UNIVERSIDADE FEDERAL DO RIO GRANDE DO SUL
INSTITUTO DE CIÊNCIAS BÁSICAS DA SAÚDE
PROGRAMA DE PÓS-GRADUAÇÃO EM MICROBIOLOGIA AGRÍCOLA E DO AMBIENTE

ÁCAROS DE AMBIENTES AQUÁTICOS E PRODUTOS ARMAZENADOS: BIOECOLOGIA E SUAS INTERAÇÕES COM MICRORGANISMOS

GUILHERME LIBERATO DA SILVA

Orientadora: Profª Drª Onilda Santos da Silva
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“Quem quiser ser líder deve ser primeiro servo. Se você quiser liderar, deve servir.”

Jesus Cristo

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ÁCAROS DE AMBIENTES AQUÁTICOS E PRODUTOS ARMAZENADOS: BIOECOLOGIA E SUAS INTERAÇÕES COM MICRORGANISMOS[1]

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RESUMO

Ácaros podem ser encontrados nos mais diferentes ambientes e podem ter um papel importante na agricultura e na área médica. Por exemplo, ácaros aquáticos têm sido relatados como bioindicadores de qualidade da água. Em ambientes domiciliares e armazéns, a espécie *Tyrophagus putrescentiae* é frequentemente associada à deterioração de alimentos e à dispersão de microrganismos. Além disso, pode servir como alimento para outras espécies de ácaros. Assim, os objetivos deste estudo foram: Avaliar padrões ecológicos de ácaros aquáticos em áreas de arroz irrigado e lagoas nativas, e conhecer o ciclo biológico de *T. putrescentiae* e seu potencial de dispersão de fungos ambientais e de importância médica. Os resultados demonstraram que os ácaros aquáticos possuem forte correlação com a turbidez, apresentando maior riqueza e abundância em lagoas nativas do que em áreas de arroz irrigado. Com relação à *T. putrescentiae*, esta espécie apresentou melhores parâmetros demográficos quando alimentada sobre levedura seca. Entretanto, a 18 e 25ºC ovipositou mais sobre ração de rato, com diferentes padrões de oviposição para as temperaturas avaliadas. Alem disso, *T. putrescentiae* apresentou maiores taxas de crescimento populacional sobre os fungos *Fusarium verticillioides*, *Penicillium citrinum* e *Trichophyton mentagrophytes*, podendo, portanto, ser considerado um disseminador de fungos ambientais e de importância médica. Além disso, nosso estudo demonstrou que *B. dentriticus* é um predador em potencial de *T. putrescentiae* e *Megninia ginglymura*. Este foi o primeiro estudo no Brasil que demonstrou a importância de ácaros de ambientes aquáticos e dispersores de fungos ambientais e de importância medica.

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MITES OF AQUATIC ENVIRONMENTS AND STORED PRODUCTS: BIOECOLOGY AND ITS INTERACTIONS WITH MICRORGANISMS[1]

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ABSTRACT

Mites can be found in many different environments and can play an important role in agriculture and in the medical field. For example, aquatic mites have been reported as bioindicators of water quality. In home environments and warehouses, *Tyrophagus putrescentiae* is often associated with food spoilage and dispersal of microorganisms. In addition, it can serve as food for other species of mites. Thus, the objectives of this study were: To evaluate ecological patterns of aquatic mites in irrigated rice and native lagoons, and to know the biological cycle of *T. putrescentiae* and its potential for dispersion of environmental fungi and of medical importance. The results showed that aquatic mites have a strong correlation with turbidity, presenting greater richness and abundance in native ponds than in irrigated rice areas. In relation to *T. putrescentiae*, this species presented better demographic parameters when fed on dry yeast. However, at 18 and 25ºC it oviposit more on rat ration, with different oviposition patterns for the evaluated temperatures. In addition, *T. putrescentiae* presented higher population growth rates on fungi *Fusarium verticillioides, Penicillium citrinum and Trichophyton mentagrophytes*, and could therefore be considered a disseminator of environmental fungi and some of medical importance. In addition, our study demonstrated that *B. dentriticus* is a potential predator of *T. putrescentiae* and *Megninia ginglymura*. This is the first study in Brazil that demonstrated the importance of mites from aquatic environments and dispersers of environmental fungi and of medical importance.

[1]Doctoral Dissertation in Agricultural and Environmental Microbiology – Instituto de Ciências Básicas da Saúde, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil. (114 p.) January, 2018.

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1. INTRODUÇÃO

Cada vez mais é necessário ampliar estudos de organismos ambientais que possuam um papel importante na agricultura e na área médica. Dessa forma, algumas espécies de ácaros são ferramentas importantes para se estudar inúmeros processos ecológicos que acontecem no meio ambiente e que possuam relação direta com áreas antropizadas e consequente disseminação de microrganismos. Um exemplo de ecossistema antropizado é a cultura do arroz irrigado. O Brasil é o nono maior produtor de arroz do mundo e o Estado do Rio Grande do Sul representa aproximadamente 69% da produção deste produto no país (IBGE, 2015). Com o crescimento das áreas de produção de arroz irrigado, o monitoramento da qualidade da água adquiriu importância crescente. No Brasil, as áreas de Restinga no Sul são uma das regiões onde ocorre um cultivo massivo de arroz irrigado. Esta região inclui uma parte do bioma da Floresta Atlântica e está sob uma pressão antropogênica extrema, que modifica as paisagens naturais (AB'SABER, 2003). Os declínios da qualidade da água, resultantes de uma intensa contaminação, diminuem os índices de diversidade e, em alguns casos, levam a um colapso da fauna de ácaros aquáticos (KOWALIK & BIESIADKA, 1981). Em geral, a água utilizada para irrigar estas culturas é proveniente de lagos e lagoas de água doce localizados nas proximidades dos campos de cultivo. Portanto, as comparações entre as composições de ácaros aquáticos de lagos nativos e áreas de arroz irrigado podem ser utilizadas para mensurar a qualidade da água, permitindo uma avaliação das alterações ocorridas no meio ambiente.

Por outro lado, ácaros de produtos armazenados são encontrados principalmente no meio rural, em grãos, farinhas e fenos. Entretanto, também podem ser encontrados na poeira domiciliar, desde que as condições de umidade e temperatura sejam favoráveis para o seu desenvolvimento. Estes ácaros se desenvolvem em ambientes úmidos, com umidade relativa ideal em torno de 80% e temperatura entre 25 e 30º C (TEE, 1994). Os gêneros mais comumente encontrados no país são pertencentes às famílias Pyroglyphidae, Glycyphagidae, Cheyletidae e Acaridae. Nesta última família, *Tyrophagus putrescentiae* (Schrank) já foi descrito em diversas regiões do país e é a principal espécie encontrada em produtos armazenados. É considerada uma praga por reduzir o teor de nutrientes e capacidade germinativa das sementes, e, consequentemente, causar prejuízos na agricultura (ROSA, 1978; KRANTZ, 2008; JORGE NETO, 1984; ŽDÁRKOVA, 1991). O Brasil é o terceiro

produtor de alimentos para animais do mundo, produzindo cerca de 68,7 milhões de toneladas em 2015 (ZANI, 2015). Desta forma, foi importante testar vários produtos armazenados, como farinha de milho, rações para ratos, farinha de arroz e fermento seco, com o objetivo de comparar o desenvolvimento biológico de *T. putrescentiae* sob as mesmas. Além disso, verificou-se o efeito da temperatura sobre a capacidade de oviposição associada a várias dietas, pelo teste com chance de escolha para oviposição.

Com relação a fungos associados a ácaros de grãos armazenados, Nesvorná et al. (2012) reportou um intenso consumo de diversas espécies de fungos por *T. putrescentiae*, tal como *Fusarium poae*, *Fusarium culmorum*, *Fusarium verticillioides* e *Fusarium avenaceum*. Essas interações promovem a disseminação de fungos em todo o grão armazenado, influenciando ambas as pragas. Os fungos filamentosos e leveduriformes são amplamente distribuídos no ar, poeira, fomites e microbiota normal. As micoses humanas são causadas por patógenos verdadeiros (ex: *Epidermophyton, Microsporum, Trichophyton, Sporothrix* spp.) e patógenos oportunistas (ex: *Aspergillus fumigatus, Candida albicans, Rhizopu*s spp.). Os seus esporos podem ser importantes agentes alergênicos humanos e os ácaros podem ser esta via de transmissão (WALSH & DIXON, 1996). A maioria dos casos alérgicos diagnosticados e também de dermatofitoses podem estar associados à presença de ácaros, principalmente de *T. putrescentiae*. Assim, nós estudamos as interações entre os ácaros e a comunidade fúngica, principalmente enfatizando os fungos patogênicos humanos.

Finalmente, o ácaro predador *Blattisocius dentriticus* (Berlese) alimenta-se de uma variedade de ovos *T. putrescentiae,* além de outros artrópodes (FENILLI & FLECHTMANN, 1990). Este predador também foi reportado em cereais, alimentos armazenados e ninhos de pássaros (BAGGIO *et al*., 1987; SILVA *et al*., 2013). Faleiro *et al.* (2015) verificaram que *B. dentriticus* era o predador mais comum em armadilhas de aviários. Silva *et al.* (2013) o reportaram em sistemas de criação livre de galinhas. A determinação da preferência de presa, e do ciclo de vida são etapas essenciais na avaliação da eficácia de um predador em programas de controle biológico. No entanto, pouco se sabe sobre a capacidade de predação de *B. dentriticus* sobre *T. putrescentiae*. Assim, este ácaro aparece como um predador em potencial para o controle biológico de diversos ácaros-praga da indústria avícola e de alimentos. Portanto, neste estudo investigou-se seu desenvolvimento biológico quando alimentado com *T. putrescentiae* e o ectoparasito *Megninia ginglymura* (Mégnin) (Analgidae).

2. OBJETIVOS

2.1 Objetivo Geral

Avaliar padrões ecológicos de ácaros aquáticos em áreas de arroz irrigado e lagoas nativas, e conhecer o ciclo biológico de *T. putrescentiae* e seu potencial de dispersão de fungos ambientais e de importância médica.

2.2 Objetivos Específicos

2.2.1 Comparar a abundância, riqueza e composição da fauna de ácaros da água de diferentes habitats.

2.2.2 Verificar a influência dos seguintes fatores abióticos sobre a composição das espécies: pH, turbidez (NTU), temperatura da água (°C) e oxigênio dissolvido (mg/L).

2.2.3 Conhecer o desenvolvimento biológico do ácaro predador *B. dentriticus* quando alimentado com *T. putrescentiae* e *M. ginglymura*.

2.2.4 Comparar o desenvolvimento biológico de *T. putrescentiae* alimentado sobre diferentes dietas e os efeitos da temperatura na capacidade de oviposição através do teste de escolha de oviposição.

2.2.5 Estudar o crescimento da população de *T. putrescentiae* sobre espécies de fungos isolados de alimentos e provenientes da coleção da UFRGS.

3. REVISÃO DA LITERATURA

3.1 Ácaros

Os ácaros pertencem ao filo Arthropoda, subfilo Chelicerata e classe Arachnida, e se caracterizam por apresentarem exoesqueleto quitinoso e pernas articuladas (WALTER *et al*., 2009). A subclasse Acari compreende os ácaros e carrapatos que diferem dos demais aracnídeos pela ausência de segmentação aparente e divisão do corpo (WALTER *et al*., 2009; EVANS, 1992). A perda da segmentação levou os autores a considerar os ácaros como artrópodes altamente especializados e membros de um grupo que se afastou muito cedo de outras linhas de evolução do filo Arthropoda (MORAES & FLECHTMANN, 2008). A subclasse Acari é dividida nas superordens Anactinotrichida e Actinotrichida. A primeira é subdividida nas ordens Ixodida, Mesostigmata, Holothyrida e Notostigmata, enquanto que a segunda é subdividida nas ordens Prostigmata, Oribatida e Astigmata (EVANS, 1992). Os ácaros podem ser divididos conforme seu hábito alimentar. Alguns são predadores, outros se alimentam de microorganismos, matéria orgânica em decomposição, diferentes partes vegetais, ou parasitam vertebrados e invertebrados. Representantes de diferentes grupos têm sido encontrados nos mais diversos ecossistemas terrestres e aquáticos (MORAES & FLECHTMANN, 2008). Os ácaros plantícolas de maior importância pertencem às ordens Prostigmata, contendo a maioria das espécies fitófagas, e Mesostigmata, composta principalmente por predadores (EVANS, 1992). Os ácaros da subordem Astigmata são, em sua maioria, de movimentos lentos, apresentando cutícula elástica, transparente, podendo variar de 0,2 a 0,5 mm de comprimento (Flechtmann 1986). Durante seu desenvolvimento pós-embrionário, os Astigmata passam pelas fases de larva, protoninfa, tritoninfa e adulto. Entretanto, uma deutoninfa pode surgir após a protoninfa, quando as condições ambientais são desfavoráveis ou na falta de alimento. Este estágio é diferente em relação à morfologia e ao comportamento dos demais estágios (MORAES & FLECHTMANN, 2008). A deutoninfa apresenta estruturas especializadas para fixar-se em outros animais, com a finalidade de atingir ambientes mais favoráveis ao seu desenvolvimento. Este transporte passivo de um organismo para outro ambiente, com o propósito de dispersão é denominado de forésia (CLAUSEN, 1976; ROFF, 1991; STEINKRAUS & CROSS, 1993). Tal comportamento é uma estratégia que possibilita ao organismo colonizar novas áreas ou fugir de locais onde as condições sejam desfavoráveis, contribuindo também para

prolongar sua sobrevivência.

3.2 Ácaros aquáticos no Brasil

Dentre as subordens de Acari, os Prostigmata são os que apresentam maior número de espécies aquáticas, divididas em dois grandes grupos: Halacaroidea e Hydracarina. Os Halacaroidea, com aproximadamente 1.000 espécies, são representados pelas famílias Halacaridae - predominantemente marinha (BARTSCH 1996, 2004). Já os Hydracarina, considerados como ácaros aquáticos “verdadeiros”, incluem atualmente por volta de 6.000 espécies, distribuídas em oito superfamílias, 57 famílias e cerca de 420 gêneros (Figura 1) (DI SABATINO *et al.,* 2008). No século XX, os esforços para o conhecimento do grupo se concentraram entre as décadas de 1920 e 1950, com especial atenção a dois pesquisadores: Karl H. Viets e Olov Lundblad.

Viets (1927, 1933, 1935, 1936a, 1936b, 1937, 1938, 1954a, 1954b, 1959) descreveu 31 novas espécies, provenientes de amostras coletadas em açudes, poças d’água e lagoas nas regiões Norte (Estado do Amazonas), Nordeste (Piauí, Ceará, Rio Grande do Norte, Paraíba, Pernambuco, Alagoas, Sergipe e Bahia) e Sul (Rio Grande do Sul).

Lundblad (1931, 1936, 1937, 1941, 1942, 1943a, 1943b, 1944) empreendeu pessoalmente uma expedição pela América do Sul, onde realizou coletas em 120 diferentes localidades, sendo 45 delas no Paraguai e 75 nas regiões Sul e Sudeste do Brasil (44 no Estado de Santa Catarina; 16 no Rio Grande do Sul; 12 no Rio de Janeiro e apenas três no estado de São Paulo). Neste último estado, foram listadas somente 20 espécies dentre as 332 encontradas em todo o país. Os últimos e mais recentes trabalhos sobre hidracarinos brasileiros trazem as descrições de *Corticacarus (Lundbladacarus) schusteri* e *Tyrrellia petrophila* , provenientes de amostras de uma cachoeira em Ubatuba, São Paulo (VIETS, 1977). Ainda, sete espécies de *Unionicola* são parasitos de moluscos bivalves coletados no Rio Jutaí, município de Jutaí, Amazonas (VIDRINE, 1985) e *Litarachna brasiliensis* encontrada na laguna Tramandaí, Imbé, Rio Grande do Sul (SMIT, 2007).

Os ácaros aquáticos têm sido utilizados como bioindicadores de qualidade ambiental em vários ecossistemas (BIESIADKA & KOWALIK, 1991; ROUSCH *et al*., 1997; DI SABATINO *et al*., 2002; DOHET *et al*., 2008; GOLDSCHMIDT, 2016). Muitos estudos revelaram que as comunidades de ácaros aquáticos podem ser afetadas por vários fatores abióticos, como agentes poluidores, variações de temperatura, oxigênio

dissolvido, condutividade e concentrações de carbonato (SCHWOERBEL, 1961; WIĘCEK *et al*., 2013). Esforços estão sendo realizados para efetivamente empregar estes ácaros como indicadores ecológicos, incluindo-os em programas de monitoramento faunístico (GOLDSCHMIDT, 2016).

Figura 1 – Um exemplar de ácaro aquático dulcícola (Hydracarina). Fonte: do autor.

3.3 O ácaro de produtos armazenados *Tyrophagus putrescentiae*

O ácaro *Tyrophagus putrescentiae* (Figura 2) é uma espécie cosmopolita comumente encontrada em ambientes domiciliares, aviários e em produtos armazenados. Nestes ambientes, geralmente prefere alimentos com alto teor de gordura e proteínas, como farinha, trigo, soja, queijo, pão de centeio, leite em pó e diversas sementes (SOLARZ *et al*., 2007; COLLOFF, 2009; HUBERT *et al*., 2013; RYBANSKA *et al*., 2016). Possui ampla distribuição, sendo relatado em um grande número de países, como por exemplo, Austrália (HIBBERSON & VOGELNEST, 2014), Brasil (BAGGIO *et al*., 1987), China (TAO *et al*., 2015), Grécia (PALYVOS *et al*., 2008), República Checa (HUBERT *et al*., 2004) e Turquia (AYGUN *et al*., 2007), apesar de ser mais comum em países tropicais e subtropicais (HUGHES, 1976). A preferência por ambientes mais quentes e úmidos foi confirmada em estudos recentes (ASPALY *et al*., 2007; SÁNCHEZ-RAMOS *et al*., 2007; HUBERT *et al*., 2010) que mostraram que esta espécie possui metabolismo ativo entre 0,8 a 48,1ºC, sendo a temperatura ótima em torno de 32ºC (HUBERT *et al*., 2010) e limiar superior de atividade de 49ºC (ASPALY *et*

al., 2007). Altas infestações desta espécie promovem a queda da qualidade do produto, acelerando sua deterioração (DUEK *et al.,* 2001). Por isto é importante elucidar a perda nutricional ocorrida pela infestação de ácaros em grãos e seus derivados lácteos, uma vez que estes são a base da alimentação da população brasileira, caracterizando as reações alérgicas decorrentes.

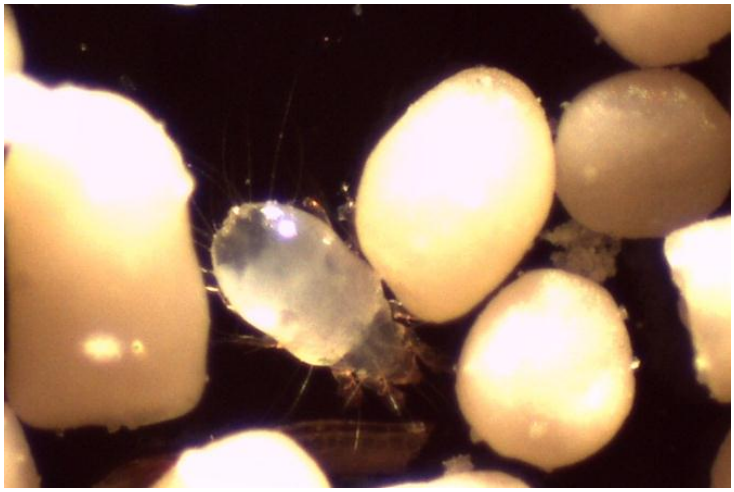

Figura 2 - *Tyrophagus putrescentiae* com pellets de levedura que são utilizados como fonte de alimento. Fonte: do autor.

A legislação brasileira estabelece uma tolerância de insetos e artrópodes em alimentos com o objetivo de manter um padrão e garantir a qualidade dos produtos. Entretanto, esta legislação não cita um limite para ácaros em produtos armazenados (AGÊNCIA NACIONAL DE VIGILÂNCIA SANITÁRIA, 2014).

Nos últimos anos os ácaros vêm recebendo atenção para a área da saúde, visto que são uma das principais causas de asma, rinite e demais doenças alérgicas devido ao seu potencial alergênico (ARLIAN *et al*., 1984; VAN HAGE-HAMSTEN & JOHANSSON, 1998; BRAVO *et al*., 1999; ELDER *et al*., 2012; LIAO *et al*., 2013a, b; YU *et al.,* 2014). A ingestão de alimentos contaminados por esta espécie também pode levar a vários casos de anafilaxia (LIAO *et al*., 2013a, b). Nuñez *et al*. (2016) descreveram uma forte indução de inflamação alérgica nos pulmões de camundongos tanto em modelos agudos e crônicos. Sabe-se até o momento que *T. putrescentiae* pode abrigar diversas bactérias (ERBAN & HUBERT, 2008; SMRŽ & ČATSKÁ, 2010; ERBAN *et al*., 2016), podendo hospedar comunidades bacterianas no seu intestino, tecidos do parênquima e trato reprodutivo (HUBERT *et al*., 2012; KOPECKY *et al*., 2014a,b). Erban *et al*. (2016) detectaram em fezes de *T. putrescentiae* uma associação simbiótica com *Bacillus cereus*, que coexiste em equilíbrio e benefício com este ácaro.

No entanto, isto aumenta a importância médica e veterinária dos alérgenos detectados nas suas fezes. As enzimas e toxinas de *B. cereus* são importantes componentes de alérgenos de ácaros. Além disso, esta espécie é associada com bactérias parasitas ou simbiontes (*Cardinium* e *Wolbachia*) que habitam o trato reprodutivo e a gordura corporal destes acaros (KOPECKY *et al*., 2013; BROWN & LLOYD, 2015). Essas bactérias associadas afetam o desempenho dos ácaros, ocasionando efeitos indiretos sobre a colonização de novos habitats.

3.3.1 Saúde humana e animal

Ácaros de produtos armazenados desencadeiam reações alérgicas, tais como: asma, rinite alérgica, urticária e dermatite de contato, através da inalação, ingestão ou contato com ácaros vivos ou mortos, subprodutos do metabolismo ou fezes (SINHA, 1979).

Casos de urticária e dermatite de contato são comuns em agricultores e trabalhadores de padarias e indústrias (REVSBECH & DUEHOLM, 1990; ARLIAN *et al*., 1997; STORAAS *et al*., 2005; ESTÉVEZ, 2006; STEJSKAL & HUBERT, 2008). Além disso, a ingestão de alimentos preparados com farinhas infestadas por ácaros pode desencadear episódios de anafilaxia (ERBEN *et al*., 1993; POSTHUMUS & BORISH, 2012; SANCHEZ-BORGES *et al*., 2013; TAKAHASHI *et al*., 2014; BARRERA *et al*., 2015). O primeiro caso de anafilaxia sistêmica ocorrida pela ingestão de alimentos infestados por ácaros foi relatado em 1993 tendo como agente causador *Dermatophagoides farinae* Trouessart (ERBEN *et al*., 1993). Após este relato, muitos outros casos foram documentados, inclusive tendo como agente causador *T. putrescentiae* (TAKAHASHI *et al*., 2014). Estima-se que a incidência de episódios de anafilaxia seja em torno de 0,5 a 2%, sendo a maioria pela ingestão de cereais (especialmente o trigo) contaminados por ácaros e seus alergênicos (POSTHUMUS & BORISH, 2012). Em geral, os episódios de anafilaxia ocorrem em pacientes com rinite alérgica ou asma, que sensibilizados para ácaros da poeira doméstica, desenvolvem reações alérgicas, mediadas ou não por IgE, após ingestão de farinha contaminada por ácaros de armazenamento (POSTHUMUS & BORISH, 2012). A sensibilização cruzada com ácaros da poeira doméstica já foi descrita na literatura (ARLIAN *et al*., 2009; LIAO *et al*., 2010).

Em relação a animais, *T. putrescentiae* é considerada a espécie mais abundante em ração de animais, alcançando o nível de praga, podendo estar

associada com casos de dermatite atópica canina (HIBBERSON & VOGELNEST, 2014).

3.3.2 *Tyrophagus putrescentiae* x Fungos

A associação entre ácaros e fungos já é estabelecida na literatura, apesar de não se conhecer os mecanismos envolvidos. *Tyrophagus putrescentiae* se alimenta de esporos e hifas de uma grande variedade de microorganismos dermatófitos (*Trichophyton mentagrophytes*, *Trichophyton rubrum*, *Trichophyton violaceum*, *Microsporum canis*, *Microsporum gypseum*, *Epidermophyton floccosum*, *Penicillium notatum*, *Cochliobolus heterostrophus*) e leveduras (*Candida albicans*, *Candida glabrata*, *Candida tropicalis*, *Candida krusei*, *Candida guilliermondii*, *Candida parapsilosis*, *Saccharomyces cerevisiae*, *Cryptococcus neoformans*) (DUEK et al. 2001). Esta espécie demostrou que quando esporos de *Aspergillus flavus* estavam aderidos ao corpo do ácaro houve maior crescimento e multiplicação deste fungo. Concluiu-se que esta espécie acarina ajuda a dispersar esporos de fungos em amostras não contaminadas de milho, demostrando assim haver simbiose entre determinadas espécies de ácaros e fungos (FRANZOLIN *et al*., 1999; JEON *et al*., 2009).

Entretanto, bolores e leveduras, quando presentes em número elevado, podem provocar a deterioração do alimento ou a redução de sua vida de prateleira. Além disso, alguns gêneros de bolores, ao se multiplicarem podem produzir metabólitos tóxicos conhecidos por micotoxinas que podem prejudicar a saúde animal e humana, e também por prejuízos econômicos (ATUÍ & LÁZZARI, 1998). A legislação brasileira determina os padrões de micotoxinas que devem ser avaliadas bem como os limites máximos permitidos em cada tipo de alimento/produto (AGÊNCIA NACIONAL DE VIGILÂNCIA SANITÁRIA, 2011). Espécies do gênero *Tyrophagus* podem colonizar ambientes criados pelo homem, tal como casas, poeira em ambientes urbanos, estruturas laboratoriais e médicas, fazendas, indústrias de alimentos (FRANZ *et al*., 1997; SPIEKSMA, 1997; SOLARZ *et al*., 2007). Alimentos armazenados de forma imprópria fornecem condições para o crescimento de diversas espécies de fungos (SINHA *et al*., 1979; MAGAN *et al*., 2003; HOPE *et al*., 2005; OSBORNE & STEIN, 2007). Sobre estas condições insatisfatórias de armazenamento, os gêneros fúngicos *Fusarium*, *Penicillium* e *Aspergillus* são os mais comuns (BALIUKONIENE *et al*., 2003; NESVORNÁ *et al*., 2012). As constantes migrações de populações de ácaros entre

ecossistemas de armazenamento contribuem eficientemente para a dispersão de esporos fúngicos viáveis, como *Aspergillus* spp. e *Penicillium* spp., sendo assim considerados como carreadores através da superfície de seu corpo ou pelas suas fezes (VAN BRONSWIJK & SINHA, 1973; LUNG-SHU, 1984; SAINT, 1984; FRANZOLIN *et al*., 1999). Fungos filamentosos e leveduras são amplamente distribuídos pelo ar, poeira, flora e fauna e podem levar a doenças em diferentes vertebrados. Micoses humanas são causadas por diversos patógenos (ex: *Epidermophyton* spp., *Microsporum* spp., *Trichophyton* spp., *Sporothrix* spp.) e outros considerados oportunistas (ex: *Aspergillus fumigatus*, *Candida albicans, Rhizopus* spp.). Seus esporos são importantes agentes alergênicos e os ácaros são uma rota de transmissão. Fungos filamentosos (bolores) produzem micotoxinas que são um conjunto complexo de substâncias tóxicas, que dependendo da concentração presente nos alimentos e rações podem levar a graves problemas à saúde humana e animal (MOSS, 1998).

Tyrophagus putrescentiae é geralmente disseminado por roedores, aves e insetos nos depósitos e armazéns, ou infestam os grãos vindos do campo a esses ambientes (OLSEN, 1983). *Tyrophagus putrescentiae* pode alimentar-se de diferentes estágios evolutivos de insetos, assim como de ácaros e de nematoides, mas normalmente é fungívoro (GERSON *et al*., 2003). O controle destes acaros em armazéns é geralmente dificultado pelo fato deles passarem despercebidos, devido ao tamanho reduzido (SOUSA *et al*., 2005). Quando detectados podem ser controlados através da alteração das condições de temperatura e umidade nas estruturas de armazenamento (LORINI, 1998). Esses ácaros também podem ser controlados com inseticidas fumigantes, como a fosfina (FIELDS & WHITE, 2002; KIM *et al*., 2004), além de piretróides e organofosforados (ŽDÁRKOVA, 1994). O controle com ácaros predadores das famílias Cheyletidae e Ascidae tem sido estudado também para controle biológico em grãos armazenados (ŽDÁRKOVA & FEIT, 1999; REZK, 2000; GRANICH *et al*., 2016), bem como o tratamento de grãos com óleos vegetais (SÁNCHEZ RAMOS & CASTAÑERA, 2001). Inseticidas e acaricidas sintéticos como a fosfina, benzil benzoato e *N,N*-dietil-*m*-toluamida têm sido amplamente usados no controle de pragas em grãos armazenados devido sua alta eficiência, e são a principal forma de controle curativo de ácaros em armazéns (LEE *et al*., 2006). Entretanto, a utilização frequente por longos períodos de tempo de alguns inseticidas tem resultado na seleção de populações resistentes de ácaros a inseticidas como pirimifós metil e

fenitrotion (SZLENDAK *et al.,* 2000). Além disso, o mau uso destes produtos também tem causado efeitos indesejáveis sobre organismos não-alvo (HAYS Jr. & LAWS Jr., 1991; ROEL, 2001). Desta forma, é muito importante que estudos sobre o controle biológico destes ácaros sejam intensificados para contribuir com mais conhecimentos científicos na área.

4. MATERIAL E MÉTODOS

4.1 Metodologias aplicadas

Toda metodologia empregada nesta tese está inserida nos artigos abaixo.

5. ARTIGOS

5.1. ARTIGO 1 - Silva GL, Metzelthin MH, Da-Costa T, Rocha MS, Silva DE, Ferla NJ, Silva OS. 2017. Responses of water mite assemblages (Acari) to environmental parameters at irrigated rice cultivation fields and native lakes. Zoologia (Curitiba). 34: e19988.

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RESEARCH ARTICLE

Responses of water mite assemblages (Acari) to environmental parameters at irrigated rice cultivation fields and native lakes

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http://zoobank.org/962071A9-11FA-4DE9-90DF-D80AD3493C69

ABSTRACT. Many studies have revealed that water mite communities can be affected by the physical and chemical parameters of the water. The similarity between the water 'mite assemblages in local water bodies and in irrigated rice areas can be a way to measure the water conditions, enabling an assessment of the anthropic impact in the environment. The aim of this study was to evaluate the distribution of water mites in lakes and irrigated rice fields in south Brazil. To accomplish that we characterized the distinctive environments using physical and chemical variables such as pH, turbidity (NTU), water temperature (ºC) and dissolved oxygen (mg/L), in order to verify the influence of these abiotic factors on the species composition of water mite communities; and to compare water mite abundance, richness and composition among different habitats. We assessed three native lakes and four sites with irrigated rice cultivation. Our results showed, for the first time in Brazil, strong correlations between the water mite fauna and turbidity. In addition, native lakes were richer and had greater mite abundance when compared with the irrigated rice areas.

KEY WORDS. Coastal plain, habitat, physical and chemical variables, rice field, water mite.

INTRODUCTION

Most freshwater mites belong to Hydrachnidiae (Acari: Prostigmata), which are represented by ca. 6,000 species. These mites can live in wetlands, temporary pools, springs, marine habitats, torrential waterfalls, ponds, streams and lakes (Smith and Cook 1991, Goldschmidt 2016). Another important acarine group that inhabits aquatic zones includes members of the suborder Oribatida. These mites can be abundant, but their species richness is very low (Fernandez and Athias-Binche 1986). Water mites have been used as environmental quality bioindicators in several ecosystems (Biesiadka and Kowalik 1991, Rousch et al. 1997, Di Sabatino et al. 2002, Dohet et al. 2008, Goldschmidt 2016). Many studies have revealed that water mite communities can be affected by several abiotic factors, such as pollution agents and variations in temperature, dissolved oxygen, conductivity and carbonate concentrations (Schwoerbel 1961, Wi cek et al. 2013). Many efforts are being made to effectively employ mites as ecological indicators, including faunal monitoring programs (Goldschmidt 2016).

Brazil is the ninth leading rice grower in the world, and the state of Rio Grande do Sul accounts for approximately 69% of country's rice production (IBGE 2015). With the addition and growth of areas of irrigated crop production such as rice, monitoring water quality has acquired growing importance. In Brazil, the Restinga in the South is one of the regions where there is massive irrigated rice cultivation. This region includes a portion of the Atlantic forest biome and it is under extreme anthropogenic pressure, which has modified the natural landscapes (Ab'Saber 2003). The decline in water quality resulting

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from intense contamination has resulted in decreasing numbers of water mites, lower water mite diversity and in some cases, a collapse of the water mite fauna (Kowalik and Biesiadka 1981).

In general, the water used to irrigate crops comes from several freshwater lakes and ponds located in the vicinities of crop fields. Therefore, comparisons between the water mite assemblages from native lakes and irrigated rice areas can be used to measure water quality, enabling an assessment of the impact of agriculture on the environment.

The composition of water mite species in irrigated rice areas is generally poorly understood. This work is the first trying to ascertain the water mite community in such an environment. This study has two goals: (1) to compare the abundance, richness and composition of the water mite fauna from different habitats; (2) to verify the influence of the following abiotic factors on species composition: pH, turbidity (NTU), water temperature (°C), and dissolved oxygen (mg/L). Our hypotheses are: (1) Different habitats should influence the abundance, richness and species composition of the water mite community; (2) water mites should respond to the physical and chemical parameters measured, reflecting the different conditions of their sample sites.

MATERIAL AND METHODS

Our study was conducted in the municipality of Mostardas, Rio Grande do Sul, Brazil. The study area is situated in the coastal plains (Table 1) (Assis et al. 2011, Magnago et al. 2010). The original vegetation cover is open. It is dominated by herbaceous plants and shrubs that have xerophytic adaptations (Assis et al. 2011, Lima et al. 2011), together with other associated Atlantic forest elements (Magnago et al. 2010).

All rice areas from which samples were taken for this survey had been cultivated for three months, and were in the mid to late vegetative phase (tillering to stem elongation) to mid to late reproductive phase (heading to flowering), when the rice fields were flooded. We chose seven sampling areas: four at an irrigated rice cultivation area (R1, R2, R3, R4), in which sampling was carried out during the rice growing season (January-March/15); and three in native lakes (L1, L2, L3) (Figs 1–9). Both L2 and L3 are connected by a narrow water channel. The rice areas R1 and R2 are supplied by L2 and L3 (distance among rice areas to native lakes: 1 km) placed in Farm 1, whereas R3 and R4 are supplied by the northern portion of a lagoon known as Lagoa dos Patos. This lagoon is separated from the Atlantic Ocean by a barrier beach system. According to Madeira-Falceta (1974), the salinity levels in the northern part of this lagoon are very low (freshwater). The L1 area is a permanent native lake, isolated from human disturbances and away from rice areas. The distance from R3, R4 to L1 is 2 km, the former two located in Farm 2. The distance between these two farms (Farm 1 and 2) is 5 km.

The native lake areas had the following characteristics: permanent lakes, and surrounding vegetation consisting basically of grasses. Cattle rearing and fishing are a very common practice near L2 and L3. Additionally, there are aquatic macrophytes (*Salvinia* spp., *Pistia* spp., *Eleocharis* spp., *Lemma* spp.) often covering the surface of the water.

Table 1. Seven sampled areas with geographical coordinates and numbers of samples collected between January to March 2015.

Site name	Longitude	Latitude	Number of samples	Altitude (m)
Rice area 1 (R1)	30°33'55.29"S	50°36'39.76"W	4	4
Rice area 2 (R2)	30°34'14.75"S	50°36'35.52"W	4	5
Rice area 3 (R3)	30°35'25.13"S	50°39'25.91"W	4	9
Rice area 4 (R4)	30°35'30.68"S	50°39'18.85"W	4	7
Lake 1 (L1)	30°35'11.29"S	50°40'38.92"W	4	5
Lake 2 (L2)	30°33'15.40"S	50°36'51.10"W	4	0
Lake 3 (L3)	30°33'39.88"S	50°36'49.02"W	4	1

Figure 1. Schematic map of Brazil and Rio Grande do Sul State illustrating the study area.

Samplings were carried out from January to March 2015, two per month. In March, only one sampling was performed, on the first week of the month. A total of five samplings were carried out at each area. For the assessment of species' composition, only adult mites were evaluated. For this reason, L1, L2, L3, R1 were used in all data analyses, whereas only three samplings from rice areas R2 and R3, and four from R4, were analyzed.

Samples were collected five meters from the margin of the lake and rice field at maximum of 40 cm depth. Each sample consisted of 10 liters of water collected using a plastic tray (50 x 30 cm), and filtered through a net (mesh size 250 µm). The water mites caught in the net were transported to the laboratory in water gallons, and were later preserved in Koenike's fluid (Mitchell and Cook 1952, Barr 1973).

