

LINUS PAULING: MOLECULAR DISEASE AND THE ORIGINS OF MOLECULAR BIOLOGY

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During the first quarter of the twentieth century chemistry had a deep influence on the development of medicine. Pauling¹ pointed out how some medical investigations had taken into account the last developments in Chemistry. An example could be the investigation around the use of chemical substances in medical therapies. The first works were, from a methodological point of view, not very orthodox; the investigations were dark and the progress low. Fortunately the scenario changed before 1950 and the chemists began to believe that there could be significant progress in the development of the pharmacology and the medical practice, once understood the molecular basis of the chemotherapeutic activity. To this end, it became necessary to investigate three fundamental concepts / aspects:

- *The detailed molecular structure of the chemotherapeutic substances.*
- *The detailed molecular structure of some constituents in the organisms against which they are directed (the bacteria, rickettsial bodies, viruses) and the human organism, with which the agents get in contact.*
- *The nature of the forces involved in the intermolecular interactions between the substances and the organisms.*²

¹ Pauling, L., (December 7, 1949).

² *Idem*, p. 4.

There were very significant progress around the first problem during the second quarter of the twentieth century; the development of methods of investigation on the structure of molecules, as the X-ray diffraction method, the progress in organic chemistry and the finding of structural formulas of many organic compounds helped to understand how atoms are linked in molecules such as penicillin. However, the knowledge of living organisms' molecular structure was much slower. The most important step was the determination in detail of the structure of some protein molecule. We must highlight Robert Coray and his collaborators' work, who worked on the crystal structure of amino acids and simple peptides. This works showed the relevance of the hydrogen bond and the hydrogen bond forces both in the configuration of the structure of a polypeptide chain and between adjacent molecules.

Step by step the knowledge of proteins and the forces between large molecules became more important; the configuration, the order of amino acids in a polypeptide chain, the nature of forces between atoms and other molecules and the nature of forces between large molecules, became the main questions to be solved.

The model of inspiration to explain the interaction between molecules of a chemotherapeutic agent and the protein molecules of the organism was the interaction between haptens and antibodies. H. Campbell, David Pressman and other Scientists had made some experiments in this field in Pasadena's laboratories. They found quantitative data about the inhibiting effects of haptens on the precipitation of precipitating antigens and antibodies and the requirements for the formation of a good bond between the hapten and the antibody.

Pauling began to show interest in the antibodies' structure and the nature of serologic reaction between 1936 and 1939. The reason was the special relation that Pauling had with Kart Landsteiner³ during these 3 years. It meant the

³ From *Rockefeller Institute for Medical Research*

beginning of a new kind of medicine. Pauling, with his works about sickle cell anemia and other investigations, had a principal role. When “Sickle-cell Anemia, a Molecular Disease” appeared in *Science* (1949), it had a strong impact on the biomedical community. The work played a crucial role at the beginning of molecular biology and molecular medicine. Pauling *et al.* showed that in anemia patient’s haemoglobin molecules had an electrical charge different from the molecules’ electrical charge in a healthy person. However, at that moment it was already known that human haemoglobin in adults and fetals differed from electrophoretically and that some diseases were related with the altered structure of blood proteins. So, what was important and novel in Pauling’s paper?

First of all, it demonstrated for the first time that a disease might be provoked by an alteration in molecular structure. Secondly, since it was known that this disease was hereditary, it was concluded that genes determined the structure of proteins. For this reason, several investigators understood that Pauling’s work meant the official inauguration of molecular medicine and molecular biology.

To understand the crucial role that placed Pauling at the origin of molecular biology and molecular medicine, we must review his previous and subsequent works. We could begin by referring to a text that Pauling published with Itano in January 1949, “A Rapid diagnostic test for sickle cell anemia”. In this paper, the sickle cell anemia was defined as «*a congenital chronic haemolytic type of anemia characterized haematologically by the development of oat-shaped and sickle-shaped erythrocytes*»⁴. The sickling process had been observed under microscope by several Scientists and it had distinguished *sickle cell anemia* from *sickle cell trait*. It was known that the erythrocytes of ill persons become sickled when the partial presion of oxygen or carbon monoxide is reduced. Then the haemoglobins begin to appear to aggregate into one or more foci, and the cell membranes collapse. The erythrocytes become birefringent and quite rigid. But

⁴ Itano, Harvey A., and Linus Pauling (January 1949), p. 1.

when oxygen or carbon monoxide is added to the haemoglobin, the cell recovers its original form. It was necessary to distinguish between the two forms. The form of the normal erythrocytes was designated *promeniscocyte* and the form of the sickle cell anemia *meniscocyte*.

The experiments indicated that the anemia was a result from the physical changes on the haemoglobin. For this reason the scientists had a pending question: «*Knowledge about the physical processes involved in sickling*».

The purpose of the paper “Sickle-cell Anemia, a Molecular Disease” was to solve this matter; «*to examine the physical and chemical properties of the haemoglobins of individuals with sickle cell trait and cell anemia, and to compare them with the haemoglobin of normal individuals to determine whether any significant differences might be observed*⁵».

