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CAMPUS DE ILHA SOLTEIRA

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Triazole-Resistance in *Aspergillus fumigatus* in Brazilian Dry Food: Focus on Yerba Mate, Green Coffee, and Black Pepper

Ilha Solteira - SP
2025

PROGRAMA DE PÓS-GRADUAÇÃO EM AGRONOMIA

MARIA CLARA SHIROMA BURI

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Dissertação apresentada à Universidade Estadual Paulista (UNESP), Faculdade de Engenharia de Ilha Solteira, Ilha Solteira – SP, como parte dos requisitos para obtenção do título de Mestra em Agronomia (Sistemas de Produção)

Orientador: Prof. Dr. Paulo Cezar Ceresini

Coorientadora: Profª. Drª. Katherin Castro-Rios

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POTENTIAL IMPACT OF THIS RESEARCH

The expected impact on society should be written succinctly, considering the following aspects: the scientific, technical, social, innovative, economic, educational and cultural potential; internationalization; local, regional and national insertion; sustainable development, knowledge and research themes.

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RESUMO

Aspergillus fumigatus é um fungo sapróñto e o principal agente causal da aspergilose, uma doença que geralmente se desenvolve após a inalação de esporos. Embora o ambiente seja reconhecido como a principal fonte de exposição, produtos alimentícios secos podem atuar como reservatórios secundários, pois os esporos podem se aerosolizar durante o manuseio. Este estudo teve como objetivo determinar a prevalência e a sensibilidade aos triazois em *A. fumigatus* de alimentos secos comuns no Brasil, a fim de avaliar seu potencial papel na exposição humana. Analisamos 25 amostras comerciais de erva-mate, pimenta do reino e café verde, e isolamos *A. fumigatus* em 68% dos produtos, obtendo 42 isolados. Todos os isolados apresentaram valores de EC_{50} abaixo dos limites clínicos de resistência para tebuconazol (0,04–0,70 µg/mL), itraconazol (0,06–0,59 µg/mL) e voriconazol (0,08–0,15 µg/mL). O sequenciamento do gene CYP51A revelou apenas o polimorfismo M172V, que não está associado à resistência. A genotipagem por microssatélites revelou alta diversidade genética, com 33 genótipos multilocus identificados entre os 42 isolados. A rede de mínima expansão mostrou alta conectividade e estrutura populacional limitada, sugerindo fontes ambientais compartilhadas e troca genética contínua. A elevada prevalência e a substancial diversidade genética *de A. fumigatus* nesses alimentos secos confirmam que eles atuam como carreadores de esporos de origem ambiental. Esses achados ressaltam a importância de uma vigilância integrada em *One Health* para detectar mudanças nas populações de *A. fumigatus* e antecipar riscos emergentes na interface entre agricultura, segurança alimentar e saúde pública.

Palavras-chave: *Aspergillus; one health*; alimentos secos; resistência a azóis; diversidade genética.

ABSTRACT

Aspergillus fumigatus is a ubiquitous saprotrophic fungus and the primary etiological agent of aspergillosis, a disease that typically arises following inhalation of airborne spores. Although the environment is recognized as the main source of exposure, dry food products may function as secondary reservoirs, as spores can become aerosolized during handling. This study aimed to determine the prevalence and triazole susceptibility of *A. fumigatus* in common Brazilian dry foods to evaluate their potential contribution to human exposure. We analyzed 25 commercial samples of yerba mate, black pepper, and green coffee, and successfully isolated *A. fumigatus* from 68% of them, yielding 42 isolates. All isolates exhibited EC_{50} values below clinical resistance thresholds for tebuconazole (0.04–0.70 μg/mL), itraconazole (0.06–0.59 μg/mL), and voriconazole (0.08–0.15 μg/mL). CYP51A sequencing revealed only the M172V polymorphism, which is not associated with azole resistance. Microsatellite genotyping uncovered high genetic diversity, identifying 33 multilocus genotypes among the 42 isolates. A minimum-spanning network showed high connectivity and limited population structure, suggesting shared environmental sources and ongoing genetic exchange. The high prevalence and substantial genetic diversity of *A. fumigatus* in these dry foods confirm their role as carriers of environmentally derived spores. These findings underscore the importance of integrated *One Health* surveillance to detect shifts in *A. fumigatus* populations and anticipate emerging risks at the intersection of agriculture, food safety, and public health.

Keywords*: Aspergillus; one health;* dry food; azole resistance; genetic diversity.

