Three Centuries of Protozoology: A Brief Tribute to its Founding Father, A. van Leeuwenhoek of Delft*

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SYNOPSIS. It was exactly 300 years ago this month (August 1974) that the 17th century modest draper from Delft, Holland-Antony van Leeuwenhoek-discovered protozoa. Describing them, often with amazing accuracy considering the optical equipment he was using (simply a home-made "glorified" hand lens), in letters to the Royal Society of London, he established himself, certainly, as the founding father of protozoology. It is particularly appropriate for an assemblage of protozoologists to pay homage to this intrepid "philosopher in little things," a man with an insatiable curiosity about his wee animalcules, on the tricentenary of his discovery of them, since it was an event of such long-lasting significance.

Index Key Words: Leeuwenhoek; discovery of protozoa, tricentenary; protozoology and microscopy, history.

T seems particularly fitting, at this very meeting, to pay homage to our founding father, Antony van Leeuwenhoek (Fig. 1). It was exactly 300 years ago—possibly this very week in August of 1674-that he discovered protozoa with his remarkable hand-lens "microscope." Thus he started a science-'protozoa-watching"-which has grown and expanded mightily, until today it enjoys the professional attention of thousands of biologists around the world, embracing workers whose particular specialties may range from chemotherapy of malaria to dating oil beds by means of foraminiferan fossils, from electron microscopy of ciliates to investigation of DNA in dinoflagellate nuclei, from biochemical research on carcinogens to use of protozoa as biologic indicators in pollution studies.

The Man from Delft

But Leeuwenhoek was the 1st. He was the 1st human being who combined adroit inventiveness, insatiable curiosity, and extremely good eyesight in successful detection of species of protozoa, watching them in action over a period of time, reflecting on what he saw, and then looking further into such unbelievable sights for which there was no precedent in his times. He was also perceptive enough as a scholar to appreciate the value of recording his observations for posterity, sending more than 200 handwritten (sometimes perhaps more aptly described as handscribbled), conversation-like letters, occasionally illustrated (with the help of a draughtsman) to whoever was Secretary of the Royal Society of London at the time. Many of these were published in part or in full, after their translation into English, in the Philosophical Transactions. Largely on the basis of this happy rapport, Leeuwenhoek's work became well enough known in scientific circles to bring about his election, in 1680, as a regular ("Foreign Members" hadn't yet been invented) Fellow of the Royal Society, a much-deserved honor which brought him lasting pleasure.

Born in Delft, Holland, on 24 October 1632, into a large family, Antony certainly did not seem destined for a scientific career. His father was a basket-maker; his mother's family were brewers. At 16, after little formal schooling, he himself

learned the business of a draper in Amsterdam; a few years later he set up shop in Delft and never left home again. He also held the post of Chamberlain to the Sheriffs of Delft, and did a little surveying on the side. He was married twice, outliving both wives and all children except a loving daughter, Maria. Finally, mention might be made of one other municipal function: Leeuwenhoek was the town's official wine-gauger, assaying all wines and spirits entering Delft (a task perhaps contributing to his longevity?!?).



Antony van Leeuwenhoek, "Father of Protozoology. Drawing based on a portion of the widely reproduced mezzotint engraving by the Dutch artist Johannes Verkolje, a work produced later in the same year, 1686, in which he painted in oils the equally well-known original portrait of his great contemporary.

* This commemorative note represents the substance of a short address delivered, on special invitation of President William C. Marquardt, before the Society of Protozoologists at the 27th An-

Marquardt, before the Society of Protozoologists at the 27th Annual Meeting, Middletown, Connecticut, 13-16 August 1974. An abstract (15) had been submitted "By Title." † I am deeply indebted to Miss Lois Reid for her excellent and artistic production of all of the figures, and to Mr. Fred Dickson for his indispensable photographic assistance. I wish to acknowledge with gratitude the assistance of National Science Foundation grant GB-41172.

