### BRIEF ACCOUNT

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OF

# **MICROSCOPICAL OBSERVATIONS**

Made in the Months of June, July, and August, 1827,

# ON THE PARTICLES CONTAINED IN THE POLLEN OF PLANTS;

AND

# ON THE GENERAL EXISTENCE OF ACTIVE MOLECULES

IN ORGANIC AND INORGANIC BODIES.

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#### MICROSCOPICAL OBSERVATIONS.

THE observations, of which it is my object to give a summary in the following pages, have all been made with a simple microscope, and indeed with one and the same lens, the focal length of which is about  $\frac{1}{52}$ nd of an inch\*.

The examination of the unimpregnated vegetable Ovulum, an account of which was published early in 1826<sup>+</sup>, led me to attend more minutely than I had before done to the structure of the Pollen, and to inquire into its mode of action on the Pistillum in Phænogamous plants.

In the Essay referred to, it was shown that the apex of the nucleus of the Ovulum, the point which is universally the seat of the future Embryo, was very generally brought into contact with the terminations of the probable channels of fecundation; these being either the surface of the placenta, the extremity of the descending processes of the style, or more rarely, a part of the surface of the umbilical cord. It also appeared, however, from some of the facts noticed in the same Essay, that there were cases in which the Particles contained in the grains

\* This double convex Lens, which has been several years in my possession, I obtained from Mr. Bancks, optician, in the Strand. After I had made considerable progress in the inquiry, I explained the nature of my subject to Mr. Dollond, who obligingly made for me a simple pocket microscope, having very delicate adjustment, and furnished with excellent lenses, two of which are of much higher power than that above mentioned. To these I have often had recourse, and with great advantage, in investigating several minute points. But to give greater consistency to my statements, and to bring the subject as much as possible within the reach of general observation, I continued to employ throughout the whole of the inquiry the same lens with which it was commenced.

+ In the Botanical Appendix to Captain King's Voyages to Australia, vol. ii. p. 534. et seq.

of pollen could hardly be conveyed to that point of the ovulum through the vessels or cellular tissue of the ovarium; and the knowledge of these cases, as well as of the structure and economy of the antheræ in Asclepiadeæ, had led me to doubt the correctness of observations made by Stiles and Gleichen upwards of sixty years ago, as well as of some very recent statements, respecting the mode of action of the pollen in the process of impregnation.

It was not until late in the autumn of 1826 that I could attend to this subject; and the season was too far advanced to enable me to pursue the investigation. Finding, however, in one of the few plants then examined, the figure of the particles contained in the grains of pollen clearly discernible, and that figure not spherical but oblong, I expected, with some confidence, to meet with plants in other respects more favourable to the inquiry, in which these particles, from peculiarity of form, might be traced through their whole course: and thus, perhaps, the question determined whether they in any case reach the apex of the ovulum, or whether their direct action is limited to other parts of the female organ.

My inquiry on this point was commenced in June 1827, and the first plant examined proved in some respects remarkably well adapted to the object in view.

This plant was Clarckia pulchella, of which the grains of pollen, taken from antheræ full grown, but before bursting, were filled with particles or granules of unusually large size, varying from nearly  $\frac{1}{4000}$  th to about  $\frac{1}{5000}$  th of an inch in length, and of a figure between cylindrical and oblong, perhaps slightly flattened, and having rounded and equal extremities. While examining the form of these particles immersed in water, I observed many of them very evidently in motion; their motion consisting not only of a change of place in the fluid, manifested by alterations in their relative positions, but also not unfrequently of a change of form in the particle itself; a contraction or curvature taking place repeatedly about the middle of one side, accompanied by a corresponding swelling or convexity on the opposite side of the particle. In a few instances the particle was seen to turn on its longer axis. These motions were such as to satisfy me, after frequently repeated observation, that they arose neither from currents in the fluid, nor from its gradual evaporation, but belonged to the particle itself.

Grains of pollen of the same plant taken from antheræ immediately after bursting, contained similar subcylindrical particles, in reduced numbers however, and mixed with other particles, at least as numerous, of much smaller size, apparently spherical, and in rapid oscillatory motion.

