JOURNAL OF ECOLOGY AND CONSERVATION
Jul - Sep 2026, VOL. 4, ISSUE 3, pp. 22-30
https://doi.org/10.61577/jec.2026.1000014

REVIEW ARTICLE

Plant bioluminescence for sustainable horticulture: Living biosensors, crop sentinels and precision crop monitoring

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ABSTRACT

Bioluminescence, although frequently framed as an ecological curiosity of fireflies, marine macrofauna and glow-in-the-dark fungi, has become a real-world tool for understanding plant biology. Over many decades, firefly luciferase-based reporter systems have facilitated sensitive and quantitative measurements of gene expression, circadian rhythms and signalling in living plants. The ability to achieve bioluminescence within a single organism, where both luciferase and its substrate are produced on demand, has only recently been realised through the construction of bacterial, for example *Vibrio*, and fungal light-producing pathways. This transition is particularly relevant to agriculture and horticulture, where repeated, non-destructive monitoring can be achieved without external luciferin application. This article connects the molecular basis of different bioluminescence systems with their recent applications in crop and horticultural research. Particular focus is placed on autonomous reporters for defence hormones, pattern-triggered immunity and viral infection, sentinel plants for protected cultivation, high-throughput phenotyping and breeding, and lux-based microbial biosensors for agriculturally relevant contaminants. An ecological perspective is also important. Natural fungal bioluminescence may influence insect behaviour, while artificial light at night can disrupt plant–pollinator networks. Therefore, engineered luminous plants must be optically and ecologically characterised rather than presumed biologically neutral. Overall, bioluminescence currently holds its strongest value as an investigational and diagnostic platform rather than routine field-scale agronomic practice, where signal intensity, metabolic context, imaging conditions, genetic containment and regulatory acceptance remain major limitations.

KEYWORDS

Autonomous bioluminescence; Crop sentinels; Fungal bioluminescence pathway; Plant biosensors; Precision horticulture; Sustainable crop protection

ARTICLE HISTORY

Received 01 August 2026;
Accepted 22 August 2026;
Published 30 August 2026

Introduction

Bioluminescence is the production of visible light by a living organism as the result of a biochemical reaction. It has evolved through chemically diverse luciferin–luciferase systems over evolutionary time and is the natural basis of signaling, defense (some luciferins have antibacterial properties), hunting/attracting prey, and possibly spore dispersal in some fungi. Trained on data until October 2023, Free Writing Moment. For plant scientists, though, the main attraction of bioluminescence is a different property: Light can be measured continuously at very low background without destroying the tissue. An example is the firefly luciferase gene, which was used as a reporter in plant cells and transgenic plants nearly four decades ago, establishing a method that later became instrumental in studies of plant promoters, circadian rhythms and signalling [1,2,3]. Luciferase-based imaging subsequently became particularly valuable because reporter activity could be followed repeatedly in intact plants, allowing temporal changes in gene expression to be monitored without destructive sampling [4,5].

The agricultural significance of this technology has grown as crop management is progressively reliant on early diagnosis. Yields, as well as quality, often start to fall before a single spot on the plant shows symptoms. Such as, a virus might move through a nursery before leaf distortion is observable; or a root-zone problem may show itself primarily as a hormonal or transcriptional response; likewise, a greenhouse malady can be very costly by the time conventional scouting identifies it. Imaging platforms e.g., hyperspectral cameras, thermal sensors, and chlorophyll fluorescence, are powerful, but their detection of stress is indirect because changes in optical or physiological traits can be caused by multiple factors. With such a genetically encoded bioluminescent circuit, one could even entertain an alternative possibility: engineering the plant to emit light only when a defined molecular event occurs. Essentially, the plant is integrated within the sensor. The broader development of image-based phenotyping has further strengthened the potential for repeated optical measurements of plant responses, particularly when spatial and temporal information is required [6,7].

That concept has shifted from theory to practical demonstration. Autonomously luminescent tobacco chloroplasts were generated from the bacterial lux operon [8]. Since caffeic acid is already incorporated in plant phenylpropanoid metabolism, discovery of the fungal caffeic-acid bioluminescence cycle subsequently enabled nuclear expression of a self-sustained pathway in plants [9,10]. Further work has optimized brightness, expanded dynamic range and reformed the pathway to programmable reporters [11,12,13]. Most notably for crop protection, autonomous reporters now visualise defence responses and can even be wired to virus-responsive circuits [12,14].

All in all, this needs careful framing. Bioluminescence is not to be confused with ultraweak photon emission or 'biophotons' detected from stressed plant tissues independent of a defined luciferase–luciferin system. While these phenomena can be informative, their physical mechanisms and instrumentation differ distinctly from genetically encoded bioluminescent reporters. Here I review enzymatic bioluminescence, natural, engineered or implemented as a

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reporter and why this technology could find value at the agriculture–horticulture–ecology interface. It also considers ecological risk as part of technological development. The glowing ornamental or sentinel plant is more than a laboratory tool; after passing through a greenhouse, garden, or landscape, it becomes an entirely new night-time optical character in an ecological community. Representative natural and engineered contexts are shown in Figure 1.