Specimens were identified to species using a phase-contrast light microscope (Leica DM750) with the help of identification keys (Rosso de Ferradás and Fernández 2009, Smith et al. 2009). Specimens collected were deposited in the mite reference collection of the Museum of Natural Sciences of the University Center UNIVATES (ZAUMCN), Lajeado, Rio Grande do Sul, Brazil.

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Figures 2–9. View of the study sites: (2) Detailed view of the rice-water samples, (3) Rice area 1 – R1, (4) Rice area 2 – R2, (5) Rice area 3 – R3, (6) Rice area 4 – R4, (7) Lake 1 – L1, (8) Lake 2 – L2, (9) Lake 3 – L3.

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We measured the values of water temperature, turbidity, pH and dissolved oxygen using portable instruments (DM-2P; DM-4P; DM-TU: Digimed) and all variables were measured when and where samples were collected. There were no records of phytosanitary treatments with pesticides during the study, in the sampling areas.

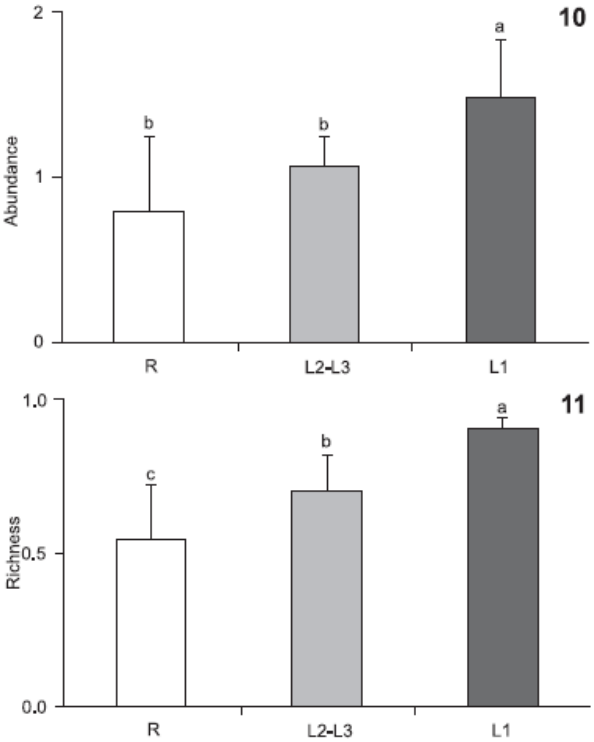
At each sampling point, species composition was analyzed considering the abundance of each species per location (quantitative data). In order to investigate whether abundance and richness of mite species varied according to the different areas, factor analyses of variance (Factorial ANOVA) were performed. These areas were characterized in three clusters of environments (R1-R4, L2-L3, L1). Based on these quantitative data, we obtained an association matrix between sampling points using Bray-Curtis similarity indices.

Using distance matrices calculated based on mite species composition, we performed an ordination using a Non-Metric Multidimensional Scaling (NMDS) with Bray-Curtis (quantitative) and Jaccard (qualitative) distance and two dimensions to visualize how species composition varied between environments (Rice Areas R1-R4 vs. Native lakes L2-L3 vs. L1). Species abundance and values of abiotic and biotic factors were Log transformed (x+1) and subsequently normalized and centralized through vectored transformations. Additionally, in order to reveal the effects of the environment on community dissimilarity, we tested whether abiotic and biotic factors adjusted to the ordination model (NMDS) by using the Envfit function. The One-factor Similarity Analyses (ANOSIM) (Clarke and Warwick 2001) tested for statistical differences in the composition of communities from different environments. SIMPER analysis (Similarity Percentage) was applied to assess which species contributed to the similarity/dissimilarity among environments (Clarke and Warwick 2001). The contribution amounts of each species were expressed as a percentage. Transformations of abundances and values of environmental factors were calculated using MULTIV software (Pillar 2004). Factorial ANOVA's and linear regressions were verified using the statistical program SYSTAT 13 (Systat Inc.). ANOSIM and SIMPER multivariate analyses were carried out using the PASt program (Paleontological Statistics, version 1.97) (Hammer et al. 2009). Mantel's, NMDS and Envfit analyses were performed using the R program, "vegan" package. The significance level was $p < 0.05$ for all statistical tests.

RESULTS

We found a total of 514 water mites, 477 were adults, distributed in 9 families, 10 genera and 19 species/morphospecies (Table 2).

The most abundant water mite morphospecies were *Koenikea* sp. 1 (Unionicolidae) (179 specimens), followed by *Limnesia* sp. 1 (84) (Limnesiidae) and *Koenikea* sp. 3 (38). Among the areas evaluated, L1 was the richest (15 species), followed by L2 (10) and L3 (9); and the order of abundance was L1 (197 specimens) followed by R2 (88) and L2 (61).



Figures 10–11. Abundance and richness of mites in rice areas cultivation and native lakes: (10) abundance adults (± SD) (Log10 X+1); (11) richness (± SD) (Log10 X+1). Different letters indicate significant differences, Tukey test, $p < 0.05$.

The abundance (N) of adult mites differed among the environments surveyed ($F_{2,27} = 6.871$, $p = 0.004$). The Tukey post-hoc test revealed differences between L1 and the rice areas (R1-R4) ($p = 0.003$), but not between L1 and L2, L1 and L3 ($p = 0.117$); thus, R1-R4 and L2-L3 ($p = 0.178$) (Fig. 10). Regarding richness, there was a significant difference among environments ($F_{2,27} = 11.663$, $p < 0.001$). The Tukey post-hoc test revealed a difference in richness between L1 and the rice areas (R1-R4) ($p < 0.001$), L1 with L2-L3 ($p = 0.005$) and R1-R4 with L2-L3 ($p = 0.003$) (Fig. 11).

The mite fauna composition among environments was significantly different (Bray-Curtis: $R2 = 0.2657$, $p = 0.01$; Jaccard: $R2 = 0.1551$, $p < 0.001$) (Figs 12, 13). The post-hoc test showed differences in mite composition among L1 and the rice areas (R1-R4) (Bray-Curtis, $p = 0.002$; Jaccard, $p = 0.003$) and among L1 and L2, L3 (Bray-Curtis, $p = 0.004$; Jaccard, $p = 0.002$), but not among R1-R4 and L2-L3 (Bray-Curtis, $p = 0.2946$; Jaccard, $p = 0.3934$). Through SIMPER analysis, the *Limnesia* sp. 4 (Limnesiidae) (Contrib.% 20.1), Oribatulidae (Contrib.% 16.44) and *Arrenurus* sp. 1 (Arrenuridae) (Contrib.% 14.35) with higher val-

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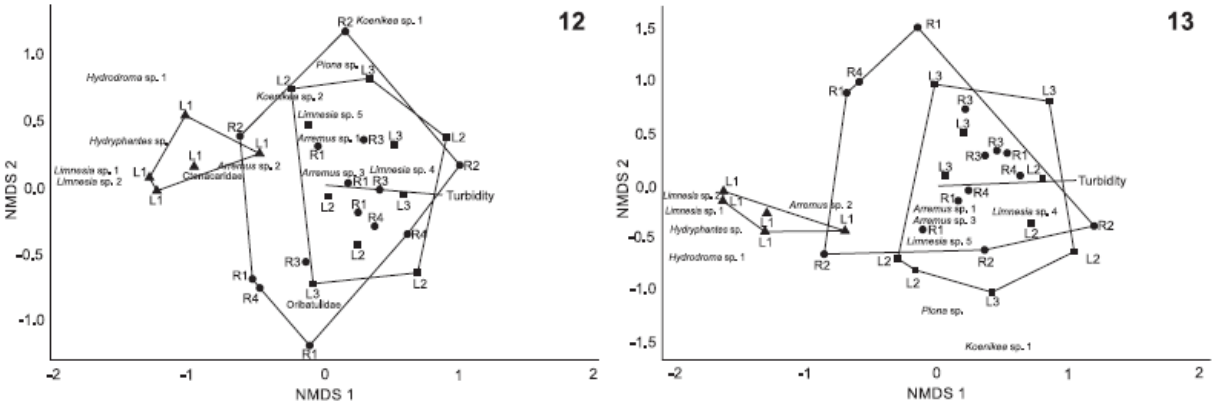
Table 2. List of water mite species and number of individuals collected (January to March/2015) in three lakes (L1, L2, L3) and four irrigated rice area cultivation (R1, R2, R3, R4) in Southern Brazil.

Order	Family	Species	Rice area 1 (R1)	Rice area 2 (R2)	Rice area 3 (R3)	Rice area 4 (R4)	Lake 1 (L1)	Lake 2 (L2)	Lake 3 (L3)	Total
Trombidiformes	Arrenuridae	*Arrenurus* sp. 1	7	3	6	–	5	4	7	32
		Arrenurus sp. 2	–	–	–	1	5	2	–	8
		Arrenurus sp. 3	–	–	–	–	1	1	–	2
	Eylaidae	*Eylais* sp.	–	–	–	–	1	–	–	1
	Hydrodromidae	*Hydrodroma* sp. 1	–	–	–	–	36	–	–	36
		Hydrodroma sp. 2	–	–	–	–	1	–	–	1
	Hydryphantidae	*Hydryphantes* sp.	–	–	–	–	5	–	–	5
	Limnesiidae	*Limnesia* sp. 1	6	2	18	11	2	21	24	84
		Limnesia sp. 2	–	–	–	–	1	2	4	7
		Limnesia sp. 3	–	1	1	–	–	1	4	7
		Limnesia sp. 4	–	–	–	–	–	–	4	4
		Limnesia sp. 5	–	–	–	–	–	–	2	2
	Pionidae	*Piona* sp.	5	–	3	8	–	11	3	30
	Unionicolidae	*Koenikea* sp. 1	3	47	1	2	121	3	2	179
		Koenikea sp. 2	–	4	–	–	8	–	–	12
		Koenikea sp. 3	1	31	1	3	1	1	–	38
		Neumania sp.	–	–	–	–	1	-	–	1
Sarcoptiformes	Oribatulidae	sp.	–	–	–	–	6	15	4	25
	Ctenacaridae	sp.	–	–	–	–	3	–	–	3
	Total		22	88	30	25	197	61	54	477



Figures 12–13. Ordination diagram (first two axes) of Non-Metric Multidimensional Scaling (NMDS) using (12) Bray-Curtis and (13) Jaccard indexes with Envfit function for the evaluated environments. (▲ Lake 1, ■ Lake 2 and 3, ● Rice Area). Stress: 0.15.

ues of abundance R1-R4 and at L2-L3, Ctenacaridae (Contrib.% 16.07) and *Hydryphantes* sp. (Hydryphantidae) (Contrib.% 10.31) contributed the most to the dissimilarity among environments. Thus, *Arrenurus* sp. 2 (Contrib.% 4.721) and *Limnesia* sp. 2 (Contrib.% 2.085) had higher values of abundance in L1.

Among the evaluated environmental parameters (Table 3), the Envfit analysis demonstrated that turbidity (r2 = 0.2725, p = 0.01) was the only factor that contributed substantially to variations in the water mite communities of the aquatic environment. Additionally, among the environmental parameters evaluated only turbidity was statistically significant ($F_{2,27}$ = 6.292, p = 0.006). The greatest turbidity levels were measured in L2, L3, followed by R1-R4 and finally, L1, which had the lowest turbidity levels.

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Table 3. Physiochemical variables (mean ± SE) evaluated from different habitats in this study (Jan-Mar/15).

Site name	pH	Water temperature (°C)	O_2 dissolved (mg/l)	Turbidity (NTU)
Rice area 1 (R1)	7.73 ± 0.5	26.13 ± 1.49	5.08 ± 0.86	41.43 ± 20.33
Rice area 2 (R2)	7.56 ± 0.5	26.5 ± 1.58	4.8 ± 0.88	46.89 ± 46.05
Rice area 3 (R3)	7.51 ± 0.9	28.38 ± 2.14	5.55 ± 1.64	31.42 ± 33.48
Rice area 4 (R4)	7.67 ± 0.7	29.88 ± 3.28	6.25 ± 1.37	7.05 ± 2.46
Lake 1 (L1)	8.31 ± 0.2	28.5 ± 1.58	5.2 ± 0.84	5.80 ± 6.51
Lake 2 (L2)	8.09 ± 0.6	25.5 ± 1.68	3.48 ± 0.71	29.14 ± 10.67
Lake 3 (L3)	8.01 ± 0.9	26.63 ± 1.89	5 ± 1.07	43.1 ± 18.65

DISCUSSION

We found differences in the composition of the water mite communities between rice areas and the isolated native lake in which the parameter turbidity influenced the composition of the population of water mites. In addition, it was possible to observe that the native lakes carry a greater richness of water mite species when compared with irrigated rice areas. The greatest number of species was found in native ponds, not in rice areas. *Eylais* sp., *Hydrodroma* species, *Neumania* sp. and Ctenacaridae were exclusively collected in L1, *Limnesia* sp. 3 and *Limnesia* sp. 4 in L3. Only two species were present in all samples sites, *Koenikea* sp. 1 and *Limnesia* sp. 1, while all species that were present in rice areas were also found simultaneously in one of the native lakes. No species occurred exclusively in rice areas.

The greater abundance and richness of water mites in L1 may be due to the low levels of turbidity, and the fact that this lake is isolated and does not suffer the impact of human action. The turbidity of the water is caused by the suspended sediment. These can originate from the organic input from microorganisms, including bacteria and algae, and external input from leaf litter and debris, and decaying carcasses of invertebrates (Laessle 1961, Maguire Jr 1971). In addition, high turbidity levels can decrease the penetration of light and reduce the water quality, significantly reducing the water resources utilized by these mites (Copatti et al. 2013). Some of the water mite species are strongly positively phototropic; however, each family responds differently to various wavelengths (Roberts et al. 1978). No relationships were found between the composition of water mite species and the other physical and chemical variables such as pH, dissolved O_2 and water temperature. Więcek et al. (2013) found a strong correlation between the water mite fauna, and the environment variables such as conductivity and pH gradient; however, those authors did not measure turbidity. Both pH and conductivity can be used to assess the mineral richness of the water.

An explanation for a higher richness of water mites at certain sites than at others is the differing dispersion ability of these arthropods. The dispersion process is very important, since it allows the expansion of mite populations, the colonization of different areas, and escaping from natural enemies (Binns 1982, Zawal et al. 2013, Knee et al. 2013). Most larval water mites are parasites of insect, which facilitates their dispersal (Smith et al. 2009, Williams and Proctor 2002). The phoretic behavior observed in several families of water mites allows for a passive transport of dispersion (Zawal et al. 2013). There are several records of *Arrenurus*, *Hydrachna* and *Limnochares* involving parasitized insects (Munchberg 1954, Mullen 1975, Stechmann 1980, Smith and Cook 1991, Snell and Heath 2006, Milne et al. 2009, Kirkhoff et al. 2013). It is possible that the absence of mite host taxa could be responsible for the low number of mites. Another, simpler explanation, is that richness has decreased in response to environmental changes.

Furthermore, we suggest that the dispersion pathways used by water mites to colonize adjacent environments are related to the route of the flow paths, as it can be observed in many species collected in this study (*Arrenurus* sp. 1, *Limnesia* sp. 1, *Limnesia* sp. 3, *Piona* sp., *Koenikea* sp. 1, *Koenikea* sp. 3) and which were found simultaneously in the native lake areas (L2-L3) that supply the rice areas (R1-R2).

Several species of water mites exploit aquatic plants, since these plants create the substrate required for the life cycles of water mites and their hosts. Several water mites lay eggs and transform from deutonymphs to tritonymphs among aquatic mosses and macrophytes (Smith et al. 2009). Macrophyte plants offer suitable conditions for various host taxa, playing a large role in the dispersion and colonization of new patches by water mites (Martin 2008). Therefore, the presence of macrophytes in general in lakes probably is probably a factor influencing water mite diversity. Wi cek et al. (2013) reported that some species of *Arrenurus* are associated with abundant growth of macrophytes.

The conservation of native lakes is very important to preserve biodiversity. Thus, aquatic mites might be used as diversity bioindicators when comparing natural and anthropized environments. Young (1969) described that water mite communities (diversity, abundance and community structure) are sensitive to contamination. Several sensitive species are restricted to very clean water and immediately respond to the early contamination of their environment. Thus, water mites are excellent indicators of pure water conditions and provide a powerful early warning system (Zawal 1996, Miccoli et al. 2013). Species of the genus *Limnesia* are very sensitive to altered sites (Van der Hammen and Smit 1996) and, according to our study, *Limnesia* sp. 4 showed greater dissimilarity in R1-R4 and L2-L3 environments while Ctenacaridae, *Hydryphantes* sp., *Arrenurus* sp. 2 and *Limnesia* sp. 2 to L1.

The abundance and diversity of water mites was significantly higher in unpolluted sites (Growns 2001). Water mites can be powerful biomonitoring tools, fulfilling all adequate requirements for a bioindicator (Goldschmidt 2016). Our results confirm that anthropized areas have lower richness of water mites than native areas, as suggested by Katayama et al. (2015). They observed a decline in richness and abundance at a consolidated rice field. In the future, it is possible that water mites will be implemented in faunal surveys as an important group for biomonitoring, be-

coming an important tool in areas of environmental protection where there are watercourses. The water mite community is not sufficiently known yet. Considering Brazil's vast territory with large affluent and unexplored areas, it would be important to carry out more studies in the country. This study compared, for the first time, the water mite species from native lakes associated with irrigated rice areas in southern Brazil.

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LITERATURE CITED

Ab'Saber AN (2003) Os domínios de natureza no Brasil: potencialidades paisagísticas. Ateliê Editorial, São Paulo.

Assis MA, Prata EMB, Pedroni F, Sanchez M, Eisenlohr PV, Martins FR, Santos FAM, Tamashiro JY, Alves LF, Vieira SA, Piccolo MC, Martins SC, Camargo PB, Carmo JB, Simões E, Martinelli LA, Joly CA (2011) Florestas de restinga e de terras baixas na planície costeira do sudeste do Brasil: vegetação e heterogeneidade ambiental. Biota Neotropica 11: 103–121. https://doi.org/10.1590/S1676-06032011000200012

Barr D (1973) Methods for the Collection, Preservation and study of Water Mites (Acari: Parasitengona). Royal Ontario Museum, Life Sciences Division, Toronto.

Biesiadka E, Kowalik W (1991) Water mites (Hydracarina) as indicators of trophy and pollution in lakes. In: Dusbabek F, Bukva V (Eds) Modern Acarology 1. SPB Academic Publishing BV, The Hague, 475–481.

Binns ES (1982) Phoresy as migration some functional aspects of phoresy in mites. Biological Reviews 57: 571–620. https://doi.org/10.1111/j.1469-185X.1982.tb00374.x

Clarke KR, Warwick RM (2001) Change in marine communities: an approach to statistical analysis and interpretation. PRIMER-E, Plymouth, UK.

Copatti CE, Fagundes LS, Quaini JB, Copatti BR (2013) Diversity of aquatic arthropods on *Eichhornia crassipes* (Mart.) Solms roots before and after removal of substrate in a reservoir in southern Brazil. Pan-American Journal of Aquatic Sciences 8: 265–275.

Di Sabatino A, Martin P, Gerecke R, Cicolani B (2002) Hydrachnidia (water mites). In: Rundle SD, Robertson AL, Schmid-Araya JM (Eds) Freshwater meiofauna: biology and ecology. Backhuys Publishers, Leiden, 105–133.

Dohet A, Ector L, Cauchie H, Hoffmann L (2008) Identification of benthic invertebrate and diatom indicator taxa that distinguish different stream types as well as degraded from reference conditions in Luxembourg. Animal Biology 58: 419–472. https://doi.org/10.1163/157075608X383719

Fernandez NA, Athias-Binche F (1986) Analyse demographique d'une population d'Hydrozetes lemnae Coggi, Acarien Oribate infeode a la lentille d'eau *Lemna gibba* L. en Argentine. I. Methodes et techniques, demographie d' *H. lemnae* comparaisons avec d'autre Oribates. Zoologisches Jahrbuch Systematik 113: 213–228.

Goldschmidt T (2016) Water mites (Acari, Hydrachnidia): powerful but widely neglected bioindicators – a review. Neotropical Biodiversity 2: 12–25. https://doi.org/10.1080/23766808.2016.1144359

Growns JE (2001) Aquatic mites as bioindicators, with an Australian example. In: Halliday RB, Walter DE, Proctor H et al. (Eds) Acarology: proceedings of the 10th International Congress. CSIRO Publishing, Melbourne, 136–142.

Hammer O, Harper DAT, Ryan PD (2009) Past: Palaeontological Statistics software package for education and data analysis. Palaeontologia Electronica 4: 1–9.

IBGE (2015) Produção agrícola. Available online at: http://www.ibge.gov.br [Accessed 12/11/2015]

Katayama N, Baba YG, Kusumoto Y, Tanaka K (2015) A review of post-war changes in rice farming and biodiversity in Japan. Agricultural Systems 132: 73–84. https://doi.org/10.1016/j.agsy.2014.09.001

Kirkhoff CJ, Simmons TW, Hutchinson M (2013) Adult mosquitoes parasitized by larval water mites in Pennsylvania. Journal of Parasitology 99: 31–39. https://doi.org/10.1645/GE-3105.1

Knee W, Fober MR, Beaulieu F (2013) Diversity and host use of mites (Acari: Mesostigmata, Oribatida) phoretic on bark beetles (Coleoptera: Scolytinae): global generalists. Annals of the Entomological Society of America 106: 339–350. https://doi.org/10.1603/AN12092

Kowalik W, Biesiadka E (1981) Occurence of water mites (Hydracarina) in the river Wieprz polluted with domestic industry sewage. Acta Hydrobiologica 23: 331–348.

Laessle AM (1961) A micro-limnological study of Jamaican bromeliads. Ecology 42: 499–517. https://doi.org/10.2307/1932236

Lima RAF, Oliveira AA, Martini AMZ, Sampaio D, Souza VC, Rodrigues RR (2011) Structure, diversity, and spatial patterns in a permanent plot of a high Restinga forest in Southeastern Brazil. Acta Botanica Brasilica 25: 633–645. https://doi.org/10.1590/S0102-33062011000300017

Madeira-Falceta M (1974) Ecological distribution of thecamoebal and foraminiferal associations in the mixohaline environments of the Southern Brazilian Littoral. Anais da Academia Brasileira de Ciências 46: 667–687.

Magnago LFS, Martins SV, Schaefer CEGR, Neri AV (2010) Gradiente fito fisionômico-edáfico em formações florestais de Restinga no Sudeste do Brasil. Acta Botanica Brasilica 24: 734–746. https://doi.org/10.1590/S0102-33062010000300017

Maguire Jr B (1971) Phytotelmata: Biota and community structure determination in plant-held waters. Annual Review of Ecology and Systematics 2: 439–464. https://doi.org/10.1146/annurev.es.02.110171.002255

Martin P (2008) Water mites (Hydrachnidia, Acari) and insects: a survey of a seldom considered relationship. Entomologie heute 20: 45–75.

Miccoli FP, Lombardo P, Cicolani B (2013) Indicator value of lotic water mites (Acari: Hydrachnidia) and their use in macroinvertebrate-based indices for water quality assessment purposes. Knowledge and management of aquatic ecosystems 411: 1–28. https://doi.org/10.1051/kmae/2013075

Milne MA, Townsend VJ, Smelser P, Felgenhauer BE, Moore MK, Smyth FJ (2009) Larval Aquatic and Terrestrial Mites Infesting a Temperate Assemblage of Mosquitoes. Experimental and Applied Acarology 47: 19–33. https://doi.org/10.1007/s10493-008-9194-2

Mitchell RD, Cook DR (1952) The preservation and mounting of water-mites. Turtox News 30: 1–4.

Mullen GR (1975) Acarine Parasites of Mosquitoes I. A Critical Review of All Know Records of Mosquitoes Parasitized by Mites (Acarina: Hydrachnellae). Journal of Medical Entomology 12: 27–36. https://doi.org/10.1093/jmedent/12.1.27

Munchberg P (1954) Zur Kenntnis der an culiciden (Diptera) schmarotzenden *Arrenurus*-Larven (Hydracarina), so-wie ueber die Bedeutung dieser Parasitenfuer Wirt und Mensch. Zeitschrift für Parasitenkunde 16: 298–312. https://doi.org/10.1007/BF00260194

Pillar VDP (2004) MULTIV: Aplicativo para análise multivariada e testes de hipóteses. Porto Alegre, Departamento de Ecologia, UFRGS. Available online at: http//:ecologia.ecoqua.ufrgs.br [Accessed 15/07/2016]

Roberts EA, Dimock Jr RV, Forward Jr RB (1978) Positive and Host-Induced Negative Phototaxis of the Symbiotic Water Mite *Unionicola formosa*. Biological Bulletin of the Marine Laboratory 155: 599–607. https://doi.org/10.2307/1540793

Rosso de Ferradás B, Fernández HR (2009) Acari, Parasitengona, Hydrachnidia. In: Domínguez E, Fernández HR (Eds) Macroinvertebrados bentônicos sudamericanos. Fundación Miguel Lillo, Tucumán, 497–549.

Rousch JM, Simmons TW, Kerans BL, Smith BP (1997) Relative acute effects of low pH and high iron on the hatching and survival of the water mite, *Arrenurus manubriator* and aquatic insect, *Chironomus riparius*. Environmental Toxicology and Chemistry 16: 2144–2150. https://doi.org/10.1002/etc.5620161022

Schwoerbel J (1961) Die Bedeutung der Wassermilbenfür die biozönotische Gliederung. Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte Limnologie 14: 355–361.

Smith IM, Cook DR (1991) Water Mites. In: Covich T (Ed.) Freshwater Invertebrates from North American. Academic Press, Toronto, 523–592.

Smith IM, Cook DR, Smith BP (2009) Water mites (Hydrachnida) and other arachnids. In: Thorp JH, Covich AP (Eds) Ecology and classification of North American freshwater invertebrates (3. ed). Academic Press, San Diego, 485–586.

Snell AE, Heath ACG (2006) Parasitism of Mosquitoes (Diptera: Culicidae) by Larvae of Arrenuridae and Microtrombidiidae (Acari: Parasitengona) in the Wellington Region, New Zealand. New Zealand Journal of Zoology 33: 9–15. https://doi.org/10.1080/03014223.2006.9518426

Stechmann DH (1980) Zum Wirtskreis syntopischer *Arrenurus*-Arten (Hydrachnellae, Acari) mit parasitischer Entwicklung an Nematocera (Diptera). Zeitschrift für Parasitenkunde 62: 267–283. https://doi.org/10.1007/BF00926567

Van der Hammen H, Smit H (1996) The water mites (Acari: Hydrachnidia) of streams in The Netherlands: distribution and ecological aspects on a regional scale. Netherland Journal of Aquatic Ecology 30: 175–185. https://doi.org/10.1007/BF02272237

Young WC (1969) Ecological distribution of Hydracarina in North Central Colorado. The American Midland Naturalist Journal 82: 367–401. https://doi.org/10.2307/2423785

Więcek M, Martin P, Gąbka M (2013) Distribution patterns and environmental correlates of water mites (Hydrachnidia, Acari) in peatland habitats. Experimental and Applied Acarology 61: 147–160. https://doi.org/10.1007/s10493-013-9692-8

Williams CR, Proctor HC (2002) Parasitism of Mosquitoes (Diptera: Culicidae) by Larval Mites (Acari: Parasitengona) in Adelaide, South Australia. Australian Journal of Entomology 41: 161–163. https://doi.org/10.1046/j.1440-6055.2002.00274.x

Zawal A (1996) Wodopójki (Hydracarina) jakobioindykatory. Water mites as bioindicators. Uniwersytet Szczeciński Materiały Konferencje 19: 229–235.

Zawal A, Çamur-Elipek B, Fent M, Kirgiz T, Dzierzgowska K (2013) First Observations in Turkish Thrace on Water Mite Larvae Parasitism of *Ranatra linearis* by *Hydrachna gallica* (Acari: Hydrachnidia). Acta Parasitologica 58: 57–63. https://doi.org/10.2478/s11686-013-0106-1

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Comparison of biological development of *Blattisocius dentriticus* (Blattisocidae) fed on *Tyrophagus putrescentiae* (Acaridae) and *Megninia ginglymura* (Analgidae)

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ABSTRACT

This study evaluated *Blattisocius dentriticus* (Berlese) as a candidate predator of two mite species of public-health importance for humans and animals. We observed the development time, reproduction, survival and sex ratio of this predatory mite when fed on one of two pest mites, *Tyrophagus putrescentiae* (Schrank) and *Megninia ginglymura* (Mégnin), under laboratory conditions, at a temperature of 25 ± 1°C and 80 ± 5% relative humidity, in the dark. The study started with 30 experimental units containing *M. ginglymura* as a food source and 30 containing *T. putrescentiae*, each with an egg of *B. dentriticus* obtained from fertilized females. The length of the pre-oviposition period was significantly longer when *M. ginglymura* was used as food, compared to *T. putrescentiae*. The life-table parameters for *B. dentriticus* differed on the two diets. These parameters indicated that the population of *B. dentriticus* fed on *T. putrescentiae* increased about 7.53 times (R_o = 7.53) every 14.3 days (T = 14.3), corresponding to a daily population growth of about 15% (λ = 1.15), i.e. a production of 0.14 female per female per day (r_m = 0.14). *B. dentriticus* fed on *M. ginglymura* showed lower values (R_o = 2.79; T = 23.76; λ = 1.04; r_m = 0.04). The gross reproductive rate (GRR) and the proportion of female offspring were higher on *T. putrescentiae* (GRR = 10.73; ratio of females to males = 0.87). Our study demonstrated that *B. dentriticus* is a potential predator, developing and reproducing successfully on the two mites *T. putrescentiae* and *M. ginglymura*. *Blattisocius dentriticus* performed better against *T. putrescentiae*.

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Introduction

Certain species of predatory mites of the family Blattisocidae are candidates for biological-control programmes, particularly *Blattisocius keegani* (Fox), *Blattisocius tarsalis* (Berlese) and *Blattisocius dentriticus* (Berlese) (Acari: Blattisocidae) (Gerson et al. 2003). Species of *Blattisocius* are often reported as predators of pests of stored food, including insects (Britto et al. 2012). *Blattisocius keegani* attacks beetles in stored products and shows potential for the control of the navel orangeworm *Amyelois transitella* Walker (Lepidoptera: Pyralidae) (Thomas et al. 2011). In a flour mill, *B. tarsalis* effectively attacked the flour moth *Ephestia kuehniella* Zeller (Pyralidae) (Nielsen 1998). This mite is also a predator of insect larvae in stored seeds (Treat 1969). *Blattisocius dentriticus* feeds on a variety of arthropod eggs and on *Tyrophagus putrescentiae* (Schrank) (Acari: Acaridae) (Fenilli and Flechtmann 1990). This predator has also been reported in cereals, stored foods and bird nests (Baggio et al. 1987; Silva et al. 2013). Faleiro et al. (2015) found that *B. dentriticus* was the most common predator in traps located in battery cages of poultry houses, predating populations of *Megninia ginglymura* (Mégnin) (Analgidae) that were infesting the hens. *B. dentriticus* has also been found in nests of free-range chickens (Silva et al. 2013).

Determination of the prey type and life cycles are essential steps in evaluating the efficacy of a predator for use in a biological-control programme. However, little is known about the predation capacity and prey type of *B. dentriticus*. Two species of pest mites were chosen in order to evaluate the biological development of *B. dentriticus* when parasitizing its hosts.

One of these pest mites is *M. ginglymura*, an ectoparasite of chicken feathers. This species has been reported mainly from several countries of South America (Cerny 1970, 1973; D'Souza et al. 2001; Sczypel et al. 2003; Quintero et al. 2010). Quintero et al. (2010) reported massive mite infestations that caused extensive feather loss in laying hens. In Brazil, it was registered *M. ginglymura* parasitizing commercial laying hens (Tucci et al. 2005). Among the consequences of the parasitism is particularly the appearance of lesions caused by saliva, itching and scabs formed from serous exudate. Fungal contamination on lesions is also quite common (Tucci et al. 2005). A 20% decrease in yield in egg production has been reported to be caused by parasitism by this group of mites (Van Emous et al. 2005). The other pest mite used was the storage mite *T. putrescentiae*, which is common worldwide in nests of free-range chickens and in stored foods such as flour, wheat, soy, cheese, rye bread, dried eggs, dried fish, dried fruits, edible mushrooms, powdered milk and various seeds (Chmielewski 1999; Duek et al. 2001; Garcia 2004; Kucerova and Horak 2004; Aygun et al. 2007; Palyvos et al. 2008; Mullen and OConnor 2009; Qu et al. 2015). High infestations of this mite degrade the quality of a product, accelerating deterioration (Zakhvatkin 1959; Kucerova and Horak 2004). In addition, due to its allergenic potential, *T. putrescentiae* has received attention as a major cause of allergic asthma and allergic diseases and rhinitis in humans (Arlian et al. 1984; Liao et al. 2013a, 2013b; Yu et al. 2014). *Tyrophagus putrescentiae* can also cause anaphylaxis through ingestion of contaminated foods (Matsumoto et al. 1996; Guerra Bernd et al. 2001; Sanchez-Borges et al. 2009; Liao et al. 2013a, 2013b). Nuñez et al. (2016) described a strong induction of allergic inflammation in the lungs of mice.

As *B. dentriticus* appears to be a candidate predator for biological control of pest mites, the present study investigated its biological development when foraging on *T. putrescentiae* and *M. ginglymura*.

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Materials and methods

The study was conducted from October 2015 through February 2016. The experiments were conducted in a climate chamber (EletroLab EL 202) at 25 ± 1°C and 80 ± 5% relative humidity (RH), in the dark. Voucher specimens of each species were deposited in the mite reference collection of the Museu de Ciências Naturais of the Centro Universitário UNIVATES (ZAUMCN), Lajeado, Rio Grande do Sul, Brazil.

Stock colonies of Blattisocius dentriticus

The specimens of *Blattisocius dentriticus* (Figure 1(A)) were obtained from corn meal provided by an animal-feed producer in Teutônia, Rio Grande do Sul state, Brazil. The stock colony was initiated about three months before the study began. The colonies were maintained in a plastic Petri dish (12 × 10 cm) placed on a layer of foam matting that was continually moistened with distilled water, inside the dish. The edges of the dish were covered with a layer of cotton wool to prevent the mites from escaping. Each colony of *B. dentriticus* was offered the prey mite *T. putrescentiae* and filled up with corn meal. The corn meal was utilized to serve as food for *T. putrescentiae* populations.

Stock colonies of Tyrophagus putrescentiae

The colonies of *T. putrescentiae* were established and maintained under the same conditions as *B. dentriticus* (Figure 1(B)).

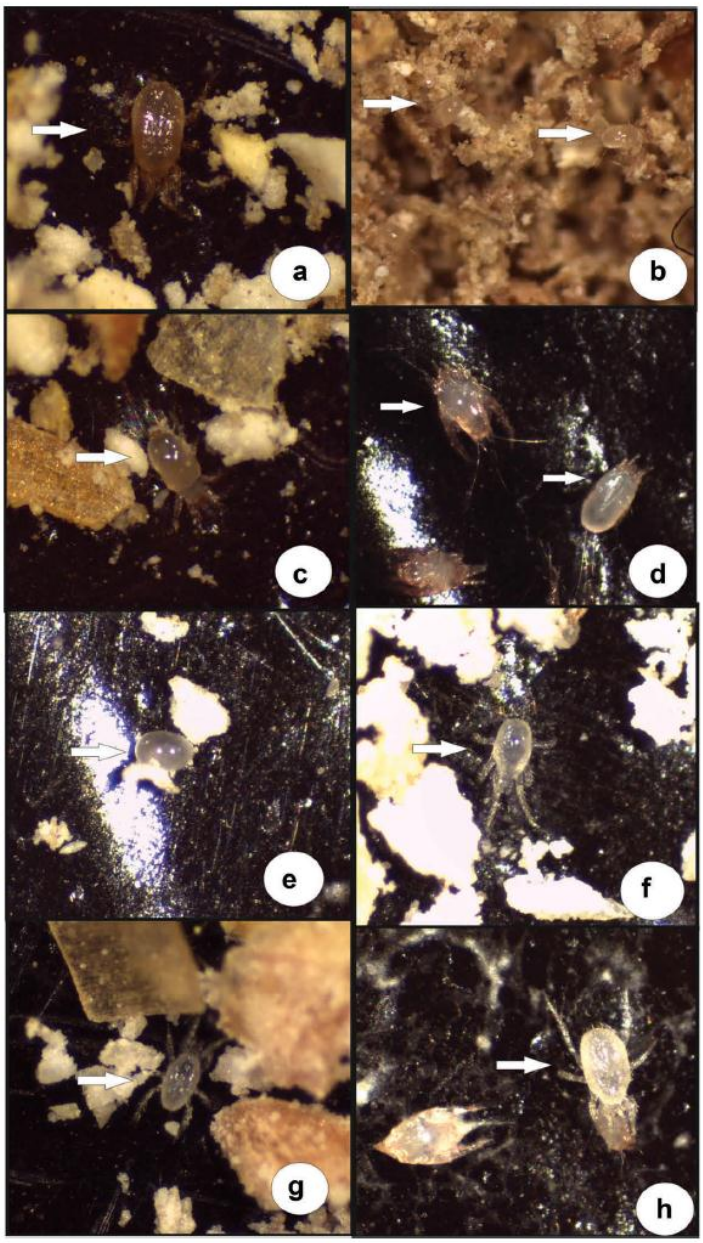

Figure 1. Indicated by arrows: (A) Adult of *Blattisocius dentriticus* in experimental unit, with corn meal; (B) Stock colony of *Tyrophagus putrescentiae*; (C) *T. putrescentiae* in experimental unit, serving as food; (D) From left to right, a male and female of *M. ginglymura*; Immature stages of *Blattisocius dentriticus*: (E) Egg; (F) Larva; (G) Protonymph; (H) Deutonymph feeding on female of *Megninia ginglymura*.

