Environmental issues by Heimen Julius

Abstract. Soil residues of glyphosate from spraying all plants in a field could severely harm soil fertility first and in a later stage make most plant life impossible. Such a disaster would be caused by the destruction of mycorrhizal networks.

1. Introduction

Glyphosate

Glyphosate, a herbicide produced by Monsanto since 1971, is more widely known as 'Roundup.' It is available as a weed killer for gardens from supermarkets and hardware stores. To be effective it must be sprayed onto the leaves of the plant.

Glyphosate is also used in agriculture. Many weeds are deep-rooted perennials with tubers and rootstocks. This makes them very difficult to eradicate. Through glyphosate all these problems were solved. Because, once past the leaf surface glyphosate moves throughout the plant, reaches deep into the roots and kills.

The kill-all problem

With such an all-round plant killer, you have to be very careful not to hit your crop as well. At this point a bright spark came up with the idea to make all crops glyphosate resistant. Then you could spray indiscriminately and only weeds would be killed. With the new genetic engineering techniques this idea was becoming feasible. It would result in convenient agricultural practices for farmers, who would also use much more glyphosate. And guess what, this was good for Monsanto's bottom line. So, for the last ten years genetic engineers have been making a wide variety of crops glyphosate resistant. However, a few essentials were overlooked. To understand glyphosate's potential for disaster, we need to know more about plants first.

The structure of plants

Despite their great variety, the structure of plants is very much the same everywhere: a stem, branches with leaves (and often flowers) and roots. That's all there is to it. Even branches are not needed, oaks have them, palms not. There are many different roots: pen roots, spread out roots, rootstocks, tubers, and so on (1, 3).

The internal structure of plants is also rather uniform. They all have a tubular system of two vessel systems bundled together. The xylem vessels transport water upwards with dissolved minerals from the soil. The phloem vessels distribute water throughout the plant with self made food (glucose products).

In dicotyl plants (oaks, geraniums) the bundled vessels are in the stem arranged in a circle right under the bark with the phloem vessels on the outside. With ring-barking you destroy the phloem system and the tree dies. On the other hand in monocotyl plants (palms, grasses) the bundled vessels are placed ad random in the stem (1).

There are many points of interaction between both vessel systems and water with dissolved minerals can freely move between them (2). In leaves the bundled vessels are visible as the leaf veins. So much for plant **structure**. Now, how do plants operate?

The workings of plants

Leaves are bathed in air and sunlight while water evaporation is going on at their surfaces. Water in the xylem system ends up in the leaf veins. The same xylem vessels go right down the stem into the roots, which are always seeking out sources of water. Roots tapping into water combined with evaporation at the leaf surfaces causes water to be drawn up into the leaves. Hence, the xylem flow has also been called the transpiration flow.

Chlorophyl gives plants their green colour. In the presence of water and sunlight it takes up carbon dioxide from the air and transforms it into glucose, which is a sugar. This process is called photosynthesis. Glucose flowing through the phloem vessels reaches all parts of the plant. So, both vessel systems provide water and food supply throughout the plants.

Glucose from photosynthesis turns out to be **the** basic chemical in nature and through many transformations all other natural substances are made. Transformed as starch it is stored in roots (potatoes). As sucrose, it is stored throughout the plant (sugar cane). As cellulose it forms the structural skeleton of plants and combined with lignin it becomes wood; cotton is a form of cellulose. Glucose in combination with nitrogen forms amino acids, which are the building blocks of protein. And oils too are transformations of glucose.

How plants grow

Plants grow not like animals. In animals the whole body participates. But plants grow only from specific growth centres, the meristems, where you find ongoing cell division. This means in practice that plants grow only from the tip of the stem and if this has branched from the tips of the branches. With roots it is the same thing: growth occurs only from the tips. But in dicotyl plants the roots and stems can become thicker. This happens through a tissue called cambium which forms inside the stem a kind of cylinder together with the xylem/phloem vessels. So, the cambium is another meristem. On the other hand in monocotyl plants there is no cambium and they stay slim (palms). Some monocotyl plants like grasses have nodes and bamboo which is a giant grass, has very visible nodes. These nodes contain meristem tissue and grasses grow from their nodes.