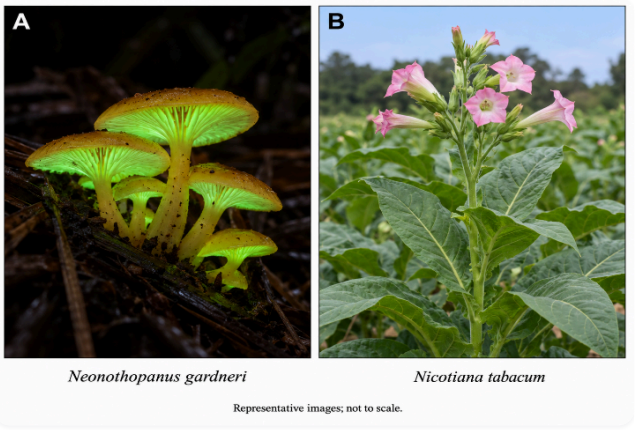


Figure 1. (A) Natural fungal bioluminescence, represented by Neonothopanus gardneri; and (B) An engineered plant reporter, represented by Nicotiana tabacum.

Scope and Approach of this Review

This is a critical narrative review and not a systematic review. It focuses on peer-reviewed primary studies based on the main criteria of establishing chemistry (as in a bioluminescent system), introducing it as a plant-compatible reporter, or using it in a context relevant to ecology or agriculture. The foundational work is retained from the 1980s onward to explain how luciferase reporters developed into increasingly autonomous systems [1,2,8]. More recent studies published between 2020 and 2026 were given precedence where they described fungal pathway engineering, genetically encoded autoluminescence in plants, improved autonomous bioluminescence pathways, plant defence reporters, multicolour systems, and related plant imaging applications [10,13,15]. Environmental biosensing methods are included only if the output is a true bioluminescent signal and the measured endpoint applies to contaminant uptake in agricultural soils, irrigation water, or biological availability of contaminants [16,17,18]. Any evidence of bioluminescent sensing was distinguished from conventional fluorescence- or colour-based biosensors, which were outside the primary scope of this review [19].

The review also distinguishes between demonstrated capability and what is merely proposed for deployment. The definitions of key terms in colour are as follows: 'established' is a function demonstrated experimentally in peer-reviewed literature; 'proof-of-concept' is a validated biological blueprint that has not yet shown repeatable agronomic benefit; and 'prospective' refers to a technical-sounding use requiring crop-scale validation. This matters because progress has been fast in the molecular world, but evidence that a luminescent reporter improves a real management decision is much thinner. No new dataset is produced, and no quantitative meta-analysis is attempted. The broader field of genetically encoded biosensors provides a useful conceptual framework for distinguishing molecular sensing from the optical reporter used to visualize the response [19,20].

Another limit is the difference between engineered bioluminescence and ultraweak photon emission (or biophoton emission) related to oxidative processes present in unstressed or stressed plant tissues. Another area that could be addressed non-invasively employing ultraweak photon emission, rather than a defined genetically encoded luciferase–luciferin circuit, is stress sensing (which falls outside the scope of this paper). Similarly, we present literature on artificial light at night as an ecological analogue for decomposing how nocturnal optical cues can influence interactions; dim plant bioluminescence is not proposed as a direct replacement for lamps with similar effects.

The overarching question guiding the review is thus not whether a plant can emit visible light, but where bioluminescent signals add to information that cannot be obtained as straightforwardly through conventional imaging or molecular diagnostics. Value can be evaluated using three metrics: specificity to a molecular target, functionality and salience. This structure lets the conversation focus on computable biological information rather than the aesthetic novelty of a glowing plant.

Biological Foundations: Three Routes to Biological Light

Bioluminescent systems all couple an oxidation reaction to the formation of an electronically excited product that emits a photon as it returns to a lower-energy state. Lineages have disparate chemistry, cofactors and genes. Three systems are of note to plant and agroecological studies: firefly luciferase, the bacterial lux system, and the fungal caffeic-acid pathway (Figure 2). The brightness of synthetic biosensors matters, but more critically their practical differences, especially between substrate dependence and metabolic burden.

Firefly luciferase remains a standard reporter. Early plant studies established firefly luciferase as a sensitive reporter for promoter activity and circadian regulation, while subsequent work demonstrated its utility for continuous monitoring of gene expression in intact [2,4]. It oxidizes D-luciferin in an ATP-and oxygen-dependent manner. The reporter affords a particularly high signal-to-noise ratio as plants have almost no endogenous background at the relevant wavelengths. It is particularly useful for monitoring promoter activity over extended periods in whole seedlings or leaves. Its main drawback in whole plants is that D-luciferin must be provided. Low signal can also be ambiguous due to uneven uptake: weak light could reflect poor gene expression, poor substrate delivery, or both. Repeated luciferins use also adds cost and operational steps for long-duration imaging [11].

