A female neriid fly (bottom right) lays eggs on rotten tree bark while her mate fights off an interloper.
Credit: Russell Bonduriansky

The Evolving Story of Heredity

ANGELA CREAN & RUSSELL BONDURIANSKY

Biologists are discovering that there is a lot more to heredity than genes. In the latest twist, it turns out that offspring size in an Australian fly species can be determined by the diet of its mother's previous mating partner.

As evolutionary biologists, we are interested in variability. What causes similarities and differences among individuals? Where does this variation come from? How is variation transmitted across generations?

According to the textbooks, these questions were answered with the discovery of Mendel's laws, genetic inheritance and DNA. But despite having the technology to sequence entire genomes, we are still faced with "missing heritability", and discoveries made in recent years have greatly complicated the picture. Simply put, 21st century biologists are discovering that there is a lot more to heredity than genes.

Contrary to popular belief, the path of scientific progress rarely resembles a straight line. More often it twists and turns, and sometimes doubles back on itself like a meandering stream.

In the 19th century, the most widely-accepted theory of inheritance –commonly referred to as Lamarckian inheritance after the French naturalist Jean-Baptiste Lamarck – was the idea that features acquired throughout an individual's lifetime can be passed to offspring. For example, Lamarckians proposed that giraffes have such long necks because giraffe ancestors stretched their necks to reach the tasty leaves on the highest branches, and then passed on their stretched necks to their offspring. This sounds ridiculous to our modern ears, because

the inheritance of acquired characters does not fit with our modern understanding of genetic inheritance.

Back in the mid-19th century, the monk Gregor Mendel discovered that he could predict the colour and texture of pea plants based on the features of parental plants, and early 20th century geneticists were shocked and delighted to discover that Mendel's "laws" could explain the inheritance of a variety of characters in animals and plants. The age of classical genetics had dawned and, a century later, students still learn that an individual's features are determined by genes inherited from its mother and father in accordance with Mendelian principles.

While genetic inheritance is a beautiful truth, scientists are now discovering that it is not the whole story. We now know that features that run in families are not just influenced by the genes that are passed down from parents to their offspring. Various non-genetic forms of inheritance make it possible for maternal and paternal environments to also influence the features of their offspring. Given these alternative forms of inheritance, we need to revisit previously discredited theories such as the inheritance of acquired characters.

The Inheritance of Acquired Characters

We all know that what a mother eats during pregnancy can influence the growth, development and health of her child. This is easy to understand, as a mother provides the environment and nutrients for her developing child.

But you may be surprised to learn that what a father eats can also influence the development and health of his child. This finding is less intuitive because there need not be any direct link between a father's environment and his offspring.

In species that have no paternal care, males seem to contribute nothing more than sperm to their offspring. Yet, even in these species, the acquired characters of males can influence offspring features – showing that Lamarck was at least partially correct!

In humans, research shows that children are more likely to develop cardiovascular disease and diabetes if their father had plenty of food to eat between the ages of 9 and 12 – the years just before puberty when a boy's reproductive system matures. Even more surprisingly, these effects can cross multiple generations – grandfathers who ate less during these pre-pubescence years had grandkids with a longer life expectancy! Paternal smoking and chewing of betel-quid (a stimulant popular in parts of South-East Asia) have also been linked to adverse health outcomes in children.

Experiments on rats have shown that such effects may be explained by a recently discovered form of heredity called epigenetic inheritance. Epigenetic factors are molecules that adhere to the DNA, and control how our cells use the DNA's genetic information. Variation in epigenetic factors is the reason why genetically identical cells in our bodies are able to develop into a variety of different cell types (such as a skin cell versus a neuron), and part of the reason why identical twins (who share all the same genes) can nonetheless differ from each other in many ways.

Most epigenetic variation is erased when eggs and sperm are produced, allowing the fertilised egg to start life with a clean slate. However, scientists have recently discovered that some of the epigenetic variation survives erasure and is transmitted from parent to child.

This is important because, unlike the DNA itself, epigenetic factors can be influenced in predictable ways by the environment. For example, what you eat can influence the epigenome in cells throughout your body. If you happen to pass this change to your offspring, then some of their features – their physiology, health or life expectancy – may be influenced by what you ate before having kids.

