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RITA LEVI-MONTALCINI: A REMARKABLE GENIUS WHO INSPIRES GENERATIONS

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Rita Levi-Montalcini, a renowned neuroscientist and brilliant scientist, left an unforgettable mark on science. She made history not only through her groundbreaking research but also as a figure with a unique and extraordinary life journey. Her courage, perseverance, and determination inspire new generations of scientists and individuals. Rita Levi-Montalcini became a symbol of success in a scientific world dominated by men. In 1986, her collaborative work with biochemist Stanley Cohen was awarded the Nobel Prize in Physiology or Medicine for her research on "growth factors," namely nerve growth factor (NGF) and epidermal growth factor (EGF). This discovery fundamentally changed our understanding of nervous system development and laid the foundation for understanding many neurological diseases.

K e y w o r d s: Nobel Prize, Rita Levi-Montalcini, nerve growth factors (NGF), neurological diseases, scientific legacy.

Rita Levi-Montalcini, as a distinguished scientist and Nobel Prize laureate in Physiology or Medicine in 1986, left a profound mark on the history of science, not only through her discoveries but also through her personality and life journey. The combination of her scientific contributions, personal example, and deep reflections makes Rita Levi-Montalcini a figure worth writing about time and again, despite the already well-known facts. Research and articles about Rita Levi-Montalcini are not only an attempt to preserve her name in history but also an opportunity to revisit her contributions to science and life, which continue to inspire and remain relevant to this day.

An extraordinary woman and renowned neurobiologist, she was born in Turin on April 22, 1909, as one of a pair of twins. She was the youngest of four children in a wealthy Italian Jewish family, led by Adamo Levi, an electrical engineer, and Adele Montalcini, an artist. At that time, societal expectations for women centered on becoming good wives and mothers, so her father refused to let Rita and her sisters, Paola and Anna, attend college. However, thanks to her family's support, both Rita and her sister Paola received an education at home, defying conventional norms. Initially, Rita dreamed of becoming a philosopher, but later decided that she lacked the logical thinking required and instead resolved to become a writer. She dreamed of telling an Italian saga in the style of Selma Lagerlöf, a Swedish author she greatly admired. But life took an unexpected turn, guiding her down a different path.

Rita Levi-Montalcini [1]

In her twenties, Rita realized that she was unlikely to conform to the traditional female role her father had envisioned, so she asked his permission to enroll in the University of Turin to become a doctor. In just eight months, she caught up in Latin, Greek, and mathematics, finished high school, and in 1930 entered the university in Turin. There, Rita develo-

ped close friendships with two classmates, Salvador Luria and Renato Dulbecco, both of whom studied under the esteemed Italian histologist Giuseppe Levi. Levi, a strict yet inspiring mentor, provided them with a solid foundation in biological sciences. Remarkably, all three went on to win the Nobel Prize in Physiology or Medicine–Luria and Dulbecco received theirs seventeen and eleven years, respectively, before Rita received hers.

In 1936, Rita graduated from medical school with honors in medicine and surgery and enrolled in a three-year residency in neurology and psychiatry, still unsure whether she would devote herself fully to the medical profession or pursue basic research in neurology at the same time.

While Rita was dreaming of a professional career, war broke out. Just before the invasion of the German army in Belgium, in the spring of 1940, Rita returned to Turin from Brussels, where she was a guest of the neurological institute. Her family had two alternatives: either to emigrate to the United States or to engage in some activity that required neither support nor contact with the outside Aryan world where they lived. The family chose the second option. Rita then decided to build a small research space in her bedroom. She was motivated by Victor Hamburger's 1934 paper on the consequences of limb extirpation in chicken embryos. She was motivated to undertake this research by Viktor Hamburger's study on the effects of limb extirpation in chicken embryos. At this time, Giuseppe Levi, who had fled Nazi-occupied Belgium, returned to Turin and joined Rita's research efforts [2].