To overcome the substrate problem, researchers have genetically encoded the native bacterial lux system to include light-producing machinery and aldehyde synthesis. The canonical setup uses a LuxA/LuxB (*LuxA and LuxB are the two subunits of bacterial luciferase. Together, they form the active luciferase enzyme that catalyses the bioluminescent reaction, producing visible light*) heterodimer to make bacterial luciferase; LuxCDE (*LuxC, LuxD and LuxE are enzymes involved in the fatty-acid reductase pathway that regenerates the long-chain aldehyde substrate required by bacterial luciferase. This enables the lux system to continuously produce light without repeated external addition of the aldehyde substrate*) reverts the long-chain aldehyde substrate to its original form; however, reduced flavin and oxygen take part in the reaction

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[8,21]. The pathway was localized in tobacco chloroplasts, thereby achieving autonomous light that was visible to the naked eye. This accomplishment drove key advances in plastid transformation while also highlighting that many crop and ornamental species remain, and continue to be, difficult or impossible to transform. This early discovery has consequently made bacterial lux especially useful for deployment in microbial whole-cell biosensors, where light output might signify toxicity, contaminant bioavailability, or promoter activation in environmental matrices. The autonomous nature of bacterial lux systems has also made them useful in whole-cell biosensors, where changes in luminescence can report cellular responses to environmental contaminants and toxic compounds [17,22].

Fungi evolved to alter the prospects of luminous plants by changing plant metabolism through their chemistry. A cycle results in the formation of fungal luciferin from caffeic acid via hispidin synthase (HispS), hispidin-3-hydroxylase (H3H), and two recycling steps involving caffeylpyruvate hydrolase (CPH) and caffeylacetone. Research into the chemistry and genetics of luminescent fungi led to the identification and evolutionary organization of this pathway [9,23-25]. The development of autonomous fungal-pathway reporters represents a broader shift from substrate-dependent reporter systems toward self-sustained plant-compatible [11,12,10].

Autonomy does not mean unlimited bioluminescence. Light production competes with basal cellular metabolism for metabolic precursors and depends on stimulus-dependent pathway induction, tissue optical properties, oxygen availability, and downstream substrate turnover. Consequently, metabolic engineering has focused on enhancing precursor supply and optimizing pathway architecture. Recent studies have demonstrated several approaches to improve bioluminescent output, including the optimization of fungal bioluminescence pathways across eukaryotic contexts, metabolic engineering to increase the availability of caffeic acid and hispidin, key precursors for fungal bioluminescence, and the development of hybrid biosynthetic routes for self-perpetuating luminescence [13,15,26]. These advances have important implications for plant bioluminescence imaging, as increased photon yield can reduce camera exposure time, improve temporal resolution, and extend the practical range of bioluminescent sensing.

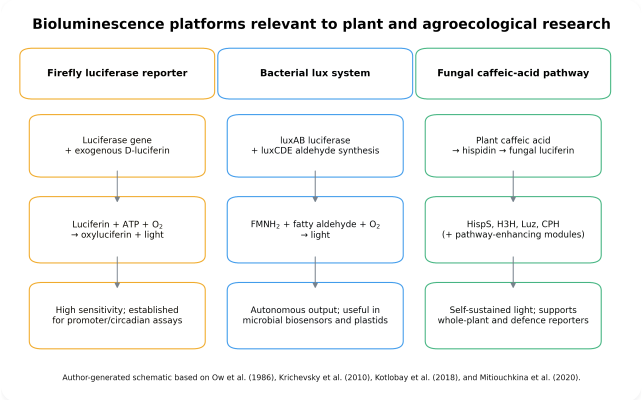


Figure 2. Mechanistic comparison of major bioluminescence systems used in plant and agroecological research. The firefly system is highly sensitive but usually requires exogenous luciferin; bacterial lux and fungal pathways can be configured for autonomous light production. Author-generated original schematic.

Table 1. Comparison of bioluminescence systems relevant to agriculture, horticulture and plant ecology.

System	Substrate source	Autonomous?	Typical emission	Main strengths	Key limitations	Plant/agro use
Firefly luciferase (FLuc)	D-luciferin supplied externally	No	~560 nm; condition-dependent	High sensitivity; mature reporter; strong temporal assays	Substrate delivery and cost; ATP dependence	Promoter assays, circadian biology, stress signalling
Bacterial luxCDABE	Endogenously regenerated aldehyde + reduced flavin	Yes	Blue-green (~490 nm)	Fully autonomous; compact microbial biosensing logic	Shorter wavelength is more absorbed by plant tissue; plastid engineering challenging	Microbial soil/water biosensors; transplastomic proof-of-concept
Fungal caffeic-acid pathway	Plant caffeic acid converted to fungal luciferin	Yes	Green (~520–530 nm)	Nuclear-encoded; substrate-free whole-plant imaging; programmable	Coupled to host phenylpropanoid metabolism; tissue-dependent output	Autonomous gene-expression, defence and virus reporters
Improved / hybrid autonomous pathways	Engineered precursor supply and pathway modules	Yes	Green or multicolor variants	Higher brightness, improved dynamic range and spectral options	More genetic components; calibration and metabolic burden	Quantitative synthetic biology, future sentinel and multicolor systems

