

Original article

# Gamal E. B. El Ghazali<sup>1</sup> and Wai'l E. Abd Alla<sup>1</sup>: Pollen morphological study on *Nymphaea lotus* L. (Nymphaeaceae) with emphasis on zonisulcate apertures

Gamal E. B. El Ghazali<sup>1</sup> and Wai'l E. Abd Alla<sup>1</sup>:スイレン科 *Nymphaea lotus* L. の花粉形態と zonisulcate 発芽口

**Abstract** Pollen morphology of *Nymphaea lotus* L. (Nymphaeaceae) was described by light and scanning electron microscopy. Pollen grains extracted from different types of stamens (including petaloid ones) of *N. lotus* exhibited a wide range of pollen morphological variation in form, size, aperture, and sculpture. Special emphasis was given to the presence of the primitive monocotyledonoid aperture type referred to as 'zonisulcate'. The significance of this aperture type in delimiting the phylogeny of the family was discussed. The family Nymphaeaceae was compared with other dicotyledonous (Eupomatiaceae and Monimiaceae) and monocotyledonous families (Araceae, Arecaceae, Liliaceae, Philydraceae, Rapateaceae, and Xanthorrhoeaceae) possessing zonisulcate pollen grains. No direct relationships were considered to exist between Nymphaeaceae and these families when the present pollen morphological study was combined with other morphological characters as revealed by previous taxonomic treatments.

**Key words:** intraspecific variation, Nymphaeaceae, pollen morphology, primitive angiosperms, zonisulcate apertures

**要旨** *Nymphaea lotus* L. の花粉形態を光学顕微鏡および走査型電子顕微鏡をもちいて記載した。花弁状のものを含む様々な雄ずいから花粉粒を採取して比較したところ、その形や大きさ、発芽口、彫紋には大きな変異が認められた。また原始的な単子葉植物に見られる zonisulcate 型の発芽口が認められた。これをもとにスイレン科を、zonisulcate 型の発芽口をもつ他の双子葉植物(エウポマティア科、モニミア科)および単子葉植物(サトイモ科、ヤシ科、ユリ科、タヌキアヤメ科、ラパテア科、ススキノキ科)と比較した。しかし他の形態学的な特徴とあわせて考慮しても、これらの分類群の間に直接的な類縁は認められなかった。

キーワード：花粉形態、原始被子植物、種内変異、スイレン科、zonisulcate 型の発芽口

## Introduction

Nymphaeaceae is an aquatic dicotyledonous family with stout perennial rhizomes, floating leaves, and many petals which pass gradually into numerous stamens (Clapham et al., 1987). This family has been placed in different orders, viz., Ranales (Caspary, 1891), Rhoeadales (Bessey, 1915), Magnoliales (Tippo, 1942), and Nymphaeales (Cronquist, 1981), but is generally accepted as one of the most primitive families of angiosperms. This treatment is also supported by pollen morphology (Jones & Clarke, 1981).

The family Nymphaeaceae may represent a group of unrelated genera and is often split into several families: Nymphaeaceae, Barclayaceae, Euryalacaceae, Cabombaceae, Nupharaceae, and Nelumbonaceae. Moreover, the genus *Nelumbo* is often placed in a different order,

Nelumbanales (Takhtajan, 1969).

Nymphaeaceae is diverse in pollen morphology and holds a key position palynologically (Nilsson & Praglowski, 1992: 304). It possesses the monocotyledonoid type of pollen grains (Erdtman, 1963: 188), rendering its assignment to dicotyledons difficult on the basis of pollen morphology (Traverse, 1988: 261).

According to Erdtman (1952), the pollen grains of genera of Nymphaeaceae are tricolpate (*Nelumbo*), non-aperturate (*Barclaya*), 1-sulcate (*Brasenia*, *Cabomba*, and *Nuphar*), or 2-, 3-, 4- or zonisulcate.

