

Contents

Articles

| | |
|--------------------------------|----|
| Lamarckism | 1 |
| Transmutation of species | 14 |
| DNA methylation | 18 |
| Chromatin | 26 |
| Saltation (biology) | 32 |
| Histone | 40 |
| CRISPR | 48 |
| Piwi-interacting RNA | 61 |
| Hologenome theory of evolution | 65 |
| Epigenetics | 70 |
| Pangensis | 85 |
| Gemmule (pangensis) | 86 |

References

| | |
|------------------------------------------|----|
| Article Sources and Contributors | 88 |
| Image Sources, Licenses and Contributors | 90 |

Article Licenses

| | |
|---------|----|
| License | 91 |
|---------|----|

Lamarckism

Lamarckism (or **Lamarckian inheritance**) is the idea that an organism can pass on characteristics that it acquired during its lifetime to its offspring (also known as heritability of acquired characteristics or soft inheritance). It is named after the French biologist Jean-Baptiste Lamarck (1744–1829), who incorporated the action of soft inheritance into his evolutionary theories as a supplement to his concept of an inherent progressive tendency driving organisms continuously towards greater complexity, in parallel but separate lineages with no extinction. Lamarck did not originate the idea of soft inheritance, which proposes that individual efforts during the lifetime of the organisms were the main mechanism driving species to adaptation, as they supposedly would acquire adaptive changes and pass them on to offspring.

| Theories of Evolution |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <ul style="list-style-type: none"> • Based on Essentialism: <ul style="list-style-type: none"> • Transmutationism (saltationism) • Transformationism • Orthogenesis • Lamarckism • Based on population biology: <ul style="list-style-type: none"> • Darwinian evolution |

When Charles Darwin published his theory of evolution by natural selection in *On the Origin of Species*, he continued to give credence to what he called "use and disuse inheritance", but rejected other aspects of Lamarck's theories. Later, Mendelian genetics supplanted the notion of inheritance of acquired traits, eventually leading to the development of the modern evolutionary synthesis, and the general abandonment of the Lamarckian theory of evolution in biology. Despite this abandonment, interest in Lamarckism has continued (2009) as studies in the field of epigenetics have highlighted the possible inheritance of behavioral traits acquired by the previous generation.

History

Between 1794 and 1796 Erasmus Darwin wrote *Zoonomia* suggesting "that all warm-blooded animals have arisen from one living filament... with the power of acquiring new parts" in response to stimuli, with each round of "improvements" being inherited by successive generations. Subsequently Jean-Baptiste Lamarck repeated in his *Philosophie Zoologique* of 1809 the folk wisdom that characteristics which were "needed" were acquired (or diminished) during the lifetime of an organism then passed on to the offspring. He incorporated this mechanism into his thoughts on evolution, seeing it as resulting in the adaptation of life to local environments.

Lamarck founded a school of French *Transformationism* which included Étienne Geoffroy Saint-Hilaire, and which corresponded with a radical British school of anatomy based in the extramural anatomy schools in Edinburgh which included the surgeon Robert Knox and the comparative anatomist Robert Edmund Grant. In addition, the Regius Professor of Natural History, Robert Jameson, was the probable author of an anonymous paper in 1826 praising "Mr. Lamarck" for explaining how the higher animals had "evolved" from the "simplest worms" – this was the first use of the word "evolved" in a modern sense. As a young student, Charles



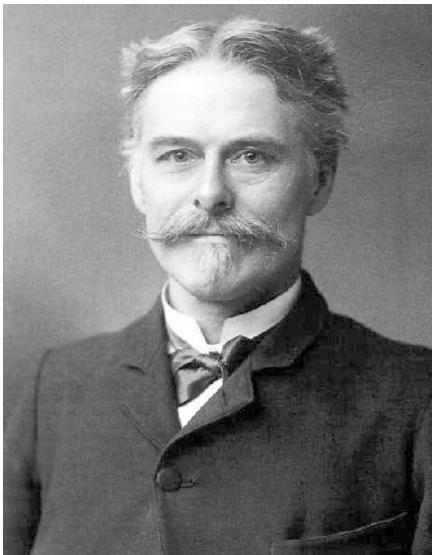
Jean-Baptiste Lamarck

Darwin was tutored by Grant, and worked with him on marine creatures.

The *Vestiges of the Natural History of Creation*, authored by Robert Chambers in St Andrews and published anonymously in England in 1844, proposed a theory which combined radical phrenology with Lamarckism, causing political controversy for its radicalism and unorthodoxy, but exciting popular interest and preparing a huge and prosperous audience for Darwin.

Darwin's *Origin of Species* proposed natural selection as the main mechanism for development of species, but did not rule out a variant of Lamarckism as a supplementary mechanism.^[2] Darwin called his Lamarckian hypothesis Pangenesis, and explained it in the final chapter of his book *Variation in Plants and Animals under Domestication*, after describing numerous examples to demonstrate what he considered to be the inheritance of acquired characteristics. Pangenesis, which he emphasised was a hypothesis, was based on the idea that somatic cells would, in response to environmental stimulation (use and disuse), throw off 'gemmules' or 'pangenes' which travelled around the body (though not necessarily in the bloodstream). These pangenes were microscopic particles that supposedly contained information about the characteristics of their parent cell, and Darwin believed that they eventually accumulated in the germ cells where they could pass on to the next generation the newly acquired characteristics of the parents. Darwin's half-cousin, Francis Galton carried out experiments on rabbits, with Darwin's cooperation, in which he transfused the blood of one variety of rabbit into another variety in the expectation that its offspring would show some characteristics of the first. They did not, and Galton declared that he had disproved Darwin's hypothesis of Pangenesis, but Darwin objected, in a letter to *Nature*, that he had done nothing of the sort, since he had never mentioned blood in his writings. He pointed out that he regarded Pangenesis as occurring in Protozoa and plants, which have no blood.

1880 to 1930



Edward Drinker Cope

The period of the history of evolutionary thought between Darwin's death in the 1880s and the foundation of population genetics in the 1920s, and the beginnings of the modern evolutionary synthesis in the 1930s is sometimes called the eclipse of Darwinism by some historians of science, because during that time many scientists and philosophers accepted the reality of evolution but doubted whether natural selection was the main evolutionary mechanism. Theories involving the inheritance of acquired characteristics were among the most popular alternatives to natural selection, and scientists who felt that such Lamarckian mechanisms were the key to evolution were called neo-Lamarckians. Proponents included the British botanist George Henslow who studied the effects of environmental stress on the growth of plants in the belief that such environmentally induced variation might explain much of plant evolution, and the American entomologist Alpheus Packard who studied blind animals living in caves and wrote a book in 1901 about Lamarck and his work. Also included were a number of paleontologists like Edward Drinker Cope and Alpheus

Hyatt who felt that the fossil record showed orderly, almost linear, patterns of development that they felt were better explained by Lamarckian mechanisms than by natural selection. Some people including Cope and Darwin critic Samuel Butler felt that inheritance of acquired characteristics would let organisms shape their own evolution, since organisms that acquired new habits would change the use patterns of their organs, which would kick-start Lamarckian evolution. They considered this philosophically superior to Darwin's mechanism of random variation

acted on by selective pressures. Lamarckism also appealed to those, like the philosopher Herbert Spencer and the German anatomist Ernst Haeckel, who saw evolution as an inherently progressive process. The German zoologist

Theodor Eimer combined Lamarckism with ideas about orthogenesis. With the development of the modern synthesis of the theory of evolution and a lack of evidence for either a mechanism or even the heritability of acquired characteristics, Lamarckism largely fell from favor.

In the 1920s, experiments by Paul Kammerer on amphibians, particularly the midwife toad, appeared to find evidence supporting Lamarckism, but his specimens with supposedly acquired black foot-pads were found to have been tampered with. In *The Case of the Midwife Toad* Arthur Koestler surmised that the tampering had been done by a Nazi sympathiser to discredit Kammerer for his political views, and that his research might actually have been valid. However most biologists believe that Kammerer was a fraud and even among those who believe he was not dishonest most believe that he misinterpreted the results of his experiments.

After 1930

A form of Lamarckism was revived in the Soviet Union of the 1930s when Trofim Lysenko promoted Lysenkoism which suited the ideological opposition of Joseph Stalin to genetics. This ideologically driven research influenced Soviet agricultural policy which in turn was later blamed for crop failures.^[3]

The biologist Ernest MacBride was a supporter of Lamarckian evolution. Herbert Graham Cannon a British zoologist defended Lamarckism in his book *Lamarck and Modern Genetics* in 1959.^[4]

Since 1988 certain scientists have produced work proposing that Lamarckism could apply to single celled organisms. A version of Lamarckian acquisition in higher order animals is still posited in certain branches of psychology, as, for example, in the Jungian racial memory.

Neo-Lamarckism is a theory of inheritance based on a modification and extension of Lamarckism, essentially maintaining the principle that genetic changes can be influenced and directed by environmental factors.

Lamarck's theory

The identification of Lamarckism with the inheritance of acquired characteristics is regarded by some as an artifact of the subsequent history of evolutionary thought, repeated in textbooks without analysis. Stephen Jay Gould wrote that late 19th century evolutionists "re-read Lamarck, cast aside the guts of it ... and elevated one aspect of the mechanics - inheritance of acquired characters - to a central focus it never had for Lamarck himself."^[5] He argued that "the restriction of "Lamarckism" to this relatively small and non-distinctive corner of Lamarck's thought must be labelled as more than a misnomer, and truly a discredit to the memory of a man and his much more comprehensive system". Gould

advocated defining "Lamarckism" more broadly, in line with Lamarck's overall evolutionary theory.

Lamarck incorporated two ideas into his theory of evolution, in his day considered to be generally true:

1. Use and disuse – Individuals lose characteristics they do not require (or use) and develop characteristics that are useful.
2. Inheritance of acquired traits – Individuals inherit the traits of their ancestors.



The long neck of the giraffe is often used as an example in explanations of Lamarckism.

Examples of what is traditionally called "Lamarckism" would include:

- Giraffes stretching their necks to reach leaves high in trees (especially Acacias), strengthen and gradually lengthen their necks. These giraffes have offspring with slightly longer necks (also known as "soft inheritance").
- A blacksmith, through his work, strengthens the muscles in his arms. His sons will have similar muscular development when they mature.

Lamarck stated the following two laws:

1. *Première Loi. Dans tout animal qui n'a point dépassé le terme de ses développements, l'emploi plus fréquent est soutenu d'un organe quelconque, fortifié peu à peu, cet organe le développe, l'agrandit, et lui donne une puissance proportionnée à la durée de cet emploi ; tandis que le défaut constant d'usage de tel organe, l'affaiblit insensiblement, le détériore, diminue progressivement ses facultés, et finit par le faire disparaître.*
2. *Deuxième Loi. Tout ce que la nature a fait acquérir ou perdre aux individus par l'influence des circonstances où leur race se trouve depuis long-temps exposée, et, par conséquent, par l'influence de l'emploi prédominant de tel organe, ou par celle d'un défaut constant d'usage de telle partie ; elle le conserve par la génération aux nouveaux individus qui en proviennent, pourvu que les changements acquis soient communs aux deux sexes, ou à ceux qui ont produit ces nouveaux individus.*^[6]

English translation:

1. *First Law. In every animal which has not passed the limit of its development, a more frequent and continuous use of any organ gradually strengthens, develops and enlarges that organ, and gives it a power proportional to the length of time it has been so used; while the permanent disuse of any organ imperceptibly weakens and deteriorates it, and progressively diminishes its functional capacity, until it finally disappears.*
2. *Second Law. All the acquisitions or losses wrought by nature on individuals, through the influence of the environment in which their race has long been placed, and hence through the influence of the predominant use or permanent disuse of any organ; all these are preserved by reproduction to the new individuals which arise, provided that the acquired modifications are common to both sexes, or at least to the individuals which produce the young.*^[7]

In essence, a change in the environment brings about change in "needs" (*besoins*), resulting in change in behavior, bringing change in organ usage and development, bringing change in form over time — and thus the gradual transmutation of the species.

However, as historians of science such as Michael Ghiselin and Stephen Jay Gould have pointed out, none of these views were original to Lamarck.^{[8][9]} On the contrary, Lamarck's contribution was a systematic theoretical framework for understanding evolution. He saw evolution as comprising two processes;

1. *Le pouvoir de la vie (a complexifying force)* - in which the natural, alchemical movements of fluids would etch out organs from tissues, leading to ever more complex construction regardless of the organ's use or disuse. This would drive organisms from simple to complex forms.
2. *L'influence des circonstances (an adaptive force)* - in which the use and disuse of characters led organisms to become more adapted to their environment. This would take organisms sideways off the path from simple to complex, specialising them for their environment.

Neo-Lamarckism

Unlike neo-Darwinism, the term neo-Lamarckism refers more to a loose grouping of largely heterodox theories and mechanisms that emerged after Lamarck's time, than to any coherent body of theoretical work.

In a series of experiments from 1869 to 1891, Charles-Édouard Brown-Séquard cut the sciatic nerve of the leg and spinal cord in the dorsal regions of guinea pigs, causing an abnormal nervous condition resembling epilepsy; these were then bred and produced epileptic offspring.^[10] Although some scientists considered this evidence for Lamarckian inheritance, the experiments were not Lamarckian, as they did not address the *use and disuse* of characteristics in response to the environment.^[11] The results from the experiment were not duplicated by other scientists.^[12] One explanation for the results was that they show a transmitted disease, and not evidence for the inheritance of an acquired characteristic.^[13] Brown-Séquard's experiments are now considered anomalous and alternative explanations have been suggested.

The French botanist Gaston Bonnier, conducting experiments in the French Alps in 1884 and the Pyrenees in 1886, studied structural changes induced by growing plants at various altitudes and transplanting them to others. Bonnier believed he had proven acquired adaptive characteristics; however, he did not weed, cultivate, fertilize or protect his plant specimens from native vegetation. In the 1920s his experiments were analysed and attributed to genetic contamination rather than Lamarckian inheritance.^[14]

In a series of experiments (in 1891, 1893 and 1895) on the action of light on the coloration of flatfish, the British marine biologist Joseph Thomas Cunningham directed light upon the lower sides of flatfishes by means of a glass-bottomed tank placed over a mirror. He discovered the influence of light in producing pigments on the lower sides of flatfishes and gave his results a Lamarckian interpretation. Other scientists wrote that Cunningham had received some definite results, but that they were open to more than one interpretation.^[15] The geneticist William Bateson was not convinced that the cause of the increase in pigmentation was from the illumination. George Romanes wrote approvingly of Cunningham's interpretation.^[16] Thomas Hunt Morgan (1903) criticized the experiments and did not believe the results were evidence for Lamarckism.^[17]

In the late 19th century Frederick Merrifield exposed caterpillars and chrysalids to significantly high and low temperatures, and discovered permanent changes in some offspring's wing patterns. Swiss biologist Max Standfuss led 30 years of intensive breeding experiments with European butterflies and after several generations, found similar preserved variations even generations after the cessation of exposing them to low temperatures.^[18] Standfuss was a Lamarckian and attributed the results of his experiments as direct changes to the environment.^[19] In 1940 Richard Goldschmidt interpreted these results without invoking Lamarckian inheritance, and in 1998 Ernst Mayr wrote that results reported by Standfuss and others on the effects of abnormal temperatures on Lepidoptera are difficult to interpret.^[20]

In 1910 the American zoologist Charles Rupert Stockard (1879-1939) tested the effects of alcohol intoxication on the offspring of pregnant guinea pigs. Stockard discovered that repeated alcohol intoxication in the guinea pigs produced defects and malformations in their offspring that was passed down to two or more generations. His results were challenged by the biologist Raymond Pearl who performed the same experiments with chickens. Pearl discovered that the offspring of the chickens that had been exposed to alcohol were not defected but were healthy.^[21] He attributed his findings to the detrimental effects of alcohol only on the eggs and sperm which were already weak, the strong eggs and sperm were unaffected by alcohol intoxication. Pearl argued that his results had a Darwinian, not a Lamarckian explanation.^[22]

The French zoologist Yves Delage in his book *The Theories of Evolution* (1912) reviewed experiments into Lamarckism concluded the evidence "is not of uniform value and is more or less open to criticism; very little of it is convincing... [due to] difficulties of experimentation and, above all, of interpretation."^[23]

In a series of experiments Francis Bertody Sumner (1874-1945) reared several generations of white mice under different conditions of temperature and relative humidity.^[24] Sumner discovered that the white mice at 20 °C to

30 °C developed longer bodies, tails and hind feet which were also transmitted to their offspring over a number of generations, however, later results were not entirely consistent and the experiments ended in uncertainty.^[25]

In the 1920s, Harvard University researcher William McDougall studied the abilities of rats to correctly solve mazes. He found that offspring of rats that had learned the maze were able to run it faster. The first rats would get it wrong 165 times before being able to run it perfectly each time, but after a few generations it was down to 20. McDougall attributed this to some sort of Lamarckian evolutionary process. Oscar Werner Tiegs and Wilfred Eade Agar later showed McDougall's results to be incorrect, caused by poor experimental controls.

The Russian physiologist Ivan Pavlov claimed to have observed a similar phenomenon in white mice being subject to a conditioned reflex experiment involving food and the sound of a bell. He wrote that with each generation, the mice became easier to condition. In 1926 Pavlov announced that there had been a fatal flaw in his experiment and retracted his claim to have demonstrated Lamarckian inheritance.^[26] In other experiments Coleman Griffith (1920, 1922) and John Detlefson (1923, 1925) reared rats in cages on a rotating table for three months. The rats adapted to the rotating condition to such an extent that when the rotation was stopped they showed signs of disequilibration and other physiological conditions which were inherited for several generations. In 1933 Roy Dorcus replicated their experiments but obtained different results as the rotated rats did not manifest any abnormalities of posture described by Griffith and Detlefson. Other studies revealed that the same abnormalities could occur in rats without rotation if they were suffering from an ear infection thus the results were interpreted as a case of infection, not Lamarckian inheritance.^[27]

Between 1918 and 1924 two American scientists Michael F. Guyer and Elizabeth A. Smith performed experiments in which fowl serum antibodies for rabbit lens-protein were injected into pregnant rabbits which resulted in defects in the eyes of some of their offspring that were inherited through eight generations. Their experiments were criticized and were not repeated by other scientists.^[28] In the 1930s the German geneticist Victor Jollos (1887-1941) in a series of experiments claimed evidence for inherited changes induced by heat treatment in *Drosophila melanogaster*. His experiments were described as Lamarckian however Jollos was not an advocate of Lamarckian evolution and attributed the results from his experiments as evidence for directed mutagenesis. American scientists were unable to replicate his results.^[29]

The American paleontologist Edward Drinker Cope was a neo-Lamarckian as he believed that an organism could respond to any challenge by choosing to adapt to a new way of life.^[30] The British anthropologist Frederic Wood Jones and the South African paleontologist Robert Broom supported a neo-Lamarckian view of human evolution as opposed to the Darwinian view. The German anthropologist Hermann Klaatsch relied on a neo-Lamarckian model of evolution to try and explain the origin of bipedalism. Neo-Lamarckism remained influential in biology until the 1940s when the role of natural selection was reasserted in evolution as part of the modern evolutionary synthesis.^[31]

In the 1970s the immunologist Ted Steele, formerly of the University of Wollongong, and colleagues, proposed a neo-Lamarckian mechanism to try to explain why homologous DNA sequences from the VDJ gene regions of parent mice were found in their germ cells and seemed to persist in the offspring for a few generations. The mechanism involved the somatic selection and clonal amplification of newly acquired antibody gene sequences that were generated via somatic hyper-mutation in B-cells. The mRNA products of these somatically novel genes were captured by retroviruses endogenous to the B-cells and were then transported through the blood stream where they could breach the soma-germ barrier and retrofect (reverse transcribe) the newly acquired genes into the cells of the germ line. Although Steele was advocating this theory for the better part of two decades, little more than indirect evidence was ever acquired to support it. An interesting attribute of this idea is that it strongly resembles Darwin's own theory of pangenesis, except in the soma to germ line feedback theory, pangenes are replaced with realistic retroviruses.^[32] Regarding Steele's research Peter J. Bowler wrote "his work was bitterly criticized at the time by biologists who doubted his experimental results and rejected his hypothetical mechanism as implausible."^[33]

Neo-Lamarckism was dominant in French biology for more than a century. French scientists who supported neo-Lamarckism included Edmond Perrier (1844-1921), Alfred Giard (1846-1908), Gaston Bonnier (1853-1922) and

Pierre-Paul Grassé (1895-1985).

Within the discipline of history of technology, Lamarckism has been used in linking cultural development to human evolution by classifying artefacts as extensions of human anatomy: in other words, as the acquired cultural characteristics of human beings. Ben Cullen has shown that a strong element of Lamarckism exists in sociocultural evolution.^[34]

Ideological neo-Lamarckism

Neo-Lamarckian versions of evolution were widespread in the late 19th century. The idea that living things could to some degree *choose* the characteristics that would be inherited allowed them things to be in charge of their own destiny as opposed to the Darwinian view, which made them puppets at the mercy of the environment. Such ideas were more popular than natural selection in the late 19th century as it made it possible for biological evolution to fit into a framework of a divine or naturally willed plan, thus the neo-Lamarckian view of evolution was often advocated by proponents of orthogenesis.^[35] According to historian of biology Peter J. Bowler:

One of the most emotionally compelling arguments used by the neo-Lamarckians of the late nineteenth century was the claim that Darwinism was a mechanistic theory which reduced living things to puppets driven by heredity. The selection theory made life into a game of Russian roulette, where life or death was predetermined by the genes one inherited. The individual could do nothing to mitigate bad heredity. Lamarckism, in contrast, allowed the individual to choose a new habit when faced with an environmental challenge and shape the whole future course of evolution.^[36]

Supporters of neo-Lamarckism such as George Bernard Shaw and Arthur Koestler claimed that Lamarckism is more humane, and optimistic than Darwinism.^[37]

Current views

Epigenetic inheritance

Forms of 'soft' or epigenetic inheritance within organisms have been suggested as neo-Lamarckian in nature by such scientists as Eva Jablonka and Marion J. Lamb. In addition to 'hard' or genetic inheritance, involving the duplication of genetic material and its segregation during meiosis, there are other hereditary elements that pass into the germ cells also. These include things like methylation patterns in DNA and chromatin marks, both of which regulate the activity of genes. These are considered "Lamarckian" in the sense that they are responsive to environmental stimuli and can differentially affect gene expression adaptively, with phenotypic results that can persist for many generations in certain organisms.^[38] In 2008, Jablonka and Lamb published a paper



Illustration of a DNA molecule that is methylated at the two center cytosines. DNA methylation plays an important role for epigenetic gene regulation in development and disease.

which claimed there is evidence for Lamarckian epigenetic control systems causing evolutionary changes and the mechanisms underlying epigenetic inheritance can also lead to saltational changes that reorganize the epigenome.

Interest in Lamarckism has increased, as studies in the field of epigenetics have highlighted the possible inheritance of behavioral traits acquired by the previous generation. A 2009 study examined foraging behavior in chickens as a function of stress, concluding:

Our findings suggest that unpredictable food access caused seemingly adaptive responses in feeding behavior, which may have been transmitted to the offspring by means of epigenetic mechanisms, including regulation of immune genes. This may have prepared the offspring for coping with an unpredictable environment.... Transmissions of information across generations which does not involve traditional inheritance of DNA-sequence alleles is often referred to as soft inheritance or 'Lamarckian inheritance'.

The evolution of acquired characteristics has also been shown in human populations who have experienced starvation, resulting in altered gene function in both the starved population and their offspring. The process of DNA methylation is thought to be behind such changes.

In October 2010, further evidence linking food intake to traits inherited by the offspring were shown in a study of rats conducted by several Australian universities. The study strongly suggested that fathers can transfer a propensity for obesity to their daughters as a result of the fathers' food intake, and not their genetics (or specific genes), prior to the conception of the daughter. A "paternal high-fat diet" was shown to cause cell dysfunction in the daughter, which in turn led to obesity for the daughter. Felicia Nowak, et al. reported at The Endocrine Society meeting in June 2013 that obese male rats passed on the tendency to obesity to their male offspring.^[39]

Several studies, one conducted by researchers at MIT and another by researchers at the Tufts University School of Medicine, have rekindled the debate once again. As reported in MIT's Technology Review in February 2009, "The effects of an animal's environment during adolescence can be passed down to future offspring ... The findings provide support for a 200-year-old theory of evolution that has been largely dismissed: Lamarckian evolution, which states that acquired characteristics can be passed on to offspring."^[40] A report investigating the inheritance of resistance to viral infection in the nematode *Caenorhabditis elegans* suggests that small RNA molecules may be inherited in a non Mendelian fashion and provide resistance to infection.

A scientific study (Akimoto *et al.* 2007) on epigenetic inheritance in rice plants came to the conclusion that "gene expression is flexibly tuned by methylation, allowing plants to gain or lose particular traits which are heritable as far as methylation patterns of corresponding genes are maintained. This is in support of the concept of Lamarckian inheritance, suggesting that acquired traits are heritable." Another study (Sano, 2010) wrote that observations suggest that acquired traits are heritable in plants as far as the acquired methylation pattern is stably transmitted which is consistent with Lamarckian evolution. Handel and Ramagopalan, 2010 have written that there is evidence that epigenetic alterations such as DNA methylation and histone modifications, are transmitted transgenerationally as a mechanism for environmental influences to be passed from parents to offspring. According to Handel and Romagopalan "epigenetics allows the peaceful co-existence of Darwinian and Lamarckian evolution."

In their book *An Introduction to Zoology* (2012) Joseph Springer and Dennis Holley wrote:

Lamarck and his ideas were ridiculed and discredited. In a strange twist of fate, Lamarck may have the last laugh. Epigenetics, an emerging field of genetics, has shown that Lamarck may have been at least partially correct all along. It seems that reversible and heritable changes can occur without a change in DNA sequence (genotype) and that such changes may be induced spontaneously or in response to environmental factors - Lamarck's "acquired traits". Determining which observed phenotypes are genetically inherited and which are environmentally induced remains an important and on going part of the study of genetics, developmental biology, and medicine.^[41]

Eugene Koonin has written that the prokaryotic CRISPR system and Piwi-interacting RNA could be classified as Lamarckian and came to the conclusion "Both Darwinian and Lamarckian modalities of evolution appear to be important, and reflect different aspects of the interaction between populations and the environment."

A study in 2013 reported that mutations caused by a father's lifestyle can be inherited by his children through multiple generations. A Swedish study showed that exercise changes the epigenetic pattern of genes that affect fat storage in the body.

Commenting on this, *Science Daily* explained,

The cells of the body contain DNA, which contains genes. We inherit our genes and they cannot be changed. The genes, however, have 'methyl groups' attached which affect what is known as 'gene expression' -- whether the genes are activated or deactivated. The methyl groups can be influenced in various ways, through exercise, diet and lifestyle, in a process known as 'DNA methylation'.

A 2013 study in *Nature* reported that mice trained to fear the smell of a chemical called acetophenone passed their fear onto at least two generations. An article on the study in the *New Scientist* titled *Mouse memory inheritance may revitalise Lamarckism* wrote "While it needs to be corroborated, this finding seems consistent with Lamarckian inheritance. It is, however, based on epigenetics: changes that tweak the action of genes, not the genes themselves. So it fits with natural selection – and may yet give Lamarck's name a sheen of respectability."

Guy Barry in a neuroscience paper in 2013 has written Darwin's hypothesis pangenesis coupled with "Lamarckian somatic cell-derived epigenetic modifications" and de novo RNA and DNA mutations can explain the evolution of the human brain.

Lamarckian elements also appear in the hologenome theory of evolution.

Critical reception

George Gaylord Simpson in his book *Tempo and Mode in Evolution* (1944) claimed that experiments in heredity have failed to corroborate any Lamarckian process.^[42] Simpson noted that neo-Lamarckism "stresses a factor that Lamarck rejected: inheritance of direct effects of the environment" and neo-Lamarckism is more closer to Darwin's pangenesis than Lamarck's views.^[43] Simpson wrote "the inheritance of acquired characters, failed to meet the tests of observation and has been almost universally discarded by biologists."^[44]

The botanist Conway Zirkle (1946) pointed out that Lamarck did not originate the hypothesis that acquired characters were heritable, therefore it is incorrect to refer to it as Lamarckism:

What Lamarck really did was to accept the hypothesis that acquired characters were heritable, a notion which had been held almost universally for well over two thousand years and which his contemporaries accepted as a matter of course, and to assume that the results of such inheritance were cumulative from generation to generation, thus producing, in time, new species. His individual contribution to biological theory consisted in his application to the problem of the origin of species of the view that acquired characters were inherited and in showing that evolution could be inferred logically from the accepted biological hypotheses. He would doubtless have been greatly astonished to learn that a belief in the inheritance of acquired characters is now labeled "Lamarckian," although he would almost certainly have felt flattered if evolution itself had been so designated.

Peter Medawar (1985) wrote regarding Lamarckism "very few professional biologists believe that anything of the kind occurs — or can occur — but the notion persists for a variety of nonscientific reasons." Medawar stated there is no known mechanism by which an adaption acquired in an individual's lifetime can be imprinted on the genome and Lamarckian inheritance is not valid unless it excludes the possibility of natural selection but this has not been demonstrated in any experiment.^[45]

Martin Gardner wrote in his book *Fads and Fallacies in the Name of Science*:

A host of experiments have been designed to test Lamarckianism. All that have been verified have proved negative. On the other hand, tens of thousands of experiments— reported in the journals and carefully checked and rechecked by geneticists throughout the world— have established the correctness of the gene-mutation theory beyond all reasonable doubt... In spite of the rapidly increasing evidence for

natural selection, Lamarck has never ceased to have loyal followers.... There is indeed a strong emotional appeal in the thought that every little effort an animal puts forth is somehow transmitted to his progeny.^[46]

According to Ernst Mayr (1997) any Lamarckian theory involving the inheritance of acquired characters has been refuted as "DNA does not directly participate in the making of the phenotype and that the phenotype, in turn, does not control the composition of the DNA."^[47] Peter J. Bowler has written although many early scientists took Lamarckism seriously, it was discredited by genetics in the early twentieth century.^[48]

Epigenetics

The evolutionary biologist T. Ryan Gregory has written that epigenetic inheritance should not be considered "Lamarckian". According to Gregory; Lamarck did not claim the environment imposed direct effects on organisms, instead Lamarck "argued that the environment created needs to which organisms responded by using some features more and others less, that this resulted in those features being accentuated or attenuated, and that this difference was then inherited by offspring." Gregory has stated that "Lamarckian" evolution in the context of epigenetics is actually closer to the view held by Darwin rather than by Lamarck.^[49]

Although the reality of epigenetic inheritance is not doubted (as many experiments have validated it), its significance to the evolutionary process is uncertain. Most neo-Darwinians consider epigenetic inheritance modifications to not be inherited past one or two generations, so are not a stable basis for evolutionary change.^{[50][51][52]}



Jerry Coyne

In a paper titled *Weismann Rules! Epigenetics and the Lamarckian Temptation* (2007), David Haig writes that research into epigenetic processes does allow a Lamarckian element in evolution but the processes do not challenge the main tenets of the modern evolutionary synthesis like modern Lamarckians have claimed. Haig argued for the primacy of DNA and evolution of epigenetic switches by natural selection. Haig has also written there is a "visceral attraction" to Lamarckian evolution from the public and some scientists as it posits the world with a meaning, in which organisms can shape their own evolutionary destiny.

Jerry Coyne has stated that "lots of studies show us that Lamarckian inheritance doesn't operate" and epigenetic changes are rarely passed on to future generations, thus do not serve as the basis of evolutionary change.^[53] Coyne has also written:

Lamarckism is not a "heresy," but simply a hypothesis that hasn't held up... If "epigenetics" in the second sense is so important in evolution, let us have a list of, say, a hundred adaptations of organisms that evolved in this Lamarckian way as opposed to the old, boring, neo-Darwinian way involving inherited changes in DNA sequence... I can't think of a single entry for that list.^[54]

Thomas Dickens and Qazi Rahman (2012) have written epigenetic mechanisms such as DNA methylation and histone modification are genetically inherited under the control of natural selection and do not challenge the modern synthesis. Dickens and Rahman have taken issue with the claims of Eva Jablonka and Marion Lamb on Lamarckian epigenetic processes.

Edith Heard and Robert Martienssen (2014) in a *Cell* review were not convinced that epigenetics has revived Lamarckism as there is no evidence epigenetic changes are passed on to multiple generations in mammals. They concluded the characteristics that are thought to be the result of epigenetic inheritance may be caused by other factors such as behavioral changes, undetected mutations, microbiome alterations or the transmission of metabolites.^[55]

References

- [1] Bonduriansky, R. (2012). *Rethinking heredity, again*. *Trends in Ecology & Evolution* 27: 330-336.
- [2] Desmond A. & Moore, J. (1991) *Darwin* Penguin Books p.617 "Darwin was to let go of the notion that a well-used and strengthened organ could be inherited"
- [3] Davis Baird, Eric R. Scerri, Lee C. McIntyre. (2005). *Philosophy of Chemistry: Synthesis of a New Discipline*. Springer. p. 166
- [4] Herbert Graham Cannon. (1975). *Lamarck and Modern Genetics* Greenwood Press Reprint. ISBN 0-8371-8173-9
- [5] Gould, Stephen J. "Shades of Lamarck", reprinted in *The Panda's Thumb* (1980) pp.65-71. Quote from page 66.
- [6] Jean-Baptiste Lamarck *Philosophie zoologique* (http://www.lamarck.cnrs.fr/ice/ice_book_detail.php?lang=fr&type=text&bdd=lamarck&table=ouvrages_lamarck&bookId=29&typeofbookId=1&num=0) ch.7, p.235
- [7] Jean-Baptiste Lamarck *Zoological Philosophy* trans. Hugh Elliot, 1914, p.113
- [8] The Imaginary Lamarck: a look at bogus "history" in schoolbooks (<http://www.textbookleague.org/54marck.htm>) by Michael Ghiselin
- [9] Gould, S.J. (2002) *The Structure of Evolutionary Theory*
- [10] Fredrick Blackmar Mumford. (1921). *The Breeding of Animals*. Macmillan. p. 209
- [11] Stephen Finney Mason. (1956). *Main Currents of Scientific Thought: A History of the Sciences*. Abelard-Schuman. p. 343. Also see Lamarck's Laws cited in Richard Burkhardt. (1995). *The Spirit of System: Lamarck and Evolutionary Biology*. Harvard University Press. p. 166
- [12] Martin Raitiere. (2012). *The Complicity of Friends: How George Eliot, G. H. Lewes, and John Hughlings-Jackson Encoded Herbert Spencer's Secret*. Bucknell University Press. p. 299
- [13] Henry Richardson Linville, Henry Augustus Kelly. (1906). *Text Book Of General Zoology*. Ginn & Company. p. 108
- [14] Robert E. Kohler. (2002). *Landscapes and Labscapes: Exploring the Lab-Field Border in Biology*. University Of Chicago Press. p. 167. ISBN 978-0-226-45010-0
- [15] The Spectator. (1928). Volume 141. p. 25
- [16] Alan G. Cock, Donald R. Forsdyke. (2008). *Treasure Your Exceptions: The Science and Life of William Bateson*. Springer. pp. 132-133. ISBN 978-0-387-75687-5
- [17] Thomas Hunt Morgan. (1903). *Evolution and Adaptation*. The Macmillan company. pp. 257-259
- [18] Auguste Forel. (1929). *The Sexual Question: A Scientific, Psychological, Hygienic and Sociological Study for the Cultured Classes*. Physicians and Surgeons Book Company. p. 36
- [19] John Michels. (1896). *Science*: Volume 4. Moses King. p. 53
- [20] Richard Goldschmidt (1940). *The Material Basis of Evolution*. Yale University. pp. 266-267. Ernst Mayr. (1998). *The Evolutionary Synthesis: Perspectives on the Unification of Biology*. Harvard University Press. p. 348
- [21] L. Doncaster. (1910). *Heredity in the Light of Recent Research*. Cambridge University Press. p. 98
- [22] Mark S. Blumberg. (2010). *Freaks of Nature: And what They Tell Us about Evolution and Development*. Oxford University Press. pp. 69-70
- [23] Yves Delage. (1912). *The Theories of Evolution*. B. W. Huebsch. pp. 210-235
- [24] Robert Thompson Young. (1922). *Biology in America*. R.G. Badger. p. 249
- [25] Charles Manning Child. (1947). *Biographical Memoir of Francis Bertold Sumner, 1874-1945*. In National Academy of Sciences of the United States of America Biographical Memoirs, Vol. 25. 146-73.
- [26] William McDougall. (1934). *Religion and the Sciences of Life: With Other Essays on Allied Topics*. Methuen & Co., Ltd. p. 180
- [27] Studies from the Otho S. A. Sprague Memorial Institute. (1940). Volume 25. p. 162
- [28] Peter Medawar. (1985). *Aristotle to Zoos: A Philosophical Dictionary of Biology*. Harvard University Press. p. 169
- [29] Jonathan Harwood. (1993). *Styles of Scientific Thought: The German Genetics Community, 1900-1933*. The University of Chicago Press. pp. 121-131.
- [30] Peter J. Bowler. (2009). *Evolution: The History of an Idea*. University of California Press. p. 227. ISBN 978-0-520-26128-0
- [31] Bernard Wood. (2013). *Wiley-Blackwell Encyclopedia of Human Evolution*. Wiley-Blackwell. ISBN 978-1-118-65099-8
- [32] Edward J. Steele, Robyn A. Lindley, Robert V. Blanden. (1998). *Lamarck's Signature : How Retrogenes Are Changing Darwin's Natural Selection Paradigm*. Perseus Books
- [33] Peter J. Bowler. (1989). The Mendelian Revolution: The Emergence of Hereditarian Concepts in Modern Science and Society. The Johns Hopkins University Press. p. 179
- [34] Ben Cullen. (2000). *Contagious Ideas: On evolution, culture, archaeology, and Cultural Virus Theory*. Oxbow Books. pp. 31-60
- [35] Peter J. Bowler. (1992). *The Eclipse of Darwinism: Anti-Darwinian Evolution Theories in the Decades around 1900*. The Johns Hopkins University Press. ISBN 978-0-8018-4391-4
- [36] Peter J. Bowler. (2003). *Evolution: The History of an Idea*. University of California Press p. 367. ISBN 978-0-520-26128-0
- [37] James Richard Moore. (2002). *History, Humanity and Evolution: Essays for John C. Greene*. Cambridge University Press. p. 330. ISBN 978-0-521-52478-0
- [38] Eva Jablonka, Marion J. Lamb. (1995). *Epigenetic Inheritance and Evolution: The Lamarckian Dimension*. Oxford University Press.
- [39] Obese male mice father offspring with higher levels of body fat (http://www.ohio.edu/research/communications/obese_paternal.cfm)
- [40] <http://www.technologyreview.com/biomedicine/22061>
- [41] Joseph Springer, Dennis Holley. (2012). *An Introduction to Zoology*. Jones & Bartlett Learning. p. 94
- [42] Simpson, George Gaylord. (1944). *Tempo and Mode in Evolution*. Columbia University Press. p. 75

- [43] Simpson, George Gaylord. (1964). *This View of Life: The World of an Evolutionist*. Harcourt, Brace & World. pp. 14-60
- [44] Simpson, George Gaylord. (1965). *Life: An Introduction to Biology*. Harcourt, Brace & World. p. 451
- [45] Medawar, Peter. (1985). *Aristotle to Zoos: A Philosophical Dictionary of Biology*. Harvard University Press. pp. 166-169
- [46] Gardner, Martin. (1957). *Fads and Fallacies in the Name of Science*. Dover Publications. pp. 142-143. ISBN 0-486-20394-8
- [47] Mayr, Ernst. (1997). *Evolution and the Diversity of Life: Selected Essays*. Harvard University Press. p. 222. "The recognition that DNA does not directly participate in the making of the phenotype and that the phenotype, in turn, does not control the composition of the DNA represents the ultimate invalidation of all theories involving the inheritance of acquired characters. This definitive refutation of Lamarck's theory of evolutionary causation clears the air."
- [48] Bowler, Peter. (2013). *Darwin Deleted: Imagining a World Without Darwin*. The University of Chicago Press. p. 21
- [49] Gregory, T. Ryan. (2008). "Lamarck didn't say it, Darwin did" (<http://www.genomicron.evolverzone.com/2009/03/lamarck-didnt-say-it-darwin-did/>). Evolver Zone.
- [50] Coyne, Jerry. (2010). "Epigenetics: the light and the way?" (<http://whyevolutionistrue.wordpress.com/2010/10/24/epigenetics-the-light-and-the-way/>). Why Evolution is True.
- [51] Coyne, Jerry. (2013). "Epigenetics smackdown at the Guardian" (<http://whyevolutionistrue.wordpress.com/2013/09/23/epigenetics-smackdown-at-the-guardian/>). Why Evolution is True.
- [52] Gorski, David. (2013). "Epigenetics: It doesn't mean what quacks think it means" (<http://www.sciencebasedmedicine.org/epigenetics-it-doesnt-mean-what-quacks-think-it-means/>). Science-Based Medicine.
- [53] Coyne Jerry. (2013). "More puffery about epigenetics, and my usual role as go-to curmudgeon" (<http://whyevolutionistrue.wordpress.com/2013/01/12/more-puffery-about-epigenetics-and-my-usual-role-as-go-to-curmudgeon/>). Why Evolution is True.
- [54] Coyne, Jerry. (2011). "Is "epigenetics" a revolution in evolution?" (<http://whyevolutionistrue.wordpress.com/2011/08/21/is-epigenetics-a-revolution-in-evolution/>). Why Evolution is True.
- [55] Heard, Edith; Martienssen, Robert. (2014). *Transgenerational Epigenetic Inheritance: Myths and Mechanisms*. Cell 157: 95-109.