The experimental works showed that there was a significant difference in the electrophoretic mobilities between the haemoglobin of normal individuals and that of sickle cell anaemic individuals. It was verified that the carbonmonoxyhemoglobin of a sickle cell anemia moves as a positive ion, while the normal compound moves as a negative ion. What was the reason for this difference? The differences were not in the particle weights or shapes of the two haemoglobins in solution. The experiments suggested the existence of a difference in the number or kind of ionisable groups in the two haemoglobins.

Only the carboxyl groups in the heme, and the carboxyl, imidazole, amino, phenolic hydroxyl, and guanidino groups in the globin, were the groups capable of forming ions in the carbonmonoxyhemoglobin.

Step by step the Scientists could draw the mechanism of the sickling process. The experiments indicated the globin region as the area to be searched.

⁵ Pauling, L. (November 25, 1949), p. 543.

In the globin region of the ill molecule there was a zone that did not exist in the normal molecule. This zone was complementary to other molecule's zone. This process happened when the partial pressure of the oxygen or carbon monoxide fell, consequently this zone should be near to an iron atom. Sickle cell anemia hemoglobin molecules could be capable of interacting with one another at these sites and to cause the partial alignment of molecules within the cell and his rigidity.

Although some details of this process were still mere assumptions the mechanism, but the mechanism was consistent with the experiments. The most important was that for the first time there was evidence that a disease may be provoked by an alteration in the molecular structure.

We have seen how Pauling's text was fundamental to understand a molecular disease for the first time, for the molecular medicine's origin. But, what about biology?

Since the very beginning, scientists had tried to give a genetic basis to the sickle cell anemia. Taliaferro and Huck had suggested that a dominant gene could be implied. Unfortunately the distinction between the sickle cell anemia and the sickle cell trait was not clear. Moreover, the existing literature in 1949 about the nature of the mechanism involved was still very confusing. Only Neel⁶ had made several experiments some time before Pauling had published "Sickle-cell Anemia, a Molecular Disease". Neel's investigation indicated *«that the gene responsible for the sickling characteristic is in heterozygous condition in individuals with sickle cell trait, and homozygous in those with sickle cell anemia»*⁷

⁶ Neel, J.V. (1949), p. 64.

⁷ Pauling, L. (November 25, 1949), p. 547.

Pauling held that his investigation had indicated the same before Neel had published his paper. Pauling affirmed that if the proposed mechanism was correct, then it was possible to identify the responsible gene.

Pauling's results on the investigation were consistent with the hypothesis of two genes responsible for the mechanism. This investigation revealed, for the first time, that a change in a protein molecule was caused by a change in a gene implied in the synthesis.

The results obtained in the experiments shown in the paper of 1949, suggested that the erythrocytes of other hereditary haemolytic anaemia had to be examined, with the aim to find the presence of other abnormal haemoglobins. For this reason, the famous paper concludes in this way: *"This we propose do"*.

This proposal had his results. Two kinds of new hemoglobin were discovered very soon. The second abnormal haemoglobin, the haemoglobin c (the letter *a* and *b* was applied to normal haemoglobin and sickle cell anemia haemoglobin respectively) was discovered by Itano y Neel⁸, and the hemoglobin *d* was discovered just by Itano⁹. After these discoveries (haemoglobins *a*, *b*, *c* and *d*) Scientists discovered 7 kinds of genetic combinations. It were the following:

Aa → normal individuals

Ab → individuals carrying sickle-cell trait

Bb → patients with sickle-cell-anemia

Ac → second abnormal hemoglobin c

Bc → patients with the first new disease, involving the inheritance of a sickle-cell-anemia allele and an allele for the second abnormal hemoglobin, c

Ad → carriers of the third abnormal hemoglobin, d

⁸ Itano, H. A., and J. V. Neel (1950).

⁹ Itano, H. A. (1951).

Bd → patients with the second new disease, resulting from the inheritance of a sickle-cell-anemia allele and an allele of the third abnormal hemoglobin, d.

While Scientists were studying the different combinations, other analyzed the related disease.

CONCLUSIONS

We can observe how these works from the late 40's and beginning of the 50's, physics and chemistry were implied in medicine and biology; the molecular medicine and molecular biology were a fact. 10 years after the publication of "*Sickle-cell Anemia, a Molecular Disease*", in the lecture "*Molecular Disease*"¹⁰, Pauling confirmed the discovery of 20 kinds of abnormal human haemoglobins associated with different diseases. The most important fact was that these diseases had a molecular cause. Pauling indicated that *«about two percent of viable children born have gross physical or mental defect because of their inheritance of defective genes. Many more suffer from minor hereditary defects»*¹¹. For this motive Pauling claimed constantly the need to increase the efforts in order to understand the molecular nature of these diseases. If we comprehend the molecular structure of this disease, we can develop effective therapies.

