

Keeping plants short: Natural gibberellin inhibitors

In this series of posts, we have discussed the different techniques and synthetic chemical substances that can be used to keep plants short. We discussed why [keeping plants short is important](#), how this can be done with [synthetic gibberellin inhibitors](#) and how this can also be achieved using [day/night temperature differentials](#). However, there are also a lot of natural substances that can be used to inhibit gibberellins, which can be used to help us achieve this same objective. In this post, we will be talking about the research around natural gibberellin inhibitors, the plant extracts that have shown this activity and what we have discovered these plant extracts contain.



Dried seeds and fruits of the carob plant

Research around plant extracts that could inhibit gibberellins started in the late 1960s. Many different plant extracts were tested for inhibitory activity. The tests were simple, a control plant was not sprayed, a second gibberellin control plant was sprayed with gibberellins and a third plant was

sprayed with a mixture of gibberellins and the tested plant extract. Whenever inhibitory activity was present, the third plant would show very similar characteristics to the control while the gibberellin sprayed plant would usually stretch significantly. You usually see graphs like the one showed below, where the plant sprayed with the pure gibberellins is the control while the extract contains both the gibberellins and the plant extract. When an extract inhibits the gibberellins the plant grows less under the same gibberellin concentration although as the gibberellin concentration is increased the inhibitory effect of the extract is surpassed and the plants reach similar points.

When doing this research, one of the plants that showed the most promise was the carob plant. Cold-pressed extracts of green carob fruits were studied quite extensively and showed this effect repeatedly ([1](#), [2](#), [3](#)). Different fractions extracted showed the effect and researchers sought to find the specific substances responsible for the inhibition. Eventually, researchers found that the culprit was abscisic acid ([4](#)), also known as ABA. Other plant extracts that had gibberellin inhibitory effects, such as lima beans, also proved to contain significant amounts of ABA ([5](#)). So why are we not using ABA as a safe and environmentally friendly gibberellin inhibitor?

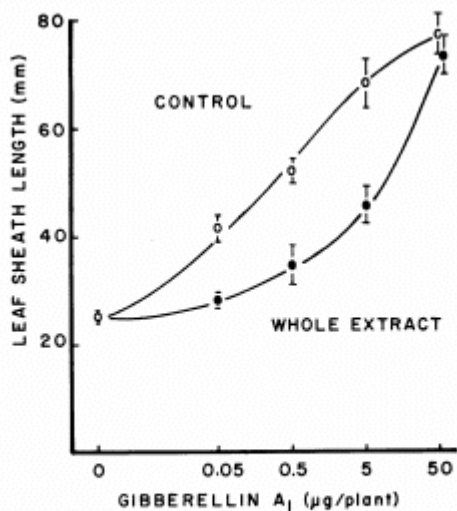


FIG. 3. The effect of gibberellin A₁ on the growth of maize seedlings in the presence and absence of whole extract. Each seedling treated with inhibitor received the extract from 5 mg fresh weight of carob fruit. Each point represents the average and standard error of 10 plants.

Sample graph showing the gibberelins inhibitory effect of a natural extract obtained from carob (taken from [here](#))

It boils down to the chemistry of ABA, which is quite complicated. First of all, ABA contains a chiral center (1' in the image below), making it the first chiral plant hormone to be discovered. This means that its mirror images are not equivalent – like your right hand is not equivalent to your left hand – which means that these two chemical forms will behave differently in biological systems. This complicates the synthesis of the molecule substantially. Furthermore, ABA contains several double bonds, which, depending on their configuration, can make the molecule completely inactive. Unfortunately, ABA goes through a double bond rearrangement under UV light that causes the molecule to deactivate, making it unstable for everyday use. So while ABA was great on paper, in practice it was never used widely. Several chemical analogs of ABA were developed and a lot of chemistry surrounding ABA and the proteins it binds to have been explored (you can read more in [this book](#)).

Phenolic compounds were also of great interest in the 1970s since many of the plant extracts that showed inhibitory activity also contained many of these molecules. These belong to a family of compounds called “tannins” and were then explored in pure form as potential gibberellin inhibitors,

with many of them showing substantial activity (6, 7, 8). This showed that extracts coming from fruits like carob had an inhibitory activity that was independent of the activity they got from ABA, although the phenolic compounds were significantly less active compared to the pure plant hormone.