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LIST OF ABREVIATIONS AND ACRONYMS

IA	Invasive Aspergillosis
PDA	Potato Dextrose Agar
DNA	Deoxyribonucleic Acid
PCR	Polymerase Chain Reaction
EC_{50}	Effective Concentration Required to Inhibit 50% of Fungal Growth
STRs	Short Tandem Repeats
M13F	Primer M13 Forward
FAM	Fluorescein Amidite
TET	Tetrachloro-fluorescein
Cy3	Cyanine 3
Cy5	Cyanine 5
MLGs	Unique Multilocus Genotypes
MSN	Minimum Spanning Network
EUCAST	European Committee on Antimicrobial Susceptibility Testing
MIC	Minimum Inhibitory Concentration
ATU	Area of Technical Uncertainty
CPA	Chronic Pulmonary Aspergillosis
IPA	Invasive Pulmonary Aspergillosis
NWT	Non-Wild-Type
AR*Af*	Azole-Resistant *Aspergillus fumigatus*

SUMMARY

1. INTRODUCTION

Deeply rooted in history, culture, and culinary traditions, the spices, teas, and coffee hold enduring significance (Mutalib et al., 2022). Among them, green coffee (*Coffea*) black pepper (*Piper nigrum L.)* and yerba smate (*Ilex paraguariensis)* have become increasingly prominent in consumer's daily lives (García-Cordero et al., 2023; Song et al., 2025). These products are valued not only for their distinctive flavors and sensory properties but also for their various health benefits, such as antioxidant activity, antiinflammatory properties, and metabolic support (Dellalibera; Lemaire; Lafay, 2006; Santiano et al., 2023). Amid a growing interest in healthier lifestyles, the demand for natural and functional foods has driven the consumption of these products. Furthermore, green coffee, black pepper, and yerba mate hold significant socioeconomic relevance in Brazil, serving as important sources of income for thousands of families and playing a key role in the national agribusiness sector (Costa; Pauletto, 2021; Maguire-Rajpaul et al., 2020).

The black pepper (*Piper nigrum* L.) is a species of a climbing vine native to Asia whose fruit offers differentiated aroma and flavor, which underlies its use as a spice (Hammouti et al., 2019). In Brazil, the introduction of the crop occurred only in 1930 in the Amazon region; since then, due to its economic importance and high market value, the condiment has become an opportunity, especially for family farmers (Moreira et al., 2021). This spice contains approximately 5–9% of piperamide alkaloids, primarily accumulating in the fruit peel and seeds that piperine stands out as the most biologically relevant compound, traditionally utilized for medicinal purposes, it demonstrates multiple pharmacological and ethnomedicinal properties (Batubara; Rafi; Yolanda, 2020; Dewi et al., 2025) including the relief of pain, digestive disorders, and fever (Ansari et al., 2023), the treatment of asthma (Usmani; Jain; Yadav, 2023), and its role in supporting anticancer therapies (Wiraswati et al., 2025).

Brazil stands as the world's foremost coffee producer, a position underpinned by its exceptionally favorable agroclimatic conditions. Currently, an estimated 330.000 rural establishments are dedicated to coffee cultivation, approximately 78% of which are classified as family farms, wherein coffee constitutes the primary source of livelihood (Santana et al., 2025). Furthermore, Coffee is a notable source of bioactive compounds, including chlorogenic acids, caffeine, cafestol, and kahweol. While it is predominantly consumed in its roasted form, green coffee retains significantly higher levels of chlorogenic acids, which have been linked to antioxidant and antiinflammatory effects. Consequently, green coffee has garnered increasing interest in scientific research and the nutraceutical sector due to its superior preservation of functional properties compared to roasted coffee beans (Humaryanto; Ave, 2019; Santana et al., 2025).

Just like green coffee, yerba mate (*Ilex paraguariensis*) is traditionally used in “Erva mate”, (is a cold aqueous infusion widely consumed in southern Brazil, Paraguay, and Argentina) notably rich in chlorogenic acids and has been garnering increasing scholarly and commercial interest due to its diverse array of bioactive properties (Kautzmann et al., 2024). Prepared from dried leaves of yerba mate and typically served in a “cuia”— a cup typically lined with leather or made from bull horn — with cold water, it offers a refreshing counterpart to chimarrão, the hot infusion made from the same plant. Owing to its complex phytochemical composition. Composing flavonoids, caffeine, theophylline, chlorogenic acids, saponins, vitamins, and trace elements, yerba mate demonstrates a broad spectrum of health-promoting properties. These include potent antioxidant capacity, metabolic stimulation, and antiinflammatory activity, positioning it as a beverage of growing relevance within the context of functional foods and nutraceuticals (Proch et al., 2023; Silvana et al., 2020).

