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The role of hyperspectral imaging in forest seedling phenotyping

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Abstract: In recent years, hyperspectral imaging has been widely adopted in agriculture and plant phenotyping, while its application in forestry has been increasing. From that point onward, hyperspectral imaging has become a valuable tool for plant phenotyping, enabling the assessment of a broad range of plant traits. Given that seedlings of forest trees are one of the most widely used types of forest planting stock, advancements in hyperspectral technology have created new possibilities for improving seedling quality assessment. High-quality forest seedlings are important for the successful establishment of forest stands, especially after outplanting within restoration initiatives. Even though hyperspectral imaging brings numerous advantages, continued technological improvements are necessary to address its several limitations and challenges. Despite its widespread use in agricultural phenotyping, applications in forest nursery production remain limited. Therefore, this review focuses on research involving hyperspectral imaging in forest seedling production and its potential for assessing seedling quality parameters.

Keywords: plant evaluation; remote sensing; research gaps; technological approaches; spectral imaging techniques

Imaging spectrometry, or hyperspectral imaging (HSI), emerged as the integration of traditional spectroscopy technology with modern imaging systems (Goetz 2009; Qian 2021; Huber et al. 2008). The term 'hyperspectral imaging' was introduced by Goetz et al. (1985), emphasising a deterministic physics-based approach to multispectral data analysis, as opposed to the statistical methods used previously, aiming to drive the development of new digital image-processing techniques. Developed in the 1980s, it was initially applied in the military (Goetz 2009; Ančić et al. 2019) and later expanded into civilian scientific fields (Qian 2021, 2022; Miljković, Gajski 2016; Lucas et al. 2004; Banerjee et al. 2020).

Due to its high efficiency and ability to perform non-destructive, real-time, and rapid measurements, advanced hyperspectral technology has been widely used in recent years (Banerjee et al. 2020; Ruett et al. 2022; Liu et al. 2023; Luo

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et al. 2024; Miljković et al. 2025). It enables the detection of a large quantity of detailed information that is invisible to the naked eye or to other methods (Qian 2021; Wahabzada et al. 2016; Miljković et al. 2017; Mahlein et al. 2018; Ruett et al. 2022; Miljković et al. 2025). It became a promising and valuable tool for plant phenotyping (Ruett et al. 2022; Awada et al. 2024; Detring et al. 2024) that measures and analyses plant morphological, physiological, and pathological traits (Fahlgren et al. 2015; Behmann et al. 2016; Banerjee et al. 2020; Stejskal et al. 2023; Awada et al. 2024). Thus, it may represent a valuable tool for improving the efficiency of forest seedling assessment and production (Lu et al. 2021; Stejskal et al. 2023), which could support more effective silvicultural operations. The application of HSI in forest seedling production is limited despite its widespread use in agriculture.

This research aims to: (*i*) summarise principles of HSI applicable in forest seedling phenotyping; (*ii*) determine its use in seedling quality parameter assessment; (*iii*) suggest implementation potential in forest nursery production.

HISTORICAL OVERVIEW OF HYPERSPECTRAL IMAGING

In the 1970s, Landsat-1 Multispectral Scanner (MSS) images were used for mineral identification, when geologists realised that, in the four spectral band data, many morphological clues of the minerals were absent (Goetz 2009; Qian 2021; Xing et al. 2019). This led to the conclusion that the spectral reflectance was the major key to mapping geological units (Qian 2021). The integration of spectroscopy technology with modern imaging systems resulted in imaging spectrometry, also known as hyperspectral imaging (Goetz 2009; Qian 2021), to address this limitation. The definition of imaging spectrometry was proposed by Goetz et al. (1985), as 'the acquisition of images in hundreds of contiguous, registered, spectral bands such that for each pixel a radiance spectrum can be derived' (Goetz 2009).

Even though the year 1979 may be viewed as the beginning of the development of spaceborne hyperspectral imagers, the first successful hyperspectral data were acquired in 1983, showing significant variations in spectral reflectance, revealing band-to-band differences (Qian 2021).

HYPERSPECTRAL IMAGING PRINCIPLES

Due to the differences in material chemical composition, physical structure, and surface properties, electromagnetic energy is reflected in specific wavelengths and patterns (Paulus, Mahlein 2020; Detring et al. 2024; Miljković et al. 2025). That reflected energy is considered as reflected light, which forms characteristic spectral bands (Detring et al. 2024). That is the principle on which HSI is based.

HSI is collecting information for several hundred narrow continuous spectral bands within a specific range of wavelengths (Bian et al. 2022). Each spectral band is mapped at different spatial positions (Babić et al. 2023), represented by individual pixels for quantitative analyses and qualitative assessments (Luo et al. 2024). The spectral bands are being measured between 350 nm and 2500 nm of the electromagnetic spectrum, at nm-level resolution for each image pixel (Fahlgren et al. 2015; Detring et al. 2024). It combines both spectral and spatial information, similar to a digital Red, Green, and Blue (RGB) camera but with higher spectral resolution (Paulus, Mahlein 2020; Mahlein et al. 2018). This technology has the ability to capture spectral bands from the visible (VIS) range (400–700 nm), to the near-infrared (NIR) range (700–1000 nm), and the shortwave infrared (SWIR) range (1000–2500 nm) (Paulus, Mahlein 2020; Mahlein et al. 2018) and to some extent the ultraviolet (UV) range (100–400 nm) (Detring et al. 2024). Given that HSI uses a large number of spectral bands (> 20), the use of only a few spectral bands (typically 3 to 15) is usually defined as multispectral imaging (Paulus, Mahlein 2020; Cotrozzi 2022).

A hyperspectral image consists of vector pixels where each pixel holds both spatial and spectral information (Čirjak et al. 2022). The x- and y-axes represent spatial dimensions, and the z-axes (λ-axes) represent the spectral dimension (Mahlein et al. 2018; Ančić et al. 2019), often referred to as an image cube (Miljković, Gajski 2016; Long et al. 2023). A hyperspectral cube, or a 3D data cube, can contain absorption, reflectance, or fluorescence spectrum data for each image pixel (Goetz 2009; Fahlgren et al. 2015; Čirjak et al. 2022). Hyperspectral data can also be portrayed as a one-dimensional hyperspectral signal as a signature vector (Chang 2013).

