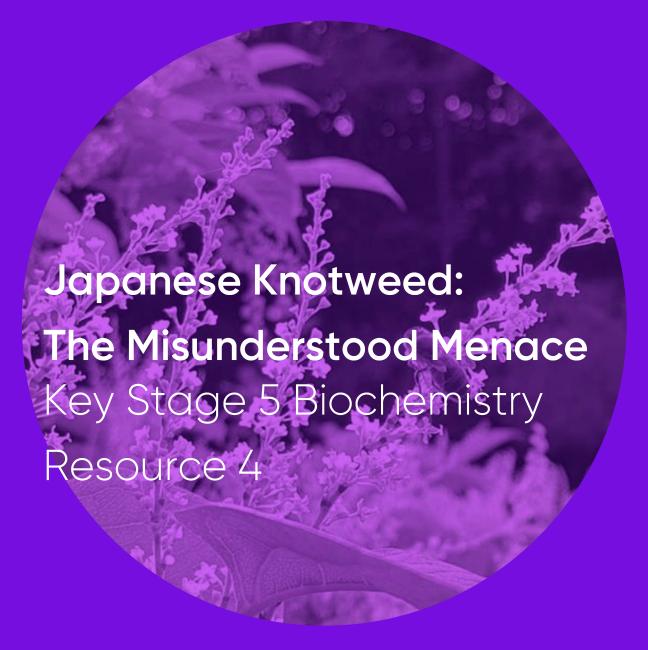
Research Based Curricula





Resource Four Overview



Topic Glyphosate Translocation and Transportation in Plants

A-Level Modules

Biology: Mass transport in plants

Chemistry: Atomic Structure, Mass numbers and isotopes

Objectives

By the end of this resource, you will be able:

- ✓ To define the phloem and xylem, and the process of mass transport in plants.
- ✓ To explain the use of tracers to investigate translocation in plants
- ✓ To define the meaning of an isotope, and explain how it can be detected using mass spectrometry.
- ✓ To understand what a herbicide does

Instructions

- 1. Read the data source
- 2. Complete the activities
- 3. Explore the further reading

Context

Japanese knotweed can be treated by a method known as chemical control. Chemical control uses chemical substances called herbicides which are toxic to the weed plants that they target. Herbicides can be sprayed onto the leaves or injected into the stem, In order to understand how herbicides move around the plant, this resource explains the two main plant transportation systems; the phloem and the xylem. Labelling experiments can be used to trace the movement of substances through these systems.



Resource Four Data Source

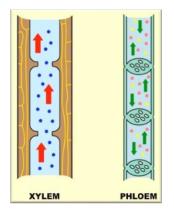


Section A

Plant Transport Systems

Just like us, plants need water and food to survive. Like our circulatory system, there are two main transport systems in plants; the phloem for food, and the xylem for water. The xylem transports water and mineral salts from the roots up to other parts of the plant, whilst the phloem transports sucrose and amino acids between the leaves and other parts of the plant.

The phloem moves food substances from where they are made, known as the sources, and where they are used or stored, known as the sinks. Photosynthesis uses the light energy which lands on the leaves of the plant to form energy-storing sugars, generally referred to as photoassimilates. These energy sources are transported in the phloem to other parts of the plant where they are needed for processes such as:



- growing parts of the plant for immediate use
- storage organs such as bulbs, tubers and rhizomes.
- developing seeds

Transport in the phloem is therefore both up and down the stem. Transport of substances in the phloem is called translocation. The movement of sucrose from mesophyll cells to phloem is active and requires energy from ATP.

Section B

Protein Synthesis

Plants absorb nitrogen from the soil through their roots in the form of nitrate (NO3-) and ammonium (NH4+). Nitrogen is transported from the root to the shoot via the xylem in the form of nitrate, dissolved ammonia and amino acids. Nitrogen is a key component of proteins. The DNA code for a protein remains in the nucleus, but a copy, called mRNA, moves from the nucleus to the organelle known as a ribosome where proteins are synthesised in the cytoplasm. Each sequence of three nitrogenous bases of mRNA that

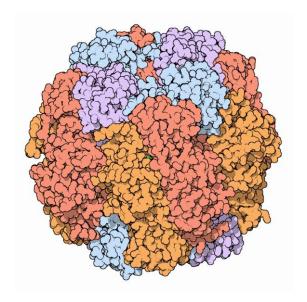
Resource Four Data Source



codifies one amino acid is called a codon. The protein produced depends on the template used, and if this sequence changes a different protein will be made. Carrier molecules bring specific amino acids to add to the growing protein in the correct order. Twenty different kinds of naturally occurring amino acids are converted from the nitrogen taken up into the plant through the roots. Each protein molecule has hundreds, or even thousands, of amino acids joined together in a unique sequence, known as the primary structure. It then folds either into an alpha-helix or a beta-sheet, known as its secondary structure. It is then folded into the correct unique shape, called a tertiary structure. When multiple folded chains of a protein join together they form a quaternary structure.