Even though these products are sources of many desirable substances, them may also contain toxic metals (Chore et al., 2024; Górka; Baran; Słowik-Borowiec, 2025), pesticide residues above regulatory limits (Da Silva et al., 2022; Kandaswamy et al., 2021; Nolasco et al., 2022), mycotoxins such as aflatoxin B1 and ochratoxin A (Al Khoury et al., 2022; Attiya et al., 2021; Costa da Silva et al., 2021; Demirhan; Demirhan, 2023) and microorganism harmful to health (Idris et al., 2020; KoohyKamaly-Dehkordy et al., 2013). Dry food products, such as spices, teas, and coffee beans, are often contaminated with high levels of microorganisms (Maman et al., 2021). This contamination can occur at various stages of the production chain and may involve bacteria, viruses, and fungi, which pose significant health risks, particularly when these products are used in the preparation of ready-to-eat foods (Gryczka et al., 2020). Among pathogenic fungi, the genus *Aspergillus* stands out for including xerophilic and thermotolerant species capable of thriving in low-moisture and hightemperature environments. Some of these species not only produce mycotoxins but can also act as opportunistic pathogens in humans, as is the case with *Aspergillus* *fumigatus* (Abdel-Azeem et al., 2016; Brandl; Andersen, 2017). The clinical importance of *Aspergillus fumigatus* lies in its ability to cause a wide range of respiratory conditions, from localized airway inflammation to severe infections, notably invasive aspergillosis (IA). Although this cosmopolitan fungus is not typically a primary human pathogen, it has emerged as a significant opportunistic agent, particularly due to the increasing prevalence of conditions that require immunosuppressive therapies, including but not limited to cancer and organ transplantation (Resendiz-Sharpe et al., 2021). Aspergillosis is an environmentally acquired respiratory disease that encompasses a spectrum of clinical and radiological manifestations, largely determined by the host's immune status (Koren Fernández et al., 2014). It is estimated that approximately 4.8 million people worldwide are affected by some form of aspergillosis, with an estimated 300,000 cases of IA occurring annually, and associated mortality rates ranging between 30% and 95% (Brown et al., 2012; Garcia et al., 2018; Wang et al., 2022).

The first-line treatment for aspergillosis relies primarily on triazole-based antifungal agents, with itraconazole, voriconazole, and posaconazole being the most commonly used (Russo et al., 2020). However, increasing reports of reduced efficacy have been linked to the emergence of triazole-resistant *A. fumigatus* strains. These resistant isolates often harbor mutations in the CYP51 gene, which encodes lanosterol 14α-demethylase, an essential enzyme in the ergosterol biosynthesis pathway, responsible for producing the primary sterol component of the fungal cell membrane (Simon et al., 2021; Sueth-Santiago et al., 2015). Furthermore, resistance mechanisms to triazoles in *Aspergillus spp*. can be broadly categorized into two types: those associated with CYP51A alterations and those that are independent of this gene (Garcia-Rubio; Cuenca-Estrella; Mellado, 2017).

CYP51A-related resistance mechanisms are the most widespread globally (Macedo et al., 2021). Since their first identification in 1998 at a hospital in the Netherlands, an increasing number of mutations in the CYP51A gene have been reported. These alterations have been found in *A. fumigatus* isolates from patients undergoing long-term triazole therapy, as well as from triazole-naïve patients and environmental samples (Gyurtane Szabo et al., 2023; Snelders et al., 2008). Mutations within the coding sequence of the CYP51A gene such as TR34/L98H, TR34/L98H/S297T/F495I, TR46/Y121F/T289A, and TR53 are commonly associated with pan-azole resistance, conferring reduced susceptibility to all triazole antifungals

(Lucio et al., 2022). Additionally, certain amino acid substitutions, including M220, G54, and G138, have been shown to impair drug–target interaction by diminishing the binding affinity between the antifungal agent and the fungal enzyme (Howard et al., 2009; Verweij et al., 2016). In contrast, other substitutions such as F46Y, M127V, and E427K are typically linked to lower levels of azole resistance (Verweij et al., 2020).

The detection of TR34/L98H and TR46/Y121F alleles in both environmental and clinical isolates of *A. fumigatus* suggests that CYP51A mutations may arise not only in response to prolonged antifungal therapy, but also as a consequence of triazole fungicide application in agriculture, given the structural similarity between medical and agricultural triazoles (Burks et al., 2021; Kang et al., 2021). Although the use of such fungicides is restricted in certain crops—such as black pepper and yerba mate, which are the focus of our investigation—adjacent areas under intensive agricultural practices may act as resistance *hotspots*. Strains selected under fungicide pressure in these regions can subsequently contaminate nearby crops at various points in the production chain. Moreover, the study by Cui et al. (2019) demonstrated that preexposure to the agricultural fungicide tebuconazole in *A. fumigatus* isolates can induce cross-resistance to itraconazole and voriconazole. Therefore, the emergence of azoleresistant *A. fumigatus* strains in the environment should be regarded as an unintended consequence of triazole use in agriculture, especially considering that this fungal species is not a plant pathogen. Although *Aspergillus fumigatus* strains resistant to triazoles have been isolated from soil, plant debris, air, and compost samples (Kang et al., 2021; Kano et al., 2015), only the study by Viegas et al. (2023) reported isolates from the Fumigati section resistant to posaconazole in tea samples. Nonetheless, this study did not investigate potential mutations associated with the observed resistance profile. Yet, the occurrence of *Aspergillus fumigatus* in food products is frequent, posing a potential risk (Castro-Ríos et al., 2025).

