

Original article

Arata Momohara¹, Yuichiro Kudo², Nao Miyake³, Toshio Nakamura⁴, Fuyuki Tokanai⁵ and Minoru Tsukagoshi⁶: Diversity of temperate flora at the Tado site, central Japan, during the last glacial stage, reconstructed from the Dr. Shigeru Miki collection

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三木茂博士採集の三重県多度産標本から復元した最終氷期の温帯性フロラの多様性

Abstract We describe plant macrofossils that comprise the collection made by Dr. Sigeru Miki at the Tado site, Mie Prefecture, central Japan, and stored in the Osaka Museum of Natural History. The specimens were dated to two age spans, 40,300–39,070 and 21,920–20,270 cal BP, corresponding to marine isotopic stage 3 (MIS 3) and the later stage of the last glacial maximum (LGM), respectively. Macrofossils from the LGM bed were dominated by temperate conifers such as *Tsuga sieboldii*, *Picea* sect. *Picea*, *Larix kaempferi*, and *Chamaecyparis obtusa* and also included diverse temperate broadleaf trees such as *Cercidiphyllum japonicum*, *Pterocarya rhoifolia*, *Fagus japonica*, and *Ostrya japonica*. Results of the pollen analysis of sediments adhered to a *Tsuga* cone indicated an expansion of the temperate coniferous forest around mixed coniferous and deciduous broadleaf forests that had developed along the Tado River. Occurrence of subalpine conifers at this site indicated the distribution of subalpine forests in the montane zone, contiguous with the mixed coniferous and deciduous broadleaf forest. Inland areas around the Tado site located mid-way between the coasts of the Pacific Ocean and Japan Sea were assumed to be refugia for temperate trees during the LGM when pinaceous conifers prevailed in central Japan. This study showed that reinvestigation of museum collections is important to reconstruct the distribution of paleovegetation during the last glacial stage.

Keywords: last glacial maximum, museum collection, plant macrofossil, refugia, temperate deciduous broadleaf tree

要 旨 本論文では三木茂博士が三重県桑名市多度で採取し、大阪市立自然史博物館に保管されている大型植物化石標本を再検討し、形態を記載した。標本の放射性炭素年代測定を実施し、MIS 3に相当する40,300–39,070 cal BPと最終氷期最寒冷期後半の21,920–20,270 cal BPの2つの時代に形成された植物化石群に由来することが明らかになった。最終氷期最寒冷期の化石群はツガ、トウヒ属バラモミ節、カラマツ、ヒノキといった温帯性針葉樹が優占し、カツラ、サワグルミ、イヌブナ、アサダなどの温帯性広葉樹の多様性が高かった。ツガ属球果と同定された化石標本に付着した堆積物の花粉分析結果は、多度川沿いに発達した針広混交林の周囲に温帯性針葉樹林が広がっていたことを示していた。亜高山針葉樹林は付近の山地帯に分布しており、針広混交林とは隣接していたと考えられる。多度周辺の地域は最終氷期最寒冷期には太平洋と日本海の中間の内陸に位置していたが、その温帯性落葉広葉樹を含む植生はマツ科針葉樹が優占していた当時の中部日本での温帯性樹木のレフュージアだったと考えられる。博物館標本の再検討は最終氷期の古植生分布を復元する上で重要である。

キーワード：大型植物化石、温帯性落葉広葉樹、最終氷期最寒冷期、博物館標本、レフュージア

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Introduction

The present-day distribution and local genetic structure of temperate tree taxa are influenced by their distribution during the last glacial maximum (LGM: ca. 30–19.5 ka; Lambeck et al., 2002) (Hattori, 1985; Fujii et al., 2002). Information on the distribution and communities of plant species during the LGM is provided by plant macrofossil records, but macrofossil records are still limited (Tsumura & Momohara, 2011), whereas pollen records obtained by boring in lakes and wetlands increased in recent years (Ooi, 2016). In the Kinki region in central Japan, macrofossil assemblages dated to ages in and around the LGM have been reported from only three sites (Fig. 1B): Tado in northern Mie Prefecture (Miki, 1948; Minaki & Matsuba, 1985), Itai-Teragatani in Hyogo Prefecture (Ooi et al., 1991), and Tonda in Osaka Prefecture (Itihara, 1960; Itihara & Kigoshi, 1962). There is also a record for a single species, *Menyanthes trifoliata*, collected from the Nagai site (Fig. 1B; Itihara & Kigoshi, 1962). Because the fossil assemblages at the Tado site include numerous temperate tree taxa (Miki, 1948, 1957; Minaki & Matsuba, 1985), reinvestigation of these assemblages is

important to elucidate the distribution and communities of temperate trees that survived in refugia during the LGM, when pinaceous conifers were dominant in vegetation throughout the Japanese archipelago.

The plant macrofossil assemblages at the Tado site, south of the Daimon Bridge of Tado Shrine (Fig. 1; 35°07'50"N, 136°37'27"E, alt. 42 m a.s.l.) in Kuwana City, Mie Prefecture, were first described based on 14 fossil taxa and classified into the “*Larix* Bed” by Miki (1948). Miki (1956, 1957) subsequently reported additional seven taxa, including subalpine conifers such as *Abies veitchii* and *Tsuga diversifolia*. About 30 years later, Matsuba (1983) reinvestigated the terrace deposits, including the fossil bed originally described by Miki (1948) and correlated these with the Middle Pleistocene (Minaki & Matsuba, 1985), whereas Kimura et al. (1984) dated the bed to 18,340 ± 430 yBP (23,160–21,020 cal BP), indicating a later stage of the LGM. From the outcrop dated by Kimura et al. (1984), Minaki & Matsuba (1985) reported plant macrofossil assemblages that were dominated by pinaceous conifers, including *Tsuga sieboldii* and *Larix kaempferi* (as *L. leptolepis*), with an admixture of temperate broad-

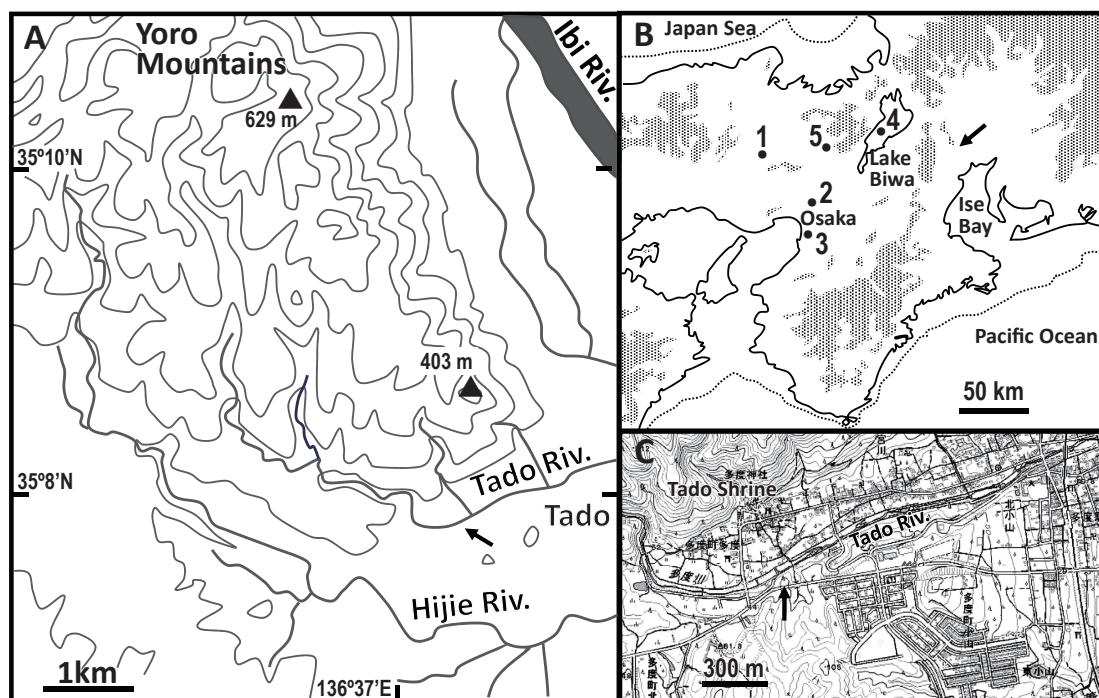


Fig. 1 The location of the Tado site (arrow) and the other fossil localities correlated with the last glacial maximum (LGM) and MIS 3 in the Kinki region, central Japan. — A, C: Topographic location of the Tado site (arrow), made from the 1:25,000 topographic map published by the Geospatial Information Authority of Japan. B: The fossil localities correlated with the LGM (1: Itai-Teragatani site (Ooi et al., 1991), 2: Tonda site (Itihara, 1960), 3: Nagai site (Itihara & Kigoshi, 1962)), and MIS 3 (4: Lake Biwa (Hayashi et al., 2010), 5: Kamiyoshi Basin (Hayashi et al., 2009)). Dashed lines and shaded areas indicate the location of the seashore during the LGM (-120 m a.s.l.) and the present montane zone above 500 m a.s.l., respectively.

leaf trees such as *Fagus crenata*, *Aesculus turbinata*, and *Stewartia* sp. Based on the composition, Minaki & Matsuba (1985) correlated the assemblages with the fossil bed described by Miki (1948) and reconstructed a mixed coniferous broadleaf tree forest in the cool-temperate zone. However, the presence of subalpine conifers described by Miki (1956, 1957) indicate the distribution of subalpine forests in and around the site during the LGM. Because plant macrofossil assemblages dominated by subalpine conifers tend to be widely distributed and reported from lower altitudinal areas in western Chugoku region (Nishiuchi et al., 2017), we thought that a more detailed investigation of plant macrofossil assemblages in western and central Japan is necessary to reconstruct the distribution of biomes during the LGM.

