

MÉRCIA SOARES DE OLIVEIRA CARDOSO

**AVALIAÇÃO DA QUALIDADE DO SOLO CULTIVADO COM
CANA-DE-AÇÚCAR E DE FRAGMENTOS DE FLORESTA
ATLÂNTICA POR MEIO DE ÍNDICES ECOLÓGICOS E ATIVIDADE
METABÓLICA DE NEMATOIDES**

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METABÓLICA DE NEMATOIDES**

Tese apresentada ao Programa de Pós-Graduação em Engenharia Agrícola na Área de Concentração em Engenharia de Água e Solo, da Universidade Federal Rural de Pernambuco (UFRPE), como requisito para obtenção do título de Doutora em Engenharia Agrícola.

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RESUMO

Os bioindicadores sinalizam o estado ambiental de determinado ecossistema, podendo ser utilizados para monitorar a qualidade do solo. Por sua abundância, especificidade alimentar, curto ciclo reprodutivo e resposta rápida a mudanças ambientais e manejo do solo, os nematoides são considerados bons indicadores. Apesar disso no Brasil, estudos que relacionem os atributos do solo com as comunidades de nematoides em áreas cultivadas e naturais são escassos. Entretanto, estudos com esta ênfase podem contribuir para a busca de estratégias de manejo do solo que minimizem os prejuízos causados pelos nematoides de importância econômica na região canavieira de Pernambuco. No período de 2009 a 2012, foram estudadas 12 áreas cultivadas com cana-de-açúcar e 13 áreas remanescentes de Floresta Atlântica (variando entre 1,5 e 298,78 ha). As amostragens foram realizadas em áreas, regiões e tipos de solo distintos, e mesmo quando se comparou áreas de uma mesma usina, cada área foi caracterizada por uma estrutura de comunidade de nematoides distinta. Os resultados obtidos neste estudo ressaltam a capacidade que as comunidades de nematoides têm de responder a alterações no ambiente. As práticas de manejo da cana-de-açúcar resultaram em alterações na densidade e porosidade do solo, na atividade biológica e nas comunidades de nematoides quando comparada a áreas de vegetação nativa. O uso da análise de pegada metabólica dos nematoides para medir a magnitude de seus serviços na cadeia trófica do solo, bem como, a avaliação da conectância funcional para observar as relações entre predadores e presas foram o diferencial neste estudo, fornecendo subsídios para a utilização dos nematoides como bioindicadores da condição da cadeia alimentar do solo. A geoestatística não possibilitou a detecção da distribuição espacial dos taxa dos nematoides, e os atributos do solo apresentaram baixos valores de alcance. Alternativamente, o padrão espacial dos nematoides foi descrito pelo *Taylor's power law model*, com valores de b variando de 1,80 a 2,41, confirmando o padrão agregado dos nematoides nas áreas de estudo.

Palavras-chave: qualidade do solo, monocultivo, vegetação nativa, degradação física do solo.

ABSTRACT

Bioindicators analyze environmental state in an ecosystem and can be used to monitor the soil quality. Due to abundance, feed specificity, short reproductive cycle and fast response to environmental and soil manage changes, nematodes presents high potential as excellent indicator. However, in the Brazil there are few studies that relate nematode to soil properties in cultivated and native soils. Understanding this relationship might, therefore, contribute to prevent losses caused by plant-parasitic nematodes in the sugarcane zone of Pernambuco State, Brazil. In the period from 2009 to 2012 were studied 12 sugarcane fields and 13 Atlantic Forest remnants (ranging in size from 1.5 to 298.78 ha), all of which enterely surrounded by a uniform matrix of sugarcane crop. Soil samplings were carried out in different areas, region and soil type. However, the structure and composition of the nematode assemblage and soil properties differed between forest and sugarcane soils, even in the same locality. The results of this study highlights the ability of nematodes to respond to changes in the environment. For the management practices applied in the sugarcane soils result in changes in bulk density, total porosity, biological activity and nematodes assemblages structure and composition when compared to native vegetation. This thesis also provided insights to enhance the utility of nematodes as bioindicators of soil food webs. The use of metabolic footprints of nematoides in order to measure the magnitude of the services of the nematodes in the soil food web and the assessment of functional connectance to observe relationships between predators and prey nematodes were relevant factors in this study. Geostatistical analysis did not allow assessment of the spatial distribution of nematodes and the soil attributes had a low range of spatial parameters. We conclude that the scale of sampling was too great for detection of spatial distribution patterns of the nematodes; alternatively, their aggregated spatial patterns were described by a Taylor's power law model, with b-values ranged from 1.80 to 2.41, confirming the aggregated pattern of nematode distribution in the study sites.

Keywords: soil quality, monoculture, native vegetation, soil physical degradation.

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LISTA DE PUBLICAÇÕES

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I Cardoso, M.S.O., Pedrosa, E.M.R., Rolim, M.M., Costa, C.S., Leitão, D.A.H.S., Oliveira, A.K. Relationships between nematode assemblages and physical properties across land use types. (Submetido a *Environmental, Monitoring and Assessment*).

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IV Cardoso, M.S.O., Pedrosa, E.M.R., Ferris, H., Rolim, M.M., Oliveira, L.S.C., David, M.F.L. Spatiotemporal distribution of nematodes and soil food web interactions in Atlantic Forest. (Submetido a *Forest Ecology and Management*).

INTRODUÇÃO GERAL

Estudos que relacionem os atributos do solo com as comunidades de nematoides são escassos no Brasil. Recentemente, vêm sendo desenvolvidos em áreas canavieiras em Pernambuco, bem como, relacionando as comunidades de nematoides com a compactação do solo (CARDOSO et al., 2012), com as propriedades químicas (MATOS et al., 2011), com a umidade e a densidade de partículas do solo (RODRIGUES et al., 2011). Estudos com esta ênfase contribuem para o entendimento das relações existentes entre os nematoides e os atributos do solo e para buscar estratégias de manejo do solo que minimizem os prejuízos causados pelos nematoides de importância econômica na região canavieira de Pernambuco.

Além disso, pesquisas realizadas comparando ecossistemas naturais e agrícolas enfatizam apenas populações de nematoides parasitas de plantas (SILVA et al., 2008a, 2008b; TOMAZINI et al., 2008) ou apenas uma família (Longidoridae) de parasitas de plantas (JESUS, 2011). Entretanto, as populações de nematoides de vida livre são tão importantes quanto os nematoides parasitas de plantas. Recentemente, ARIEIRA et al., (2013) avaliaram a ocorrência de nematoides de vida livre e parasitas de plantas em áreas cultivadas com cana-de-açúcar em comparação à remanescente de Floresta Atlântica situadas no Estado do Paraná. Contudo, observaram que a textura do solo é fator de interferência na estrutura das comunidades de nematoides.

Os resultados obtidos nas pesquisas aqui apresentadas ressaltam a capacidade que as comunidades de nematoides têm de responder a alterações no ambiente. Ao todo, foram estudadas 12 áreas cultivadas com cana-de-açúcar e 13 áreas remanescentes de Floresta Atlântica (com tamanho variando de 1,5 a 298,78 ha). As amostragens foram realizadas em áreas, regiões e tipos de solo distintos, e mesmo quando se comparou áreas em mesma usina, cada área foi caracterizada por uma estrutura de comunidade de nematoides distinta. As práticas de manejo nas áreas cultivadas com cana-de-açúcar resultaram em alterações na densidade e porosidade do solo, na atividade biológica e nas comunidades de nematoides quando comparada à áreas de vegetação nativa (BELL; STIRLING; PANKHUST 2007; STIRLING et al. 2010; BAQUERO et al. 2012).

O uso dos *metabolic footprints* dos nematoides (FERRIS, 2010) para medir a magnitude dos serviços destes organismos na cadeia trófica do solo e a avaliação da conectância funcional (SÁNCHEZ-MORENO et al. 2011; FERRIS et al. 2012) para observar as relações entre predadores e presas foram o diferencial neste estudo. A

geoestatística não possibilitou a detecção da distribuição espacial dos *taxa* dos nematoides, e os atributos do solo apresentaram baixos valores de alcance. Assim, a variabilidade espacial dos nematoides tem sido baseado no efeito pepita (DINARDO-MIRANDA; FRACASSO, 2010; CARDOSO et al., 2012). Alternativamente, o padrão espacial agregado dos nematoides pode ser descrito pelo Taylor's power law model (TAYLOR, 1961; FERRIS, 1985; ALLSOPP, 1990; GHADERI et al., 2012; PARK et al., 2013). Ao aplicar este modelo, o padrão de distribuição dos nematoides foi constatado como agregado nas áreas de estudo.

Diante do exposto, as hipóteses deste estudo foram: (a) a abundância e a diversidade dos nematoides parasitas de plantas diferem entre solos de floresta e cultivados com cana-de-açúcar; (b) as propriedades físicas do solo são afetadas pelas práticas de manejo; (c) as alterações nas propriedades físicas do solo influenciam a abundância e a diversidade dos nematoides parasitas de plantas e predadores; (d) os solos cultivados com cana-de-açúcar possui cadeia alimentar enriquecida, mas desestruturada e são conduktivos à decomposição bacteriana. Enquanto os solos de floresta possuem cadeia alimentar estruturada e são conduktivos à decomposição fúngica; (e) a distribuição espaço-temporal dos nematoides é influenciada pelos atributos do solo; (f) a baixa abundância das presas está relacionada a alta abundância dos predadores. Assim, esta pesquisa teve os seguintes objetivos: (1) descrever a diversidade de nematoides em áreas cultivadas com cana-de-açúcar e áreas de floresta; (2) analisar as relações entre os atributos do solo (densidade, porosidade e respiração do solo) e as comunidades de nematoides; (3) avaliar a condição da cadeia alimentar do solo por meio da *faunal analysis* e *metabolic footprints* dos nematoides; (4) observar a distribuição espaço-temporal dos nematoides e relacionar com os atributos do solo; (5) avaliar as interações predador-presa.

Esta tese avaliou a estrutura e composição das comunidades de nematoides em diferentes níveis de distúrbio: áreas cultivadas com cana-de-açúcar sob diferentes manejos e em áreas remanescentes de Floresta Atlântica, observando as relações entre a nematofauna e as propriedades do solo (umidade do solo, porosidade total, densidade do solo, resistência mecânica do solo e textura) (Capítulo 2). No Capítulo 3, foram estudadas as comunidades de nematoides associadas a dez áreas cultivadas com cana-de-açúcar e oito fragmentos de Floresta Atlântica, observando a diversidade dos nematoides e avaliando as relações destes com a densidade, porosidade e respiração do solo. Além disso, a estrutura e a condição das cadeias tróficas de solos cultivados com

cana-de-açúcar foram avaliadas por meio de índices ecológicos e *metabolic footprints* dos nematoides (Capítulo 4). No Capítulo 5, a estrutura e a composição da nematofauna em áreas de floresta conservada foram descritas, analisando a sua distribuição espaço-temporal, a conectância funcional (interação entre predadores e presas) e a estrutura e condição da cadeia trófica do solo. Por fim, o Capítulo 6 sintetiza os resultados deste estudo e delineia as perspectivas.

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Revisão de Literatura

CAPÍTULO I

Cana-de-Açúcar

A cana-de-açúcar é uma gramínea semi perene, geralmente cultivada em áreas tropicais ou subtropicais, sendo originária do Sudeste Asiático. Pertence à classe Liliopsida, sub-classe Commelinidae, ordem Cyperales, família Poaceae, tribo Andropogoneae, sub-tribo Saccharininaeae e gênero *Saccharum* (CASTRO; KLUGE, 2001).

O Brasil é considerado atualmente o maior produtor mundial de cana-de-açúcar. Em Pernambuco, para a safra 2014/2015, serão cultivados 277,74 mil hectares, nos quais se estima obter uma produção de 14.634,10 mil toneladas de cana-de-açúcar para todos os usos. E a produtividade de $52.690 \text{ kg ha}^{-1}$, a qual se estima que seja utilizada como matéria-prima na produção de, aproximadamente, 1.157,93 mil toneladas de açúcar e mais de 300 mil litros de álcool (CONAB, 2014).

A cana-de-açúcar vem sendo cultivada no Brasil desde a época da colonização. Atualmente, assume grande importância econômica, social e ambiental, pelas grandes áreas plantadas, por gerar matéria prima como base para as agroindústrias do açúcar e álcool, além de representar para o nosso país uma fonte de geração de empregos e renda no meio rural. No Nordeste brasileiro, assim como em outras regiões produtoras de cana-de-açúcar, significativos impactos positivos no setor socioeconômico estão sendo alcançados com o desenvolvimento do setor energético proveniente do etanol (MARTÍNEZ et al., 2013).

