



Investigating Carrot Colours to Produce Healthier Crops

Dr Philipp W. Simon

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Carrots display a wide range of different colours – orange, purple, white, red and yellow – driven by the accumulation of various compounds. These compounds affect the nutritional value and health benefits of the roots, making them prime targets for breeding better varieties. **Dr Philipp Simon** and his colleagues at the United States Department of Agriculture investigate the genetics of carrot colours to help breeders develop even more nutritious strains.

A Nutritious & Colourful Root

Versatile, tasty, and full of vitamins, carrots can be found in cuisines across the globe. And their popularity is rising – the number of carrots grown worldwide has quadrupled in the last few decades, with farmers currently producing more than 42 million tonnes every year.

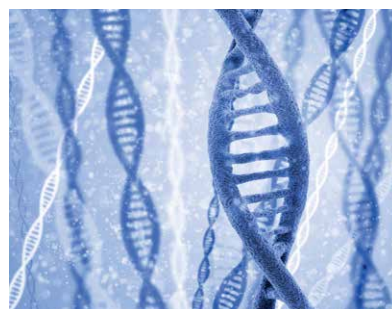
However, the orange carrots we see at the supermarket are a comparatively recent variety. In fact, for most of history, farmers have been digging purple roots out of their fields. ‘Orange carrots were only first described around 500 years ago,’ notes Dr Philipp Simon, researcher at the United States Department of Agriculture. ‘Purple and yellow were the first colours, reported 1100 years ago. Red and white carrots have also been developed within the last several hundred years.’

The reason behind the widespread switch to orange carrots is still rather controversial. Many people claim that it was a political statement by Dutch farmers supporting the Prince of Orange, who led the struggle for Dutch independence. Others dismiss this as entertaining but factually unsupported. Regardless of the reason, the choice of

colourful carrots was a rather fortunate one for humanity as a whole – many of the root’s health benefits can be traced back to these cheerful pigments. ‘Brightly coloured carrots are important for nutrition,’ explains Dr Simon. ‘The most important of these are the orange carotenoid compounds which are sources of vitamin A. The red, yellow, and purple pigments also provide key nutrients as antioxidants.’

This is excellent news for carrot lovers, but researchers are always interested in the ‘why’ behind facts. Why are carrots such excellent sources of these compounds? How are these compounds formed? And can we make carrots produce them in higher quantities?

Crop breeders are always seeking to develop healthier strains of fruit and vegetables, which have better nutritional value or contain higher levels of vitamins and antioxidants. For this reason, Dr Simon is bringing the many well-hidden secrets of carrot genetics into the light, to aid breeders in their quest to create highly nutritious varieties.



The Carrot Genome

The basis of this work comes from the carrot genome – its complete genetic code – which Dr Simon and his collaborators published in 2016. Determining a genome is no simple process; while genetic sequencing technology has advanced by leaps and bounds, we are still limited to ‘reading’ around 1000 letters of the genetic code at a time. For comparison, the length of the carrot genome is roughly 470 million ‘letters’ (or, more correctly, ‘base pairs’).

This means that any sequence has to be created from thousands of different ‘reads’, each of which need to be placed into context like jigsaw pieces. Specialised computer software and very expensive machines are required to gather this information and bring it together in the right way. Even with

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a wealth of knowledge behind them, problems can arise – for example, some parts of the genome are filled with repeated sequences, which makes it impossible to determine how they fit together. The carrot genome published by Dr Simon and his collaborators provided a map of around 90% of the complete genetic code – not perfect, but enough to make it one of the most complete genomes available.

From this basis, the researchers branched out into the world of carrot colours.

Food for Your Eyes

Carrots are one of the best dietary sources of a carotenoid compound called beta-carotene, which is converted into vitamin A in the human body. As vitamin A is vital for our vision, genetic processes, immune responses and skin health, carrots are an important part of a healthy diet.

Beta-carotene is a bright orange-red chemical, providing much of the bright orange colour we associate with carrots. Yet, as we've mentioned, orange carrots are themselves a fairly recent innovation. Why do they contain

so much beta-carotene, and such a blazingly orange colour? What genetic changes happened across centuries of breeding to reach this point?

This is the sort of question that Dr Simon loves to investigate. ‘Carrot colours provide insights into the development and domestication of carrot as a root crop,’ he explains, ‘allowing better understanding of the molecular and biochemical mechanisms underlying carotenoid biosynthesis, and are useful for nutritionists to study pigments in whole foods.’