Labeled diagram of the active form of ABA

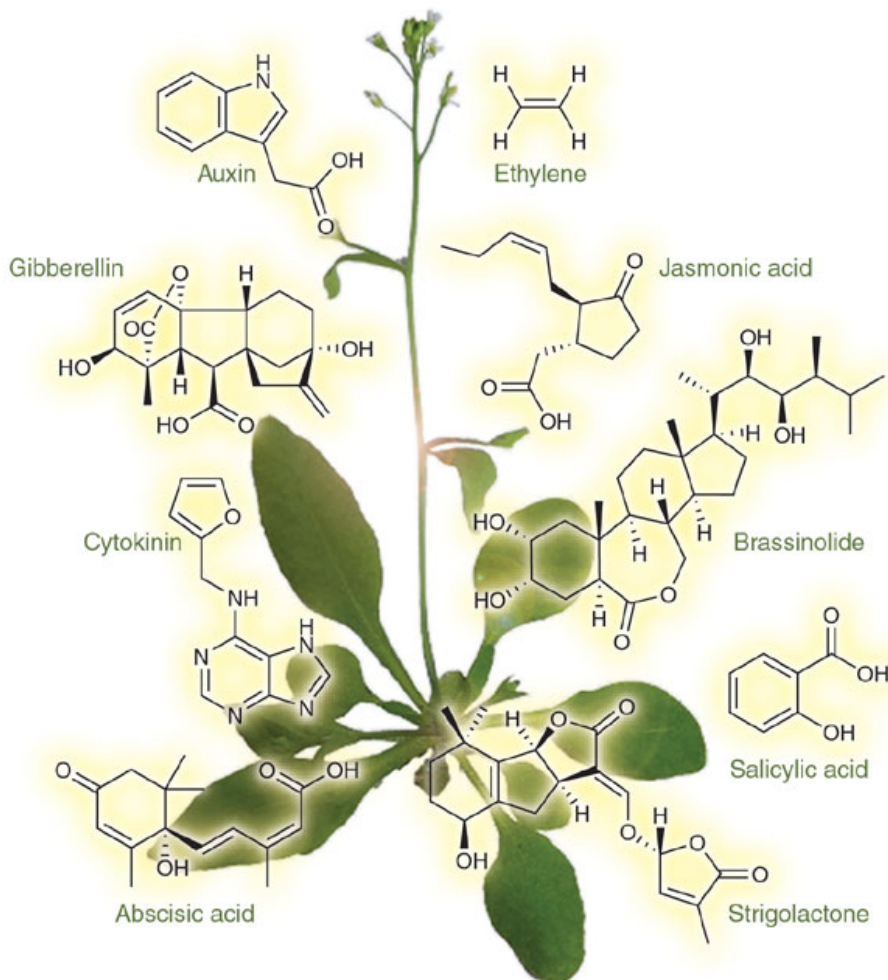
In the late 1970s, the research into these natural gibberellin inhibitors stopped as the first successful synthetic gibberellin synthesis inhibitors started to surface. These were much more effective since they did not deal with the gibberellin once produced but mostly attacked the paths that were used to form the chemical within the plants. Substances such as Chloromequat and Paclobutrazol made most of this research into naturally source inhibitors irrelevant, as these were cheap to produce in mass quantities and much more effective.

With the return towards safer and more natural alternatives and advances in chemical synthesis, the direct use of ABA or phenolic substances in order to inhibit gibberellins to prevent shoot elongation starts to become attractive. If you're interested in this path, looking at past research from the 1970s to come up with test formulations for foliar spray or root drench products would be a good initial approach. If you want to avoid the use of pure substances and all chemical synthesis, using direct extracts from plants like lima beans and carob is also a potential approach, although care needs to be taken to ensure the conditions of the extraction processes and extract storage do not destroy their active properties.

Six things you need to know before using plant hormones

Plant hormones are small molecules with no nutritional value that are used as chemical signalers within plants. A hormone will trigger a chemical signaling cascade that will cause the plant to carry out certain specific behavior. This fact has made them one of the most useful tools to manipulate plant growth and improve the yields and quality of many crops, especially flowering plants. This has also made them a key target for hype, with many products promising significant gains without much talk about interactions with other hormones or other fundamental aspects. In this post I want to talk about six things you should know about plant hormones, both to use them more effectively and to adequately manage your expectations when you use them. Note that although plant hormones are considered plant growth regulators (PGRs), this broad class includes other molecules – such as gibberellin synthesis inhibitors – that are not being considered in this post.

Know specifically what you want. A hormone will affect a plant in a very specific way, to achieve a specific purpose. Hormones can help you manipulate plant growth but which one you use depends fundamentally on what you want to achieve. Do you want the plant to be bigger or shorter? Do you want to have more water content in your product? More solid content? More terpenes? Do you want to fight drought conditions? Excess salinity? Insects? The specifics of what you want will guide you into choosing an appropriate hormone for your specific needs.