The genus *Nymphaea* is of cosmopolitan distribution and exhibits many unique pollen morphological features that deserve special consideration. Interspecific pollen morphological variations exist among *Nymphaea* species which appear to represent intermediate stages in

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the evolution of the family Nymphaeaceae (Walker, 1974: 1123).

The main objective of the present work is to closely examine the unique pollen aperture of *Nymphaea lotus*. The taxonomic significance of the observed characters is briefly reviewed.

### Material and methods

Pollen grains contained in various types of stamens (petaloid and otherwise) were collected from different plant populations of *Nymphaea lotus* in Kosti-Rabak (Sudan) along the White Nile (El Ghazali 1, 2, 3, Kosti, Jan. 1997, KHU). They were extracted, subjected to Erdtman's (1960) acetolysis method for recent pollen grains, and stained with basic fuchsin. The voucher slides were mounted in glycerol, sealed with nail polish, and deposited at the National Centre for Research, Khartoum, Sudan (Wai'l 79, Kosti, 5 Feb. 1997). To measure pollen parameters, polar axis length (P) and equatorial diameter (E), at least 50 symmetrical unbroken grains, that were fully expanded in equatorial view, were used. Light microscopy (LM) was done with a Zeiss microscope using a phase contrast (Ph) objective lens of  $\times 100$  magnification and an eyepiece of  $\times 10$  magnification. Scanning electron microscopy (SEM) was done with a JEOL JSM-7200 scanning electron microscope, using an accelerating voltage of 25 kV and 8 mm working distance.

### Results

#### Pollen morphology of *Nymphaea lotus* L. (Figs. 1, 2)

*Form.* Single, but sometimes 2–4-celled.

*Polarity.* Iso-polar or para-isopolar (Fig. 2A).

*Symmetry.* Bilaterally symmetric.

*Shape in polar view.* Circular or elliptic.

*Shape in equatorial view.* Biconvex or broadly elliptic with rounded, acute, or emarginate ends.

*Shape class.* Sub-oblately.

*Dimensions.* P = 37.45  $\mu\text{m}$  (29.47–42.37); E = 47.93  $\mu\text{m}$  (42.37–55.26); P/E = 0.78.

*Pollen class.* Zonisulculate.

*Apertures.* Single ring or band-like (Fig. 1B) sulculus, continuously encircling the grain at or parallel to the equator and slightly shifted towards the distal pole (Fig. 2A); fairly broad, narrow, or slit-shaped; margins usually thickened and sometimes thicker along one side (proximal) (Figs. 2C–2F); membranes smooth or with baculate, papillate, or scattered minor warts (Fig. 2B); edges not sharply defined, ragged or tattered.

*Exine.* Imperforate or perforate; perforations distinct or indistinct, regularly or irregularly distributed (Fig. 1D), sometimes concentrated near the sulculus (Fig. 1F),

of varying diameters, dense or widely spaced (Fig. 2A), sometimes provided with grooves or thin areas (Fig. 1A).

*Columellae.* Absent or indistinct.

*Sculpture.* Psilate or scabrate; scabrae distinct with LM, of varying diameters and sometimes elongated, irregularly distributed.

### Discussion

#### Intraspecific variation

The pollen morphology of *Nymphaea lotus* was previously examined by El Ghazali (1993), Bonnefille & Riollet (1980), Walker (1974), and Maley (1970). Their results agree with each other, but never indicated significant variation within species. SEM and LM observation of this study clearly revealed the existence of distinct and recurrent intraspecific pollen morphological variation in *N. lotus*, that was not recognized by the previous studies. The sulculus of this species may be ring-like or band-like and oriented equatorially or rarely distally with well-defined or ill-defined edges and smooth or warted membranes (Figs. 1, 2). This species is also heterogeneous in other morphological characters. Worsley (cited in Verdcourt, 1989) also reported considerable intraspecific variations in *N. lotus* in the number of petals, stamens, and carpels, as well as in the size of flowers and leaves. Further studies are necessary to clarify the sources of variation (within and between stamens, plants, or populations), which was not the objective of the present study.

