

Lighting Van Leeuwenhoek's samples

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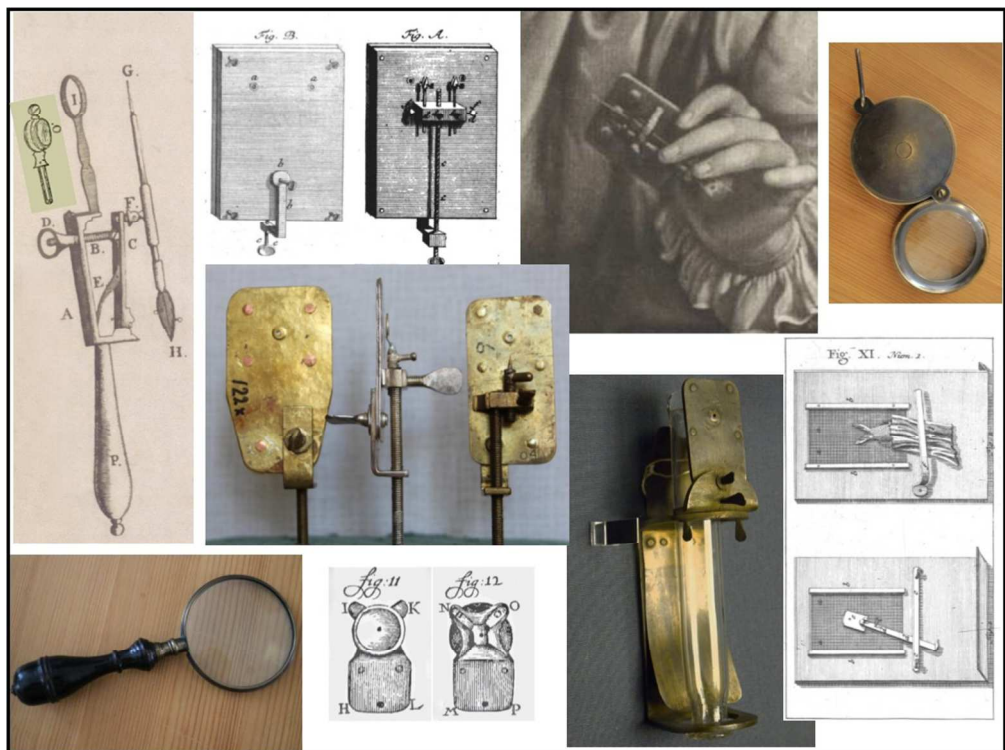
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Lighting Van Leeuwenhoek's samples

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1 Title: Lighting Van Leeuwenhoek's samples

2 Running title: Lighting Van Leeuwenhoek's samples.

3

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14 Keywords: single lens microscopes, Van Leeuwenhoek, opaque samples

15

16 Single sentence summary: The problems associated with viewing opaque samples using
17 Van Leeuwenhoek microscopes are demonstrated, possible solutions are reviewed, and the
18 possible identity of his "particular method of observing" is discussed.

19

1
2
3 **20 Abstract**
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5
6 21 Possible techniques for lighting opaque samples while using Van Leeuwenhoek
7
8 22 microscopes have been tested, and the results are presented in relation to published
9
10 23 material. The design of the microscope causes the sample to be in shadow with any form of
11
12 24 top-lighting. It is therefore suggested that Van Leeuwenhoek's hinted "particular method of
13
14 25 observing" might refer to a different style of microscope as shown in the frontispiece of the
15
16 26 sale catalogue for his microscopes, and available at that time for purchase from sellers of
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18 27 optical equipment.
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3 284
5 29 **Introduction**

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7 30 Despite the many descriptions of his experimental methods scattered throughout his
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9 31 letters, Antoni van Leeuwenhoek has a reputation for being secretive about his experimental
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11 32 methods. This might partly be because the versions of his letters published by the Royal
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13 33 Society (Phil Trans) were generally only edited highlights. If one reads letters by other
14
15 34 scientists published in the same volumes, their methods are also rarely described in detail.
16
17 35 To find more detailed descriptions of some of his work, the reader must either seek out his
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19 36 privately-published collections, most of which can be downloaded as pdf files from the
20
21 37 Internet Archive and other sites (generally in Dutch or Latin), or consult the volumes of
22
23 38 Collected Letters, volumes 1-15 of which can be downloaded as pdf files (DNBL). Of course,
24
25 39 perhaps to avoid plagiarism, he did not describe everything.