Accordingly, the aim of this study was to investigate the resistance status of *Aspergillus fumigatus* isolates sampled from commercial products of yerba mate, black pepper, and green coffee to the medical azoles itraconazole and voriconazole, and to the agricultural azole tebuconazole. We compared the phenotypic and genotypic profiles of fungicide susceptibility. Additionally, we performed sequencing of genes encoding fungicide target proteins, and microsatellite typing based on short tandem environmental populations, underscoring the potential role of foods as reservoirs for genetically diverse strains that may also circulate in human environments.

5. CONCLUSION

Despite the absence of phenotypically resistant isolates, our study demonstrates that *Aspergillus fumigatus* is consistently present in minimally processed plant products commonly consumed in Brazil, including black pepper, yerba mate, and green coffee. The variability observed in EC_{50} values for tebuconazole and itraconazole, particularly among isolates from plant-based substrates, suggests early shifts in susceptibility that may reflect environmental selective pressures rather than established resistance. Although voriconazole susceptibility remained uniform across isolates, the presence of subtle differences in sensitivity highlights the importance of continuous monitoring, especially considering the fungus's remarkable adaptive capacity.

Genetic characterization revealed high multilocus genotypic diversity among isolates, alongside evidence of clonal expansion within specific substrates. The coexistence of unique and recurrent genotypes, coupled with the absence of major CYP51A mutations, underscores the evolutionary plasticity *of A. fumigatus* populations circulating in food products. Environmental exposure to agricultural triazoles, whether through fungicide drift, soil contamination, or indirect contact in production environments, may be driving early-stage adaptive responses. These findings reinforce that even food items not directly treated with azoles can act as reservoirs for diverse fungal populations that mirror patterns previously reported in clinical and environmental settings worldwide.

From a One Health perspective, the detection of *A. fumigatus* across multiple food matrices, combined with its genetic diversity and evidence of early susceptibility shifts, highlights the need for integrated surveillance strategies spanning agricultural, environmental, and clinical sectors. Ensuring safe production chains, rational fungicide use, and harmonized monitoring policies is essential to mitigate the risk of selecting for azole-resistant fungal populations. As global consumption of plant-derived foods continues to grow, proactive surveillance of *A. fumigatus* in food commodities represents a critical step toward safeguarding public health, supporting sustainable agriculture, and reducing the potential emergence and dissemination of triazoleresistant strains.

REFERENCES

ABDEL-AZEEM, A. M. *et al.* Biodiversity of the genus *Aspergillus* in different habitats. In:
GUPTA, Vijai Kumar B. T.-New and Future Developments in Microbial Biotechnology and Bioengineering (Org.). **New and Future Developments in Microbial Biotechnology and Bioengineering**. Amsterdam: Elsevier, 2016. p. 3–28.

AGROFIT. Ministério da Agricultura, Pecuária e Abastecimento**. Agrofit: sistema de agrotóxicos fitossanitários**. Disponível em: <https://agrofit.agricultura.gov.br/agrofit_cons/principal_agrofit_cons>. Acesso em: 29 jul. 2025.

AL KHOURY, A. *et al.* Inhibition of aflatoxin B1 synthesis in *Aspergillus* flavus by mate (Ilex *paraguariensis), Rosemary (Rosmarinus officinalis)* and green tea *(Camellia sinensis)* Extracts: Relation with Extract Antioxidant Capacity and Fungal Oxidative Stress Response Modulation. **Molecules**, v. 27, n. 23, 2022.

ANSARI, P. *et al.* Protective effects of medicinal plant-based foods against diabetes: a review on pharmacology, phytochemistry, and molecular mechanisms. **Nutrients**, v. 15, n. 14, p. 3266, 2023.

ATTIYA, W. A. *et al.* Prevalence of toxigenic fungi and mycotoxins in Arabic coffee (*Coffea arabica*): protective role of traditional coffee roasting, brewing and bacterial volatiles. **PLoS ONE**, v. 16, n. 10, 2021.

BATUBARA, I.; RAFI, M.; YOLANDA, M. L. Antioxidant, antibacterial, and degradation of Streptococcus mutans biofilms activities of black pepper (*Piper nigr*um) seed extract. **AIP Conference Proceedings**, v. 2243, n. 1, p. 030003, 2020.