#### Zonisulculate apertures

A considerable number of pollen morphological terms were used by various authors to describe the apertures of *Nymphaea*. The terms encountered to corroborate our observation on the apertures of *Nymphaea* include circumploid and ring-furrow (Traverse, 1988), equatorial-sulculate and sulcate-operculate (Thanikaimoni, 1978), zonisulculate (Walker & Doyle, 1975), monocolpate (Nair, 1970), monosulcate (Erdtman, 1947), porate-operculate (Faegri & Iversen, 1950), syncolpate (Iversen & Troels-Smith, 1950), zonate (Wodehouse, 1935), and zonisulculate (Erdtman, 1952).

In the present study, the term 'zonisulculate' (sensu Erdtman, 1952) is used to describe the aperture type of *Nymphaea lotus*. This term is of wide usage and devoid of unnecessary syllables and perhaps covers most of the primary characters needed for the precise taxonomic identification and phylogenetic understanding.

In dicotyledons, the zonisulculate aperture type occurs in Nymphaeaceae, Eupomatiaceae, and Monimiaceae (Erdtman, 1952; Walker, 1974). Bessey (1915), Takhtajan (1969), Hutchinson (1973), Stebbins (1974),

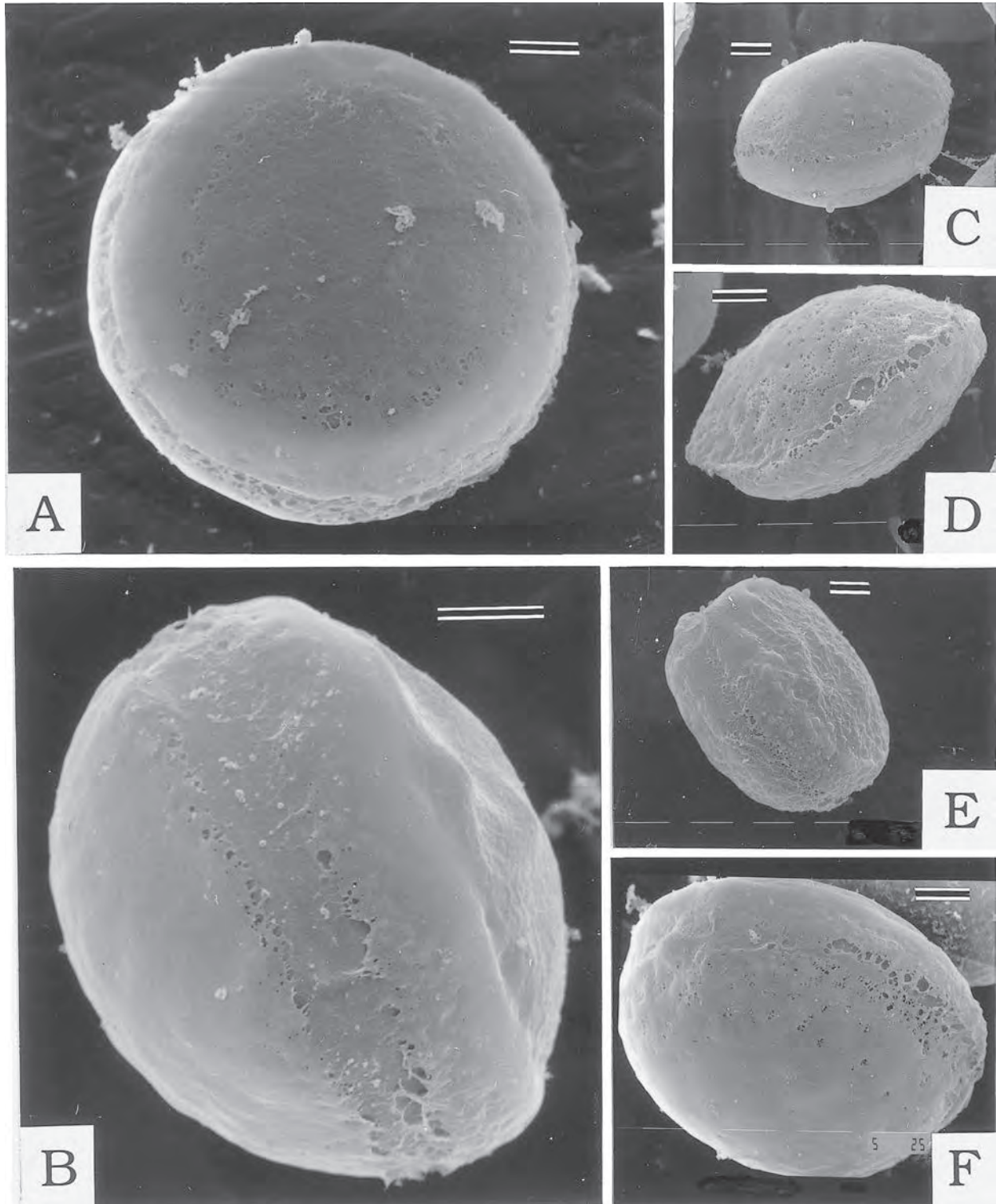


Fig. 1 Pollen grains of *Nymphaea lotus* L. (Jan Berge, SEM 15/98, Bergen). Scale bars = 5  $\mu$ m. — A: Polar view showing psilate sculpture and an exine provided with grooves or thin areas. — B: Equatorial view showing a band-like sulculus with ragged or tattered edges. — C–F: Oblique equatorial views showing a ring-like sulculus. — C: Sculpture is psilate-perforate. — D: Sculpture is psilate and densely perforate. — E: Sculpture is scabrate. — F: Sculpture is psilate with perforations more or less concentrated near the sulculus.