Examples of widely used plant hormones

Plan your hormone applications strategically. Different hormones can stimulate different processes that are needed at different points of a plant's life. If you plan the use of hormones carefully you can stimulate root growth when plants are transplanted, then stimulate flowering or other behavior when you want the plant to express that behavior more strongly. Plants take some time to steer, they react to their environment, hormone applications at the right times can give a plant a strong signal that it should follow certain behavior and you – as a grower – can ensure that the environmental conditions are perfect for the processes the plant will be carrying out next. Hormones are the flares telling the plant where to go, you should ensure you make that a smooth ride.

There is no free lunch. Plant hormones act to cause a certain behavior to happen, but this behavior comes at a specific cost. A plant that is stimulated to produce more flowers will

often grow smaller fruits, a plant that is stimulated to produce more terpenes might produce lower yields because of the additional energy spent in these molecules, a plant that grows more roots, grows less shoots while it's doing that, etc. A plant does not magically get access to more energy because it has been stimulated with a hormone, it simply chooses to act differently with the energy it is receiving.

Hormones interact with each other. A given hormone can behave in a way when it's applied and in a very different way when it's applied with another hormone. As different hormones signal different paths, the net effect is often related with how these different paths are activated. Some are synergistic, the total is more than the sum of the parts, while others are antagonistic, meaning you get less than the sum of the parts. Growers interested in hormones will often make the mistake of applying a lot of things at the same time, but they have no idea what the net effects are going to be like. When dealing with hormones introduce them one at a time and make sure you're getting a measurable positive effect before you venture into using another one with it. Incremental gains is the name of the game not "apply every hormone under the sun that has a peer reviewed paper published where it increases yields in a plant".

Concentration is everything. To make things even more complicated, a hormone might activate one signaling path when it's present at a given concentration but a different one when it's present at a much larger concentration. Using the wrong concentration for the hormone might end up causing a completely different effect or an effect so pronounced that it's negative side effects are going to out-do the positive effects. Furthermore, this can also be genetic dependent, so when using hormones on new varieties or species it is always advisable to do a concentration trial across 2-3 orders of magnitude to see where the "sweet spot" for the desired effect is. Sometimes hormones are most effective at surprisingly low

concentrations – even 0.1 to 1 ppm – while other times they need to be applied in very significant amounts (100-300 ppm).

The application route and vehicle is very important. A hormone might be very effective when applied in a foliar spray, while completely ineffective when applied in a root drench. Sometimes the hormone requires specific additives or solvents to be used in order to ensure its absorption and others it needs to be applied at a very specific pH range or even just by itself. Knowing the particular application conditions of the hormone you want to use is also important to achieve the expected results.

These are some simple guidelines to consider when using plant hormones in your crop. Hormones are no miracle but they can certainly provide amazing improvements in yields and quality if used appropriately. Formulating a good hormonal regime, with adequately formulated foliar/root drenches, applied at the right times, with the right hormones, can provide amazing results. This however requires a lot of testing, a lot of effort and a lot of understanding about the plant being grown and its crop cycle. Every crop has its own genetic and environmental conditions and requires significant experimentation to achieve the best possible results.

Keeping plants short: Synthetic gibberellin inhibitors

Plants grow both vertically and horizontally. A plant will develop branches along its stem – expanding horizontally – and the stem will grow towards the sun, making the plant taller.

This vertical growth is almost always an undesirable quality, both in extensive and intensive crops, which creates an opportunity to improve plant cultures by attempting to reduce the height of plants. You can read more about why making short plants is important in [this post](#). Although there are many potential ways to achieve this – which I will discuss in detail in future posts – this post will deal with the most powerful tools that have been developed for this purpose, a class of plant growth regulators (PGRs) known as gibberellin inhibitors or more commonly as “growth retardants”.

