

Safety risk assessment of edible fungi

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Abstract: Edible fungi are a typical type of food microorganism, hold significant nutritive value and are considered valuable resources. However, ensuring the safety of these fungi is a crucial concern that must not be underestimated, encompassing both food safety and human health. Tackling this issue is paramount to safeguarding consumer well-being and upholding the integrity of the edible fungi industry. The safety risk assessment of edible fungi encompasses numerous facets, and a variety of technical approaches are applied to these areas. For instance, Convolutional Neural Networks and Spectroscopy technology have been utilized to identify the species of edible fungi to ascertain their suitability for human consumption. Bioinformatics methods, such as transcriptome data mining and gene sequencing, have been employed to identify toxic substances and pathogenic microorganisms present in edible fungi, thereby mitigating the risk of food poisoning. Atomic absorption spectroscopy has been employed to assess the heavy metal content in edible fungi, effectively preventing excessive intake of heavy metals by the human body. This paper offers a comprehensive review of the research advancements in safety risk assessment of edible fungi, covering various facets such as edible fungi species identification, toxicity assessment, microbiological safety evaluation, and heavy metal detection. It offers valuable insights for evaluating safety issues related to edible fungi and furnishes theoretical underpinning for mitigating risks of edible fungi poisoning and other safety concerns.

1. Introduction

Edible fungi, also called edible mushrooms, are a valuable category of fungi that offer significant intrinsic value and potential as a natural food source. Celebrated for their delightful flavors and exceptional nutritional content, edible fungi are highly regarded for their rich reserves of high-quality protein, carbohydrates, minerals, vitamins, and other essential nutrients. Particularly notable for their high protein content and low levels of sugar, cholesterol, and fats, edible fungi emerge as an excellent dietary option [1]. Edible fungi have demonstrated efficacy in managing a spectrum of health conditions, encompassing diabetes, obesity, cancer, and cardiovascular diseases. Their therapeutic attributes play a pivotal role in enhancing holistic human wellness [2, 3]. Food, medicine, agriculture and the environment are all areas where edible fungi are widely used (Figure 1).

Edible fungi are cherished for their exquisite flavors and varied nutritional compositions. However, the safety of edible fungi raises considerable concerns regarding food safety and human health, necessitating diligent

oversight. It is widely recognized that specific types of edible fungi contain harmful compounds. Furthermore, the cultivation and storage environments of edible fungi may introduce pathogenic microorganisms or heavy metals (HMs). It illustrates the pivotal factors that impact the safety of edible fungi in Figure 2. Therefore, it is crucial to conduct safety risk evaluations for wild edible fungi.

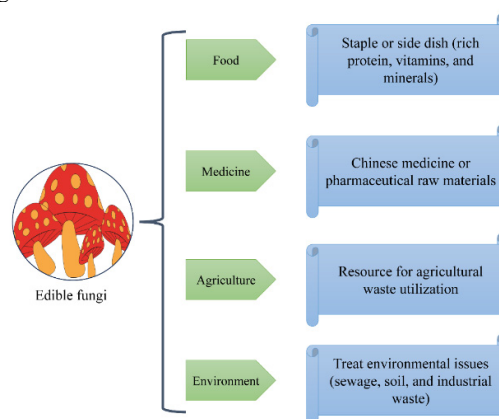


Figure 1. Application fields of edible fungi.

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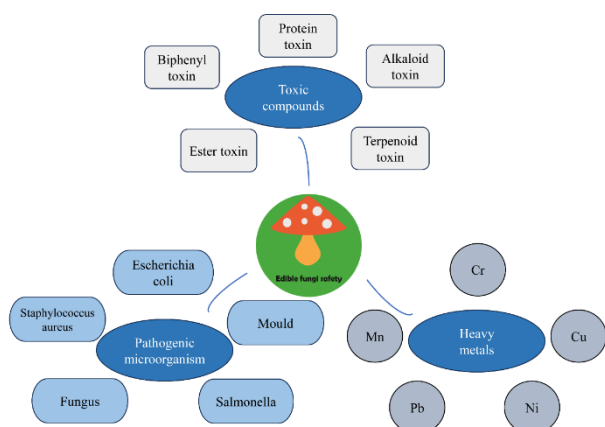


Figure 2. Important factors affecting the safety of edible fungi.

