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Rice false smut (*Ustilaginoidea virens*): Advances in pathogen biology, innovative detection tools and next-generation management strategies

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Abstract

Rice false smut (RFS), caused by *Ustilaginoidea virens* (teleomorph: *Villosiclava virens*), has rapidly transitioned from a minor disease to a major global threat to rice production. The pathogen infects rice spikelets during the booting and flowering stages, forming green to black smut balls that replace grains and contaminate harvests with ustiloxins and ustilaginoidins - mycotoxins harmful to humans and livestock. Yield losses of 5–85% have been reported, with severe economic and food security implications across Asia and beyond. Advances in pathogen biology reveal effector proteins and mycotoxins that manipulate host immunity and grain development, while epidemiological studies highlight the role of warm, humid climates, nitrogen fertilization, and intensive cultivation in disease spread. Traditional diagnostics are limited, but innovative tools such as qPCR, LAMP, CRISPR-based assays, nanopore sequencing, hyperspectral imaging, and AI-driven models enable early detection and precise surveillance. Management strategies are shifting from chemical reliance toward integrated approaches combining resistant cultivars, CRISPR/Cas-mediated gene editing, host-induced gene silencing, biocontrol agents, microbiome engineering, nanopesticides, RNAi-based biopesticides and cultural practices. The convergence of genomics, precision agriculture, and climate-smart forecasting provides new opportunities for sustainable control of RFS. This review synthesizes recent advances in biology, detection and next-generation management strategies, offering a framework for mitigating yield loss, safeguarding food safety and ensuring resilient rice production under changing climate conditions.

Keywords: *Ustilaginoidea virens***,** Rice false smut**,** molecular diagnostics**,** integrated disease management**,** CRISPR and RNAi technologies

Introduction

Rice stands as a fundamental staple crop, nourishing over half of the global population and serving as a cornerstone for food security, particularly across Asian, African and Latin American continents. Its extensive cultivation spans more than 150 countries, covering diverse environments from temperate zones to sub-humid and humid climatic conditions, underscoring its adaptability and crucial role in global agricultural systems [1]. But its productivity is significantly hampered by numerous biotic stresses, including emerging diseases such as false smut [2]. False smut, caused by the ascomycetous fungus *Ustilaginoidea virens* (teleomorph: *Villosiclava virens*), has emerged as a significant threat to rice production worldwide, particularly in major rice-producing regions like India [3]. This devastating disease can lead to yield losses ranging from 5% to 85%, depending on environmental factors and varietal susceptibility, posing a substantial challenge to global food security [4].

Historically the RFS were known as "Lakshmi disease" in India as it is considered as the sign of bumper harvests, later it was reported as minor disease in Tamil Nadu, India [5]. *U. virens* is a unique hemibiotrophic fungus that infects rice spikelets during the booting to flowering stages by bypassing the plant surface defences and colonizes the floral organs, replacing grains with velvety green smut balls that later turn orange or black. These smut balls not only reduce grain yield and quality but also harbour potent mycotoxins such as ustiloxins and ustilaginoidins, which are toxic to humans and livestock by inhibiting the cell division and disrupt microtubule formation in host tissues. This disease has been now spread to over 56 rice-growing countries including China, Japan, Pakistan, Bangladesh, Vietnam and the Philippines [6]. In India alone, yield losses of up to 44% have been documented in states like Punjab and Uttar Pradesh [7]. The pathogen thrives in warm, humid conditions and is particularly aggressive in high-yielding, irrigated rice systems. Climate change, with its erratic rainfall and rising temperatures, has further expanded the geographical range and intensity of RFS outbreaks.

Recent genomic studies have revealed that *U. virens* possesses a rich repertoire of effector proteins that manipulate host immunity and hormonal pathways. Transcriptomic analyses have identified key genes involved in pathogenicity, such as UvHOG1 and UvPRO1, which regulate fungal development and stress responses [8].

The impact of RFS extends beyond yield loss. In severe outbreaks, up to 44% of spikelets may be infected, leading to significant reductions in marketable grain. The presence of smut balls and mycotoxins compromises grain quality, posing risks to food safety and international trade. In India, economic losses due to RFS have been estimated in the millions, especially in states like Punjab, Uttar Pradesh and Bihar.

Traditional visual inspection is often inadequate due to the cryptic nature of early infection. Modern tools such as PCR and qPCR assays for pathogen DNA detection [5], Loop-mediated isothermal amplification (LAMP) for rapid field diagnostics, Remote sensing and hyperspectral imaging allow large-scale monitoring of disease incidence and severity. These technologies enable early detection and targeted intervention, reducing reliance on blanket fungicide applications.

Managing RFS requires a holistic approach that integrates cultural, biological, chemical and genetic strategies. Resistant cultivars are being developed through QTL mapping and molecular breeding, though durable resistance remains elusive [8], Biocontrol agents such as *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens* have shown efficacy in suppressing *U. virens* under field conditions [9]. Fungicides, particularly triazoles and strobilurins, are effective when applied at the booting stage, though concerns about resistance and environmental impact persist [6]. CRISPR/Cas9-based gene editing is being explored to enhance host resistance by targeting susceptibility genes and silencing pathogen effector.