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HSI data contains a vast amount of information (Wahabzada et al. 2016), and it is represented as a 3D data cube (Goetz 2009; Fahlgren et al. 2015). As the amount of data increases, so does the complexity (Moghimi et al. 2018; Luo et al. 2024). To find useful information in that vast amount of data, it is essential to remove the unnecessary information by selecting the important ones (Chang 2013; Luo et al. 2024; Xing et al. 2024). This can be achieved through reduction operations such as feature selection or feature extraction (Luo et al. 2024).

Data processing and classification. Before data processing in a hyperspectral workflow, both data preprocessing and image processing must be conducted. Preprocessing begins after hardware calibration and measurement validation have been completed (Paulus, Mahlein 2020). Calibration is important, among other things, for standardisation of the spectral axis (z-axis) and data accuracy (Paulus, Mahlein 2020). For improving the accuracy of information extracted from hyperspectral data, effective data processing and analysis algorithms are necessary (Luo et al. 2024). Furthermore, addressing issues such as noise and atmospheric interference is also important. The overall HSI workflow in plant phenotyping typically consists of data acquisition, data preprocessing, analysis, and information extraction. A general hyperspectral imaging workflow in forest seedling phenotyping is presented in Figure 1.

Types of hyperspectral imaging systems. Hyperspectral imaging systems are constructed of several components, including the light source, objective lenses, hyperspectral sensor, imaging spectrograph, and a computer (Sarić et al. 2022). Systems can be generally classified into four types based on their imaging methods: line scanning (push broom), point scanning (whisk broom), area scanning (filter-based systems), and snapshot (Sarić et al. 2022; Detring et al. 2024; Luo et al. 2024), each of which has advantages and limitations. Additionally, there is a classification based on the platforms on which they are mounted: satellite, airborne, and ground-based/laboratory (Ančić et al. 2019). Common hyperspectral imaging methods are illustrated in Figure 2.

Furthermore, it can be classified based on different spectral dispersion methods: (a) the dispersive element-based approach, (b) the spectral filter-based approach, (c) the Fourier transform imaging interferometer, (d) snapshot hyperspectral imagers (Qian 2021).

(a) The dispersive element-based approach is the most popular one, and it uses dispersive elements (e.g. prisms, diffraction gratings) to separate light from an object into spectral components. The scene can be scanned using a ground scanner, by aircraft, or by satellite. Based on the imaging methods, the line scanning (push broom) and point scanning (whisk broom) are used (Qian 2021).

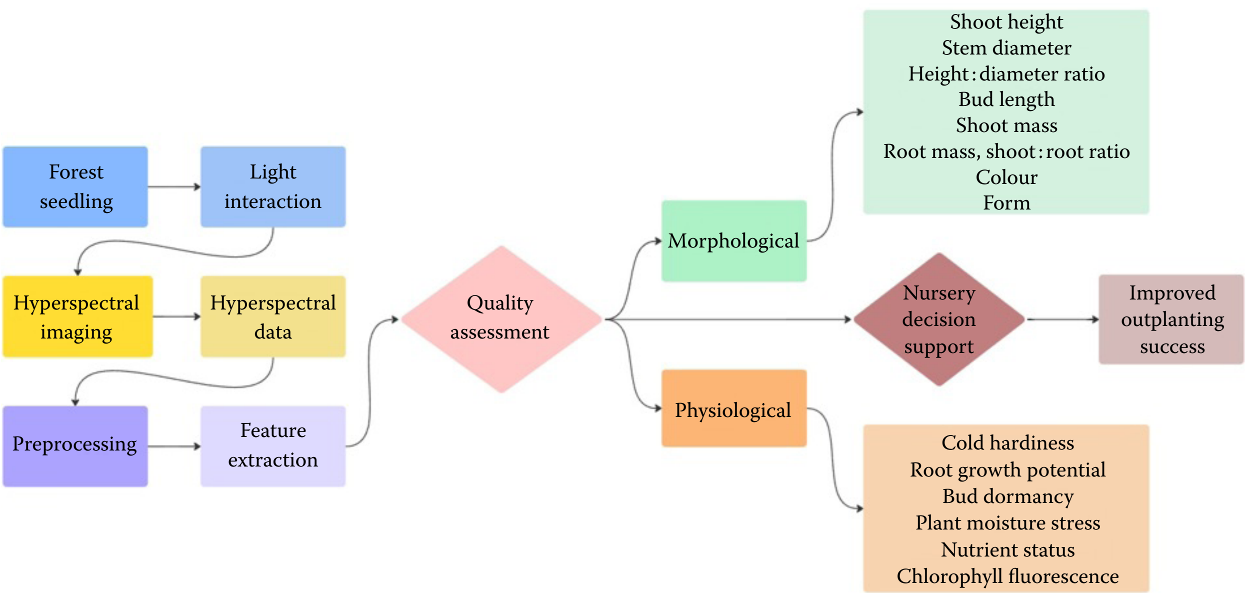

Figure 1. General hyperspectral imaging workflow in forest seedling phenotyping

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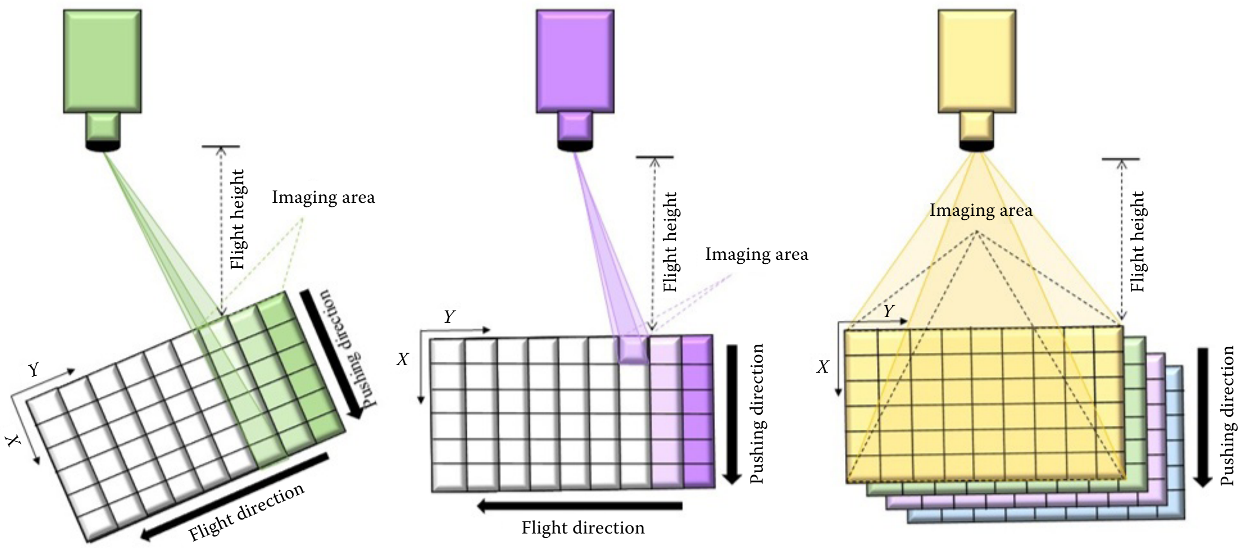