2. Identification of edible fungi species

2.1. Convolutional Neural Network (CNN)

Traditional classification methods often rely on specialized expertise and experience, which may introduce subjectivity and errors. Therefore, leveraging technological tools becomes essential for the accurate classification of edible fungi. One such technique, CNN, has demonstrated remarkable success in image classification tasks [4]. When applied to edible fungi classification, CNN can autonomously extract image features and achieve precise categorization through the analysis of extensive edible fungi image datasets. Zahan et al. employed CNNs to classify mushrooms into edible, inedible, and poisonous categories [5]. Ketwongsa et al. utilized AlexNet, a CNN, to distinguish between toxic and edible fungi among the 5 most frequently found types in Thailand [6]. Similarly, Gupta et al. used CNNs to rank the edibility of mushrooms in the *Agaricus* and *Lepiota* families [7]. Zhang et al. also developed a classifier based on deep CNNs to classify toxic and edible fungi discovered in China from mushroom images [8]. Recently, Xu et al. enhanced the existing CNN model ShuffleNetV2, developing the ShuffleNetV2-Lite+SE model. This model strikes a delicate balance between functionality, parameter quantity, and real-time capability, making it ideal for use on resource-limited devices like portable gadgets. This advancement facilitates instantaneous and accurate identification of edible fungi toxicity [9].

This approach automates the classification of edible fungi species by creating a database comprising a vast array of edible fungi images. This database aids individuals in determining the toxicity and edibility of fungi. Using CNNs for identifying edible fungi species has emerged as a novel technology theory in recent years. However, the current knowledge framework necessitates a substantial amount of labeled data for training CNNs, especially in complex classification scenarios. Therefore, we believe that the future trend of CNN development will focus more on reducing the reliance on labeled data by leveraging innovative learning methods and technologies to enhance model performance and generalization capabilities.

2.2. Spectroscopy technique

There is increasing interest in the possible applications of spectroscopy techniques in the food industry due to their ability to provide rapid analysis, real-time monitoring, minimal or no sample preparation, and non-destructive measurements. Among the vibrational spectroscopic methods commonly used in the food sector, infrared (IR) spectroscopy stands out. Esteves et al. conducted a study using Attenuated Total Reflectance Fourier Transform Infrared (FTIR-ATR) Spectroscopy to examine 64 mushroom samples from different genera, which included hallucinogenic, poisonous, and edible species. The acquired IR spectra were analysed using Orthogonal Partial Least Squares Discriminant Analysis (OPLS-DA) with SIMCA chemometric software, allowing the accurate identification of the fundamental molecule vibration of the fungal components [10]. On the flip side, the differentiation analysis of mushrooms has extensively utilized the combination of near-infrared (NIR) spectroscopy and advanced chemometric methodologies, leading to the development of several effective NIR-based analytical approaches [11]. For example, in a study by Chen et al, Fourier transform near-infrared (FT-NIR) spectroscopy was coupled with data-centric versions of Soft Independent Modelling of Class Analogy (DD-SIMCA) and Random Forests (RF) models to discriminate the species and the edibility of wild bolete mushrooms. The resulting models showed promising ability to discriminate species and edibles of bolete mushrooms [12].

Currently, spectroscopy techniques are extensively utilized in the realm of food analysis. Nonetheless, the processing and interpretation of spectral data demand specialized expertise and experience. Furthermore, spectral data can be susceptible to environmental factors such as light, humidity, temperature, among others, potentially impacting the accuracy of the data. Therefore, the handling of spectral data is evolving towards the development of robust models that are resilient to environmental variations. Future advancements will involve the integration of techniques that can mitigate the impact of environmental factors on spectral data accuracy, thereby enhancing the reliability and applicability of spectroscopy techniques in the identification of edible fungi species.