To improve this understanding, Dr Simon and his colleagues investigated the genomes of different carrot strains, ranging in colour from white through to bright orange. They demonstrated that orange colour was correlated to changes in a single area of the genome – one that contains 72 genes.

To narrow down this region further, the researchers performed transcriptome analysis, which indicates whether a gene is ‘on’ or ‘off’ – whether it is actively being used to create proteins or not.

They showed that four genes have different activities at different stages in the carrot lifecycle, while another 13 exhibit different activity depending on the local conditions. With this as a basis, the team then examined 25 different carrot varieties, showing that the genetic sequence in that area is much more variable between orange strains than between white or yellow ones.

This is important for several reasons. First, the variability could be linked to beta-carotene levels – thus plant breeders could use particular genetic markers to determine if their new strains would be more or less orange. Second, the list of variable genes would then be the basis for further research, with in-depth studies discovering how precisely beta-carotene synthesis is controlled.

The Or Gene

As previously mentioned, carrots are one of the best sources of carotenes in our diet. However, carrots have *much higher* carotene levels than we would expect, given what scientists know about the biochemical pathways involved.



Dr Simon and his collaborators looked at the genomes of many different wild and domesticated carrot varieties, looking for differences that might explain this. They were able to find a mutation within a single gene, known as the *Or* gene, which is never found in wild carrots but is almost always present in domestic varieties. The mutation leads to increased formation of chromoplasts – cellular compartments that produce and store coloured chemicals such as carotenes.

Interestingly, the team was also able to show that all Western domestic carrots are genetically very similar, despite the wide variety in colour and appearance. The mutant *Or* gene, with its beneficial impact on carotene levels, appears to have been an early trait that was selected when carrots were domesticated, many centuries ago.

Purple is the New Orange

While orange carrots contain large amounts of carotenoid compounds such as beta-carotene, purple carrots have very high concentrations of anthocyanins, which exhibit antioxidant and anti-inflammatory effects. Diets rich in anthocyanin-containing fruits and vegetables are associated with a reduced level of diabetes, heart disease, arthritis and even some cancers.

This is quite interesting for crop breeders, who are always seeking to develop strains with both additional health benefits and visual appeal. In order to do so, however, they need to understand the biochemical basis of the purple carrots – how are these anthocyanins generated, and how can we enhance their production?

This is where Dr Simon enters the picture once more. His team had previously looked into the genome of purple carrots and

had shown that the genes for anthocyanin expression in the root and petiole (the stalk that attaches a leaf to the stem) could be narrowed down to a specific region of the genome. Further experiments narrowed this down even further, allowing the researchers to identify a cluster of transcription factors that control anthocyanin biosynthesis.

Transcription factors are a special form of protein, which control the expression of other genes. One transcription factor may turn a set of genes on in response to certain stimuli, while another may turn them off if a cellular signal comes in. In this way, the fixed ‘instructions’ present in the genome can be read in a specific, context-dependent manner.

In this case, a cluster of transcription factors belong to the *MYB* gene family are the main candidates for purple colouring in all carrots examined during the studies. Dr Simon and his colleagues demonstrated that these influence colouring in the roots and petioles, respectively, by controlling a number of other genes and biochemical processes that lead to anthocyanin accumulation.

The Carrot of the Future

So where will the researchers go from here? Dr Simon is interested in the development of new carrot strains that contain increased levels of beneficial nutrients such as carotenoids and anthocyanins. They are currently working with several experts in agriculture to bring these ideas to the market.

Beyond this, he is enthusiastic about the next steps for his research. ‘Sequencing of the carrot genome has significantly stimulated the development of high-pigment carrots,’ says Dr Simon, ‘and this story is only beginning to unfold!’

Meet the researcher



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Dr Philipp Simon began his scientific studies with a PhD in Genetics from the University of Wisconsin, Madison in 1977. Since 1979, he has been working as a Research Geneticist, and since 1986 a Research Leader, in the Vegetable Crops Research Unit of the USDA Agricultural Research Service (USDA-ARS), and Professor of Horticulture at the University of Wisconsin, Madison. Over the course of his successful career, Dr Simon has published over 200 papers, trained more than 30 PhD students, and won a multitude of prestigious awards, including an Honorary Doctorate from the Agricultural University of Krakow, Poland, and the National Association of Plant Breeders Lifetime Achievement Award. His research in vegetable genetics and breeding focuses on fresh market carrot improvement, targeting improved flavour and nutritional quality, nematode, disease and abiotic stress resistance, and genetic mapping.

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FURTHER READING

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