26
27 40 Since Van Leeuwenhoek's time, people have speculated about how he lit his
28
29 41 samples. Anyone who has used one (or a copy) will know that they are very effective with
30
31 42 transparent samples. Van Leeuwenhoek commented that they are best used with diffuse
32
33 43 light from the sky, a lamp or a candle, but never the sun because of distortion (Van
34
35 44 Leeuwenhoek, 1694, 1699). He also mentioned using a curved mirror or a magnifying glass
36
37 45 to improve the lighting, and this is probably how he achieved dark field views (Robertson,
38
39 46 2015a; Robertson et al, 2016). However, despite the fact that many of the samples in his
40
41 47 huge body of research were opaque, his microscopes are not very effective with them.
42
43 48 Opaque samples only appear as silhouettes with transmitted lighting, and the level of detail
44
45 49 shown in the drawings that accompanied his letters cannot be seen without top (or reflected)
46
47 50 lighting (see, for example, the drawing of a parasitic wasp, *Aphidius* sp., published by Van
48
49 51 Leeuwenhoek in 1702a).

52
53 52 "Living history", where experts use historical equipment such as agricultural
54
55 53 implements or adopt domestic lifestyles from the past rather than theorizing, can provide a
56
57 54 great deal of useful information and eliminate (or even confirm) theories. Historical
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2
3 55 microbiology is a limited form of this where historically important experiments are repeated
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5 56 with original equipment or accurate copies to test how such experiments could have been
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7 57 done in the days before detailed method descriptions in publications (Robertson, 2015a). It
8
9 58 seems reasonable to expect that repeating selected experiments from Van Leeuwenhoek's
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11 59 work would also shed light on his lighting techniques.
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17 **Materials and methods.**

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19 62 The following magnifiers were used:

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21 63 A facsimile Van Leeuwenhoek microscope (Loncke, 2006a, b) with a magnification of
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23 64 about 65x.

24
25 65 A late 19th century Carl Zeiss Jena "jug handled" compound microscope (Anon,
26
27 66 1906) as a control.

28
29 67 A 20th century botanist's hand-lens with a magnification of 30x. This hand-lens was
30
31 68 chosen to represent the 17th century originals because, like the microscopes sold in
32
33 69 Van Leeuwenhoek's time for viewing opaque samples, its lens is mounted in a simple
34
35 70 metal ring, rather than in a metal plate.

36
37 71 As Van Leeuwenhoek frequently remarked, artificial lighting is necessary for work in the
38
39 72 evenings or bad weather. Photography also imposes small compromises in experimental
40
41 73 design. It has previously been shown (Robertson, 2015A) that similar results are obtained
42
43 74 with different light sources, including a candle. For continuity with previous experiments and
44
45 75 ease of photography, all magnifiers were lit using an LED light and a thin diffusing screen (to
46
47 76 give even lighting for the sensor in the camera), unless otherwise specified.

48
49 77 Photography was done using a Canon EOS M10 digital camera body fitted with a Bresser
50
51 78 microscope adaptor which had been modified by lining the stainless steel inner tub with
52
53 79 black adhesive material to eliminate internal reflection. To simulate the focusing controls of
54
55 80 the two types of microscope, samples for the hand-lens were mounted on a glass rod
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1
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3 81 attached to a macro focusing slide. All photographs are the result of focus stacking between
4
5 82 4 and 15 images to correct for the limited depth of field imposed by the camera.

6
7 83 To allow the moth's wing to be moved between the microscopes and the hand-lens, it was
8
9 84 attached to a glass coverslip using a drop of colourless adhesive.

10
11 85 Sample selection was governed by the availability of Van Leeuwenhoek drawings for
12
13 86 comparison as well as the ability to mount the samples in all three magnifiers. Moth and
14
15 87 butterfly wings were convenient as representatives of the many samples Van Leeuwenhoek
16
17 88 examined, especially because they are transparent at their edges and opaque over the rest
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19 89 of the surface. Similar results were obtained with both, so only the results with moth wings
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21 90 are presented.
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26

92 **Results**

27
28 93 It is not possible to top-light samples using a classical Van Leeuwenhoek microscope
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30 94 (Fig. 1A) because the metal lens mount obstructs the light path, casting a shadow. Several
31
32 95 researchers (Baker, 1739; Harting, 1859; Clay & Court, 1932) have suggested that Van
33
34 96 Leeuwenhoek lit opaque samples from the side, using a lens or curved mirror. However,
35
36 97 while the use of a lens or mirror to focus light on the sample works well for transparent
37
38 98 samples and to achieve dark field microscopy (Robertson et al, 2016), it does not work from
39
40 99 the top or the side. Depending on the strength of the lens in use, the distance between it and
41
42 100 the sample pin when the sample is focused varies from 8mm -1mm with magnifications
43
44 101 between 68x -303x, respectively (Fig. 1A, centre microscope). Light from the side reached
45
46 102 the sample perpendicular to the light path between the sample and the observer's eye, and
47
48 103 gave little or no improvement in visible detail.