Further reading

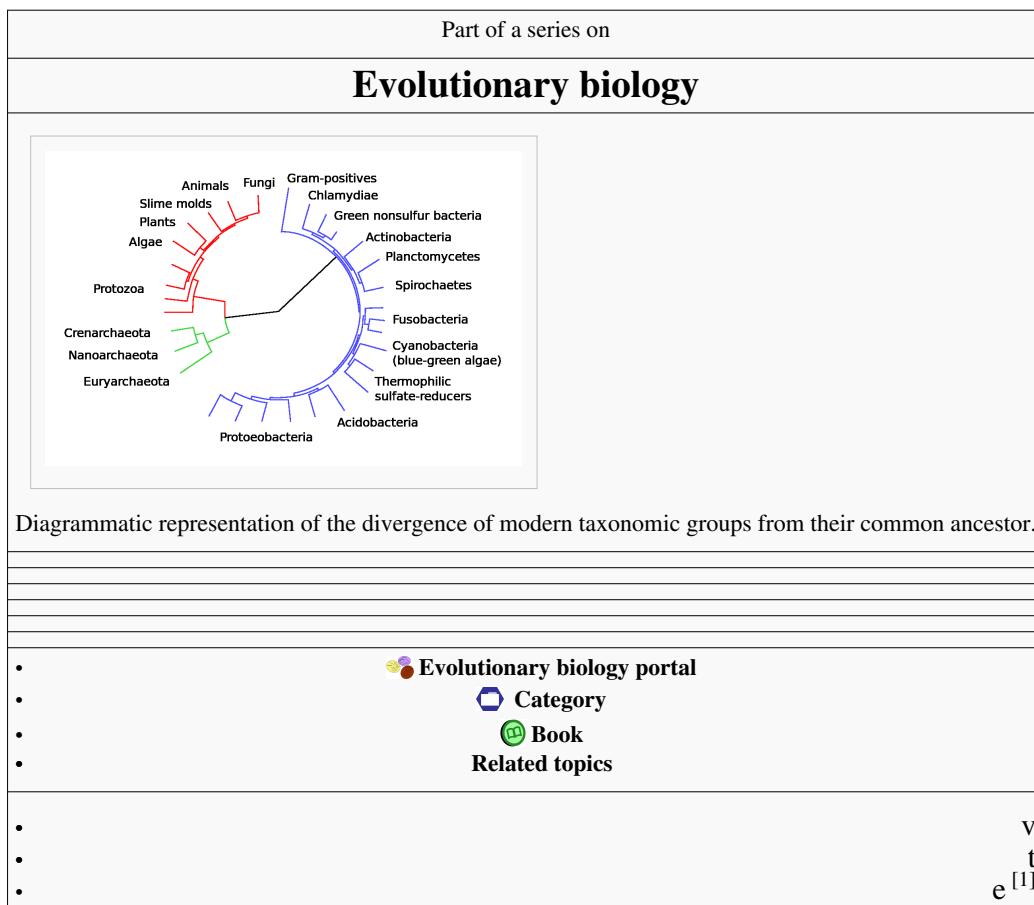
- Burkeman, Oliver. *Why everything you've been told about evolution is wrong* (<http://www.guardian.co.uk/science/2010/mar/19/evolution-darwin-natural-selection-genes-wrong>). *The Guardian*, March 19, 2010.
- Cook, George M. (December 1999). "Neo-Lamarckian Experimentalism in America: Origins and Consequences". *The Quarterly Review of Biology* **74** (4): 417–37. doi: 10.1086/394112 (<http://dx.doi.org/10.1086/394112>). JSTOR 2664721 (<http://www.jstor.org/stable/2664721>). PMID 10672643 (<http://www.ncbi.nlm.nih.gov/pubmed/10672643>).
- Cunningham, J. T. (2009). "Additional Evidence on the Influence of Light in producing Pigments on the Lower Sides of Flat Fishes". *Journal of the Marine Biological Association of the United Kingdom* **4**: 53. doi: 10.1017/S0025315400050761 (<http://dx.doi.org/10.1017/S0025315400050761>).
- Desmond, Adrian (1989). *The Politics of Evolution: Morphology, Medicine, and Reform in Radical London*. Chicago: University of Chicago Press. ISBN 0-226-14374-0.
- Gould, Stephen J. (2002). *The Structure of Evolutionary Theory*. Harvard: Belknap Harvard. pp. 170–197 on Lamarck. ISBN 0-674-00613-5.
- Medawar, Peter (1959). "The threat and the glory". BBC Reith Lectures No. 6.
- Molino, Jean (2001). "Toward an Evolutionary Theory of Music and Language" (<http://books.google.com/books?id=vYQEakqM4I0C&pg=PA165>). In Wallin, Nils L.; Merker, Björn; Brown, Steven. *The Origins of Music*. Cambridge: The MIT Press. pp. 165–76. ISBN 978-0-262-73143-0.
- Waddington, Conrad. (1961). "The human evolutionary system". In: Michael Banton (Ed.), *Darwinism and the Study of Society*. London: Tavistock.
- Seki, Yoshiyuki (2013). "Serum-mediated transgenerational effects on sperm: Evidence for lamarckian inheritance?". In Groszmann, Roberto J; Iwakiri, Yasuko; Taddei, Tamar H. *Hepatology* **57** (4): 1663–5. doi: 10.1002/hep.26240 (<http://dx.doi.org/10.1002/hep.26240>). PMID 23568276 (<http://www.ncbi.nlm.nih.gov/pubmed/23568276>).
- Honeywill, Ross (2008). *Lamarck's Evolution: two centuries of genius and jealousy*. Murdoch Books, Sydney See website (<http://lamarcksevolution.com/>)
- Fecht, Sarah. (2011). *Longevity Shown for First Time to Be Inherited via a Non-DNA Mechanism* (<http://www.scientificamerican.com/article.cfm?id=longevity-inheritance-epigenetics>). Scientific American.

- Ward, Lester Frank. (1891). *Neo-Darwinism and Neo-Lamarckism* (<http://archive.org/stream/neodarwinismand00washgoog#page/n6/mode/2up>). Washington Biological Society, Proceedings, VI.
- Jablonka, Eva. Lamb, Marion. (2008). *The Epigenome in Evolution: Beyond The Modern Synthesis* (http://www.bionet.nsc.ru/vogis/pict_pdf/2008/t12_1_2/vogis_12_1_2_21.pdf). VOGis Herald. Volume 12: 242–254.
- Persell, Stuart. (1999). *Neo-Lamarckism and the evolution controversy in France, 1870-1920*. Edwin Mellen Press.
- Burkhardt, R. W. (2013). "Lamarck, Evolution, and the Inheritance of Acquired Characters" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3730912>). *Genetics* **194** (4): 793–805. doi: 10.1534/genetics.113.151852 (<http://dx.doi.org/10.1534/genetics.113.151852>). PMC 3730912 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3730912>). PMID 23908372 (<http://www.ncbi.nlm.nih.gov/pubmed/23908372>).
- Burkhardt, Richard. (1995). *The Spirit of System: Lamarck and Evolutionary Biology*. Harvard University Press.
- Madaule, Madeleine Barthélémy. (1982). *Lamarck, the Mythical Precursor*. Mit Press.
- Liu, Yongsheng (2007). "Like father like son. A fresh review of the inheritance of acquired characteristics" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1973965>). *EMBO Reports* **8** (9): 798–803. doi: 10.1038/sj.embo.7401060 (<http://dx.doi.org/10.1038/sj.embo.7401060>). PMC 1973965 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1973965>). PMID 17767188 (<http://www.ncbi.nlm.nih.gov/pubmed/17767188>).
- Gissis, Snait. Jablonka, Eva. Zeligowski, Anna. (2011). *Transformations of Lamarckism: From Subtle Fluids to Molecular Biology*. MIT Press. ISBN 978-0262015141
- Peng, Wayne (2011). "Lamarckian viral defense in worms". *Nature Genetics* **44**: 15. doi: 10.1038/ng.1062 (<http://dx.doi.org/10.1038/ng.1062>).
- Pennisi, E. (2013). "Evolution Heresy? Epigenetics Underlies Heritable Plant Traits". *Science* **341** (6150): 1055. doi: 10.1126/science.341.6150.1055 (<http://dx.doi.org/10.1126/science.341.6150.1055>). PMID 24009370 (<http://www.ncbi.nlm.nih.gov/pubmed/24009370>).

External links

- Nonsense in Schoolbooks - The Imaginary Lamarck (<http://www.textbookleague.org/54marck.htm>): Michael T. Ghiselin recounts Lamarck's times and writings.
- Jean-Baptiste Lamarck : works and heritage (<http://www.lamarck.cnrs.fr/?lang=en>): an English/French web site edited by Pietro Corsi (Oxford Univ.) and realised by CNRS (France - IT team of CRHST). This web site contents all books, texts, manuscripts and Lamarck's Herbarium
- Guralnick, Rob, et. al (2006). "Jean-Baptiste Lamarck (1744-1829)" (<http://www.ucmp.berkeley.edu/history/lamarck.html>). *A History of Evolutionary Thought*. University of California Museum of Paleontology. Retrieved 3 July 2010.
- The Sins of the Fathers, Take 2 (<http://web.archive.org/web/20100104110615/http://www.newsweek.com/id/180103/output/print>): "At tributes to Darwin, Lamarckism — inheritance of acquired traits — will be the skunk at the party." Sharon Begley, *Newsweek*, Jan 17, 2009, from the magazine issue dated January 26, 2009
- Essay on Meta-Lamarckism (<http://lamarcksevolution.com/the-case-for-meta-lamarckism/>) by Ross Honeywill

Transmutation of species



Transmutation of species or **Transformism** are terms often used to describe 19th century evolutionary ideas for the altering of one species into another that preceded Charles Darwin's theory of natural selection.^[2] The French *Transformisme* was a term used by Jean Baptiste Lamarck in 1809 for his theory, and other 19th century proponents of pre-Darwinian evolutionary ideas included Étienne Geoffroy Saint-Hilaire, Robert Grant, and Robert Chambers who anonymously published the book *Vestiges of the Natural History of Creation*. Opposition in the scientific community to these early theories of evolution, led by influential scientists like the anatomists Georges Cuvier and Richard Owen and the geologist Charles Lyell, was intense. The debate over them was an important stage in the history of evolutionary thought and would influence the subsequent reaction to Darwin's theory.

Terminology

Transmutation was one of the names commonly used for evolutionary ideas in the 19th century before Charles Darwin published *On The Origin of Species* (1859). Transmutation had previously been used as a term in alchemy to describe the transformation of base metals into gold. Other names for evolutionary ideas used in this period include *the development hypothesis* (one of the terms used by Darwin) and *the theory of regular gradation*, used by William Chilton in the periodical press such as *The Oracle of Reason*.^[3] Transformation is another word used quite as often as transmutation in this context. These early 19th century evolutionary ideas played an important role in the history of evolutionary thought.

The proto-evolutionary thinkers of the 18th and early 19th century had to invent terms to label their ideas, but it was first Joseph Gottlieb Kölreuter who used the term "transmutation" to refer to species who have had biological changes through hybridization.^[4]

The terminology did not settle down until some time after the publication of the *Origin of Species*. The word *evolution* was quite a late-comer: it can be seen in Herbert Spencer's *Social Statics* of 1851,^[5] and there is at least one earlier example, but it was not in general use until about 1865-70.

Historical development

See also: Lamarckism

Jean-Baptiste Lamarck proposed a theory on the transmutation of species in *Philosophie Zoologique* (1809). Lamarck did not believe that all living things shared a common ancestor. Rather he believed that simple forms of life were created continuously by spontaneous generation. He also believed that an innate life force, which he sometimes described as a nervous fluid, drove species to become more complex over time, advancing up a linear ladder of complexity that was related to the great chain of being. Lamarck also recognized that species were adapted to their environment. He explained this observation by saying that the same nervous fluid driving increasing complexity, also caused the organs of an animal (or a plant) to change based on the use or disuse of that organ, just as muscles are affected by exercise. He argued that these changes would be inherited by the next generation and produce slow adaptation to the environment. It was this secondary mechanism of adaptation through the inheritance of acquired characteristics that became closely associated with his name and would influence discussions of evolution into the 20th century.

A radical British school of comparative anatomy that included the surgeon Robert Knox and the anatomist Robert Grant was closely in touch with Lamarck's school of French *Transformationism*, which contained scientists such as Étienne Geoffroy Saint-Hilaire. Grant developed Lamarck's and Erasmus Darwin's ideas of transmutation and evolutionism, investigating homology to prove common descent. As a young student Charles Darwin joined Grant in investigations of the life cycle of marine animals. He also studied geology under professor Robert Jameson whose journal published an anonymous paper in 1826 praising "Mr. Lamarck" for explaining how the higher animals had "evolved" from the "simplest worms" – this was the first use of the word "evolved" in a modern sense. Jameson's course closed with lectures on the "*Origin of the Species of Animals*".

The computing pioneer Charles Babbage published his unofficial *Ninth Bridgewater Treatise* in 1837, putting forward the thesis that God had the omnipotence and foresight to create as a divine legislator, making laws (or programs) which then produced species at the appropriate times, rather than continually interfering with *ad hoc* miracles each time a new species was required. In 1844 the Scottish publisher Robert Chambers anonymously published an influential and extremely controversial book of popular science entitled *Vestiges of the Natural History of Creation*. This book proposed an evolutionary scenario for the origins of the solar system and life on earth. It claimed that the fossil record showed a progressive ascent of animals with current animals being branches off a main line that leads progressively to humanity. It implied that the transmutations lead to the unfolding of a preordained plan that had been woven into the laws that governed the universe. In this sense it was less completely materialistic than the ideas of radicals like Robert Grant, but its implication that humans were just the last step in the ascent of animal life incensed many conservative thinkers. Both conservatives like Adam Sedgwick, and radical materialists

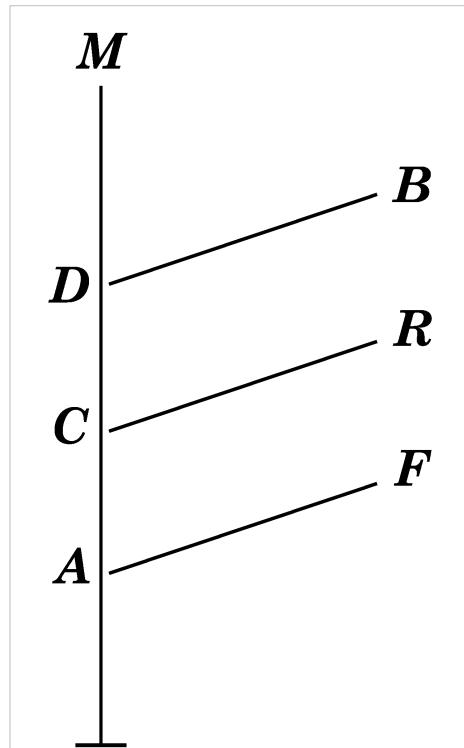


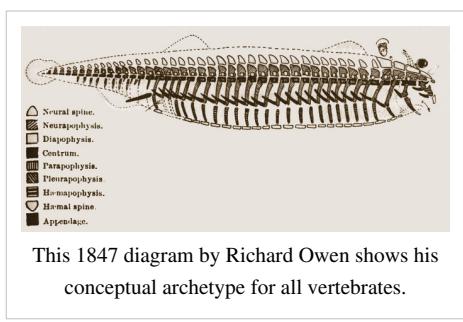
Diagram from the 1844 book *Vestiges of the Natural History of Creation* by Robert Chambers shows a model of development where fish (F), reptiles (R), and birds (B) represent branches from a path leading to mammals (M).

like Thomas Henry Huxley, who disliked Chambers' implications of preordained progress, were able to find scientific inaccuracies in the book that they could disparage. Darwin himself openly deplored the author's "poverty of intellect", and dismissed it as a "literary curiosity." However, the high profile of the public debate over *Vestiges*, with its depiction of evolution as a progressive process, and its popular success, would greatly influence the perception of Darwin's theory a decade later. It also influenced some younger naturalists, including Alfred Russel Wallace, to take an interest in the idea of transmutation.

Opposition to transmutation

Ideas about the transmutation of species were strongly associated with the radical materialism of the enlightenment and were greeted with hostility by more conservative thinkers. Cuvier attacked the ideas of Lamarck and Geoffroy Saint-Hilaire, agreeing with Aristotle that species were immutable. Cuvier believed that the individual parts of an animal were too closely correlated with one another to allow for one part of the anatomy to change in isolation from the others, and argued that the fossil record showed patterns of catastrophic extinctions followed by re-population, rather than gradual change over time. He also noted that drawings of animals and animal mummies from Egypt, which were thousands of years old, showed no signs of change when compared with modern animals. The strength of Cuvier's arguments and his reputation as a leading scientist helped keep transmutational ideas out of the scientific mainstream for decades.

In Britain, where the philosophy of natural theology remained influential, William Paley wrote the book *Natural Theology* with its famous watchmaker analogy, at least in part as a response to the transmutational ideas of Erasmus Darwin. Geologists influenced by natural theology, such as Buckland and Sedgwick, made a regular practice of attacking the evolutionary ideas of Lamarck and Grant, and Sedgwick wrote a famously harsh review of *The Vestiges of the Natural History of Creation*. Although the geologist Charles Lyell opposed scriptural geology he also believed in the immutability of species, and in his *Principles of Geology* (1830–1833), criticized and dismissed Lamarck's theories of development. Instead, he advocated a form of progressive creation, in which each species had its "centre of creation" and was designed for this particular habitat, but would go extinct when this habitat changed.



Another source of opposition to transmutation was a school of naturalists who were influenced by the German philosophers and naturalists associated with idealism, such as Goethe, Hegel and Lorenz Oken. Idealists such as Louis Agassiz and Richard Owen believed that each species was fixed and unchangeable because it represented an idea in the mind of the creator. They believed that relationships between species could be discerned from developmental patterns in embryology, as well as in the fossil record: but that these relationships represented an underlying pattern of divine thought, with progressive

creation leading to increasing complexity and culminating in humanity. Owen developed the idea of "archetypes" in the Divine mind that would produce a sequence of species related by anatomical homologies, such as vertebrate limbs. Owen was concerned by the political implications of the ideas of transmutationists like Robert Grant, and he led a public campaign by conservatives that successfully marginalized Grant in the scientific community. In his famous 1841 paper, which coined the term dinosaur for the giant reptiles discovered by Buckland and Gideon Mantell, Owen argued that these reptiles contradicted the transmutational ideas of Lamarck because they were more sophisticated than the reptiles of the modern world. Darwin would make good use of the homologies analyzed by Owen in his own theory, but the harsh treatment of Grant, along with the controversy surrounding *Vestiges*, would be factors in his decision to ensure that his theory was fully supported by facts and arguments before publishing his ideas.

References

- [1] http://en.wikipedia.org/w/index.php?title=Template:Evolutionary_biology&action=edit
- [2] Sloan, Phillip, "Evolution", The Stanford Encyclopedia of Philosophy (Fall 2010 Edition), Edward N. Zalta (ed.), online (<http://plato.stanford.edu/archives/fall2010/entries/evolution/>)
- [3] Secord, James A. 2000. *Victorian sensation: the extraordinary publication, reception, and secret authorship of the Vestiges of the Natural History of Creation*. Chicago, p311
- [4] <http://plorenzano.files.wordpress.com/2012/08/xxi-joseph-gottlieb-kc3b6lreuter-plorenzano.pdf>
- [5] There are three examples of the word 'evolution' in *Social Statics*, but none in the sense that is used today in biology. See (http://oll.libertyfund.org/index.php?option=com_staticxt&staticfile=advanced_search.php)

Bibliography

- Bowler, Peter J. (2003). *Evolution: The History of an Idea*. University of California Press. ISBN 0-520-23693-9.
- Desmond, Adrian; Moore, James (1994). *Darwin: The Life of a Tormented Evolutionist*. W. W. Norton & Company. ISBN 0-393-31150-3.
- Bowler, Peter J.; Morus, Iwan Rhys (2005). *Making Modern Science*. The University of Chicago Press. ISBN 0-226-06861-7.
- Larson, Edward J. (2004). *Evolution: The Remarkable History of Scientific Theory*. Modern Library. ISBN 0-679-64288-9.
- van Wyhe, John (27 March 2007). "Mind the gap: Did Darwin avoid publishing his theory for many years?" (http://darwin-online.org.uk/people/van_Wyhe_2007_Mind_the_gap_did_Darwin_avoid_publishing_his_theory.pdf) (PDF). *Notes and Records of the Royal Society* **61** (2): 177–205. doi: 10.1098/rsnr.2006.0171 (<http://dx.doi.org/10.1098/rsnr.2006.0171>). Retrieved 2008-02-07..

External links

- Darwin's Precursors and Influences (<http://www.talkorigins.org/faqs/precursors/precursortrans.html>)
- The lead up to *The Origins of Species* (<http://mechanism.ucsd.edu/~bill/teaching/philbiology/Darwin1.pdf>)

DNA methylation

DNA methylation is a biochemical process where a methyl group is added to the cytosine or adenine DNA nucleotides. The rate of cytosine DNA methylation differs strongly between species, e.g. absolute quantification by mass spectrometry revealed 14% of cytosines methylated in *Arabidopsis thaliana*, 8% in *Mus musculus*, 2.3% in *Escherichia coli*, 0.03% in *Drosophila*, and virtually none (< 0.0002%) in yeast species. DNA methylation may stably alter the expression of genes in cells as cells divide and differentiate from embryonic stem cells into specific tissues. The resulting change is normally permanent and unidirectional, preventing a cell from reverting to a stem cell or converting into a different cell type. DNA methylation is typically removed during zygote formation and re-established through successive cell divisions during development. However, the latest research shows that hydroxylation of methyl groups occurs rather than complete removal of methyl groups in the zygote. Some methylation modifications that regulate gene expression are heritable and cause genomic imprinting.

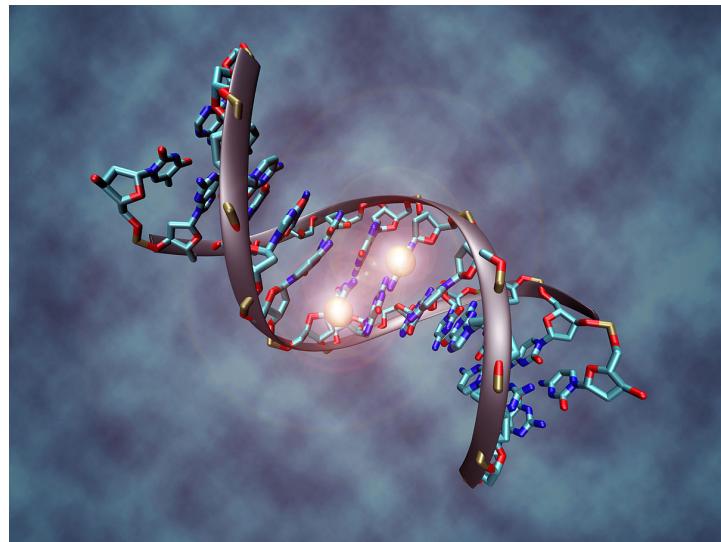


Illustration of a DNA molecule that is methylated at the two center cytosines. DNA methylation plays an important role for epigenetic gene regulation in development and disease.

DNA methylation suppresses the expression of endogenous retroviral genes and other harmful stretches of DNA that have been incorporated into the host genome over time. DNA methylation also forms the basis of chromatin structure, which enables a single cell to grow into multiple organs or perform multiple functions. DNA methylation also plays a crucial role in the development of nearly all types of cancer.

DNA methylation at the 5 position of cytosine has the specific effect of reducing gene expression and has been found in every vertebrate examined. In adult somatic cells (cells in the body, not used for reproduction), DNA methylation typically occurs in a CpG dinucleotide context; non-CpG methylation is prevalent in embryonic stem cells, and has also been indicated in neural development.

In mammals

DNA methylation is essential for normal development and is associated with a number of key processes including genomic imprinting, X-chromosome inactivation, suppression of repetitive elements, and carcinogenesis.

Between 60% and 90% of all CpGs are methylated in mammals. Methylated C residues spontaneously deaminate to form T residues over time; hence CpG dinucleotides steadily deaminate to TpG dinucleotides, which is evidenced by the under-representation of CpG dinucleotides in the human genome (they occur at only 21% of the expected frequency). (On the other hand, spontaneous deamination of unmethylated C residues gives rise to U residues, a change that is quickly recognized and repaired by the cell.)

Unmethylated CpGs are often grouped in clusters called *CpG islands*, which are present in the 5' regulatory regions of many genes. In many disease processes, such as cancer, gene promoter CpG islands acquire abnormal hypermethylation, which results in transcriptional silencing that can be inherited by daughter cells following cell division. Alterations of DNA methylation have been recognized as an important component of cancer development.

Hypomethylation, in general, arises earlier and is linked to chromosomal instability and loss of imprinting, whereas hypermethylation is associated with promoters and can arise secondary to gene (oncogene suppressor) silencing, but might be a target for epigenetic therapy.

DNA methylation may affect the transcription of genes in two ways. First, the methylation of DNA itself may physically impede the binding of transcriptional proteins to the gene, and second, and likely more important, methylated DNA may be bound by proteins known as methyl-CpG-binding domain proteins (MBDs). MBD proteins then recruit additional proteins to the locus, such as histone deacetylases and other chromatin remodeling proteins that can modify histones, thereby forming compact, inactive chromatin, termed heterochromatin. This link between DNA methylation and chromatin structure is very important. In particular, loss of methyl-CpG-binding protein 2 (MeCP2) has been implicated in Rett syndrome; and methyl-CpG-binding domain protein 2 (MBD2) mediates the transcriptional silencing of hypermethylated genes in cancer.

Research has suggested that long-term memory storage in humans may be regulated by DNA methylation.

DNA methylation levels can be used to estimate age, forming an accurate biological clock in humans and chimpanzees.

In cancer

DNA methylation is an important regulator of gene transcription and a large body of evidence has demonstrated that genes with high levels of 5-methylcytosine in their promoter region are transcriptionally silent, and that DNA methylation gradually accumulates upon long-term gene silencing. DNA methylation is essential during embryonic development, and in somatic cells, patterns of DNA methylation are generally transmitted to daughter cells with a high fidelity. Aberrant DNA methylation patterns – hypermethylation and hypomethylation compared to normal tissue – have been associated with a large number of human malignancies. Hypermethylation typically occurs at CpG islands in the promoter region and is associated with gene inactivation. A lower level of leukocyte DNA methylation is associated with many types of cancer. Global hypomethylation has also been implicated in the development and progression of cancer through different mechanisms. Typically, there is hypermethylation of tumor suppressor genes and hypomethylation of oncogenes.

In atherosclerosis

Epigenetic modifications such as DNA methylation have been implicated in cardiovascular disease, including atherosclerosis. In animal models of atherosclerosis, vascular tissue as well as blood cells such as mononuclear blood cells exhibit global hypomethylation with gene-specific areas of hypermethylation. DNA methylation polymorphisms may be used as an early biomarker of atherosclerosis since they are present before lesions are observed, which may provide an early tool for detection and risk prevention.^[1]

Two of the cell types targeted for DNA methylation polymorphisms are monocytes and lymphocytes, which experience an overall hypomethylation. One proposed mechanism behind this global hypomethylation is elevated homocysteine levels causing hyperhomocysteinemia, a known risk factor for cardiovascular disease. High plasma levels of homocysteine inhibit DNA methyltransferases, which causes hypomethylation. Hypomethylation of DNA affects genes that alter smooth muscle cell proliferation, cause endothelial cell dysfunction, and increase inflammatory mediators, all of which are critical in forming atherosclerotic lesions.^[2] High levels of homocysteine also result in hypermethylation of CpG islands in the promoter region of the estrogen receptor alpha (ER α) gene, causing its down regulation.^[3] ER α protects against atherosclerosis due to its action as a growth suppressor, causing the smooth muscle cells to remain in a quiescent state.^[4] Hypermethylation of the ER α promoter thus allows intimal smooth muscle cells to proliferate excessively and contribute to the development of the atherosclerotic lesion.^[5]

Another gene that experiences a change in methylation status in atherosclerosis is the monocarboxylate transporter (MCT3), which produces a protein responsible for the transport of lactate and other ketone bodies out of many cell types, including vascular smooth muscle cells. In atherosclerosis patients, there is an increase in methylation of the

CpG islands in exon 2, which decreases MCT3 protein expression. The down regulation of MCT3 impairs lactate transport, and significantly increases smooth muscle cell proliferation, which further contributes to the atherosclerotic lesion. An ex vivo experiment using the demethylating agent Decitabine (5-aza-2'-deoxycytidine) was shown to induce MCT3 expression in a dose dependant manner, as all hypermethylated sites in the exon 2 CpG island became demethylated after treatment. This may serve as a novel therapeutic agent to treat atherosclerosis, although no human studies have been conducted thus far.^[6]

In aging

A longitudinal study of twin children, showed that between the ages of 5 and 10 there was divergence of methylation patterns due to environmental rather than genetic influences. There is a global loss of DNA methylation during aging. But some genes become hypermethylated with age, including genes for the estrogen receptor, p16, and insulin-like growth factor 2. Biological clocks such as an epigenetic clock, are promising biomarkers of aging.

In exercise

High intensity exercise has been shown to result in reduced DNA methylation in skeletal muscle. Promoter methylation of PGC-1 α and PDK4 were immediately reduced after high intensity exercise, whereas PPAR- γ methylation was not reduced until three hours after exercise. By contrast, six months of exercise in previously sedentary middle-age men resulted in increased methylation in adipose tissue. One study showed a possible increase in global genomic DNA methylation of white blood cells with more physical activity in non-Hispanics.

DNA methyltransferases

In mammalian cells, DNA methylation occurs mainly at the C5 position of CpG dinucleotides and is carried out by two general classes of enzymatic activities – maintenance methylation and *de novo* methylation.^[7]

Maintenance methylation activity is necessary to preserve DNA methylation after every cellular DNA replication cycle. Without the DNA methyltransferase (DNMT), the replication machinery itself would produce daughter strands that are unmethylated and, over time, would lead to passive demethylation. DNMT1 is the proposed maintenance methyltransferase that is responsible for copying DNA methylation patterns to the daughter strands during DNA replication. Mouse models with both copies of DNMT1 deleted are embryonic lethal at approximately day 9, due to the requirement of DNMT1 activity for development in mammalian cells.

It is thought that DNMT3a and DNMT3b are the *de novo* methyltransferases that set up DNA methylation patterns early in development. DNMT3L is a protein that is homologous to the other DNMT3s but has no catalytic activity. Instead, DNMT3L assists the *de novo* methyltransferases by increasing their ability to bind to DNA and stimulating their activity. Finally, DNMT2 (TRDMT1) has been identified as a DNA methyltransferase homolog, containing all 10 sequence motifs common to all DNA methyltransferases; however, DNMT2 (TRDMT1) does not methylate DNA but instead methylates cytosine-38 in the anticodon loop of aspartic acid transfer RNA.

Since many tumor suppressor genes are silenced by DNA methylation during carcinogenesis, there have been attempts to re-express these genes by inhibiting the DNMTs. 5-Aza-2'-deoxycytidine (decitabine) is a nucleoside analog that inhibits DNMTs by trapping them in a covalent complex on DNA by preventing the β -elimination step of catalysis, thus resulting in the enzymes' degradation. However, for decitabine to be active, it must be incorporated into the genome of the cell, which can cause mutations in the daughter cells if the cell does not die. In addition, decitabine is toxic to the bone marrow, which limits the size of its therapeutic window. These pitfalls have led to the development of antisense RNA therapies that target the DNMTs by degrading their mRNAs and preventing their translation. However, it is currently unclear whether targeting DNMT1 alone is sufficient to reactivate tumor suppressor genes silenced by DNA methylation.

In plants

Significant progress has been made in understanding DNA methylation in the model plant *Arabidopsis thaliana*. DNA methylation in plants differs from that of mammals: while DNA methylation in mammals mainly occurs on the cytosine nucleotide in a CpG site, in plants the cytosine can be methylated at CpG, CpHpG, and CpHpH sites, where H represents any nucleotide but guanine. Overall, *Arabidopsis* DNA is highly methylated, i.e. an exemplary mass spectrometry analysis estimated 14% of cytosines to be modified.

The principal *Arabidopsis* DNA methyltransferase enzymes, which transfer and covalently attach methyl groups onto DNA, are DRM2, MET1, and CMT3. Both the DRM2 and MET1 proteins share significant homology to the mammalian methyltransferases DNMT3 and DNMT1, respectively, whereas the CMT3 protein is unique to the plant kingdom. There are currently two classes of DNA methyltransferases: 1) the *de novo* class, or enzymes that create new methylation marks on the DNA; and 2) a maintenance class that recognizes the methylation marks on the parental strand of DNA and transfers new methylation to the daughters strands after DNA replication. DRM2 is the only enzyme that has been implicated as a *de novo* DNA methyltransferase. DRM2 has also been shown, along with MET1 and CMT3 to be involved in maintaining methylation marks through DNA replication. Other DNA methyltransferases are expressed in plants but have no known function (see the Chromatin Database [8]).

It is not clear how the cell determines the locations of *de novo* DNA methylation, but evidence suggests that, for many (though not all) locations, RNA-directed DNA methylation (RdDM) is involved. In RdDM, specific RNA transcripts are produced from a genomic DNA template, and this RNA forms secondary structures called double-stranded RNA molecules. The double-stranded RNAs, through either the small interfering RNA (siRNA) or microRNA (miRNA) pathways direct de-novo DNA methylation of the original genomic location that produced the RNA. This sort of mechanism is thought to be important in cellular defense against RNA viruses and/or transposons, both of which often form a double-stranded RNA that can be mutagenic to the host genome. By methylating their genomic locations, through an as yet poorly understood mechanism, they are shut off and are no longer active in the cell, protecting the genome from their mutagenic effect.

In fungi

Many fungi have low levels (0.1 to 0.5%) of cytosine methylation, whereas other fungi have as much as 5% of the genome methylated. This value seems to vary both among species and among isolates of the same species. There is also evidence that DNA methylation may be involved in state-specific control of gene expression in fungi. Wikipedia:Citation needed However, at a detection limit of 250 attomoles by using ultra-high sensitive mass spectrometry DNA methylation was not confirmed in single cellular yeast species such as *Saccharomyces cerevisiae* or *Schizosaccharomyces pombe*, indicating that yeasts do not possess this DNA modification.

Although brewers' yeast (*Saccharomyces*) and fission yeast (*Schizosaccharomyces*) have no detectable DNA methylation, the model filamentous fungus *Neurospora crassa* has a well-characterized methylation system. Several genes control methylation in *Neurospora* and mutation of the DNA methyl transferase, *dim-2*, eliminates all DNA methylation but does not affect growth or sexual reproduction. While the *Neurospora* genome has very little repeated DNA, half of the methylation occurs in repeated DNA including transposon relics and centromeric DNA. The ability to evaluate other important phenomena in a DNA methylase-deficient genetic background makes *Neurospora* an important system in which to study DNA methylation.

In insects

DNA methylation is debated in insects, *Drosophila melanogaster* for instance seems to possess a very low level of DNA methylation only that is however too low to be studied by methods such as bisulphite sequencing. Takayama et al.^[9] developed a sensitive method that allowed to find that the fly genome DNA sequence patterns that associate with methylation are very different from the patterns seen in humans, or in other animal or plant species to date. Genome methylation in *D. melanogaster* was found at specific short motifs (concentrated in specific 5-base sequence motifs that are CA- and CT-rich but depleted of guanine) and is independent of DNMT2 activity.

Main article: Epigenetics in insects

In bacteria

Adenine or cytosine methylation is part of the restriction modification system of many bacteria, in which specific DNA sequences are methylated periodically throughout the genome. A methylase is the enzyme that recognizes a specific sequence and methylates one of the bases in or near that sequence. Foreign DNAs (which are not methylated in this manner) that are introduced into the cell are degraded by sequence-specific restriction enzymes and cleaved. Bacterial genomic DNA is not recognized by these restriction enzymes. The methylation of native DNA acts as a sort of primitive immune system, allowing the bacteria to protect themselves from infection by bacteriophage.

E. coli DNA adenine methyltransferase (Dam) is an enzyme of ~32 kDa that does not belong to a restriction/modification system. The target recognition sequence for *E. coli* Dam is GATC, as the methylation occurs at the N6 position of the adenine in this sequence (G meATC). The three base pairs flanking each side of this site also influence DNA–Dam binding. Dam plays several key roles in bacterial processes, including mismatch repair, the timing of DNA replication, and gene expression. As a result of DNA replication, the status of GATC sites in the *E. coli* genome changes from fully methylated to hemimethylated. This is because adenine introduced into the new DNA strand is unmethylated. Re-methylation occurs within two to four seconds, during which time replication errors in the new strand are repaired. Methylation, or its absence, is the marker that allows the repair apparatus of the cell to differentiate between the template and nascent strands. It has been shown that altering Dam activity in bacteria results in increased spontaneous mutation rate. Bacterial viability is compromised in dam mutants that also lack certain other DNA repair enzymes, providing further evidence for the role of Dam in DNA repair.

One region of the DNA that keeps its hemimethylated status for longer is the origin of replication, which has an abundance of GATC sites. This is central to the bacterial mechanism for timing DNA replication. SeqA binds to the origin of replication, sequestering it and thus preventing methylation. Because hemimethylated origins of replication are inactive, this mechanism limits DNA replication to once per cell cycle.

Expression of certain genes, for example those coding for pilus expression in *E. coli*, is regulated by the methylation of GATC sites in the promoter region of the gene operon. The cells' environmental conditions just after DNA replication determine whether Dam is blocked from methylating a region proximal to or distal from the promoter region. Once the pattern of methylation has been created, the pilus gene transcription is locked in the on or off position until the DNA is again replicated. In *E. coli*, these pilus operons have important roles in virulence in urinary tract infections. It has been proposedWikipedia:Manual of Style/Words to watch#Unsupported attributions that inhibitors of Dam may function as antibiotics.

On the other hand, DNA cytosine methylase targets CCAGG and CCTGG sites to methylate cytosine at the C5 position (C meC(A/T) GG). The other methylase enzyme, EcoKI, causes methylation of adenines in the sequences AAC(N₆)GTGC and GCAC(N₆)GTT.

Molecular cloning

Most strains used by molecular biologists are derivatives of *E. coli* K-12, and possess both Dam and Dcm, but there are commercially available strains that are dam-/dcm- (lack of activity of either methylase). In fact, it is possible to unmethylate the DNA extracted from dam+/dcm+ strains by transforming it into dam-/dcm- strains. This would help digest sequences that are not being recognized by methylation-sensitive restriction enzymes.

The Restriction enzyme DpnI can recognize 5'-GmeATC-3' sites and digest the methylated DNA. Being such a short motif, it occurs frequently in sequences by chance, and as such its primary use for researchers is to degrade template DNA following PCR reactions (PCR products lack methylation, as no methylases are present in the reaction). Similarly, some commercially available restriction enzymes are sensitive to methylation at their cognate restriction sites, and must as mentioned previously be used on DNA passed through a dam-/dcm- strain to allow cutting.

Detection

DNA methylation can be detected by the following assays currently used in scientific research:

- Mass spectrometry is a very sensitive and reliable analytical method to detect DNA methylation. MS in general is however not informative about the sequence context of the methylation, thus limited in studying the function of this DNA modification.
- Methylation-Specific PCR (MSP), which is based on a chemical reaction of sodium bisulfite with DNA that converts unmethylated cytosines of CpG dinucleotides to uracil or UpG, followed by traditional PCR. However, methylated cytosines will not be converted in this process, and primers are designed to overlap the CpG site of interest, which allows one to determine methylation status as methylated or unmethylated.
- Whole genome bisulfite sequencing, also known as BS-Seq, which is a high-throughput genome-wide analysis of DNA methylation. It is based on aforementioned sodium bisulfite conversion of genomic DNA, which is then sequenced on a Next-generation sequencing platform. The sequences obtained are then re-aligned to the reference genome to determine methylation states of CpG dinucleotides based on mismatches resulting from the conversion of unmethylated cytosines into uracil.
- The HELP assay, which is based on restriction enzymes' differential ability to recognize and cleave methylated and unmethylated CpG DNA sites.
- ChIP-on-chip assays, which is based on the ability of commercially prepared antibodies to bind to DNA methylation-associated proteins like MeCP2.
- Restriction landmark genomic scanning, a complicated and now rarely used assay based upon restriction enzymes' differential recognition of methylated and unmethylated CpG sites; the assay is similar in concept to the HELP assay.
- Methylated DNA immunoprecipitation (MeDIP), analogous to chromatin immunoprecipitation, immunoprecipitation is used to isolate methylated DNA fragments for input into DNA detection methods such as DNA microarrays (MeDIP-chip) or DNA sequencing (MeDIP-seq).
- Pyrosequencing of bisulfite treated DNA. This is sequencing of an amplicon made by a normal forward primer but a biotinylated reverse primer to PCR the gene of choice. The Pyrosequencer then analyses the sample by denaturing the DNA and adding one nucleotide at a time to the mix according to a sequence given by the user. If there is a mis-match, it is recorded and the percentage of DNA for which the mis-match is present is noted. This gives the user a percentage methylation per CpG island.
- Molecular break light assay for DNA adenine methyltransferase activity – an assay that relies on the specificity of the restriction enzyme DpnI for fully methylated (adenine methylation) GATC sites in an oligonucleotide labeled with a fluorophore and quencher. The adenine methyltransferase methylates the oligonucleotide making it a substrate for DpnI. Cutting of the oligonucleotide by DpnI gives rise to a fluorescence increase.
- Methyl Sensitive Southern Blotting is similar to the HELP assay, although uses Southern blotting techniques to probe gene-specific differences in methylation using restriction digests. This technique is used to evaluate local

methylation near the binding site for the probe.