In light of its expanding geographic footprint, complex pathogenic mechanisms and the dual threat of yield loss and mycotoxin contamination, *Ustilaginoidea virens* has become a focal point of rice pathology research. The integration of molecular biology, genomics and precision agriculture has significantly advanced our understanding of the pathogen’s biology, enabling the development of sensitive detection tools and predictive surveillance systems. Simultaneously, the emergence of biocontrol agents, resistant cultivars and gene-editing technologies marks a paradigm shift in how Rice False Smut is managed moving from reactive chemical control to proactive, sustainable strategies. This review synthesizes these multidisciplinary advances to provide a comprehensive framework for tackling RFS, aligning with the broader goals of food safety, environmental stewardship and resilient rice production in the face of climate variability and evolving pathogen dynamics.

Pathogen Biology and Epidemiology

Taxonomy and classification of *Ustilaginoidea virens*

Ustilaginoidea virens (Cooke) Takahashi is a filamentous ascomycete fungus responsible for rice false smut, a globally significant floral disease of rice. Taxonomically, it belongs to the phylum Ascomycota, class Sordariomycetes, order Hypocreales and family Clavicipitaceae. The teleomorph (sexual stage) of the fungus is classified as *Villosiclava virens*, a designation confirmed through molecular phylogenetic analyses using ribosomal RNA and β-tubulin gene sequences. These studies resolved longstanding confusion with ergot fungi such as *Claviceps* spp., which share similar morphological traits but differ significantly in genetic lineage and pathogenic behaviour. Historically, *U. virens* was misclassified under various synonyms including *Ustilago virens*, *Tilletia oryzae* and *Claviceps virens*, due to its superficial resemblance to smut and ergot fungi [10]. However, unlike true smuts, *U. virens* does not replace the rice kernel with teliospores but instead forms false smut balls composed of chlamydospores that erupt through the palea and lemma. Morphological and cultural studies have further supported its distinct identity, with characteristic colony pigmentation, conidial morphology and sporulation patterns observed across diverse isolates [7] (Figure 1).

Recent molecular characterizations using internal transcribed spacer (ITS) regions and simple sequence repeat (SSR) markers have reinforced its taxonomic placement and revealed moderate genetic diversity among Indian isolates [7]. These findings are consistent with its classification within the Clavicipitaceae and support its inclusion in the anamorphic tribe Ustilaginoideae of Hypocreales. The availability of genomic resources and phylogenetic tools has enabled more precise identification and differentiation of *U. virens* from closely related taxa, facilitating improved diagnostics and epidemiological tracking.

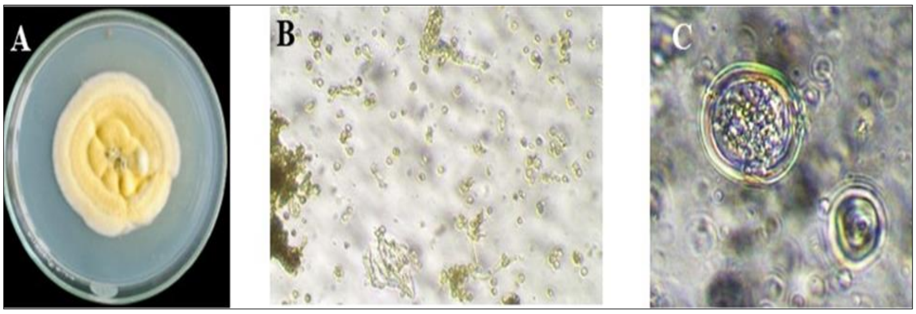

Fig 1: *Cultural and morphological characteristics of* Ustilaginoidea virens *(Cooke) Takahashi.* (A) Colony morphology on potato dextrose agar (PDA) showing creamy yellow, circular growth; (B) Single-celled conidia observed under light microscopy; (C) Thick walled, globose chlamydospore typical of mature smut structures.

Life Cycle and Infection Process

The life cycle of *Ustilaginoidea virens*, comprises both sexual and asexual stages. The pathogen survives between cropping seasons through chlamydospores and sclerotia, which act as overwintering structures in soil and crop debris [11]. During the booting stage of rice, conidia germinate and hyphae colonize the spikelets intercellularly, particularly targeting the upper stamen filaments located between the ovary and lodicules. Notably, the fungus does not penetrate host cell walls directly and does not form haustoria, confirming its biotrophic lifestyle [11]. Following successful colonization, false smut balls begin to emerge approximately 10 - 15 days post-anthesis. These structures initially appear white, then transition through yellow-green to black as chlamydospores mature on their surface. The ovary remains alive and uninfected, indicating that the pathogen manipulates host physiology without inducing cell death [12]. Toward the end of the season, sclerotia form either on or beneath the soil surface. It has been reported that both sclerotia and chlamydospores survive under laboratory field condition for almost ten months [12]. The sclerotia mature to form ascocarps under light condition after 2-to-5-month dormancy period, which release ascospores - the primary inoculum for initiating new infections in subsequent seasons (Figure 2). The ascospores are usually trapped using spore trap in the paddy fields before and after planting this indicate that sclerotia successfully overwinter and produce ascospores frequently [13]. This dual reproductive strategy enhances the pathogen's persistence and epidemiological success across diverse rice-growing environments. In addition to rice, alternative hosts such as weeds (*Digitaria marginata*, *Panicum trypheron, Echinochola crusgalli* and *Imperata cylindria*) may also be involved in the life cycle of *U. virens* with rare infections observed [14].