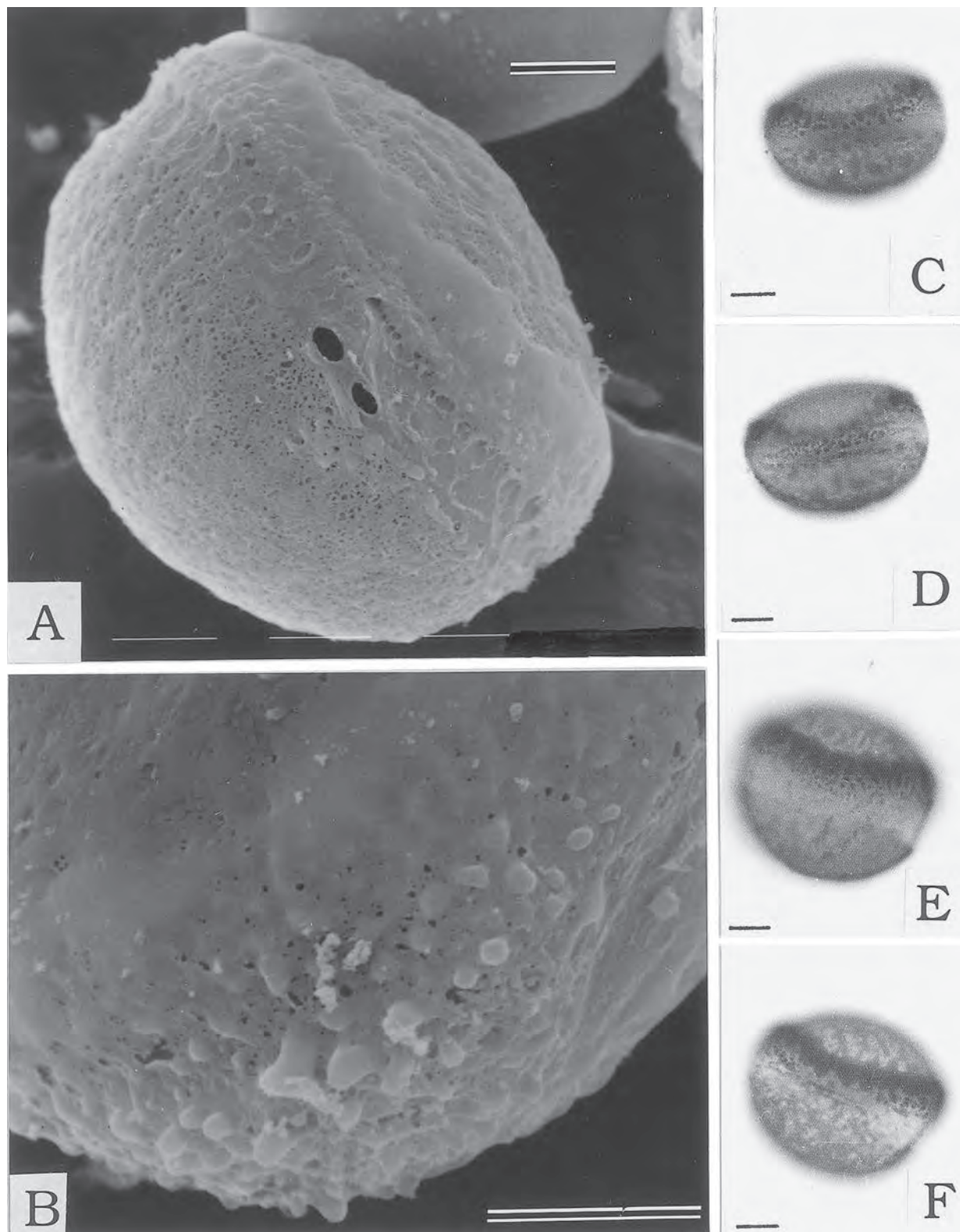


Fig. 2 Pollen grains of *Nymphaea lotus* L. (Jan Berge, SEM 15/98, Bergen and Wai'l 78, Kosti, 5 Feb. 1997). — A–B: SEM. Scale bars = 5 µm. — A: Equatorial view showing scabrate sculpture, a band-like sulculus, a densely perforate exine, and parasopolar grains. — B: Detailed micrograph showing the sulculus membrane provided with baculate, papillate, or scattered minor warts. — C–F: LM. Scale bars = 10 µm. Equatorial views with scabrate sculpture and sulculus margins thicker along the proximal side.

and Cronquist (1981) placed Nymphaeaceae in different order(s) from that of Eupomatiaceae and Monimiaceae, hence excluding any possible close relationships between these families. In monocotyledons, zonisulcate apertures exist in Liliaceae, Xanthorrhoeaceae, Phillydraceae, Araceae, Arecaceae, and Rapateaceae (Goldberg, 1989). According to Takhtajan (1969), the first three families belong to