These smaller particles, or Molecules as I shall term them, when first seen, I considered to be some of the cylindrical particles swimming vertically in the fluid. But frequent and careful examination lessened my confidence in this supposition; and on continuing to observe them until the water had entirely evaporated, both the cylindrical particles and spherical molecules were found on the stage of the microscope.

In extending my observations to many other plants of the same natural family, namely *Onagrariæ*, the same general form and similar motions of particles were ascertained to exist, especially in the various species of Œnothera, which I examined. I found also in their grains of pollen taken from the antheræ immediately after bursting, a manifest reduction in the proportion of the cylindrical or oblong particles, and a corresponding increase in that of the molecules, in a less remarkable degree, however, than in Clarckia.

This appearance, or rather the great increase in the number of the molecules, and the reduction in that of the cylindrical particles, before the grain of pollen could possibly have come in contact with the stigma,—were perplexing circumstances in this stage of the inquiry, and certainly not favourable to the supposition of the cylindrical particles acting directly on the ovulum; an opinion which I was inclined to adopt when I first saw them in motion. These circumstances, however, induced me to multiply my observations, and I accordingly examined numerous species of many of the more important and remarkable families of the two great primary divisions of Phænogamous plants.

In all these plants particles were found, which in the different families or genera varied in form from oblong to spherical, having manifest motions similar to those already described; except that the change of form in the oval and oblong particles was generally less obvious than in Onagrariæ, and in the spherical particle was in no degree observable\*. In a great proportion of these plants I also remarked the same reduction of the larger particles, and a corresponding increase of the molecules after the bursting of the antheræ: the molecule, of apparently uniform size and form, being then always present; and in some cases indeed, no other particles were observed, either in this or in any earlier stage of the secreting organ.

In many plants belonging to several different families, but especially to Gramineæ, the membrane of the grain of pollen is so transparent that the motion of the larger particles within the entire grain was distinctly visible; and it was manifest also at the more transparent angles, and in some cases even in the body of the grain in Onagrariæ.

In Asclepiadeæ, strictly so called, the mass of pollen filling each cell of the anthera is in no stage separable into distinct grains; but within, its tesselated or cellular membrane is filled with spherical particles, commonly of two sizes. Both these kinds of particles when immersed in water are generally seen in vivid motion; but the apparent motions of the larger particle might in these cases perhaps be caused by the rapid oscillation of the more numerous molecules. The mass of pollen in this tribe of plants never bursts, but merely connects itself by a determinate point, which is not unfrequently semitransparent, to a process of nearly similar consistence, derived from the gland of the corresponding angle of the stigma.

In *Periploceæ*, and in a few *Apocineæ*, the pollen, which in these plants is separable into compound grains filled with spherical moving particles, is applied to processes of the stigma, analogous to those of Asclepiadeæ. A similar economy exists in *Orchideæ*, in which the pollen masses are always, at least in the early stage, granular; the grains, whether simple or compound, containing minute, nearly spherical particles, but

\* In Lolium perenne, however, which I have more recently examined, though the particle was oval and of smaller size than in Onagrariæ, this change of form was at least as remarkable, consisting in an equal contraction in the middle of each side, so as to divide it into two nearly orbicular portions. the whole mass being, with very few exceptions, connected by a determinate point of its surface with the stigma, or a glandular process of that organ.

Having found motion in the particles of the pollen of all the living plants which I had examined, I was led next to inquire whether this property continued after the death of the plant, and for what length of time it was retained.

In plants, either dried or immersed in spirit for a few days only, the particles of pollen of both kinds were found in motion equally evident with that observed in the living plant; specimens of several plants, some of which had been dried and preserved in an herbarium for upwards of twenty years, and others not less than a century, still exhibited the molecules or smaller spherical particles in considerable numbers, and in evident motion, along with a few of the larger particles, whose motions were much less manifest, and in some cases not observable\*.

In this stage of the investigation having found, as I believed, a peculiar character in the motions of the particles of pollen in water, it occurred to me to appeal to this peculiarity as a test in certain families of Cryptogamous plants, namely Mosses, and the genus Equisetum, in which the existence of sexual organs had not been universally admitted.