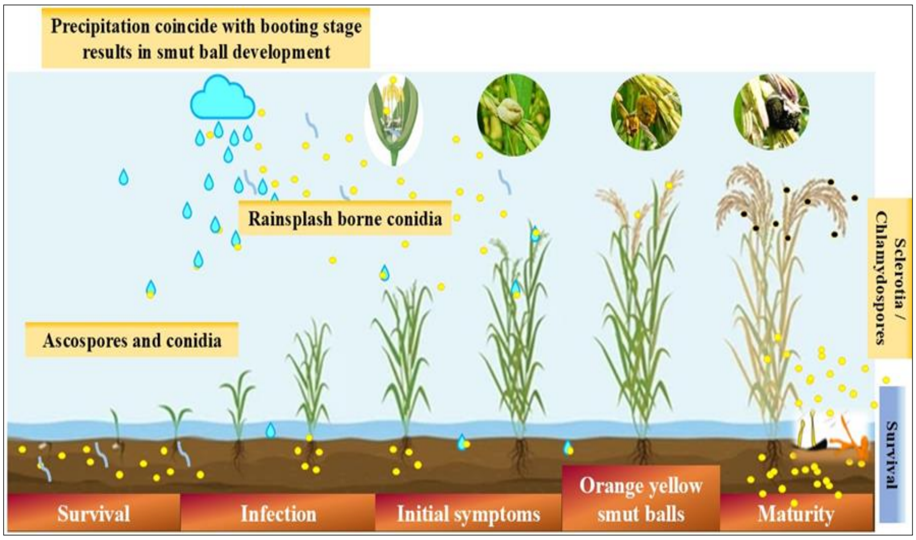

Fig 2: Schematic representation of the life cycle and epidemiology of *Ustilaginoidea virens* causing rice false smut. The pathogen survives in soil and crop residues as sclerotia and chlamydospores, which germinate to produce ascospores and conidia that infect rice spikelets during the booting stage. Rain splash-borne conidia facilitate secondary spread. Favourable environmental conditions such as high humidity and rainfall coincide with the booting stage, resulting in smut ball development. Mature smut balls release chlamydospores that return to the soil, completing the cycle.

Host-Pathogen Interaction Mechanisms

Ustilaginoidea virens, maintains a biotrophic relationship with rice by colonizing floral organs particularly stamen filaments without inducing host cell death, allowing it to exploit host resources while evading strong immune responses [15]. A central virulence strategy involves the secretion of effector proteins such as UvCBP1, a chitin-binding protein that competes with the rice receptor OsCEBiP, thereby suppressing chitin-triggered immunity including reactive oxygen species (ROS) burst, callose deposition, and defense gene expression [15]. This immune evasion facilitates fungal colonization and the formation of false smut balls mycelial masses laden with chlamydospores that replace fertilized grains and redirect host grain-filling pathways [16]. The pathogen's infection strategy is further supported by its reduced arsenal of glycoside hydrolases, enabling stealthy invasion without extensive cell wall degradation. In addition to structural manipulation, *U. virens* produces ustiloxins and ustilaginoidins, potent mycotoxins that inhibit microtubule polymerization and mitosis in eukaryotic cells, posing serious food safety risks to humans and animals [17]. These toxins accumulate in infected grains and are resistant to conventional processing, raising concerns about their persistence in the food chain. Recent genomic and proteomic analyses have revealed a compact effector repertoire and transcriptional regulation of virulence genes, offering insights into the molecular basis of pathogenicity and potential targets for host-induced gene silencing and resistance breeding [17]. Collectively, these findings underscore the sophisticated interplay between *U. virens* and rice, highlighting the pathogen's ability to subvert host immunity, redirect developmental pathways and produce toxic metabolites that compromise both yield and safety.

Environmental Factors Influencing Disease Development

Environmental conditions play a pivotal role in modulating the severity and epidemiology of rice false smut (RFS). The pathogen thrives under optimal temperatures ranging from 25–30 °C, high relative humidity exceeding 95%, and frequent rainfall during the flowering stage, which collectively enhance sporulation, conidial dispersal, and floral organ colonization [18] (Figure 3). These conditions are particularly conducive during the Rabi season in subtropical rice-growing regions, where dew formation and prolonged leaf wetness further facilitate infection. Additionally, light exposure and acidic pH have been shown to stimulate the biosynthesis of sorbicillinoids, secondary metabolites linked to fungal virulence and pigmentation. Agronomic practices also influence disease dynamics; notably, excessive nitrogen fertilization (>160 lbs/acre) correlates with increased disease incidence, likely due to enhanced canopy density and delayed flowering, which create microclimates favourable for pathogen establishment [18]. Wind speed and direction appear to have minimal correlation with disease progression, whereas temperature and humidity show strong positive associations with disease severity and area under disease progress curve (AUDPC) values [18]. These environmental and agronomic factors collectively determine the timing, intensity, and geographic spread of RFS outbreaks, underscoring the need for integrated forecasting models and calendar-based fungicidal schedules to mitigate yield losses.