From Reporter Gene to Living Plant Sensor

Light alone, considered as information and not just a source of illumination, reveals the scientific value of plant bioluminescence most clearly. A reporter can encode promoter activity, whether or when a signaling pathway is induced, how strong the defense response is, or whether a pathogen-responsive molecular event has occurred. More broadly, genetically encoded plant biosensors can couple molecular recognition to readily detectable outputs and have been proposed for monitoring developmental, hormonal, environmental and stress-related processes in vivo [19,20]. This means one of the most important design decisions is not just which luciferase to use, but where in the biology to insert the sensor.

The use of promoter–luciferase fusions to monitor transcription dates back to the early days of mammalian gene expression studies. The classic CAB2::LUC system addressed circadian and phytochrome regulation in Arabidopsis while also providing a rationale for using luminescence to repeatedly measure the same living individual [3]. This concept, taken further in autonomous fungal systems, does not require the application of any substrate. The fungal pathway was reconstructed in a variety of plants, showing that it could reveal time-dependent gene expression patterns and hormone responses. They then modified the pathway to expand its quantitative dynamic range, making autonomous light much

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more useful for comparing regulatory sequences or synthetic circuits that simply confirm whether a construct is 'on' versus 'off'. Earlier defence-related reporter studies also demonstrated that luciferase promoter fusions could reveal both local and systemic activation of plant defence pathways [27,28].

A particularly attractive target is plant immunity. Garcia and Steinbrenner constructed a genetically encoded bioluminescent reporter of pattern-triggered immunity in Nicotiana benthamiana [29]. The reporter translates activation of defence-associated transcription into visible output, providing a functional readout for receptor activity. In 2026, Balakireva et al. enabled long-term, substrate-independent imaging of salicylic acid- and jasmonic acid-related defence responses through extended autonomous luminescence [14]. Despite poor resolution, responses to pests and pathogens could be tracked in Arabidopsis or N. benthamiana using local and systemic signals supported by consumer-grade cameras [14]. Why this matters for horticulture: defence dynamics are typically spatial, as an arrival can signal a lesion, feeding site, or infected sector and elicit responses from distances far beyond any visible point of attack. Luciferase reporters have also been used in forward genetic and high-throughput approaches to identify components of pathogen-triggered immunity, illustrating how luminescence can function not only as a diagnostic output but also as a screening phenotype [30].

The most recent strong link developed is the field of autonomous viral sentinels, which bridge crop diagnostics. Calvache et al. developed plants whose infection with a potyvirus generates an observable difference in the bioluminescent signal, such as a spectral change that can be imaged using inexpensive hardware [12]. The work shows a broader point beyond the particular virus: a plant can embody a molecular recognition circuit, an amplifying biological reporter, and an observable output in the same living organism. Although such systems still cannot replace laboratory diagnostics, if one sentinel plant can continuously monitor a greenhouse compartment or nursery batch and alert when confirmatory testing is needed, surveillance economics could be transformed.

Bioluminescent outputs also provide complementary information to other plant sensing modalities. Fluorescence relies on excitation light and suffers from autofluorescence and illumination artefacts; spectral imaging (including thermal) can help infer plant status through physical traits; electrochemical or nanosensor systems can provide very specific measurements but require introducing material into the plant. Bioluminescence: Bioluminescence is atypical in that the signal can be produced from within the tissue in darkness, and it can be genetically coupled to a desired regulatory event. The flip side of this advantage is its disadvantage: biology must be engineered, so sensor performance depends on genotype, transformation efficacy, expression stability, and regulatory acceptance.

Agriculture and Horticulture Applications: What is Established and What Remains Prospective

For agricultural translation, it is important to distinguish experimentally supported applications from uses that remain plausible but unvalidated. Table 2 makes that distinction explicit. At present, the strongest evidence lies in plant research, controlled imaging, and proof-of-concept diagnostics; bioluminescence itself has not been shown to directly improve yield and should not be claimed to do so.

Protected cultivation is the most realistic environment for early deployment. Greenhouses, screenhouses and vertical farms already use fixed infrastructure, defined crop blocks, controlled lighting schedules and routine imaging. A sentinel genotype placed among commercial plants could be engineered to respond to a pathogen class, vector-transmitted virus or hormone signature associated with infection. The camera does not need to image every crop plant if sentinel plants are intentionally distributed as biological sampling points. Figure 3 shows a practical pipeline: a biological event activates a sensor circuit, the circuit changes light output, a low-light camera records the signal, software applies a threshold or spatial rule, and the grower receives a management cue. The management action might be targeted scouting, confirmatory molecular testing, rogueing of infected plants or intensified vector control—not automatic pesticide application.