- MethylCpG Binding Proteins (MBPs) and fusion proteins containing just the Methyl Binding Domain (MBD) are used to separate native DNA into methylated and unmethylated fractions. The percentage methylation of individual CpG islands can be determined by quantifying the amount of the target in each fraction.^[10] Extremely sensitive detection can be achieved in FFPE tissues with absorption-based detection.
- High Resolution Melt Analysis (HRM or HRMA), is a post-PCR analytical technique. The target DNA is treated with sodium bisulfite, which chemically converts unmethylated cytosines into uracils, while methylated cytosines are preserved. PCR amplification is then carried out with primers designed to amplify both methylated and unmethylated templates. After this amplification, highly methylated DNA sequences contain a higher number of CpG sites compared to unmethylated templates, which results in a different melting temperature that can be used in quantitative methylation detection.

Differentially methylated regions (DMRs)

Differentially methylated regions (DMRs), are genomic regions with different methylation statuses among multiple samples (tissues, cells, individuals or others), are regarded as possible functional regions involved in gene transcriptional regulation. The identification of DMRs among multiple tissues (T-DMRs) provides a comprehensive survey of epigenetic differences among human tissues. DMRs between cancer and normal samples (C-DMRs) demonstrate the aberrant methylation in cancers. It is well known that DNA methylation is associated with cell differentiation and proliferation. Many DMRs have been found in the development stages (D-DMRs) and in the reprogrammed progress (R-DMRs). In addition, there are intra-individual DMRs (Intra-DMRs) with longitudinal changes in global DNA methylation along with the increase of age in a given individual. There are also inter-individual DMRs (Inter-DMRs) with different methylation patterns among multiple individuals.

QDMR (Quantitative Differentially Methylated Regions) is a quantitative approach to quantify methylation difference and identify DMRs from genome-wide methylation profiles by adapting Shannon entropy (<http://bioinfo.hrbmu.edu.cn/qdmr>). The platform-free and species-free nature of QDMR makes it potentially applicable to various methylation data. This approach provides an effective tool for the high-throughput identification of the functional regions involved in epigenetic regulation. QDMR can be used as an effective tool for the quantification of methylation difference and identification of DMRs across multiple samples.

Gene-set analysis (a.k.a pathway analysis; usually performed tools such as DAVID, GoSeq or GSEA) has been shown to be severely biased when applied to high-throughput methylation data (e.g. MeDIP-seq, MeDIP-ChIP, HELP-seq etc.), and a wide range of studies have thus mistakenly reported hyper-methylation of genes related to development and differentiation; it has been suggested that this can be corrected using sample label permutations or using a statistical model to control for differences in the numbers of CpG probes / CpG sites that target each gene.

Computational prediction

DNA methylation can also be detected by computational models through sophisticated algorithms and methods. Computational models can facilitate the global profiling of DNA methylation across chromosomes, and often such models are faster and cheaper to perform than biological assays. Such up-to-date computational models include Bhasin, *et al.*, Bock, *et al.*, and Zheng, *et al.* Together with biological assay, these methods greatly facilitate the DNA methylation analysis.

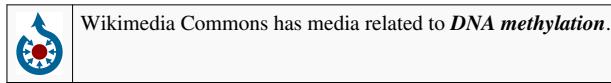
References

- [1] Lund, G.L., Andersson, L., Lauria, M., Lindholm, M., Fraga, M.F., Villar-Garea, A., Ballestar, E., Estellar, M. and Zaina, S. (2004). DNA methylation polymorphisms precede any histological sign of atherosclerosis in mice lacking Apolipoprotein E. *J Biol Chem.* **279**:29147-29154.
- [2] Castro, R., Rivera, I., Struys, E.A., Jansen, E.E., Ravasco, P., Camilo, M.E., Blom, H.J., Jakobs, C. and Tavares de Almeida, T. (2003). Increased homocysteine concentrations and S-adenosylhomocysteine concentrations and DNA hypomethylation in vascular disease. *Clin Chem.* **49** (8): 1292-1296.
- [3] Huang, Y.S., Zhi, Y.F. and Wang, S.R. (2009). Hypermethylation of estrogen receptor- α gene in atheromatosis patients and its correlation with homocysteine. *Pathophysiology.* **16**: 259–265.
- [4] Dong, C.D., Yoon, W. and Goldschmidt-Clermont, P.J. (2002). DNA methylation and atherosclerosis. *J Nutr.* **132** (8): 2406S-2409S.
- [5] Ying, A.K., Hassanain, H.H., Roos, C.M., Smiraglia, D.J., Issa, J.J., Michler, R.E., Caligiuri, M., Plass, C. and Goldschmidt-Clermont, P.J. (2000). Methylation of the estrogen receptor- α gene promoter is selectively increased in proliferating human aortic smooth muscle cells. *Cardiovas Res.* **46**: 172-179.
- [6] Zhu, S., Goldschmidt-Clermont, P.J. and Dong, C. (2005). Inactivation of Monocarboxylate Transporter MCT3 by DNA methylation in atherosclerosis. *Circulation.* **112**:1353-1361.
- [7] Gratchev, Alexei. Review on DNA Methylation. (n.d.) Retrieved from http://www.methods.info/Methods/DNA_methylation/Methylation_review.html
- [8] <http://chromdb.org>
- [9] S. Takayama, J. Dhahbi, A. Roberts, G. Mao, S.-J. Heo, L. Pachter, D. I. K. Martin, D. Boffelli (2014). Genome methylation in *D. melanogaster* is found at specific short motifs and is independent of DNMT2 activity. *Genome Research,*
- [10] ^ David R. McCarthy, Philip D. Cotter, and Michelle M. Hanna (2012). MethylMeter(r): A Quantitative, Sensitive, and Bisulfite-Free Method for Analysis of DNA Methylation, DNA Methylation - From Genomics to Technology, Dr. Tatiana Tatarinova (Ed.), ISBN 978-953-51-0320-2, InTech, DOI: 10.5772/36090. Available from: <http://www.intechopen.com/books/dna-methylation-from-genomics-to-technology/methylmeter-a-quantitative-sensitive-and-bisulfite-free-method-for-analysis-of-dna-methylation>

Further reading

- Law J, Jacobsen SE (2010). "Establishing, maintaining and modifying DNA methylation patterns in plants and animals" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3034103>). *Nat. Rev. Genet.* **11** (3): 204–220. doi: 10.1038/nrg2719 (<http://dx.doi.org/10.1038/nrg2719>). PMC 3034103 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3034103>). PMID 20142834 (<http://www.ncbi.nlm.nih.gov/pubmed/20142834>).
- Straussman R, Nejman D, Roberts D, et al. (2009). "Developmental programming of CpG island methylation profiles in the human genome". *Nat. Struct. Mol. Biol.* **16** (5): 564–571. doi: 10.1038/nsmb.1594 (<http://dx.doi.org/10.1038/nsmb.1594>). PMID 19377480 (<http://www.ncbi.nlm.nih.gov/pubmed/19377480>).
- Patra SK (2008). "Ras regulation of DNA-methylation and cancer". *Exp Cell Res* **314** (6): 1193–1201. doi: 10.1016/j.yexcr.2008.01.012 (<http://dx.doi.org/10.1016/j.yexcr.2008.01.012>). PMID 18282569 (<http://www.ncbi.nlm.nih.gov/pubmed/18282569>).
- Patra SK, Patra A, Ghosh TC, et al. (2008). "Demethylation of (cytosine-5-C-methyl) DNA and regulation of transcription in the epigenetic pathways of cancer development". *Cancer Metast. Rev.* **27** (2): 315–334. doi: 10.1007/s10555-008-9118-y (<http://dx.doi.org/10.1007/s10555-008-9118-y>). PMID 18246412 (<http://www.ncbi.nlm.nih.gov/pubmed/18246412>).

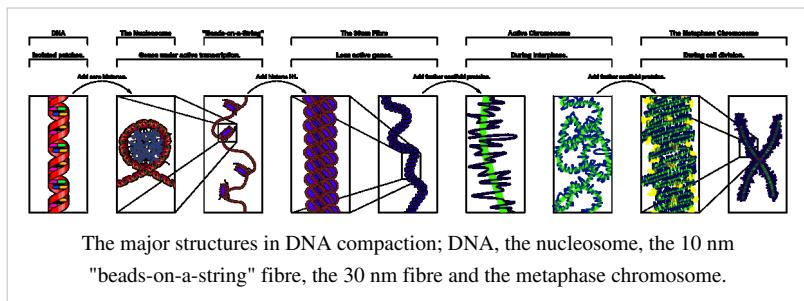
External links



- DNA Methylation (<http://www.nlm.nih.gov/cgi/mesh/2011/MB.cgi?mode=&term=DNA+Methylation>) at the US National Library of Medicine Medical Subject Headings (MeSH)
- ENCODE threads explorer (<http://www.nature.com/encode/#/threads/dna-methylation>) Non-coding RNA characterization. Nature (journal)
- PCMdB (<http://crdd.osdd.net/raghava/pcmdb/>) Pancreatic Cancer Methylation Database. Nature Scientific Report 4:4197 (<http://www.nature.com/srep/2014/140226/srep04197/full/srep04197.html>)

Chromatin

Chromatin is the combination or complex of DNA and proteins that make up the contents of the nucleus of a cell. The primary functions of chromatin are 1) to package DNA into a smaller volume to fit in the cell, 2) to strengthen the DNA to allow mitosis, 3) to prevent DNA damage, and 4) to control gene expression and DNA replication. The primary protein components of chromatin are histones that compact the DNA. Chromatin is only found in eukaryotic cells, (a cell with a defined nucleus). Prokaryotic cells have a very different organization of their DNA, which is referred to as a genophore (a chromosome without chromatin).



The structure of chromatin depends on several factors. The overall structure depends on the stage of the cell cycle. During interphase, the chromatin is structurally loose to allow access to RNA and DNA polymerases that transcribe and replicate the DNA. The local structure of chromatin during interphase depends on the genes present on the DNA: DNA coding genes that are actively transcribed ("turned on") are more loosely packaged and are found associated with RNA polymerases (referred to as euchromatin) while DNA coding inactive genes ("turned off") are found associated with structural proteins and are more tightly packed (heterochromatin). Epigenetic chemical modification of the structural proteins in chromatin also alter the local chromatin structure, in particular chemical modifications of histone proteins by methylation and acetylation. As the cell prepares to divide, i.e. enters mitosis or meiosis, the chromatin packages more tightly to facilitate segregation of the chromosomes during anaphase. During this stage of the cell cycle this makes the individual chromosomes in many cells visible by optical microscope.

In general terms, there are three levels of chromatin organization:

1. DNA wraps around histone proteins forming nucleosomes; the "beads on a string" structure (euchromatin).
2. Multiple histones wrap into a 30 nm fibre consisting of nucleosome arrays in their most compact form (heterochromatin). (Definitively established to exist in vitro, the 30-nanometer fibre was not seen in recent X-ray studies of human mitotic chromosomes.)
3. Higher-level DNA packaging of the 30 nm fibre into the metaphase chromosome (during mitosis and meiosis).

There are, however, many cells that do not follow this organisation. For example, spermatozoa and avian red blood cells have more tightly packed chromatin than most eukaryotic cells, and trypanosomatid protozoa do not condense their chromatin into visible chromosomes for mitosis.

During interphase

The structure of chromatin during interphase is optimized to allow easy access of transcription and DNA repair factors to the DNA while compacting the DNA into the nucleus. The structure varies depending on the access required to the DNA. Genes that require regular access by RNA polymerase require the looser structure provided by euchromatin.

Change in structure

Chromatin undergoes various forms of change in its structure. Histone proteins, the foundation blocks of chromatin, are modified by various post-translational modification to alter DNA packing. Acetylation results in the loosening of chromatin and lends itself to replication and transcription. When certain residues are methylated, they hold DNA together strongly and restrict access to various enzymes. A recent study showed that there is a bivalent structure present in the chromatin: methylated lysine residues at location 4 and 27 on histone 3. It is thought that this may be involved in development; there is more methylation of lysine 27 in embryonic cells than in differentiated cells, whereas lysine 4 methylation positively regulates transcription by recruiting nucleosome remodeling enzymes and histone acetylases.

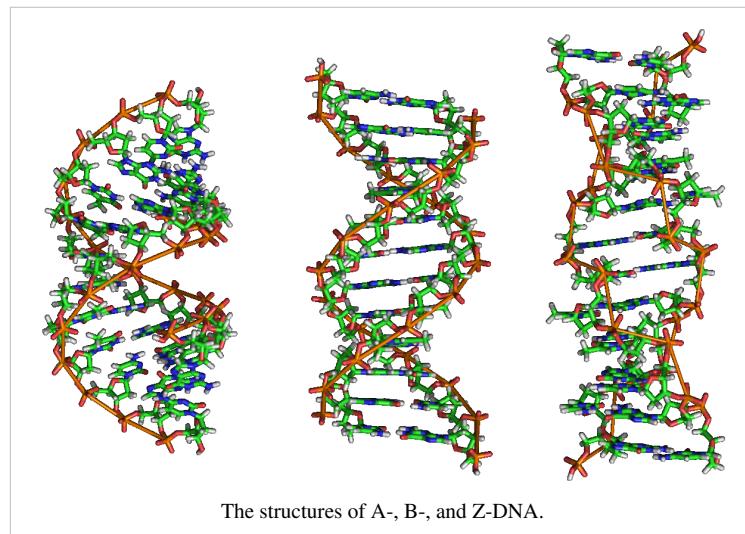
Polycomb-group proteins play a role in regulating genes through modulation of chromatin structure.

For additional information, see Histone modifications in chromatin regulation and RNA polymerase control by chromatin structure.

DNA structure

Main articles: Mechanical properties of DNA and Z-DNA

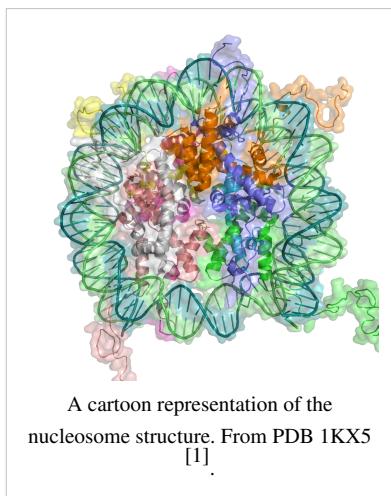
The vast majority of DNA within the cell is the normal DNA structure. However, in nature, DNA can form three structures, A-, B-, and Z-DNA. A and B chromosomes are very similar, forming right-handed helices, whereas Z-DNA is a more unusual left-handed helix with a zig-zag phosphate backbone. Z-DNA is thought to play a specific role in chromatin structure and transcription because of the properties of the junction between B- and Z-DNA.



At the junction of B- and Z-DNA, one pair of bases is flipped out from normal bonding. These play a dual role of a site of recognition by many proteins and as a sink for torsional stress from RNA polymerase or nucleosome binding.

The nucleosome and "beads-on-a-string"

Main articles: Nucleosome, Chromatosome and Histone



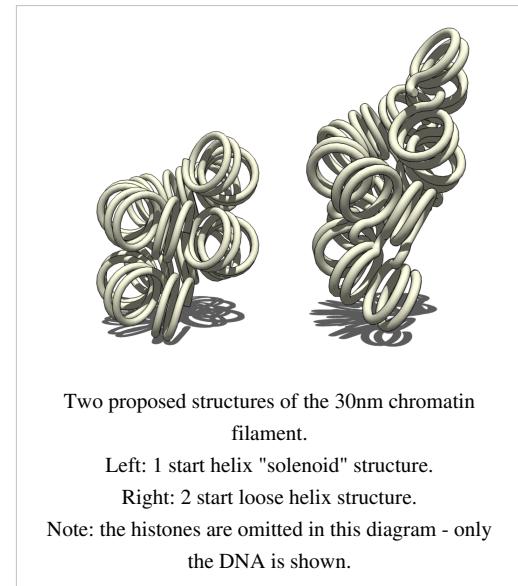
The basic repeat element of chromatin is the nucleosome, interconnected by sections of linker DNA, a far shorter arrangement than pure DNA in solution. In addition to the core histones, there is the linker histone, H1, which contacts the exit/entry of the DNA strand on the nucleosome. The nucleosome core particle, together with histone H1, is known as a chromatosome. Nucleosomes, with about 20 to 60 base pairs of linker DNA, can form, under non-physiological conditions, an approximately 10 nm "beads-on-a-string" fibre. (Fig. 1-2).

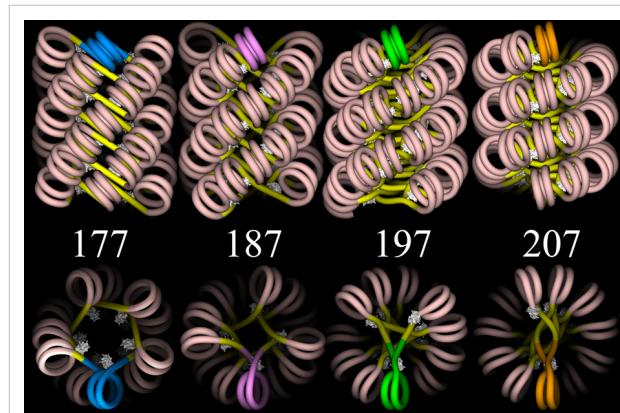
The nucleosomes bind DNA non-specifically, as required by their function in general DNA packaging. There are, however, large DNA sequence preferences that govern nucleosome positioning. This is due primarily to the varying physical properties of different DNA sequences: For instance, adenine and thymine are more favorably compressed into the inner minor grooves. This means nucleosomes can bind preferentially at one position approximately every 10 base pairs (the helical repeat of DNA)- where the DNA is rotated to maximise the number of A and T bases that will lie in the inner minor groove. (See mechanical properties of DNA.)

30-nanometer chromatin fibre

With addition of H1, the "beads-on-a-string" structure in turn coils into a 30 nm diameter helical structure known as the 30 nm fibre or filament. The precise structure of the chromatin fibre in the cell is not known in detail, and there is still some debate over this. Wikipedia:Citation needed

This level of chromatin structure is thought to be the form of euchromatin, which contains actively transcribed genes. EM studies have demonstrated that the 30 nm fibre is highly dynamic such that it unfolds into a 10 nm fiber ("beads-on-a-string") structure when transversed by an RNA polymerase engaged in transcription.





Four proposed structures of the 30 nm chromatin filament for DNA repeat length per nucleosome ranging from 177 to 207 bp.

Linker DNA in yellow and nucleosomal DNA in pink.

The existing models commonly accept that the nucleosomes lie perpendicular to the axis of the fibre, with linker histones arranged internally. A stable 30 nm fibre relies on the regular positioning of nucleosomes along DNA. Linker DNA is relatively resistant to bending and rotation. This makes the length of linker DNA critical to the stability of the fibre, requiring nucleosomes to be separated by lengths that permit rotation and folding into the required orientation without excessive stress to the DNA. In this view, different length of the linker DNA should produce different folding topologies of the chromatin fiber. Recent theoretical work, based on electron-microscopy images of reconstituted fibers support this view.

Spatial organization of chromatin in the cell nucleus

The layout of the genome within the nucleus is not random - specific regions of the genome have a tendency to be found in certain spaces. Specific regions of the chromatin are enriched at the nuclear membrane, while other regions are bound together by protein complexes. The layout of this is not, however, well-characterised apart from the compaction of one of the two X chromosomes in mammalian females into the Barr body. This serves the role of permanently deactivating these genes, which prevents females from getting a 'double dose' relative to males. The extent to which the inactive X is actually compacted is a matter of some controversy.

Chromatin and bursts of transcription

Chromatin and its interaction with enzymes has been researched, and a conclusion being made is that it is relevant and an important factor in gene expression. Vincent G. Allfrey, a professor at Rockefeller University, stated that RNA synthesis is related to histone acetylation. The lysine amino acid attached to the end of the histones is positively charged. The acetylation of these tails would make the chromatin ends neutral, allowing for DNA access.

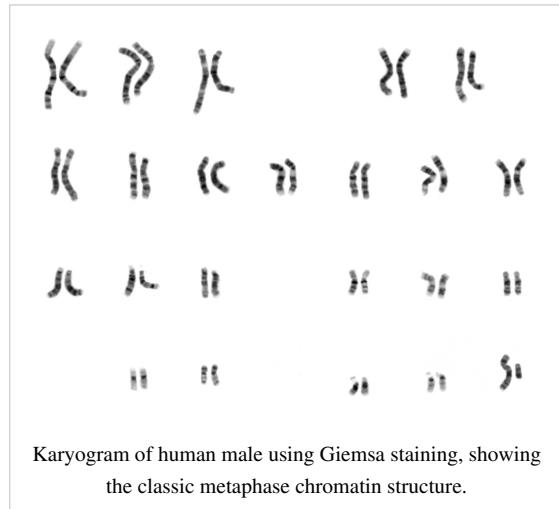
When the chromatin decondenses, the DNA is open to entry of molecular machinery. Fluctuations between open and closed chromatin may contribute to the discontinuity of transcription, or transcriptional bursting. Other factors are probably involved, such as the association and dissociation of transcription factor complexes with chromatin. The phenomenon, as opposed to simple probabilistic models of transcription, can account for the high variability in gene expression occurring between cells in isogenic populations^[2]

Metaphase chromatin

The metaphase structure of chromatin differs vastly to that of interphase. It is optimised for physical strength and manageability, forming the classic chromosome structure seen in karyotypes. The structure of the condensed chromosome is thought to be loops of 30 nm fibre to a central scaffold of proteins. It is, however, not well-characterised.

The physical strength of chromatin is vital for this stage of division to prevent shear damage to the DNA as the daughter chromosomes are separated. To maximise strength the composition of the chromatin changes as it approaches the centromere, primarily through alternative histone H1 analogues.

It should also be noted that, during mitosis, while most of the chromatin is tightly compacted, there are small regions that are not as tightly compacted. These regions often correspond to promoter regions of genes that were active in that cell type prior to entry into mitosis. The lack of compaction of these regions is called bookmarking, which is an epigenetic mechanism believed to be important for transmitting to daughter cells the "memory" of which genes were active prior to entry into mitosis. This bookmarking mechanism is needed to help transmit this memory because transcription ceases during mitosis.



Karyogram of human male using Giemsa staining, showing the classic metaphase chromatin structure.

Chromatin: alternative definitions

- Simple and concise definition:** Chromatin is DNA plus the proteins (and RNA) that package DNA within the cell nucleus.
- A biochemists' operational definition:** Chromatin is the DNA/protein/RNA complex extracted from eukaryotic lysed interphase nuclei. Just which of the multitudinous substances present in a nucleus will constitute a part of the extracted material partly depends on the technique each researcher uses. Furthermore, the composition and properties of chromatin vary from one cell type to the another, during development of a specific cell type, and at different stages in the cell cycle.
- The DNA + histone = chromatin definition:** The DNA double helix in the cell nucleus is packaged by special proteins termed histones. The formed protein/DNA complex is called chromatin. The basic structural unit of chromatin is the nucleosome.

Alternative chromatin organizations

During metazoan spermiogenesis, the spermatid's chromatin is remodelled into a more spaced-packaged, widened, almost crystal-like structure. This process is associated with the cessation of transcription and involves nuclear protein exchange. The histones are mostly displaced, and replaced by protamines (small, arginine-rich proteins).

Nobel Prizes

The following scientists were recognized for their contributions to chromatin research with Nobel Prizes:

| Year | Who | Award |
|------|----------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1910 | Albrecht Kossel (University of Heidelberg) | Nobel Prize in Physiology or Medicine for his discovery of the five nuclear bases: adenine, cytosine, guanine, thymine, and uracil. |
| 1933 | Thomas Hunt Morgan (California Institute of Technology) | Nobel Prize in Physiology or Medicine for his discoveries of the role played by the gene and chromosome in heredity, based on his studies of the white-eyed mutation in the fruit fly <i>Drosophila</i> . ^[3] |
| 1962 | Francis Crick, James Watson and Maurice Wilkins (MRC Laboratory of Molecular Biology, Harvard University and London University respectively) | Nobel Prize in Physiology or Medicine for their discoveries of the double helix structure of DNA and its significance for information transfer in living material. |
| 1982 | Aaron Klug (MRC Laboratory of Molecular Biology) | Nobel Prize in Chemistry "for his development of crystallographic electron microscopy and his structural elucidation of biologically important nucleic acid-protein complexes" |
| 1993 | Richard J. Roberts and Phillip A. Sharp | Nobel Prize in Physiology "for their independent discoveries of split genes," in which DNA sections called exons express proteins, and are interrupted by DNA sections called introns, which do not express proteins. |
| 2006 | Roger Kornberg (Stanford University) | Nobel Prize in Chemistry for his discovery of the mechanism by which DNA is transcribed into messenger RNA. |

References

- [1] <http://www.rcsb.org/pdb/explore/explore.do?structureId=1KX5>
- [2] *Gatekeepers of chromatin: Small metabolites elicit big changes in gene expression, Kaochar, Salma; Tu, Benjamin P. Trends in biochemical sciences doi:10.1016/j.tibs.2012.07.008 (volume 37 issue 11 pp.477 - 483)
- [3] "Thomas Hunt Morgan and His Legacy". (http://www.nobelprize.org/nobel_prizes/medicine/laureates/1933/morgan-article.html) Nobelprize.org. 7 Sep 2012

Other references

- Cooper, Geoffrey M. 2000. The Cell, 2nd edition, A Molecular Approach. Chapter 4.2 Chromosomes and Chromatin. (<http://www.ncbi.nlm.nih.gov/bookshelf/br.fcgi?book=cooper&part=A618#A620>)
- Corces, V. G. 1995. Chromatin insulators. Keeping enhancers under control. Nature 376:462-463.
- Cremer, T. 1985. Von der Zellenlehre zur Chromosomentheorie: Naturwissenschaftliche Erkenntnis und Theorienwechsel in der frühen Zell- und Vererbungsforschung, Veröffentlichungen aus der Forschungsstelle für Theoretische Pathologie der Heidelberger Akademie der Wissenschaften. Springer-Vlg., Berlin, Heidelberg.
- Elgin, S. C. R. (ed.). 1995. Chromatin Structure and Gene Expression, vol. 9. IRL Press, Oxford, New York, Tokyo.
- Gerasimova, T. I., and V. G. Corces. 1996. Boundary and insulator elements in chromosomes. Current Op. Genet. and Dev. 6:185-192.
- Gerasimova, T. I., and V. G. Corces. 1998. Polycomb and Trithorax group proteins mediate the function of a chromatin insulator. Cell 92:511-521.
- Gerasimova, T. I., and V. G. Corces. 2001. CHROMATIN INSULATORS AND BOUNDARIES: Effects on Transcription and Nuclear Organization. Annu Rev Genet 35:193-208.
- Gerasimova, T. I., K. Byrd, and V. G. Corces. 2000. A chromatin insulator determines the nuclear localization of DNA [In Process Citation]. Mol Cell 6:1025-35.
- Ha, S. C., K. Lowenhaupt, A. Rich, Y. G. Kim, and K. K. Kim. 2005. Crystal structure of a junction between B-DNA and Z-DNA reveals two extruded bases. Nature 437:1183-6.
- Pollard, T., and W. Earnshaw. 2002. Cell Biology. Saunders.

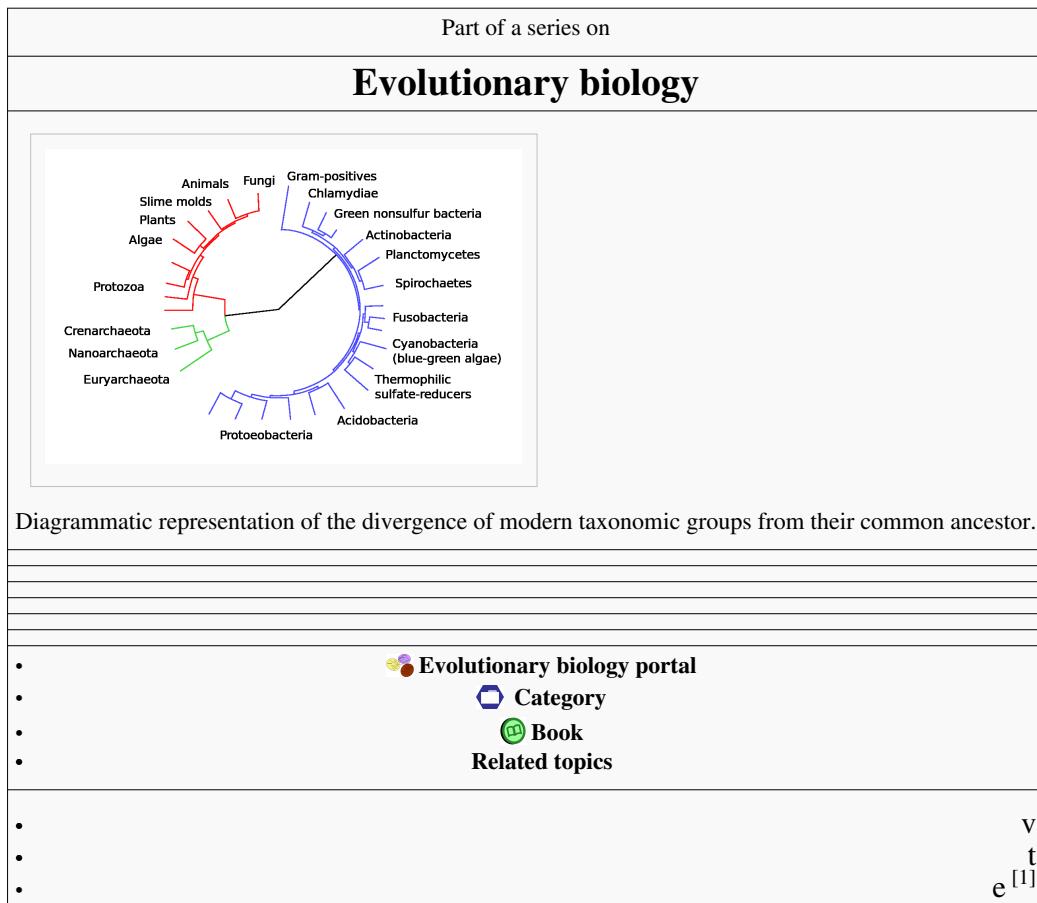
- Saumweber, H. 1987. Arrangement of Chromosomes in Interphase Cell Nuclei, p. 223-234. In W. Hennig (ed.), Structure and Function of Eucaryotic Chromosomes, vol. 14. Springer-Verlag, Berlin, Heidelberg.
- Sinden, R. R. 2005. Molecular biology: DNA twists and flips. Nature 437:1097-8.
- Van Holde KE. 1989. Chromatin. New York: Springer-Verlag. ISBN 0-387-96694-3.
- Van Holde, K., J. Zlatanova, G. Arents, and E. Moudrianakis. 1995. Elements of chromatin structure: histones, nucleosomes, and fibres, p. 1-26. In S. C. R. Elgin (ed.), Chromatin structure and gene expression. IRL Press at Oxford University Press, Oxford.

External links

- Chromatin, Histones & Cathepsin (<http://www.youtube.com/watch?v=eYrQ0EhVCYA>); PMAP The Proteolysis Map-animation
- Recent chromatin publications and news (<http://www.chromatin.co.uk>)
- Protocol for *in vitro* Chromatin Assembly (<http://www.activemotif.com/documents/134.pdf>)
- ENCODE threads Explorer (<http://www.nature.com/encode/#/threads/chromatin-patterns-at-transcription-factor-binding-sites>) Chromatin patterns at transcription factor binding sites. Nature (journal)

Saltation (biology)

For other uses, see Saltation (disambiguation).



In biology, **saltation** (from Latin, *saltus*, "leap") is a sudden change from one generation to the next, that is large, or very large, in comparison with the usual variation of an organism. The term is used for nongradual changes

(especially single-step speciation) that are atypical of, or violate gradualism - involved in modern evolutionary theory.

History

Prior to Charles Darwin most evolutionary scientists had been saltationists.^[1] Jean-Baptiste Lamarck was a gradualist but similar to other scientists of the period had written that saltational evolution was possible. Étienne Geoffroy Saint-Hilaire endorsed a theory of saltational evolution that "monstrosities could become the founding fathers (or mothers) of new species by instantaneous transition from one form to the next."^[2] Geoffroy wrote that environmental pressures could produce sudden transformations to establish new species instantaneously.^[3] In 1864 Albert von Kölliker revived Geoffroy's theory that evolution proceeds by large steps, under the name of heterogenesis.^[4]

With the publication of *On the Origin of Species* in 1859 Charles Darwin had denied saltational evolution by writing that evolutionary transformation always proceeds gradually and never in jumps. Darwin insisted on slow accumulation of small steps in evolution and wrote "natural selection acts solely by accumulating slight successive favourable variations, it can produce no great or sudden modification; it can act only by very short steps".^[5]

From 1860 to 1880 saltation had a minority interest but by 1890 had become a major interest to scientists.^[6] In their paper on evolutionary theories in the 20th century (Levit *et al.* 2008) wrote;

The advocates of saltationism deny the Darwinian idea of slowly and gradually growing divergence of character as the only source of evolutionary progress. They would not necessarily completely deny gradual variation, but claim that cardinally new 'body plans' come into being as a result of saltations (sudden, discontinuous and crucial changes, for example, the series of macromutations). The latter are responsible for the sudden appearance of new higher taxa including classes and orders, while small variation is supposed to be responsible for the fine adaptations below the species level.^[7]

In the early 20th century a mechanism of saltation was proposed as large mutations. It was seen as a much faster alternative to the Darwinian concept of a gradual process of small random variations being acted on by natural selection. It was popular with early geneticists such as Hugo de Vries, who along with Carl Correns helped rediscover Gregor Mendel's laws of inheritance in 1900, William Bateson, a British zoologist who switched to genetics, and early in his career Thomas Hunt Morgan. Some of these geneticists developed it into the mutation theory of evolution. There was also a debate over accounts of the evolution of mimicry and if they could be explained by gradualism or saltation. The geneticist Reginald Punnett supported a saltational theory in his book *Mimicry in Butterflies* (1915).^[8]

The mutation theory of evolution held that species went through periods of rapid mutation, possibly as a result of environmental stress, that could produce multiple mutations, and in some cases completely new species, in a single generation. This mutationist view of evolution was later replaced by the reconciliation of Mendelian genetics with natural selection into a gradualistic framework for the neo-Darwinian synthesis.^[9] It was the emergence of population thinking in evolution which forced many scientists to adopt gradualism in the early 20th century. According to Ernst Mayr, it wasn't until the development of population genetics in the neo-Darwinian synthesis in the 1940s that demonstrated the explanatory power of natural selection that saltational views of evolution were largely abandoned.^[10]

Saltation was originally denied by the "modern synthesis" school of neo-Darwinism which favoured gradual evolution but has since been accepted due to recent evidence in evolutionary biology (see the current status section).^{[11][12][13][14]} In recent years there are some prominent proponents of saltation, including Carl Woese. Woese, and colleagues, suggested that the absence of RNA signature continuum between domains of bacteria, archaea, and eukarya constitutes a primary indication that the three primary organismal lineages materialized via one or more major evolutionary saltations from some universal ancestral state involving dramatic change in cellular organization that was significant early in the evolution of life, but in complex organisms gave way to the generally

accepted Darwinian mechanisms.^[15] The geneticist Barbara McClintock introduced the idea of "jumping genes", chromosome transpositions that can produce rapid changes in the genome.^[16]

Saltational speciation, also known as abrupt speciation, is the discontinuity in a lineage that occurs through genetic mutations, chromosomal aberrations or other evolutionary mechanisms that cause reproductively isolated individuals to establish a new species population. Polyploidy, karyotypic fission, symbiogenesis and lateral gene transfer are possible mechanisms for saltational speciation.^[17]

Confusion with punctuated equilibrium

It is a popular misconception that punctuated equilibrium is a saltationist theory, often mistaken for Richard Goldschmidt's hypothesis of "Hopeful Monsters." However, punctuated equilibrium refers instead to a pattern of evolution where most speciation occurs relatively rapidly from a geological perspective (tens of thousands of years instead of millions of years), but through neo-Darwinian evolution, not by saltations.

Stephen Jay Gould

In 1977 Stephen Jay Gould argued that the recent discovery of regulatory genes offered new evidence which supported some of Goldschmidt's postulates. Gould argued that instances of rapid evolution neither undermine Darwinian theory (as Goldschmidt believed) nor await immediate discreditation (as many neo-Darwinians thought).^[18] Gould insisted that Darwin's belief in gradualism—which was largely inherited from the anti-catastrophic views of Charles Lyell—was never an essential component to Darwin's theory of evolution. Thomas Henry Huxley also warned Darwin that he had loaded his work "with an unnecessary difficulty in adopting *Natura non facit saltum* so unreservedly."^[19] Huxley feared this assumption could discourage naturalists who believed that major leaps and cataclysms played a significant role in the history of life. Gould continued:

As a Darwinian, I wish to defend Goldschmidt's postulate that macroevolution is not simply microevolution extrapolated, and that major structural transitions can occur rapidly without a smooth series of intermediate stages. . . . In his infamous book of 1940, Goldschmidt specifically invokes rate genes as a potential maker of hopeful monsters: 'This basis is furnished by the existence of mutants producing monstrosities of the required type and the knowledge of embryonic determination, which permits a small rate change in early embryonic processes to produce a large effect embodying considerable parts of the organism.' In my own, strongly biased opinion, the problem of reconciling evident discontinuity in macroevolution with Darwinism is largely solved by the observation that small changes early in embryology accumulate through growth to yield profound differences among adults.

Nevertheless, Gould argued that Goldschmidt's "hopeful monster" concept was incorrect:

The developmental theme of the 'hopeful monster' (despite its inappropriate name, virtually guaranteed to inspire ridicule and opposition), based on the important concept of 'rate genes,' came first in Goldschmidt's thought, and always occupied more of his attention and research. Unfortunately, he bound this interesting challenge from development, a partially valid concept that could have been incorporated into a Darwinian framework as an auxiliary hypothesis (and now has been accepted, to a large extent, if under different names), to his truly oppositional and ultimately incorrect theory of systemic mutation, therefore winning anathema for his entire system. Goldschmidt may have acted as the architect of his own undoing, but much of his work should evoke sympathetic attention today.^[20]

Macromutation theory

The German geneticist Richard Goldschmidt was the first scientist to use the term "hopeful monster". Goldschmidt thought that small gradual changes could not bridge the hypothetical divide between microevolution and macroevolution. In his book *The Material Basis of Evolution* (1940) he wrote "the change from species to species is not a change involving more and more additional atomistic changes, but a complete change of the primary pattern or reaction system into a new one, which afterwards may again produce intraspecific variation by micromutation." Goldschmidt believed the large changes in evolution were caused by macromutations (large mutations). His ideas about macromutations became known as the hopeful monster hypothesis which is considered a type of saltational evolution.^[21]

Goldschmidt's thesis however was universally rejected and widely ridiculed within the biological community, which favored the neo-Darwinian explanations of R.A. Fisher, J. B. S. Haldane and Sewall Wright.^[22] However there has been a recent interest in the ideas of Goldschmidt in the field of evolutionary developmental biology as some scientists are convinced he was not entirely wrong.^[23]

Otto Schindewolf, a German paleontologist, also supported macromutations as part of his evolutionary theory. He was known for presenting an alternative interpretation of the fossil record based on his ideas of orthogenesis, saltational evolution and extraterrestrial impacts opposed to gradualism but abandoned the view of macromutations in later publications.^[24]

Søren Løvstrup, a biochemist and embryologist from Denmark, advocated a similar hypothesis of macromutation to Goldschmidt's in 1974.^[25] Lovstrup believed that macromutations interfered with various epigenetic processes, that is, those which affect the causal processes in biological development. This is in contrast to the gradualistic theory of micromutations of Neo-Darwinism, which claims that evolutionary innovations are generally the result of accumulation of numerous very slight modifications. Lovstrup also rejected the punctuated equilibria of Stephen Gould and Niles Eldredge, claiming it was a form of gradualism and not a macromutation theory. Lovstrup defended many of Darwin's critics including Schindewolf, Mivart, Goldschmidt, and Himmelfarb.^[26] Mae Wan Ho described Lovstrup's theory as similar to the hopeful monster theory of Richard Goldschmidt.