In this study, we reinvestigated the specimens collected from the Tado site by Shigeru Miki, which are currently stored at the Osaka Museum of Natural History (Kokawa et al., 2006), and identified several taxa that were not previously described by Miki (1948, 1956, 1957). Because Miki's reports lacked descriptions and images of the collected fossils, we examined their morphology to ascertain the floristic features of the assemblages. We dated some of these fossils and analyzed the pollen in sediments adhering to macrofossils to reconstruct the paleovegetation in greater detail.

Materials and methods

The fossil specimens collected from the Tado site by Shigeru Miki are registered at the Osaka Museum of Natural History (Kokawa et al., 2006). The collection includes macrofossils preserved in ethanol that are maintained in three bottles, 3-1, 3-6, and 3-11, and specimens mounted on 66 glass slides. The bottles include labels describing the respective localities and collection dates, i.e., bottle 3-1: "Tado Pref. Mie, March 28, 1942" and "Tado Daimon", bottle 3-11: "Tado 28 III 42", and bottle 3-6 "Tado upper 6 March 1966." Because the label in bottle 3-6 shows a different locality name (Tado upper) and a collection date 24 years later, the collection in bottle 3-6 is distinguished from those in the other two bottles, 3-1 and 3-11. Bottle 3-1 contains mainly coniferous taxa, whereas broadleaf tree taxa predominate in bottle 3-11. Glass slides bearing the locality names "Tado Daimon," "Tado," or "Tado (upper)" correspond to the localities indicated for bottled specimens. When Kokawa et al. (2006) identified and arranged the collection, registration numbers (F-) were assigned based on taxa and plant parts in bottles and on glass slides (Table 2, 3). For the newly identified specimens, we added registration num-

bers F24866–F24873 and F24901–F24917. For specimens with pre-existing registration numbers, we added suffix numbers (e.g., F17651-1) to specify the material used for radiocarbon dating and photographing.

Plant macrofossils were identified by comparing with modern reference materials stored in the Herbarium of the Graduate School of Horticulture, Chiba University (MTDO) and were counted under a binocular microscope. Fragmented plant parts were numerically represented by estimating the number of whole parts that could be reconstructed from these fragments. We counted the number of leaf bases for conifer needles and calculated the volume of shoots by using a length of 1 cm to represent a single specimen. A *Pinus* subgen. *Haploxylon* leaf (F17653-3) was sliced by hand, and the self-luminescent walls of epidermal and endodermal cells was observed under a fluorescence microscope. Sizes of fossils smaller than ca. 10 mm were measured by micrometer and indicated in 0.1 mm increment.

Samples used for pollen analysis were obtained from silt adhering to a *Tsuga sieboldii* cone (F17645-1), which was taken from bottle 3-1. In order to assess the possibility of contamination with modern pollen, we also analyzed the silty dregs deposited at the bottom of a vial (F17645) containing the sample cone, which was maintained in 70% ethanol solution for six months along with three other *Tsuga* cones (two of *T. sieboldii* and one of *T. diversifolia*). These pollen/spore specimens were treated with a solution of 10% KOH, centrifuged with ZnCl₂ solution, and finally treated with an acetolysis solution. The specimens were then mounted on slides in glycerol jelly. All pollen and spore samples obtained from the *Tsuga* cone were identified and counted. Percentages of arboreal pollen and those of herbaceous pollen and spore were based on the total arboreal pollen sum and the total pollen and spore sum, respectively.

Taxonomic nomenclature of fossils in this paper generally follows Birks & Birks (1980) that illustrates examples used in Quaternary paleoecology. For example, "*Picea*" indicates plural specimens in that genus, whose generic determination is certain, but whose types or species are undetermined or indeterminable. "*Picea* sp." indicates one specimen or fossil type, consisting of only one species. "*Picea alcoquiana*-*P. koyamae*" indicates one fossil type including two probable species that cannot be distinguished from fossil morphology alone. "*Picea alcoquiana* type" indicates one fossil type including three or more probable species.

Following the acid-alkali-acid (AAA) treatment at the National Museum of Japanese History, radiocarbon dating by the accelerator mass spectrometry (AMS)

was conducted for a *Chamaecyparis obtusa* shoot (F17648-1) and *Larix kaempferi* shoot (F17651-1) from bottle 3-1, an *Alnus* twig (F24903-1) from bottle 3-11, and a *Juglans ailanthifolia* nut (F17678) from bottle 3-6 at the Institute for Space-Earth Environmental Research, Nagoya University, and for a *Picea* cone (F17679) from bottle 3-6 at the High Sensitivity Accelerator Center, Yamagata University. The carbon ages were calibrated using OxCal version 4.2 (Bronk Ramsey, 2009) based on IntCal 20 (Reimer et al., 2020).

Results

1. AMS radiocarbon dating

The dated 2σ calibrated ages of *Larix kaempferi* shoot (F17651-1), *Chamaecyparis obtusa* shoot (F17648-1), and *Alnus* twig (F24903-1) were 21,920–21,390, 21,090–20,640, and 20,650–20,270 cal BP, respectively (Table 1). These ages correspond to the latter stages of the LGM and are slightly younger than the calibrated age of 23,160–21,020 cal BP calculated from the $18,340 \pm 430$ yBP date reported by Kimura et al. (1984). The dated 2σ calibrated ages of *Juglans ailanthifolia* nut (F17678) and *Picea* cone (F17679) from bottle 3-6 were 40,020–39,070 and 40,300–39,410 cal BP, respectively, that approximately overlap at 39.5–40 ka during the MIS 3 stage (ca. 60–30 ka).

2. Description of plant macrofossils

The Miki collection comprises a total of 37 taxa, including 25 woody and 12 herbaceous taxa (Table 2, 3). The descriptions of these taxa are provided below.

Pinaceae

Abies homolepis Siebold et Zucc. Upper part of seed scale (Fig. 2-2a) fan-shaped, 8 mm long, 12 mm wide, woody; lateral and upper margin flat, thinning gradually to the margin; abaxial surface striate. Bract scale (Fig. 2-2b) circular, 4.6 mm long, 5.3 mm wide; apex cuspidate; upper margin serrate.

Abies veitchii Lindl. Seed scale (Fig. 2-3) fan-shaped, 6.3–9.0 mm long, 9–10 mm wide; base cuneate; upper part transversely elliptic; upper margin curved shortly to adaxial side. Bract scale abraded.

Abies. Leaf (Fig. 2-1) oblanceolate, dorsoventrally flat, 10–21 mm long, 1.2 (avg. 1.5)–1.7 mm wide, with stomatal bands on ventral side of lamina; apex retuse; lower part of lamina tapering off to the base bulging above circular abscission scar.

Larix kaempferi (Lamb.) Carrière. Female cone (Fig. 2-7) broadly ovate, 18 mm long, 13.8 mm wide, with a short shoot attached to the base; cone scale woody, broadly obovate, with the upper margin reflected shortly. Seed (Fig. 2-6) liginous, trigonous-obovoid, dorsoventrally flat, 3.7–4.0 mm long, 2.3–2.4 mm wide, 1.2–1.5 mm thick, ridged along upper margin, with membranous wing (Fig. 2-5); seed wing obovate, 6.0 mm long, 3.6 mm wide, adhering to abaxial side of seed body, marginally covering its adaxial base. Short shoot (Fig. 2-4) cylindrical, 3.0–6.0 mm long, 2.3–2.5 mm wide, banded by scales and leaf abscission scars alternately at interval of 0.5–0.6 mm. Leaf (Fig. 2-8) membranous, linear–oblanceolate, 11 (avg. 15)–18 mm long, 0.8–1.0 mm wide; apex obtuse; lateral margin curved shortly to ventral side; lower part of lamina tapering off to base with an abscission scar of 0.2 mm wide.