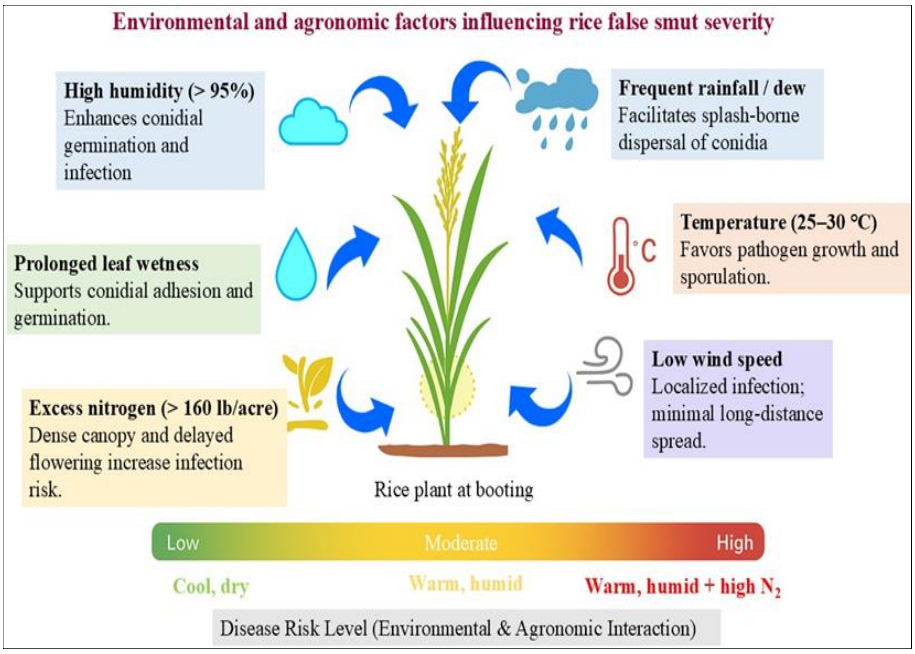

Fig 3: Environmental and agronomic factors influencing rice false smut development. High humidity, frequent rainfall, optimal temperature and excessive nitrogen application create microclimatic conditions favourable for conidial germination, infection, and smut ball formation on rice panicles. Disease risk intensifies as these factors align during the booting stage.

Geographic Distribution and Outbreak Trends

Rice false smut (RFS), has emerged as a globally significant disease, particularly in Asia's major rice-producing regions including India, China, Pakistan, Bangladesh, and Vietnam, where conducive agro-climatic conditions and intensive cultivation practices have accelerated its spread. In India, recent epidemiological surveys have revealed high disease incidence in northern states such as Uttar Pradesh, with Varanasi district reporting up to 53.64% infected tillers, while lower incidence rates (20–27%) were observed in cooler, less humid regions like Kangra (Himachal Pradesh) and Tehri Garhwal (Uttarakhand) [19] (Figure 4). The disease tends to peak during the flowering stage under warm, humid conditions, and its severity is influenced by cultivar susceptibility, with varieties like Pusa Basmati showing up to 57% yield loss due to grain replacement and smut ball formation. Molecular characterization of isolates from different Indian states has revealed high intra-population genetic diversity, suggesting rapid adaptation and local evolution, which complicates resistance breeding and disease forecasting [7]. Globally, RFS outbreaks have become more frequent and severe over the past decade, driven by climate variability, excessive nitrogen fertilization, and expansion of hybrid rice cultivation, which often lacks durable resistance. These trends underscore the need for region-specific surveillance, predictive modeling, and integrated disease management strategies to mitigate the expanding threat of *U. virens*.

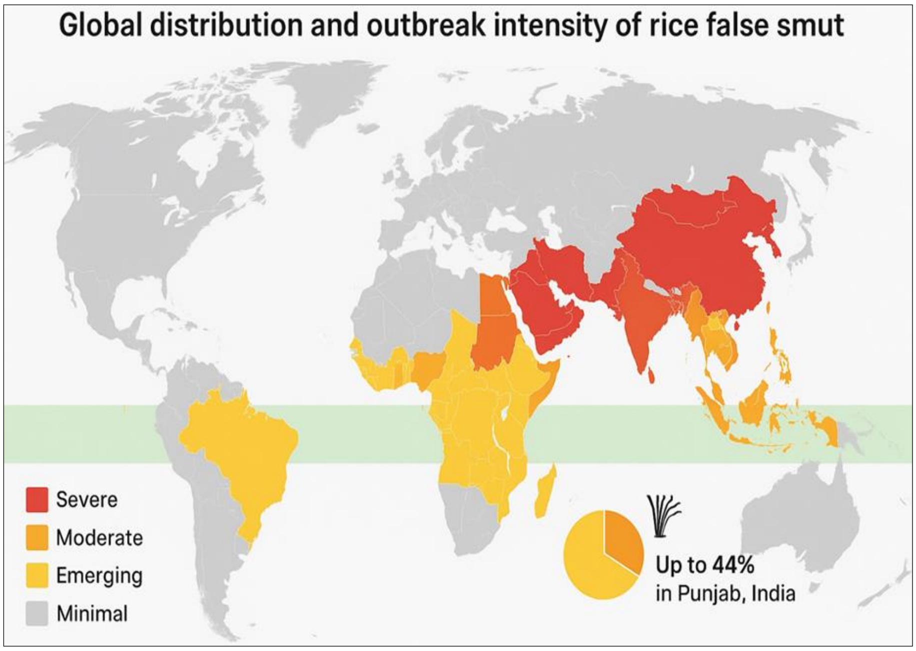

Fig 4: Global distribution of rice false smut (*Ustilaginoidea virens*) showing high prevalence in Asia and expanding occurrence in other rice-growing regions.

Innovative Detection Tools

Accurate and timely detection of *Ustilaginoidea virens*, the causal agent of rice false smut, is critical for effective disease surveillance and management. Advances in molecular diagnostics have enabled the development of sensitive, specific, and field-deployable tools that surpass conventional methods in speed and reliability (Table 1, Figure 5).