A partir do final da década de 1950 e início da de 1960, para melhorar a produtividade, todas as fases da produção da cana-de-açúcar se modernizaram com a mecanização, uso de defensivos agrícolas e introdução de novas variedades. Este processo foi bastante heterogêneo entre as regiões, sendo que ainda existem usinas que cultivam a cana com tecnologia baseada em mão-de-obra e outras com mecanização intensiva (VIAN et al., 2007).

A implantação de políticas públicas, que também se refletiu no surgimento de inovações institucionais e técnicas/científicas, na década de 1970 foi o fator que mais impulsionou a produção e a produtividade da cana-de-açúcar no Brasil. O PROÁLCOOL foi o marco da transformação do sistema agroindustrial da cultura. A desregulamentação do setor sucroalcooleiro, na década de 1990, voltou a impulsionar uma nova fase de aumento de produtividade da cana-de-açúcar, desta vez firmado em um modelo de forte tendência privada e baseado na maior eficiência e competitividade do setor, com ênfase na sustentabilidade (SANTIAGO et al., 2006).

Com isso, as práticas de manejo empregadas no sistema de produção da cana-de-açúcar se iniciam no plantio e vão até o final do corte. Desse modo, inúmeras operações de preparo do solo, colheita e transporte da matéria prima são conduzidas diversas vezes em uma mesma área. Esse intenso tráfego de máquinas agrícolas provoca modificações na estrutura física do solo (LANZANOVA et al., 2007). Sendo assim, um preparo inadequado do solo pode reduzir a produção, refletindo em todo o ciclo da cana-de-açúcar, já que, a produção das soqueiras está atrelada à produção do corte anterior (CAMARGO; ALLEONI 1997).

Estudos relatam que o manejo empregado em áreas cultivadas com cana-de-açúcar alteram a densidade e porosidade do solo, a atividade biológica e as comunidades de nematoides quando comparada à áreas de florestas (BELL et al., 2007; STIRLING et al., 2010; BAQUERO et al., 2012). Além disso, a conversão de solos de floresta em áreas agrícolas impactam negativamente os atributos de qualidade do solo, reduzindo a biodiversidade e compactando o solo (ROUSSEAU et al., 2013; GAJI , 2013).

De fato, um dos efeitos mais comuns do manejo em áreas cultivadas é a compactação do solo, a qual é caracterizada pelo aumento da densidade do solo e da resistência à penetração ao crescimento das raízes e pela redução dos espaços porosos, bem como refletindo negativamente na fauna do solo (CHEESMAN, 2004; BATEY; MCKENZIE, 2006; BELL et al., 2007) e alterando a distribuição dos nematoides nos níveis tróficos, aumentando a abundância dos parasitas de plantas e reduzindo a abundância dos bacteriófagos, onívoros e predadores (BOUWMAN; ARTS, 2000; CARDOSO et al., 2012). Além de muitas vezes favorecer a dominância de um único gênero (PATTISON et al., 2008) ou aumentando a abundância de gêneros parasitas de plantas, com comunidades dominadas, principalmente, por *Pratylenchus* e *Helicotylenchus* (ARIEIRA et al., 2013).

Floresta Atlântica

De acordo com o ponto de vista da legislação brasileira (Lei Federal nº 11.428/2006), a Floresta Atlântica é composta por formações bem distintas, incluindo a Floresta Ombrófila Densa, Floresta Ombrófila Mista (Mata de Araucárias), Floresta Ombrófila Aberta, Floresta Estacional Semidecidual e Floresta Estacional Decidual, bem como os manguezais, restingas, campos de altitude, brejos interioranos e encraves florestais do Nordeste.

É considerada uma das florestas tropicais mais ameaçadas do mundo, restam apenas 11,73% de toda a extensão territorial brasileira (RIBEIRO et al., 2009). E o que restou encontra-se fragmentados, em estádio de sucessão secundária, bem como alterados e

empobrecidos quanto a sua composição (SOUZA et al., 2002). Sendo considerada por MYERS et al., (2000) o quarto *hotspot* mais importante em termos de endemismo de plantas e invertebrados. Porém, apesar de reduzida a poucos fragmentos, na sua maioria descontínuos, a sua biodiversidade é uma das maiores do planeta.

No Nordeste brasileiro, a perda de áreas florestadas está intrinsecamente relacionada com as formas de uso da terra e com o modo de produção estabelecido nas áreas convertidas. Pernambuco é o estado que detém os menores fragmentos de Floresta Atlântica; ao todo são 76 unidades de conservação, dentre as quais 39 são reservas de proteção integral e 37 são de uso sustentável (CPRH, 2014). Esses fragmentos relativamente conservados encontram-se em propriedades particulares, sobretudo em áreas pertencentes às usinas de cana-de-açúcar (TABARELLI; RODAL, 2005).

Considerando-se os estádios avançados de degradação da Floresta Atlântica, as informações sobre a biodiversidade permanecem incipientes e limitadas, principalmente quando se trata da nematofauna. Estudos sobre a diversidade de nematoides, tanto em áreas cultivadas como naquelas cobertas por vegetação nativa se justificam, em termos econômicos, pelos seus possíveis efeitos sobre a vegetação, uma vez que são organismos originários das áreas naturais que se adaptaram as áreas cultivadas.

Os nematoides como bioindicadores da qualidade do solo

A qualidade do solo é definida como «a capacidade do solo funcionar» e refere-se às propriedades físicas, químicas e biológicas, as quais consideravelmente influenciam o crescimento vegetal, a disponibilidade de nutrientes para as plantas e reduz o efeito do estresse ambiental na planta (ELLIOTT et al., 1996; KARLEN et al., 1997). A avaliação da qualidade do solo é feita por meio de diferentes atributos do solo, incluindo atributos físicos, químicos e biológicos. Entretanto, estes atributos necessitam ser considerados bons indicadores e serem capazes de diferenciar o tipo de manejo, ter sensibilidade a condições adversas associadas a ações antropogênicas, ser de fácil mensuração, ter curto tempo de resposta e acessibilidade (ANDREWS et al., 2004; BATISDA et al., 2008; LIMA et al., 2013).

Em busca de melhores produtividades, os solos agrícolas são, comumente submetidos às práticas convencionais de cultivo, tais como: aplicação de pesticidas, revolvimento do solo, irrigação, dentre outras. Contudo, estas práticas de manejo, geralmente ocasionam compactação, salinização, acidificação a longo prazo, levando a alterações na estrutura

biológica, física e/ou química do solo (CHEESMAN, 2004; BRIAR et al., 2007). A perda da qualidade do solo está associada a diversos processos desempenhados pelos microrganismos do solo, por exemplo. Desse modo, por meio da avaliação da densidade, atividade, estrutura e composição das comunidades dos microrganismos é possível determinar se um sistema de manejo mantém, melhora ou degrada o solo (BATISDA et al., 2008; VALLEJO-QUINTERO, 2013).

As propriedades físicas do solo referem-se à capacidade estrutural, a qual é considerada um fator importante para a qualidade, pois um solo bem estruturado é sinônimo de boa produtividade. Dessa forma, os poros são indiscutivelmente a característica mais importante da física do solo, devido ao fato da maioria dos processos edáficos terem consequência imediata para a atividade microbiana ou conservação (CRAWFORD et al., 2012; HELLIWELL et al., 2014). Tais processos ocorrem dentro dos poros ou nas superfícies das partículas que formam as suas paredes. Contudo, não é apenas o tamanho ou a quantidade de poros que são importantes, mas, também, a sua continuidade; obviamente poros isolados desempenharão papéis menos centrais nos processos edáficos (CÁRDENAS et al., 2012; NAVCEED et al., 2014).

As práticas de manejo empregadas em sistemas de produção da cana-de-açúcar, por exemplo, afetam negativamente a qualidade do solo, incluindo alterações em diversos componentes físicos, afetando, principalmente, a estrutura e, consequentemente a biota (CHEESMAN, 2004; BELL et al., 2007). A compactação é comum em áreas canavieiras, aumentando a densidade do solo e a resistência à penetração ao crescimento das raízes e reduzindo a porosidade (CHEESMAN, 2004; BATEY; MCKENZIE, 2006), incluindo efeitos negativos na estrutura da comunidade da macrofauna (RADFORD et al., 2001), de nematoides (BOUWMAN; ARTS, 2000) e nas atividades microbianas (JORDAN et al., 2003).

As alterações na estrutura das comunidades de nematoides estão associadas às características intrínsecas do ambiente, bem como às alterações nas condições do solo. Assim, os fatores edáficos que influenciam o total de nematoides e a abundância dos grupos tróficos incluem a conservação de carbono e as mudanças na estrutura física ocasionadas pelo manejo, tais como: a densidade e o teor de água no solo (GUPTA; YEATES, 1997; PATTISON et al., 2008; NIELSEN et al., 2014).

Dentro das limitações do tipo de solo, umidade e temperatura, a natureza do recurso alimentar também influenciam a dominância de espécies e dominância dentro de cada grupo trófico de nematoides (YEATES, 2003). Além disso, o declínio dos recursos alimentares e

espaço poroso com a profundidade do solo geralmente é relacionado à redução da densidade e diversidade de nematoides em maiores profundidades (SOHLENIUS; SANDOR, 1987, YEATES et al., 2008).

Em áreas cultivadas com cana-de-açúcar a longo prazo no Paraná, a estrutura das comunidades de nematoides é afetada, favorecendo o aumento da abundância de parasitas de plantas, com maior dominância de *Pratylenchus* e *Helicotylenchus* (ARIEIRA et al., 2013). Por conseguinte, a compactação do solo altera a distribuição dos nematoides nos níveis tróficos, aumentando a abundância dos parasitas de plantas e reduzindo a abundância dos bacteriófagos, onívoros e predadores (BOUWMAN; ARTS, 2000; CARDOSO et al., 2012). E muitas vezes, leva a dominância de um único gênero (PATTISON et al., 2008).

Assim, torna-se um desafio a sustentabilidade e o aumento da produtividade em áreas com solos degradados. O aumento ou, ao menos, a manutenção da qualidade do solo aliadas aos fatores de produção agrícola é essencial para alcançar este desafio (CARTER, 2002). Diversos estudos tem investigado a melhor forma de avaliar a qualidade (KARLEN et al., 2008). Comumente, esse processo inicia-se com a escolha dos indicadores.

A demanda por indicadores de qualidade ambiental é resultante do aumento da pressão em áreas disponíveis em face da expansão da urbanização, perda de áreas com vegetação nativa e de área degradada. Atualmente, tem-se observado um crescente interesse na avaliação e interpretação da qualidade do solo (LI et al., 2013; TESFAHUNEGN, 2013; EPELDE et al., 2014). Para isso, indicadores biológicos, químicos e físicos têm sido estudados e utilizados para monitorá-la (MARINARI et al., 2006; MONCADA et al., 2013). Contudo, a escolha de parâmetros que sejam realmente sensíveis para detectar alterações em solos submetidos a diferentes sistemas de manejo é uma tarefa difícil (LIMA et al., 2013).

Os micro-organismos do solo residem dentro da estrutura do poro e entre as partículas do solo. Por isso, a comunidade destes organismos pode ser usada como um eficiente indicador da qualidade do solo e estresses ecológicos devido a sua adaptação. E com isso, eles vêm sendo estudados como potenciais bioindicadores para monitorar a qualidade do solo (BONGERS; BONGERS, 1998; HOFMAN et al., 2003; CORDEIRO et al., 2004; NEHER et al., 2005; FIGUEIRA et al., 2011; CARDOSO et al., 2012; ZHANG et al., 2012; MELLONI et al., 2013; VALLEJO-QUINTERO, 2013).

Na década de 1970, os nematoides começaram a ser utilizados para avaliar a qualidade da água. Durante a década de 1980, as preocupações com a vulnerabilidade dos solos aumentaram, com isso, a nematofauna passou a ser estudada para avaliação de ecossistemas. Isso porque os nematoides possuem uma ampla diversidade trófica, a qual reflete a estrutura

da cadeia trófica do solo em resposta a alterações na disponibilidade de recursos (YEATES et al., 1993; FERRIS et al., 2001). Comumente, os cinco principais grupos reconhecidos são: parasitas de plantas, bacteriófagos, micófagos, predadores e onívoros (YEATES et al., 1993).

