Research for Practice LAIMBURG JOURNAL

Short Paper

‘Pink Floyd’, cauliflowers and everything else you need to know about hyperspectral technology and plant stress

"Pink Floyd", Blumenkohl und alles, was Sie sonst noch über Hyperspektraltechnologie und Pflanzenstress wissen müssen

"Pink Floyd", cavolfiori e tutto ciò che c’è da sapere sulla tecnologia iperspettrale e lo stress delle piante

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ABSTRACT

Light and electromagnetic radiation possess properties that allow it to interact with matter. By using hyperspectral technology that measures electromagnetic radiation and therefore its interaction with matter, we can infer lots about the quality of things made up of matter (i.e., everything). This technique can be and is being applied in the field of agriculture particularly for the detection of plant stress. Effective, efficient and sustainable management of stress in crops is critically important, not only because stress in crops limits crop production, but also because the methods in which stress is managed are often associated with negative economic, environmental and human health consequences. This article provides an introduction to electromagnetic radiation, measuring it with hyperspectral technology and how the technique is used in agriculture for the detection of plant stress and the consequent sustainable management thereof. Importantly, it is intended to be entertaining and easy-to-read, particularly for those who do not have extensive backgrounds in the fields of physics and remote sensing.

KEYWORDS

Light, Electromagnetic Radiation, Plant Stress, Spectroradiometer, Hyperspectral camera, Precision Agriculture, Beginner’s Introduction

CITE ARTICLE AS

Cullinan Cameron Brodrick (2023). ‘Pink Floyd’, cauliflowers and everything else you need to know about hyperspectral technology and plant stress. Laimburg Journal 05, 2023.002, DOI:10.23796/LJ/2023.002.

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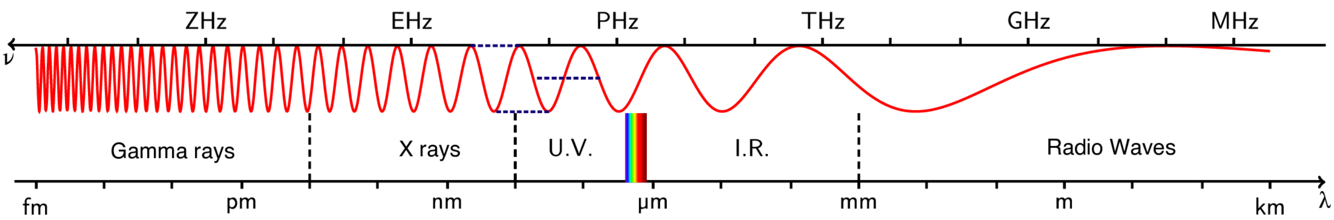

Abb. 1: Electromagnetic radiation. Gamma rays, x rays, ultraviolet light, visible light, infrared light, and radio waves all exert forces on electric and magnetic particles and so form part of the electromagnetic spectrum. The wavelength, shown here with dashed blue lines, determines how much energy the light carries and whether or not it will be absorbed by a particular material. Source: Wikimedia Commons [1]. Words originally presented in French were translated to their English equivalents. Dashed blue lines indicating wavelength have been added to the original work.

LIGHT, ELECTROMAGNETIC RADIATION AND THEIR RESEMBLANCE TO SUB-ATOMIC CAULIFLOWERS

Light is energy. Light is made of waves. Light is also made of particles. Some light is visible to the human eye and some light isn't. It has no mass and yet somehow it has momentum. Light is clearly complicated, so complicated that even the brightest modern quantum physicists in the world aren't sure of what it is at its most basic level [2] and yet it forms such a central part of almost everyone's everyday lives. Using the light that travels to our eyes, our brains are able to gain an unimaginably large amount of information about our surrounding environment: not only how big or how far away something is but also a rough guess as to what it is made of. For instance, we can usually tell if something is made of metal or wood or whether it is wet or dry just by looking at it. This is made possible because of light's unique properties and particularly those that allow it to interact with matter.