Line scanning (push broom) Point scanning (whisk broom) Area scanning (filter-based)

Figure 2. Common hyperspectral imaging (HSI) methods: line scanning (push broom), point scanning (whisk broom), and area scanning (filter-based)

(b) The spectral filter-based approach is newer and less popular, and it works by using one or more spectral filters to transmit the selected spectral bands of interest. The selected spectral bands are transmitted, and some are blocked through an absorption or interference process as the beam passes through the filter (Qian 2021).

(c) The Fourier transform imaging interferometer is based on interferometers that are scanned to obtain an interferogram, on which Fourier transform-based algorithm is applied to derive the spectrum of each pixel. It is used to obtain a spectrum using a Michelson interferometer (two-beam) or an FPI (multibeam) (Qian 2021).

(d) Snapshot hyperspectral imagers are used to generate a hyperspectral cube by using whisk broom or push broom imaging methods. In order to generate a hypercube, this method requires a wavelength scanning using spectral filters (Qian 2021).

APPLICATIONS IN FORESTRY AND PLANT PHENOTYPING

Since the early application for mineral identification, the implementation of HSI has increased significantly, especially in recent years. The first mention of HSI in environmental science was in the early 1980s, more specifically in the identification of vegetation stressed by metals in the substrate (Goetz et al. 1985). The development of AIS (Airborne Imaging Spectrometer) and AVIRIS (Airborne Visible/Infrared Imaging Spectrometer) has revealed the potential to provide information over large areas of the Earth for vegetation mapping, health assessment, stress detection, and characteristics (Goetz et al. 1985). From that point forward, this high-end technology has had many applications in various scientific fields, including remote sensing and environmental monitoring (Qian 2021; Moghimi et al. 2018; Detring et al. 2024), medicine (Miljković et al. 2017; Detring et al. 2024), astronomy (Ančić et al. 2019), archaeology (Miljković, Gajski 2016), agriculture (Qian 2021; Moghimi et al. 2018; Babić et al. 2023; Detring et al. 2024), forestry (Qian 2021, 2022; Babić et al. 2023), etc.

The application in forestry began in the 1990s with the mapping of forest physical and structural characteristics, as well as the measurement of forest ecosystem characteristics for forest ecosystem health and sustainability prediction (Treitz, Howarth 1999). Following its initial application, nowadays it has a large variety of uses in forestry, ranging from chlorophyll content estima-

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tion (Luo et al. 2024) to species determination (Ballanti et al. 2016; Fassnacht et al. 2016; Ančić et al. 2019), as summarised in Table 1.

While HSI has been applied to a wide range of forestry applications, some areas are well-studied, whereas others remain comparatively underexplored (Table 1). Thanks to its versatility, HSI has become a promising and valuable tool for both plant phenotyping (Ruett et al. 2022; Awada et al. 2024; Detring et al. 2024) and for forestry (Qian 2021, 2022; Babić et al. 2023).

Although HSI enables the measurement and analysis of a wide range of plant traits, it has been mainly used in forestry and phenotyping studies, focusing on short-term traits; therefore, traits related to long-term performance remain largely unexplored. The current focus of HSI applications in assessing forest seedling traits is summarised in Table 2.

Table 1. General forestry applications of hyperspectral imaging (HSI)

Applications	Reference(s)
Changes of chlorophyll, ascorbic acid, glutathione, and soluble protein in foliage and roots	Luo et al. 2024
Content of photosynthetic pigments and their spatial distribution in leaves	Luo et al. 2024
Biochemistry of forest canopies	Qian 2021
Leaf optical properties	Mahlein et al. 2018; Stejskal et al. 2023; Luo et al. 2024
Leaf reflectance	Jones et al. 2025
Plant reflectance characteristics	Behmann et al. 2016
Canopy reflectance response	Banerjee et al. 2020; Babić et al. 2023
Insect/pest outbreaks	Ančić et al. 2019; Bian et al. 2022
Plant species/biodiversity	Ballanti et al. 2016; Fassnacht et al. 2016; Ančić et al. 2019
Differences between broadleaved and coniferous forests	Ančić et al. 2019
Forest monitoring	Ančić et al. 2019

Table 2. Examples of hyperspectral imaging (HSI) applications of forest seedling traits

Traits	Reference(s)
Structural traits	Wahabzada et al. 2016; Moghimi et al. 2018
Green biomass	Behmann et al. 2018; Ančić et al. 2019
Leaf Area Index (LAI)	Behmann et al. 2018; Miljković et al. 2025
Nutrient status (N, P, K)	Banerjee et al. 2020; Yang et al. 2021; Long et al. 2023; Luo et al. 2024; Miljković et al. 2025
Water status/content	Paulus, Mahlein 2020; Ančić et al. 2019; Ruett et al. 2022; Luo et al. 2024; Xing et al. 2024; Miljković et al. 2025
Changes/status in plant physiology	Wahabzada et al. 2016; Ruett et al. 2022; Stejskal et al. 2023
Freeze tolerance	Lu et al. 2021
Light-use efficiency (LUE)	Behmann et al. 2018
Vitality/plant health	Behmann et al. 2016; Roscher et al. 2016; Ruett et al. 2022
Abiotic stress (heavy metal, cold, drought, water)	Paulus, Mahlein 2020; Stejskal et al. 2023; Luo et al. 2024; Felix et al. 2025
Biotic stress (insect, virus, pathogen)	Cotrozzi 2022; Paulus, Mahlein 2020; Roscher et al. 2016; Mahlein et al. 2018; Ančić et al. 2019; Hoeppner et al. 2020; Pandey et al. 2021; Ruett et al. 2022; Stejskal et al. 2023; Detring et al. 2024

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WHICH PARAMETERS ARE IMPORTANT FOR FOREST PLANTING STOCK AND WHY?

