

there is no requirement for an ad hoc initial chemical layering of the above type. High temperatures move the stability field of plagioclase deep into the interior of the moon and this serves to decrease the mean density. A small core composed of the high-pressure phase assemblage can be tolerated without violating the mean density or moment of inertia. This removal of a constraint on the internal composition of the moon has been discussed by Anderson (39).

References and Notes

1. R. B. Baldwin, *Science* **170**, 1297 (1970).
2. M. G. Langseth, S. P. Clark, J. L. Chute, S. J. Keihm, A. E. Wechsler, *Moon* **4**, 390 (1972).
3. T. Hanks and D. L. Anderson, *Phys. Earth Planet. Interiors* **5**, 409 (1972).
4. P. W. Gast, *Moon* **5**, 121 (1972).
5. H. Urey and G. J. F. MacDonald, in *Physics and Astronomy of the Moon*, Z. Kopal, Ed. (Academic Press, New York, ed. 2, 1971), p. 213.
6. W. M. Kaula, *J. Geophys. Res.* **74**, 4807 (1969); D. L. Anderson and R. L. Kovach, in *Proceedings of the Caltech-JPL Lunar and Planetary Conference, 13-18 September 1965* (Jet Propulsion Laboratory, Pasadena, Calif., 1966), p. 84.
7. W. M. Kaula, *Phys. Earth Planet. Interiors* **2**, 123 (1969).
8. ———, in *Trajectories of Artificial Celestial Bodies*, J. Kovalevsky, Ed. (Springer-Verlag, Berlin, 1966), p. 247.
9. J. Lorell, G. Born, E. Christensen, J. Jordan, P. Laint, W. Martin, W. Sjogren, I. Shapiro, R. Reasenberg, G. Slater, *Science* **175**, 317 (1972).
10. M. Caputo, *J. Geophys. Res.* **70**, 3993 (1965).
11. J. E. Conel and G. B. Holstrom, *Science* **161**, 680 (1968).
12. W. L. Sjogren, P. M. Muller, W. R. Wollenhaupt, *Moon* **4**, 411 (1972).
13. G. Hulme, *Nature* **238**, 448 (1972).
14. R. B. Baldwin, *Icarus* **8**, 401 (1968).
15. A useful tabulation of laboratory measurements of rock strengths appears in the article by J. Handin in "Handbook of Physical Constants" [S. P. Clark, Jr., Ed. *Geol. Soc. Amer. Mem.* **97** (1966), pp. 223-289]. A few representative values are given here.

Material	T (°C)	Confining pressure (kbar)	Ultimate strength (kbar)
Anorthosite	150	1.0	5.9
	500	5.1	9.4
Basalt	300	5.0	13.8
	800	5.1	2.6
Granite	150	1.0	3.3
	500	5.1	8.3

16. F. Birch has estimated the strength of the earth's mantle to be of the order of 100 bars by using data from geodesy, gravity, and geology. Under some high mountains it may reach several kilobars. Caputo (17) estimated the minimum strength required to maintain the global departures from hydrostatic equilibrium to be 20 to 70 bars. Kaula (18) has also analyzed the gravity field of the earth and derived maximum stress differences of 97 and 300 bars for the lower mantle and the crust, respectively. See F. Birch, in *State of Stress in the Earth's Crust*, W. R. Judd, Ed. (Elsevier, New York, 1964), pp. 55-80; H. Jeffreys, *The Earth* (Cambridge Univ. Press, London, ed. 4, 1959), p. 420.
17. M. Caputo, *J. Geophys. Res.* **70**, 955 (1965).
18. W. M. Kaula, *ibid.* **68**, 4967 (1963).
19. Y. Shimazu, *Icarus* **11**, 455 (1966).
20. J. Weertman, *Rev. Geophys. Space Phys.* **8**, 145 (1970); R. I. Walcott, *J. Geophys. Res.* **75**, 3941 (1970).
21. C. B. Raleigh and S. H. Kirby, in *Mineral. Soc. Amer. Spec. Pap.* **3**, B. A. Morgan, Ed. (1970), pp. 113-124.
22. This estimate for stress differences now being supported in the lunar crust is close to the yield strength calculated by Baldwin (14). From an analysis of variations in lunar crater dimensions as a function of diameter, he calculated that lunar rocks would behave elastically below stresses of 30 to 50 bars and distort viscously at higher stresses. His analysis covered crater diameters from fractions of a kilometer to 100 km. Therefore, the inferred strength of the crust is relatively constant for loads varying by four orders of magnitude in effective wavelength. This suggests that loads have decayed to this value and that the present implied stress differences are relatively permanent features of the moon. The estimates of viscosity in this article and elsewhere, where zero strength is assumed for long-term processes, are therefore upper bounds. If the moon's crust has a finite permanent strength, then the viscosity of the crust can be arbitrarily low.
23. C. P. Sonett, D. S. Colburn, P. Dyal, C. W. Parkin, B. F. Smith, G. Schubert, K. Schwartz, *Nature* **230**, 359 (1971).
24. P. Dyal and C. W. Parkin, *J. Geophys. Res.* **76**, 5947 (1971).