Goldschmidt presented two mechanisms for how hopeful monsters might work. One mechanism, involved "systemic mutations", rejected the classical gene concept and is no longer considered by modern science; however, his second mechanism involved "developmental macromutations" in "rate genes" or "controlling genes" that change early development and thus cause large effects in the adult phenotype. These kind of mutations are similar to the ones considered in contemporary evolutionary developmental biology.^[27]

On the subject of Goldschmidt Donald Prothero in his book *Evolution: What the Fossils Say and Why It Matters* (2007) wrote:

The past twenty years have vindicated Goldschmidt to some degree. With the discovery of the importance of regulatory genes, we realize that he was ahead of his time in focusing on the importance of a few genes controlling big changes in the organisms, not small-scales changes in the entire genome as neo-Darwinians thought. In addition, the hopeful monster problem is not so insurmountable after all. Embryology has shown that if you affect an entire population of developing embryos with a stress (such as a heat shock) it can cause many embryos to go through the same new pathway of embryonic development, and then they all become hopeful monsters when they reach reproductive age.^[28]

In 2008 evolutionary biologist Olivia Judson in her article *The Monster Is Back, and It's Hopeful* listed some examples which may support the hopeful monster hypothesis^[29] and an article published in the journal Nature in 2010 titled *Evolution: Revenge of the Hopeful Monster* reported that studies in stickleback populations in a British Columbia lake and bacteria populations in a Michigan lab have shown that large individual genetic changes can have vast effects on organisms "without dooming it to the evolutionary rubbish heap". According to the article "Single-gene changes that confer a large adaptive value do happen: they are not rare, they are not doomed and, when

competing with small-effect mutations, they tend to win. But small-effect mutations still matter — a lot. They provide essential fine-tuning and sometimes pave the way for explosive evolution to follow.^[30]

A paper by (Page *et al.* 2010) have written that the Mexican axolotl (*Ambystoma mexicanum*) could be classified as a hopeful monster as it exhibits an adaptive and derived mode of development that has evolved rapidly and independently among tiger salamanders. According to the paper there has been an interest in aspects of the hopeful monster hypothesis in recent years:

Goldschmidt proposed that mutations occasionally yield individuals within populations that deviate radically from the norm and referred to such individuals as "hopeful monsters". If the novel phenotypes of hopeful monsters arise under the right environmental circumstances, they may become fixed, and the population will found a new species. While this idea was discounted during the Modern synthesis, aspects of the hopeful monster hypothesis have been substantiated in recent years. For example, it is clear that dramatic changes in phenotype can occur from few mutations of key developmental genes and phenotypic differences among species often map to relatively few genetic factors. These findings are motivating renewed interest in the study of hopeful monsters and the perspectives they can provide about the evolution of development. In contrast to mutants that are created in the lab, hopeful monsters have been shaped by natural selection and are therefore more likely to reveal mechanisms of adaptive evolution.^[31]

Guenter Theissen, a professor of genetics, has classified homeotic mutants as hopeful monsters and documented in his scientific publications (Theissen, 2005 and Theissen *et al.* 2006) many examples for animal and plant lineages that may have originated as hopeful monsters.^{[32][33]} American biologist Michael Freeling has proposed "balanced gene drive" as a saltational mechanism in the mutationist tradition, which could explain trends involving morphological complexity in plant and animal eukaryotic lineages.^[34]

Current status

Examples of saltational evolution include cases of stabilized hybrids that can reproduce without crossing (such as allotetraploids) and cases of symbiogenesis. Both gene duplication and lateral gene transfer have the capacity to bring about relatively large changes that are saltational.^[35] Evidence of phenotypic saltation has been found in the centipede^[36] and some scientists have suggested there is evidence for independent instances of saltational evolution in Sphinx moths.^[37] Saltational changes have occurred in the buccal cavity of the roundworm *Caenorhabditis elegans*.^[38] Some processes of epigenetic inheritance can also produce changes that are saltational.^[39] There has been a controversy over if mimicry in butterflies and other insects can be explained by gradual or saltational evolution.^[40] According to (Norrstrom *et al.* 2007) there is evidence for saltation in some cases of mimicry.^[41] The endosymbiotic theory is considered to be a type of saltational evolution.^[42] Symonds and Elgar, 2004 have suggested that pheromone evolution in bark beetles is characterized by large saltational shifts.^[43] The mode of evolution of sex pheromones in *Bactrocera* has occurred by rapid saltational changes associated with speciation followed by gradual divergence thereafter.^[44]

Saltational speciation has been recognized in the genus *Clarkia* (Lewis, 1966).^[45] It has been suggested (Carr, 1980, 2000) that the *Calycadenia pauciflora* could have originated directly from an ancestral race through a single saltational event involving multiple chromosome breaks.^[46] Specific cases of homeosis in flowers can be caused by saltational evolution. In a study of divergent orchid flowers (Bateman and DiMichele, 2002) wrote how simple homeotic morphs in a population can lead to newly established forms that become fixed and ultimately lead to new species.^[47] They described the transformation as a saltational evolutionary process, where a mutation of key developmental genes leads to a profound phenotypic change, producing a new evolutionary lineage within a species.^[48] Polyploidy (most common in plants but not unknown in animals) is considered a type of saltation. Polyploidy meets the basic criteria of saltation in that a significant change (in gene numbers) results in speciation in just one generation. Mammalian liver cells are typically polyploidal, but they are not part of the germ line.

Use by creationists

Some creationists have associated Goldschmidt's "hopeful monsters" with the theory of punctuated equilibrium, as proposed by Eldredge and Gould.^[49] Punctuated equilibrium differs from hopeful monsters in that the former acts on populations rather than individuals, is theoretically more gradual (which proposes to take 50,000 to 100,000 years), functions by the evolution of reproductive isolation (through mechanisms such as allopatric speciation), and the latter says nothing of stasis. Creationists such as Luther Sutherland claim that both theories inadvertently appeal to the absence of fossil evidence for evolution and thereby undermining the theory of Darwinian evolution. This predicament is used by creationists to argue that "there are no transitional fossils." Paleontologists such as Niles Eldredge, Stephen Jay Gould, and Steven M. Stanley avoid this by explaining that transitional forms may be rare between species, but "they are abundant between larger groups",^[50] and none of these paleontologists support Goldschmidt's "hopeful monster" hypothesis. Steven M. Stanley argued that some of Goldschmidt's views err mainly in exaggerating the importance of "chromosomal rearrangements" leading to "rapid changes in growth gradients or developmental sequences, and on what we now call quantum speciation."^[51]

References

- [1] Henry Fairfield Osborn. (1894). *From the Greeks to Darwin: An outline of the development of the evolution idea*. New York, London, Macmillan and Co.
- [2] Benedikt Hallgrímsson, Brian K. Hall. (2011). *Variation: A Central Concept in Biology*. Academic Press. p. 18
- [3] Peter J. Bowler. (2003). *Evolution: The History of an Idea*. University of California Press. p. 127
- [4] Sewall Wright. (1984). *Evolution and the Genetics of Populations: Genetics and Biometric Foundations Volume 1*. University of Chicago Press. p. 10
- [5] Charles Darwin. (1859). *On the Origin of Species*. p. 471
- [6] Gregory Radick. (2008). *The Simian Tongue: The Long Debate about Animal Language*. University Of Chicago Press. p. 368
- [7] Levit, G. S., Meister, K., Hößfeld, U. (2008). *Alternative Evolutionary Theories: A Historical Survey*. Journal of Bioeconomics 10.1. pp. 71–96.
- [8] Reginald Punnett. (1915). *Mimicry in Butterflies* (<http://archive.org/stream/cu31924003093162#page/n7/mode/2up>). Cornell University Library.
- [9] Peter J. Bowler. (2003). *Evolution: The History of an Idea*. University of California Press.
- [10] Ernst Mayr. (2007). *What Makes Biology Unique?: Considerations on the Autonomy of a Scientific Discipline*. Cambridge University Press; 1 edition
- [11] Bateman, R. M. and DiMichele, W. A. (1994). *Saltational evolution of form in vascular plants: a neoGoldschmidtian synthesis*. In *Shape and Form in Plants and Fungi* (eds D. S. Ingram and A. Hudson), Academic Press, London. pp. 61-100.
- [12] Gregory, T. R. and Hebert, P. D. N. (1999). *The modulation of DNA content: proximate causes and ultimate consequences* (<http://genome.cshlp.org/content/9/4/317.full>). Genome Res. 9, 317–324.
- [13] Eva Jablonka and Marion J. Lamb. (2005). *Evolution in Four Dimensions: Genetic, Epigenetic, Behavioral, and Symbolic Variation in the History of Life*. A Bradford Book. ISBN 0262600692
- [14] Serres MH, Kerr AR, McCormack TJ, Riley M. (2009). *Evolution by leaps: gene duplication in bacteria* (<http://www.biologydirect.com/content/4/1/46>). Biology Direct 4: 46.
- [15] Roberts, E., A. Sethi, J. Montoya, C.R. Woese and Z. Luthey-Schulten. (2008). *Molecular signatures of ribosomal evolution* (<http://www.pnas.org/content/105/37/13953.full?sid=f3651397-00e9-4a57-802b-f41c6ef6cf5a>). Proc. Natl. Acad. Sci. USA, 105: 13953–13958.
- [16] McClintock, B. (1984). *The significance of responses of the genome to challenge*. Science Vol. 226, pp. 792-801.
- [17] Oladele Ogunseitan. (2004). *Microbial Diversity: Form and Function in Prokaryotes*. Wiley-Blackwell. p. 13. ISBN 978-0632047086
- [18] Gould, S. J. (1977). "The Return of Hopeful Monsters." (http://www.stephenjaygould.org/library/gould_hopeful-monsters.html) *Natural History* 86 (June/July): 24, 30.
- [19] Thomas Henry Huxley. (1859). Letter to Charles Darwin. (<http://www.darwinproject.ac.uk/darwinletters/calendar/entry-2544.html>) Nov. 23, 1859.
- [20] Gould, S. J. (2002). *The Structure of Evolutionary Theory*. Cambridge, MA: Harvard Univ. Press, p. 68. (<http://books.google.com/books?id=nhII7e61WOUC&pg=PP100>)
- [21] Verne Grant. (1963). *The Origin of Adaptations*. Columbia University Press.
- [22] Gould, S. J. (1982). "The uses of heresy: an introduction to Richard Goldschmidt's *The Material Basis of Evolution*." pp. xiii-xlii (<http://books.google.com/books?id=kAPLvAnp7KAC&lpg=PP1&pg=PR13>). New Haven: Yale University Press.
- [23] Scott F. Gilbert. (2000). *Developmental Biology* Sinauer Associates; 6th edition. ISBN 0878932437
- [24] Otto Schindewolf. (1969). *Über den "Typus" in morphologischer und phylogenetischer Biologie*. Mainz: Akademie der Wissenschaften und der Literatur.

- [25] Kathryn E. Hood, Carolyn Tucker Halpern and Gary Greenberg. (2010). *Handbook of Developmental Science, Behavior, and Genetics*. Wiley-Blackwell. p. 70
- [26] Review of Lovtrup's book in the New Scientist, Oct 15, 1988 (http://books.google.com/books?id=2O7iz2mGeSkC&pg=PA66&dq=soren+lovtrup&hl=en&ei=UcfRTZfoDMyVswbArJC4CQ&sa=X&oi=book_result&ct=book-preview-link&resnum=1&ved=0CC4QuwUwAA#v=onepage&q=soren%20lovtrup&f=false)
- [27] Guenter Theissen. (2010). *Homeosis of the angiosperm flower: Studies on three candidate cases of saltational evolution* (http://www.palaeodiversity.org/pdf/03Suppl/Supplement_Theissen.pdf). Palaeodiversity 3, Supplement: 131-139.
- [28] Donald Prothero. (2007). *Evolution: What the Fossils Say and Why It Matters*. Columbia University Press. p. 100
- [29] Olivia Judson. (2008). *The Monster Is Back, and It's Hopeful* (<http://opinionator.blogs.nytimes.com/2008/01/22/the-monster-is-back-and-its-hopeful/>). The New York Times.
- [30] Tanguy Chouard. (2010). *Evolution: Revenge of the hopeful monster* (<http://www.nature.com/news/2010/100217/full/463864a.html>). Nature 463, 864-867.
- [31] Robert B. Page, Meredith A. Boley, Jeramiah J. Smith, Srikrishna Putta and Stephen R. Voss. (2010). *Microarray analysis of a salamander hopeful monster reveals transcriptional signatures of paedomorphic brain development* (<http://www.biomedcentral.com/1471-2148/10/199>). BMC Evolutionary Biology, 10:199.
- [32] Guenter Theissen. (2006). *The proper place of hopeful monsters in evolutionary biology* (<http://evolocus.com/Publications/Theissen2006.pdf>). Theory in Biosciences, 124: 349–369.
- [33] Hintz, M.; Bartholmes, C.; Nutt, P.; Ziermann, J.; Hameister, S.; Neuffer, B.; Theissen, G. (2006). *Catching a 'hopeful monster': shepherd's purse (*Capsellabursa-pastoris*) as a model system to study the evolution of flower development* (<http://jxb.oxfordjournals.org/content/57/13/3531.full.pdf>). Journal of Experimental Botany, 57: 3531-3542.
- [34] Freeling M, Thomas BC. (2006). *Gene-balanced duplications, like tetraploidy, provide predictable drive to increase morphological complexity*. Genome Res. 16: 805-814. Freeling M. (2009). *Bias in plant gene content following different sorts of duplication: tandem, whole-genome, segmental, or by transposition*. Annu Rev Plant Biol 60: 433–453.
- [35] Serres MH, Kerr AR, McCormack TJ, Riley M. (2009). *Evolution by leaps: gene duplication in bacteria* (<http://www.biologydirec.com/content/4/1/46>). Biology Direct 4: 46.
- [36] Minelli, A, Chagas Junior, A, & Edgecombe, G D. (2009). *Saltational evolution of trunk segment number in centipedes* (<http://www.nhm.ac.uk/resources-rx/files/minelli-et-al-2009-saltation-94095.pdf>). Evolution & development. 11: 318-322.
- [37] Rubinoff, D. and J. J. Le Roux. (2008). *Evidence of Repeated and Independent Saltational Evolution in a Peculiar Genus of Sphinx Moths (*Proserpinus: Sphingidae*)* (<http://www.plosone.org/article/info:doi/10.1371/journal.pone.0004035>). PLoS ONE 3 (12): e4035.doi:10.1371/journal.pone.0004035.
- [38] Knight CG, Patel MN, Azevedo RB, Leroi AM. (2002). *A novel mode of ecdysozoan growth in Caenorhabditis elegans* (<http://www.mcisb.org/people/knight/docs/WormSize.pdf>). Evolution & development. 4: 16–27.
- [39] Eva Jablonka. (2012). *Epigenetic inheritance and plasticity: The responsive germline* (<http://www.ncbi.nlm.nih.gov/pubmed/22975443>). Prog Biophys Mol Biol. 1-0.
- [40] Olof Leimar, Birgitta S. Tullberg and James Mallet. *Mimicry, Saltational Evolution, and the Crossing of Fitness Valleys* (https://docs.google.com/viewer?a=v&q=cache:yhegI8wPURgJ:www.ucl.ac.uk/taxome/jim/pap/leimar_et.al_2012.pdf+mimicry+saltation&hl=en&gl=uk&pid=bl&srcid=ADGEEShEeNvgfCL-j1mOo7K52ogmuBsdQiGy6KI0py4navgNaH_6pcokMaSKXXZZ-hFCVD9ekaddXv_h3l9MkLKhmnVvaK31YhL7p2R7BXM12Wjsig=AHIEtbRSMHsCj-8pmstF2u0TD15sCeaT2KA). In E. I. Svensson & R. Calsbeek eds. (2012). *The Adaptive Landscape in Evolutionary Biology*. Oxford University Press.
- [41] Norrström, N., Getz, W. M., & Holmgren, N. M. A. (2006). *Coevolution of exploiter specialization and victim mimicry can be cyclic and saltational* (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2674650/>). Evolutionary Bioinformatics Online. 2: 35-43.
- [42] Michael Syvanen, Clarence I. Kado. (2002). *Horizontal Gene Transfer* Academic Press. p. 405. ISBN 978-0126801262
- [43] Symonds, M.R.E., and M. A. Elgar. (2004). *The mode of pheromone evolution: evidence from bark beetles* (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1691658/>). Proc. R. Soc. Lond. B. Biol. Sci. 271: 839–846.
- [44] Symonds, Matthew R. E., Moussalli, Adnan and Elgar, Mark A. (2009). *The evolution of sex pheromones in an ecologically diverse genus of flies* (<http://www.utsc.utoronto.ca/~mandrade/reprints/flies.pdf>). Biological journal of the Linnean Society. Vol. 97, no. 3. pp. 594-603.
- [45] Lewis, H. (1966). *Speciation in flowering plants*. Science 152: 167–172.
- [46] Carr, G. D. (1980). *Experimental evidence for saltational chromosome evolution in Calycadenia pauciflora Gray (Asteraceae)*. Heredity 45: 107–112. Carr GD, Carr RL. (2000). *A new chromosome race of Calycadenia pauciflora (Asteraceae: Heliantheae-Madiinae) from Butte County, California*. Amer. J. Bot. 87. (10): 1459-1465.
- [47] Bateman RM, WA DiMichele. (2002). *Generating and filtering major phenotypic novelties: neoGoldschmidtian saltation revisited*. pp. 109–159 in QCB Cronk, RM Bateman, JA Hawkins, eds. *Developmental genetics and plant evolution*. Taylor & Francis, London
- [48] Louis P. Ronse De Craene. (2002). *The Evolutionary Significance of Homeosis in Flowers: A Morphological Perspective* (<http://www.jstor.org/discover/10.1086/376878>). International Journal of Plant Sciences. Vol. 164, No. 5, Flowers—Diversity, Development, and Evolution A conference organized and held at the Institute of Systematic Botany, University of Zurich, Switzerland, July 5–7. pp. 225-235.
- [49] Eldredge, Niles and S. J. Gould (1972). "Punctuated equilibria: an alternative to phyletic gradualism" (<http://www.blackwellpublishing.com/ridley/classictexts/eldredge.asp>) In T.J.M. Schopf, ed., *Models in Paleobiology*. San Francisco: Freeman Cooper. pp. 82-115.

-
- [50] Stephen Jay Gould. (1981). *Evolution as Fact and Theory* (http://www.stephenjaygould.org/library/gould_fact-and-theory.html). *Discover* 2 (May): 34-37.
 - [51] Steven M. Stanley. (1981). *The New Evolutionary Timetable*. New York: Basic Books. p. 135

Further reading

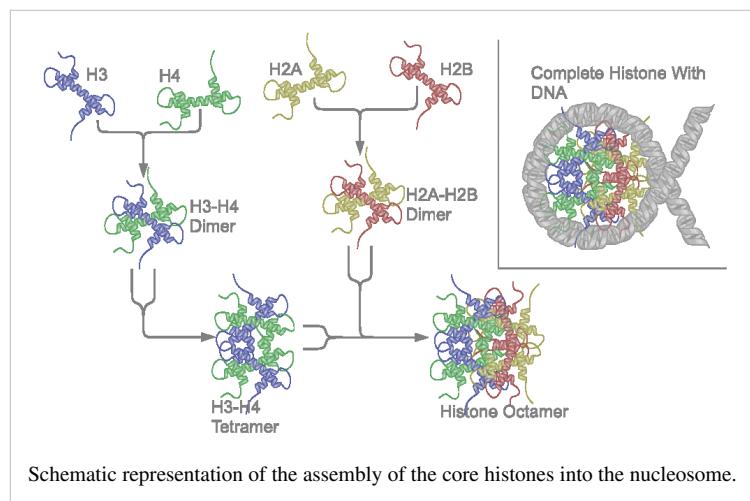
- Thomas C. Baker. (2002). *Mechanism for saltational shifts in pheromone communication systems* (<http://www.pnas.org/content/99/21/13368.full>). Proceedings of the National Academy of Sciences. USA 99: 13368-13370.
- Richard M. Bateman, William A. DiMichele. (2002). *Generating and filtering major phenotypic novelties: neoGoldschmidtian saltation revisited*. In: Cronk QCB, Bateman RM, Hawkins JA, eds. *Developmental genetics and plant evolution*. London: Taylor & Francis. pp. 109–159.
- Brian K. Hall, Roy D. Pearson, Gerd B. Müller. (2004). *Environment, Development, and Evolution: Toward a Synthesis*. MIT Press. ISBN 978-0262083195
- Ulrich Kutschera, Karl J. Niklas. (2008). *Macroevolution via secondary endosymbiosis: a Neo-Goldschmidtian view of unicellular hopeful monsters and Darwin's primordial intermediate form*. Theory in Biosciences 127: 277-289.
- David J. Merrell. (1994). *The Adaptive Seascape: The Mechanism of Evolution*. University of Minnesota Press. ISBN 978-0816623488
- Jeffrey H. Schwartz. (2006). *Sudden origins: a general mechanism of evolution based on stress protein concentration and rapid environmental change* (http://www.pitt.edu/~jhs/articles/Maresca_Schwartz_sudden_origins.pdf). The Anatomical Record. 289: 38–46.
- Gamberale-Stille G, Balogh AC, Tullberg BS, Leimar O. (2012). *Feature saltation and the evolution of mimicry* (<http://www.ncbi.nlm.nih.gov/pubmed/22380441>). Evolution 66: 807-17.
- Guenter Theissen. (2009). *Saltational evolution: hopeful monsters are here to stay* (<http://www.evolocus.com/publications/theissen2009.pdf>). Theory in Bioscience. 128, 43-51.

External links

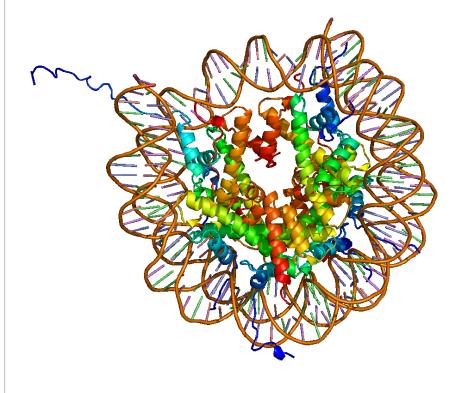
- New species evolve in bursts by Kerri Smith (<http://www.nature.com/news/2009/091209/full/news.2009.1134.html>)

Histone

In biology, **histones** are highly alkaline proteins found in eukaryotic cell nuclei that package and order the DNA into structural units called nucleosomes. They are the chief protein components of chromatin, acting as spools around which DNA winds, and play a role in gene regulation. Without histones, the unwound DNA in chromosomes would be very long (a length to width ratio of more than 10 million to 1 in human DNA). For example, each human cell has about 1.8 meters of DNA, but wound on the histones it has about 90 micrometers (0.09 mm) of chromatin, which, when duplicated and condensed during mitosis, result in about 120 micrometers of chromosomes.

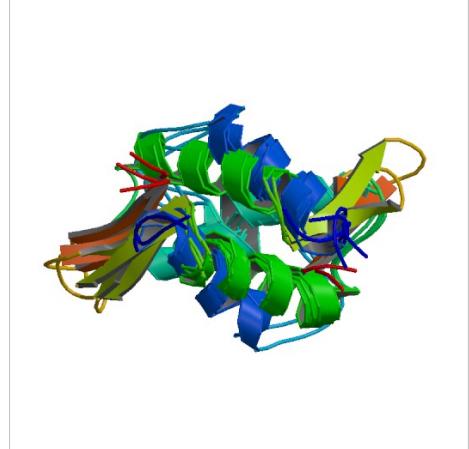


Schematic representation of the assembly of the core histones into the nucleosome.

| Core histone H2A/H2B/H3/H4 | |
|-----------------------------------------------------------------------------------------------------------------------|---------------|
|  | |
| PDB rendering of Complex between nucleosome core particle (h3,h4,h2a,h2b) and 146 bp long DNA fragment based on 1aoi. | |
| Identifiers | |
| Symbol | Histone |
| Pfam | PF00125 [1] |
| Pfam clan | CL0012 [2] |
| InterPro | IPR007125 [3] |
| SCOP | 1hio [4] |
| SUPERFAMILY | 1hio [5] |

| Available protein structures: | |
|-------------------------------|----------------------------------|
| Pfam | structures [6] |
| PDB | RCSB PDB [7]; PDBe [8]; PDBj [9] |
| PDBsum | structure summary [10] |

linker histone H1 and H5 family



PDB rendering of HIST1H1B based on 1ghc.

| Identifiers | |
|--------------------|----------------|
| Symbol | Linker_histone |
| Pfam | PF00538 [11] |
| InterPro | IPR005818 [12] |
| SMART | SM00526 [13] |
| SCOP | 1hst [14] |
| SUPERFAMILY | 1hst [15] |

| Available protein structures: | |
|-------------------------------|-------------------------------------|
| Pfam | structures [16] |
| PDB | RCSB PDB [17]; PDBe [18]; PDBj [19] |
| PDBsum | structure summary [20] |

Classes

Five major families of histones exist: H1/H5, H2A, H2B, H3 and H4. Histones H2A, H2B, H3 and H4 are known as the core histones, while histones H1 and H5 are known as the linker histones.

Two of each of the core histones assemble to form one octameric nucleosome core, approximately 63 Angstroms in diameter (a solenoid (DNA)-like particle). 147 base pairs of DNA wrap around this core particle 1.65 times in a left-handed super-helical turn to give a particle of around 100 Angstroms across. The linker histone H1 binds the nucleosome at the entry and exit sites of the DNA, thus locking the DNA into place and allowing the formation of higher order structure. The most basic such formation is the 10 nm fiber or beads on a string conformation. This involves the wrapping of DNA around nucleosomes with approximately 50 base pairs of DNA separating each pair of nucleosomes (also referred to as linker DNA). Higher-order structures include the 30 nm fiber (forming an irregular zigzag) and 100 nm fiber, these being the structures found in normal cells. During mitosis and meiosis, the condensed chromosomes are assembled through interactions between nucleosomes and other regulatory proteins.

The following is a list of human histone proteins:

| Super family | Family | Subfamily | Members |
|--------------|--------|-----------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Linker | H1 | H1F | H1F0, H1FNT, H1FOO, H1FX |
| | | H1H1 | HIST1H1A, HIST1H1B, HIST1H1C, HIST1H1D, HIST1H1E, HIST1H1T |
| Core | H2A | H2AF | H2AFB1, H2AFB2, H2AFB3, H2AFJ, H2AFV, H2AFX, H2AFY, H2AFY2, H2AFZ |
| | | H2A1 | HIST1H2AA, HIST1H2AB, HIST1H2AC, HIST1H2AD, HIST1H2AE, HIST1H2AG, HIST1H2AI, HIST1H2AJ, HIST1H2AK, HIST1H2AL, HIST1H2AM |
| | | H2A2 | HIST2H2AA3, HIST2H2AC |
| | H2B | H2BF | H2BFM, H2BFS, H2BFWT |
| | | H2B1 | HIST1H2BA, HIST1H2BB, HIST1H2BC, HIST1H2BD, HIST1H2BE, HIST1H2BF, HIST1H2BG, HIST1H2BH, HIST1H2BI, HIST1H2BJ, HIST1H2BK, HIST1H2BL, HIST1H2BM, HIST1H2BN, HIST1H2BO |
| | | H2B2 | HIST2H2BE |
| | H3 | H3A1 | HIST1H3A, HIST1H3B, HIST1H3C, HIST1H3D, HIST1H3E, HIST1H3F, HIST1H3G, HIST1H3H, HIST1H3I, HIST1H3J |
| | | H3A2 | HIST2H3C |
| | | H3A3 | HIST3H3 |
| | H4 | H4I | HIST1H4A, HIST1H4B, HIST1H4C, HIST1H4D, HIST1H4E, HIST1H4F, HIST1H4G, HIST1H4H, HIST1H4I, HIST1H4J, HIST1H4K, HIST1H4L |
| | | H44 | HIST4H4 |

Structure

The nucleosome core is formed of two H2A-H2B dimers and a H3-H4 tetramer, forming two nearly symmetrical halves by tertiary structure (C2 symmetry; one macromolecule is the mirror image of the other). The H2A-H2B dimers and H3-H4 tetramer also show pseudodyad symmetry. The 4 'core' histones (H2A, H2B, H3 and H4) are relatively similar in structure and are highly conserved through evolution, all featuring a 'helix turn helix turn helix' motif (which allows the easy dimerisation). They also share the feature of long 'tails' on one end of the amino acid structure - this being the location of post-translational modification (see below).

It has been proposed that histone proteins are evolutionarily related to the helical part of the extended AAA+ ATPase domain, the C-domain, and to the N-terminal substrate recognition domain of Clp/Hsp100 proteins. Despite the differences in their topology, these three folds share a homologous helix-strand-helix (HSH) motif.

Using an electron paramagnetic resonance spin-labeling technique, British researchers measured the distances between the spools around which eukaryotic cells wind their DNA. They determined the spacings range from 59 to 70 Å.

In all, histones make five types of interactions with DNA:

- Helix-dipoles from alpha-helices in H2B, H3, and H4 cause a net positive charge to accumulate at the point of interaction with negatively charged phosphate groups on DNA
- Hydrogen bonds between the DNA backbone and the amide group on the main chain of histone proteins
- Nonpolar interactions between the histone and deoxyribose sugars on DNA
- Salt bridges and hydrogen bonds between side chains of basic amino acids (especially lysine and arginine) and phosphate oxygens on DNA
- Non-specific minor groove insertions of the H3 and H2B N-terminal tails into two minor grooves each on the DNA molecule

The highly basic nature of histones, aside from facilitating DNA-histone interactions, contributes to their water solubility.

Histones are subject to post translational modification by enzymes primarily on their N-terminal tails, but also in their globular domains Wikipedia:Citation needed. Such modifications include methylation, citrullination, acetylation, phosphorylation, SUMOylation, ubiquitination, and ADP-ribosylation. This affects their function of gene regulation (see "Function" section).

In general, genes that are active have less bound histone, while inactive genes are highly associated with histones during interphase Wikipedia:Citation needed. It also appears that the structure of histones has been evolutionarily conserved, as any deleterious mutations would be severely maladaptive. All histones have a highly positively charged N-terminus with many lysine and arginine residues.

History

Histones were discovered in 1884 by Albrecht Kossel. The word "histone" dates from the late 19th century and is from the German "Histon", of uncertain origin: perhaps from Greek *histanai* or from *histos*. Until the early 1990s, histones were dismissed by most as inert packing material for eukaryotic nuclear DNA, based in part on the "ball and stick" models of Mark Ptashne and others who believed that transcription was activated by protein-DNA and protein-protein interactions on largely naked DNA templates, as is the case in bacteria. During the 1980s, work by Michael Grunstein demonstrated that eukaryotic histones repress gene transcription, and that the function of transcriptional activators is to overcome this repression. It is now known that histones play both positive and negative roles in gene expression, forming the basis of the histone code. The work of Vincent Allfrey on histone modification was a pioneering one, he is regarded as father of epigenetics.^[21]

The discovery of the H5 histone appears to date back to 1970s, and in classification it has been grouped with H1.

Conservation across species

Histones are found in the nuclei of eukaryotic cells, and in certain Archaea, namely Thermoproteales and Euryarchaea, but not in bacteria. The unicellular algae known as dinoflagellates are the only eukaryotes that are known to completely lack histones.

Archaeal histones may well resemble the evolutionary precursors to eukaryotic histones. Histone proteins are among the most highly conserved proteins in eukaryotes, emphasizing their important role in the biology of the nucleus.^[939] In contrast mature sperm cells largely use protamines to package their genomic DNA, most likely because this allows them to achieve an even higher packaging ratio.

Core histones are highly conserved proteins; that is, there are very few differences among the amino acid sequences of the histone proteins of different species. Linker histone usually has more than one form within a species and is

also less conserved than the core histones. [Wikipedia:Citation needed](#)

There are some *variant* forms in some of the major classes. They share amino acid sequence homology and core structural similarity to a specific class of major histones but also have their own feature that is distinct from the major histones. These *minor histones* usually carry out specific functions of the chromatin metabolism. For example, histone H3-like CenpA is associated with only the centromere region of the chromosome. Histone H2A variant H2A.Z is associated with the promoters of actively transcribed genes and also involved in the prevention of the spread of silent heterochromatin. Furthermore, H2A.Z has roles in chromatin for genome stability. Another H2A variant H2A.X binds to the DNA with double-strand breaks and marks the region undergoing DNA repair. Histone H3.3 is associated with the body of actively transcribed genes.

Function

Compacting DNA strands

Histones act as spools around which DNA winds. This enables the compaction necessary to fit the large genomes of eukaryotes inside cell nuclei: the compacted molecule is 40,000 times shorter than an unpacked molecule.

Chromatin regulation

Histones undergo posttranslational modifications that alter their interaction with DNA and nuclear proteins. The H3 and H4 histones have long tails protruding from the nucleosome, which can be covalently modified at several places. Modifications of the tail include methylation, acetylation, phosphorylation, ubiquitination, SUMOylation, citrullination, and ADP-ribosylation. The core of the histones H2A, H2B, and H3 can also be modified. Combinations of modifications are thought to constitute a code, the so-called "histone code". Histone modifications act in diverse biological processes such as gene regulation, DNA repair, chromosome condensation (mitosis) and spermatogenesis (meiosis).

The common nomenclature of histone modifications is:

- The name of the histone (e.g., H3)
- The single-letter amino acid abbreviation (e.g., K for Lysine) and the amino acid position in the protein
- The type of modification (Me: methyl, P: phosphate, Ac: acetyl, Ub: ubiquitin)
- The number of modifications (only Me is known to occur in more than one copy per residue. 1, 2 or 3 is mono-, di- or tri-methylation)

So H3K4me1 denotes the monomethylation of the 4th residue (a lysine) from the start (i.e., the N-terminal) of the H3 protein.

Examples of histone modifications in transcription regulation include:

| Type of modification | Histone | | | | | | | |
|----------------------|------------|------------|------------|------------|------------------------|------------|------------|------------|
| | H3K4 | H3K9 | H3K14 | H3K27 | H3K79 | H3K36 | H4K20 | H2BK5 |
| mono-methylation | activation | activation | | activation | activation | | activation | activation |
| di-methylation | | repression | | repression | activation | | | |
| tri-methylation | activation | repression | | repression | activation, repression | activation | | repression |
| acetylation | | activation | activation | activation | | | | |

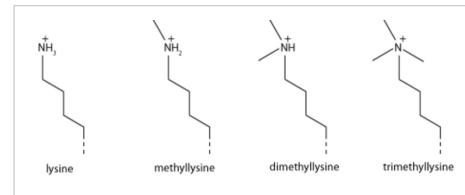
Functions of histone modifications

A huge catalogue of histone modifications have been described, but a functional understanding of most is still lacking. Collectively, it is thought that histone modifications may underlie a histone code, whereby combinations of histone modifications have specific meanings. However, most functional data concerns individual prominent histone modifications that are biochemically amenable to detailed study.

Chemistry of histone modifications

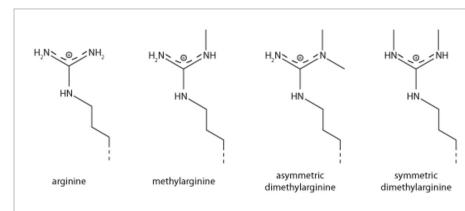
Lysine methylation

The addition of one, two or three methyl groups to lysine has little effect on the chemistry of the histone; methylation leaves the charge of the lysine intact and adds a minimal number of atoms so steric interactions are mostly unaffected. However, proteins containing Tudor, chromo or PHD domains, amongst others, can recognise lysine methylation with exquisite sensitivity and differentiate mono, di and tri-methyl lysine, to the extent that, for some lysines (e.g.: H4K20) mono, di and tri-methylation appear to have different meanings. Because of this, lysine methylation tends to be a very informative mark and dominates the known histone modification functions.



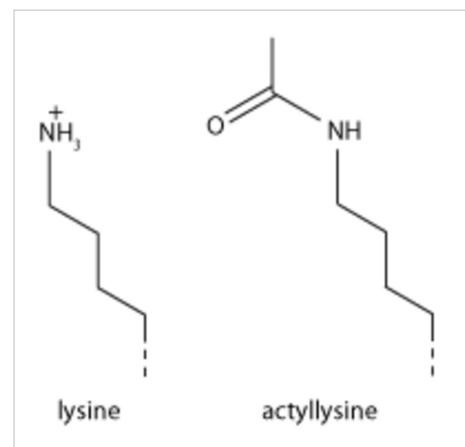
Arginine methylation

What was said above of the chemistry of lysine methylation also applies to arginine methylation, and some protein domains—e.g., Tudor domains—can be specific for methyl arginine instead of methyl lysine. Arginine is known to be mono- or di-methylated, and methylation can be symmetric or asymmetric, potentially with different meanings.



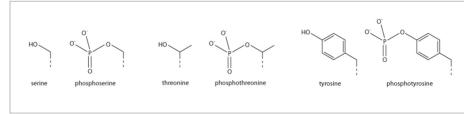
Lysine acetylation

Addition of an acetyl group has a major chemical effect on lysine as it neutralises the positive charge. This reduces electrostatic attraction between the histone and the negatively charged DNA backbone, loosening the chromatin structure; highly acetylated histones form more accessible chromatin and tend to be associated with active transcription. Lysine acetylation appears to be less precise in meaning than methylation, in that histone acetyltransferases tend to act on more than one lysine; presumably this reflects the need to alter multiple lysines to have a significant effect on chromatin structure.



Serine/Threonine/Tyrosine phosphorylation

Addition of a negatively charged phosphate group can lead to major changes in protein structure, leading to the well-characterised role of phosphorylation in controlling protein function. It is not clear what structural implications histone phosphorylation has, but histone phosphorylation has clear functions as a post-translational modification, and binding domains such as BRCT have been characterised.



Functions in transcription

Most well-studied histone modifications are involved in control of transcription.

Actively transcribed genes

Two histone modifications are particularly associated with active transcription:

Trimethylation of H3 lysine 4 (H3K4Me3) at the promoter of active genes

H3K4 trimethylation is performed by the COMPASS complex. Despite the conservation of this complex and histone modification from yeast to mammals, it is not entirely clear what role this modification plays. However, it is an excellent mark of active promoters and the level of this histone modification at a gene's promoter is broadly correlated with transcriptional activity of the gene. The formation of this mark is tied to transcription in a rather convoluted manner: early in transcription of a gene, RNA polymerase II undergoes a switch from initiating' to 'elongating', marked by a change in the phosphorylation states of the RNA polymerase II C terminal domain (CTD). The same enzyme that phosphorylates the CTD also phosphorylates the Rad6 complex, which in turn adds a ubiquitin mark to H2B K123 (K120 in mammals). H2BK123Ub occurs throughout transcribed regions, but this mark is required for COMPASS to trimethylate H3K4 at promoters.

Trimethylation of H3 lysine 36 (H3K36Me3) in the body of active genes

H3K36 trimethylation is deposited by the methyltransferase Set2. This protein associates with elongating RNA polymerase II, and H3K36Me3 is indicative of actively transcribed genes. H3K36Me3 is recognised by the Rpd3 histone deacetylase complex, which removes acetyl modifications from surrounding histones, increasing chromatin compaction and repressing spurious transcription. Increased chromatin compaction prevents transcription factors from accessing DNA, and reduces the likelihood of new transcription events being initiated within the body of the gene. This process therefore helps ensure that transcription is not interrupted.

Repressed genes

Three histone modifications are particularly associated with repressed genes:

Trimethylation of H3 lysine 27 (H3K27Me3)

This histone modification is deposited by the polycomb complex PRC2. It is a clear marker of gene repression, and is likely bound by other proteins to exert a repressive function. Another polycomb complex, PRC1, can bind H3K27Me3 and adds the histone modification H2AK119Ub which aids chromatin compaction. Based on this data it appears that PRC1 is recruited through the action of PRC2, however, recent studies show that PRC1 is recruited to the same sites in the absence of PRC2.

Di and tri-methylation of H3 lysine 9 (H3K9Me2/3)

H3K9Me2/3 is a well-characterised marker for heterochromatin, and is therefore strongly associated with gene repression. The formation of heterochromatin has been best studied in the yeast *Schizosaccharomyces pombe*, where it is initiated by recruitment of the RNA-induced transcriptional silencing complex to double stranded RNAs produced from centromeric repeats. RITS recruits the Clr4 histone methyltransferase which deposits H3K9Me2/3. This process is called histone methylation. H3K9Me2/3 serves as a binding site for the recruitment of Swi6

(heterochromatin protein 1 or HP1, another classic heterochromatin marker) which in turn recruits further repressive activities including histone modifiers such as histone deacetylases and histone methyltransferases.

Trimethylation of H4 lysine 20 (H4K20Me3)

This modification is tightly associated with heterochromatin, although its functional importance remains unclear. This mark is placed by the Suv4-20h methyltransferase, which is at least in part recruited by heterochromatin protein 1.

Bivalent promoters

Analysis of histone modifications in embryonic stem cells (and other stem cells) revealed many gene promoters carrying both H3K4Me3 and H3K27Me3, in other words these promoters display both activating and repressing marks simultaneously. This peculiar combination of modifications marks genes that are poised for transcription; they are not required in stem cells, but are rapidly required after differentiation into some lineages. Once the cell starts to differentiate, these bivalent promoters are resolved to either active or repressive states depending on the chosen lineage.

Other functions

DNA damage

Marking sites of DNA damage is an important function for histone modifications.

Phosphorylation of H2AX at serine 139 (γ H2AX)

Phosphorylated H2AX (also known as gamma H2AX) is a marker for DNA double strand breaks, and forms part of the response to DNA damage. H2AX is phosphorylated early after detection of DNA double strand break, and forms a domain extending many kilobases either side of the damage. Gamma H2AX acts as a binding site for the protein MDC1, which in turn recruits key DNA repair proteins (this complex topic is well reviewed in) and as such, gamma H2AX forms a vital part of the machinery that ensures genome stability.