Table 1: Comparative overview of conventional, molecular and next-generation detection tools developed for early and accurate identification of *Ustilaginoidea virens*

Category	Detection Tool / Platform	Target / Principle	Key Features	Advantages	Limitations	Reference
Conventional	Microscopy & cultural isolation	Colony morphology and conidial observation	Basic laboratory identification	Simple, low-cost	Low specificity; time-consuming	[5]
Molecular (PCR)	PCR / qPCR assays	ITS region of fungal rDNA	Quantitative detection of pathogen DNA	Highly specific and sensitive; early detection	Requires lab equipment	[20]
Isothermal Amplification	LAMP (Loop-mediated isothermal amplification)	ITS gene amplification at constant temperature	Rapid field detection within 1 h	No thermal cycler needed; user-friendly	Primer design complexity	[21, 22]
Advanced Genomic	Metagenomic or nanopore sequencing	Whole-genome / direct DNA sequencing	Comprehensive profiling; strain-level ID	Culture-independent; real-time sequencing	High cost; data analysis expertise	[23, 24]
Immunological	ELISA & Lateral flow immunoassays	Detection of ustiloxins A & B	Monoclonal antibody-based quantification	Rapid, cost-effective; post-harvest screening	May cross-react; semi-quantitative	[25, 26]
Metabolite Profiling	LC–MS/MS	Ustiloxin and ustilaginoidin metabolites	Targeted metabolomics	High sensitivity and precision	Needs instrumentation	[27]
Nanotechnology Sensors	AuNP-based electrochemical biosensor; QD-based fluorescent probes	Detection of ustiloxins or pathogen DNA	Nanomaterial-enhanced signal transduction	Ultra-sensitive; portable	Still under development	[28, 29]
AI & Imaging	UAV-based hyperspectral imaging; Deep learning (CNN)	Spectral reflectance and image analysis	Remote canopy-level detection	Non-invasive, large-scale monitoring	Needs model training, high-cost sensors	[30, 31]
CRISPR-based Diagnostics	SHERLOCK / DETECTR (Cas12/13)	Pathogen-specific nucleic acids	Programmable nuclease-based fluorescence / strip read-out	Ultra-specific, rapid, field-deployable	Early research stage	[32, 33]

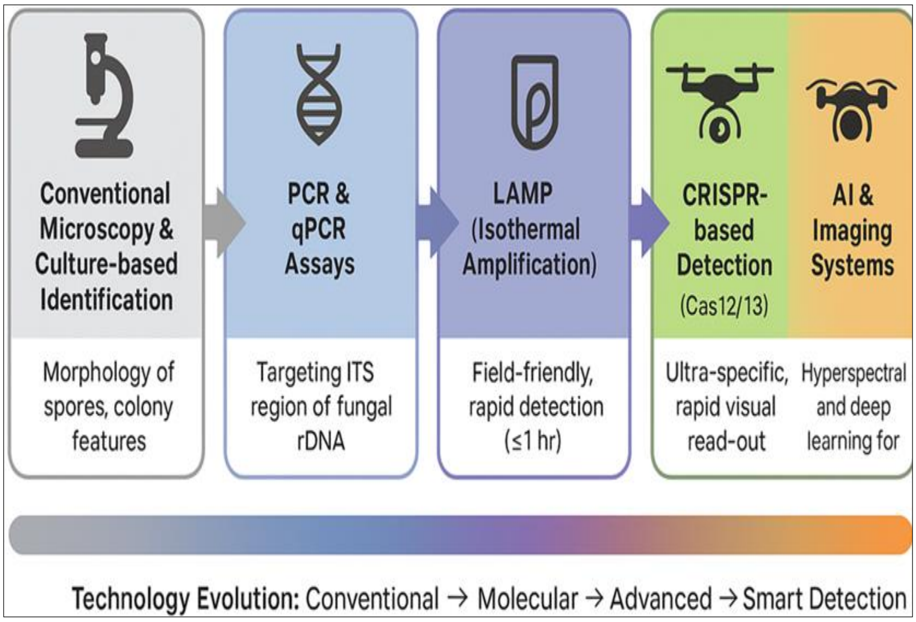

Fig 5: Overview of modern diagnostic tools for detection of *U. virens*, highlighting the transition from conventional microscopy to advanced molecular assays including qPCR, LAMP, nanopore sequencing, CRISPR-based diagnostics, and AI-powered imaging. The diagram illustrates the evolution of detection technology from conventional to molecular, advanced, and smart systems.

Conventional and Molecular Tools

Quantitative PCR (qPCR)

qPCR enables precise quantification of fungal DNA in infected plant tissues, offering high sensitivity and specificity. Likewise, [20] developed a qPCR assay targeting the internal transcribed spacer (ITS) region of *U. virens*, allowing early detection before symptom manifestation and facilitating epidemiological studies.

Loop-Mediated Isothermal Amplification (LAMP)

LAMP is a rapid, cost-effective, and field-adaptable technique that amplifies DNA under isothermal conditions. The study by [34] reported a LAMP assay for *U. virens* detection using primers targeting the ITS region, achieving results within 60 minutes without the need for thermal cyclers. Its robustness and simplicity make it ideal for on-site diagnostics in rice-growing regions.

