The Reception of Abbe's Theory in England

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In the nineteenth century microscopy in England and on the Continent (especially in Germany) followed rather different lines. The English microscopists were largely empiricists, and for many of them theory counted but little - indeed if they were aware of it! Perhaps they may best be categorised as 'gentlemen/amateurs', often professional men in the Church, banking, insurance and the law, not trained scientists. They were on average well-off and often almost obsessed by possessing the latest equipment from the best makers such as Ross and Powell & Lealand. John Mayall writing in 1881 in a letter (now in the archives of Carl Zeiss, Jena) to Abbe noted this lack of interest in English microscopist, commenting:

only ocular demonstration will convince the great mass who won't read any paper.

Their main interest was in achieving the ultimate in magnification and resolution chiefly using siliceous diatom frustules with their very fine detail as their test objects. As a result they were often dubbed 'diatom-dotters'! They were happy with their images if the centre of the field of view was sharp and free from colour. They used principally 'dry' objectives often of very short focal lengths such as 1/14th or 1/16th of an inch. The lenses were often provided with correction collars but generally the numerical aperture of the objectives did not exceed 0.8. Water and oil immersion objectives were gaining increased vogue in the latter half of the century but many well-known microscopists (e.g. Francis Wenham, as we know from the correspondence of Stephenson) could not appreciate why an immersion objective could have an aperture greater than the equivalent of 180° in air. Condensers were commonly used especially those of Gillet (Fig 1.) and Powell & Lealand. There was a fashion for the use of extreme obliquity of illumination which led to several microscopes being empirically designed with this in mind. Two examples are those of Ross (c. 1878) with its swinging substage and the Ross 'Radial' microscope in which almost every possible movement was provided both for the substage and for the limb (Figs 2, 3).

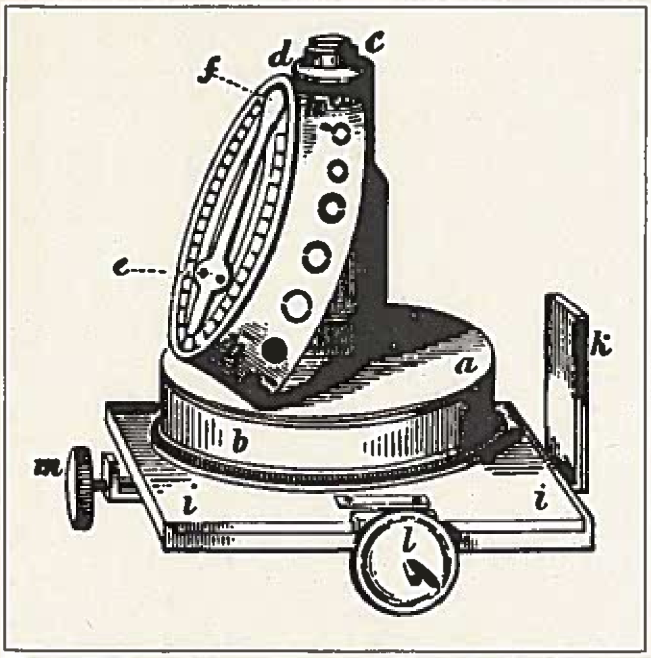

Fig. 1. Gillet's condenser, as illustrated in Hogg's book on the microscope (1867). This condenser, developed in the 1860s, was achromatic, provided with a centring mechanism and had a ring of diaphragms to allow extensive modification of the illumination.

In contrast the Continental microscopist was also a professional man, but one who was a trained scientist or medical doctor. They were applying the microscope to the study not of diatom frustules but to material which was relevant to pathology or cytology. To this end they developed the use of specific dyes and stains. Altmann introduced his acid fuchsin/picric acid technique for staining the mitochondria in cells; Koch developed alkaline methylene blue used with vesuvin (now known as Bismark brown) to stain the bacilli of tuberculosis and Flemming was using iron haematoxylin to colour chromosomes in cell division. In the latter case a counterstain was not used so that he would have had a problem with seeing the tissue itself, although this is not so important in the studies of mitosis and meiosis. Prior to c. 1870 a narrow cone of illumination from a concave mirror and from a pin-hole diaphragm in the microscope substage (Fig. 4) would have been used to provide contrast if the actual tissue structure had to be observed. In 1869 Abbe, by now working with Carl Zeiss, invented his ex-centring substage with condenser and variable iris diaphragm (Fig. 5). This could be closed and moved off the microscope optical axis to provide the contrast needed to observe both the stained inclusions or bacteria and, at the same time, the unstained tissue of the section. Indeed, the corrections of Abbe's original condenser were poor and this led those using it to adopt narrow cones in order to minimize aberrations introduced by the condenser itself. The Abbe substage and condenser allowed (as Koch put it)

a 'structural picture' of the tissue [unstained] and a 'colour picture' of the bacilli.