Phosphate chemistry

None of the chemical substances participating in cell growth are particularly reactive. Left by themselves nothing would happen. But in the presence of phosphate all transformations occur at blinding speed, whereby phosphate groups switch from chemical to chemical like monkeys swinging from tree to tree. In the process chemicals are joint together and split apart forming new molecules needed for cell growth and normal living processes. Phosphate moves throughout the plant and when in short supply, phosphate in older leaves is often mobilised and transferred to young rapidly growing leaves (4). This moving to new locations is called translocation.

The cell nucleus is a chapter apart as far as phosphate is concerned. Here phosphate groups are permanently built into the DNA, which is the carrier of the genetic code. It determines the sequence of the genes. The same goes for RNA, which is essential for protein building. It determines the sequence of the building blocks of protein. So, in growth centres (meristems) phosphate is needed in large amounts (4). If phosphate is so essential for all life processes, where is it coming from? To find out we have to delve into the soil.

Soil, roots and phosphate

Soil consists of disintegrated rock with organic material on top of it. Rock particles are classified by size. Sand particles (2 to 0.05 mm), silt particles (0.05 to 0.002 mm), and clay particles (less than 0.002 mm), are found in varying amounts forming different kinds of loam (5). But this does not say anything about their chemical composition. In general, phosphate levels are low and it is firmly adsorbed to clay. Over time phosphate becomes even more insoluble through slow, ongoing chemical reactions (8). No phosphate is leached from soil during a water pour-down through this strong adsorption

Soil structure is very important. Tiny clay particles adhere to much larger sand and silt particles. Claycoated particles are clustered into micro-aggregates and in their turn into larger aggregates. This all happens through electrostatic forces and the gluing of polysaccharides leaked (exuded) from plant roots. To complete the picture: water forms a film on the clay surfaces and here live all soil bacteria. The spaces in and between aggregates are generally filled with air (6, 7).

This is the environment in which plant roots extract phosphate from soil. They do this through normal diffusion processes, which are far too slow and inefficient for the amounts needed. Even root hairs assist only marginally. Fortunately nature has developed a second way to get phosphate into plants. This is through a close cooperation with certain moulds or fungi which extract phosphate from soil.

Mycorrhizas

Mycorrhiza means literally fungus-root. Most plants have a number of fungi hooking into their roots. Mycorrhizas are very good at penetrating the soil structure. With their thin threads (their hyphen) they can get into the fine pores of micro-aggregates. Here mycorrhizas dissolve the available phosphate, which is transported via their hyphen to the plant roots (9). In return mycorrhizas have easy access to plant food flowing through phloem vessels as part of normal nutrients supply to roots (11).

Mycorrhizas are not specific about the roots they hook into. So, they hook into the roots of different plants and form underground networks connecting many unrelated plants. They transport nutrients to all 'their' plants so that a situation of host and recipient plants is created, whereby excess nutrients in one plant is passed to plants with a shortfall (10). A wide range of minerals is transported through these networks such as sodium, calcium, zinc, sulfur, phosphorus and potassium to name some important ones. Also, substantial amounts of water flow through these networks and carbon (sugar etc.) is freely moving among neighbouring plants (12, 13, 14). This nutrient transport occurs through concentration gradients.

That phosphate is free to move between plants was demonstrated when radioactive phosphorus applied to the leaves of one plant, was shortly after application discovered in the shoots of neighbouring plants (13, 19). Phosphorus was apparently transported via underground mycorrhizas, as phosphate in direct contact with soil is immediately adsorbed to soil particles and does not move any more. Although the entrance point of this radioactive phosphorus was unusual (taken up by the leaves instead of the roots), the phosphorus was normally processed and any excess was passed onto neighbouring plants.

Mycorrhizal structure

A closer look at the structure of mycorrhizal networks revealed that not all their threads (hyphen) were the same. Hyphen with a large diameter and without cytoplasm were wrapped in a sheath of hyphen with smaller diameter and full of cytoplasm. It was realised that hyphen with a large diameter and without inside 'hindrances' were very suitable for water transport over longer distances. In other words that they were the equivalent of xylem vessels in plants. On the other hand, the narrower hyphen with

cytoplasm inside corresponded to phloem vessels in plants. And so it was concluded that mycorrhizas were structurally and functionally comparable to plant roots (15, 16). In fact they were their natural extension, hence the name fungus-root.

The deep penetration of soil structure by mycorrhizal hyphen is of special importance in 'mining' scarce resources like nitrate and phosphate (17, 18). What has been repeatedly found is that after infection with mycorrhizas most plants gain more phosphate and grow better.

