The Plasma Membrane, with Notes on the History of Botany

By Homer W. Smith, Sc.D.

The origin of the concept of the plasma membrane is rather ambiguously identified. It antedated Pfeffer and de Vries, and, after searching the literature, the writer attributes it to Carl Nägeli (1855), who had been working with algae, fungi, mosses, and unicellular plants. He noted particularly that the surface of discontinuity of the cell is impermeable to pigments contained within the cytoplasm or added to the solution around the cell. He found the protoplasmic surface to be more dense, viscous, and otherwise distinguishable from the cytoplasm, and hence he called the surface layer the plasma membrane. From experiments on osmosis he concluded that the plasma membrane endows the cell with its osmotic properties.

A FEW WEEKS back Dr. Francis O. Schmitt was in my office, and I talked with him about the plasma membrane, expressing my regret that he could not participate in this symposium, especially in what the jurist would call the summation. Then he asked me: Would I rather open a symposium or close it? My answer was almost automatic—I would rather open it, because then I can sit back and relax.

The questions before us are chiefly two: Is there a plasma membrane, and if so, what is it?

As the terms are commonly used today, the plasma membrane designates something at the surface of discontinuity of protoplasm; it differs in physical consistency and in many other respects from the underlying cytoplasm, but often it is indistinguishable cytologically from the latter. Its most important characteristics have been well epitomized by Dr. Schmitt:

'The plasma membrane serves not only to enclose the cell and to direct the molecular traffic into and out of the cell but also, presumably, to mount the biochemical mechanisms by which solutes, such as sodium ions, may be transported or "pumped" against an activity gradient.'

As Otto Loewi frequently said to me, 'Homer, it's all in the plasma membrane,' echoing a long-standing pharmacological sus-

From the Department of Physiology and Biophysics, New York University School of Medicine, New York, N. Y.
used in 1665 by Robert Hooke (1653–1703) (Micrographia) to describe the minute cavities which he discovered in cork and other vegetable tissues. The 'cell' walls were regarded by him and others after him as composed of the 'interstitial substance.' Marcellus Malpighi (1628–1694) (De viscerum structura: exercitatio anatomica, 1666; Anatome plantarum, 1672) and Nehemiah Grew (1641–1712) (Anatomy of Vegetables Begun, 1672) added many new details and recognized that neither animal nor vegetable cells were just empty holes; but better microscopes were needed before Ludolph Christian Traversius (1779–1864) in 1806 could observe, even if he could not accurately interpret, the elongation and duplication of plant cells. The modern cell theory was presciently forecast in 1824 by the French physiologist and botanist Rene Jocachim Henri Dutrochet (1776–1847) (who introduced the manometer into the study of osmotic pressure) when he said in his Recherches Anatomique et Physiologique that 'all organic tissues are actually globular cells of exceedingly smallness, which appear to be united only by simple adhesive forces.' Dutrochet's statement was at best an informed guess, but it had the supreme virtue of being right.

The nucleus in the ovule of a lily was described in 1831 by the English botanist Robert Brown (1773–1858) (whose name is known to you in connection with the movements of microscopic particles which he had reported in 1827), and the nucleolus was described in 1836 by the anatomist and embryologist, Gabriel Gustav Valentine (1810–1883). These cell 'inclusions' were at the time, however, only matters for speculation.

It remained for Matthias Jacob Schleiden (1804–1881), then an undergraduate student in biology at Jena, to show, first, that a nucleated cell is the only original constituent of the plant embryo (c. 1837); that all plant cells come from pre-existing plant cells (as against the doctrine of spontaneous generation) (1838); and third, to contribute his monumental Grundzüge der wissenschaftlichen Botanik (Principles of Scientific Botany; 1842–43) which supplied students with the first modern textbook in this science.* This work went through several editions and was translated into English, and it did much to shake the purely systematic Linnean school, whose accumulations Schleiden irreverently described as 'hay.' By his speculative activity and by the introduction of improved technical methods, he gave so vivid an impulse to the younger botanists of his time that he has been called the 'reformer of scientific botany.'

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Schleiden was a great botanist, but he was also something of a prig. The very title of his textbook presumptuously implied that all botany before his time had been unscientific. If he was out to influence botanists, he apparently had little desire to make them his friends, as will be shown by one paragraph from his book:

'If we consider the attempts that have hitherto been made to subject the life of plants to scientific observation, we shall find that all those who have conducted them have brought to their works groundless prejudices, and, following the old beaten track, have not even paused to inquire whether or not it were right, and whether or not their prejudices were just; and they have even taken these latter as leading maxims to form the basis of all their investigations. I have already discussed the fanciful analogy between the physiology of

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*Previous to 1840, the student of botany had no general textbook other than the Théorie Élémentaire de Botanique (1813) and Physiologie Végétale of Augustine Pyrame de Candolle. 2, 5

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animals and of plants. In consequence of the use of this absurd analogy, almost all the works which have hitherto appeared on vegetable physiology are perfectly worthless, for in no instance have they adopted the only true fundamental position, namely, the essential peculiarity of vegetable life; nay, the larger number of writers have not even given a comprehensive view of the facts already known, as such would have destroyed their assumed principles. Each branch of natural science, if it would lay claim to such name, must have its own peculiar independent principle of development, which must be drawn from its own data, and only thence. It is not until considerable advance had been made towards perfection that it is safe to begin to inquire whether analogies exist between itself and some other branch of natural science, and, if so, what they are. The manner in which science is usually pursued is not following it out gradually through a long course of original investigations, but by grasping hastily at all statements and dogmas that are afloat respecting it, seeking to participate in its treasures as an inheritance from strangers, rather than by examining into its foundations and building up its structure; this is the reason that we find even more dangerous prejudices to combat in science than in practical life.

Thus it has been with botany: books have been written when plants should have been examined, conjectures have been made when investigations should have been pursued. Hence for about a century we have but revolved in a circle, without making the least advance or discovering new facts; and new laws are given us which are only the result of the play of chances, whilst correct fundamental maxims and correct methods of advance would have guaranteed the solution of various problems, and secured the progress of the science.

Let us inquire quickly: Just who was Schleiden animadverting so acidulously in his 'scientific' textbook? Prehistoric man must have studied plants both as a source of food and drugs, and this distinction is itself just on the threshold of scientific observation. King Solomon spoke of 'the cedar tree that is in Lebanon, and even of the hyssop that springeth out of the wall,' and, according to my Oxford Concordance, plants, trees, and flowers are mentioned over forty times in the Old Testament. Nor was Solomon the first to make discriminating botanical observations; the ancient Mesopotamians, Egyptians, Hindus and Chinese knew one flower from another, and the flowers from the bees. The use of herbs had early become welded to medicine, and the Greek satirist Lucian (c. 120–180), in his Dialogues of the Gods, has Hercules call Aesculapius 'a root digger and a wandering quack.'

Theophrastus (b. c. 372–287 B.C.) of Eresus, friend of Plato and successor to Aristotle, who presided over the Peripatetic school for thirty-five years and was guardian of Aristotle's children, in his two large botanical treatises Historia Plantarum and De Causis Plantarum, written about 300 B.C., defined root, stem, branch, twig, leaf, flower, and fruit, mentioning some 500 types of plants; he knew, as did the early Mesopotamians, that the fruit-bearing fig and date trees remain barren unless the nonfruit-bearing (male) palm is in flower and its dust is shaken over the flower of the female; and he rejected as fabulous the belief that wheat can turn into barley or vice versa. He recognized the essential differences between stem, leaf, and seed in the monocotyledons and dicotyledons. Nothing substantial was added to his work until the days of Grew and Malpighi in the seventeenth century. The Greek writer Dioscorides, who lived about the time of Nero, wrote a treatise on materia medica (c. 64) which became one of the most assiduously studied of all textbooks and, like Galen's works, carried such authority that the greater part of the new botanical matter published during the whole of the sixteenth and part of the seventeenth centuries appeared as annotations upon it. It was Dioscorides, not Linnaeus, who first recognized natural families among the plants. Pliny the Elder (c. 23–79 C.E.), author of a Naturalis Historia, described about a thousand plants, many of them esteemed for their me-
dicinal virtues, but, like many compilers, he was content with a hodgepodge of good and bad information.

If we omit the herbalists who wrote about plants incidentally and with no intent to improve our knowledge of plant life, after Theophrastus eighteen centuries elapsed before, in the general renaissance of knowledge, there emerged a botanist worthy of the name. When the botanical renaissance did come, the subject evoked the almost simultaneous interest of numerous students who were determined to take a new look at things.

