IMPACT

ADDING TO THE AGRICULTURAL TOOLKIT

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bioscience for the future

Adding to the agricultural toolkit

*A team of researchers based at the **University of Warwick**, UK, has been exploring the potential of plant auxins and is helping to usher in a new generation of safe, selective and low dosage agricultural compounds*

Hormones control plant growth and development, coordinating growth in response to environmental cues. One such hormone is auxin, which is involved in almost every part of a plant's life. Synthetic auxins are herbicides, used to help protect the world's crop yield but there is a growing problem of resistance to current active compounds. There is a need to protect existing actives and develop new ones in order to continue to produce more food to feed the growing population.

This is where auxin comes in. Auxin herbicides are generally active against broad-leaved weeds and inactive against cereals and grasses, which makes them desirable for use in arable agriculture on a global scale. With the identity of their target receptor only having been discovered around a decade ago, there is much to be explored about the potential of auxins, and one research group in particular is building upon this knowledge in an impressive way.

SHARING KNOW-HOW

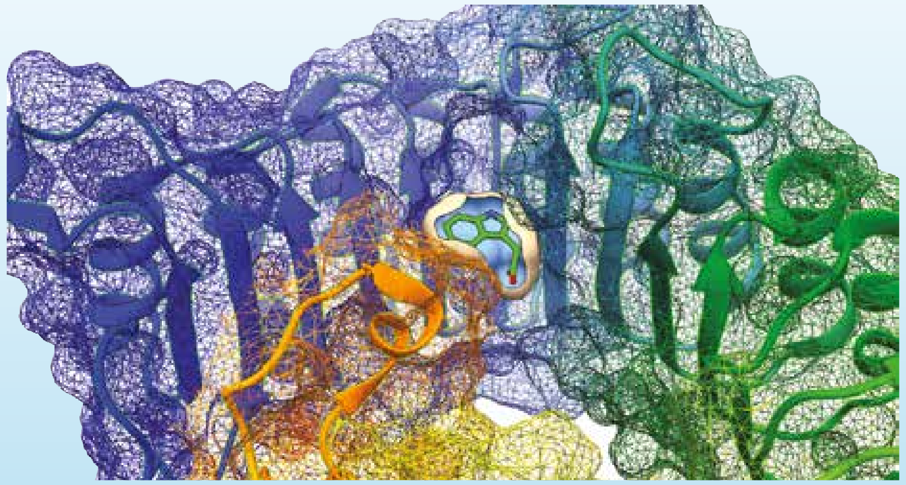
A three-year project studying this very topic and supported by the Biotechnology and Biological Sciences Research Council (BBSRC) called 'Next generation auxins and anti-auxins: principles for binding and design' has recently concluded. The project was led by Professor Richard Napier, who is based in the School of Life Sciences at the University of Warwick, and was focused on the development of agricultural compounds and creation of selective weed killers and agrochemicals to make a new generation of safe, selective and low-dosage herbicides.

With 20 years' experience working on plant hormone perception and signalling, and prior experience in animal membrane biochemistry, Napier was well placed to lead the work. The project united the expertise of five world-class research groups, including specialists in chemometrics, chemical biology libraries and synthetic chemistry, as Napier explains: 'We collaborated very productively with a number of research groups with common interests. In particular, we worked with Dr Stefan Kepinski from the University of Leeds, UK, who was one of the scientists who discovered auxin receptor proteins. The agrochemical companies Syngenta (UK) and Dow AgroSciences (USA) were also valued collaborators, with each providing herbicide know-how and access to some of their chemical collections. Both proved enormously helpful in our project.'

THE IMPORTANCE OF AUXIN

The project is focused on auxin, a hormone that is involved in most aspects of a plant's life. In order for auxin to trigger responses, it needs a receptor (a protein to which it binds in a very specific way). Auxin binding acts as a molecular switch, initiating a chain of events that leads to changes in which whole groupings of the plant's genes are switched on or off to change developmental decisions. The researchers used their new results to improve the resolution of what they understand to be the basis of the mechanism of action of auxins.

In the project, the researchers performed a set of experiments to quantify the differences between family members of TIR1, a protein that the team is studying as an auxin receptor, along with other members of its family called AFB proteins. This allows the researchers to describe the special features that determine the molecular rules of good auxins. The researchers are aware that in their main application as herbicides, auxins already present a certain element of selectivity, but they expect that there are more layers of selectivity to be explored and exploited. They believe this will enable them to design a new generation of selective plant growth regulators. The team has already shown that some auxins are selective for one or other receptor type. 'We have been able to demonstrate that the two newest, youngest members of the auxin herbicide family target a subclass of receptors with high affinity, distinguishing them from most of the older, established auxin herbicides against which resistance is increasing,' Napier reveals.



Natural auxin deep inside its binding pocket

THE LATEST TECHNIQUES

The project employed the latest biophysical techniques to measure the speeds of binding and the energy changes on binding. The researchers were then able to compare these values with computer-driven calculations of the auxin molecules themselves to derive design features specific for each template.