In the supposed stamina of both these families, namely, in the cylindrical antheræ or pollen of Mosses, and on the surface of the four spathulate bodies surrounding the naked ovulum, as it may be considered, of Equisetum, I found minute spherical particles, apparently of the same size with the molecule described in Onagrariæ, and having equally vivid

\* While this sheet was passing through the press I have examined the pollen of several flowers which have been immersed in weak spirit about eleven months, particularly of *Viola tricolor*, *Zizania aquatica*, and *Zea Mays*; and in all these plants the peculiar particles of the pollen, which are oval or short oblong, though somewhat reduced in number, retain their form perfectly, and exhibit evident motion, though I think not so vivid as in those belonging to the living plant. In *Viola tricolor*, in which, as well as in other species of the same natural section of the genus, the pollen has a very remarkable form, the grain on immersion in nitric acid still discharged its contents by its four angles, though with less force than in the recent plant. motion on immersion in water; and this motion was still observable in specimens both of Mosses and of Equiseta, which had been dried upwards of one hundred years.

The very unexpected fact of seeming vitality retained by these minute particles so long after the death of the plant, would not perhaps have materially lessened my confidence in the supposed peculiarity. But I at the same time observed, that on bruising the ovula or seeds of Equisetum, which at first happened accidentally, I so greatly increased the number of moving particles, that the source of the added quantity could not be doubted. I found also that on bruising first the floral leaves of Mosses, and then all other parts of those plants, that I readily obtained similar particles, not in equal quantity indeed, but equally in motion. My supposed test of the male organ was therefore necessarily abandoned.

Reflecting on all the facts with which I had now become acquainted, I was disposed to believe that the minute spherical particles or Molecules of apparently uniform size, first seen in the advanced state of the pollen of Onagrariæ, and most other Phænogamous plants,-then in the antheræ of Mosses and on the surface of the bodies regarded as the stamina of Equisetum,-and lastly in bruised portions of other parts of the same plants, were in reality the supposed constituent or elementary molecules of organic bodies, first so considered by Buffon and Needham, then by Wrisberg with greater precision, soon after and still more particularly by Müller, and, very recently, by Dr. Milne Edwards, who has revived the doctrine and supported it with much interesting detail. I now therefore expected to find these molecules in all organic bodies: and accordingly on examining the various animal and vegetable tissues, whether living or dead, they were always found to exist; and merely by bruising these substances in water, I never failed to disengage the molecules in sufficient numbers to ascertain their apparent identity in size, form, and motion, with the smaller particles of the grains of pollen.

I examined also various products of organic bodies, particularly the gum resins, and substances of vegetable origin, extending my inquiry even to pit-coal; and in all these bodies Molecules were found in abundance. I remark here also, partly as a caution to those who may hereafter engage in the same inquiry, that the dust or soot deposited on all bodies in such quantity, especially in London, is entirely composed of these molecules.

One of the substances examined, was a specimen of fossil wood, found in Wiltshire oolite, in a state to burn with flame; and as I found these molecules abundantly, and in motion in this specimen, I supposed that their existence, though in smaller quantity, might be ascertained in mineralized vegetable remains. With this view a minute portion of silicified wood, which exhibited the structure of Coniferæ, was bruised, and spherical particles, or molecules in all respects like those so frequently mentioned, were readily obtained from it; in such quantity, however, that the whole substance of the petrifaction seemed to be formed of them. But hence I inferred that these molecules were not limited to organic bodies, nor even to their products.

To establish the correctness of the inference, and to ascertain to what extent the molecules existed in mineral bodies, became the next object of inquiry. The first substance examined was a minute fragment of window-glass, from which, when merely bruised on the stage of the microscope, I readily and copiously obtained molecules agreeing in size, form, and motion with those which I had already seen.

I then proceeded to examine, and with similar results, such minerals as I either had at hand or could readily obtain, including several of the simple earths and metals, with many of their combinations.

Rocks of all ages, including those in which organic remains have never been found, yielded the molecules in abundance. Their existence was ascertained in each of the constituent minerals of granite, a fragment of the Sphinx being one of the specimens examined.

