

Characterization of soil, vegetation, and soil fungal community in Cheongsu Gotjawal Forest

Jong-Shik Kim^{1,†} · Dae-Shin Kim²

*Marine Industry Research Institute for East sea rim, Uljin 36315, Republic of Korea
World Heritage Office, Jeju Special Self-Governing Provincial Government,
Jeju 63341, Republic of Korea*

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Abstract : Gotjawal, Jeju Island, Korea is a lava-formed forest with low soil content that is unique in the world. Around 6 percent of the Jeju land is classified as a lava-based specific forest. The forest has been gradually disappearing during the last several decades, with approximately one half having been destroyed. Our study undertakes a detailed analysis of the landscape of the lava subsidence, and describes the fungi, vegetation, and soils of Cheongsu Gotjawal. Soil samples from the Gotjawal were collected, and soil analyses as well as pyrosequencing of the internal transcribed spacer gene for fungal communities were performed. Soil fungal communities are represented by *Discisedars*, *Fusarium*, *Pleochaeta*, and *Fuscoporia* genera. Endemic vegetation of the Gotjawal includes the plants *Pteris critical*, *Machilus japonica*, *Quercus glauca*, *Arachniodes aristata*, and *Neocheiropteris ensata*. Results of soil analysis indicate sandy loam with 31.70% organic matter, and 1.36 mg/kg of total nitrogen. This fundamental information can help understand the invaluable and unique nature of Cheongsu Gotjawal, and the necessity for more studies on Gotjawal.

Keywords : Soil, Fungi, Vegetation, Gotjawal, Cheongsu Gotjawal forest

1. Introduction

Gotjawal is a low-soil forest that is unique in the world [1, 2, 3]. Representative Gotjawal microbial diversity was analyzed; in addition to analyzing soil microbial diversity, soil characterization was elucidated for the first time [4]. The study showed high values for organic matter (OM), NO₃, iron, and Al concentrations.

Hankyeong-Andeok (HA) Gotjawal is located in the southwest region of Jeju at 70–350 m above sea level (a.s.l.), and was formed by Pahoehoe Lava. Cheongsu Gotjawal is located in the center of HA Gotjawal (Fig. 1), and exhibits a topography involving discontinuously formed valleys which is the result of the lava flowing one lobe at a time (Fig. 2).

Currently, some areas of the Gotjawal are protected, but approximately 50% of the original forests have suffered losses from deforestation, and have been replaced with agriculture, and urbanization [5]. Gotjawal

[†]Corresponding author
(E-mail: soilmicrobiome@gmail.com)

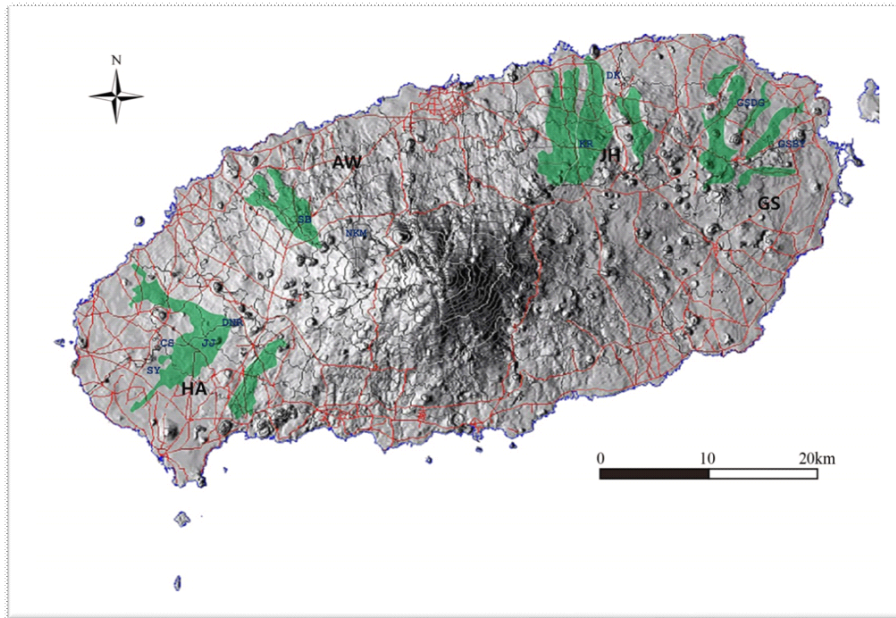


Fig. 1. Cheongsu Gotjawal forest on the Jeju map and sampling site (blue arrow).