Light can exert a force on electric and magnetic particles and because the direction of this force alternately pushes these particles between opposite directions (at very rapid rates), we can think of light as a wave - an electromagnetic wave. The distance between the closest two points being pushed in the same direction with the same force by a light wave, such as for instance, the distance between two consecutive peaks or troughs of a wave, is called the wavelength (Fig. 1). Depending on this wavelength, light has particular qualities. The light we see, called visible light, has wavelengths of between 380 nm and 700 nm, which correspond to blue and red light, respectively, with all the other colours falling in between. Light with wavelengths in this visible range actually forms only a tiny amount of all the possible wavelengths. All the energy that exerts an electromagnetic force, like light, from the radioactive gamma rays with wavelengths at ten billionths of a metre to radio waves with wavelengths of several kilometres is called electromagnetic radiation. This electromagnetic radiation can have wavelengths at any value between the two extremes of gamma rays and radio waves and we say that all this electromagnetic radiation belongs to a spectrum - the electromagnetic spectrum (the subsequent use of the word spectrum to describe entities being continuous in nature is derived from the continuous nature of electromagnetic radiation and not viceversa [3]). The word 'spectral' is used to describe things, processes, methods, and other entities that are connected to this 'spectrum'.

We say electromagnetic radiation is made up of waves because of the way it behaves. Electromagnetic radiation, however, also sometimes behaves like particles. That is, as one guy in his bath [4], put it: electromagnetic radiation sometimes behaves in the way we would expect tiny cauliflowers to behave. This is chiefly because of the way they stack into two piles when we shoot them one by one through two narrow gaps onto a detecting screen - but only when we are watching. When we do the same thing but don't watch, these tiny particles mysteriously behave much more like waves again and what we see on the detection screen is a pattern characteristic of waves, known as a diffraction pattern. Why the stage fright? No one really knows. In any case, these tiny electromagnetic, occasionally cauliflower-like, particles are called photons. A peculiar property of these photons is that they are sometimes absorbed by matter when the two collide and other times they simply bounce straight back or pass straight through it. Whether or not they are absorbed depends on what the matter is made of and the wavelength of the photon.

The matter-specific and wavelength-dependent absorption of electromagnetic radiation is what allows us to use light to tell us about our environment because, as more of a particular material is present more of the light at the right wavelength is absorbed and less of that light is reflected. Our eyes receive light that is reflected by an object - the light with a wavelength that isn't the right wavelength for it to be absorbed. Our brains interpret the wavelength of light as colour. Over time we learn to associate different combinations of light colour (wavelength) and brightness (amount) with different types of matter, and we subconsciously use this learnt knowledge to tell us something about the make-up of an object.