Picea alcoquiana (Veitch ex Lindl.) Carrière–*P. koyamae* Shirasawa. Apical part of a female cone (Fig. 2-10) ovoid, 23 mm long, 14 mm wide; seed scale, woody, thick, obovate, ca. 12 mm long, ca. 8 mm wide, with the upper part abraded and broken; cone axis, 3.0 mm wide, with solid pith.

Among *Picea* distributed in central Honshu, *P. alcoquiana* (synonym of *P. bicolor*), *P. koyamae*, and *P. jezoensis* are distinguished by having a solid pith in the cone axis from *P. maximowiczii* and *P. polita* having a soft pith (Miki, 1957). Cone scales of this specimen are shorter than those of *P. alcoquiana* that are longer than ca. 20 mm in the apical part and thicker than those of *P. jezoensis* with thin and undulated margins. *Picea* sect. *Picea* cone morphotypes similar to *P. alcoquiana* and *P. koyamae* are often found in the last glacial sediments and are assigned to *P. tomizawaensis* by Suzuki (1991) or *P. aff. shirasawae* by Minaki (1987). Minaki (1987) suggested existence of a wider variation in the cone morphology of *Picea* sect. *Picea* than in the cones

Table 1 Radiocarbon ages of plant macrofossils from the Tado site, Mie Prefecture, central Japan

| Dating number | Taxa and plant part | Registration no. | Collection | $\delta^{13}\text{C}$ (‰) | ^{14}C age (yrBP) | 2σ calibrated age (calBP) |
|---------------|-----------------------------------|------------------|------------|---------------------------|----------------------------|----------------------------------|
| NUTA2-14922 | <i>Larix kaempferi</i> shoot | F17651-1 | 3-1 | -26.2 ± 0.11 | $17,820 \pm 70$ | 21,920–21,390 |
| NUTA2-14922 | <i>Chamaecyparis obtusa</i> shoot | F17648-1 | 3-1 | -24.5 ± 0.12 | $17,320 \pm 70$ | 21,090–20,640 |
| NUTA2-14923 | <i>Alnus</i> twig | F24903-1 | 3-11 | -29.0 ± 0.12 | $16,940 \pm 70$ | 20,650–20,270 |
| NUTA2-14928 | <i>Juglans ailanthifolia</i> nut | F17678 | 3-6 | -26.5 ± 0.10 | $34,320 \pm 230$ | 40,020–39,070 |
| YU-9445 | <i>Picea</i> cone | F17679 | 3-6 | -26.43 ± 0.38 | $34,670 \pm 150$ | 40,300–39,410 |

Table 2 List of plant macrofossils from the LGM bed at the Tado site

| Taxa | Plant part | Collection* | Registration number** | Number | % |
|---|-----------------|-------------|--|------------|------------|
| Arboreal taxa | | | | | |
| <i>Abies homolepis</i> | cone scale | s | F9470 | 1 | 0.2 |
| <i>Abies veitchii</i> | cone scale | s | F9532 | 2 | 0.4 |
| <i>Abies</i> | leaf | 1, s | F24869, F9510, F9653, F9759 | 12 | 2.6 |
| <i>Larix kaempferi</i> | female cone | 1 | F24866 | 1 | 0.2 |
| | seed | 1, s | F24871, F10030 | 3 | 0.7 |
| | short shoot | 1, s | F17651, F10008 | 11 | 2.4 |
| | leaf | 1, s | F24872, F9958 | 40 | 8.7 |
| <i>Picea alcoquiana</i> type | leaf | 1 | F17649 | 40 | 8.7 |
| | shoot with leaf | 1 | F17650 | 4 | 0.9 |
| <i>Picea jezoensis</i> var. <i>hondoensis</i> | leaf | 1 | F24873 | 1 | 0.2 |
| <i>Picea polita</i> | leaf | 1 | F17649-2 | 20 | 4.4 |
| | seed | 2 | F24904 | 1 | 0.2 |
| <i>Picea</i> | shoot | 2 | F17693 | 3 | 0.7 |
| <i>Picea-Tsuga</i> | cone scale | s | F10006 | 2 | 0.4 |
| <i>Pinus koraiensis</i> | seed | 1 | F17652 | 1 | 0.2 |
| | leaf | 1 | F17653-2, 3 | 1 | 0.2 |
| | shoot with leaf | 1 | F17653 | 3 | 0.7 |
| <i>Pinus</i> subgen. <i>Haploxyylon</i> | shoot with leaf | s | F2815, F3007 | 4 | 0.9 |
| <i>Tsuga diversifolia</i> | female cone | 1 | F17645-2 | 1 | 0.2 |
| | shoot | 1 | F17646-1 | 1 | 0.2 |
| <i>Tsuga sieboldii</i> | female cone | 1 | F17645-1, 3, 4 | 3 | 0.7 |
| <i>Tsuga</i> | seed | 2 | F24867, F24902 | 7 | 1.5 |
| | shoot | 1, s | F17646, F10278, F10355 | 16 | 3.5 |
| | leaf | 1, s | F17647, F10143, F10147 | 149 | 32.5 |
| <i>Chamaecyparis obtusa</i> | seed | 1 | F24870 | 1 | 0.2 |
| | shoot | 1, 2, s | F17648, F24905, F10557, F12478, F12524, F12525, F12532, F12533, F12535, F12557, F12563, F12564, F12565, F12567, F12568, F12569 | 29 | 6.3 |
| conifer | male cone | s | F2938 | 1 | 0.2 |
| <i>Corylopsis</i> sp. | seed | s | F8073 | 1 | 0.2 |
| <i>Cercidiphyllum japonicum</i> | fruit | s | F7562 | 1 | 0.2 |
| <i>Cerasus maximowiczii</i> | stone | 2, s | F17697, F1114 | 3 | 0.7 |
| <i>Rubus</i> sp. | stone | 2 | F24907 | 1 | 0.2 |
| <i>Fagus japonica</i> | cupule | 2, s | F17696-1, F15215 | 2 | 0.4 |
| | nut | 2 | F17696-2 | 1 | 0.2 |
| <i>Fagus</i> sp. | nut | s | F15205 | 1 | 0.2 |
| <i>Pterocarya rhoifolia</i> | nutlet | 2 | F17694 | 5 | 1.1 |
| <i>Alnus hirsuta</i> | infructescence | 2 | F17699 | 5 | 1.1 |
| | fruiting scale | s | F13603, F13605, F13609, F13610, F13651 | 8 | 1.7 |
| | fruit | 2, s | F24901, F13642, F13691 | 6 | 1.3 |
| <i>Alnus</i> | twig | 2 | F24903 | 35 | 7.6 |
| <i>Betula grossa</i> | fruiting scale | s | F13842, F13843, F13844 | 3 | 0.7 |
| | fruit | s | F13577 | 3 | 0.7 |
| <i>Carpinus cordata</i> | fruit | 2, s | F17695, F3736 | 4 | 0.9 |
| <i>Ostrya japonica</i> | fruit | s | F13860 | 1 | 0.2 |
| | fruiting bract | s | F13953 | 1 | 0.2 |
| <i>Acer japonicum</i> | fruit | 2, s | F17698, F4813, F4968 | 3 | 0.7 |
| <i>Aralia elata</i> | stone | s | F9044 | 1 | 0.2 |
| unknown | bud scale | s | F2817 | 1 | 0.2 |
| | spine | s | F9010 | 1 | 0.2 |
| Herbaceous taxa | | | | | |
| <i>Ranunculus</i> sp. | fruit | s | F16027 | 1 | 0.2 |
| <i>Persicaria nepalensis</i> | fruit | 1, 2 | F24868, F24908 | 2 | 0.4 |
| <i>Persicaria thunbergii</i> | fruit | 2, s | F24906, F3679 | 3 | 0.7 |
| Unknown plant part | | s | F4107, F4161, F10019 | 5 | 1.1 |
| Unknown | fruit | 2 | F24909 | 2 | 0.4 |
| Total number | | | | 458 | 100 |

*1: bottle 3-1, 2: bottle 3-11, s: slides.