49
50 104 It has also been suggested that Van Leeuwenhoek made silver microscopes so that
51
52 105 they could be polished and reflect light onto the upper surface of the sample, or else used a
53
54 106 thin mirror between the microscope and the sample (Baker, 1739; Anderson, 2017). This
55
56 107 was tested by covering the facsimile microscope (leaving a small hole for the lens) with a flat
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3 108 piece of aluminium foil with the highly reflective side facing the sample. Again, this gave little
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5 109 or no improvement. The lens mount is flat and therefore does not focus reflected light onto
6
7 110 the sample. With the lens and sample necessarily so close together, there is no room to
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9 111 curve the lens mount in the manner of the cup-shaped reflector that first appeared early in
10
11 112 the 18th century (Baker, 1739, 1742), but eventually became known as a Lieberkühn.

12
13 113 Fig. 2A shows the backlit wing of a large yellow underwing moth, as photographed
14
15 114 with the facsimile Van Leeuwenhoek microscope. Van Leeuwenhoek's drawing of scales
16
17 115 from a silkworm moth is inset. At the upper edge of the wing, where the light only had to
18
19 116 travel through a single layer, the appearance of the scales in the two images is similar. With
20
21 117 thicker layers of scales, little or no detail can be seen. Figs 2B and C show the same wing
22
23 118 under the Zeiss microscope with top and back lighting, respectively. B resembles A, but only
24
25 119 C shows all of the scales. As Fig. 3A shows, Van Leeuwenhoek was also able to see the
26
27 120 scales covering a butterfly wing (Van Leeuwenhoek, 1678). When the hand-lens was used,
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29 121 results similar to those in Fig 2B were obtained by backlighting the wing. However, it was
30
31 122 also possible to position the light at the front, beside the eye or camera (Figs 3B and C),
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33 123 allowing the surface of the wing and the scales, to be lit. The distance between the hand-
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35 124 lens and the sample was approximately 20mm which permitted a lighting angle greater than
36
37 125 90° because the narrow metal band mount for the lens did not interrupt the light path and did
38
39 126 not cast a shadow.

127 **Discussion**

128 Antoni van Leeuwenhoek is generally remembered for his discovery of
129 microorganisms (Van Leeuwenhoek, 1676), for which the level of magnification delivered by
130 his traditional microscopes (Fig. 1A) was certainly necessary. Aqueous samples are
131 generally transparent, and it is even possible to use dark field lighting (Robertson et al,
132 2016). However, his research covered a wealth of subjects, often with samples that were not
133 transparent, did not require such strong magnification, or both. It is clear from his own words

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2
3 134 (below) that he viewed his microscopes as tools for a particular job rather than objects in
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5 135 their own right:
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7 136 “I have said heretofore how I composed my instruments, which some people would have
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9 137 made far finer and more accurate. I have so far trained myself that I have for many years
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11 138 made the tools that I needed for several matters. And that is why what I required for my use
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13 139 was only made a bit roughly by myself.” (Van Leeuwenhoek, 1689).

14
15 140 Robert Hooke mentioned the impossibility of lighting samples with a single lens
16
17 141 microscope in one of his Cutlerian Lectures (Hooke, 1679):
18
19 142 “The only inconvenience in these kinds of Microscopes, is, that the object is necessarily
20
21 143 brought so near the glass*, that none but such as are transparent, and to be viewed by a
22
23 144 through light are capable of examination by them” (* “glass” in this context in the 17th century
24
25 145 meant “lens”).

26
27 146 Van Leeuwenhoek must have found a way around the problem. His approach to the
28
29 147 apparatus he called an *aalkijker* for viewing blood circulation in eels and various fish shows
30
31 148 that he was willing to adapt his tools as necessary (1689, 1695). The original version of the
32
33 149 *aalkijker* used the same lens holder as his microscopes (Fig. 4A). However, in his second
34
35 150 paper on the subject, he described how, in order to be able to shine more light on his
36
37 151 sample, he removed most of the metal on the lens holder (Fig 4B). To protect his eye from
38
39 152 the light, it was then necessary to add an eye cup. Some writers (e.g. Baker, 1739; Priestly,
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41 153 1772; Harting, 1850) described this cup as a reflector surrounding the lens to direct light onto
42
43 154 the sample, the arrangement which eventually became known as a “Lieberkuhn”. They even
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45 155 suggested that Van Leeuwenhoek should be credited with its invention. However, as pointed
46
47 156 out above, the very small distance between the sample and the lens on a Van Leeuwenhoek
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49 157 microscope does not allow a curved reflector to be used. There is no room. If one considers
50
51 158 Van Leeuwenhoek’s own description (his *figs 11 and 12* are shown in Fig 4B), it is clear that
52
53 159 those authors were looking at the apparatus from the wrong side (Van Leeuwenhoek, 1689):
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3 160 "...screwed on to this instrument is the brass plate into which, again, a magnifying glass has
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5 161 been riveted, as is shown here in fig: 11, HIKL., over which magnifying glass I have soldered
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7 162 a small cup, in order that the eye might the better see the objects, for I had filed away the
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9 163 brass around the magnifying glass as much as it could bear, to bring as much light as was
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11 164 practicable on to the objects that one might wish to see.... on fig: 12. MNOP, where one can
12
13 165 see the same instrument from the other side".