Liliales, whereas the other families belong to Arales, Arecales, and Commelinales, respectively. On the other hand, Takhtajan (1969) regarded Nymphaeaceae to exhibit definite links with Alismales none of whose members possesses zonisulcate pollen grains (Goldberg, 1989). It seems that the unique zonisulcate pollen aperture type of Nymphaeaceae and that of the other families have originated parallelly.

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書 評：小野 昭・小池裕子・福澤仁之・山田昌久・2000．環境と人間 自然の中に歴史を読む．179 pp. ISBN 4-254-18005-5. 朝倉書店．2900 円 + 税．

本書は東京都立大学教養部の学際系講義の教科書にすることを想定してつくられたということである。題名のとおり人を取り巻く自然環境の変遷と人類の歴史の関連性を総合的に捉えようとしている。いくつかの短い総論以外は、具体的な研究例が多岐にわたって示されている。基礎編・通史編・環境と歴史の3部に分かれており、基礎編は、自然史と人類史、堆積作用と環境、食料資源環境と人間、遺跡形成論からなり、通史編は、グローバルな自然環境変化と人類進化の関係、狩猟・採集民は環境の影響の一方的な受け手か、後氷期の環境の多様化と定住社会の工夫、農耕の出現と環境を扱い、環境と歴史は、環境という用語とその条件、研究の精度の向上と人間、環境と歴史からみる「地球温暖化」で構成されている。そのほかところどころにコラムがちりばめられ、最後には用語集もある。それぞれの節は独立しているの、どこから読んでもよいだろう。