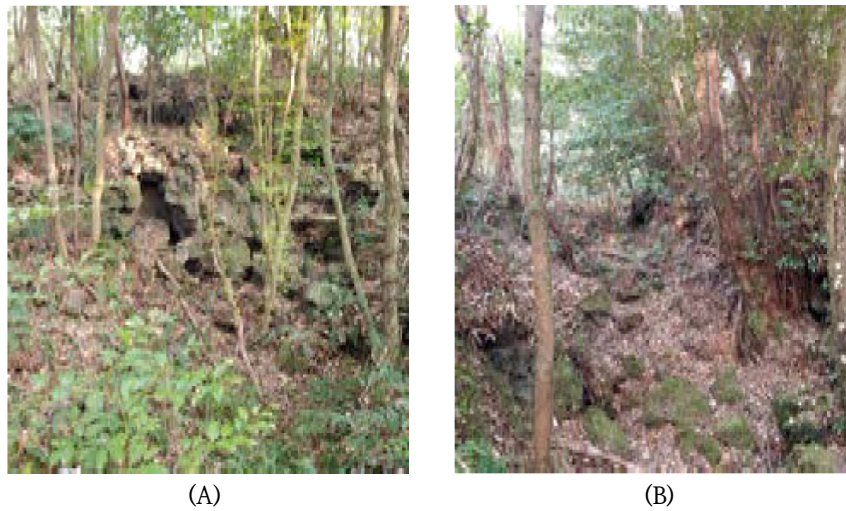


Fig. 2. Photographs showing the Cheongsu Gotjawals forest in Jeju, Korea (A: CS1, B: CS2).

soils are considered highly fertile due to their high OM and nitrogen content, but there is relatively limited knowledge about their microbiology [6–13]. Soil fungi can play important roles as biological controllers,

ecosystem regulators, and species decomposing in organic matter and participating compound transformations. Fungi are the major part of microbial biomass in soil, so they represent a great store of nutrients for potential life

growth and remain in soil through diverse mechanisms including spores and resting structures [14–16].

The aim of the present study was (1) to analyze the fungal diversity for the first time; (2) to prepare vegetation; and (3) to evaluate soil properties of Cheongsu Gotjawal in HA Gotjawal.

2. Materials and Methods

2.1. Soil sampling and DNA extraction

Samples were collected at Cheongsu Gotjawal (N33, 18, 19.7° , E126. 15, 44.9°) in October 2018 (Fig. 1). At each sampling location, samples collected from beneath the lava and tree roots as well as between them were considered as one sample; additionally, six mixed subsamples were also collected. The two samples at each sampling point were collected at distances of 10 m to 30 m in the field. Samples were collected aseptically using ethanol-disinfected shovels, and were placed in clean, sealable plastic bags. Sampled soils were sieved using 2-mm mesh sieves, which were stored in a 4 °C cooler during transfer to a laboratory (Uljin, South Korea).

2.2. ITS gene pyrosequencing

DNA from the soil was extracted using a FastDNA SPIN Kit for soil (QBiogene Inc., Vista, CA), for ITS gene sequencing. The fungal ITS2 gene amplicons were generated and pyro sequenced using a Roche 454 GS-FLX with titanium reagents, as per the manufacturer's instructions (Chunlab Inc, Seoul, South Korea). The sequencing data were analyzed by DNA link (Seoul, Korea). The sequence data were deposited in the National Center for Biotechnology Information (NCBI) Sequence Read Archive under BioSample accession numbers SAMN15366406 (soil CS1) and SAMN15366407 (soil CS2; BioProject ID PRJNA612949).

2.3. Soil analysis

Soil properties were measured at the National Instrumentation Center for Environmental Management (NICEM, Seoul, Korea) based on standard Soil Science Society of America protocols (Table 1). The parameters analyzed were pH, electrical conductivity (EC), OM, total organic carbon content (TOC), total nitrogen content (T-N), NH_4^+ , NO_3^- , CEC (cation exchange capacity: Mg, Na, Ca, K), P_2O_5 , clay, silt, and sand (Table 1).

2.4. Topography

Cheongsu Gotjawal CS1 is L-shaped, with a topography similar to a valley. It is 8–18 m wide and 7–8 m deep. Gotjawal CS2 has a funnel-shaped topography, 5–10 m wide, and approximately 10 m deep.