Zeiss had asked Abbe to determine the formulae for rationalising the construction of microscope objectives and this work led Abbe on to his determination of expressions for the resolution of the microscope, of the concept of objectives of high numerical aperture and later of very low aberrations - the Zeiss apochromats. In 1873 Abbe produced his first major paper in German on the theory of the microscope and this was rapidly followed by an English translation undertaken by Fripp in 1874 and published in a rather unlikely journal. During the preparation of his work Abbe had learned of similar studies by Helmholtz and it was only with difficulty that Abbe could be persuaded to publish his own work.

In its original form Abbe's theory considered resolution of large details in the image to be the result of what he called an absorption or 'dioptric image' and that diffraction was only responsible for the resolution of the fine detail. Later, of course, he abandoned this view. In

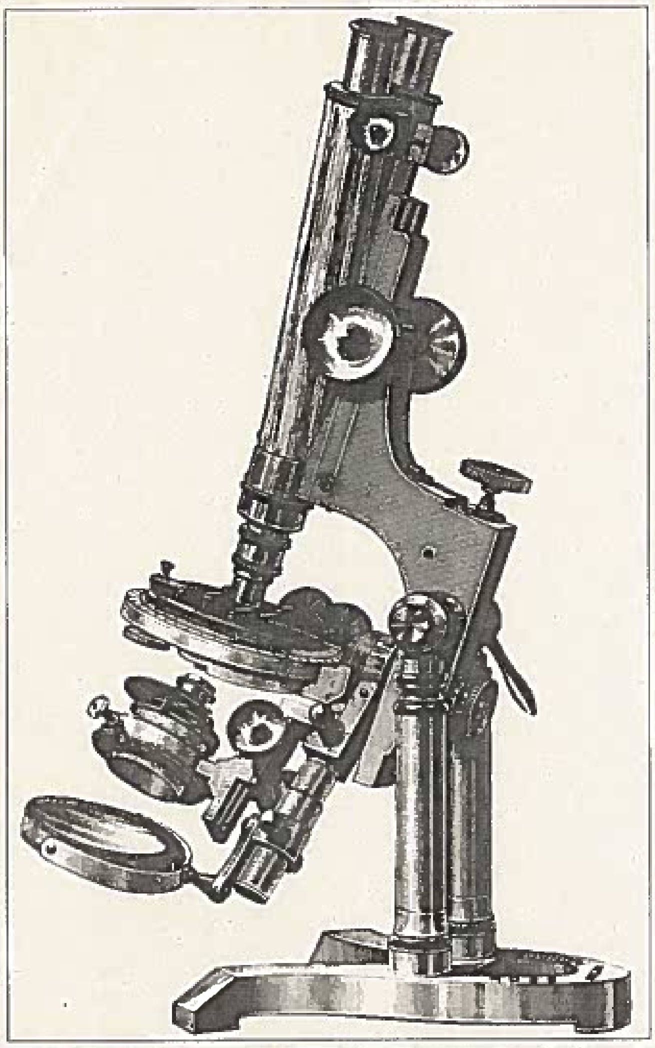

Fig. 2. Ross microscope (c. 1878) with Zentmayer's swinging substage designed to provide oblique illumination.

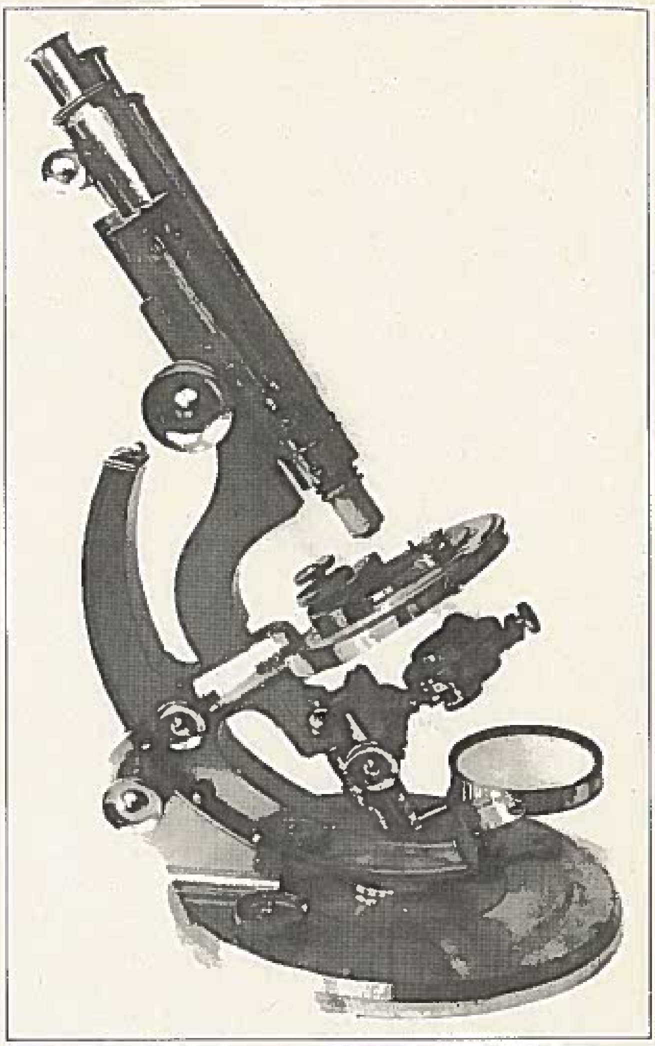

Fig. 3. Ross Radial microscope of the early 1880s. This instrument not only allowed obliquity of substage illumination, but also of the microscope tube itself.