Megninia ginglymura

These mites were obtained directly from chickens (*Gallus gallus* L.) in poultry houses. Their feathers were examined under a stereoscopic microscope, in order to collect the mites, which were placed in the experimental units.

Experimental units

The experimental units consisted of a round dark plastic Petri dish (4 cm diameter), inside another plastic dish. To optimize the oviposition of predators, a plastic strip folded to form a "house" was placed in each experimental unit (Figure 2). The edges of the dish were covered with a layer of cotton wool to prevent the mites from escaping. The units were kept in a climate chamber at 25 ± 1°C and 80 ± 5% (RH), in the dark. A small amount of corn meal was added to each experimental unit, in order to facilitate egg laying by the predators, as noted in preliminary tests. As a control, 10 replicates without prey item and with corn meal, and another 10 replicates with no prey item or corn meal were tested. These controls were performed in order to check if corn meal might serve as a food source or affect the development time of the predators.

This work was carried out based on two types of treatments, in the first was offered *T. putrescentiae* and the second was *offered M. ginglymura*. The study began with eggs of *B. dentriticus* obtained individually from 30 females fed on *M. ginglymura* and 30 females fed on *T. putrescentiae* from the experimental unit. These females were removed after oviposition, leaving a single egg in each unit, totalling a number of 30 replicates for each food type. For each experimental unit, the post-embryonic stages (larvae, protonymphs, deutonymphs and adults) of the predators were fed with a mixture of all prey stages of *M. ginglymura* and *T. putrescentiae* during the study; these mixtures were considered as "free-living forms".

Immature stages of *B. dentriticus* were evaluated three times a day (08:00, 14:00 and 20:00 h). Events that occurred after 20:00 h were estimated to have occurred midway to the next observation, i.e. at 02:00 h. During the oviposition phase, a single observation was made daily, at 14:00 h. After the mites reached adulthood, their sex was determined visually, a male was introduced to the leaf disc when determined that was female. If necessary, an additional male was added to ensure successful mating. The sex ratio was determined on the basis of a count of adults originating from those eggs. Sex ratio is described as the proportion of females in the progeny. In the first treatment, ten specimens of *T. putrescentiae* (Figure 1(C)) and in the second treatment, six of *M. ginglymura* (Figure 1(D)) were offered per experimental unit. The eaten preys were replenished once per day. The following parameters were evaluated: reproductive period, duration of developmental stages, oviposition rate (number of eggs/female/day), adult longevity and sex ratio of the progeny.

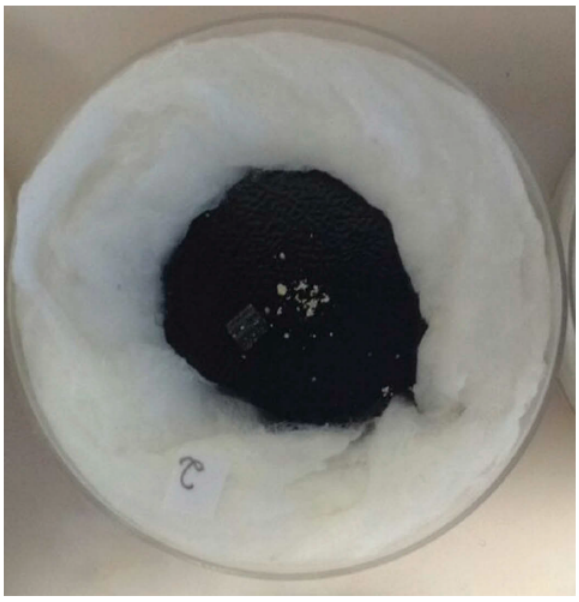

Figure 2. Front view of experimental unit consisting of a round dark plastic plate (4 cm diameter), inside another plastic plate.

The data obtained were organized for life-table calculations, according to Silveira Neto et al. (1976). Net reproductive rate ($Ro = \Sigma mx \times lx$–mx: total eggs/number of females; lx: live specimens/total specimens), mean length of a generation ($T = mx \times lx \times x/mx \times lx\ \Sigma$), innate capacity for increase (rm = log Ro/T.0.4343), finite increase rate (λ = antilog rm), gross reproductive rate (GRR = Σmx) and doubling time (DT = ln(2)/rm) were calculated. Significant differences and means were compared by the Kruskal–Wallis test ($p < 0.05$) and H and degrees of freedom (df) are provided. Differences in sex ratio were analysed by a chi-square ($\chi 2$) test. For all tests, p-values <0.05 were considered statistically significant.

Preliminary tests showed that the mites offered as prey could survive for at least one week in the experimental unit, under the experimental conditions. These preliminary tests also helped to determine the density of each kind of prey items offered to the predator, in order to assure a more than sufficient amount for the predator to consume each day.

Results

Under the conditions used, *Blattisocius dentriticus* developed from the egg through the three immature stages (larva, protonymph and deutonymph) to the adult. The eggs were opaque white, protonymphs were white and deutonymphs as well as adults were light brown (Figure 1(E–H)). The larval stage appeared to be non-feeding, since only this stage reached the next quiescent phase without needing food. The mortality of all predators occurred in the larval or protonymph stages, with or without corn meal (control studies).

The development of eggs and the quiescent protonymph, protonymph, deutonymph and teliochrysalis periods differed according to the different foods offered (Table 1). The egg-to-adult period when the predator fed on *M. ginglymura* was much longer than on *T. putrescentiae*, and the life span was shorter with *M. ginglymura*. Survivorship of the egg-to-adult phase when the predator fed on *T. putrescentiae* was 100% ($N = 30$), while on *M. ginglymura* the survivorship was 80% ($N = 24$).

Females of *B. dentriticus* fed on *T. putrescentiae* laid more eggs per day and required a shorter pre-oviposition period (Table 2). In addition, *T. putrescentiae* supported higher fecundity than did *M. ginglymura*: on average, *B. dentriticus* oviposited more than double the number of eggs when fed on *T. putrescentiae* (251) than on *M. ginglymura* (107), although the difference was not statistically significant ($H = 1.494$; $df = 1$; $p = 0.22$).

The maximum number of eggs was observed at Day 7 on *T. putrescentiae* and at Day 17 on *M. ginglymura* (Figure 3).

The life-table parameters for *B. dentriticus* differed between the two diets. The population of *B. dentriticus* fed on *T. putrescentiae* increased about 7.53 times ($Ro = 7.53$) every 14.3 days ($T = 14.3$), corresponding to a daily population growth of about 15% ($\lambda = 1.15$), i.e., a production of 0.14 female per female per day ($rm = 0.14$). *B. dentriticus* fed on *M. ginglymura* showed lower values ($Ro = 2.79$; $T = 23.76$; $\lambda = 1.04$; $rm = 0.04$). GRR and sex ratio were higher on *T. putrescentiae* (GGR = 10.73; female: male sex ratio = 0.87) than on *M. ginglymura* (GGR = 7.31; female: male sex ratio = 0.66). A chi-squared test showed statistically significant in sex ratio ($\chi 2 = 12.26$; $p < 0.05$) (Table 3). From the sex ratio of total

Table 1. Mean development time (days ± SEM) and survivorship (%) of the immature stages of *Blattisocius dentriticus* fed on *Megninia ginglymura* and *Tyrophagus putrescentiae* at 25 ± 1°C and 80 ± 5% relative humidity, in the dark, under laboratory conditions.

	N	Eggs	Larvae	Quiescent protonymph	Protonymph	Quiescent Deutonymph	Deutonymph	Teliochrysalid	Egg-adult	S (%)
Megninia ginglymura	30	0.87 ± 0.05a*	0.98 ± 0.03a	0.25 ± 0.01a	2.19 ± 0.2b	0.21 ± 0.01a	6.55 ± 1.02a	0.24 ± 0.01b	10.46 ± 1.04a	80
Tyrophagus putrescentiae	30	0.71 ± 0.05b	0.94 ± 0.02a	0.23 ± 0.01b	3.11 ± 0.22a	0.22 ± 0.00a	3.97 ± 0.52b	0.26 ± 0.01a	9.51 ± 0.55a	100
H/df		12.06/1	1.187/1	13.679/1	6.626/1	2.585/1	6.428/1	18.777/1	0.015/1	

SE – Standard Error of the Mean; *N* – Number of mites evaluated; S – Survivorship; *df* – degrees of freedom.
*Different letters indicate significant difference between immature stages ($p < 0.05$, Kruskal–Wallis test).

Table 2. Duration (days, mean ± SEM) of longevity, pre-oviposition, oviposition and post-oviposition periods, and fecundity (total number eggs/female) of *Blattisocius dentriticus* fed on *Megninia ginglymura* and *Tyrophagus putrescentiae* at 25 ± 1°C and 80 ± 5% relative humidity, in the dark, under laboratory conditions.

Parameters	*M. ginglymura* (*N* = 16)	*T. putrescentiae* (*N* = 26)	*H/df*
Fecundity	6.69 ± 1.38a	10.46 ± 1.86a*	1.494/1
Total eggs of females	107	251	
Adult longevity	32.44 ± 3.38a	25.5 ± 2.84a	2.35/1
Pre-oviposition	11.5 ± 1.62a	8.17 ± 1.46b	3.822/1
Oviposition	16.38 ± 3.62a	9.5 ± 1.54a	1.164/1
Post-oviposition	4.56 ± 1.34a	7.83 ± 1.47a	2.675/1
Eggs/day/female	0.76 ± 0.16b	1.13 ± 0.01a	6.971/1

SE – Standard Error of the Mean; *N* – Number of mites evaluated; *df* – degrees of freedom.
*Different letters indicate significant difference between immature stages ($p < 0.05$, Kruskal–Wallis test).

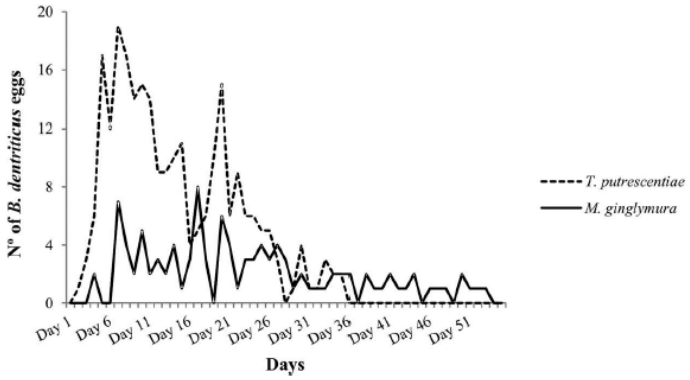

Figure 3. Oviposition peaks (+SE) of *Blattisocius dentriticus* feeding on *Tyrophagus putrescentiae* (*n* = 26) and *Megninia ginglymura* (*n* = 16) at 25 ± 1°C and 80 ± 5% relative humidity, in the dark, under laboratory conditions.

Table 3. Mean generation time (T), net reproductive rate (*Ro*), innate capacity for increase (*rm*), finite rate of increase (λ), doubling time (DT), gross reproduction rate (GRR) and sex ratio (females: males) of *Blattisocius dentriticus* fed on *Megninia ginglymura* and *Tyrophagus putrescentiae* at 25 ± 1°C and 80 ± 5% relative humidity, in the dark, under laboratory conditions.

Parameters	*M. ginglymura* (*N* = 26)	*T. putrescentiae* (*N* = 30)
T	23.76	14.3
R_0	2.79	7.53
r_m	0.04	0.14
λ	1.04	1.15
DT	16.05	4.91
GRR	7.31	10.73
Sex ratio	0.66a*	0.87b

* Different letters indicate significant difference between sex ratio ($p < 0.05$, chi-square test).

30 replicates for each different food type, sixteen females (*N* = 16) that fed on *M. ginglymura* and 26 females on *T. putrescentiae* were used to assess the fertility.

Similar fecundity and survival values were observed for each food variety. Fertility peaks occurred 4–7, 15–19 and 29–32 days after the start of the adult stage.

Discussion

Our study demonstrated that *B. dentriticus* is a potential predator for the two mite species. Individuals of *B. dentriticus* were able to develop and reproduce when fed on both *T. putrescentiae* and *M. ginglymura*. However, the life-table parameters differed, and *B. dentriticus* performed better on *T. putrescentiae*. This result may be related to the age of colony, which was initially established using this prey as food, and was maintained for several months before the study began. Similarly, Castilho et al. (2009) reported high rates of prey consumption for *Protogamasellopsis posnaniensis* Wisniewski and Hirschmann (Rhodacaridae) fed on the same prey used in the stock colony, compared with other food sources tested.

Mohamed (2013) examined the life cycle of *B. dentriticus* at 30° C under laboratory conditions. Fecundity and daily oviposition rate (eggs/female/day) were relatively low when fed on *T. putrescentiae* and *M. ginglymura* compared to other food sources. However, in our study the egg-to-adult time, oviposition period and longevity were similar to each other (Table 4). Perhaps the temperature affected the life parameters, mainly the oviposition capacity and longevity of the predator, as reported by

Table 4. Development times (mean ± SEM, in days) of *Blattisocius dentriticus* females, compared with previously published results (longevity, fecundity, oviposition period, daily rate (eggs/female/day) and egg-adult period).

Reference	Prey species	Longevity	Fecundity	Oviposition period	Daily rate (eggs/female/day)	Egg-adult
Mohamed (2013)[a]	*Rhizoglyphus robini*	21.4 ± 4.67	47.8 ± 11.17	10.4 ± 3.36	4.6	6.8 ± 0.41
	Fusarium moniliforme (Fungi)	36.8 ± 0.77	26.0 ± 2.35	17.8 ± 0.99	1.5	19.95 ± 0.65
	Botrytis allii (Fungi)	32.2 ± 1.6	33.2 ± 2.49	18.0 ± 2.0	1.84	14.15 ± 2.08
	Musca domestica	24.8 ± 0.83	43.2 ± 3.42	13.8 ± 0.84	3.13	10.35 ± 0.96
This study[b]	*T. putrescentiae*	25.5 ± 3.38	10.46 ± 1.86	9.5 ± 1.54	1.13 ± 0.01	9.51 ± 0.55
	M. ginglymura	32.44 ± 2.84	6.69 ± 1.38	16.38 ± 3.62	0.76 ± 0.16	10.46 ± 1.04

[a]*Blattisocius dentriticus* studied in Dokki, Egypt, at 30 + 2°C and 80 + 5% RH.
[b]*Blattisocius dentriticus* studied in Lajeado, Rio Grande do Sul state, Brazil, at 25 ± 1°C and 80 ± 5% relative humidity.

Table 5. Mean generation time (T), net reproductive rate (*Ro*), innate capacity for increase (*rm*), finite rate of increase (λ), doubling time (DT) and sex ratio (females: males) of *Blattisocius dentriticus* fed on different types of prey.

Parameters	*Rhizoglyphus robini*[a]	*Fusarium moniliforme*[a] (Fungi)	*Botrytis allii*[a] (Fungi)	Larvae of *Musca domestica*[a]	*T. putrescentiae*[b]	*M. ginglymura*[b]
T	10.35	26.81	20.4	14.85	14.3	23.76
DT	2.34	0.07	5.23	3.48	4.91	16.05
Ro	21.42	11.79	14.95	19.17	7.53	2.79
λ	1.34	1.09	1.14	1.22	1.15	1.04
rm	0.29	0.09	0.13	0.19	0.14	0.04
Sex ratio	0.5	0.5	0.5	0.5	0.86	0.66

[a]*Blattisocius dentriticus* studied in Dokki, Egypt, at 30 + 2°C and 80 + 5% RH (Mohamed 2013).
[b]*Blattisocius dentriticus* studied in Lajeado, Rio Grande do Sul state, Brazil, at 25 ± 1°C and 80 ± 5% relative humidity.

McClanahan (1968), Nordenfors et al. (1999) and Gotoh et al. (2004), becoming the key factor for these different results.

The data in Table 5 show that *Rhizoglyphus robini* Claparède (Acaridae) appears to be the most appropriate food source to support the development of *B. dentriticus*, followed by *T. putrescentiae* and larvae of *Musca domestica* Linnaeus (Muscidae). The development time reported by Mohamed (2013) was longer than our results, mainly with *R. robini*. The egg-adult period of *B. dentriticus* fed on the fungi *Fusarium moniliforme* Sheldon and *Botrytis allii* (Munn) is slightly longer than periods on other foods (19.95 + 0.65; 14.15 + 2.08, respectively; Table 5). These differences could be associated with the nutritional value of the food or the preference of the predator for other acarid prey such as *R. robini* and *T. putrescentiae* (Goleva and Zebitz 2013). Generalist predators may differ in their prey preferences and ability to utilize the prey as food source, resulting in their species-specific suitability to control particular target pests (Schausberger and Walzer 2001).

The sex ratio of offspring produced by *B. dentriticus* fed on *T. putrescentiae* was more female-biased than on the other foods offered. Although a female-biased sex ratio is normally reported for invertebrates, the infected females produce more daughters in order to colonize new patches (Hamilton 1967; Walter and Proctor 2013). As prey, *T. putrescentiae* may induce *B. dentriticus* to produce more females since the environmental conditions and food are especially appropriate for their colonization. Silva et al. (2013) reported that in poultry houses, *B. dentriticus* was found in nests that also contained high densities of *T. putrescentiae*, whereas *M. ginglymura* was found mostly on feathers. In the present study, the better performance of this predator when fed on *T. putrescentiae* could be related to both species occupying the same environment. The several food sources utilized by *B. dentriticus* indicate that it is a generalist. Mashaya (2002) concluded that *B. dentriticus* is a natural enemy of the booklouse *Liposcelis entomophila* (Enderlein) (Liposcelididae). It proved to be very susceptible to the insecticides deltamethrin and fenitrothion, when these were applied together on the predator feeding on *L. entomophila*.

Further studies are required to assess the possibility of using *B. dentriticus* in biological-control programmes. The most efficient temperature for hens in poultry houses is 25°C ± 1°C, and the ideal RH is 60–80% (Czarick and Lacy 1997), as used in our study. No previous studies have evaluated other predators of *M. ginglymura*, and this is the first study reporting a predatory mite feeding on *M. ginglymura*.

Other predators are reported as potential predators tested on *T. putrescentiae*. They are the cheyletids mites, that are known to have a predatory habit and used in biological control of pests in stored products, among them *Cheyletus eruditus* and *Cheyletus malaccensis* (Oudemans) (Ždárkova and Horák 1990; Ždárkova 1998; Lukas et al. 2007; Cebolla et al. 2009; Palyvos and Emmanouel 2009, 2011). However, Cebolla et al. (2009) proved highest control efficacy on *Aleuroglyphus ovatus* Troupeau (Acaridae) and *Glycyphagus destructor* (Schrank) (Glycyphagidae) and less on *T. putrescentiae*.

These findings suggest that large-scale field experiments on this potential predator are needed. Predation tests investigating the capacity to eliminate the prey are the next step. In addition, strategic releases of mass-produced predaceous mites into poultry houses should be performed. These field releases should not occur only in poultry houses with *M. ginglymura* infestations, but also in homes and storehouses in order to suppress *T. putrescentiae* in these locations. The prey consumption and behaviour of *B. dentriticus* in field conditions is not sufficiently known yet. Ždárkova and Horák (1990) reported that storehouses are not a tightly closed space, suggesting that other mites can invade it from natural habitats; being mostly brought into stores by man with plant materials on machines, or by animals such as insects, birds and rodents. Pests and predators often co-occurred, but the differences in their environmental requirements usually affected the outcome of the interaction (Gerson et al. 2003). Some predatory species are influenced by the presence of other predators. For example, the performance of *C. eruditus*, during their interactions with *B. dentriticus* in the laboratory, caused the elimination of the latter when they co-occurred in the same container (Burnett 1977); however, the outcome could differ in a real stored food situation.

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References

Arlian L, Geis D, Vyszenski-Moher D, Bernstein I, Gallagher J. 1984. Antigenic and allergenic properties of the storage mite *Tyrophagus putrescentiae*. Journal of Allergy and Clinical Immunology 74:166–171.

Aygun O, Yaman M, Durmaz H. 2007. A survey on occurrence of *Tyrophagus putrescentiae* (Acari: Acaridae) in Surk, a traditional Turkish dairy product. Journal of Food Engineering 78:878–881.

Baggio D, Figueiredo SM, Flechtmann CHW, Zambon GQ, Miranda SHG. 1987. Avaliação da presença de ácaros em cereais armazenados na grande São Paulo. Anais da Escola Superior de Agricultura Luiz de Queiroz 44:617–626.

Britto EPJ, Lopes PC, de Moraes GJ. 2012. *Blattisocius* (Acari, Blattisociidae) species from Brazil, with description of a new species, redescription of *Blattisocius keegani* and a key for the separation of the world species of the genus. Zootaxa 3479:33–51.

Burnett T. 1977. Biological models of two acarine predators of the grain mite, *Acarus siro* L. Canadian Journal of Zoology 55:1312–1323.

Castilho RC, de Moraes GJ, Silva ES, Silva LO. 2009. Predation potential and biology of *Protogamasellopsis posnaniensis* Wisniewski & Hirschmann (Acari: Rhodacaridae). Biological Control 48:164–167.

Cebolla R, Pekár S, Hubert J. 2009. Prey range of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) and its efficacy in the control of seven stored-product pests. Biological Control 50:1–6.

Cerny V. 1970. Feather mites of domestic fowl in Cuba. Folia Parasitologica 17:233–238.

Cerny V. 1973. The feather mite genus *Megninia* of some gallinaceous birds. Folia Parasitologica 20:255–261.

Chmielewski W. 1999. Acceptance of buckwheat grain as a food by *Tyrophagus putrescentiae* (Schrank) (Acari: Acaridae). Fagopyrum 16:95–97.

Czarick M, Lacy M 1997. Average house temperature. Poultry Housing Tips, University of Georgia Cooperative Extension Service, 9 (11). Available from: https://www.poultryventilation.com/sites/default/files/tips/1997/vol9n11.pdf

D'Souza PE, Murthy KS, Jagannath MS. 2001. Feather mite infestation in a broiler breeder farm. Veterinary Record 149:777.

Duek L, Kaufman G, Palevsky E, Berdicevsky I. 2001. Mites in fungal cultures. Mycoses 44:390–394.

Faleiro DCC, Toldi M, Silva GL, Ferla NJ. 2015. The ectoparasites Dermanyssus gallinae and Megninia ginglymura: bioecology and natural enemies in commercial egg-laying hens. Systematic and Applied Acarology 20:861–874.

Fenilli R, Flechtmann CHW. 1990. Ácaros do pinheiro-do-Paraná em Lages, Santa Catarina. Anais da Escola Superior de Agricultura Luiz de Queiroz, Piracicaba 47:243–250.

Garcia N. 2004. Efforts to control mites on Iberian ham by physical methods. Experimental and Applied Acarology 32:41–50.

Gerson U, Smiley RL, Ochoa R. 2003. Mites (Acari) for pest control. Oxford: Blackwell Science.

Goleva I, Zebitz CPW. 2013. Suitability of different pollen as alternative food for the predatory mite *Amblyseius swirskii* (Acari, Phytoseiidae). Experimental and Applied Acarology 61:259–283.

Gotoh T, Yamaguchi K, Mori K. 2004. Effect of temperature on life history of the predatory mite *Amblyseius* (*Neoseiulus*) *californicus* (Acari: Phytoseiidae). Experimental and Applied Acarology 32:15–30.

Guerra Bernd LA, Arruda LK, Barros Antunes HB. 2001. Oral anaphylaxis tomites. Allergy 56:83–84.

Hamilton WD. 1967. Extraordinary sex ratios. Science 156:477–488.

Kucerova Z, Horak P. 2004. Arthropod infestation in samples of stored seeds in the Czech Republic. Czech Journal of Genetics and Plant Breeding 40:11–16.

Liao E, Ho C, Yin S, Tsai J. 2013a. Immune responses to *Tyrophagus putrescentiae*-induced airway inflammation in mice. Journal of Investigational Allergology and Clinical Immunology 23:20–29.

Liao EC, Lin YH, Chiu CL, Lin TC, Tsai JJ. 2013b. Identification of allergenic component Tyr p 8 from *Tyrophagus putrescentiae* and cross-reactivity with Der p 8. Clinical and Vaccine Immunology 20:506–512.

Lukás J, Stejskal V, Jarosík V, Hubert J, Zdárková E. 2007. Differential natural performance of four Cheyletus predatory mite species in Czech grain stores. Journal of Stored Products Research 43:97–102.

Mashaya N. 2002. Predation of the booklouse *Liposcelis entomophila* (Enderlein) by *Blattisocius dentriticus* (Berlese) and their susceptibility to deltamethrin and fenitrothion. Insect Science and Its Application 22:75–79.

Matsumoto T, Hisano T, Hamaguchi M, Miike T. 1996. Systemic anaphylaxis after eating storage-mite-contaminated food. International Archives of Allergy and Immunology 109:197–200.

McClanahan RJ. 1968. Influence of temperature on the reproductive potential of two mite predators of the two-spotted spider mite. The Canadian Entomologist 100:549–556.

Mohamed AEM. 2013. Biological aspects and life table parameters of predator gamasid ascid mite, *Blattisocius dentriticus* (Berlese) (Acari: Gamasida:Ascidae). Egyptian Academic Journal of Biological Sciences 6:97–105.

Mullen GR, OConnor BM. 2009. Mites. In: Mullen GR, Durden L, editors. Medical and veterinary entomology. 2nd ed. London (UK): Academic Press; p. 423–482.

Nielsen S. 1998. The use of *Blattisocius tarsalis* (Acari: Ascidae) for biological control in flour mills. Proceedings of the 7th International Working Conference on Stored-product Protection 2:1265–1268.

Nordenfors H, Höglund J, Uggla A. 1999. Effects of temperature and humidity on oviposition, molting, and longevity of *Dermanyssus gallinae* (Acari: Dermanyssidae). Journal of Medical Entomology 36:68–72.

Nuñez NK, da Cunha AA, dos Santos Dutra M, Barbosa GL, Morassutti AL, de Souza RG, Vargas MHM, Antunes GL, Silveira JS, Silva GL, et al. 2016. Acute and chronic exposure to *Tyrophagus putrescentiae* induces allergic pulmonary response in a murine model. Asia Pacific Allergy 6:48–55.

Palyvos NE, Emmanouel NG. 2009. Temperature-dependent development of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae). Experimental and Applied Acarology 47:147–158.

Palyvos NE, Emmanouel NG. 2011. Reproduction, survival, and life table parameters of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) at various constant temperatures. Experimental and Applied Acarology 54:139–150.

Palyvos NE, Emmanouel NG, Saitanis CJ. 2008. Mites associated with stored products in Greece. Experimental and Applied Acarology 44:213–226.

Qu SX, Li HP, Ma L, Song JD, Hou LJ, Lin JS. 2015. Temperature-dependent development and reproductive traits of *Tyrophagus putrescentiae* (Sarcoptiformes: Acaridae) reared on different edible mushrooms. Environmental Entomology 44:392–399.

Quintero MT, Itza M, Juarez G, Eleno A. 2010. Seasonality of *Megninia ginglymura*: a one-year study in a hen farm in Yucatan, Mexico. Trends in Acarology: Proceedings of the 12th International Congress. 537–538.

Sczypel B, Larramendy R, Hernández M. 2003. Evaluación de distintos insecticidas frente a ectoparásitos de la gallina doméstica. Revista Cubana de Ciencia Avícola 27:121–124.

Sanchez-Borges M, Suarez-Chacon R, Capriles-Hulett A, Caballero-Fonseca F, Iraola V, Fernandez-Caldas E. 2009. Pancake syndrome (oral mite anaphylaxis). World Allergy Organization Journal 2:91–96.

Schausberger P, Walzer A. 2001. Combined versus single species release of predaceous mites: predator–predator interactions and pest suppression. Biological Control 20:269–278.

Silva GL, Ferla NJ, Toldi M, Faleiro DCC. 2013. Mite fauna (Acari) associated to commercial laying hens and bird nests in Vale do Taquari, Rio Grande do Sul, Brazil. Revista Biotemas 26:255–264.

Silveira Neto S, Nakano O, Barbin D, Villa Nova NA. 1976. Manual de ecologia dos insetos. Piracicaba: Agronômica Ceres.

Thomas HQ, Zalom FG, Nicola NL. 2011. Laboratory studies of *Blattisocius keegani* (Fox) (Acari: Ascidae) reared on eggs of navel orangeworm: potential for biological control. Bulletin of Entomological Research 101:499–504.

Tucci EC, Guastali EAL, Rebouças MM, Mendes MC, Gama NMSQ. 2005. Infestação por *Megninia* spp. em criação industrial de aves produtoras de ovos para consumo. Arquivos do Instituo Biológico 72:121–124.

Treat AE. 1969. Association of the Mite *Blattisocius tarsalis* with the Moth *Epizeuxis aemula*. Journal of the New York Entomological Society 77:171–175.

Van Emous RA, Fiks-van Niekerk TGCM, Mul MF. 2005. Bloedluizen (vogelmijten) op papier en in depraktijk. PraktijkRapport17 ISSN 15708624 (in Dutch, English summary).

Walter DE, Proctor HE. 2013. Mites: ecology, evolution & behaviour: life at a microscale. 2nd ed. Dordrecht: Springer; 486 pp.

Yu SJ, Liao EC, Tsai JJ. 2014. House dust mite allergy: environment evaluation and disease prevention. Asia Pacific Allergy 4:241–252.

Zakhvatkin AA. 1959. A translation of Fauna of U.S.S.R.: Arachnoidea: Vol. VI, No. 1: Tyroglyphoidea [Acari]. In A. Ratcliffe, and A. M. Hughes (eds.) and translated, The American Institute of Biological Sciences, Washington, DC.

Ždárkova E, Horák E. 1990. Preventive biological control of stored food mites in empty stores using *Cheyletus eruditus* (Schrank). Crop Protection 9:378–382.

Ždárkova E. 1998. Biological control of storage mites by *Cheyletus eruditus*. Integrated Pest Management Reviews 3:111–116.

5.3. ARTIGO 3 - Influence of various diets on development, life table parameters and choice oviposition test of *Tyrophagus putrescentiae*: An illustration using scanning electron microscopy (SEM) – Artigo aceito para publicação no periódico “Journal of Stored Products Research”.

Manuscript Details

Manuscript number	SPR_2017_333_R1
Title	Influence of various diets on development, life table parameters and choice oviposition test of Tyrophagus putrescentiae (Acari: Acaridae): An illustration using scanning electron microscopy (SEM)
Article type	Research Paper

Abstract

This study evaluated the life table parameters development, reproduction and oviposition of Tyrophagus putrescentiae (Schrank) (Acari: Acaridae) on five different diets (dry yeast, corn meal, mouse food, blood meal and rice flour) at 25±1ºC, 80±5% r.h.. Oviposition choice tests were carried out at 18±1, 25±1 and 30±1ºC at 80±5% r.h. for 10 days. Scanning electron microscopy (SEM) was used to observe the major morphological characters. The shortest periods for immature development were T. putrescentiae fed on mouse food. The diets had a significant effect on the development times. Daily oviposition rate (eggs/female/day) was highest on dry yeast (14.5±1.9 eggs) and lowest on rice flour (3.3±0.97 eggs). Net reproductive rate, innate capacity for increase and finite rate of increase were higher on dry yeast. The diet did not influence the oviposition at 18ºC. However, the oviposition rate was statistically significant among the three temperatures studied. The life-table parameters for T. putrescentiae differed on the five diets. Our study demonstrated that T. putrescentiae performed better on dry yeast, but when different diets were offered simultaneously, females oviposited significantly more eggs on mouse food (at 18 and 25ºC) and corn meal (at 30ºC). SEM images provided detailed descriptions of several structures, highlighting the egg surface, mouth, solenidion ωI, and genital and anal apertures.

Keywords	dry yeast; life cycle; mouse food; storage mite; storehouse.
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Porto Alegre, 10/11/2017

Dear Editor;

Please, we would like to submit our manuscript entitled "**Influence of various diets on development, life table parameters and choice oviposition test of Tyrophagus putrescentiae (Acari: Acaridae): An illustration using scanning electron microscopy (SEM)**" by Silva et al., to be considered for publication in the Journal of Stored Products Research.

This manuscript contains material that has not been published yet, and is not being considered for publication by any other print or electronic journal.

All authors have contributed significantly to the manuscript. They have taken part in the bioassays, writing the manuscript, reviewing it, and revising its intellectual and technical content.

Thanks for your attention.

Best regards,

The Authors

Highlights

- *T. putrescentiae* developed and reproduced successfully when fed on five diets
- *T. putrescentiae* performed better on dry yeast
- Females oviposited significantly more eggs on mouse food and corn meal
- The diet did not influence the oviposition at 18°C.

1 **Influence of various diets on development, life table parameters and choice**
2 **oviposition test of *Tyrophagus putrescentiae* (Acari: Acaridae): An illustration**
3 **using scanning electron microscopy (SEM)**
4
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Abstract

This study evaluated the life table parameters development, reproduction and oviposition of *Tyrophagus putrescentiae* (Schrank) (Acari: Acaridae) on five different diets (dry yeast, corn meal, mouse food, blood meal and rice flour) at 25±1°C, 80±5% r.h.. Oviposition choice tests were carried out at 18±1, 25±1 and 30±1°C at 80±5% r.h. for 10 days. Scanning electron microscopy (SEM) was used to observe the major morphological characters. The shortest periods for immature development were *T. putrescentiae* fed on mouse food. The diets had a significant effect on the development times. Daily oviposition rate (eggs/female/day) was highest on dry yeast (14.5±1.9 eggs) and lowest on rice flour (3.3±0.97 eggs). Net reproductive rate, innate capacity for increase and finite rate of increase were higher on dry yeast. The diet did not influence the oviposition at 18°C. However, the oviposition rate was statistically significant among the three temperatures studied. The life-table parameters for *T. putrescentiae* differed on the five diets. Our study demonstrated that *T. putrescentiae* performed better on dry yeast, but when different diets were offered simultaneously, females oviposited significantly more eggs on mouse food (at 18 and 25°C) and corn meal (at 30°C). SEM images provided detailed descriptions of several structures, highlighting the egg surface, mouth, solenidion ωI, and genital and anal apertures.

Keywords: dry yeast · life cycle · mouse food · storage mite · storehouse.

1. Introduction

The cosmopolitan species, *Tyrophagus putrescentiae* (Schrank), widely known as the mould mite, is found in various habitats in subtropical and tropical regions (Colloff, 2009; Robertson, 1961). This mite can develop and reproduce at a wide range of temperatures (10–34°C) and relative humidities (60–100%) using complex food

46 resources (Cunnington, 1969; Duek et al., 2001; El-Atta and Osman, 2016; Hubert et
47 al., 2004, 2014; Sánchez-Ramos and Castañera, 2001, 2005; Sánchez-Ramos et al.,
48 2007; Walter et al., 1986) This species has been associated with dried fruits, cultured
49 cheeses and other high-value foods (Hagstrum et al., 2013). Also, it is the most
50 abundant and predominant mite species found in organic materials and cereal-based
51 food products (Kheradmand et al., 2007; Thind and Clarke, 2001), and in processing
52 and packing areas of dry pet food factories (Brazis et al., 2008; Rybanska et al., 2016;
53 Thind, 2005). Potential health hazards include anaphylaxis through ingestion of
54 contaminated foods (Guerra Bernd et al., 2001; Matsumoto et al., 1996; Sanchez-Borges
55 et al., 2009). A high infestation by this mite causes degradation of food and feed
56 products, accelerating their deterioration in storage facilities (Kucerova and Horak,
57 2004; Zakhvatkin, 1959). Brazil is the third leading animal feed producer in the world,
58 and produces around 68.7 million tons in 2015 (Zani, 2015). Migration of *T.*
59 *putrescentiae* populations within a wide range of ecosystems contributes to the dispersal
60 of viable spores of several fungal species that can be transported on the vector's body
61 surface or deposited with their faeces, rapidly distributing the spores throughout the
62 substrate in which they live (Franzolin et al., 1999; Gazêta, 2000; Griffiths, 1959;
63 Hubert et al., 2004; Nesvorná et al., 2012). Furthermore, Nesvorná et al. (2012) reported
64 intense consumption of several fungal species by *T. putrescentiae*, such as *Fusarium*
65 *poae* (Peck) Wollenw, *F. culmorum* (Wm.G. Sm.) Sacc., *F. verticillioides* (Sacc.) and
66 *F. avenaceum* (Fr.) Sacc. These interactions promote the dissemination of fungi
67 throughout the stored grain, influencing both pests. Hence, if the grain is improperly
68 stored the fungi continue to grow and produce mycotoxins (Hope et al., 2005; Osborne
69 and Stein, 2007). In stored food and poultry houses it is known that *T. putrescentiae* can
70 serve as prey to predators *Blattisocius dentriticus* (Berlese) (Acari: Blattisocidae)

71 (Fenilli and Flechtmann, 1990; Silva et al., 2016), and *Cheyletus malaccensis*
72 (Oudemans) (Acari: Cheyleidae) (Cebolla et al., 2009; Palyvos and Emmanouel, 2011;
73 Silva et al., 2013).

