance were different across the studies reserves as well as within the reserve zones. During the study period, Ajloun and Dibbeen (except tourism zone) had constantly higher soil respiration values than Dana and Yarmouk reserves. In term of soil saprophytics, the mean value (number g^{-1}) was 120 at Dibbeen, 86 at Ajloun, 377 at Dana and 204 at Yarmouk reserve. In addition, the mean AMF (spores $100g^{-1}$ soil) at Dibbeen Forest Reserve was 516; Ajloun, 877; Dana, 350 and at Yarmouk was 501. Considering the soil respiration and biota results, management practices should be carried out in Yarmouk (all zones), Dibbeen and Ajloun (tourism region), and Dana (all zones) to enhance soil microbial abundance and activity.

4.4. Plant and Soil Pathogens

Diseases, insects, fire, and bad weather have a significant impact on the look and commercial and recreational use of forested land. Of these, diseases and wood decay outrank all other agents in destructiveness, causing the highest losses and posing the biggest threat to tree growth [34]. Diseases can be brought on by a wide variety of living and non-living things, and they can affect a tree's growth, quality, ability to survive, or susceptibility to further pest attacks [35]. Tree disease is typically caused by infectious or non-infectious agents. However, the vulnerable tree, predisposing environmental factors, or a live, infectious agent like a fungus, interact in a complex way before disease symptoms manifest. Greater understanding of the dynamics underlying disease outbreaks can be achieved by being aware of this interaction between living and non-living agents [35].

In this study, and based on growth and morphological traits, the most significant fungal diseases in four Jordanian reserves were identified. However, future work should consider molecular techniques for confirmation of the identification. *Diplodia* sp. and *Phomopsis* sp. were identified from Dibbein and Dana reserves. There are two different species of *Diplodia* cause diplodia shoot blight and diplodia canker on pines. *D. pinea* is more aggressive and capable of doing more serious harm compared to *D. scrobiculata*, *Diplodia* destroys young needles [36]. This is the first report of *Diplodia* sp. diseases on pines in Jordan. Diplodia canker of *Juniperus phoenicea* caused by *Diplodia africana*, may cause extensive crown dieback, and severely infected plants eventually died [37]. This fungus is first reported in this study.

Phomopsis juniperovora or *Kabatina juniperi* are two fungi that can cause juniper tip blight. Both types of fungi result in tip dieback and twig cankers, which weaken and deform the plant. The plant may die from the disease if the infection is sufficiently severe. It primarily affects juniper, although it can also damage cedar, cypress, and certain other conifers [38]. In this study Phomopsis was reported on pines and junipers. The fungus was previously reported on oak (*Q. ithaburensis)* in Jordan in Mamlouk et al. checklist in 1984 [39], but it is the first time reported on juniper. For *Kabatina*, it is reported for the first time in this study either on pines or junipers.

The genus *Mycosphaerella dearnessii* causes brown spot needle blight of pine. Small trees and lower branches on large trees are the most affected by the fungus. Young long leaf pine is especially vulnerable because of its distinctive growth pattern (grass stage) [40]. This fungus is reported for the first time on pines in Jordan.

Cercospora Blight of Juniper is reported for the first time in Jordan. Cercospora needle blight can cause devastation and mortality to juniper especially with repeated years of infection. The foliage of the inner branches of the lower crown becomes bronzed, then necrotic, and eventually sheds, leaving the inner crown devoid of foliage. The extremities remain green [41].

The two genera of *Gnomonia* and *Ceratocystis* on oak in Yarmouk reserve are reported for the first time. *Gnomonia* causes a leaf spot disease called oak anthracnose. The disease is common but not harmful to oak. Anthracnose can cause complete defoliation and it is most dangerous to white oak trees. *Ceratocystis* causes oak wilt disease that is characterized by necrotic leaves. The fungus is spread through grafting and by insects that feed on tree sap and infects the xylem tissues and kill the foliage [42].

Hypoxylon canker is reported for the first time on oak in Ajloun reserve in this study. The disease is most often associated with trees that are under environmental stress such as drought or mechanical damage.

Fusarium spp. was isolated from pines and junipers and is reported for the first time on these trees. The species *circinatum* is usually associated with pine pitch canker disease and is among the most devastating pine diseases in the world [43]. In Jordan, *Fusarium oxysporum* was reported on *Pinus pinea* in 2021 and caused wilting and damping-off symptoms in 50% of *P. pinea* seedling nurseries [44].

Taphrina caerulescens was identified in Yarmouk reserve on oak trees *Q. ithaburensis* with almost 100% incidence. The disease was previously reported in the checklist of Mamlouk et al., 1984 [39].

Depending on the underlying cause, many disorders are managed differently. Cultural activities, including pruning and removing infected leaves and branches, are quite beneficial and effective, though. Additionally, fungicides that are both preventative and curative can be used in chemical applications.