Os nematoides também possuem papel-chave na decomposição e na ciclagem de nutrientes do solo (FERRIS et al., 2004) e são excelentes bioindicadores: por sua abundância, adaptação a quase todos os tipos de ambiente, especificidade alimentar (YEATES et al., 1993), curto ciclo reprodutivo e resposta rápida a mudanças ambientais, demonstrando sensibilidade às ações de manejo do solo (MATTOS, 2002; BRIAR et al., 2007; ZHANG et al., 2012) e à presença de contaminantes no solo (RUDEL et al., 2013; ZHAO; NEHER, 2013).

Índices ecológicos baseados na nematofauna do solo

Diversos índices desenvolvidos e implementados para a avaliação da qualidade ambiental e estrutura da cadeia trófica do solo indicam o potencial para as funções e serviços dos ecossistemas baseados na abundância relativa das guildas estruturais e funcionais dos nematoides (BONGERS, 1990; BONGERS; FERRIS, 1999; FERRIS; BONGERS, 2009). Entretanto, os referidos índices não indicam a provável magnitude dos serviços, a qual é fornecida pela análise da pegada metabólica dos nematoides (FERRIS et al., 2010).

Em estudos ecológicos, índices de diversidade, tais como, riqueza e equabilidade de espécies (Índice de Simpson e Índice de Shannon-Weaver) são amplamente utilizados para avaliar alterações ecológicas, efeitos de distúrbios, poluição no ecossistema e de alterações climáticas (LEVI et al., 2012). Esses índices são, também, usados em estudos de ecologia de nematoides. Contudo, o uso dos nematoides é potencialmente mais informativo que outros grupos, porque o filo Nematoda é um grupo amplo e comprehende diferentes estratégias de vida que responde de diferentes formas a alterações no ambiente.

A diversidade de nematoides vem sendo utilizada para modelar o tipo de vegetação e a qualidade do solo (EISENHAUER et al., 2011; WILLERSLEV et al., 2014). Em um experimento de longo prazo, EISENHAUER et al., (2011) observaram que a densidade de nematoides predadores aumenta significativamente com a diversidade de plantas, apontando para o aumento da complexidade da cadeia alimentar em comunidades vegetais ricas em espécies. Em comunidades vegetais complexas a comunidade de nematoides foi dominada por micófagos, bacteriófagos e predadores, em contraposição a abundância dos nematoides parasitas de plantas.

Considerando as especificidades dos nematoídeos, Bongers (1990) desenvolveu o Índice de Maturidade (MI), o qual se baseia na abundância relativa de taxa de nematoídeos não parasitas de plantas como medida de distúrbios ambientais. Em sua essência, o MI indica o estado de sucessão ecológica de uma comunidade com baixos valores indicando distúrbios (BONGERS, 1990; BONGERS; KORTHALS, 1993; BONGERS et al., 1997). Neste índice, cada família de nematoide é alocada em uma escala cp (1-5) baseada em espécies estritamente relacionadas mostrando similaridade em relação aos traços de vida devido ao parentesco na linhagem filogenética, morfologia, anatomia, fisiologia e genética (Tabela 1, BONGERS, 1990).

Tabela 1. Descrição dos valores cp e as características do ciclo de vida dos nematoídeos correspondentes (adaptado de BONGERS, 1990).

cp	Descrição	Exemplos
1	Curto tempo de geração, ovos pequenos, alta fecundidade, se alimentam continuamente em meios enriquecidos, principalmente bacteriófagos.	<i>Bunonema, Diploscapter, Monhystera, Rhabditis...</i>
2	Tempo de geração maior e menor fecundidade que o grupo cp1, muito tolerante a condições adversas e podem tornar-se criptobióticos.	<i>Wilsonema, Acrobelus, Aphelenchus, Seinura...</i>
3	Continuam se alimentando quando o recurso está em declínio, principalmente bacteriófagos e micófagos.	<i>Prismatolaimus, Criconemoides, Hemicycliophora, Pratylenchus, Rotylenchus...</i>
4	Tempo de geração maior, menor fecundidade, maior sensibilidade a distúrbios que cp3. Além de outras funções tróficas, espécies de pequenos onívoros.	<i>Alaimus, Cryptonchus, Dorylillum, Trichodorus, Dorylaimus, Thornia, Mononchus...</i>
5	Maior tempo de geração que os demais grupos, corpos maiores, menor fecundidade, maior sensibilidade a distúrbios, principalmente predadores e onívoros.	<i>Isolaimium, Xiphinema, Mesodorylaimus, Prodorylaimus, Laimydorus...</i>

* Os números 1-5 são correspondentes aos escores cp.

O MI foi inicialmente desenvolvido utilizando dados de nematoídeos de estudos em rio poluído (ZULLINI, 1976). Existem diversos estudos utilizando o MI dos nematoídeos como um indicador ecológico (KIMENJU et al., 2009; BIEDERMAN; BOUTTON, 2010; ZHANG et al., 2013; LIU et al., 2013; WU et al., 2014). O desenvolvimento deste índice foi um significante avanço na interpretação das relações entre a ecologia de comunidades de nematoídeos e funções do solo, o que faz as comunidades de nematoídeos amplamente utilizadas como indicadores em biomonitoramento (NEHER, 2001).

A classificação colonizador-persistente (escala cp) permite o cálculo do MI por meio da frequência média ponderada das classes cp (BONGERS, 1990). A progressão das condições do solo de estressado ou poluído a intacto não é exatamente congruente com as

características do ciclo de vida dos nematoides na classificação cp como definido por Bongers (1990). Preferencialmente, os mais abundantes *taxa* de nematoides sob condições estressadas são aqueles em cp2, enquanto os oportunistas de enriquecimento (cp1) respondem positivamente a distúrbios que resultam no enriquecimento em qualquer nível de qualidade ambiental (BONGERS, 1999; BONGERS; FERRIS, 1999).

Além do MI, existem outros índices: Índice de Maturidade Modificado₂₋₅ (MI₂₋₅) neste índice são excluídos os nematoides com valor cp1; Índice de Parasitas de Plantas (PPI) o qual lista apenas os parasitas de plantas, baseado numa escala cp de 2 a 5. O MI, o PPI e o MI₂₋₅ são calculados pela mesma fórmula, cujo o valor cp varia de 1 (colonizador) a 5 (persistentes) do táxon "i".

$$MI = \frac{v(i) \cdot f(i)}{n_i}$$

$$n_i = 1$$

Em que: v(i) é o valor cp do táxon; e

f(i) é a frequência daquele táxon na amostra.

Após distúrbios ambientais, a diversidade e abundância de nematoides decrescem (YEATES, 2007; CARRASCOSA et al., 2014) originando uma comunidade tipicamente dominada por pequenos nematoides, *taxa* oportunistas cp2 e passa a ser considerada 'basal' no MI. Se após o distúrbio o solo for adubado, nematoides menores, *taxa* cp1, vão aumentar em abundância relativa e passarão a dominar a comunidade, e assim, o solo será considerado 'enriquecido'. Como a comunidade está propícia a se recuperar ao longo do tempo e os recursos tornam-se disponíveis, a complexidade dos recursos aumenta, e com ou sem adubação, o solo se tornará 'estruturado' e nematoides cp3 a cp5 começam a colonizar o solo e diversificar a comunidade por meio do incremento de mais ligações tróficas (FERRIS et al., 2001).

Um aprimoramento do MI foi feito por meio da criação de um quadro conceitual por Ferris et al., (2001), no qual foram combinados a escala cp e os hábitos alimentares (Figura 1). Dentro desse quadro, as guildas funcionais dos nematoides dão origem a um Índice de Enriquesimento (EI) e a um Índice de Estrutura (SI), baseados nas trajetórias de enriquecimento e estrutura na cadeia trófica. (FERRIS et al., 2001; 2004) redefiniram os conceitos desses índices para fornecer uma maior resolução para os efeitos do enriquecimento, distúrbio e contaminação nos ecossistemas do solo.

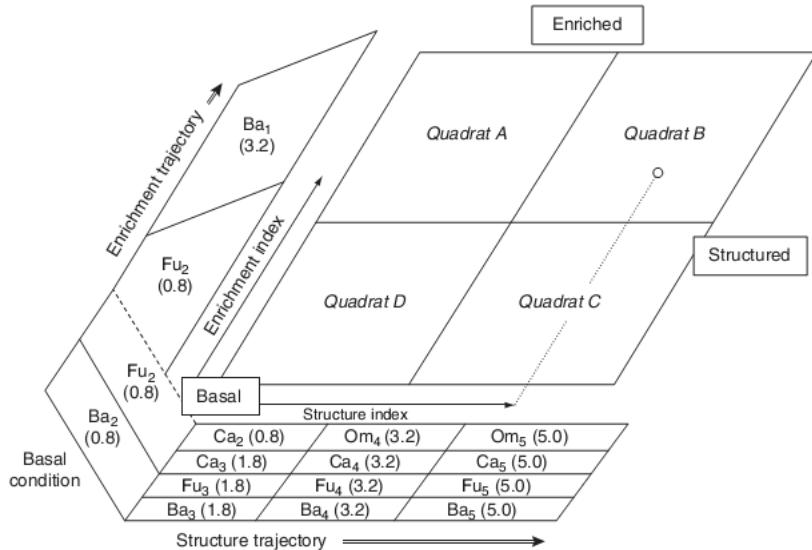


Figura 1. Representação gráfica do *faunal analysis* dos nematoides indicando se a comunidade do solo está enriquecida, mas desestruturada (Quadrante A), enriquecida e estruturada (Quadrante B), limitada de recursos e estruturada (Quadrante C) ou com recursos esgotados e com estrutura mínima (Quadrante D) (Adaptado de FERRIS et al., 2001).

Os índices da cadeia trófica do solo baseados na abundância das guildas funcionais dos nematoides (escala cp integrada com as fontes de alimentação) (FERRIS et al., 2001) tem sido utilizados para examinar o efeito da poluição, manejo e vegetação em agroecossistemas (LIANG et al., 2005; STIRLING; LOGDE, 2005; WANG et al., 2006; CULMAN et al., 2010; BRIAR et al., 2012), pois fornecem informações qualitativas da cadeia trófica do solo.

Na *faunal analysis*, os nematoides são analisados como indicadores da condição da cadeia trófica do solo, classificada como basal, enriquecida, estruturada ou estável. Os índices indicadores da condição do ecossistema são calculados de acordo com Ferris et al., (2001): Índice de Enriquecimento (EI), Índice de Estrutura (SI), Índice Basal (BI) e Índice de Canal (CI). Desse modo, quando o EI é plotado contra o SI, o resultado é um gráfico dividido em quatro quadrantes, os quais são descritivos das características da cadeia trófica do solo: perturbada, maturando, estruturada ou degradada (Figura 1; FERRIS et al., 2001; FERRIS e MATUTE, 2003).

O Índice de Estrutura (SI) baseia-se na abundância relativa ponderada de guildas sensíveis; O Índice de Enriquecimento (EI) é alto quando a atividade microbiana aumenta seguido de distúrbio e os oportunistas de enriquecimento, bacteriófagos e micófagos,

predominam dentro da comunidade. Na base ambos os índices estão os *taxa* tolerantes à condições adversas e basal para todas as comunidades de nematoides (FERRIS et al., 2001).

Altos valores de Índices de Maturidade e Estrutura (MI e SI) indicam estabilidade, caracterizada por uma comunidade de nematoides dominada por nematoides maiores com ciclos de vida mais longo. Liu et al., (2013), confirmam a eficiência destes índices para avaliação do manejo de conservação na qualidade do solo.

O Índice de Estrutura (SI) indica o estado da cadeia trófica afetada por estresse ou distúrbio (FERRIS et al., 2001). O SI é um indicador de comprimento e conectância da cadeia trófica do solo. A sensibilidade do SI é determinada, principalmente, pelas populações de nematoides onívoros e predadores, os quais necessitam de muito mais tempo para se estabelecer que os nematoides micófagos e bacteriófagos que tem crescimento rápido (LIANG et al., 2009). Além disso, os nematoides onívoros e predadores são suscetíveis a distúrbios e podem se reestabelecer quando a intensidade do manejo é reduzida (BRIAR et al., 2007).

Em cultivos anuais os valores de SI são maiores na zona de maior influência das raízes, reduzindo em profundidade. Essa redução coincide com as reduções na biomassa das raízes (BUYANOVSKY et al., 1987; JACKSON et al., 1996), refletindo o potencial das raízes em manter estruturada a cadeia trófica do solo.