For the moment we have made a brief review of Pauling's work that had so much influence in the origins and subsequent development of molecular medicine and the molecular biology. This has been an exposition centered on historical and descriptive questions. However, if we want to give a complete image of a chapter or episode in the history of science, we should analyze the epistemological, ontological and conceptual questions that are at stake. In this case we want to pay attention to a problem that permeates all Pauling's work and

¹⁰ Pauling, L., (March 29, 1959).

¹¹ Pauling, L. (March 29, 1959), p. 4.

lots of his colleague's works. It is the problem of continuum space and three-dimensional space.

During this lecture we have been able to verify that in molecular medicine and molecular biology Scientists have made use of pictorial representation. The notion of complementary, very important to understand the sickle cell anemia mechanism and the DNA's functioning, needs a three-dimensional pictorial representation.

It was necessary to have a representation of the haemoglobin molecule and the DNA to understand their functioning and problems respectively. However, we must remind that the starting-point of molecular biology and molecular medicine is the quantum physics, where causality, the continuo space and the possibility of pictorial representations had disappeared. The Quantum World is an uncontinuous world where it is not possible to make three-dimensional pictorial representations. Are there two spaces with different ontological spaces? Or, is it only a space and a spatial pattern with a pragmatic purpose?

These two options must be studied according to pragmatism, idealism and realism. These ontological and epistemological problems that arised from the quantum physics reached the field of biology. There was a debate, not recognized by their main protagonists, in which N. Bohr and E. Schrödinger participated. In his work "What is life?" Schrödinger answered these questions and set the basis of the molecular biology that Pauling would use later.

The nature of a congress like this and the limited time we have, make impossible to carry out an accurate study of this problem. Although it is enough for our purpose to indicate that Pauling, consciously or unconsciously, had a major role when choosing Schrödinger's pattern; the influence of Pauling's work on the origin and development of the molecular medicine and molecular biology did not finish with his scientific investigations, we must also emphasize his

influence on the molecular biology's epistemology, ontology and methodology; the recovery of a continuous space, of three-dimensional descriptions, a new methodology and a new vision of natural life.

BIBLIOGRAPHY

- **González Recio, J. L.** (editor, 2005), "El taller de las ideas. 10 lecciones de historia de la ciencia", Madrid, Plaza y Valdés.
- **Itano, Harvey A., and Linus Pauling** (January 1949), "A Rapid Diagnostic Test for Sickle Cell Anemia." *Blood: The Journal of Hematology* IV, 1, pp. 66-68.
- **Itano, H. A., and J. V. Neel** (1950), "A new inherited abnormality of human hemoglobin", *Proc. Nat. Acad. Sci.* **36**: 613-617.
- **Itano, H. A.**, (1951), "A third abnormal hemoglobin associated with hereditary hemolytic anemia", *Proc. Nat. Acad. Sci.* **37**: 775-784, 1951.
- **Pauling, Linus** 26 September 1941, "Properties of Antibodies".
- **Pauling, Linus**, (25 May 1946), "Molecular Architecture and Biological Reactions." *Chemical and Engineering News* 24, 10, pp. 1375-1377.
- **Pauling, Linus** (17 July 1947), "Molecular Structure and Biological Specificity".
- **Pauling, Linus** (27 February 1948), "The Nature of Forces Between Large Molecules of Biological Interest." *Proceedings of the Royal Institution of Great Britain* 34, pp. 181-187.
- **Pauling, Linus**, Harvey A. Itano, S. J. Singer, and Ibert C. Wells (25 November 1949), "Sickle Cell Anemia, A Molecular Disease." *Science* 110, 2865, pp. 543-548.
- **Pauling, L.** (December 7, 1949), "Structural Chemistry in Relation to Biology and Medicine", in *Second Bicentennial Science Lecture of the City College Chemistry Alumni Association*, New York.
- **Pauling, Linus** (October 1952), "The Hemoglobin Molecule in Health and Disease." *Proceedings of the American Philosophical Society* 96, 5, pp. 556-565.
- **Pauling, Linus** (1955), "Abnormality of Hemoglobin Molecules in Hereditary Hemolytic Anemias." *Harvey Lectures* XII, pp. 216-241.
- **Pauling, Linus** (2 May 1956), "The Molecular Basis of Genetics".
- **Pauling, Linus** (1 May 1958), "Current Opinion: Molecular Disease." *Pfizer Spectrum* 6, 9.
- **Pauling, Linus** (29 March 1959), "Molecular Disease", lecture address to the *American Orthopsychiatry Association*, San Francisco.

- **Pauling, Linus** (1959), "Molecular Structure in Relation to Biology and Medicine." *Ciba Foundation Symposium on Significant Trends in Medical Research*. Ciba Foundation, p. 3-10.
- **Pauling, Linus** (1974), "Some Aspects of Orthomolecular Medicine" (pages 1-25).
- **Pauling, Linus** (1974), "Some Aspects of Orthomolecular Medicine" (pages 26-29, References).
- **Schrödinger, E.** (1967), *What is life? Mind and matter*, Cambridge, Cambridge University Press. En español (1997) *¿Qué es la vida?*, Tusquets, Barcelona.