3. Results and Discussion

From the FLX Gold sequence data, we examined a total of 140,716 reads from Cheongsu Gotjawal soils. The read count per sample was 2798 and 4078. The operational taxonomy units were 758 and 922, while the Shannon index was 6.93 and 7.88. Rarefaction curves were obtained with most samples approaching a plateau.

For the CS1 and CS2 samples, the Shannon index values were 6.93 and 7.88, the evenness values were 0.72 and 0.83, and the Chao1 values were 2347.88 and 1424.32, respectively.

Species analysis showed representative values of fungi *Discisedars*, *Fusarium*, *Pleochaeta*, and *Fuscoporia* genera to be 26.9%, 5.7%, 4.0%, and 3.57%, respectively, in sample CS1, while fungi *Fusarium*, *Pleochaeta*, *Discisedars*, and *Fuscoporia* genera represented 13.6%, 9.39%, 3.8%, and 3.11%, respectively, in sample CS2 (Fig. 3, Fig. 4).

The results indicate 5, 5.3 of pH, 30.86, 31.70% of OM, 1.35, 1.37% of TN, 203, 315 mg/kg of ammonia, 57.13, 76.61 mg/kg of

Table 1. Soil characteristics and diversity index in Cheongsu Gotjawal. Two samples were statistically significant ($p < 0.05$)

Gotjawal		CS1	CS2
pH		5	5.3
EC	dS/m	0.79	1.03
OM	%	30.86	31.7
TN	%	1.35	1.37
NH ₄ (+)	mg/kg	315	203.01
NO ₃ (-)	mg/kg	57.13	76.61
CEC	cmol/kg	66.32	77.32
Exch. Cation (mg/kg)	Ca	3737.07	4380.46
	Mg	868.86	1005.47
	K	255.82	329.39
	Na	96.6	154.48
Avail. P ₂ O ₅	mg/kg	67.71	38.89
Soil texture		sandy loam	sandy loam
Sand	%	60.04	79.08
Silt		22.00	6.64
Clay		17.96	14.28
Chao1	Diversity	2347.88	1424.32
Shannon		6.93	7.88
Simpson		0.96	0.98
OTU		758	922

NO₃, 3737.07, 4380.46 mg/kg of calcium, and 67.71, 38.89 mg/kg of available P; both samples are sandy loam soil with high concentration of OM. Our group elucidated microbial diversity [1, 2, 8] including new species and new genera of microbes recovered so far [3–7]; some of these microbes are not found in other soils, and only specific microbes were dominant in Gotjawal (unpublished data). Thus, the Gotjawal soil ecosystem is found to possess a very unique microbial diversity compared to soil ecosystems that were studied earlier. Particularly, Cheongsu Gotjawal exhibited high OM and T–N concentration (Table 1).

The vegetation of this area is a *Quercus glauca*-dominated tree layer on the upper lava lobe, with the slope consisting of a low-tree layer with sparse growth of *Machilus japonica*

and *Acer palmatum* among others. At the herbaceous layer, *Arachniodes aristata* dominates the upper area of the lava lobe, a mixture of *Neochiropteris ensata* and *Arachniodes aristata* dominates the middle to lower areas, and *Pteris cretica* dominates the bottom.

Gotjawal CS2 has a funnel-shaped topography with a smaller size than Gotjawal CS1 at 5–10 m wide and approximately 10 m deep. The vegetation is a *Quercus glauca* dominated tree layer at the upper areas of the lava lobe with a rare growth of *Machilus japonica* on the slope. At the herbaceous layer, *Arachniodes aristata* dominates upper to middle areas of the lava lobe while *Neochiropteris ensata* dominates the middle to bottom areas (Fig. 2).