それぞれの筆者の得意分野を中心に据えており、内容も、時間・空間のスケールもさまざまな例によって、いろいろな側面から人間とそれをとりまく環境との関係が描き出されている。人類の進化と気候変動との関係のように長い時間を扱ったものもあれば、諏訪高島城の築城を記録した諏訪湖底

堆積物のような短い時間を扱ったものもある。狩猟・採集民の生活を扱ったものもあれば、食料資源に着目したものもある。河内平野の形成と弥生時代農耕集落の展開のように環境変遷と人間活動の関係を議論したものもある。

このように内容が多岐にわたることもあって、全体の統一感はあまりない。具体的な研究例が多いため、図も引用が多く不揃いで、なかには見づらいものもある。途中に挿入されるコラムや巻末の用語集も、やや中途半端で散漫な印象を強めてしまったように感じた。

そのほか、主題とはあまり関係がないが、Taphonomyに対する「埋蔵学」という訳の提唱もある。最近はタフォミーと仮名書きすることが多いようだが、「埋蔵学」は中国で実際に使われているということで、ためしに使ってみてもよいかなと思わせる訳である。

著者に植生史研究を主とする人がいないため、植物に関わる部分はやや希薄ではある。また、人間を取り巻く環境を意識しているものが多い。しかし、かえってそれぞれの例を植物の側から研究するならどうするかと考えさせられた。それは、植生史研究のいろいろな方向への展開を示唆しているように思えた。

(大井信夫)

書 評：化石研究会編．2000．化石の研究法．採集から最新の解析法まで．x + 388 pp. 共立出版，東京．ISBN 4-320-04635-8．本体 8500 円．

第四紀の化石の研究方法は、『第四紀試料分析法1・2』(日本第四紀学会編，1993．東京大学出版会，東京)で一通り網羅されている。しかしここでは化石の研究は、あくまでも第四紀学という学際的な学問の一部として扱われており、個別の生物群を第四紀という枠の中で研究するうえでは問題はないものの、生物の進化を直接扱う正統的な古生物学の観点からは物足りない点も多々あった。

本書は1971年に出版された『化石の研究法』をまったく新たに書きあらためたものであり、いわゆる古生物学の対象となる生物群とその研究方法をほぼ網羅したものである。対象とされている分類群は、細菌類からはじまって、動物化石、植物化石、および生痕化石まで、ほとんどの生物群とその痕跡に触れている。たとえば微少な植物化石では、概論からはじまって、石灰質ナンノ化石と、珪藻類、珪質鞭毛藻類、渦鞭毛藻類、花粉、植物珪酸体を取り上げられている。また大型植物化石では、概説のあと、葉・シュート、鉱化化石、印象化石、球果・果実・種子、材を対象として標本作製方法や

形態観察法が紹介されている。項目によっては、より具体的な研究例や方法に触れており、たとえば材の項ではシダ植物の茎と根における維管束の復原が紹介されているほか、化石と比較対照するための現生植物のさく葉標本やクリアード・リーフ標本、木材標本の作り方が記述されている。

またこうした個別の生物群の研究法をのべた各論の前には、基礎編として野外調査と室内処理の記述があり、産状の記載や採集、クリーニング、薄片作製、複製標本の作製、統計処理、標本の整理と保管、文献検索などに簡単に触れている。また各論のあとには、形態解析や鉱物・無機分析、有機分析といった機器をもちいた分析方法が紹介され、最後の章では旧版以降に発展した研究方法として、分子化石の研究法から、コンピュータ・シミュレーション、多変量解析法、系統復原法、ロコモーション復原法、飼育実験などが紹介される。

このように本書は古生物学の正統的な研究方法と考え方を紹介するものであって、第四紀の化石を扱っているものにとっても資するところが大きい。

(能城修一)