Notable among the pioneers of the botanical renaissance was Valerius Cordus (b. 1515) of Wittenberg, whose Historia Plantarum was written when he was twenty-five years old but not published (1561) until seventeen years after his death. Son of poor but informed parents who squeezed ends tight to send him to the famous university, it has been said of him that 'to the best possible education of an intellect naturally keen there was united that happy temperament to which nothing is impossible or even difficult of attainment. To these gifts he added a truly marvelous industry and assiduity in research; and above all a most retentive memory for everything he either saw in nature or read in books.\(^2\)\(^{,}\)\(^14\) Cordus has been called the Harvey of botany, 'the first to teach men,' as Haller said, 'to cease dependence on the poor [and so frequently inaccurate] descriptions of the ancients and to describe plants anew from Nature.' In his Historia Plantarum he described between 400 and 500 types, and to perfect the book before giving it to the printer he went on a natural history pilgrimage to Padua, Ferrara, Pisa, Bologna, Florence, Lucca, and finally to Rome where he contracted malaria and died at the age of twenty-nine. His works were published posthumously by Konrad von Gesner (1516–1565), a famous Alpinist and encyclopedist and a botanist and zoologist of no mean merit.\(^2\)\(^{,}\)\(^14\)

Because Cordus's work was for so long unpublished, actual priority in the botanical renaissance goes to three herbalists, Brunfels, Fuchs, and Bock. Otho Brunfels, a Carthusian monk, said in his Herbarum Vivae Icones (c. 1530), 'In this whole work I have no other end in view than that of giving a prop to fallen botany; to bring back to life a science almost extinct. And because this has seemed to me to be in no other way possible than by thrusting aside all the old herbals, and publishing new and really life-like engravings, and along with them accurate descriptions extracted from ancient and trustworthy authors, I have attempted both, using the greatest care and pains that both shall be faithfully done.' The book is illustrated by some 300 figures of variable excellence, but the figures and the descriptions do not always agree, and the historian Lee Greene remarks that Brunfels probably did not advance the art of plant description by a syllable. However, Brunfels did reform the nomenclature of genera by the exclusion of names made up of two words, 220 years before Linnaeus who is usually credited with this principle.

Leonhard Fuchs's Historia Stirpium appeared in 1542; the book was illustrated with excellent woodcuts, but it added nothing new. Fuchs's interest was not in plants as plants but only in plants as drugs. Hieronymus Bock's Kreuterbuch (1546) was apparently aimed at describing plants in such a manner as to make them identifiable by the description only because he could not afford the elaborate woodcuts which illustrated the works of Brunfels and Fuchs. Again it added little to true botanical knowledge and in some ways was retrogressive: For example, he believed that orchids had no seeds but arose from the excreta of birds and that fungi were 'merely the superfluous moisture of trees, of rotten wood, and other rotten things.' He accepted spontaneous generation and the transmutation of one cereal into another, notions which had been rejected by Theophrastus as fabulous, but he alone of these three herbalists went directly to nature for his information. Despite the fact that Cordus's work did not reach publication during his lifetime, he towers above the three other men. He had the genius of the real founder of botany who had lived eighteen centuries before.\(^2\)

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In this period the only English botanist of note was William Turner (c. 1515–1568), who published a History of Plants in 1551 and succeeding years. The later years of the century saw the appearance of the Stirpium Adversaria Nova by Lobelius (1538–1616), published in 1576, which attempted to classify plants into groups, some of which, such as the Cruciferae and the Labiatae, are recognized to this day. The Stirpium Historiae Pemptades Sex, published in 1583 by a Dutch herbalist called Dodonaeus, formed the basis of several English herbals. More important is De Plantis of Andreas Caesalpinus (1519–1603), which also appeared in 1583. Caesalpinus’s book is notable because it separated botany from materia medica for the first time, the two having been wedded from the days of Theophrastus and before; it had the fault that its author settled on his system of classification first and then twisted experience around to make it fit his system. He distributed the 1,520 plants described by him into 15 classes, the distinguishing characteristics being taken from the fruit. But De Plantis advances some novel ideas: The chief function of leaves is to protect young buds and fruits from air and light; since plants need less food than animals, they have no blood vessels; and after careful thought he localized the plant soul at the junction of the stem and root.

In searching for the location of the vegetable soul Caesalpinus was following in the steps of Aristotle, whose grip on the world could not easily be shaken off. Giordano Bruno (1548–1600), the Italian philosopher who was a contemporary of Caesalpinus, relates that at Oxford in his day masters and bachelors who did not follow Aristotle faithfully were liable to a fine of five shillings for every point of divergence and for every fault committed against the logic of the Organon. Even so, Oxford let heretics off lightly; after seven years of confinement, Bruno was burned at the stake in Rome in 1600 for scoffing at Aristotle, for expressing his low opinions of monks, miracles, prayer, the Jewish records, etc., and for his contemptuous attitude toward the Roman Church generally. Perhaps Caesalpinus was fortunate to be a botanist, since the Inquisitors did not take an interest in the souls of plants.

One positive thing can be said of the late sixteenth century: Men who now wrote books on botany had gardens which they tended with loving care, either their own or belonging to some philanthropist or to a civic center. Their gardens were their botanical laboratories, and plant life had come into its own as worthy of study, a matter the cogency of which every microbiologist today will recognize. John Gerard (1545–1612), a barber-surgeon, had a garden in Holborn where he maintained a collection of British plants and from which he issued his Herbal in 1597, which offered a crude system of classification based on the external appearance of plants and their uses to man. The garden at Padua had been started in 1545, at Pisa in 1547, at Bologna in 1567, and soon Leyden in the Netherlands, Montpellier in France, Heidelberg in Germany, and many other cities followed suit. The Royal Botanic Gardens at Kew were taken under the aegis of the Crown in 1840, and the New York Botanical Garden in the Bronx was founded in 1891.

While taxonomists Kaspar Bauhin (1560–1624), Joachim Jung (1587–1657), and others were seeking to discover a ‘natural’ method of classifying plants, lenses were coming into use as aids to eyesight. A double-lens magnifier may have been invented before the time of Roger Bacon (c. 1294), but it first emerges from historical obscurity in the 1590’s, when Dutch spectacle makers greatly improved the art of grinding lenses. The two-lens system reached the physicist Galileo in Padua in June of 1609, and he quickly devised a lead tube, fitted at one end with a planoconvex, at the other with a planoconcave lens, bringing the instrument to an optical efficiency which it could not have hitherto possessed. By appropriate focusing, Galileo’s ‘trunk’ or ‘cylinder’ could be used either as a telescope or microscope. It was of course as a telescope that Galileo himself put it to its most effective use, as described in his Siderius Nuncius.
(1610), a short pamphlet which, so it has been said, records more world-shaking discoveries than any other book of whatever length ever written. However, Galileo also used his cylinder as a microscope and in 1610 observed the legs and eyes and other anatomical features of minute animals, such as insects. Great labor and skill are required to grind good lenses, and through the first half of the seventeenth century the use of the microscope was limited to a small, intimate, and wealthy coterie of Italians, who called themselves the Accademia dei Lincei (Companions of the Lynx). Among the notable achievements of the Lincei was a work entitled the *Apiarium* (1625), containing descriptions of every bee and wasp known to the members of the Academy and including some American species. This seems to be the first recorded treatise on microscopic anatomy. Another discovery, more important to us here, was the identification of the spores of a fern, hitherto thought to be seedless, by Cesi in 1625, perhaps the first objects beyond the range of unaided vision to be revealed by the microscope. Galileo’s ‘cylinder’ was probably well known before 1620, but it was not until the middle of the century that it was used, to the enrichment of all biology, by Malpighi of Italy, Robert Hooke (1635–1703) and Nehemiah Grew of London, Jan Swammerdam (1637–1680) of Amsterdam, and Antony van Leeuwenhoek (1632–1723) of Delft.6

When Hooke used the word *cellula* (1665), he failed to consider what might be, or might have been, contained within his ‘cells.’ But in the hands of Malpighi and Grew the microscope revealed that the cells were not empty. Essays by these two men were submitted to the Royal Society at nearly the same time, and they both bear the publication date of 1671, though Grew is entitled to claim priority because his essay reached the Royal Society first.* Malpighi and Grew never had any argument over priority, however, and indeed Grew proposed discontinuing his own researches in order to leave the field open for his rival in Italy, but ‘luckily for science the Royal Society dissuaded him from such an exhibition of self-sacrificing renunciation.’4 p. 30

You have heard much about Malpighi’s anatomy, if not in botany, but permit me one quotation from Grew, who apologizes in advance, as it were, for his possible shortcomings:

‘I know it will be difficult,’ he says, ‘to make observations of this kind upon the organical parts of plants,’ but nevertheless he takes heart in the following words: ‘For what we obtain of Nature, we must not do it by commanding, but by courting of her; . . . wherever men will go beyond phansie and imagination, depending upon the conduct of Divine wisdom, they must labour, hope and persevere. And as the means propounded, are all necessary, so they may, in some measure, prove effectual. How far I promise not; the way is long and dark; . . . the way of Nature, is so impervious, and, as I may say, downhill and uphill, that how far so ever we go, yet the surmounting of one difficulty, is wont still to give us the prospect of another . . . To conclude, if but little should be effected, yet to design more, can do us no harm; for a man shall never be able to hit stars by shooting at them; yet he shall come much nearer to them, than another that throws at apples.’7 p. 31

All copies of the Transactions available have been bound, and in the process the date-bearing covers have been stripped. As pointed out elsewhere this leaves a moot point as to whether Carl Ludwig had seen Bowman’s article before Ludwig gave his two famous Marburg addresses on the kidney. The writer is convinced that Ludwig had not.*

Harvey-Gibson incorrectly says that Grew first introduced the word *parenchyma*. On the contrary, this word was used by Erasistratus (340–280 B.C.), of the Alexandrian school of medicine, to characterize the lungs, liver, and other soft tissues (as distinct from the bones, tendons, and blood vessels), which were conceived to be gelatinous masses formed by coagulation of the blood between the ends of the large arteries and veins. The word *parenchyma* is formed from *para* (between) + *en* (in) + *chein* (to pour) and hence literally means ‘poured in between.’