BRANDL, J.; ANDERSEN, M. R. *Aspergilli*: models for systems biology in filamentous fungi. **Current Opinion in Systems Biology**, p. 67–73, 2017.

BROWN, G. D. *et al.* Hidden killers: human fungal infections. **Science Translational Medicine**, v. 4, n. 165, p. 165rv13, 2012.

BURKS, Caroline *et al.* Azole-resistant *Aspergillus fumigatus* in the environment: Identifying key reservoirs and hotspots of antifungal resistance. **PLOS Pathogens**, v. 17, n. 7, p. e1009711, 2021.

CASTRO-RÍOS, Katherin *et al.* Aspergillus fumigatus in the food production chain and azole resistance: a growing concern for consumers. **Journal of Fungi**, v. 11, n. 4, p. 252, 2025.

CERESINI, P. C. *et al.* Strategies for managing fungicide resistance in the Brazilian tropical agroecosystem: safeguarding food safety, health, and the environmental quality. **Tropical Plant Pathology**, v. 49, n. 1, p. 36–70, 2024.

CHORE, Eunice *et al.* Spicing up nutrition: investigation of trace elements in some spices locally sold in two markets in Kisumu-Kenya. **Environmental Monitoring and Assessment**, v. 196, n. 12, p. 1247, 2024.

COSTA DA SILVA, M. *et al.* Ochratoxin A levels in fermented specialty coffees from Caparaó, Brazil: is it a cause of concern for coffee drinkers? **Food Additives and Contaminants - Part A Chemistry, Analysis, Control, Exposure and Risk Assessment**, v. 38, n. 11, p. 1948–1957, 2021.

COSTA, D. M. C.; PAULETTO, D. Importância dos sistemas agroflorestais na composição de renda de agricultores familiares: estudo de caso no município de Belterra, Pará. **Nativa**, v. 9, n. 1, p. 92–99, 2021.

CUI, Ning *et al.* Tebuconazole induces triazole resistance in Asp*ergillus fumigatus* in liquid medium and soil. **Science of The Total Environment**, v. 648, p. 1237–1243, 2019.

DA SILVA, R. C. *et al.* Commercial yerba mate (I*lex paraguariensis*) produced in South America: determination of dithiocarbamate residues by gas chromatography-mass spectrometry. **Food Chemistry**, v. 394, 2022.

DE VALK, H. A. *et al.* Use of a novel panel of nine short tandem repeats for exact and high-resolution fingerprinting of A*spergillus fumigatus* isolates. **Journal of Clinical Microbiology**, v. 43, n. 8, p. 4112–4120, ago. 2005.

DELLALIBERA, O.; LEMAIRE, B.; LAFAY, S. Svetol®, green coffee extract, induces weight loss and increases the lean to fat mass ratio in volunteers with overweight problem. 2006. Disponível em: <https://www.semanticscholar.org/paper/Svetol%C2%AE%2C-green-coffeeextract%2C-induces-weight-loss-Dellalibera-Lemaire/fb7662abe61237a11977251edcf661debdb74e1b>. Acesso em: 9 abr. 2025

DEMIRHAN, B.; DEMIRHAN, B. E. Analysis of multi-mycotoxins in commonly consumed spices using the LC-MS/MS method for assessing food safety risks. **Microorganisms**, v. 11, n. 7, 2023.

DEWI, R. T. *et al.* Enhancement of dissolution rate of piperine using solid dispersion approach and evaluation of Its antioxidant activity. **Trends in Sciences**, v. 22, n. p. 8757, 2025.

DIESTE-PÉREZ, L. *et al.* Azole resistance in *Aspergillus* isolates from animals or their direct environment (2013–2023): a systematic review. **Frontiers in Veterinary Science**, v. 12, 2025.

FRAAIJE, B. *et al.* The multi-fungicide resistance status of a*spergillus fumigatus* populations in arable soils and the wider european environment. **Frontiers in Microbiology**, v. 11, 2020.

GARCIA, M. V. *et al.* Fungi in spices and mycotoxigenic potential of some *Aspergilli* isolated. **Food Microbiology**, v. 73, p. 93–98, 2018.

GARCÍA-CORDERO, J. *et al.* Dietary supplements containing oat beta-glucan and/or Green coffee (poly)phenols showed limited effect in modulating cardiometabolic risk biomarkers in overweight/obese patients without a lifestyle intervention. **Nutrients**, v. 15, n. 9, p. 2223, 2023.

GARCIA-RUBIO, R.; CUENCA-ESTRELLA, M.; MELLADO, E. Triazole resistance in *Aspergillus* species: an emerging problem. **Drugs**, v. 77, n. 6, p. 599–613, 2017.

