A Note on the Lichen Genus *Ramalina* (Ramalinaceae, Ascomycota) in the Hengduan Mountains in China

Soon-Ok Oh¹, Xin Yu Wang², Li Song Wang², Pei Gui Liu² and Jae-Seoun Hur^{1,*}

¹Korean Lichen Research Institute, Sunchon National University, Suncheon 540-742, Korea ²Key Laboratory of Biodiversity and Biogeography, Kunming Institute of Botany, Chinese Academy of Sciences, Heilongtan, Kunming, Yunnan 650204, China

Abstract On the basis of extensive field investigation and a series of herbarium specimen identifications, we present and discuss the descriptions and distribution of 22 species of *Ramalina* found in the Hengduan Mountains of southwestern China. In this revisionary study, representatives of the *Ramalina* genus, including *R. americana, R. confirmata, R. dendriscoides, R. obtusata, R. pacifica, R. pentecostii, R. peruviana, R. shinanoana,* and *R. subcomplanata* are found for the first time in this area. In addition, *R. holstii* is reported for the first time China. Finally, a newly described species identified as *Ramalina hengduanshanensis* S. O. Oh & L. S. Wang is reported. It is characterized as growing from a narrow holdfast, solid, sparsely or richly and irregularly dichotomously branched, palmate and flattened lobes with distinctly dorsiventral appearance, surface rugose to reticulate, surface rugosely cracked, dense chondroid tissue, helmet shaped soralia at the tip. The species grows on rock and tree at the highest elevations in this area. Although very few lichen species belonging to the genus *Ramalina* have been collected above 4,000 m, this new species is found at this elevation. We present detailed morphological, anatomical, and chemical descriptions of this species along with molecular phylogenetic analysis of the internal transcribed spacer rDNA sequences.

Keywords Hengduanshan, Key, Molecular phylogeny, New species

The Hengduan Mountains are the biggest series of parallel north-south ranges in China, extending from Sichuan and Yunnan provinces to eastern Tibet, lying between $22^{\circ} \sim 32^{\circ}05'$ N and $97^{\circ} \sim 103^{\circ}$ E. Alternating high ridges and deep valleys range from 2,000 to 6,000 m above the sea level. The genus *Ramalina* belongs to the family Ramalinaceae and is a widespread fruticose lichen genus containing about 200 species worldwide, with about 50 taxa described to date in China [1]. The genus *Ramalina* has been studied in the Northern Hemisphere for over two hundred years and

Mycobiology 2014 September, **42**(3): 229-240 http://dx.doi.org/10.5941/MYCO.2014.42.3.229 plSSN 1229-8093 • elSSN 2092-9323 © The Korean Society of Mycology

*Corresponding author E-mail: jshur1@sunchon.ac.kr

Received May 5, 2014 Revised July 10, 2014 Accepted August 7, 2014

© This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http:// creativecommons.org/licenses/by-nc/3.0/) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

several hundred taxa have been named [2]. In the Southern Hemisphere, by contrast, very little research has been undertaken. The genus Ramalina was first described by Acharius when it was distinguished from Parmelia by its cartilaginous thallus and apothecial characteristics [3]. It was later subjected to monographic treatment on a global basis by Nylander [4]. Although several groups within the genus have been revised by Kashiwadani and other lichenologists [5-11], certain species found in Japan and its neighboring countries still require more precise study. Recently, Kashiwadani et al. [12, 13] have described seven new taxa, including R. almquistii Vain., R. hokkaidensis Kashiw., R. intermedia (Delise ex Nyl.) Nyl., R. obtusata (Arnold) Bitt., R. pertusa Kashiw., Ramalina pentecostii Krog & Swinsc., and R. sekika from China, and in addition have revised the genus Ramalina in Taiwan. Zhao et al. [14] described two new species, R. americana Hale. and R. aspera Räsänen Ann. from the Qinling Mountains in China. Ohmura et al. [15] revised three species, *R. pollinaria* (Westr.) Ach., R. sekika Asahina, and R. yasudae Räsänen based on morphological and chemical characteristics as well as a molecular phylogenetic analysis using internal transcribed spacer (ITS) rDNA sequences. Since 1991, there have been few or no studies pertinent to China, and no revisional work had been carried out in the Hengduan Mountains before the present study. As a result, many taxonomic and

nomenclatural problems remain in the description of the *Ramalina* genus in China. Therefore, the purpose of the present study is to revise the taxonomy of the *Ramalina* species found in the Hengduan Mountains.

MATERIALS AND METHODS

Characterization of phenotype. A total of 354 specimens were collected from Sichuan, Tibet, and Yunnan provinces of China between 1980 and 2010 and stored at the Cryptogamic Herbarium of the Kunming Institute of Botany (HKAS) at the Chinese Academy of Sciences. Description of the external morphology was based on air-dried material observed under a dissecting microscope (C-PS 1068908; Nikon, Tokyo, Japan). To evaluate anatomical variation within the thallus and the apothecia, hand-cut sections were prepared. The anatomy of the specimens was examined with standard light microscopic procedures (BX 50F4; Olympus, Tokyo, Japan). The microcrystallization and color tests of lichen substances were conducted as described previously [16]. Thin-layer chromatography (TLC) was carried out by a standardized method [17] using a DC Platten Kieselgel 60 F254 instrument (Merck, Darmstadt, Germany) and a three solvent system: Solvent A (benzene : dioxane : acetic acid = 180:45:5), Solvent B (hexane: methyl tertbutyl ether : formic acid = 140 : 72 : 18), Solvent C (toluene : acetic acid = 200:30).

Molecular phylogeny. Sequences of the ITS were amplified from selected specimens of Ramalina and a total of 17 new sequences were generated for this study. DNA was extracted using the QIAGEN DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). Polymerase chain reaction (PCR) amplification of the ITS1, ITS2, and 5.8S rDNA region was performed using the previously described primer sets ITS4 [18] and ITS5 [18]. Amplifications were performed using the Amplitaq DNA polymerase with buffer conditions recommended by the manufacturer and the following reaction parameters: 30 cycles of 45 sec denaturation at 94°C, 45 sec annealing at 55°C, and 1 min extension at 72°C, followed by 1 cycle of 5 min extension at 72°C. The PCR products were electrophoresed on an agarose gel to verify product size, and the remaining product was then purified using the PCR Purification Kit (Qiagen) and was sequenced.

The sequences obtained from each sample were aligned with selected sequences of *Ramalina* from GenBank, by using *Usnea diffracta* (DQ232663, DQ394374) as the outgroup. Details of the GenBank accession numbers are presented in Table 1. Alignments for each sequence were assembled separately in BIOEDIT 7.0.9 [19] and the sequences were initially aligned using CLUSTALX 1.83 [20]. The ITS rDNA phylogenetic analysis was conducted using neighbor-joining (NJ), maximum parsimony (MP), and maximum likelihood (ML) methods in MEGA ver. 5 [21]. For the NJ analysis, the Tamura-Nei model was used.