New Surprises

While early 20th century geneticists were busy explaining why acquired characters could not be inherited, they also tossed out a theory called telegony – the idea that offspring can inherit the characteristics of a previous mate of their mother, and thus resemble a male who is not the father.

The classic example of telegony was Lord Morton's mare. This pure-bred Arabian horse was first mated to a quagga (an extinct relative of the zebra) and produced hybrid foals. Later, she was mated to another pure-bred horse and, to Lord Morton's great surprise, the two foals produced by these horses had striped legs and a stiff short mane that stuck straight up – the foals seemed to have some of the features of a quagga! In all future matings with horses, the mare produced foals that had quagga-like features, so it seemed that the initial mating with the quagga left some kind of imprint on the mare that affected all of her future offspring.

Many other putative examples of telegony surfaced during the 19th century, but the Mendelian geneticists were unconvinced. By the 1920s, the theory of telegony was reduced to a historical footnote.

But scientists have learned a great deal about cells, genes and heredity since 1920. As we've seen, new evidence of non-genetic forms of inheritance has necessitated a reappraisal of the possibility that acquired characteristics could be inherited. Could telegony be possible as well?

Male neriid flies (*Telostylinus angusticollis*) engage in spectacular battles for control of territories near egg-laying sites on rotting tree bark. Credit: Russell Bonduriansky

In addition to epigenetics, there are several other biological pathways through which the paternal environment may influence offspring features. For example, it is well-known that seminal fluid – the liquid medium in which sperm cells are transported from male to female — can carry sexually transmitted disease and infections. But semen also carries a cocktail of proteins, sugars, enzymes and hormones. These molecules can alter the physiology and behaviour of the male's mating partner, and could potentially influence offspring development. Thus, males may influence the development of offspring even if their sperm does not fertilise the egg.

New Evidence for Telegony in Flies

Previous research in our laboratory has shown that an Australian species of male neriid flies passes acquired characters to their offspring. We found that male flies fed a larval diet high in nutrients grow up to become large adults that produce larger offspring than male parents fed a diet that is low in nutrients.

This effect of paternal diet on offspring is especially interesting because there is no evidence of any conventional form of paternal care in this species – males transfer a tiny quantity of sperm, and matings only last for an average of 43 seconds. Thus there doesn't seem to be any opportunity for male flies to transfer nutrients to their offspring.

Instead, the effect of paternal diet on offspring size seems likely to be mediated by epigenetic factors associated with DNA in the sperm or by molecules in the seminal fluid. We set out to try to determine which of these two plausible mechanisms was involved.

We designed an experiment exploiting the reproductive physiology of flies. We fed male larvae a diet that was high or low in nutrients, and then mated these small and large males to immature females.

In flies, eggs are not fertilised until just before they are laid, so this initial mating exposed the developing eggs to seminal fluid but did not result in fertilisation. Two weeks later, we re-mated each female to a second small or large male to fertilise her mature eggs, and measured the size of these offspring.

We used genetic paternity analysis to confirm that the second male had fertilised the eggs, and indeed found that the vast majority of offspring were the second male's progeny. However, when we examined the body size of the offspring we discovered a surprising pattern: offspring size was determined by the diet of the first male that the females mated with. If a female was initially mated to a small male, her offspring were small even if their father was large (and vice-versa).

The most plausible explanation for this mind-boggling effect is that a male's diet influences the qualities of his seminal fluid, and his seminal fluid can in turn influence the development of eggs that are later fertilised by a different male. Since we did not find any effect from the diet of the second male whose sperm fertilised the egg, our results cannot be explained by the transmission of epigenetic factors in sperm.

But now comes the hard part. There are potentially hundreds of compounds in semen that could be driving this effect, and at this point we have no idea which one(s) may be responsible for the effect we've discovered.

We do not yet know whether similar patterns of inheritance occur in other species, but the potential for telegony exists in any species where females mate with more than one male and eggs are fertilised internally.

As evolutionary biologists, we get slightly dizzy just thinking about the potential implications of such effects for male and female mating behaviour. Our findings extend potential sources of variation in offspring features from the maternal and paternal environment to the environment of other males with whom female has previously mated.

Our study raises more questions than it answers, but that is the most exciting aspect of research. When you find something that was truly unexpected, a new fork in the road to discovery begins.

Angela Crean is an ARC DECRA Fellow and Russell Bonduriansky is an Associate Professor in the Evolution & Ecology Research Centre at UNSW Australia.