GÓRKA, A.; BARAN, D.; SŁOWIK-BOROWIEC, M. Assessment of heavy metals, PAHs, and pesticide levels in yerba mate on the European market. **Environmental Science and Pollution Research**, v. 32, n. 2, p. 603–616, 2025.

GRYCZKA, U. *et al.* Efficacy of low energy electron beam on microbial decontamination of spices. **Radiation Physics and Chemistry**, v. 170, p. 108662, 2020.

GUINEA, J. Updated EUCAST clinical breakpoints against *Aspergillus*, implications for the clinical microbiology laboratory. **Journal of fungi,** Basel, v.6, n. 4, p. 343, 2020. Disponível em: https://www.mdpi.com/2309608X/6/4/343. Acesso em: 29 jul. 2025.

GYURTANE SZABO, Nikolett *et al.* Comparison of the Micronaut-AM system and the EUCAST broth microdilution reference method for MIC determination of four antifungals against *Aspergillus fumigatus*. **Journal of Fungi** (Basel, Switzerland), v. 9, n. 7, p. 721, 2023. DOI: 10.3390/jof9070721. Disponível em: https://www.mdpi.com/2309-608X/9/7/721. Acesso em: 29 jul. 2025.

HAMMOUTI, Belkheir *et al.* Black pepper, the “King of Spices”: chemical composition to applications. *Arabian Journal of Chemical and Environmental Research*, Vol. 6, n. 1, p. 12–56, 2019

HARISH, Ety *et al.* Triazole priming as an adaptive response and gateway to resistance in *Aspergillus fumigatu*s. **Antimicrobial Agents and Chemotherapy**, Washington, v. 66, n. 8, p. e00458-22, 2022. DOI: 10.1128/aac.00458-22

HOWARD, Susan J. *et al. Frequency and evolution of Azole resistance in Aspergillus fumigatus associated with treatment failure.* **Emerging infectious diseases**, Atlanta, v. 15, n. 7, p. 1068–1076, 2009. DOI: 10.3201/eid1507.090043.

HUMARYANTO; AVE, O. R. Exploring the potential of green coffee extract for wound healing treatment. **IOP Conference Series: Earth and Environmental Science**, Bristol, v. 391, n. 1, p. 012057, 2019. DOI: 10.1088/1755-1315/391/1/012057.

IDRIS, A. L. *et al.* Ecologically controlling insect and mite pests of tea plants with microbial pesticides: a review. **Archives of Microbiology**, v. 202, n. 6, p. 1275–1284, 2020. DOI: 10.1007/s00203-020-01862-7.

JØRGENSEN, Karin Meinike *et al.* Dissection of the activity of agricultural fungicides against clinical *Aspergillus* isolates with and without environmentally and medically induced azole resistance. **Journal of Fungi**, Basel, v. 7, n. 3, p. 205, 2021. DOI: 10.3390/jof7030205.

KANDASWAMY, C. *et al.* Comparative evaluation of multi-residue methods for analysis of pesticide residues in black pepper by gas chromatography tandem mass spectrometry:
critical evaluation of matrix co-extractives and method validation. **Journal of Food Science and Technology**, New Delhi, v. 58, n. 3, p. 911–920, 2021. DOI: 10.1007/s13197-020-04694-8.

KANG, S. Earl *et al.* Evidence for the agricultural origin of resistance to multiple antimicrobials in Aspergillus fumigatus, a fungal pathogen of humans. **G3: Genes|Genomes|Genetics**, Raleigh, v. 12, n. 2, p. jkab427, 2021. DOI: 10.1093/g3journal/jkab427.

KANO, Rui *et al.* Does farm fungicide use induce azole resistance in *Aspergillus fumigatus*? **Medical Mycology**, Oxford, v. 53, n. 2, p. 174–177, 2015. DOI: 10.1093/mmy/myu076.

KAUTZMANN, Charles *et al.* Roasted yerba mate (*Ilex paraguariensis*) infusions in bovine milk model before and after *in vitro* digestion: Bioaccessibility of phenolic compounds, antioxidant activity, protein–polyphenol interactions and bioactive peptides. **Food Research International**, Amsterdam, v. 183, p. 114206, 2024. DOI: 10.1016/j.foodres.2024.114206.

KOOHY-KAMALY-DEHKORDY, P. *et al.* Microbiological quality of retail spices in Tehran, Iran. **Journal of Food Protection**, Des Moines,, v. 76, n. 5, p. 843–848, 2013. DOI: 10.4315/0362-028X.JFP-12-180.

KOREN FERNÁNDEZ, L. *et al.* [The different manifestations of pulmonary aspergillosis: multidetector computed tomography findings]. **Radiologia**, Barcelona, v. 56, n. 6, p. 496–504, 2014. DOI: 10.1016/j.rx.2013.09.007.