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If the writer were asked to date the beginnings of modern botany, he would name Grew's paper, which reflected the introduction of the microscope. Attempts to describe plants are unquestionably older than Theophrastus, and all efforts to attain a scientific taxonomy had to remain more or less fruitless until after the publication and assimilation of the Origin of Species (1859)—which leaves even Linnaeus in the botanical Dark Ages. An adequate taxonomy was ultimately to come, but the effort at 'classification' long presented a jungle to get lost in.*

It was John Ray (1627-1705) who laid the foundations of the 'natural system' of taxonomy in his Methodus Plantarum (1682): He separated the flowering from the flowerless plants and divided the former into dicotyledons and monocotyledons, and his classes were founded to some extent on a correct idea of the affinities of plants. Ray's Synopsis Methodica Stirpium Britannicarum, though a small volume, long served as the vade mecum of the naturalist on his botanical excursions.† Jakob Rudolf Camerarius (1665-1721) is generally given credit for the demonstration of sex in plants in 1694, but Ray really has the priority. Initially at Cambridge, Ray had worked the Fenland, the Midlands, North Wales, Cornwall, and the southern counties; he was a Puritan divine and found himself unable to accept the Act of Uniformity, and he and a friend traveled over the continent for three years, after which he retired to Essex to write his Historia Plantarum Generalis (1686). He has been called the greatest European botanist of the seventeenth century.

Robert Morison (1620-1683), the first professor of botany at the University of Oxford, in his taxonomy (Practidia Botanica, 1672, and Plantarum Historia Universalias, 1680) largely followed Casalpinus, dividing plants into 18 classes, distinguishing them according as they were woody or herbaceous, and taking into account the nature of the flowers and fruit. Augustus Quirinus Rivinus (1652-1723) (Introductio Generalis in Rem Herbarium, 1690) promulgated a classification based on the forms of the flowers. J. P. de Tournefort (1656-1708) (Élémens de Botanique, 1694; Institutiones Rei Herbariae, 1700), who was long at the head of the French school, described about 8,000 'species' of plants and distributed them into 22 classes, chiefly according to the form of the corolla, distinguishing herbs and undershrubs on the one hand from trees and true shrubs on the other.

The ultimate in taxonomic effort was achieved by Carl von Linne, or Linnaeus (1707-1778).§ There are almost as many different opinions of Linnaeus as there are writers. Of him, Harvey-Gibson says:

'Linnaeus' outstanding characteristic is his power of describing and systemizing. Sachs [the historian of botany] calls him a classifying, coordinating, and subordinating machine; he even classified the very botanists whose works he studied. He collected all the works of his predecessors, picked out all he thought best in them, and with scissors and paste, welded the scraps into one concrete whole. Not that he palmed off the compilation as his own original production—far from it. Each author received due credit for what he had accomplished. He picked out a beam here, a tile there, a block of stone somewhere else, and out of the total mass of constructive material he built a house. His pre-eminent skill as a literary architect enabled him to erect a building that was hailed as a masterpiece both by his contemporaries and by generations of admiring pupils; a house perhaps convenient to dwell in at the time it was erected but unadaptable and quite unsuited to more modern requirements. It was as though the Hall of Cedric the Saxon had been offered to you as a residence in the twentieth century; where would you find accommodation for your modern furniture, even if you were successful in obtaining permission from sanitary, municipal, and other authorities to live in it at all?"**

This satiric masterpiece may be balanced by

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*One of the writer's colleagues has an aphorism to the effect that when a man starts classifying disease(s), it is a sign that he doesn't know anything about it (them).

† There have been attempts to make use of Linnaeus' system, partly according to the ideas of the times. The attempt of Amedeo, the last of the systematists, to incorporate plants in the animal kingdom, seems to have been a misunderstanding, and it was not until J. H. M. C. Parke (1737-1812) and F. T. James (1780-1828) that a real attempt was made to adapt the Linnaean system to the requirements of modern botany. Parke's Systema Vegetabilum (1789) was the first comprehensive work on the subject, and James' Systema Vegetabilium (1813) was the last attempt to make the system a practical one.

§ The system that Linnaeus established is still the basis of botanical classification. It is based on the form of the flower, and is divided into three classes: flowering plants, coniferous plants, and non-vascular plants. The classes are divided into orders, the orders into families, the families into genera, and the genera into species.

** The Hall of Cedric the Saxon is a building in the English countryside, located at the village of Cedric in the county of Lincolnshire. It is said to be the oldest inhabited building in England, and is said to have been built by Cedric the Saxon, a famous Saxon warrior. It has been the subject of much debate, with some historians claiming that it is a genuine medieval building, while others believe it to be a modern construction.
some favorable comments. Linnaeus is credited with the permanent establishment of the binomial nomenclature (for genus and species) by means of a terminology largely founded on, if not borrowed from, the botanist Jung, replacing the long-winded and confused descriptions of the herbalists by clear and succinct diagnosis. Linnaeus did not invent the binomial nomenclature; the germ of the idea is to be found in Bauhin’s *Pinax* or even in the *Enquiry* of Theophrastus. In the opinion of Bentham, his great successor in taxonomy, ‘it was reserved for the master mind of the immortal Swede to fix, by the establishment of genera and species, upon sound philosophical principles, a firm stage to serve as a basis and starting point for further progress and exploration. By his accurate discrimination by genera and species he really made possible the subsequent generalization of De Jussieu and De Candolle.’ Linnaeus, however, showed an utter incapacity for careful investigation of any object at all difficult to observe, and apparently he made not a single discovery of the slightest import. By those who are not enthusiastic about him, it has been said that his *Philosophia Botanica* (1751) and earlier works set botany back by a full century.2 p. 52

The pre-Darwinian period of plant taxonomy closes with the work of three men, Antoine Laurent de Jussieu (1748–1836), Joseph Gottlieb Koelreuter (1733–1806), and Conrad Sprengel (1766–1833). De Jussieu (*Genera Plantarum*, 1789) was, like most of his predecessors, a firm believer in the constancy of species, and consequently his work consisted mainly of shuffling the old cards around. His interest in botany was stimulated by the fact that his uncle, Bernard de Jussieu, was the custodian of the gardens at Versailles; here Antoine laid out the plants according to a plan that was adapted from Ray and more nearly approached a natural classification than any hitherto proposed.3 Where Ray had strongly suspected sexuality in plants but required more decided proofs, Jakob Rudolf Camerarius (1665–1721) a few years afterwards provided some of these proofs by experiments on castrated flowers, and Koelreuter, about 1760, discovered the functions of nectar and the part played by insects and by the wind in flower pollination. He also made notable observations on the structure of the pollen grain, a difficult task considering the imperfect state of the microscope in his day. He practiced artificial pollination and established the prepotency of the pollen taken from flowers of the same species. He was successful also in obtaining hybrids between plants of different species and thus ‘driving the first nail into the coffin of the dogma of the constancy of species.”2 p. 60 The real connection between floral morphology and insect visitation was described by Kurt Sprengel (1766–1833) in 1795. Sprengel’s main thesis is that the structure of the flower can be interpreted only by considering the duty of each part in relation to the visits of insects. The color and scent are the signboards held out to attract the visitor’s attention; the markings on the corolla are guides showing the way to the hidden nectar supplies. He raised the question of why a flower cannot be fertilized by its own pollen: The answer to the problem of cross-pollination was to be given by Charles Darwin and Gregor Mendel in the next century. Of all works mentioned, that of Sprengel is the most notable up to this date.

We have dwelt with the problem of taxonomy at such length that other matters must be indicated only by name: Christian Wolff’s studies on plant nutrition had to remain vitalistic and unprofitable until Friedrich Wöhler’s (1800–1882) synthesis of urea (1830) had dealt vitalism its death blow,* and until Justus von Liebig (1803–1873) showed that carbon dioxide, ammonia, and water are the primary requisites for plant nutrition, disposing of the ‘humus’ theory.

*Harvey-Gibson*2 p. 560 says, ‘After 1840 we hear no more of this *deus ex machina* [vitalism] that [had been] dragged in to account for everything that could not be explained by a reference to the known laws of physics and chemistry.’ One wishes that were true!39

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Stephen Hales (1677–1761) (who demonstrated and measured the blood pressure in the horse in 1733), observing the continued bleeding of a badly pruned vine (Vegetable Staticks, 1727), undertook extensive studies of the loss of water in plants by evaporation, the rate of growth of shoots and leaves, variations in root force at different times of the day, etc. In fact, up to Hales’s time, practically all that was known of plant physiology could almost have been written on a sheet of note paper. Hales has been called the founder of experimental physiology of plants.

Plant respiration had to wait for the development of the chemistry of gases. In the early seventeenth century, Jan Baptista van Helmont (1577–1644), a pupil of Paracelsus, initiated this development by some simple but ingenious experiments. Van Helmont was, in a sense, a modernized Paracelsus; he had Paracelsus’ mystic impulses and propounded equally fantastic schemes wherein invisible, supernatural forces ruled the world; for the greater and lesser archet of Paracelsus he substituted blas, by which he meant a hierarchy of forces governing the heavens and the body; minor blas presided over muscular movements, the nerves, chemical processes, and other manifestations of living things.