3. Toxicity assessment

Numerous proteins and peptides discovered in edible fungi exhibit cytotoxic properties, targeting specific entities. Moreover, fungal viruses are prevalent among various fungal groups. Understanding the abundance and distribution of these toxins in edible fungi is essential, as they possess the potential to produce a range of human cell cytotoxic effects. In a study by Landi et al., two novel ribotoxin-like proteins (RL-Ps) were isolated from *Boletus edulis* fruit bodies, commonly known as porcini mushrooms. One of these proteins displayed significant resistance to proteolysis, with its toxicity being eliminated only after exposure to heat treatment (90°C) with

subsequent proteolysis. These findings underscore the presence of toxins in porcini mushrooms and underscore the significance of thorough cooking to ensure detoxification prior to consumption [13]. Numerous fungal viruses have been successfully detected and characterized in edible fungi through the application of advanced deep sequencing techniques. For example, Deakin et al. used RNA profiling to identify 18 RNA virus and 8 non-host RNAs (ORFans) in the fruit bodies of *Agaricus bisporus* [14]. Additionally, the adoption of an in silico analysis approach, involving transcriptome data-mining, has emerged as a popular method for fungal virus discovery. Gilbert et al. developed a data mining pipeline that interrogates publicly available fungal transcriptome datasets at the National Center for Biotechnology Information (NCBI) to identify mycoviruses [15]. This strategy provides a robust and dependable means to detect novel viral type from the raw sequencing data. While bioinformatics methods offer high sensitivity and resolution in fungal virus identification within edible fungi, challenges related to cost and operational complexity persist. In summary, the emerging trend is towards the simplification and cost-effectiveness of bioinformatics tools for fungal virus identification in edible fungi. Future developments will focus on streamlining workflows, reducing operational complexities, and optimizing cost-efficient solutions.

4. Microbiological safety assessment

In the last few decades, the ingestion of raw edible mushrooms has emerged as a significant contributor to foodborne illnesses. This trend is primarily linked to the specific growing conditions that foster the proliferation of pathogenic bacteria. Fresh mushrooms undergo a series of stages before reaching the consumer, encompassing activities such as picking, sorting, cleaning, chopping, packaging, storing and transporting (Figure 3), all of which can potentially facilitate the growth of harmful microorganisms. In recent years, instances of severe infections, hospitalizations, and fatalities have been attributed to microbial contamination of mushrooms. Consequently, it is imperative to identify and address the presence of pathogenic micro-organisms in edible fungi to safeguard public health.

In the last few years, considerable research effort has been devoted to investigating the microbiological safety of edible mushrooms. In a study conducted by Li et al., it was found that 15 out of 104 samples of oyster mushrooms from conventional and supermarket retail outlets in China tested positive for *Cronobacter* spp. This discovery underscores the prevalence of *Cronobacter* spp. in oyster mushrooms available in these markets, underscoring the essentiality of stringent food safety protocols to mitigate contamination risks and uphold consumer well-being [16]. The objective of a study conducted by Meng et al. was to assess the safety and microbiological quality of four distinct edible fungi available in the Korean market. The study also investigated the prevalence of *Listeria* spp., particularly *Listeria monocytogenes*. Results revealed that 16% of the

samples analyzed tested positive for *Listeria* spp. [17]. These results underline the value of continuous monitoring of the microbiological safety of edible fungi and provide important data for risk assessment. However, the current accuracy, speed, and level of automation in detecting pathogenic microorganisms in edible fungi need to be improved. The future trend is to employ more advanced technologies and methods to enhance the sensitivity, specificity, and efficiency of pathogen detection and identification, ultimately improving food safety and quality control in the cultivation and consumption of edible fungi.

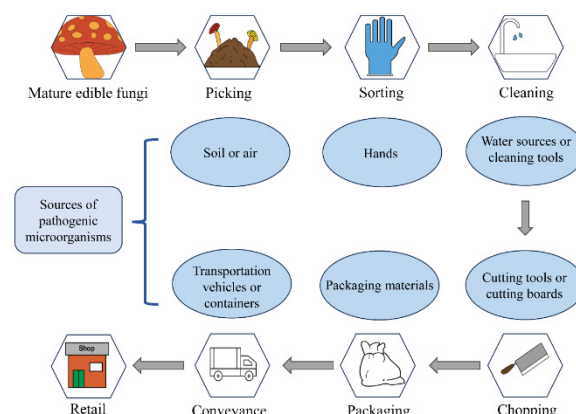


Figure 3. The process of edible fungi reaching consumers.