(For more information on mycorrhizas see also references 20, 21)

Interim summary: plants in action

Plants have a double vessel system. The xylem brings water up from the soil with dissolved minerals. Through evaporation at the leaf surfaces water is pulled upwards from the roots, which are always seeking out sources of water. The chlorophyl in leaves enables plants in the presence of sunlight to convert carbon dioxide from the air into glucose, which is **the** basic chemical of all plant biochemistry. Glucose and its products are distributed throughout the plant as food via the phloem. Phloem and xylem are at many points in open connection with each other.

Phosphate is needed for all biochemical processes in plants, but is in short supply because of its strong adsorption to soil. Most plant roots are insufficiently developed to extract sufficient phosphate from soil. However, a close cooperation with specific fungi, the mycorrhizas, overcomes this problem. The mycorrhizas are good at mining minerals, including phosphate and transport this back to the roots they hook into. Mycorrhizas do this to get easy access to the flow of plant food nourishing the roots. Mycorrhizas are not choosy and hook into many unrelated plants. In doing so they form networks and they transport minerals, water and carbon (sugar etc.) from plants with an excess to plants with a shortfall. This happens through concentration gradients.

Let us now see what glyphosate is up to.

2. Glyphosate

Some characteristics

Its correct chemical name is a mouthful: [N-(phosphonomethyl)glycine]. The 'phosphono' bit refers to the presence of a phosphate group. It is an organo-phosphate herbicide (22) and an extremely stable one. Pure glyphosate dissolved in distilled water is stable for many years. Roundup, a diluted formulation of glyphosate, hardly undergoes any changes for 7 months when kept in glass bottles at room temperature (23). And glyphosate of 98% purity is biologically and chemically stable to moist heat sterilisation (75).

Glyphosate in soil is quickly inactivated through adsorption to soil particles, especially to clay and organic material (22, 23, 48). Clay soils with their very fine structure and large surface area have a high capacity for glyphosate adsorption. Sandy soils which are much coarser have less surface area and adsorb therefore less glyphosate. In addition soil minerals like iron and aluminium influence this adsorption (22, 52). It turns out that glyphosate and phosphate are competing for the same soil binding sites (24, 25), but phosphate always wins as its adsorption to soil is much stronger. Glyphosate is pushed away when phosphate is added to soil and this makes glyphosate adsorption reversible (22, 24).

To be effective glyphosate must be applied to the leaves of a plant. When applied to a leaf around half the glyphosate gets past its surface and joins the ploem flow of photosynthetic products (25, 26, 27, 28, 29). Glyphosate can also be taken up by the roots. Then it moves with the regular water intake via the xylem vessels (22, 24, 25, 30, 31).

Glyphosate acts like a false phosphate. While real phosphate moves freely throughout the plant to assist in biochemical processes, this false phosphate accumulates in the meristems. Here it blocks a specific biochemical reaction.

Glyphosate toxicity

Once arrived in the meristems, glyphosate starts to compete with a specific enzyme. When glyphosate wins, a biochemical chain reaction grinds to a halt. This is the shikimate pathway, which is a major biochemical path in plants and microorganisms (33). The result of this blocked pathway is that three aromatic amino acids are not formed. This brings the protein synthesis to a halt like a biochemical stranglehold and the plant begins to die (32, 33, 34, 35, 36). Typically, the protein synthesis is resumed when the missing amino acids are added to glyphosate treated plant cells (32, 75, 76).

Glyphosate resistance

Some plants have more of this specific enzyme than other plants. It turns out that the more specific enzyme a plant has, the more glyphosate is needed for a lethal effect (37, 38, 39). Thus, more enzyme makes the plant more glyphosate resistant. Enzyme levels can be artificially manipulated. When carrot cells were grown in increasing, non-lethal concentrations of glyphosate, they were stimulated to produce more enzyme, which made them more glyphosate resistant (40, 41).

Genetic engineering

At this point genetic engineers moved in. First, they selected cells from a Petunia plant, which were grown stepwise on nutrients with increasing amounts of glyphosate. This made these cells more glyphosate resistant. In the end these cells overproduced the desired enzyme 15- to 20-fold. Then, this genetic material was transferred to other plants. The resulting transgenic plants were also glyphosate

resistant (42, 43). Also, bacteria with a high glyphosate tolerance were used. Their genetic material transferred to crop plants gave glyphosate resistant crops such as cotton (44. 45).