Nursery and propagation systems offer another strong use case because losses can spread rapidly through clonally propagated material. A small number of sentinel plants could provide continuous warning in mother blocks, plug-production areas or hardening chambers. Autonomous reporters are especially attractive here because repeated substrate application would be inconvenient on a commercial scale. However, a useful nursery sensor would need to demonstrate low false-positive rates under changes in temperature, humidity, nutrient status and lighting. A defense promoter that responds to several stresses may be excellent for basic biology but too nonspecific for quarantine decisions.

High-throughput phenotyping and breeding may be nearer to routine use than field diagnostics. This perspective is consistent with the broader development of high-throughput plant phenotyping, in which imaging-based approaches are increasingly used to quantify plant traits across large populations and over time [6,7]. Reporter lines can convert otherwise hidden physiological events into a scalable optical phenotype. Breeders and molecular biologists could use luminescent reporters to compare alleles, promoters, immune receptors or stress-response pathways across hundreds of lines. Autonomous systems reduce labour for time-series phenotyping and support repeated measurements of the same plant. A powerful future design would combine a crop diversity panel with a standardized reporter background, allowing quantitative comparison of defense timing or hormonal dynamics rather than only final symptom scores.

Bioluminescence also has a role outside the plant itself. Bacterial lux reporters can be engineered to change light in response to a bioavailable contaminant or a toxic effect. Importantly, biosensor-based measurements can provide information about biological availability rather than simply total chemical concentration, making whole-cell reporter systems complementary to conventional analytical chemistry [17,31]. This is relevant to agricultural soils, irrigation water, composts and food-chain matrices because total chemical concentration does not necessarily represent the fraction experienced by living cells. Ivask et al., for example, assembled a suite of luxCDABE-based 'lights-on' bacterial strains for quantifying bioavailable heavy metals, including cadmium, mercury, zinc, lead and copper, alongside constitutively luminous strains that help distinguish target-specific induction from general toxicity [32]. Earlier work showed that

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bioluminescent bacterial sensors could also be applied to heavy metals in soil extracts [16]. More recent reviews similarly describe luminescent bacterial assays as rapid tools for environmental toxicity and pollutant screening, while emphasizing limitations related to reproducibility, matrix effects and field translation [18,22].

These assays can complement analytical chemistry by answering a biological question—how much of a contaminant is biologically accessible under the test conditions—but matrix colour, turbidity and the metabolic state of the reporter cells can influence light output. They should therefore be treated as rapid biological screens, not replacements for validated regulatory chemistry. This distinction is consistent with the broader biosensor literature, which emphasizes that biological reporters measure organism-level responses and bioavailable fractions rather than necessarily providing the same quantitative information as conventional chemical analysis [33].

Ornamental horticulture occupies a different space. Visible autoluminescent flowers demonstrate public-facing feasibility and create obvious commercial interest. Scientifically, ornamentals such as petunia are also useful because flower development and phenylpropanoid metabolism provide informative spatial patterns. The history of genetically engineered orange petunias entering commercial breeding without complete regulatory traceability reminds us that ornamental supply chains can move genetic traits widely [34,35]. Bioluminescent ornamentals therefore provide both an innovation opportunity and a governance case study: novelty traits need event characterization, labelling, stewardship and a plan for cross-border movement.

A recurring limitation across all these applications is the need for darkness. The bioluminescent signal is easiest to detect in low ambient light. Greenhouses can schedule imaging during darkness, but open-field deployment must contend with moonlight, skyglow, headlights and variable backgrounds. Optical filters, spectral-ratio reporters, enclosed camera hoods and brief dark adaptation could help. The 2026 viral sentinel study is particularly relevant because a change in spectral composition can be more robust than a simple increase in total brightness [36]. Future crop sensors should therefore be designed around signal discrimination, not maximal glow alone.

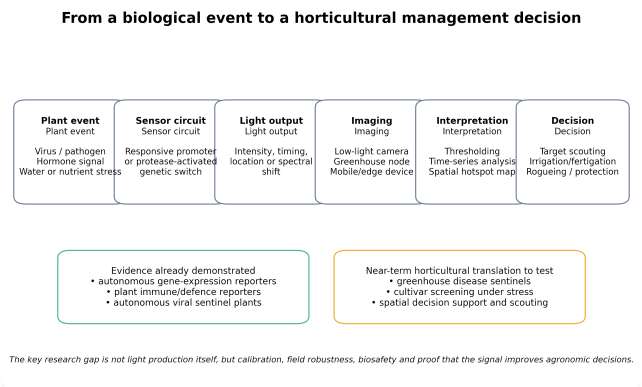


Figure 3. Conceptual pipeline linking an engineered plant response to a management decision in protected horticulture. Green boxes indicate applications already demonstrated at proof-of-concept level; orange boxes indicate translational steps that require agronomic validation. Author-generated original schematic.

Table 2. Current and prospective applications of bioluminescence at the agriculture–horticulture interface.