Acetylation of H3 lysine 56 (H3K56Ac)

H3K56Ac is required for genome stability. H3K56 is acetylated by the p300/Rtt109 complex, but is rapidly deacetylated around sites of DNA damage. H3K56 acetylation is also required to stabilise stalled replication forks, preventing dangerous replication fork collapses. Although in general mammals make far greater use of histone modifications than microorganisms, a major role of H3K56Ac in DNA replication exists only in fungi, and this has become a target for antibiotic development.

Chromosome condensation

Phosphorylation of H3 at serine 10 (phospho-H3S10)

The mitotic kinase aurora B phosphorylates histone H3 at serine 10, triggering a cascade of changes that mediate mitotic chromosome condensation. Condensed chromosomes therefore stain very strongly for this mark, but H3S10 phosphorylation is also present at certain chromosome sites outside mitosis, for example in pericentric heterochromatin of cells during G2. H3S10 phosphorylation has also been linked to DNA damage caused by R loop formation at highly transcribed sites.

Phosphorylation H2B at serine 10 in yeast or serine 14 in mammalian cells (phospho-H2BS10/14)

Phosphorylation of H2B at serine 10 (yeast) or serine 14 (mammals) is also linked to chromatin condensation, but for the very different purpose of mediating chromosome condensation during apoptosis. This mark is not simply a late acting bystander in apoptosis as yeast carrying mutations of this residue are resistant to hydrogen peroxide-induced apoptotic cell death.

References

- [1] <http://pfam.xfam.org/family?acc=PF00125>
- [2] <http://pfam.xfam.org/clan/CL0012>
- [3] <http://www.ebi.ac.uk/interpro/entry/IPR007125>
- [4] <http://scop.mrc-lmb.cam.ac.uk/scop/search.cgi?tlev=fa;&pdb=1hio>
- [5] http://supfam.org/SUPERFAMILY/cgi-bin/search.cgi?search_field=1hio
- [6] <http://pfam.sanger.ac.uk/family/PF00125?tab=pdbBlock>
- [7] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF00125>
- [8] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF00125>
- [9] <http://pdjbj.org/searchFor?query=PF00125>
- [10] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF00125
- [11] <http://pfam.xfam.org/family?acc=PF00538>
- [12] <http://www.ebi.ac.uk/interpro/entry/IPR005818>
- [13] http://smart.embl-heidelberg.de/smart/do_annotation.pl?DOMAIN=SM00526
- [14] <http://scop.mrc-lmb.cam.ac.uk/scop/search.cgi?tlev=fa;&pdb=1hst>
- [15] http://supfam.org/SUPERFAMILY/cgi-bin/search.cgi?search_field=1hst
- [16] <http://pfam.sanger.ac.uk/family/PF00538?tab=pdbBlock>
- [17] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF00538>
- [18] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF00538>
- [19] <http://pdjbj.org/searchFor?query=PF00538>
- [20] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF00538
- [21] Histones, Calf Thymus. "Vincent Allfrey's Work on Histone Acetylation." (2012)

External links

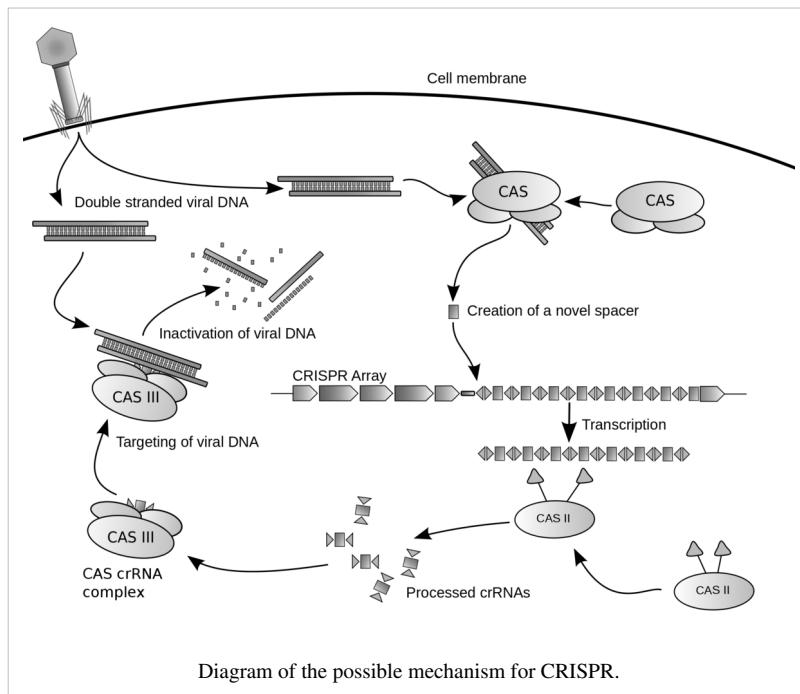
- Chromatin, Histones & Cathepsin (<http://www.youtube.com/watch?v=eYrQ0EhVCYA>); PMAP The Proteolysis Map-animation

CRISPR

CRISPRs (clustered regularly interspaced short palindromic repeats) are DNA loci containing short repetitions of base sequences. Each repetition is followed by short segments of "spacer DNA" from previous exposures to a virus.

CRISPRs are found in approximately 40% of sequenced eubacteria genomes and 90% of sequenced archaea.^[1]

CRISPRs are often associated with **cas genes** that code for proteins related to CRISPRs. The CRISPR/Cas system is a prokaryotic immune system that confers resistance to foreign genetic elements such as plasmids and phages and provides a form of acquired immunity. CRISPR spacers recognize and cut these exogenous genetic elements in a manner analogous to RNAi in eukaryotic organisms.



Since 2013, the CRISPR/Cas system has been used for gene editing (adding, disrupting or changing the sequence of specific genes) and gene regulation in species throughout the tree of life. By delivering the Cas9 protein and appropriate guide RNAs into a cell, the organism's genome can be cut at any desired location.

It may be possible to use CRISPR to build RNA-guided gene drives capable of altering the genomes of entire populations.

History

Bacteria may incorporate foreign DNA in other circumstances and even scavenge damaged DNA from their environment.

Repeats were first described in 1987 for the bacterium *Escherichia coli*. In 2000, similar clustered repeats were identified in additional bacteria and archaea and were termed Short Regularly Spaced Repeats (SRSR). SRSR were renamed CRISPR in 2002. A set of genes, some encoding putative nuclease or helicase proteins, were found to be associated with CRISPR repeats (the *cas*, or *CRISPR-associated* genes).

In 2005, three independent researchers showed that CRISPR spacers showed homology to several phage DNA and extrachromosomal DNA such as plasmids. This was an indication that the CRISPR/cas system could have a role in adaptive immunity in bacteria. Koonin and colleagues proposed that spacers serve as a template for RNA molecules, analogously to eukaryotic cells that use a system called RNA interference.

In 2007 Barrangou, Horvath (food industry scientists at Danisco) and Moineau's group at Université Laval (Canada) showed that they could alter the resistance of *Streptococcus thermophilus* to phage attack with spacer DNA.

Doudna and Charpentier had independently been exploring CRISPR-associated proteins to learn how bacteria deploy spacers in their immune defenses. They jointly studied a simpler CRISPR system that relies on a protein called Cas9. They found that bacteria respond to an invading phage by transcribing spacers and palindromic DNA into a long RNA molecule that the cell then uses tracrRNA and Cas9 to cut it into pieces called crRNAs.

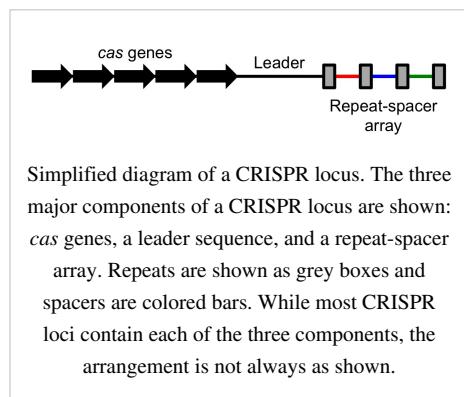
Cas9 is a nuclease, an enzyme specialized for cutting DNA, with two active cutting sites, one for each strand of the double helix. The team demonstrated that they could disable one or both sites while preserving Cas9's ability to home in on its target DNA. Jinek combined tracrRNA and spacer RNA into a "single-guide RNA" molecule that, mixed with Cas9, could find and cut the correct DNA targets. Jinek *et al* proposed that such synthetic guide RNAs might be able to be used for gene editing.

CRISPR was first shown to work as a genome engineering/editing tool in human cell culture by 2012. It has since been used in a wide range of organisms including baker's yeast (*S. cerevisiae*), zebra fish (*D. rerio*), flies (*D. melanogaster*), nematodes (*C. elegans*), plants, mice, and several other organisms.

Additionally CRISPR has been modified to make programmable transcription factors that allow scientists to target and activate or silence specific genes.

Libraries of tens of thousands of guide RNAs are now available.

The first evidence that CRISPR can reverse disease symptoms in living animals was demonstrated in March 2014, when MIT researchers cured mice of a rare liver disorder.



Gene-editing predecessors

In the early 2000s, researchers developed zinc finger nucleases, synthetic proteins whose DNA-binding domains enable them to cut DNA at specific spots. Later, synthetic nucleases called TALENs provided an easier way to target specific DNA and were predicted to surpass zinc fingers. They both depend on making custom proteins for each DNA target, a more cumbersome procedure than guide RNAs. CRISPRs are more efficient and can target more genes than these earlier techniques.

Locus structure

Repeats and spacers

CRISPR loci range in size from 24 to 48 base pairs. They usually show some dyad symmetry, implying the formation of a secondary structure such as a hairpin, but are not truly palindromic. Repeats are separated by spacers of similar length. Some CRISPR spacer sequences exactly match sequences from plasmids and phages, although some spacers match the prokaryote's genome (self-targeting spacers). New spacers can be added rapidly in response to phage infection.

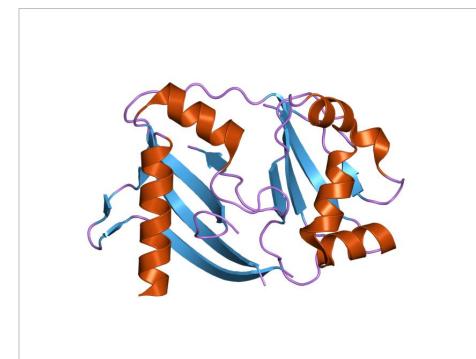
Cas genes and CRISPR subtypes

CRISPR-associated (*cas*) genes are often associated with CRISPR repeat-spacer arrays. Extensive comparative genomics have identified many different *cas* genes; an initial analysis of 40 bacterial and archaeal genomes suggested that there may be 45 *cas* gene families, with only two genes, *cas1* and *cas2*, universally present. The current CRISPR classification groups *cas* operons into three major divisions, each with multiple subdivisions based on *cas1* phylogeny and *cas* operon gene complement. Aside from *cas1* and *cas2*, the three major divisions have vastly different sets of constituent genes, with each of the subdivisions characterised by a 'signature gene' found exclusively in that subdivision. Many organisms contain multiple CRISPR-Cas systems suggesting that they are compatible and may even share components. The sporadic distribution of the CRISPR/Cas subtypes suggests that the system is subject to horizontal gene transfer during microbial evolution.

Signature genes and their putative functions for the major and minor CRISPR-cas types.

| Cas type | Signature gene | Function | Reference |
|----------|----------------|--------------------------------------------------------------------------------------------------|-----------|
| I | Cas3 | Single-stranded DNA nuclease (HD domain) and ATP-dependent helicase | |
| IA | Cas8a | Subunit of the interference module | |
| IB | Cas8b | | |
| IC | Cas8c | | |
| ID | Cas10d | contains a domain homologous the palm domain of nucleic acid polymerases and nucleotide cyclases | |
| IE | Cse1 | | |
| IF | Csy1 | Not Determined | |
| II | Cas9 | RuvC and HNH domain containing nuclease | |
| IIA | Csn2 | Not Determined | |
| IIB | Cas4 | Not Determined | |
| IIC | | Characterized by the absence of either Csn2 or Cas4 | |
| III | Cas10 | Homolog of Cas10d and Cse1 | |
| IIIA | Csm2 | Not Determined | |
| IIIB | Cmr5 | Not Determined | |

CRISPR associated protein

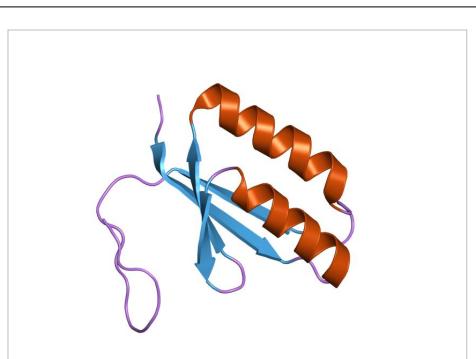


crystal structure of a crispr-associated protein from *thermus thermophilus*

| Identifiers | |
|------------------|---------------|
| Symbol | CRISPR_assoc |
| Pfam | PF08798 [2] |
| Pfam clan | CL0362 [3] |
| InterPro | IPR010179 [4] |
| CDD | cd09727 [5] |

| Available protein structures: | |
|-------------------------------|----------------------------------|
| Pfam | structures [6] |
| PDB | RCSB PDB [7]; PDBe [8]; PDBj [9] |
| PDBsum | structure summary [10] |

CRISPR associated protein Cas2



crystal structure of a hypothetical protein tt1823 from *thermus thermophilus*

| Identifiers | |
|-----------------|----------------|
| Symbol | CRISPR_Cas2 |
| Pfam | PF09827 [11] |
| InterPro | IPR019199 [12] |

| | |
|--------------------------------------|-------------------------------------|
| CDD | cd09638 [13] |
| Available protein structures: | |
| Pfam | structures [14] |
| PDB | RCSB PDB [15]; PDBe [16]; PDBj [17] |
| PDBsum | structure summary [18] |

| CRISPR-associated protein Cse1 | |
|---------------------------------------|-------------------------------------|
| Identifiers | |
| Symbol | CRISPR_Cse1 |
| Pfam | PF09481 [19] |
| InterPro | IPR013381 [20] |
| CDD | cd09729 [21] |
| Available protein structures: | |
| Pfam | structures [22] |
| PDB | RCSB PDB [23]; PDBe [24]; PDBj [25] |
| PDBsum | structure summary [26] |

| CRISPR-associated protein Cse2 | |
|---------------------------------------|-------------------------------------|
| Identifiers | |
| Symbol | CRISPR_Cse2 |
| Pfam | PF09485 [27] |
| InterPro | IPR013382 [28] |
| CDD | cd09670 [29] |
| Available protein structures: | |
| Pfam | structures [30] |
| PDB | RCSB PDB [31]; PDBe [32]; PDBj [33] |
| PDBsum | structure summary [34] |

Mechanism

Acquisition of Spacers into CRISPR loci

Capturing invading DNA into a CRISPR locus in the form of a spacer is the first stage in the immune response. The prevalence of cas1 and cas2 was the first clue that they were involved in spacer acquisition as all CRISPRs shared the regular repeating structure. Mutation studies confirmed this hypothesis as removal of cas1 or cas2 abrogated spacer acquisition, without affecting CRISPR immune response. The exact function of Cas1 and Cas2 is unknown, however a number of Cas1 proteins have been biochemically characterised and their structures resolved. Cas1 proteins have very diverse amino acid sequences, however their crystal structures are strikingly similar and all purified Cas1 proteins are metal-dependent nucleases that bind to DNA in a sequence-independent manner. Representative Cas2 proteins have also been characterised and possess either ssRNA or dsDNA specific endoribonuclease activity. The functional data and genetic mutation studies suggests that Cas1 and Cas2 excise fragments of invading DNA and insert them into CRISPR arrays.

Bioinformatic analysis of regions of phage genomes that were excised as spacers (termed protospacers) revealed that they were not randomly distributed in but instead were found adjacent to short (3 – 5 bp) DNA sequences termed PAMs (protospacer adjacent motifs). Analysis of CRISPR-Cas systems from the three major divisions have shown PAMs to be important for type I, type II but not type III systems during the spacer acquisition process. In type I and type II systems, protospacers are excised at positions adjacent to a PAM sequence, with the other end of the spacer cut using a ruler mechanism inherent to the Cas1 protein, thus maintaining the regularity of the spacer size in the CRISPR array. The conservation of the PAM sequence differs between CRISPR-Cas systems and appears to be evolutionarily linked to cas1 and the leader sequence.

New spacers are added to a CRISPR array in a directional manner, occurring preferentially but not exclusively adjacent to the leader sequence. Analysis of the type I-E system from *E. coli* have demonstrated that the first direct repeat, adjacent to the leader sequence is copied, with the newly acquired spacer inserted between the first and second direct repeats. The PAM sequence also appears to be important during spacer insertion in type I-E systems. The PAM sequence of the I-E system contains a strongly conserved final nucleotide (adjacent to the first nucleotide of the protospacer) and it has been shown that this nucleotide becomes the final base in the first direct repeat. This suggests that the spacer acquisition machinery generates single stranded overhangs in the second-to-last position of the direct repeat and in the PAM during spacer insertion. However, not all CRISPR-Cas systems appear to share this mechanism as PAMs characterised in other organisms do not show the same level of conservation in the final position. It is likely that in those systems, a blunt end is generated at the very end of the direct repeat and the protospacer during acquisition. Recent analysis of *Sulfolobus solfataricus* CRISPRs revealed further complexities to the canonical model of spacer insertion as one of its six CRISPR loci inserted new spacers randomly throughout its CRISPR array, as opposed to inserting closest to the leader sequence.

It has been noted in a number of CRISPRs that they contain many spacers to the same phage. The mechanism that causes this phenomenon has recently been elucidated in the type I-E system of *E. coli*. A significant enhancement in spacer acquisition has been detected where there are already spacers targeting the phage, even mismatches to the protospacer. This ‘priming’ requires both the Cas proteins involved in acquisition and interference to interact with each other. Newly acquired spacers that result from the priming mechanism are always found on the same strand as the original spacer that caused the priming. This observation has led to the hypothesis that the acquisition machinery slides along the foreign DNA after priming to find a new protospacer.

Interference stage

The CRISPR immune response occurs through two steps: CRISPR-RNA (crRNA) biogenesis and crRNA-guided interference. A CRISPR array is transcribed from a promoter in the leader into a single long transcript. This transcript is processed by cleavage inside the repeat sequence to form crRNAs. The mechanisms to produce mature crRNAs differ greatly between the three main CRISPR-Cas systems. In both type I-E and type I-F systems, the proteins Cas6e and Cas6f respectively, recognise stem-loops created by the palindromic nature of the direct repeats. These proteins cleave the primary transcript at the junction between double-stranded and single-stranded RNA, leaving an 8 nt 5'-handle originating from the repeat on mature crRNAs along with a single spacer sequence. Type III systems also use Cas6, however the repeats found in type III systems do not produce stem-loops, instead cleavage occurs by the primary transcript wrapping around the Cas6 to allow cleavage 8 nt upstream of the repeat spacer junction. Type II systems lack the Cas6 gene and instead utilize RNaseIII for cleavage. Functional type II systems encode an extra small RNA that is complementary to the repeat sequence, known as a trans-activating RNA (tracrRNA). Transcription of the tracrRNA and the primary CRISPR transcript results in base pairing and the formation of dsRNA at the repeat sequence, which is subsequently targeted by RNaseIII to produce crRNAs. Unlike the other two systems the crRNA does not contain the full spacer but instead is truncated at one end by 10 nt.

crRNAs associate with Cas proteins to form ribonucleotide complexes that recognize foreign nucleic acids. A number of phage and plasmid challenge experiments have shown that crRNAs show no preference between coding and non-coding strand, which is indicative of an RNA-guided DNA-targeting system. The type I-E complex (commonly referred to as Cascade) requires five Cas proteins arranged in a 'seahorse' conformation, bound to a single crRNA that runs down the spine. During the interference stage in type I systems the PAM sequence is recognized on the crRNA-complementary strand and is required along with crRNA annealing. In type I systems correct base pairing between the crRNA and the protospacer signals a conformational change in Cascade that recruits Cas3 for DNA degradation.

Type II systems rely on a single multifunctional protein, Cas9, for the interference step. Cas9 requires both the crRNA and the tracrRNA to function and cleaves DNA using its dual HNH and RuvC/RNaseH-like endonuclease domains. Basepairing between the PAM and the phage genome is also required in type II systems, however the PAM is recognized on the same strand as the crRNA (the opposite strand to type I systems).

Type III systems, like type I require a multi-protein complex to associate with the crRNA. Biochemical and structural analyses of complexes from *S. solfataricus* and *Pyrococcus furiosus* have elucidated that six or seven cas proteins bind to crRNAs, respectively. Surprisingly, the type III systems analysed from *S. solfataricus* and *P. furiosus* have both target the mRNA of phage/plasmids, which may make these systems uniquely capable of targeting RNA based phage genomes.

The mechanism for distinguishing self from foreign DNA during interference is built into the crRNAs and is therefore inferred to be common to all three systems. Even through the distinctive maturation process of each major type, all crRNAs contain a spacer sequence and some portion of the repeat at one or both ends. It is the partial repeat sequence that prevents the CRISPR-Cas system from targeting the chromosome as base pairing beyond the spacer sequence signals self and prevents DNA cleavage of the chromosome. RNA-guided CRISPR enzymes are classified as type V restriction enzymes.

Evolution and diversity

Studies of *Streptococcus thermophilus* first indicated how CRISPRs drive phage and bacterial evolution. A CRISPR spacer must correspond perfectly to the sequence of the target phage gene. Phages can continue to infect their hosts where there are point mutations in the spacer. Similar stringency is required in PAM or the strain will remain phage sensitive. The basic model of CRISPR evolution is one where newly incorporated spacers drive phages to mutate their genomes creating diversity in both the phage and host populations.

CRISPR evolution has been studied using comparative genomics of many strains of *S. thermophilus*, *Escherichia coli* and *Salmonella enterica*. A study of 124 strains of *S. thermophilus* showed that 26% of all spacers were unique and that different CRISPR loci showed different rates of new spacer acquisition. The results showed that particular CRISPR loci evolve more rapidly than others, which allowed the strains' phylogenetic relationships to be determined. A similar analysis of *E. coli* and *S. enterica* strains revealed that they evolved much slower than *S. thermophilus*. The latter's strains that had diverged 250 thousand years ago still contained the same spacer complement.

CRISPR diversity was studied in multiple environmental communities using metagenomics. Analysis of two acid mine drainage biofilms showed that one of the analyzed CRISPRs contained extensive deletions and spacer additions in comparison to the other biofilm, suggesting a higher phage activity/prevalence in one community compared to the other. In the oral cavity, a temporal study determined that 7-22% of spacers were shared between timepoints over 17 months within an individual and less than 2% of spacers were shared between different individuals at any single timepoint. From the same environment a single strain was tracked using PCR primers specific to its CRISPR. Unlike the broad-level results of spacer presence/absence, which showed significant diversity, this CRISPR added 3 spacers over 17 months, suggesting that even in an environment with significant CRISPR diversity some loci evolve slowly. CRISPRs have also been analysed from the metagenomes produced for the human microbiome project. Although most CRISPRs were body-site specific, some CRISPRs within a body site are widely shared among individuals. One of these CRISPR loci originated from streptococcal species and contained ~15,000 spacers, 50% of which were unique. Similar to the targeted studies of the oral cavity, some of the CRISPRs showed little evolution between timepoints.

CRISPR evolution has been studied in chemostats using *S. thermophilus* to explicitly examine the rate of spacer acquisition. Over a period of one week, strains of *S. thermophilus* acquired up to three spacers when challenged with a single phage. During the same time period the phage developed a number of single nucleotide polymorphisms that became fixed in the population, suggesting that CRISPR targeting had prevented all other phage types from replicating if they did not contain these mutations. Other experiments, also with *S. thermophilus*, showed that phages can still infect and replicate in hosts that have only one targeting spacer and that sensitive hosts can exist in environments with high phage titres. The chemostat results combined with the observational studies of CRISPRs suggest many nuances to the outcome of CRISPR and phage evolution.

Bioinformatic identification of CRISPRs in genomes and metagenomes

CRISPRs are widely distributed amongst the bacteria and archaea and show some sequence similarities, however their most notable characteristic is their repeating spacers and direct repeats. This characteristic makes CRISPRs easily identifiable in long sequences of DNA, since the number of repeat copies decreases the likelihood of a false positive match. There are currently three programs used for CRISPR repeat identification that search for regularly interspaced repeats in long sequences: CRT, PILER-CR and CRISPRfinder.

Analysis of CRISPRs in metagenomic data is more challenging, as CRISPR loci do not typically assemble due to their repetitive nature or through strain variation, which confuses assembly algorithms. Where there are many reference genomes available, PCR can be used to amplify CRISPR arrays and analyse spacer content. However, this approach will only yield information for CRISPRs specifically targeted and for organisms with sufficient representation in public databases to design reliable PCR primers.

The alternative approach is to extract and reconstruct CRISPR arrays from shotgun metagenomic data. Identification of CRISPR arrays from metagenomic reads is computationally more difficult, particularly with second generation sequencing technologies (e.g. 454, Illumina), as the short read lengths prevent more than two or three repeat units being present in a single read. CRISPR identification in raw reads has been achieved using purely de novo identification or by using direct repeat sequences in partially assembled CRISPR arrays from contigs and direct repeat sequences from published genomes as a hook for identifying direct repeats in individual reads.

Evolutionary significance

A bioinformatic study showed that CRISPRs are evolutionarily conserved and cluster into related types. Many show signs of a conserved secondary structure.

Through the CRISPR/Cas mechanism, bacteria can acquire immunity to certain phages and thus halt further transmission of targeted phages. For this reason, CRISPR/Cas can be described as a Lamarckian inheritance mechanism. Others investigated the coevolution of host and viral genomes by analysis of CRISPR sequences.

Cas9 proteins are highly enriched in pathogenic and commensal bacteria. CRISPR/Cas-mediated gene regulation may contribute to the regulation of endogenous bacterial genes, particularly during bacterial interaction with eukaryotic hosts. For example, Cas protein Cas9 of *Francisella novicida* uses a unique, small, CRISPR/Cas-associated RNA (scaRNA) to repress an endogenous transcript encoding a bacterial lipoprotein that is critical for *F. novicida* to dampen host response and promote virulence.

Applications

The proof-of-principle demonstration of selective engineered redirection of the CRISPR/Cas system in 2012 provided a first step toward realization of proposals for CRISPR-derived biotechnology:

- Artificial immunization against phage by introduction of engineered CRISPR loci in industrially important bacteria, including those used in food production and large-scale fermentation
- Genome engineering at cellular or organismic level by reprogramming a CRISPR/Cas system to achieve RNA-guided genome engineering. Proof of concept studies demonstrated examples both *in vitro* and *in vivo*
- Discrimination of bacterial strains by comparison of spacer sequences

Therapeutics

Editas Medicine, a \$43 million startup, aims to develop treatments that employ CRISPR/Cas to make edits to single base pairs and larger stretches of DNA. Inherited diseases such as cystic fibrosis and sickle-cell anemia are caused by single base pair mutations; CRISPR/Cas technology has the potential to correct these errors. The "corrected" gene remains in its normal location on its chromosome, which preserves the way the cell normally activates and/or inhibits its expression.

After harvesting blood cell precursors called hematopoietic stem cells from a patient's bone marrow, CRISPR gene surgery would correct the defective gene. Then the gene-corrected stem cells would be returned to the patient's marrow, which would then produce healthy red blood cells. Replacing 70% of the sickle cells would produce a cure.

Before it can be used clinically, the company must be able to guarantee that only the targeted region will be affected and determine how to deliver the therapy to a patient's cells.

Other pathologies potentially treatable by CRISPR include Huntington's disease, aging, schizophrenia and autism, not to mention modifying DNA in living embryos.

Improved targeting is required before CRISPR can be used in medical applications. Current guide RNAs may target sequences that differ by multiple base pairs from the intended sequence.

In 2014, UCSF researchers used CRISPR to create disease-free versions of induced pluripotent stem cells of beta thalassemia patients.

Mouse models

CRISPR simplifies creation of mouse models and reduces the time required to a matter of weeks from months or longer. Knockdown of endogenous genes has been achieved by transfection with a plasmid that contains a CRISPR area with a spacer, which inhibits a target gene. Injecting mouse zygotes with Cas9 and two guide RNAs was able to disable two genes with 80% efficiency. So-called homology-directed repair involves using Cas9 to "nick" DNA, to introduce new gene parts to the zygote.[Wikipedia:Citation needed](#)

Agriculture

In 2014, Chinese researcher Gao Caixia filed patents on the creation of a strain of wheat that is resistant to powdery mildew. The strain lacks genes that encode proteins that repress defenses against the mildew. The researchers deleted all three copies of the genes from wheat's hexaploid genome. The strain promises to reduce or eliminate the heavy use of fungicides to control the disease. Gao used the TALENs and CRISPR gene editing tools without adding or changing any other genes. No field trials are yet planned.

Functions

Editing

CRISPRs can add and delete base pairs at specifically targeted DNA loci. CRISPRs have been used to cut as many as five genes at once.

Reversible knockdown

Main article: CRISPR interference

"CRISPRi" like RNAi, turns off genes in a reversible fashion by targeting but not cutting a site. In bacteria, the presence of Cas9 alone is enough to block transcription, but for mammalian applications, a section of protein is added. Its guide RNA targets regulatory DNA, called promoters that immediately precede the gene target.

Activation

Main article: CRISPR interference

Cas9 was used to carry synthetic transcription factors (protein fragments that turn on genes) that activated specific human genes. The technique achieved a strong effect by targeting multiple CRISPR constructs to slightly different spots on the gene's promoter.

The genes included some tied to human diseases, including those involved in muscle differentiation, cancer, inflammation and producing fetal hemoglobin.

Use by phages

Another way for bacteria to defend against phage infection is by having chromosomal islands. A subtype of chromosomal islands called phage-inducible chromosomal island (PICI) is excised from bacterial chromosome upon phage infection and can inhibit phage replication. The mechanisms that induce PICI excision and how PICI inhibits phage replication are not well understood. One study showed that lytic ICP1 phage, which specifically targets *Vibrio cholerae* serogroup O1, has acquired a CRISPR/Cas system that targets a *V. cholera* PICI-like element. The system has 2 CRISPR loci and 9 Cas genes. It seems to be homologous to the 1-F system found in *Yersinia pestis*. Moreover, like the bacterial CRISPR/Cas system, ICP1 CRISPR/Cas can acquire new sequences, which allows the phage to co-evolve with its host.

Automation and library support

Free software is available to design RNA to target any desired gene. The Addgene repository offers academics the DNA to make their own CRISPR system for \$65. In 2013 Addgene distributed more than 10,000 CRISPR constructs. The facility has received CRISPR-enabling DNA sequences from 11 independent research teams.

Patent

A provisional US patent application on the use of the CRISPR system for editing genes and regulating gene expression was filed by the inventors on May 12, 2012. Subsequent applications were combined and on March 6, 2014 the resulting patent application was published by the USPTO. The patent rights have been assigned by the inventors to the Regents of the University of California and to the University of Vienna.

References

- [1] 71/79 Archaea, 463/1008 Bacteria CRISPRdb (<http://crispr.u-psud.fr/crispr/CRISPRdatabase.php>), Date: 19.6.2010
- [2] <http://pfam.xfam.org/family?acc=PF08798>
- [3] <http://pfam.xfam.org/clan/CL0362>
- [4] <http://www.ebi.ac.uk/interpro/entry/IPR010179>
- [5] <http://www.ncbi.nlm.nih.gov/Structure/cdd/cddsrvc.cgi?uid=cd09727>
- [6] <http://pfam.sanger.ac.uk/family/PF08798?tab=pdbBlock>
- [7] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF08798>
- [8] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF08798>
- [9] <http://pdjb.org/searchFor?query=PF08798>
- [10] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF08798
- [11] <http://pfam.xfam.org/family?acc=PF09827>
- [12] <http://www.ebi.ac.uk/interpro/entry/IPR019199>
- [13] <http://www.ncbi.nlm.nih.gov/Structure/cdd/cddsrvc.cgi?uid=cd09638>
- [14] <http://pfam.sanger.ac.uk/family/PF09827?tab=pdbBlock>
- [15] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF09827>
- [16] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF09827>
- [17] <http://pdjb.org/searchFor?query=PF09827>
- [18] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF09827
- [19] <http://pfam.xfam.org/family?acc=PF09481>
- [20] <http://www.ebi.ac.uk/interpro/entry/IPR013381>
- [21] <http://www.ncbi.nlm.nih.gov/Structure/cdd/cddsrvc.cgi?uid=cd09729>
- [22] <http://pfam.sanger.ac.uk/family/PF09481?tab=pdbBlock>
- [23] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF09481>
- [24] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF09481>
- [25] <http://pdjb.org/searchFor?query=PF09481>
- [26] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF09481
- [27] <http://pfam.xfam.org/family?acc=PF09485>
- [28] <http://www.ebi.ac.uk/interpro/entry/IPR013382>
- [29] <http://www.ncbi.nlm.nih.gov/Structure/cdd/cddsrvc.cgi?uid=cd09670>
- [30] <http://pfam.sanger.ac.uk/family/PF09485?tab=pdbBlock>
- [31] <http://www.rcsb.org/pdb/search/smartsSubquery.do?smartSearchSubtype=PfamIdQuery&pfamID=PF09485>
- [32] <http://www.ebi.ac.uk/pdbe-srv/PDBeXplore/pfam/?pfam=PF09485>
- [33] <http://pdjb.org/searchFor?query=PF09485>
- [34] http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/pdbsum/GetPfamStr.pl?pfam_id=PF09485

Further reading

- Sander, J. D.; Joung, J. K. (2014). "CRISPR-Cas systems for editing, regulating and targeting genomes". *Nature Biotechnology* **32** (4): 347–55. doi: 10.1038/nbt.2842 (<http://dx.doi.org/10.1038/nbt.2842>). PMID 24584096 (<http://www.ncbi.nlm.nih.gov/pubmed/24584096>).
- Terns, R. M.; Terns, M. P. (2014). "CRISPR-based technologies: Prokaryotic defense weapons repurposed". *Trends in Genetics* **30** (3): 111–8. doi: 10.1016/j.tig.2014.01.003 (<http://dx.doi.org/10.1016/j.tig.2014.01.003>). PMID 24555991 (<http://www.ncbi.nlm.nih.gov/pubmed/24555991>).
- Westra, E. R.; Buckling, A.; Fineran, P. C. (2014). "CRISPR–Cas systems: Beyond adaptive immunity". *Nature Reviews Microbiology*. doi: 10.1038/nrmicro3241 (<http://dx.doi.org/10.1038/nrmicro3241>).
- Horvath, P.; Romero, D. A.; Coûté-Monvoisin, A. -C.; Richards, M.; Deveau, H.; Moineau, S.; Boyaval, P.; Fremaux, C.; Barrangou, R. (2007). "Diversity, Activity, and Evolution of CRISPR Loci in *Streptococcus thermophilus*" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2238196>). *Journal of Bacteriology* **190** (4): 1401–1412. doi: 10.1128/JB.01415-07 (<http://dx.doi.org/10.1128/JB.01415-07>). PMC 2238196 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2238196>). PMID 18065539 (<http://www.ncbi.nlm.nih.gov/pubmed/18065539>).
- Deveau, H.; Barrangou, R.; Garneau, J. E.; Labonté, J.; Fremaux, C.; Boyaval, P.; Romero, D. A.; Horvath, P.; Moineau, S. (2007). "Phage Response to CRISPR-Encoded Resistance in *Streptococcus thermophilus*" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2238228>). *Journal of Bacteriology* **190** (4): 1390–1400. doi: 10.1128/JB.01412-07 (<http://dx.doi.org/10.1128/JB.01412-07>). PMC 2238228 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2238228>). PMID 18065545 (<http://www.ncbi.nlm.nih.gov/pubmed/18065545>).
- Andersson, A. F.; Banfield, J. F. (2008). "Virus Population Dynamics and Acquired Virus Resistance in Natural Microbial Communities". *Science* **320** (5879): 1047–1050. doi: 10.1126/science.1157358 (<http://dx.doi.org/10.1126/science.1157358>). PMID 18497291 (<http://www.ncbi.nlm.nih.gov/pubmed/18497291>).
- Hale, C.; Kleppe, K.; Terns, R. M.; Terns, M. P. (2008). "Prokaryotic silencing (psi)RNAs in *Pyrococcus furiosus*" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2590957>). *RNA* **14** (12): 2572–2579. doi: 10.1261/rna.1246808 (<http://dx.doi.org/10.1261/rna.1246808>). PMC 2590957 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2590957>). PMID 18971321 (<http://www.ncbi.nlm.nih.gov/pubmed/18971321>).
- Carte, J.; Wang, R.; Li, H.; Terns, R. M.; Terns, M. P. (2008). "Cas6 is an endoribonuclease that generates guide RNAs for invader defense in prokaryotes" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2607076>). *Genes & Development* **22** (24): 3489–3496. doi: 10.1101/gad.1742908 (<http://dx.doi.org/10.1101/gad.1742908>). PMC 2607076 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2607076>). PMID 19141480 (<http://www.ncbi.nlm.nih.gov/pubmed/19141480>).
- Shah, S. A.; Hansen, N. R.; Garrett, R. A. (2009). "Distribution of CRISPR spacer matches in viruses and plasmids of crenarchaeal acidothermophiles and implications for their inhibitory mechanism". *Biochemical Society Transactions* **37** (Pt 1): 23–28. doi: 10.1042/BST0370023 (<http://dx.doi.org/10.1042/BST0370023>). PMID 19143596 (<http://www.ncbi.nlm.nih.gov/pubmed/19143596>).
- Lillestøl, R. K.; Shah, S. A.; Brügger, K.; Redder, P.; Phan, H.; Christiansen, J.; Garrett, R. A. (2009). "CRISPR families of the crenarchaeal genus *Sulfolobus*: Bidirectional transcription and dynamic properties". *Molecular Microbiology* **72** (1): 259–272. doi: 10.1111/j.1365-2958.2009.06641.x (<http://dx.doi.org/10.1111/j.1365-2958.2009.06641.x>). PMID 19239620 (<http://www.ncbi.nlm.nih.gov/pubmed/19239620>).
- Mojica, F. J. M.; Diez-Villasenor, C.; Garcia-Martinez, J.; Almendros, C. (2009). "Short motif sequences determine the targets of the prokaryotic CRISPR defence system". *Microbiology* **155** (3): 733–740. doi: 10.1099/mic.0.023960-0 (<http://dx.doi.org/10.1099/mic.0.023960-0>). PMID 19246744 (<http://www.ncbi.nlm.nih.gov/pubmed/19246744>).
- Van Der Ploeg, J. R. (2009). "Analysis of CRISPR in *Streptococcus mutans* suggests frequent occurrence of acquired immunity against infection by M102-like bacteriophages". *Microbiology* **155** (6): 1966–1976. doi: 10.1099/mic.0.027508-0 (<http://dx.doi.org/10.1099/mic.0.027508-0>). PMID 19383692 (<http://www.ncbi.nlm.nih.gov/pubmed/19383692>).

- nlm.nih.gov/pubmed/19383692).
- Hale, C. R.; Zhao, P.; Olson, S.; Duff, M. O.; Graveley, B. R.; Wells, L.; Terns, R. M.; Terns, M. P. (2009). "RNA-Guided RNA Cleavage by a CRISPR RNA-Cas Protein Complex" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2951265>). *Cell* **139** (5): 945–956. doi: 10.1016/j.cell.2009.07.040 (<http://dx.doi.org/10.1016/j.cell.2009.07.040>). PMC 2951265 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2951265>). PMID 19945378 (<http://www.ncbi.nlm.nih.gov/pubmed/19945378>).
 - Van Der Oost, J.; Brouns, S. J. J. (2009). "RNAi: Prokaryotes Get in on the Act". *Cell* **139** (5): 863–865. doi: 10.1016/j.cell.2009.11.018 (<http://dx.doi.org/10.1016/j.cell.2009.11.018>). PMID 19945373 (<http://www.ncbi.nlm.nih.gov/pubmed/19945373>).
 - Marraffini, L. A.; Sontheimer, E. J. (2010). "Self versus non-self discrimination during CRISPR RNA-directed immunity" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2813891>). *Nature* **463** (7280): 568–571. doi: 10.1038/nature08703 (<http://dx.doi.org/10.1038/nature08703>). PMC 2813891 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2813891>). PMID 20072129 (<http://www.ncbi.nlm.nih.gov/pubmed/20072129>).
 - Karginov, F. V.; Hannon, G. J. (2010). "The CRISPR System: Small RNA-Guided Defense in Bacteria and Archaea" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2819186>). *Molecular Cell* **37** (1): 7–19. doi: 10.1016/j.molcel.2009.12.033 (<http://dx.doi.org/10.1016/j.molcel.2009.12.033>). PMC 2819186 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2819186>). PMID 20129051 (<http://www.ncbi.nlm.nih.gov/pubmed/20129051>).
 - Pul, Ü.; Wurm, R.; Arslan, Z.; Geissen, R.; Hofmann, N.; Wagner, R. (2010). "Identification and characterization of *E. Coli* CRISPR-cas promoters and their silencing by H-NS". *Molecular Microbiology* **75** (6): 1495–1512. doi: 10.1111/j.1365-2958.2010.07073.x (<http://dx.doi.org/10.1111/j.1365-2958.2010.07073.x>). PMID 20132443 (<http://www.ncbi.nlm.nih.gov/pubmed/20132443>).
 - Diez-Villasenor, C.; Almendros, C.; Garcia-Martinez, J.; Mojica, F. J. M. (2010). "Diversity of CRISPR loci in *Escherichia coli*". *Microbiology* **156** (5): 1351–1361. doi: 10.1099/mic.0.036046-0 (<http://dx.doi.org/10.1099/mic.0.036046-0>). PMID 20133361 (<http://www.ncbi.nlm.nih.gov/pubmed/20133361>).
 - Deveau, H. L. N.; Garneau, J. E.; Moineau, S. (2010). "CRISPR/Cas System and Its Role in Phage-Bacteria Interactions". *Annual Review of Microbiology* **64**: 475–493. doi: 10.1146/annurev.micro.112408.134123 (<http://dx.doi.org/10.1146/annurev.micro.112408.134123>). PMID 20528693 (<http://www.ncbi.nlm.nih.gov/pubmed/20528693>).
 - Koonin, E. V.; Makarova, K. S. (2009). "CRISPR-Cas: An adaptive immunity system in prokaryotes" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2884157>). *F1000 Biology Reports* **1**: 95. doi: 10.3410/B1-95 (<http://dx.doi.org/10.3410/B1-95>). PMC 2884157 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2884157>). PMID 20556198 (<http://www.ncbi.nlm.nih.gov/pubmed/20556198>).
 - Touchon, M.; Rocha, E. P. C. (2010). "The Small, Slow and Specialized CRISPR and Anti-CRISPR of *Escherichia* and *Salmonella*" (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2886076>). In Randau, Lennart. *PLoS ONE* **5** (6): e11126. doi: 10.1371/journal.pone.0011126 (<http://dx.doi.org/10.1371/journal.pone.0011126>). PMC 2886076 (<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2886076>). PMID 20559554 (<http://www.ncbi.nlm.nih.gov/pubmed/20559554>).