74 The development and oviposition capacity of *T. putrescentiae* is closely
75 associated with its diet and environmental condition. It is frequently found at high
76 densities in stored foods with a high fat and protein content (Hughes, 1976). A series of
77 food preference tests performed by Zdarková (1969) showed that groundnuts, sunflower
78 seed and rapeseed were preferred by *T. putrescentiae* when compared with linseed,
79 palm kernel, poppy and cotton seeds, whereas soybean was not attractive to these mites.
80 *Tyrophagus putrescentiae* has also been found feeding on nematodes, for example,
81 *Meloidogyne* sp., an important nematode pest of ornamental and bulb crops (El-Atta and
82 Osman, 2016). Another notable observation is that this mite species has received
83 attention as one of the leading causes of allergic asthma and allergic diseases and
84 rhinitis in humans (Arlian et al., 1984; Liao et al., 2013a, b; Yu et al., 2014). Nuñez et
85 al. (2016) described a strong induction of allergic inflammation in the lungs of mice.
86 Considering that the type of diet can influence the response in *T. putrescentiae* fitness,
87 we assumed that mite development times are influenced by the different diets. Also,
88 from oviposition tests, it is expected that *T. putrescentiae* prefers to lay more eggs when
89 fed on a diet that provides its optimum development. So, this research was conducted to
90 compare the biological development of *T. putrescentiae* fed on five diets and the effects
91 of temperature on the oviposition capacity associated with various diets. We used
92 scanning electron microscopy (SEM) to observe unique surface features using our
93 adapted technique and photographed the major morphological characters of *T.*
94 *putrescentiae*. Finally, the utilization of SEM can provide support to taxonomy of *T.*
95 *putrescentiae* because this mite has been frequently studied and its taxonomy

96 classification widely discussed (Fan and Zhang, 2007; Jorrín-Novo et al., 2003; Klimov
97 and O'Connor; 2009, 2015). Thus, the results will provide additional data on
98 development of *T. putrescentiae*.

99 **2. Material and methods**

100 The life cycle observations of *T. putrescentiae* were carried out in a climate
101 chamber (EletroLab EL 202) at 25±1°C and 80±5% relative humidity (r.h.), in total
102 darkness. The oviposition tests were performed in a climate chamber at three
103 temperatures (18±1, 25±1 and 30±1°C) at 80±5% r.h.. Voucher specimens of the
104 specimens were deposited in the mite reference collection of the Museu de Ciências
105 Naturais of the Centro Universitário UNIVATES (ZAUMCN), Lajeado, Rio Grande do
106 Sul, Brazil.

107 2.1 *Stock colonies of T. putrescentiae*

108 The colonies of *T. putrescentiae* were obtained from corn meal (CM) provided
109 by an animal-feed producer in Teutônia, Rio Grande do Sul, Brazil. The stock colony
110 was initiated about 6 months before the study began. The colonies were maintained in
111 plastic Petri dishes (12 x 10 cm) placed on a layer of foam matting that was continually
112 moistened with distilled water, inside the dish. The edges of the dish were covered with
113 a layer of cotton wool to prevent the mites escaping. Each colony unit was filled with
114 dry yeast (DY), and environmental conditions were not controlled (23-26°C and 65-85%
115 r.h.).

116 *2.2 Experimental units*

117 The experimental units consisted of a circular, dark, plastic Petri dish (4 cm
118 diameter). The margins of the plate were covered with a layer of cotton wool to prevent
119 the mites escaping. The units were kept in a climate chamber at 25±1°C and 80±5% r.h.,
120 in the dark. Thirty experimental units per diet were studied. Thus, five diets were

121 offered to the mites [CM, rice flour (RF), blood meal (BM), mouse food (MF) and DY].
122 Whole foods offered were presented in form of small granules. Also, a couple of each
123 diet (small granules) was added in each experimental unit. Ten replicates without food
124 were tested as a study control, introducing a single egg within each of the ten replicates
125 too. The mites were tested before of start the experiment to verify if they are
126 nutritionally adapted to each diet used in the experiment.

127 *2.3 Biological Parameters*

128 The study began with eggs of *T. putrescentiae* obtained from 30 fertilized
129 females per diet to each bioassay unit from the stock colony. These fertilized females
130 were collected from the observations of mating with males. Females were removed after
131 oviposition when laying a single egg in each unit. Immature stages were evaluated three
132 times a day (08:00, 14:00 and 20:00 h). Events that occurred after 20:00 h were
133 estimated to have occurred midway to the next observation, i.e. at 02:00 h. During the
134 oviposition phase, a single observation was made daily at 14:00 h. After the mites had
135 reached adulthood, their sex was determined visually. So, when a female was identified,
136 a male was introduced to her leaf disc. A male was added to ensure successful mating.
137 The sex ratio was determined by counting the adult males and females originating from
138 those eggs. Sex ratio is described as the proportion of females in the progeny. To
139 recognize females and males, the mites were flipped with paintbrush to observe the
140 genital area. The following parameters were evaluated: reproductive period (pre-
141 oviposition, oviposition and post-oviposition), duration of developmental stages,
142 oviposition rate (number of eggs/female/day), adult longevity, and sex ratio of the
143 progeny.

144

145 The data obtained were organised for life table calculations, according to
146 Silveira-Neto et al. (1976). Net reproductive rate (*Ro* = Σmx.lx; mx: total eggs/number
147 of females; lx: live specimens/total specimens), mean length of a generation (T =
148 Σmx.lx.x/ Σmx.lx), innate capacity for increase (*rm* = log Ro/T.0.4343) (Krebs 1994),
149 finite increase rate (λ = antilog rm), gross reproductive rate (GRR = Σmx), and doubling
150 time (DT = ln(2)/rm) were calculated.

151 *2.4 Oviposition choice tests*

152 The experimental units consisted of a circular, dark, plastic Petri dish (8 cm
153 diameter). The margins of the plate were covered with a layer of cotton wool to prevent
154 the mites escaping. The oviposition choice tests were performed in a climate chamber at
155 18±1, 25±1 and 30±1°C, at 80±5% r.h., in the dark. Initially, 20 *T. putrescentiae* gravid
156 females dated were transferred to the centre of each experimental unit. Six experimental
157 units for each temperature, containing four diets (CM, RF, MF and DY) were studied
158 (Fig. 1). During 10 consecutive days, the units were examined daily to determine the
159 number of eggs laid by *T. putrescentiae* fed on each diet. After the daily count, the laid
160 eggs were discarded.

161 2.5 *Data analysis*

162 For all tests, P values (≤0.05) were considered statistically significant. The
163 differences among the immature stages and life table parameters were analyzed using
164 ANOVA followed by post-hoc Tukey tests using BioEstat 5.0 software (Ayres et al.,
165 2007). Oviposition rates of *T. putrescentiae* were described by abundance (number of
166 eggs at various temperatures and diets). Finally, to evaluate the effect of diet on
167 developmental and reproductive parameters of *T. putrescentiae* for a diet for the various
168 temperatures studied, randomisation analyses with contrasts and 1000 permutations
169 were made with MultiV 3.47 software (Pillar, 2004). For these analyses, a random data

set is generated by shuffling the observations in each variable among the sampling units, the resemblance matrix is computed and each resemblance value for a variable pair is compared to the corresponding value found in the observed data set (Pillar, 2004). The Bray - Curtis distance was used as a measure of dissimilarity and the pseudo F-ratio as a test criterion (Anderson, 2001).

2.6 SEM Methods

Specimens of *T. putrescentiae* were collected from a stock colony to study the morphology of its external structures by SEM. General morphological terms for the leg- and idiosomal-chaetotaxy were based on Gaud and Atyeo (1996) with the modifications proposed by Norton (1998). For SEM, mites were fixed in 70% alcohol and dehydrated in an increasing series of ethanol concentrations (80, 90 and 100%) for 15 min each. After dehydration, the samples were critical-point-dried, mounted on SEM stubs, and coated with gold/palladium (20 nm). The specimens were viewed under a Zeiss EVO LS10 microscope using high-vacuum mode at 10 kV. All measurements were in micrometres (µm).

3. Results

3.1 Development

Mortality of *T. putrescentiae* was observed in the larval or protonymph stages without food (control). Only 20% reached protonymph stages. Also, our control study appeared to show that the larval stage seemed to be non-feeding because it reached the next quiescent phase without needing any food. Except for all the dormant stages, the development of eggs, protonymph, tritonymph and egg-to-adult seemed to differ significantly according to the diets offered (Table 1). Among the diets offered, the development of *T. putrescentiae* from egg-to-adult was much longer when the mites were fed on BM and DY ($P<0.05$) and shorter when fed on MF ($P<0.05$).

195 *3.2 Reproduction*

196 The pre-oviposition period of *T. putrescentiae* fed on MF was significantly
197 shorter than that of mites fed on other diets, such as RF ($P<0.05$; Table 2). A longer
198 oviposition time was observed when females were fed CM and BM, and the shorter
199 oviposition was on MF diet. However, the post-oviposition time was similar among the
200 diets. Fecundity was significantly higher when mites fed on DY ($P<0.05$), while the
201 other diets were comparable. When mites fed on DY, a significantly higher number of
202 eggs/female/day was produced (14.5 eggs/female/day; $P<0.05$), totalling 4,480. The sex
203 ratio among the diets was practically equal. Fertility peaks occurred 2–20 days after the
204 start of the adult stage (Fig. 2).

205 *3.3 Life table parameters*

206 The life table parameters of *T. putrescentiae* were different on each diet (Table
207 3). The mites fed on DY increased about 102.2 times (Ro = 102.2) every 25.1 days (T =
208 25.1), corresponding to a daily population growth of about 20% (λ = 1.2), i.e., a
209 production of 0.18 females per female per day (rm = 0.18). The gross reproductive rate
210 was highest on DY (GRR = 126.1) and lowest on RF (GRR =39.2). All life parameters
211 provided a better performance when the mites were fed on DY than other diets.

212 **3.4 Oviposition choice**

213 Females of *T. putrescentiae* oviposited on all diets offered. At 18 and 25°C, they
214 oviposited more on MF than other diets. However, at 30°C the females oviposited more
215 on CM ($P<0.05$) (Fig. 3). There were differences in oviposition rate for all diets offered
216 at 25 and 30°C, i.e., at these temperatures, *T. putrescentiae* showed different patterns of
217 oviposition to each diet. In contrast, there was no difference at 18°C, irrespective of the
218 diet. However, among the three temperatures studied, the oviposition rate was
219 statistically significant. On DY and CM, different oviposition rates were observed at the

three temperatures evaluated. While on MF and RF at 25 and 30ºC, the oviposition rates were not different. Considering all foods tested at 30ºC, *T. putrescentiae* oviposited a total of 7290 eggs during the 10 days, followed by 4537 eggs at 25ºC and 1210 eggs at 18ºC.

3.5 SEM photographs

SEM displayed the surface features of the egg, solenidion ωI, the main dorsal anterior parts, and genital and anal apertures of the ventral region (Fig. 4). All eggs of *T. putrescentiae* were covered only by spherical-shaped agglomerations along their surface. The solenidion ωI, at the base of the tarsus, presented multiple orifices distributed across its surface.

4. Discussion

Our study demonstrated that *T. putrescentiae* developed and reproduced successfully when fed on five diets tested in our study. We observed the same stages of development reported for other acarid mites in previous studies (Arlian and Dippold, 1996; El-Atta and Osman, 2016). However, the life table parameters differed, and *T. putrescentiae* performed better on DY, corroborating our first hypothesis. Yet, when different diets were offered simultaneously, *T. putrescentiae* females oviposited significantly more on MF (18 and 25ºC) and CM (30ºC). The DY seemed to display the worst indices and hence did not support our second hypothesis. However, females prefer to oviposit in places where their eggs could remain hidden from predators. These most appropriate places might be powdery foods and granules, such as MF and CM, with small particle sizes and cavities (fractures) that serve as oviposition sites.

The ability to feed on various diets could be associated with their mouthparts that are fundamental feeding organs and adapted to several activities like piercing, chewing, tearing and sucking (Alberti and Coons, 1999). Alberti and Coons (1999)

245 described the pharynx (buccal cavity) as reniform to these acariform mites, as well as
246 the dorsal setae and solenidion presenting mechanoreceptors and olfactory functions,
247 respectively. It's worth mentioning that the diet needs to be semi-fluid before ingestion,
248 to provide the lubrication required for it to pass down the narrow pharynx and
249 oesophagus. Thus, saliva-associated carbohydrases mix with the food in the pre-buccal
250 cavity, before the diet is ingested (Brody et al., 1972). These mites penetrate into the
251 hard grain and cause internal changes, the fragments of food are bitten off with the help
252 of the chelicerae and other mouthparts, adapted to assimilating small particles (Akimov,
253 1977; Sinha, 1979). The diet preference of *T. putrescentiae* might be influenced by their
254 ability to ingest the food particles. According to Colloff (2009), the pharynx can be
255 distended from 2–3 μm up to 20 μm to accommodate solid structures. The
256 chemosensory seta solenidion ωI could be an olfactory receptor that aids to recognise
257 the diets. The longer periods spent in development of the egg-to-adult presented by the
258 BM diet might be associated with the ability of *T. putrescentiae* to identify the diet,
259 even at the immature stages, possibly because of texture, shape or even odour of the
260 food. Therefore, some food sources are more easily assimilated by mites than others.
261 Using various scanning electron microscopy (SEM) techniques, Murillo et al.
262 (2013) observed and analysed the surface features of *T. putrescentiae*, displaying that
263 alcohol methods were useful for preserving the morphological structures of the
264 specimens. In this regard, Murillo et al. (2013) utilized seven treatments to SEM, thus
265 showing that the better treatments were based on dehydration in an increasing series of
266 ethanol concentrations, however we did not placed the mites more than 15 minutes
267 before start the dehydration period. Additionally, it was not used concentrations less
268 than 70% (dehydration) and terbuthanol. Our adapted technique, based on Murillo et al.

269 (2013), detailed some surface features never seen before in this way, highlighting the
270 egg surface, mouth, solenidion ωI, and genital and anal apertures.

271 Kucerova and Stejskal (2009) described the egg surface from several species of
272 stored product mites, including *Tyrophagus* spp. The authors detailed the egg surface
273 with large locular chambers and small granules between them. While our eggs samples
274 showed a kind of spherical-shaped agglomerations (rugosity) along the egg surface that
275 could serve a respiratory function (Callaini and Mazzini, 1984) or to reduce water loss,
276 according to Witalinski (1993) the rugosity has no adhesive function. To the best of our
277 knowledge, our study provides a detailed description of the egg surface of *T.*
278 *putrescentiae* which is different than the one proposed by Kucerova and Stejskal (2009).
279 Although it is the same species, the *T. putrescentiae* strains could be different, and
280 environmental factors may also influence the surface features of the egg.

281 The wide range of stored products infested by *T. putrescentiae* demands further
282 attention. Our results demonstrate the successful development of this mite fed various
283 food sources and its capacity to oviposit at various temperatures. The temperature and
284 type of diet are considered the keys parameters for development when estimating their
285 reproductive capacity (Qu et al., 2015). The modification of temperature could be a
286 useful tool to limit infestations and reduce mobility to different habitats. These factors
287 could explain the fact that at low temperatures *T. putrescentiae* development was not
288 affected by the type of diet. However, this control technique using temperature only
289 affects the mobile stages (Barker, 1967). Previously 10ºC at 85% r.h. was reported to be
290 the lower threshold the *rm* was negative indicating that at lowest temperatures the *T.*
291 *putrescentiae* populations tend to disappear (Aspaly et al., 2007; Sánchez-Ramos and
292 Castañera, 2005). The egg stage is freeze tolerant (supports approx. -48ºC) (Eaton and

293 Kells, 2011). Thus, the prevalence of eggs represents a problem in stored products
294 because egg survival at low temperatures might provide a potential re-infestation.
295 The quality of the stored food has a strong effect on the population increase of
296 storage mites (Collins, 2012). Quality includes the nutritional content and the grain
297 condition. In the current research, all diets promoted successful mite development.
298 However, among the tested diets, RF had lower indices for fecundity, egg/day/female
299 and development parameters compared to other diets. Erban et al. (2015) demonstrated
300 accelerated population growth of *T. putrescentiae* when fed a high-fat, high-protein diet
301 compared to a low-fat, low-protein diet. Thus, this study partially corroborated that *T.*
302 *putrescentiae* is relatively poorly-adapted to carbohydrate-type substrates, as suggested
303 by Erban et al. (2009). However, this species has diverse feeding habits and probably
304 fed on several fungi presence in stored foods that are rich in carbohydrates (Hubert et
305 al., 2003, 2004). Even if the BM is rich in protein (about 80%), the lack of lipids and
306 water could affect the mite nutrition. The DY, MF and CM diets had a balanced
307 concentrations of lipids, carbohydrates and protein, thus providing a more nutritious diet
308 for the species than the BM. The mite digestive physiology varies among mite strains
309 (Bowman, 1981), thus, our populations presented digestives activities that could affect
310 the performance on each diet. Under equivalent conditions, our study presented on dry
311 yeast, the net reproductive rate (*Ro*), mean generation time (T) and doubling time (DT)
312 similar with the Button Mushroom (*Agaricus bisporus*) while that *T. putrescentiae* fed
313 on Oyster Mushroom *Pleurotus ostreatus* was similar the innate capacity for increase
314 (*rm*), finite rate of increase (λ) in comparing when fed on rice flour (RF) and blood meal
315 (BM) (Kheradmand and Zamani, 2008). However the results obtained by Qu et al.
316 (2015) reported better demographic parameters when fed on *A. bisporus* than our
317 outcomes, while that *P. ostreatus* showed the worst parameters. The total

318 developmental times mentioned by Pakyari and Maghsoudlo (2011) for *T. putrescentiae*
319 feeding on two different diets composed of mushroom *Pleurotus ostreatus* (Jacq. ex Fr.)
320 P. Kumm and phytonematode (*Ditylenchus destructor* Thorne), were similar to those in
321 our study. In our study the demographic parameters (T, *Ro*) were higher than the results
322 obtained by Pakyari and Maghsoudlo (2011). We suggest that our tested diets improve
323 the performance of this mite species when compared with phytonematodes and some
324 mushrooms.

325 Sánchez-Ramos et al. (2007) tested *T. putrescentiae* feeding on Brewer's yeast
326 flakes at the same conditions as our study, but our demographic parameters T, λ, *rm* and
327 DT, with exception *Ro*, showed better performance. El-Atta and Osman (2016)
328 examined the life cycle of *T. putrescentiae* specimens fed on egg-masses, adult females
329 of the nematode *Meloidogyne incognita* Chitwood and *Rotylenchulus reniformis*
330 Linford and Oliveira, roumy cheese and yeast under the same conditions. These authors
331 observed the intrinsic rate of natural increase (*rm*), GRR and finite rate of increase (λ)
332 highest for mites feeding on yeast, when compared to other prey/diet. Our results may
333 be related to the age of the colony, which was initially established using DY before the
334 study began. The time (days) spent by females ovipositing while feeding on BM was
335 longer than when feeding on nematodes *M. incognita* and *R. reniformis*. Fecundity and
336 daily oviposition rate (eggs/female/day) when feeding on CM, MF, RF and BM were
337 relatively low compared to El-Atta and Osman (2016), when feeding on egg masses of
338 *M. incognita* and *R. reniformis*. Nonetheless, in our study, the egg-to-adult time,
339 oviposition period and longevity were similar when compared with such nematodes.
340 Furthermore, *T. putrescentiae* fed on DY and BM displayed longer larva, protonymph
341 and tritonymph phases than the other diets and nematodes *M. incognita* and *R.*
342 *reniformis*.

343 This study substantially contributes to knowledge about development and
344 reproduction of *T. putrescentiae* on a diverse range of foods. Also, the preference of
345 oviposition regarding various diets at different temperatures was elucidated. In
346 accordance with El-Atta and Osman (2016), our results suggest that DY seems to be the
347 most appropriate diet to mass rearing, development and reproduction. Nevertheless,
348 animal-feed producers, storehouses and warehouses where DY, MF, CM or even RF are
349 exposed to suitable environment conditions for the mite, successful infestations of *T.*
350 *putrescentiae* will occur between 18–30ºC.
351

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359

360 **References**

361 Akimov, I.A., 1977. Determination of the injuriosness of storage mites. Zashch Rast
362 (Moscow) 2, 44.

363 Alberti, G., Coons, L.B., 1999. Acari - Mites. In: Harrison, F. W. (Ed.) Microscopic
364 Anatomy of Invertebrates. New York, John Wiley & Sons, pp. 515–1265.

365 Anderson, M.J., 2001. A new method for non-parametric multivariate analysis of
366 variance. Austral Ecology 26, 32–46.

367 Arlian, L.G., Dippold, J.S., 1996. Development and fecundity of *Dermatophagoides*
368 *farinae* (Acari: Pyroglyphidae). Journal of Medical Entomology 33, 257–260.

369 Arlian, L., Geis, D., Vyszenski-Moher, D., Bernstein, I., Gallagher, J., 1984. Antigenic
370 and allergenic properties of the storage mite *Tyrophagus putrescentiae*. Journal of
371 Allergy and Clinical Immunology 74, 166–171.

372 Aspaly, G., Stejskal, V., Pekar, S., Hubert, J., 2007. Temperature-dependent population
373 growth of three species of stored product mites (Acari: Acaridida). Experimental and
374 Applied Acarology 42, 37–46.

375 Ayres, M., Ayres, J.M., Ayres, D.L., Santos, A.A.S., 2007. Bioestat 5.0: Aplicações
376 estatísticas nas áreas das ciências biomédicas. Versão 5.0. Belém, Pará: Sociedade Civil
377 Mamirauá, MCT-CNPq.

378 Barker, P.S., 1967. The effects of high humidity and different temperatures on the
379 biology of *Tyrophagus putrescentiae* (Schrank) (Acarina: Tyrogliphidae). Canadian
380 Journal of Zoology 45, 91–96.

381 Bowman, C.E., 1981. Hide protease in stored product mites (Astigmata: Acaridae).
382 Comparative Biochemistry and Physiology - Part B 70, 803–805.

383 Brazis, P., Serra, M., Selles, A., Dethioux, F., Biourge, V., Puigdemont, A., 2008.
384 Evaluation of storage mite contamination of commercial dry dog food. Veterinary
385 Dermatology 19, 209–214.
386 Brody, A.R., McGrath, J.C., Wharton, G.W., 1972. *Dermatophagoides farinae*, the
387 digestive system. Journal of the New York Entomological Society 80, 152–177.
388 Callaini, C., Mazzini, M., 1984. Fine structure of the egg shell of *Tyrophagus*
389 *putrescentiae* (Schrank) (Acarina, Acaridae). Acarologia 25, 359–364.
390 Cebolla, R., Pekár, S., Hubert, J., 2009. Prey range of the predatory mite *Cheyletus*
391 *malaccensis* (Acari: Cheyletidae) and its efficacy in the control of seven stored-product
392 pests. Biological Control 50, 1–6.
393 Collins, D.A., 2012. A review on the factors affecting mite growth in stored grain
394 commodities. Experimental and Applied Acarology 56, 191–208.
395 Colloff, M.J., 2009. Dust Mites. Collingwood, Australia: CSIRO Publishing, and
396 Dordrecht: Springer.
397 Cunnington, A.M., 1969. Physical limits for the complete development of the copra
398 mite, *Tyrophagus putrescentiae* (Shrank) (Acarina, Acaridae) in relation to its world
399 distribution. In: Evans, G.O. (Ed.), Proceedings of the Second International Congress of
400 Acarology, Akademai Kiado, Budapest, pp. 241–248.
401 Duek, L., Kaufman, G., Palevsky, E., Berdicevsky, I., 2001. Mites in fungal cultures.
402 Mycoses 44, 390–394.
403 Eaton, M., Kells, S.A., 2011. Freeze Mortality Characteristics of the Mould mite
404 *Tyrophagus putrescentiae*, a Significant Pest of Stored Products. Journal of Economic
405 Entomology 104, 1423–1429.

406 El-Atta, D.A., Osman, M.A., 2016. Development and reproductive potential of
407 *Tyrophagus putrescentiae* (Acari: Acaridae) on plant-parasitic nematodes and artificial
408 diets. Experimental and Applied Acarology 68, 477–483.
409 Erban, T., Erbanova, M., Nesvorna, M, Hubert, J., 2009. The importance of starch and
410 sucrose digestion in nutritive biology of synanthropic acaridid mites: Alpha-amylases
411 and alpha-glucosidases are suitable targets for inhibitor-based strategies of mite control.
412 Archives of Insect Biochemistry and Physiology 71, 139–158.
413 Erban, T., Rybanska, D., Hubert, J., 2015. Population Growth of the Generalist Mite
414 *Tyrophagus putrescentiae* (Acari: Acaridida) Following Adaptation to High- or Low-
415 Fat and High- or Low-Protein Diets and the Effect of Dietary Switch. Environmental
416 Entomology 44, 1599–1604.
417 Erban, T., Klimov, P.B., Smrz, J., Phillips, T.W., Nesvorna, M., Kopecky, J., Hubert, J.,
418 2016. Populations of Stored Product Mite *Tyrophagus putrescentiae* Differ in Their
419 Bacterial Communities. Frontiers in Microbiology 7, 1046.
420 Fan, Q.H., Zhang, Z.Q., 2007. *Tyrophagus* (Acari: Astigmata: Acaridae). Fauna of New
421 Zealand. New Zealand, NZ: Manaaki Whenua Press, Lincoln.
422 Fenilli, R., Flechtmann, C.H.W., 1990. Ácaros do pinheiro-do-Paraná em Lages, Santa
423 Catarina. Anais da Escola Superior de Agricultura Luiz de Queiroz 47, 243–250.
424 Franzolin, M.R., Gambale, W., Cuero, R.G., Correa, B., 1999. Interaction between
425 toxigenic *Aspergillus flavus* Link and mites (*Tyrophagus putrescentiae* Schrank) on
426 maize grains: effect on fungal growth and aflatoxin production. Journal of Stored
427 Products Research 35, 215–224.
428 Gaud, J., Atyeo, W.T., 1996 Feather mites of the World (Acarina, Astigmata). The
429 supraspecific taxa. Annales du Muse´e Royale de l'Afrique centrale, Sciences
430 Zoologiques, 277, 1–193.

431 Gazêta, G.S., Norberg, A.N., Aboud-Dutra, A.E., Serra-Freire, N.M., 2000. *Tyrophagus*
432 *putrescentiae* (Schrank, 1781) as a vector of pathogenic bacteria: laboratory
433 observation. Entomología y Vectores 7, 49–59.
434 Guerra Bernd, L.A., Arruda, L.K., Barros, Antunes, H.B., 2001. Oral anaphylaxis to
435 mites. Allergy 56, 83–84.
436 Griffiths, D.A., Hodson, A.C., Christensen, C.M., 1959. Grain storage fungi associated
437 with mites. Journal of Economic Entomology 52, 514–518.
438 Hagstrum, D.W., Klejdysz, T., Subramanyam, B., Nawrot, J., 2013. Atlas of stored-
439 product insects and mites. AACC International. St Paul, Minnesota.
440 Hope, R., Aldred, D., Magan, N., 2005. Comparison of environmental profiles for
441 growth and deoxynivalenol production by *Fusarium culmorum* and *F. graminearum* on
442 wheat grain. Letters in Applied Microbiology 40, 295–300.
443 Hubert, J., Stejskal, V., Kubatova, A., Munzbergova, Z., Vanova, M., Zdarkova, E.,
444 2003. Mites as selective fungal carriers in stored grain habitats. Experimental and
445 Applied Acarology 29, 69–87.
446 Hubert, J., Jarosik, V., Mourek, J., Kubatova, A., Zdarkova, E., 2004. Astigmatid mite
447 growth and fungi preference (Acari: Acaridida): comparisons in laboratory experiments.
448 Pedobiologia 48: 205–214.
449 Hubert, J., Pekár, S., Aulický, R., Nesvorná, M., Stejskal, V., 2013. The effect of stored
450 barley cultivars, temperature and humidity on population increase of *Acarus siro*,
451 *Lepidoglyphus destructor* and *Tyrophagus putrescentiae*. Experimental and Applied
452 Acarology 60, 241–252.
453 Hubert, J., Nesvorná, M., Kopecký, J., 2014. The effect of *Tyrophagus putrescentiae* on
454 *Fusarium poae* transmission and fungal community in stored barley in a laboratory
455 experiment. Insect Science 21, 65–73.

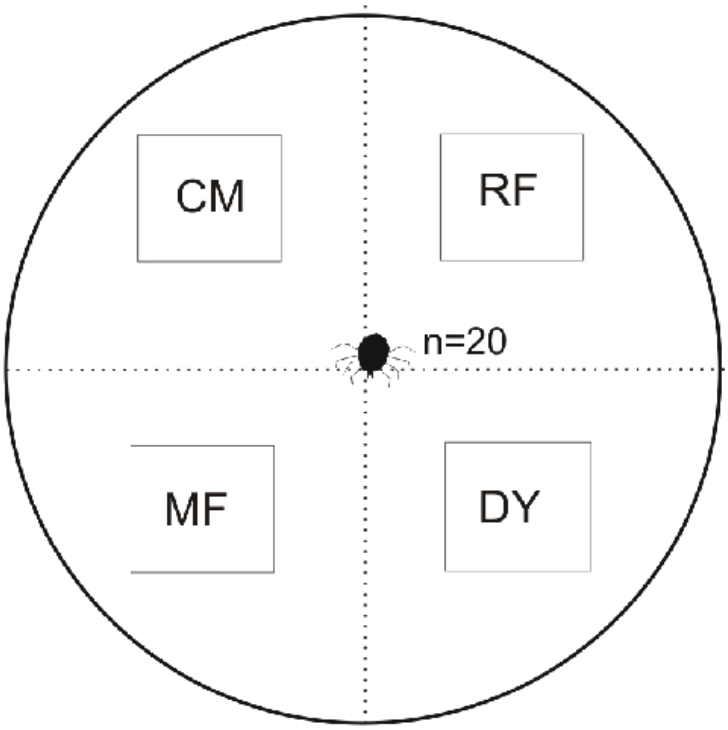
456 Hughes, A.M., 1976. The mites of stored food and houses. Technical Bulletin, Ministry
457 of Agriculture, Fisheries and Food.
458 Jorrín-Novo, J., Acosta-García, M.I., Vargas-Piqueras, P., 2003. Morphometry and
459 intrapopulational variability of *Tyrophagus putrescentiae* (Acari, Acaridae). Acarologia
460 43, 307–322.
461 Kheradmand, K., Zamani, A.A., 2008. Life table and reproductive parameters of two
462 mushroom pests, *Tyrophagus putrescentiae* (Acari: Astigmata) and *Pediculaster*
463 *flechtmanni* (Acari: Prostigmata). Journal of the Entomological Research Society 32,
464 99–102.
465 Kheradmand, K., Kamali, K., Fathipour, Y., Goltapeh, E.M., 2007. Development, life
466 table and thermal requirement of *Tyrophagus putrescentiae* (Astigmata: Acaridae) on
467 mushrooms. Journal of Stored Products Research 43, 276–281.
468 Klimov, P.B., O'Connor, B.M., 2009. Conservation of the name *Tyrophagus*
469 *putrescentiae*, a medically and economically important mite species (Acari: Acaridae).
470 International Journal of Acarology 35, 95–114.
471 Klimov, P.B., O'Connor, B.M., 2015. Comment on *Acarus putrescentiae* Schrank, 1781
472 (currently *Tyrophagus putrescentiae*; Acariformes, ACARIDAE): proposed
473 conservation of usage by designation of a replacement neotype. Bulletin of zoological
474 nomenclature 72, 50–56.
475 Krebs, C.J., 1994. Ecology: the experimental analysis of distribution and abundance.
476 New York: Harper & Row.
477 Kucerova, Z., Horak, P., 2004. Arthropod infestation in samples of stored seeds in the
478 Czech Republic. Czech Journal of Genetics and Plant Breeding 40, 11–16.
479 Kucerova, Z., Stejskal, V., 2009. Morphological diagnosis of the eggs of stored-
480 products mites. Experimental and Applied Acarology 49, 173–183.

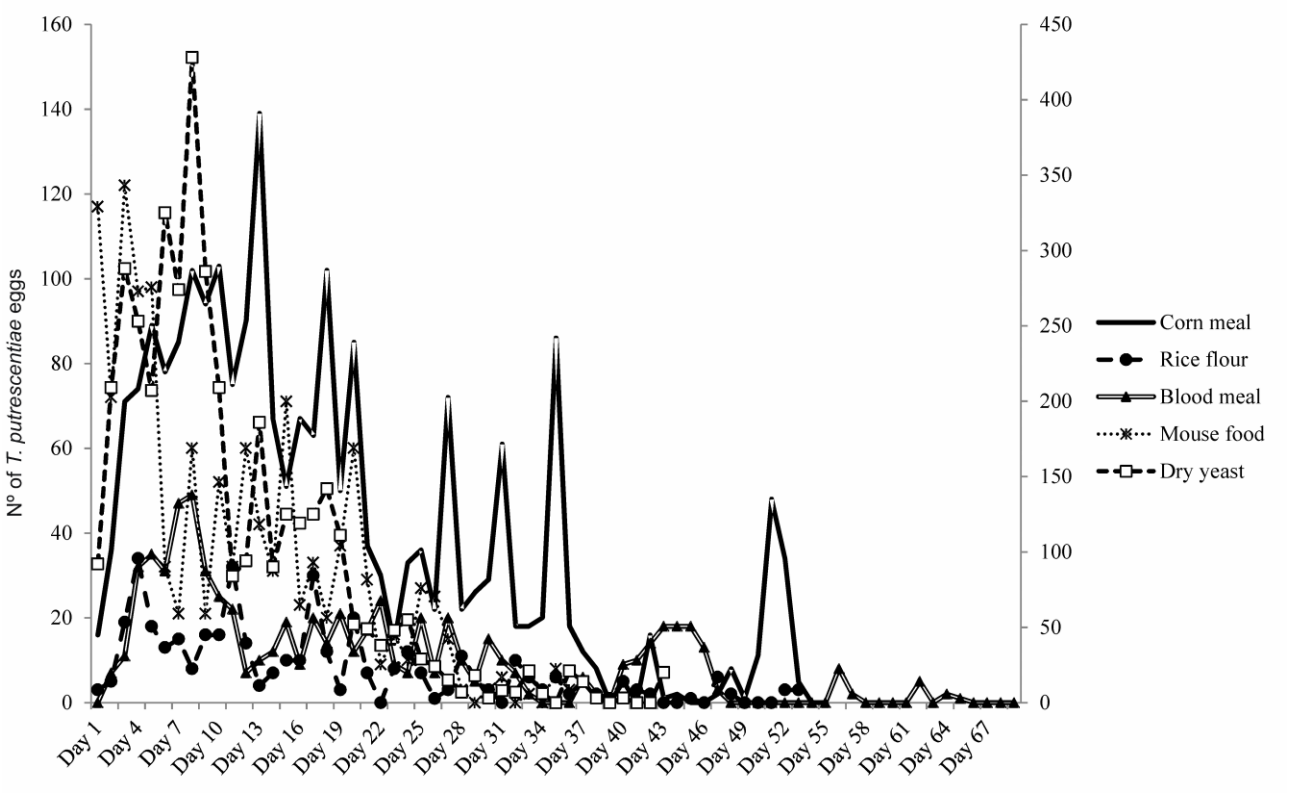
481 Matsumoto, T., Hisano, T., Hamaguchi, M., Miike, T., 1996. Systemic anaphylaxis after
482 eating storage-mite-contaminated food. International Archives of Allergy and
483 Immunology 109, 197–200.
484 Murillo, P., Aguilar, H., Sanchez, E., 2013. Use of different SEM techniques in the
485 study of *Tyrophagus putrescentiae* (Acari: Acaridae) in Costa Rica. Cuadernos de
486 Investigación UNED 5, 201–208.
487 Nesvorná, M., Gabrielova, L., Hubert, J., 2012. *Tyrophagus putrescentiae* is able to
488 graze and develop on *Fusarium* fungi of mycotoxins importance under laboratory
489 conditions. Journal of Stored Products Research 48, 37–45.
490 Norton, R., 1998. Morphological evidence for the evolutionary origin of Astigmata
491 (Acari: Acariformes). Experimental and Applied Acarology 22, 559–594.
492 Nuñez, N.K., da Cunha, A.A., dos Santos Dutra, M., Barbosa, G.L., Morassutti, A.L.,
493 de Souza, R.G., Vargas, M.H.M., Antunes, G.L., Silveira, J.S., Silva, G.L., Pitrez, P.M.,
494 2016. Acute and chronic exposure to *Tyrophagus putrescentiae* induces allergic
495 pulmonary response in a murine model. Asia Pacific Allergy 6, 48–55.
496 Liao, E.C, Ho, C., Yin, S., Tsai, J., 2013a. Immune Responses to *Tyrophagus*
497 *putrescentiae*-Induced Airway Inflammation in Mice. Journal of Investigational
498 Allergology and Clinical Immunology 23, 20–29.
499 Liao, E.C., Lin, Y.H., Chiu, C.L., Lin, T.C., Tsai, J.J., 2013b. Identification of
500 allergenic component Tyr p 8 from *Tyrophagus putrescentiae* and cross-reactivity with
501 Derp 8. Clinical and Vaccine Immunology 20, 506–512.
502 Osborne, L.E., Stein, J.M., 2007. Epidemiology of *Fusarium* head blight on small-grain
503 cereals. International Journal of Food Microbiology 119, 103–108.