Ta	ble	 Vouche 	er specimens	and Ger	nBank acces	ssion n	um	bers
of	the	internal	transcribed	spacer	sequences	used	in	the
ph	yloge	enetic ana	lysis					

phylogenetic unarysis	Species and herbarium	GenBank
Lichen species	accession No.	accession No.
Usnea diffracta	_	DQ232663
Usnea diffracta	-	DQ394374
Ramalina americana	-	AF109235
R. americana	-	AF109238
R. aspera	06-26100	JF923601
R. aspera	06-26284	HQ845884
R. aspera	KoLRI No. 003908	JF937044
R. aspera	KoLRI No. 004051	JF937043
R. calicaris	KoLRI No. 003686	JF937045
R. calicaris	KoLRI No. 003714	JF937042
R. conduplicans	06-26202	JF923600
R. conduplicans	-	AB362789
R. conduplicans	-	DQ394391
R. confirmata	06-26195	JF923609
R. confirmata	07-28833	JF923606
R. fraxinea	-	AF249907
R. fraxinea	-	AY462054
R. hengduanshanensis	02-21466	JF923596
R. pollinaria	07-29001	JF923612
R. pollinaria	-	AB362794
R. pollinaria	-	AB362795
R. pollinaria	-	EF432560
R. sinensis	06-26194	JF923611
R. sinensis	KoLRI No. 003711	JF937050
R. sinensis	KoLRI No. 003747	JF937053
R. sinensis	KoLRI No. 003780	JF937051
R. sinensis	KoLRI No. 003903	JF937052
R. sinensis	KoLRI No. 004049	JF937049
R. sinensis	-	AB362797
R. yasudae	-	AB362799
R. yasudae	-	AB362800
R. yasudae	-	AB362801
R. yasudae	-	AB362802
R. yasudae	-	AB362803
R. yasudae	-	AB362804
R. yasudae	-	AB362809
R. yasudae	-	AB362810
R. yasudae	-	AB362811
R. yasudae	-	AB362812
R. yasudae	-	DQ394394
R. yasudae	-	EF544573

Accession numbers in bold font represent new sequences generated in this study.

One thousand bootstrap replications were performed to assess confidence values for each tree. The MP analysis was obtained using the subtree pruning regrafting method with a search level of 1, in which the initial trees were obtained with the random addition of sequences (10 replicates). All positions containing gaps and missing data were eliminated from the dataset (Complete Deletion option). The ML analysis was performed using parameters that included uniform rates among sites, the nearest neighbor interchange heuristic method, and all gaps being treated as missing data.

RESULTS AND DISCUSSION

The newly derived sequence of Ramalina hengduanshanensis

was aligned with sequences obtained from GenBank as listed in Table 1. The ML tree based on the *Ramalina* sequences is shown in Fig. 1. The operational taxonomic units (OTUs) of all species formed a monophyletic clade (>95% in ML, NJ, and MP). The *R. pollinaria* clade was not related to the OTUs of *R. yasudae*, suggesting that *R.*

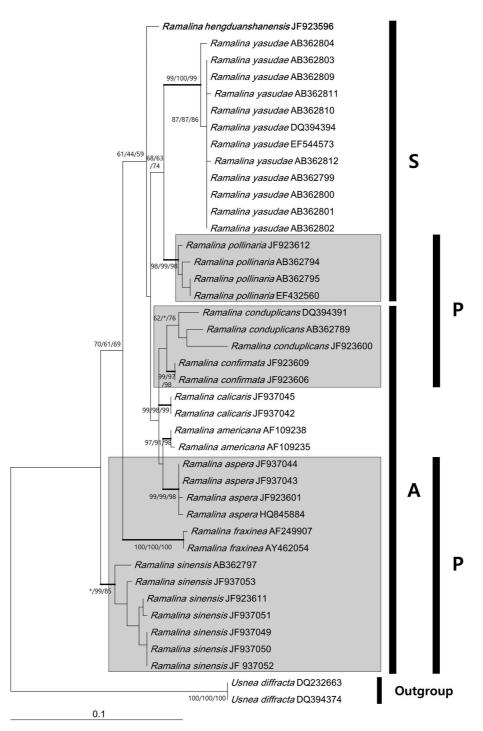


Fig. 1. Phylogeny of *Ramalina* species as inferred from rDNA analysis to investigate the phylogenetic placement of *R. hengduanshanensis*. Maximum likelihood tree obtained using the program MEGA ver. 5. Thick branches indicate bootstrap support >95% and support values given (bootstrap values from maximum likelihood neighbor-joining maximum parsimony). Characters are indicated as follows: A, apothecial clade; S, soralial clade; P, pseudocyphellae clade.

232 Oh et al.

pollinaria and R. yasudae should be treated as different species [15]. R. hengduanshanensis resembles R. sinensis in having similar branches. However, it can be distinguished from the latter species by the absence of soralia. R. hengduanshanensis also resembles R. pollinaria upon molecular analysis, but it can be distinguished from the latter species by the sublabriform powdery soralia at the apices of the branches. Neither the R. pollinaria clade nor the R. sinensis clade was related to the OTUs of R. hengduanshanensis. The support value for the branching of the clade, including for R. hengduanshanensis and the other species, is relatively lower compared to that of other monophyletic clades (61%, 44%, and 59% by ML, NJ, and MP, respectively). The monophyly of other species could not be confirmed within the clade. Hence, R. hengduanshanensis was determined to be a different species by both morphological and molecular phylogenetic characteristics, and therefore, is reported as such.

Key to the *Ramalina* species in the Hengduan Mountains in China

1. Thallus hollow 2
1a. Thallus solid 3
2. Saxicolous, soredia absent R. almquistii
2a. Corticolous, soredia present R. obtusata
3. Soredia present 4
3a. Soredia absent 14
4. Pseudocyphellae absent R. hengduanshanensis
4a. Pseudocyphellae present 5
5. Marginal, linear pseudocyphellae
5a. Punctiform, ellipsoid, rounded pseudocyphellae 7
6. Cracked chondroid tissue, soralia parietal, marginally in
cracks between the upper and lower cortex R. pentecostii
6a. Not cracked chondroid tissue R. pacifica
7. Soralia very irregular in shape, at or close to the lobe
tips
7a. Soralia round, elliptical, or elongate; branch tips
tapering
8. Branches narrow or broad, 0.5~3 mm wide; soredia
mainly on the lower surface of the branch tips, not developing
within hood-shaped expansions; soralia subterminal,
terminal R. pollinaria
8a. Branches narrow, 1 mm wide; soredia terminal, laminal,
bilateral laciniae R. yasudae
9. Isidia-like branchlets 10
9a. Lacking isidia-like branchlets 11
10. Chondroid tissue cracked R. hossei
10a. Chondroid tissue not cracked R. peruviana
11. Soredia granular, usually concentrated at the branch tips 12
11a. Soredia marginal or laminal, not concentrated at the
branch tips
12. Common holdfast, shrubby, weakly canaliculated;
chondroid tissue not cracked <i>R. dendriscoides</i>
12a. Delimited holdfast; chondroid tissue cracked
R. intermedia

13. Soralia mainly elliptical farinose, on the branch margins,
and occasionally on the surface; branches mostly 0.5~3-
mm wide R. farinacea
13a. Soralia subgranular, laminal, subterminal; finely
dissected branchlets ending in nodules; branches mostly
1 mm wide R. shinanoana
14. Apothecia on the branch surface or margins 15
14a. Apothecia on or close to the tips of the branches $\cdots16$
15. Warts or tubercles abundant on the branches; branches
broad, up to 5 mm wide R. aspera
15a. Warts or tubercles abundant on the margin; Branches
narrow, up to 2 mm wide R. holstii
16. Branches canaliculated 17
16a. Branches with depressions and ridges, or with long
grooves, but without tubercles 19
17. Pseudocyphellae common, short linear pseudocyphellae
along branches, cracked chondroid tissue, spore curved
18
17a. Pseudocyphellae rare, chondroid tissue not cracked,
spore ellipsoid R. calicaris
18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>20</i> 20. Ellipsoid, short spores 12.5 μm long and 5 μm wide
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 20 20. Ellipsoid, short spores 12.5 μm long and 5 μm wide <i>R. americana</i> 20a. Broad spores 15 μm long and 6 μm wide <i>R. americana</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 20 20. Ellipsoid, short spores 12.5 μm long and 5 μm wide <i>R. americana</i> 20a. Broad spores 15 μm long and 6 μm wide <i>R. americana</i>
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide
 18. Narrow curved spore ca. 12.5 μm long and 3.8 μm wide <i>R. confirmata</i> 18a. Broad curved spore ca. 15 μm long and 5 μm wide <i>R. subcomplanata</i> 19. Laciniae 1~1.5 cm wide; palmately branched; small curved spores 5 μm long and 2.5 μm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 19a. Laciniae 1~5 mm wide <i>R. sinensis</i> 20 20. Ellipsoid, short spores 12.5 μm long and 5 μm wide <i>R. americana</i> 20a. Broad spores 15 μm long and 6 μm wide <i>R. americana</i>

The new species.