Application	Evidence status	Bioluminescent function	Potential value	Main validation need
Promoter and circadian phenotyping	Established	Repeated luminescence from living plants	High-throughput time-series screening	Genotype and imaging normalization
Plant immunity / hormone imaging	Established proof-of-concept	Defence-responsive autonomous reporters	Screen receptors, elicitors and resistance mechanisms	Specificity; developmental baseline
Viral sentinel plants	Recent proof-of-concept	Virus-responsive spectral/intensity output	Earlier targeted scouting in greenhouse/nursery	Host range, field validation, false positives
Protected-crop sentinel network	Prospective translational use	Distributed engineered indicator plants + cameras	Reduce scouting area and trigger confirmatory tests	Regulatory approval, containment, camera design
Breeding and genotype screening	Near-term research use	Reporter kinetics as phenotype	Select faster/stronger defence or stress responses	Reporter introgression and background effects
Soil/water contaminant biosensing	Established mainly in bacteria	lux whole-cell response to bioavailable chemicals	Rapid biological screening of irrigation water or extracts	Matrix effects; not a replacement for regulatory chemistry
Bioluminescent ornamentals	Technically established trait class	Visible autonomous luminescence	Novel ornamental trait and public engagement	Stewardship, gene flow, ecological effects, regulation

Ecological Relevance of Natural and Engineered Bioluminescence

Light can mediate interactions between organisms, so bioluminescence belongs here in ecology. Light emission in luminescent fungi is not just a biochemical side effect. Oliveira et al. Neonothopanus gardneri demonstrated circadian regulation of bioluminescence (strongest emission timing at night, but experimental observations are also consistent with insect attraction to enhance spore dispersal [37]. Comparative genomics shows that fungal bioluminescence is linked to a highly conserved gene cluster but is also evolutionary complex among lineages of luminous mushrooms [25]. Light-emitting fungi have not been studied in relation to the ecological roles of dead wood, mulch, and humid microhabitats that support fungal communities in agroforestry landscapes relevant to decomposition and insect–fungus interactions.

Because engineered luminous plants put a permanent light source inside a photosynthetic organism, they raise an ecological question that has not been possible before. It would be a mistake to assume this is like street lighting, as engineered plant light tends to be much lower, spatially constrained, and spectrally different. Nor would it be wrong to think that the same is safe simply because the light is also biologically emitted. Ecological consequences depend on

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photon flux, wavelength, timing, and contrast against background, as well as the visual sensitivity of the exposed organisms. These variables ought to be measured directly. More broadly, ecological responses to artificial illumination depend on intensity, spectral composition, timing and the biological sensitivity of exposed organisms, rather than on light presence alone [38].

Research on artificial light at night (ALAN) establishes a framework for caution. Street-level illumination influenced nocturnal pollinator visitation and plant reproductive success in field experiments, and effects carried over into daytime plant–pollinator interactions [39,40]. The spectral characteristics of artificial lighting can also alter visual interactions between flowers, pollinators and predators, indicating that wavelength composition should be considered alongside photon intensity when evaluating ecological exposure [41,42]. They affect the interactions between flowers and pollinators as well as predators [41]. The light intensities employed in these studies were orders of magnitude different from those of all currently glowing plants, meaning that effect sizes cannot be directly transferred. It is a matter of principle: nocturnal light can alter interactions, and relevant patterning is the ecological web, not structure. Experimental and meta-analytic evidence indicates that artificial night lighting can influence behaviour, physiology, life-history traits and species interactions across taxa, although the magnitude and direction of responses vary among organisms and ecological contexts [38].

Thus, the next priority experiments in bioluminescent ornamentals should examine moths, beetles, and nocturnal bees (if present), predatory arthropods, and herbivores at more natural brightnesses. Choice tests allow quantification of attraction or avoidance; pollen-transport studies help determine if visitor composition changes; and seed-set measurements indicate whether optical novelty influences reproductive success. An ideal design would compare a luminous line with its non-luminous near-isogenic control under true darkness and common urban night skyglow. This would show whether bioluminescence contributes an ecological signal that molecular background light pollution does not.

Gene flow and trait retention constitute a second ecological axis. The exposure scenario may differ, e.g., sentinel plants containing greenhouses and fertile ornamental plants for outdoor use. A risk assessment should consider pollen movement, seed escape, the persistence of volunteers, and any introgression into compatible relatives. The orange-petunia evidence shows that ornamental germplasm can diffuse through traditional breeding networks before all stakeholders detect the genetic event [34,35]. In this Opinion, we outline a conceptual ecological framework that connects optical characterization, organismal exposure, behavioural response, and ecosystem outcome and containment (Figure 4).