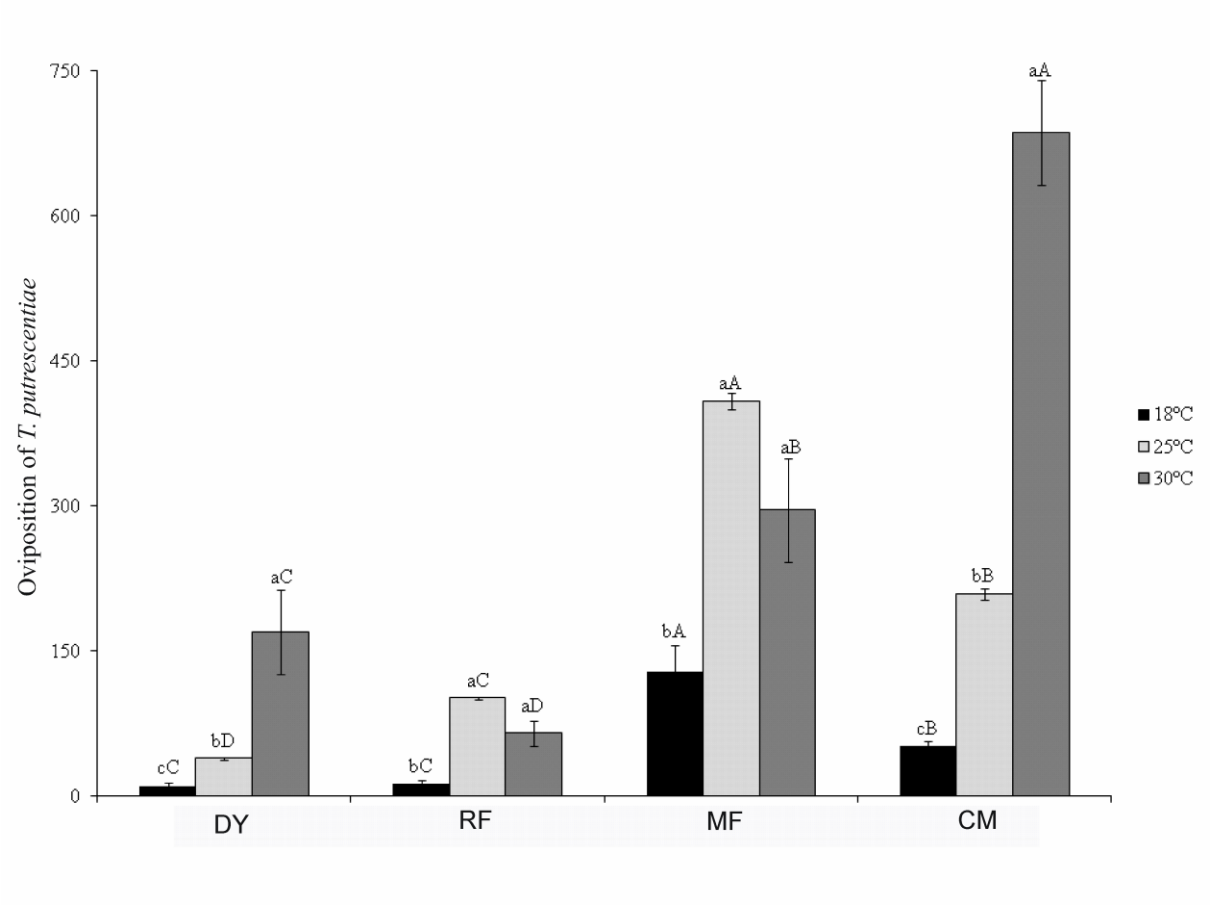
504 Pakyari, H., Maghsoudlo, M., 2011. Development and life table of *Tyrophagus*
505 *putrescentiae* (Astigmata: Acaridae) on mushroom and phytonematode. Academic
506 Journal of Entomology 4, 59–63.
507 Palyvos, N.E., Emmanouel, N.G., 2011. Reproduction, survival, and life table
508 parameters of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) at various
509 constant temperatures. Experimental and Applied Acarology 54, 139–150.
510 Pillar, V.D.P., 2004. MULTIV: Aplicativo para análise multivariada e testes de
511 hipóteses. Porto Alegre. Depto. de Ecologia. UFRGS. http//:ecologia.ecoqua.ufrgs.br.
512 Qu, S.X., Li, H.P., Ma, L., Song, J.D., Hou, L.J., Lin, J.S., 2015. Temperature-
513 dependent development and reproductive traits of *Tyrophagus putrescentiae*
514 (Sarcoptiformes: Acaridae) reared on different edible mushrooms. Environmental
515 Entomology 44, 392–399.
516 Robertson, P.L., 1961. A morphological study of variation in *Tyrophagus* (Acarina)
517 with particular reference to populations infesting cheese. Bulletin of Entomological
518 Research 52, 501–529.
519 Rybanska, D., Hubert, J., Markovic, M., Erban, T., 2016. Dry Dog Food Integrity and
520 Mite Strain Influence the Density-Dependent Growth of the Stored-Product Mite
521 *Tyrophagus putrescentiae* (Acari: Acaridida). Journal of Economic Entomology 109,
522 454–460.
523 Sánchez-Borges, M., Suarez-Chacon, R., Capriles-Hulett, A., Caballero-Fonseca, F.,
524 Iraola, V., Fernandez-Caldas, E., 2009. Pancake syndrome (oral mite anaphylaxis).
525 World Allergy Organization Journal 2, 91–96.
526 Sánchez-Ramos, I., Castañera, P., 2001. Development and survival of *Tyrophagus*
527 *putrescentiae* (Acari: Acaridae) at constant temperatures. Environmental Entomology
528 30:1082–1089.

529 Sánchez-Ramos, I., Castañera, P., 2005. Effect of temperature on reproductive
530 parameters and longevity of *Tyrophagus putrescentiae* (Acari: Acaridae). Experimental
531 and Applied Acarology 36, 93–105.
532 Sánchez-Ramos, I., Alvarez-Alfageme, F., Castañera, P., 2007. Effects of relative
533 humidity on development, fecundity and survival of three storage mites. Experimental
534 and Applied Acarology 41, 87–100.
535 Silva, G.L., Ferla, N.J., Toldi, M., Faleiro, D.C.C., 2013. Mite fauna (Acari) associated
536 to commercial laying hens and bird nests in Vale do Taquari, Rio Grande do Sul, Brazil.
537 Revista Biotemas 26, 255–264.
538 Silva, G.L., Radaelli, T.F.S., Esswein, I.Z., Ferla, N.J., Silva, O.S., 2016. Comparison
539 of Biological development of *Blattisocius dentriticus* (Blattisocidae) fed on *Tyrophagus*
540 *putrescentiae* (Acaridae) and *Megninia ginglymura* (Analgidae). International Journal
541 of Acarology 42, 405–411.
542 Silveira-Neto, S., Nakano, O., Barbin, D., Nova, A.V., 1976. Manual de ecologia e dos
543 insetos. São Paulo: Agronomia Ceres.
544 Sinha, R.N., 1979. Role of Acarina in the stored grain ecosystem. In: Rodriguez J.G.
545 (Ed.) Recent Advances in Acarology. Vol. I Academic Press, London, pp. 263–272.
546 Thind, B.B., 2005. A new versatile and robust trap for detection and monitoring of
547 stored product pests in the cereal and allied industries. Experimental and Applied
548 Acarology 35, 1–15.
549 Thind, B.B., Clarke, P.G., 2001. The occurrence of mites in cereal-based foods destined
550 for human consumption and possible consequences of infestation. Experimental and
551 Applied Acarology 25, 203–215.
552 Yu, S.J., Liao, E.C., Tsai, J.J., 2014. House dust mite allergy: environment evaluation
553 and disease prevention. Asia Pacific Allergy 4, 241–252.

554 Walter, D.E., Hudgens, R.A., Freckman, D.W., 1986. Consumption of nematodes by
555 fungivorous mites *Tyrophagus* spp. (Acarina: Astigmata: Acaridae). Oecologia 70, 357–
556 361.
557 Witalinski, W., 1993. Egg shells in mites: vitelline envelope and chorion in Acaridida
558 (Acari). Experimental and Applied Acarology 17, 321–344.
559 Zakhvatkin, A.A., 1959. A translation of Fauna of U.S.S.R.: Arachnoidea: Vol. VI, No.
560 1: Tyroglyphoidea [Acari]. In: Ratcliffe, A., Hughes A. M. (Eds.), The American
561 Institute of Biological Sciences, Washington, DC.
562 Zani, A., 2015. Boletin informativo do setor de alimentação animal.
563 http://www.sindirações.com.br.
564 Zdarková, E., 1969. Mites of stored food in Czechoslovakia. In: Evans G.O. (Ed.),
565 Proceeding of 2nd International Congress of Acarology 1967. (Akad Kaido) Budapest,
566 pp. 261–264.


CM
RF
n=20
MF
DY


Nº of T. putrescentiae eggs
160
140
120
100
80
60
40
20
0
450
400
350
300
250
200
150
100
50
0
Day 1
Day 4
Day 7
Day 10
Day 13
Day 16
Day 19
Day 22
Day 25
Day 28
Day 31
Day 34
Day 37
Day 40
Day 43
Day 46
Day 49
Day 52
Day 55
Day 58
Day 61
Day 64
Day 67
Corn meal
Rice flour
Blood meal
Mouse food
Dry yeast


Oviposition of T. putrescentiae
750
600
450
300
150
0
cC
bD
aC
bC
aC
aD
bA
aA
aB
cB
bB
aA
DY
RF
MF
CM
18°C
25°C
30°C

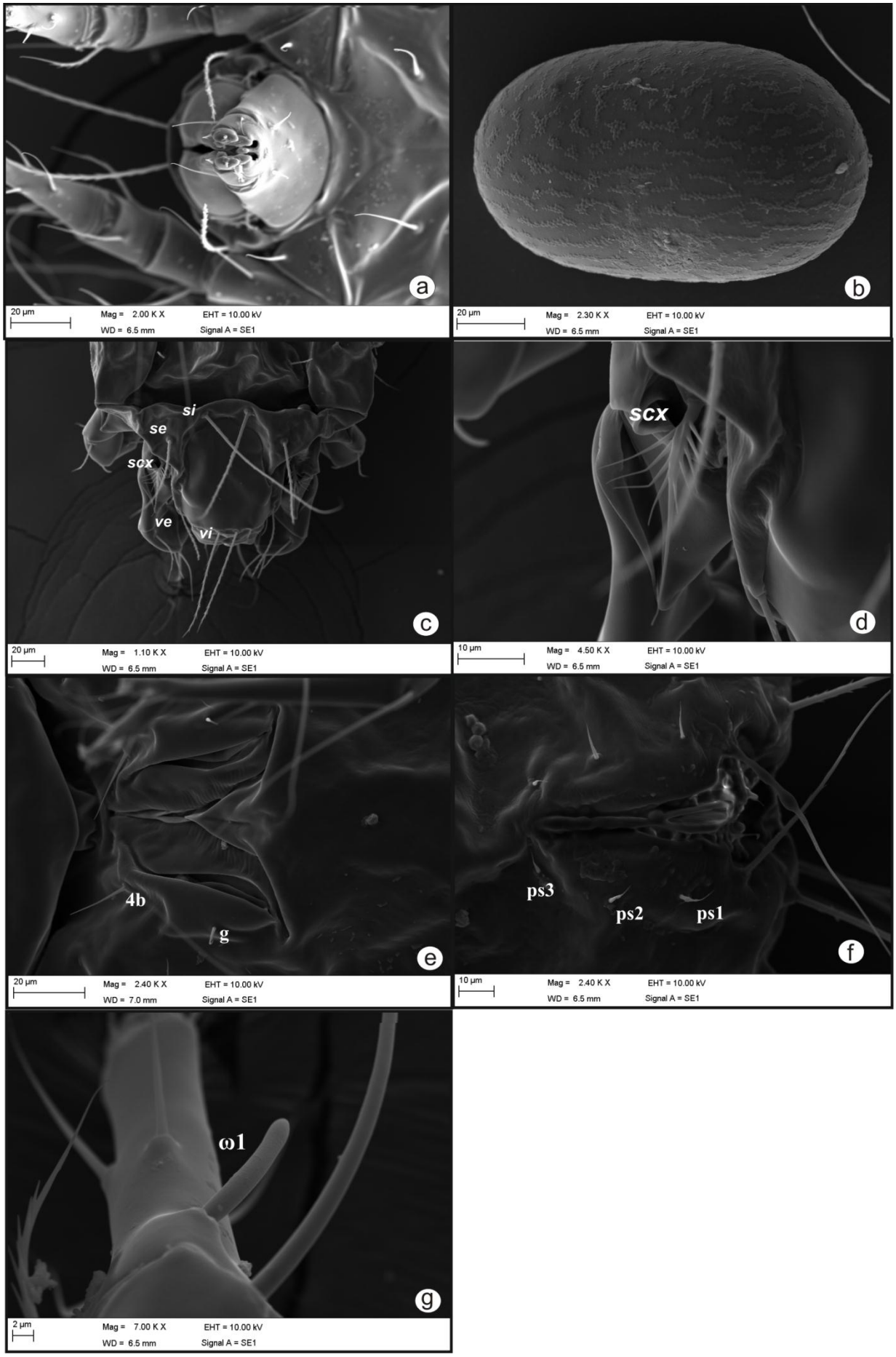

a
20 µm
Mag = 2.00 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1
b
20 µm
Mag = 2.30 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1
si
se
scx
ve
vi
c
20 µm
Mag = 1.10 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1
scx
d
10 µm
Mag = 4.50 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1
4b
g
e
20 µm
Mag = 2.40 K X
EHT = 10.00 kV
WD = 7.0 mm
Signal A = SE1
ps3
ps2
ps1
f
10 µm
Mag = 2.40 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1
ω1
g
2 µm
Mag = 7.00 K X
EHT = 10.00 kV
WD = 6.5 mm
Signal A = SE1

Figure Captions:

Fig. 1 Oviposition choice test of *Tyrophagus putrescentiae* when different diets were offered simultaneously (corn meal - CM, rice flour - RF, mouse food - MF and dry yeast - DY) at 18±1°C, 25±1°C and 30±1°C at 80± 5% r.h.. Dotted line represents the space limit of each diet; Square represents the position of each diet

Fig. 2 Daily fecundity of *Tyrophagus putrescentiae* feeding on five diets: CM, RF, BM, MF (first axis) and DY (secondary axis) at 25±1°C and 80±5% r.h., in the dark, under laboratory conditions.

Fig. 3 Oviposition (total number of eggs) of *Tyrophagus putrescentiae* at three temperatures (18±1°C, 25±1°C and 30±1°C at 80±5% r.h.). Values followed by the same letters are not significantly different. Note: Small letters indicate differences of same diet at different temperatures and the values followed by different capital letters indicate differences of same temperature with different diets ($p<0.05$; permutation tests).

Fig. 4 SEM photographs of *Tyrophagus putrescentiae*. (a) Frontal view of gnathosoma; (b) egg; (c) Propodossoma showing vertical setae "*vi*" and "*ve*"; scapular setae "*si*" and "*se*"; supracoxal seta "*scx*"; (d) supracoxal seta "*scx*" maximized; (e) genital opening; (f) anal region; (g) Tarsi I with solenidion ωI.

Table 1 Mean development time (days ± SE; N= 30; degrees of freedom= 4) and survivorship (S%) of the immature stages of *Tyrophagus putrescentiae* fed on five diets (CM, RF, BM, MF and DY) at 25±1°C and 80±5% r.h., in the dark, under laboratory conditions. For each stage, the table indicates the F-ratio and the P value.

	Egg	Larva	Quiescent larva	Protonymph	Quiescent protonymph	Tritonymph	Quiescent tritonymph	Egg-Adult	S (%)
CM	2.9±0.2a*	3.3±0.3b	0.27±0a	3.5±0.28b	0.3±0a	3.9±0.4cd	0.29±0a	14.5±0.58bc	100
RF	3±0.1a	3±0.3bc	0.28±0a	2.7±0.32bc	0.3±0a	4.1±0.3cd	0.29±0a	13.7±0.6bc	100
BM	2.9±0.3ab	4.2±0.2a	0.29±0a	4±0.44ab	0.3±0a	5.6±0.4ab	0.29±0a	17.7±0.7a	100
MF	3.4±0.2a	2.2±0.3c	0.29±0a	2.5±0.27c	0.3±0a	3.5±0.2d	0.29±0a	12.5±0.34c	100
DY	2.4±0.1b	3.3±0.3b	0.29±0a	4.7±0.4a	0.3±0a	4.9±0.4ac	0.29±0a	16.2±0.43ab	100
F-ratio	5.1	7.4	0.83	5.9	0.00	4.5	0	13.6	
P value	0.04	0.001	0.54	0.003	1	0.001	1	0.001	

* Values followed by the same letters are not significantly different in column among immature stages for each diet ($P<0.05$, ANOVA followed by post-hoc Tukey tests).

Table 2 Duration (days, mean ± SE; N=15; degrees of freedom= 4) of longevity, pre-oviposition, oviposition and post-oviposition periods, fecundity, total number eggs of *Tyrophagus putrescentiae* fed on five diets (CM, RF, BM, MF and DY) at 25±1°C and 80±5% r.h., in the dark, under laboratory conditions. For each parameter, the table indicates the F-ratio and the P value.

Parameters	CM	RF	BM	MF	DY	F-ratio	P value
			Mean number of days ± SE				
Fecundity	111.6±20.7b*	47.2±18.1b	99.4±40.6b	73.9±16.3b	224±31.2a	6.1	0.001
Total eggs of females	2232	425	696	1257	4480	-	-
Adult longevity	29.5±2.7a	28.8±5.3a	36.7±6.7a	18.3±2.1b	24.6±2.6ab	1.3	0.03
Pre-oviposition	4.3±1.3bc	9.1±2.2ab	4.6±1.3bc	3.4±1.2c	4.5±1.2bc	5	0.13
Oviposition	20.2±2.4a	15.8±4.2ab	28±8.4a	11.6±2b	16.6±1.9ab	1.9	0.02
Post-oviposition	4.9±1.7a	3.9±1.7a	4.1±1.9a	3.4±1.4a	4.2±1.7a	0.11	0.98
Eggs/day/female	8±0.72b	3.3±0.97b	3.9±1.2b	6.4±1.1ab	14.5±1.9a	2	0.40

* Values followed by the same letters are not significantly different in the row among each parameter ($P<0.05$, ANOVA followed by post-hoc Tukey tests).

Table 3 Mean generation time (T), net reproductive rate (*Ro*), innate capacity for increase (*rm*), finite rate of increase (λ), population doubling time in days (DT), gross reproduction rate (GRR) and sex ratio (females: males) of *Tyrophagus putrescentiae* (N=30) fed on five diets (CM, RF, BM, MF and DY) at 25±1°C and 80±5% r.h., in the dark, under laboratory conditions.

Parameters	CM	RF	BM	MF	DY
T	31.3	29.3	35.4	22.1	25.1
Ro	55.8	22.6	49.7	37	102.2
rm	0.13	0.11	0.11	0.16	0.18
λ	1.14	1.11	1.12	1.18	1.2
DT	5.4	6.5	6.3	4.2	3.8
GRR	125.9	39.2	76.5	72.3	126.1
Sex ratio	0.5	0.5	0.5	0.5	0.5

5.4. ARTIGO 4 - *Tyrophagus putrescentiae* (Acaridae): population development on fungal community – A ser submetido ao periódico “Journal of Stored Products Research“

***Tyrophagus putrescentiae* (Acari: Acaridae): population development on fungal community**

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Abstract

The food storage mite *Tyrophagus putrescentiae* (Schrank) (Acari) has been associated with the presence of several fungal species. The aims of this work were to assess *T. putrescentiae* population growth associated to environmental and medically important fungal species, to determine on which fungal species the *T. putrescentiae* populations display the best fitness, and to evaluate its ability to disperse each fungal species. Firstly, 24 fungal species were inoculated separately in Petri dishes containing Sabouraud agar. After one week from the inoculation, 50 mites were added to each plate. At the 28th evaluation day, mites and eggs were counted in each plate, and 50 randomly collected mites from each replicate were transferred to new plates containing only Sabouraud agar. Thus, again at the 28th day, mites, eggs and fungal population were evaluated in each plate. Only eight fungal species were dispersed by *T. putrescentiae* to the new Petri dishes: *Aspergillus clavatus, Candida tropicalis, Candida albicans, Fusarium verticillioides, Hyphopichia burtonii*, *Penicillium citrinum*, *Rhizophus azygosporus*, and *Trichophyton mentagrophytes.* The best fitness of *T. putrescentiae* was feeding on *F. verticillioides*, *P. citrinum*, and *T. mentagrophytes.* Therefore, *T.*

putrescentiae was successful in utilizing fungi as a food source, as well as demonstrated to be an important tool for dissemination of both environmental and medically important fungi.

Keywords: Dispersion · Mycophagy · *Fusarium verticillioides* · *Trichophyton mentagrophytes* · Storage mite.

Introduction

The cosmopolitan *Tyrophagus putrescentiae* (Schrank) (Acari: Acaridae) is one of the most abundant and frequent mite species found in stored foods (wheat, soy, cheese etc.), and is a mite species with economic and medical-veterinary importance (Duek et al. 2001; Mullen and OConnor 2009). These mites can also colonize human-created habitats, such as homes, dust in urban environments, medical and laboratory facilities, farms, and the food industry (Franz et al. 1997; Spieksma 1997; Solarz et al. 2007). A high infestation by this mite causes degradation of food and feed products, accelerating their deterioration in storage facilities (Zakhvatkin 1959; Kucerova & Horak 2004).

Under unsatisfactory sanitary conditions, for example, if the food is improperly stored, it may provide conditions for growth of fungi (Hope et al. 2005; Osborne and Stein 2007), which can be concomitantly infested by mites (Stejskal & Hubert 2008). Studies concerning mite-fungus interaction have demonstrated 1) fungal consumption by fungivorous mite species, such as *T. putrescentiae*, which can develop and reproduce on exclusively fungal diets (Nesvorná et al. 2012, Smrz et al. 2016), 2) selective transfer of fungal species by mites, with fungi attractive to mites being more dispersed (Hubert et al. 2003), and 3) grain contamination with fungi pathogenic to mites (Armitage and George 1986). It is known that the migration of *T. putrescentiae* populations within a wide range of ecosystems contributes to the dispersal of viable spores of several fungal species, that can be transported on the vector's body surface or deposited with their faeces, rapidly distributing the spores throughout the substrate in which they live (Lung-Shu 1984; Franzolin et al. 1999; Hubert et al. 2004; Nesvorná et al. 2012). The fungi may profit from the dispersal of spores attached on the body surface or undigested spores in the excreta (Griffiths et al. 1959; Hubert et al. 2003). In addition, the fungi either attract mite grazers by kairomones, or repel them with volatiles (Vanhaelen et al. 1980). These interactions promote the dissemination of fungi throughout the stored grain, influencing both pests.

Molds and yeasts are widely distributed in air, dust, fomites and normal flora. Although there are few reports of human pathogenic fungi in stored food (Téllez et al. 2014), these fungi live normally in environment and substrata and cause infections in susceptible hosts (Téllez et al. 2014). Therefore, cross-contamination of stored food and feed by human pathogenic fungi transported by mites from their primary environmental habitats is a feasible situation. Furthermore, the presence of mycotoxigenic fungi in stored grains is well-known (Mediavilla et al. 1996; Zulkifli &

Zakaria 2017), as well as the presence of fungal species whose spores may be important as human allergenic agents, and could be transmitted by mites (Walsh & Dixon 1996, Hubert et al, 2004). In addition, *T. putrescentiae* has received attention as one of the leading causes of allergic asthma and allergic diseases and rhinitis in humans (Arlian et al. 1984; Liao et al. 2013; Nuñez et al. 2016). Therefore, interaction studies between mites and human pathogenic fungi are urgently needed.

Our main objective was to expand the current knowledge concerning the interaction between mites, in particular *T. putrescentiae*, and fungi. We tried to elucidate if I) *T. putrescentiae* fitness is influenced by the fungal species or cell morphology (filamentous x yeast-like fungi), II) *T. putrescentiae* preferentially disperses the fungal species that provide it the best fitness, and III) *T. putrescentiae* can disperse and sustain population growth on human pathogenic fungi as well as on fungi isolated from stored food.

Methods

Stock colonies of Tyrophagus putrescentiae

Tyrophagus putrescentiae individuals were obtained from corn meal provided by an animal-feed producer in Teutônia, Rio Grande do Sul State, Brazil. The stock colony was initiated about eight months before the study began. The colonies were maintained in a plastic Petri dish (15 cm × 2.5 cm) placed on a layer of foam matting that was continually moistened with distilled water, inside the dish. The edges of the dish were covered with a layer of cotton wool to prevent the mites from escaping. Each colony unit were filled up with dry yeast (*Saccharomyces cerevisiae*), that was utilized as a food source for the mites, and environmental conditions were not controlled. Unsexed adults were used for the assays.

Fungal cultures

Two groups of fungi were used in the experiments: environmental fungal species, isolated from samples of cornmeal and rice flour through the method stipulated by Normative Instruction (IN) no. 62 of 26 August 2003 (Brazilian Ministry of Agriculture, Livestock and Food Supply - MAPA: Ministério da Agricultura, Pecuária e Abastecimento), and medically relevant fungal species maintained at a Culture Collection at Federal University of Rio Grande do Sul (UFRGS), Brazil.

One week before the experiments, a space suspension of each fungus was spread in Petri dishes containing Sabouraud agar with chloramphenicol (0.24g/l) and cultures were incubated in climate chamber at 25±1ºC. The fungal species list is described in Table 1.

Identification of Fungal species

Clinical isolates identification was maintained as received. Identification of stored food fungi at the genus level was performed by the macro and micromorphological observations of the isolates. Identification at the species level was done by sequencing of a DNA region according to each genus. The genomic DNA of the isolates was extracted using Power Soil DNA Isolation Kit® (MoBio, USA), following the manufacturer's guidelines. The regions of DNA were amplified and sequenced based on recent molecular taxonomic studies for each genus according to Table 2. The PCR consisted of: primers described in Table 2; 1× buffer PCR (Invitrogen); 1.5 mM magnesium chloride for *rpoB*, 2 mM for *β–tub* and *TEF*1α, and 3 mM for ITS and *Calm*; 0.012mM dNTP mix for ITS and 0.04 mM for the other regions; 0.04 U Taq DNA polymerase for ITS and 0.03 U Taq platinum DNA polymerase (Invitrogen) for the other regions; and 5µL of DNA. The PCR products were purified using ExoSAP-IT (Affymetrix, USA), according to the manufacturer's instructions. The sequencing was performed in ABIPRISM 3100 Genetic Analyzer (Applied Biosystems). The sequences were assembled and compared with sequences of type strains deposited in GenBank using the Basic Local Alignment Search Tool (BLAST) algorithm.

Assessment of T. putrescentiae growth on the fungal species

Twenty four (24) fungal species (eight environmental and 16 medically important species) were inoculated separately on Petri dishes (9 cm x 1.5 cm) containing Sabouraud agar with chloramphenicol, and incubated at 25±1ºC for one week, in triplicate. The study was divided in two phases. The first phase started after one week of fungal inoculation, when 50 mites were added to each previously inoculated Petri dish. The dishes were covered by Parafilm M® (Bemis NA, Neenah, Wisconsin, USA) to prevent mites from escaping. The Petri dishes were incubated at 25ºC for 28 days and observed at weekly intervals using a stereoscopic microscope Leica EZ4 HD with a digital camera (Leica). After the 28th day, the mites (immature stages and adults) and eggs were counted in each Petri dish (Fig. 1). Afterwards, the second phase of the study was initiated.

T. putrescentiae spread of fungal species to new Petri dishes

After the end of the first phase, fifty mites (maximum; if there were less than 50 mites in the Petri dish, we took them all) from each replicate of fungal species were collected using a sterile brush. The mites were collected outside visible fungal colonies and transferred into Petri dishes containing Sabouraud agar with chloramphenicol (0.24g/l) (Hubert et al. 2013). The Petri dishes were covered by parafilm, incubated at 25ºC for 28 days, and observed at weekly intervals through photos. At the 28th day, the mites (immature stages and adults) and eggs were counted in each

Petri dish.

Control studies

Two types of control studies were performed: (1) One replicate containing each fungal species and no mites, and (2) A triplicate containing only mites (no fungus) to check if *T. putrescentiae* was contaminated by a different microorganism than the one it was fed (dry yeast – *S. cerevisiae*). Both controls were done in Petri dishes containing Sabouraud agar with chloramphenicol (0.24g/l). The plates of control type (1) were monitored for 21 days to evaluate fungal growth in the absence of the mite. The aim of type control (1) was to observe the normal fungal growth without mites' interference, and then when compared with our experiments to prove that the fungi were consumed by mites. Control (2) was monitored for seven days to evaluate mite's contamination. Photos were taken to show that no contamination occurred by other microorganisms, and to display fungal growth without mites.

All experiments were conducted in a climate chamber (EletroLab EL 202) at 25±1ºC and 80±5% relative humidity (RH), in the dark. All material utilized during the study was sterilized by autoclave or UV ray.

Data analysis

The differential density-independent model ($N_t = N_0 \cdot e^{rt}$) was used to estimate the observed rate of increase (*r*) of the mite's population for each replicate (McCallum, 2000). N_t was the final mite density (eggs and mobile forms), N_0 was the initial density of mites (50) and *t* was the duration of the experiment (28 days). The response of population growth to the diet was studied using analyses of variance (one-way ANOVA) (Nesvorná et al. 2012). The final population density of mites was the dependent variable and the fungal species was the independent variable. The differences among the fungal species were compared with Tukey's pairwise *post-hoc* and Fisher's least significant difference (LSD) test. ANOVA's, Fisher's (LSD), and test for normality (K-S Test–Lilliefors) were verified using the statistical program SYSTAT 13 (Systat Inc.), and Tukey's pairwise *post-hoc* was verified using PAST 3 (Paleontological Statistics Software Package) (Hammer et al. 2001). The data in the first phase were Log-transformed for homogeneity of variance (homoscedasticity). For tests, p values <0.05 were considered statistically significant.

Results

T. putrescentiae population development on fungi

The mite population increased when fed with most fungal species, while reduced or showed a non-significant increase in the presence of *Rhizophus azygosporus*, *Sporothrix* sp., *Aspergillus terreus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Aspergillus oryzae*, and *Aspergillus flavus* (Table 3). The growth results were confirmed by the mean rate of increase (*r*) of *T. putrescentiae* (Table 3). The highest population increases were on *Trichophyton*

mentagrophytes, *Alternaria* sp., *Microsporum gypseum*, and *Aspergillus chevalieri* ($p < 0.05$) (Table 3). For *Fusarium verticillioides* and the medically important fungi *Microsporum canis*, *M. gypseum*, *T. mentagrophytes* and *Trichophyton rubrum*, it was observed at the 28th day that mites fed on whole mycelium, "cleaning" the plates (Fig. 2). Total mortality of individuals was observed only on *A. flavus*, and mite's population did not survive until the end of 28th day (Fig. 3a, b).

From the 24 fungi species tested, only eight formed colonies in the new Petri dishes after transfer by *T. putrescentiae* (2nd phase), therefore mite's population increase was calculated only for them (Table 4). The fungal species dispersed, presenting positive mite growth rates (r) and utilized as food by the mite were *Aspergillus clavatus*, *Candida tropicalis*, *Candida albicans*, *F. verticillioides*, *Hyphopichia burtonii*, *Penicillium citrinum*, *R. azygosporus*, and *T. mentagrophytes* (Table 4, Fig. 4a, b). *T. putrescentiae* populations showed better performances on *F. verticillioides*, *P. citrinum*, *T. mentagrophytes* when compared with the others ($p < 0.05$) (Table 4). Although it had negative growth rates (*r*) in the 1st phase when fed with *A. fumigatus* and *A. niger*, *T. putrescentiae* dispersed these fungi to new plates (Fig. 5b), but could not keep its colonies on them. *T. putrescentiae* showed negative growth rate on *R. azygosporus* in the 2nd phase, reinforcing the poor growth on this fungal species obtained in the 1st phase. The controls proved the mite stock was not contaminated with fungi other than *S. cerevisiae*, with which it was fed during storage, and experimental conditions were suitable for fungal development (data not shown).

Discussion

This is the first study approaching the specific population growth of *T. putrescentiae* on several fungi, in special some medically important species. *T. putrescentiae* increased its population when fed on various fungi species, dispersed and kept feeding on the environmental fungi *F. verticillioides*, *H. burtonii*, *P. citrinum*, and *R. azygosporus* (belonging to food samples) and the medically important fungi *A. clavatus*, *C. tropicalis*, *C. albicans*, and *T. mentagrophytes*. The consumption of these fungal species resulted in a population explosion of the mites. The growth on other tested fungal species was not so expressive, but mite's population increased on most fungi. It is known that fungi can benefit the populational growth of mites, in special the fungivorous ones (Hubert 2004; Nesvorná 2012).

Medically Important Fungi

Although mite growth on medically-important fungi maintained in Culture Collections is known, and *T. putrescentiae* has already been demonstrated to feed on the spores and hyphae of all the dermatophytes, moulds and yeasts tested (Duek et al. 2001), it was quite surprising to observe the explosive populacional growth of the mites on

several human pathogenic fungi (Table 3). The huge development of *T. putrescentiae* on *T. mentagrophytes*, aethiological agent of dermatophytosis, is worrying. *T. mentagrophytes* is considered zoonotic, and can be spread from animals to humans through direct contact, therefore the relation among animal-mite-human at home environments may potentiate clinical cases of dermatomycosis caused by this fungus. The association of *T. mentagrophytes* and the mite genus *Cheyletiella* (Acari: Cheyletidae) was shown to cause intensely itchy papules with necrotic areas on skin locations in contact with the animal (Dobrosavljevic et al. 2007). Thus, the bites of *T. putrescentiae* could transmit dermatophytes to the human hosts, and this study highlights the great potential of *T. putrescentiae* to act as a host to human pathogenic fungi.

The positive growth of *T. putrescentiae* feeding on *A. clavatus* at the two phases of study represent a potential risk as both pests develop on cereals and their products used in animal feed (Varga et al. 2003). As *T. putrescentiae* is the most abundant and predominant mite species found in organic materials, in special cereal-based food products, the association with *A. clavatus* could occur in the stored products (Kheradmand et al. 2007), processing and packing areas of dry pet food factories (Rybanska et al. 2016), and might cause anaphylaxis through ingestion of contaminated foods (Matsumoto et al. 1996; Sanchez-Borges et al. 2009).

Duek et al. (2011) reported an association between *T. putrescentiae* and species belonging to the yeast genus *Candida*. Our study demonstrated that *T. putrescentiae* was able to grow on and mainly disperse human opportunistic yeast pathogens.

Even pathogenic or mycotoxigenic fungal species that resulted in no or negative growth of mite populations could be disseminated to the second phase. It is the case of two *Aspergillus* species (*A. fumigatus* and *A. niger*) tested in this study. Sinha (1966) showed that *A. niger* was rejected by astigmatid mites, corroborating with our results, since *T. putrescentiae* also is classified in cohort Astigmatina. A possible explanation is that *T. putrescentiae* cannot feed on these species or their nutritional value is very low to sustain the mite population. The dissemination of fungi which are not used as food by the mite point to a fortuitous dispersion of fungus spores attached to the mite's body, independent of any interaction among the mite and the fungus. It is curious that *T. putrescentiae* grew well on *A. chevalieri* and *A. clavatus*, but could not disperse *A. chevalieri* (Tables 3 and 4), it's worth mentioning, that this species was reported causing opportunistic cutaneous aspergillosis (Naidu and Singh 1994). Future studies focused on *A. chevalieri* survival through the mite's digestive tract should elucidate this issue.

T. putrescentiae could not survive in the presence of *A. flavus* in our experimental conditions. Franzolin et al. (1999) verified the growth of *T. putrescentiae* in the presence of *A. flavus* and maize grains as food source, and verified an efficient dispersion of viable fungal spores from the inoculated compartment to the uncontaminated one. The same

authors reported the increase of aflatoxins concentrations whenever the mite was present. Notwithstanding, according to Rodrigues et al. (1984), the aflatoxins can prevent or reduce mite oviposition in general, but do not affect *T. putrescentiae*. We believe mycotoxin production by *A. flavus* is not the explanation for the surveillance failure, since it presented an astonishing populational increase in the presence of *A. clavatus*, which is also a mycotoxigenic fungus. The opposed growth behavior of *T. putrescentiae* population on the several *Aspergillus* species evaluated does not seem to be related to the phylogeny of the fungi, as both *A. clavatus* and *A. fumigatus* belong to *Aspergillus* section *Fumigati*, and the mite showed almost no growth in the presence of the latter.

Theron-De Bruin et al. (2017) reported fungus-mite mutualism, evidencing birds transporting mites with *Sporothrix* fungal spores to colonise new Protea trees. The mite species *Glycyphagus* sp. and *Proctolaelaps vandenbergi* carried fungal spores *Sporothrix* spp. on *Protea* plants, utilizing this fungus as food source too. However, our study demonstrated that *T. putrescentiae* has difficulty to develop its populations.