Ramalina hengduanshanensis S. O. Oh & L. S. Wang sp. nov. (Fig. 2)

MycoBank no.: MB 809857.

Type: Sichuan Prov., Xiangcheng County, Mt. Daxue, 12 Sep 2002, L. S. Wang, 18823 (KUN: holotype).

Brief description of morphology and anatomy: Habitat: corticolous, saxicolous, fruticose; Growth form: caespitose, growing from a narrow holdfast; Thallus: solid, sparsely, or richly and irregularly dichotomously branched, erect, palmate, and flattened lobes with distinctly dorsiventral appearance, surface rugose to reticulate, surface rugosely cracked, up to 1.5 mm wide, up to 3~5 cm high; Cortex: distinct, ca. 12.5 µm thick; Medulla: lacking, dense; Chondroid tissue: irregular in thickness, ca. 75 µm thick, not cracked; Pseudocyphellae: absent; Soralia: apical on lower side, helmet-shaped; Soredia: granular; Apothecia: not seen.

Chemistry: Medulla K-, C-. TLC: usnic acid

Etymology: The specific epithet "hengduanshanensis" refers to the Hengduan Mountains.

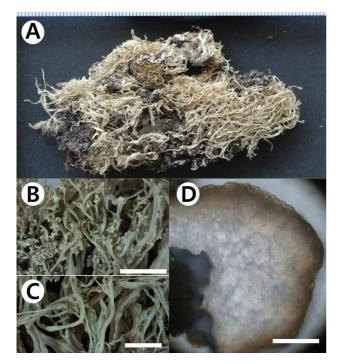


Fig. 2. Ramalina hengduanshanensis, holotype. A, Habitus; B, C, Thalli; D, Chondroid tissue (scale bars: A = 1 mm, B, C = $2 \text{ mm}, \text{ D} = 100 \text{ }\mu\text{m}$).

Remark: Ramalina hengduanshanensis resembles Ramalina sinensis in having similar branches. However, it can be distinguished from the latter species by the absence of soralia. R. hengduanshanensis also resembles R. pollinaria, although it can be distinguished from the latter species by the sublabriform powdery soralia at the apices of branches. Most lichens have specific requirements with regard to moisture, light, and temperature, effectively restricting their distribution range. In the Hengduan Mountains, the variation in altitude is from 2,000 m to almost 6,000 m. At about 2,000 m R. dendriscoides is found, which is another species mostly collected between 2,000 and 3,500 m, up to 4,000 m. From about 4,000 m upward, the new species R. hengduanshanensis is found. Since very few lichens have been collected above 4,000 m, the upper distribution limit for these species is not yet known.

Selected specimens examined: Sichuan Prov.: Kangding Co., Mt. Zheduo, on rock, elev. 4,000 m, 7 Sep 1996, L. S. Wang, 96-16339; Xiangcheng Co., Mt. Daxue, on bark of Larix sp., N 31°13', E 102°46', elev. 3,800 m, 2 Jun 2006, L. S. Wang, 06-26065; Tibet: Basu Co., Ranwu village, on bark of Juniperus sp., N 29°29', E 096°42', elev. 4,050 m, 19 Aug 2007, L. S. Wang, 07-28103.

Other Ramalina spp.

Ramalina almquistii Vain., Ark. Bot. 8: 17 (1909).

Brief description of morphology and anatomy: Thallus saxicolous, fruticose, shrubby and fistulose thallus forming indistinct holdfast at the base, absence of pseudocyphellae

and soredia, chondroid tissue not cracked, common apicalterminal apothecia, shortly fusiform ascospores are 18 µm long and 8 µm wide in size.

Chemistry: Medulla K-, C-. TLC: usnic acid, divaricatic acid.

Ecology and distribution: This species has been previously reported in China [13], Kuril Islands [22], Japan [23], and South Korea [24]. In China, it was found growing on rock in mountainous regions at altitudes ranging from 3,230 to 3,700 m.

Selected specimens examined: Yunnan Prov.: Huize Co., Dahai, on rock, elev. 3,700 m, L. S. Wang, 96-17149, L. S. Wang, 96-16702, L. S. Wang, 96-16721; Binchuan Co., Mt. Jizhu, Jinding, on rock, alt. 3,200 m, L. S. Wang, 02-21070 and on rock, elev. 3,230 m, L. S. Wang, 06-26143.

Ramalina americana Hale. Bryologist 81: 599 (1978).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, shrubby, delimited holdfast, solid, with strong ridges and channels, flat, side branches common, pseudocyphella rare, laminal and marginal, chondroid tissue not cracked, apothecia common, subterminal, ellipsoid ascospores are 12.5 µm long and 5 µm wide in size. Chemistry: Medulla K-, C-. TLC: usnic acid.

Ecology and distribution: This species has been previously reported in North America [25] and China [14] where it grows on bark in mountainous regions at altitudes ranging from 1,900 to 2,900 m.

Selected specimens examined: Yunnan Prov.: Gongshan Co., Binzhongluo, Zhongdi village, elev. 1,900 m, L. S. Wang, 05-24282; Chuxiong Co., Mt. Zixi, N 25°04', E 101°24', elev. 2,300 m, L. S. Wang, D. L. Niu, Luo Heng, 05-25253; Songming Co., A-ziving village, Shujie, elev. 2,200 m, L. S. Wang, Xin-yu Wang, Meng-meng Liang, 10-31340.

Remark: Small specimens of R. sinensis are difficult to distinguish from R. americana. The latter species is more abundantly branched and never produces fan-shaped thalli.

Ramalina aspera Räsänen Ann. Bot. Soc. Zool. Bot. Fenn. Vanamo 20: 5 (1944).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, sparingly branched, growing from a narrow holdfast, bilateral, flat, branches solid, pseudocyphellae common, laminal, raised, tuberculate, chondroid tissue not cracked, apothecia common, subterminal, straight or slightly curved, shortly fusiform ascospores are 13 µm long and 5 µm wide in size.

Chemistry: Medulla K-, C-. TLC: usnic acid, divaricatic acid.

Ecology and distribution: This species has been previously reported in North America [25] and China [14] where only two collections have been made on bark in mountainous regions at 3,750 m.

Selected specimens examined: Sichuan Prov.: Luquan Co., Mt. Jiaozixue, N 26°03', E 102°05', elev. 3,245 m, L. S. Wang, S. O. Oh, X. Y. Wang, 06-26284; Yunnan Prov.: **234** Oh *et al.*

Yunlong Co., Mt. Ziben, N 25°44', E 99°03', L. S. Wang, 00-18924; Luquan Co., Zhuanglong village, Mt. Jiaozixue, N 26°04', E 102°50', elev. 2,567 m, L. S. Wang, S. O. Oh, 06-26100; Lijiang Co., Mt. Laojun, N 25°59', E 99°53', elev. 2,955 m, L. S. Wang, S. O. Oh, CH050258.

Remark: *R. aspera* might be confused with *R. complanata*, which differs in having a more flattened surface of the branches with cracked chondroid tissue and in containing salazinic acid.

Ramalina calicaris (L.) Fr. Fries, Sched, Critic: 17 (1824).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, shrubby, growing from a delimited holdfast, branches solid, flat, channeled towards base, pseudocyphellae rare, lateral, marginal, tuberculate, chondroid tissue not cracked, apothecia common, terminal or subterminal, ellipsoid ascospores are $12.5 \,\mu$ m long and $5 \,\mu$ m wide in size.

Chemistry: Medulla K–, C–. TLC: usnic acid, homosekikaic acid, sekikaic acid.

Ecology and distribution: This species has been previously reported in Europe [26] and China [14] where it grows on bark at elevations between 1,350 and 3,500 m.