To mention all the mineral substances in which I have found these molecules, would be tedious; and I shall confine myself in this summary to an enumeration of a few of the most remarkable. These were both of aqueous and igneous origin, as travertine, stalactites, lava, obsidian, pumice, vol-

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canic ashes, and meteorites from various localities<sup>\*</sup>. Of metals I may mention manganese, nickel, plumbago, bismuth, antimony, and arsenic. In a word, in every mineral which I could reduce to a powder, sufficiently fine to be temporarily suspended in water, I found these molecules more or less copiously; and in some cases, more particularly in siliceous crystals, the whole body submitted to examination appeared to be composed of them.

In many of the substances examined, especially those of a fibrous structure, as asbestus, actinolite, tremolite, zeolite, and even steatite, along with the spherical molecules, other corpuscules were found, like short fibres somewhat moniliform, whose transverse diameter appeared not to exceed that of the molecule, of which they seemed to be primary combinations. These fibrils, when of such length as to be probably composed of not more than four or five molecules, and still more evidently when formed of two or three only, were generally in motion, as least as vivid as that of the simple molecule itself; and which from the fibril often changing its position in the fluid, and from its occasional bending, might be said to be somewhat vermicular.

In other bodies which did not exhibit these fibrils, oval particles of a size about equal to two molecules, and which were also conjectured to be primary combinations of these, were not unfrequently met with, and in motion generally more vivid than that of the simple molecule; their motion consisting in turning usually on their longer axis, and then often appearing to be flattened. Such oval particles were found to be numerous and extremely active in white arsenic.

As mineral bodies which had been fused contained the moving molecules as abundantly as those of alluvial deposits, I was desirous of ascertaining whether the mobility of the particles existing in organic bodies was in any degree affected by the application of intense heat to the containing substance. With this view small portions of wood, both living and dead, linen, paper, cotton, wool, silk, hair, and muscular fibres, were exposed to the flame of a candle or burned in

\* I have since found the molecules in the sand-tubes, formed by lightning, from Drig in Cumberland. platina forceps, heated by the blowpipe; and in all these bodies so heated, quenched in water, and immediately submitted to examination, the molecules were found, and in as evident motion as those obtained from the same substances before burning.

In some of the vegetable bodies burned in this manner, in addition to the simple molecules, primary combinations of these were observed, consisting of fibrils having transverse contractions, corresponding in number, as I conjectured, with that of the molecules composing them; and those fibrils, when not consisting of a greater number than four or five molecules, exhibited motion resembling in kind and vivacity that of the mineral fibrils already described, while longer fibrils of the same apparent diameter were at rest.

The substance found to yield these active fibrils in the largest proportion and in the most vivid motion, was the mucous coat interposed between the skin and muscles of the haddock, especially after coagulation by heat.

The fine powder produced on the under surface of the fronds of several Ferns, particularly of *Acrostichum calomelanos*, and the species nearly related to it, was found to be entirely composed of simple molecules and their primary fibrelike compounds, both of them being evidently in motion.

There are three points of great importance which I was anxious to ascertain respecting these molecules, namely, their form, whether they are of uniform size, and their absolute magnitude. I am not, however, entirely satisfied with what I have been able to determine on any of these points.

As to form, I have stated the molecule to be spherical, and this I have done with some confidence; the apparent exceptions which occurred admitting, as it seems to me, of being explained by supposing such particles to be compounds. This supposition in some of the cases is indeed hardly reconcileable with their apparent size, and requires for its support the further admission, that, in combination, the figure of the molecule may be altered. In the particles formerly considered as primary combinations of molecules, a certain change of form must also be allowed; and even the simple molecule itself has sometimes appeared to me when in motion to have been slightly modified in this respect. My manner of estimating the absolute magnitude and uniformity in size of the molecules, found in the various bodies submitted to examination, was by placing them on a micrometer divided to five thousandths of an inch, the lines of which were very distinct; or more rarely on one divided to ten thousandths, with fainter lines, not readily visible without the application of plumbago, as employed by Dr. Wollaston, but which in my subject was inadmissible.