14
15 166 The cup is there to protect the viewer's eye from the light aimed at the sample. The
16
17 167 credit for inventing the "Lieberkühn" belongs to Descartes (1637).

18
19 168 Van Leeuwenhoek was obviously aware of the advantages of being able to light his
20
21 169 sample from all angles. If he adapted his *aalkijkers* to do this, why would he not also adapt
22
23 170 his microscopes for opaque samples? He could possibly have used the reduced lens holders
24
25 171 shown in Fig 4B, but when he needed a weaker lens, is it possible that he was using a
26
27 172 different style of microscope? This might explain the inclusion of something that strongly
28
29 173 resembles a microscope commonly sold at that time for viewing opaque samples (Fig. 5A) in
30
31 174 the frontispiece of the sale catalogue for his microscopes after his daughter's death (Fig. 6H;
32
33 175 Rees, 1747).

34
35 176 Most of Van Leeuwenhoek's microscopes have vanished, and the assumption that
36
37 177 they were all alike is common despite the fact that he clearly adapted his tools as required.
38
39 178 He twice altered his original *aalkijker* (the 3rd time to make viewing easier for visitors by
40
41 179 holding the fish against a piece of glass) but versions 2 and 3 (Figs 4B and C) have not
42
43 180 survived except as written descriptions or illustrations (Van Leeuwenhoek, 1695, 1708; Von
44
45 181 Uffenbach, 1754). The catalogue for the sale of his microscopes (Rees, 1747) mentions
46
47 182 microscopes with two and three lenses, something which some authors have believed to
48
49 183 indicate that he was also using compound microscopes (e.g. Harting, 1850) even though
50
51 184 pictures of such microscopes made at the time clearly show two or three lenses side by side
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53 185 (Fig. 1B and C, Verkolje, 1686; Rees, 1747; Von Uffenbach, 1754). Those microscopes have
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55 186 also not survived, neither have those mentioned in the catalogue as being able to hold two
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3 187 samples, those with a hole to mount a capillary tube in the sample block (Fig. 1C) or those
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5 188 made from gold.

6
7 189 Van Leeuwenhoek frequently hinted (Dobell, 1932) that he had another “particular
8
9 190 method of observing” which he did not allow guests to use, but which allowed him to see
10
11 191 more. The “burning glasses” (e.g. Fig 5B, C) mentioned in the inventory of his house after
12
13 192 his daughter’s death (Geesteranis, 1745) would not have been strong enough for a lot of his
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15 193 work, although they would have permitted top lighting. We can never be certain, but it is
16
17 194 attractive to speculate that he was talking about top lighting samples using the type of
18
19 195 microscope shown in Figs. 5A and 6H. Top lighting reveals a lot of additional detail (Figs 2C
20
21 196 and 3), but it would have been more complicated to set up than his traditional microscopes,
22
23 197 and he was famously impatient with visitors and the time they cost him. In addition, would he
24
25 198 have wanted his visitors to report that for some of his work he was using microscopes of a
26
27 199 type readily available from opticians, even if he improved the lenses?

28
29 200 There was clearly more variation in Van Leeuwenhoek’s magnifying toolkit than can
30
31 201 be seen from the few surviving microscopes. All of the equipment shown in Fig. 6 can be
32
33 202 identified in his letters or the reports of his visitors (Robertson et al, 2015b) except the
34
35 203 magnifier, H, which resembles that shown in Fig. 5A.

36
37 204 The digital abstract for this paper shows Van Leeuwenhoek’s magnifying toolkit as it
38
39 205 might have been.

40
41 206

42 43 207 **Acknowledgement**

44
45 208 The author appreciates constructive comments from the reviewers.