That glyphosate resistant crops will stimulate glyphosate usage is generally accepted as the following quotes illustrate. 'The recent development of soybean genetically engineered to be resistant to postemergence applications of glyphosate will expand glyphosate use in conventional and no-till soybean production' (46). 'Recently, genetically-engineered glyphosate tolerant crops, notably soybean and cotton have been marketed in North America under the Roundup Ready label. This development will undoubtably increase glyphosate use' (47).

What will be the effect on the environment? The Agricultural Research Department of Monsanto claims that glyphosate is completely and rapidly degraded in soil and water (49). But this turns out to be incorrect. It is therefore unfortunate that this assertion has been repeated in a number of textbooks (33, 50). A startling admission from a Monsanto research team 20 years later is the following: ... "the residual effect of glyphosate in soil is not expected to provide selection pressure on local weed populations" (86). Quite an admission after maintaining for years that glyphosate was quickly and completely degraded by soil bacteria! Let us have a closer look at this degradation.

Glyphosate degradation

Glyphosate is only degraded by bacteria, which vary in number and kind from location to location depending on the soil. Many bacteria do not degrade glyphosate and how complete it is degraded depends on the kind of bacteria that are present (51).

How speedily is this breakdown? It took 28 days to degrade 45 to 48 % of the original glyphosate in a sandy loam and/or sandy clay loam (49, 51). As a microbiological process this is at a snail's pace. Contrast this with food poisoning where bacteria reach dangerous levels in a few hours under the right circumstances. Rapid microbiological breakdown would mean 'done in a few days.' However, complete degradation of glyphosate took 112 days in a shake flask culture. This is almost 4 months (49). Especially in a **shake** flask culture a rapid microbiological breakdown should not take more than a few hours, as this method provides maximal exposure of glyphosate to bacteria. This situation never occurs in the field.

Because of the slowness of degradation, microbiologists think that glyphosate breakdown occurs through 'co-metabolism.' This means that glyphosate is not essential for bacterial growth and most of the time bacteria leave it alone. This enables glyphosate to accumulate in soil even in the presence of bacteria that could degrade it (55).

Interim Summary: glyphosate

Glyphosate acts like a false phosphate in soil and plants. It blocks a biochemical pathway (shikimate pathway) and as a result the plant dies. Glyphosate is very stable, not rapidly degraded by bacteria and leaves residues in the soil.

The average reader can skip here and start reading again at the end of the wide left margin under 'the mycorrhizas revisited' on page 8.

Slow degradation confirmed

Monsanto's report (49) mentions also research from others. It concerned field studies of eleven different soils, which covered a full range of soil types and geographical areas. The results showed that **half** the amount of glyphosate was degraded in 2 months on average (49). Slow degradation rates were also found in other studies. In Hawaiian sugarcane soils, glyphosate was broken down by half in times varying from 18 days to 22.8 **years** (54).

A later review on glyphosate breakdown showed that glyphosate **half-lives** varied from 18 days to 270 days in agricultural soils. For forest soils this varied from 14 days to 45 days and in some cases from 65 to 200 days (55). These data of laboratory trials from different soils compared with field data showed that there was good agreement (56).

Glyphosate residues in soil

Given the real situation, it is not surprising that glyphosate residues were found if people cared to look for them.

In Finland the persistence of the herbicides MCPA, glyphosate, maleic hydrazide and tri-allate and the insecticide parathion in crop land were measured. Within a short time most residues were very low, except for glyphosate. Parathion was below 0.02 mg per kg in 11 days, while MCPA was below 0.06 mg per kg in 7 days. But glyphosate settled to a level of 0.2 mg per kg during the following summer (57). This was ten times the level of parathion in 11 days.

In arctic forest soils something similar was found. Radioactive glyphosate was applied to soil samples. From measuring the various soil fractions it was concluded that glyphosate residues could remain at high concentrations for more than 1 year after application (58).

After aerial spraying glyphosate was found to settle in the bottom sediments of water flows and was considered biologically unavailable through soil adsorption (59). Soil samples from nearby land showed for glyphosate an estimated half-life of 45-60 days. After 360 days the total soil residue was still 6 -18% of the initial glyphosate levels (60).

Soil in dry irrigation canals was also investigated. Samples were taken in spring the day before the canals were filled. These samples contained still 0.35 ppm glyphosate. This was 23 weeks after the canals were sprayed in autumn to keep the weeds down when in winter the canals stood dry (61).

An entirely different kind of research found how to extract glyphosate from soil and clay minerals. Glyphosate extraction was possible for up to 1 part per million from clays and organic matter, and 0.5 ppm from sandy soils (77).