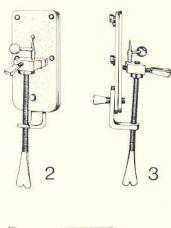
His Simple "Microscope"

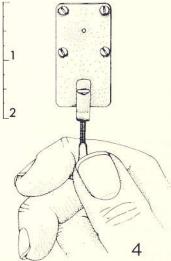
Leeuwenhoek's simple "microscope" is not to be thought of as the Ist microscope, nor is he, in my opinion, to be given the title of "father of microscopy"-although he is properly known as the "father" of protozoology, bacteriology, parasitology, histology, hematology, and a host of other subjects requiring microscopy as their major tool of descriptive investigation. But Galileo, Campani, the Janssens, Borel, Malpighi, Robert Hooke, and others were earlier (or contemporary) microscopists and, furthermore, worked with or in the development of the compound microscope with its multiple-lens system, destined to give rise, ultimately, to our Leitz, Zeiss, Spencer, and other models of today.1 Leeuwenhoek's "scopes" were simply "glorified" hand lenses, or, as he preferred to call them, "magnifying-glasses." But it is true that they were mounted with a well-ground and skillfully polished lens, allowing a magnification sometimes up to an estimated maximum of 300 diameters (or perhaps even more: see 32).

Leeuwenhoek made hundreds2 of his instruments, some of gold and silver but most of brass and not at all ornate or larger than necessary, few-surprisingly enough-measuring much more than 3 inches in length. The emphasis was always on the resolving power of the single tiny biconvex lens, ground only by the man himself: it may have reached a figure of 1.0 μm, according to Rooseboom (31), better than that of many compound microscopes for years beyond his time. The mechanical 'gears" on his microscope (Figs. 2-4) were ingenious, too, allowing, in effect, coarse and fine adjustments, a mechanical stage, etc. For viewing living protozoa, a tiny, handmade capillary glass tube was probably mounted or stuck some way onto the tip or point (possibly modified?) of the object-carrier, then manipulated into proper position just opposite the aperture leading to the lens, with the viewer's eye appressed to the opposite side of the opening. Whew, what patience and what eyesight, to see what he saw!

Dobell (17) and others have concluded that Leeuwenhoek must also have discovered some simple means of darkfield illumination, in order to have seen so clearly flagella on bacteria, etc. Another point about the man, often overlooked, is that he was the 1st microscopist to actually *measure* the objects he saw.

I hardly need to mention to readers of this journal that we have the late Clifford Dobell of England, a pioneering protistologist himself in many ways, to thank for the 1st truly lasting biography of the "father of protozoology" (17). Carried out with dogged persistence, often into the wee hours of the night, with unimpeachable precision and exactitude, and, above all, with an ardent love for his subject, Dobell's completion of the task resulted in an authoritative yet exciting and intimate account which, I am pleased to say, has been on college reading lists in fields far from protozoology and on bedside bookshelves





Figs. 2-4. Leeuwenhoek's microscope. Drawings, printed at ~\mathbb{\gamma}_3 actual size, represent the back (Fig. 2), side (Fig. 3), and front (Fig. 4) views of the author's brass copy of an original instrument. In all figures note: the various "gears," briefly explained in the text, and the minute aperture leading to the tiny biconvex lens (not shown) housed between the 2 brass plates. The small size of the instrument is emphasized by that of the author's fingers included in Fig. 4. Scale, reduced by ~\mathbb{\gamma}_3, is in inches.

of people who have never enjoyed a course in science. Incidentally, the original hardback 1932 (purposely published on the tricentenary of Leeuwenhoek's birth) edition may be difficult to find, but 2 (and perhaps more) inexpensive paperback editions have appeared since; and I believe that the Dover edition of 1960, at least, is still easily available. In more recent years, Schierbeek's (32-34; and see 24a) contributions have represented notable additions to our knowledge of the pioneering Dutchman and his works—now surely never to fall into near-oblivion again.