25. The dependence of electrical conductivity on sample quality, sample size, oxidation state, and impurity level are well known. A conductivity of 10^{-4} (ohm-cm) $^{-1}$, the value found by Sonett *et al.* (23) at a lunar radius of 1500 km, yields temperatures from 920° to 1790°K, even if we restrict attention to olivine single crystals (26). The electrical conductivity of lunar basalt is two or three orders of magnitude greater than that of typical terrestrial olivine (24), although it is unlikely that the intrinsic bulk conductivity was being measured. The range of temperature inferred at a radius of 1500 km, under the unlikely assumption that the material at this depth is basalt, is 700° to 900°K. These low temperatures are typical of those inferred in (23) and (24).
26. F. C. Schwerer, G. P. Huffman, R. M. Fuher, T. Nagata, *Moon* **4**, 187 (1972).
27. A. W. England, G. Simmons, D. Strangway, *J. Geophys. Res.* **73**, 3219 (1968).
28. A. Duba, H. C. Heard, R. N. Schock, *Earth Planet. Sci. Lett.*, in press; A. Duba, *J. Geophys. Res.* **77**, 2483 (1972).
29. D. L. Anderson, *Nature* **239**, 263 (1972).
30. G. Latham, M. Ewing, J. Dorman, D. Lammlein, F. Press, N. Toksoz, G. Sutton, F. Duennebie, Y. Nakamura, *Moon* **4**, 373 (1972).
31. Lunar Science Analysis Planning Team, *Science* **176**, 975 (1972).
32. L. Thomsen, *Nature*, in press.
33. D. L. Anderson, *Sci. Amer.* **207**, 52 (July 1962).
34. D. A. Papanastassiou and G. J. Wasserburg, *Earth Planet. Sci. Lett.* **11**, 37 (1971).
35. Europium is enriched in the moon relative to carbonaceous chondrites differently than the other rare earth elements.
36. N. J. Hubbard, P. W. Gast, C. Meyer, L. E. Nyquist, C. Shik, H. Wiesmann, *Earth Planet. Sci. Lett.* **13**, 71 (1971); N. J. Hubbard and P. W. Gast, *Geochim. Cosmochim. Acta* **2** (Suppl. 2), 999 (1971).
37. D. L. Anderson and R. L. Kovach, *Phys. Earth Planet. Interiors*, in press.
38. G. W. Wetherill, *Science* **160**, 1256 (1968); A. E. Ringwood and E. Essene, in *Proceedings of the Apollo 11 Lunar Science Conference*, A. A. Levinson, Ed. (Pergamon, New York, 1970), p. 769.
39. D. L. Anderson, *J. Geophys. Res.*, in press.
40. T. J. Shankland, in *The Application of Modern Physics to the Earth and Planetary Interiors*, S. K. Runcorn, Ed. (Wiley-Interscience, New York, 1969), pp. 175-211; R. M. Hamilton, *J. Geophys. Res.* **70**, 5679 (1965); S. P. Mitoff, *J. Chem. Phys.* **31**, 1261 (1959). See also (24-28).
41. Supported by NASA grant NGL 05-002-069. Contribution No. 2188, Division of Geological and Planetary Sciences, California Institute of Technology.

Louis Pasteur Sesquicentennial (1822-1972)

J. R. Porter

Born on 27 December 1822, Louis Pasteur spent his early life in Dôle and Arbois, France. The most unique feature of this period was his rigorous schooling, with his father—a tanner by trade—checking his lessons every evening. Between the ages of 13 and 18, Pasteur demonstrated great artistic

ability, and he received a certificate in arts in 1840 at Besançon. His drawings were precise, and his attractive pastels and lithographs were so well done that art critics even today say he could have made a great reputation for himself in the arts (Fig. 1). Years later, from 1863 to 1867, he did serve

at the École des Beaux Arts in Paris as professor of geology, physics, and chemistry in relation to the arts.