order to convince sceptics of the existence of the diffracted light and its importance in microscopic resolution, Abbe devised what was to become perhaps the most famous experiment in microscope optics. This involved the use of his 'Diffractionsplatte', a special slide constructed by Zeiss and containing lines ruled on an opaque silvered background. The plate had parallel lines at two different spacings, a set of lines ruled at right angles and a set of lines at an angle of 60° to each other. It was intended to be used with the Zeiss aa objective of c. 1″ focal length and this was to be mounted on a carrier which had provision for rotation and for the insertion of thin opaque metal diaphragms which had various arrangements of holes and slits. By the use of the parallel line grating and a corresponding very narrow axial cone of light the appearance of the first and second orders of diffracted light could be seen in the objective's back focal plane (Fig. 6). These orders occupied the 'dark space', surrounding the image of the central axial light (the so-called dioptric beam). This demonstration and the various experiments related to it involving occlusion of various parts of the diffracted light in the back focal plane, with the consequent effects on the resolution of the microscope, was first published in English by Fripp (1876). This was followed a year later by a paper by J.W. Stephenson, at that time Treasurer of the RMS and to whom 57 letters from Abbe (preserved in the RMS archives) were written. Fortunately many of the queries from Stephenson to Abbe, and to which the latter's letters relate, have remained in the Zeiss archives and it is hoped that these will be collated and published in the near future with the RMS Abbe letters (and others from microscopists such as Crisp, Mayall, Edmunds and others). The images from the Diffractionsplatte experiments are perhaps the most spectacular ever devised to illustrate microscopy. In order to demonstrate the diffracted light, however, a very narrow axial cone from a small stop in the condenser must be used. It may perhaps be this single requirement, more than any writings of any microscopist, which prejudiced the English microscopists against the ideas of Abbe; after all, everyone involved in the study of diatom frustules KNEW that small stops should never be placed in either the condenser or objective focal planes! The English microscopists (with one or two noted exceptions) did not

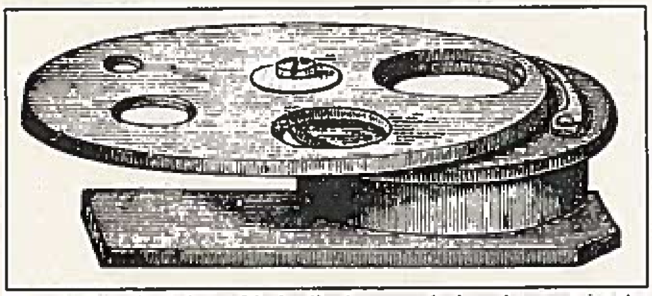

Fig. 4. A typical ring of hole diaphragms designed to restrict the aperture of the illuminating cone provided by the concave mirror of the microscope. These diaphragms would have been used without any form of substage condenser.

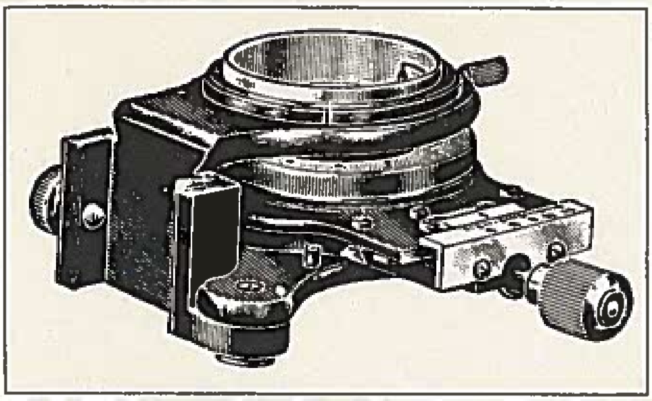

Fig. 5. Abbe's ex-centring substage shown here without the condenser. This mount was fitted with an iris diaphragm and a rackwork mechanism to displace the whole mount out of the optical axis of the microscope. It was also designed to rotate so that light of varying degrees of azimuth or obliquity could be obtained on the specimen.

understand that the use of small stops were only needed for the demonstration of the effect and that they were not being advocated for general use. They could not seem to understand the importance of numerical aperture and that the image resembles the original object according to the amount of diffracted light accepted. Furthermore, English users did not understand that *large* objects were imaged faithfully because even an objective of low numerical aperture could accept nearly all the diffracted rays from a large object because of their relatively close spacing.