External links

- E-CRISP.org A comprehensive software for CRISPR gRNA design (<http://www.e-crisp.org>)
- CRISPR Design Tool (http://www.broadinstitute.org/mpg/crispr_design/faq.php)
- CRISPR Design Tool with scoring algorithms (<http://www.dna20.com/eCommerce/startCas9>)
- Tool for finding CRISPRs (<http://crispr.u-psud.fr/Server/CRISPRfinder.php>)
- Tool for finding CRISPR targets in other nucleic acids (<http://bioanalysis.otago.ac.nz/CRISPRTarget>)
- Rfam page for the CRISPR entries (<http://rfam.sanger.ac.uk/search?type?paths=Gene;+CRISPR;>)

Piwi-interacting RNA

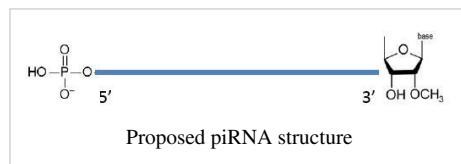
"piRNA" redirects here. For the software package, see Partition function for Interacting RNAs.

Piwi-interacting RNA (piRNA) is the largest class of small non-coding RNA molecules expressed in animal cells.^{[1][2]} piRNAs form RNA-protein complexes through interactions with piwi proteins. These piRNA complexes have been linked to both epigenetic and post-transcriptional gene silencing of retrotransposons and other genetic elements in germ line cells, particularly those in spermatogenesis.^[3] They are distinct from microRNA (miRNA) in size (26–31 nt rather than 21–24 nt), lack of sequence conservation, and increased complexity.

It remains unclear how piRNAs are generated, but potential methods have been suggested, and it is certain their biogenesis pathway is distinct from miRNA and siRNA, while rasiRNAs are a piRNA subspecies.^[4]

Characteristics

piRNAs have been identified in both vertebrates and invertebrates, and although biogenesis and modes of action do vary somewhat between species, a number of features are conserved. piRNAs have no clear secondary structure motifs, the length of a piRNA is, by definition, between 26 and 31 nucleotides, and the bias for a 5' uridine is common



to piRNAs in both vertebrates and invertebrates. piRNAs in *Caenorhabditis elegans* have a 5' monophosphate and a 3' modification that acts to block either the 2' or 3' oxygen,^[5] and this has also been confirmed to exist in *Drosophila melanogaster*,^[6] zebrafish,^[7] mice^[8] and rats. This 3' modification is a 2'-O-methylation; the reason for this modification is not clear, but it has been suggested to increase piRNA stability.^[9] It is thought that there are many hundreds of thousands of different piRNA species found in mammals.^[10] Thus far, over 50,000 unique piRNA sequences have been discovered in mice and more than 13,000 in *D. melanogaster*.^[11]

Location

piRNAs are found in clusters throughout the genome; these clusters may contain as few as ten or up to many thousands of piRNAs^[12] and can vary in size from one to one hundred kb. The detection and annotation of piRNA clusters in genomes based on bioinformatic methods became more and more sophisticated during the past years. While the clustering of piRNAs is highly conserved across species, the sequences are not.^[13] While *D. melanogaster* and vertebrate piRNAs have been located in areas lacking any protein coding genes,^[14] piRNAs in *C. elegans* have been identified amidst protein coding genes.

In mammals, piRNAs are found both in testes^[15] and ovaries,^[16] although they only seem to be required in males. In invertebrates, piRNAs have been detected in both the male and female germlines.

At the cellular level, piRNAs have been found within both nuclei and cytoplasm, suggesting that piRNA pathways may function in both of these areas and, therefore, may have multiple effects.^[17]

Biogenesis

The biogenesis of piRNAs is not yet fully understood, although possible mechanisms have been proposed. piRNAs show a significant strand bias, that is, they are derived from one strand of DNA only, and this may indicate that they are the product of long single stranded precursor molecules. A primary processing pathway is suggested to be the only pathway used to produce pachytene piRNAs; in this mechanism, piRNA precursors are transcribed resulting in piRNAs with a tendency to target 5' uridines.^{[18][19]} Also proposed is a 'Ping Pong' mechanism wherein primary piRNAs recognise their complementary targets and cause the recruitment of piwi proteins. This results in the cleavage of the transcript at a point ten nucleotides from the 5' end of the primary piRNA, producing the secondary piRNA. These secondary piRNAs are targeted toward sequences that possess an adenine at the tenth position. Since the piRNA involved in the ping pong cycle directs its attacks on transposon transcripts, the ping pong cycle acts only at the level of transcription. One or both of these mechanisms may be acting in different species; *C. elegans*, for instance, does have piRNAs, but does not appear to use the ping pong mechanism at all.

A significant number of piRNAs identified in zebrafish and *D. melanogaster* contain adenine at their tenth position, and this has been interpreted as possible evidence of a conserved biosynthetic mechanism across species. Ping-pong signatures have been identified in very primitive animals such as sponges and cnidarians, pointing to the existence of the ping-pong cycle already in the early branches of metazoans.^[20]

Function

The wide variation in piRNA sequences and piwi function over species contributes to the difficulty in establishing the functionality of piRNAs.^[21] However, like other small RNAs, piRNAs are thought to be involved in gene silencing, specifically the silencing of transposons. The majority of piRNAs are antisense to transposon sequences, suggesting that transposons are the piRNA target. In mammals it appears that the activity of piRNAs in transposon silencing is most important during the development of the embryo, and in both *C. elegans* and humans, piRNAs are necessary for spermatogenesis.

RNA silencing

piRNA has a role in RNA silencing via the formation of an RNA-induced silencing complex (RISC). piRNAs interact with piwi proteins that are part of a family of proteins called the Argonautes. These are active in the testes of mammals and are required for germ-cell and stem-cell development in invertebrates. Three piwi subfamily proteins - MIWI, MIWI2 and MILI - have been found to be essential for spermatogenesis in mice. piRNAs direct the piwi proteins to their transposon targets. A decrease or absence of PIWI protein expression is correlated with an increased expression of transposons. Transposons have a high potential to cause deleterious effect on their host, and, in fact, mutations in piRNA pathways are found to reduce fertility in *D. melanogaster*. However, piRNA pathway mutations in mice do not demonstrate reduced fertility; this may indicate redundancies to the piRNA system. Further, it is thought that piRNA and endogenous small interfering RNA (endo-siRNA) may have comparable and even redundant functionality in transposon control in mammalian oocytes.

piRNAs appear to have an impact on particular methyltransferases that perform the methylations which are required to recognise and silence transposons, but this relationship is not well understood.

Epigenetic effects

piRNAs can be transmitted maternally, and based on research in *D. melanogaster*, piRNAs may be involved in maternally derived epigenetic effects. The activity of specific piRNAs in the epigenetic process also requires interactions between piwi proteins and HP1a, as well as other factors.

Recent discovery also show, the existence of snoRNA, microRNA, piRNA characteristics in a novel non-coding RNA: x-ncRNA and its biological implication in *Homo sapiens*.

Accessory Proteins of the piRNA Pathway

Genetic screens examining fertility defects identified a number of proteins that are not Piwi-clade Argonautes, yet produce the same sterility phenotypes as Piwi mutants.

Drosophila Tudor Domain Proteins

Many factors required for the piRNA pathway in *Drosophila* contain Tudor domains that are known to bind symmetrically dimethylated arginine residues (sDMA) present in methylation motifs of Piwi proteins. Piwi proteins are symmetrically dimethylated by the PRMT5 methylosome complex, consisting of Valois (MEP50) and Capsulène (dart5; PRMT5).

- Tudor
- Qin/Kumo
- Spindle-E
- Krimper
- Tejas
- Vreteno
- Papi
- Yb (*fs(1)Yb*)
- BrotherOfYb
- SisterOfYb

Non-Tudor *Drosophila* piRNA pathway proteins

- Vasa
- Maelstrom

***Drosophila* Nuclear piRNA pathway proteins**

- Rhino (HP1D)
- Deadlock
- Cutoff
- SetDB1
- SuVar3-9

Investigation

Major advances in the piRNA field have been achieved thanks to the use of next-generation sequencing techniques, such as Solexa and 454. These techniques allow analysis of highly complex and heterogeneous RNA populations like piRNAs. Due to their small size, expression and amplification of small RNAs can be challenging, so specialised PCR-based methods have been developed in response to this difficulty.^{[22][23]}

References

- [1] Molecular Biology Select. Cell, 2006. 126(2): p. 223, 225-223, 225.
- [2] Seto, A.G., R.E. Kingston, and N.C. Lau, The Coming of Age for Piwi Proteins. Molecular Cell, 2007. 26(5): p. 603-609.
- [3] Siomi MC, Sato K, Pezic D, Aravin AA: PIWI-interacting small RNAs: the vanguard of genome defence. Nat Rev Mol Cell Biol 2011, 12:246-258.
- [4] Klattenhoff, C. and W. Theurkauf, Biogenesis and germline functions of piRNAs. Development, 2008. 135(1): p. 3-9.
- [5] Ruby, J.G., et al., Large-Scale Sequencing Reveals 21U-RNAs and Additional MicroRNAs and Endogenous siRNAs in *C. elegans*. 2006. 127(6): p. 1193-1207.
- [6] Vagin, V.V., et al., A Distinct Small RNA Pathway Silences Selfish Genetic Elements in the Germline. Science, 2006. 313(5785): p. 320-324.
- [7] Houwing, S., et al., A Role for Piwi and piRNAs in Germ Cell Maintenance and Transposon Silencing in Zebrafish. Cell, 2007. 129(1): p. 69-82.
- [8] Kirino, Y. and Z. Mourelatos, Mouse Piwi-interacting RNAs are 2[prime]-O-methylated at their 3[prime] termini. Nat Struct Mol Biol, 2007. 14(4): p. 347-348.

- [9] Faehnle, C.R. and L. Joshua-Tor, Argonautes confront new small RNAs. *Current Opinion in Chemical Biology*, 2007. 11(5): p. 569-577.
- [10] Das, P.P., et al., Piwi and piRNAs Act Upstream of an Endogenous siRNA Pathway to Suppress Tc3 Transposon Mobility in the *Caenorhabditis elegans* Germline. *Molecular Cell*, 2008. 31(1): p. 79-90.
- [11] Lin, H., et al., The role of the piRNA pathway in stem cell self-renewal. *Developmental Biology*, 2008. 319(2): p. 479-479.
- [12] O'Donnell, K.A. and J.D. Boeke, Mighty Piwis Defend the Germline against Genome Intruders. *Cell*, 2007. 129(1): p. 37-44.
- [13] Malone, C.D. and G.J. Hannon, Small RNAs as Guardians of the Genome. *Cell*, 2009. 136(4): p. 656-668.
- [14] Brennecke, J., et al., An Epigenetic Role for Maternally Inherited piRNAs in Transposon Silencing. *Science*, 2008. 322(5906): p. 1387-1392.
- [15] Aravin, A., et al., A novel class of small RNAs bind to MILI protein in mouse testes. *Nature*, 2006. 442(7099): p. 203-207.
- [16] Tam, O. H. et al. Pseudogene-derived small interfering RNAs regulate gene expression in mouse oocytes. *Nature* 453, 534–538 (2008).
- [17] Ruvkun, G., Tiny RNA: Where do we come from? What are we? Where are we going? *Trends in Plant Science*, 2008. 13(7): p. 313-316.
- [18] Aravin , A.A., et al., A piRNA Pathway Primed by Individual Transposons Is Linked to De Novo DNA Methylation in Mice. *Molecular Cell*, 2008. 31(6): p. 785-799.
- [19] Brennecke, J., et al., Discrete Small RNA-Generating Loci as Master Regulators of Transposon Activity in Drosophila. *Cell*, 2007. 128(6): p. 1089-1103.
- [20] Grimson, A., Srivastava, M., Fahey, B., Woodcroft, B.J., Chiang, H.R., King, N., Degnan, B.M., Rokhsar, D.S., and Bartel, D.P. (2008). Early origins and evolution of microRNAs and Piwi-interacting RNAs in animals. *Nature* 455, 1193–1197.
- [21] Wang, G. and V. Reinke, A *C. elegans* Piwi, PRG-1, Regulates 21U-RNAs during Spermatogenesis. *Current Biology*, 2008. 18(12): p. 861-867.
- [22] Ro, S., et al., A PCR-based method for detection and quantification of small RNAs. *Biochemical and Biophysical Research Communications*, 2006. 351(3): p. 756-763.
- [23] Tang, F., et al., A sensitive multiplex assay for piRNA expression. *Biochemical and Biophysical Research Communications*, 2008. 369(4): p. 1190-1194.

Further reading

- N.C. Lau et al., "Characterization of the piRNA Complex from Rat Testes," *Science* 313, 363 (2006)
- V.N. Kim, "Small RNAs Just Got Bigger: Piwi-Interacting RNAs (piRNAs) in Mammalian Testes," *Genes Dev.* 20, 1993 (2006)
- A. Girard et al., "A Germline-Specific Class of Small RNAs Binds Mammalian Piwi Proteins," *Nature* 442, 199 (2006)
- S.T. Grivna et al., "A Novel Class of Small RNAs in Mouse Spermatogenic Cells," *Genes Dev.* 20, 1709 (2006)
- T. Watanabe et al., "Identification and characterization of two novel classes of small RNAs in the mouse germline," *Genes Dev.* 20, 1732 (2006)
- M.A. Carmell et al., "MIWI2 Is Essential for Spermatogenesis and Repression of Transposons in the Mouse Male Germline," *Dev Cell.* 12, 503 (2007)

External links

- PingPongPro (<http://sourceforge.net/projects/pingpongpro/>) - a software for finding ping-pong signatures and ping-pong cycle activity
- piRNA Bank (<http://pirnabank.ibab.ac.in/>) A web resource on classified and clustered piRNAs
- proTRAC (<http://www.biomedcentral.com/1471-2105/13/5>) - a software for probabilistic piRNA cluster detection, visualization and analysis
- piRNA cluster - database (http://www.uni-mainz.de/FB/Biologie/Anthropologie/492_ENG_HTML.php)

Hologenome theory of evolution

The **hologenome theory of evolution** proposes that the object of natural selection is not the individual organism, but the holobiont, i.e. the organism together with its associated microbial communities.

Precursor: coral probiotic hypothesis

The hologenome theory of evolution originated in studies on coral reefs. Coral reefs are the largest structures created by living organisms, and contain abundant and highly complex microbial communities. A coral "head" is a colony of genetically identical polyps, which secrete an exoskeleton near the base. Depending on the species, the exoskeleton may be hard, based on calcium carbonate, or soft and proteinaceous. Over many generations, the colony creates a large skeleton that is characteristic of the species. Diverse forms of life take up residence in a coral colony, including photosynthetic algae such as *Symbiodinium*, as well as a wide range of bacteria including nitrogen fixers, and chitin decomposers, all of which form an important part of coral nutrition. The association between coral and its microbiota is species dependent, and different bacterial populations are found in mucus, skeleton and tissue from the same coral fragment.



Over the past several decades, major declines in coral populations have occurred. Climate change, water pollution and overfishing are three stress factors that have been described as leading to disease susceptibility. Over twenty different coral diseases have been described, but of these, only a handful have had their causative agents isolated and characterized.

Coral bleaching is the most serious of these diseases. In the Mediterranean Sea, the bleaching of *Oculina patagonica* was first described in 1994 and, through a rigorous application of Koch's Postulates, determined to be due to infection by *Vibrio shiloi*. From 1994 to 2002, bacterial bleaching of *O. patagonica* occurred every summer in the eastern Mediterranean. Surprisingly, however, after 2003, *O. patagonica* in the eastern Mediterranean has been resistant to *V. shiloi* infection, although other diseases still cause bleaching.

The surprise stems from the knowledge that corals are long lived, with lifespans on the order of decades, and do not have adaptive immune systems. Their innate immune systems do not produce antibodies, and they should seemingly not be able to respond to new challenges except over evolutionary time scales. Yet multiple researchers have documented variations in bleaching susceptibility that may be termed 'experience-mediated tolerance'. The puzzle of how corals managed to acquire resistance to a specific pathogen led Eugene Rosenberg and Ilana Zilber-Rosenberg to propose the Coral Probiotic Hypothesis. This hypothesis proposes that a dynamic relationship exists between corals and their symbiotic microbial communities. Beneficial mutations can arise and spread among the symbiotic microbes much faster than in the host corals. By altering its microbial composition, the "holobiont" can adapt to changing environmental conditions far more rapidly than by genetic mutation and selection in the host species alone. Extrapolating the Coral Probiotic Hypothesis to other organisms, including higher plants and animals, led to the proposal of the Hologenome Theory of Evolution.

Hologenome theory

Definition

The principles of the hologenome theory of evolution are as follows (condensed from Rosenberg *et al.*, 2007):

- "All animals and plants establish symbiotic relationships with microorganisms."
- "Different host species contain different symbiont populations and individuals of the same species can also contain different symbiont populations."
- "The association between a host organism and its microbial community affect both the host and its microbiota."
- "The genetic information encoded by microorganisms can change under environmental demands more rapidly, and by more processes, than the genetic information encoded by the host organism."
- "... the genome of the host can act in consortium with the genomes of the associated symbiotic microorganisms to create a hologenome. This hologenome...can change more rapidly than the host genome alone, thereby conferring greater adaptive potential to the combined holobiont evolution."
- "Each of these points taken together [led Rosenberg *et al.* to propose that] the holobiont with its hologenome should be considered as the unit of natural selection in evolution."

Some authors supplement the above principles with an additional one. If a given holobiont is to be considered a unit of natural selection:

- The hologenome must be heritable from generation to generation.

Horizontally versus vertically transmitted symbionts

Many case studies clearly demonstrate the importance of an organism's associated microbiota to its existence. (For example, see the numerous case studies in the Microbiome article.) However, horizontal *versus* vertical transmission of endosymbionts must be distinguished. Endosymbionts whose transmission is predominantly vertical may be considered as contributing to the heritable genetic variation present in a host species.

In the case of colonial organisms such as corals, the microbial associations of the colony persist even though individual members of the colony, reproducing asexually, live and die. Corals also have sexual mode of reproduction, resulting in planktonic larva; it is less clear whether microbial associations persist through this stage of growth. Also, the bacterial community of a colony may change with the seasons.

Many insects maintain heritable obligate symbiosis relationships with bacterial partners. For example, normal development of female wasps of the species *Asobara tabida* is dependent on *Wolbachia* infection. If "cured" of the infection, their ovaries degenerate. Transmission of the infection is vertical through the egg cytoplasm.

In contrast, many obligate symbiosis relationships have been described in the literature where transmission of the symbionts is via horizontal transfer. A well-studied example is the nocturnally feeding squid *Euprymna scolopes*, which camouflages its outline against the moonlit ocean surface by emitting light from its underside with the aid of the symbiotic bacterium *Vibrio fischeri*. The Rosenbergs cite this example within the context of the hologenome theory of evolution. Squid and bacterium maintain a highly co-evolved relationship. The newly hatched squid collects its bacteria from the sea water, and lateral transfer of symbionts between hosts permits faster transfer of beneficial mutations within a host species than are possible with mutations within the host genome.

Primary versus secondary symbionts

Another traditional distinction between endosymbionts has been between primary and secondary symbionts. Primary endosymbionts reside in specialized host cells that may be organized into larger, organ-like structures (in insects, the bacteriome). Associations between hosts and primary endosymbionts are usually ancient, with an estimated age of tens to hundreds of millions of years. According to endosymbiotic theory, extreme cases of primary endosymbionts include mitochondria, plastids (including chloroplasts), and possibly other organelles of eukaryotic cells. Primary endosymbionts are usually transmitted exclusively vertically, and the relationship is always mutualistic and generally obligate for both partners. Primary endosymbiosis is surprisingly common. An estimated 15% of insect species, for example, harbor this type of endosymbiont. In contrast, secondary endosymbiosis is often facultative, at least from the host point of view, and the associations are less ancient. Secondary endosymbionts do not reside in specialized host tissues, but may dwell in the body cavity dispersed in fat, muscle, or nervous tissue, or may grow within the gut. Transmission may be via vertical, horizontal, or both vertical and horizontal transfer. The relationship between host and secondary endosymbiont is not necessarily beneficial to the host; indeed, the relationship may be parasitic.



Green Vegetable Bug (*Nezara viridula*)
in Fronton, France

The distinction between vertical and horizontal transfer, and between primary and secondary endosymbiosis is not absolute, but follows a continuum, and may be subject to environmental influences. For example, in the stink bug *Nezara viridula*, the vertical transmission rate of symbionts, which females provide to offspring by smearing the eggs with gastric caeca, was 100% at 20 °C, but decreased to 8% at 30 °C. Likewise, in aphids, the vertical transmission of bacteriocytes containing the primary endosymbiont *Buchnera* is drastically reduced at high temperature. In like manner, the distinction between commensal, mutualistic, and parasitic relationships is also not absolute. An example is the relationship between legumes and rhizobial species: N₂ uptake is energetically more costly than the uptake of fixed nitrogen from the soil, so soil N is preferred if not limiting. During the early stages of nodule formation, the plant-rhizobial relationship actually resembles a pathogenesis more than it does a mutualistic association.

Neo-Lamarckism within a Darwinian context

Lamarckism, the concept that an organism can pass on characteristics that it acquired during its lifetime to its offspring (also known as inheritance of acquired characteristics or soft inheritance) incorporated two common ideas of its time:

- Use and disuse – individuals lose characteristics they do not require (or use) and develop characteristics that are useful.
- Inheritance of acquired traits – individuals inherit the traits of their ancestors.

Although Lamarckian theory was rejected by the neo-Darwinism of the modern evolutionary synthesis in which evolution occurs through random variations being subject to natural selection, the hologenome theory has aspects that harken back to Lamarckian concepts. In addition to the traditionally recognized modes of variation (*i.e.* sexual recombination, chromosomal rearrangement, mutation), the holobiont allows for two additional mechanisms of variation that are specific to the hologenome theory: (1) changes in the relative population of existing microorganisms (*i.e.* amplification and reduction) and (2) acquisition of novel strains from the environment, which may be passed on to offspring.

Changes in the relative population of existing microorganisms corresponds to Lamarckian "use and disuse", while the ability to acquire novel strains from the environment, which may be passed on to offspring, corresponds to

Lamarckian "inheritance of acquired traits". The hologenome theory, therefore, is said by its proponents to incorporate Lamarckian aspects within a Darwinian framework.

Additional case studies

The pea aphid *Acyrtosiphon pisum* maintains an obligate symbiotic relationship with the bacterium *Buchnera aphidicola*, which is transmitted maternally to the embryos that develop within the mother's ovarioles. Pea aphids live on sap, which is rich in sugars but deficient in amino acids. They rely on their *Buchnera* endosymbiotic population for essential amino acids, supplying in exchange nutrients as well as a protected intracellular environment that allows *Buchnera* to grow and reproduce. The relationship is actually more complicated than mutual nutrition; some strains of *Buchnera* increases host thermotolerance, while other strains do not. Both strains are present in field populations, suggesting that under some conditions, increased heat tolerance is advantageous to the host, while under other conditions, decreased heat tolerance but increased cold tolerance may be advantageous. One can consider the variant *Buchnera* genomes as alleles for the larger hologenome. The association between *Buchnera* and aphids began about 200 million years ago, with host and symbiont co-evolving since that time; in particular, it has been discovered that genome size in various *Buchnera* species has become extremely reduced, in some cases down to 450 kb, which is far smaller even than the 580 kb genome of *Mycoplasma genitalium*.



Pea aphids extracting sap from the stem and leaves of garden peas

Development of mating preferences, *i.e.* sexual selection, is considered to be an early event in speciation. In 1989, Dodd reported mating preferences in *Drosophila* that were induced by diet. It has recently been demonstrated that when otherwise identical populations of *Drosophila* were switched in diet between molasses medium and starch medium, that the "molasses flies" preferred to mate with other molasses flies, while the "starch flies" preferred to mate with other starch flies. This mating preference appeared after only one generation and was maintained for at least 37 generations. The origin of these differences were changes in the flies' populations of a particular bacterial symbiont, *Lactobacillus plantarum*. Antibiotic treatment abolished the induced mating preferences. It appears that the symbiotic bacteria changed the levels of cuticular hydrocarbon sex pheromones.

Zilber-Rosenberg and Rosenberg (2008) have tabulated many of the ways in which symbionts are transmitted and their contributions to the fitness of the holobiont, beginning with mitochondria found in all eukaryotes, chloroplast in plants, and then various associations described in specific systems. The microbial contributions to host fitness included provision of specific amino acids, growth at high temperatures, provision of nutritional needs from cellulose, nitrogen metabolism, recognition signals, more efficient food utilization, protection of eggs and embryos against metabolism, camouflage against predators, photosynthesis, breakdown of complex polymers, stimulation of the immune system, angiogenesis, vitamin synthesis, fiber breakdown, fat storage, supply of minerals from the soil, supply of organics, acceleration of mineralization, carbon cycling, and salt tolerance.

Criticism

The hologenome theory is debated. A major criticism by Ainsworth *et al.* has been their claim that *V. shiloi* was misidentified as the causative agent of coral bleaching, and that its presence in bleached *O. patagonica* was simply that of opportunistic colonization.

If this is true, the original observation that led to the theory would be invalid. On the other hand, Ainsworth *et al.* performed their samplings in 2005, two years after the Rosenberg group discovered *O. patagonica* no longer to be susceptible to *V. shiloi* infection; therefore their finding that bacteria are not the primary cause of present-day bleaching in Mediterranean coral *O. patagonica* should not be considered surprising. The rigorous satisfaction of Koch's postulates, as employed in Kushmaro *et al.* (1997), is generally accepted as providing a definitive identification of infectious disease agents.

Baird *et al.* (2009) have questioned basic assumptions made by Reshef *et al.* (2006) in presuming that (1) coral generation times are too slow to adjust to novel stresses over the observed time scales, and that (2) the scale of dispersal of coral larvae is too large to allow for adaptation to local environments. They may simply have underestimated the potential rapidity of conventional means of natural selection. In cases of severe stress, multiple cases have been documented of ecologically significant evolutionary change occurring over a handful of generations. Novel adaptive mechanisms such as switching symbionts might not be necessary for corals to adjust to rapid climate change or novel stressors.

Organisms in symbiotic relationships evolve to accommodate each other, and the symbiotic relationship increases the overall fitness of the participant species. Although the hologenome theory is still being debated, it has gained a significant degree of popularity within the scientific community as a way of explaining rapid adaptive changes that are difficult to accommodate within a traditional Darwinian framework.

References

Epigenetics

For the unfolding of an organism or the theory that plants and animals (including humans) develop in this way, see epigenesis (biology). For epigenetics in robotics, see developmental robotics.

Epigenetics is the study of changes in gene expression caused by certain base pairs in DNA, or RNA, being "turned off" or "turned on" again, through chemical reactions. In biology, and specifically genetics, epigenetics is mostly the study of heritable changes that are *not* caused by changes in the DNA sequence; to a lesser extent, epigenetics also describes the study of stable, long-term alterations in the transcriptional potential of a cell that are not necessarily heritable. Unlike simple genetics based on changes to the DNA sequence (the genotype), the changes in gene expression or cellular phenotype of epigenetics have other causes, thus use of the term *epi-* (Greek: *επί*- over, outside of, around) -*genetics*.

The term also refers to the changes themselves: functionally relevant changes to the genome that do not involve a change in the nucleotide sequence. Examples of mechanisms that produce such changes are DNA methylation and histone modification, each of which alters how genes are expressed without altering the underlying DNA sequence. Gene expression can be controlled through the action of repressor proteins that attach to silencer regions of the DNA. These epigenetic changes may last through cell divisions for the duration of the cell's life, and may also last for multiple generations even though they do not involve changes in the underlying DNA sequence of the organism; instead, non-genetic factors cause the organism's genes to behave (or "express themselves") differently. (There are objections to the use of the term epigenetic to describe chemical modification of histone, since it remains unclear whether or not histone modifications are heritable.)

One example of an epigenetic change in eukaryotic biology is the process of cellular differentiation. During morphogenesis, totipotent stem cells become the various pluripotent cell lines of the embryo, which in turn become fully differentiated cells. In other words, as a single fertilized egg cell – the zygote – continues to divide, the resulting daughter cells change into all the different cell types in an organism, including neurons, muscle cells, epithelium, endothelium of blood vessels, etc., by activating some genes while inhibiting the expression of others.

In 2011, it was demonstrated that the methylation of mRNA plays a critical role in human energy homeostasis. The obesity-associated FTO gene is shown to be able to demethylate N6-methyladenosine in RNA. This discovery launched the subfield of RNA epigenetics.

Historical usage of term

Epigenetics (as in "epigenetic landscape") was coined by C. H. Waddington in 1942 as a portmanteau of the words *epigenesis* and *genetics*. *Epigenesis* is an old^[1] word that has more recently been used (see *preformationism* for historical background) to describe the differentiation of cells from their initial totipotent state in embryonic development. When Waddington coined the term the physical nature of genes and their role in heredity was not known; he used it as a conceptual model of how genes might interact with their surroundings to produce a phenotype; he used the phrase "epigenetic landscape" as a metaphor for biological development. Waddington held that cell fates were established in development much like a marble rolls down to the point of lowest local elevation.^[2] Waddington suggested visualising increasing irreversibility of cell type differentiation as ridges rising between the valleys where the marbles (cells) are travelling.^[3] In recent times Waddington's notion of the epigenetic landscape has been rigorously formalized in the context of the systems dynamics state approach to the study of cell-fate.^[4]

The term "epigenetics" has also been used in developmental psychology to describe psychological development as the result of an ongoing, bi-directional interchange between heredity and the environment.^[5] Interactivist ideas of development have been discussed in various forms and under various names throughout the 19th and 20th centuries. An early version was proposed, among the founding statements in embryology, by Karl Ernst von Baer and

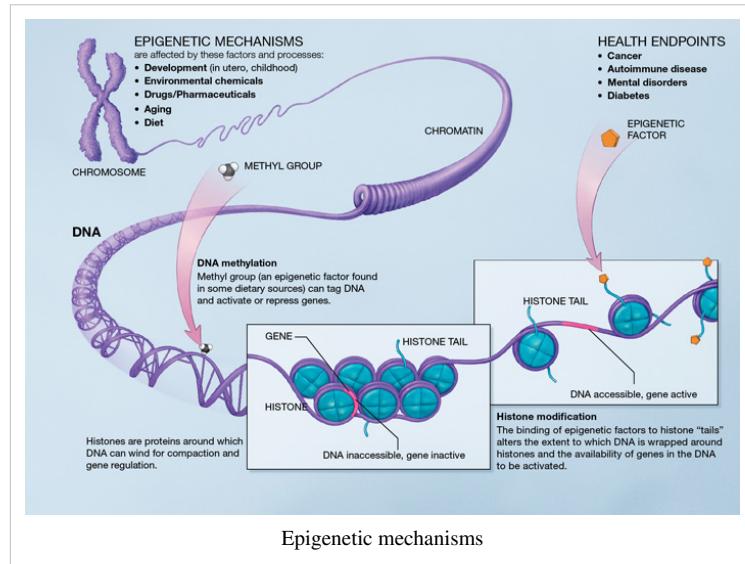
popularized by Ernst Haeckel. A radical epigenetic view (physiological epigenesis) was developed by Paul Wintrebert. Another variation, probabilistic epigenesis, was presented by Gilbert Gottlieb in 2003.^[6] This view encompasses all of the possible developing factors on an organism and how they not only influence the organism and each other but how the organism also influences its own development.

Noted developmental psychologist Erik Erikson used the term **epigenetic principle** in his book *Identity: Youth and Crisis* (1968), and used it to encompass the notion that we develop through an unfolding of our personality in predetermined stages, and that our environment and surrounding culture influence how we progress through these stages. This biological unfolding in relation to our socio-cultural settings is done in stages of psychosocial development, where "progress through each stage is in part determined by our success, or lack of success, in all the previous stages."^[7]

Contemporary usage of term

Robin Holliday defined epigenetics as "the study of the mechanisms of temporal and spatial control of gene activity during the development of complex organisms." Thus *epigenetic* can be used to describe anything other than DNA sequence that influences the development of an organism.

The more recent usage of the word in science has a stricter definition. It is, as defined by Arthur Riggs and colleagues, "the study of mitotically and/or meiotically heritable changes in gene function that cannot be explained by changes in DNA sequence." The Greek prefix *epi-* in *epigenetics* implies features that are "on top of" or "in addition to" genetics; thus *epigenetic* traits exist on top of or in addition to the traditional molecular basis for inheritance.



The term "epigenetics", however, has been used to describe processes which have not been demonstrated to be heritable such as histone modification; there are therefore attempts to redefine it in broader terms that would avoid the constraints of requiring heritability. For example, Sir Adrian Bird defined epigenetics as "the structural adaptation of chromosomal regions so as to register, signal or perpetuate altered activity states." This definition would be inclusive of transient modifications associated with DNA repair or cell-cycle phases as well as stable changes maintained across multiple cell generations, but exclude others such as templating of membrane architecture and prions unless they impinge on chromosome function. Such redefinitions however are not universally accepted and are still subject to dispute. The NIH "Roadmap Epigenomics Project," ongoing as of 2013, uses the following definition: "Epigenetics is an emerging frontier of science that involves the study of changes in the regulation of gene activity and expression that are not dependent on gene sequence. For purposes of this program, epigenetics refers to both heritable changes in gene activity and expression (in the progeny of cells or of individuals) and also stable, long-term alterations in the transcriptional potential of a cell that are not necessarily heritable. While epigenetics refers to the study of single genes or sets of genes, epigenomics refers to more global analyses of epigenetic changes across the entire genome."^[8]

In 2008, a consensus definition of the epigenetic trait, "stably heritable phenotype resulting from changes in a chromosome without alterations in the DNA sequence", was made at a Cold Spring Harbor meeting.

The similarity of the word to "genetics" has generated many parallel usages. The "epigenome" is a parallel to the word "genome", and refers to the overall epigenetic state of a cell. The phrase "genetic code" has also been adapted—the "epigenetic code" has been used to describe the set of epigenetic features that create different phenotypes in different cells. Taken to its extreme, the "epigenetic code" could represent the total state of the cell, with the position of each molecule accounted for in an *epigenomic map*, a diagrammatic representation of the gene expression, DNA methylation and histone modification status of a particular genomic region. More typically, the term is used in reference to systematic efforts to measure specific, relevant forms of epigenetic information such as the histone code or DNA methylation patterns.

Molecular basis

Epigenetic changes can modify the activation of certain genes, but not the sequence of DNA. Additionally, the chromatin proteins associated with DNA may be activated or silenced. This is why the differentiated cells in a multi-cellular organism express only the genes that are necessary for their own activity. Epigenetic changes are preserved when cells divide. Most epigenetic changes only occur within the course of one individual organism's lifetime, but, if gene inactivation occurs in a sperm or egg cell that results in fertilization, then some epigenetic changes can be transferred to the next generation. This raises the question of whether or not epigenetic changes in an organism can alter the basic structure of its DNA (see Evolution, below), a form of Lamarckism.

Specific epigenetic processes include paramutation, bookmarking, imprinting, gene silencing, X chromosome inactivation, position effect, reprogramming, transvection, maternal effects, the progress of carcinogenesis, many effects of teratogens, regulation of histone modifications and heterochromatin, and technical limitations affecting parthenogenesis and cloning.

DNA damage can also cause epigenetic changes. DNA damages are very frequent, occurring on average about 10,000 times a day per cell of the human body (see DNA damage (naturally occurring)). These damages are largely repaired, but at the site of a DNA repair, epigenetic changes can remain. In particular, a double strand break in DNA can initiate unprogrammed epigenetic gene silencing both by causing DNA methylation as well as by promoting silencing types of histone modifications (chromatin remodeling) (see next section). In addition, the enzyme Parp1 (poly(ADP)-ribose polymerase) and its product poly(ADP)-ribose (PAR) accumulate at sites of DNA damage as part of a repair process.^[9] This accumulation, in turn, directs recruitment and activation of the chromatin remodeling protein ALC1 that can cause nucleosome remodeling. Nucleosome remodeling has been found to cause, for instance, epigenetic silencing of DNA repair gene MLH1. DNA damaging chemicals, such as benzene, hydroquinone, styrene, carbon tetrachloride and trichloroethylene, cause considerable hypomethylation of DNA, some through the activation of oxidative stress pathways.

Foods are known to alter the epigenetics of rats on different diets. Some food components epigenetically increase the levels of DNA repair enzymes such as MGMT and MLH1 and p53. Other food components can reduce DNA damage, such as soy isoflavones and bilberry anthocyanins.

Epigenetic research uses a wide range of molecular biologic techniques to further our understanding of epigenetic phenomena, including chromatin immunoprecipitation (together with its large-scale variants ChIP-on-chip and ChIP-Seq), fluorescent in situ hybridization, methylation-sensitive restriction enzymes, DNA adenine methyltransferase identification (DamID) and bisulfite sequencing. Furthermore, the use of bioinformatic methods is playing an increasing role (computational epigenetics).

Computer simulations and molecular dynamics approaches revealed the atomistic motions associated with the molecular recognition of the histone tail through an allosteric mechanism.

Mechanisms

Several types of epigenetic inheritance systems may play a role in what has become known as cell memory, note however that not all of these are universally accepted to be examples of epigenetics.

DNA methylation and chromatin remodeling

Because DNA methylation and chromatin remodeling play such a central role in many types of epigenetic inheritance, the word "epigenetics" is sometimes used as a synonym for these processes. However, this can be misleading. Chromatin remodeling is not always inherited, and not all epigenetic inheritance involves chromatin remodeling.

Because the phenotype of a cell or individual is affected by which of its genes are transcribed, heritable transcription states can give rise to epigenetic effects. There are several layers of regulation of gene expression. One way that genes are regulated is through the remodeling of chromatin. Chromatin is the complex of DNA and the histone proteins with which it associates. If the way that DNA is wrapped around the histones changes, gene expression can change as well. Chromatin remodeling is accomplished through two main mechanisms:

1. The first way is post translational modification of the amino acids that make up histone proteins. Histone proteins are made up of long chains of amino acids. If the amino acids that are in the chain are changed, the shape of the histone might be modified. DNA is not completely unwound during replication. It is possible, then, that the modified histones may be carried into each new copy of the DNA.

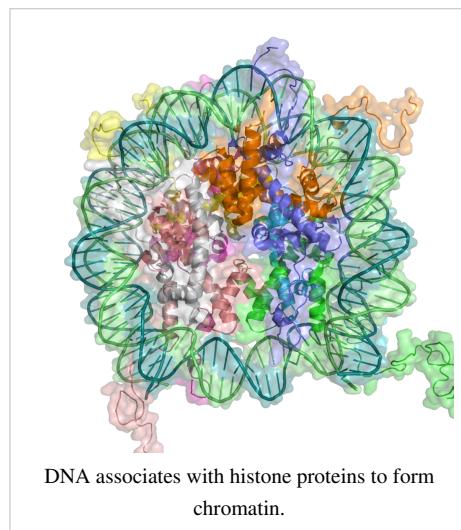
Once there, these histones may act as templates, initiating the surrounding new histones to be shaped in the new manner. By altering the shape of the histones around them, these modified histones would ensure that a lineage-specific transcription program is maintained after cell division.

2. The second way is the addition of methyl groups to the DNA, mostly at CpG sites, to convert cytosine to 5-methylcytosine. 5-Methylcytosine performs much like a regular cytosine, pairing with a guanine in double-stranded DNA. However, some areas of the genome are methylated more heavily than others, and highly methylated areas tend to be less transcriptionally active, through a mechanism not fully understood. Methylation of cytosines can also persist from the germ line of one of the parents into the zygote, marking the chromosome as being inherited from one parent or the other (genetic imprinting).