**Italic registration numbers indicate specimen on slides.

Table 3 List of plant macrofossils from the MIS3 bed, i.e., Tado (upper) bed, at the Tado site

| Taxa | Plant part | Collection* | Registration number** | Number |
|---|-------------|-------------|-----------------------|--------|
| Arboreal taxa | | | | |
| <i>Picea alcoquiana</i> – <i>P. koyamae</i> | female cone | 3 | F17679 | 3 |
| <i>Picea</i> | seed | 3 | F24910 | 4 |
| <i>Chamaecyparis obtusa</i> | shoot | 3 | F24917 | 1 |
| <i>Chamaecyparis pisifera</i> | seed | 3, s | F24911, F12714 | 2 |
| <i>Juglans ailanthifolia</i> | nut | 3, s | F17678, F13410 | 2 |
| <i>Betula maximowicziana</i> | fruit | 3 | F24912 | 1 |
| Herbaceous taxa | | | | |
| <i>Alisma canaliculatum</i> | fruit | s | F16022 | 4 |
| <i>Carex dickinsii</i> | fruit | 3, s | F17675, F3695, F24862 | 21 |
| <i>Carex maximowiczii</i> | fruit | 3, s | F17676, F3530-1 | 10 |
| <i>Carex vesicaria</i> | fruit | 3 | F24914 | 2 |
| <i>Carex</i> sp. | fruit | 3 | F24915 | 1 |
| <i>Eleocharis</i> sp. | fruit | 3 | F24916 | 1 |
| <i>Schoenoplectiella botarui</i> type | fruit | 3, s | F17674, F3530-2 | 9 |
| <i>Viola</i> | seed | 3 | F17677 | 2 |
| <i>Mosla dianthera</i> | fruit | 3 | F24913 | 1 |
| Unknown plant part | | s | F3951 | 1 |

*3: bottle 3-6, s: slides. **Italic registration numbers indicate specimen on slides.

of modern relatives (*P. koyamae*, *P. shirasawae*, and *P. alcoquiana*). However, we assigned this fossil cone to *P. alcoquiana*–*P. koyamae*, because it is a part of a cone lacking detailed characteristics comparable to reported fossil species and because *P. shirasawae* is treated as a synonym of *P. koyamae* (Katsuki et al., 2008).

Picea alcoquiana (Veitch ex Lindl.) Carrière type. Leaf (Fig. 2-11) linear, 8.0–10.5 mm long, 0.9–1.0 mm wide, quadrangular in transverse section with stomatal band on each face, base truncate, apex acute–obtuse. This leaf morphotype is consistent with the leaf morphology of *Picea alcoquiana*, *P. glehnii*, *P. koyamae*, and *P. maximowiczii* and is distinguished from that of *P. jezoensis* having narrowly elliptic transverse sections and that of *P. polita* having acuminate apex.

Picea jezoensis (Siebold et Zucc.) Carrière var. *hondoensis* (Mayr) Rehder. Leaf (Fig. 2-12) linear, apex acute, base truncate, 10–11 mm long, 1.3–1.5 mm wide, dorsoventrally flat with narrowly elliptic transverse section; adaxial surface (Fig. 2-12b) with stomatal bands; abaxial surface (Fig. 2-12a) with slightly swollen midrib and without stomatal bands.

Picea polita (Siebold et Zucc.) Carrière. Leaf (Fig. 2-13) linear–narrowly oblong, laterally compressed quadrangular in transverse section, with stomatal bands on each face, dorsoventrally curved, 9.8 (avg. 12.0)–15.0 mm long, 1.2 (avg. 1.5)–1.8 mm wide; apex acuminate; base truncate. Seed (Fig. 2-14) obovoid, 5.5 mm long, 2.9 mm wide, 2.0 mm thick, ligneous; apex round; base obtuse. The size of seeds is the largest among *Picea* species distributed in Japan.

Picea. Shoot (Fig. 2-9) cylindrical, with pulvini pro-

truded perpendicularly, occasionally with bud scales at the top and spirally arranged leaves on pulvini; pulvinus ca. 1.0 mm long. This shoot has leaves belonging to *P. alcoquiana* type. Seed (Fig. 2-15) obovoid; apex round; base obtuse–round, 2.4–4.0 mm long, 1.6–2.4 mm wide, 1.0–1.4 mm thick, ligneous.

Pinus koraiensis Siebold et Zucc. Seed (Fig. 2-21) obovoid, 14.0 mm long, 11.5 mm wide; seed coat 1.0–1.5 mm thick, sclerous; outer epidermis membranous with striated tissues. Short shoot (Fig. 2-23) cylindrical, ca. 1.5 mm long, 2.4–2.9 mm wide, with horizontal furrows; apex with a fascicle of five leaves or triangular leaf scars. Leaf (Fig. 2-22) linear, with three ridges serrated, with stomatal bands on two ventral faces; apex acute; base with a trigonal abscission scar. Transverse section of leaf (Fig. 4-1, 2) triangular, 1.0–1.2 mm wide, with a vascular bundle enclosed by a single layer of 26 endodermal cells at the center; three resin ducts at mid-portion between ridgetop and endodermal cell layer; hypodermal cell layer single (Fig. 4-2).

Tsuga sieboldii Carrière. Female cone (Fig. 2-19) ellipsoidal, 15–18 mm long, 7–13 mm wide, with spirally arranged seed scales; seed scale very broadly obovate, with truncate or slightly emarginate apex.

Tsuga diversifolia (Maxim.) Mast. Female cone (Fig. 2-20) ellipsoidal, 20 mm long, 12 mm wide, with spirally arranged seed scales; seed scale transversely broadly elliptic, 6.0–9.0 mm long, 11 mm wide, with round apex. Shoot (Fig. 2-18) linear with pulvini arranged spirally, 10 mm long, 1 mm wide; surface with short hairs (Fig. 4-3); pulvinus diverging gradually from twig, 1.2–1.5 mm long, 0.3 mm wide, with semi-



Fig. 2 Plant macrofossils from the Tado site (1). — 1: *Abies* leaf (F24869-1). — 2: *A. homolepis* (a: seed scale (F9470-2), b: bract scale (F9470-1)). — 3: *A. veitchii* cone scale (F9532). — 4–8: *Larix kaempferi* (4: short shoot (F17651-2), 5: seed (F10030, dorsal view), 6: seed (F24871-1, ventral view), 7: female cone (F24866), 8: leaf (F24872-1)). — 9, 11: *Picea alcoquiana* type (9: shoot (17650-1), 11: leaf (F17649-1)). — 10: *P. alcoquiana*–*P. koyamae* female cone (F17679). — 12: *P. jezoensis* var. *hondoensis* leaf (F24873, a: abaxial side, b: adaxial side). — 13, 14: *P. polita* (13: leaf (F17649-2), 14: seed (F24904)). — 15: *Picea* seed (F24910-1). — 16–17: *Tsuga* (16: seed (F24867-1), 17: leaf (F17647-1)). — 18, 20: *T. diversifolia* (18: shoot (F17646-1), 20: female cone (F17645-2)). — 19: *T. sieboldii* female cone (F17645-3). — 21–23: *Pinus koraiensis* (21: seed (F17652), 22: leaf (F17653-2), 23: short shoot (F17653-1)). — 24–25: *Chamaecyparis obtusa* (24: shoot (a: F17648-2, b: F17648-3), 25: seed (F24870)). — 26: *Chamaecyparis pisifera* seed (F24911). — 27: *Alisma canaliculatum* fruit (F16022-1). — 28: *Carex dickinsii* fruit (F17675-1). — 29: *C. vesicaria* fruit (F24914-1). Thick scale = 5 mm (2, 7, 9, 10, 19, 20, 21, 24); thin scale = 1 mm (others).

circular leaf scar, 0.2 mm long, 0.4 mm wide, facing diagonally upwards.

Tsuga. Seed (Fig. 2-16) trigonal obovate, flat, 4.3–4.5 mm long, 2.3–2.5 mm wide, 1.5–1.8 mm thick, with small resin vesicles on surface, with membranous wing adhering to abaxial side of seed and covering adaxial base of seed. Shoot 1.0–1.5 mm wide, glabrous. The surface of current year shoot of *T. sieboldii* is glabrous, whereas that of *T. diversifolia* has short hairs (Fig. 4-3). Because hairs of *T. diversifolia* are lost through aging, taphonomic process, and cleaning treatment, the glabrous shoots include both species of *Tsuga*. Leaf (Fig. 2-17) linear–narrowly oblong, petiolate, 8 (avg. ca. 13)–16 mm long, 1.3–1.8 mm wide, dorsoventrally flat, with stomatal bands on ventral side; apex retuse; base round, curved to ventral side; petiole straight, ca. 1 mm long.