His Letters on Protozoa

Dobell (17) conveniently numbered Leeuwenhoek's numerous letters written over a span of some 50 years. Letter No. 6, dated 7 September 1674, contained the 1st observations ever made on protistan (sensu lato) and other very minute aquatic plant and animal forms (24). Here, for example, we find the now classical succinct yet almost poetical, characterization of what probably was Euglena viridis: "green in the middle, and before and behind white." Ciliates were surely seen, too, in

¹The development of (descriptive) protozoology has, obviously, depended on the parallel, or antecedent, development of the field of microscopy (7, 12, 20, 38). It may be interesting to note, in passing, that in the 1950's nearly 3 full centuries after Leeuwenhoek and his "magnifying-glass," the late Dr. Ludwig H. Bretschneider, a modern-day protozoologist, pioneered in the early use of the transmission electron microscope in the very same town of Delft (4).

² Alas, nearly all of which have disappeared over the centuries. Dobell (17) hints that skulduggery was involved in the case of the beautiful collection once the property of the Royal Society. Figures 2-4 are drawings of a very accurate brass copy (typical size, construction, etc., but with a lens of only 30× magnification) kindly given to me some 15 years ago by Dr. A. Schierbeek of Amsterdam.

his lake-water samples: only a few, but the 1st to be observed. What a contrast with the multifarious forms recognizable today—nearly 7,500 described species of ciliates alone, plus probably countless thousands still awaiting discovery (14)!

The celebrated letter (Dobell's No. 18), which Kent (21) and many other protozoologists and textbook writers subsequently have long been erroneously citing as the "first" on protozoa, was dated 9 October 1676. Actually, 1 or 2 additional notes had appeared on microorganisms between that time and September 1674, date of the historic and truly 1st report. But Dobell's (17) accurate and lengthy account (occupying 54 pages of his book), a personal translation from the original and difficult Dutch of that century, does establish this letter for all time as the most extensive of Leeuwenhoek's early observations on microscopic organisms abounding in such natural and experimental habitats as fresh-water, sea-water, rain-water, pepper-water, vinegar, ginger-water, clove-water, and nutmegwater. And we now have Schierbeek's (33) illustrated 31-page booklet (unfortunately entirely in the Dutch language) devoted solely to this October 1676 landmark in the history of pro-

Although the founding father of our discipline never gave latinized names to his discoveries-the artful science of nomenclature did not start up officially until 1758 (11, 25), 35 years after Leeuwenhoek's passing-it is clear that he identified or offered acceptable descriptions of many protozoan species familiar to us today. In his famous Letter No. 18, alone, he offered confirmable data on members of such present-day genera as Vorticella,3 Bodo, Chilodonella, Monas, Cyclidium, Oxytricha, Stylonychia, Colpidium, and probably our ubiquitous friend Tetrahymena (13). Later letters contained recognizable descriptions of additional free-living flagellates and ciliates from soil and aqueous habitats: for example, Chlamydomonas, Haematococcus, Cercomonas, Polytoma, Dileptus, Coleps, Euplotes, Kerona, Trichodina, Cothurnia (in its lorica), Paramecium (in modern times probably the most popular ciliate ever known), and such colonial forms as Carchesium, among the ciliates, and Volvox and Anthophysa, among the flagellates.

Leeuwenhoek described reproduction in *Volvox* in detail; he also gave the 1st account of conjugation in ciliates, although O. F. Müller (28) is usually credited (see 10) with the 1st correct interpretation of the phenomenon not again appreciated until the latter years of the 19th century (by Balbiani, Bütschli, Hertwig, Maupas, and contemporaries), more than 175 years after the good Dutchman's letters on the subject. In the interim, it had been commonly supposed that pairing of ciliates was a second or "longitudinal" (as opposed to "transverse") kind of binary fission.

Curiously enough, Leeuwenhoek never saw, or at least, never seemed to describe in his letters in a recognizable fashion, any living sarcodinids.⁴ He did, however, offer a quite accurate

figure of a foraminiferan test, probably a species of the genus *Elphidium* (Fig. 13), taken from the stomach of a shrimp.