Selected specimens examined: Sichuan Prov.: Huili Co., Mt. Longzhou, on bark of *Rhododendron* sp., elev. 3,000~ 3,500 m, L. S. Wang, 96-18074 and 96-18053; Yunnan Prov.: Baoshan Co., Baihualin, Jinchanghe, Mt. Gaoligong, elev. 2,400 m, L. S. Wang, 03-22954; Dali Co., Mt. Cang, elev. 3,250 m, L. S. Wang, 01-20517 and N 25°42', E 100°06', elev. 3,460 m, L. S. Wang, CH050038 and N 25°41', E 100°06', elev. 2,925 m, L. S. Wang, CH050064; Lijiang Co., Mt. Yulong, S part, Yu-feng Temple, elev. 2,600~2,800 m, L. S. Wang, 87-46266; Shangrila Co., Geza village, Mt. Xiaoxue, N 28°30', E 99°49', elev. 3,000 m, L. S. Wang, 00-20041; Bingchuan Co., Mt. Jizu, elev. 3,200 m, L. S. Wang, 06-26199.

Remark: *R. calicaris* is a corticolous lichen similar to *R. elegans* and *R. fastigiata*. *R. calicaris* is morphologically similar to *R. fastigiata* but has a hollow thallus with a very irregular surface and many perforations. *R. calicaris* resembles *R. elegans*, from which it can be distinguished by its solid thallus, a regular, sulcate surface towards the base, and its mainly marginal apothecia.

Ramalina conduplicans Vain. Ann. Soc. Zool. Bot. Fenn. 1: 35 (1921).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, shrubby, caespitose, shrubby branches developing from a delimited holdfast, solid, forked branches ending with nodules surface flat to weakly reticulately ridged, pseudocyphellae abundant on lower side of branches, punctiform, chondroid tissue not cracked, apothecia common, laminal, fusiform ascospores are $15 \,\mu$ m long and $6 \,\mu$ m wide in size.

Chemistry: Medulla K–, C–. TLC: sekikaic acid, homosekikaic acid, salazinic acid.

Ecology and distribution: *R. conduplicans* is widely distributed in eastern Asia, having been collected in China [1], Japan [23], Korea [24], and the Himalayas [1]. In China, it was collected on bark at elevations between 1,200 and 3,814 m.

Selected specimens examined: Sichuan Prov.: Huili Co., Mt. Longzhou, on bark of *Rhododendron* sp., elev. 3,000~ 3,500 m, L. S. Wang, 97-17946; Yunnan Prov.: Baoshan Pref., Longlin Co., Mt. Liang, elev. 2,300 m, L. S. Wang, 06-26002; Chuxiong Co., Mt. Zixi, N 25°04', E 101°24', elev. 2,330 m, L. S. Wang, 05-25270; Dali Co., Mt. Cang, N 25°42', E 100°06', elev. 3,200~3,465 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26855; Dongchuan Co., Yaoshan village, Mt. Jiaozixue, elev. 2,322 m, L. S. Wang, 07-27781; Lijiang Pref., Mt. Tiejia, elev. 2,600 m, L. S. Wang, 85-0226; Yongde Co., Wumulong village, Mt. Daxue, elev. 3,500 m, L. S. Wang, 07-27631.

Remark: *R. conduplicans* is widely distributed in eastern Asia, having been collected in China, Japan, Korea, and Himalayas. This species is highly variable morphologically and chemically. The holotype of this species is a fragmental specimen having dorsiventral main lobes with sparse secondary branches tapering towards the apices. Most of the specimens from China have wide and canaliculated main branches.

Ramalina confirmata (Nyl.) Elix Lichenes Australasici Exsiccati 5: 219 (1990).

Brief description of morphology and anatomy: Habitat: corticolous, fruticose; Growth form: erect to subpendulous; Thallus: branching subdichotomous, sparse to moderate, branches compressed, narrow and canaliculated to broad and flat, apices attenuate or blunt, surface smooth to rugose, up to 1~5 mm wide, up to 6 cm high; Cortex: distinct, ca. 25μ m thick; Medulla: dense; Chondroid tissue: discontinuous, cracked, ca. 75μ m thick; Pseudocyphellae: short linear pseudocyphellae; Soralia: absent; Apothecia: common, marginal, subterminal, disc concave to plane, margin entire; Ascospores: ellipsoid, straight, or curved, 12.5 μ m long and 3.8 μ m wide.

Chemistry: Medulla K–, C–. TLC: usnic acid.

Ecology and distribution: This species has previously been reported in Australia [3]. In China, it was collected on bark of various species (*Quercus, Salix*) at elevations between 1,460 and 3,400 m.

Selected specimens examined: Sichuan Prov.: Dukou Co., Dabaoding, on bark of *Quercus* sp., elev. 2,000 m, L. S. Wang, 83-221; Tibet: Shan'an Qulin Jiebala, Mu Zang 88; Yunnan Prov.: Gongshan Co., Bingxhongluo, Mt. Songtaxue, N 28°09', E 98°33', elev. 2,500 m, L. S. Wang, 00-19205; Lijiang Co., Mt. Laojun, on bark of *Salix* sp., N 26°39', E 99°46', elev. 3,000 m, L. S. Wang, 05-25104; Pingbian Co., Mt. Dawei, N 22°58', E 103°42', elev. 1,530 m, L. S. Wang, 05-23909; Yunlong Co., Caojian village, Ziben Mt., on bark of *Salix* sp., N 27°44', E 99°03', elev. 3,000 m, L. S. Wang, 05-24390; Bingchuan Co., Mt. Jizu, on bark of *Quercus* sp.,

N 25°58', E 100°21', elev. 3,200 m, L. S. Wang, 06-26195.

Remark: *R. confirmata* closely resembles *R. calicaris*, though it can be distinguished from the latter species by strongly pseudocyphellate lobes and the distinctly cracked chondroid tissue.

Ramalina dendriscoides Nyl. Flora 59: 412 (1876).

Brief description of morphology and anatomy: Habitat: corticolous, fruticolous; Growth form: shrubby, growing from a restricted or common holdfast; Thallus: richly and irregularly branched, up to 3 cm high, solid, less than 1 mm wide, weakly canaliculated in main branches, sorediate; Cortex: distinct, ca. 10 μ m thick; Medulla: dense; Chondroid tissue: not cracked, discontinuous, dissected by the medullary hyphae, ca. 12.5~65 μ m thick; Pseudocyphellae: small, elliptic, flat; Soralia: terminal, apical, restricted to short, secondary branches; Soredia: granular; Apothecia: not seen.

Chemistry: Medulla K-, C-. TLC: salazinic acid.

Ecology and distribution: This species has been reported in Brazil [4] and East Africa [27]. In China, only two collections were made on bark at elevations between 1,840 and 2,450 m.

Selected specimens examined: Sichuan Prov.: Luding Co., Gongga Mt., N 29°20', E 101°30', elev. 2,450 m, L. S. Wang, 96-17327; Yunnan Prov.: Luchun Co., Fenshuiling, N 23°59', E 102°26', elev. 1,840 m, L. S. Wang, 05-23992.

Remark: *R. dendriscoides* has been confused with *R. peruviana*, a species with very similar subterete branches and mode of branching, with a primary difference being the lateral rather than mainly apical soralia. In addition, *R. peruviana* is known to produce sekikaic acid. *R. tenella*, a coastal species, also resembles *R. dendriscoides* in its apical, subcapitate soralia and in the production of salazinic acid, but it has shorter and distinctly flattened main branches.

Ramalina farinacea (L.) Ach. Lichenogr. Univers.: 606 (1810).