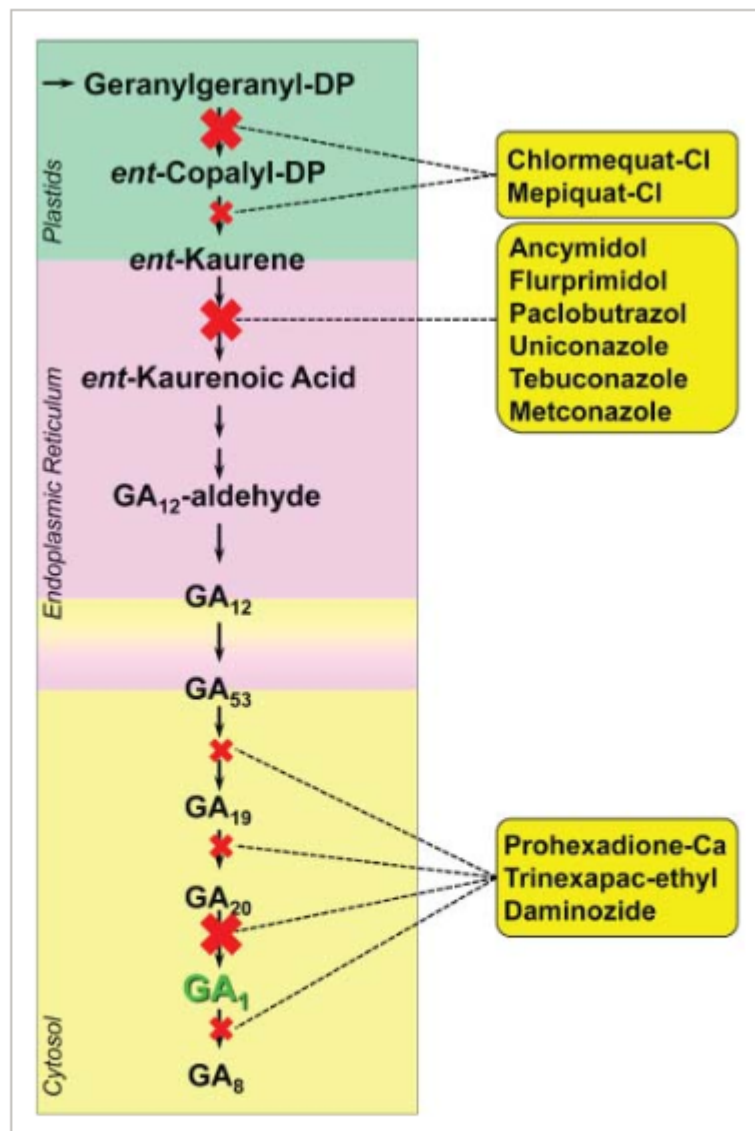


Figure 12.2

[Open in figure viewer](#) | [PowerPoint](#)

Main steps of gibberellin biosynthesis leading to biologically active GA₁ and points of inhibition by plant growth retardants. The cellular locations of the reactions is indicated by different greyscales. (The conversion of GA₁₂ into GA₅₃ can be located in both the endoplasmic reticulum or the cytosol.)

This figure was taken from [this article](#).

Making a plant grow shorter is no trivial task. This is because we do not want to make the plant less productive, but we want the same productivity of a tall plant in a much bushier and compact package. We therefore need to inhibit vegetative growth without affecting the flowering stages of our plant. Scientists figured out around 30 years ago that a set of plant hormones called gibberellins played a critical role in the vegetative growth of plants – especially the elongation of a plant -so these became a prime target to stop

growth. If you can disrupt the gibberellin creation pathway right when the plant is supposed to stretch, then the plant will stop growing vertically without the flowering development of the plant being affected at all.

We have found several different types of compounds that can do this. The figure above shows you the gibberellin synthesis path and the steps where different molecules have been shown to disrupt it. Among the most powerful and commonly used were the ones that disrupted the conversion of kaurene to kaurenoic acid, with the most famous one being paclobutrazol. In the other groups the most commonly used ones were chlormequat and daminozide. These molecules are all part of the first generation of gibberellin inhibitors and they did exactly what they were supposed to, proving to be extremely powerful growth retardants that were able to keep plants compact and strongly increased yields in several different crops.

However it soon became evident that their toxicity and retention in plant tissue is significant. Paclobutrazol has been shown to be toxic, having developmental and reproductive effects in rats ([1](#)) although it has been shown not to be carcinogenic in humans but still very toxic to aquatic life ([2](#)). The use of paclobutrazol on food crops is therefore not recommended, but whether or not it's actually allowed or not depends on the legislation of the country where you're in. Some countries will allow paclobutrazol to be used as long as enough time is given between application and the development of the edible parts of the crop and then again this usually only applies to a limited number of crops where the time between use and harvest can be guaranteed to be long enough. Chlormequat and daminozide follow similar stories, although in the case of daminozide it was discovered that it was carcinogenic and its use in edible crops was completely banned world wide in the late 1980s.