At the age of 21, Pasteur left his native province and went to Paris, where he came in contact at the Sorbonne with one of the most distinguished scientists of the time, Jean-Baptiste Dumas, who encouraged him to study chemistry at the École Normale Supérieure from 1843 to 1846. From this time until 1885, he made great and ingenious discoveries in at least nine different areas of science, and he influenced scientific thought and investigation in many fields. His major fields of research are carved in

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marble on the walls of the little chapel where he is buried at the Institut Pasteur in Paris: 1848, dissymétrie moléculaire; 1857, fermentations; 1862, générations dites spontanées; 1863, études sur le vin; 1865, maladies des vers à soie; 1871, études sur la bière; 1877, maladies virulentes; 1880, virus-vaccins; 1885, prophylaxie de la rage.

Molecular Asymmetry

Pasteur's scientific career probably began in 1844, when as a student at École Normale he read and questioned a published note on the rotatory properties of tartaric acid. Soon afterward, in 1847, he successfully defended his physics and chemistry theses before the faculty of sciences at the University of Paris (1) and continued his investigations in crystallography.

Although he reported to the Académie des Sciences (1) his great discovery on the separation of dextrotartaric and levotartaric acids in 1848, it required nearly 10 years to establish definitely the molecular asymmetry of tartaric acid. In describing a biological means of separating the asymmetric forms of the compound he wrote (2, p. 21):

I have discovered a method of fermenting tartaric acid which works readily with dextro-tartaric acid and poorly or not at all with levo-tartaric acid. It is a curious fact, but one which could have been predicted, that if racemic acid is subjected to the same mode of fermentation it is split into a dextro-acid which ferments and into a levo-acid that remains intact.

Molecular asymmetry continued to interest Pasteur for many years. He thought about ways to transform nature and how to develop techniques for producing asymmetrical substances by artificial means. In 1874, for example, he wrote (1, p. 361): "Do such asymmetrical qualities, caused perhaps by cosmic influences, exist in light, electricity, magnetism, and heat? Do these qualities relate to the movement of the earth, to electric currents by which physicists explain the existence of the magnetic and terrestrial poles?"

Based on his work in crystallography, Pasteur was nominated in 1856 to the Academy of Sciences, but when the votes were counted in 1857 he lost. Later, however, after his outstanding work on fermentations, his discovery of anaerobiosis, and his

disproof of spontaneous generation, he was elected to the section of mineralogy of the academy in 1862.

Fermentations

When Pasteur was appointed dean and professor in the new faculty of sciences at Lille in 1854, he became interested in the variable amounts of the two asymmetric forms of amyl alcohol that developed as trace end products during the alcoholic fermentation of various substrates. This was a logical development of his thinking in earlier work on the crystallography of tartrate. But at the time, no one realized that such phenomena as lactic, alcoholic, tartaric, butyric, and acetic acid fermentations may be caused by different organisms, some of which grow without free oxygen. Furthermore, there were no explanations for the source of the microbes, how they grow and multiply, what substrates they attack, and how different chemical end products arise in fermentations.

In 1857, Pasteur reported his discovery that living bacteria cause the lactic fermentation (3). This outstanding discovery opened an entirely new field of investigation, for it explained one of science's most puzzling mysteries; it eventually led to an improvement in the health and welfare of mankind.

Pasteur left Lille in 1857 to become director of scientific studies at École Normale in Paris. There he continued his experiments on how yeasts cause alcoholic fermentation and on how other fermentations are produced by different microorganisms. Indeed, being in Paris, he had a better opportunity to enter into active debates with such leading adversaries of his theories as Claude Bernard, Jöns Jacob Baron Berzelius, Félix Pouchet, and Justus Liebig, who believed mainly in abiological theories of fermentation.

The controversies that developed over various fermentations and anaerobiosis were among the more intense and passionate in scientific history, and it took Pasteur several years to convince his opponents of the validity of his conclusions. In 1861, he wrote to the Academy of Sciences (2, pp. 142 and 146):

In the various communications that I have had the honor of addressing to the Academy on the subject of fermentations . . . all my efforts were directed toward

demonstrating that fermentations were correlative to the presence and proliferation of living organisms, with a different organism corresponding to each type of fermentation. . . . The purpose of my inquiries had been to study the products of fermentations more closely than had been done before, to isolate the ferments, and to discover experimental proofs of their structure and mode of life. . . .