Of at least equal importance to his many charming and accurate accounts of free-living protozoa were Leeuwenhoek's several descriptions of symbiotic or parasitic species, forms found primarily in the digestive tract or feces of hosts which included invertebrates and such vertebrates as the frog, rabbit, and even man. He was the 1st to see what were probably representatives of Opalina, Nyctotherus, Eimeria (in the oocyst stage), Chilomastix, Giardia (in his own stools), Crithidia, and "Trichomonas" sensu lato. Surely some other common protozoan parasites, though again apparently no amebas, were seen, but were either not recorded or too scantily described to warrant definite recognition today.

Leeuwenhoek submitted sketches or drawings of some protozoa with his letters: for example, of species which today we feel may be assigned to the genera Coleps, Vorticella, Volvox, Anthophysa, Carchesium, Cothurnia, Opalina, Nyctotherus, and Elphidium (called "Polystomella" by Dobell). Simple but generally sufficient—if we are generous enough to make some allowances—for such distinctive genera were these particular figures, as may be noted in their reproduction here (Figs. 5-13).

In 1716, according to Dobell (17), Leeuwenhoek penned his last letter to the Royal Society containing any data on protozoological topics. But additional letters on other subjects were written and sent across the Channel even into the year of his death, in 1723; in fact, he was dictating a scientific communication 36 hours before his passing. In due time, the world learned from his devoted daughter Maria (by then 67 herself) that her father and the "father of protozoology" had passed away quietly, in Delft, on 26 August of that year, at the ripe old age of nearly 91.

His Insatiable Curiosity

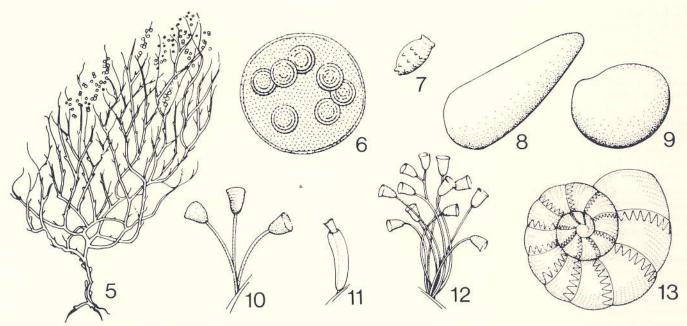
In an abbreviated sketch such as this, many items of great interest in the scientific and personal life of this remarkable yet modest microscopist from The Netherlands have had to be omitted. Although Dobell's (17) book has served admirably as my principal source of information, the reader is also referred to Beltrán (1), Bender (2), Bradbury (3), Bulloch (5), Bütschli (6), Cole (7-9), Corliss (10), Dobell (16), Kent (21), Kudo (22), Lechevalier & Solotorovsky (23), Locy (26), Manwell (27), Nordenskiöld (29), Purtle (30), Richards (30a), Rooseboom (31), Schierbeek (32-34), Singer (35), Singer et al. (36), and Woodruff (37-39) for additional fascinating details in the life of Leeuwenhoek⁵ and/or for the more or less related history and development of microscopy and protozoology.

Of all the outstanding characteristics which Antony van Leeuwenhoek possessed, his insatiable curiosity about the natural wonders of the microscopic world seems to me the single most important trait of this "proto-protozoologist." The same sort of interest in his little "animalcules" has furnished the motivating power for scores of workers in the field since his discoveries of 300 years ago. Today, armed with such optical and mechanical advantages as phase microscopy, microcinematography, electron microscopy, and the sophisticated tools of the biochemist and

^a For this genus, his description included the oft-cited passage which offers such a vivid account of the coiling and uncoiling of the contractile stalk (called a "tail" by Leeuwenhoek) of these fascinating ciliates (see 18).

⁴It is thus rather amusing to note that the cover of the 1960 Dover paperback edition of Dobell (17) is illustrated with drawings of rhizopod amebae "pseudopod-ing" their way across the page! Errors of a rather similar kind may be found in the nonetheless delightful and otherwise most cleverly illustrated classic by Hegner (19); in his Fig. 10 Leeuwenhoek is in possession of a compound microscope (unquestionably Robert Hooke's), being bowled over by the marvelous sight of testaceous amoebae, trypanosomes, and some other species—few of which apparently were ever actually among those described by the Dutch protozoologist, although undoubtedly favorites of Dr. Hegner.