Mechanisms of heritability of histone state are not well understood; however, much is known about the mechanism of heritability of DNA methylation state during cell division and differentiation. Heritability of methylation state depends on certain enzymes (such as DNMT1) that have a higher affinity for 5-methylcytosine than for cytosine. If this enzyme reaches a "hemimethylated" portion of DNA (where 5-methylcytosine is in only one of the two DNA strands) the enzyme will methylate the other half.

Although histone modifications occur throughout the entire sequence, the unstructured N-termini of histones (called histone tails) are particularly highly modified. These modifications include acetylation, methylation, ubiquitylation, phosphorylation, sumoylation, ribosylation and citrullination. Acetylation is the most highly studied of these modifications. For example, acetylation of the K14 and K9 lysines of the tail of histone H3 by histone acetyltransferase enzymes (HATs) is generally related to transcriptional competence.

One mode of thinking is that this tendency of acetylation to be associated with "active" transcription is biophysical in nature. Because it normally has a positively charged nitrogen at its end, lysine can bind the negatively charged phosphates of the DNA backbone. The acetylation event converts the positively charged amine group on the side chain into a neutral amide linkage. This removes the positive charge, thus loosening the DNA from the histone.



When this occurs, complexes like SWI/SNF and other transcriptional factors can bind to the DNA and allow transcription to occur. This is the "cis" model of epigenetic function. In other words, changes to the histone tails have a direct effect on the DNA itself.

Another model of epigenetic function is the "trans" model. In this model, changes to the histone tails act indirectly on the DNA. For example, lysine acetylation may create a binding site for chromatin-modifying enzymes (or transcription machinery as well). This chromatin remodeler can then cause changes to the state of the chromatin. Indeed, a bromodomain — a protein domain that specifically binds acetyl-lysine — is found in many enzymes that help activate transcription, including the SWI/SNF complex. It may be that acetylation acts in this and the previous way to aid in transcriptional activation.

The idea that modifications act as docking modules for related factors is borne out by histone methylation as well. Methylation of lysine 9 of histone H3 has long been associated with constitutively transcriptionally silent chromatin (constitutive heterochromatin). It has been determined that a chromodomain (a domain that specifically binds methyl-lysine) in the transcriptionally repressive protein HP1 recruits HP1 to K9 methylated regions. One example that seems to refute this biophysical model for methylation is that tri-methylation of histone H3 at lysine 4 is strongly associated with (and required for full) transcriptional activation. Tri-methylation in this case would introduce a fixed positive charge on the tail.

It has been shown that the histone lysine methyltransferase (KMT) is responsible for this methylation activity in the pattern of histones H3 & H4. This enzyme utilizes a catalytically active site called the SET domain (Suppressor of variegation, Enhancer of zeste, Trithorax). The SET domain is a 130-amino acid sequence involved in modulating gene activities. This domain has been demonstrated to bind to the histone tail and causes the methylation of the histone.

Differing histone modifications are likely to function in differing ways; acetylation at one position is likely to function differently from acetylation at another position. Also, multiple modifications may occur at the same time, and these modifications may work together to change the behavior of the nucleosome. The idea that multiple dynamic modifications regulate gene transcription in a systematic and reproducible way is called the histone code, although the idea that histone state can be read linearly as a digital information carrier has been largely debunked. One of the best-understood systems that orchestrates chromatin-based silencing is the SIR protein based silencing of the yeast hidden mating type loci HML and HMR.

DNA methylation frequently occurs in repeated sequences, and helps to suppress the expression and mobility of 'transposable elements': Because 5-methylcytosine can be spontaneously deaminated (replacing nitrogen by oxygen) to thymidine, CpG sites are frequently mutated and become rare in the genome, except at CpG islands where they remain unmethylated. Epigenetic changes of this type thus have the potential to direct increased frequencies of permanent genetic mutation. DNA methylation patterns are known to be established and modified in response to environmental factors by a complex interplay of at least three independent DNA methyltransferases, DNMT1, DNMT3A, and DNMT3B, the loss of any of which is lethal in mice. DNMT1 is the most abundant methyltransferase in somatic cells, localizes to replication foci, has a 10–40-fold preference for hemimethylated DNA and interacts with the proliferating cell nuclear antigen (PCNA).

By preferentially modifying hemimethylated DNA, DNMT1 transfers patterns of methylation to a newly synthesized strand after DNA replication, and therefore is often referred to as the 'maintenance' methyltransferase. DNMT1 is essential for proper embryonic development, imprinting and X-inactivation. To emphasize the difference of this molecular mechanism of inheritance from the canonical Watson-Crick base-pairing mechanism of transmission of genetic information, the term 'Epigenetic templating' was introduced. Furthermore, in addition to the maintenance and transmission of methylated DNA states, the same principle could work in the maintenance and transmission of histone modifications and even cytoplasmic (structural) heritable states.^[10]

Histones H3 and H4 can also be manipulated through demethylation using histone lysine demethylase (KDM). This recently identified enzyme has a catalytically active site called the Jumonji domain (JmjC). The demethylation

occurs when JmjC utilizes multiple cofactors to hydroxylate the methyl group, thereby removing it. JmjC is capable of demethylating mono-, di-, and tri-methylated substrates.

Chromosomal regions can adopt stable and heritable alternative states resulting in bistable gene expression without changes to the DNA sequence. Epigenetic control is often associated with alternative covalent modifications of histones. The stability and heritability of states of larger chromosomal regions are suggested to involve positive feedback where modified nucleosomes recruit enzymes that similarly modify nearby nucleosomes. A simplified stochastic model for this type of epigenetics is found here.

It has been suggested that chromatin-based transcriptional regulation could be mediated by the effect of small RNAs. Small interfering RNAs can modulate transcriptional gene expression via epigenetic modulation of targeted promoters.

RNA transcripts and their encoded proteins

Sometimes a gene, after being turned on, transcribes a product that (directly or indirectly) maintains the activity of that gene. For example, Hnf4 and MyoD enhance the transcription of many liver- and muscle-specific genes, respectively, including their own, through the transcription factor activity of the proteins they encode. RNA signalling includes differential recruitment of a hierarchy of generic chromatin modifying complexes and DNA methyltransferases to specific loci by RNAs during differentiation and development. Other epigenetic changes are mediated by the production of different splice forms of RNA, or by formation of double-stranded RNA (RNAi). Descendants of the cell in which the gene was turned on will inherit this activity, even if the original stimulus for gene-activation is no longer present. These genes are often turned on or off by signal transduction, although in some systems where syncytia or gap junctions are important, RNA may spread directly to other cells or nuclei by diffusion. A large amount of RNA and protein is contributed to the zygote by the mother during oogenesis or via nurse cells, resulting in maternal effect phenotypes. A smaller quantity of sperm RNA is transmitted from the father, but there is recent evidence that this epigenetic information can lead to visible changes in several generations of offspring.

MicroRNAs

MicroRNAs (miRNAs) are members of non-coding RNAs that range in size from 17 to 25 nucleotides. As indicated by Wang et al.,^[11] miRNAs regulate a large variety of biological functions in plants and animals. So far, in 2013, about 2000 miRNAs have been discovered in humans and these can be found online in an miRNA database.^[12] Each miRNA expressed in a cell may target about 100 to 200 messenger RNAs that it downregulates.^[13] Most of the downregulation of mRNAs occurs by causing the decay of the targeted mRNA, while some downregulation occurs at the level of translation into protein.^[14]

It appears that about 60% of human protein coding genes are regulated by miRNAs.^[15] Many miRNAs are epigenetically regulated. About 50% of miRNA genes are associated with CpG islands, that may be repressed by epigenetic methylation. Transcription from methylated CpG islands is strongly and heritably repressed.^[16] Other miRNAs are epigenetically regulated by either histone modifications or by combined DNA methylation and histone modification.

sRNAs

sRNAs are small (50-250 nucleotides), highly structured, non-coding RNA fragments found in bacteria. They control gene expression including virulence genes in pathogens and are viewed as new targets in the fight against drug-resistant bacteria. They play an important role in many biological processes, binding to mRNA and protein targets in prokaryotes. Their phylogenetic analyses, for example through sRNA–mRNA target interactions or protein binding properties, are used to build comprehensive databases.^[17] sRNA-gene maps based on their targets in microbial genomes are also constructed.^[18]

Prions

For more details on this topic, see Fungal prions.

Prions are infectious forms of proteins. In general, proteins fold into discrete units that perform distinct cellular functions, but some proteins are also capable of forming an infectious conformational state known as a prion. Although often viewed in the context of infectious disease, prions are more loosely defined by their ability to catalytically convert other native state versions of the same protein to an infectious conformational state. It is in this latter sense that they can be viewed as epigenetic agents capable of inducing a phenotypic change without a modification of the genome.

Fungal prions are considered by some to be epigenetic because the infectious phenotype caused by the prion can be inherited without modification of the genome. PSI+ and URE3, discovered in yeast in 1965 and 1971, are the two best studied of this type of prion. Prions can have a phenotypic effect through the sequestration of protein in aggregates, thereby reducing that protein's activity. In PSI+ cells, the loss of the Sup35 protein (which is involved in termination of translation) causes ribosomes to have a higher rate of read-through of stop codons, an effect that results in suppression of nonsense mutations in other genes. The ability of Sup35 to form prions may be a conserved trait. It could confer an adaptive advantage by giving cells the ability to switch into a PSI+ state and express dormant genetic features normally terminated by stop codon mutations.

Structural inheritance systems

For more details on this topic, see Structural inheritance.

In ciliates such as *Tetrahymena* and *Paramecium*, genetically identical cells show heritable differences in the patterns of ciliary rows on their cell surface. Experimentally altered patterns can be transmitted to daughter cells. It seems existing structures act as templates for new structures. The mechanisms of such inheritance are unclear, but reasons exist to assume that multicellular organisms also use existing cell structures to assemble new ones.

Functions and consequences

Development

Somatic epigenetic inheritance through epigenetic modifications, particularly through DNA methylation and chromatin remodeling, is very important in the development of multicellular eukaryotic organisms. The genome sequence is static (with some notable exceptions), but cells differentiate into many different types, which perform different functions, and respond differently to the environment and intercellular signalling. Thus, as individuals develop, morphogens activate or silence genes in an epigenetically heritable fashion, giving cells a "memory". In mammals, most cells terminally differentiate, with only stem cells retaining the ability to differentiate into several cell types ("totipotency" and "multipotency"). In mammals, some stem cells continue producing new differentiated cells throughout life, such as in neurogenesis, but mammals are not able to respond to loss of some tissues, for example, the inability to regenerate limbs, which some other animals are capable of. Unlike animals, plant cells do not terminally differentiate, remaining totipotent with the ability to give rise to a new individual plant. While plants do utilise many of the same epigenetic mechanisms as animals, such as chromatin remodeling, it has been hypothesised that some kinds of plant cells do not use or require "cellular memories", resetting their gene expression patterns using positional information from the environment and surrounding cells to determine their fate.

Epigenetics can be divided into predetermined and probabilistic epigenesis. Predetermined epigenesis is a unidirectional movement from structural development in DNA to the functional maturation of the protein. "Predetermined" here means that development is scripted and predictable. Probabilistic epigenesis on the other hand is a bidirectional structure-function development with experiences and external molding development.

Medicine

Epigenetics has many and varied potential medical applications as it tends to be multidimensional in nature. Congenital genetic disease is well understood, and it is also clear that epigenetics can play a role, for example, in the case of Angelman syndrome and Prader-Willi syndrome. These are normal genetic diseases caused by gene deletions or inactivation of the genes, but are unusually common because individuals are essentially hemizygous because of genomic imprinting, and therefore a single gene knock out is sufficient to cause the disease, where most cases would require both copies to be knocked out.

Evolution

See also: Transgenerational epigenetics

Epigenetics can impact evolution when epigenetic changes are heritable. A sequestered germ line or Weismann barrier is specific to animals, and epigenetic inheritance is more common in plants and microbes. Eva Jablonka and Marion Lamb have argued that these effects may require enhancements to the standard conceptual framework of the modern evolutionary synthesis.^[19] Other evolutionary biologists have incorporated epigenetic inheritance into population genetics models or are openly skeptical.

Two important ways in which epigenetic inheritance can be different from traditional genetic inheritance, with important consequences for evolution, are that rates of epimutation can be much faster than rates of mutation and the epimutations are more easily reversible. An epigenetically inherited element such as the PSI+ system can act as a "stop-gap", good enough for short-term adaptation that allows the lineage to survive for long enough for mutation and/or recombination to genetically assimilate the adaptive phenotypic change. The existence of this possibility increases the evolvability of a species.

Current research findings and examples of effects

Epigenetic changes have been observed to occur in response to environmental exposure—for example, mice given some dietary supplements have epigenetic changes affecting expression of the agouti gene, which affects their fur color, weight, and propensity to develop cancer.

One study indicates that traumatic experiences can produce fearful memories which are passed to future generations via epigenetics. A study carried out on mice in 2013 found that mice could produce offspring which had an aversion to certain items which had been the source of negative experiences for their ancestors.^{[20][21]} Reports stated that:

For the study, author Brian Dias and co-author Kerry Ressler trained mice, using foot shocks, to fear an odour that resembles cherry blossoms. Later, they tested the extent to which the animals' offspring startled when exposed to the same smell. The younger generation had not even been conceived when their fathers underwent the training, and had never smelt the odour before the experiment.

The offspring of trained mice were "able to detect and respond to far less amounts of odour... suggesting they are more sensitive" to it, Ressler told AFP of the findings published in the journal Nature Neuroscience. They did not react the same way to other odours, and compared to the offspring of non-trained mice, their reaction to the cherry blossom whiff was about 200 percent stronger, he said.

The scientists then looked at a gene, M71, that governs the functioning of an odour receptor in the nose that responds specifically to the cherry blossom smell. The gene, inherited through the sperm of trained mice, had undergone no change to its DNA encoding, the team found. But the gene did carry epigenetic marks that could alter its behaviour and cause it to be "expressed more" in descendants, said Dias. This in turn caused a physical change in the brains of the trained mice, their sons and grandsons, who all had a larger glomerulus—a section in the olfactory (smell) unit of the brain.

In the case of humans with different environmental exposures, Fraga et al.^[22] studied young monozygotic (identical) twins and older monozygotic twins. They found that although such twins were epigenetically indistinguishable

during their early years, older twins had remarkable differences in the overall content and genomic distribution of 5-methylcytosine DNA and histone acetylation. The twin pairs who had spent less of their lifetime together and/or had greater differences in their medical histories were those who showed the largest differences in their levels of 5methylcytosine DNA and acetylation of histones H3 and H4.

More than 100 cases of transgenerational epigenetic inheritance phenomena have been reported in a wide range of organisms, including prokaryotes, plants, and animals. For instance, Mourning Cloak butterflies will change color through hormone changes in response to experimentation of varying temperatures.^[23]

Recent analyses have suggested that members of the APOBEC/AID family of cytosine deaminases are capable of simultaneously mediating genetic and epigenetic inheritance using similar molecular mechanisms.

Epigenetic effects in humans

Genomic imprinting and related disorders

Some human disorders are associated with genomic imprinting, a phenomenon in mammals where the father and mother contribute different epigenetic patterns for specific genomic loci in their germ cells. The best-known case of imprinting in human disorders is that of Angelman syndrome and Prader-Willi syndrome—both can be produced by the same genetic mutation, chromosome 15q partial deletion, and the particular syndrome that will develop depends on whether the mutation is inherited from the child's mother or from their father. This is due to the presence of genomic imprinting in the region. Beckwith-Wiedemann syndrome is also associated with genomic imprinting, often caused by abnormalities in maternal genomic imprinting of a region on chromosome 11.

Transgenerational epigenetic observations

See main article Transgenerational epigenetics

In the Överkalix study, Marcus Pembrey and colleagues observed that the paternal (but not maternal) grandsons^[24] of Swedish men who were exposed during preadolescence to famine in the 19th century were less likely to die of cardiovascular disease. If food was plentiful, then diabetes mortality in the grandchildren increased, suggesting that this was a transgenerational epigenetic inheritance.^[25] The opposite effect was observed for females—the paternal (but not maternal) granddaughters of women who experienced famine while in the womb (and therefore while their eggs were being formed) lived shorter lives on average.

Cancer and developmental abnormalities

A variety of compounds are considered as epigenetic carcinogens—they result in an increased incidence of tumors, but they do not show mutagen activity (toxic compounds or pathogens that cause tumors incident to increased regeneration should also be excluded). Examples include diethylstilbestrol, arsenite, hexachlorobenzene, and nickel compounds.

Many teratogens exert specific effects on the fetus by epigenetic mechanisms. While epigenetic effects may preserve the effect of a teratogen such as diethylstilbestrol throughout the life of an affected child, the possibility of birth defects resulting from exposure of fathers or in second and succeeding generations of offspring has generally been rejected on theoretical grounds and for lack of evidence. However, a range of male-mediated abnormalities have been demonstrated, and more are likely to exist. FDA label information for Vidaza, a formulation of 5-azacitidine (an unmethylatable analog of cytidine that causes hypomethylation when incorporated into DNA) states that "men should be advised not to father a child" while using the drug, citing evidence in treated male mice of reduced fertility, increased embryo loss, and abnormal embryo development.^[26] In rats, endocrine differences were observed in offspring of males exposed to morphine. In mice, second generation effects of diethylstilbestrol have been described occurring by epigenetic mechanisms.

Recent studies have shown that the mixed-lineage leukemia (MLL) gene causes leukemia by rearranging and fusing with other genes in different chromosomes, which is a process under epigenetic control.

Other investigations have concluded that alterations in histone acetylation and DNA methylation occur in various genes influencing prostate cancer. Gene expression in the prostate can be modulated by nutrition and lifestyle changes.

In 2008, the National Institutes of Health announced that \$190 million had been earmarked for epigenetics research over the next five years. In announcing the funding, government officials noted that epigenetics has the potential to explain mechanisms of aging, human development, and the origins of cancer, heart disease, mental illness, as well as several other conditions. Some investigators, like Randy Jirtle, PhD, of Duke University Medical Center, think epigenetics may ultimately turn out to have a greater role in disease than genetics.

DNA methylation in cancer

DNA methylation is an important regulator of gene transcription and a large body of evidence has demonstrated that aberrant DNA methylation is associated with unscheduled gene silencing, and the genes with high levels of 5-methylcytosine in their promoter region are transcriptionally silent. DNA methylation is essential during embryonic development, and in somatic cells, patterns of DNA methylation are in general transmitted to daughter cells with a high fidelity. Aberrant DNA methylation patterns have been associated with a large number of human malignancies and found in two distinct forms: hypermethylation and hypomethylation compared to normal tissue. Hypermethylation is one of the major epigenetic modifications that repress transcription via promoter region of tumour suppressor genes. Hypermethylation typically occurs at CpG islands in the promoter region and is associated with gene inactivation. Global hypomethylation has also been implicated in the development and progression of cancer through different mechanisms.

DNA repair epigenetics in cancer

Germ line (familial) mutations have been identified in 34 different DNA repair genes that cause a high risk of cancer, including, for example BRCA1 and ATM. These are listed in the article DNA repair-deficiency disorder. However, cancers caused by such germ line mutations make up only a very small proportion of cancers. For instance, germ line mutations cause only 2% to 5% of colon cancer cases.^[27]

Epigenetic reductions in expression of DNA repair genes, however, are very frequent in sporadic (non-germ line) cancers, as shown among some representative cancers in the table in this section, while mutations in DNA repair genes in sporadic cancer are very rare.^[28]

Epigenetic changes in DNA repair genes in sporadic cancers

| Cancer | Gene | Epigenetic change | Frequency | Ref. |
|----------------|--------|------------------------|-----------|---------|
| Breast | BRCA1 | CpG island methylation | 13% | 1 |
| | WRN | CpG island methylation | 17% | 2 |
| Ovarian | WRN | CpG island methylation | 36% | 3 |
| | BRCA1 | CpG island methylation | 5%-30% | 1,11,12 |
| | FANCF | CpG island methylation | 21% | 11 |
| | RAD51C | CpG island methylation | 3% | 11 |

| | | | | |
|------------------------------|-------|-------------------------|---------|----------|
| Colorectal | MGMT | CpG island methylation | 40%-90% | 4-8 |
| | WRN | CpG island methylation | 38% | 2 |
| | MLH1 | CpG island methylation | 2%-65% | 2,5,9 |
| | MSH2 | CpG island methylation | 13% | 6 |
| | ERCC1 | epigenetic type unknown | 100% | 10 |
| | Xpf | epigenetic type unknown | 55% | 10 |
| Head and neck | MGMT | CpG island methylation | 35%-57% | 13-16 |
| | MLH1 | CpG island methylation | 27%-33% | 17,19,20 |
| | NEIL1 | CpG island methylation | 62% | 13 |
| | FANCB | CpG island methylation | 46% | 13 |
| | MSH4 | CpG island methylation | 46% | 13 |
| | ATM | CpG island methylation | 25% | 18 |

References in the table are given here: 1,^[29] 2,^[30] 3,^[31] 4,^[32] 5,^[33] 6,^[34] 7,^[35] 8,^[36] 9,^[37] 10,^[38] 11,^[39] 12,^[40] 13,^[41] 14,^[42] 15,^[43] 16,^[44] 17,^[45] 18,^[46] 19,^[47] 20^[48]

Deficiencies in expression of DNA repair genes cause increased mutation rates. Mutations rates increase in mice defective for mismatch DNA repair genes PMS2, MLH1, MSH2, MSH3 or MSH6^{[49][50]} or for DNA repair gene BRCA2,^[51] while chromosomal rearrangements and aneuploidy are noted to increase in humans defective in DNA repair gene BLM.^[52] Thus, deficiency in DNA repair causes genome instability and this genome instability is likely the main underlying cause of the genetic alterations leading to cancer. In fact, as indicated by Nowak et al.^[53] through a mathematical calculation, the first event in many sporadic neoplasias is a heritable alteration that affects genetic instability, and we note that epigenetic defects in DNA repair are somatically heritable.

Variant histones H2A in cancer

The histone variants of the H2A family are highly conserved in mammals, playing critical roles in regulating many nuclear processes by altering chromatin structure. One of the key H2A variants, H2A.X, marks DNA damage, facilitating the recruitment of DNA repair proteins to restore genomic integrity. Another variant, H2A.Z, plays an important role in both gene activation and repression. A high level of H2A.Z expression is ubiquitously detected in many cancers and is significantly associated with cellular proliferation and genomic instability. Histone variant macroH2A1 is important in the pathogenesis of many types of cancers, for instance in hepatocellular carcinoma.

Cancer treatment

Current research has shown that epigenetic pharmaceuticals could be a putative replacement or adjuvant therapy for currently accepted treatment methods such as radiation and chemotherapy, or could enhance the effects of these current treatments. It has been shown that the epigenetic control of the proto-onco regions and the tumor suppressor sequences by conformational changes in histones directly affects the formation and progression of cancer. Epigenetics also has the factor of reversibility, a characteristic that other cancer treatments do not offer.

Drug development has focused mainly on histone acetyltransferase (HAT) and histone deacetylase (HDAC), and has included the introduction to the market of the new pharmaceutical vorinostat, an HDAC inhibitor. HDAC has been shown to play an integral role in the progression of oral squamous cancer.

Current front-runner candidates for new drug targets are histone lysine methyltransferases (KMT) and protein arginine methyltransferases (PRMT).

Twin studies

Recent studies involving both dizygotic and monozygotic twins have produced some evidence of epigenetic influence in humans.

Direct comparisons between identical twins constitute the ideal experimental model for testing environmental epigenetics, because DNA sequence differences that would be abundant in a singleton-based study do not interfere with the analysis. Research has shown that a difference in the environment can produce long-term epigenetic effects, and different developmental monozygotic twin subtypes may be different with respect to their susceptibility to be discordant from an epigenetic point of view.^[54]

One of the first high-throughput studies of epigenetic differences between monozygotic twins focused in comparing global and locus-specific changes in DNA methylation and histone modifications in a sample of 40 monozygotic twin pairs.^[55] In this case, only healthy twin pairs were studied, but a wide range of ages was represented, between 3 and 74 years. One of the major conclusions from this study was that there is an age-dependent accumulation of epigenetic differences between the two siblings of twin pairs. This accumulation suggests the existence of epigenetic “drift”.

A more recent study, where 114 monozygotic twins and 80 dizygotic twins were analyzed for the DNA methylation status of around 6000 unique genomic regions, concluded that epigenetic similarity at the time of blastocyst splitting may also contribute to phenotypic similarities in monozygotic co-twins. This supports the notion that microenvironment at early stages of embryonic development can be quite important for the establishment of epigenetic marks.^[56]

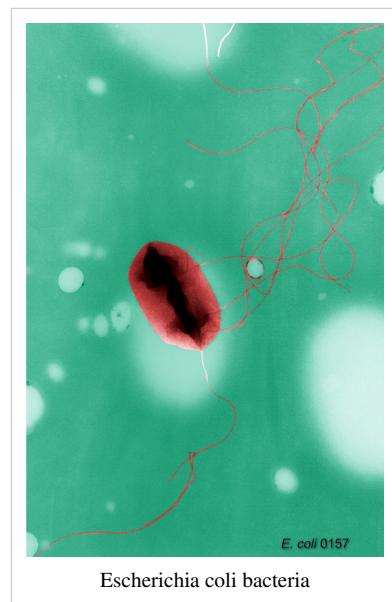
Epigenetics in microorganisms

Bacteria make widespread use of postreplicative DNA methylation for the epigenetic control of DNA-protein interactions. Bacteria make use of DNA adenine methylation (rather than DNA cytosine methylation) as an epigenetic signal. DNA adenine methylation is important in bacteria virulence in organisms such as *Escherichia coli*, *Salmonella*, *Vibrio*, *Yersinia*, *Haemophilus*, and *Brucella*. In *Alphaproteobacteria*, methylation of adenine regulates the cell cycle and couples gene transcription to DNA replication. In *Gammaproteobacteria*, adenine methylation provides signals for DNA replication, chromosome segregation, mismatch repair, packaging of bacteriophage, transposase activity and regulation of gene expression.

The filamentous fungus *Neurospora crassa* is a prominent model system for understanding the control and function of cytosine methylation. In this organism, DNA methylation is associated with relics of a genome defense system called RIP (repeat-induced point mutation) and silences gene expression by inhibiting transcription elongation.

The yeast prion PSI is generated by a conformational change of a translation termination factor, which is then inherited by daughter cells. This can provide a survival advantage under adverse conditions. This is an example of epigenetic regulation enabling unicellular organisms to respond rapidly to environmental stress. Prions can be viewed as epigenetic agents capable of inducing a phenotypic change without modification of the genome.

Direct detection of epigenetic marks in microorganisms is possible with single molecule real time sequencing, in which polymerase sensitivity allows for measuring methylation and other modifications as a DNA molecule is being sequenced. Several projects have demonstrated the ability to collect genome-wide epigenetic data in bacteria.



Notes and references

- [1] According to the Oxford English Dictionary: It is also worth quoting this adumbration of the definition given there (viz., "The formation of an organic germ as a new product"):
- [2] Waddington, Conrad H. 1953. The Epigenetics of birds. Cambridge University Press
- [3] (<http://www.ncbi.nlm.nih.gov/pubmed/14760651>) [url=<http://dx.doi.org/10.1002/jez.b.20002>] Hall BK. 2004. In search of evolutionary developmental mechanisms: the 30-year gap between 1944 and 1974. *J Exp Zool B Mol Dev Evol.* 15 January 2004;302(1):5-18.
- [4] (<http://www.plosone.org/article/info:doi/10.1371/journal.pone.0003626>) [url=<http://dx.doi.org/10.1371/journal.pone.0003626>] Álvarez-Buylla ER, Chaos Á, Aldana M, Benítez M, Cortes-Poza Y, et al. 2008. Floral Morphogenesis: Stochastic Explorations of a Gene Network Epigenetic Landscape. *PLoS ONE* 3(11): e36265.
- [5] Gottlieb,G., (1991). Epigenetic systems view of human development. *Developmental Psychology*, 27(1), 33-34.
- [6] Gilbert Gottlieb. Probabilistic epigenesis (http://chd.ucsd.edu/_files/winter2009/Gottlieb.probabilistic.pdf), *Developmental Science* 10:1 (2007), 1-11
- [7] Boeree, C. George, (1997/2006), *Personality Theories, Erik Erikson* (http://www.social-psychology.de/do/pt_ekirkson.pdf)
- [8] NIH Roadmap Epigenomics Project Overview (<http://www.roadmapepigenomics.org/overview>)
- [9] Malanga M, Althaus FR (2005). The role of poly(ADP-ribose) in the DNA damage signaling network. *Biochem Cell Biol* 83(3) 354-364. PMID 15959561
- [10] Ogrzykow VV. Erwin Schrödinger, Francis Crick and epigenetic stability. *Biol Direct.* 2008 Apr 17;3:15. (<http://www.biology-direct.com/content/3/1/15>)
- [11] Wang Z, Yao H, Lin S, Zhu X, Shen Z, Lu G, Poon WS, Xie D, Lin MC, Kung HF (2012). Transcriptional and epigenetic regulation of human microRNAs" *Cancer Lett* 331(1) 1-10. doi: 10.1016/j.canlet.2012.12.006. PMID 3246373
- [12] Browse miRBase by species (<http://www.mirbase.org/cgi-bin/browse.pl>)
- [13] Lim LP, Lau NC, Garrett-Engele P, Grimson A, Schelter JM, Castle J, Bartel DP, Linsley PS, Johnson JM (2005). Microarray analysis shows that some microRNAs downregulate large numbers of target mRNAs" *Nature* 433(7027) 769-773. PMID 15685193
- [14] Lee D, Shin C (2012). MicroRNA-target interactions: new insights from genome-wide approaches" *Ann N Y Acad Sci* 1271:118-28. doi: 10.1111/j.1749-6632.2012.06745.x. Review. PMID 23050973
- [15] Friedman RC, Farh KK, Burge CB, Bartel DP (2009). Most mammalian mRNAs are conserved targets of microRNAs" *Genome Res* 19(1) 92-105. doi: 10.1101/gr.082701.108. PMID 18955434
- [16] Goll MG, Bestor TH (2005). Eukaryotic cytosine methyltransferases" *Annu Rev Biochem* 74:481-514. PMID 15952895
- [17] sRNATarBase 2.0 A comprehensive database of bacterial SRNA targets verified by experiments (<http://ccb.bmi.ac.cn/srnatarbase/>)
- [18] Genomics maps for small non-coding RNA's and their targets in microbial genomes (<http://srnamap.mbc.nctu.edu.tw/>)
- [19] See also Denis Noble *The Music of Life* see esp pp. 93-8 and p. 48 where he cites Jablonka & Lamb and Massimo Pigliucci's review of Jablonka and Lamb in *Nature* 435, 565-566 (2 June 2005)
- [20] Fearful Memories Passed Down to Mouse Descendants: Genetic imprint from traumatic experiences carries through at least two generations (<http://www.scientificamerican.com/article.cfm?id=fearful-memories-passed-down&print=true>), By Ewen Callaway and Nature magazine | Sunday, 1 December 2013.
- [21] Mice can 'warn' sons, grandsons of dangers via sperm (<http://medicalxpress.com/news/2013-12-mice-sons-grandsons-dangers-sperm.html#ajTabs>), by Mariette Le Roux, 12/1/13.
- [22] Fraga MF, Ballestar E, Paz MF, Ropero S, Setien F, Ballestar ML, Heine-Suñer D, Cigudosa JC, Urioste M, Benitez J, Boix-Chornet M, Sanchez-Aguilera A, Ling C, Carlsson E, Poulsen P, Vaag A, Stephan Z, Spector TD, Wu YZ, Plass C, Esteller M (2005)" *Proc Natl Acad Sci U S A* 102(30) 10604-10609. PMID 16009939
- [23] Davies, Hazel (2008). Do Butterflies Bite?: Fascinating Answers to Questions about Butterflies and Moths (Animals Q&A). Rutgers University Press.
- [24] A person's paternal grandson is the son of a son of that person; a maternal grandson is the son of a daughter.
- [25] Robert Winston refers to this study in a lecture (<http://www.dundee.ac.uk/externalrelations/events/lectures.html>); see also discussion at Leeds University, here (<https://web.archive.org/web/20061003060251/http://www.fbs.leeds.ac.uk/staff/pm/epigenetics.htm#exciting2>)
- [26] WebCite query result (<http://www.webcitation.org/5QeAQ5n5o>)
- [27] Jasperson KW, Tuohy TM, Neklason DW, Burt RW (2010). Hereditary and familial colon cancer" *Gastroenterology* 138(6) 2044-2058. doi: 10.1053/j.gastro.2010.01.054. PMID 20420945
- [28] Wood LD, Parsons DW, Jones S, Lin J, Sjöblom T, Leary RJ, Shen D, Boca SM, Barber T, Ptak J, Silliman N, Szabo S, Dezso Z, Ustyanksky V, Nikolskaya T, Nikolsky Y, Karchin R, Wilson PA, Kaminker JS, Zhang Z, Croshaw R, Willis J, Dawson D, Shipitsin M, Willson JK, Sukumar S, Polyak K, Park BH, Pethiyagoda CL, Pant PV, Ballinger DG, Sparks AB, Hartigan J, Smith DR, Suh E, Papadopoulos N, Buckhaults P, Markowitz SD, Parmigiani G, Kinzler KW, Velculescu VE, Vogelstein B (2007). The genomic landscapes of human breast and colorectal cancers" *Science* 318(5853) 1108-1113. PMID 17932254
- [29] Esteller M, Silva JM, Dominguez G, Bonilla F, Matias-Guiu X, Lerma E, Bussaglia E, Prat J, Harkes IC, Repasky EA, Gabrielson E, Schutte M, Baylin SB, Herman JG (2000). Promoter hypermethylation and BRCA1 inactivation in sporadic breast and ovarian tumors. *J Natl Cancer Inst* 92(7) 564-569. PMID 10749912

- [30] Agrelo R, Cheng WH, Setien F, Ropero S, Espada J, Fraga MF, Herranz M, Paz MF, Sanchez-Cespedes M, Artiga J, Guerrero D, Castells A, von Kobbe C, Bohr VA, Esteller M (2006). Epigenetic inactivation of the premature aging Werner syndrome gene in human cancer" *Proc Natl Acad Sci U S A* 2006;103(23) 8822-8827. PMID 16723399 PMCID: PMC1466544
- [31] Baldwin RL, Nemeth E, Tran H, Shvartsman H, Cass I, Narod S, Karlan BY (2000). BRCA1 promoter region hypermethylation in ovarian carcinoma: a population-based study" *Cancer Res* 60(19) 5329-5333. PMID 11034065
- [32] Shen L, Kondo Y, Rosner GL, Xiao L, Hernandez NS, Vilaythong J, Houlihan PS, Krouse RS, Prasad AR, Einspahr JG, Buckmeier J, Alberts DS, Hamilton SR, Issa JP (2005). MGMT promoter methylation and field defect in sporadic colorectal cancer. *J Natl Cancer Inst* 97(18) 1330-1338. PMID 16174854
- [33] Psofaki V, Kalogera C, Tzambouras N, Stephanou D, Tsianos E, Seferiadis K, Kolios G (2010). Promoter methylation status of hMLH1, MGMT, and CDKN2A/p16 in colorectal adenomas. *World J Gastroenterol* 16(28) 3553-3560. PMID 20653064 PMCID: PMC2909555
- [34] Lee KH, Lee JS, Nam JH, Choi C, Lee MC, Park CS, Juhng SW, Lee JH (2011). Promoter methylation status of hMLH1, hMSH2, and MGMT genes in colorectal cancer associated with adenoma-carcinoma sequence. *Langenbecks Arch Surg* 396(7) 1017-1026. PMID 21706233
- [35] Amatu A, Sartore-Bianchi A, Moutinho C, Belotti A, Bencardino K, Chirico G, Cassingena A, Rusconi F, Esposito A, Nichelatti M, Esteller M, Siena S (2013). Promoter CpG Island Hypermethylation of the DNA Repair Enzyme MGMT Predicts Clinical Response to Dacarbazine in a Phase II Study for Metastatic Colorectal Cancer. *Clin Cancer Res* [Epub ahead of print] PMID 23422094
- [36] Mokarram P, Zamani M, Kavousipour S, Naghibalhossaini F, Irajie C, Moradi Sarabi M, Hosseini SV (2012). Different patterns of DNA methylation of the two distinct O6-methylguanine-DNA methyltransferase (O(6)-MGMT) promoter regions in colorectal cancer. *Mol Biol Rep* December 28. [Epub ahead of print] PMID 23271133
- [37] Truninger K, Menigatti M, Luz J, Russell A, Haider R, Gebbers JO, Bannwart F, Yurtsever H, Neuweiler J, Riehle HM, Cattaruzza MS, Heinimann K, Schär P, Jiricny J, Marra G. Immunohistochemical analysis reveals high frequency of PMS2 defects in colorectal cancer" *Gastroenterology* 2005;128(5) 1160-1171. PMID 15887099
- [38] Facista A, Nguyen H, Lewis C, Prasad AR, Ramsey L, Zaitlin B, Nfonsam V, Krouse RS, Bernstein H, Payne CM, Stern S, Oatman N, Banerjee B, Bernstein C (2012). Deficient expression of DNA repair enzymes in early progression to sporadic colon cancer. *Genome Integr* 3(1) 3. PMID 22494821
- [39] Rigakos G, Razis E (2012). BRCAness: finding the Achilles heel in ovarian cancer" *Oncologist* 17(7) 956-962. doi: 10.1634/theoncologist.2012-0028. Review. PMID 22673632
- [40] Stefansson OA, Villanueva A, Vidal A, Martí L, Esteller M (2012). BRCA1 epigenetic inactivation predicts sensitivity to platinum-based chemotherapy in breast and ovarian cancer. *Epigenetics* 7(11) 1225-1229. doi: 10.4161/epi.22561. PMID 23069641
- [41] Chaisaingmongkol J, Popanda O, Warta R, Dyckhoff G, Herpel E, Geiselhart L, Claus R, Lasitschka F, Campos B, Oakes CC, Bermejo JL, Herold-Mende C, Plass C, Schmezer P (2012). Epigenetic screen of human DNA repair genes identifies aberrant promoter methylation of NEIL1 in head and neck squamous cell carcinoma" *Oncogene* 31(49) 5108-16. doi: 10.1038/onc.2011.660. PMID 22286769
- [42] Fan CY (2004). Epigenetic alterations in head and neck cancer: prevalence, clinical significance, and implications. *Curr Oncol Rep* 6(2) 152-161. Review. PMID 14751093
- [43] Koutsimpelas D, Pongsapich W, Heinrich U, Mann S, Mann WJ, Brieger J (2012). Promoter methylation of MGMT, MLH1 and RASSF1A tumor suppressor genes in head and neck squamous cell carcinoma: pharmacological genome demethylation reduces proliferation of head and neck squamous carcinoma cells. *Oncol Rep* 27(4) 1135-41. doi: 10.3892/or.2012.1624. PMID 22246327
- [44] Sun W, Zaboli D, Liu Y, Arnaoutakis D, Khan T, Wang H, Koch W, Khan Z, Califano JA (2012). Comparison of promoter hypermethylation pattern in salivary rinses collected with and without an exfoliating brush from patients with HNSCC" *PLoS One* 7(3) e33642. doi: 10.1371/journal.pone.0033642. PMID 22438973
- [45] Puri SK, Si L, Fan CY, Hanna E (2005). Aberrant promoter hypermethylation of multiple genes in head and neck squamous cell carcinoma. *Am J Otolaryngol* 26(1) 12-17. PMID 15635575
- [46] Ai L, Vo QN, Zuo C, Li L, Ling W, Suen JY, Hanna E, Brown KD, Fan CY (2004). Ataxia-telangiectasia-mutated (ATM) gene in head and neck squamous cell carcinoma: promoter hypermethylation with clinical correlation in 100 cases. *Cancer Epidemiol Biomarkers Prev* (1) 150-6. PMID 14744748
- [47] Zuo C, Zhang H, Spencer HJ, Vural E, Suen JY, Schichman SA, Smoller BR, Kokoska MS, Fan CY (2009). Increased microsatellite instability and epigenetic inactivation of the hMLH1 gene in head and neck squamous cell carcinoma. *Otolaryngol Head Neck Surg* 141(4) 484-490. doi: 10.1016/j.otohns.2009.07.007. PMID 19786217
- [48] Tawfik HM, El-Maqsood NM, Hak BH, El-Sherbiny YM (2011). Head and neck squamous cell carcinoma: mismatch repair immunohistochemistry and promoter hypermethylation of hMLH1 gene. *Am J Otolaryngol* 32(6) 528-536. doi: 10.1016/j.amjoto.2010.11.005. PMID 21353335
- [49] Narayanan L, Fritzell JA, Baker SM, Liskay RM, Glazer PM. (1997). Elevated levels of mutation in multiple tissues of mice deficient in the DNA mismatch repair gene Pms2" *Proc Natl Acad Sci U S A* 94(7) 3122-3127. PMID 9096356
- [50] Hegan DC, Narayanan L, Jirik FR, Edelmann W, Liskay RM, Glazer PM. (2006). Differing patterns of genetic instability in mice deficient in the mismatch repair genes Pms2, Mlh1, Msh2, Msh3 and Msh6" *Carcinogenesis* 2006 Dec;27(12) 2402-2408. PMID 16728433
- [51] Tutt AN, van Oostrom CT, Ross GM, van Steeg H, Ashworth A. (2002). Disruption of Brca2 increases the spontaneous mutation rate in vivo: synergism with ionizing radiation" *EMBO Rep* 3(3) 255-260. PMID 11850397 PMCID: PMC1084010
- [52] German J. (1969). Bloom's syndrome. I. Genetical and clinical observations in the first twenty-seven patients" *Am J Hum Genet* 1969 Mar;21(2) 196-227. PMID 5770175 PMCID: PMC1706430

- [53] Nowak MA, Komarova NL, Sengupta A, Jallepalli PV, Shih IeM, Vogelstein B, Lengauer C (2002). The role of chromosomal instability in tumor initiation" *Proc Natl Acad Sci U S A* 99(25) 16226-16231. PMID 12446840
- [54] Ballestar, E. (2009). Epigenetics Lessons from Twins: Prospects for Autoimmune Disease. Clinical Reviews in allergy & immunology, 39(1), 30-41. Retrieved 6 December 2013, from http://download.springer.com/static/pdf/554/art%253A10.1007%252Fs12016-009-8168-4.pdf?auth66=1386532566_ad74522baf84f602ad13988b1c4b3c7&ext=.pdf
- [55] Fraga MF, Ballestar E, Paz MF et al (2005) Epigenetic differences arise during the lifetime of monozygotic twins. *Proc Natl Acad Sci U S A* 102:10604–10609
- [56] Kaminsky ZA, Tang T, Wang SC et al (2009) DNA methylation profiles in monozygotic and dizygotic twins. *Nat Genet* 41:240–245

External links

- Haque FN, Gottesman II, Wong AH (May 2009). "Not really identical: epigenetic differences in monozygotic twins and implications for twin studies in psychiatry". *Am J Med Genet C Semin Med Genet* **151C** (2): 136–41. doi: 10.1002/ajmg.c.30206 (<http://dx.doi.org/10.1002/ajmg.c.30206>). PMID 19378334 (<http://www.ncbi.nlm.nih.gov/pubmed/19378334>).
- The Human Epigenome Project (HEP) (<http://www.epigenome.org/>)
- The Epigenome Network of Excellence (NoE) (<http://www.epigenome-noe.net/index.php>)
- Canadian Epigenetics, Environment and Health Research Consortium (CEEHRC) (<http://www.epigenomes.ca/>)
- The Epigenome Network of Excellence (NoE)- public international site (<http://www.epigenome.eu/>)
- DNA Is Not Destiny (<http://discovermagazine.com/2006/nov/cover>) – Discover Magazine cover story
- BBC – Horizon – 2005 – The Ghost In Your Genes (<http://www.bbc.co.uk/sn/tvradio/programmes/horizon/ghostgenes.shtml>)
- Epigenetics article (<http://www.hopkinsmedicine.org/press/2002/november/epigenetics.htm>) at Hopkins Medicine
- Towards a global map of epigenetic variation (http://genome.wellcome.ac.uk/doc_WTX036556.html)

Pangenesis

Pangenesis was Charles Darwin's hypothetical mechanism for heredity. He presented this 'provisional hypothesis' in his 1868 work *The Variation of Animals and Plants under Domestication* and felt that it brought 'together a multitude of facts which are at present left disconnected by any efficient cause'. The etymology of the word comes from the Greek words *pan* (a prefix meaning "whole", "encompassing") and *genesis* ("birth") or *genos* ("origin"). The hypothesis was eventually replaced by Mendel's laws of inheritance.