To sum up, besides all previously known organisms, which without exception can breathe and feed only by absorbing free oxygen, there exists another class of organisms whose respiration is sufficiently active that it can live in an airless environment by extracting oxygen from certain compounds, with the result that the latter are slowly and progressively decomposed.

This second category of organisms [anaerobes] consists of the ferments, similar in every way to the members of the first category—assimilating carbon, nitrogen, and phosphate, and requiring oxygen—but differing from them by possessing the ability of utilizing oxygen extracted from relatively stable compounds, in the absence of free oxygen.

Although the confusion and controversy over the mechanisms of fermentation did not end for several years after 1861, Pasteur's experiments and explanations were sufficiently convincing to establish the biological theory of fermentation and to discard the purely mechanical or chemical theories. For this work he was awarded the Prix Jecker by the Academy of Sciences. Finally, when Edüard Buchner demonstrated in 1897 that the ferments of living cells were enzymes, capable of catalyzing chemical reactions in cell-free preparations, the true explanation of Pasteur's theory became apparent.

Spontaneous Generation

How and why microscopic forms of life appear in rainwater or infusions or organic matter when exposed to air were being hotly debated by many learned persons in the middle of the last century. In 1858 and 1859, Pouchet, director of the Natural History Museum at Rouen, presented results on "Vegetable and animal proto-organisms generated spontaneously in artificial air and in oxygen" (2, p. 222). He stated that he conducted his experiments with extreme caution, and he claimed that his results were final proof that microbes could originate anew from inanimate solutions. Pasteur was prepared by his studies in fermentation to subject this claim to rigorous experimentation. He was convinced

that an answer to the problem was essential, despite Biot, Dumas, and other colleagues who tried to discourage him from working on this difficult and apparently unsolvable problem.

Pasteur's innovative work on this topic—his strategy of attack and his design and performance of certain key experiments—is fascinating. He acknowledged that somewhere in our universe life may arise spontaneously, but he raised the question: What can be done to evaluate the claims of those who pretend to have seen life emerge anew or to have brought about conditions under which its spontaneous generation is possible?

He began by designing an aspirator that would concentrate on a cotton filter the microscopic particles in atmospheric air (4). With this he demonstrated that microbes of various kinds exist in the atmosphere—but not everywhere and not in the manner his opponents claimed. He then conducted experiments with various liquids and found that he could induce the appearance of organisms in flasks of sterile liquids by introducing atmospheric air. In order to refute the argument that filtering or heating the air was adverse to the development of microbes, Pasteur made balloon flasks with open, but drawn-out, wavy, or curved, necks. Thus the microbes entering with the air settled in the bends and did not pass into the liquid in the flask; the liquid remained clear and sterile. But if the curved necks of such flasks were cut off and the liquid exposed directly to the atmosphere, microbes soon entered, grew in the solution, and turned it cloudy and putrid (4).

As a result of this research, Pasteur was awarded the Alhumbert Prize in 1862 by the Academy of Sciences. John Tyndall (5) wrote: "Clearness, strength, and caution with consummate experimental skill for their minister were rarely more strikingly displayed than in this imperishable work." "Spontaneous generation?" wrote Pasteur in 1878, "I have been looking for it for 20 years but I have not yet found it, although I do not think it is an impossibility" (6, p. 30).

Studies on Wine and Vinegar

Although it was well known by 1860 that wine and cider turn to vinegar when they come in contact with air and that aeration hastened the

process, no one knew what caused the change. Liebig, Berzelius, and others were convinced that the transformation was not produced by microorganisms. Pasteur visited many vinegar factories in Orleans to check on contradictory statements. He examined with a microscope the surface film, the wines undergoing change, and the sediment that formed, which he thought might be responsible for the change. He soon found that the taking up of oxygen from air and the acetification process were caused by the ferment *Mycoderma aceti* (*Acetobacter aceti*) rather than by the nitrogenous constituents of the wine, as most persons believed (7). He demonstrated that wooden racks or beechwood shavings in fermenters acted as physical supports for the ferment rather than only as carriers of the ferment. With this knowledge he developed a practical process

for vinegar-making that could be standardized and controlled (8).

By 1863, the wines of France were in great demand in markets throughout the world, but many kinds would not survive transportation or long storage and others were subject to a number of hazards such as turning sour (acetification). These problems were causing great economic losses to France, and Emperor Napoleon III asked Pasteur for help.