Though in the matter of his blas Van Helmont did not advance beyond his master, he made a real step forward in another direction. To describe an invisible and seemingly imponderable substance in the air, he coined the word gas; in his notion of this medium and its relation to life, he was not only the first of the modern chemists, but the first of the chemical physiologists. He chose the word gas because it sounded to him like ‘chaos’ and because his imponderable substance had much in common with the latter. Actually, what Van Helmont meant by gas is now recognized to be carbon dioxide; he did not suspect that there were several different kinds of gases. He thought that there were only two elements, air and water, and he performed many experiments to show that neither of these can ever be changed into the other, but gas he believed to be a form of water. Apart from his experiments with gas, he is justly famous for his experiments with the willow. He planted a young willow which weighed 16 pounds in 200 pounds of dry earth and watered it for a period of five years. The vessel containing the earth was carefully covered so that no dust could accumulate in it. As one might expect, at the end of five years the earth still weighed 200 pounds, but the willow had gained in weight 164 pounds. Obviously, said Van Helmont, 164 pounds of wood, bark, and roots had been elaborated out of water. His conclusion was, of course, erroneous, but one knows he would have been delighted to learn that the willow had been elaborated in part from water and in part from his gas, carbon dioxide. He also demonstrated the liberation of carbon dioxide when wood and gunpowder were burned and during the fermentation of grapes and malt. He was so impressed by the latter process that he was led to search for ‘ferments’ at the heart of everything, these ferments operating, of course, under the direction of his blas. Even though his blas contributed to vitalism, Van Helmont made signal contributions to botany by emphasizing the application of chemistry to plant physiology, by drawing attention to the process of fermentation, and by the discovery of a specific kind of matter, his gas.

In 1754, Joseph Black (1728–1799) ‘rediscovered’ Van Helmont’s gas as a specific chemical compound, which he called fixed air and which Lavoisier later called carbonic acid gas. John Mayow (1645–1679) in 1674 had discovered a gas which he called spiritus nitroaerus (oxygen), but it was not until 1774 that Joseph Priestley (1733–1804) made pure oxygen and, under phlogiston theory, called it dephlogisticated air. Historians debate as to who discovered ‘oxygen’—the English flatly answer that Priestley did, while others enter a forcible reservation in favor of Lavoisier. That Priestley had oxygen in his flask of ‘dephlogisticated air’ is undeniable, but so did Mayow, exactly a century before; and it is equally patent that beyond the fact that dephlogisticated air was useful to keep the phlogiston theory alive, Priestley had no
proper idea of the role of oxygen in calcination and combustion or of its true chemical properties. Lavoisier showed that during calcination (and combustion), when heat is given off, something is added to the substrate to increase the weight of the product and that this added substance is oxygen, which he so named because the products of oxidation are generally acids (oxy acid + gen to make).

Priestley, however, made one distinct positive contribution: In his famous Experiments and Observations on Different Kinds of Air, he showed that air which had been vitiated by the burning of a candle could be restored to ‘normal’ by growing vegetation in it. This opened the way for the real founder of the physiology of plant respiration, Jan Ingen-Housz (1730–1799), to show in his Experiments on Vegetables (1779) that in the sunlight plants absorb carbon and give off oxygen, while in the dark they exhale an excess of carbon, as animals do. The exhalation of oxygen begins at sunrise and increases in amount with the intensity of sunlight and duration of exposure. Only the leaves and petioles possess this respiratory function (not the flowers, fruits, and roots). Plants obtain their juices from the soil by their roots, their carbon from the air, and from these elaborate the substances of the stems, leaves, flowers, fruits, etc. After Lavoisier’s work, Ingen-Housz was able to formulate his respiratory cycle specifically in terms of carbon dioxide and oxygen. Neither Ingen-Housz nor his contemporary Senebier, who published between 1782 and 1790, realized that the exhaled oxygen was derived from the inhaled carbon dioxide, that the respiratory process depended on the green pigment in the leaves and petioles, or that the whole of the animal kingdom was dependent on the products of plant respiration and nutrition.

Nicholas Théodore de Saussure (1767–1845) is credited with the first quantitative observation on plant respiration. In his Recherches Chimique sur la Végétation (1804), he recognized the important role of the green pigment (chlorophyll) in the carbon dioxide: oxygen cycle, but he erroneously thought that other, incidental pigments participated in respiration. He held the erroneous theory that plants derived their nitrogen from animal and plant wastes or from ammonia compounds formed therefrom, and thus he started the ‘humus theory,’ which remained popular until it was disproved about the middle of the century. In the studies of De Saussure, we meet the experimental method for the first time, and botany begins to take on a modern look.2, p. 79

* * *

Everything within the cell wall had been lumped together as ‘cell contents,’ ‘nutrient sap,’ ‘vital juice,’ etc., which was of little aid to plant physiology and particularly plant movements. Every observer—even the most ignorant—had been aware how tendrils twine around immovable stems in order to support a climbing vine; how some plants keep their leaves or flowers turned toward the sun; how from the seedling the stems grow upwards, the roots downwards; and how the rootlets sought a moister soil—heliotropism, geotropism, and hydrotropism; and botanists, at least, were familiar with mimosa, the Venus’s-flytrap, and other sensitive plants, but they had been content to take it for granted that that is the way plants grow. The first to challenge this easy answer was Thomas Andrew Knight (1759–1838). He hesitated over the word sensitivity because it had too many animal connotations, and according to Aristotle only animals possessed sensitivity and the power of motion, so he attributed plant motions to mechanical forces. In 1810, he defied the force of gravity by raising seeds on the circumference of a wheel rotating in the vertical plane, saw them sprout contrary to nature, their rootlets growing outward under centrifugal force, their buds [plumules] growing inward to meet at the center of rotation. He also demonstrated that hydrotropism could overcome geotropism. The first experiment was afterwards extended by Dutrochet to leaves, whereby he showed that these are also subject

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to the effect of gravity, turning their under-
surface towards the periphery of the wheel.*

As we enter the nineteenth century, the
development of botany divides into several
paths, the one of immediate interest to us
being that leading to the cell theory. Dutro-
chet's prediction was coming true, if but
slowly, because even at the middle of the cen-
tury the botanists' armamentarium consisted
of a microscope and some plants, and perhaps
a barber's razor, which could be purchased
for one shaving and with which he could cut
reasonably thin sections, or he could use Val-
entin's knife, which consisted of two parallel
blades that, by means of a screw, could be
brought close to each other. Paraffin embed-
ding was introduced by Theodor Albrecht
Edwin Klebs (1834-1913) in 1860; an ade-
quate microtome was first introduced by Wil-
helm His, Sr., (1831-1904) in 1866 but not
perfected until 1875, and the rotary micro-
tome was not developed until 1885. The essen-
tial point was staining the cells, for which
purpose they had to be killed or 'fixed,' be-
cause dyes, as we later shall see, do not enter
the living cell, and fixation led to the prob-
lem of artifacts, a problem which is still with
us.

A few natural pigments had long been avail-
able: saffron, cinnabar, carmine, lead chro-
mate, indigo, suspensions of India ink, the
juice of blueberries, popeberries, red cabbage,
purpurin-alizarin of madder root, indigo, lit-
mus, gamboge, vermilion, logwood, and, most
important of all, cochineal, but these pigments
were of no avail on living cells. Chromic
acid, alum, lead acetate, and other fixatives had
been in use before 1850, but it was not until
1865 that Schultz introduced osmium tetrox-
ide. The first aniline dyes were introduced in
1862.\(^\text{3}\)

To study unstained living cells without such
ancillary methods of microscopy as were avail-
able from 1870 on was no easy task, and that
investigators made the progress they did is
something to be marveled at. Study of com-
plex animal tissues was scarcely advanced
beyond the days of Malpighi; and botanical
microscopy was practically confined to dia-
toms, Algae, Fungi, Spirogyra, and the mis-
cellaneous little plants (Confera glomerata)
that comprise the scum on the surface of a
fresh-water pond. The introduction in 1812,
by Johann Jakob Paul Moldenhauer (1766-
1827), of the technique of macerating tissues
in water, by means of which he was able to
isolate cells and fibers and investigate them
separately under the microscope, can in retro-
spect be seen to be an almost dramatic advance
over the older method of 'doing as best one
could.' The 'teased' preparation was used by
the early anatomists (with considerable
success). With this technique Moldenhauer
made many discoveries and was enabled to
present a new conception of the architecture
of the plant organs. More importantly, he
supplied the technique which was to prove so
important in the hands of von Mohl, Prings-
heim, Nägeli, and later botanists.

At the middle of the century two men, apart
from Schleiden, were most prominent among
botanists, Hugo von Mohl and Carl Nägeli.
In his earlier period von Mohl had, simulta-
neously with Dutrochet, studied the move-
ments of tendrils, twining stems, and mobile
leaves.\(^\text{2, p. 106}\) In 1838, he made the first serious
attempt to examine the chlorophyll apparatus
and showed that the pigment did not occur in
solution in the cell but in the form of minute
green masses (chloroplasts) of various shapes,
which also contained granules of starch—the
latter point proving to be important in the
subsequent studies of carbohydrate metabo-
lism. From the beginning of his career, von
Mohl had been interested in the development
(differentiation) of tissue elements out of un-

\(^*\)This sounds teleological, which it is; but non-Aristotelian, naturalistic teleology is useful if used in the proper way. 'It is not consonant with the scientific mind to investigate the structure of a piece of mechanism and refrain from asking what it is for, what are its functions.'\(^\text{2, p. 71}\) how is it in harmony with the grand teleology of evolution. This process in 5 billion years has left some blind alleys, some vestiges, but by a ratio of untold thousands to one it has decorated living organisms with a mul-
titude of exquisite adaptations.