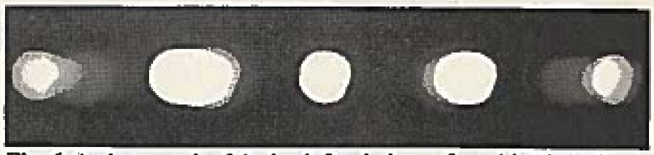

Fig. 6. A photograph of the back focal plane of an objective when a grating on the stage of the microscope is illuminated by a narrow cone of light. The direct light is in the centre of the picture and the diffracted orders of light may be seen on either side. The area which is not illuminated was called the 'dark space'.

Abbe wrote very good English (although most of his later papers in the Journal of the RMS were subjected to much heavy editing by Frank Crisp, who was for many years the Secretary of the RMS and the editor of its Journal). To today's readers much Victorian writing is very prosy and hard to follow; one wonders how readers at that time, to whom the ideas being put forward were completely strange and new, reacted! Spitta (1853-1921) another leading microscopist and photomicrographer was also of the view that one of the major obstacles to the acceptance of Abbe's theory was the manner in which it was explained.

As already mentioned, Abbe's theory in its original form considered the resolution of large details in the image to be the result of what he called an absorption or 'dioptric image' and that diffraction was responsible only for the resolution of the fine detail. The actual image in the image plane was a secondary phenomenon, the result of

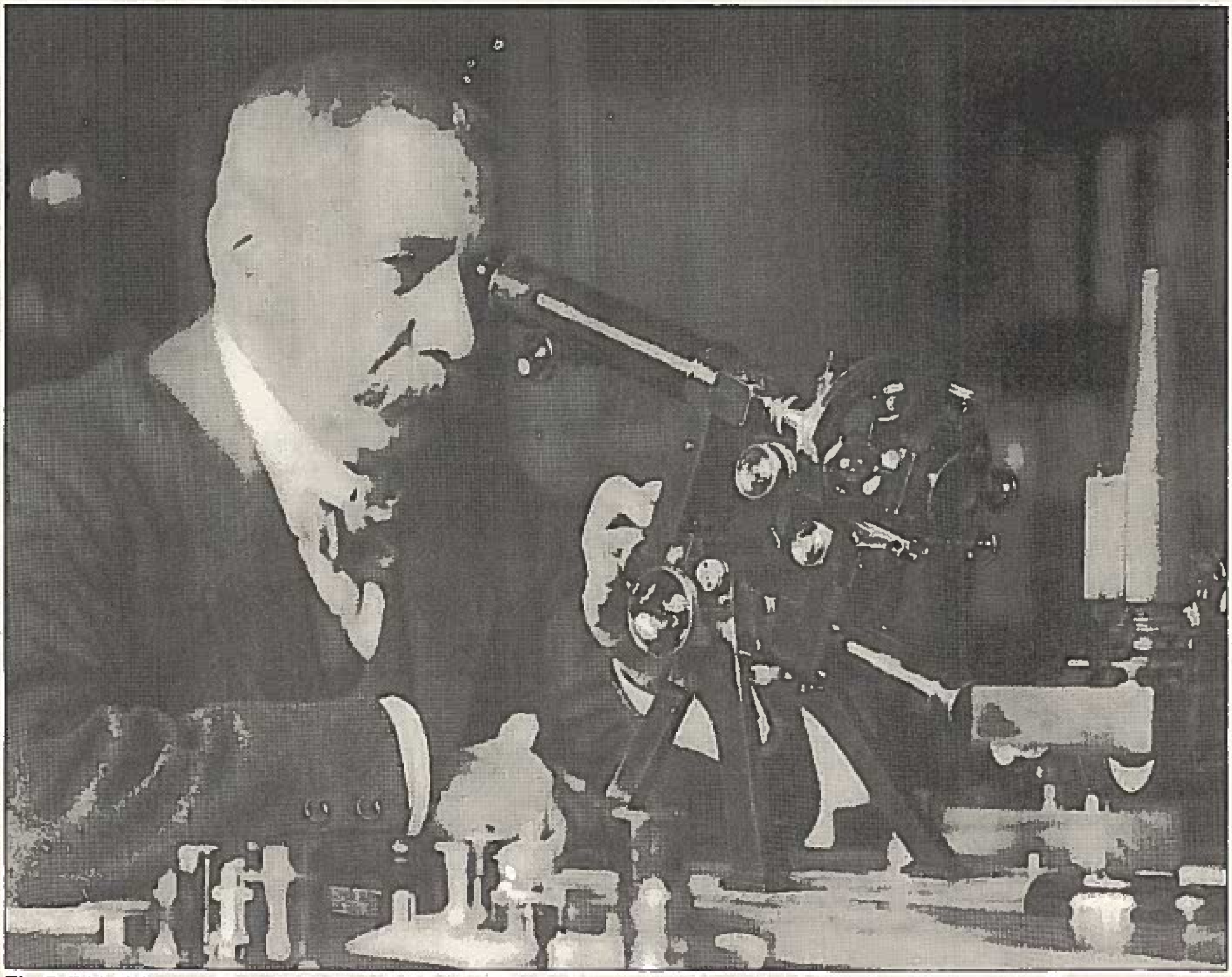

Fig. 7. E.M. Nelson working with his Powell & Lealand No. 1 microscope; a picture taken in 1910 by A.E.E. Eliot-Merlin.