Glyphosate activity in soil

As long as people think that glyphosate is always firmly bound to soil and inactive, no one really seems to care. But there is evidence that glyphosate even in soil can be active. Salazar et al. found glyphosate applied to soil, especially to moist soil, could be toxic to plants up to 5 days after application (63). It is possible that in really moist soil glyphosate did not get completely adsorbed to soil particles and remained in solution with toxic effects.

In sandy loam of normal humidity glyphosate was not completely inactivated as measured by the root weight of clover and nitrogen fixation after 120 days or 4 months (64). This of course raised the question whether glyphosate in soil could be taken up by plant roots.

This issue was researched by Sprankle et al. They planted soybean and corn seeds together in one pot with sandy loam, which was treated with glyphosate. After 16 days the plants were examined and small amounts of glyphosate were found in the shoots and roots of the plants (24). The small amounts taken up by the plants led Hance to conclude through comparisons with other herbicides that the low activity of glyphosate in soil was due to its low intrinsic toxicity when applied to roots (79). This missed the point totally as it had been well established that glyphosate in nutrient solutions was readily taken up by roots and toxic as ever (24). A researcher from Monsanto held the same view: "Plants were treated via the roots in hydroponic solution. This was considered to be the most efficient means of loading the plant with radioactive glyphosate" (53).

It is therefore puzzling that it is always Monsanto that is trotting out this Hance research with the claimed intrinsic low toxicity of glyphosate when applied to roots. It could well be that Monsanto's own dissipation study has something to do with this endorsing of Hance's research.

Monsanto's dissipation study

Monsanto had made the claim that the glyphosate dissipation rate of 2 soils was 90% in less than 12 weeks (49). Thus, in less than 3 months 90% of glyphosate had disappeared from the researched soil. This was supposedly through bacterial activity. But anyone looking up this research will see that also corn seeds were planted in the trays with glyphosate treated soil. So, obviously the corn seedlings had taken up some glyphosate from the soil through their roots. While Sprankle et al's trial (24) took only 16 days, Monsanto's trial took three months (49). Besides the used glyphosate concentrations in the soil were low. So, in Monsanto's research two agents would have contributed to glyphosate dissipation from soil. These were soil bacteria that break down glyphosate and seedling roots that take up glyphosate. It is therefore impossible to claim that all glyphosate dissipation was from bacterial activity.

Glyphosate residues in water

Glyphosate residues persist also in water. This was discovered as follows. In two filled irrigation canals glyphosate was injected at a calculated rate (61). About 70% of this glyphosate was found at 1.6 km downstream from the injection point. So, it did not readily dissipate in water. About 58% of glyphosate was found at the end of the canals, which were respectively 8 and 14.4 km long.

In a follow up study was claimed that no damage was done to plants watered with irrigation water contaminated with glyphosate (73). However, this water was piped through copper tubing. Later research found that glyphosate could be strongly adsorbed to copper oxide and copper ions (74). Thus, copper tubing would protect plants irrigated with water contaminated with glyphosate. The generalisation that irrigation water polluted with glyphosate was safe to use on crops turned out to be too optimistic as the next research showed. This research found that glyphosate was not well adsorbed to the suspended solids of turbid irrigation water and remained toxic to safflower plants when their roots were placed in it (62).

Conclusion

Glyphosate leaves residues in soil and water, which can be taken up by plant roots under certain circumstances.

The mycorrhizas revisited

Mycorrhizas hook into any plant within reach and connect weeds with crop plants. Through mycorrhizas excess nutrients in one plant are shunted towards plants with a shortfall. This happens through concentration gradients. Mycorrhizas seem to handle glyphosate the same way. This was illustrated by Rodrigues et al. They took sandy loam and planted wheat and soybean in one pot side by side. When the wheat plants were 2 weeks old, soybean seeds were put in the soil and the wheat plants were treated with glyphosate. During this procedure the soil was carefully protected from glyphosate contamination. By varying the number of wheat plants and the concentration of glyphosate, they could cause growth effects in the soybean seedlings. This experiment was repeated with wheat plants and corn seedlings. They got similar results. In the corn seedlings they found radioactive glyphosate in all its parts.

Their interpretation was that glyphosate had been leaked from the roots of the wheat plants and taken up by the soybean and corn seedlings (78). A more likely explanation is that mycorrhizas connected all plants and shunted glyphosate from wheat plants with a high glyphosate concentration to seedlings without any glyphosate. Rodrigues et al. were not aware of the existence of mycorrhizas and this is also Monsanto's blind spot.