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differentiated cells. Here was perhaps his greatest work, and here Moldenhawer’s maceration technique proved to be invaluable. Von Mohl was the first to argue the secondary nature of the cell wall, precipitating an argument which persisted for a decade. Some microscopists asserted that the protozoa, which had no rigid cell wall, should not even be called cells. The true structure and development of the plant epidermis, cork, and bark were first described by him. His best known work, The Vegetable Cell (Anatomische und Physiologische Studien zur Vegetabilische Zellen), first published in 1851 and translated into English in 1852, became the standard reference work on all problems in cytology.

It was to the century 1740–1840 that Schleiden was specifically referring when he let out the burp which I have quoted earlier. Schleiden may have been justified in damning with no praise the purely systematic Linnaean school, who were persistently arbitrary in their classification of plants, and whose works he irreverently called ‘hay’—if a plant did not conform to Linnaeus’s arrangement, it had been incorrectly described, or it could not be a proper plant but must be an unnatural monster. And Schleiden was also right on many other matters pertaining to the botany of the preceding century. But that the work of de Jessieu, Camerarius, Kohlreuter, Sprengel, Stephen Hales, Joseph Black, Ingen-Housz, de Saussure, Thomas Knight, Dutrochet, Moldenhawer, and von Mohl consisted of ‘groundless prejudices,’ ‘fanciful analogy,’ and were ‘perfectly worthless’ is an unwarranted exaggeration, and certainly these men should not all be put out to graze with Linnaeus.

Despite the fact that he was so critical of the work of others, Schleiden was not too critical of his own observations. He had come to the conviction that all tissues are composed of cells, but how these were formed remained a moot question. In 1838, he described the formation of cells in the embryo sac of flowering plants as a sort of precipitation or crystallization of ‘gum’ around definite centers, which, with other observations, led him to the erroneous theory of free-cell formation. The structureless substance or fluid from which the cell crystallized he called cytolasma (cyto cell + Gr. blastema bud). In von Mohl’s words, during free-cell formation . . . the cell-membrane is developed over the surface of a mass of nitrogenous substance swimming in a fluid which contains formative matter, without the co-operation of a parent cell. In the regular course of vegetation this process of cell-formation occurs only in the interior of cells; it may occur independently of the life of the parent plant in the creation of parasitic Fungi, yeast cells, etc., both in the decomposing fluid of cells and in the excreted or expressed juices. In normal free cell-formation the secondary cells usually possess but a very small size in comparison with the parent-cell, and stand in no connection, or, at least, not a necessary one, with the walls of the latter. 18, p. 57–58 It remained for Nägeli to show that the ‘gum’ was nitrogenous and hence wrongly named from a chemical standpoint, but Nägeli compounded Schleiden’s error by proposing that new cells were formed by crystallization out of the interstitial substance which separates pre-existing cells, and which he also called cytolasma. The cell-division theory should therefore be credited to Schwann.

Schleiden (1842) had called the contents of the plant cell plant slime (Pflanzenschleim), and Nägeli referred to it as the slimy layer (Schleimschicht); but both words were more or less displaced by von Mohl’s name Primordialschlauch (1844). 9, 10 This word presents the translator with difficulties: In Adler’s German dictionary of 1861 the English equivalents of Schlauch are given as: skin, leather bag or bottle, leather pipe, water conduit, or drunkard; in Lucas’s German dictionary of 1868 the English equivalents are given as leather bag or bottle, leather pipe or hose, water conduit, wooden pipe and funnel; and, in botany, utricle or ampulla; in connection with fishing, a bag, net, or sheath; and, in the vulgate and figurative usage, it meant a fat,
corpulent person, a drunkard, or a glutton. A frivolous historian could say that Schleiden was a Schlauch.

In 1835 Dujardin (1801–1860) had designated the contents of animal cells as sarcode (Gr. sarx, sarkos, meaning flesh); in 1846, von Mohl took the great step forward in equating Pflanzenschleim with sarcode under the term protoplasm (Gr. protos first + plasma to form or mold),* taking this word from the Greek physiologist Johannes Evangelista Purkinje (1787–1869), who had first used it in 1840 to designate the granular formative substance of the animal ovum.

We have already said some good things about Schleiden, and we must add one impersonal detail: In 1837 he dined with the younger Theodor Schwann (1810–1882), German physiologist and erstwhile pupil of Johannes Müller, and talk turned to the nuclei of vegetable cells, which Brown had described in 1831. The conversation set Schwann to thinking about similar structures he had seen in the cells of the notochord, and he immediately realized the parallel significance of the two phenomena. The results soon appeared in his famous Microscopic Investigations on the Accordance in the Structure and Growth of Plants and Animals (1839), which extended Schleiden’s cell theory to animal tissues—a difficult task in view of the differences between the cells in a filament of Spirogyra and those of the notochord. As against Schleiden’s free-cell formation theory, however, Schwann adhered to the theory of cell division. He traversed the whole field of biology, proving the cellular origin of the most highly differentiated tissues—nails, feathers in animals, nerves, etc.† Thus the modern ‘cell-division’ theory crystallized in Schwann’s hands from a ‘cytoblastema’ of theoretical speculation and often confusing microscopy, until it was well established by the late 1850’s. The most difficult hazards it had to overcome were the complexities offered by pathology, but after the classic researches (1858) of Rudolf Ludwig Virchow (1821–1902), the founder of cellular pathology, it could be said, for both botany and medicine, omnis cellula e cellula—every cell comes from a pre-existing cell.* And Max Johann Sigismund Schultz (1825–1874) could assert in 1861 that the term protoplasm could appropriately be applied to the living matrix of all animals and plants, unicellular and multicellular; also in 1861, the physiologist Ernst Wilhelm Ritter von Brücke (1819–1892) emphasized that protoplasm is not an amorphous liquid but a complex structure—truly an elementary organism—the details of which are invisible only because of the limitations in our methods of observation. In 1868, Thomas Henry Huxley (1825–1895) introduced protoplasm to that stronghold of conservatism, Edinburgh, in a Sunday evening lecture entitled ‘On the Physical Basis of Life,’ and even the orthodox, failing to read between the lines, were undisturbed by his assertion, ‘If the phenomena exhibited by water are its properties, so are those presented by protoplasm, living or dead, its properties.’†

*In the Oxford English Dictionary (VII) the earliest English usage cited is plasmation (1888), meaning molding or forming. In the seventeenth, eighteenth, and early nineteenth centuries, the formative meaning is frequently encountered, plasm indicating something molded, formed, or an image, hence a mold or matrix; or plasmatical, the capacity to mold or give shape or form; and the transitive verb plasmate, to form or mold. It is unquestionable that both Purkinje the physiologist and embryologist and von Mohl the botanist and cytologist had the formative and creative meaning of plasm in mind when they applied the word protoplasm to the living contents of cells.

The physicists and their plasma of charged particles constrained by an electromagnetic field seem to be way out on an etymologic limb.

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* Nordenskold, 25. p. 196 in noting that von Mohl called the fundamental stuff within the cell protoplasm says, ‘... he thereby establishes’ that the cell content is an element by itself and not merely ‘slime’ of some independent kind, as Schleiden had supposed. The name, which in spite of its clumsiness, has come into perpetual use, is, as a matter of fact, based on a false assumption that all the component parts of the cell, even (and above all) the nucleus, originate in this element, ‘the primal slime.’ On the contrary, it seems that von Mohl, Nägeli, and others accepted that omnis nucleus e nucleo.
you know in connection with the 'Brownian' movement of microscopic particles under thermal agitation (1827), had discovered the nucleus (Zellenkern or cytoplasm of Schleiden) in 1831, Valentine the nucleolus in 1836, but these cell 'inclusions' were meaningless at the moment. You probably do not know that Brown was an eminent botanist and virtually destroyed the artificial Linnaean taxonomic system for plants by the simple expedient of ignoring it. Brown also developed what has since been called the natural system of taxonomy, the one under which Darwin worked, and in his hands anatomy and morphology of plants and their mode of reproduction fell naturally into place. After Brown's discovery (1831), the cell nucleus became an object of intensive study and has subsequently produced a whole library of memoirs and remains today the subject of much chemistry, biophysics, and electron microscopy relative to nucleoproteins, genetics, the theory of evolution, and goodness knows what else, and some of which must certainly come under discussion in this symposium.

After Schleiden and Schwann, the nucleus was recognized as an integral part of the vegetable and animal cell, and in the 'cell theory,' as formulated after 1839, it was held that 'all the tissue elements, no matter whatever their ultimate forms and functions, were derived from primary isodiametric [having equal diameters] cells and that every cell arose from a pre-existing cell and in the long run from the fertilized ovum.' In the Schleiden-Schwann theory, however, the cell wall, composed of cellulose with secondary organic and inorganic stuffs, was still required to contain the green, granular slime (Pflanzenchleim) adhering to the cellulose wall, as well as the fluid contents of the vacuole, and to control their composition.