46 47 209 **CONFLICT OF INTEREST**

48
49 210 None

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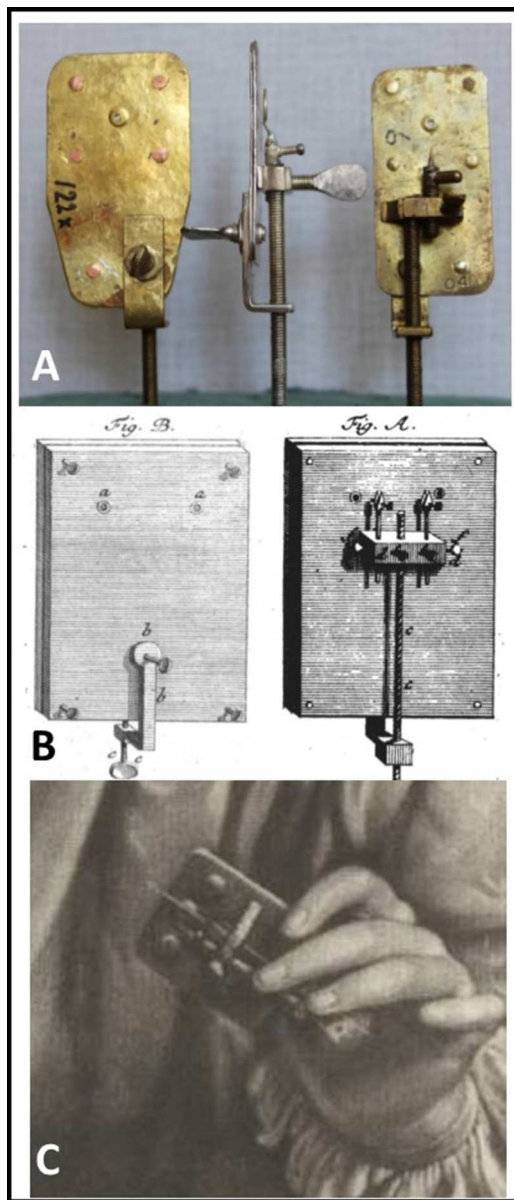


Figure 1: Three versions of Van Leeuwenhoek's microscope. A: Facsimiles of the well-known form. B: Version shown by Von Uffenbach (1754) with 2 lenses and 2 sample holders. C: Version shown by Verkolje (1686) with 3 lenses, 1 sample pin and a holder for a capillary tube.

70x163mm (150 x 150 DPI)

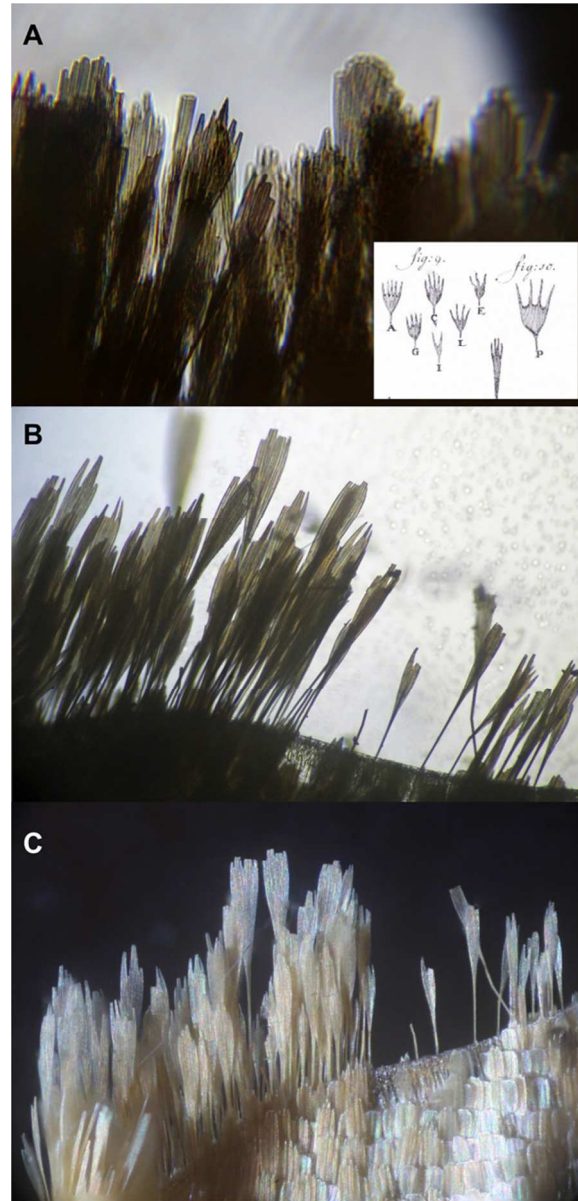


Figure 2. A: The edge of the hindwing of a large yellow underwing moth as photographed through the facsimile Van Leeuwenhoek microscope, inset: Van Leeuwenhoek's drawings of the scales from the wing of a silkworm moth (Van Leeuwenhoek, 1702b). B and C: the same sample photographed through the Zeiss microscope under back and top-lighting, respectively.