The results so obtained can only be regarded as approximations, on which perhaps, for an obvious reason, much reliance will not be placed. From the number and degree of accordance of my observations, however, I am upon the whole disposed to believe the simple molecule to be of uniform size, though as existing in various substances and examined in circumstances more or less favourable, it is necessary to state that its diameter appeared to vary from  $\frac{1}{15,000}$ th to  $\frac{1}{20,000}$ th of an inch\*.

I shall not at present enter into additional details, nor shall I hazard any conjectures whatever respecting these molecules, which appear to be of such general existence in inorganic as well as in organic bodies; and it is only further necessary to mention the principal substances from which I have not been able to obtain them. These are oil, resin, wax and sulphur, such of the metals as I could not reduce to that minute state of division necessary for their separation, and finally, bodies soluble in water.

In returning to the subject with which my investigation commenced, and which was indeed the only object I originally had in view, I had still to examine into the probable mode of action of the larger or peculiar particles of the pollen, which, though in many cases diminished in number before the grain could possibly have been applied to the stigma, and particularly in Clarckia, the plant first examined, were yet

\* While this sheet was passing through the press, Mr. Dollond, at my request, obligingly examined the supposed pollen of *Equisetum virgatum* with his compound achromatic microscope, having in its focus a glass divided into 10,000ths of an inch, upon which the object was placed; and although the greater number of particles or molecules seen were about  $\frac{1}{20}, \frac{1}{200}$ , yet the smallest did not exceed  $\frac{1}{30}, \frac{1}{1000}$  th of an inch. in many other plants found in less diminished proportion, and might in nearly all cases be supposed to exist in sufficient quantity to form the essential agents in the process of fecundation.

I was now therefore to inquire, whether their action was confined to the external organ, or whether it were possible to follow them to the nucleus of the ovulum itself. My endeavours, however, to trace them through the tissue of the style in plants well suited for this investigation, both from the size and form of the particles, and the development of the female parts, particularly Onagrariæ, was not attended with success; and neither in this nor in any other tribe examined, have I ever been able to find them in any part of the female organ, except the stigma. Even in those families in which I have supposed the ovulum to be naked, namely, Cycadeæ and Coniferæ, I am inclined to think that the direct action of these particles, or of the pollen containing them, is exerted rather on the orifice of the proper membrane than on the apex of the included nucleus; an opinion which is in part founded on the partial withering confined to one side of the orifice of that membrane in the larch,-an appearance which I have remarked for several years.

To observers not aware of the existence of the elementary active molecules, so easily separated by pressure from all vegetable tissues, and which are disengaged and become more or less manifest in the incipient decay of semitransparent parts, it would not be difficult to trace granules through the whole length of the style: and as these granules are not always visible in the early and entire state of the organ, they would naturally be supposed to be derived from the pollen, in those cases at least in which its contained particles are not remarkably different in size and form from the molecule.

It is necessary also to observe that in many, perhaps I might say in most plants, in addition to the molecules separable from the stigma and style before the application of the pollen, other granules of greater size are obtained by pressure, which in some cases closely resemble the particles of the pollen in the same plants, and in a few cases even exceed them in size : these particles may be considered as primary combinations of

the molecules, analogous to those already noticed in mineral bodies and in various organic tissues.

From the account formerly given of Asclepiadeæ, Periploceæ, and Orchideæ, and particularly from what was observed of Asclepiadeæ, it is difficult to imagine, in this family at least, that there can be an actual transmission of particles from the mass of pollen, which does not burst, through the processes of the stigma; and even in these processes I have never been able to observe them, though they are in general sufficiently transparent to show the particles were they present. But if this be a correct statement of the structure of the sexual organs in Asclepiadeæ, the question respecting this family would no longer be, whether the particles in the pollen were transmitted through the stigma and style to the ovula, but rather whether even actual contact of these particles with the surface of the stigma were necessary to impregnation.

Finally, it may be remarked that those cases already adverted to, in which the apex of the nucleus of the ovulum, the supposed point of impregnation, is never brought into contact with the probable channels of fecundation, are more unfavourable to the opinion of the transmission of the particles of the pollen to the ovulum, than to that which considers the direct action of these particles as confined to the external parts of the female organ.