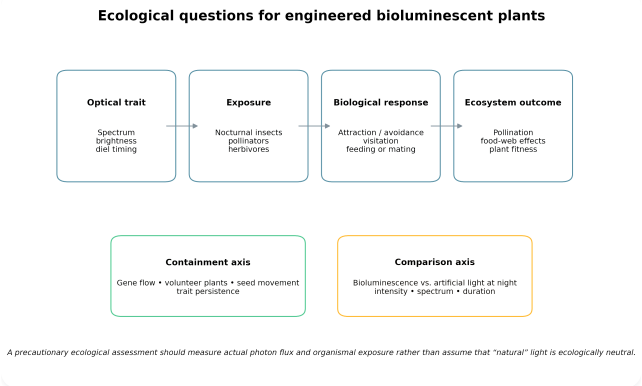


Figure 4. Ecological assessment framework for engineered bioluminescent plants. Optical traits should be linked experimentally to organismal exposure, behavior, ecosystem function and gene-flow scenarios rather than inferred from visual appearance. Author-generated original schematic.

Table 3. Priority ecological questions before outdoor deployment of engineered bioluminescent plants.

Question domain	Core question	Suggested experiment	Primary endpoints
Nocturnal arthropod response	Does plant-generated light attract or repel insects?	Choice tests with luminous vs. near-isogenic control; quantify spectrum and photon flux	Visitor abundance, residence time, orientation
Pollination service	Does the optical trait alter pollen transfer or fruit/seed set?	Semi-field cages and outdoor plots with matched background illumination	Pollen deposition, visitation network, fruit/seed set
Interaction with ALAN	Is the signal detectable against rural/urban skyglow?	Factorial bioluminescence × background light experiment	Behavioural effect size under realistic night lighting
Plant fitness / metabolism	Does pathway flux change growth or reproduction?	Multi-season comparison of luminous and control lines	Biomass, flowering, seed set, phenylpropanoid traits
Gene flow and persistence	Can the trait move into volunteers or compatible relatives?	Pollen-flow and seed-persistence studies appropriate to species	Outcrossing distance, volunteer survival, trait frequency
Sentinel containment	Can diagnostic value be retained with reduced reproductive risk?	Sterile/non-flowering lines or contained production	Sensor performance plus escape probability

Technical Limitations, Calibration and Standardization

The key question for practical agricultural applications is no longer whether plants can emit light, but whether the signal titer can be set so that the same biological state yields similar output across tissues (roots and shoots), plant lines, days, and environments. Bioluminescence is limited by the number of enzymes, substrate or precursor at a given time, oxygen and ATP in some systems (e.g., fireflies), as well as temperature and optical tissue attenuation. Because the metabolic and optical environments can differ between a leaf and a flower, the linear relationship between reporter transcription and detected intensity may not be identical at the per-organ level.

Autonomous fungal reporters address substrate-delivery variability, but they come with metabolic coupling. Caffeic acid ($C_9H_8O_4$) is a reactive molecule that functions within the cellular context of phenylpropanoid metabolism, incorporating into cell wall polymers such as lignin and pigments, as well as playing a role in defense. By diverting metabolic flux into luciferin biosynthesis, it is therefore

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possible to couple promoter activity to light output. Better-designed pathways have increased signal intensity, but very bright constitutive expression may not be the best strategy for sensing. An ideal diagnostic reporter should have a low baseline level, a large inducible change in expression after the event, and, above all, appropriate threshold kinetics and a faithful return to baseline after the event. Calvache et al. highlighted how these issues can be addressed in a recent study by reconfiguring the transcriptional entry point of the fungal pathway, expanding on the idea that, in mammalian multicistronic circuits, circuit architecture can be more important than raw pathway brightness [36].

Imaging standards are equally important. Camera model, lens and aperture, exposure time, sensor gain/ISO, working distance, optical filters, dark adaptation, temperature, and signal normalization should be standardized. Expressing imaging performance in terms of photon output allows meaningful comparison between laboratories. In plant diagnostics, detection thresholds should be validated against an independent statistical reference, such as quantitative polymerase chain reaction (qPCR), enzyme-linked immunosorbent assay (ELISA)-based pathogen load, or hormone quantification. The importance of independent validation is also recognized in genetically encoded biosensor development, where sensor specificity, dynamic range, reversibility, and interference from endogenous pathways are important determinants of practical utility [19,20].

High sensitivity without high specificity is insufficient, and diagnostic performance depends on both positive and negative predictive efficiency at the relevant disease prevalence. Standardized image acquisition and quantitative phenotyping are therefore essential for separating biological variation from variation introduced by camera systems, illumination, acquisition settings, and image-processing pipelines [6,7].

In addition, field robustness should also be evaluated throughout the crop cycle. Developmental stage and transgene silencing can affect stable expression; Pathway precursors can be affected by tissue damage, senescence, and even carbon status. The spectral attributes of leaves are related to their contents in chlorophyll and water. It may take years for, like, a sensor to remain interpretable, rather than just weeks, by the nature of perennial horticultural crops. An additional complication arises if events in the scion are being reported: a reporter in the scion may miss rootstock-localized responses unless the signal or regulatory response is systemic.

Finally, biological containment must be built into the design of data interpretation. You could minimize the risk of gene flow in a protected cultivation system by preventing flowering in the pollen parent or compensating with a sterile or non-flowering sentinel genotype. For a commercial ornamental, however, you may need to implement fertility management, event-related detection methods, and supply-chain traceability. Thus, the optimal engineering approach depends on the type of ecological exposure you wish to create.