Forest planting stock plays an important role in afforestation, forest restoration, and all activities related to forest preservation and regeneration. Quality planting stock is often a key condition for the successful implementation of forest and habitat restoration programs, and other development programs (Grossnickle 2012; Đodan et al. 2023). The quality of forest planting stock is determined by genetics, physiological and morphological traits (Mataruga et al. 2023), and is assessed through corresponding morphological and physiological traits.

Forest seedlings, as one of the most widely used types of forest planting stock, are vital for the successful establishment of forest stands. Morphological traits for seedling quality assessment include e.g. height, stem diameter, bud length, shoot and root mass, shoot : root ratio, colour, and form; and physiological traits include cold hardiness, root growth potential (RGP), bud dormancy, plant moisture stress (PMS), nutrients, and chlorophyll fluorescence (Haase 2007, 2008; Đodan, Barišić 2026). The most commonly measured morphological traits are height and stem diameter (Haase 2010; Đodan, Barišić 2026), which can be ineffective for evaluating physiological traits, particularly under stress conditions (Haase 2008). Several parameters affecting seedling quality are linked to field survival and productivity:

– quick root development,
– quick anchoring of the seedling in the ground and initiation of assimilation and growth after planting,
– a developed root system,
– foliage adapted to the sunlight,
– a large root collar diameter,
– a balanced ratio between root and shoot,
– high carbohydrate reserves,
– optimal level of nutrients,
– development of adequate mycorrhizal or *Rhizobium* infection (Jaenicke 1999).

Important parameters for seedling production include seed quality, hardening, temperature and light, water and nutrient status, pest and disease control, growth and biomass (Grossnickle 2012, 2018).

Some of these quality parameters are difficult to assess non-destructively, thus pointing out to the potential of advanced technologies such as HSI.

CAN HYPERSPECTRAL IMAGING ASSESS SEEDLING QUALITY PARAMETERS?

HSI provides a non-destructive approach to evaluate seedling quality parameters by capturing information across the visible (VIS), near-infrared (NIR), and shortwave infrared (SWIR) ranges (Paulus, Mahlein 2020; Mahlein et al. 2018). That is possible because HSI relies on electromagnetic energy reflection of plant tissues across specific wavelengths, due to differences in their chemical composition, internal physical structure, and surface properties, resulting in characteristic spectral signatures (Paulus, Mahlein 2020).

The use of HSI compared to traditional measurement methods for forest seedling quality assessment has been synthesised in terms of measurement feasibility, methodological requirements, and the extent to which HSI can directly or indirectly replace conventional assessments. The following section expands on this synthesis by presenting a detailed narrative description of each seedling quality parameter and its corresponding traditional and HSI-based assessment approach.

HIS-based assessment approach indicates how a parameter is evaluated using hyperspectral imaging, which can be direct, indirect, or derived. The direct approach is measured primarily from spectral data alone; the indirect approach is measured using modelling, calibration, or complementary technologies; and the derived approach is calculated from predicted traits.

Descriptions of the morphological and physiological quality parameters and their traditional measurement methods are based on Haase (2007, 2008).

Morphological seedling quality parameters

Since hyperspectral imaging (HSI) captures hundreds of continuous spectral bands, and not dimensional/physical information, most morphological parameters cannot be directly measured using HSI alone. Therefore, its role in morphological assessment is typically supportive, relying on integration with complementary technologies such as LiDAR, photogrammetry, and surface modelling approaches.

Shoot height. Shoot height is an indicator of photosynthetic and transpirational capacity. Traditionally, it is measured non-invasively using a meter stick. Shoot height could be estimated in a manner like plant height. Tao et al. (2020) used struc-

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tural models such as Digital Surface Model (DSM), Digital Elevation Model (DEM), and Crop Surface Model (CSM) to estimate winter wheat plant height. Plant height can be accurately estimated using UAV-based hyperspectral remote sensing data derived from hyperspectral-derived structural information (H_{CSM}), where it is strongly correlated with the height H (R^2 = 0.97). This indicates that UAV-based hyperspectral remote sensing data can be used to provide an estimation of crop height and may help address the limitations of traditional methods. Therefore, although shoot height is not directly measured using HSI, it can be derived indirectly through structural models (DSM, DEM, CSM). This parameter is therefore classified as an indirect HSI-based assessment.

Stem diameter. Stem diameter is a predictor of seedling survival and growth in the field. Traditionally, it is evaluated using a calliper, which is a non-invasive method. Hyperspectral imaging does not directly enable the measurement of stem diameter because it provides spectral rather than structural information. It is instead derived from 3D data, such as structural LiDAR data, for obtaining an estimation of the stem diameter. LiDAR alone can estimate stem diameter, but combining it with HSI can improve predictive models. This parameter is therefore classified as an indirect HSI-based assessment.

Height : diameter ratio. Height : diameter ratio is an indicator of balance between height and stem diameter. Using the traditional method, it is measured by calculating height and stem diameter. Height : diameter ratio is not directly measurable using HSI and is instead calculated from a previous prediction of height and diameter. This parameter is therefore classified as derived HSI-based assessment.

Bud length. Bud length is an indicator of seedling vigour and field growth potential. It can be evaluated with two traditional tools: a ruler or a calliper. Hyperspectral imaging alone does not enable the measurement of bud length; however, it may be indirectly estimated using approaches similar to those applied for plant height. Tao et al. (2020) used structural models such as Digital Surface Model (DSM), Digital Elevation Model (DEM), and Crop Surface Model (CSM) to estimate winter wheat plant height. Strong correlation of H_{CSM} and H (R^2 = 0.97) shows that plant height can be used to provide an estimation of bud length and may help address the limitations of traditional methods. Bud length could also be derived indirectly by integrating hyperspectral data with structural models (DSM, DEM, CSM), although studies for bud length estimation remain limited. This parameter is therefore classified as an indirect HSI-based assessment.