130x271mm (125 x 125 DPI)

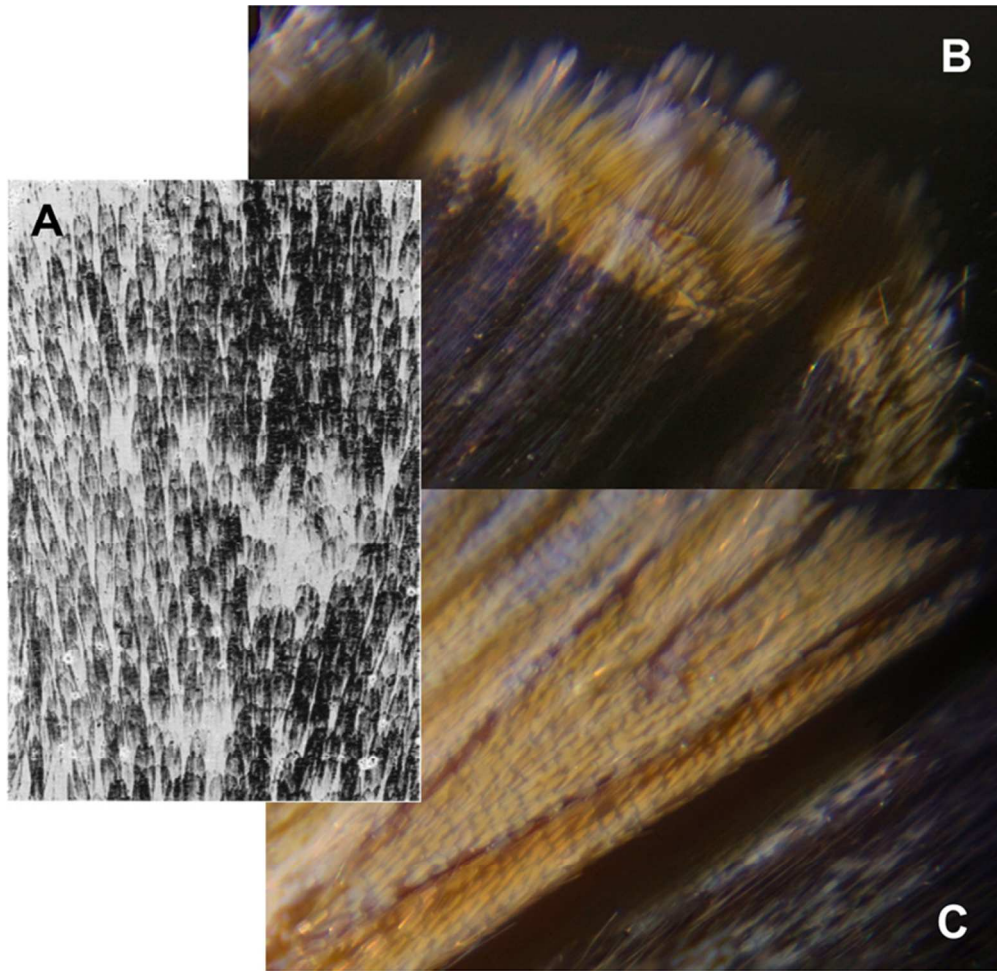


Figure 3. A: The scales seen by Van Leeuwenhoek on the surface of a butterfly's wing (Van Leeuwenhoek, 1678). B and C: The same moth's wing as in Fig. 2, photographed through the modern hand lens with the light positioned beside the camera to give top lighting.

140x135mm (150 x 150 DPI)