O Índice Basal (BI), baseado na abundância de nematoides oportunistas, é um indicador de basal, condição perturbada da cadeia trófica do solo (FERRIS et al., 2001), com elevados valores indicando uma comunidade composta por nematoides resistentes à perturbações, principalmente, de baixos níveis tróficos. Okada e Harada (2007) verificaram que menores entradas de nutrientes e menor ciclagem da matéria orgânica decresce a abundância de nematoides bacteriófagos.

O Índice de Canal (CI) indica se a via de decomposição predominante é fúngica (CI alto) ou bacteriana (CI baixo) (FERRIS et al., 2001). Além disso, fornece uma medida quantitativa do fluxo de carbono e energia através dos canais de decomposição fúngicos e bacterianos.

Enquanto o EI avalia a resposta da cadeia trófica a disponibilidade de recursos, o CI indica a predominância das vias de decomposição. Desse modo, estes índices, em conjunto, fornecem base para a avaliação da fertilidade do solo, disponibilidade de nutrientes, potencial de lixiviação de nutrientes, bem como, os ajustes necessários de carbono ou nitrogênio para alterar estas condições. DuPont et al. (2009), observaram que em solos cobertos com cobertura morta o EI é alto e o CI e BI apresentam baixos valores, sugerindo dominância de

bactérias no sistema com recursos abundantes e rápida ciclagem de nutrientes. Estas qualidades, geralmente, associadas a elevada produtividade agrícola.

A *faunal analysis* juntamente com a análise dos *metabolic footprints* informam a condição do solo analisado. A avaliação dos *metabolic footprints* dos nematoides (NMF) é baseada na biomassa, atividade metabólica e magnitude dos serviços dos componentes da comunidade de nematoides. O cálculo é feito através dos parâmetros ecofisiológicos dos nematoides em níveis funcionais e taxonômicos (FERRIS, 2010). O NMF tem um componente de produção e outro de respiração. No componente de produção, a quantidade de carbono é particionada no crescimento e produção de ovos. Enquanto no componente de respiração está o carbono utilizado na atividade metabólica (FERRIS et al., 2012). Os atributos ecofisiológicos dos nematoides, agrupados a níveis de espécies, gêneros e famílias estão disponíveis no NEMAPLEX (<http://plpnemweb.ucdavis.edu/nemaplex>).

O *footprint* de enriquecimento (EF) é o NMF dos nematoides oportunistas de enriquecimento (BONGERS; FERRIS, 1999; FERRIS, 2010). O *footprint* de estrutura (SF) é o NMF dos nematoides os quais podem ter uma função regulatória na cadeia alimentar e os quais, também, são indicativos da abundância dos *taxa* de não-nematoides com similares funções. É um indicador dos maiores níveis tróficos de predação, refletindo a atividade metabólica dos nematoides de altos níveis tróficos, incluindo as espécies oportunistas de predadores. Esse *footprint* representa a utilização de carbono da suposta presa dos nematoides indicadores de estrutura. O *footprint* funcional é a área total dos *footprints* de enriquecimento e de estrutura.

Os NMF dos parasitas de plantas, bacteriófagos e micófagos são baseados na avaliação dos NMF para nematoides indicadores de carbono e energia que entram na cadeia alimentar do solo por meio dos seus respectivos canais (FERRIS et al., 2001; FERRIS, 2010).

As comunidades de nematoides em ecossistemas agrícolas e naturais

Comparações entre ecossistemas agrícolas e naturais revelam a magnitude dos efeitos das práticas agrícolas na diversidade e abundância dos nematoides do solo (ZAMITH; LORDELLO, 1957; GOULART et al., 2003; SÁNCHEZ-MORENO; FERRIS, 2007; PATTISON et al., 2008; TOMAZINI et al., 2008; STIRLING et al., 2010). Consequentemente, Neher (2010) enfatizou a necessidade de testar hipóteses ecológicas, especificamente em relação a ecologia de nematoides em solos naturais e agrícolas afim de

buscar estratégias para o controle biológico, monitoramento ambiental e para desenvolver modelos preditivos para a tomada de decisão.

O manejo de áreas agrícolas altera a estrutura do solo degradando-o, e, consequentemente, desfavorece os nematoides predadores e a regulação biótica que ocorre em ecossistemas menos manejados, podendo resultar no aumento das populações de patógenos (SYLVAIN; WALL, 2011). Dessa forma, as comunidades de nematoides que se desenvolvem sob áreas agrícolas se diferenciam daquelas que se desenvolvem em áreas de vegetação nativa.

A estrutura da comunidade de nematoides é afetada pelo manejo do solo talvez devido a heterogeneidade de espécies vegetais nas áreas de vegetação nativa, desfavorecendo, assim, os nematoides parasitas de plantas. Além disso, o baixo teor de nutrientes, alto teor de matéria orgânica e a mínima perturbação do solo pode contribuir para menores populações deste grupo trófico. A complexidade da cadeia trófica do solo e a maior interação entre os organismos explicam a elevada diversidade de nematoides sob solos de ecossistemas naturais em relação a solos sob monocultivos (PATTISON et al., 2008).

Sánchez-Moreno e Ferris (2007) avaliaram a estrutura da fauna do solo em áreas naturais e agrícolas e testaram a supressividade a *Meloidogyne incognita*. Para isso, observaram os efeitos de cinco tratamentos (dois fertilizantes nitrogenados, dois herbicidas e manejo simulado) na supressividade. Calcularam os índices de cadeia trófica para inferir a condição do solo, observando que ao contrário das áreas agrícolas, as áreas naturais foram caracterizadas por cadeias tróficas longas e complexas, com predominância de nematoides predadores e onívoros, efetivamente suprimindo populações de parasitas de plantas. Por fim, concluíram que o manejo agrícola leva a redução da capacidade supressiva da cadeia trófica do solo.

Kimenju et al., (2009) estudaram os efeitos do uso da terra e a intensidade do cultivo na estrutura das comunidades de nematoides. Para isso, analisaram áreas de floresta nativa, floresta plantada, chá, café, capim napier, sistema agroflorestal, pousio e cultivo anual intercalando milho e feijão. Observaram que a abundância e a densidade de nematoides decresceu com a intensidade do cultivo, ao comparar com a área de floresta nativa que utilizaram como referência. A diversidade refletiu as alterações físicas, químicas biológicas do solo. E os maiores índices de maturidade foram registrados em floresta e os maiores índices de parasitos de plantas em área intensivamente cultivada sob cultivo anual (milho/feijão), com nematoides de vida livre predominando em floresta e parasitas de plantas em sistema anual de cultivo.

Li et al. (2007), observaram que similares comunidades de plantas e condições do solo podem suportar diferentes comunidades de nematoides, com diferentes grupos tróficos respondendo ao uso da terra de diferentes formas. Todavia, similares comunidades de nematoides foram observadas após anos de cultivo. O que indica que essa discrepância pode se tornar menor ao longo do tempo. Assim, a diversidade trófica varia pouco ao longo de uma cronosequência. Os autores observaram também que a adição de um mesmo recurso pode propiciar diferentes respostas de componentes da cadeia trófica dependendo de sua estrutura original. E, assim, essa discrepança diminui ao longo do tempo apenas para nematoides parasitas de plantas, mas persiste para a maioria dos grupos tróficos tal como bacteriófagos, micófagos e predadores.

Em um experimento de longo prazo, Eisenhauer et al. (2011) observaram que a densidade de nematoides predadores aumenta significativamente com a diversidade de espécies vegetais, apontando para o aumento da complexidade da cadeia alimentar em decorrência da maior riqueza de espécies vegetais. Além disso, revelaram que em formações vegetais complexas a comunidade de nematoides é alterada e reduz a abundância de parasitas de plantas, enquanto favorece micófagos, bacteriófagos e predadores.

No Brasil, estudos que relacionem ecossistemas agrícolas e naturais com intuito de avaliar as diferenças na estrutura e composição das comunidades de nematoides são escassos. Entretanto, quase todas as pesquisas que foram realizadas comparando Floresta Atlântica e áreas cultivadas enfatizam apenas as populações de nematoides parasitas de plantas (SILVA et al., 2008a, 2008b; TOMAZINI et al., 2008) ou apenas uma família deste grupo trófico (Longidoridae: JESUS, 2011), com exceção apenas para uma pesquisa recente, Arieira et al., (2013), na qual avaliaram tanto a ocorrência de nematoides de vida livre quanto de parasitas de plantas em áreas cultivadas com cana-de-açúcar em comparação a remanescente de Floresta Atlântica situadas no Paraná.

No Brasil, Zamith e Lordello (1957) foram os pioneiros no estudo de comunidades de nematoides em solos cobertos com vegetação nativa, comparando-os com solos sob cultivos agrícolas. Ao final do estudo os autores concluíram que os solos cobertos com vegetação nativa apresentavam maior diversidade que os solos cultivados e que ao converter áreas florestadas em áreas cultivadas, algumas espécies tiveram a sua abundância reduzida enquanto outras permaneceram.

No bioma Cerrado, Goulart et al., (2003) avaliaram a estrutura das comunidades de nematoides em solos sob vegetação nativa, cultivo anual (milho) e cultivo perene (goiaba) e

observaram menor similaridade entre as áreas com cultivo anual e com vegetação nativa, além de observar maior número de táxons em vegetação nativa que em cultivados.

Tomazini et al., (2008) analisaram a estrutura de comunidades de nematoides em remanescente de floresta e de culturas perenes (citrus, pessegueiro e bananeira) e anuais (milho e leguminosas). Ao fim do estudo concluíram que a menor profundidade a riqueza de gênero foi maior em remanescente de floresta e a maior profundidade em área cultivada com milho, leguminosas e citrus. Além de observar que a conversão de vegetação nativa para cultivos anuais e perenes influenciaram as comunidades de nematoides, resultando na ausência de *Discocronemella* e *Xiphidorus* (antes abundantes), na estabilidade de *Helicotylenchus* e *Rotylenchus* e na introdução de *Pratylenchus* e *Radopholus*.

Silva et al., (2008a) avaliaram as comunidades de nematoides parasitas de plantas em diferentes áreas de Floresta Atlântica, São Paulo. Observaram a presença de 17 espécies, sendo que as famílias de maior abundância e riqueza taxonômica de espécies foram Hoplolaimidae, Criconematidae e Tylenchulidae. Além disso, os autores verificaram que os nematoides parasitas de plantas de maior importância econômica na região de estudo (*Meloidogyne javanica* e *Radopholus similis*) não foram encontrados nas áreas de Floresta Atlântica analisadas, sugerindo que estes foram introduzidos nas áreas de cultivo.

Silva et al., (2008b) analisaram a diversidade de nematoides parasitas de plantas em áreas preservadas da Floresta Amazônica (Mato Grosso) e avaliaram o efeito do uso da terra sobre a comunidade destes nematoides. A comparação entre as áreas revelou baixa similaridade entre as duas áreas de preservação, provável reflexo do endemismo vegetal do bioma, e ausência de similaridade entre as áreas com vegetação primária e as contíguas cultivadas, demonstrando forte influência da atividade agrícola sobre a comunidade dos nematoides parasitas de plantas.

Estudos realizados na Nigéria confirmaram que a substituição de ecossistemas nativos por cultivos agrícolas ocasiona reduções nas densidades de nematoides (CAVENESS, 1972). Embora, essas densidades cresçam nos cultivos anuais e decresçam em períodos de pousio, nas áreas cultivadas *Pratylenchus*, *Meloidogyne* e *Helicotylenchus* predominaram, em detrimento dos gêneros *Scutellonema*, *Xiphinema*, *Paratylenchus* e *Criconemoides*.

Na Austrália, as comunidades de nematoides foram analisadas em solo sob cultivo de banana por Pattison et al., (2008). Para determinar o impacto do cultivo nas propriedades do solo utilizaram áreas de floresta e pastagem para compará-las. Neste estudo, os seguintes atributos do solo foram determinados: densidade do solo, teor de matéria orgânica, taxa de infiltração de água, nitrogênio total, pH e condutividade elétrica. Os autores concluíram que o

cultivo de banana reduziu a diversidade de nematoides quando comparado aos outros manejos menos intensivos.