5. Heavy metal detection

Metals such as Cr, Mn, Cu, Ni, Pb, Hg, Cd, Fe, and Co are widely recognized as highly hazardous pollutants. Numerous studies have been dedicated to investigating the capacity of fungal fruiting bodies to accumulate these metals. A study conducted on Nigerian mushrooms utilizing atomic absorption spectrometry (AAS) revealed that the examined metals, including Cd, Cr, Ca, Cu, Co, Fe, K, Ni, Mn, Pb, Na, and Mg, did not bioaccumulate in the mushrooms [18]. Additionally, in a study by Soceanu et al., the metal content of five distinct species of edible fungi sourced from the Romanian was compared. Metal contents were measured using graphite atomic absorption spectrometry (GTAAS), and it was observed that the content of poisonous metals in all samples were notably low [19]. However, the safety of HMs in various other wild, edible fungi available in the market remains to be evaluated to ensure their consumption poses no health risks. In a study employing AAS, researchers examined the concentrations of six HMs (Cd, Cu, Cr, Zn, Fe, and Mn) in two wild oyster mushroom species (*Pleurotus djamor* and *P. ostreatus*) collected from India's Rajaji National Park in Haridwar. The findings indicated that *P. ostreatus* contained higher levels of these elements compared to *P. djamor* [20]. In a study by Wang et al., a hierarchical identification system utilizing FT-NIR spectroscopy was proposed to assess the safety of consuming porcini mushrooms in the Yunnan region. The research revealed that *Leccinum rugosiceps* from the Midu area exceeded the recommended limits for Cd concentration [21].

The analytical methods of AAS and FT-NIR mentioned above are commonly used to determine the heavy metal content in edible fungi. While AAS offers high sensitivity and precision, it can only analyze a single element at a time. On the other hand, FT-NIR can simultaneously analyze multiple components but with lower sensitivity. Therefore, the selection of an appropriate analytical method should be based on specific analytical requirements and experimental conditions. In summary, the emerging trend in the analysis of heavy metals in edible fungi is towards the development of hybrid analytical approaches that leverage the strengths of both AAS and FT-NIR. This trend aims to combine the high sensitivity and precision of AAS with the capability of simultaneous multi-component analysis provided by FT-NIR. By integrating these techniques, researchers can enhance the efficiency and accuracy of heavy metal analysis in edible fungi, leading to improved food safety and quality control measures in the cultivation and consumption of edible fungi. Assessing the risks associated with HMs in edible fungi is crucial for ensuring food safety, safeguarding human health, and protecting the environment. Such assessments also aid in formulating effective management strategies and monitoring protocols to mitigate the potential risks of HM contamination to individuals.

6. Conclusions and future prospects

This review introduces the methods and technologies utilized to ensure the safety of edible fungi, encompassing species identification, toxicity assessment, microbiological safety evaluation, and HM detection. It suggests that risk assessment tools can be employed for efficient identification of edible fungi types and detection of toxic compounds. Furthermore, it proposes evaluating the safety by identifying pathogenic microorganisms and heavy metal elements present within these edible fungi. Through multiple application examples, it is evident that food safety policies, regulatory practices, and industry standards related to edible fungi need to be detailed and developed based on these aspects to establish comprehensive safety standards for edible fungi. For instance, implementing stringent toxicity assessment protocols for edible fungi and establishing comprehensive standards for HM content in edible fungi. This review enhances our understanding of edible fungi safety while providing valuable research insights to promote advancements in risk assessment studies.

This review highlights the significant efforts that researchers have dedicated to studying the safety risks associated with edible fungi, resulting in notable advancements. However, many methods and techniques have limitations and imperfections. For example, the analysis of unique compounds in edible fungi using large instruments can be highly complex, restricting its practical application in this field. Future scientific research should focus on exploring more suitable methods to assess and ensure the safety of edible fungi from diverse perspectives to address current limitations. To proactively address potential challenges, it is crucial to

carefully select the most appropriate research methods for investigating edible fungi, considering their specific characteristics and requirements. By leveraging the strengths of each technology, a comprehensive and effective approach to edible fungus research can be developed, while minimizing any adverse outcomes.

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