Pasteur responded to the request by studying the various "diseases of wine" and the role of oxygen in wine-making. He reported (8) that oxygen in the air causes some wine to spoil and that the most effective means of preserving wine was Nicholas Appert's method of applying heat (50° to 60°C) for a few minutes to prevent oxidation. Unlike wine connoisseurs, who claimed that wines so "mummified" could not mel-



Fig. 1. Four pastel portraits done by Pasteur between the ages of 16 and 17.

low with age, Pasteur was convinced that most delicate wines would be improved by heating. He said, "The aging of wines is due, not to fermentation, but to a slow oxidation which is favored by heat" (8, p. 409).

Pasteur designed special equipment for heating large quantities of wine and for preserving it in tightly stoppered bottles. These practical methods were of great value to the wine industry in France and throughout the world. The common technique of pasteurization developed from these studies.

Diseases of Silkworms

By the 19th century, silk production was a major French industry, as well as a great source of wealth. Beginning in about 1845, however, diseases of the silkworm began to spread through nurseries in southern France, reaching such epidemic proportions in 1865 that estimated economic losses to the country were over 100 million francs. Since similar epidemics were occurring at the time in most other silk-producing countries, this was a problem of major proportions.

At the request of his old friend Dumas and of the minister of agriculture, Pasteur agreed to investigate pébrine (9), a disease of the silkworm. But he admitted that he had never seen a silkworm, had only a vague idea of sericulture, and knew nothing about the disease. He began his 5-year study on the subject in 1865.

In most of his earlier research Pasteur worked alone, and few people knew the details of his results until he published them. But the investigations into the silkworm disease were different because by then he was a man of fame and great things were expected of him. He had the help of his colleague Émile Duclaux and three students, and many people in France, including Napoleon III, were continually inquiring or requesting results.

When Pasteur began his work, he found the silkworm industry in complete confusion, with few leads on how to attack the problem. Pasteur's intuition first led him to determine whether he was dealing with one or several causes of the disease and to evaluate the significance and nature of the corpuscles in the various stages of the silkworm's life. He visited silkworm nurseries, examined numerous eggs, worms, pupae, and moths, and designed many unique and carefully controlled experi-

ments. After much arduous research, he found that (i) infected insects are usually spotted, but spotted insects are not always infected; (ii) the disease develops mainly in pupae and moths; (iii) moths showing no corpuscles (parasites) produce eggs without them, but when moths do contain such bodies, they may or may not lay eggs with corpuscles; (iv) worms that contain corpuscles on hatching die in the moulting stages, before they become moths; and (v) corpuscles kept outside of eggs for a year, or dried, are incapable of reproduction. Eventually he discovered that this disease of the silkworm and other caterpillars was caused by a microsporidian, and he was able to work out a method for its control (10).

During his studies on pébrine, Pasteur investigated another silkworm disease, flacherie. His experiments showed one form of this disease to be a communicable intestinal infection caused by a "vibrion." The infection was spread by feces, contaminated mulberry leaves and food, or dust in the insect nursery. The incubation period varied from a day to a few weeks, and infected worms died before they could spin cocoons. He suggested ways to prevent this disease (10). Pasteur found at the conclusions of his researches that he had not only solved the problems that were destroying sericulture in France, but he had also pointed out the importance of practical, experimental research on microorganisms in microbiology and pathology.

Studies on Beer

Pasteur performed his studies on beer during a transition period in his life and career. In 1868, at the age of 46, he suffered a stroke that paralyzed the left side of his body, although his mind remained clear and lucid. He was soon able to return to his research and writing, even though discouraged over partial paralysis. In 1870, he was further disturbed, and his work interrupted, by the opening of the Franco-German war; he was so intensely loyal to his country that he returned his honorary doctor of medicine diploma to the University of Bonn at that time. Because of the war and his health, he was encouraged to return with his family to his old home in Arbois.

In 1871, while waiting to return to Paris and engage in greater scientific activities, Pasteur began studying beer.

He had no definite aim in mind at first; rather, he was humiliated by the war, and he set about to use his knowledge to help rehabilitate his country by raising the question: Was it not possible to make beer in France as good as that in Germany and free the country through science from paying tribute to breweries across the Rhine? Such was the ambition that led him to study the manufacture and preservation of beer (11).

After visiting breweries and studying the problems, he began an investigation in Duclaux's laboratory at Clermont-Ferrand on why beer undergoes spontaneous spoilage, becoming acid and even putrid, especially in warm climates. He demonstrated that these changes were always caused by microorganisms, and he showed brewers in France and England how the disorders could be prevented. He designed laboratory experiments, using pure cultures of selected yeast, that could be adopted to improve the brewing industry. His practical suggestions on the production, preservation, and improvement of plant equipment were so useful that he was granted several patents (11).