Com base no exposto acima pode-se inferir que a estrutura e a composição das comunidades de nematoides que se desenvolvem em solos agrícolas se diferenciam daquelas que se desenvolvem em solos de vegetação nativa. Com o manejo do solo, as comunidades de nematoides são afetadas com consequente redução da diversidade e da abundância de nematoides (YEATES, 2007; CARRASCOSA et al., 2014). Além do mais, a conversão de áreas de florestas em agricultura influencia negativamente diversos parâmetros do solo, aumentando a compactação do solo e, consequentemente, reduzindo a biodiversidade do solo (ROUSSEAU et al., 2013).

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***Relationships between nematode assemblages and physical properties across
land use types****

CAPÍTULO II

Relationships between nematode assemblages and physical properties across land use types

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Abstract

Sugarcane is one of the most important crops in Brazil and its productivity is associated with several factors, among these the presence of plant-parasitic nematodes leads to high losses. Soil nematode assemblages were investigated in three land use types (forest, sugarcane under two management) in order to evaluate relationships with soil physical properties. Relationships between soil physical properties (water content, bulk density, total porosity, soil mechanical resistance, sand, silt and clay) and the distribution of the nematodes were evaluated by canonical correspondence analysis. Cluster analysis was used to assess similarity relations between nematode communities across areas. Soil samples were collected at 0-10, 10-20, 20-30, 30-40, and 40-50 cm deep along two 200-m transects with points at 20-m intervals from each other. *Prismatolaimus*, *Tyloclorus*, *Paratrichodorus*, *Rotylenchulus* and *Tylenchus* were detected only in forest remnant. *Hoplolaimus* was detected exclusively in sugarcane under non-intensive management. Plant-parasitic and fungivores nematodes were associated with soil physical properties. Canonical Correspondence and Cluster Analyses set out that the composition and structure of the nematode communities differed among studied sites.

Keywords: sugarcane, Atlantic Forest, biodiversity, soil conservation, Canonical Correspondence Analysis, Cluster.

Introduction

Sugarcane is one of the most important crops in Brazil. In northeastern Brazil, sugarcane cultivation plays an important socio-economic role but productivity is associated to several factors, among these the presence of plant-parasitic nematodes leads to high losses. Usually, conventional management practices such as pesticide and fertilizer application, soil tillage, and irrigation have been applied to increase yields; however, these management practices lead to changes in soil biological, physical and/or chemistry properties (Bell et al. 2007; Stirling et al. 2010; Baquero et al. 2012; Godefroid et al. 2013). Thus, the adopted land use can affect the soil fauna, including the nematodes communities by altering their natural occurrence (Yeates 2003; Figueira et al. 2011).

Even though indicators based on nematode community composition are among the best developed metrics of soil health, little research has been done to support their application to management (Ugarte et al. 2013). Likewise, studies that relate soil physical properties to nematode community structure are scarce. In sugarcane fields nematode community have been correlated to soil physical properties in different environments (Cardoso et al. 2012), chemical properties (Matos et al. 2011), water content and particle density of soil (Rodrigues et al. 2011). Research understanding relationships between sugarcane management strategies and the occurrence and density population of plant-parasitic nematodes allows improving strategies to minimize yield losses.

Ordination and cluster techniques allow identify environmental factors that most influence the structure and composition of the nematodes communities. Canonical correspondence analysis, widely used for characterize community ecology, permits relate nematodes to soil condition (Ficus and Neher 2002; Neher et al. 2005), while cluster analysis is performed in order to evaluate relations similarity between nematodes communities in areas with different soil managements. Therefore, this research investigated the influence of land use types on nematode community structure in the sugarcane zone of Pernambuco State, Brazil.

Material and Methods

Study sites

Were selected for this study three land use types (forest and sugarcane under two management) located in Pernambuco State, Brazil (Table 1). The area under management 1 is a Ultisol located at northern Pernambuco with mechanized harvesting, irrigated, burning, 87% of sand, and higher soil bulk density. The area under management 2 is an Oxisol located at southern of Pernambuco with manual harvesting, non-irrigated, and 95% of sand. Pesticides and fertilizers are applied in both cultivated areas. According to Köppen Climate Classification (Köppen 1948), climate at all sites is humid tropical type As' or pseudotropicalis, characterized by hot and humid, with rains from autumn to winter, and annual averages temperature ranging around 24°C.

Table 1. Localization of the study sites.

Sites	Farms	Zona da Mata, PE, Brazil	Geographic coodenates	Sampling date
Atlantic Forest remnant	Santa Teresa, Goiânia	North	7°36'45,7"S 35°00'47,6"W	November 2009
Sugarcane (management 1)	Santa Teresa, Goiânia	North	7°36'48"S 34°58'45,9"W	September 2009
Sugarcane (management 2)	Salgado, Ipojuca	South	8°31'29"S 35°03'26"W	September 2010

Soil sampling

Soil samples (600g) were collected at depths of 0-10, 10-20, 20-30, 30-40, and 40-50 cm along two 200-m transects with points maintained in approximately 20-m intervals from each other. Samples were placed in identified plastic bags and taken to the laboratory, kept under refrigeration ($\pm 6^{\circ}\text{C}$).

Nematodes extraction

Samples were processed, homogenized and nematodes extracted per 300 cm³ of soil through centrifugal flotation method in sucrose solution, Jenkins (1964). Population was estimated by counting on Petersø slides, under an optical microscope, with two replications, and results computed per 300 cm³ of soil. Nematodes were classified according to feeding

habits in five trophic groups (plant-parasitic, bacterivores, fungivores, predators and omnivores), based on the stoma and esophagus morphology, according to Yeates et al. (1993). Plant-parasitic nematodes were identified to genus level according to Mai et al. (1996).

Physical properties analyses

Soil physical analyses were performed at the Laboratory of Soil Mechanics and Waste Utilization, Department of Rural Technology, UFRPE, using methods detailed in EMBRAPA (1997):

- Particle size analysis (hydrometer method);
- Water content (thermogravimetric method);
- Particle density (volumetric flask method);
- Bulk density (volumetric ring method);
- Total porosity (calculation by particle density and bulk density values);
- Mechanical resistance (digital penetrometer).

Statistical analyses

Relationships between soil physical properties and nematodes taxa were evaluated by Canonical Correspondence (CCA) and Cluster (Sorensen Bray-Curtis) Analyses. Canonical Correspondence Analysis was carried out to identify patterns of association between the nematode community composition and physical properties in soils with different stages of conservation: sugarcane cropping under non-intensive management, sugarcane cropping under intensive management and an Atlantic Forest remnant. Monte Carlo permutation test, with 999 permutations, was conducted to estimate the significance of first and second axes. Cluster analysis was performed in order to assess relationships of similarity between the nematodes communities in the areas. Because it is less affected by outliers than simple and complete linkage methods (Norusis 2011), Unweighted Pair Group Method with Arithmetic Mean - UPGMA was employed as a clustering algorithm based on the Sorensen (Bray-Curtis) distance. Multivariate analyses described were performed by PC-Ord software Version 6.07 (McCune and Mefford 2011).

Results and Discussion

Nematode communities structure and composition

Eighteen genera were identified; eleven were common to all areas (Table 2). However, *Prismatolaimus*, *Tyloclorus*, *Paratrichodorus*, *Rotylenchus* and *Tylenchus* were detected only in forest remnant, whereas *Hoplolaimus* was found, exclusively, in sugarcane under management 2. This is the first report of *Prismatolaimus* in Atlantic Forest in Pernambuco State. There are few reports in Brazil, such as in the biome cerrado in São Carlos (SP) (Goulart et al. 2003); in the contiguous areas of preserved natural forest, banana, citrus, peach and corn x legumes on rotation in Piracicaba (SP) (Tomazini et al. 2008) and in beans cropping, Seropédica (RJ) (Mondino et al. 2009).

Total abundance was lower in sugarcane under management 2. *Pratylenchus* was dominant in sugarcane under management 1, Dorylaimidae in forest remnant and *Hoplolaimus* in sugarcane under management 2. Data corroborate Sánchez-Moreno et al. (2006) that assure different management practices and cropping systems determine soil properties and hence nematodes abundance.

Table 2. Abundance, means and dominance of the nematodes in sugarcane cropping and in Atlantic Forest remnant located in Zona da Mata de Pernambuco.

Trophic groups	Sugarcane (management 1)			Forest remnant			Sugarcane (management 2)		
	A	Means ± SD	D (%)	A	Means ± SD	D (%)	A	Means ± SD	D (%)
Bacterivores									
<i>Acrobelus</i>	320	2.46 ± 10.95	1.05	404	1.55 ± 7.06	1.42	1,599	14.54 ± 36.95	9.08
<i>Rhabditidae</i>	853	6.55 ± 16.32	2.80	5,555	21.36 ± 32.79	19.51	518	4.71 ± 13.92	2.94
<i>Prismatolaimus</i>	0	0.00	0.00	111	0.60 ± 4.14	0.39	0,00	0.00	0.00
Fungivores									
Aphelenchidae	704	5.41 ± 15.38	2.31	346	1.33 ± 6.20	1.21	254	2.31 ± 7.14	1.44
Omnivores									
Dorylaimidae	2,382	18.32 ± 35.37	7.83	8,439	32.45 ± 44.23	29.65	5,743	52.21 ± 67.67	32.62
Predators									
Mononchidae	1,545	11.88 ± 47.00	5.08	3,341	12.85 ± 25.14	11.74	583	5.3 ± 14.33	3.31
Plant-parasitic									
<i>Criconemella</i>	658	5.06 ± 14.89	2.16	884	3.40 ± 12.58	3.11	347	3.15 ± 11.81	1.97
<i>Helicotylenchus</i>	7,774	59.80 ± 81.43	25.56	704	2.70 ± 11.95	2.47	115	1.04 ± 4.87	0.65
<i>Hemicyclophora</i>	36	0.27 ± 2.27	0.12	99	0.38 ± 4.59	0.35	0,00	0.00	0.00
<i>Hoplolaimus</i>	0	0.00	0.00	0,00	0.00	0.00	6,402	58.02 ± 81.66	36.37
<i>Meloidogyne</i>	1,074	8.26 ± 19.00	3.53	716	2.75 ± 9.84	2.52	184	1.67 ± 15.49	1.04
<i>Pratylenchus</i>	11,788	90.67 ± 107.56	38.76	510	1.96 ± 8.34	1.79	731	6.64 ± 18.49	4.15
<i>Paratrichodorus</i>	0	0.00	0.00	20	0.07 ± 1.24	0.07	0,00	0.00	0.00
<i>Rotylenchulus</i>	0	0.00	0.00	293	1.12 ± 6.69	1.03	0,00	0.00	0.00
<i>Tylenchus</i>	0	0.00	0.00	75	0.28 ± 3.28	0.26	0,00	0.00	0.00
<i>Trichodorus</i>	1,229	9.45 ± 24.66	4.04	41	0.15 ± 1.83	0.14	168	1.53 ± 7.54	0.95
<i>Tyloclorus</i>	0	0.00	0.00	3,321	12.77 ± 27.33	11.67	0,00	0.00	0.00
<i>Xiphinema</i>	2,046	15.73 ± 30.96	6.73	3,566	13.71 ± 24.15	12.53	960	8.72 ± 18.31	5.45
Total	30,408	233.9 ± 177.4		28,425	109.5 ± 88.1		17,604	160 ± 142.81	

A (abundance) sum of nematodes number in 110 samples from each area by taxa per 300 cm³ of soil, *Means ± SD* Means number and standard deviation of nematodes per 300 cm³ of soil in each area, *D(%)* Dominance of each trophic group and taxa expressed as a percentage.

Relationships between nematofauna and soil physical properties

Relations between nematode community and soil physical properties are described in the ordination diagram (Figure 1). Soil physical properties (water content, total porosity, bulk density, mechanical resistance, silt, sand and clay) are represented by straight lines. Results of CCA presented eigenvalues: $\lambda_1 = 0.312$ ($p = 0.002$) and $\lambda_2 = 0.140$ ($p = \text{n.a.}$), considered low (< 0.5) according to ter Braak (1995).

Correlations between nematode-environment were 0.825 to the axis 1 and 0.777 to the axis 2. Canonical axes explained 19.5% of the variation, indicating much variance unexplained. Thus other factors are affecting the structure and composition of nematode communities in each vegetation type. Nevertheless, the significance of nematodes-environment relationships was not affected, because the Monte Carlo permutation test was significant in the axis 1 at a probability level of 1%, indicating nematode genera are correlated with soil variables provided.

As set out in Table 3, the summary of canonical correspondence analysis, which shows the correlation values between soil attribute and two first ordination axes.