interference between the direct and diffracted beams. The more diffracted beams accepted by the aperture of the objective, the closer the correspondence of the image with the real structure of the object. Furthermore, no fine structure could be deduced until at least one diffracted order was admitted to the objective. Later, after discussions with various English microscopists, Abbe altered his views somewhat and, as outlined in later editions of e.g. Carpenter and Dallinger (1901) he said:

> I no longer maintain, in principle, the distinction between the absorption image (the direct dioptrical image) and the diffraction image, nor do I hold that the microscopical image of an object consists of two superimposed images of different origin or different modes of production.

This was almost certainly as a result of a paper in 1889 by E.M.Nelson (Fig. 7) to the Royal Microscopical Society. This paper was never published in the Journal but a copy was sent to Abbe on whom it obviously had considerable influence. Many microscopists in England found it hard to appreciate Abbe's work. There was considerable apathy to theoretical studies at that time, and Abbe's writings were not easy to follow. From the surviving letters of Crisp to Abbe we know that Crisp, who was the Editor of the *Journal*, made many alterations to the text of Abbe's various papers, but even so the language was that which was fashionable at that time with rather convoluted sentence structure and in consequence the end result was often very long, rather turgid papers which would not be easy reading. At the same time there seems to have been considerable difficulty in the understanding of Abbe's work. In the letters preserved in the RMS archives from Abbe to Stephenson there are several attempts to explain the theory. Crisp in his letters to Abbe also commented several times on the lack of understanding of the English microscopists.

> The longer I live the more I am astonished at the ignorance of English microscopists (20 January 1881)

> Now you cannot comprehend the profound state of ignorance in England among microscopists on optical matters.
>
> Look at our books
> Look at Shadbolt [ex-President!]
> Look at Edmunds
> Look at Wenham
> Look at me! - I class myself as a man of fair average intelligence - supposed to be devoted to the Microscope - Secretary of the Royal Microscopical Society! ... What labour you have had over and over again to get me to comprehend what you meant, although I was anxious to understand. How I struggled to master what you said "a child knew" and which it took me weeks to grasp!
>
> If so it be with me - 'with fair average intelligence !' - what must it be with others. (7 March 1881)

A few years later Crisp (1878) went even further in public, writing in the Royal Microscopical Society Journal

> It may I think be truly said that out of the entire scientific world there is probably no body of men who devote so little real attention to the principles that lie at the root of that branch of science of which they are disciples, as do the English microscopists.
>
> As a particular instance I refer to the apathy that has been shown in regard to the researches of Professor Abbe on the theory of the microscope, researches undoubtedly as important as any that can be found in the whole of its history and rivalling even the discovery of achromatism, or that of Lister on aplanatic foci.

It is clear from this that Crisp regarded apathy as the most immediate reaction to Abbe's work; it must be said that this reaction must be closely followed by incomprehension and misunderstanding. Later as a result of the efforts of Stephenson (1877) and Crisp (1881), the theory was expounded again in greater detail and Abbe himself (1889) helped its acceptance by writing on illumination by wide cones and considering the wide angled cone as a multitude of 'diffraction pencils' at all angles which formed the image and that therefore the diffracted orders in the back focal plane of the objective overlap and so only an area of full illumination was seen here with no 'dark space'. Other well-known microscopists such as Dallinger came to accept Abbe's theory, but it is clear from his writings that Dallinger did not fully understand it in detail as he knew from experience that a large cone was vital and hence "theory and practice were at variance". The diffraction theory of microscopical vision came to be discussed in many papers over the following years and at meetings of both the Royal Microscopical Society and the Quekett Microscopical Club. Indeed a controversy resulted which continued for nearly 50 years, largely fuelled by microscopists who had seen the diffraction plate experiments and had misunderstood them to mean that they applied only to narrow cones of illumination. Abbe's own views that the use of wide cones was **not** inconsistent with the acceptance of his theory seemed to be ignored.