Advanced Genomic Approaches

Recent advances in high-throughput sequencing technologies have revolutionized pathogen detection, enabling comprehensive profiling of microbial communities and real-time identification of plant pathogens, including *Ustilaginoidea virens*. These approaches offer unprecedented sensitivity, resolution, and adaptability for both laboratory and field diagnostics.

Metagenomic Sequencing

Metagenomic sequencing bypasses the need for prior culturing or targeted amplification, allowing unbiased detection of *U. virens* within complex rice-associated microbiomes. The study by [23] demonstrated the utility of shotgun metagenomics in identifying *U. virens* DNA directly from symptomatic and asymptomatic rice tissues, revealing its ecological interactions and potential co-occurrence with other microbial taxa. This approach facilitates early detection, pathogen surveillance, and insights into disease ecology.

Portable Nanopore Sequencing

Oxford Nanopore Technologies (ONT) have introduced portable, real-time sequencing platforms such as the MinION, which enable on-site pathogen diagnostics. Originally developed for rapid human pathogen detection [35] this technology has been adapted for plant pathology applications. They applied nanopore sequencing to detect *U. virens* in rice samples, achieving real-time identification with minimal sample preparation. The portability, speed, and long-read capabilities of ONT platforms make them ideal for field-based diagnostics, especially in resource-limited agricultural settings.

Immunological and Metabolite-Based Detection

Beyond nucleic acid-based diagnostics, immunological and metabolite profiling techniques offer complementary strategies for detecting *Ustilaginoidea virens*, particularly through its mycotoxins – ustiloxins which contaminate rice grains and pose risks to food safety.

ELISA and Lateral Flow Assays

Enzyme-linked immunosorbent assays (ELISA) and lateral flow immunoassays have been developed to detect ustiloxins in rice samples with high specificity and throughput. Similar study by [36] reported monoclonal antibody-based ELISA and portable lateral flow strips capable of identifying ustiloxin A and B in contaminated grains. These tools are valuable for post-harvest screening and regulatory monitoring, offering rapid, cost-effective detection without the need for complex instrumentation.

LC-MS/MS Metabolomics

Liquid chromatography-tandem mass spectrometry (LC-MS/MS) enables precise quantification and profiling of ustiloxins and related metabolites [27]. utilized targeted metabolomics to identify ustiloxins as early biomarkers of *U. virens* infection, even before visible symptoms emerged. This approach supports early warning systems and enhances understanding of pathogen-host interactions at the biochemical level.

Nanotechnology-Enabled Sensors

Nanotechnology has opened new frontiers in plant pathogen diagnostics by enabling ultra-sensitive, rapid and miniaturized detection platforms. These sensors leverage the unique physicochemical properties of nanomaterials to enhance signal transduction, specificity, and portability, making them ideal for both laboratory and field applications.

Electrochemical Biosensors with Gold Nanoparticles

Electrochemical biosensors functionalized with gold nanoparticles (AuNPs) have emerged as powerful tools for detecting ustiloxins - mycotoxins produced by *Ustilaginoidea virens* [28]. developed an AuNP-based sensor that exploits the high surface area and conductivity of gold nanostructures to amplify electrochemical signals. This platform achieved femtomolar-level sensitivity for ustiloxin A, offering a promising solution for food safety monitoring in rice supply chains. Complementing this, [26] introduced an immuno-magnetic bead-based ELISA system coupled with nanomaterial enrichment, further enhancing assay sensitivity and specificity for ustiloxin detection in complex grain matrices.

Fluorescent Quantum-Dot Probes

Quantum dots (QDs), semiconductor nanocrystals with tenable fluorescence, are being explored for DNA-based detection of *U. virens*. These probes can be conjugated with oligonucleotides targeting fungal-specific sequences, enabling real-time visualization of hybridization events. Their high photostability and multiplexing capability make them ideal for high-resolution pathogen diagnostics. Although still under development, QD-based biosensors represent a promising avenue for portable, rapid, and multiplexed detection platforms in plant pathology [29].

AI and Imaging Technologies

Artificial intelligence (AI) and advanced imaging platforms are transforming plant disease diagnostics by enabling non-invasive, scalable, and high-throughput detection of *Ustilaginoidea virens* at the field level. These technologies facilitate early identification of false smut symptoms, often before visible signs emerge, thereby supporting precision agriculture and timely intervention.

Hyperspectral Imaging and UAV-Based Surveillance

Hyperspectral imaging, when deployed via unmanned aerial vehicles (UAVs), enables detection of canopy-level spectral anomalies associated with early smut infection [30]. demonstrated that specific reflectance patterns in rice canopies particularly in the near-infrared and red-edge bands correlate with *U. virens* infection, allowing remote sensing of disease hotspots across large cultivation areas. This approach supports non-destructive, real-time monitoring and spatial mapping of disease progression.

Deep Learning for Automated Detection

Deep learning models trained on annotated UAV imagery have shown high accuracy in identifying rice false smut incidence [31]. developed a convolutional neural network (CNN) architecture capable of distinguishing infected panicles from healthy ones under variable field conditions. The model achieved robust performance across diverse lighting and canopy structures, highlighting the potential of AI-driven image analytics for automated disease surveillance and decision support.

CRISPR-Based Diagnostics

The advent of CRISPR/Cas systems has revolutionized molecular diagnostics, offering programmable, highly specific, and rapid detection platforms for plant pathogens. For *Ustilaginoidea virens*, CRISPR-based assays represent a promising frontier in field-deployable diagnostics.