Virulent Maladies

By 1877, there was a great awakening of thought among physicians and scientists about infectious diseases of animals and human beings, and the scientific world was on the threshold of many exciting and beneficial discoveries in the field. The actinomycete of lumpy jaw in cattle, the spirochete of relapsing fever in human beings, and the anthrax bacillus in sheep had already been seen and studied briefly. Joseph Lister had made his first contribution to wound infection and aseptic surgery. As for Pasteur, he had published the results of his extensive study on silkworm diseases, which Émile Roux regarded as a veritable guide for anyone wishing to study contagious diseases.

During the next few years, Pasteur conducted exciting research related to such important diseases as anthrax, gaseous gangrene, chicken cholera, furunculosis, osteomyelitis, puerperal infection, swine erysipelas, and rabies. All of these studies caused many passionate controversies, but in the end they improved the health and welfare of mankind (12).

When Pasteur decided to enter the controversy over the cause of anthrax,

he had had experience in studying microbes, and he approached the disease from a practical point of view: How can the tremendous losses of sheep, cattle, and occasionally other animals be avoided?

As early as 1850, Casimir Davaine had seen a bacillus in the blood of sheep dying from anthrax, and he concluded that the bacillus was the cause. He found that blood was only infectious in the late stages of the disease, when the organism first appeared in the circulation. This relationship was disputed by persons who claimed that, in his crude experiments, Davaine was transferring poisonous products of the blood, or even an invisible agent (virus), from sick sheep to healthy animals. Somewhat later, when studying the etiology of anthrax, Robert Koch found that spores were formed by the bacillus seen by Davaine, but only in the blood and tissues of animals that had died of anthrax and providing that the temperature was suitable and there was sufficient oxygen. Koch also found that natural infection in animals came from the soil via their food.

In collaboration with Jules Joubert, Pasteur conducted his first experiments on anthrax in 1877. These consisted of finding a suitable medium in which the organism from a sick animal could be isolated and cultivated in the laboratory. After a medium was found and the organisms isolated, the properties of the bacillus were studied extensively, both in the laboratory and in experimental animals. They also isolated *Bacillus anthracis* from the soil in areas where sheep were dying and even implicated the earthworm as helping to spread the organism. They demonstrated that domestic fowl, contrary to widespread opinion, can be infected with anthrax if their normal body temperature (42°C) is reduced by about 4°C. Also, if the temperature of a diseased chicken is raised to normal again before the animal dies, the growth and virulence of the organism is decreased to such a level that the defense mechanisms of the body can bring the infection under control and the animal will survive.

After these experiments, Pasteur and his associates Émile Roux and Charles Chamberland turned their attention to studies on the virulence of the bacillus and the prophylaxis of anthrax. They found that it is possible to attenuate virulence and even to suppress it entirely by cultivating the bacillus through several transfers at 42° to 43°C. With such an attenuated culture, it then

seemed possible to vaccinate animals against anthrax, and one of Pasteur's most famous experiments was conducted in May 1881 at Pouilly-le-Fort (near Melun) before a large group of distinguished people. This experiment consisted simply of first immunizing one group of 25 sheep with the attenuated culture of the anthrax bacillus and keeping a similar group as nonvaccinated controls. Fifteen days after the last of two inoculations were given to the sheep being immunized, these animals and the controls were infected with a highly virulent anthrax culture. The nonimmunized sheep developed anthrax and died, whereas the immunized sheep remained healthy.

Pasteur therefore not only isolated and characterized the agent responsible for anthrax, but he discovered a way to prevent it. From this time on, he committed himself to a new path of research—the study of infectious diseases in animals and man.

While Pasteur was conducting his experiments on anthrax in poultry, his advice was asked on a disease known as "chicken cholera," which was causing great losses to French farmers. He was soon able to isolate the responsible bacterium from sick fowl and study it in the laboratory. He noted that the organism assumed several shapes, and he thought he might be dealing with an intermediate group of microbes. When cultures were transferred infrequently to a fresh medium and exposed to the air, they lost their virulence for chickens. From this observation, Pasteur suspected that he might have made a discovery of practical significance. This proved to be the case when he immunized chickens with an old, attenuated culture of the bacterium and then found later that the birds could not be infected with a highly virulent strain of the organism.