The overall 3D structure is very important, as it allows the protein to do its job. Proteins perform lots of important functions within cells. For example, proteins called enzymes catalyse reactions within the cell, and proteins called chaperones protect the cell from damaging heavy metals. The process of protein synthesis can be disrupted in plants by chemicals called herbicides, in order to cause damage to the plant.

This is the structure of RuBisCO, a plant protein essential for photosynthesis



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Section C Herbicides The growth of unwanted plants can be controlled by the application of chemicals known as herbicides. Herbicides may be selective and species specific, or be non-selective and target a broad-range of plant species. The persistence, or 'residual action' of an herbicide refers to the length of time it remains active in the soil for before degradation into its breakdown products. Herbicides have different uptake methods, for example through the stomata on the leaves or through the roots, as well as different transportation methods once in planta. Some are phloem mobile and some are xylem mobile. The phloem transports photo-assimilates from the leaves to the rest of the plant, however, this translocation can be altered by sink-source relationships. The xylem transports water and solutes, and transpiration drives the flow upwards from the roots to the leaves.



Herbicides all have different mechanisms of action, however, the safest ones for use have plant-specific target domains. Plant-specificity is an important quality for a Japanese knotweed herbicide owing to its usual proximity to water courses, with the preservation of native aquatic life in mind. Japanese knotweed is also found in parks, railway lines, and other public areas where treated-plants could be easily encountered by dogs or children. Plant-specific herbicidal target domains may have mechanisms of action related to function and biosynthesis of chlorophylls, leaf pigments, amino acids, fatty acids and photosynthetic electron transport and other metabolic processes. (Wakabayashi 2002)

Section D
Glyphosate

The herbicide known as 'glyphosate' is an example of an herbicide which works by inhibiting protein synthesis.

Glyphosate, or (N-(phosphonomethyl)glycine), is the most often recommended treatment for Japanese knotweed. This

Resource Four Data Source



is a non-persistent herbicide, with both phloem and xylem mobility. It is broad range, and not selective to a specific plant species. Glyphosate has a number of favourable environmental features, such as rapid soil inactivation, degradation to relatively harmless products, and low toxicity to non-plant life. Therefore, it has approval for use near water.



Limited effectiveness - Japanese knotweed can be controlled by glyphosate, however, it often fails to kill it. Environmental factors can render glyphosate less effective. These factors include accumulation of roadway dust (Figueroa 1989), a high concentration of nitrogen in soil (Hunter et al. 1993), and rainfall (Bariuan et al. 1999; Miller et al. 1998). Dusty conditions prevent the glyphosate from entering the plant via its leaf stomata. If it rains soon after application then the rain can simply wash the glyphosate into the soil, where it is broken down by soil microbes.

In high doses glyphosate has been shown effective at killing plants (Dewar et al. 2000). However, in sub-lethal doses, glyphosate affects growth and the partitioning of photoassimilates in many species, but does not kill them (Baur et al. 1977; Coupland and Casely 1975; Geiger et al. 1986; Scorza et al. 1984). Changes in aboveground parts of the plant has little effect on Japanese knotweed's secret weapon, its rhizomethe massive underground energy storage organ (Geiger et al. 1986; Haidar et al. 2005).

Sink-source relations are likely to play a large part in the ineffectiveness of glyphosate on Japanese Knotweed. Diversion of herbicide to different sinks, such as growing leaves or flowers, can divert herbicide away from the rhizomes and cause dilution in planta. The time of year when glyphosate is effectively allocated to the rhizome, autumn,

Resource Four Data Source



and when the glyphosate is most effective due to an increased rate of protein synthesis, summer, do not coincide. In autumn, the plant is preparing for dormancy and so the levels of protein synthesis are very low and glyphosate is unlikely to kill. The rate of glyphosate allocation to the rhizomes in summer is unlikely to provide a lethal dose. In combination with other herbicides, or regulators of sinksource relationships, glyphosate may be more effective.

In addition to the limited effectiveness of glyphosate on Japanese knotweed there is also a growing demand to find an alternative control solution due to concerns about its safety. Glyphosate has received increased press coverage in Europe recently due to the accumulation of both glyphosate and its degradation product, AMPA, in the environment. Although it has been used widely in agriculture for the past forty years, only recently have chronic low dose effects on animals and humans have been documented. Glyphosate was determined a 'probable carcinogen' by the International Agency for Research on Cancer REF, and has since sparked outrage with many campaigners calling for a ban.

Section E
Isotopic labelling

As part of my project, I will be using a labelled version of glyphosate to trace its movement around the plant. I will be using a method called isotopic labelling. To understand isotopic labelling, you must first understand what isotopes are.