⁵ Space does not permit critical comments on discrepancies, differences, and inaccuracies, which are sometimes quite amazing and occasionally amusing, discoverable in various accounts of the life of Leeuwenhoek. For 1 example, in a very recent biographical sketch, which is deliberately not cited in this paper, the out-of-context statement is made that the Dutch microscopist was, early in life, "appointed janitor in the Delft City Hall, a sinecure which he held for the remainder of his life." (Enough to cause poor Dobell to turn over in his grave!)



Figs. 5-13. [Redrawings (not at a common scale) of original engravings (reproduced in 17) which accompanied publication (in the *Philosophical Transactions* or other outlets) of several of Leeuwenhoek's letters on protozoologic topics. The brief descriptions prethe Philosophical Transactions or other outlets) of several of Leeuwenhoek's letters on protozoologic topics. The brief descriptions presented in quotation marks are excerpts, sometimes slightly paraphrased, from (Dobell's translation of) Leeuwenhoek's more complete accounts of the structure and activities of each organism.] 5. "Like a bough off a tree, with its many twigs...each with extreme tips beset with round transparent globules, which can detach and move about... Its root, fastened to the glass, is the color of oaken wood, and is encrusted with little round granules..." (from Letter No. 150, year 1703). (No doubt the well known arboroid colonial chrysomonad flagellate Anthophysa vegetans, whose stalk is indeed encrusted with brown particles of ferric hydroxide, giving the species the common appellation of "iron-protozoon." 6. "Green round particle... with many little projecting particles... swimming with a rolling motion... and containing little round globules of like structure..." (from Letter No. 122, year 1700). (Surely a species of the spheroid colonial phytomonad flagellate Volvox. Specimen portrayed clearly has seven daughter colonies within it.) 7. "[Barrellike form] whose helly is flat and from which little instruments stick out wherewith it effects its progress... It has little round rolling motion...and containing little round globules of like structure.." (from Letter No. 122, year 1700). (Surely a species of the spheroid colonial phytomonad flagellate Volvox. Specimen portrayed clearly has seven daughter colonies within it.) 7. "(Barrellike form) whose belly is flat, and from which little instruments stick out, wherewith it effects its progress... It has little round globules in its body..." (from Letter No. 144, year 1702). (Very likely a species of the common gymnostome ciliate Coleps.) 8. "The biggest sort of a great company of living animalcules from the gut of a frog..." (from Letter No. 38, year 1633). (Very likely a species of an opalinid, probably of Opalina, an organism common in the large intestine of frogs and toads.) 9. "Not as large [as the preceding] and more rounded... and very few in numbers..." (from Letter No. 38, year 1633). (Probably a member of the heterotrich ciliate genus Nyctotherus), although various authors have implicated the trichostome ciliate Balantidium, to the extent that the latter genus continues to be mistakenly cited as a Lecuwenhoek discovery in textbooks, etc. But Dobell (17), was most likely correct, and nyctotherans easily found today in the frog Rana do indeed, in outline, resemble Lecuwenhoek's drawing.) 10. "Fashioned like a bell... moving very gently on their long tails... yet in an instant contracting... (from Letter No. 149, year 1702). (Vorticelled peritrich ciliates, probably Vorticella, very similar to ones described some 25 years earlier in his celebrated Letter No. 18 (see text), attached to a rootlet of the small aquatic plant known as duckweed. Three solitary individuals are depicted.) 11. "Very small cases, part of its body out..." (from Letter No. 149, year 1702). (Lovely partial description of a species of Cothurnia, a solitary loricate peritrich ciliate which could be confused, superficially, with small tubicolous rotifers, but Lecuwenhoek, who also described the latter with accuracy, recognized the difference. The "wheels" were th

molecular biologist, we continue the quest for more knowledge about these fascinating microorganisms. But it is doubtful if present-day or even future "philosophers in little things" will ever be able to satiate completely their curiosity about the protozoa.

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