CRISPR/Cas systems such as SHERLOCK (Specific High-sensitivity Enzymatic Reporter unLOCKing) and DETECTR (DNA Endonuclease Targeted CRISPR Trans Reporter) utilize Cas enzymes Cas13 and Cas12 respectively to recognize pathogen-specific nucleic acid sequences and trigger collateral cleavage of reporter molecules [33]. proposed the application of these platforms for *U. virens* detection, highlighting their potential for ultra-sensitive, equipment-free diagnostics in rice fields. These systems can be integrated with lateral flow strips or fluorescence-based readouts, enabling visual confirmation within 30–60 minutes and eliminating the need for thermal cyclers or complex laboratory infrastructure.

The programmability of CRISPR diagnostics allows rapid adaptation to emerging strains and multiplexed detection of co-infecting pathogens, making them ideal for integrated disease surveillance. Although still in early development for plant pathology, CRISPR-based tools are poised to complement existing qPCR and LAMP assays with enhanced specificity and field utility.

Next-Generation Management Strategies

Host Resistance

Host resistance remains a cornerstone of sustainable disease management and recent advances in genomics and biotechnology have enabled precision breeding strategies against *Ustilaginoidea virens*. These approaches aim to either enhance innate resistance or suppress pathogen virulence through targeted genetic interventions.

QTL Mapping

Quantitative trait loci (QTL) mapping has facilitated the identification of genomic regions associated with partial resistance to rice false smut [37]. mapped resistance loci in diverse rice germplasm, providing molecular markers for marker-assisted selection (MAS). These loci offer a foundation for breeding programs aimed at pyramiding resistance traits without compromising agronomic performance.

CRISPR/Cas9 Genome Editing

Genome editing using CRISPR/Cas9 has emerged as a powerful tool to engineer resistance by targeting host susceptibility (S) genes [38]. demonstrated that editing rice S genes involved in fungal accommodation can significantly reduce disease incidence. They further refined this approach by multiplexing guide RNAs to simultaneously edit multiple loci, accelerating the development of resistant rice lines with minimal off-target effects.

RNAi-Based Host-Induced Gene Silencing (HIGS)

Host-induced gene silencing (HIGS) leverages RNA interference (RNAi) to suppress pathogen virulence genes from within the host plant [39]. successfully applied HIGS in model systems to silence *U. virens* effector genes, resulting in reduced fungal colonization and symptom severity. This strategy exemplifies trans-kingdom RNAi and offers a non-transgenic route for resistance deployment via spray-induced gene silencing (SIGS) or transgenic expression.

Biological Control and Microbiome Engineering

Biological control strategies and microbiome engineering offer eco-friendly, sustainable alternatives to chemical fungicides for managing *Ustilaginoidea virens.* These approaches harness beneficial microorganisms to suppress pathogen activity and enhance host immunity, aligning with integrated disease management principles.

Antagonistic Microorganisms

Several antagonistic microbes have demonstrated efficacy in inhibiting *U. virens* growth through mechanisms such as competition, antibiosis, and induced systemic resistance. Notably, *Trichoderma spp., Bacillus subtilis* and *Pseudomonas fluorescens* have shown strong antagonistic activity *in vitro* and in field trials [40]. reported that these biocontrol agents significantly reduced false smut incidence when applied as seed treatments or foliar sprays, highlighting their potential for scalable deployment in rice cultivation systems.

Microbiome Engineering with Endophytes

Endophytic fungi and bacteria residing within rice tissues play a pivotal role in modulating host immunity and stress responses [41]. explored the functional diversity of rice endophytes and their interactions with *U. virens*, identifying microbial taxa capable of priming defense pathways and suppressing pathogen colonization. These findings pave the way for designing synthetic microbial communities (SynComs) tailored to enhance disease resistance, nutrient uptake and overall plant health. Microbiome engineering thus represents a frontier in precision agriculture, integrating host genetics with microbial ecology [42].

Chemical Innovations

Chemical control remains a key component of rice false smut management, but emerging innovations are reshaping its sustainability, precision and environmental footprint. Traditional fungicides are being complemented by nanotechnology and RNA-based biopesticides, offering new avenues for targeted and resistance-aware interventions.

Conventional Fungicides

Triazoles and strobilurins are widely used fungicides for managing *Ustilaginoidea virens*. These compounds inhibit ergosterol biosynthesis and mitochondrial respiration, respectively and have shown efficacy in reducing disease incidence when applied at booting or early heading stages. However, prolonged use has led to concerns over resistance development and environmental persistence [43]. reported declining sensitivity of *U. virens* isolates to triazole-based treatments, underscoring the need for rotation strategies and alternative chemistries.

Nanopesticides

Nanotechnology has enabled the development of nanopesticides, where active ingredients are encapsulated within nanocarriers such as liposomes, polymers or silica matrices [44]. demonstrated that nanoformulated fungicides enhance delivery efficiency, reduce required dosages and prolong field persistence through controlled release mechanisms. These formulations also improve adhesion to plant surfaces and reduce off-target effects, aligning with precision agriculture goals.

RNA-Based Biopesticides

RNA interference (RNAi) has emerged as a novel strategy for silencing pathogen virulence genes. Sprayable double-stranded RNA (dsRNA) formulations can be applied directly to rice plants, triggering gene silencing in *U. virens* upon uptake [45]. pioneered this approach in plant pathology, showing that topical RNAi can suppress fungal infection without genetic modification of the host. This technology offers a non-transgenic, environmentally benign alternative to conventional fungicides and is currently under development for field-scale deployment.