Environmental fungi

Nesvorná et al. (2012) demonstrated that *F. verticillioides* is highly suitable to support mite growth and dispersion of spores is expected for all *Fusarium* fungi consumed by *T. putrescentiae* in stored grain. This fungus can contaminate the stored grains with mycotoxins (Bottalico & Perrone 2002). Our study confirms the findings of Nesvorná et al. (2012) proving the suitainability of *T. putrescentiae* with *F. verticillioides*, mainly as a food source. *Fusarium verticillioides* is renowned as plant pathogen but still it is hard to determine whether in stored grain such interaction would lead to the suppression or acceleration of *F. verticillioides* growth. From our findings, we suggest that infestations of *T. putrescentiae* could disseminate the fungus on stored food reaching unprecedented economic damage level.

Penicillium citrinum produce mycotoxins responsible for intoxication in humans and animals because of eating contaminated food (Rosa 1986). We can suggest that *T. putrescentiae* could support the dissemination of this fungus in the storehouses, mainly when kept under unsatisfactory sanitary conditions.

The yeast-like *H. burtonii* is known as ‘chalky mould’, and can spoil foods by means of powdery and filamentous colonies due to the fragmentation of hyphae into short lengths (Deschuyffeleer et al. 2011). This is the first work reporting the interaction between a mite and *H. burtonii*. It is known that *H. burtonii* produce powerful amylases (Takeuchi et al. 2006), and could give support to the mite digestion on stored products like cereals and grains. It is a clear interaction of benefit to the mite while it gives support to fungal dispersion, then probably this type of benefits, through the enzymes production, might occur with another fungal species. The experiments in the present study were

carried on Sabouraud agar, which has glucose as the main carbon source; it would be interesting to evaluate the interaction between the mite and the yeast in a medium containing starch as carbon source to see whether amylase production by the yeast could really benefit the mite upon stored products. We know that yeasts have an important role on development of several arthropods, as example, yeast spores can survive digestion by *Drosophila melanogaster* (Diptera), which suggests that these flies can serve as effective vectors of yeasts under natural conditions (Reuter et al. 2007). Those insects' larvae show preference for yeast species that lead to faster development time and increased adult body weight (Anagnostou et al. 2010).

In this study, the fungus *R. azygosporus* was isolated from food sample, which has been reported associated with zygomycosis, frequently reported in immunocompromised host, mainly by inhaling spores (Ribes et al. 2000). Patients presenting diabetic ketoacidosis, cancer, antibiotic or prednisolone use, deferoxamine and desferrioxamine therapy, transplantation, renal failure, extensive burns, trauma, and intravenous drug abuse could be associated with zygomycosis (Schipper et al. 1996). *Rhizopus azygosporus* is closely related to the *Rhizopus microsporus* group, because when *Rhizopus microsporus* var. *microsporus* mated with *R. microsporus* var. *rhizopodiformis*, *azygosporus* strains are produced which have very similar morphology to *R. azygosporus*, suggesting that *R. azygosporus* resulted in nature from the spontaneous mating between these two varieties of *R. microsporus* (Papasian et al. 1991). Therefore *R. azygosporus* was given its own species designation despite its morphologic similarity to the *R. microsporus* group due to its obligate azygosporic nature (Yuan & Jong 1984). The weak population growth rate at 1stphase and the negative growth at 2nd phase of *T. putrescentiae* could be explained by the fact of *R. azygosporus* presents high density of filaments, possibly preventing the motility and the digestive ability of mite when feed on mycelium. Notwithstanding, the most important was the ability to successfull dispersion through the mite. We know that it is possible to verify several fungi associated with the faeces and the salivary glands of the mites, therefore *T. putrescentiae* demonstrated potential of *R. azygosporus* transmission and could infect immunosuppressed hosts either in home environments or in hospitals under unsatisfactory sanitary conditions.

Fungal palatability

Two factors are determinants for a successful interaction; it is the mite's ability to utilize the fungus and the digestibility of a particular fungal species (Maraun et al. 2003; Pankiewicz-Nowicka et al. 1984). The reason *T. putrescentiae* did not develop its populations on various fungi might be associated with the fungal digestibility. The understand of the fungal digestibility, it is needed to highlight that the ancestors of storage mites were mycophagous (OConnor 1982), and it has been found to host bacterial communities in the gut, parenchymal tissues and reproductive

tract (Kopecky et al. 2014a, b). Also, it is known that *T. putrescentiae* acquires bacteria and fungi (*Trichoderma* spp.) with chitinolytic activity to digest chitin from fungal cell walls (Smrz et al. 1991; Smrz & Catska 2010), and its feeding habits are switched due to the bacterial community associated with the gut and parenchymal tissues (Smrz 2003). The mite produces bacteriolytic enzymes which hydrolyze the cell walls of gram positive bacteria, showing that the microorganisms can benefit or not, both mite fitness and population growth (Erban & Hubert 2008; Erban et al. 2016). Several fungi differ in the chemical structures of their cell walls, presenting a diversity of chitin and many types of chitinolytic enzymes that are specific for such substrates (Gow et al. 2017). Smrz et al. (2016), utilizing histological methods and fluorescence microscopy, proved that exist a nearly symbiotic relationship between chitinolytic bacteria and digested fungi (spores and mycelium) exist in *T. putrescentiae*. We can suggest that many cases when the mite did not feed on fungi could be related both chitinolytic bacteria efficiently and the resistance of fungal spores. The ascospores of *A. fumigatus* are protected by an extraordinarily thick wall and tolerate stresses imposed by the environment (Kwon-Chung & Sugui 2013), and determined situations might cause structural or functional alteration of the cell wall in *A. flavus* (Seo et al. 1999). It is possible to infer that these factors could influence in digestibility of fungi by mites.

Although the mites have increased their populations at the expenses of most fungi tested, the dispersion was selective, and can vary according to the mite species (Hubert et al. 2003). For example, on *Alternaria* sp., *M. canis*, *M. gypseum* and *A. chevalieri*, the mite showed good performances of population growth at the 1st phase, but did not demonstrate ability to disperse this fungal species.

According to Hubert et al. (2003), there is no difference in the number of fungal species at the mite surfaces or guts, with a lower number of fungal species being isolated from mite when compared with the environment that they are living, demonstrating a kind of selectivity. We believe that some fungi may present toxic activity for mites; the presence of these mites on its mycelium may lead to a response of the fungus in an attempt to eliminate what would be a threat to its development. Meanwhile, certain interactions will occur simultaneously in nature, as well as have antagonistic interactions and there are microorganisms that possible benefit the mite, as suggested in relation to *T. putrescentiae* and *H. burtonii*.

Our conclusions are that *T. putrescentiae* carry some fungal species, only the dispersed fungal species, developing successfully when utilize those fungi as a food source, as well as demonstrated to be an important tool of dissemination of both environmental fungi and of medical importance.

Tyrophagus putrescentiae did not show distinction on its habits neither for fungal of medical interest, nor environmental fungi collected from food samples. The same situation was observed about filamentous and yeasts-like

fungi, the mite did not show feeding preferences for any particular form.

From those findings it is possible to suggest that several fungal human pathogens could be transmitted by mite. Our studies indicate that further research is needed to understand the processes of simultaneous dispersion of microorganisms in home environments and hospitals, where a higher density of immunosuppressed patients can be found and more medically important fungal species must be tested in contact with this mite. A survey of dust mites in order to know the associated microbiota associated could improve the management of hospital sanitary conditions. It's a satisfactory method and specific to check if stored foods are in good conditions.

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References

Abe A, Asano K, Sone T (2010) A molecular phylogeny-based taxonomy of the genus *Rhizopus*. Biosci Biotechnol Biochem 74:1325-1331.

Anagnostou C, Dorsch M, Rohlfs M (2010) Influence of dietary yeasts on *Drosophila melanogaster* life-history traits. Entomologia Experimentalis et Applicata. 136:1-11.

Andriole VT (1993) Infections with *Aspergillus* species. Clin Infect Dis17: S481-S486.

Arlian L, Geis D, Vyszenski-Moher D, Bernstein I, Gallagher J (1984) Antigenic and allergenic properties of the storage mite *Tyrophagus putrescentiae*. J Allergy Clin Immunol 74: 166-171.

Baliukoniene V, Bakutis B, Stankevicius H (2003) Mycological and mycotoxicological evaluation of grain. Ann Agric Environ Med 10: 223-227.

Bottalico A, Perrone G (2002) Toxigenic *Fusarium* species and mycotoxins associated with head blight in small-grain cereals in Europe. Eur J Plant Pathol 108: 611-624.

Bush RK, Prochnau JJ (2004) *Alternaria*-induced asthma. J Allergy Clin Immunol 113: 227-234

Cantani A, Ciaschi V (2004) Epidemiology of *Alternaria alternata* allergy: a prospective study in 6840 Italian asthmatic children. Eur Rev Med Pharmacol Sci 8: 289-294.

Deschuyffeleer N, Audenaert K, Samapundo S, Ameye S, Eeckhout M, Devlieghere F (2011)

Identification and characterization of yeasts causing chalk mould defects on par-baked bread. Food Microbiol. 28:1019-1027.

Deutsch D, Adler S, Teller J, Savir H (1989) Endogenous candidal endophthalmitis. Ann Ophthalmol 21:260-265.

Dixon DM, McNeil MM, Cohen ML, Gellin BG, La Montagne JR (1996) Fungal infections: a growing threat. Public Health Rep 111:226-236.

Dobrosavljevic DD, Popovic ND, Radovanovic SS (2007) Systemic manifestations of *Cheyletiella* infestation in man. Int J Dermatol 46: 397-399.

Druzhinina IS, Komoń-Zelazowska M, Kredics L, Hatvani L, Antal Z, Belayneh T, Kubicek CP (2008) Alternative reproductive strategies of *Hypocrea orientalis* and genetically close but clonal *Trichoderma longibrachiatum*, both capable of causing invasive mycoses of humans. Microbiology 154:3447-3459..

Duek L, Kaufman G, Palevsky E, Berdicevsky I (2001) Mites in fungal cultures. Mycoses 44: 390-394.

Erban T, Hubert J (2008) Digestive function of lysozyme in synanthropic acaridid mites enables utilization of bacteria as a food source. Exp Appl Acarol 44, 199-212.

Erban T, Klimov PB, Smrz J, Phillips TW, Nesvorna M, Kopecky J, Hubert J (2016) Populations of Stored Product Mite *Tyrophagus putrescentiae* Differ in Their Bacterial Communities. Frontiers in Microbiology 7: 1046.

Franz J-T, Masuch G, Musken H, Bergmann K-C (1997) Mite fauna of German farms. Allergy 52: 1233-1237.

Franzolin MR, Gambale W, Cuero RG, Correa B (1999) Interaction between toxigenic *Aspergillus flavus* Link and mites (*Tyrophagus putrescentiae* Schrank) on maize grains: effect on fungal growth and aflatoxin production. J Stored Prod Res 35: 215-224.

Glass NL, Donaldson GC (1995) Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Appl Environ Microbiol 61:1323-1330.

Gow NAR, Latge JP, Munro CA(2017) The Fungal Cell Wall: Structure, Biosynthesis, and Function. Microbiology Spectrum 5. doi: 10.1128/microbiolspec.FUNK-0035-2016.

Griffiths DA, Hodson AC, Christensen CM (1959) Grain storage fungi associated with mites. J Econ Entomol 52: 514-518.

Groenewald M, Smith MT (2010) Re-examination of strains formerly assigned to *Hyphopichia burtonii*, the phylogeny of the genus *Hyphopichia*, and the description of *Hyphopichia pseudoburtonii* sp. nov. Int J Syst Evol

Microbiol 60:2675-2680.

Hammer Ø, Harper DAT, Ryan PD (2001) PAST: Paleontological Statistics Software Package for Education and Data Analysis. Palaeontologia Electronica 4: 1-9.

Hara KS, Ryu JH, Lie JT, Roberts GD (1989) Disseminated *Aspergillus terreus* infection in immunocompromised hosts. Mayo Clin Proc 64:770-775.

Hope R, Aldred D, Magan N (2005) Comparison of environmental profiles for growth and deoxynivalenol production by *Fusarium culmorum* and *F. graminearum* on wheat grain. Lett Appl Microbiol 40: 295-300.

Hubert J, Stejskal V, Kubátová A, Munzbergová Z, Vánová M, Zd'árková E (2003) Mites as selective fungal carriers in stored grain habitats. Exp Appl Acarol 29:69-87.

Hubert J, Jarosik V, Mourek J, Kubatova A, Zdarkova E (2004) Astigmatid mite growth and fungi preference (Acari: Acaridida): comparisons in laboratory experiments. Pedobiologia 48: 205-214.

Hubert J, Nesvorna M, Hujslova M, Stara J, Hajslova J, Stejskal V (2013) *Acarus siro* and *Tyrophagus putrescentiae* (Acari: Acarididae) transfer of *Fusarium culmorum* into germinated barley increases mycotoxin deoxynivalenol content in barley under laboratory conditions. Int J Acarol 39: 235-238.

Iwen PC, Rupp ME, Langnas AN, Reed EC, Hinrichs SH (1998) Invasive pulmonary aspergillosis due to *Aspergillus terreus*: 12-year experience and review of the literature. Clin Infect Dis 26:1092-1097.

Kellerman TS, Pienaar JG, Van der Westhuizen GCA, Anderson LAP, Naudé TW (1976) A highly fatal tremorgenic mycotoxicosis of cattle caused by *Aspergillus clavatus*. Onderstepoort J Vet Res 43:147-154.

Kheradmand K, Kamali K, Fathipour Y, Goltapeh EM (2007) Development, life table and thermal requirement of *Tyrophagus putrescentiae* (Astigmata: Acaridae) on mushrooms. J Stored Prod Res 43:276-281.

Kokollari F, Daka A, Blyta Y, Ismajli F, Haxhijaha-Lulaj K (2015) Tinea Corporis, Caused by *Microsporum canis* - a Case Report From Kosovo. Medical Archives 69:345-346.

Kopecky J, Nesvorna M, Hubert J (2014a) *Bartonella*-like bacteria carried by domestic mite species. Exp Appl Acarol 64: 21-32.

Kopecky J, Nesvorna M, Mareckova-Sagova M, Hubert J (2014b). The effect of antibiotics on associated bacterial community of stored product mites. PLoS ONE 9:e112919.

Kwon-Chung KJ, Sugui JA. (2013) *Aspergillus fumigatus*—What Makes the Species a Ubiquitous Human Fungal Pathogen? Heitman J, ed. PLoS Pathogens. 9:e1003743.

Lacaz CS, Porto E, Martins JEC, Heins-Vacari EM, Melo NT (2002) Micoses superficiais. In: Lacaz CS,

Porto E, Martins JEC, Heins-Vacari EM, Melo NT. Tratado de micologia médica. 9ª ed. São Paulo: Sarvier, p. 252-352.

Lacey J, Ramakrishna N, Hamer A, Magan N, Marfleet IC (1991) Grain fungi. In: Arora DK, Mukerji KG, Marth EH (Eds). Handbook of Applied Mycology. Foods and Feeds. Marcel Dekker, New York, pp. 120-177.

Latgé J-P (1999) *Aspergillus fumigatus* and Aspergillosis. Clin Microbiol Rev 12:310-350.

Liao E, Ho C, Yin S, Tsai J (2013) Immune Responses to *Tyrophagus putrescentiae*-Induced Airway Inflammation in Mice. J Investig Allergol Clin Immunol 23: 20-29.

López-Villavicencio M, Aguileta G, Giraud T, de Vienne DM, Lacoste S, Couloux A, Dupont J (2010). Sex in *Penicillium*: combined phylogenetic and experimental approaches. Fungal Genet Biol 47: 693-706.

Loretti AP, Colodel EM, Driemeier D, Corrêa A, Bangel Jr JJ, Ferreiro L (2003) Neurological disorder in dairy cattle associated with consumption of beer residues contaminated with *Aspergillus clavatus*. J Vet Diagn Invest 15:123-132.

Lung-Shu L (1984) Stored grain mites in the work environment of farmers. In: Griffiths, D.A., Bowman, C.E. (Eds.), Proceedings of the 6th International Congress of Acarology, vol. 2, Edinburgh, pp. 1002-1005.

Maraun M, Martens H, Migge S, Theenhaus S, Scheu S (2003) Adding to ‘‘the enigma of soil animal diversity’’: fungal feeders and saprophagous soil invertebrates prefer similar food substrates. Eur J Soil Biol 39: 85-95.

Matsumoto T, Hisano T, Hamaguchi M, Miike T (1996) Systemic anaphylaxis after eating storage-mite-contaminated food. Int Arch Allergy Immunol 109:197-200.

McCallum H (2000) Population Parameters. Estimation for Ecological Models. Blackwell Science, Oxford.

Mediavilla A, Angulo J, Rodero JM, Dominguez E, Galán C, Infante F (1996) Fungal contamination of potential medical interest in Spanish grain stores. J Invest Allergol Clin Immunol 6:196-201.

Mullen GR, OConnor BM (2009) Mites. In: Mullen GR, Durden L (eds.), Medical and veterinary entomology, 2nd ed. Academic Press, London, United Kingdom. pp. 423-482.

Naidu J, Singh SM (1994) *Aspergillus chevalieri* (Mangin) Thom and Church: a new opportunistic pathogen of human cutaneous aspergillosis. Mycoses 37:271-274.

Nesvorná M, Gabrielova L, Hubert J (2012) Suitability of a range of *Fusarium* species to sustain populations of three stored product mite species (Acari: Astigmata). J Stored Prod Res 48:37-45.

Nuñez NK, da Cunha AA, dos Santos Dutra M, Barbosa GL, Morassutti AL, de Souza RG, Vargas MHM, Antunes GL, Silveira JS, Silva GL, Pitrez PM (2016) Acute and chronic exposure to *Tyrophagus putrescentiae* induces allergic pulmonary response in a murine model. Asia Pac Allergy 6: 48-55.

OConnor BM (1982) Evolutionary ecology of astigmatid mites. Annu Rev Entomol 27: 385-409.

Osborne LE, Stein JM (2007) Epidemiology of *Fusarium* head blight on small-grain cereals. Int J Food Microbiol 119: 103-108.

Ottonelli Stopiglia CD, Magagnin CM, Castrillón MR, Mendes SD, Heidrich D, Valente P, Scroferneker ML (2014) Antifungal susceptibilities and identification of species of the *Sporothrix schenckii* complex isolated in Brazil. Med Mycol 52:56-64.

Pankiewicz-Nowicka D, Boczek J, Davis R (1984) Food selection in *Tyrophagus putrescentiae* (Schrank) (Acaria: Acarididae). J Georgia Entomol So 19: 317-321.

Papasian CJ, Zabrabi CM, Dall LH, Stanford JF, Galant JM, Parker JR (1991) Invasive polymycotic pneumonia in an uncontrolled diabetic. Arch Pathol Lab Med 115:517-519.

Person AK, Chudgar SM, Norton BL, Tong BC, Stout JE (2010) *Aspergillus niger*: an unusual cause of invasive pulmonary aspergillosis. J Med Microbiol 59:834-838.

Reuter M, Bell G, Greig D (2007) Increased outbreeding in yeast in response to dispersal by an insect vector. Current Biology 17:81-83.

Ribes JA, Vonover-Sams CL, Baker DJ (2000) Zygomycetes in human disease. Clin Microbiol Rev 13: 236-301.

Rodrigues JG, Potts MF, Patterson CG. (1984) Mycotoxin-producing fungi: effects on stored product mites. In: Griffths, D.A., Bowman, C.E. (Eds.), Proceedings of 6th International Congress of Acarology, Chichester, Vol.1, pp. 343-350.

Rosa CAR (1986) Perfil bioquímico e sérico de suínos intoxicados experimentalmente com citrinina. Rio de Janeiro, 1986. (Dissertação de Mestrado. - Instituto de Veterinária da Universidade Federal Rural do Rio de Janeiro).

Rybanska D, Hubert J, Markovic M, Erban T (2016) Dry Dog Food Integrity and Mite Strain Influence the Density-Dependent Growth of the Stored-Product Mite *Tyrophagus putrescentiae* (Acari: Acaridida). J Econ Entomol 109:454–460.

Samson RA, Seifert KA, Kuijpers AFA, Houbraken JAMP, Frisvad JC (2004) Phylogenetic analysis of *Penicillium* subgenus *Penicillium* using partial β-tubulin sequences. Stud Mycol 49: 175–200.

Samson RA, Visagie CM, Houbraken J, Hong SB, Hubka V, Klaassen CH, Perrone G, Seifert KA, Susca A, Tanney JB, Varga J, Kocsubé S, Szigeti G, Yaguchi T, Frisvad JC (2014) Phylogeny, identification and nomenclature of the genus *Aspergillus*. Stud Mycol 78:141-173.

Sanchez-Borges M, Suarez-Chacon R, Capriles-Hulett A, Caballero-Fonseca F, Iraola V, Fernandez-

Caldas E (2009) Pancake syndrome (oral mite anaphylaxis). World Allergy Organ J 2:91-96.

Schipper MAA, Maslen MM, Hogg GG, Chow CW, Samson RA (1996) Human infection by *Rhizopus azygosporus* and the occurrence of azygospores in zygomycetes. J Med Vet Mycol 34:199-203.

Scott DW, Miller WH, Griffin CE (2000) Muller & Kirk's Small Animal Dermatology. Saunders, Philadelphia.

Schwartz RA (2004) Superficial fungal infections. Lancet 364:1173-1182.

Seo K, Akiyoshi H, Ohnishi Y (1999) Alteration of cell wall composition leads to amphotericin B resistance in *Aspergillus flavus*. Microbiol Immunol 43:1017-25.

Silva GL, Ferla NJ, Toldi M, Faleiro DCC (2013) Mite fauna (Acari) associated to commercial laying hens and bird nests in Vale do Taquari, Rio Grande do Sul, Brazil. Revista Biotemas 26: 255-264.

Sinha RN (1966) Feeding and reproduction of some stored-product mites on seed-borne fungi. J Econom Entomol 59: 1227-1232.

Smrž J (2003) Microanatomical and biological aspects of bacterial associations in *Tyrophagus putrescentiae* (Acari: Acaridida). Exp Appl Acarol 31: 105-113.

Smrž J, Čatská V (2010) Mycophagous mites and their internal associated bacteria cooperate to digest chitin in soil. Symbiosis 52: 33–40.

Smrž J, Svobodová J, Čatská V (1991) Synergetic participation *Tyrophagus putrescentiae* (Schrank) (Acari, Acaridida) and its associated bacteria on the destruction of some soil micromycetes. J Appl Entomol 111: 206-210.

Smrž J, Soukalová H, Čatská V, Hubert J (2016) Feeding Patterns of *Tyrophagus putrescentiae* (Sarcoptiformes: Acaridae) Indicate That Mycophagy Is Not a Single and Homogeneous Category of Nutritional Biology. Journal of Insect Science 16: 94.

Solarz K, Senczuk L, Maniurka H, Cichecka E, Peszke M (2007) Comparisons of the allergenic mite prevalence in dwelling sand certain outdoor environments of the Upper Silesia (southwest Poland). Int J Hyg Environ Health 210: 715-724.

Solarz K, Szilman P, Szilman E (1999) "Allergenic mites associated with bird nests in Poland (Astigmata: Pyroglyphidae, Acaridae, Glycyphagidae),"in Proceedings of the 3rd Symposium of the European Association of Acarologists: Ecology and Evolution of the Acari, Series Entomologica, Vol. 55, 1–5 July 1996, Amsterdam, eds J. Bruin, L.P.S. van der Geest, and M.W. Sabelis (Boston, MA: Kluwer Academic Publishers). pp. 651–656.

Spieksma FTM (1997) Domestic mites from an acarologic perspective. Allergy 52: 360-368.

Stejskal V, Hubert J (2008) Risk of occupational allergy to stored grain arthropods and false pest-risk perception in Czech grain stores. Ann Agric Environ Med 15: 29-35.

Simpanya MF (2000) Dermatophytes: Their taxonomy, ecology and pathogenicity. Rev Iberoam Micol 17:1-12.

Svejgaard EL, Nilsson J (2004) Onychomycosis in Denmark: prevalence of fungal nail infection in general practice. Mycoses 47:131-135.

Takeuchi A, Shimizu-Ibuka A, Nishiyama Y, Mura K, Okada S, Tokue C, Arai S (2006) Purification and Characterization of an α-Amylase of *Pichia burtonii* Isolated from the Traditional Starter "Murcha" in Nepal. Biosci Biotechnol Biochem 70: 3019-3024. doi: 10.1271/bbb.60430 Theron-De Bruin N, Dreyer LL, Ueckermann EA, Wingfield MJ, Roets F (2017) Birds Mediate a Fungus-Mite Mutualism. Microb Ecol pp. 1-12.

Travassos LR (1985) *Sporothrix schenckii*. In: Szaniszlo PJ (Editor), Fungal Dimorphism, with Emphasis on Fungi Pathogenic for Humans. Plenum Press, New York.

Varga J, Rigó K, Molnár J, Tóth B, Szencz S, Téren J, Kozakiewicz Z (2003) Mycotoxin production and evolutionary relationships among species of *Aspergillus* section *Clavati*. Antonie van Leeuwenhoek 83:191-200.

Vanhaelen M, Vanhaelen–Fastré R, Geeraerts J (1980) Occurrence in mushrooms (Homobasidiomycetes) of cis and trans-octa-1,5-dien-3-ol, attractants to the cheese mite *Tyrophagus putrescentiae* (Schrank) (Acarina, Acaridae). Experentia 36: 406.

Yuan GF, Jong SC (1984) A new obligate azygosporic species of *Rhizopus*. Mycotaxon 20:397-400.

Walsh TJ, Merz WG (1986) Pathologic features in the human alimentary tract associated with invasiveness of *Candida tropicalis*. Am J Clin Pathol 85: 498-502.

Walsh TJ, Dixon DM (1996) Chapter 75: Spectrum of Mycoses. In: Baron S, editor. Medical Microbiology. 4th edition. Galveston (TX): University of Texas Medical Branch at Galveston.

Watanabe M, Yonezawa T, Lee K, Kumagai S, Sugita-Konishi Y, Goto K, Hara-Kudo Y (2011) Molecular phylogeny of the higher and lower taxonomy of the *Fusarium* genus and differences in the evolutionary histories of multiple genes. BMC Evol Biol 11:322.

Weitzman I, Summerbell RC (1995) The dermatophytes. Clin Microbiol Rev 8: 240-259.

White TJ, Bruns T, Lee S, Taylor JW (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: PCR Protocols: A Guide to Methods and Applications, eds. Innis MA, Gelfand DH, Sninsky JJ, White TJ. Academic Press, Inc., New York. pp. 315-322.

Zakhvatkin AA (1959) A translation of Fauna of U.S.S.R.: Arachnoidea: Vol. VI, No. 1: Tyroglyphoidea [Acari]. In A. Ratcliffe, and A. M. Hughes (eds.) and translated, The American Institute of Biological Sciences, Washington, DC.

Zulkifli NA, Zakaria L (2017) Morphological and Molecular Diversity of *Aspergillus* From Corn Grain Used as Livestock Feed. HAYATI Journal of Biosciences 24: 26-34.

Tables:

Table 1 List of environmental fungal species isolated from food samples and medically important fungal species from Culture Collection at Federal University of Rio Grande do Sul (UFRGS) used in the experiments.

Environmental fungal species	Date of isolation	Sample
Aspergillus chevalieri	10/03/2016	Corn meal
Aspergillus oryzae	11/03/2016	Rice flour
Fusarium verticillioides	11/03/2016	Corn meal
Hyphopichia burtonii	10/03/2016	Rice flour
Penicillium aethiopicum	13/03/2016	Corn meal
Penicillium citrinum	11/03/2016	Corn meal
Rhizopus azygosporus	10/03/2016	Corn meal
Trichoderma bissettii	11/03/2016	Corn meal

Medically important fungal species (Clinical isolates)	Clinics aspects	References
Alternaria sp.	Cause opportunistic human infections and it has been associated mainly with hypersensitivity pneumonitis, bronchial asthma, and allergic sinusitis and rhinitis.	Bush and Prochnau 2004; Cantani and Ciaschi 2004
Aspergillus clavatus	It has been associated with a neurological disease in cattle and sheep in several countries, including Brazil.	Kellerman et al. 1976; Loretti et al. 2003
Aspergillus flavus	Grains contaminate with mycotoxins, such as aflatoxins, which are highly toxic to man and animals.	Lacey et al. 1991
Aspergillus fumigatus	The most prevalent airborne fungal pathogen, causing severe and usually fatal invasive infections in immunocompromised hosts in developed countries. The inhalation	Andriole 1993; Dixon et al. 1996; Latgé 1999

	of conidia by immunocompetent individuals rarely has any adverse effect, since the conidia are eliminated relatively efficiently by innate immune mechanisms.	
Aspergillus niger	It has been rarely reported as a cause of pneumonia.	Person et al. 2010
Aspergillus terreus	Cause of opportunistic infections.	Hara et al. 1989; Iwen et al. 1998
Candida albicans, Candida parapsilosis, Candida tropicalis	Those are the most common cause of nosocomial candidaemia and candidiasis, causing infection in gastrointestinal tract. Immunosuppression, organic weakness and other factors, could become this fungus pathogenic, resulting in a severe systemic infection.	Walsh and Merz, 1986; Deutsch et al. 1989
Epidermophyton floccosum	Humans are the primary host of this, causes tinea cruris, tinea pedis, tinea corporis, and onychomycosis.	Weitzman and Summerbell, 1995
Microsporum canis, *Microsporum gypseum*	Can be transmitted to humans by anthropophilic (between people), geophilic (from soil) and zoophilic (from animals) spread causing tinea corporis and onychomycosis.	Kokollari et al. 2015
Sporothrix sp.	The sporotrichosis has several clinical forms: the most frequent being the cutaneous/subcutaneous spreading may to the bone, central nervous system and lung. It has been associated mainly with immunocompromised individuals.	Travassos 1985
Trichophyton mentagrophytes	The most causes of dermatophytosis, causing epidermophages, onychomycosis and lesions on the scalp, frequently is isolated from rabbits, rodents, cats and dogs. The obligate parasite causes a disruption of the stratum corneum which may	Scott et al. 2000

	facilitate a dermatophyte infection.	
Trichophyton rubrum	Cause in humans mainly infections of nail bed (onychomycosis); may spread to groin and hand.	Svejgaard and Nilsson 2004
Trichosporum sp.	In humans causes fungemia and white piedra, a superficial infection of the skin, and scalp and pubic hair, mainly documented mostly in patients immunocompromised.	Schwartz 2004

Table 2 Methods and results of fungal species identifications by PCR and sequencing.

Locus	Genus	Reference for locus choice	Primers	Reference for primers	Primer concentration (pmol/μL)	PCR Program	Species identification	Type strain	Identity %	Cover
β-tubulin (BenA)	*Fusarium*	Watanabe et al. 2011	Bt2a Bt2b	Glass & Donaldson 1995	0.64	95°C (4min) 95°C (45s)/66°C (30s)/72°C (2min)-35x 72°C (7min)	*Fusarium verticillioides*	CBS 576.78	100	99
Internal Transcribed Spacer (ITS)	*Rhizopus*	Abe et al. 2010	ITS1	White et al. 1990	0.32	95°C (5min) 95°C (1min)/55°C (1min)/72°C (2min)-30x 72°C (10min)	*Rhizopus azygosporus*	CBS 357.93	100	100
	Hyphopichia	Groenewald & Smith 2010	ITS1				*Hyphopichia burtonii*	CBS 2352	99	97
	Penicillium	López-Villavicencio et al. 2010	ITS4				*Penicillium citrinum*	NRRL 1841	100	100
Calmodulin (Calm)	*Penicillium*	Samson et al. 2004	CL1 CL2A	Ottonelli et al. 2015	0.8	94°C (4min) 95°C (30s)/60°C (1min)/72°C (1min)-35x 72°C (7min)	*Penicillium aethiopicum*	CBS 484.84	99	98
	Aspergillus	Samson et al. 2014					*Aspergillus oryzae*	NRRL 447	99	96
							Aspergillus chevalieri	NRRL 78	100	96
Translation elongation factor 1-α (TEF1)	*Trichoderma*	Druzhinina et al. 2008	EF1 EF2	Watanabe et al. 2011	0.24	94°C (1min) 94°C (30s)/62°C (45s)/72°C (1min)-34x 72°C (5min)	*Trichoderma bissettii*	UTHSC:08-2443	99	96

Table 3 Development of *T. putrescentiae* on environmental and medically important fungal species.

ANOVA *one way*	**F-ratio**	**p-value**
	47.23	<0.05
Test for Normality	**Test Statistic**	**p-value**
K-S Test (Lilliefors)	0.087	0.202
Fungal species	**Total mites (triplicates)***	**Mean Rate of Increase (*r*) ± SD**
Trichophyton mentagrophytes	4050±298.16 a**	0.116±0.008
Alternaria sp.	3200±208.26 ab	0.108±0.006
Microsporum gypseum	2965±18.93 abc	0.106±0.000
Aspergillus chevalieri	2960±412.04 abc	0.103±0.017
Candida tropicalis	2404±153.13 bcd	0.098±0.006
Fusarium verticilioides	2381±83.34 bcd	0.098±0.003
Candida parapsilosis	1895±112.50 bcde	0.089±0.006
Microsporum canis	1760±90.73 cdef	0.087±0.005
Trichosporum sp.	1525±93.05 defg	0.082±0.006

Epidermophyton floccosum	1230±45.82 defg	0.074±0.004
Penicillium aethiopicum	1105±48.83 efg	0.070±0.004
Trichophyton rubrum	1000±28.86 efg	0.067±0.003
Penicillium citrinum	969±110.01 efg	0.064±0.013
Aspergillus clavatus	895±36.17 efg	0.063±0.004
Hyphopichia burtonii	810±58.94 fg	0.059±0.007
Candida albicans	740±92.91 g	0.054±0.015
Trichoderma bissetti	655±92.80 gh	0.049±0.018
Rhizopus azygosporus	160±34.01 hi	0.003±0.026
Sporothrix sp.	160±14.04 hi	0.003±0.008
Aspergillus terreus	150±11.78 hi	0.000±0.007
Aspergillus fumigatus	81±6.24 i	-0.022±0.008
Aspergillus niger	27±7.54 i	-0.078±0.052
Aspergillus oryzae	6±2.64 i	-0.107±0.029
Aspergillus flavus	0	--------

* The values are the sums of each triplicate; to data analysis, the values of replicates were calculated independently.

**Different letters indicate significant differences in column among *T. putrescentiae* population growth on each fungal species ($p<0.05$, Tukey's pairwise *post-hoc* test).

SD: Standard Deviation

Table 4 Development of *T. putrescentiae* populations on dispersed fungal species.

ANOVA *one way*	**F-ratio**	**p-value**
	11.385	<0.05
Test for Normality	**Test Statistic**	**p-value**
K-S Test (Lilliefors)	0.130	0.367
Fungal species	**Total mites (triplicates)***	**Mean Rate of Increase (*r*) ± SD**
Fusarium verticillioides	2315±114.56 a**	0.097±0.005
Penicillium citrinum	1588±109 b	0.083±0.007
Trichophyton mentagrophytes	1111±248.25 bc	0.065±0.023
Candida tropicalis	824±116 cd	0.057±0.018
Hyphopichia burtonii	717±21.51 d	0.055±0.003
Candida albicans	522± 62.98d	0.042±0.013
Aspergillus clavatus	296±25.42 d	0.023±0.008
Rhizophus azygosporus	139±20.64 d	-0.004±0.014

*The values are the sums of each triplicate; to data analysis, the values of replicates were calculated independently.

**Different letters indicate significant differences in column among population growth of each fungal species ($p<0.05$, Fisher's least significant difference (LSD) test).

SD: Standard Deviation

Figures:

Fig. 1 Schematic experiment detailing the method applied to study the mites and fungus interactions.

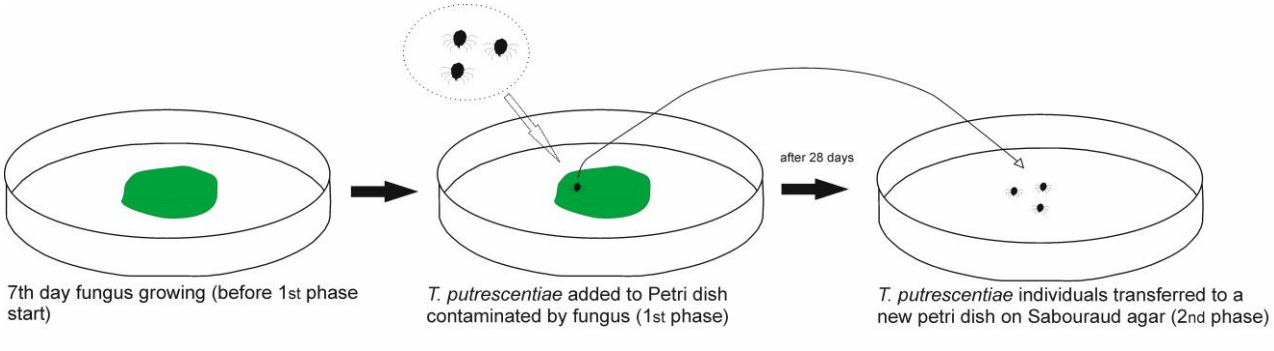


Fig. 2 Environmental fungal species utilized at the 1st phase of experiment. Initial (after one week of fungal inoculation, fifty mites were added), 7th day, 14th, 21th day and 28th day with *T. putrescentiae*.