An important technique employed by the researchers was docking, which facilitated the exploration of complex binding processes. 'The advances of computers and the science of computational chemistry allow us to test for molecules that bind in silico (in computer simulation), greatly increasing the number and diversity of compounds available,' Napier explains. 'To do this, the computer-generated structure of each molecule is bounced around in the known structure of the target binding site to see if it can bind, and how likely it is to bind. This virtual binding can reduce bewildering complexity down to a reasonable number of molecules to evaluate further in the lab. In some cases, docking can also inform us about important points of contact between binding molecule and binding site.'

TOMODOCK

In addition to novel auxins, the team produced a computer code called TomoDock to create a new tool for drug discovery, and used it to discover how deep binding pockets act as selective barriers, filtering out some molecules while allowing others to reach their binding site. The researchers used auxin and its receptors in their case study to identify amino acids that are involved as molecular filters. 'We wrote a new app which takes a whole series of snapshots as the docking box is moved from outside to inside and past the final binding site,' Napier notes. 'In this way we mimic the transit of a molecule, building a detailed picture of movements and interactions on the way.'

INSPIRING THE NEXT GENERATION

Napier also hopes to encourage more students to consider the potential of plant biology: 'To my mind, helping the world produce enough healthy food is at least as important as curing ailments, and I promote the importance of food security in my teaching and outreach activities,' he highlights. 'Once people are prepared to "test the water" of plant biology, they start to see that its challenges and opportunities are just as difficult and appealing as medicine.'

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Project Insights

FUNDING
Biotechnology and Biological Sciences Research Council (BBSRC)

LEAD ORGANISATION
University of Warwick (UK)

COLLABORATORS
University of Leeds (UK) • Syngenta (Switzerland) • Dow AgroSciences (USA)

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PRINCIPAL INVESTIGATOR BIO
Professor Richard Napier holds a BSc in Botany from the University of Reading and a PhD in Plant Physiology from the University of Leicester, UK. He joined East Malling Research Station in 1987 and subsequently worked for Horticulture Research International before joining the University of Warwick in 2005. Napier is a full professor researching the molecular recognition of plant hormones.

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Impact Objectives

- Describe the special features on each type of auxin – a natural plant hormone and a family of selective herbicides
- Learn more detail about the molecular rules of specificity to support the design of a new generation of selective plant growth regulators

Auxin adventures

***Professor Richard Napier** of Warwick University, UK, introduces us to a drug discovery platform for novel plant growth regulators, analogues of the natural plant hormone auxin*

How did you become drawn to the life sciences?

I grew up in East Anglia in the UK surrounded by arable agriculture. At school I was drawn by some inspirational teachers to biology and chemistry, but I knew that I did not want to drift into medicine and so I chose to study plant biology at university. I enjoyed the more classical topics such as plant identification, but was always more intrigued by signalling systems and biochemistry.

What is auxin and what makes it so interesting?

Auxin is a natural plant hormone, a mobile signal that is directed around plants and actively accumulated in certain tissues where it induces profound changes in growth and development. One such accumulation is in the early embryo where it is part of the mechanism that initiates polarity in the new organism and establishes what will become the shoot apical meristem, which is a population of cells that is found in the shoot tip. Very rapidly the site of accumulation changes, leading to establishment of the root at the other pole of the embryo. From these early events in every plant's life and onwards, auxin is involved at every step. Yet the natural auxin, known as indole-3-acetic acid or IAA, is only one small molecule. How can one small molecule drive and determine so many aspects of the life of plants, life on which we all depend? From my perspective that is pretty intriguing.

What techniques do you use to study auxins?

In order to validate active molecules, we test for binding against purified auxin receptors in the laboratory. For this we use modern analytical instruments, in particular one called a surface plasmon resonance spectrometer. Essentially, if binding occurs we measure this as an increase in weight on a molecular scale. We have used these very precise measurements to build up a composite image of the 'perfect auxin'. We can use this as a template for designing novel auxins.

What were the key findings unearthed through the 'Next generation auxins and anti-auxins: principles for binding and design' project?

We have shown very clearly that there are differences in molecular selectivity between members of the family of auxin receptor proteins. This is somewhat surprising since we believe that there is only one natural auxin – IAA. If there is only one, why do different versions of the receptor vary in the binding site? We have also shown that the newest generation of auxin herbicides have a preference for one subclass of these receptors, which indicates that these new active compounds differ in selectivity from many more established auxin herbicides. Their very high affinity for this subclass has illustrated why they are active at very low field dose rates, meaning that weed control is achieved using far less compound.

What are your thoughts on designer herbicides?

We have also discovered a new class of auxin by rational design. Its initial activity was poor, but with rational design we have improved its efficacy. The appeal of this compound is that it is specific for one of the other subclasses of receptor, making it a realistic prospect that we will soon have tools to switch on only a few, selected receptors at a time, and to design further target-specific molecules.

What are your plans for future research in this area?

Our project support from the Biotechnology and Biological Sciences Research Council (BBSRC) has been remarkably successful, and we will be bidding for further awards in due course. The project also continues to attract some support for focused objectives from Dow AgroSciences. Sustainable food production continues to need new herbicides and we will work hard to support these needs.