It was von Mohl who placed microscopic botany on a firm basis. His best known work, Principles of the Anatomy and Physiology of the Vegetable Cell, was published in Germany in 1851 and translated into English by Arthur Henfrey in 1852. Von Mohl's detailed description of plant cells is a delight to read. In his Principles he says, 'If we examine the texture of plants with a powerful microscope, we find that it does not consist, as appears to the naked eye or under slight magnifying power, of a homogeneous substance perforated by a greater or less abundance of cavities, but is composed of minute portions, of definite form and organization, separable from each other (the elementary organs).

'The primary form of the elementary organ of plants is that of a completely closed, globular or elongated vesicle, composed of a solid membrane, and containing the fluid [Primordialschlauch] (utricule, utriculus). If this still remains closed after its development is completed, it is called a cell, cellula; but if a row of utricles arranged in a line become combined during the course of their development, into a tube with an uninterrupted cavity, through the absorption of their cross walls, a compound elementary organ is produced—the vessel (or spiroid of Link). . . .

'If a tissue composed of young cells be left some time in alcohol, or treated with nitric or muriatic acid, a very thin, finely granular membrane becomes detached from the inside of the wall of the cells, in the form of a closed vesicle, which becomes more or less contracted [probably in consequence of exosmosis], and consequently removes all the contents of the cell, which are enclosed in this vesicle, from the wall of the cell. Reasons hereafter to be discussed have led me to call this inner cell the primordial utricle (Promordialschlauch). Iodine colors it yellow, and it is therefore probably always nitrogenous.' Von Mohl also recognized that: 'In full-grown cells the protoplasm usually forms but a very subordinate part, as to mass, of the contents of the cell; while the watery cell-sap, which at first appeared only in isolated cavities, formed by degrees in the protoplasm, fills the whole cavity of the cell. The quantity of it is subject to variation, according to the plant has absorbed or evaporated more water; the decrease, however, cannot descend below a certain limit in the cells of most organs of the higher plants, without destroying the life of the cell.' (See fig. 1, p. 1003.)
Von Mohl knew that the vegetable cell membrane (by which he meant the cell wall, in our context) is composed of cellulose. The contemporary translation\(^8,9\) for *Schlauch* reads ‘utricle or sac,’ but these words are now archaic, and I will call von Mohl’s *Schlauch*, which encloses protoplasm and which, as Nägeli was subsequently to show, endows the protoplasm with its semi permeability, the plasma membrane in our current sense. He considered this membrane to be proteinaceous, cellulose being wholly absent. I suspect that both von Mohl and Nägeli, and perhaps others among their contemporaries, saw the plasma membrane in the flesh. Except for Brown’s nucleus and Valentine’s nucelus, the contents of the cell (protoplasm) were conceived by 1846 to consist of an opaque, viscid, proteinaceous fluid of a white color in which there were suspended numerous green granules (chloroplasts) and sometimes granules of other colors (particularly in colored leaves and flower petals).

At this point I will relate a riddle gleaned by my son from the November number of *Highlights for Children*:

Q. How does a fisherman make his net?
A. He takes a hand full of holes and sews them together.

Throughout this discourse I have been using much the same method, as will become more apparent as I discuss the work of Carl Wilhelm Nägeli (1817–1891). Nägeli held a leading position in botanical science for fifty years. His activity began at Zurich in the early days of Schleiden’s predominance, but later studies at Freiburg (1852) until his death at Munich (1898) were wholly independent. His work touched every side of biology: systematic botany, morphology, anatomy, the theory of heredity and descent, histology, and chemical and physical physiology—in fact, modern botanical physiology was practically nonexistent before him. The son of a country doctor, as a child he was devoted to books and to natural history. He began the study of medicine at the recently established University of Zurich but soon lost interest in this subject and devoted himself to botany under de Candolle. At Jena he came under the influence of Schleiden, who interested him in microscopic work. In 1844, Schleiden and Nägeli founded the *Zeitschrift für wissenschaftliche Botanik* (1844–1846), a unique journal in that it never got beyond its first volume, and all but two papers were by Nägeli. The very name, *Journal of Scientific Botany*, is characteristic of Schleiden’s attitude toward his contemporaries and predecessors and expresses the somewhat arrogant claims of the enthusiastic naturalists of the day.

Nägeli’s association with Cramer, so fruitful of good work, began in 1850, but it is said that from 1855 on his work was hindered by the temporary failure of his sight, owing to too much microscopic work. In 1857, he was summoned to the professorship of botany at Munich, where King Maximilian II was striving to render his capital as distinguished in science as it already was in art. This post he held until his death thirty years later.

If Nägeli was at first misled by Schleiden’s theory of free-cell formation, it was in part because of his intense study of the cryptogams (Algae, Fungi, mosses, liverworts, etc.). No area of botany presented more puzzles and was worse confounded by confusion than these apparently seedless, sexless lower plants that so modestly concealed their reproductive cycle—here the spontaneous generation of life out of putrefying wood and leaves was almost self-evident. Cryptogamic botany was started on its scientific course by Nägeli. But it was also from the study of these lowly plants that he was led to what is perhaps the second greatest generalization in cellular biology (the cell theory being first), namely, the existence and special properties of the plasma membrane. The course of science has an inevitability about it, and one does not see how Nägeli’s discoveries could have come about except through his intense interest in the cryptogams and their physiology.

Nägeli called his major paper on the physiology of the cell, *Primordialschlauch*. Beware of the word *Schlauch*, however, because its meaning had already shifted from the *Plas-
It was undoubtedly his experience with the cryptogams that led Nägeli to his views on permeability and the plasma membrane. He at first agreed with Schleiden's theory of free-cell formation (1838), but later he accepted, and indeed fortified, the cell-division theory. Theodor Schwann in 1839 extended Schleiden's theory to animal tissues, with this yet greater error, that new cells might arise not only within the mother cell but also from the intercellular substance (or interstitium) so common in animal tissues (to which Schwann also gave the name cytoesin). In the free-cell formation theory, it was supposed that part of the cell content separates out and consists at least of protoplasm that is capable of forming the Plasmamembran directly. Nägeli brings to the support of this argument the fact that in larger cells of the lower Algae and Fungi, whose cells die here and there, the origin of the Plasmamembran is often clearly observed. The living content, where it borders on the morbidly withering content, is covered with a Plasmamembran that in all respects behaves like the original Plasmamembran, covering itself with a (cellulose) cell wall, and that can even sprout into branches and initiate top (apical) growth. Sometimes it seems that while the entire remaining content of the cell dies, free-lying parts remain vital, and over their entire surface they form a protoplasmic layer which functions like a normal Plasmamembran. If a drop of protoplasm lies free, one can see how its smooth surface is covered with this membrane. He cites evidence that the plasma membranes of the separate granules and of the various vesicles are identical. Moreover, it is very likely that these films consist of the same substance as the plasma threads which adhere to the cell wall during exosmosis. If the Plasmamembran really differs from the protoplasm, it must be sought at the outer boundary of the protoplasmic layer.

By the mid-century, thanks mainly to the efforts of von Mohl and Nägeli, botanists had come to recognize as a general law that cells arise only by the division of a pre-existing cell, but the universal acceptance of this prin-
ciple was delayed by the greater complexity of the problem of animal cells. Its ultimate acceptance by zoologists was largely the result of the work of Albert von Kolliker (1844/45), Carl Bogislaus Reichert (1841/47), Robert Remak (1852/55); and by Rudolf Virchow's (1858) studies in pathology, cellula e cellula welded, at least in one sense, the plant and animal kingdom together. It had been only as recent as 1842 that Schleiden had flatly denied any analogy between plants and animals.

Even more than von Mohl, Nägeli was the first to set out on a fruitful road of molecular mechanism in botany and perhaps the first to see in physiology a science worthy of itself without reference to medicine. It is appropriate to quote here from Rudolf Höber's Physical Chemistry of Cells and Tissues (1945): 'The subject of this book is physiology; not "physiology from above," but "physiology from below"; not physiology originated essentially to fill human needs and help suffering individuals, but physiology as a branch of physical chemical science dealing with life as a physical, though exceedingly complex system, that may be subjected to scientific analysis like any other natural object.' Such was Nägeli's objective. He was definitely opposed to any distinction between animals and plants.

If we may judge by the sequence of discussion in Nägeli's paper, Primordialschlauch, of 1855, the most important one which he published on the general properties of the cell and in which he focuses attention on the plasma membrane, he began with the gross properties of protoplasm and only secondarily considered the question of permeability, though permeability (or impermeability) to natural pigments was of course a question raised with every pigmented cell at which he looked.

Nägeli early recognized that in normal plant cells the cellulose wall is lined with a thin, membranous layer of protoplasm, which he sometimes called Plasmaschleim and sometimes Plasm for short, though he more frequently refers to it as the mucous layer, all terms being equivalent, in the end, to von Mohl's Primordialschlauch. We will call it protoplasm instead of sac, the latter term being most frequently used by Nägeli. The protoplasm forms a completely closed sac (Blase) which surrounds the entire contents of the cell (meaning the central vacuole), except for the nucleus, which Nägeli thought lay naked in the central vacuole (see fig. 1). This protoplasmic layer is, however, extremely thin in many simple fresh-water plant cells and very difficult to see, the greater part of the cell being occupied by the central vacuole.