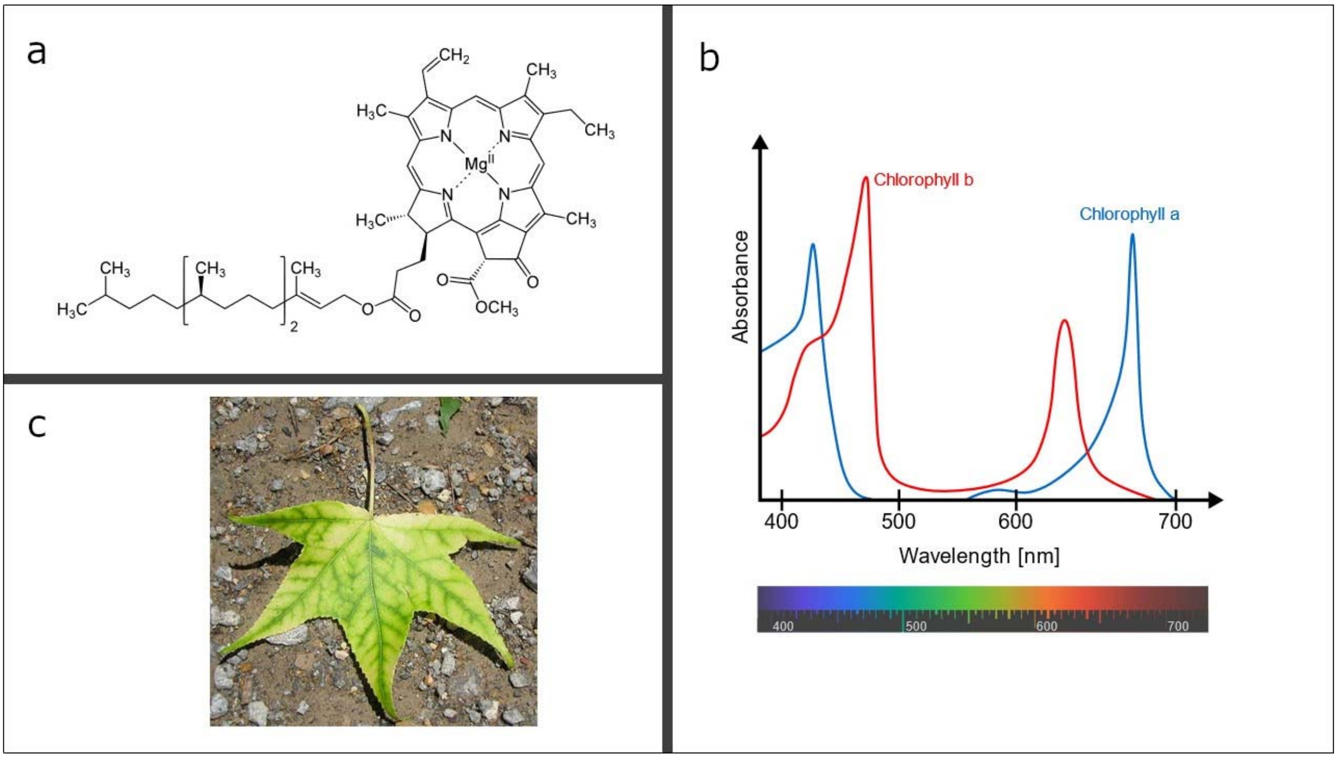

Abb. 2: a) The chemical structure of chlorophyll a. To synthesize chlorophyll, plants need nitrogen (N) and magnesium (Mg) (amongst other things), which they must obtain from the environment Source: Wikimedia Commons. b) Absorption spectra of two slightly different types of chlorophyll, chlorophyll a and chlorophyll b. Both molecules absorb red and blue light to drive photosynthesis but reflect green light. Source: Wikimedia Commons.c) Liquid Amber (*Liquidambar styraciflua* L.) showing the yellowing that results from declines in chlorophyll concentration. Chlorophyll is what makes plants look green to us. More chlorophyll makes plants look greener, while when there is less, they often look yellow. Source: Wikimedia Commons.

PLANT STRESS, MAN'S INADEQUACY, AND THE DARK SIDE OF THE MOON

This principle works for everything we can see, including plants. Any seasoned gardener, farmer or plant enthusiast will tell you that you can tell a lot about the health of plant based on what it looks like. At face value, this seems like a banal statement, but on a molecular level this is because healthy plants absorb light at certain wavelengths and reflect light in others. For instance, healthy plants are green because, when white light from the sun (consisting of all the different colours), shines on its leaves, most of the blue and red light is absorbed and used to drive photosynthesis (Fig. 2b). On the other hand, a lot more of the green light is reflected and so when this light reaches the human eye, our brains conclude that the plant is green. At a closer level, this is because of a group of molecules, collectively referred to as chlorophyll. Chlorophylls are responsible for capturing a portion of the energy in light, especially that of red and blue light, and delivering it on to the next photosynthetic processes that ultimately store it, in the form of energy rich molecules that provide or have provided almost all the energy ever used to sustain life on earth. Chlorophylls, to put it lightly, are arguably the most important molecules, not just for plants but for life on earth as we know it.

Rather frustratingly, given its importance, plants cannot simply build chlorophyll out of nothing. To produce chlorophyll, they need to use resources: sugars and energy produced through photosynthesis itself, as well as plant mineral nutrients, especially nitrogen and magnesium (Fig. 2a). If the plant is unable acquire these resources, it cannot produce chlorophyll. Without these nutrients and, therefore, without chlorophyll, the plant cannot photosynthesise properly, that is to say, the plant is stressed. Furthermore, as a consequence of this stress-induced decline in chlorophyll, the plant absorbs less blue and red light and reflects less green light. What we usually observe is a plant with a more yellowish colour (in the absence of chlorophyll we can see the yellow light reflected by other plant molecules or pigments) (Fig. 2c). Because stress has changed the amount of light that the plant reflects or absorbs, we say that the stress has changed the optical properties of the plant. We, as humans, often observe a change in optical properties as a change in colour.