Shoot mass. Shoot mass is an indicator of growth potential and photosynthetic capacity. Using traditional methods, it is measured as fresh or dry weight, commonly obtained through water displacement or as dry weight following oven drying. Some of those traditional methods are invasive to the plant. Shoot mass is not directly measured by hyperspectral imaging but is estimated through modelling approaches based on vegetation indices. Normalised Difference Vegetation Index (NDVI), as well as image-based methods such as Bootstrapping Soft Shrinkage (BOSS), have been shown to be an effective and accurate prediction of shoot and root biomass of *Arabidopsis* plants (Song et al. 2023). This parameter is therefore classified as a direct/indirect HSI-based assessment.

Root mass. Root mass is an indicator of greater potential for growth and better field survival. Traditionally, it is measured in the same way as the shoot mass. Since roots are underground and not directly accessible, root mass cannot be evaluated without exposing the roots or estimated indirectly through modelling approaches. Shoot mass is not directly measured by hyperspectral imaging but is estimated through modelling approaches based on vegetation indices. NDVI, as well as image-based methods such as BOSS, have been shown to provide accurate prediction of shoot and root biomass of *Arabidopsis* plants (Song et al. 2023). This parameter is therefore classified as an indirect HSI-based assessment.

Shoot : root ratio. Shoot : root ratio is an indicator of seedling health, where a good and balanced shoot : root ratio indicates a healthy seedling. Traditionally, it is calculated from separately measured shoot and root biomass. Shoot : root ratio is derived after the prediction of both shoot and root mass using modelling approaches. This parameter is therefore classified as a derived HSI-based assessment.

Colour. Colour varies by species and can indicate lower vigour and/or chlorophyll content. Traditionally, it is assessed through visual observation, which is a non-invasive method. While it can be evaluated through visual observation, modern methods allow more accurate assessment. HSI enables quantita-

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tive estimation of pigment composition rather than measuring colour in a conventional sense. Zhao et al. (2025) developed a deep learning model for content evaluation of chlorophyll *a* (Chl*a*), chlorophyll *b* (Chl*b*), carotenoids (Car), and total pigment content (TPC) in lettuce canopy. Spatial distribution of pigment content is achieved by combining hyperspectral reflectance with the leaf-level inversion model. Since the study was conducted under controlled laboratory conditions, different factors (e.g. water stress, plant characteristics, plant age, etc.) can influence hyperspectral reflectance characteristics, thereby affecting the model's predictive accuracy. Therefore, the model should be tested, including different factors for effective pigment estimation. This parameter is therefore classified as an indirect HSI-based assessment.

Form. Form is an indicator of morphological abnormalities and physical damage that can negatively affect seedling quality. Traditionally, it is evaluated by visual observation non-invasively. The quality parameter 'form' includes structural characteristics such as multiple or forked shoots, stem sweep, root deformity, stiff lateral roots, and physical damage. Hyperspectral imaging does not directly enable the measurement of form structural characteristics because it provides spectral rather than structural information. For obtaining an estimate of structural characteristics, LiDAR data are used instead. LiDAR alone can estimate form, but combining it with HSI can improve predictive models. This parameter is therefore classified as an indirect HSI-based assessment.

Physiological seedling quality parameters

In contrast to morphological parameters, physiological parameters are more closely linked to biochemical and functional plant properties. As a result, hyperspectral imaging plays a more direct role, although many applications still rely on modelling approaches.

Cold hardiness. Cold hardiness is an indicator of minimum temperature tolerance and freeze damage resistance. Traditionally, it is assessed through freezing the whole plant or by freeze-induced electrolyte leakage methods, which are invasive. Cold hardiness and freeze tolerance can cause physiological changes in plants; therefore, cold hardiness could be estimated in a manner like freeze tolerance by extracting spectral information from hyperspectral data.

The spectral data provided information about reductions in chlorophyll and water concentrations in the subject seedlings. To address the imbalanced data from freeze-stressed loblolly pine seedlings, a cost-sensitive learning technique was proposed for modelling, as demonstrated in Lu et al. (2021). The hyperspectral imaging approach based on the proposed learning technique had a geometric accuracy of 75% to 96% for different scanning events. Therefore, the HSI is a potentially valuable tool that offers improved efficiency for freeze-tolerance screening. This parameter is therefore classified as an indirect HSI-based assessment.

Root growth potential. Root growth potential is an indicator of a seedling's ability to develop new roots under optimal environmental conditions. Traditionally, seedlings are planted in soil or placed into hydroponic tanks in optimal conditions, and the length and number of newly formed roots are evaluated. Since roots are underground and not directly accessible, root growth potential cannot be evaluated without exposing the roots or through root monitoring. Root growth was analysed combining HSI and imec VNIR SNAPSCAN camera with the Random Forest (RF) image classification method and the Support Vector Machine (SVM) machine learning approach. The proposed method provided reliable classification between root, soil, and the root-soil interface, which can assist in monitoring of root biomass and growth (Faehn et al. 2024). This parameter is therefore classified as an indirect HSI-based assessment.

Bud dormancy. Bud dormancy is an indicator of bud activity, showing dormancy status and stress resistance. Traditionally, there are three assessment methods: (*i*) placing the seedlings in optimal conditions; (*ii*) squashing and staining buds on a microscope slide, where a Mitotic Index (MI) is evaluated; (*iii*) estimation of the number of primordia by dissecting buds. Hyperspectral imaging alone does not allow direct quantification of bud dormancy, but it can be quantified in combination with other technologies using spectral patterns. Dmitriev et al. (2024) applied a Linear Discriminant Analysis (LDA) model to hyperspectral data for identifying 'vegetation' and 'dormancy' states in coniferous plants by analysing their annual time series of spectral characteristics, achieving a classification accuracy of 97.3%. Even though they assessed dormancy at a plant or shoot scale in laboratory conditions, this method could potentially be adapted to es-

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timate bud dormancy, under the assumption that a seedling is observed under appropriate conditions and calibration. This parameter is therefore classified as an indirect HSI-based assessment.