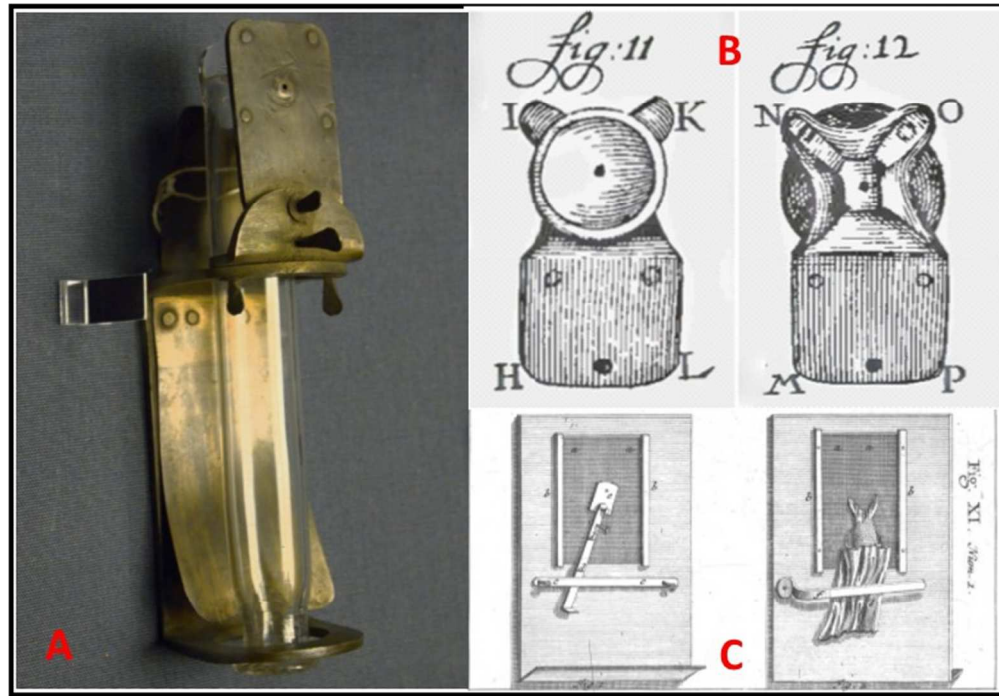


Figure 4. A: Facsimile of Van Leeuwenhoek's original aalkijker with the same lens holding plate as used in his microscope (Fig. 1A). B: The reduced lens holder with the eye cup attached to Van Leeuwenhoek's first modification of his aalkijker. This lens could be attached to the aalkijker frame in place of the familiar rectangular flat plate shown in Fig. 4A, "fig 11" shows the observer's side, and "fig 12", the sample side. C: The 3rd version of the "aalkijker" with the glass tube for holding a fish or eel replaced by a flat glass plate, a clamp and a piece of wet fabric (see also Fig 6B).

140x98mm (150 x 150 DPI)