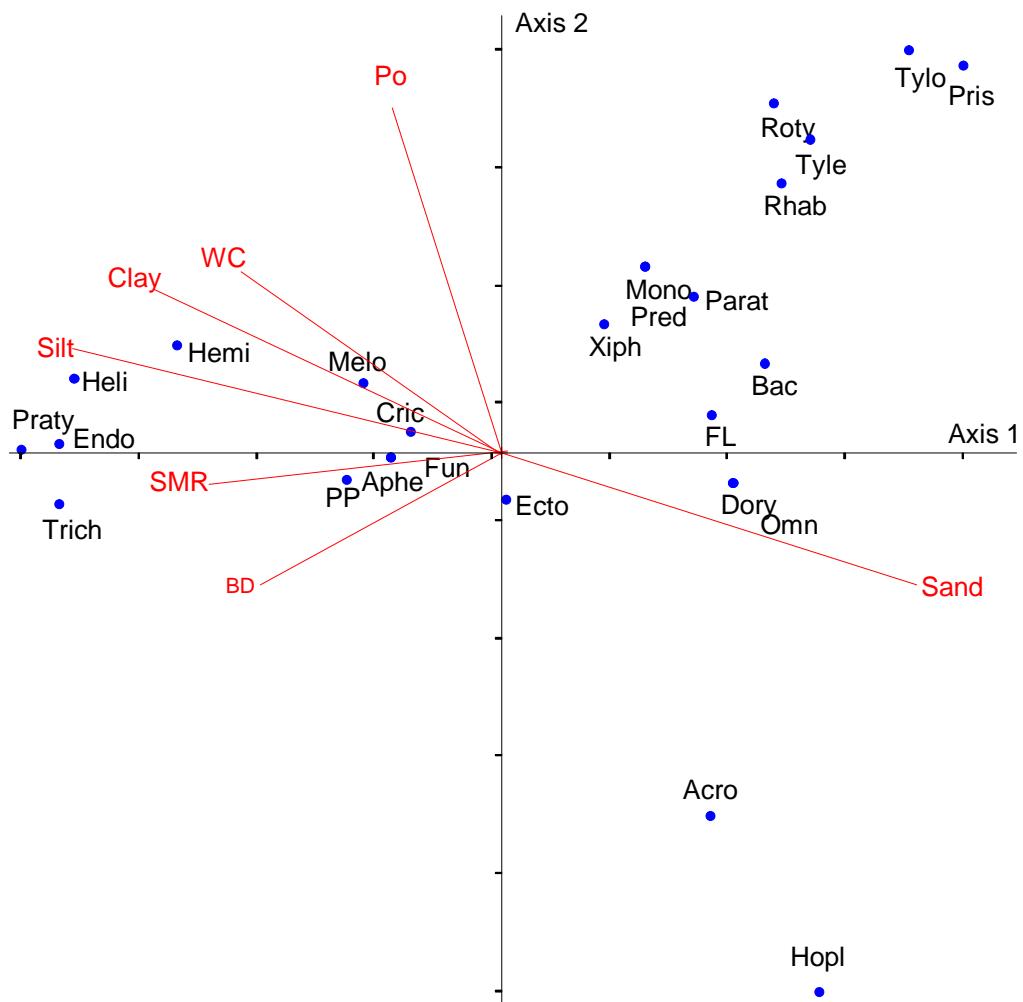
Table 3. Correlation between environmental variables and axis, significance of permutation test of Monte Carlo and variance explained.

	Axis 1	Axis 2
Correlations nematodes-environments	0.312 0.825	0.140 0.777
% of variance explained	13.5	6.0
Monte Carlo Test (p)	0.002	n.a.
Trophic groups		
Free-living	0.457	0.100
Plant-parasitic	-0.334	-0.075
Fungivores	-0.238	-0.015
Bacterivores	0.569	0.243
Omnivores	0.502	-0.083
Predators	0.312	0.508
Soil Physical Properties		
Water content	-0.565	0.500
Bulk density	-0.522	-0.362
Total porosity	-0.236	0.939
Mechanical resistance	-0.648	-0.091
Silt	-0.930	0.284
Clay	-0.757	0.446
Sand	0.898	-0.362

p = proportion of randomized runs with species-environment correlation greater than or equal to the observed species-environment correlation; that is $p = (1 + n^o \text{ permutations} \times \text{observed}) / (1 + n^o \text{ permutations})$. Values of p are not reported for axis 2 because using a simple randomization test for this axis may bias the values, hence: n.a. = not applicable.

The soil attributes silt, clay and mechanical resistance were strongly related to the first ordination axis. While soil total porosity showed lowest relative with this axis. However, the second axis was associated to mainly soil bulk density and sand content. Positives and negatives values of the correlations shows that plant-feeder and fungivore nematodes are associated to soil physical properties, except soil sandy content, which is correlated with others free-living nematodes: bacterivores, omnivores and predators. This result indicates the importance of these soil physical attributes in the study of soil nematodes.

Plant-parasitic and fungivores were correlated to soil mechanical resistance, whereas *Criconemella* and *Helicotylenchus* showed positive correlation with soil silt content. *Meloidogyne* correlated to water content and clay soil. Dorylaimidae was related to soil sand content. Nematodes in this family are usually larger and restricted by the smaller pores space, therefore they are considered sensitive to changes in soil properties and are often associated to less intensive management (Gomes et al. 2003; Goulart and Ferraz 2003; Cardoso et al. 2011). In this study, abundance and dominance values ranged from 8,439 (29.65 %) in forest to 2,382 (7.83 %) in sugarcane under intensive management. Gomes et al. (2003) consider sites with low human intervention those with Dorylaimidae dominance above 25%.



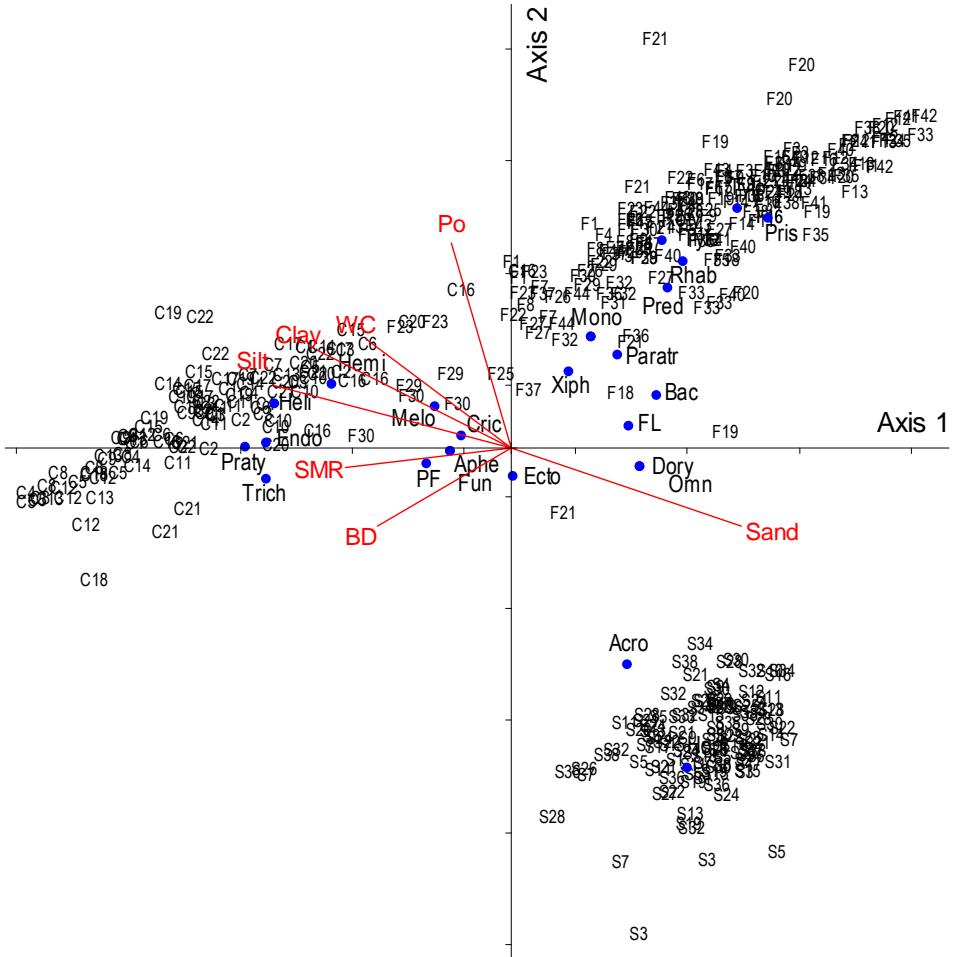
Po total porosity; *WC* water content; *SMR* soil mechanical resistance; *BD* bulk density; *Melo Meloidogyne*; *Hemi Hemicycliophora*; *Praty Pratylenchus*; *Cric Criconemella*; *Aphe Aphelenchidae*; *Trich Trichodorus*; *Tylo Tyloclorus*; *Pris Prismatolaimus*; *Acro Acrobeles*; *Hopl Hoplolaimus*; *Rhab Rhabditidae*; *Dory Dorylaimidae*; *Heli Helicotylenchus*; *Mono Mononchidae*; *Parat Paratrichodorus*; *Roty Rotylenchulus*; *Tyle Tylenchus*; *Xiph Xiphinema*; *Endo* plant endoparasites; *Ecto* plant ectoparasites; *PP* plant-parasitics; *FL* free-living; *Pred* predators; *Fun* Fungivores; *Omn* omnivores; *Bac* bacterivores.

Figure 1. Ordination diagram of the nematode communities composition and trophic groups by Canonical Correspondence Analysis (CCA).

Nematodes taxa ordination by CCA (Figure 1) suggests that *Pratylenchus*, *Hemicycliophora*, *Helicotylenchus*, *Meloidogyne*, and *Criconemella* are related to sugarcane cropping under intensive management, soil characterized by higher SMR, bulk density, water content and clay. In Forest Atlantic remnant, *Prismatolaimus*, *Tyloadorus*, *Paratrichodorus*, *Rotylenchulus*, *Tylenchus*, and *Xiphinema* were unrelated to soil physical properties. However, *Hoplolaimus* and *Acrobeles* were associated to sugarcane cropping under non-intensive management.

As set out in Figure 2, CCA ordered three distinct groups according to the sampling sites. Plant-parasitic nematodes were associated to sugarcane under intensive management.

Whereas *Acrobeles* and *Hoplolaimus* were associated to sugarcane under non-intensive management. Free-living nematodes, except Aphelenchidae, were associated to forest area, corroborating presence of *Acrobeles* sp., *Prismatolaimus* sp. and Dorylaimidae associated to preserved forest areas reported by Neher et al. (2005).



Po total porosity; *WC* water content; *SMR* soil mechanical resistance; *BD* bulk density; *Melo Meloidogyne*; *Hemi Hemicycliophora*; *Praty Pratylenchus*; *Cric Criconemella*; *Aphe Aphelenchidae*; *Trich Trichodorus*; *Tylo Tyloclorus*; *Pris Prismatolaimus*; *Acro Acrobeles*; *Hoplo Hoplolaimus*; *Rha Rhabditidae*; *Dory Dorylaimidae*; *Heli Helicotylenchus*; *Mono Mononchidae*; *Paratr Paratrichodorus*; *Roty Rotylenchulus*; *Tyle Tylechus*; *Xiph Xiphinema*; *Endo* plant endoparasites; *Ecto* plant ectoparasites; *PF* plant-feeders; *FL* free-living; *Pred* predators; *Fun* Fungivores; *Omn* omnivores; *Bac* bacterivores.

Figure 2. Ordination diagram of sampling points obtained by Canonical Correspondence Analysis (CCA) based on the distribution of the nematodes taxa and correlations with soil physical properties in sugarcane cropping under management 1 (C), in Atlantic Forest remnant (F), and sugarcane cropping under management 2 (S).

Cluster analysis

Cluster analysis dendrogram presents differences in nematode community structure of the studied areas by formation of three groups (Figure 3), with higher similarity between the forest remnant and sugarcane under management 2, suggesting that soil managements changes nematode communities.