All plant stresses, whether nutrient deficiencies, water deficits, insect infestation, pathogen infection etc., will likely similarly change the chemical make-up of the plant which will similarly alter the optical properties of the plant [5] [6] [7] [8]. A lot of these properties are visible to

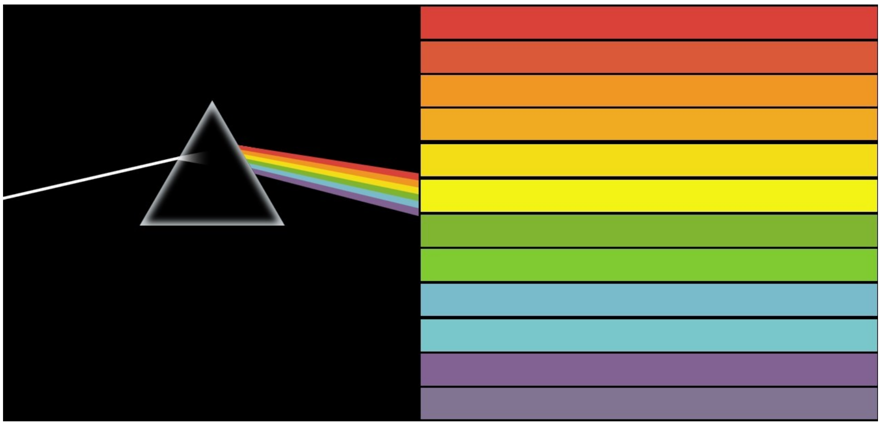

Abb. 3: Left: the dispersion of light into its component colours as it passes through a prism. Spectrometers, spectroradiometers and hyperspectral cameras all make use of this principle or similar to split light into bands with very narrow wavelength ranges which the devices can then measure independently of each other. Source: Wikimedia Commons. Right: 50 shades of everything except grey. Humans are limited in our visual abilities to see subtle or, indeed, invisible changes in wavelength. All electromagnetic radiation outside of the visible region is completely invisible to us, but even in the visible region we are not necessarily able to tell the difference between two very slightly different colours as is the case here. Visually gifted readers might be able to conclude that every second bar is a slightly lighter shade of the same colour above it. Fortunately for the others, we have a wide range of technologies that can tell the difference between these shades of colour and, indeed, between shades that are far more similar than presented here, including at wavelengths completely invisible to the human eye.

humans, especially when the stress is severe,i.e., the plants present visible symptoms. However, there are limitations to what we can physically see ourselves. For one thing, we can only see electromagnetic radiation in the visible region (with wavelengths from 350-700 nm) which is a tiny proportion of all the possible electromagnetic radiation. Some stresses cause changes in the light absorbed or reflected at wavelengths outside this region, which would be completely invisible to the human eye. A second limitation to the human observation of plant stress is that we have a limited ability to tell the difference between two very similar colours of light with wavelengths very similar to each other (Fig. 3) and to notice differences in the one colour when the overall colour remains largely the same. Therefore,even if a stress is present, especially if it is in its early stages, we may not be able to detect it visually ourselves.

To address our human limitations, we make use of spectral technology: technology that has been designed to detect electromagnetic radiation in much the same way that the human eye does but at the same time, to overcome its limitations. Spectrometers, spectroradiometers, hyperspectral cameras and thermal cameras all detect electromagnetic radiation, and can all be used to help us detect plant stress based on the fact that this stress will likely change the optical properties of the plant.

Spectroradiometers get their names from the combination of the words ‘spectral’, ‘radiation,’ and ‘meter’ (i.e., to measure). Therefore, these devices are used, unsurprisingly, to measure radiation belonging to the electromagnetic spectrum, usually in the visible and near infrared regions. They do this by funnelling incoming light through an object that splits the light into its components with different wavelengths (such as is done by a prism - famously depicted in the album cover for Pink Floyd’s, ‘The Dark Side of the Moon’) (Fig. 3) and then measures the light in each of the bands of these wavelengths. In this way, we can use spectroradiometers to tell the difference between light with very similar wavelengths (i.e., with very similar colours) in the visible regions as well as in the near infrared region (which humans can’t see).