Table 2. Pesticide analytes and their action levels

Analyte	Chemical Abstract Services (CAS) Registry number	Action level ppm	Analyte	Chemical Abstract Services (CAS) Registry number	Action level ppm
Abamectin	71751-41-2	0.5	Imazalil	35554-44-0	0.2
Acephate	30560-19-1	0.4	Imidacloprid	138261-41-3	0.4
Acequinocyl	57960-19-7	2	Kresoxim-methyl	143390-89-0	0.4
Acetamiprid	135410-20-7	0.2	Malathion	121-75-5	0.2
Aldicarb	116-06-3	0.4	Metalaxyl	57837-19-1	0.2
Azoxystrobin	131860-33-8	0.2	Methiocarb	2032-65-7	0.2
Bifenazate	149877-41-8	0.2	Methomyl	16752-77-5	0.4
Bifenthrin	82657-04-3	0.2	Methyl parathion	298-00-0	0.2
Boscalid	188425-85-6	0.4	MGK-264	113-48-4	0.2
Carbaryl	63-25-2	0.2	Myclobutanil	88671-89-0	0.2
Carbofuran	1563-66-2	0.2	Naled	300-76-5	0.5
Chlorantraniliprole	500008-45-7	0.2	Oxamyl	23135-22-0	1
Chlorfenapyr	122453-73-0	1	Paclobutrazol	76738-62-0	0.4
Chlorpyrifos	2921-88-2	0.2	Permethrins*	52645-53-1	0.2
Clofentezine	74115-24-5	0.2	Phosmet	732-11-6	0.2
Cyfluthrin	68359-37-5	1	Piperonyl butoxide	51-03-6	2
Cypermethrin	52315-07-8	1	Prallethrin	23031-36-9	0.2
Daminozide	1596-84-5	1	Propiconazole	60207-90-1	0.4
DDVP (Dichlorvos)	62-73-7	0.1	Propoxur	114-26-1	0.2
Diazinon	333-41-5	0.2	Pyrethrins†	8003-34-7	1
Dimethoate	60-51-5	0.2	Pyridaben	96489-71-3	0.2
Ethoprophos	13194-48-4	0.2	Spinosad	168316-95-8	0.2
Etofenprox	80844-07-1	0.4	Spiromesifen	283594-90-1	0.2
Etoxazole	153233-91-1	0.2	Spirotetramat	203313-25-1	0.2
Fenoxycarb	72490-01-8	0.2	Spiroxamine	118134-30-8	0.4
Fenpyroximate	134098-61-6	0.4	Tebuconazole	80443-41-0	0.4
Fipronil	120068-37-3	0.4	Thiacloprid	111988-49-9	0.2
Fonicamid	158062-67-0	1	Thiamethoxam	153719-23-4	0.2
Fludioxonil	131341-86-1	0.4	Trifloxystrobin	141517-21-7	0.2
Hexythiazox	78587-05-0	1			

Table taken from [here](#), these are substances banned for use in cannabis by the state of Oregon. You can see how several of the above mentioned growth retardants are present.

The above developments caused chemical companies to search for and develop new gibberellin synthesis inhibitors with lower toxicities and lower accumulation in plants that could be approved for use in edible crops. This led to the development of Prohexadione-Ca and Trinexapac-ethyl, which are two of the most commonly used growth retardants right now. These two have considerably lower toxicities and lower half-lives in the environment. For this reason trinexapac-ethyl has been approved for general use in places like New York (3). In this document the toxicity for mammals and aquatic life is discussed and trinexapac-ethyl is not found to be a threat to humans or animals at the maximum suggested application rate.

This is mainly due to the fact that it's quickly bio degraded in the environment. A risk assessment made by the EFSA also reached similar conclusions (4). Another EFSA risk assessment for prohexadione-Ca also points in the same direction (5). Prohexadione-Ca is currently approved by the EPA for use in apples, grass grown for seed, peanuts, pears, strawberries, sweet cherry, turf, watercress, alfalfa and corn (6).

Optimal results with these new growth retardants also require careful consideration of the application formulation, the application time and adequate pairing of the PGR with the plant being grown . For example in apple trees much larger doses of Trinexapac-ethyl are required compared to Prohexadione-Ca to achieve the same results and trees that have been treated with Trinexapac-ethyl can have important reductions of flowers in subsequent crops (7).

With the development of less toxic and still highly active growth retardants, it might seem like a no-brainer to use these in crops to prevent elongation and increase yields. However the introduction of inhibitors in the gibberellin pathway is not without further consequence as this path is also important to guide the production of important phytonutrients and essential oils. When using these growth retardants it's important to evaluate their effect in the quality of the product, as they can also lead to a change in the properties of the end product. For example in apples these PGRs can induce the production of luteoforol, a flavonoid they normally do not produce (8).