The pangenesis theory, similarly to Hippocrates's views on the topic, imply that the whole of parental organisms participate to heredity—thus the prefix *pan*—, while adapting to cell theory. Much of Darwin's model was speculatively based on inheritance of tiny heredity particles he called gemmules that could be transmitted from parent to offspring. Darwin emphasized that only cells could regenerate new tissues or generate new organisms. He posited that atomic sized gemmules formed by cells would diffuse and aggregate in the reproductive organs.

Overview

Darwin's pangenesis theory was complex as he tried to explain the process of sexual reproduction, passing of traits and complex developmental phenomena, such as cellular regeneration. His pangenesis theory was criticised for its Lamarckian premise that parents could pass on traits acquired in their lifetime. Lamarckism fell from favour after August Weismann's research in the 1880s indicated that changes from use (such as lifting weights to increase muscle mass) and disuse (such as being lazy and becoming scrawny) were not heritable. Some Lamarckian principles, however, have not been entirely discounted and some of Darwin's pangenesis principles (in this regard) do relate to heritable aspects of phenotypic plasticity, while the status of gemmules has been firmly rejected. Darwin himself had noted that "the existence of free gemmules is a gratuitous assumption"; by some accounts in modern interpretation, gemmules may be considered a prescient mix of DNA, RNA, proteins, prions, and other mobile elements that are heritable in a non-Mendelian manner at the molecular level.

Later elaboration

In his later work, *The Descent of Man*, Darwin elaborated further on the model. In a section on the "Laws of inheritance," Darwin specified that two elements in particular were most important: the *transmission* and the *development* of inherited characteristics. Darwin's insights were that characteristics could be transmitted which were not at the time of transmission actually being manifest in the parent organism, and that certain traits would manifest themselves at the same point of development (say, old age) in both the parent and child organisms. In order to make sense of his theory of sexual selection, he also stipulated that certain traits could be passed through organisms but would only develop depending on the sex of the organism in question.

Galton's experiments on rabbits

Darwin's half-cousin Francis Galton conducted wide-ranging inquiries into heredity which led him to refute Charles Darwin's hypothetical theory of pangenesis. In consultation with Darwin, he set out to see if gemmules were transported in the blood. In a long series of experiments in 1869 to 1871, he transfused the blood between dissimilar breeds of rabbits, and examined the features of their offspring [1]. He found no evidence of characters transmitted in the transfused blood (Bulmer 2003, pp. 116–118). Darwin challenged the validity of Galton's experiment, giving his reasons in an article published in 'Nature'[2] where he wrote: "Now, in the chapter on Pangenesis in my *Variation of Animals and Plants under Domestication*, I have not said one word about the blood, or about any fluid proper to any circulating system. It is, indeed, obvious that the presence of gemmules in the blood can form no necessary part of my hypothesis; for I refer in illustration of it to the lowest animals, such as the Protozoa, which do not possess blood or any vessels; and I refer to plants in which the fluid, when present in the vessels, cannot be considered as true

blood." He goes on to admit: "Nevertheless, when I first heard of Mr. Galton's experiments, I did not sufficiently reflect on the subject, and saw not the difficulty of believing in the presence of gemmules in the blood."

References

- Bulmer M. G. "Francis Galton: Pioneer of heredity and biometry" [3]

External links

- On-line Facsimile Edition of *The Variation of Animals and Plants Under Domestication* ^[4] from Electronic Scholarly Publishing
- Variation under Domestication ^[5], From: Freeman, R. B. 1977. *The Works of Charles Darwin: An Annotated Bibliographical Handlist*. 2nd edn. Dawson: Folkstone, at DarwinOnline, with links to online versions of the 1st. edition, first and second issues, and the 2nd. edition.

References

- [1] <http://www.abc.net.au/rn/science/ss/stories/s216074.htm>
- [2] <http://darwin-online.org.uk/content/frameset?itemID=F1751&viewtype=side&pageseq=1>
- [3] http://books.google.co.uk/books?id=vL0hq80XXqMC&dq=bulmer+galton&printsec=frontcover&source=bn&hl=en&ei=wWVgS5_yJYyTjAeI0bzpCg&sa=X&oi=book_result&ct=result&resnum=4&ved=0CA8Q6AEwAw#v=onepage&q=&f=false
- [4] <http://www.esp.org/books/darwin/variation/facsimile/title3.html>
- [5] http://darwin-online.org.uk/EditorialIntroductions/Freeman_VariationunderDomestication.html

Gemmule (pangenesis)

This article is about the proposed mechanism of heredity. For the internal buds of freshwater sponges, see Gemmule.

Gemmules were imagined particles of inheritance proposed by Charles Darwin as part of his Pangenesis theory. This appeared in his book *The Variation of Animals and Plants under Domestication*, published in 1868, nine years after the publication of his famous book *On the Origin of Species*.

Gemmules, also called plastitudes or pangenes, were assumed to be shed by the organs of the body and carried in the bloodstream to the reproductive organs where they accumulated in the germ cells or gametes. They thus provided a possible mechanism for the inheritance of acquired characteristics, as proposed by Jean-Baptiste Lamarck, which Darwin believed to be a cause of the observed variation in living organisms.

This was prior to Gregor Mendel's discovery of the particulate nature of inheritance becoming common knowledge among biologists after their rediscovery in 1900.

Quotes

(from *The Variation of Plants and Animals under Domestication* (1868), Charles Darwin)

It is universally admitted that the cells or units of the body increase by self-division, or proliferation, retaining the same nature, and that they ultimately become converted into the various tissues and substances of the body. But besides this means of increase I assume that the units throw off minute granules which are dispersed throughout the whole system; that these, when supplied with proper nutriment, multiply by self-division, and are ultimately developed into units like those from which they were originally derived. These granules may be called gemmules. They are collected from all parts of the system to constitute the sexual elements, and their development in the next generation forms the new being; but they are likewise capable of transmission in a dormant state to future generations and may then be developed.)

(from *Charles Darwin: The Power of Place* by E. Janet Browne):

Individual gemmules did not contain a complete microscopic blueprint for an entire creature in the way that Herbert Spencer or Carl von Nägeli described.' (p.276)

Pangenesis looked to him as if it might supply the answer. Darwin proposed that some limited effects from the environment might become embedded in an individual's constitution and thus be liable to be transmitted, via the gemmules, to the offspring. (p.281)

But Darwin now wanted to include in his scheme the possibility of the inheritance of some limited acquired characteristics. Pangenesis gave him the chance to be Lamarckian without any of Lamarck's inner strivings. As he put it, some aspects of the external environment could modify the inheritable gemmules.

In variations caused by the direct actions of changed conditions, of which several instances have been given, certain parts of the body are directly affected by the new conditions, and consequently throw off modified gemmules, which are transmitted to the offspring.¹⁸

No doubt the whole hypothesis of pangenesis was extremely complicated, he conceded. "But so are the facts." (p.283–284)

Galton was troubled because he began the work in good faith, intending to prove Darwin right; and he praised pangenesis in *Heredity Genius* in 1869. Somehow he had unintentionally proved Darwin wrong. Cautiously, he criticised his cousin's theory, although qualifying his remarks by saying that Darwin's gemmules (he called them "pangenes") might be only temporary inhabitants of the blood and that his experiments could have failed to pick them up. (p.291–292)

External links

- *The Variation of Animals and Plants under Domestication* ^[1] Full text online

References

[1] <http://www.esp.org/books/darwin/variation/facsimile/contents.htm>

Article Sources and Contributors

Lamarckism *Source:* <http://en.wikipedia.org/w/index.php?oldid=621882980> *Contributors:* ***Ria777, A little insignificant, Achmed123456789, Against the current, Aitias, Alansohn, Aliekens, Aljujla, Ams80, Anadverb, Androstachys, Anentiresleeve, Anna Lincoln, Ano-User, Anonywiki, Ashmoo, Astor, AxelBoldt, Baileynkeller, Barticus88, Bejnar, Bender235, Berton, Bifutake, Binadot, Black Kite, Bomac, CensoredScribe, Cesare Barbone, Charles Matthews, Chiswick Chap, Chocolateboy, Chris Capoccia, Chris the speller, Chris55, ChrisGualtieri, Cinik, Colonies Chris, Conty, Coricus, Csernica, Cybercobra, Cyberix, DARTH SIDIOUS 2, DadaNeem, Damneinstien, Daniel Brockman, Dave souza, Denisarona, Denispir, Discospinster, Dissemby, DrMicro, Dreaded Walrus, Dumarest, DuncanHill, Duncharris, Dyed Purple, Eekster, Ejil, Epicgenius, Ethan Mitchell, Extremophile, Fastfission, Fences and windows, Fgnievinski, Fpigerre, Fred Hsu, GCarty, Gilliam, Glacialfox, Gogo Dodo, Gomm, GreenUniverse, Grebbard, Gwern, Haeb, Haruth, Havermayer, Headbombs, Huckleboy, Human.v2.0, Huysman, Hyacinth, Icairms, Isnow, Jack Merridew, John of Reading, Jsethewards, Jsonitsac, Kvn8907, Lebensmuende, LeonardM, LilHelpa, Linas, Lyddeaa, Macdonald-ross, Madeleine Price Ball, Magda wojtyra, Magioladitis, Magnani, Mangostar, Marechal Ney, Mark Carden, Marshman, Memestream, Mercury, Mgiganteus1, Millosh, Mindmatrix, Minimac, Mr. Wheely Guy, Myles325a, Nascar1996, Nemo bis, Nepenthes, Ngio, Nneonneo, Omegatron, OmicronSSD, Onnerfors, OrangUtanUK, Orphan Wiki, PDH, Paul Barlow, Pee Tern, Pengo, Peripitus, PeterWagsatff4, PeterisP, Philip Trueman, Pwner594, Quiddity, Quuxplusone, R:128.40.76.3, Rabidfoxes, Rachell, Ragesoss, Redheylin, Reedy, Researfaker, Rich Farmbrough, Richard Arthur Norton (1958-), Richard001, RichardKennaway, Rjwilmsi, Robert K S, Robofish, Rpeh, Rusty Cashman, SCMcdanish, Saint91, Samsara, Sanfranman59, Shanel, Shouriki, Silverxxx, Skeptical24, SlimVirgin, Smeggyssmeg, SmilesALot, Snesseird, Soave, Solarra, Spylab, Steinsky, Stigmatella aurantiaca, Surv1v4l1st, SwisterTwister, Tassedethe, TheNusAbides, Theprivater83, Thibbs, Timwi, Tinz, Trueblood, Tstrobaugh, Tuckerj1976, Tuxedo junction, Twang, User99, Visite fortuitement prolongée, Voszman, Vremya, WAS 4.250, WLU, West.andrew.g, Wiki Raja, Wiki13, WikiPuppies, Wimt, Wtmitchell, XJasonZ, Xabian40409, Yuh66h, Zamphuor, Zoicon5, 459 anonymous edits

Transmutation of species *Source:* <http://en.wikipedia.org/w/index.php?oldid=597486027> *Contributors:* Alan Liefing, Armchair info guy, AuburnPilot, Calaschysm, Charles Matthews, CharlesGillingham, Cybercobra, DMR5713, Dannyno, Dave souza, Demfranchise, Dposte46, DuncanHill, Duncharris, Eequor, EncycloPetey, Exir Kamalabadi, ForestDim, Goose friend, Hairy Dude, Halenmccracken79, Havermayer, Hazmat2, JDspeeder1, JTBurman, Jagged 85, Johnbod, Johnuniq, Jullz, Kpalion, Macdonald-ross, Mausy5043, Mentifisto, Michael Hardy, Plommespiser, Realtas, Rjwilmsi, Rusty Cashman, Samsara, Sepahbash, Synchronicity66, TheParanoidOne, Tide rolls, Twas Now, TypoBoy, UnitedStatesian, WLU, WindRunner, Yankeesfan982, 20 anonymous edits

DNA methylation *Source:* <http://en.wikipedia.org/w/index.php?oldid=620058263> *Contributors:* A876, Aceofhearts1968, Achriner, Addshore, Adrian J. Hunter, Akahst, AlexR, Arcadian, Austinprince, Axel Driken, Benbest, Bigskyes, BobJones, Bobo192, Boghog, BrotherE, Ca2james, Cangrejoimmortal, Ceyockey, Charles Matthews, Chris the speller, Chrispounds, Christoph-bock, Clementine2009, ClockworkSoul, Courtney Tait, Cow4prez, Crana, Dandv, Daniel Cliff, Dayed, Dcirovic, Deniz Feneri, Dmitry Dzhagarov, Dokidok, Drphilharmonic, Duxwing, EditorInTheRye, Eef (A), Elcicsiegel, Evan.morien, Fcrivera22, Fluffernut, Gadfiim, Guillaume Filion, Hmliuic, Hongbo919, Hopefuldonor, InvictaHOG, JIP, Jacobglass, Jgreally, JonHarder, Jonnabuz, Jsnover, Julianomions, Karin sandy, Kembanggraps, Khabar2, Kodiai71, Kweekeet, Laportechicago, Lexor, Ligulem, Little Mountain 5, Lproven, Luuva, Magioladitis, Mallajaya, Mandarax, Materialscientist, Memming, Messengercro, Miguel Andrade, MikePittsburgh, Mikewax, Mikipedia2, Miltoor, Mohawkjohn, Munkitechoy, Narayanese, NawlinWiki, Nina Gerlach, Nipisiquit, Nursebhaiyan, Omics, Omnipaedista, Peak, Pgau002, Physicistjedi, Piotrus, Plantdrew, Plindenbaum, Raetschz, Raylim34, Rifleman 82, Rjwilmsi, Rod57, SBabovic, Spreuss, Scapermoya, Sean Potato Business, Shellymah, Smowntow, Specter01010, St3vo, Stevetihi, Taw, Ted.E, Teddybme, Tentinator, Tessi87, The Thing That Should Not Be, Thetunicgod, Thorwald, Tim1357, Tlabbshier, Touchstone42, TraumBa, Trikmc, Uthbirian, Von Juvalt, WLU, Was a bee, Wavelength, WereSpielChequers, Wickey-nl, WikHead, Wiki1911, Wikipelli, Wikipolonus, Woohookitty, Yashgaroth, Yworo, 197 anonymous edits

Chromatin *Source:* <http://en.wikipedia.org/w/index.php?oldid=62220592> *Contributors:* 2004-12-29T22:45Z, 30lkra, ARUNKUMAR P.R, Aadhart, Abstraktn, Addshore, Aehrens, Agathman, Ahoerstemeier, Alex4223, AlexiusHoratius, Alpha Quadrant, Amourindian, Andres, AndyWu, Andybuckle, Animeronin, Anna Lincoln, Antandrus, Antiqueight, Apparition11, Arakunem, Arcadian, Arthena, Avenged Eightfold, Aznguy14, Babbage, Bantman, Bejnar, Ben Moore, Bendzh, Bensaccount, Bhadani, Bigger1, Blafreniere, Bluemoose, Budde, Bulba2036, Bursting74, Can't sleep, clown will eat me, Carrp, Cgtdk, Clicketyclack, Cnickelfr, Commdor, Conversion script, Cooldude7273, Cordell, Crazy sane, Dabomb87, DannyWilde, Dbfirs, Delldot, Dfrg.msc, Dgw, Discospinster, Djplummer, Dmr2, Donner60, Drmies, Drphilharmonic, Duagloth, Dysprosia, ESKog, Eleassar, Electriccatfish2, EnSamulili, Eola2, EtymAestheto, Excirial, Explodingdoggies, FaeriellGrey, Fezz, Finell, Flewiss, Flies 1, FlyingToaster, Forluvoft, Gabo71, Gecm, Gdarin, Gene Nygaard, Giftite, Gjd001, Glen, Gogo Dodo, Gouerouz, Graham87, Grayshi, Grtrnidad, Hac13, Hairy Dude, Hannes Röst, Hmaina, Hopefuldonor, Hordaland, HorsePunchKid, Ian Pitchford, Iced Kola, Indiana123, JNW, JSpuung, JSquish, JackAidley, James086, Jimwhoisfat, Jj137, JoanneB, John254, Jschwart37, JuanitaJP, Jóna Pórumn, K. Annoyomous, KDSKDS, Kalathalan, KayEss, Kazkaskazkasako, Khb3rd, Keilana, Koshatul, Kosigrim, Kpj, LLDMart, Lankenau, Lar, LeilaniLad, Lexor, Loopy1894, Luk, Luuva, MBproj, MDG38, MacroDaemon, Magioladitis, Magnus Manske, MarcelM, Marek69, Markus Kuhn, Martarius, Materialscientist, Maury Markowitz, Mdill12, MichaK, Micron2, Misull, Mr Stephen, MrOllie, Mszeve, MuteRussian, NCurse, NE Ent, Namsed, Natisto, Nat11, NawlinWiki, Nbauman, Nephron, Newlynorth, Neøn, Nina, NitRav, Noosh-X, NuclearWarfare, OcciMoron, Ohconfucius, Opelio, Oxymoron83, Pdeitiker, Pgan002, Phcompeau, Philip Trueman, Pinethicket, Poopdaddy1234, ProtoFire, Quilt6, RJaguar3, RadioKirk, RedHillian, Redheylin, Remotelysensed, Riana, Rimas Letap, RockyRaccoon, Romanm, Salix alba, Sango123, Sasper16, Scriber, Sean Heron, Seans Potato Business, Shanes, Shrimp wong, Shureg, Siddhant, Sifaka, Snowolf, Some jerk on the Internet, Spaully, Steinsky, Super48paul, SwitcherCat, SyntaxError55, TaintedCherub, Tassedethe, TestPilot, Tgeairn, The Random Editor, The Red, The Thing That Should Not Be, The infinite, Tide rolls, TimVickers, Tito4000, Titodutta, Tony1, Touchstone42, VMS Mosaic, Velella, Vespristiano, Vrenator, Wackywendell, Webclient101, Weien, Wid, Wikimachine, Wikkipediarox, Williamb, Yersinia, Youssefsan, Yuividct, Zamftb, Zephyris, Александр, 570 anonymous edits

Saltation (biology) *Source:* <http://en.wikipedia.org/w/index.php?oldid=593955402> *Contributors:* Adzyaye, Arno Matthias, Beasterline, BobEnyart, Brooklyn-alvin, Digitata, Donner60, Dysmordorepanis, Edward, Embryomystic, Everyking, Evnchrist, Excirial, Extremophile, Fama Clamosa, Fodor Fan, Furrykef, Garygromet, Graham87, Greeneto, Gutterpunks, Hadrian89, Harizotoh9, Havermayer, Hrafna, HumbleGod, Insidiae, Janto, Johnuniq, Laurang, Leonardorejorge, Leptictidium, Liveinthe forests, Magic5ball, Mchavez, Merzul, Mgerb, Mitteldorf, Mmcannis, Mogism, Paul Barlow, Portillo, Proxima Centauri, Rusty Cashman, SteveChervitzTrutane, Tagus, Tbjablkin, Tktkt, Trilobitealive, Unimaginative Username, Woohookitty, 144 anonymous edits

Histone *Source:* <http://en.wikipedia.org/w/index.php?oldid=621324013> *Contributors:* 168..., A2-33, Aadhart, Abergabe, Abstraktn, Agathman, Arcadian, Audriusa, Banus, Bensaccount, Bgwhite, Boghog, Boracium, CBM, Can't sleep, clown will eat me, Carandraug, Ceyockey, Chodges, Conversion script, Cubaz, D6, DMacks, Darwinius, DragonflySixtyseven, Drphilharmonic, Dxnihilo, Dysprosia, Ebabye, Elem3nt, EnSamulili, Forluvoft, Fragle81, Gensdei, Giacomo Consalez, Giants27, Glipton1, Gould363, Grey Geezer, Gzornenplatz, Headbomb, Heathmoor, Idiobrett, Irishtgey, Jack Greenmaven, Jesse Vv, JonathonSimster, Jonsprofile, Karakal, Kkmurray, Koafv, Kosigrim, Kuru, Kvalva, Lambiam, Laz, Lexor, LilHelpa, MIRROR, Magioladitis, Mani1, Maristoddard, Marj Tiefert, Martarius, Mdmeguin, Meters, Mgiganteus1, Mikeblas, Miketwardos, Mintleaf, Misull, Mmeinstein, Movado73, Mr handsome, Mr. Stradivarius, MrOllie, NewEnglandYankee, Nicolas.Wu, Nono64, PatrickStar LaserPants, Pauli133, Pgan002, RDBrown, RE73, Raetschz, Raymondwinn, Redeemer079, Reinotr, Reo On, Rich Farmbrough, Rjwilmsi, Rmky87, Roadnottaken, Rocher86, RockyRaccoon, Rod57, Salvio giuliano, Salvateama2008, Schekn, Seans Potato Business, Shadowjams, Silvrous, Sjavitt84, Solon.KR, Stewartcock, Stillnotel, Tariqb, Terrycojones, Tgeairn, TimVickers, Ugur Basak, Unfree, Vanisaac, Vespristiano, Victorandrelemonmorales, Wavelength, WeigelaPen, Welsh, Wikid77, Wtowtn, Wtmitchell, X CheshireCat x, Yersinia, Zephyris, Zeunas, 207 anonymous edits

CRISPR *Source:* <http://en.wikipedia.org/w/index.php?oldid=624043336> *Contributors:* A2-33, Afbillin, Ajh20, Amkilpatrick, AnnaJune, AxelBoldt, Bejnar, Ben Moore, Benbest, Betucinas, Bgwhite, Boghog, Canhelp, Ceyockey, CharlesHBennett, Crooveck, CtSkennerton, Daniel.hohle, Dmitry Dzhagarov, Dmneedha, El Mayimbe, Emble64, Fascinoma, Frp, Frze, Genomeengineering, Gimmetrow, Giraffedata, Gregoriev, Imareaver, India thelandof monuments, James atmos, Jamesikim, Jenkins10, Jeyradan, John of Reading, Jonesey95, Justbrowsing2, JustinDSmith, Kalafati, Kelsvlt, Laurawgenesyn, Leegr, Lfstevens, Marodoc, Michaplot, Pakapramen, Panoramix303, Pgardne, Quercus solaris, RE73, Retired username, Rjwilmsi, SA UBC, SchreiberBike, SyleontJohn, Spirit of Eagle, Sunrise, SylviaStanley, Tanyajayne7, TheJJunk, Thorwald, TimVickers, Tinz, TransControl, Trumpstermator, Upsud, Wavelength, WikiAllele, Wjfox2005, 79 anonymous edits

Piwi-interacting RNA *Source:* <http://en.wikipedia.org/w/index.php?oldid=619127266> *Contributors:* Antonov86, Aonangfind, Ashgene, Banus, Ben Moore, Blix000, Ediacara, Felix Tritschler, Hrcitisz, Jsc0, Keizer51, Ketiltrout, Khazar, L5tardust, Madurai1982, Michal Sobkowski, Narayanese, NickelShoe, Nina, Nono64, Nurg, Opabinia regalis, Sriram sh, Thebignile, Valentine, Woohookitty, 25 anonymous edits

Hologenome theory of evolution *Source:* <http://en.wikipedia.org/w/index.php?oldid=609575734> *Contributors:* Alan Liefing, Archaeodontosaurus, DadaNeem, Dave souza, Fgegypt, Magioladitis, Neodop, NotWith, Plantdrew, Rjwilmsi, SandyGeorgia, Stigmatella aurantiaca, Syp, 5 anonymous edits

Epigenetics *Source:* <http://en.wikipedia.org/w/index.php?oldid=621398707> *Contributors:* 7mike5000, 83d40m, A-Day, A., B., A876, AThing, Aadhart, Adina cappell, Adrin.jalali, AdultSwim, Agathman, Alex.muller, Alexbrn, Amatulic, Andrej, AnnieWoo, Anrusna, Anthere, Anthonyhcole, Antoni Barau, Apers0n, Arcadian, Arkelweis, Arthur Rubin, Ashraf boss, AubreyEllenShomo, AxelBoldt, BD2412, Badagnani, Balzamon85, Ben Ben, Bencherlite, Bender235, Bensaccount, Bernstein0275, Beyond My Ken, Billman119, Bobfreshwater, Boghog, Bojo-is-the-man, Br'er Rabbit, Braincricket, Bryan Derksen, Bus stop, Butwhatdoiknow, Capmo, Caroline1981, Cbock, Cgingold, Charles Matthews, Chris walsh, Chris Capoccia, Chris Howard, Christian75, Christopherin, Ckatz, Cmglee, D taz R, D6, DMacks, DadaNeem, Danfischer313, Daniel.Cardenas, Danielgrad, David D., Davydhortman, Dbrregister, Delldot, Der Zeitgeist, DerHexer, Desellers, Dhfreedman, Diracseawave, Discospinster, Dogface, Don Gosiewski, Dougher, Dr Oldekop, DrKenHo, Drphilharmonic, Dumpster muffin, Duncan.france, Dysmordorepanis, EPM, EPAdmirateur, Eaglizard, Eahd201, EdH, Edison, Eleassar, Electricmuffin11, Electron9, Eric Kvaalen, ErkDemon, Erud, Ervinn, Espoo, Ettrig, Ex gratia, Excentrifuge, Eyoste, Faizan, False vacuum, Fat Cigar, Fd88ar, Fgnievinski, Forluvoft, Gaius Cornelius, Galapah, Gomm, Gondola, Gpokela, Gschliss, Guillaume2303, H0riz0n, H2g2bob, Hambleton, HansPopper, Heathhunnicutt, Hodja Nasreddin, Hwttdz, Hzh, IMKatgrrl, Ian Pitchford, Ichooxu, Id711, InverseHypercube, Irrbloss, JForget, Jakecarver2010, Jason Quinn, Jcmg1964, Jeronimo,

Jethero, Jetspeed11, Jhd2, Jjalexand, Joannamasel, Joeckingbling, Johannordholm, John Foley, John0101ddd, Johnuniq, Jon the id, Jonkerz, Joriki, Jraffe0404, Jules.LT, JustinWick, Jytdog, KDSKDS, KYPark, Keenan Pepper, Kembangrapz, Kevin Luethy, Khym Chanur, Kintetsubuffalo, Kkmurray, Kyrsgo, Lankenau, Leadwind, Lensicon, Lexor, Lighthead, Looie496, Lova Falk, LudicrousTripe, Lunarian, M farouk2000, Madeleine Price Ball, Mama meta modal, Marj Tiefert, MarkinBoston, Marooned Morlock, Martin.Budden, Matthew of Hamburg, Mauritsmaartendejong, Mbolcar, Mdsam2, Memestream, Menchi, Merzul, Mgiganteus1, Michael Hardy, Michael K. Duke, Mike Serfas, Mike2vil, Mild Bill Hiccup, Mindmatrix, MistyMorn, Mlewan, Modify, Mortense, MuteRussian, Mwsal, NBeale, NYCJosh, Narayaneese, Naysteam, Nbauman, Nhhswm, Nightwriter50, Nikpapag, NizoDino, Nomadna, Nouse4aname, O'Q, Oldekop, Osirus1701, Ost316, Osterluzei, OttoMäkelä, Owleye769, Oxymoron83, PDH, PFHLai, Passportguy, Paul venter, Peak, Pete.Hurd, Pgr94, Pianomanjeff, Pjozsi, Plehn, Plumbago, Poa, Polsmeth, Polyhedron, Pseudomonas, Pseudomyrmex, Psytranscience, Ptr123, Qwerty53, Qxz, R'n'B, R. S. Shaw, RDBrown, RK, Rambatino, Raymond1922A, Redpillbluepill, Reesei, Reindra, Rich Farmbrough, Richjoo, Rigadoun, Rjwilmsi, Robert P. O'Shea, RomanSpa, Ronald.snijder, RoyGoldsmith, Rubywine, RustineS, Sam Hocevar, Samsara, Satirev, SattvaBodhi, Sbharris, Shoehring, Seans Potato Business, Semple1994, Shyamal, Slaporte, Sleepdoc1, SlimVirgin, Sm8900, Sms0610, Snalwibma, Snowolf, Sociocerebral, Somecdguy4, Squidonius, Srasher, Steinsky, Stevetihi, Stigmella aurantiaca, StoptheDatabaseState, Strangecow, StuRat, Stuckinski, SvenskaJohannes, Sylvain Mielot, Symbiogenesis, Szwedkowski, T-borg, TAConsta, Taka, Tambone, Tdadademem, Teamcytostorm, TedPavlic, Tenbergen, Thumperward, TijanaP, TimVickers, Tito4000, Tlabshier, Tobeprecise, Todash61, Tomcorsonknowles, Touchstone42, TraumB, Travis Martin, Trilobitealive, Tiam25, Tiotsw, Twas Now, Ukepat, Universal Life, Unsautsa, Vala M, VeryVerily, Vitund, Wapondaaponda, WatchAndObserve, Watcher0911, Wavelength, Werlop, Whoisjohngalt, Wikipolonus, Will Beback Auto, William Avery, Winterst, Wolf O'Donnell, Wolfkeeper, Woodrowvitz, Woohookity, WriterHound, Wuming77, Yonatan, Zacharylewis, Zaslav, Zzyzx11, Солярист, 359 anonymous edits

Pangenesis *Source:* <http://en.wikipedia.org/w/index.php?oldid=621843066> *Contributors:* Aa77zz, AlphaEta, Amaltheus, Anna Lincoln, Arcadian, Bubba73, Calaschysm, Conversion script, Courcelles, Crystallina, Cicero, DCDuring, Dave souza, Delusion23, Denispir, Duncharis, EagleFan, Editor2020, Emperorhma, Fanchon e, Fastfission, Fences and windows, Fill, Fredrik, JamesHilt62, Jarble, JoeSperrazza, Johnuniq, Keithbyrd, Kpjias, Krauss, Lankenau, Lexor, Lumos3, Maestlin, Margaretta, Memestream, Onsetuntil, Osarius, Ragesoss, Reywas92, Rich Farmbrough, Rjwilmsi, Robert1947, Samsara, Spotty11222, Sunrise, Tdadademem, The catr, Thomspsma, Titoxd, Tkttk, Tuxedo junction, Vegaswikian, 29 anonymous edits

Gemmule (pangenesis) *Source:* <http://en.wikipedia.org/w/index.php?oldid=588686280> *Contributors:* Aa77zz, AndyZ, Bluemoose, Bueller 007, Gurch, Jdvelasc, JoeSperrazza, John of Reading, Maestlin, Memestream, Moe Epsilon, PDH, Ranolev, Rjwilmsi, Spotty11222, SwiftGus, Tdadademem, 4 anonymous edits

Image Sources, Licenses and Contributors

Image:Jean-baptiste lamarck2.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:Jean-baptiste_lamarck2.jpg *License:* Public Domain *Contributors:* User:Valérie75

File:PSM V49 D527 Edward Drinker Cope.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:PSM_V49_D527_Edward_Drinker_Cope.jpg *License:* Public Domain *Contributors:* Ineuw, Kilom691

Image:Giraffe23.jpg *Source:* <http://en.wikipedia.org/w/index.php?title=File:Giraffe23.jpg> *License:* Public Domain *Contributors:* Ltshears

File:DNA methylation.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:DNA_methylation.jpg *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* User:Christoph-bock

File:Jerry Coyne, American professor of biology at the University of Chicago.jpg *Source:*

http://en.wikipedia.org/w/index.php?title=File:Jerry_Coyne,_American_professor_of_biology_at_the_University_of_Chicago.jpg *License:* Creative Commons Attribution-Sharealike 3.0
Contributors: David Gerard, Fuzheado

File:CollapsedtreeLabels-simplified.svg *Source:* <http://en.wikipedia.org/w/index.php?title=File:CollapsedtreeLabels-simplified.svg> *License:* Public Domain *Contributors:* Original uploader was User:TimVickers, SVG conversion by User:User_A1. Original uploader was User A1 at en.wikipedia

File:Tree of life.svg *Source:* http://en.wikipedia.org/w/index.php?title=File:Tree_of_life.svg *License:* GNU Free Documentation License *Contributors:* User:Samsara

File:Folder Hexagonal Icon.svg *Source:* http://en.wikipedia.org/w/index.php?title=File:Folder_Hexagonal_Icon.svg *License:* GNU Free Documentation License *Contributors:* Anomie, Mifter

File:Symbol book class2.svg *Source:* http://en.wikipedia.org/w/index.php?title=File:Symbol_book_class2.svg *License:* Creative Commons Attribution-Sharealike 2.5 *Contributors:* Lokal_Profil

Image:Vestiges dev diag.svg *Source:* http://en.wikipedia.org/w/index.php?title=File:Vestiges_dev_diag.svg *License:* Public Domain *Contributors:* Vestiges_dev_diag.jpg: Robert Chambers derivative work: Gregors (talk) 07:40, 9 March 2011 (UTC)

Image:Vertebrate archetype.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:Vertebrate_archetype.jpg *License:* Public Domain *Contributors:* Richard Owen

Image:Commons-logo.svg *Source:* <http://en.wikipedia.org/w/index.php?title=File:Commons-logo.svg> *License:* logo *Contributors:* Anomie

File:Chromatin Structures.png *Source:* http://en.wikipedia.org/w/index.php?title=File:Chromatin_Structures.png *License:* GNU Free Documentation License *Contributors:* Original uploader was Richard Wheeler at en.wikipedia Later version(s) were uploaded by Seans Potato Business at en.wikipedia

File:A-DNA, B-DNA and Z-DNA.png *Source:* http://en.wikipedia.org/w/index.php?title=File:A-DNA,_B-DNA_and_Z-DNA.png *License:* GNU Free Documentation License *Contributors:* Original uploader was Richard Wheeler (Zephyris) at en.wikipedia

File:Nucleosome 1KX5 2.png *Source:* http://en.wikipedia.org/w/index.php?title=File:Nucleosome_1KX5_2.png *License:* GNU Free Documentation License *Contributors:* By Richard Wheeler (Zephyris) 2005.

File:30nm Chromatin Structures.png *Source:* http://en.wikipedia.org/w/index.php?title=File:30nm_Chromatin_Structures.png *License:* GNU Free Documentation License *Contributors:* Original uploader was Zephyris at en.wikipedia

File:ChromatinFibers.png *Source:* <http://en.wikipedia.org/w/index.php?title=File:ChromatinFibers.png> *License:* Public Domain *Contributors:* Julien Mozziconacci

File:NHGRI human male karyotype.png *Source:* http://en.wikipedia.org/w/index.php?title=File:NHGRI_human_male_karyotype.png *License:* Public Domain *Contributors:* Courtesy: National Human Genome Research Institute

Image:Nucleosome structure.png *Source:* http://en.wikipedia.org/w/index.php?title=File:Nucleosome_structure.png *License:* GNU Free Documentation License *Contributors:* Richard Wheeler (Zephyris)

File:Protein H2AFJ PDB 1aoi.png *Source:* http://en.wikipedia.org/w/index.php?title=File:Protein_H2AFJ_PDB_1aoi.png *License:* Creative Commons Attribution-Sharealike 3.0
Contributors: Emw

File:PBB Protein HIST1H1B image.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:PBB_Protein_HIST1H1B_image.jpg *License:* Public Domain *Contributors:* . Original uploader was ProteinBoxBot at en.wikipedia

File:methyl lysine.tif *Source:* http://en.wikipedia.org/w/index.php?title=File:Methyl_lysine.tif *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* User:Jonsprofile

File:methyl arginine.tif *Source:* http://en.wikipedia.org/w/index.php?title=File:Methyl_arginine.tif *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* User:Jonsprofile

File:acetyl lysine.tif *Source:* http://en.wikipedia.org/w/index.php?title=File:Acetyl_lysine.tif *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* User:Jonsprofile

File:amino acid phosphorylations.tif *Source:* http://en.wikipedia.org/w/index.php?title=File:Amino_acid_phosphorylations.tif *License:* Creative Commons Attribution-Sharealike 3.0
Contributors: User:Jonsprofile

Image:Crispr.png *Source:* <http://en.wikipedia.org/w/index.php?title=File:Crispr.png> *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* James atmos

File:SimpleCRISPR.jpg *Source:* <http://en.wikipedia.org/w/index.php?title=File:SimpleCRISPR.jpg> *License:* Creative Commons Attribution-Sharealike 3.0 *Contributors:* AnnaJune

File:PDB_1wj9 EBI.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:PDB_1wj9_EBI.jpg *License:* unknown *Contributors:* -

File:PDB_1zpw EBI.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:PDB_1zpw_EBI.jpg *License:* unknown *Contributors:* -

Image:piRNA.jpg *Source:* <http://en.wikipedia.org/w/index.php?title=File:piRNA.jpg> *License:* Public Domain *Contributors:* Original uploader was Aonangfind at en.wikipedia

File:CoralBleaching.jpg *Source:* <http://en.wikipedia.org/w/index.php?title=File:CoralBleaching.jpg> *License:* Public Domain *Contributors:* Lfstevens, Liné1, Martin H.

File:Nezara viridula MHNT verte.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:Nezara_viridula_MHNT_verte.jpg *License:* Creative Commons Attribution-Sharealike 3.0
Contributors: Didier Descouens

File:Acyrtosiphon pisum (pea aphid)-PLoS.jpg *Source:* [http://en.wikipedia.org/w/index.php?title=File:Acyrtosiphon_pisum_\(pea_aphid\)-PLoS.jpg](http://en.wikipedia.org/w/index.php?title=File:Acyrtosiphon_pisum_(pea_aphid)-PLoS.jpg) *License:* Creative Commons Attribution 2.5
Contributors: Shipher Wu (photograph) and Gee-way Lin (aphid provision), National Taiwan University

File:Epigenetic mechanisms.jpg *Source:* http://en.wikipedia.org/w/index.php?title=File:Epigenetic_mechanisms.jpg *License:* Public Domain *Contributors:* National Institutes of Health

File:Escherichia coli flagella TEM.png *Source:* http://en.wikipedia.org/w/index.php?title=File:Escherichia_coli_flagella_TEM.png *License:* unknown *Contributors:* E. H. White, Content Provider: Peggy S. Hayes

License

Creative Commons Attribution-Share Alike 3.0
[//creativecommons.org/licenses/by-sa/3.0/](http://creativecommons.org/licenses/by-sa/3.0/)