Cupressaceae

Chamaecyparis obtusa (Siebold et Zucc.) Endl. Shoot (Fig. 2-24) with decussate scale leaves, compressed dorsoventrally; lateral leaves boat-shaped, with blunt and slightly incurved apex, 1.5–3.0 mm long, 0.8–1.5 mm wide; facial leaves triangular, 1.0–2.0 mm long, 1.0–1.5 mm wide; stomata distributed along leaf margin and leaf base. Seed (Fig. 2-25) somewhat skewed circular, flat, 3.3 mm long, 3.5 mm wide, 1.2 mm thick, with thick lateral wings of ca. 0.6 mm wide; micropylar apex round with two short tips; base truncated with semicircular hilum; surface shiny, wrinkled longitudinally, with two large fusiform resin vesicles.

Chamaecyparis pisifera (Siebold et Zucc.) Endl. Seed (Fig. 2-26) ovate, flat, 2.2 mm long, 1.1 mm wide, 0.4 mm thick, with several fusiform–narrow elliptic resin vesicles on surface.

Alismataceae

Alisma canaliculatum A. Braun et C. D. Bouché. Fruit (Fig. 2-27) winged achene, obovate, laterally compressed with wedge-shaped transverse section, 2.8 mm long, 2.0 mm wide, yellowish brown, translucent; apex round with a projection of style base at adaxial side; base notched; adaxial wing membranous; abaxial wing thick, spongy with two transverse lobes; seed of amphitropous ovule origin horseshoe-shaped, reddish brown.

Cyperaceae

Carex dickinsii Franch. et Sav. Fruit (Fig. 2-28) achene, fusiform with regular triangular transverse section, broadly rhombic from frontal view, 2.4–3.0 mm long, 1.8–2.3 mm wide; beak of style thick, straight at lower part, crooked at upper part, 3.0–5.0 mm long; surface smooth, slightly lustrous, reddish brown, with conspicuous hexagonal epidermal cells.

Carex maximowiczii Miq. Fruit (Fig. 3-1) achene, lenticular, circular from frontal view, 1.9–2.2 mm long, 1.6–2.0 mm wide, 0.8 mm thick; apex with lignified and straight style base ca. 0.3 mm long; base truncate with abscission scar of 0.5 mm in width; surface smooth, dull, black, with large and conspicuous hexagonal epidermal cells (Fig. 4-6). This species is distinguished from other species of *Carex* sect. *Phacocystis* by large and circular shape, lignified and straight style base, wide abscission scar, and the largest epidermal cells in the section *Phacocystis*.

Carex vesicaria L. Fruit (Fig. 2-29) achene, fusiform with swelled triangular transverse section, 2.1–2.3 mm long, 1.2–1.6 mm wide; apex acute with long and crooked beak of style; base cuneate with small abscission scar; surface smooth, slightly lustrous, reddish brown.

Carex sp. Fruit (Fig. 3-2) achene, obovoid with swelled triangular transverse section, 2.1 mm long, 1.6–1.9 mm wide; apex truncate with short and lignified base of style; base truncate with wide abscission scar; surface slightly lustrous, reddish brown, with conspicuous hexagonal epidermal cells (Fig. 4-7).

Eleocharis sp. Fruit (Fig. 3-3) achene, lenticular, broadly obovate from frontal view, 1.5 mm long, 1.2 mm wide, 0.4 mm thick; apex cuspidate with spongy stylopodium; base truncate with papillate scars of bristle surrounding abscission scar; surface smooth, yellowish brown.

Schoenoplectiella hotarui (Ohwi) J. D. Jung et H. K. Choi type. Fruit (Fig. 3-4) achene, lenticular, broad-obovoid with swollen abaxial face, 2.3–2.8 mm long, 1.9–2.1 mm wide; apex mucronate with short beak of style; base truncate with bristles of perianth; pericarp black, lustrous, rough with irregular transverse rows of low humps; epidermal cells longitudinally linear, very fine (Fig. 4-8). The achene morphotype larger than 2.0 mm in length includes *S. hotarui*, *S. juncooides*, and *S. triangulata*.

Ranunculaceae

Ranunculus sp. Fruit (Fig. 3-5) achene, obovate, laterally compressed, 2.4 mm long, 2.0 mm wide; exocarp membranous, translucent, yellowish gray; endocarp obovate, 2.2 mm long, 1.5 mm wide, brown.

Hamamelidaceae

Corylopsis sp. Seed of anatropous ovule origin (Fig. 3-7) ellipsoidal, 4.2 mm long, 2.9 mm wide; apex obtuse; base truncate with narrowly ovoid hilum depressed at both lateral faces; seed coat black, shiny, sclerous.

Cercidiphyllaceae

Cercidiphyllum japonicum Siebold et Zucc. Up-



Fig. 3 Plant macrofossils from the Tado site (2). — 1: *Carex maximowiczii* fruit (F17676-1). — 2: *Carex* sp. fruit (F24915). — 3: *Eleocharis* fruit (F24916). — 4: *Schoenoplectiella hotarui* type fruit (17674-1). — 5: *Ranunculus* sp. fruit (F16027). — 6: *Cercidiphyllum japonicum* fruit (F7562). — 7: *Corylopsis* sp. seed (F8073). — 8: *Cerasus maximowiczii* stone (F17697-1). — 9: *Rubus* sp. stone (F24907). — 10–11: *Fagus japonica* (10: cupule (F17696-1), 11: nut (F17696-2)). — 12: *Pterocarya rhoifolia* nutlet (F17694-1). — 13: *Juglans ailanthifolia* nut (F17678, a: frontal view, b: transverse section). — 14: *Betula maximowicziana* fruit (F24912). — 15–16: *Alnus hirsuta* (15: fruit (F24901-1), 16: infructescence (F17699-1)). — 17: *Alnus* twig (F24903-2). — 18–19: *Betula grossa* (18: fruit (F13577-1), 19: fruiting scale (F13843)). — 20: *Carpinus cordata* fruit (F17695-1). — 21–22: *Ostrya japonica* (21: fruiting bract (F13953), 22: fruit (F13860)). — 23: *Acer japonicum* fruit (F17698). — 24: *Persicaria thunbergii* fruit (F24906-1). — 25: *Persicaria nepalensis* fruit (F24868-1). — 26: *Viola* sp. seed (F17677-1). — 27: *Aralia elata* stone (F9044). — 28: *Mosla dianthera* fruit (F24913). Thick scale = 5 mm (13); thin scale = 1 mm (others).