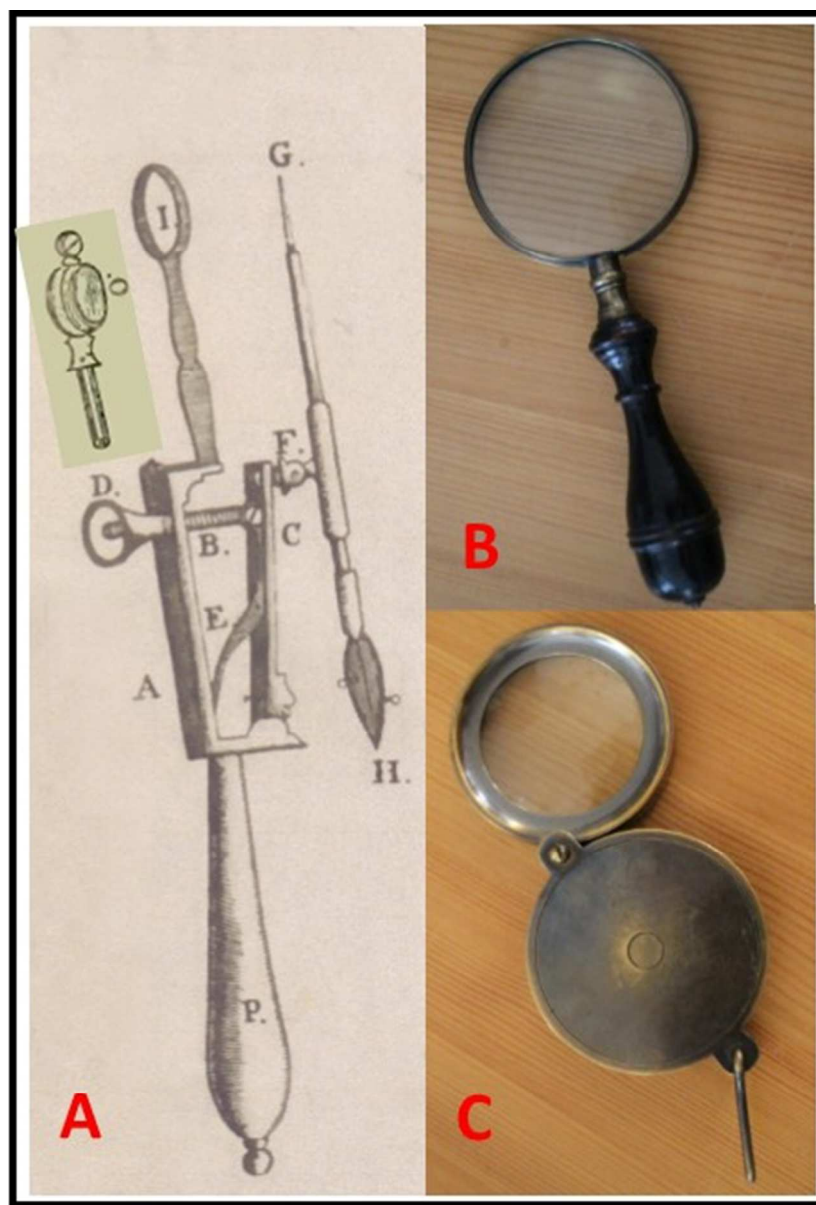


Figure 5: A: Low powered microscope for opaque samples (Baker, 1739) which strongly resembles the instrument marked B in Fig 6. It could be used with larger lenses (O) mounted in a metallic ring (I), or fitted with a smaller, stronger lens surrounded by a reflector (often called a "Lieberkuhn") which could also be attached to the microscope at (I). B and C: Facsimiles of "burning glasses" in use in the 17th and 18th centuries, as sold by Museum Boerhaave in Leiden.

75x109mm (150 x 150 DPI)

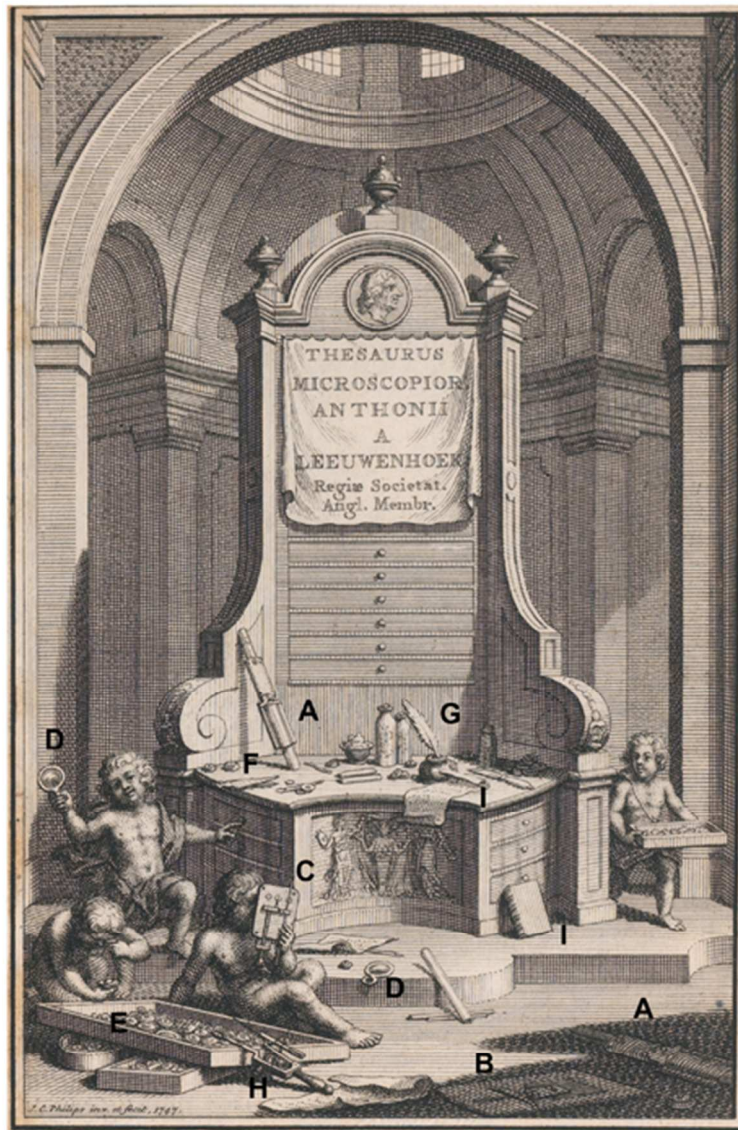


Figure 6. The frontispiece of the catalogue for the sale of Van Leeuwenhoek's microscopes after the death of his daughter, Maria (Rees, 1747, Robertson 2015b). (A) original aalkijker; (B) second modification of the aalkijker; (C) microscope with three lenses side by side, a sample pin and a capillary sample tube; (D) magnifying glass; (E) loose lenses, tweezers; (G) quill pen and ink; (H) microscope for opaque samples; (I) bound book.

106x149mm (125 x 125 DPI)