With the experience and wisdom gained from his past research, Pasteur directed his attention more and more to human diseases. His contributions were of great significance in explaining the role of anaerobic bacilli in gaseous gangrene and of cocci in boils, osteomyelitis, and puerperal infections. Not only was the work primarily responsible for the final proof of the germ theory of infectious diseases, but it helped establish the important discipline of immunology. Of even greater consequence, however, was the impact that this research had on improving hygiene in the practice of medicine and surgery.

Rabies Studies and Prophylaxis

The work (12) by Pasteur on rabies (hydrophobia) was the last in his long and famous scientific career, and it did more to promote his fame throughout the world than all his other research combined. This was partly because the studies ended with the use of his rabies vaccine in treating human patients.

In his many controversies about diseases with physicians and scientists of his day, Pasteur could not help thinking that human beings are exposed to the same dangers and are subject to the same disorders and diseases as animals. He had already observed these relationships with certain bacteria that cause disease in both animals and man, and he suspected the same was true for rabies. In 1880, the cause of rabies in dogs was unknown. The disease was mysterious in its incubation period, ranging from days to months; alarming in its symptoms when transmitted to human beings through dog bites; and impossible to cure once symptoms appeared.

When Pasteur decided to attack the confusing problem, his first attempts to discover the causative agent in the saliva and blood of mad dogs failed. In his reading, however, he noted that a veterinarian in Lyon was able to transmit the agent from dogs to rabbits by means of their saliva and brain tissue. With this information, and assisted by his colleagues Chamberland, Roux, and Louis Thuillier, Pasteur decided to continue his studies on rabies. He thought he might be able to isolate the agent, attenuate its virulence as he had done with the chicken-cholera microbe, and thereby prevent the disease.

In his 4 years of reading and raising questions about the source of the agent, designing crucial experiments to study the variable incubation period, and examining animal tissues and fluids for the agent, Pasteur obtained the following results (12): (i) symptoms of the disease were found to vary greatly, but in all cases (animal or human) the causative agent was the same; (ii) saliva of human patients may contain other microbial pathogens, a fact that complicated experimental results with rabbits; (iii) the brain, spinal cord, and probably the entire nerve system of persons who succumb to rabies contained the same agent, as was the case in animals with the disease; (iv) the incubation period of the disease could be shortened from weeks to several days by injecting rabid material through the skull and di-

rectly onto the brain of animals; and (v) the intravenous injection into healthy animals of the saliva or blood from a diseased animal did not confer immunity on the healthy one.

Further research demonstrated that the virulence of the rabies virus could be altered by passing it through animals. In passing material from the brain or spinal cord of a dog to a monkey and then from monkey to monkey, Pasteur and his colleagues found that the virulence decreased with each succeeding passage in the monkeys. When the virulence was reduced for the monkey, it also remained attenuated when inoculated back into the dog, rabbit, or guinea pig; furthermore, animals so treated remained refractory to rabies. On the other hand, the virulence of the virus increased when it was passed from rabbit to rabbit, and after 20 to 25 such passages the incubation period was reduced to a week and remained constant during further passages. When this virus from the rabbit was inoculated into dogs, it was found to be more virulent than the virus present in an ordinary rabid dog.

Pasteur and his colleagues then discovered that the virulence, but not the immunizing ability, of the virus in the spinal cord of a rabbit would, if suspended and stored in dry air, gradually decrease until it disappeared; the lower the temperature, the longer the virulence remained. They realized that they had discovered a possible method of immunization. Pasteur next designed a series of experiments in which he used dogs. The animals were inoculated under the skin on succeeding days with a suspension of rabbit spinal cord material that had been stored in the dry state for various periods. The first inoculations began with 15-day-old material known to be avirulent, 2 days later fresher cord material was used, and so on, at 2-day intervals, until fresh, highly virulent material was inoculated. Dogs so treated were resistant to rabies, even when inoculated on the surface of the brain. These experiments with dogs were repeated many times without failure, and the results became widely known.

One of the greatest decisions in scientific and medical history came on 6 July 1885, when 9-year-old Joseph Meister, who had been bitten 2 days

before by a rabid dog, was brought to Pasteur's laboratory. Pasteur was appalled at the thought of what might happen to the boy, and at the question of whether he had the right to apply his method of immunization to human beings. After Pasteur consulted with colleagues at the Academy of Medicine, who also examined Joseph and were convinced that he would become rabid within 2 to 3 weeks, it was decided that Jacques Grancher, a physician, should immunize the boy, using Pasteur's method. On each of the next 10 days, inoculations were made with progressively fresher emulsions of rabbit spinal cords. After this series of inoculations, the boy was observed for 3 months and was then considered out of danger. This dramatic triumph spread around the world and aroused both great enthusiasm and great controversy everywhere. During the next 15 months, nearly 2500 people who had been bitten by rabid dogs or wolves underwent preventive inoculations for rabies. In December 1886, Pasteur was awarded the Reynard Prize for his work on rabies, "one of the most magnificent in the records of science" (12, p. 897).