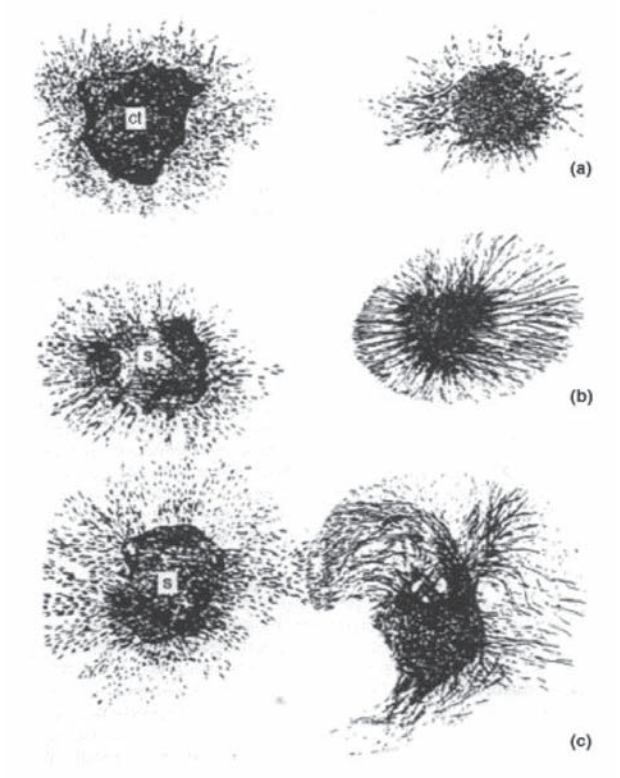
Rita Levi-Montalcini in her laboratory [3]

In 1941, the family moved to a rural cottage in Piedmont, where Rita constantly asked the peasants for a couple of eggs. She did not need them for food; Rita used chicken embryos for her experiments. In the fall of 1943, the invasion of Italy by the German army forced the family to flee to Florence, where they lived underground until the end of the war [2]. After the end of the war in May 1945, Rita and her family returned to scientific work at the university in Turin. In 1947, Professor Viktor Hamburger invited her to his laboratory in Washington University in St. Louis (WUSTL, Missouri) to continue the experiments they had conducted many years earlier on the chick embryos.

The research in Professor Viktor Hamburger's laboratory focused on the study of proliferation, differentiation, and degeneration in the spinal ganglia of chicken embryos under normal and experimental conditions [4]. Special attention was given to the modifications in the development of primary nerve center by changes occuring in tyhe peripheral areas which they innervate. During *in vitro* experiments examining the effects of mouse sarcomas 180 and 37 on the spinal and sympathetic ganglia of the chicken embryo [5], Rita discovered the outgrowth of certain nerve fibers in the chicken embryo. Sensory and sympathetic ganglia, explanted from 8-day-old chicken embryos into a semi-solid medium near–but not in contact with–fragments of mouse sarcomas 180 or 37, produced within 24 hours a dense halo of nerve fibers on the side facing the tumor [5]. Below is a figure drawn by Rita Levi-Montalcini [6].

Together with the young biochemist S. Cohen, who joined the research team in the early 1950s, and V. Hamburger, Rita investigated the effects of mouse sarcomas 37 and 180 on the sensory and sympathetic ganglia of chicken embryos. Tumor transplantation into 3-4-day-old chicken embryos caused numerical hyperplasia, cellular hypertrophy, and accelerated ganglia differentiation. *In vitro* tumor explantation in close proximity to sensory or sympathetic ganglia led to an exceptional outgrowth of nerve fibers. The *in vivo* and *in vitro* effects confirmed that in both cases, the researchers were dealing with the same stimulating agent (a nerve growth-promoting protein, later named nerve growth factor, or NGF). Thus, it was concluded that cell-free tumor homogenates could replicate the effect of actively growing tissue cultures [7].