Brief description of morphology and anatomy: Habitat: corticolous, fruticose; Growth form: shrubby, polymorphic-tufted, subpendulous, irregularly or dichotomously branched from a narrow holdfast; Thallus: solid, greenish gray, smooth, shiny, sorediate, irregular in thickness or plane, $1{\sim}4$ mm wide, up to 3.5 cm high; Cortex: continuous, distinct, ca. 12.5 µm thick; Medulla: loose; Chondroid tissue: not continuous, weakly cracked or not cracked, irregular thickness of 40~90 µm; Pseudocyphellae: almost lacking in distal branches, more common near the base of branches, ellipsoid to short linear; Soralia: farinose, marginal, laminal; Soredia: rounded; Apothecia: rare.

Chemistry: Medulla K-, C-. TLC: usnic acid, sekikaic acid.

Ecology and distribution: This species is widely distributed in the world, having been reported in Chile [27], Europe [26], and North America [25]. In China, it was collected on the bark of various species (*Sorbus, Picea*) at elevations

Lichen Genus Ramalina in China 235

between 2,050 and 3,600 m.

Selected specimens examined: Sichuan Prov.: Tianquan Co., Mt. Erlang, Labahe, elev. 2,050 m, L. S. Wang, 06-26073; Yunnan Prov.: Zhongdian Co., Xiaozhongdian village, on bark of *Sorbus* sp., N 27°26', E 99°49', elev. 3,210 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26812; Gongshan Co., Binzhongluo to Tongda Yakou, on bark of *Picea* sp., N 28°04', E 98°41', elev., 600 m, L. S. Wang, 99-18557.

Remark: *R. farinacea* resembles *R. intermedia*, a welldefined species which can be clearly distinguished from other sorediate *Ramalina* species by its larger, broader lobed, and more abundant soredia.

Ramalina fraxinea (L.) Ach. Lich. Univ.: 602 (1810).

Brief description of morphology and anatomy: Habitat: corticolous, fruticose; Growth form: coarsely tufted, rarely monophyllous, subpendulous; Thallus: laciniae compressed, twisted, longitudinally and partly reticulate, variable in width, more or less irregularly tapered towards the base and the apices, planar or variously channeled, up to $1\sim$ 3 mm wide, more than 7 cm high; Cortex: distinct ca. 12.5 µm thick; Medulla: loose; Chondroid tissue: discontinuous, not cracked, ca. 50 µm thick; Pseudocyphellae: frequent, pale, rounded or ovate-elongate; Apothecia: usually frequent, marginal or laminal, disc concave to plane; Ascospore: ellipsoid, 15 µm long and 6.25 µm wide.

Chemistry: Medulla K-, C-. TLC: usnic acid.

Ecology and distribution: This species has been reported previously in Europe [26]. The distribution range now includes China where it grows on various barks (*Quercus, Pinus,* and *Rhododendron*) at elevations between 1,760 and 3,500 m.

Selected specimens examined: Sichuan Prov.: Huili Co., Mt. Longzhou, on bark of *Rhododendron* sp., elev. 3,000~ 3,500 m, L. S. Wang, 96-18036; Yunnan Prov.: Dali Co., Mt. Cang, on bark of *Quercus* sp., elev. 2,500 m, L. S. Wang, Y. Q. Xiao, 04-23450 and 04-23420, on bark of *Pinus armandii*, N 25°41', E 100°06', elev. 3,400 m, L. S. Wang, 05-24657 and 05-24658; Songming Co., Guo dong, Mt. Zhaobi, elev. 2,400 m, Y. G. Su, 11; Yuanyang Co., on bark of *Cinnamomum* sp., N 23°07', E 102°44', elev. 1,760 m, L. S. Wang, 05-23940.

Remark: The narrow-lobed forms of *R. fraxinea* can resemble *R. calicaris*. The latter species has smoother, less wrinkled lobes, with uncommon pseudocyphellae. The spores are straight, and are longer and wider than *R. calicaris*.

Ramalina holstii Krog & Swinsc. Sp. Nov. T. D. V. Nor J Bot 22: 275 (1975).

Brief description of morphology and anatomy: Habitat: corticolous, fruticose; Growth form: shrubby branches developing from a delimited holdfast; Thallus: greenish yellow, solid, up to 2 mm wide, up to $3\sim5$ cm high, flat or partially canaliculated; Cortex: distinct, $10\sim12.5$ µm thick; Medulla: loose; Chondroid tissue: not cracked, layer discontinuous, $20\sim70$ µm thick; Pseudocyphellae: predominantly marginal, tuberculate common; Apothecia: lateral, marginal, on geniculate branches, disc concave to flat, reticulately ridged, common; Hymenium: ca. 50 μ m thick; Subhymenium: ca. 12.5 \sim 17.5 μ m thick; Ascospore: fusiform, slightly curved, two-celled, 12.5 μ m long and 2.5 \sim 3.7 μ m wide.

Chemistry: Medulla K-, C-. TLC: salazinic acid.

Ecology and distribution: This species has been reported previously in East Africa [27]. In China, only two collections were made on bark at elevations between 2,000 and 3,800 m.

Selected specimens examined: Yunnan Prov.: Gongshan Co., Binzhongluo to Tongda Yakou, on bark of *Abies* sp., N 28°05', E 98°41', elev. 3,800 m, L. S. Wang, 99-18503; Lijiang Co., Shigu to Xionggu village, elev. 2,000 m, L. S. Wang, 09-30062.

Remark: *Ramalina* specimens with tuberculate pseudocyphellae and salazinic acid are often identified as *R. denticulata* (Eschw.) Nyl., described in Brazil [4]. The latter species has predominantly laminal pseudocyphellae and larger spores, $5 \sim 6 \mu m$ long and $15 \sim 17 \mu m$ wide, and is lacking in cryptochlorophaeic acid.

Ramalina hossei Vain. Ann. Soc. Zool-Bot. Fenn, Vanamo 1: 36 (1921).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, tufted, richly branched near the base, narrow branches growing from a common holdfast, solid, anisotomic, dichotomous or irregular, often with short side branches ending in granular soredia or tiny branchlets, pseudocyphellae common, orbicular, more or less raised, punctiform, granular soredia, chondroid tissue cracked, apothecia rare, laminal or submarginal, short fusiform ascospores of 15 μ m long and 5 μ m wide in size, slightly curved.

Chemistry: Medulla K–, C–. TLC: usnic acid, homosekikaic acid, sekikaic acid.

Ecology and distribution: This species has been previously reported in Taiwan [12] and Bhutan [8]. In China, it was collected on various barks (*Larix, Picea,* and *Quercus*) at elevations between 2,400 and 3,950 m.

Selected specimens examined: Sichuan Prov.: Daofu Co., Renguo village, on bark of *Larix* sp., elev. 3,950 m, L. S. Wang, 07-28327; Luding Co., Mt. Gongga, N 29°20', E 101°30', elev. 2,800 m, L. S. Wang, 96-17367 and on bark of *Picea* sp., N 29°20', E 101°30', elev. 2,450 m, L. S. Wang, 96-16159; Yunnan Prov.: Gongshan Co., Binzhongluo to Tongda Yakou, N 28°05', E 98°41', elev. 3,500 m, L. S. Wang, 99-18578; Lijiang Co., Mt. Yulong, on bark of *Quercus* sp., elev. 3,200 m, L. S. Wang, 01-20704; Zhongdian Co., Napa Lake, on bark of Bamboo, elev. 3,540 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26624; Bingchuan Co., Mt. Jizu, on bark of *Quercus* sp., N 25°58', E 100°21', elev. 3,200 m, L. S. Wang, 06-26201.

Remark: *R. hossei* apparently is closely related to *R. shinanoana* Kashiw., a species reported in Japan and China, because they both have similar branches and branchlets

with granular soredia and outgrowth, and short fusiform spores. However, it can be clearly separated from the latter species by its cracked chondroid tissue. The present species resembles *R. peruviana* Ach., a species widely distributed in the Pacific regions, from which it can be distinguished by the rather smooth surface of its branches, cracked chondroid tissue, and short fusiform spores. In *R. peruviana* the chondroid tissue is never cracked and spores are narrow fusiform [11].