The cellulose wall (cell membrane) is visible separately from the plasma membrane only when the latter is detached naturally or

![Diagram](image_url)
artificially from the cell wall. This occurs in consequence of drying out, in consequence of artificially produced exosmosis* or endosmosis, by mechanical injury of the cell, or in the natural course of certain types of reproductive processes. The cell wall has considerable rigidity, whereas the plasma membrane is soft and apparently elastic. During drying, air may penetrate the cell wall and thus come to surround the plasma membrane, as in certain spores and pollen.

*The observation that the protoplasmic contents of plant cells contract in strong sugar or salt solutions (exosmosis) was made by Nathanael Pringsheim (1823–1894) in 1854 and by Nägeli in 1855. Dutrochet** had coined the words endosmosis and exosmosis in 1827; the terms plasmolysis and deplasmolysis remained to be introduced by Hugo de Vries several years later. Since the plasmolytic method (as de Vries called it) was used in many studies of this period, it should be noted that it was not until 1861 that Thomas Graham distinguished between crystals and colloids, and not until de Vries's²⁷ classical studies on isotonic solutions and van't Hoff's theory²⁸ of osmotic pressure that the plasmolytic method became more than a rough tool.

Chambers²⁶, p. 35² says, 'Just as Virchow had announced the dictum omnis cellula e cellula et Fleming omnis nucleus e nucleo, so de Vries claimed that all plasma membranes originate from a previous membrane.' The writer has not encountered de Vries's statement, but it can be true only in the sense that any bit of protoplasm that does not cytolize forms its own plasma membrane. There is no evidence of genetic continuity in the case of the nucleus.

The question of penetration into, or escape from, a cell appeared simpler in Nägeli's day than it does now, because he was unaware of the fine distinctions between diffusion, facilitated diffusion, active transport, etc. Diffusion may be defined as a motion of solvent and solute relative to one another throughout a definite region of space in which mixing is taking place. However, diffusion should not be applied to the circumstances where the solute itself is going into solution in a solvent, as, for example, at the surface of a crystal; this process can, to my knowledge, be called only solution; diffusion begins only after the salt which has dissolved in the liquid is transported from the surface of the crystal throughout the solvent by diffusion.² Gross passage of a solute through the plasma membrane cannot safely be referred to as diffusion or solution, and perhaps we had best refer to it ambiguously as penetration.

During exosmosis, which Nägeli usually produced with sugar solutions, all the cellular contents contract; during mild exosmosis, the cell wall is left intact, though it may be collapsed by strong exosmosis (see, in fig. 2, figs. 4 and 13 of Nägeli's plate 2, opposite p. 1008). If the tissue is torn or cut, the protoplasm is usually torn along the cell wall, but occasionally it falls out undamaged, as shown in plate 2, figure 15 of figure 2.

During extreme exosmosis, the sac contracts so much that it virtually surrounds only the firm parts of the protoplasmic contents; that is, the vacuole may wholly disappear (see, in fig. 2, plate 2, fig. 10). This contraction is accompanied by only the slightest wrinkling of the plasma membrane. The contraction caused by exosmosis is reversible when the cell is returned to water, though the degree of reversibility varies with different types of cells. During contraction small punctiform spots on the membrane stick to the cell wall and are pulled into long threads (see, in fig. 2, plate 2, figs. 3–6), showing that the membrane is not uniformly attached to the cell wall in all cells nor in all parts of the same cell. Alcohol and acids cause the plasma membrane to lose its distensibility, thicken it, and reduce its surface. From such observations, Nägeli concluded that the plasma membrane behaves like a viscous, semiliquid mucus and is only attached at a few points, or not at all, to the cell wall and that it can expand considerably but has no true elasticity. Injurious stimuli (chemical or mechanical) produce loss of distensibility, thicken the membrane, and reduce its surface.

During cell division, the cellulose wall protrudes into the lumen at the same time that the protoplasmic contents divide. Nägeli recognizes that the phenomena are interrelated, but he is cautious as to which is cause and which is effect. However, at another time (and referring to a different plant species) he states that division of the protoplasm appears to precede division of the cell wall at a circular spot (or ring?); the protoplasm extrudes cellulose, and a ring-shaped disc is formed, which projects deeper and deeper.
until the two now separate parts of the cell are isolated only by a perforated (cellulose) disc (fig. 2, plate 4, figs. 3–11).

Referring to the Primordialschlauch, he says, 'I have not touched upon the question of whether it is a membrane or not. It seems to me idle to talk about it as long as we have no scientific definition of a membrane. Until now, I believe, we can define the membrane only negatively, and even so very inadequately by stating what a membrane definitely is not.' Against von Mohl, who regarded the Schlauch as a membrane of rather solid composition, Nägeli is inclined to the view that it is a mucous layer (Schleimschicht), not differing substantially from the plasma threads and plasma membranes. But he says, 'I would not like to call a layer of fluid which surrounds the drop of another fluid a membrane.'

With respect to permeability, he notes that in some plants pigment is present in solution (or as crystals) in the vacuole or in the protoplasm, or both. During exosmosis only a clear fluid is withdrawn from the cell (vacuole and protoplasm), the pigment therein becoming proportionately concentrated, even to the point of forming new crystals. Such is the case in the vacuole in Viola tricolor, Orchis mascula, and the epidermal cells of Perigonium. Yet after injury of the cell by dilute hydrochloric acid, these pigments escape rapidly from the vacuole to the protoplasm and thence to the exterior through the cell wall. If now the cell is subjected to exosmosis, the pigment can be seen in the space between the protoplasm and the cell wall, and the nucleus becomes almost colorless. Nägeli concludes that the protoplasmic membrane surrounding the vacuole is normally impermeable to the pigments contained in the vacuole.

Conversely, if normal cells are placed in solutions colored by cherry juice or other pigments, the cells do not become colored; but where exosmosis has contracted the protoplasm, the pigment can be seen in the space between the protoplasm and the cell wall. Finessing the fine point as to whether sugar solution (exosmosis) changes the permeable nature of the cell wall (these are the reviewer's words), this wall is permeable to all the pigments studied. It is also a matter of splitting hairs to inquire concerning the molecular weight of the pigments observed in Nägeli's various observations; Nägeli's experiments are too numerous and varied to challenge, and one presumes that the pigments which he observed were crystalloids with a fairly low molecular weight (anthocyanine, erythrophyll, etc.). Even during exosmosis, pigments do not enter the protoplasm, the vacuole, or the nucleus. The last would have presented no problem had Nägeli recognized that the nucleus lies within the cellular protoplasm and that the protoplasm presents a plasma membrane to ensnare the vacuole. If one puts a colorless cell into a sugar solution tinted red by the pigment of a fruit, such as cherry juice, the pigment penetrates the cell wall but the contents of the contracting protoplasm remain uncolored. Only after prolonged exposure does the pigment penetrate and color the protoplasm.

In the epidermal cells of the upper petal surface of Viola tricolor, excess anthocyanine is secreted in granules and the vacuole is reddish violet, more rarely bluish violet, though a few cells are colorless or pale. In the most intensely colored cells, two or more rarely three or four granules 4 to 7 micromillimeters in diameter, almost spherical in shape, of a violet or nearly black color, are observed. Nägeli notes that the smaller granules 0.5 to 1.4 micromillimeters are in molecular motion. (He makes no reference to Brownian movement or to Brown's paper of 1827.) Therefore he concluded that the protoplasmic surface (plasma membrane), whether facing the cell wall or the interior, is a barrier to penetration.

* * *

If, under the microscope, one stimulates one edge of a piece of Perigonium epidermis, Viola, or Orchis by applying dilute hydrochloric acid, one can observe the changes in permeability cell by cell as the acid reaches them, the pigment diffusing out and leaving the cell colorless. The nucleus, the vacuoles, and chlo-

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roplasts behave toward pigments as does the protoplasm. Sometimes two fluids are secreted in the cell, one colored and one colorless, separated only by a delicate plasma membrane. And two cells may lie side by side, one colored, the other colorless, without the pigment passing into the latter. Sometimes chlorophyll is dispersed in solution in a cell (fresh water Algae, etc.) without escaping either into the vacuole or to the exterior, unless the cell is injured. In such chlorophyll-stained cells, the plasma membrane remains colorless (fig. 2, plate 2, figs. 3 and 4).

His experiments demonstrate that the protoplasmic membrane surrounding the vacuole is normally impermeable to pigments from either side, but that, after injury of this membrane, the pigments can readily enter the vacuole or leave it.

Nägeli’s paper is long and repetitious and deals with several topics: I wonder what would have been the consequence, in our present era of short papers, if he had submitted his manuscript to one of our botanical journals and the editor had requested him to cut it in half and to write a concise summary.

* * *

I cannot cut the paper in half, but I can attempt a summary of sorts: Working with Algae, Fungi, mosses, and unicellular plants, Nägeli discovered that when protoplasm is torn the wound heals itself. A new protoplasmic surface, which behaves in all ways like the original, forms quickly around bits of protoplasm which have been extruded mechanically from the cell. The protoplasmic surface is dense, more viscous, and otherwise distinguishable from the protoplasm as a whole, and hence he called the surface layer the *Plasmamembran*. This membrane is impermeable to several naturally occurring plant pigments, which therefore can not enter the cell from the outside nor escape from the vacuole. From experiments with exosmosis and endosmosis, he concluded that the plasma membrane endows the cell with its osmotic properties.