Further studies were aimed at purifying and identifying NGF. When snake venom was used to purify the extract, it was unexpectedly discovered to contain its own active properties [8]. Assuming the venom originated from the snake's salivary glands, Cohen also examined the salivary glands of mice and found that they contained high levels of



Drawings illustrating the in vitro "halo" effect on 8-day chick embryo sensory ganglia culturedin the presence of fragments of mouse sarcoma 180 for 24 hours (b) or 48 hours (c). In (a), the ganglion that faces a fragment of chick embryonic tissue (cl) shows fibroblasts but few nerve fibers. In (b) and (c), the ganglia, facing fragments of sarcoma 180 (s) show the typical 'halo' effect elicited by the growth factor released from the sarcoma. Note in (c) the first evidence of a neurotropic effect of the growth factor [6]

NGF activity, a crucial step for protein purification [9]. Cohen produced rabbit antiserum that inhibited NGF activity in tissue culture and exhibited specific neurocytotoxic activity when injected into various mammal species.

To determine if NGF was physiologically important, Levi-Montalcini and graduate student Barbara Booker injected anti-NGF antiserum into newborn mice. The results of these studies were presented in articles [10, 11] in the same March 1960 issue of PNAS as Cohen's article [9]. The studies demonstrated how the growth factor stimulated nerve growth across a broad range of mammalian embryonic ganglia (including human tissue) and the effect of NGF on stimulating the growth of sympathetic ganglia when injected into newborn, young, and adult mice *in vivo* [10]. Additionally, it was shown that injecting anti-NGF serum into newborn mice, rats, rabbits, and kittens led to the near-complete destruction of the developing sympathetic nervous system without damaging other organs and tissues [11].

The use of antibodies (a type of phenotypic knockout) set the direction for further studies of growth factors and other proteins with the application of antibodies.In addition to describing the isolation of NGF and the preparation of antiserum, Cohen also observed that the treatment caused mice's eyes to open earlier and their teeth to emerge sooner compared to the controls, but the effects on eyes and teeth disappeared when he used a more highly purified NGF fraction [9]. Cohen suspected that the crude extract contained not only NGF but likely another factor responsible for the non-neuronal effects on newborn mice. Several years later, Cohen identified this second factor as epidermal growth factor (EGF).

Thus, NGF and EGF were the first among many signaling substances that regulate the growth of cells and organs to be discovered and characterized. For this discovery, Italian biologist Rita Levi-Montalcini and American biochemist Stanley Cohen received the Nobel Prize in 1986.

Many researchers began searching for additional factors that might influence the growth and survival of other types of nerves. However, the hunt for additional factors proved long and challenging, and it was only in the 1980s that Yves Barde, Hans Thoenen, and their colleagues isolated a factor from the brain, which they called brain-derived neurotrophic factor (BDNF). It soon became clear that NGF also acts in the brain, and two additional neurotrophins were subsequently discovered. As molecular neuroscience developed, it became evident that neurotrophins also play a role in the adult human brain. They promote learning and memory by supporting the survival of new synaptic connections. There is compelling evidence that reduced neurotrophic factor levels coincide with the onset of neurodegenerative diseases, such as Alzheimer's and Parkinson's, and these proteins are actively being studied as therapeutic tools for such diseases.

For the first time, chemically defined signaling substances could be used to investigate mechanisms regulating nervous system development. With modern gene technology, it has now become possible to characterize the NGF gene in humans and ani-

mals. Hybridization methods that identify messenger RNA for NGF have mapped the tissues that synthesize NGF [12].

Although Rita Levy planned to remain in St. Louis for only ten to twelve months, the excellent results of her research caused her to delay her return to Italy. Ultimately, she spent 30 years at Washington University. In 1956, she was offered the position of associate professor, and in 1958, she was offered a full professor position, which she held until her retirement in 1977. In 1962, Rita Levy established a research unit in Rome, dividing her time between that city and St. Louis. From 1969 to 1978, she also served as director of the Institute of Cell Biology of the Italian National Research Council in Rome, and after her retirement in 1979, became visiting professor at the same institute [13].