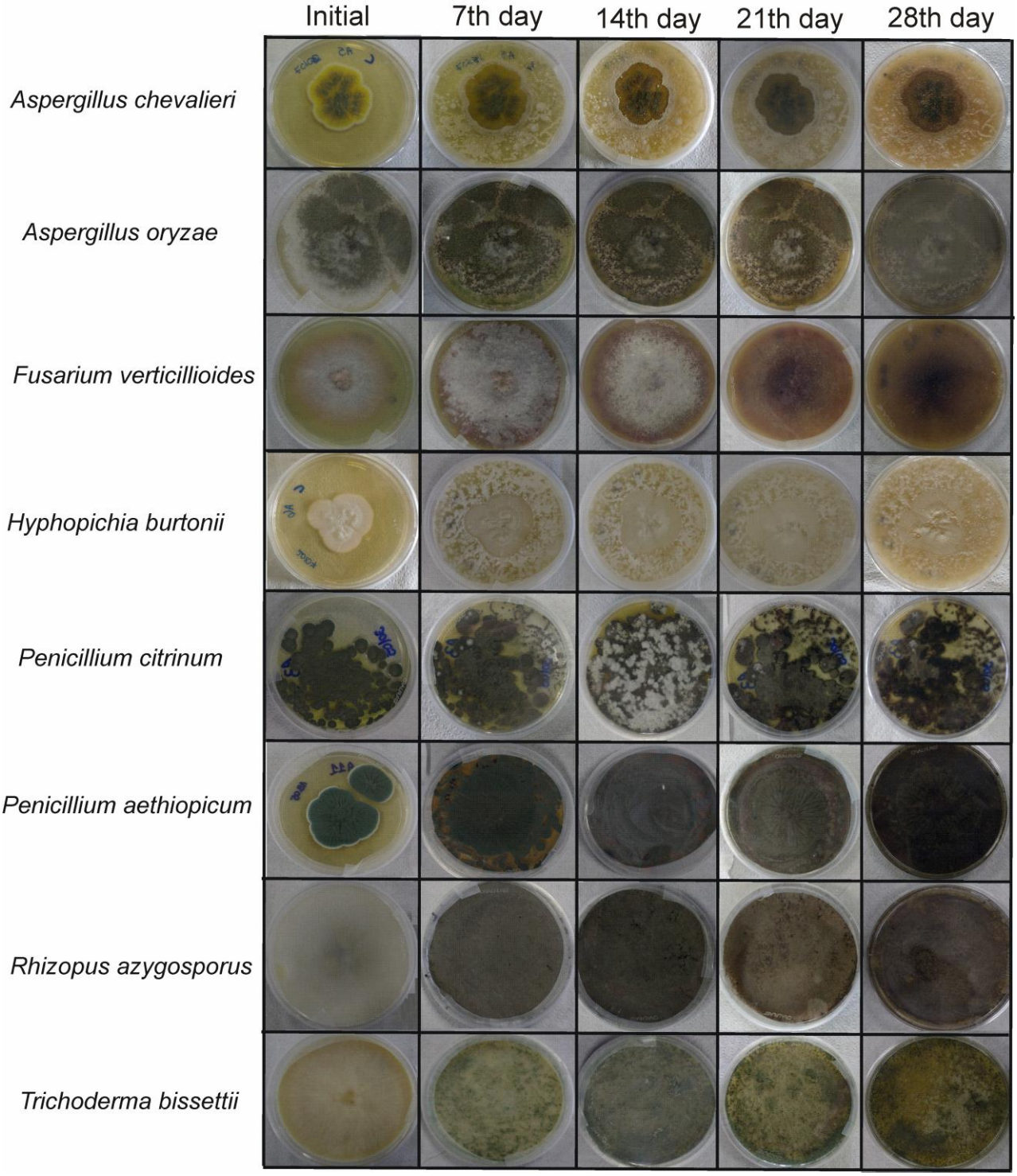


Fig. 3a Medically important fungal species utilized at the 1st phase of experiment. Initial (after one week of fungal inoculation, fifty mites were added), 7th day, 14th, 21th day and 28th day with *T. putrescentiae*.

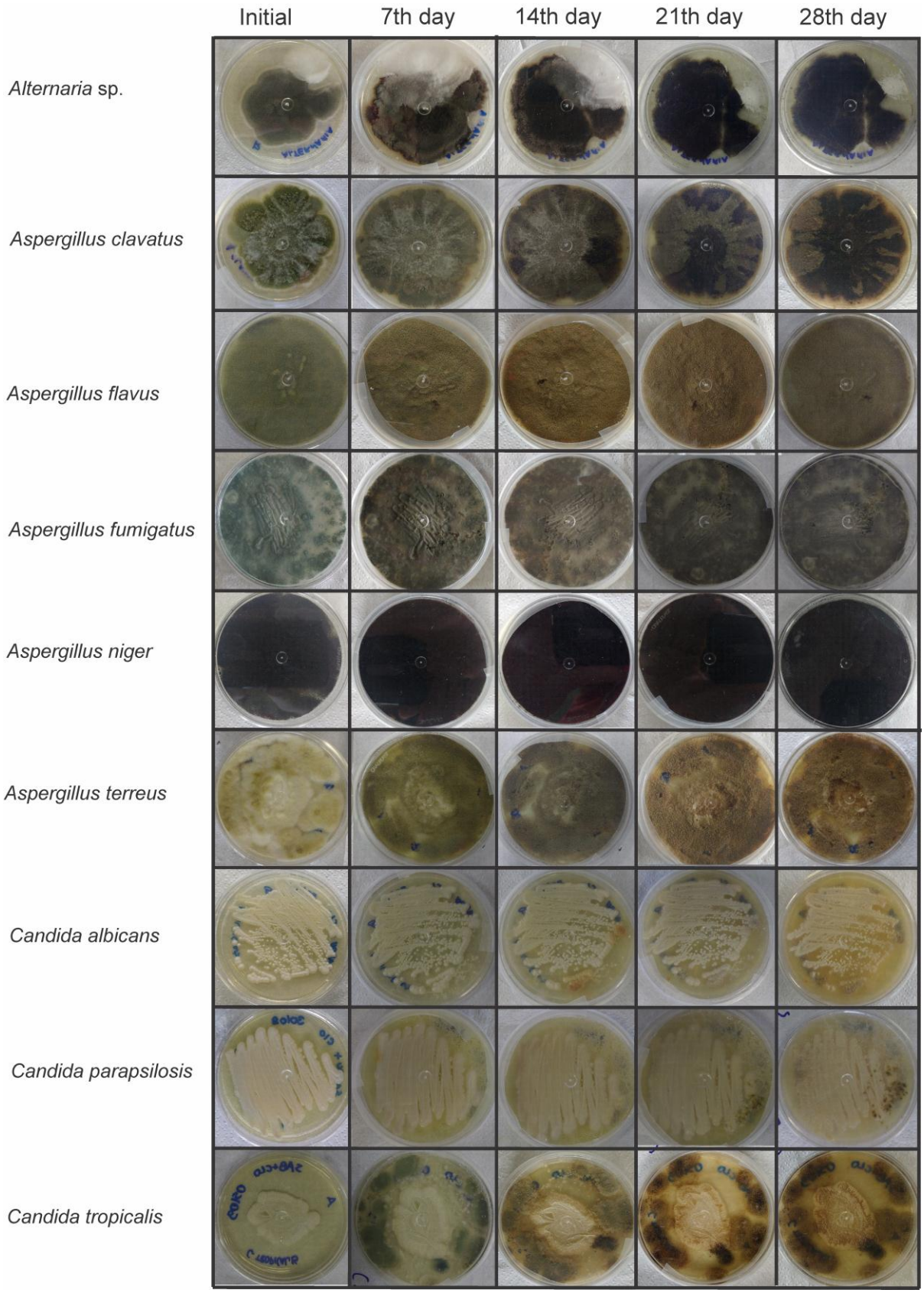


Fig. 3b Medically important fungal species utilized at the 1st phase of experiment. Initial (after one week of fungal inoculation, fifty mites were added), 7th day, 14th, 21th day and 28th day with *T. putrescentiae*.

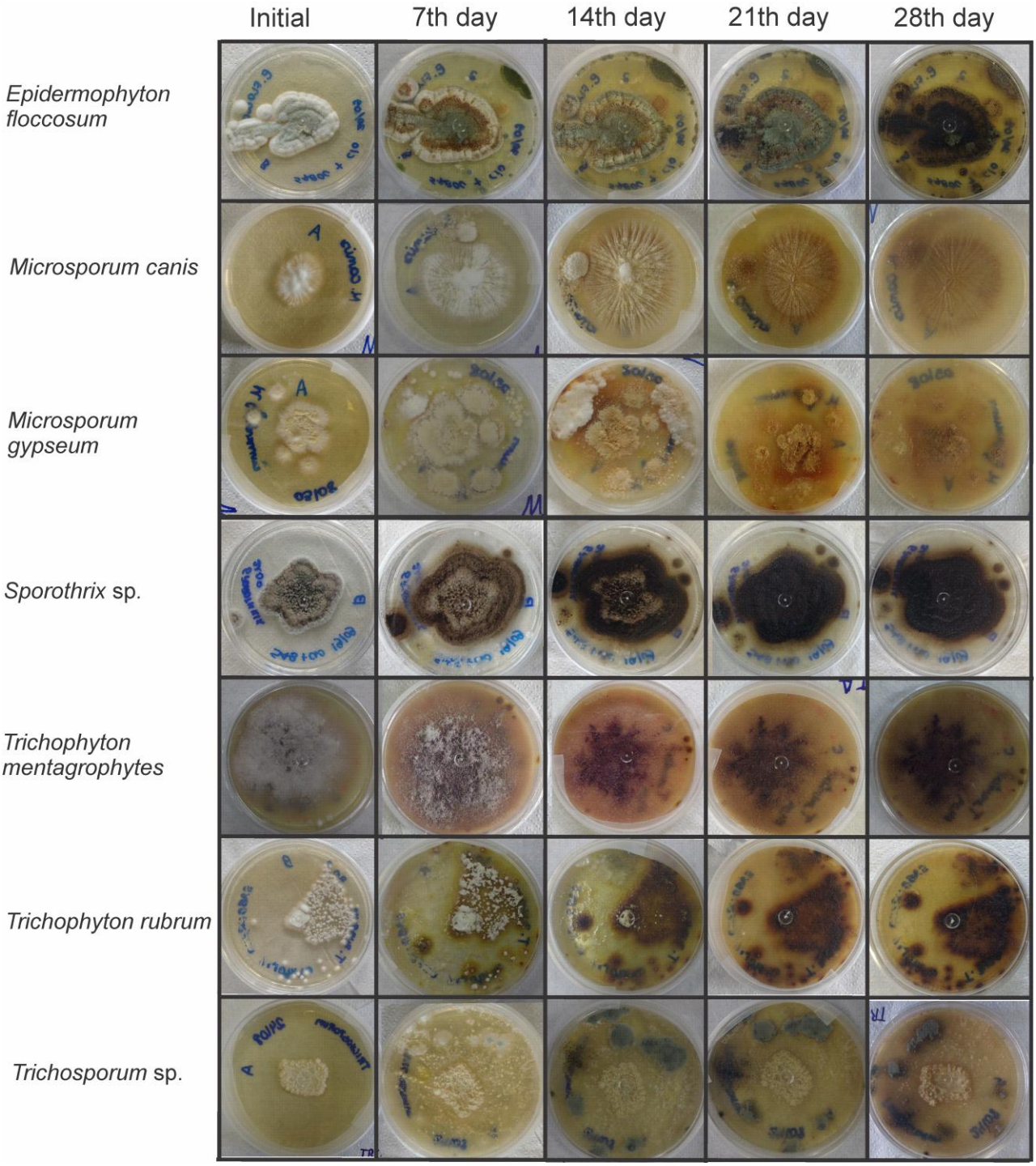


Fig. 4a Environmental fungal species utilized at the 2nd phase of experiment. Showing the effects of *T. putrescentiae* grazing on fungal species 7th day, 14th, 21th day and 28th day.

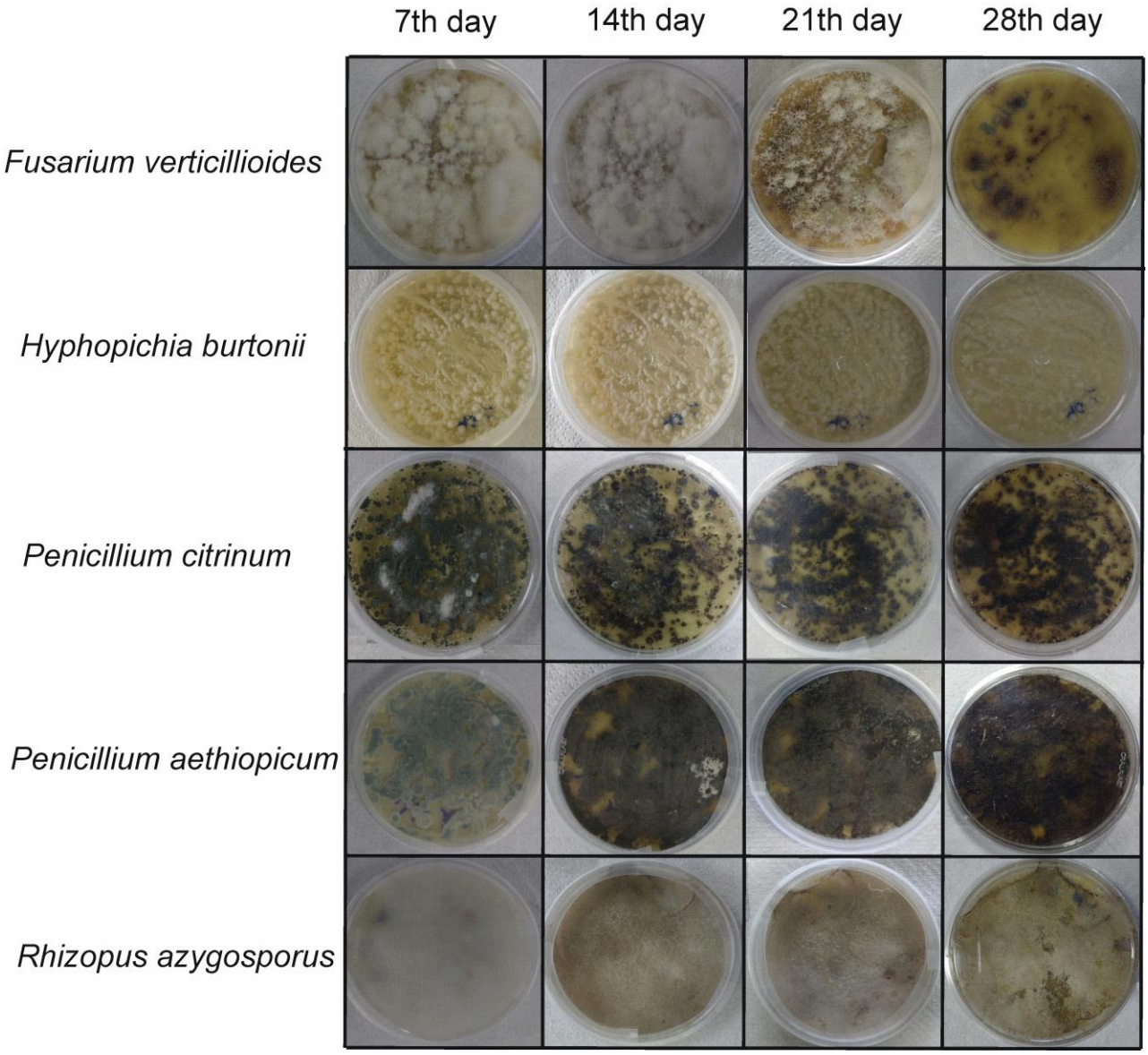
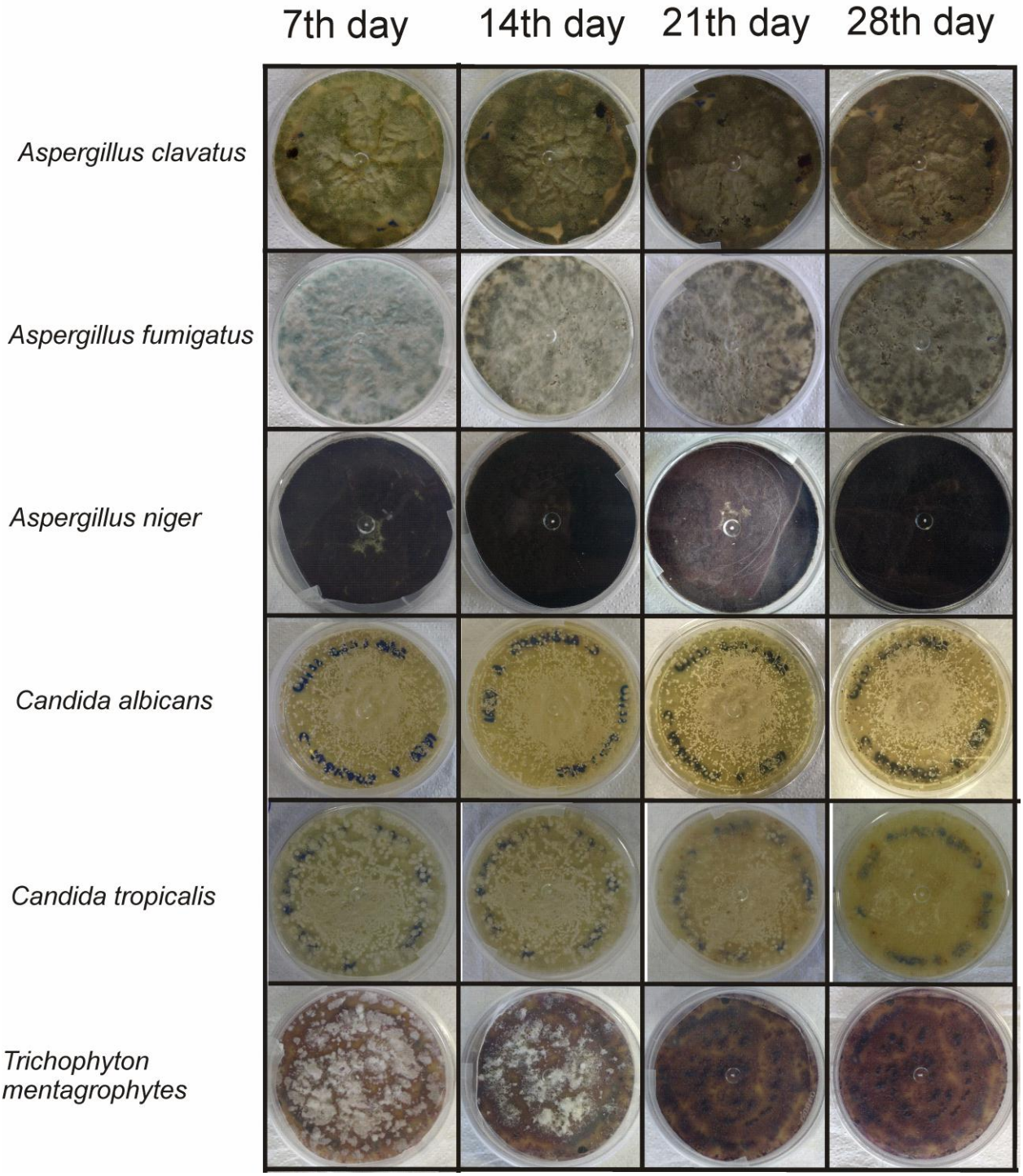


Fig. 4b Medically important fungal species utilized at the 2nd phase of experiment. Showing the effects of *T. putrescentiae* grazing on fungal species at 7th day, 14th, 21th day and 28th day.



5.5. RELATÓRIO DO DOUTORADO SANDUÍCHE - University of Michigan

No período de 01/Abril até 01/Agosto de 2017, realizei um estagio de Doutorado-Sanduíche na conceituada *University of Michigan,* Estados Unidos, financiado pela CAPES.

A *University of Michigan* (UMICH) está situada na cidade de Ann Arbor, sendo um dos mais importantes centros de pesquisa universitários dos Estados Unidos. Foi fundada em 1817, é pública e possui sedes em Ann Arbor, em Flint e Dearborn. Conta com mais de 500 grandes edifícios, três hospitais universitários, dezenas de ambulatórios, muitos centros de cuidados médicos, pesquisas e educação.

Ann Arbor é considerada uma cidade universitária, onde grande parte da economia da cidade gira em torno da universidade, empregando a maioria da população. A universidade administra um dos maiores orçamentos anuais de pesquisa colegiada de qualquer universidade nos Estados Unidos, totalizando cerca de $ 1 bilhão em 2009.

As 19 bibliotecas espalhadas pela cidade, o maior estádio de futebol de faculdade do país (um dos maiores estádios de futebol do mundo) e as residências estudantis chamam a atenção na cidade. O sistema residencial dos estudantes é organizado em "comunidades temáticas", onde os alunos têm a oportunidade de estar cercada por estudantes em uma sala residencial que compartilham interesses semelhantes, estimulando a liderança global, experiência de transição da faculdade e internacionalismo.

O pesquisador Dr. Pavel B. Klimov foi meu orientador nesta universidade. Ele é pesquisador do programa de Pós-Graduação em *Ecology and Evolutionary Biology* na *College of Literature, Science and the Arts - University of Michigan*. Seus interesses de pesquisa atuais incluem filogenia molecular, relações parasito-hospedeiro, co-evolução e sistemática de ácaros. Klimov é uma *forte referência* com diversos artigos publicados em revistas conceituadas na área de ecologia e evolução.

Figura 3. Entrada do Laboratório de Acarologia da UMICH.

O Laboratório de Acarologia faz parte do *Ruthven Alexander G Museums Building* representado pelo Departamento de *Ecology and Evolutionary Biology* (Figura 3 e 4) e conta com diversos pesquisadores. O laboratório é uma das principais referências na área de acarologia no mundo. O eminente acarologista Barry O'Connor é o chefe do laboratório e coordenador das pesquisas. No período que estive no laboratório acompanhei todas as suas atividades. A qualidade dos laboratórios e acesso a infraestrutura da universidade de estudos ou centro de pesquisa são excelentes.

Figura 4. Visão externa do *Ruthven Alexander G Museums Building*.

O objetivo principal do meu projeto de pesquisa no exterior foi trabalhar na incompatibilidade citoplasmática e ferramentas de bioinformática sobre o ácaro *T. putrescentiae*. O foco principal foi realizar cruzamentos entre populações de diferentes lugares do mundo e caracterizar a nível molecular cada população com intuito de reconhecer quais microrganismos (principalmente bactérias endossimbiontes) podem estar causando a incompatibilidade citoplasmática (se comprovada). Ainda na minha estadia trabalhei com ferramentas de bioinformática a fim de comparar a microbiota no genoma de *T. putrescentiae* e *Dermatophagoides farinae*, avaliando a diversidade de microrganismos que estão associados ao gene COI destas duas espécies. Desta forma podem-se compreender mais as interações dos microrganismos e os ácaros.

Nos quatro meses de laboratório, fiquei responsável pelas diversas culturas de *T. putrescentiae* coletadas de vários lugares do mundo (Figura 5).

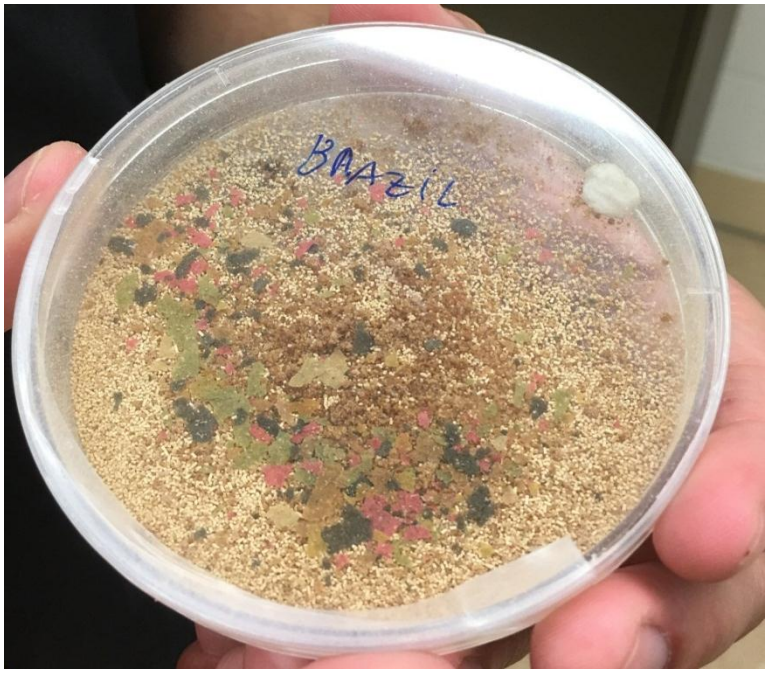


Figura 5. Culturas de *T. putrescentiae* em placas de Petri: comida de peixe e pellets de levedura seca oferecidas como fonte de alimento.

Para o estudo de comparação da microbiota entre *T. putrescentiae* e *D. farinae* foram utilizados os programas FASTQC, BBDUK, CLARK, KRONA e SPADES – Assembly. O trabalho ainda está em fase de estudo, abaixo segue resultado parcial da microbiota utilizando o programa KRONA (Figura 6):

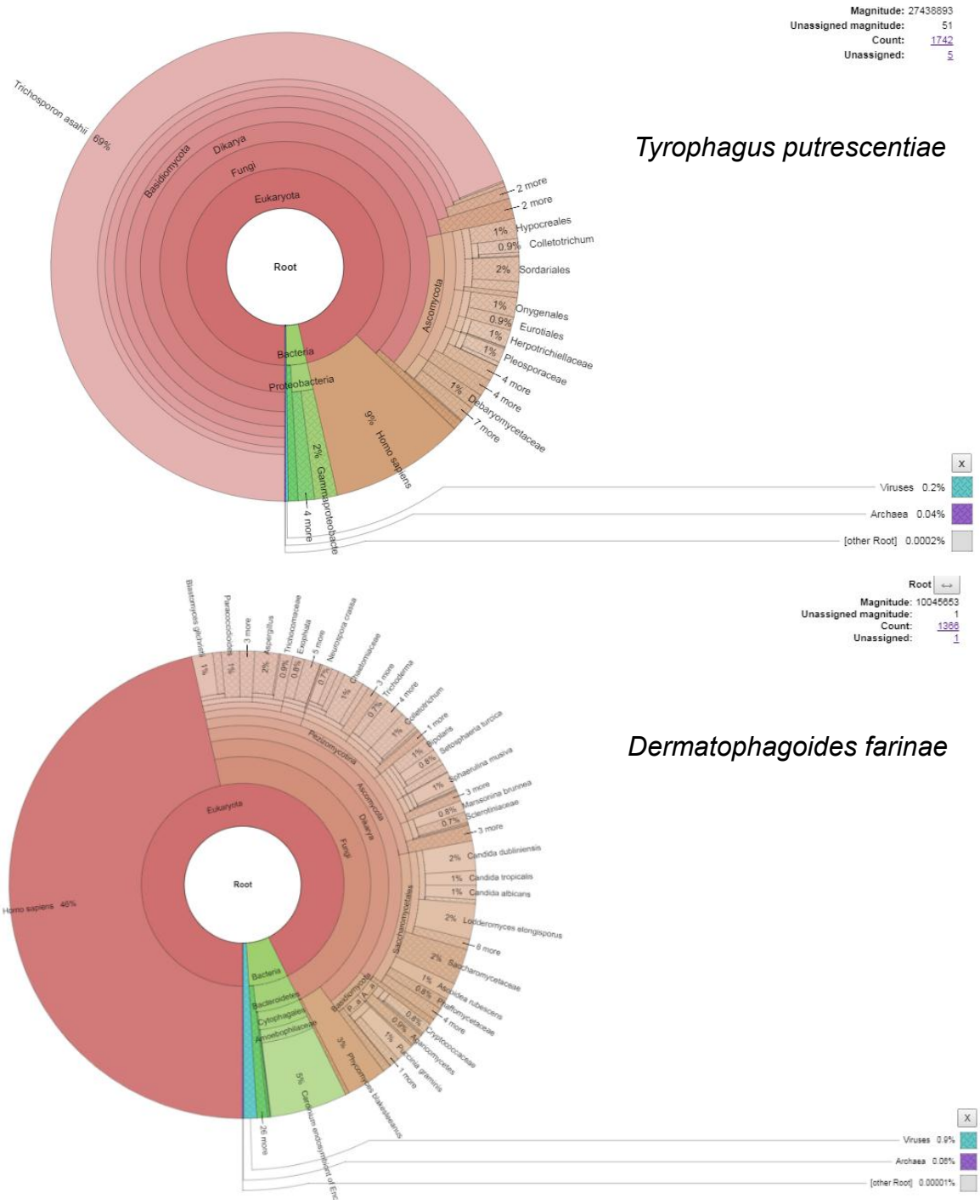


Figura 6. Imagens geradas pelo programa KRONA.

A partir destes resultados, possivelmente haverá a publicação de um artigo com o objetivo de conhecer os microrganismos associados a estas duas espécies de importância mundial para a saúde e alimentos. Outra publicação será relacionada aos cruzamentos de populações de *T. putrescentiae,* sendo que durante minha estadia

foram sequenciados o DNA destas populações e os cruzamentos estão sendo realizados aqui no Brasil.

Mais informações relacionadas ao Dr. Pavel Klimov podem ser encontradas no site https://lsa.umich.edu/eeb/people/research-scientists/pklimov.html.

Minha adaptação à cultura e à experiência, no geral, foram extremamente enriquecedoras.

6. DISCUSSÃO GERAL

Na presente tese são apresentados dois temas distintos dentro da acarologia. São eles: Ácaros de ecossistemas aquáticos e Ácaros de ambientes associados à indústria de alimentos. Embora os assuntos sejam diferentes, ambos apresentam algo grau de importância para o aprofundamento no estudo dos ácaros, principalmente aqui no Brasil.

Esta tese contribuirá significativamente para elucidar determinadas lacunas no conhecimento destes ácaros, bem como fomentar novas ideias para futuros estudos mais aplicados.

Pela primeira vez no Brasil foi realizado um levantamento faunístico da diversidade de ácaros aquáticos utilizando-se como foco de estudo alguma cultura arrozeira, de grande importância econômica. Foram encontradas diferenças na composição das comunidades de ácaros aquáticos entre as áreas de arroz e as lagoas nativas em que a turbidez representou um dos parâmetros ambientais que influenciou a composição da comunidade de ácaros aquáticos nas áreas avaliadas. Além disso, as lagoas nativas apresentaram uma maior riqueza de espécies em comparação com as áreas de arroz irrigado. A turbidez da água é causada pelo sedimento suspenso. Estes podem ser originários a partir do aumento de microorganismos, incluindo bactérias e algas, e a entrada externa de lixo, detritos e carcaças de invertebrados em decomposição (LAESSLE, 1961; MAGUIRE JR, 1971). Além disso, os altos níveis de turbidez podem diminuir a penetração da luz e reduzir a qualidade da água, reduzindo significativamente os recursos hídricos utilizados por esses ácaros (COPATTI *et al*., 2013). Algumas das espécies de ácaros aquáticos são fortemente fototrópicas positivas. No entanto, os representantes de cada família de ácaros responde de forma diferente aos vários comprimentos de onda (ROBERTS *et*

al., 1978). Várias espécies de ácaros aquáticos exploram plantas aquáticas, uma vez que estas plantas criam o substrato necessário para os seus ciclos de vida e hospedeiros. Vários ácaros aquáticos depositam seus ovos e se transformam de deutoninfas para tritoninfa entre os musgos aquáticos e macrófitas (SMITH *et al*., 2009). As macrófitas oferecem condições adequadas para vários tipos de hospedeiro, desempenhando um grande papel na dispersão e colonização de novos pontos por estes ácaros (MARTIN, 2008). Portanto, a presença de macrófitas em geral em lagoas provavelmente é um fator que influencia a diversidade destes ácaros. Więcek *et al*., (2013) relataram que algumas espécies de *Arrenurus* estão associadas à abundância de macrófitas. A conservação de lagoas nativas é muito importante para preservar a biodiversidade. Assim, os ácaros aquáticos podem ser utilizados como bioindicadores de diversidade ao comparar ambientes naturais e antropizado. Young (1969) descreveu que as comunidades de ácaros aquáticos (diversidade, abundância e estrutura da comunidade) são sensíveis à contaminação. Várias espécies sensíveis são restritas a água muito limpa e respondem imediatamente à contaminação precoce de seu ambiente. Outro estudo descreveu que a diversidade de ácaros aquáticos foi significativamente maior em locais não poluídos (GROWNS, 2001). Nossos resultados corroboram que as áreas antropizadas têm menor riqueza de ácaros aquáticos do que as áreas nativas, como sugerido por Katayama *et al.,* (2015). Os autores observaram um declínio na riqueza e abundância em um campo de arroz bem consolidado. A partir deste estudo, poderemos desenvolver mais trabalhos relacionados ao uso destes organismos como bioindicadores de qualidade ambiental.

Não obstante, em outra área emergente na acarologia faz-se necessário ampliar os estudos, como exemplo o ácaro de produtos armazenados e suas interações com microrganismos e alimentos. Sabe-se que *T. putrescentiae* causa sérios problemas na indústria de alimentos, asma alérgica em seres humanos e ainda possui um histórico de interações com diversos microrganismos. A capacidade de oviposição sobre diferentes temperaturas e o seu desenvolvimento biológico quando alimentado com várias dietas, ressalta o fato desta espécie facilmente colonizar inúmeros habitats (ERBAN *et al.*, 2015). Nosso estudo demonstrou que *T. putrescentiae* se desenvolveu e reproduziu com sucesso quando alimentado sobre as cinco dietas. Os parâmetros da tabela de vida diferiram entre as dietas, apresentando melhor desempenho sobre levedura seca. Todavia, quando diferentes dietas foram oferecidas simultaneamente, as fêmeas de *T. putrescentiae* ovipositaram

significativamente mais em ração de rato (aos 18 e 25ºC) e farinha de milho (a 30ºC). A levedura seca exibiu os piores índices quanto à preferência por oviposição, demonstrando que esta espécie não escolhe a dieta que melhor desenvolve seu ciclo, mas sim, o alimento que apresenta as melhores condições de abrigo, seja devido pela forma ou textura.

A capacidade deste ácaro de se alimentar de várias dietas pode ser associada com suas peças bucais que são adaptadas a várias atividades como perfuração, mastigação, além de rasgar e sugar (ALBERTI & COONS, 1999). Estes ácaros penetram nos grãos mais duros e causam mudanças internas. Os fragmentos de alimentos são mastigados com a ajuda das quelíceras e outras partes bucais, adaptados para assimilar pequenas partículas (AKIMOV, 1977; SINHA, 1979).

Além disso, a temperatura e o tipo de dieta são considerados os parâmetros chaves para desenvolvimento e capacidade reprodutiva desta espécie de ácaro (QU *et al.*, 2015). A modificação da temperatura pode ser uma ferramenta útil para limitar infestações e reduzir a mobilidade em diferentes habitats. Esses fatores poderiam explicar o fato de que, a baixas temperaturas, o desenvolvimento de *T. putrescentiae* não foi afetado pelo tipo de dieta. Assim, este estudo corroborou parcialmente com Erban *et al*., (2009), afirmando que *T. putrescentiae* é relativamente mal adaptado aos substratos ricos em carboidratos. Dietas como levedura seca, ração de rato e farinha de milho apresentaram concentrações equilibradas de lipídios, carboidratos e proteínas, proporcionando assim uma dieta mais nutritiva para as espécies do que as demais dietas testadas, como farinha de arroz e farinha de sangue.

O sucesso de *T. putrescentiae* em diferentes habitats é relacionado a sua capacidade de utilizar diversas fontes de alimento. Um exemplo desta amplitude na sua habilidade de utilizar outras fontes de alimento foi a partir do estudo desenvolvido na tese quando foi avaliado o desenvolvimento das populações sobre 24 espécies de fungos, sendo oito isolados de amostras de alimentos e 16 provenientes da Coleção do laboratório de Micologia da UFRGS. *Tyrophagus putrescentiae* é capaz de se desenvolver utilizando exclusivamente fungos como fonte de alimento. Os melhores índices de crescimento ocorreram sobre os fungos *Fusarium verticillioides*, *Penicillium citrinum* e *Trichophyton mentagrophytes,* provavelmente devido à facilidade com que o ácaro conseguiu digerir suas hifas, bem como apresentar propriedades nutricionais extremamente satisfatórias para o ácaro. Além dos fungos citados acima, outros fungos obtiveram sucesso na sua dispersão pelos ácaros, estes são: *Candida*

tropicalis, *Hyphopichia burtonii*, *Candida albicans*, *Aspergillus clavatus*, *Rhizophus* sp., respectivamente.

O esplendoroso desenvolvimento de *T. putrescentiae* sobre *T. mentagrophytes*, agente etiológico de dermatomicoses é preocupante. *Trichophyton mentagrophytes* é considerado zoonótico e pode ser transmitido de animais para humanos através do contato direto. Portanto, as interações entre animal-ácaro-humano em domicílios poderá potencializar casos clínicos de dermatomicose causada por esse fungo.