Ramalina intermedia (Delise ex Nyl.) Nyl. Flora 56: 66 (1873).

Brief description of morphology and anatomy: Thallus saxicolous, corticolous, fruticose, growing from an delimited holdfast, sparingly branched, irregularly or dichotomously branched, solid, pseudocyphellae common, ellipsoid, often forming soredia. Granular soredia, chondroid tissue cracked, apothecia not seen in Chinese materials.

Chemistry: Medulla K–, C–. TLC: usnic acid, homosekikaic acid, sekikaic acid.

Ecology and distribution: This species has been previously reported in China [13, 14], Europe [26], South Korea [24], and the Kuril Islands [22]. In China, it was collected on various barks (*Abies, Rhododendron* etc.) at elevations between 1,900 and 4,000 m.

Selected specimens examined: Sichuan Prov.: Huili Co., Mt. Longzhou, elev. $3,000 \sim 3,500$ m, L. S. Wan, 97-17905; Yunnan Prov., Caojian Co., Mt. Ziben, L. S. Wang, 00-18886; Dali Co., Mt. Cang, elev. 3,400 m, L. S. Wang, Y. Q. Xiao, 04-23445; Deqen Co., Mt. Baimaxue, elev. 3,400 m, L. S. Wang, 93-13568 and on rock, elev. 3,400 m, L. S. Wang, 93-13436; Lijiang Co., Jiuhe village, Mt. Laojun, on bark of *Rhododendron* sp., N 26°39', E 99°46', elev. 3,450 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26468 and on bark of *Abies* sp., N 26°39', E 99°46', elev. 3,600 m, L. S. Wang, 05-24738; Zhongdian Co., Dabaosi, on bark of *Rhododendron* sp., N 27°46', E 99°46', elev. 3,650 m, L. S. Wang, 04-23367 and 04-23381.

Remark: *R. intermedia* might be confused with *R. pollinaria*, as they have similar branching pattern and soredia. However, *R. intermedia* can be easily distinguished from the latter species by its laciniae that develop from a narrow holdfast and by having homosekikaic and sekikaic acid as major chemical substances. *R. intermedia* resembles *R. exilis* in having similar branches with soredia. However, it can be distinguished from the latter species by the laminal and marginal soredia, the conspicuously cracked chondroid tissue below the cortex, and by the presence of homosekikaic and sekikaic acids.

Ramalina obtusata (Arnold) Bitt. Pingsheim Jahrb. Wiss. Bot. 36: 435 (1901).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, shrubby, shortly tufted, growing from a narrow holdfast, fistulose thallus, inflated and more or less pellucid thalli without side branches, consisting of

one single or several palmate or unorientated laciniae, pseudocyphellae absent, farinose soredia, soralia common, mainly developed within terminal or subterminal vesicles, labriform, helmet-shape, chondroid tissue not cracked, apothecia unknown.

Chemistry: Medulla K–, C–. TLC: obtusatic acid, evernic acid.

Ecology and distribution: This species has been previously reported in China [13], Europe [26], and North America [25]. In China, the only two collections were made on bark at elevations between 2,000 and 3,000 m.

Selected specimens examined: Sichuan Prov.: Nanping Co., Jiuzhaigou, on bark, elev. 2,000 m, L. S. Wang, 86-2525; Yunnan Prov., Binchuan Co., Mt. Jizhu, elev. 3,000 m, L. S. Wang, 96-15946.

Remark: *R. pollinaria* is very similar but does not develop inflated tips. *R. obtusata* can be distinguished by the soralia that are very irregular in size and shape appearing where the branch tips. *R. intermedia* is much more slender, dissolving into irregular sorediate patches near the tips.

Ramalina pacifica Asah. J. Jpn. Bot. 15: 213 (1939).

Brief description of morphology and anatomy: Habitat: corticolous, fruticose; Growth form: subpendulous to pendulous, tufted; Thallus: branching dichotomous, sparse to moderately dense, branches compressed, flat, solid, usually with longitudinal striae or cracks on the surface, sorediate, up to 1 mm wide, up to 4 cm high; Cortex: distinct, ca. 12.5 μ m thick; Medulla: dense; Chondroid tissue: under the cortex not continuous, not cracked, 25~60 μ m thick; Pseudocyphellae: linear pseudocyphellae slightly or strongly developed; Soralia: marginal, ellipsoid, well-delimited; Apothecia: rare, stipitate, apical; disc flat to convex; Hymenium: ca. 50 μ m thick; Subhymenium: ca. 25 μ m thick; Ascospore: fusiform or ellipsoid, 12.5 μ m long and 7.5 μ m wide.

Chemistry: Medulla K-, C-. TLC: usnic acid, salazinic acid.

Ecology and distribution: This species has been reported in Australian [3] and Japan [5]. In China, it was collected on various barks (*Quercus, Rhododendron*) at elevations between 400 and 3,300 m.

Selected specimens examined: Sichuan Prov.: Tongjiang Co., Fuyangqu Chenhe village, on branch of *Quercus* sp., elev. 400 m, L. S. Wang, 89-529 and L. S. Wang, 89-519 and X. J. Li, 56 and L. S. Wang, 89-524; Yunnan Prov.: Dali Pref., Huadianba, Ganchaiqing, on bark of *Rhododendron* sp., elev. 2,900 m, X. J. Xun, 372; Gongshan Co., Qinatong, Songtaxueshan, N 28°11', E 98°31', elev. 3,300 m, L. S. Wang, 00-19681.

Remark: *R. pacifica* may be confused with *R. intermediella* because both have similar sorediate branches and non-cracked chondroid tissues. However, this species has coarser thalli growing from a narrow holdfast and wide lobes. In addition, the soralia of this species is farinose and crateriform with limited margins.

Ramalina pentecostii Krog & Swinsc. Nor. J. Bot. 23: 167 (1976).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, pendulous, repeatedly dichotomously branched, branches stramineous, solid, pseudocyphellae common, marginal, linear pseudocyphellae, parietal soralia, erupting marginally in cracks between the upper and lower cortex, apices often recurved, chondroid tissue cracked, apothecia not seen.

Chemistry: Medulla K–, C–. TLC: no medullary substances.

Ecology and distribution: This species has previously been reported in China [13] and in East Africa [27]. In China it was collected on various barks (*Quercus, Rhododendron*) at an elevation of 3,300 m.

Selected specimens examined: Sichuan Prov.: Xiaojin Co., Shuangqiaogou, on trunk, elev. 3,300 m, L. S. Wang, 96-17717.

Remark: *R. pentecostii* may be confused with *R. disparate* because they both have similar soralia branches. It can be distinguished from the latter species in its parietal, mainly marginal soralia that never spread irregularly on to the lamina, and in the lack of medullary lichen substances.

Ramalina peruviana Ach. Lich. Univ.: 1599 (1810).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, subpendulous, tufted thallus growing from a common holdfast, solid, pseudocyphellae rare, narrowly elongated, marginal or laminal soralia with granular soredia and tiny branchlets, chondroid tissue not cracked, apothecia not seen in Chinese materials.

Chemistry: Medulla K–, C–. TLC: usnic acid, homosekikaic acid, sekikaic acid.

Ecology and distribution: This species has previously been reported in China [13], but is also widely distributed throughout the world, including reports from Australia [3], Brazil [4], Chile [28], East Africa [27], Indonesia [29], Japan [23], South Korea [24], and North America [25]. In China, it was collected on various barks (*Camellia, Rhododendron*) at elevations between 2,000 and 3,450 m.

Selected specimens examined: Sichuan Prov.: Hueili Co., Mt. Longzhou, on bark of *Rhododendron* sp., elev. 3,450 m, L. S. Wang, 97-24264; Miyi Co., Malong village, Mt. Beipo, elev. 2,800 m, L. S. Wang, 83-995; Yunnan Prov.: Lincang Co., Nouling village, on bark of *Camellia* sp., N 23°19', E 99°22', elev. 2,000 m, Cong-ren Yang, 05-33; Qiaojia Co., Dagiao, elev. 2,700 m, L. S. Wang, 96-16688.