Other objections to Abbe's theories appeared to arise from misunderstandings and misinterpretations. For example, it was realised that false images would result from the acceptance of some diffraction orders by the objective and the exclusion of other, *lower*, orders. The admission of the second order and the exclusion of the first order by the physical presence of a stop in the back focal plane gave a false image with the appearance of double periodicity. This was understood and accepted. The fact that even **without** the presence of a physical stop in the back focal plane, zonal spherical aberration at some foci could exclude the first order and cause the second order to combine with the direct light, was not. Hence if this occurred, appearances which were unreal might be interpreted as genuine. This is illustrated by the problem set by the mathematical faculty at Jena which postulated a given pattern of diffracted orders (Fig.8) accepted by an objective of a specified numerical aperture and required the calculation of the appearance of the image. The arrangement of orders in the back focal plane, as set by the Jena faculty was, in fact, very similar to that actually produced by the diatom *Pleurosigma angulatum*. The problem was solved by a mathematician called Eichhorn (who had never seen a diatom under the microscope); he suggested that the image would be composed of circles encompassed within hexagons, with small circular markings within the triangular interstices between three adjacent hexagons (Fig. 9). The circular markings between the puncta of the diatom *Pleurosigma angulatum* had not actually been seen at that time but nevertheless they came to be called the 'Eichhorn intercostals'. In 1909 E.J.Spitta produced a remarkable photomicrograph (reproduced here as Fig. 10) which undoubtedly shows shadowy markings in the correct position. Spitta in the legend to the photo indicated that he suspected that these markings were probably spurious, since they appeared "when a certain focus was obtained". E,M Nelson was strongly of the opinion that the Eichhorn intercostals could only have arisen from

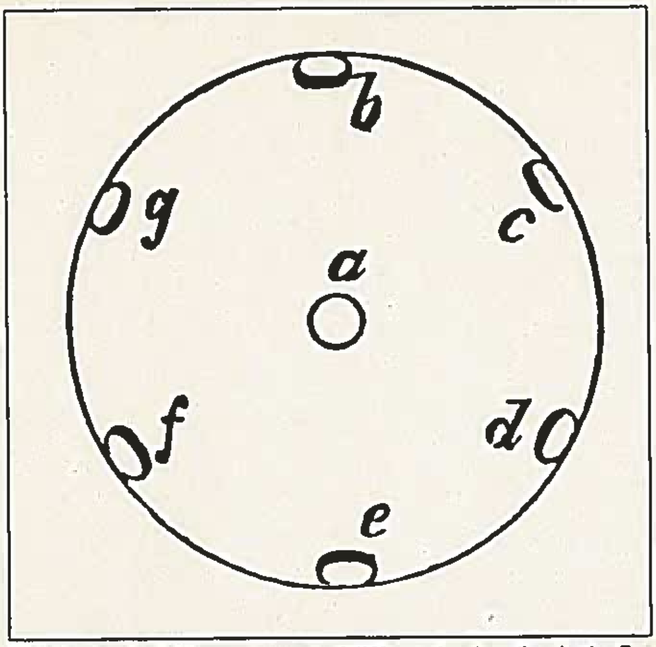

Fig. 8. The six-fold arrangement of diffracted orders (b-g in the figure, taken from Crisp, 1881) which was set by the Jena mathematical faculty for the student problem.

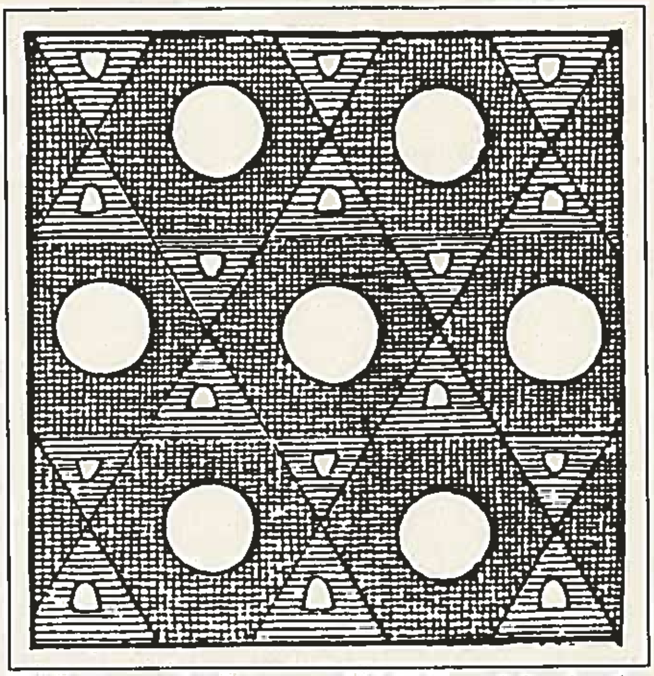

Fig. 9. The postulated structure which Eichhorn deduced mathematically, showing the hexagonal structure with small markings in the triangular areas between them (from Crisp, 1881).

the fact that the objective was accepting the edges of the *second* orders of diffracted light because zonal spherical aberration at one particular focus completely annulled the effects of the *first* order at the particular focus chosen. If a cone of light which occupies at least three quarters of the aperture of the objective is used or a slightly different focal plane is chosen, then diffracted light of the first order will inevitably pass through the same zone of the objective as that from the second order and prevent such false images due to zonal aberrations.