Upon learning that she had been awarded the Nobel Prize, she commented, "It was a great honor. But no award can compare with the moment of discovery." Despite the remarkable recognition of her work, awards were never her main goal. Notably, she was only the fourth woman to receive a Nobel Prize in Physiology or Medicine.

Rita Levi-Montalcini's career was truly extraordinary, and the discovery of NGF is only part of her story. In 1992, she established a foundation with her sister Paola to provide mentorship and guidance to children. In a scientific world long dominated by men, Rita Levi-Montalcini became a role model as a woman who reached the highest achievements. Her accomplishments are significant for advancing gender equality in science, making her an inspirational symbol for many researchers today. Her success story has inspired numerous young scientists, showing that despite obstacles and gender stereotypes, women can achieve great things in science.

In a 1993 interview with Margaret Holloway for Scientific American, Levi-Montalcini stated, "If I hadn't faced discrimination or persecution, I would never have won the Nobel Prize." In this interview, it was also highlighted how her recognition allowed her to support scientific initiatives and empower underprivileged communities, particularly through her ongoing support for education and research [14].

She founded the Rita Levi-Montalcini Foundation [15], an organization dedicated to supporting the education of young women, particularly in Africa, where her foundation offered scholarships to help them fulfill their academic dreams. In 2001, she expanded this foundation, which now provides educational support and scholarships for women and children from Africa.

In 2001, Italy appointed Levi-Montalcini as a lifetime senator, and in 2006 she made headlines during a clash with right-wing Italian politicians over budget cuts that reduced science funding. At age 97, she was a decisive vote in the Italian parliament in support of a budget that backed the government of Romano Prodi. She threatened to withdraw support unless the government reversed the recent science funding cuts. The funding was restored, and the budget was approved despite opposition leader Francesco Storace's attempts to silence her. Storace mockingly sent her crutches, claiming she was too old to vote and was a "crutch" for a struggling government [15].

In 2002, with her close collaborator Pietro Calissano and entrepreneur Luigi Amadio, owner of the Santa Lucia Foundation, Rita Levi-Montalcini founded the European Brain Research Institute (EBRI) in Rome. The EBRI project was born from the desire to create a high-level research center with international outreach in Italy, dedicated to studying the brain and its diseases. This project was presented by Professor Levi-Montalcini at a conference in Cernobbio in September 2001. Several cities (Varese, Turin, Trieste, and Rome), involving public and private institutions, competed for the project. A committee of external evaluators, including Nobel laureate Renato Dulbecco, selected Rome as the location. After its founding in 2002, EBRI hired a team of researchers selected by an International Scientific Council and began operations in 2005 [16].

EBRI facilitated the co-location of the Institute of Neurobiology and Molecular Medicine (INMM) and the laboratories of the Santa Lucia Foundation. Soon, the European Brain Research Center (CERC) was created, where around 150 researchers worked together closely, united in their mission to study the brain. CERC, with EBRI as its main driver and catalyst, evolved into a scientific campus that quickly gained international recognition thanks to its critical mass, shared experimental resources, know-how, and the creation of a scientific community. At CERC, Rita Levi-Montalcini often walked the institute's long corridors, conversing with researchers and opening the door to her office, listening attentively to their ideas. EBRI's international reach was demonstrated by frequent visits from foreign colleagues.

After the expiration of the ten-year lease, the CERC project was discontinued due to changes in

the management objectives of the Santa Lucia Foundation IRCCS. EBRI found new spaces, scientific horizons, and opportunities for scientific and clinical collaborations at its current location at Sapienza University in Rome. At the entrance to the new institute, visitors are greeted by a large framed photograph of Rita's smiling face [15].