A related issue is whether mycorrhizas could be affected by glyphosate transport through their networks as the shikimate pathway is present in all microorganisms including mycorrhizas. Therefore, glyphosate is also toxic to them.

Glyphosate toxicity to microorganisms

Monsanto claimed that glyphosate had a minimal effect on microorganisms in soil (49). However, a later review of Monsanto's soil testing found that soil containing 4 and 8 ppm glyphosate was a low concentration for field applications even before the introduction of genetically engineered crops (68). Thus Monsanto's generalisations based on this research are irrelevant for the amounts that will be needed on genetically engineered crops.

Monsanto was not the only one who researched this issue. Research on glyphosate effects on soil microorganisms (lasting 214 days or almost 7 months), found that stimulation and inhibition occurred depending on glyphosate concentrations (69). Also, research with two soils treated with glyphosate found that glyphosate inhibited microorganisms, but also that some adaptation to glyphosate occurred (65).

Soil carefully treated with glyphosate remains of course an artificial thing. But what about glyphosate that is leaked from plant roots? Grossbard pointed out in 1985 that there was a great gap in our knowledge on the response of soil microorganisms to glyphosate exuded from roots (72). This ignorance persists.

Laboratory research without soil found that 50 ppm (parts per million) glyphosate reduced bacterial growth by 73%, fungal growth by 91% and actinomycetes' growth by 94% (66). This last category are bacteria with some characteristics of fungi. So, a clear limit was found to glyphosate tolerance in microorganisms (from arable land). The absence of soil in this research was criticised (67).

A similar research was done with five identified mycorrhizas. Here, a glyphosate concentration of 50 ppm and more led to significant reductions in growth (71). In addition it became clear that each mycorrhiza had its own individual glyphosate tolerance, just like plants. The absence of soil in this research reflected reality as mycorrhizas hook directly into plant roots and get glyphosate full strength from the plant's phloem flow.

Research on Rhizobium bacteria in shake flask culture without soil, suggested that the ability to degrade glyphosate could be widespread among Rhizobium bacteria. Here too, an upper glyphosate limit was found above which bacterial growth was inhibited (70). What amazed these researchers was that by 1991 so few glyphosate-degrading bacteria had been isolated.

Interim conclusion

Clear upper limits have been found to glyphosate resistance in microorganisms.

Interim summary

Glyphosate acts like a false phosphate, is chemically very stable and poorly degraded by bacteria. Around half the glyphosate applied to a leaf gets past its surface and moves with the phloem flow to the growth centres (meristems). Here it competes with an enzyme and if it wins this competition, then a biochemical pathway is blocked. This is the shikimate path, which is present in microorganisms and plants alike. When the Shikimate pathway gets blocked the protein synthesis grinds to a halt and the plant dies. By increasing the level of this particular enzyme through genetic engineering, plants can become glyphosate resistant. Such genetic engineered crops open the way to spray all plants in a field with glyphosate. Only the weeds will die. This would bring large amounts of glyphosate into the environment and result in increased residues in soil and water. Increased accumulation of glyphosate could affect all soil microorganisms.

Glyphosate and the mycorrhizal network

How will the higher glyphosate levels used on genetically engineered plants affect the mycorrhizal networks? To give an idea let us take a research with glyphosate-tolerant soybean and weed control. Johnsongrass (Sorghum halepense) is one of the most common and troublesome weeds in the south of the United States. It can reduce soybean yield by more than 50%. Glyphosate is effective in controlling Johnsongrass, but it is equally toxic to soybean. So, to avoid contact with soybean, the herbicide had to be applied with selective application equipment, usually after Johnsongrass had grown taller than soybean.

With glyphosate-tolerant soybean however, glyphosate spraying can be done any time throughout the soybean flowering stage. For a particular research a 130- horsepower tractor with a 18.29 m boom sprayer was used (80). This resulted in all plants on that field, soybean and Johnsongrass alike being dosed with glyphosate. Above the ground everything had been figured out: soybean would not be harmed, Johnsongrass would be killed. But what about underground?

When only weeds were sprayed with glyphosate, extras in glyphosate would have been channelled away via mycorrhizas to many other plants in line with concentration gradients. In the process glyphosate would have been so diluted that probably no harm would have come to other plants.