Who then were the English microscopists who were pro-Abbe? In the 1880s Stephenson, Mayall and Crisp were undoubtedly his main champions at the RMS and the Quekett Club. Later, around the turn of the century, Julius Rheinberg (now remembered for his illumination technique) was very much a supporter of Abbe and stressed how much misinterpretation had occurred. He wrote in 1901:

I wish to distinctly dissociate myself from many of the chief deductions which have been drawn from this theory - notably that narrow cones of light are preferable to wide ones, and that there are no reasons why a wide cone should yield a more correct projection of the object.

That a number of deductions harmful to the progress of Microscopy have been drawn from the Abbe diffraction theory, particularly the uncertainty as to the correctness of the image and the desirability of using narrow cones of light, seems undeniable in the face of the practical experience of today. But, whilst parting with the deductions which are erroneous, we must be very careful not to throw away valuable fundamental principles.

Similarly Conrady (quoted in Spitta, 1909, p.428) was very much in favour of Abbe's work:

all the most striking nightmares produced with Abbe's plate require the use of suitable diaphragms in the back focal plane by which certain diffraction spectra are deliberately excluded (which would otherwise enter) and which if admitted would at once cause the "nightmares" to vanish.

Conrady (1904, 1905) had previously produced two papers in which he wrote strongly in support of Abbe's theory. Spitta himself, in the same work, wrote:

Whilst the Abbe theory has never been attacked by a competent physicist, there have appeared from time to time elaborate papers by others trying to prove that it was entirely erroneous; the "it", however, was never the Abbe theory as put forward by Abbe; "it" represented the particular author's way of misunderstanding Abbe's papers or accounts of them in books.

Perhaps, as Spitta points out elsewhere, if this had been stressed in the 1880s,

A vast amount of misunderstanding and scepticism would have been nipped in the bud.

What of the other great English microscopist of the period - E.M.Nelson? At first he seemed to think that the real Abbe theory turned on the use of small cones of light and, as Nelson could empirically demonstrate the value of large cone illumination for resolution, he was sceptical. Later when Abbe emphasised his own belief in large angles of both illumination and objective aperture, Nelson became very supportive. Indeed Eliot Merlin comments in Nelson's obituary:

The most strenuous part of Nelson's life-work consisted in correcting the harmful observational methods then upheld in consequence of the erroneous deductions based on Abbe's celebrated diffraction theory of microscopical vision.

The first major attack on Abbe's work by an English microscopist was by Gordon in 1901, when in a long paper he came out strongly in opposition to Abbe. This paper by Gordon was much criticised at the meeting at which it was delivered, especially by Julius Rheinberg. The latter contended that Gordon had made several fundamental errors especially in confusing diffraction spectra in the back focal plane of the objective with patterns produced by interference of these waves in the primary image plane.

In the following years interest in the subject seemed to abate and the Abbe theory seemed to become almost universally accepted, although some microscopists still seemed to retain doubts as to its universal validity. In particular an influential paper by Berek in 1926 proposed an alternative view formulating the ideas that the microscope image was composed of 'consonant' and 'dissonant' light. Berek later dismissed the Abbe theory on the grounds (summarised by Moore, 1940) that:

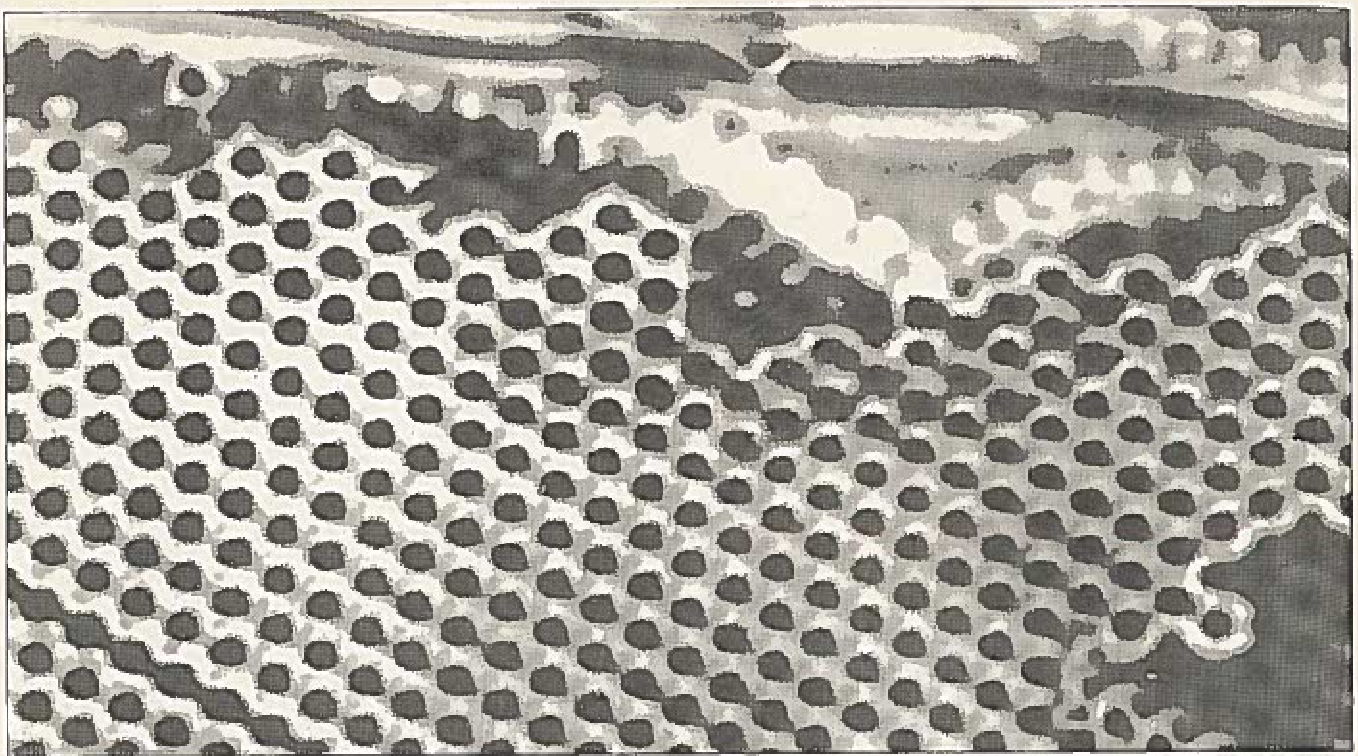

Fig. 10. Spitta's micrograph (published in his 1909 book) of *Pleurosigma angulatum* showing the Eichhorn intercostals (the small dark areas in the frustule wall between the puncta). These are almost certainly spurious effects due to zonal aberrations in the objective.

the interference effects, to which so much importance was attached by Abbe, are shown to be a non-focused phenomenon which has none of the properties of an optical image. It is also shown that they are not prominently visible except when the illumination is limited to a cone of very small angle, and it is finally concluded that the theory has no general applicability and has no bearing on practical microscopy.

In his 1940 paper, Moore reiterates Berek's views by saying that:

in all ordinary observation of non-luminous objects under the microscope there is an image formed in the same way as the image of a luminous body. Superposed on this image is an 'Abbe' interference effect which is prominently visible with very narrow angle illumination, but which can be made to disappear completely when wide-angle illumination is employed. It is thus impossible to consider that the Abbe theory is of any importance in practical microscopy; the theory merely deals with a phenomenon which impairs the true image given by the microscope and which every microscopists does his best to eliminate by proper adjustment of the illumination which he employs.

Shortly after Berek's work had appeared in the 1920s, Moore (1928) produced a paper on image formation in the microscope, upon which Conrad Beck suggested that the Society should have a full discussion of image formation; this suggestion was accepted and the meeting took place in March 1929. The papers were published in the Journal of the Society (*J.Roy. Micr. Soc.*, 1929, **XLIX**, pp123-142 and pp.228-264.). As might be expected there was a wide range of opinions but Moore, looking back in his 1940 paper, singled out two main trends. These were that although many contributors found it difficult to reject the Abbe theory entirely, they had some difficulty in accepting all its postulations, and that in some papers there was an

attempt to shield Abbe himself at the expense of his theory, since it was claimed that in 1903 he had modified his views so far as to admit that image formation in the microscope takes place according to ordinary dioptric processes.

Berek considered that the 'dissonant' component of light in the microscope image was that focused by the objective which gave an image free from visible fringes with a precisely defined focal plane. The image detail corresponded point for point with the object detail and the processes of image formation were exactly the same as those forming images of luminous points. Any lack of correspondence between object and image detail was attributed to i) the finite aperture of the objective and ii) residual aberrations in the objective.

The 'consonant' component produces interference effects over considerable distances on either side of the true focal plane of the image in the microscope tube and nothing allows one individual plane to be identified as the true plane in preference to another. The quality of the final image Berek believed depended on the relative proportions of consonant/dissonant light - what he termed the 'degree of consonance'. Conditions of complete consonance (i.e. a point source at infinity) are not of course realisable, but an approximation is given by a narrow-angled cone of light. As the angle of the illuminating cone increases, the degree of consonance decreases and the image quality improves. This approach seems to have been generally accepted as it covers all illumination conditions from narrow cone (with pronounced interference effects) to full illumination by wide cone when the interference effects are not seen.

It thus seemed that after fifty or more years of controversy, with misunderstandings on all sides, that there was a return to views held by physicists such as Rayleigh in the latter years of the 19th century (and neglected because they were presented in such a detailed mathematical way!). He held that if a non-self-luminous object is illuminated by a cone of a medium angle it can then be regarded as intermediate between a truly self-luminous or incoherent source and a coherently-illuminated object as considered by Abbe.

A full and rather mathematical treatment of modern views on image formation in the microscope will be found in Martin (1966).

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