Overall, soil of the four reserves contains a variety of saprotrophic organisms that decompose organic matter and their various products in appropriate quantities, in addition to free-living nematodes and mycorrhizal fungi. This is a good indication of the fertility of the soil in general. On the other hand, these soils contain pathogens such as *Fusarium* spp. The soil contains pathogens that may cause seed rot and seedling rot before or after emergence until late stages of growth. This might threaten the natural regeneration process of reservation trees, especially in some specific locations in Dibben coniferous reserves. Despite the presence of wilt and spot diseases in Dibbeen, Ajloun, and Yarmouk reserves, they are considered in relatively good condition because these trees have a high leaf density and can continue to grow well in general. But this does not prevent monitoring and managing the newly grown trees. There is a serious infection of wilt diseases in the Phoenician juniper in Dana reserve, specifically zone A, numerous trees were dead. This infection stopped the natural renewal of trees permanently. The fluctuation in the rainfall rate during successive years may be a reason for enhancing the incidence of some diseases.

4.5. Limitations

Despite the fact that this study yielded substantial information regarding the condition of the forest, including aspects such as soil (respiration, microorganisms, etc.) and plant health (physiology, regeneration, etc.), further information is necessary, specifically regarding the structure and diversity of the forest community. While the evaluation of tree physiology and soil parameters is important in assessing the health of a forest ecosystem, indicators of forest community structure and biodiversity can provide valuable insights into the forest's health status.

5. Conclusions

This study measured several plant and soil variables to evaluate the forest health in four Jordanian forest reserves. These forests are grown in different bio-ecological zones including Mediterranean, Irano-Turanian, and Sudanian (sub-tropical). The results revealed that the fluctuation of rainfall and human activities (grazing, hiking) negatively impacted tree and soil health indicators. The low rainfall in 2021 (81 mm) at Dana Biosphere Reserve resulted in an extreme high Ψsmd values in *Juniperus phoenicea* (−4.4 to −5.3 MPa), low regeneration and lichens and the presence of soil borne pathogens including wilt diseases. In Dibbeen Forest reserve, the main threaten factor was human activities. At hiking zone, soil health indicators (respiration) as well as regeneration and lichens were significantly lower than the restricted zones. In Yarmouk Forest Reserve, high grazing intensity resulted in extreme reduction in soil respiration, seedling regeneration and lichens. In term of forest tree species, the native species *Quercus ithaburensis* and *Ceratonia silique* showed better water relations and gas exchange performance than the introduced species (*Pinus halepensis*) at Yarmouk Reserve. But, at Dibbeen reserve where *Pinus halepensis* is considered a native species, the performance was excellent. Across the forest reserves, *Arbutus andrachne* in Dibbeen and *Juniperus phoenicea* in Dana were the most stressed species based on water relations (Ψsmd, RWC) and gas exchange component. Overall, human activities (grazing and hiking) as well as the frequent drought period due to climate change are detrimental damaging factors to Jordanian forests. Urgent interventions are required to sustain several native historical forest trees including *Juniperus phoenicea* (>600-year-old), *Quercus ithaburensis* (>500 year old), and *Pinus halepensis* (>100 year old). Future work using, remote sensing can aid in forest health assessment. Moreover, a strategic plan for forest health is required for at least the next 20 years in order to increase forest health, help forests in adapting to climatic change, and understand the ecological, economic, and social benefits associated with forests.

Author Contributions: Conceptualization, K.M.A., Y.A.O. and M.M.T.; methodology, K.M.A., Y.A.O., M.M.T. and H.A.-D. and A.A.Y.; validation, K.M.A., Y.A.O. and M.M.T.; formal analysis, K.M.A., Y.A.O.; investigation, K.M.A., Y.A.O., M.M.T. and H.A.-D. and A.A.Y.; resources, B.A., T.A.-S., S.A., R.A.-Z., M.A., S.M. and M.B.A.; data curation, K.M.A. and Y.A.O.; writing—original draft preparation, K.M.A., Y.A.O.; writing—review and editing, K.M.A., Y.A.O. and M.M.T.; visualization, K.M.A., Y.A.O., M.M.T. and H.A.-D.; supervision, K.M.A., Y.A.O. and M.M.T.; project administration, K.M.A., Y.A.O., M.M.T., H.A.-D., A.A.Y., B.A., T.A.-S., S.A., R.A.-Z., M.A., S.M. and M.B.A. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The teamwork would like to thank the continuous support of all colleagues, both in the field and in the office, and appreciate their continuous help in contributing to the development this survey. We are grateful to Nashat Hamidan the director of Nature Conservation Monitoring Center, for his support and continuous help. Special thanks to Basheer Ayasrah the director of Dibbeen Forest Reserve, Mohammad Malkawy the director of Yarmouk Forest Reserve, Othman Tawalbeh the director of Ajloun Forest Reserve, Amer Roufo the director of Dana Biosphere Reserve for their unlimited support and logistic facilities and hospitality. The team would like to thank all other staff in all reserves and sites who help us during the fieldwork and special thanks to Jalal J. Alassaf for his help as well.

Conflicts of Interest: The authors declare no conflict of interest.

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