Cultural and Ecological Management

Cultural practices and ecological interventions play a pivotal role in reducing the inoculum pressure and environmental conduciveness for *Ustilaginoidea virens* infection. These strategies are cost-effective, scalable, and compatible with integrated disease management frameworks.

Balanced Nitrogen Management:

Excessive nitrogen fertilization has been linked to increased susceptibility to rice false smut. The study by [46] demonstrated that optimizing nitrogen input particularly during panicle initiation significantly reduces disease incidence by minimizing lush canopy growth that favours pathogen establishment. Complementary findings by [47] suggest that integrating nitrogen data into predictive models further enhances disease management by aligning fertilization schedules with pathogen risk profiles.

Straw Residue Decomposition and Crop Rotation

Post-harvest straw residue serves as a reservoir for *U. virens* sclerotia, which overwinter and initiate primary infection. Accelerated decomposition through microbial inoculants or mechanical incorporation, combined with crop rotation (e.g., with legumes or non-host cereals), effectively lowers inoculum load and disrupts the pathogen life cycle [48]. reported that strategic residue management and rotational cropping significantly reduce sclerotial survival and subsequent infection rates, reinforcing the ecological value of these interventions.

Precision Agriculture & Forecasting

Emerging technologies in precision agriculture are enhancing the predictive capacity and spatial resolution of rice false smut management.

AI-Driven Disease Prediction Models

Advancements in machine learning have enabled the integration of UAV imagery, nitrogen application data, and meteorological variables to forecast false smut outbreaks in rice. Such models support proactive management through spatially targeted fungicide applications, reducing excessive chemical use. Building on this foundation, later studies combined artificial intelligence with IoT sensor networks to improve predictive modeling and automate decision-support systems for rice disease surveillance [49].

Climate-Smart Forecasting Tools

Advanced forecasting platforms now incorporate climate change scenarios to model future risks of rice false smut under shifting temperature and precipitation regimes [50]. constructed climate-resilient models that simulate pathogen dynamics under warming trends, supporting long-term planning for cultivar selection, sowing schedules and regional disease preparedness.

Future Perspectives

Future management of *Ustilaginoidea virens* must embrace a convergence of cutting-edge biotechnologies, ecological insights, and predictive analytics to achieve sustainable control under intensifying cultivation and climate variability. Integrative multi-omics approaches encompassing genomics, transcriptomics, proteomics and metabolomics will be instrumental in decoding the molecular dialogue between rice and the pathogen, enabling precise identification of virulence effectors and host resistance genes. These discoveries can accelerate resistance breeding and inform RNAi target selection. On the diagnostic front, portable platforms that combine CRISPR-based systems (e.g., SHERLOCK, DETECTR) with nanopore sequencing offer real-time, field-deployable solutions for strain-level detection without reliance on laboratory infrastructure [32]. Such tools promise rapid, on-site decision-making and early warning capabilities. In parallel, RNAi-based biopesticides particularly sprayable double-stranded RNA (dsRNA) formulations are emerging as environmentally friendly alternatives to chemical fungicides [45]. demonstrated that clay nanosheet carriers can deliver dsRNA effectively, silencing pathogen genes and offering sustained protection without genetic modification of the host. Another promising frontier is microbiome engineering, where synthetic microbial communities (SynComs) tailored to rice floral tissues could enhance immunity and suppress pathogen colonization [41]. These communities may be designed to prime defense pathways, compete with *U. virens*, and improve overall plant resilience. Finally, AI-driven forecasting models that integrate UAV imagery, nitrogen application data, and weather variables are revolutionizing precision disease management. When coupled with climate-smart forecasting tools that simulate future disease risks under warming scenarios, these platforms enable proactive, spatially targeted interventions and long-term planning. Together, these innovations chart a path toward holistic, adaptive and sustainable management of rice false smut in the face of evolving agricultural and environmental challenges.

Conclusion

Rice false smut has transitioned from a minor sporadic disease to a major threat under intensive cultivation and climate variability. Significant progress in detection from qPCR and LAMP to nanopore sequencing and AI-driven imaging along with innovative management tools such as CRISPR diagnostics, RNAi-based biopesticides and microbiome engineering, offers hope for sustainable control. A coordinated global effort integrating biotechnology, precision agriculture, and eco-friendly practices will be essential to safeguard rice productivity and food safety in the decades ahead

CRediT authorship contribution statement

A.G. Meena: Writing – original draft, Resources, Methodology, Conceptualization. S. Bharathi: Supervision, Methodology, Conceptualization.: Supervision, Methodology, Conceptualization. N. Shiva: Writing – review & editing, Project administration, Conceptualization. K. Selvam: Writing – review & editing, Supervision, Conceptualization. A. Yogapriya: Visualization, Supervision, Project administration, Methodology. Ragul S: Resource support, Supervision and administration.

Declaration of competing interest

I hereby declare that the manuscript entitled "Rice False Smut (*Ustilaginoidea virens*): Advances in Pathogen Biology, Innovative Detection Tools and Next-Generation Management Strategies" is original and has not been published previously. It is not under consideration for publication elsewhere, in whole or in part. All authors have read and approved the manuscript, and the submission is on behalf of all the authors. We also confirm that all appropriate ethical standards and guidelines have been followed in the preparation of this article.

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