With genetically engineered crops however, the glyphosate entry points of the mycorrhizal network might have been doubled or more, as **all** plants of that field were sprayed. As a result all plants of that field leaked glyphosate into the soil and tried to get rid of it through the mycorrhizal networks. However, this time with all plants sprayed there were no longer concentration gradients. So, the mycorrhizal networks would have been filled up with glyphosate and the question arrises: could mycorrhizal networks be poisoned? This seems to be a distinct possibility in view of Orobanche research

Orobanche controlled by Glyphosate

The plants from the Orobanche family, the Broomrapes, are root parasites. They are totally invisible as they have no parts above the ground. They are like clumps of wirwar roots attached to the roots of other plants. All you see is that those plants infected with Orobanche are not thriving. Underground however these parasites hook into the roots of any broadleaf plant in sight, connect up with their vessel system (phloem and xylem) and suck their juices.

Orobanche hooked into carrot roots could be controlled with glyphosate, as carrots are rather glyphosate resistant. By spraying the carrots, sufficient glyphosate was passed onto Orobanche to poison it. With the introduction of glyphosate resistant crops this Orobanche control was further extended (81, 82).

Mycorrhizas again

The mycorrhizal networks are in a similar boat as Orobanche. Only mycorrhizas are glyphosate **resistant** and Orobanche is glyphosate **sensitive**. Under normal circumstances mycorrhizas grow rapidly and their degradation rates are high as is the case for fine root hairs (83). This rapidly shedding protects plants and mycorrhizas. High glyphosate input will further accelerate this shedding process. But what then?

Soil biomass

The biomass in soil from mycorrhizas and fine roots can be considerable. In two temperate forest stands it was claimed that fine root production was roughly equivalent to leaf biomass production (92). It has also been claimed that the mycorrhizal biomass is the largest microbiological component of many forest soils and contains large reserves of nitrogen, phosphorus, potassium and magnesium (83, 84). Taken together, fine roots and mycorrhizas contribute between 84 and 78 percent of the total tree organic matter to the soil.

We are not particularly concerned about forests at this stage, because of the absence of genetically engineered crops there. But, what to expect in arable land where each year with genetically engineered crops a few more showers of glyphosate come down? We have here a mechanism whereby over the years glyphosate is accumulating in soil without being firmly adsorbed to soil particles, but locked into biomass. What then happens is any one's guess and depends on local circumstances. Bacteria feeding on this biomass would find that this time the filling in the cake was highly toxic.

Glyphosate mining?

One particular aspect is also totally unknown. The phosphate group of glyphosate seems to trick plants. As a result fake-phosphate is handled as if it were real phosphate. So, glyphosate passes all plant membranes and goes with the flow till it reaches the meristems, gets locked in and then the stranglehold starts to work. Could it be that mycorrhizas are also tricked in accepting glyphosate as if it were phosphate? Given the fact that glyphosate is easier to dislodge from soil particles than phosphate, could this lead to glyphosate being mined and channelled back to plants instead of phosphate? No one really knows.

A dark scenario

There is a possibility that eventually so much glyphosate will accumulate in soils that arable land will get too toxic to grow anything. This will not happen overnight. But say over twenty or thirty years. Take note, we don't know what is there already after thirty years of weed spraying. Now add another

thirty years of all plants spraying and what will then be the situation? After a period of lowered soil fertility we might wake up one day seeing plants withering in the fields because soils have become too toxic for any plant life. Given the fact that glyphosate is presently the most widely used herbicide throughout the world, this new situation would mean that we were staring in the face of world famine and that arable land throughout the world would have been turned into biological deserts.

Before people start to sneer at such a dramatic scenario do remember that people in the 1950's and 60's would not have believed that a water volume the size of the Baltic Sea (Scandinavia) could become biologically dead. But by the mid 1980's this was the case.

Also, in the 1950's and 60's no one would have believed that motor cars, trucks and power generation plants could cause so much air pollution that it could cause climatic change. But, from the mid 1990's this is what we are facing.

"Data from pesticide research have already accounted for the appearance of growth irregularities in field plots and commercial fields where VAM fungi (=mycorrhizas) were killed." (95). The particular pesticide was not mentioned, but this is less relevant here because any effective pesticide will do. So, the signs are on the wall.