Plant moisture stress. Plant moisture stress is the most damaging stress and an indicator of low seedling survival and growth. Traditionally, it is determined by placing a seedling in a pressure chamber. Water stress levels were estimated by analysing the spectral signatures of plant leaves using HSI. The plants were monitored with both hyperspectral and RGB cameras, and stress levels were correlated with calculated spectral indices, such as the Normalised Difference Vegetation Index (NDVI) (Kim et al. 2011). In outdoor applications, the hyperspectral camera must be carefully calibrated to account for illumination changes so that the hyperspectral image data is not distorted. Neuwirthová et al. (2026) demonstrated the potential of HSI and machine learning for assessing the physiological state of Scots pine seedlings. Water stress during the stress period and during the recovery phase was successfully detected using hyperspectral data and the Random Forest classification algorithm. The achieved accuracy was 83% during the stress period and 79% during the recovery phase.

Since water stress and plant moisture stress are closely related, as they both exhibit insufficient water availability and are often used without distinction, the methodology for water stress can also be applied to plant moisture stress. This parameter is therefore classified as a direct/indirect HSI-based assessment.

Nutrient status. Nutrient status is important for optimal physiological processes and is an indicator of seedling health. Traditionally, it is evaluated through laboratory tissue analysis. Long et al. (2023) applied a Visible Near-Infrared HSI technique in the detection of nitrogen (N), phosphorus (P), and potassium (K) concentrations from *P. elliottii* × *P. caribaea* saplings. The 38 wavelengths were removed at both ends, and MSC-CARS was used to extract the characteristic wavelengths related to N, P, and K. Several learning regression algorithms were used as prediction models. Nutrition evaluation was based on spectral information that is related to the content of internal compounds composed of nutrient elements. Hyperspectral imaging enables nutrient estimation with moderate accuracy for potassium (R^2 = 0.697) and phosphorus (R^2 = 0.622), and high accuracy for nitrogen (R^2 = 0.833). This parameter is therefore classified as a direct HSI-based assessment.

Chlorophyll fluorescence. Chlorophyll fluorescence is an indicator of photosynthetic activity and plant responses to disturbances. Traditionally, it is measured using fluorometers, a non-invasive method. Hyperspectral imaging cannot directly measure chlorophyll fluorescence, as it relies on detecting reflected light in specific narrow spectral bands, rather than fluorescence light. However, Yang et al. (2021) demonstrated that vegetation indices derived from hyperspectral data can be used to quantify leaf-scale chlorophyll fluorescence (ChlF) parameters. ChlF parameters consist of dark-adapted parameters: *Fo* (minimal fluorescence), *Fm* (maximal fluorescence), *Fv*/*Fm* [maximum quantum efficiency of photosystem II (PS II), where $Fv = Fm - Fo$]; and light-adapted parameters: *Fo*' (minimal fluorescence), *Fm*' (maximal fluorescence), *Y*(II) (effective quantum yield of PS II photochemistry). For the purpose of quantifying ChlF parameters, new vegetation indices were developed. This approach achieved moderate to high prediction, especially in distinguishing species *Camellia oleifera* and eucalyptus with high accuracy (*OA* = 97.56%, *AA* = 97.47%, Kappa = 0.9665) (Yang et al. 2025). The accuracy was assessed using overall accuracy (*OA*), average accuracy (*AA*), and Kappa coefficient, where *OA* represents the percentage of correctly classified reference samples; *AA* refers to the average accuracy across all classes; and the Kappa measures the degree of agreement between the classification results and the samples (Yang et al. 2025). This methodology could be used to quantify ChlF parameters on forest seedlings by adapting the vegetation indices depending on species-specific leaf structures. This parameter is therefore classified as an indirect HSI-based assessment.

Some traditional methods for seedling quality assessment, such as measuring shoot height, stem diameter, bud length, root growth, cold hardiness, and chlorophyll content, are often invasive or destructive. These include techniques such as squashing and staining buds on microscope slides or freezing the entire plant. In contrast, HSI offers a non-destructive approach and the ability to perform real-time and rapid measurements of plant traits. HSI can directly assess physiological and biochemical traits, such as shoot mass, plant moisture stress, and nutrient content, by analysing characteristic spectral signatures. However, HSI cannot directly measure struc-

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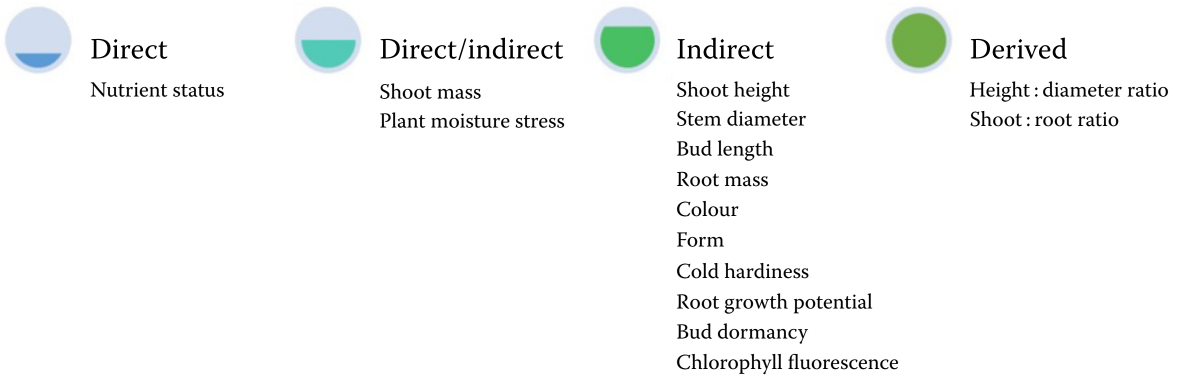

Figure 3. Classification of morphological and physiological seedling quality parameters according to the hyperspectral imaging (HSI) assessment approach