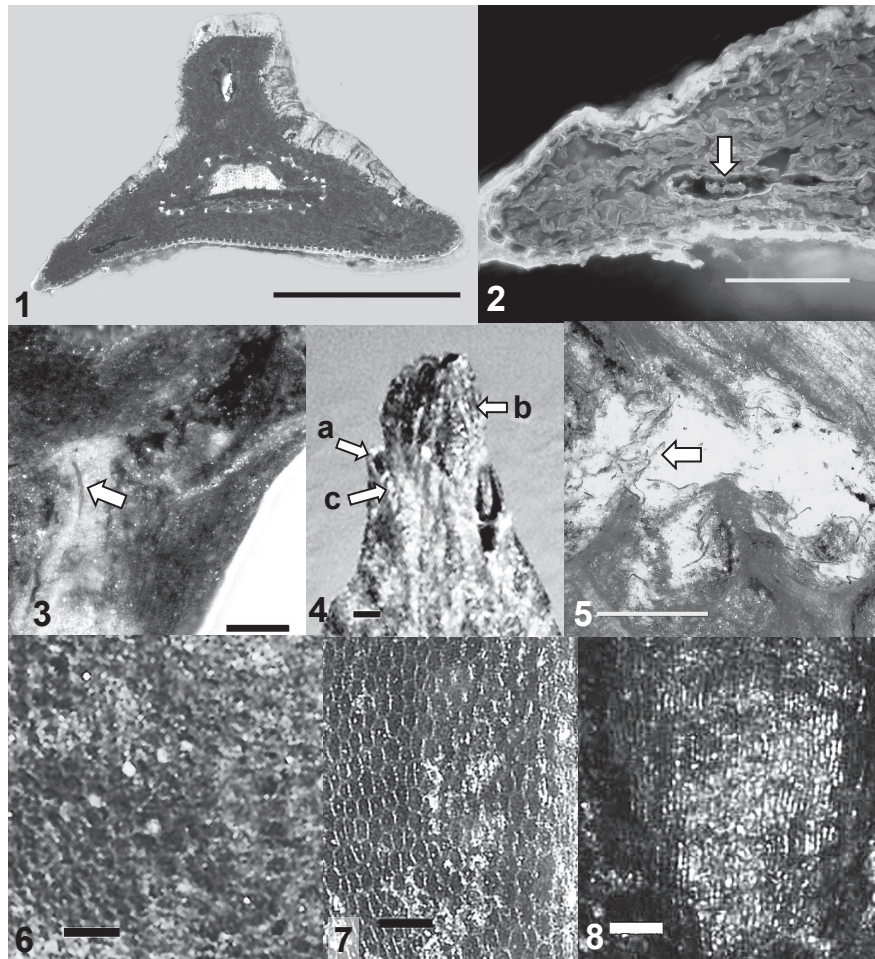


Fig. 4 Magnified and anatomical images of plant macrofossils from the Tado site. — 1–2: Transverse sections of a *Pinus koraiensis* leaf (F17653-3, fluorescence microscope image; arrow in 2: a resin duct). — 3: *Tsuga diversifolia* shoot (F17646-1; arrow: a hair borne on the shoot). — 4: Apex of *Fagus japonica* nut (F17696-2; arrow a: tepal, arrow b: style, arrow c: hairs). — 5: Upper part of an *Acer japonicum* fruit (F17698; arrow: crisp hairs). — 6–8: Surface of Cyperaceae fruits showing epidermal cells (6: *Carex maximowiczii* (F17676-1), 7: *Carex* sp. (F24915), 8: *Schoenoplectiella hotarui* type (17674-1)). Scales in 1 and 5 = 0.5 mm; scales in others = 0.1 mm.

per part of follicle (Fig. 3-6) lanceolate, 3.0–3.3 mm wide; exocarp chartaceous, dark brown; endocarp yellow, membranous, finely striated, with adaxial margin thickened with a width of 0.1 mm.

Rosaceae

Cerasus maximowiczii (Rupr.) Kom. Stone of drupe (Fig. 3-8) globose, 3.8 mm long, 4.8–5.3 mm wide, 4.5 mm thick; apex acute; base round with a small circular hollow of hilum; surface with protuberant and roughly reticulate ridges on lateral faces and fine grooves along suture line; endocarp woody and thick.

Rubus sp. Stone of drupe (Fig. 3-9) skewed semicircular, laterally compressed, 2.0 mm long, 1.3 mm wide, 1.1 mm thick; abaxial margin ridged; adaxial face round with small hilum; lateral face with reticulate

ridges; endocarp woody.

Fagaceae

Fagus japonica Maxim. Cupule (Fig. 3-10) ovoid, 7 mm long, ca. 6 mm wide, with four valves and peduncle; valve ovate, 4.0–6.0 mm long, 3.0–4.0 mm wide, with many small triangular scales of ca. 1.0 mm long, 0.3–0.4 mm wide; peduncle 0.8 mm wide, with small scale scars. Nut (Fig. 3-11) ovoid with regular triangle transvers section, 6.8 mm long, 3.3 mm wide, wingless; apex tapering with a persisted style base and small tepals covered by dense hairs (Fig. 4-4); base truncate with a triangle abscission scar.

Juglandaceae

Juglans ailanthifolia Carrière. Upper half of nut (Fig. 3-13) of estimated diameter ca. 20 mm; apex round;

surface furrowed slightly and somewhat abraded; nut wall of transverse section (Fig. 3-13b) thick and sclerous with large lacunae; primary septum with a large lacuna.

Pterocarya rhoifolia Siebold et Zucc. Fruit (Fig. 3-12) nutlet, rhombic ellipsoid from lateral view, 4.5–6.8 mm long, 5.0–6.5 mm wide, with 8–11 sharp longitudinal ribs converging into narrow abscission point; apex blunt.

Betulaceae

Alnus hirsuta Turcz. Axis of infructescence (Fig. 3-16) cylindrical, 9 mm long, 2 mm wide, with fruiting scales arranging spirally; fruiting scale, fan shaped, 2–3 mm long, 1.5–2.0 mm long, apex lobed. Fruit (Fig. 3-15) broadly obovate–rounded pentagonal, thin and chartaceous, 2.3–3.3 mm long, 2.7–3.1 mm wide, fringed by wing up to 0.5 mm wide along lateral and upper side; apex with two narrowly triangular pistils of 0.7–0.8 mm long; base truncate–cordate.

Alnus. Twig (Fig. 3-17) cylindrical, 1.2–2.5 mm in diameter; bark shiny, smooth with horizontal shallow furrows; leaf scar rounded semi-circular.

Betula grossa Siebold et Zucc. Fruit (Fig. 3-18) nutlet, broadly elliptic, thin and chartaceous, 3.4 mm long, 2.8 mm wide, brown; apex emarginate; base truncate; fruit body elliptic, with translucent pericarp showing the inside with median septum and two elliptic seeds, fringed by membranous wings of ca. 0.5 mm wide along the lateral sides. Fruiting scale (Fig. 3-19) trident-shaped, 5.1–7.5 mm long, 4.6–4.8 mm wide; middle lobe 1.2–3.1 mm long, 1.2 mm wide; lateral lobe 1.2–2.0 mm long, 1.7 mm wide; joint of lobes creased and bulging on abaxial face; abscission point with a small beak projecting at the base of adaxial face.

Betula maximowicziana Regel. Fruit (Fig. 3-14) nutlet, obovate, 1.8 mm long, 1.2 mm wide, 0.4 mm thick; apex round; base round with a circular abscission scar; surface black, rough, dimly lustrous; style base yellowish.

Carpinus cordata Blume. Fruit (Fig. 3-20) nutlet, ellipsoidal, 3.8–4.0 mm long, 2.4 mm wide, 1.8 mm thick; apex cuspidate with two styles; base with circular abscission scar; perianth adhering to exocarp entirely, brown; surface slightly lustrous, with ca. 10 fine vertical ribs.

Ostrya japonica Sarg. Base of saccate fruiting bract (Fig. 3-21), chartaceous, 10 mm long, 9 mm wide, with 20 fine primary veins diverging radially from the base, finely reticulate between the primary veins; base thickened and truncate with circular abscission scar of 0.7 mm in diameter. Fruit (Fig. 3-22) nutlet, ovate, dorsoventrally compressed, 5.1 mm long, 2.6 mm wide;

apex truncate; base round with abscission scar of 0.2 mm long, 0.8 mm wide; perianth membranous, brown, enclosing exocarp except for the basal part, partly separated from exocarp, with several fine vertical ribs on the surface.

Violaceae

Viola. Seed of anatropous ovule origin (Fig. 3-26) obovoid, 1.3 mm long, 0.9 mm wide; base mucronate; apex round with circular chalazal thickening; pericarp reddish brown, with very fine vertical striations.

Sapindaceae

Acer japonicum Thunb. Fruit (Fig. 3-23) samara, 5.0 mm long, 3.5 mm wide, 4.0 mm thick, oblong; base truncate with lanceolate hilum; surface with thick longitudinal veins diverging thin veins, with crisp hairs at upper part of fruit body (Fig. 4-5); wing developing from base of style.

Polygonaceae

Persicaria nepalensis (Meisn.) H.Gross. Fruit (Fig. 3-25) achene, lenticular, broadly ovate from frontal view, 2.1–2.2 mm long, 1.7–1.9 mm wide, 0.8 mm thick, apex cuspidate, base with circular hilum of 0.2 mm diameter; pericarp hard, black, with very fine scalariform network of ridges on the surface.

Persicaria thunbergii (Siebold et Zucc.) H.Gross. Fruit (Fig. 3-24) achene, ovoid, with triangular transverse section, 4.1 mm long, 2.5–2.7 mm wide; apex acute; base truncate, with remnant of perianth; pericarp soft, smooth, light brown.

Lamiaceae

Mosla dianthera (Hamilt.) Maxim. Fruit (Fig. 3-28) achene, skewed hemi-globose bulging upward on abaxial face, 1.2 mm long, 1.1 mm wide, 0.8 mm thick; base truncate; pericarp brown, with faint reticulation of ridges on the surface.

Araliaceae

Aralia elata (Miq.) Seem. Stone of drupe (Fig. 3-27) rounded semicircular, 2.0 mm long, 1.4 mm wide, laterally compressed, furrowed along abaxial margin; endocarp reddish brown, translucent, finely striated.