O crescimento positivo de *T. putrescentiae* sobre *A. clavatus* nas duas fases do estudo representa um risco em potencial à medida que as duas espécies são pragas que se desenvolvem em cereais e outros produtos utilizados na alimentação animal (VARGA *et al*., 2003). Como já de conhecimento, *T. putrescentiae* é a espécie acarina mais abundante e predominante em materiais orgânicos e produtos alimentares à base de cereais. Assim, esta associação com *A. clavatus* pode ocorrer em produtos armazenados (KHERADMAND *et al*., 2007), áreas de processamento e embalagem de alimentos para animais (RYBANSKA *et al*., 2016), e ainda podem causar anafilaxia em humanos através da ingestão de alimentos contaminados (SANCHEZ-BORGES *et al*., 2009).

Foi demonstrado que *T. putrescentiae* é capaz de dispersar patógenos leveduriformes oportunistas de humanos (DUEK et al., 2001). Assim, nosso estudo corroborou com Nesvorná *et al*., (2012), que abordou uma forte interação entre fungos do gênero *Fusarium* e *T. putrescentiae*, sendo observado em nosso estudo o consumo de hifas por parte do ácaro sobre as placas avaliadas. As infestações do ácaro poderiam disseminar o fungo em produtos armazenados atingindo níveis de danos econômicos sem precedentes.

Algumas espécies de fungos foram dispersas pelos ácaros. No entanto, o crescimento em ambas as fases das populações não foram positivas. Esta disseminação de fungos não utilizados como alimento aponta para uma dispersão fortuita de esporos anexados ao corpo do ácaro, independente de qualquer interação entre o ácaro e o fungo.

T. putrescentiae demonstrou ser uma importante vetor de disseminação de fungos, não evidenciando uma distinção em seus hábitos nem para fungos de interesse médico, ou fungos ambientais coletados de amostras de alimentos. Além disso, não demonstrou preferência de alimentação sobre qualquer morfologia fúngica específica (filamentosa ou leveduriforme). Assim, sugere-se que *T. putrescentiae*

possua uma microbiota com atividade quitinolítica que auxilia através de uma simbiose a digestão de paredes de células de fungos (SMRŽ *et al*., 1991; SMRŽ & ČATSKÁ, 2010), pois os hábitos alimentares são alterados devido à microbiota associada aos tecidos intestinais e parenquimatosos (SMRŽ, 2003).

Sugerimos que vários casos clínicos fúngicos podem ser transmitidos pelo ácaro e não por outras vias de transmissão já descritas na literatura. São necessários mais estudos para entender os processos de dispersão simultânea de microorganismos em ambientes domiciliares e hospitais, onde há uma maior densidade de pacientes imunossuprimidos e registros de casos clínicos fúngicos. Outras espécies de fungos clínicos devem ser testadas em contato com este ácaro, sendo necessário conhecer a microbiota associada a ácaros em locais com hospedeiros imunossuprimidos, para avaliar a gestão das condições sanitárias.

Devido à dada importância de *T. putrescentiae* como uma relevante praga de produtos armazenados e causadores de alergias, diversas alternativas de suprimir as suas infestações foram realizadas. Neste trabalho, apresentamos uma alternativa utilizando o ácaro predador *B. dentriticus* com potencial de utilizar *T. putrescentiae* exclusivamente como fonte de alimento. Como forma de comparação, uma fonte de alimento secundária foi testada: o ectoparasito de galinhas poedeiras *M. ginglymura*. Este predador apresentou sucesso no seu desenvolvimento e capacidade de predação sobre *T. putrescentiae* e *M. ginglymura*. Corroborando com Mohamed (2013), os melhores índices de desenvolvimento para este predador são apresentados quando alimentados por astigmatídeos. Nossos estudos sugerem que são necessárias experiências de campo em larga escala sobre o potencial deste predador. Os testes de predação que investigam a capacidade de eliminar a presa seriam os próximos passos. O consumo, como o número de presas, e o comportamento de *B. dentriticus* em condições de campo ainda não são suficientemente conhecidos. Ždárkova & Horák (1990) relataram que os armazéns, onde os produtos são armazenados, não é um espaço completamente fechado, sugerindo que outros ácaros poderiam invadir este habitat, trazidos através do homem, animais como insetos, aves e roedores. Alguns predadores frequentemente podem conviver simultaneamente no mesmo ambiente, porem, podem ser influenciadas negativamente. Por exemplo, em condições de laboratório, o ácaro predador *Cheyletus eruditus* (Schrank) influenciou negativamente nas populações de *B. dentriticus*, causando a eliminação de *B. dentriticus* localizados no mesmo

recipiente (BURNETT, 1977). Contudo, os resultados podem diferir em uma situação real, como em armazéns.

7. CONCLUSÃO

Lagoas nativas apresentaram maior riqueza e abundância de ácaros aquáticos quando comparados com as áreas de arroz irrigado.

A turbidez demonstrou ser o parâmetro influenciador na composição de ácaros aquáticos.

O ácaro *B. dentriticus* é um predador em potencial, desenvolvendo e reproduzindo com sucesso quando se alimenta de *T. putrescentiae* e *M. ginglymura*, apresentando melhor desempenho quando se alimenta de *T. putrescentiae*.

No estudo do ciclo de vida, *T. putrescentiae* apresentou melhor desempenho alimentando-se de levedura seca. No entanto, quando oferecidas várias dietas simultaneamente, demonstrou preferência em ovipositar mais sobre ração de rato (em 18ºC e 25ºC) e em farinha de milho (30ºC).

Populações de *T. putrescentiae* desenvolveram-se com sucesso, bem como serviram como agente de dispersão dos fungos *Fusarium verticillioides*, *Penicillium citrinum*, *Trichophyton mentagrophytes, Candida tropicalis*, *Hyphopichia burtonii*, *Candida albicans*, *Aspergillus clavatus* e *Rhizopus azygosporus,* demonstrando que estes ácaros são um importante vetor de disseminação de fungos clínicos.

8. REFERÊNCIAS

AGÊNCIA NACIONAL DE VIGILÂNCIA SANITÁRIA. Resolução RDC nº 7, de 18 de fevereiro de 2011. **Diário Oficial da União**, Brasília, DF, 22 fev. 2011. Seção 1, p. 72.

AGÊNCIA NACIONAL DE VIGILÂNCIA SANITÁRIA. Resolução RDC n° 14, de 28 de março de 2014. **Diário Oficial da União**, Brasília, DF, 31 mar. 2014. Seção 1, p. 58.

AKIMOV, I.A. Determination of the injuriosness of storage mites. Zashch Rast (Moscow) v. 2, p. 44, 1977.

ALBERTI, G.; COONS, L.B. Acari - Mites. In: Harrison, F. W. (Ed.) **Microscopic Anatomy of Invertebrates**. New York, John Wiley & Sons, p. 515-1265, 1999.

ARLIAN, L.G.; GEIS, D.; VYSZENSKI-MOHER, D.; BERNSTEIN, I.; GALLAGHER, J. Antigenic and allergenic properties of the storage mite *Tyrophagus putrescentiae*. **J Allergy Clin Immunol**, v. 74, n. 2, p. 166-71, 1984.

ARLIAN, L.G.; MORGAN, M.S.; VYSZENSKI-MOHER, D.L.; SHARRA, D. Cross-reactivity between storage and dust mites and between mites and shrimp. **Exp Appl Acarol**, v. 47, n. 2, p. 159-72, 2009.

ARLIAN, L.G.; VYSZENSKI-MOHER, D.L.; JOHANSSON, S.G.O.; HAGE-HAMSTEN, M. Allergenic characterization of *Tyrophagus putrescentiae* using sera from occupationally exposed farmers. **Ann Allergy Asthma Immunol**, v. 79, n. 6, p. 525-29, 1997.

ASPALY, G.; STEJSKAL, V.; PEKÁR, S.; HUBERT, J. Temperature-dependent population growth of three species of stored product mites (Acari: Acaridida). **Exp Appl Acarol**, v. 42, n. 1, p. 37-46, 2007.

ATUÍ, M.B.; LÁZZARI, F.A. Monitoramento de fungos em milho em grão, grits e fubá. **Ciênc Tecnol Aliment**, v. 18, n. 4, p. 363-67, 1998.

AYGUN, O.; YAMAN, M.; DURMAZ, H. A survey on occurrence of *Tyrophagus putrescentiae* (Acari: Acaridae) in Surk, a traditional Turkish dairy product. **J Food Eng**, v. 78, n. 3, p. 878-81, 2007.

BAGGIO, D.; FIGUEIREDO, S.M.; FLECHTMANN, C.H.W.; ZAMBON, G.Q.; MIRANDA, S.H.G. Avaliação da presença de ácaros em cereais armazenados na Grande São Paulo. **An Esc Super Agric Luiz de Queiroz Univ São Paulo**, v. 44, n. 1, p. 617-26, 1987.

BALIUKONIENE, V.; BAKUTIS, B.; STANKEVICIUS, H. Mycological and mycotoxicological evaluation of grain. **Ann Agric Environ Med**, v. 10, p. 223-27, 2003.

BARRERA, O.M.; MURGAS, I.L.; BERMÚDEZ, S.; MIRANDA, R.J. Oral anaphylaxis by ingestion of mite contaminated food in Panama City, 2011-2014. **Rev Alerg Mex**, v. 62, n. 2, p. 112-17, 2015.

BARTSCH, I. Halacarids (Halacaroidea, Acari) in freshwater. Multiple invasions from the Paleozoic onwards. **J Nat Hist**, v. 30, p. 67-99, 1996.
BARTSCH, I. 2004. Geographical and ecological distribution of marine halacarid genera and species (Acari: Halacaridae). **Exp Appl Acarol**, v. 34, p. 37-58.

BIESIADKA, E.; KOWALIK, W. Water mites (Hydracarina) as indicators of trophy and pollution in lakes. In: Dusbabek F, Bukva V (Eds) **Modern Acarology** 1. SPB Academic Publishing BV, The Hague, p. 475-481, 1991.

BRAVO, C.M.; ORTIZ, I.L.; SOTO, A.O.; VAZQUEZ, R.G. 1999. Allergy to storage mites. **Allergy**, v. 54, n. 7, p. 769-70.

BROWN, A.N.; LLOYD, V.K. Evidence for horizontal transfer of *Wolbachia* by a *Drosophila* mite. **Exp Appl Acarol**, v. 66, n.3, p. 301-11, 2015.

BURNETT, T. Biological models of two acarine predators of the grain mite, *Acarus siro* L. **Can J Zool**, v. 55, p. 1312-23, 1977.

CLAUSEN, C.P. Phoresy among entomophagous insects. **Annu Rev Entomol.** v. 21, p. 343-367, 1976.

COLLOFF, M.J. 2009. **Dust Mites**. Collingwood, Australia: CSIRO Publishing, and Dordrecht: Springer.

COPATTI, C.E.; FAGUNDES, L.S.; QUAINI, J.B.; COPATTI, B.R. Diversity of aquatic arthropods on *Eichhornia crassipes* (Mart.) Solms roots before and after removal of substrate in a reservoir in southern Brazil. **Panam J Aquat Sci,** v. 8, p. 265-75, 2013.

DI SABATINO, A.; MARTIN, P.; GERECKE, R.; CICOLANI, B. Hydrachnidia (water mites). In: Rundle SD, Robertson AL, Schmid-Araya JM (Eds) **Freshwater meiofauna: biology and ecology.** Backhuys Publishers, Leiden, p. 105-133, 2002.

DI SABATINO, A.; SMIT, H.; GERECKE, R.; GOLDSCHMIDT, T.; MATSUMOTO, N.; CICOLANI, B. Global diversity of water mites (Acari, Hydrachnidia; Arachnida) in freshwater. **Hydrobiologia**, v. 595, p. 303-15, 2008.

DOHET, A.; ECTOR, L.; CAUCHIE, H.; HOFFMANN, L. Identification of benthic invertebrate and diatom indicator taxa that distinguish different stream types as well as degraded from reference conditions in Luxembourg. **Animal Biol**, v. 58, n. 4, p. 419-72, 2008.

DUEK, L.; KAUFMAN, G.; PALEVSKY, E.; BERDICEVSKY, I. Mites in fungal. **Micoses**, v. 44, p. 390-94, 2001.

ELDER, B.L.; MORGAN, M.S.; ARLIAN, L.G. Effect of stored product mite extracts on human dermal microvascular endothelial cells. **J Med Entomol**, v. 49, n. 6, p. 1411-18, 2012.

ERBAN, T.; HUBERT, J. Digestive function of lysozyme in synanthropic acaridid mites

enables utilization of bacteria as a food source. **Exp Appl Acarol**, v. 44, n. 3, p. 199-212, 2008.

ERBAN, T.; ERBANOVA, M.; NESVORNA, M.; HUBERT, J. The importance of starch and sucrose digestion in nutritive biology of synanthropic acaridid mites: Alpha-amylases and alpha-glucosidases are suitable targets for inhibitor-based strategies of mite control. **Arch Insect Biochem Physiol**, v. 71, p. 139-58, 2009.

ERBAN, T.; RYBANSKA, D.; HUBERT, J. Population Growth of the Generalist Mite *Tyrophagus putrescentiae* (Acari: Acaridida) Following Adaptation to High- or Low-Fat and High- or Low-Protein Diets and the Effect of Dietary Switch. **Environ Entomol**, v. 44, p. 1599-604, 2015.

ERBAN, T.; KLIMOV, P.B.; SMRZ, J.; PHILLIPS, T.W.; NESVORNA, M.; KOPECKY, J.; HUBERT, J. Populations of Stored Product Mite *Tyrophagus putrescentiae* Differ in Their Bacterial Communities. **Front Microbiol**, v. 7, p. 1046, 2016.

ERBEN, A.M.; RODRIGUEZ, J.L.; MCCULLOUGH, J.; OWNBY, D.R. 1993. Anaphylaxis after ingestion of beignets contaminated with *Dermatophagoides farinae*. **J Allergy Clin Immunol**, v. 92, n. 6, p. 846-49.

ESTÉVEZ, M.D. Occupational contact urticaria-dermatitis by *Tyrophagus putrescentiae*. **Contact Derm**, v. 55, n. 5, p. 308-09, 2006.

EVANS, G.O. **Principles of acarology**. CAB International, Wallingford. 1992.

FIELDS, P.G.; WHITE, N.D.G. Alternatives to methyl bromide treatments for stored-product and quarantine insects. **Annu Rev Entomol,** v. 47, p. 331-59, 2002.

FLECHTMANN, C.H.W. **Ácaros em produtos armazenados e na poeira domiciliar**. FEALQ, Piracicaba. 97 p. 1986.

FRANZ, J-T.; MASUCH, G.; MUSKEN, H.; BERGMANN, K-C. Mite fauna of German farms. **Allergy**, v. 52, n. 12, p. 1233-37, 1997.

FRANZOLIN, M.R.; GAMBALE, W.; CUERO, R.G.; CORREA, B. Interaction between toxigenic *Aspergillus flavus* Link and mites (*Tyrophagus putrescentiae* Schrank) on maize grains: effects on fungal growth and aflatoxin production. **J Stored Prod Res**. v. 35, n. 3, p. 215-24, 1999.

GERSON, U.; SMILEY, R.L.; OCHOA, R. **Mites (Acari) for pest control**. Oxford, Blackweell Publishing, UK, 539 p., 2003.

GOLDSCHMIDT, T. Water mites (Acari, Hydrachnidia): powerful but widely neglected bioindicators – a review. **Neotrop Biodivers**, v. 2, n. 1, p. 12-25, 2016.

GRANICH, J.; HORN, T.B.; KÖRBES, J.H.; TOLDI, M.; SILVA, G.L.; FERLA, N.J. 2016. Development of *Cheyletus malaccensis* (Acari: Cheyletidae) feeding on mite species found in commercial poultry systems: *Megninia ginglymura* (Acari: Analgidae) and *Tyrophagus putrescentiae* (Acari: Acaridae). **Syst Appl Acarol**, v. 21, n. 12, p. 1604-13.

GROWNS, J.E. Aquatic mites as bioindicators, with an Australian example. In: Halliday RB, Walter DE, Proctor H et al. (Eds) **Acarology: proceedings of the 10th International Congress.** Melbourne, CSIRO Publishing, p. 136-142, 2001.

HAYS, JR. J.B.; LAWS, J.R. E.R. **Handbook of Pesticide Toxicology**. v. 1. Academic Press, San Diego, 1571 p., 1991.

HIBBERSON, C.E.; VOGELNEST, L.J. Storage mite contamination of commercial dry dog food in south-eastern Australia. Aust Vet J. v. 96, n. 6, p. 219-24, 2014.

HOPE, R.; ALDRED, D.; MAGAN, N. Comparison of environmental profiles for growth and deoxynivalenol production by *Fusarium culmorum* and *F. graminearum* on wheat grain. **Lett Appl Microbiol**, v. 40, p. 295-300, 2005.

HUBERT, J.; JAROSIK, V.; MOUREK, J.; KUBATOVA, A.; ŽDÁRKOVA, E. Astigmatid mite growth and fungi preference (Acari: Acaridida): comparisons in laboratory experiments. **Pedobiologia**, v. 48, n. 3, p. 205-14, 2004.

HUBERT, J.; PEKÁR, S.; NESVORNÁ, M.; SUSTR, V. Temperature Preference and Respiration of Acaridid Mites. **J Econ Entomol**, v. 103, n. 6, p. 2249-57, 2010.

HUBERT, J.; KOPECKY, J.; PEROTTI, M.A.; NESVORNA, M.; BRAIG, H.R.; SAGOVA-MARECKOVA, M.; MACOVEI, L.; ZUREK, L. Detection and identification of species-specific bacteria associated with synanthropic mites. Microb Ecol. v. 63, n. 4, p. 919-28, 2012.

HUBERT, J.; PEKÁR, S.; AULICKÝ, R.; NESVORNÁ, M.; STEJSKAL, V. The effect of stored barley cultivars, temperature and humidity on population increase of *Acarus siro*, *Lepidoglyphus destructor* and *Tyrophagus putrescentiae*. **Exp Appl Acarol**, v. 60, n. 2, p. 241-52, 2013.

HUGHES, A.M. **The mites of stored food and houses**. Tech Bull Minist Agric Fish Food. 400 p., 1976.

JEON, J.H.; LEE, C.H.; LEE, H.S. Food protective effect of geraniol and its congeners against stored food mites. **J Food Prot**, v. 72, n. 7, p. 1468-71, 2009.

KATAYAMA, N.; BABA, Y.G.; KUSUMOTO, Y.; TANAKA, K. A review of post-war changes in rice farming and biodiversity in Japan. **Agricultural Systems**, v. 132, p. 73-84, 2015.

KHERADMAND, K.; KAMALI, K.; FATHIPOUR, Y.; GOLTAPEH, E.M. Development, life table and thermal requirement of *Tyrophagus putrescentiae* (Astigmata: Acaridae) on mushrooms. **J Stored Prod Res,** v. 43, p. 276-81, 2007.

KIM, H.K.; KIM, J.R.; AHN, Y.J. Acaricidal activity of cinnamaldehyde and its congeners *against Tyrophagus putrescentiae* (Acari: Acaridae). **J Stored Prod Res**, v. 40, n. 1, p. 55-63, 2004.

KOPECKY, J.; PEROTTI, M.A.; NESVORNA, M.; ERBAN, T.; HUBERT, J. *Cardinium* endosymbionts are wide spread in synanthropic mite species (Acari: Astigmata). **J Invertebr Pathol**, v. 112, p. 20-23, 2013.

KOPECKY, J.; NESVORNA, M.; HUBERT, J. Bartonella-like bacteria carried by domestic mite species. **Exp Appl Acarol**, v. 64, n. 1, p. 21-32, 2014a.

KOPECKY, J.; NESVORNA, M.; MARECKOVA-SAGOVA, M.; HUBERT, J. The effect of antibiotics on associated bacterial community of stored product mites. **PLoS ONE**, v. 9, p. e112919, 2014b.

LAESSLE, A.M. A micro-limnological study of Jamaican bromeliads. **Ecology**, v. 42, p. 499-517, 1961.

LEE, C.H.; SUNG, B.K.; LEE, H.S. Acaricidal activity of fennel seed oils and their main components against *Tyrophagus putrescentiae*, a stored-food mite. **J Stored Prod Res**, v. 42, n. 1, p. 8-14, 2006.

LIAO, E-C.; HO, C.M.; LIN, M.Y.; TSAI, J.J. *Dermatophagoides pteronyssinus* and *Tyrophagus putrescentiae* Allergy in Allergic Rhinitis Caused by Cross-reactivity Not Dual-Sensitization. **J Allergy Clin Immunol**, v. 30, n. 6, p. 830-39, 2010.

LIAO, E.; HO, C.; YIN, S.; TSAI, J. Immune Responses to *Tyrophagus putrescentiae*-Induced Airway Inflammation in Mice. **J Investig Allergol Clin Immunol**, v. 23, n. 1, p. 20-29, 2013a.

LIAO, E.C.; LIN, Y.H.; CHIU, C.L.; LIN, T.C.; TSAI, J.J. 2013b. Identification of allergenic component Tyr p 8 from *Tyrophagus putrescentiae* and cross-reactivity with Derp 8. **Clin Vaccine Immunol**, v. 20, p. 506-12, 2013b.

LORINI I. 1998. Controle integrado de pragas de grãos armazenados. Passo Fundo, EMBRAPA-CNPT, 52p. (EMBRAPA-CNPT. Documentos, 48).

LUNDBLAD, O. Südamerikanische Hydracarinen. **Zoologiska Bidrag**, v. 13, p. 1-86, 1931.

LUNDBLAD, O. Weitere neue Wassermilben aus Santa Catharina in Südbrasilien. **Zoologischer Anzeiger**, v. 116, p. 14-24, 1936.

LUNDBLAD, O. Vierte Mitteilung über neue Wassermilben aus Santa Catharina in Südbrasilien. **Zoologischer Anzeiger**, v. 118, p. 9-10, 1937.

LUNDBLAD, O. Neue Wassermilben aus Paraguay. **Zoologischer Anzeiger**, v. 122, p. 7-19, 1938.

LUNDBLAD, O. Die Hydracarinenfauna Südbrasiliens und Paraguays. Erster Teil. **Kungliga Svenska Vetenskapsakademiens Handlingar**, v. 19, n. 7, p. 1-183, 1941.

LUNDBLAD, O. Die Hydracarinenfauna Südbrasiliens und Paraguays. Zweiter Teil. **Kungliga Svenska Vetenskapsakademiens Handlingar**, v. 20, n. 2, p. 3-175, 1942.

LUNDBLAD, O. Die Hydracarinenfauna Südbrasiliens und Paraguays. Dritter Teil. **Kungliga Svenska Vetenskapsakademiens Handlingar**, v. 20, n. 5, p. 1-148, 1943a.

LUNDBLAD, O. Die Hydracarinenfauna Südbrasiliens und Paraguays. Vierter Teil. **Kungliga Svenska Vetenskapsakademiens Handlingar**, v. 20, n. 8, p. 1-171, 1943b.

LUNDBLAD, O. Die Hydracarinenfauna Südbrasiliens und Paraguays. Fünfter Teil. **Kungliga Svenska Vetenskapsakademiens Handlingar**, v. 20, n. 13, p. 1-182, 1944.

LUNG-SHU, L. Stored grain mites in the work environment of farmers. In: Griffiths, D.A., Bowman, C.E. (Eds.), **Proceedings of the 6th International Congress of Acarology**, vol. 2, Edinburgh, pp. 1002-05, 1984.

MAGAN, N.; HOPE, R.; CAIRNS, V.; ALDRED, D. Post-harvest fungal ecology: Impact of fungal growth and mycotoxin accumulation in stored grain. **Eur J Plant Pathol**, v. 109, n. 7, p. 723-30, 2003.

MAGUIRE Jr, B. Phytotelmata: Biota and community structure determination in plant-held waters. **Annu Rev Ecol Evol Syst,** v. 2, p. 439-64, 1971.

MARTIN, P. Water mites (Hydrachnidia, Acari) and insects: a survey of a seldom considered relationship. **Entomologie heute**, v. 20, p. 45-75, 2008.

MORAES, G.J.; FLECHTMANN, C.H.W. **Manual de Acarologia, Acarologia Básica e Ácaros de Plantas Cultivadas no Brasil**. Ribeirão Preto, Holos Editora, 308p. 2008.

MOHAMED, A.E.M. Biological aspects and life table parameters of predator gamasid ascid mite, *Blattisocius dentriticus* (Berlese) (Acari: Gamasida: Ascidae). **Egyptian Academic Journal of Biological Sciences**, v. 6:97–105. 2013.

MOSS, M.O. Recent studies of mycotoxins. **J Appl Microbiol**. v.84, n. 27, p. 62-76, 1998.

NESVORNÁ, M.; GABRIELOVA, L.; HUBERT, J. *Tyrophagus putrescentiae* is able to graze and develop on *Fusarium* fungi of mycotoxins importance under laboratory conditions. **J Stored Prod Res,** v. 48, p. 37-45, 2012.

NUÑEZ, N.K.; DA CUNHA, A.A.; DUTRA, M.D.S.; BARBOSA, G.L.; MORASSUTTI, A.L.; DE SOUZA, R.G.; VARGAS, M.H.M.; ANTUNES, G.L.; SILVEIRA, J.S.; SILVA, G.L.; PITREZ, P.M. Acute and chronic exposure to *Tyrophagus putrescentiae* induces allergic pulmonary response in a murine model. **Asia Pac Allergy**, v. 6, n. 1, p. 48-55, 2016.

OLSEN, A.R. Food-contaminating mites from imported foods entering the United States through southern California. **Int J Acarol**, v. 9, p. 189-93, 1983.

OSBORNE, L.E.; STEIN, J.M. Epidemiology of *Fusarium* head blight on small-grain cereals. **Int J Food Microbiol**, v. 119, n. 1-2, p. 103-08, 2007.

PALYVOS, N.E.; EMMANOUEL, N.G.; SAITANIS, C.J. Mites associated with stored products in Greece. **Exp Appl Acarol**, v. 44, n. 3, p. 213-26, 2008.

POSTHUMUS, J.; BORISH, L. A 71-year-old man with anaphylaxis after eating grits. **Allergy & Asthma Proceedings**, v. 33, n. 1, p. 1110-113, 2012.

QU, S.X.; LI, H.P.; MA, L.; SONG, J.D.; HOU, L.J.; LIN, J.S. Temperature-dependent development and reproductive traits of *Tyrophagus putrescentiae* (Sarcoptiformes: Acaridae) reared on different edible mushrooms. **Environ Entomol**, v. 44, p. 392-99, 2015.

REVSBECH, P.; DUEHOLM, M. Storage mite allergy among bakers. **Allergy**, v. 45, n. 3, p. 204-08, 1990.

REZK, H.A. Mites associated with stored dried-dates in Egypt and the role of *Blattisocius keegani* Fox as a biological control agent. **Alexandria J Agric Res**, v. 45, p. 179-91, 2000.

ROBERTS, E.A.; DIMOCK JR, R.V.; FORWARD JR, R.B. Positive and Host-Induced Negative Phototaxis of the Symbiotic Water Mite *Unionicola formosa*. **Biol Bull**, v. 155, p. 599-607, 1978.

ROEL, A.R. Utilização de plantas com propriedades inseticidas: uma contribuição para o desenvolvimento rural sustentável. **Rev Int Desenv Local**, v. 1, p. 43-50, 2001.

ROFF, D.A. 1991. Life history consequences of bioenergetic and biomechanical constrains on migration. **Amer Zool**, v. 31, n. 1, p. 205-15.

ROUSCH, J.M.; SIMMONS, T.W.; KERANS, B.L.; SMITH, B.P. Relative acute effects of low pH and high iron on the hatching and survival of the water mite, *Arrenurus manubriator* and aquatic insect, *Chironomus riparius*. **Environ Toxicol Chem**, v. 16, n. 10, p. 2144-150, 1997.

RYBANSKA, D.; HUBERT, J.; MARKOVIC, M.; ERBAN, T. Dry dog food integrity and mite strain influence the density-dependent growth of the stored-product mite *Tyrophagus putrescentiae* (Acari: Acaridida). **J Econ Entomol**, v. 109, n. 1, p. 454–60, 2016.

SAINT, G-GD. Effects of dietary lipids on the population growth of *Dermatophagoides pteronyssinus*. In: Griffiths, D.A., Bowman, C.E. (Eds.), **Proceedings of 6th International Congress of Acarology**, Chichester, v. 1, p. 351-57, 1984.

SANCHEZ-BORGES, M.; SUAREZ-CHACON, R.; CAPRILES-HULETT, A.; CABALLERO-FONSECA, F.; IRAOLA, V.; FERNANDEZ-CALDAS, E. Pancake syndrome (oral mite anaphylaxis). **World Allergy Organ J,** v. 2, p. 91-6, 2009.

SANCHEZ-BORGES, M.; CHACÓN, R.S.; CAPRILES-HULETT, A.; CABALLERO-FONSECA, F.; FERNÁNDEZ-CALDAS, E. Anaphylaxis from ingestion of mites: pancake anaphylaxis. **J Allergy Clin Immunol**, v. 131, n. 1, p. 31-35, 2013.

SÁNCHEZ RAMOS, I.; CASTAÑERA, P. Acaricidal activity of natural monoterpenes on *Tyrophagus putrescentiae* (Schrank), a mite of stored food. **J Stored Prod Res**, v. 37, n. 1, p. 93-101, 2001.

SÁNCHEZ-RAMOS, I.; ÁLVAREZ-ALFAGEME, F.; CASTANERA, P. Effects of relative humidity on development, fecundity and survival of three storage mites. **Exp Appl Acarol**, v. 41, n. 1-2, p. 87-100, 2007.

SCHWOERBEL, J. Die Bedeutung der Wassermilbenfür die biozönotische Gliederung. Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte. Limnologie. **Verhandlungen**, v. 14, p. 355–61, 1961.

SINHA, R.N. Role of Acarina in the stored grain ecosystem. **Recent Advances in Acarolog**, v. 1, p. 263-72. 1979.

SMIT, H. *Litarachna brasiliensis* n. sp., the first member of the water mite family Pontarachnidae (Acari: Hydrachnidia) from South America. **Syst Appl Acarol**, v. 12, n. 2, p. 141-46, 2007.

SMITH, I.M.; COOK, D.R.; SMITH, B.P. Water mites (Hydrachnida) and other arachnids. In: Thorp JH, Covich AP (Eds) **Ecology and classification of North American freshwater invertebrates**. San Diego, Academic Press, 3rd ed, p. 485-586, 2009.

SMRŽ, J. Microanatomical and biological aspects of bacterial associations in *Tyrophagus putrescentiae* (Acari: Acaridida). **Exp Appl Acarol**, v. 31, p. 105-13, 2003.

SMRŽ, J.; SVOBODOVÁ, J.; ČATSKÁ, V. Synergetic participation *Tyrophagus putrescentiae* (Schrank) (Acari, Acaridida) and its associated bacteria on the destruction of some soil micromycetes. **J Appl Entomol**, v. 111, p. 206-10, 1991.

SMRŽ, J.; ČATSKÁ, V. Mycophagous mites and their internal associated bacteria cooperate to digest chitin in soil. **Symbiosis**, v. 52, n. 1, p. 33-40, 2010.

SOLARZ, K.; SENCZUK, L.; MANIURKA, H.; CICHECKA, E.; PESZKE, M. Comparisons of the allergenic mite prevalence in dwelling sand certain outdoor environments of the Upper Silesia (southwest Poland). **Int J Hyg Environ Health**, v. 210, p. 715-24, 2007.

SOUSA, J.M.; GONDIM, J.R. M.G.C.; BARROS, R.; OLIVEIRA, J.V. Ácaros em produtos armazenados comercializados em supermercados e feiras livres da cidade do Recife. **Neotrop Entomol**, v. 34, n. 2, p. 303-09, 2005.

SPIEKSMA, F.T.M. Domestic mites from an acarologic perspective. **Allergy**, v. 52, n. 4, p. 360-68, 1997.

STEINKRAUS, D.C.; CROSS, E.A. Description and life history of *Acarophenax mahunkai,* n. sp. (Acari, Prostigmata: Acarophenacidae), an egg parasite of the lesser mealworm (Coleoptera: Tenebrionidae). **Ann Entomol Soc Am**, v. 86, p. 239-249, 1993.

STEJSKAL, V.; HUBERT, J. Risk of occupational allergy to stored grain arthropods and false pest-risk perception in Czech grain stores. **Ann Agric Environ Med**, v. 15, n. 1, p. 29-35, 2008.

STORAAS, T.; STEINSVÅG, S.K.; FLORVAAG, E.; IRGENS, Å.; AASEN, T.B. Occupational rhinitis: diagnostic criteria, relation to lower airway symptoms and IgE sensitization in bakery workers. **Acta Otolaryngol**, v.125, n.11, p. 1211-17, 2005.

SZLENDAK, E.; CONYERS, C.; MUGGLETON, J.; THIND, B.B. Pirimiphos-methyl resistance in two stored product mites, *Acarus siro* and *Acarus farris*, as detected by impregnated paper bioassay and esterase activity assays. **Exp Appl Acarol**, v. 24, p. 45-54, 2000.

TAKAHASHI, K.; TANIGUCHI, M.; FUKUTOMI, Y.; SEKIYA, K.; WATAI, K.; MITSUI, C.; TANIMOTO, H.; OSHIKATA, C.; TSUBURAI, T.; TSURIKISAWA, N.; MINOGUCHI, K.; NAKAJIMA, H.; AKIYAMA, K. Oral mite anaphylaxis caused by mite-contaminated okonomiyaki/ pancake-mix in Japan: 8 case reports and a review of 28 reported cases. **Allergology international: official journal of the Japanese Society of Allergology**, v. 63, n. 1, p. 51-56, 2014.

TAO, N.; ZHAN, X.D.; SUN, E.T.; LI, C.P. Investigation of Acaroid mites breeding in stored dry fruits. **Chinese journal of schistosomiasis control**, v. 27, n. 6, p. 634-37, 2015.

VAN BRONSWIJK, J.E.M.H.; SINHA, R.N. Role of fungi in the survival of *Dermatophagoides* (Acarina: Pyroglyphidae) in house dust environment. **Environ Entomol**, v. 2, n. 1, p. 142-145, 1973.

VAN HAGE-HAMSTEN, M.; JOHANSSON, E. Clinical and immunologic aspects of storage mite allergy. **Allergy**, v. 53, n. 48, p. 49-53, 1998.

VARGA, J.; RIGÓ, K.; MOLNÁR, J.; TÓTH, B.; SZENCZ, S.; TÉREN, J.; KOZAKIEWICZ, Z. Mycotoxin production and evolutionary relationships among species of *Aspergillus* section *Clavati*. **Antonie van Leeuwenhoek,** v. 83, p. 191-200, 2003

VIDRINE, M.F. Nine new species in the subgenus Atacella (Acari: Unionicolidae: *Unionicola*) from Mexico and Brazil. **Int J Acarology**, v. 11, n. 4, p. 255-71, 1985.

VIETS, K.H. Neue Hydrachna- und Eylais-Arten (Hydrachnellae, Acari) aus Porto Alegre, Brasilien. **Zool Anz,** v. 103, n. 7-8, p. 161-171, 1933.

VIETS, K.H. Neue brasilianische Wassermilben. **Zool Anz**, v. 112, n. 11-12, p. 273-83, 1935.

VIETS, K.H. Weitere brasilianische Wassermilben aus dem Staate Pernambuco. **Zool Anz**, v. 113, n. 9-10, p. 209-18, 1936a.

VIETS, K.H. Brasilianische Wassermilben III. **Zool Anz**, v. 115, n. 11-12, p. 273-87, 1936b.

VIETS, K.H. Brasilianische Wassermilben IV. **Zool Anz**, v. 117, p. 168-76, 1937.

VIETS, K.H. Brasilianische Wassermilben V. **Zool Anz**, v. 121, p. 21-24, 1938.

VIETS, K.O. Zwei neue Arten von Wassermilben (Acari, Hydrachnellae) aus Brasilien. **Stud Neotrop Fauna E**, v. 12, p. 253-61, 1977.

WALTER, D.E.; LINDQUIST, E.E.; SMITH, I.M.; COOK, D.R.; KRANTZ, G.W. Order Trombidiformes. In: KRANTZ GW. and WALTER DE. (eds.), **A Manual of Acarology. Third Edition**. Texas Tech University Press, Lubbock. 233-420 p., 2009.

WIĘCEK, M.; MARTIN, P.; GĄBKA, M. Distribution patterns and environmental correlates of water mites (Hydrachnidia, Acari) in peatland habitats. **Exp Appl Acarol**, v. 61, n. 2, p.147-60, 2013.

YOUNG, W.C. Ecological distribution of Hydracarina in North Central Colorado. **Am Midl Nat**, v. 82, p. 367-401, 1969.

YU, S.J.; LIAO, E.C.; TSAI, J.J. House dust mite allergy: environment evaluation and disease prevention. **Asia Pac Allergy**, v. 4, n. 4, p. 241-52, 2014.

ŽDÁRKOVA, E. The effectiveness of organophosphate acaricides on stored product mites interacting in biological control. **Exp Appl Acarol**, v. 18, n. 11-12, p. 747-51, 1994.

ŽDÁRKOVA, E.; HORÁK, E. Preventive biological control of stored food mites in empty stores using *Cheyletus eruditus* (Schrank). **Crop Protection**, v. 9, p. 378-82, 1990.

ŽDÁRKOVA, E.; FEIT, R. Biological control of stored mites on oilseeds using the mites predator *Cheyletus eruditus* (Schrank). **Pl Prot Sci**, v. 35, n. 4, p. 136-38, 1999.