Another observation is also highly relevant, which concerns crop rotation. Maize (a strongly mycorrhizal plant) was grown after oil-seed rape (a non-mycorrhizal plant). Maize grown after rape grew poorly with typical phosphate deficiency symptoms. But maize grown after maize did well. Root samples collected in early summer showed 12% mycorrhizal infection in maize after rape and 71% infection in maize after maize. So, mycorrhizal networks are not indestructible and do depend on their environment for survival.

Increasing glyphosate resistance in weeds

Another factor is the development of glyphosate resistant weeds. With Monsanto's sledge hammer approach the obvious next step would be to raise the bar further and make crop plants even more glyphosate resistant followed by spraying with higher concentrations of glyphosate. This however, would give us only a comparatively short respite.

How futile this approach would be in the long run is illustrated by the fact that the 'world's worst weeds' are mainly facultative mycrotrophic. This means they are thriving with and without mycorrhizas. This offers them the advantages of adequate nutrient uptake in soils without mycorrhizas plus the ability to tap into mycorrhizal networks and use the juices from other plants (97, 98).

The roots of most agronomic crops however, rely on mycorrhizas for adequate nutrition. Such as: corn, cotton, wheat, potatoes, soybeans, alfalfa, sugarcane, cassava, dryland rice and most vegetables and fruits such as apples, grapes, and citrus. Many forest trees fall in this category including maple, yellow poplar, and redwood as well as important tree crops as cacao, coffee, and rubber. Two important groups of crop plants that do not form mycorrhizal connections are the Cruciferae such as cabbage, mustard, canola, and broccoli, and the Chenopodiaceae such as sugar beet, red beet and spinach. (91, 93, 94).

Monsanto, obviously totally ignorant about mycorrhizas is not aware that every time glyphosate is entering a mycorrhizal network, all weeds tapping into this network are handed the blueprint of this toxin and are given ample opportunity to come up with an answer.

A research team of Monsanto claimed in 1997 that it was hardly credible that weeds could ever develop glyphosate resistance (86). But in 1999 a research paper reported just that. A biotype of

Lolium rigidum from a field in northern Victoria (Australia) had developed glyphosate resistance. It turned out that glyphosate had been used for the past 15 years on that field. The new biotype was nearly 10-fold more resistant than the original glyphosate susceptible biotype (87).

It is obvious that the more glyphosate residues are left in soil, the more weeds have access to this poison on a permanent basis and the more weeds can develop glyphosate resistance. So, the only sane way to use this herbicide is on weeds only and not on whole crops. It is well worth to protect glyphosate in this way from weeds' capacity to develop resistance, as glyphosate is one of the very best herbicides we have at present.

New developments

Spray systems apply a single herbicide rate over an entire area and provide adequate weed control, but result in herbicide waste and environmental pollution. Weeds are mostly not evenly distributed over an entire field, but tend to occur sporadically. Therefore, selective spraying of only the weed portions would result in significant reductions in herbicide usage (88). This was achieved with a new weed sensing technology. Originally designed for non-crop situations, it was also made available to crop production. The reductions in herbicide usage were considerable (89, 90).

This new technology could well make glyphosate resistant crops superfluous. However, the sophistication of the equipment would seem to limit its use to developed countries. This leaves us still with the developing countries where farmers would be encouraged to use increasing amounts of glyphosate in combination with glyphosate resistant crops.

Conclusions

- 1 Monsanto's sledgehammer approach with glyphosate resistant crops is based on ignorance about the existence of mycorrhizas.
- 2 Glyphosate is chemically very stable and degraded only by bacteria at a snail's pace
- **3** The dosing of whole crops with glyphosate year after year, will lead to glyphosate accumulation in soil biomass through shed mycorrhizas and root hairs saturated with glyphosate.
- 4 It is unknown whether mycorrhizas would mine glyphosate from soil particles as if it were phosphate and channel it back to crop plants.
- 5 Increasing levels of glyphosate in soil will initially harm soil fertility and could later lead to an environment too toxic for any plant to grow.
- 6 Increasing levels of glyphosate in soil accelerate the development of glyphosate resistance in weeds. Some weeds have done this already.
- 7 the only sane approach to glyphosate use is to spray only the weeds. The use of weed sensing equipment should be promoted.

A final note: I read in a newspaper article that genetically engineered crops yielded less than normal crops. It could well be that this is already an indication of damaged mycorrhizal networks.

The overall conclusion must be that spraying whole crops with herbicide is a mad hatters approach steeped in ignorance and should be banned. This practice should also be banned for public health reasons as no one really knows what the effects will be of consuming food over a life time containing high levels of herbicides.

See further the paper on health issues

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