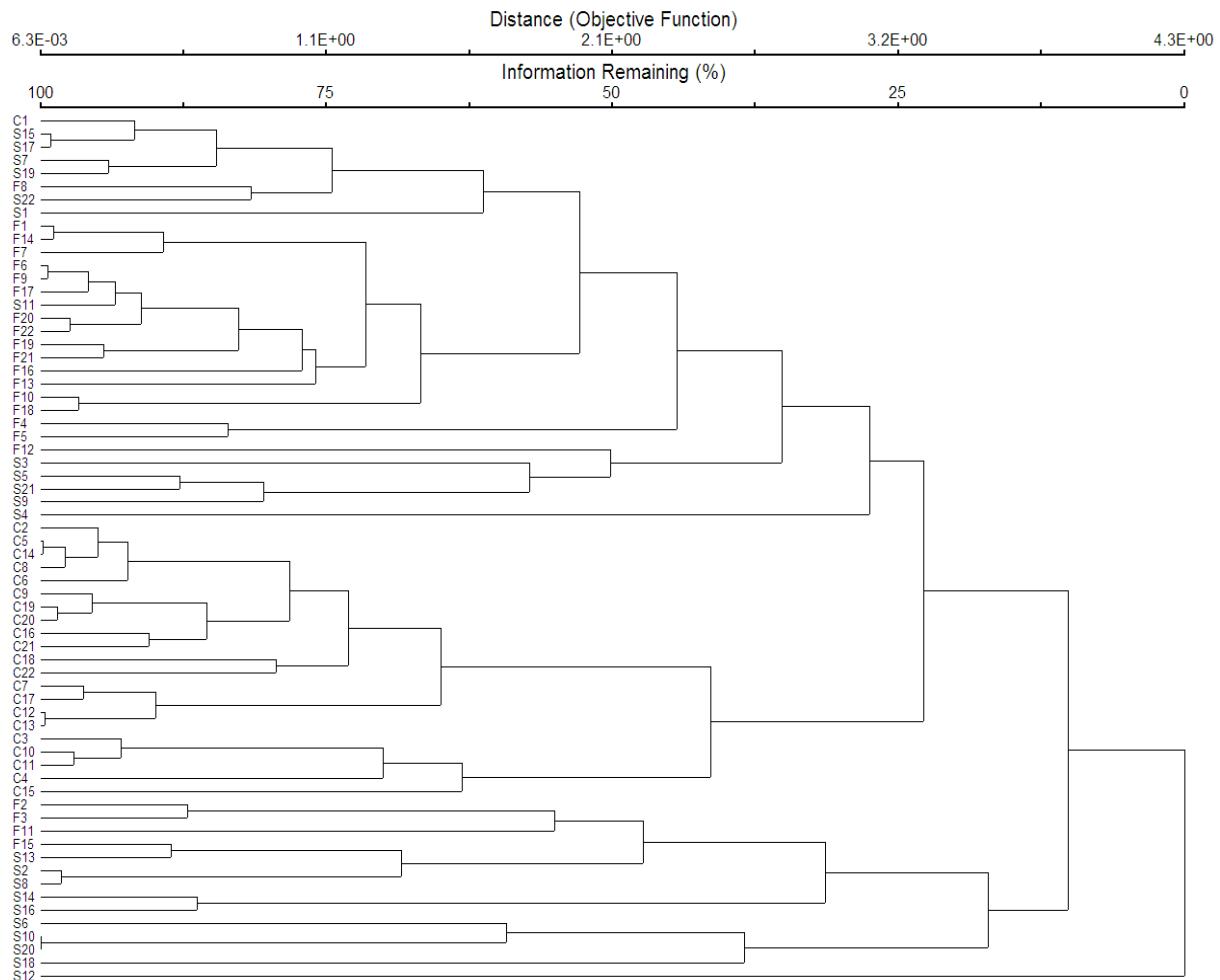


Figure 3. Dendrogram of cluster analysis using the Sorenson (Bray-Curtis) distance measure and UPGMA linkage method showing the groups obtained in sugarcane cropping under management 1 (C), in Atlantic Forest remnant (F), and sugarcane cropping under management 2 (S).

According to multivariate analyses, difference within nematode communities depended on environmental conditions; corroborating Lazarova et al. (2004) that applying similar techniques concluded the environmental heterogeneity influences distribution patterns of nematodes, where each environment can be characterized by a particular community.

Conclusion

The present study shows difference among land-use forest and sugarcane soils with regard to soil properties and the structure and composition of the nematode assemblages. Soil bulk density and abundances of all nematodes and of plant-parasitic nematodes tended to be higher in sugarcane soils. In contrast, the total porosity, soil respiration, predator nematodes were at lower levels in the sugarcane soils. Ordination and cluster analyses revealed differences in the nematode assemblage composition of study sites. Thus, different edaphic conditions support different nematode communities.

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*Comparing sugarcane fields and forest fragments: the effect of disturbance on
soil physical properties and nematode assemblages**

CAPÍTULO III

Comparing sugarcane fields and forest fragments: the effect of disturbance on soil physical properties and nematode assemblages

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Highlights

- Structure and composition of the nematode assemblage and soil properties differed between forest and sugarcane soils, even in the same locality;
- The soil bulk density, the abundance of all nematodes and of plant-parasitic nematodes were greater while soil porosity, soil respiration, and abundance of predator nematodes were lower in sugarcane than in forest areas;
- We suggest that sugarcane management practices result in changes in the soil properties, and concomitantly alter the composition and structure of the nematode assemblages;
- Canonical correspondence analysis indicated that others environmental factors also might be affecting the nematofauna.

Abstract

Comparisons of agricultural and natural ecosystems reveal the magnitude of the effects of agricultural practices on the diversity and abundance of soil nematodes. Consequently, there is the need for testing ecological hypotheses, specifically with regard to nematode ecology, in natural and agricultural soils in order to seek strategies for biological control, environmental monitoring and for developing predictive models for policy decisions. We studied soil nematode assemblages and soil physical attributes of five soil layers (0-10; 10-20; 20-30; 30-40 and 40-50-cm) from sugarcane plantations and forest remnants in the sugarcane zone of Pernambuco State, Brazil. Structure and composition of the nematode assemblage and soil properties differed between forest and sugarcane soils, even in the same locality. The soil bulk density, the abundance of all nematodes and of plant-parasitic nematodes were greater while soil porosity, soil respiration, and abundance of predator nematodes were lower in sugarcane than in forest areas. We suggest that sugarcane management practices result in changes in the soil properties, and concomitantly alter the composition and structure of the nematode assemblages. Canonical correspondence analysis indicated that others environmental factors also might be affecting the nematofauna.

Keywords: sugarcane, Atlantic Forest, Cluster Analysis, bulk density, total porosity.

1. Introduction

The Atlantic Forest of Brazil is considered one of the world's biological diversity hotspots (Mittermeier et al. 2005; Myers et al. 2000). However, vast areas of Atlantic Forest have been converted to agricultural production so that it is now one of the earth's most threatened tropical biomes with only 11.73% of its original area remaining (Ribeiro et al. 2009). In the northeast of Brazil, government economic incentives during the 1980s led to expansion of sugarcane farming and concomitant reduction of native vegetation. However, the sugarcane crop plays important agronomic, economic and societal roles. During 2013/2014, in Pernambuco State, 14,400 tonnes of sugarcane were harvested from 284,600 ha with production of 50,600 Kg.ha⁻¹ (CONAB, 2012).

In conventional, high-yielding production systems, agricultural soils are commonly subjected to long-term monoculture, uncontrolled traffic of heavy machinery and excessive tillage. Such practices eventually contribute to yield decline due to their effects on the biological, physical and/or chemical characteristics of the soil (Garside et al. 2005; Briar et al. 2007; Lanzanova et al. 2007; Barthès et al. 2011; Briar et al. 2011). Biological effects of farming system practices are observed in the soil fauna, including the nematode communities, with important implications for the functioning and stability of soil food webs (Powell et al. 2007; Sánchez-Moreno et al. 2008; Ferris, 2010; Briar et al. 2011; Sánchez-Moreno et al. 2011). Moreover, forest conversion to conventional agriculture impacts soil quality, reduces biodiversity, alters the soil physical properties and increases compaction (Gaji , 2013; Rousseau et al. 2013)

Comparisons of agricultural and natural ecosystems reveal the magnitude of the effects of agricultural practices on the diversity and abundance of soil nematodes (Zamith and Lordello, 1957; Goulart et al. 2003; Sánchez-Moreno and Ferris, 2007; Pattison et al. 2008; Tomazini et al. 2008; Stirling et al. 2010). Consequently, Neher (2010) emphasized the need for testing ecological hypotheses, specifically with regard to nematode ecology, in natural and agricultural soils in order to seek strategies for biological control, environmental monitoring and for developing predictive models for policy decisions.

Studies have examined the effects of soil physical properties on nematode assemblages in Brazil, however there is a need for better understanding of their roles and relevance in both natural and agricultural ecosystems. The challenge and opportunity are to develop management systems that sustain and improve sugarcane yields while minimizing environmental impact. For example, soil compaction (Cardoso et al. 2012), changes in soil

chemistry (Matos et al. 2011), and changes in soil moisture and particle density (Rodrigues et al. 2011) resulting from agricultural practices all affect the structure of soil nematode assemblages.

In considering previous studies, we hypothesized that: (1) abundance and diversity of plant-parasitic nematodes differs in forest and sugarcane fields; (2) soil physical properties are affected by management practices; and (3) changes in soil physical properties affect the abundance and diversity of plant-parasitic and predator nematodes. Therefore, we aimed to: (a) describe the nematode diversity of native forest fragments and sugarcane fields; and (b) evaluate the relationships between soil attributes (bulk density, total porosity and soil respiration) and nematode assemblages.

2. Material and Methods

2.1 Study sites

This research was carried out in ten sugarcane plantations and eight forest fragments. Each forest fragment was entirely surrounded by one of the sugarcane plantations (Table 1). Soil samples were collected between 2010 and 2012 from four farms (Estreliana, União & Indústria, Trapiche and Salgado) located in Zona da Mata Sul of Pernambuco. The local climate is humid tropical type Amsø according to Köppen Climate Classification (Köppen, 1948), with precipitation in autumn and winter and average annual rainfall between 1,000 and 2,200 mm.

2.2 Soil sampling

At each site, soil samples were collected at five 10-cm depth increments from the four corners of a 10 x 10 m square. Soil samples were taken by a modified TORSOL® sampler for assessment of bulk density, water content, and soil respiration and stored in parafilm-sealed plastic containers. About 600g of soil was collected at each depth for nematode and texture analyses. The samples were packaged in labeled plastic bags and immediately transported to the Phytonematology Laboratory, Agronomy Department, UFRPE.

Table 1 Location and soil characteristics of ten sugarcane fields and eight forest fragments at Zona da Mata Sul of Pernambuco.

Label	Coordinates	Farm	Municipality	Date Sampling	Soil texture	Soil type
Sugarcane						
ST1	8°33'52.4"S; 35°08'36.2"W	Trapiche	Sirinhaém	Jun 2010	Sandy clay loam	Yellow Ultisol
ST2	8°34'57.3"S; 35°07'01.7"W	Trapiche	Sirinhaém	Jun 2010	Sandy clay loam	Yellow Ultisol
SE1	8°43'12.7"S; 35°07'17.7"W	Estreliana	Ribeirão	Nov 2010	Loamy sand	Yellow Oxisol
SE2	8°42'46.3"S; 35°07'38.1"W	Estreliana	Ribeirão	Nov 2010	Sandy loam	Yellow Oxisol
SE3	8°42'25.3"S; 35°07'14.4"W	Estreliana	Ribeirão	Jan 2011	Sandy clay loam	Yellow Oxisol
SE4	8°43'49.3"S; 35°07'13.9"W	Estreliana	Ribeirão	Jan 2011	Sandy clay loam	Yellow Oxisol
SU1	8°19'57.1"S; 35°18'11.9"W	União e Indústria	Primavera	Jun 2010	Clay	Yellow Ultisol
SU2	8°18'49.3"S; 35°19'39.6"W	União e Indústria	Primavera	Jun 2010	Sandy clay loam	Yellow Ultisol
SS1	8°26'50.1"S; 35°06'21.9"W	Salgado	Ipojuca	Jun 2010	Sandy	Yellow Oxisol
SS2	8°26'53.3"S; 35°06'02.0"W	Salgado	Ipojuca	Jun 2010	Sandy	Yellow Oxisol
Forest						
FA	8°34'07.4"S; 35°08'33.4"W	Trapiche	Sirinhaém	Mar 2012	Sandy loam	Yellow Ultisol
FB	8°33'57.4"S; 35°07'44.6"W	Trapiche	Sirinhaém	Mar 2012	Sandy	Yellow Ultisol
FC	8°34'59.5"S; 35°22'00.5"W	Estreliana	Ribeirão	Nov 2011	Loamy sand	Yellow Oxisol
FD	8°35'28.1"S; 35°20'30.7"W	Estreliana	Ribeirão	Nov 2011	Loamy sand	Yellow Oxisol
FE	8°33'05.4"S; 35°08'48.4"W	Trapiche	Sirinhaém	Set 2012	Sandy clay	Yellow Ultisol
FF	8°33'30.8"S; 35°09'31.2"W	Trapiche	Sirinhaém	Set 2012	Sandy clay loam	Yellow Ultisol
FG	8°40'33.4"S; 35°25'06.9"W	Estreliana	Ribeirão	Set 2012	Sandy clay loam	Yellow Oxisol
FH	8°39'20.7"S; 35°25'12.3"W	Estreliana	Ribeirão	Set 2012	Sandy clay loam	Yellow Oxisol

2.3 Nematodes extraction

Nematodes were extracted by the sucrose centrifugation (Jenkins, 1964). The suspensions were stored in a refrigerator for no more than 3 days before counting and identification. Nematodes were counted at x20 magnification in two replicate aliquots on

Peters glass counting slides (1 ml of capacity). All nematodes counted were identified to genus/family level at x40 and x100 magnification. For identification to the genus level, temporary slides were prepared. Nematode abundance was expressed per 300 cm³ of soil.