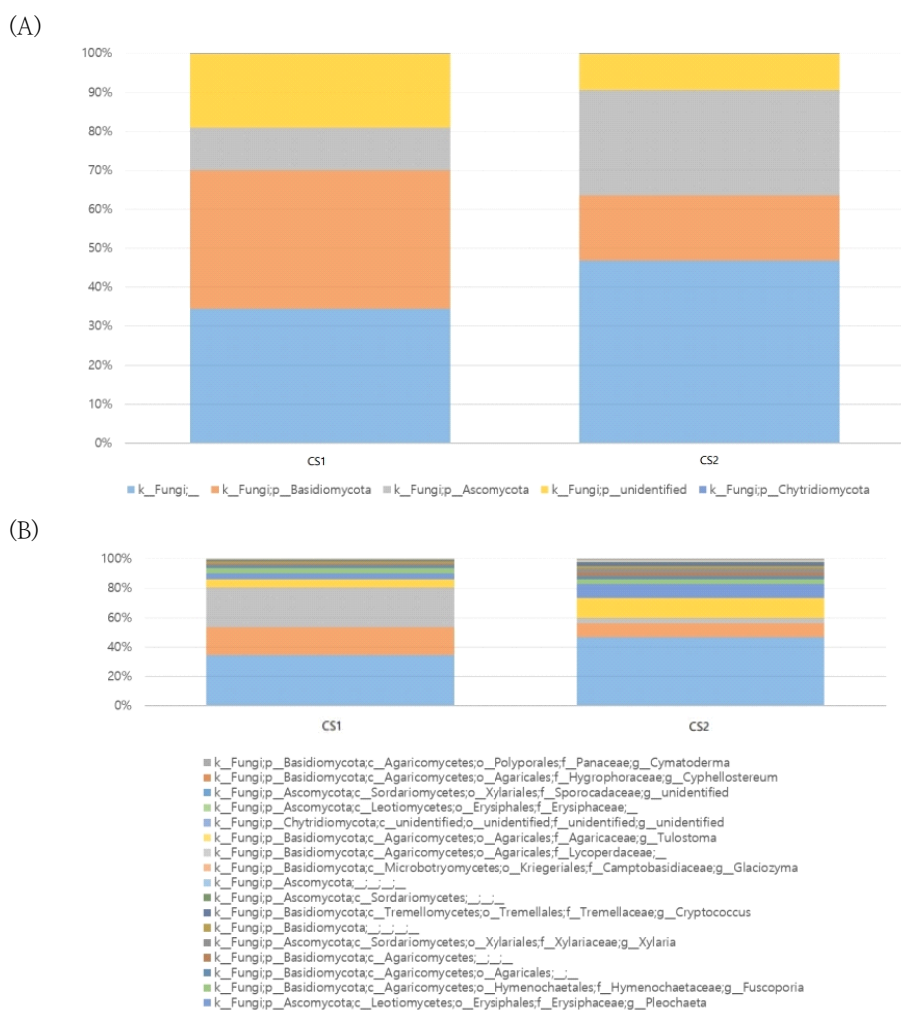


Fig. 3. Fungal Taxonomy (A: Phylum, B: genera) of the Cheongsu Gotjawals (CS1, CS2).

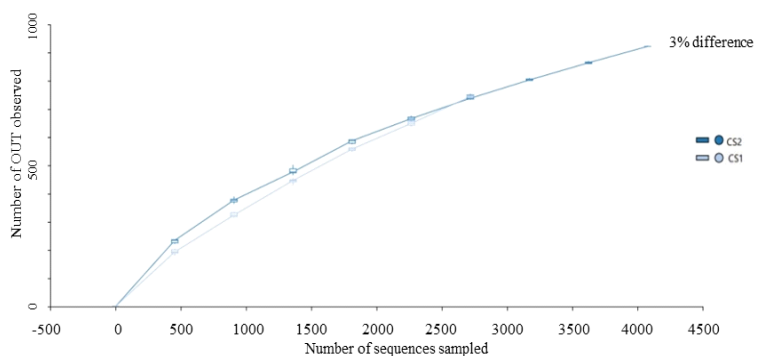


Fig. 4. Rarefaction curve of FLX-pyrosequences indicating the observed number of OTUs in the Cheongsu Gotjawal forest soil.

4. Conclusions

The results describe the Cheongsu Gotjawal forest and its fungal community, with plant and soil characteristics analyzed for the first time. Lee et al. [8] found that there is an unexpected unique nitrogen fixer in the soils of Gotjawal forest compared with that of other forests. With limited soil, how best to support the forest to form and maintain on the lava rocks and to be a self-supporting system remains to be answered. Further studies are required to help protect and conserve this invaluable forest.

There are some limitations to this study. The data gathered and the analyses carried out are not sufficient to compare different areas of the Gotjawal forest. Nevertheless, this study describes the fungal community of Cheongsu Gotjawal for the first time, suggesting a previously undescribed 'Gotjawal soil' with low soil content, high OM (over 30%) and TN concentration, and the dominant vegetation of Cheongsu Gotjawal.

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