HYPERSPECTRAL DATA AND THINKING MACHINES FOR A UTOPIAN AGRICULTURAL FUTURE (WITH APOLOGIES TO FRANK HERBERT)

If we draw a graph of the light reflected in each of the bands that are produced when light is split into its components, we get what is called a hyperspectral profile of whatever the device is looking at. Generally, the hyperspectral profiles of plants have a very particular shape with strong absorbance in the visible region and a strong reflection in the near infrared region (Fig. 4a). If a plant is experiencing stress and its optical properties are changed, we expect the shape of its hyperspectral profile to deviate from the normal shape. However, identifying these deviations is not as easy as it seems: there are several other factors that influence the reflectance recorded by the instrument and sophisticated data analysis is required to distinguish true signals from those pesky external influences. The use of machine learning algorithms is at the forefront of this data analysis because they can look in a theoretically unlimited number of dimensions (humans are frustratingly limited to a measly three) to find the important differences in the hyperspectral profiles of healthy and stressed plants.

In much the same way that children learn to associate a particular look with a material such as the shininess we associate with metal, or the brown textured look of wood, these machine learning algorithms need to be trained. We do this by giving the algorithm some hyperspectral profiles and telling it what kind of stress, if any, the tree was experiencing when we took them. The algorithm then learns what patterns to associate with different stresses so that when we don't know what kind of stress a plant is experiencing, we can give the hyperspectral profile of that plant to the algorithm, and it can give us its best guess on what kind of stress it is experiencing and possibly even to what degree.

This technique has been used to detect different types of stress in all sorts of different crops. It has proven to be particularly useful at determining the nitrogen concentration and the concentration of other nutrients of crops, including lettuce [9], maize [10] [11], wheat [12], cucumber [13] and apple [14]. Infection by bacteria, fungi and viruses can also change the optical properties of a crop and so the technique has been used to detect infection with several of these pathogens. *Xylella fastidiosa* and *Verticillium dahlia* infection in Olive and Almond [15], *Venturia inaequalis*, the pathogen that causes apple scab [16] and *Diplocarpon mali*, the pathogen that causes Marssonina disease, in Apple [17]. The Functional Genomics group at the Laimburg Research Centre is currently working on developing the technique for the detection of apple proliferation caused by '*Candidatus* Phytoplasma mali', a pathogen related to the 'Flavescence dorée'-phytoplasma that infect grape vines. 'Flavescence dorée' can also be detected using spectral techniques [18]. Spectral techniques, especially when combined with the measurement of electromagnetic radiation in the thermal infrared region (with wavelengths between 4000 and 15 000 nm), can also be used to estimate water use and detect water deficit stress in crops [19] [20].

So, we can do it. But so, what? Beyond the - perhaps obvious - answer of if we can detect a stress early, especially when symptoms are not visually detectable by humans, we can fix it before it does too much damage, there is another, far more important, consequence of this ability. As most people are probably aware, these days agriculture has a bad reputation in terms of environmental impact. In one sense, after the energy sector, the agricultural sector is the biggest contributor to greenhouse gases. A significant proportion of this comes from our agricultural soils (4.1% of total greenhouse gas emissions [21]), due to the application of synthetic nitrogen fertilisers that turn into potent greenhouse gases: nitrous oxide and other similar gases. Furthermore, when synthetic fertilisers are leached from agricultural soils, they damage downstream water systems by promoting the growth of harmful and often toxic algae and bacteria [22] [23]. Other agrochemicals, such as those we use to control pests and diseases are also of concern because their use has been associated with reduced biodiversity [24] [25] and some are even directly toxic to humans [26] [27].

Therefore, it is in everyone's interest that the resources are used as sparingly as possible. To exclude them entirely, however, is not feasible. For instance, it is estimated that almost half of the world's population owes its existence to synthetic nitrogen fertilisers and the increased production of food they make possible [28]. To meet our demands for food without depleting or damaging our natural environments (and indeed ourselves) we need to make sure that we supply our crops only with the resources they need (fertilisers, water etc.) in exactly the right amounts, in exactly the right places at exactly the right times and not more. The process of doing this is known as site-specific management [29] or precision agriculture.