After severe injury of the plasma mem-

brane, protoplasm can not form a new membrane, and the cell becomes permeable to pigments and no longer behaves as an osmotic system.

Though the greater part of Nägeli’s most important paper concerns the plasma membrane, no mention of it has been seen by the writer in any of Nägeli’s obituaries. But, throughout the next century, the plasma membrane hovers like a ghost which makes a favored visit here and there: De Vries and Klebs used the idea in osmotic studies, Pfeffer refers to it in setting up his ferrocyanide pot, and Overton used it in his theory of narcosis. Yet the plasma membrane is not in the index of William Bayliss’s *General Physiology*, Nordenskjöld’s *History of Biology*, Garrison’s *History of Medicine*, or Wilson’s *The Cell in Development and Inheritance*, or in Garrison and Morton’s *Medical Bibliography*, nor is it discussed by Harvey-Gibson in his *Outlines of the History of Botany* or included in the index of the last edition (1958) of the *Encyclopaedia Britannica*. As far as I can find out, the plasma membrane has never made an index, which is a little surprising since van’t Hoff relied so heavily on de Vries’s data, obtained by the plasmolytic method, in developing his theory of osmotic pressure, and since the functions attributed to the plasma membrane in the cell are fundamental to the entire cell theory. Yet the plasma membrane must be as old as the first bit of true protoplasm, where, as now, it served to create a favorable intracellular environment for the primeval protoplasmic contents, permitting some things to enter while keeping other things out.

It has not been wholly neglected, however: In the *Physical Chemistry of Cells and Tissues*, edited by Rudolph Höber and published in 1945, Höber himself devotes 56 pages to the section entitled, ‘The Surface of the Protoplasm, Its Properties and Its Architecture,’ and twelve pages are devoted to the ‘Chemistry and Physics of the Plasma Membrane.’ Twelve pages comprised about all that there was to say in 1945, but now look at the program today with nearly 1,100 per-

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SYMPOSIUM ON THE PLASMA MEMBRANE

persons in attendance, over 50 per cent of them from outside the Greater New York area, and seventeen speakers requiring four chairmen to hold them in line.*

In the introduction to his section on the plasma membrane Höber says, 'There is no problem in cell physiology more challenging to investigators than the conditions allowing dissolved substances to enter the protoplasm or to leave it. This problem has occupied the general interest of physiologists since Nägeli's discovery (1855) of the plasmosis and deplasmosylation of plant cells afforded a number of botanists, Pfeffer, de Vries, G. Klebs, and others, an easy way to measure the osmotic forces of the living cell and the permeability of the protoplasmic surfaces to solutes.' But Höber believed that there was a 'plasma membrane,' and thus he was in that minority who were viewed askance by dog, cat, rabbit, and white-rat physiologists as being a little queer.

On the whole, a century elapsed during which the terms 'membrane,' 'cell membrane,' and 'plasma membrane' appear in many papers, settling here and there in subordinate clauses, less rarely appearing as the grammatical subject of a sentence, before the ghost acquired bones and flesh and became a matter of proper scientific interest.

The plasma membrane was first given substantive form, in my opinion, by the exquisitely delicate microdissection method of Robert Chambers (1881–1957). I have elsewhere briefly reviewed the history of the microdissection method and some of the uses to which Chambers and his colleagues put it in relation to renal physiology, and the subject in its more general aspects has recently been reviewed in greater detail by Chambers and Chambers.**

By 1922, Chambers had developed the microdissection method to the point where he was able to impale a naked starfish egg with one needle and hold it steadily while he introduced a second needle for injection of solutions, thus avoiding the barrier presented by the plasma membrane. He found that dilute hydrochloric acid has no ill effects on the egg when the acid is confined to the environment, but, if the acid is injected, the egg quickly cytolizes. He also showed that eosin will not penetrate an ameba but, if the eosin is injected, the dye quickly permeates the entire protoplasm, to be arrested only at the surface. He concluded from his experiments that it is immaterial whether the semipermeable membrane be that of the original cortex, a film newly formed over a cut surface, or a film that surrounds an artificially produced vacuole within the cell; as long as the surface film exists neither the acid group of ammonium chloride nor the alkaline group of sodium bicarbonate can, within certain concentration limits, penetrate into the cell. However, these solutions, if injected into the cell, immediately cause cytolysis, p. 41

These early microinjection experiments were followed by a long series of similar observations in the hands of Chambers and his colleagues and in the hands of others, using a variety of cells, which served only to confirm the initial observations and which afforded definitive visual demonstration of the semipermeable properties of the surface of discontinuity of protoplasm as Nägeli had proposed (see fig. 3).

A striking demonstration which affords a suggestion regarding the nature of the plasma membrane is given by Chambers and Raznikoff, who record the following experiment on an actively flowing ameba, the plasma membrane of which is almost naked: 'On being dragged out of the water, the plasma membrane will be caught by the surface of the water and will be pulled entirely off the ameba. The ameba, which has in this way literally been flayed alive, may recover if it is immediately pushed away from the surface into the depth of the water.' p. 47

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**I am glad, as I said to Dr. Schmitt, that I am opening rather than closing this symposium. When Sir Walter Raleigh wrote his History of the World, he apologized for not writing the history of his own time—which would,' he says, 'have been more pleasing to the reader'—on the ground that 'whosoever in writing a modern history shall follow truth too near the heels, it may haply strike out his teeth.'
Plate II. Figures 1–8: The reaction of the primordial sac to anthocyan. The epidermic cells of figures 1–2, 3–6, and 7–8 are from different flower leaves. Figures 9–14: The behavior of the cells of Spirogyra under the effect of glucose solution. Figure 15: Transverse section of Porphyra vulgaris Ag. The primordial sacs have fallen out of a few cells unharmed.

Plate IV. Figures 1–15: Cell division in Cladophora. In figure 16, the upper cell half has died and disappeared; the lower, viable half of the content is clothed in a primordial sac and a membrane at the vacated plane. Figure 17 shows a viable upper cell and an injured lower and side cell. Figures 18–21: Formation of the special mother cells of pollen in Althaea rosea Car. Figures 22–24: Other examples of the behavior of the primordial sac.

Diagram to show three components of cell cortex of a sea-urchin egg. 1. Extraneous investments: (a) jelly; (b) fertilization membrane; (c) 'hyaline plasma layer or membrane.' (a and c can be removed chemically; b can be removed mechanically.) 2. Protoplasmic surface film, a fluid layer not coagulated by calcium. 3. Plasma-gel layer, underlying the protoplasmic surface film and consisting of a reversibly gelating portion of the granular cytoplasm.

Underneath the extraneous coats and distinct from them is a delicate film which invests the protoplasmic body and on which the integrity of the protoplasm depends. In the diagram this film is designated as the protoplasmic surface film.

Chambers suggests that the term protoplasmic surface film, or simply surface film, is preferable to plasma membrane, since the latter is generally used to designate the physiologically active part which may or may not be a molecular layer lying somewhere in or on the surface layer of protoplasm. All that is claimed in his paper for the protoplasmic surface film is to designate a visibly differentiated surface layer which protects the underlying cytoplasm from disintegration. However, for the purposes of this symposium, Nageli's term plasma membrane is better because it is more nearly definitive. (From Am. Naturalist 72: 146, 1938.)
And yet, as Nägeli saw, the plasma membrane will heal itself if torn and forms de novo around a bit of extruded protoplasm.

Figure 3 is a diagram of the cortex of the echinoderm egg, published by Chambers in 1838. In this paper he emphasizes the presence of the nucleus as influential in the repair of injured protoplasmic surfaces. If an amoeba is torn with microneedles the exposed cytoplasm starts to flow out and undergo degeneration. If the nucleus is at a distance from the tear, repair takes place by the formation of a film-like membrane running in a more or less straight line across the gap and excluding the outflowing, disintegrating cytoplasm. However, if the nucleus is included in the outflowing cytoplasm, the repairing surface, evidently influenced by the presence of the nucleus, tends to run in a sweeping curve around the cytoplasm so as to enclose the nucleus.

In short, Chambers could hook a microdissection needle under the surface (at least of a starfish egg) and, by giving it a microscopic yank, lift the plasma membrane off the underlying protoplasm. He really had hold of it.

At the end of his Outlines of the History of Botany, Harvey-Gibson says, 'The history of science is an aid in scientific research. It places the student in the current of scientific thought, and gives him a clue to the purpose and the necessity of the theories he is required to master. It presents science as a constant pursuit of truth rather than the formulation of truth long since revealed; it shows science as progressive rather than fixed; dynamic rather than static, a growth to which each may contribute. It does not paralyze the self-activity of youth by the record of an infallible past.'

At the end of this outline, the writer would suggest that if you put synthetic chromosomes inside a synthetic plasma membrane, you would soon have a synthetic cell.

The history of science can be a lot of hard work, but nevertheless it is fun. And you see now why the Riddle of the Fisherman is no joke.

References


The Plasma Membrane, with Notes on the History of Botany
HOMER W. SMITH

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