Research Gaps and a Roadmap for Horticultural Translation

The next round of the sector must be assessed via agronomic questions, not brightness data. This shift from reporter construction toward functional validation is consistent with current plant synthetic biology, where biosensor design is increasingly organized around standardized sensing design–build–test–learn workflows [20,33]. Five research priorities stand out. Autonomous reporters should first be developed targeting validated stress signatures relevant to crops. The promise of viral proteases, pathogen effectors, and well-characterized immune pathways make them attractive; they enable molecular recognition rather than a general stress response. Second, systems should not be limited to Nicotiana and Arabidopsis, but should also extend to horticultural crops, where the expense of sensor infrastructure is offset by protected cultivation and high crop value (i.e., tomato, pepper, cucumber, strawberry, ornamentals, and clonally propagated nursery crops offer clear opportunities).

Third, optical engineering joins in plant engineering. Whether spectral-shift reporters, ratiometric signals, or multicolour autonomous systems, these frame-of-reference strategies can decouple biological change from modulation of overall brightness. We have already demonstrated autonomous multicolour bioluminescence across biological hosts, and colour can be diagnostically useful, as shown in the viral sentinel work to combat antimicrobial resistance published this year [12,36,43]. For instance, a camera in a greenhouse might sample two wavelength bands and raise an alarm only when the ratio exceeds a pre-validated threshold.

Fourth, studies must determine whether a reporter benefits management. A credible horticultural experiment could randomize greenhouse compartments to conventional scouting versus sentinel-supporting scouting, then measure time-to-detection, number of confirmatory tests, disease spread, pesticide use, crop loss, and labour. That evidence gap between beautiful synthetic biology and a plant. However, if the system catches infections two days earlier but triggers numerous false alarms, it may not, in fact, save money or input. Even if it never helps diagnose the pathogen on its own, in an area with reliable scouting resolution at a small scale, that value could be substantial.

Fifth, pre-commercial outdoor assessments should be done using proceeds from ecological assessment. For the light, brightness and spectrum need to be measured in absolute units; for behavioural tests, place-based nocturnal taxa should be used; and for gene-flow scenarios, these must correspond with the crop or ornamental species. This matters because living light is public-facing and has a lot of constituencies behind it. Transparent evidence on metabolic impact, environmental persistence and ecological exposure will outlast an appeal to the naturalness of a lookalike technology.

The next big play is integrating bioluminescent plants into digital age. Low-light acquisition, image registration, and anomaly detection can be done locally on edge cameras, with only alerts sent over rather than continuous video transmission. From there, a network of sentinel plants could function as biological nodes spread across that area. This paradigm, however, is clearest if the plant sensor evokes molecular specificity not easily offered by traditional imaging. It is less compelling when the same decision can already be made easily and cheaply from standard RGB, thermal or multispectral data. The goal of research here should therefore be to use bioluminescence as a complement to other instrumentation: use it when a process is difficult to measure with an external sensor.

Conclusion

Bioluminescence has evolved from an eye-catching natural phenomenon to a multipurpose plant research tool and, more

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recently, to a sustainable platform for in-plant crop sensors. Firefly luciferase demonstrated the importance of low-background, non-destructive reporters; bacterial lux showed that plants and microbes can produce their own light autonomously; and the fungal caffeic-acid pathway enabled self-sustained nuclear-encoded luminescence across a variety of plant tissues. The most significant recent development is functional, not aesthetic: light can now convey signaling defense and viral infection without multiple treatments with an external substrate.

In agriculture and horticulture, the immediate opportunity is not to displace established diagnostics or remote sensing. A key aim is to develop biologically relevant early-warning tools suitable for environments with existing phenotyping infrastructure, especially in protected cultivation, nurseries, and high-throughput tissues. Future studies with the strongest methodology will link reporter activation to a biologically validated independent event, then show that this information is used effectively to alter management.

Ecology needs to keep being part of that translation. Natural fungal bioluminescence shows that biological light can mediate interactions among organisms, and studies of artificial light at night corroborate that nocturnal luminescence can reshape pollination networks. Unlike regular lamps, genetically modified glowing plants are far less bright, but their ecological impact cannot be inferred from brightness alone. Aspects to be measured include frequency, timing, magnitude, behaviour and gene flow. Bioluminescence could evolve from a glowing plant to something much more consequential, a living interface between molecular plant physiology, environmental monitoring and precision horticultural decision-making if those questions are resolved in tandem with calibration and biosafety.

Declarations

Acknowledgements

Not applicable.

Author contributions

Dr. Parbin Huda Baruah and Dr. Tanmoy Das contributed to the conceptualisation, literature synthesis, critical analysis, and writing review and editing of this review. Both authors should review and approve the final version before submission.

Funding

This review received no specific external funding.

Availability of data and materials

No new datasets were generated or analyzed for this narrative review.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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