To apply resources in exactly the right places and at exactly the right times requires knowledge on exactly where and when they should be applied. This is where spectral sensing technology comes into play. It allows us to obtain information on the health status of a crop quickly and effectively. What's more, the technology can be readily incorporated into remote sensing technologies, i.e., those that take the spectral measurements at a distance. Hyperspectral cameras or imaging spectrometers essentially measure a separate hyperspectral profile for every pixel in an image (Fig. 4b). By doing so, not only do we get the hyperspectral profiles that we can use to detect stress, but we also know exactly where each profile came from and therefore, where the plants experiencing stress are (Fig. 4c) [30]. This provides us with unprecedented opportunities for the sustainable management of plant stress because this technology can rapidly supply us with incredible information on whether a particular plant is stressed or not and furthermore, possibly even severity of the stress - all of which form a critical part of what we need for an effective precision agriculture system.

The use of spectral techniques to detect when a plant is stressed and the kind of stress it is experiencing is undoubtedly loaded with potential. Furthermore, its use will be critical if we are going to make agriculture more sustainable. However, with a single spectroradiometer easily costing around € 100 000 or worse, and a hyperspectral camera around € 170 000, price is still a limiting factor. What's more, while there has been much work on the use of the technology under relatively small-scale research settings, the way in which we apply the technology practically within large scale commercial settings still needs to be refined. Nevertheless, the price of this technology is falling and new innovations for its application, such as drones [30] [31] and robots [32], are being developed that together make the technology more and more feasible with every passing day. We can, therefore, only look forward to how it will be incorporated into the cropping systems of the future - systems that will undoubtedly be the hallmark of the beginning of a newer, more sustainable age of agriculture.