Table 3. Application of hyperspectral technology in forest seedlings studies

Applications	Plant species	Reference(s)
Moisture and nitrogen content	*Pinus massoniana* Lamb.	Huang et al. 2021
Freeze tolerance	*Pinus taeda* L.	Lu et al. 2021
Detection of fusiform rust disease	*Pinus taeda* L.	Pandey et al. 2021
Nutrient deficiency detection	*Eucalyptus grandis* W.Hill ex Maiden	Singh et al. 2021
Nutrient content estimation	*Pinus elliottii* × *Pinus caribaea*	Long et al. 2023
Genetic variation	*Pinus sylvestris* L.	Stejskal et al. 2023
Needle chlorophyll and water content	*Cunninghamia lanceolata* (Lamb.) Hook.	Xing et al. 2024
Chlorophyll content estimation	*Ginkgo biloba* L.	Yue et al. 2024
Water stress detection	*Agathis australis* (D.Don) Loudon	Felix et al. 2025
Elevated CO_2 effect on leaf reflectance	*Quercus robur* L.	Jones et al. 2025

tural traits such as shoot height, bud length, and stem diameter, or chlorophyll fluorescence, as it relies on detecting reflected light in specific narrow spectral bands, rather than fluorescence light. Therefore, it can be quantified by integrating HSI with complementary technologies such as LiDAR, structural models, vegetation indices, image-based methods, etc. Overall, HSI offers a non-invasive alternative to traditional techniques, with strong potential for forest seedling quality assessment. The classification of seedling quality parameters according to HSI assessment approach is summarised in Figure 3.

EXISTING KNOWLEDGE IN HYPERSPECTRAL IMAGING APPLICATIONS IN FOREST SEEDLING PRODUCTION

Although hyperspectral technology has been used in forestry since the 1990s (Treitz, Howarth 1999) and has a wide range of applications today, its use in forest seedling production remains unexplored. However, several studies have investigated the application of hyperspectral technology on forest seedlings (Table 3).

GUIDELINES IN FOREST NURSERY PRODUCTION

There is a need for developing more efficient seedling quality assessment methods, such as refining testing procedures for early detection of damaged plants (Haase 2010). This would increase the chances of seedling performance in the field after planting (Grossnickle 2012). Because some traditional methods, such as root growth, cold hardiness, and chlorophyll content, are invasive and destructive to plants, the adoption of non-destructive methods is essential. Technologies such as HSI could be applied either independently or in combination with other technologies to assess parameters non-destructively. HSI qual-

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ity parameter detection methods, in combination with LiDAR, could provide estimates of stem diameter and form. When integrated with structural models, it could estimate shoot height and bud length. Additionally, using vegetation indices, it is possible to estimate shoot and root mass prediction, plant moisture stress and chlorophyll fluorescence.

Effective monitoring of quality parameters is essential during seedling development. New technologies, such as HSI, are becoming increasingly accessible, allowing for more effective monitoring of seedling development and quality parameters. Grossnickle and MacDonald (2018) proposed that advanced technology, such as fluorescence-imaging systems, could be integrated with irrigation systems to automatically trigger irrigation or fertigation in response to early signs of plant stress. A similar approach could be applied using HSI, which can detect plant stress caused by pest and disease infestation or deficiencies in water and nutrients. This would enable well-timed and precise responses, including automated application of pesticides, fungicides, irrigation, and fertigation.

CHALLENGES AND LIMITATIONS

Despite being a high-end technology, HSI still faces several challenges and limitations. Large amounts of data are the result of HSI capturing hundreds of continuous spectral bands (Bian et al. 2022), which is one of the challenges in data processing (Chang 2013; Liu et al. 2023). Large data may result in high computational cost of data processing, long delay of data transmission and communications, and difficult management of data storage and archiving (Chang 2013). This can be resolved by data reduction (Chang 2013) operations, such as feature selection or feature extraction (Luo et al. 2024).

Another related challenge involves compressing spectral data resulting from highly correlated bands while keeping essential information, which can be resolved by developing methods to remove unnecessary information (Chang 2013). Even though both challenges can be resolved by data reduction, they are actually different operations and should be viewed independently (Chang 2013). Other key challenges and limitations of HSI are summarised in Table 4.

There is a need for data standardisation processes and analysis due to the different data collection methods in order to ensure data consistency between different studies (Paulus, Mahlein 2020; Luo et al. 2024). Additionally, to extract more reliable information from hyperspectral data, it is important to develop more efficient data processing and analysis algorithms (Luo et al. 2024). One of the limiting factors of widespread use of hyperspectral technology is its high cost (Fassnacht et al. 2016; Luo et al. 2024). Furthermore, multi-temporal data collection is time-consuming, and from a practitioner's perspective, potential accuracy gains may not justify the expense (Fassnacht et al. 2016). Large data volumes are another important challenge because it is necessary to remove redundant information so that relevant information can be evaluated (Awada et al. 2024).

The challenges and limitations presented in Table 4 refer to HSI in general, although several could also be applied specifically to its use in plant phenotyping, which includes illumination, plant-specific traits, large data volume, etc.

Illumination position (Detring et al. 2024) is one of the major challenges in HSI for plant phenotyping because the data quality and the level of detail depend on the position of the illumination and its angle. Additionally, when imaging 3D objects such as plants, a phenomenon of edge pixels occurs (Detring et al. 2024). The phenomenon occurs at the boundary between healthy or diseased leaves and black ground, thereby capturing mixed spectral information from both objects (Detring et al. 2024). Tissue composition, surface property, size, and consistency of the wax layer and epidermis (Behmann et al. 2016) are plant-specific traits that affect HSI of the smallest organs and deformations due to plant growth. Additional limiting factors are the wind, actor movement, and changes in leaf position due to growth, sun movement, or stress (Behmann et al. 2016). The challenge lies in the high diversity of plants and their traits, which are found in countless different environments (Pieruschka, Schurr 2022). Large volumes of data are generated during plant phenotyping; therefore, data management is necessary. This includes data storage, sharing, and analysis, which must continuously advance as the amount of data increases (Awada et al. 2024). Challenges in data standardisation could be created by different collection methods (Pieruschka, Schurr 2022;

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Table 4. Examples of challenges and limitations of hyperspectral imaging (HSI)