Remark: *R. dendriscoides* Nyl. resembles *R. peruviana* in its mode of branching, but it differs in having most of its soralia situated apically on short lateral branchlets, and in containing salazinic acid.

Ramalina pollinaria (Westr.) Ach. Lich. Univ.: 608 (1810). **Brief description of morphology and anatomy:** Thallus corticolous, fruticose, pendulous, sparingly branched, growing from a narrow holdfast, solid, flattened or partly terete, especially towards the apices, extensively branched and dissected, with numerous small nodulose proliferations, palmate margins, irregular in thickness in cross section, pseudocyphellae common, ellipsoid, laminal, terminal soralia, labriform or developed in terminal helmet-shaped soralia with granular soredia, chondroid tissue not cracked, apothecia not seen.

Chemistry: Medulla K-, C-. TLC: usnic acid.

Ecology and distribution: This species has previously been reported in China [13], but is also widely distributed throughout the world, including reports from East Africa [26], Europe [27], the Kuril Islands [22], and North America [25]. In China, it was collected on various barks (*Quercus, Sorbus*) at elevations between 2,900 and 3,540 m.

Selected specimens examined: Sichuan Prov.: Kangding Co., Mt. Zhedou, on rock, N $30^{\circ}02'$, E $101^{\circ}49'$, elev. 3,800 m, L. S. Wang, 07-29001; Yunnan Prov., Dongchuan Co., Hongtudi village, on bark of *Quercus* sp., elev. 2,900 m, L. S. Wang, 09-30680; Luquan Co., Jiaozixue Mt., on bark of *Sorbus* sp., N $26^{\circ}03'$, E $102^{\circ}05'$, elev. 3,245 m, L. S. Wang, S. O. Oh, X. Y. Wang, 06-26304; Zhongdian Co., Napa Lake, on branch, N $27^{\circ}56'$, E $99^{\circ}36'$, elev. 3,540 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26646 and 06-26653.

Remark: *R. polymorpha* may also resemble *R. pollinaria*, but the former species has a roughened cortex, coarser soredia, and no medullary substance. Flattened specimens can be confused with *R. farinacea*, but the soralia of *R. pollinaria* are at least partly laminal and the medullary chemistry is different. *R. pollinaria* also resembles *R. chihuahuana* that differs in having marginal soralia and in producing divaricatic acid.

Ramalina shinanoana Kashiw. Bull. Natl. Sci. Mus. Ser. B Bot. 12: 122 (1986).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, shrubby branches growing from a narrow holdfast, solid, the finely dissected branchlets ending in nodules, pseudocyphellae common, ellipsoid, lateral, laminal soralia with subgranular soredia, chondroid tissue not cracked, apothecia rare, laminal, broadly fusiform ascospores are 15 μ m long and 6 μ m wide.

Chemistry: Medulla K–, C–. TLC: homosekikaic acid, sekikaic acid.

Ecology and distribution: This species has previously been reported in China [12], as well as in Indonesia [28] and Japan [24]. In China, it was collected on various barks (*Juniperus, Rhododendron* etc.) or rarely on rocks at elevations between 800 and 3,850 m.

Selected specimens examined: Sichuan Prov.: Kangding Co., Yajiagen, elev. 2,680 m, L. S. Wang, 10-31858; Yanyuan Co., Bailing village, Sidadui, L. S. Wang, 83-1383; Tibet: Motou Da Mu Ga lung, on rock, elev. 1,050 m, Yong-ge Su, 82-1957@; Yunnan Prov.: Deqen Co., Mt. Baimaxue, Yakou, on bark of *Juniperus* sp., N 28°19', E 99°06', elev. 3,980 m, L. S. Wang, 03-22758; Lijiang Co., Jiuhe village, Mt. Laojun, on bark of *Rhododendron* sp., N 26°39', E 99°46', elev. 3,450 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26479; Qiaojia Co., Dagiao, elev. 2,700 m, L. S. Wang, 96-16692; Zhongdian Co., Tianchi lake, N 27°37', E 99°38', elev. 3,500 m, L. S. Wang, 01-20897.

Remark: R. shinanoana resembles R. hossei, though it can be distinguished from the latter species by the thallus growing from a narrow holdfast and the presence of soralia. R. shinanoana might also be confused with R. farinacea, a species widely distributed throughout the world, which differs in having farinose soralia and producing protocetraric acid as a major chemical substance. R. shinanoana very much resembles R. asahinae in having sorediate branches terminating in nodules and the noncracked chondroid tissues. However, it can be separated from the latter species by the slender branches. In R. asahinae, the branches are wider and the apothecia usually bear slender spurs and granular soredia. It might also be confused with R. intermediella Vain., from which it can be separated by the shrubby thallus growing from a narrow holdfast and by the finely dissected branchlets.

Ramalina sinensis Jatta Nouv. G. Bot. Ital. 9: 462 (1902).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, growing from narrow holdfast, erect, thalli with greener "dorsal" sides, and white "ventral" sides, solid, thallus with broad, flat, veined, rugose reticulate, depressed and reticulately arranged pseudocyphellae on the lower surface of lobes, chondroid tissue not cracked, apothecia common, terminal, marginal entire, fusiform ascospores are $5 \,\mu$ m long and $2.5 \,\mu$ m wide and slightly curved.

Chemistry: Medulla K–, C–. TLC: usnic acid.

Ecology and distribution: This species has been reported in China [12, 14], and is also widely distributed in Europe [26], the Kuril Islands [22], Japan [23], North America [25], and South Korea [24]. In China, it was collected on various barks (*Quercus, Rhododendron, Pinus*, etc.) at elevations between 1,800 and 4,300 m.

Selected specimens examined: Sichuan Prov.: Daofu Co., Renguo village, on bark of *Picea* sp., N 30°46', E 101°18', elev. 3,950 m, L. S. Wang, 07-28313; Tibet: Jiayu Co., Zhuenbadala, Mu Zang, 67; ZhangMu to Lixing, elev. 2,500~2,700 m, Shu-Kun Luo 50; Yunnan Prov.: Bingchuan Co., Mt. Jizu, on bark of *Quercus* sp., N 25°58', E 100°21', elev. 3,200 m, L. S. Wang, 06-26194; Dali Co., Mt. Cang, on bark of *Pinus armandii*, N 25°41', E 100°06', elev. 3,400 m, L. S. Wang, 05-24661; Deqen Co., Mt. Baimaxue, elev. 3,400 m, L. S. Wang, 93-13569; Luquan Co., Mt. Jiaozixue, elev. 3,750 m, L. S. Wang, S. O, Oh, 06-26101; Zhongdian Co., Napa Lake, on bark of *Rhododendron* sp., N 27°56', E 99°36', elev. 3,540 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26611.

Remark: *R. sinensis* is highly variable in lobe shape and branching, though specimens commonly found in China

typically have palmate and loosely tufted lobes. It might be confused with *R. fraxinea*, a species that differs in having branches without dorsiventral appearance, the ellipsoid pseudocyphellae, and distinctly curved ascospores.

Ramalina subcomplanata (Nyl.) Kashiw. Bull. Natl. Sci. Mus., Ser. B. Bot. 12: 92 (1986).

Brief description of morphology and anatomy: Thallus corticolous, fruticose, growing from a narrow holdfast, dichotomously or irregularly branched, solid, surface angular or flat, rarely canaliculated, pseudocyphellae common, marginal or ventral side, more or less convex, chondroid tissue cracked, apothecia subterminal, rarely lateral, broadly ellipsoid ascospores that are 15 μ m long and 5 μ m wide.

Chemistry: Medulla K–, C–. TLC: usnic acid, homosekikaic acid, sekikaic acid.