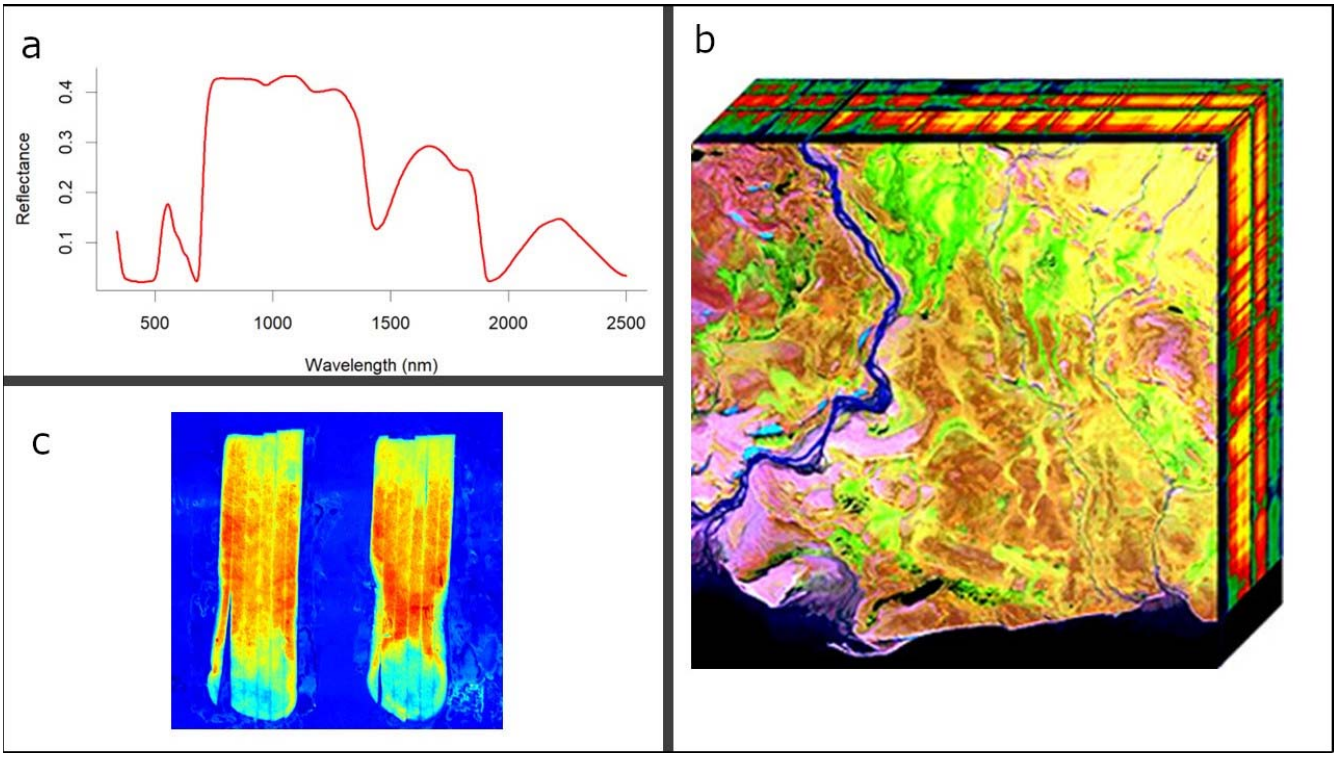

Abb. 4: a) Hyperspectral profile showing the characteristic shape for vegetation. Hyperspectral profiles are generated by plotting the proportion of incoming light (between 0 and 1) reflected at each of the hundreds of bands the incoming light is split into by a hyperspectral device. The hyperspectral profiles of vegetation have a very characteristic shape with high absorption in the visible region (380-700 nm) and strong reflectance in the near infrared region (700 nm onwards). Water in plant tissue absorbs infrared light at around 1400 nm and 1800 nm [12]. When trying to detect plant stress, the aim is to find the differences between the spectral profiles of healthy and stressed plants, something which can usually only be done through sophisticated data analysis. b) Hyperspectral image. By combining the hyperspectral profiles with spatial information, we can obtain a hyperspectral image. Each pixel in the image contains a hyperspectral profile, represented by the depth of the cube, called a hyperspectral image cube. Source: Wikimedia Commons. c) Image derived from a hyperspectral image of a potato field. Using hyperspectral imaging for site-specific management of plant stress, "sugar end," a physiological disorder of potato can be detected through hyperspectral imaging. Highly affected plants are shown in red, while unaffected plants are shown in blue. Spatial information combined with hyperspectral information is incredibly useful because it allows us to tell exactly where, when, and to possibly even to what extent a plant is stressed. Once we have this information, we can develop stress management strategies that deal with the stress effectively, while limiting the damaging effects to the environment that often result from stress management practices. Source: Wikimedia Commons.

ZUSAMMENFASSUNG

Licht und elektromagnetische Strahlung besitzen Eigenschaften, die es ihnen ermöglichen, mit Materie zu interagieren. Durch den Einsatz der Hyperspektraltechnologie, die elektromagnetische Strahlung und damit ihre Wechselwirkung mit Materie misst, können wir Rückschlüsse auf die Qualität der Materie ziehen. Diese Technik findet auch im Bereich der Landwirtschaft Anwendung. Dieser Artikel bietet eine Einführung in die elektromagnetische Strahlung, die Messung mit Hyperspektraltechnologie und die Anwendung dieser Technik in der Landwirtschaft zur Erkennung von Pflanzenstress und dessen nachhaltigen Managements. Dabei wurde auf ein leichtes Leseverständnis und einen hohen Unterhaltungswert geachtet, insbesondere für diejenigen, die über keine umfangreichen Kenntnisse in den Bereichen Physik und Fernerkundung verfügen.

RIASSUNTO

La luce e la radiazione elettromagnetica possiedono proprietà che permettono loro di interagire con la materia. Utilizzando la tecnologia iperspettrale che misura la radiazione elettromagnetica e quindi la sua interazione con la materia, possiamo dedurre informazioni sulla qualità della materia. Questa tecnica può essere applicata nel campo dell'agricoltura. Questo articolo fornisce un'introduzione alla radiazione elettromagnetica, alla sua misurazione tramite tecnologia iperspettrale e al suo utilizzo in agricoltura per il rilevamento dello stress delle piante ed alla sua gestione sostenibile. Importante è che il brano ristulti leggero, divertente e di facile lettura, in particolare per coloro che non hanno una formazione approfondita nei campi della fisica e del telerilevamento.

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