The observations, of which I have now given a brief account, were made in the months of June, July and August, 1827. Those relating merely to the form and motion of the peculiar particles of the pollen were stated, and several of the objects shown, during these months, to many of my friends, particularly to Messrs. Bauer and Bicheno, Dr. Bostock, Dr. Fitton, Mr. E. Forster, Dr. Henderson, Sir Everard Home, Captain Home, Dr. Horsfield, Mr. Kænig, M. Lagasca, Mr. Lindley, Dr. Maton, Mr. Menzies, Dr. Prout, Mr. Renouard, Dr. Roget, Mr. Stokes, and Dr. Wollaston; and the general existence of the active molecules in inorganic as well as organic bodies, their apparent indestructibility by heat; and several of the facts respecting the primary combinations of the molecules, were communicated to Dr. Wollaston and Mr. Stokes in the last week of August. None of these gentlemen are here appealed to for the correctness of any of the statements made; my sole object in citing them being to prove from the period and general extent of the communication, that my observations were made within the dates given in the title of the present summary.

The facts ascertained respecting the motion of the particles of the pollen, were never considered by me as wholly original; this motion having, as I knew, been obscurely seen by Needham, and distinctly by Gleichen, who not only observed the motion of the particles in water after the bursting of the pollen, but in several cases remarked their change of place within the entire grain. He has not, however, given any satisfactory account either of the forms or of the motions of these particles, and in some cases appears to have confounded them with the elementary molecule, whose existence he was not aware of.

Before I engaged in the inquiry in 1827, I was acquainted only with the abstract given by M. Adolphe Brongniart himself, of a very elaborate and valuable memoir, entitled "*Recherches sur le Génération et le Développement del Embryon dans les Végétaux Phanerogames*," which he had then read before the Academy of Sciences of Paris, and has since published in the *Annales des Sciences Naturelles*.

Neither in the abstract referred to, nor in the body of the memoir, which M. Brongniart has with great candour given in its original state, are there any observations, appearing of importance even to the author himself, on the motion or form of the particles; and the attempt to trace these particles to the ovulum with so imperfect a knowledge of their distinguishing characters, could hardly be expected to prove satisfactory. Late in the autumn of 1827, however, M. Brongniart having at his command a microscope constructed by Amici, the celebrated professor of Modena, he was enabled to ascertain many important facts on both these points, the result of which he has given in the notes annexed to his memoir. On the general accuracy of his observations on the motions, form, and size of the granules, as he terms the particles, I place great reliance. But in attempting to trace these particles through their whole course, he has overlooked two points of the greatest importance in the investigation.

For, in the first place, he was evidently unacquainted with the fact, that the active spherical molecules generally exist in the grain of pollen along with its proper particles; nor does it appear from any part of his memoir that he was aware of the existence of molecules having spontaneous or inherent motion, and distinct from the peculiar particles of the pollen, though he has doubtless seen them, and in some cases, as it seems to me, described them as those particles.

Secondly, he has been satisfied with the external appearance of the parts in coming to his conclusion, that no particles capable of motion exist in the style or stigma before impregnation.

That both simple molecules and larger particles of different form, and equally capable of motion, do exist in these parts, before the application of the pollen to the stigma can possibly take place, in many of the plants submitted by him to examination, may easily be ascertained; particularly in *Antirrhinum majus*, of which he has given a figure in a more advanced state, representing these molecules or particles, which he supposes to have been derived from the grains of pollen, adhering to the stigma.

There are some other points respecting the grains of pollen and their contained particles in which I also differ from M. Brongniart, namely, in his supposition that the particles are not formed in the grain itself, but in the cavity of the anthera; in his assertion respecting the presence of pores on the surface of the grain in its early state through which the particles formed in the anthera, pass into its cavity; and lastly, on the existence of a membrane forming the coat of his boyau or mass of cylindrical form ejected from the grain of pollen.

I reserve, however, my observations on these and several other topics connected with the subject of the present inquiry for the more detailed account, which it is my intention to give.

July 30, 1828.

THE END.

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