At this time, there began a drive to establish in Paris an antirabies center, to be called the Institut Pasteur. Donations came from persons around the world, including the czar of Russia, the sultan of Turkey, and the emperor of Brazil. On 14 November 1888, this now-famous institute was dedicated in the presence of the president of the Republic of France and other noted persons (6, 13, 14). Pasteur's advice to his colleagues at the time was (13), "Preserve the enthusiasm inspired by bacteriology . . . which has acquired such might from its recent conquests that it has captivated the minds of all." But, he added, "Always accompany this enthusiasm with rigid restraint. Put forward nothing that cannot be proved simply and decisively. . . . Cultivate a critical faculty. By itself it is neither an originator of ideas nor a stimulus to great things, but without it nothing is sound" (13).

What must have been one of the most glorious and touching ceremonies in the history of science took place in the amphitheater of the Sorbonne in 1892, in celebration of Pasteur's 70th birthday (15). The president of France

and other distinguished persons from many countries attended. Speaking on behalf of the French Academy of Sciences, Joseph Bertrand remarked (13): "Each of your great works shines with such brilliance that on studying it and regarding it closely, one is tempted to think that it eclipses all others; all are illuminated by the same light, the same accurate, extensive and sound science. . . ." Lister said to Pasteur (13): "Truly there does not exist in the whole world a person to whom medical science owes more than to you." And the publisher of *The Jubilee of M. Pasteur* concluded (13): "It was a unique spectacle in which a great man was . . . 'carried in triumph on the hearts of all.'"

Pasteur's last days were spent quietly, and he died on 28 September 1895.

With the great ideas, intuition, enthusiasm, experimental ability, and discoveries credited to this scientific genius, together with his loyalty and devotion to his country, family, and science, one must agree with Stephen Paget, who wrote (16, p. 509) of Pasteur that he is one of the most perfect men "who has ever entered the Kingdom of Science."

References and Notes

1. P. Vallery-Radot, *Oeuvres de Pasteur*, vol. 1, *Dissymétrie Moléculaire* (Masson, Paris, 1922), pp. 1-480.
2. ———, *Oeuvres de Pasteur*, vol. 2, *Fermentations et Générations Dites Spontanées* (Masson, Paris, 1922), pp. 1-664.
3. L. Pasteur, *Compt. Rend.* **45**, 913 (1857).
4. ———, *Ann. Sci. Natur. Ser. 4* **16**, 5 (1861).
5. J. Tyndall, *Floating Matter of the Air in Relation to Putrefaction and Infection* (Appleton, London, 1882), p. 338.
6. P. Vallery-Radot, *Oeuvres de Pasteur*, vol. 7, *Mélanges Scientifiques et Littéraires* (Masson, Paris, 1939), pp. 1-666.
7. L. Pasteur, *Compt. Rend.* **54**, 265 (1862).
8. P. Vallery-Radot, *Oeuvres de Pasteur*, vol. 3, *Études sur le Vinaigre et sur le Vin* (Masson, Paris, 1924), pp. 1-519.
9. So named because tiny black pepper-like specks appear on the body and in certain internal tissues of the silkworm.
10. P. Vallery-Radot, *Oeuvres de Pasteur*, vol. 4, *Études sur la Maladie des Vers à Soie* (Masson, Paris, 1926), pp. 1-716.
11. ———, *Oeuvres de Pasteur*, vol. 5, *Études sur la Bière* (Masson, Paris, 1928), pp. 1-361.
12. ———, *Oeuvres de Pasteur*, vol. 6, *Maladies Virulentes, Virus-Vaccins et Prophylaxie de la Rage* (Masson, Paris, 1933), pp. 1-906.
13. Quoted in L. Descour, *Pasteur and His Work* (Unwin, London, 1922), p. 238.
14. R. J. Dubos, *Louis Pasteur—Free Lance of Science* (Little, Brown, Boston, 1950), pp. 1-418.
15. On 20 November, the American Society for Microbiology held a Pasteur Commemoration Day in New Orleans. Next May, the French government, the Institut Pasteur, and the people of France, as well as other countries abroad, will honor him in Paris, as they have done in the past.
16. S. Paget, *The Spectator* (1 October 1910), pp. 509-510.