Challenges/limitations	Reference(s)
Data-related challenges	
Large data volumes, data complexity and high dimensionality	Chang (2013); Liu et al. (2023)
Data standardisation	Luo et al. (2024)
Different data collection methods	Luo et al. (2024)
Complex hyperspectral image classification	Chang (2013)
Timing differences in data collection	Behmann et al. (2016)
Difficulty in effective analysis and data interpretation	Wahabzada et al. (2016)
Shadowed, noise-filled, or mixed pixels	Chang (2013); Ballanti et al. (2016)
Instrumentation and technical limitations	
High equipment cost	Luo et al. (2024)
Limited capacity of imaging instruments	Ngadi, Liu (2010)
Slow image acquisition	Fahlgren et al. (2015)
Scanner/sensor setup issues	Behmann et al. (2016); Paulus, Mahlein (2020)
Environmental and operational factors	
Illumination position sensitivity	Paulus, Mahlein (2020); Detring et al. (2024)
Sensitivity to environmental conditions	Behmann et al. (2016)
Detection of concealed or camouflaged targets without prior knowledge	Chang (2013)
Biological and subject-specific challenges	
Plant-specific traits	Behmann et al. (2016); Paulus, Mahlein (2020); Yue et al. (2024)

Luo et al. 2024). A response to that challenge would be to develop a hyperspectral phenotyping data-sharing platform in order to keep data consistency and simplify processing methods (Luo et al. 2024). Although nursery production of forest seedlings has a much longer history than the use of HSI, it still faces its own challenges and limitations.

Even though the quality parameters are met (morphological and physiological), and the seedling appears healthy, that does not necessarily mean that the seedling will survive after planting out (Jaenicke 1999). Therefore, adapting seedling production to the specific environmental conditions of the intended planting site is essential to ensure seedlings are healthy, strong, and better adapted to harsher conditions (Jaenicke 1999).

FUTURE PERSPECTIVES

Current use of HSI in forest seedling nursery production is limited in assessing seedling quality parameters. However, recent studies by Dalponte et al. (2009), Kim et al. (2011), Tao et al. (2020), Lu et al. (2021), Long et al. (2023), Song et al. (2023), and Faehn et al. (2024) have presented effective detection methods in evaluating similar quality traits in plants. These methods could potentially be adapted for the assessment of seedling quality in forest seedling production. Since HSI is enabling non-invasive detection (Mahlein et al. 2018) of a broad range of plant traits, it represents a promising advancement in seedling quality assessment and, consequently, nursery production, ultimately supporting forest restoration efforts. Moreover, for further improvement of seedling quality assessment, it enables the simultaneous detection of multiple plant attributes and supports the advancement of modern plant phenotyping approaches. Additionally, integrating HSI technology into seedling quality assessment can lay a foundation for nursery practitioners and all stakeholders involved in the application of seedlings. Nursery practitioners could benefit from technology integration in both indoor and outdoor environments.

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Even though the use of the technology in assessing seedling quality parameters is limited, recent studies proposed detection methods for similar plant traits that could potentially be adapted for quality assessment.

For seedling quality assessment improvement and consequently seedling production effectiveness, it is necessary to establish standardised hyperspectral data processing and analysis methods (Luo et al. 2024). Furthermore, standardisation of hyperspectral data is challenging due to differences in data collection methods (Luo et al. 2024). Therefore, to improve the consistency of hyperspectral data and facilitate the exchange of raw data and processing methods, there is a need to develop a hyperspectral phenotyping data-sharing platform (Luo et al. 2024). The application of HSI technology offers significant advantages despite its high cost (Stuart et al. 2022), especially when improving seedling quality.

Multiple challenges and limitations highlight that hyperspectral technology is still in its developing stage. The current limitations and future perspectives of HSI in forest nurseries are summarised in Figure 4. Therefore, the development of more budget-friendly hyperspectral technology will help make the assessment of quality parameters and plant phenotyping more accessible.

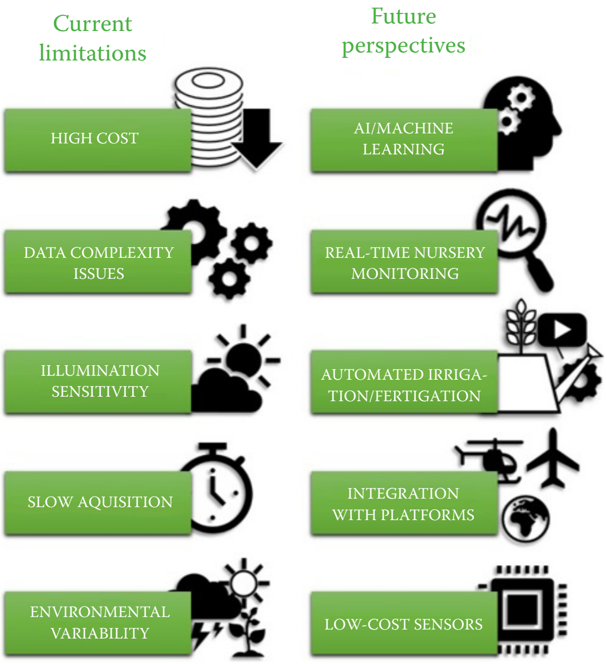

Figure 4. Current limitations and future opportunities of hyperspectral imaging in forest nurseries

CONCLUSION

Advancements in plant phenotyping and HSI detection methods have opened the possibility of improving forest seedling quality assessment. Since forest seedlings are one of the most widely used forms of forest planting stock, their quality assessment is important for the successful establishment of forest stands. Due to the extreme environmental conditions and various challenges, there is a need for forest seedling production advancement. Seedling quality can be assessed with traditional methods, of which some are invasive and damaging to the seedlings; however, with the HSI it is possible to evaluate them non-destructively.

Despite HSI being an advanced technology, there are limitations in forest seedling nursery production, especially in seedling quality assessment. HSI is based on the principle that all materials reflect light in specific narrow spectral bands and capture it in specific wavelengths. Considering that, its ability to detect dimensional/physical traits or fluorescence light is not possible without combining with other technologies. However, developed detection methods for evaluating similar quality traits in plants could potentially be adapted to assess seedling quality in forest seedling production.

Nursery production of quality seedlings using hyperspectral plant phenotyping offers significant advantages despite all the challenges the technology is facing. Improved seedling quality would contribute to a more successful establishment in forest stands under various environmental conditions, thereby contributing to greater resilience.

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