Ecology and Distribution: This species has been reported in China [14], where it was grows on various barks (*Rhododendron, Salix* etc.) at elevations between 1,700 and 3,540 m.

Selected specimens examined: Yunnan Prov.: Long-chuan Co., Tongbiguang, elev. 1,700 m, L. S. Wang, 97-18101; Zhongdian Co., Napa Lake, on bark of *Rhododendron* sp., N 27°56', E 99°36', elev. 3,540 m, L. S. Wang, S. O. Oh, D. L. Niu, 06-26607; Yunlong Co., Caojian village, Mt. Ziben, on bark of *Salix* sp., N 27°44', E 99°03', elev. 3,000 m, L. S. Wang, 05-24388 and N 25°45', E 99°06', elev. 2,800 m, L. S. Wang, Qiang Ren, 02-22438.

Remark: *R. subcomplanata* resembles *R. calicaris*, though it can be distinguished from the latter species by the strongly pseudocyphellate lobes and by the distinctly cracked chondroid tissue. In *R. calicaris*, the pseudocyphellae are very rare or completely lacking and the chondroid tissue forms no hyphal bundles.

Ramalina yasudae Räsänen J. Jpn. Bot. 16: 87 (1940).

Brief description of morphology and anatomy: Thallus saxicolous, fruticose, caespitose, erect, growing from a common holdfast, solid, simple or sparingly furcate branched towards the apices, mostly flattened, pseudocyphellae rare, soralia terminal and laminal with granular soredia, chondroid tissue not cracked, apothecia not seen.

Chemistry: Medulla K-, C-. TLC: evernic acid, obtusatic acid.

Ecology and distribution: This species has been reported in China [15], and is also distributed in Japan [23] and South Korea [24]. In China it was collected on rock or rarely on bark at elevations between 3,100 to 4,000 m.

Selected specimens examined: Sichuan Prov.: Kangding Co., Liqiu to Shade, on rock, elev. 3,200 m, L. S. Wang, 96-17546; Kangding Co., Mt. Zhedou, on rock, elev. 3,800 m, L. S. Wang, 07-28995; Muli Co., Yala Shaoxiangliangzi, elev. 3,800 m, L. S. Wang, 83-1734; Xiaojin Co., Changpinggou, elev. 3,100 m, L. S. Wang, 96-17784; Yunnan Prov.: Deqin Co., Meilishi village, Suola Ya-kou, N 28°38', E 98°36', elev. 4,000 m, L. S. Wang, 00-19764; Zhongdian Co., Wengshuei

village, Mt. Daxue, N 28°30', E 99°49', elev. 3,800 m, L. S. Wang, 00-19843.

Remark: *R. yasudae* may be confused with *R. pollinaria* because they both have similar sorediate branches. However, it can be distinguished from the latter species by bilateral laciniae, terminal and laminal soralia, and by granular and slightly corticated soredia.

ACKNOWLEDGEMENTS

This work was supported by a grant from the Korean Forest Service Program (KNA 2014) through the Korea National Arboretum and the Korean National Research Resource Center Program (NRF, 2012M3A9B8021726).

REFERENCES

- 1. Wei JC. An enumeration of lichens in China. Beijing: International Academic Publishers; 1991. p. 218-22.
- 2. Galloway DJ. Flora of New Zealand lichens. Vol. 2. Lincoln: Manaaki Whenua Press; 2007. p. 1505-22.
- Stevens GN. The lichen genus *Ramalina* in Australia. Bull Br Mus Nat Hist Bot Ser 1987;16:107-223.
- 4. Nylander W. Recognitio monographica Ramalinarum. Bull Soc Linn Normandie Ser 2 1870;4:101-181.
- Kashiwadani H. Genus *Ramalina* (lichens) in Japan: (2) On *Ramalina pacifica* ASAH. and its allies. Bull Natl Sci Mus Ser B Bot 1986;12:117-25.
- 6. Kashiwadani H, Moon KH. A new or interesting species of the genus *Ramalina* (Ascomycotina: Ramalinaceae) from Korea and Japan. Bull Natl Sci Mus Ser B Bot 2002;28:1-6.
- Kashiwadani H. Genus *Ramalina* (lichens) in Japan: (3) *Ramalina exilis* Asah. and its allies. Bull Natl Sci Mus Ser B Bot 1987;13:133-40.
- Kashiwadani H. *Ramalina hossei* Vain. (lichen) found in Bhutan and Formosa. Bull Natl Sci Mus Ser B Bot 1988; 14:129-33.
- 9. Kashiwadani H. *Ramalina siliquosa* (Huds.) A. L. Sm. and *R. subbreviuscula* Asah. in Japan. Mem Natl Sci Mus Tokyo 1992;25:63-9.
- Kashiwadani H. Ramalina kurokawae Kashiw., a new lichen species from Japan. Bull Natl Sci Mus Ser B Bot 1996;22:55-7.
- Stevens GN, Kashiwadani H. Synonymy of *Ramalina* intermediella Vain. with *Ramalina peruviana* Ach. J Jpn Bot 1987;62:373-6.
- Kashiwadani H, Moon KH, Lai MJ. The genus *Ramalina* (Ascomycotina: Ramalinaceae) in Taiwan. Mem Natl Sci Mus Tokyo 2006;44:161-73.
- Kashiwadani H, Moon KH, Guo S, Dai YC, Chen XL. Noteworthy species of the genus *Ramalina* (Ascomycotina: Ramalinaceae) in China. Bull Natl Sci Mus Ser B Bot 2006;32:161-6.
- Zhao ZT, Guo SX, Sun LY, Ren Q. The lichen genus *Ramalina* from Qinling mountains in China. Mycosystema 2007;26: 343-8.
- 15. Ohmura Y, Moon KH, Kashiwadani H. Morphology and molecular phylogeny of *Ramalina pollinaria*, *R. sekika* and *R. yasudae* (Ramalinaceae, lichenized Ascomycotina). J Jpn Bot

240 Oh et al.

2008;83:156-64.

- Orange A, James PW, White FJ. Microchemical methods for the identification of lichens. London: British Lichen Society; 2010. p. 7-50.
- 17. Culberson CF. Improved conditions and new data for identification of lichen products by standardized thin-layer chromatographic method. J Chromatogr A 1972;72:113-25.
- White TJ, Bruns T, Lee S, Taylor J. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, editors. PCR protocols: a guide to methods and applications. San Diego: Academic Press; 1990. p. 315-22.
- Hall TA. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Window 95/98/NT. Nucleic Acids Symp Ser 1999;41:95-8.
- Thompson JD, Gibson TJ, Plewniak F, Jeanmougin F, Higgins DG. The CLUSTAL_X Windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic Acids Res 1997;25:4876-82.
- 21. Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar

S. MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol Biol Evol 2011;28:2731-9.

- 22. Joneson S, Kashiwadani H, Tschabaneako S, Gage S. *Ramalina* of the Kuril Islands. Bryologist 2004;107:98-106.
- 23. Harada H, Okamoto T, Yoshimura I. A checklist of lichens and lichen-allies of Japan. Lichenology 2004;2:47-165.
- 24. Hur JS, Koh YJ, Harada H. A checklist of Korean lichens. Lichenology 2005;4:65-95.
- Brodo IM, Sharnoff SD, Sharnoff S. Lichens of North America. New Haven: Yale University Press; 2001. p. 620-32.
- Krog H, James PW. The genus *Ramalina* in Fennoscandia and The British Isles. Nor J Bot 1977;24:15-43.
- Krog H, Swinscow TD. The genus *Ramalina* in East Africa. Nor J Bot 1976;23:153-75.
- Kashiwadani H. Some chilean species of the genus *Ramalina* (lichens). Bull Natl Sci Mus Ser B Bot 1990;16:1-12.
- Kashiwadani H, Moon KH. The genus *Ramalina* (Ascomycota, Ramalinaceae) in Indonesia. Bibl Lichenol 2007;96:145-56.