3. Composition of the Miki collection

Among the specimens preserved in the Miki collection, neither *Picea jezoensis*, *P. polita*, *Chamaecyparis pisifera*, *Juglans ailanthifolia*, *Betula maximowicziana*, *Ostrya japonica*, *Corylopsis*, *Rubus*, *Aralia elata*, nor any of the herbaceous taxa except for *Schoenoplectiella hotarui* were described in the original literature (Miki, 1948, 1956, 1957, 1958, 1961). Among 21 taxa reported by Miki (1956, 1957), *Thuja standishii*, *Actinidia polygama*, *Quercus crispula*, *Potamogeton gramineus*, and *P. minimum* were not found in the Miki

collection. *Picea alcoquiana*–*P. koyamae*, *C. pisifera*, *J. ailanthifolia*, *B. maximowicziana*, and all the herbaceous plants, with the exception of two *Persicaria* species and *Ranunculus*, were collected from the “Tado (upper)” bed in 1966 after Miki’s 1956 publication (Table 2, 3). *Picea alcoquiana* cone (as *P. bicolor*) and *Schoenoplectiella* sp. (as *Scirpus* sp.) reported from the “*Larix* bed” by Miki (1956, 1957) were included in the samples from the “Tado (upper)” bed. Among the total 521 plant samples in the Miki collection, 65 samples were collected from the “Tado (upper)” bed, with the remainder being obtained from the “*Larix* bed” (Miki, 1948). The composition of samples from the “upper” bed is characterized by diverse herbaceous plants, comprising mainly wetland taxa such as six cyperaceous taxa and *Alisma*, whereas woody taxa are limited to three coniferous and two broadleaf taxa (Table.3).

The samples collected from the “*Larix* bed” comprise 22 woody and three herbaceous taxa. Among these, *Tsuga* is the most abundant taxon, representing 38.6% of the total material, and includes *T. diversifolia* and *T. sieboldii*. Numerically, *Picea* (15.1%) and *Larix kaempferi* (12.0%) are the second and third largest components, whereas *Chamaecyparis obtusa* accounts for 6.6% of the total materials. Among the deciduous broadleaf taxa, *Alnus* (11.8%) including *A. hirsuta* is the most abundant. Other deciduous broadleaf trees, accounting for 6.3% of the total fossil count, comprise 12 taxa. Most of the tree taxa are components of temperate forests characteristic of the climatic zone located between the uppermost part of the warm temperate zone and the lowermost part of the subarctic zone. Although subalpine conifers such as *Abies veitchii*, *Picea jezoensis* var. *hondoense*, and *Tsuga diversifolia* are included, plant parts of *P. jezoensis* var. *hondoense* (leaf) and *T. diversifolia* (cone) are less abundant than those of the cool temperate taxa belonging to the same genera.

4. Pollen assemblage

The pollen assemblages comprise specimens identified to 21 arboreal and six herbaceous taxa, along with a spore of *Polypodium* and other monolet-type spores (Table 4). Arboreal pollen accounts for 90.8% of the total pollen and spore samples, and 78.7% of arboreal pollen is derived from pinaceous conifers, including *Abies*, *Larix*, *Picea*, *Pinus* subgen. *Haploxyylon*, *P.* subgen. *Diploxyylon*, *Tsuga sieboldii* type, and *T. diversifolia* type. Among these, *P.* subgen. *Haploxyylon* (28.7% of total arboreal pollen) and *T. sieboldii* type pollen (26.4%) are dominant. Among the taxa of broadleaf trees, *Alnus* subgen. *Alnus* pollen (5.1%) is

the most abundant. Of the total number of pollen and spore specimens, herbaceous pollen and spores account for 8.0%. The composition of the pollen assemblage sampled from the silty dregs in ethanol preservative is similar to that determined from sediment adhering to a collected *Tsuga* cone, without any evidence of additional taxa (Table 4). Thus, the possibility of contamination with present-day pollen can be excluded.

Interestingly, whereas *Pinus* subgen. *Haploxyylon* is the dominant representative in the pollen assemblage, it accounts for only a small fraction of the macrofossil collection, in which specimens of *Larix* and *Picea* tend to predominate. Furthermore, in both pollen assemblage and number of cones, *Tsuga sieboldii* type occurs

Table 4 List of pollen obtained from sediments adhering to a *Tsuga sieboldii* cone (F17645-1) and the silty dregs accumulated in the bottom of a vial containing *Tsuga* cones

| Taxa | Cone | | Vial | |
|---|-------|------|-------|------|
| | count | % | count | % |
| Arboreal pollen | | | | |
| <i>Abies</i> | 9 | 4.2 | 3 | 5.4 |
| <i>Larix</i> | 6 | 2.8 | 1 | 1.8 |
| <i>Pinus</i> subgen. <i>Diploxyylon</i> | 4 | 1.9 | 2 | 3.6 |
| <i>Pinus</i> subgen. <i>Haploxyylon</i> | 62 | 28.7 | 17 | 30.4 |
| <i>Pinus</i> undifferentiated | 8 | 3.7 | 6 | 10.7 |
| <i>Picea</i> | 10 | 4.6 | 4 | 7.1 |
| <i>Tsuga diversifolia</i> type | 14 | 6.5 | 2 | 3.6 |
| <i>Tsuga sieboldii</i> type | 57 | 26.4 | 15 | 26.8 |
| <i>Cryptomeria</i> | 2 | 0.9 | - | - |
| Other Cupressaceae | 1 | 0.5 | - | - |
| <i>Ulmus</i> type | 3 | 1.4 | - | - |
| <i>Fagus crenata</i> type | 4 | 1.9 | - | - |
| <i>Fagus japonica</i> type | 1 | 0.5 | - | - |
| <i>Quercus</i> subgen. <i>Quercus</i> | 2 | 0.9 | - | - |
| <i>Pterocarya-Juglans</i> | 5 | 2.3 | 2 | 3.6 |
| <i>Betula</i> | 6 | 2.8 | - | - |
| <i>Carpinus</i> (except for <i>C. tschonoskii</i> type) | 6 | 2.8 | 3 | 5.4 |
| <i>Acer</i> | 1 | 0.5 | - | - |
| <i>Alnus</i> subgen. <i>Alnus</i> | 11 | 5.1 | 1 | 1.8 |
| <i>Corylus</i> | 2 | 0.9 | - | - |
| <i>Salix</i> | 1 | 0.5 | - | - |
| Araliaceae | 1 | 0.5 | - | - |
| Total | 216 | 100 | 56 | 100 |
| Herbaceous pollen and spore | | | | |
| Cyperaceae | 2 | 0.8 | - | - |
| Gramineae | 4 | 1.7 | - | - |
| Ranunculaceae | 2 | 0.8 | - | - |
| Rosaceae | 1 | 0.4 | - | - |
| <i>Artemisia</i> | 3 | 1.3 | - | - |
| Umbelliferae | 2 | 0.8 | - | - |
| <i>Polypodium</i> | 1 | 0.4 | - | - |
| Other monolet-type spores | 4 | 1.7 | 7 | 10.9 |
| Total | 19 | 8.0 | 7 | 10.9 |
| Decayed pollen | 3 | 1.3 | 1 | 1.6 |
| Total pollen and spore | 238 | - | 64 | - |

Percentages of arboreal pollen and those of herbaceous pollen and spore were based on the total arboreal pollen sum and the total pollen and spore sum, respectively.

more than *T. diversifolia* type.

Discussion

1. Stratigraphic position and sedimentary environment of assemblages in the Miki collection

The results of macrofossil dating indicate that samples in bottle 3-6 labeled “Tado (upper)” were obtained from sediments deposited in marine isotopic stage 3 (MIS 3) of the Late Pleistocene, whereas those contained in bottles 3-1 and 3-11 were from the later stages of the LGM. Different sampling dates and locality names, as well as dating results, indicate sampling from different localities or horizons, and “upper” written on the label does not signify a stratigraphically upper position. Two overlapping dates at approximately 40 ka obtained from specimens in bottle 3-6 indicate that samples collected from the “Tado (upper)” bed may represent a single fossil assemblage. However, three different non-overlapping ages at approximately 21 ka seem to indicate sampling from different layers deposited at different ages. A sketch of an outcrop from a cutting excavated during road construction (Fig. 2 in Minaki & Matsuba, 1985) shows more than three organic layers, including the uppermost layer (M1) dated by Kimura et al. (1984) to $18,340 \pm 430$ yBP (23,160–21,020 cal BP). However, because of the similarities of 2σ age ranges within ca. 1.7 thousand years (21,920–20,270 cal BP), we amalgamate the samples in bottles 3-1 and 3-11 as a single fossil assemblage of the later stage of the LGM.