Her life's work demonstrated that age was no barrier to achievement. Well into her 90s and early 100s, she often said, "My body may be showing signs of age, but my mind is sharper than ever". Her energy and intellect continued to inspire researchers globally. Despite her age, Rita Levi-Montalcini remained active in scientific and public life until her passing in 2012 at the age of 103. Her contributions to neuroscience and dedication to nurturing new generations of researchers left an indelible mark on the field. Even as she continued to advance brain research through EBRI, she also became a symbol of resilience, independence, and unwavering commitment to knowledge.

She used her platform not only to advance scientific endeavors but also to promote the importance of equal opportunities for women in science and academia, a cause she championed throughout her life. Rita's commitment to humanitarian causes extended beyond the laboratory.

Levi-Montalcini was never married, had no children, and never regretted it. In a 1988 interview

Rita Levi-Montalcini (standing) in her lab with her assistants [17]

Rita Levi-Montalcini, September 2008 [18]

with Omni magazine, she noted that marriage, even between two brilliant people, can suffer from resentment over unequal success. Levi-Montalcini was often asked about the secret to her success and longevity. She would reply that it was very simple: 'Little food, no husband, and no regrets'.

Nevertheless, she always enjoyed engaging with her students and friends, often hosting delightful gatherings. Those lucky enough to attend these gatherings enjoyed a relaxed atmosphere filled with fascinating conversations and exquisite cuisine. Rita Levi had refined taste, loved dressing elegantly, and always looked impeccable.

Rita Levi-Montalcini was actively involved in promoting science, mainly through lectures, publications, and interviews. She emphasized the importance of science for society and encouraged investment in scientific research. Her efforts helped raise awareness about neurobiology.

She was also the author or co-author of over 20 popular books, including her autobiography and dozens of scientific studies. She received numerous scientific honors, including the United States National Medal of Science, which President Ronald Reagan awarded to her at the White House in 1987.

Levi-Montalcini was not only a scientist but also a philosopher. She shared her reflections on the nature of life, the human mind, and the future of science, making her works fascinating not only for specific scientific discoveries but also as a source of profound ethical and philosophical ideas.

Here are some of her famous quotes:

"I tell young people: don't think about yourself, think about others. Think about the future awaiting you, about what you can do, and fear nothing".

"Above all, do not fear difficult moments. The best comes from them".

"Work hard in silence, let your success be your noise".

"First, find peace within yourself. Don't eat too much. Keep your mind active. Love".

In Rita Levi-Montalcini's obituary in The New York Times in 2012, a quote from her autobiography was included: "Imperfection–not perfection–is the ultimate outcome of the program embedded in this extraordinarily complex mechanism that is the human brain, as well as the result of environmental influences and those who care for us throughout the long years of our physical, psychological, and intellectual development."

Rita Levi-Montalcini's life was a testament to the boundless potential of the human spirit, and her groundbreaking work continues to fuel the drive to understand the brain, revealing mysteries that may one day revolutionize medicine as profoundly as her own discoveries did.

РИТА ЛЕВІ-МОНТАЛЬЧИНІ: ВИДАТНИЙ ГЕНІЙ, ЯКИЙ НАДИХАЄ ПОКОЛІННЯ

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Рита Леві-Монтальчині, відомий нейробіолог і блискуча вчена, залишила незабутній слід у науці. Вона увійшла в історію не лише завдяки своїм новаторським дослідженням, але й як особистість із надзвичайним життєвим шляхом. Її сміливість, наполегливість та рішучість надихають нові покоління науковців і особистостей. Рита Леві-Монтальчині стала символом успіху у світі науки, де панують чоловіки. У 1986 р. її спільна робота з біохіміком Стенлі Коеном була удостоєна Нобелівської премії з фізіології та медицини за дослідження «факторів росту», а саме фактора росту нервів (NGF) і епідермального фактора росту (EGF). Це відкриття докорінно змінило наше уявлення про розвиток нервової системи та заклало основу для розуміння багатьох неврологічних захворювань.

Ключові слова: Нобелівська премія, Рита Леві-Монтальчині, фактори росту нервів (NGF), неврологічні захворювання, наукова спадщина.

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