Atoms consist of a nucleus containing sub-atomic particles called protons and neutrons, surrounded by electrons in shells. The specific number of protons determines the type of element an atom is. All atoms of the same element must have the same number of protons, but the numbers of neutrons can vary. Atoms with the same number of protons but a different number of neutrons are called isotopes. Some of these isotopes are radioactive, and some are stable.

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Radioactive isotopes can break down releasing energy, and be dangerous to our bodies, so for this reason I will be using stable isotopes of carbon and nitrogen, Glyphosate-2-13C,15N.

There are three naturally occurring isotopes of carbon: 12, 13, and 14. 14C is radioactive. 12C and 13C are stable, occurring in a natural proportion of approximately 93:1, so almost all naturally occurring carbon is carbon-12. Nitrogen has two naturally occurring stable isotopes, nitrogen-14, which makes up the vast majority of naturally occurring nitrogen, and nitrogen-15. Therefore, the glyphosate I will be using will be doubly labelled with carbon-13 and nitrogen-15.



Carbon-12
6 protons
6 neutrons



Carbon-13 6 protons 7 neutrons



Carbon-14 6 protons 8 neutrons

The quantities of the different isotopes can be measured by a method called mass spectrometry and compared to a standard. In analytical chemistry, a standard is a precisely known concentration of an element or a substance, which can be used for calibration. Chemists can measure the mass of atoms and molecules to a high degree of accuracy in a mass spectrometer, so they can work out if the number of neutrons is different. By sampling at different parts of the plant over time, I will be able to use mass spectrometry to see how glyphosate moves around the plant. The understanding of the limitations of glyphosate would redefine the required criteria for a new herbicide, and may elucidate a solution to improve the effectiveness of glyphosate.

Resource Four Activities



Activities

1. Match up the following words with their definitions.

| Key Words | Definitions |
|---------------------------------|--|
| | A) The sites in a plant where photo-assimilates are |
| 1) Phloem | stored or used. |
| | B) The movement of materials from leaves to other |
| 2) Xylem | tissues throughout the plant |
| | C) Variants of a particular chemical element that |
| 3) Isotope 4) Herbicide | differ in neutron number, but not atomic number. |
| | D) The living tissue that transports the soluble organic |
| | compounds made during photosynthesis. |
| 5) Translocation | E) The transport tissue that transports water and |
| | mineral ions in plants from the roots to the leaves. |
| 6) Sinks | F) The parts of a plant where net fixation of carbon |
| | dioxide occurs. |
| 7) Sources 8) Mass spectrometry | G) An analytical technique that ionizes chemical |
| | species and sorts the ions based on their mass-to- |
| | charge ratio i.e. measures masses. |
| | H) A substance that is toxic to plants, used to destroy |
| | unwanted vegetation. |
| | |

- 2. What is the function of a ribosome?
 - a) To release energy from glucose
 - b) Involved in protein synthesis
 - c) To build sugars by photosynthesis
- 3. What substance(s) is transported in the xylem?
 - a) Water
 - b) Water and mineral ions
 - c) Sugars and amino acids
- 4. What is the process that transports substances in the phloem?
 - a) Transpiration
 - b) Translocation
 - c) Transcription

Resource Four Activities



Activities

- 5. In which direction are substances transported in the phloem?
 - a) Upwards only
 - b) Downwards only
 - c) Upwards and downwards
- 6. Which tissue transports sugars from the leaves to other parts of the plant including roots for storage?
 - a) Xylem
 - b) Phloem
 - c) Veins
- 7. By which process does water move into a root hair cell?
 - a) Osmosis
 - b) Diffusion
 - c) Active transport
- 8. Suggest an explanation for why translocation in the phloem may be stopped by metabolic inhibitors.



9. The table shows the destination (sink) of translocated carbohydrates in a mature strawberry plant. Suggest an explanation for the percentage of carbohydrate translocated to the strawberries

| DESTINATION | % OF TOTAL TRANSLOCATED CARBOHYDRATE |
|-------------|--------------------------------------|
| Roots | 25 |
| Stem | 23 |
| Leaves | 14 |
| Strawberry | 38 |

Resource Four Activities



Activities

- 10. Suggest how isotopic carbon could be used to investigate photosynthesis in a plant.
- 11. A complete ring of bark, which includes the phloem but not the xylem, is removed from a woody stem.
- 12. Suggest an explanation for why:
 - a) The tissue above the ring swells but the tissue beneath the ring withers.
 - b) Chemical analysis of the fluid in the swollen tissue shows high concentrations of sugars and other organic solutes, but the tissue beneath the ring is deprived of nutrients.

Resource Four Further Reading



Explore

For a recent article about the growing concern surrounding glyphosate:



https://www.independent.co.uk/life-style/roundup-weed-killer-cancer-lawsuit-wine-beer-alcohol-a8797911.html



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