In terms of the abundance of *Larix kaempferi* (in M1 and M2) and *Tsuga sieboldii* (in M3), the composition of Miki’s LGM samples is similar to that of the macrofossil assemblages described by Minaki & Matsuba (1985). However, Miki’s samples include abundant specimens of *Picea*, *Chamaecyparis*, and *Alnus*, which are rare or absent in the assemblages described by Minaki & Matsuba (1985). The corresponding pollen assemblage in M1 reported by Kimura et al. (1984), in which *Pinus* (70.7%) prevails and *Tsuga* and *Alnus* comprise 14.0% and 3.1%, respectively, had somewhat lower diversity than that in Miki’s collection comprising of *Pinus* (34.3%), *Tsuga* (32.9%), *Picea* (4.6%), *Abies* (4.2%), and *Alnus* (5.1%). This is consistent with the higher diversity of Miki’s macrofossil collection. Furthermore, the assemblages described by Minaki & Matsuba (1985) are characterized by an absence of subalpine conifers, but diverse and abundant wetland taxa, whereas Miki’s collection includes subalpine conifers and relatively few representatives of wetland plants.

The absence of subalpine conifers in the assemblages

of Minaki & Matsuba (1985) could conceivably be ascribed to differences in the sedimentary environments and paleo-altitude of the vegetation from which the fossils have been derived, although differences in climatic conditions should also be considered. Difference in sedimentary environment is suggested by the dominance of wetland plants in the assemblages described by Minaki & Matsuba (1985) that may have developed in and around lowland wetlands under cool temperate conditions. Miki’s collection is characterized by the occurrence of trees that typically grow along valleys, such as *Cercidiphyllum*, *Pterocarya*, *Alnus hirsuta*, and *Carpinus cordata*, which indicates that assemblage components were transported by rivers from higher altitudes where subalpine conifers grew.

In contrast, the fossil assemblage in the “Tado (upper)” bed of MIS 3 comprise exclusively cool temperate plants with characteristic abundance of wetland herbs and has its origin in and around the wetland site of fossil deposition. Pollen profiles from Kamiyoshi Basin in Kyoto Prefecture (Hayashi et al., 2009) and the BIW95-4 drilling core in Lake Biwa (Hayashi et al., 2010) (Fig. 1) indicate that vegetation in central Kinki region in ca. 40 ka was dominated by deciduous broadleaved trees including *Quercus* with temperate conifers such as Cupressaceae, *Cryptomeria*, and pinaceous conifers. Mixture of cool temperate conifers and deciduous broadleaf trees in Miki’s samples is consistent with paleovegetation reconstructed from those profiles.

2. Phylogeographic significance of fossil assemblages in the LGM

Minaki & Matsuba (1985) reconstructed mixed coniferous and deciduous broadleaf forests that developed under cool temperate conditions, in which *Larix kaempferi*, *Pinus koraiensis*, and *Tsuga sieboldii* are major elements, and deciduous broadleaf trees such as *Fagus crenata*, *Betula grossa*, and *Quercus serrata* are mixed as minor elements. They compared the composition with the paleovegetation maps prepared for full glacial stages by Kamei and RGBWG (1981) and Tsukada (1984). Paleovegetation in lowlands around the northern part of the present-day Ise Bay has been reconstructed as deciduous broadleaf forest by Kamei and RGBWG (1981) and temperate conifer forest by Tsukada (1984). The results of pollen analysis indicating a dominance of *Tsuga sieboldii* type and *Pinus* subgen. *Haploxylon* and macrofossils with numerous specimens of *Chamaecyparis obtusa* show an extension of temperate conifer forests around the study site. However, the presence of diverse temperate deciduous

broadleaf tree taxa in both macrofossil and pollen assemblages indicate the existence of mixed temperate conifers and deciduous broadleaf forests, that Tsukada (1984) assumed to have been distributed in coastal areas along the Pacific Ocean.

Although Minaki & Matsuba (1985) did not refer to Miki (1956, 1957) describing the presence of subalpine taxa, we ascertained the occurrence of subalpine conifers such as *Abies veitchii*, *Picea jezoensis* var. *hondoensis*, and *Tsuga diversifolia*, which are classified as components of boreal conifer forests in Tsukada's (1984) vegetation map. We speculate that if the boundary between the temperate and boreal coniferous forests was located at 400 m a.s.l. at a latitude of approximately 35°N, as proposed by Tsukada (1985), boreal coniferous forests may have been distributed in the upper reaches of the Tado River, and that sediments including macrofossil assemblages from areas up to 600 m a.s.l. in the Yoro Mountains may thus have been transported to lower-lying areas. Moreover, on the basis of the presence of specimens derived from diverse temperate tree flora, we suspect that mixed temperate conifers and deciduous forests were also distributed along the upper reaches of the Tado River, at least in the close proximity to the river itself. This evidence indicates that boreal coniferous forests were more contiguous with mixed temperate conifers and deciduous broadleaf forests than has previously been suggested by Tsukada (1984, 1985).

Tsukada (1984, 1985) also described zonal changes in biomes in the central and western regions of Japan, characterized by a transition from mixed temperate conifer and deciduous broadleaf forests along the Pacific coast, via temperate coniferous forests in the lowlands, to boreal coniferous forests in the montane zone. However, plant macrofossil and pollen records for the LGM indicate that the distribution of vegetation was determined largely by geomorphology rather than by altitudinal gradients. In this regard, Nishiuchi et al. (2017) suggested that, in the Chugoku region, subalpine trees were often predominant on slopes at lower altitudes, whereas temperate broadleaf trees commonly occurred on gentle slopes surrounding wetlands at higher altitudes. Furthermore, Momohara et al. (2016) reported the coexistence of warm-temperate plants and subalpine conifers at the same altitude in a lowland area in the central Kanto region and indicated the distribution of subarctic plants overlapped extensively with that of temperate plants in central and western Japan. These authors also suggested that water stress during the LGM may have been higher than that at present, and that this effectively confined temperate broadleaf trees

to humid locations, thereby relieving conifers from competition with broadleaf trees and thus promoting their expansion into drier areas in warmer regions.

During the LGM, when the sea level fell to between -120 and -130 m a.s.l., the inland Tado site was located mid-way between the Pacific Ocean and Japan Sea coasts (Fig. 1B). On the basis of the LGM (ca. 20 ka) pollen assemblages distributed among four sites in the lowlands around Ise Bay, Ooi (2016) classified predominant pollen physiognomical classes as *Picea*, *Abies*, and *Tsuga* dominant class at three sites and *Pinus* dominant class at one site, indicating that pinaceous dominant forests prevailed in and around Ise Bay. However, the plant macrofossil assemblages characterizing the Tado site indicate that glacial refugia of temperate broadleaf trees, including Paleogene-Neogene relicts such as *Pterocarya*, *Cercidiphyllum*, *Corylopsis*, *Ostrya*, and *Stewartia*, may have extended to inland areas, possibly along the river. This scenario is consistent with the diversity of the present-day local flora around Ise Bay, which includes the "Tokai hilly land element" comprising numerous local endemic, semi-endemic, and relict taxa (Ueda, 1989).

Plant macrofossil assemblages characterized by a diversity of temperate plants during the LGM, such as those at the Tado site, are scarce in central and western Japan. This is because most fossil assemblages tend to be autochthonous, preserved in peat deposits and comprising a relatively small number of species. Among the last glacial assemblages reported from the Kinki region, the Itai-Teragatani site in Hyogo Prefecture (Ooi et al., 1991) and the Tonda site in Osaka Prefecture (Itihara, 1960; Itihara & Kigoshi, 1962)(Fig. 1) were dated to the LGM based on the stratigraphic position with tephra and radiocarbon dating. Assemblages in peat deposits above the Aira-Tn tephra at the Itai-Teragatani site comprise specimens from only five woody taxa, including *Picea* sect. *Picea* and *Betula platyphylla*, along with numerous types of wetland plants. The assemblage from Tonda comprises 10 temperate taxa, including *Abies homolepis*, *Acer miyabei*, *Carpinus cordata* (as *C. erosa*), and *Ostrya japonica* (Itihara, 1960), and the composition of diverse deciduous broadleaf trees is similar to that seen in the assemblages from the Tado site. Radiocarbon dating of the assemblage puts their age at 26,000 ± 800 yBP (Itihara & Kigoshi, 1962), which is calibrated to 31,860–28,680 cal BP. Pollen profile from Lake Biwa indicates that deciduous broadleaf tree pollen maintained higher percentages until ca. 27 ka and decreased in percentage in the later stage of the LGM (Hayashi et al., 2010). However, deciduous broadleaf tree pollen types, including those of *Fagus*

and *Quercus*, account for high percentages in arboreal pollen at several sites throughout glacial maximum stages, even in inland areas in central and western Japan (Ooi, 2016; Nishiuchi et al., 2017). Accordingly, in order to reconstruct the temporal and spatial distribution of refugia of temperate trees during the last glacial stage, it will be necessary to reinvestigate and date further plant macrofossil collections, including those stored in museums.

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