LUCIO, Jose *et al.* An expanded agar-based screening method for azole-resistant Aspergillus fumigatus. **Mycoses**, Berlin, v. 65, n. 2, p. 178–185, fev. 2022. DOI: 10.1111/myc.13400.

MACEDO, Daiana *et al.* Emergence of triazole resistance in *Aspergillus* spp. in Latin America. **Current Fungal Infection Reports**, New York, v. 15, n. 3, p. 93–103, 2021. DOI: 10.1007/s12281-021-00418-6.

MAGUIRE-RAJPAUL, Victoria A. *et al.* Coffee certification in Brazil: compliance with social standards and its implications for social equity. **Environment, Development and Sustainability**, Dordrecht, v. 22, n. 3, p. 2015–2044, 2020. DOI: 10.1007/s10668-018-0275-z.

MAMAN, Mantana *et al.* Storage fungi and ochratoxin A associated with arabica coffee bean in postharvest processes in Northern Thailand. **Food Control**, Cambridge, v. 130, p. 108351, 2021. DOI: 10.1016/j.foodcont.2021.108351.

MOREIRA, Wendel K. O. *et al.* Production Chain of Black Pepper in Brazil. **Nucleus**, São Paulo, v. 18, n. 2, p. 101–116, 2021. DOI: 10.3738/1982.2278.3826.

MUTALIB, Sofianita *et al.* Herbal plant analysis based on leaf features using K-means clustering. **IOP Conference Series: Earth and Environmental Science**, Bristol, v. 1019, n. 1, p. 012026, 2022. DOI: 10.1088/1755-1315/1019/1/012026.

NOLASCO, A. *et al.* Coffee silverskin: chemical and biological risk Assessment and health profile for Its potential use in functional foods. **Foods**, Basel, v. 11, n. 18, 2022. DOI: 10.3390/foods11182834.

PITT, John I.; HOCKING, Ailsa D. Aspergillus and Related Teleomorphs. In: PITT, John I.; HOCKING, Ailsa D. (Orgs.). **Fungi and Food Spoilage**. Cham: Springer International Publishing, 2022. p. 351–439. DOI: 10.1007/978-3-031-13347-9_8.

PROCH, Jędrzej *et al.* Influence of Brewing Method on the Content of Selected Elements in Yerba Mate (Ilex paraguarensis) Infusions. **Foods**, Basel, v. 12, n. 5, p. 1072, 2023. DOI: 10.3390/foods12051072.

RESENDIZ-SHARPE, Agustin *et al.* Triazole-Resistance in Environmental Aspergillus fumigatus in Latin American and African Countries. **Journal of Fungi,** Basel, , v. 7, n. 4, p. 292, 2021. DOI: 10.3390/jof7040292.

RIZZO MOREIRA, T *et al.* Climatic favorability to the occurrence of Hemileia vastatrix in apt areas for the cultivation of Coffea arabica L. in Brazil. Climate, Basel, v. 12, n. 8, p. 123, 2024. DOI: 10.3390/cli12080123.

RUSSO, Alessandro *et al.* Pulmonary Aspergillosis: an evolving challenge for diagnosis and treatment. **Infectious Diseases and Therapy**, Cham, v. 9, n. 3, p. 511–524, 2020. DOI: 10.1007/s40121-020-00315-4.

RYBAK, Jeffrey M.; FORTWENDEL, Jarrod R.; ROGERS, P. David. Emerging threat of triazole-resistant Aspergillus fumigatus. **Journal of Antimicrobial Chemotherapy**,Oxford, v. 74, n. 4, p. 835–842, 2019. DOI: 10.1093/jac/dky517.

SANTANA, Derielsen Brandão *et al.* Spatiotemporal dynamics of soil and soil organic Carbon losses via water erosion in coffee cultivation in tropical regions. **Sustainability**, Basel, v. 17, n. 3, p. 821, 2025. DOI: 10.3390/su17030821.

SANTIANO, Flavia E. *et al.* Protective effects of yerba mate (*Ilex paraguariensis*) on prostate cancer development. **Nutrition**, Amsterdam, v. 108, p. 111957, 2023. DOI: 10.1016/j.nut.2022.111957.

SERRANO, Rita *et al.* Rapid identification of *Aspergillus fumigatus* within the section Fumigati. **BMC microbiology**, london, v. 11, p. 82, 2011. DOI: 10.1186/1471-2180-11-82.

SILVANA, Velázquez María *et al.* Colonization dynamics of arbuscular mycorrhizal fungi (AMF) in *Ilex paraguariensis* crops: seasonality and influence of management practices. **Journal of King Saud University - Science**, Riyadh, v. 32, n. 1, p. 183–188, 2020. DOI: 10.1016/j.jksus.2018.03.017.

SIMON, Loïc *et al.* Azole resistance in *Aspergillus fumi*gatus isolates from respiratory specimens in Lyon University Hospitals, France: prevalence and mechanisms involved. **International Journal of Antimicrobial Agents**, Oxford, v. 58, n. 6, p. 106447, dez. 2021. DOI: 10.1016/j.ijantimicag.2021.106447.

SIOPI, Maria *et al.* Nationwide surveillance of azole-resistant Aspergillus fumigatus environmental isolates in Greece: detection of pan-azole resistance associated with the TR46/Y121F/T289A cyp51A mutation. **The Journal of Antimicrobial**

Chemotherapy, Oxford, v. 75, n. 11, p. 3181–3188, 2020. DOI: 10.1093/jac/dkaa316.

SNELDERS, Eveline *et al.* Emergence of Azole Resistance in Aspergillus fumigatus and Spread of a Single Resistance Mechanism. **PLOS Medicine**, San Francisco, v. 5, n. 11, p. e219, 2008. DOI: 10.1371/journal.pmed.0050219.

SONG, Yiting *et al.* A systematic review on the yerba mate (*Ilex paraguariensis* A. St. Hil.). **Journal of Food Composition and Analysis**, Amsterdam, v. 142, p. 107466, 2025. DOI: 10.1016/j.jfca.2025.107466.

SUETH-SANTIAGO, Vitor *et al.* CYP51: Uma boa ideia? **Revista Virtual de Química**, São Paulo, v. 7, n. 2, p. 539–575, 2015. DOI: 10.5935/1984-6835.20150024.

TALHINHAS, Pedro *et al.* The coffee leaf rust pathogen Hemileia vastatrix: one and a half centuries around the tropics. **Molecular Plant Pathology**, Oxford, v. 18, n. 8, p. 1039–1051, 2017. DOI: 10.1111/mpp.12512.

USMANI, Kainat; JAIN, Subodh Kumar; YADAV, Shweta. Mechanism of action of certain medicinal plants for the treatment of asthma. **Journal of Ethnopharmacology**, Amsterdam, v. 317, p. 116828, 5 dez. 2023. DOI: 10.1016/j.jep.2023.116828.

VERWEIJ, Paul E. *et al.* Azole resistance in *Aspergillus fumigatus*: can we retain the clinical use of mold-active antifungal azoles? **Clinical Infectious Diseases**, Oxford, v. 62, n. 3, p. 362–368, 2016. DOI: 10.1093/cid/civ885.

VERWEIJ, Paul E. *et al.* The one health problem of azole resistance in *Aspergillus fumigatus*: current insights and future research agenda. **Fungal Biology Reviews**, Oxford, v. 34, n. 4, p. 202–214, 2020. DOI: 10.1016/j.fbr.2020.10.003.

VICENTINI, Samara N. C. *et al.* Monitoring of Brazilian wheat blast field populations reveals resistance to QoI, DMI, and SDHI fungicides. **Plant Pathology**, Oxford, v. 71, n. 2, p. 304–321, 2022. DOI: 10.1111/ppa.13470.

VIDAL, Maria de Fátima. Evolução do cultivo de pimenta-do-reino na área de atuação do BNB. **Caderno Setorial ETENE**, Fortaleza, v. 5, n. 146, dez. 2020. Banco do Nordeste do Brasil, 2020.

VIEGAS, Carla *et al.* Tea contamination by mycotoxins and azole-resistant mycobiota – The need of a One Health approach to tackle exposures. **International Journal of Food Microbiology**, Amsterdam, v. 385, p. 110015, 2023. DOI: 10.1016/j.ijfoodmicro.2022.110015

WANG, Feiyan *et al.* Increased triazole-resistance and cyp51A mutations in *Aspergillus fumigatus* after selection with a combination of the triazole fungicides difenoconazole and propiconazole. **Journal of Hazardous Materials**, Amsterdam, v. 400, p. 123200, 2020. DOI: 10.1016/j.jhazmat.2020.123200.

WANG, Sijiao *et al.* Resveratrol attenuates inflammation and apoptosis through alleviating endoplasmic reticulum stress via Akt/mTOR pathway in fungus-induced allergic airways inflammation. **International Immunopharmacology**, v. 103, p. 108489, 2022. DOI: 10.1016/j.intimp.2021.108489.

WIRASWATI, Hesti Lina et al. Harnessing the anticancer potential of Piper nigrum: a synergistic approach to chemotherapy enhancement and reduced side effects. **Discover Oncology**, Basel, v. 16, n. 1, p. 10, 2025. DOI: 10.1007/s12672-024-01716-4.