2.4 Trophic and functional structure of the nematode assemblages

Nematodes were assigned to five trophic groups according to feeding habits: plant-parasitic (root feeders), bacterivores (bacteria feeders), fungivores (feed on fungal spores), predators (feed on other nematodes), and omnivores (feed on a variety of food sources, including algae, fungi, bacteria, small rotifers, enchytraeids, and small nematodes). This trophic classification was based on the morphology of the stoma and esophagus according to Yeates et al. (1993). Even though the trophic habits of the Tylenchidae are uncertain, all individuals in this family were considered plant-parasitic. Nematodes also were classified in functional guilds based on feeding habits and five colonizer-persister (c-p) groups which represent life history characteristics and sensitivity to environmental perturbation (Bongers, 1990; Bongers and Bongers, 1998). Plant-parasitic nematodes were identified to genus level using keys and descriptions of Mai et al. (1996) and free-living nematodes to the family level according to keys of Tarjan et al. (1977).

The nematode assemblage structure and composition, and taxa dominance also were determined in this study. The structure refers to composition of the assemblage and the abundance of nematode taxa and dominance is the predominance of one or a few taxa within an ecosystem.

2.5 Soil respiration

Microbial activity was estimated from soil respiration as indicated by the C-CO₂ evolution rate (Grisi, 1978). Soil samples (100 g) and container with 10 ml 0.5N KOH were placed into sealed glass chambers and incubated at 25 ± 2°C for 15 days. The CO₂ absorbed by the KOH was determined by titration with HCL 0.1 N, using phenolphthalein and methyl orange as indicators.

2.6 Soil physical properties

Soil physical analyses were performed at the Laboratory of Soil Mechanics and Waste

Utilization, Department of Rural Technology, UFRPE, using methods detailed in EMBRAPA (1997):

- i. Soil texture by the hydrometer method using sodium hydroxide as dispersant.
- ii. Soil bulk density (BD) was determined for intact soil cores 5 cm in diameter, 2.5 cm in length, and 50 cm³ in volume. To determine water content (WC), soil samples were dried at 105-110°C for 24 hours and weighed before and after to determine their weight loss. BD was estimated by division between dry weight (DW) and core volume (BD=DW/50 cm³).
- iii. Soil particle density (PD) was determined in a 50-mL volumetric flask using 20 g of air-dried soil and alcohol as fluid to determine the volume occupied by the particles (PD = 20g of soil/ (50 ml ó alcohol volume)).
- iv. Total porosity (Po) calculated according to formula: Po = (1- (BD/ PD))*100.

2.7 Data analyses

Descriptive statistics were calculated to determine distribution of the data from nematode and soil analyses. Nematodes data were log-transformed log(x+1), prior to analysis to comply with assumptions of normality. Relevant data from each study area were graphed to allow visualization. In addition, two-way (ANOVA) was performed and the Scott-Knott test to assess significant differences among areas and depths ($P<0.05$). ANOVA, descriptive statistics and Pearson's correlation were carried out by software Statistica 8.0 (StatSoft, Inc., 2008).

The response of individual nematode taxa to study sites was visualized using Principal Components Analysis (PCA), while the environmental variables which best explained nematode structure and composition were identified and illustrated using Canonical Correspondence Analysis (CCA). The Monte Carlo test method was used to determine the significance of the first and second axes. PCA was also carried out by the software Statistica 8.0 (StatSoft, Inc., 2008) and only variables with loadings >0.70 were considered. CCA was performed by PC-Ord version 6.07 (McCune and Mefford, 2011)

Differences on nematode community composition from forest and agricultural soils were analyzed by Non-Metric Multidimensional Scaling (NMDS) and analysis of similarity (ANOSIM) based on Dice-Sorensen distance measure. The δ stress value from NMDS should be small, at least less than 0.20 and ideally less than 0.10, showing that the reduction to two

dimensions implies very little loss of information (Legendre and Legendre, 1998). Multivariate analyses were performed by PAST version 2.15 (Hammer et al. 2001).

3. Results and Discussion

3.1 Structure and composition of nematode assemblages

Twenty-one nematode taxa were identified; twelve (*) were common to all areas (Tab. 2). *Alaimus*, *Prismatolaimus*, *Nothotylenchus*, *Discocriconemella*, *Paratylenchus*, *Rotylenchus* and Tylenchidae were detected only in forest fragments. In contrast, *Trichodorus* and *Paratrichodorus* were found, exclusively, in sugarcane fields.

Plant-parasitic nematodes were the most abundant functional group in sugarcane, comprising 76% of the total dominance. *Pratylenchus* and *Helicotylenchus* were genera dominant in sugarcane, 41% and 23% of the total dominance, respectively. *Pratylenchus* was absent from only one of the 10 sugarcane fields.

Free-living nematodes comprised 69% of the nematodes dominance in forest fragments and the order Dorylaimida (Qudsianematidae, Thornematidae, Dorylaimidae, and Nordiidae) predominated with 40%. There was also a substantial presence in forest soils of the root associates and root feeders: Tylenchidae and *Helicotylenchus*, 6.7% and 6.4%, respectively.

Thus, the abundance of predator nematodes was significantly lower in sugarcane areas ($p < 0.05$); conversely, the plant-parasitic nematodes predominated. Total nematode abundance was significantly greater ($p < 0.05$) in sugarcane than in forest soils. Similarly, Pattison et al. (2008) observed twice as many nematodes in banana soils than in pastures or forest. Neher et al. (2005) found greater abundance in wetland and agricultural than in forest soils. Therefore, the structure and composition of nematode assemblages in sugarcane and forest soils were quite different.

Table 2 Abundance, means, standard deviation and dominance of the nematode assemblages associated with ten sugarcane fields and eight Atlantic Forest fragments in Zona da Mata Sul of Pernambuco.

Nematodes		Sugarcane fields			Forest fragments		
Trophic groups	F	A	Means ± SD	D (%)	A	Means ± SD	D (%)
Bacterivores							
Alaimidae	b4	0	0.00 ± 0.00	0	74	0.46 ± 4.19	0.17
Cephalobidae*	b2	1,300	6.50 ± 23.85	1.74	313	1.95 ± 8.21	0.71
Prismatolaimidae	b3	0	0.00 ± 0.00	0	727	4.54 ± 18.86	1.65
Rhabditidae*	b1	3,467	17.33 ± 33.43	4.63	2,862	17.88 ± 36.43	6.49
Fungivores							
Aphelenchidae*	f2	1,884	9.42 ± 18.42	2.52	355	2.21 ± 11.78	0.80
Anguinidae	f2	0	0.00 ± 0.00	0	1,598	9.98 ± 43.63	3.62
Omnivores							
Dorylamida*	o4	10,726	53.63 ± 75.07	14.33	21,682	135.51 ± 165.79	49.15
Predators							
Mononchida*	p4	669	3.34 ± 11.12	0.89	2,853	17.83 ± 48.46	6.47
Plant-parasitic							
<i>Criconemella</i> *	h3	4,179	20.89 ± 56.37	5.58	1,319	8.24 ± 43.17	2.99
<i>Discocriconemella</i>	h3	0	0.00 ± 0.00	0	1,245	7.78 ± 42.73	2.82
<i>Helicotylenchus</i> *	h3	17,516	87.58 ± 160.35	23.41	2,834	17.71 ± 43.09	6.42
<i>Hemicyclophora</i> *	h3	11	0.05 ± 0.77	0.01	16	0.10 ± 1.26	0.04
<i>Hoplolaimus</i> *	h3	2,270	11.35 ± 41.29	3.03	178	1.11 ± 6.31	0.40
<i>Meloidogyne</i> *	h3	936	4.68 ± 14.66	1.25	1,647	10.29 ± 38.63	3.73
<i>Pratylenchus</i> *	h3	30,776	153.88 ± 236.94	41.13	2,279	14.24 ± 43.09	5.17
<i>Paratrichodorus</i>	h4	18	0.09 ± 0.95	0.02	0	0.00 ± 0.00	0
<i>Paratylenchus</i>	h2	0	0.00 ± 0.00	0	47	0.29 ± 2.15	0.11
<i>Rotylenchus</i>	h3	0	0.00 ± 0.00	0	29	0.18 ± 2.29	0.07
Tylenchidae	h2	0	0.00 ± 0.00	0	2,973	18.58 ± 57.45	6.74
<i>Trichodorus</i>	h4	452	2.26 ± 6.00	0.60	0	0.00 ± 0.00	0
<i>Xiphinema</i> *	h5	629	3.14 ± 12.67	0.84	1,078	6.73 ± 22.77	2.44
Nematodes Total		74,833			44,109		

F = functional guilds; A (abundance) = sum of nematodes number in 200 samples from ten sugarcane areas and 160 samples from eight forest fragments (300 cm³ of soil in each sample), Means ± SD = Means number and standard deviation of nematodes per 300 cm³ of soil in each area, D (%) = Dominance of each taxa expressed as a percentage; *taxa common to all areas.

3.2 Effects of management practices on soil biological and physical properties

Compared to forest fragments, the soil bulk density tended to be higher ($p < 0.05$) in sugarcane areas and in two forest fragments at FC and FD. In contrast, the total porosity and C-CO₂ evolution rate were lower at those sites ($p < 0.05$).

Soil bulk density did not vary with depth, but differed significantly between areas with lower mean value in forest fragments (1.19 Mg.m⁻³) and higher mean value in sugarcane areas (1.42 Mg.m⁻³) ($p < 0.05$; Standard Error: 0.01). Similarly, Araújo et al. (2011) found bulk density values: 1.22 Mg.m⁻³ in forest soil and in pasture soil 1.34 Mg.m⁻³, in same soil type of this study. Soil total porosity also did not vary with depth and mean values were higher in forest fragments than in sugarcane areas, 57% and 46%, respectively ($p < 0.05$; Standard Error: 0.05). Thus, we observed that as bulk density increases, pore size decrease, corroborating Dörner et al. (2010).

A one-way analysis of similarity (ANOSIM) revealed significant differences between the nematode assemblages of the 18 study sites (Global R= 0.94; $p= 0.0001$). A non-metric multidimensional scaling (NMDS) highlighted three distinct groups (the sampling groupings were based on Sorensen clustering: (I) FC, FD, SU1, SU2, SE1, SE2, SE3, SE4, ST1, ST2, SS1, and SS2; (II) FE, FF, FG, and FH (III) FA and FB (Fig. 1). These groupings were also confirmed by the generally significant differences detected with the pair-wise of the ANOSIM test. ANOSIM showed that the difference in community composition in both group II and group III compared to the group I plots was significant (group I vs. group II, r = 0.9428, $p < 0.0006$; group I vs. group III, r = 0.9428, $p < 0.01$), whereas the community composition of group II and group III did not differ.

The differences among areas illustrated the effects of conversion of forest to agriculture on the structure and composition of nematode assemblages. Sugarcane areas were characterized by higher bulk density, lower total porosity, and presence of *Pratylenchus* and *Meloidogyne*. The grouping of the two forest fragments with the sugarcane plantations was attributed to presence of these nematodes genera, which may have been washed into those areas by drainage or human intervention. Moreover, these fragments are characterized by high bulk density, which can be result of the natural compaction, and may have favored the establishment these plant-parasitic nematodes.

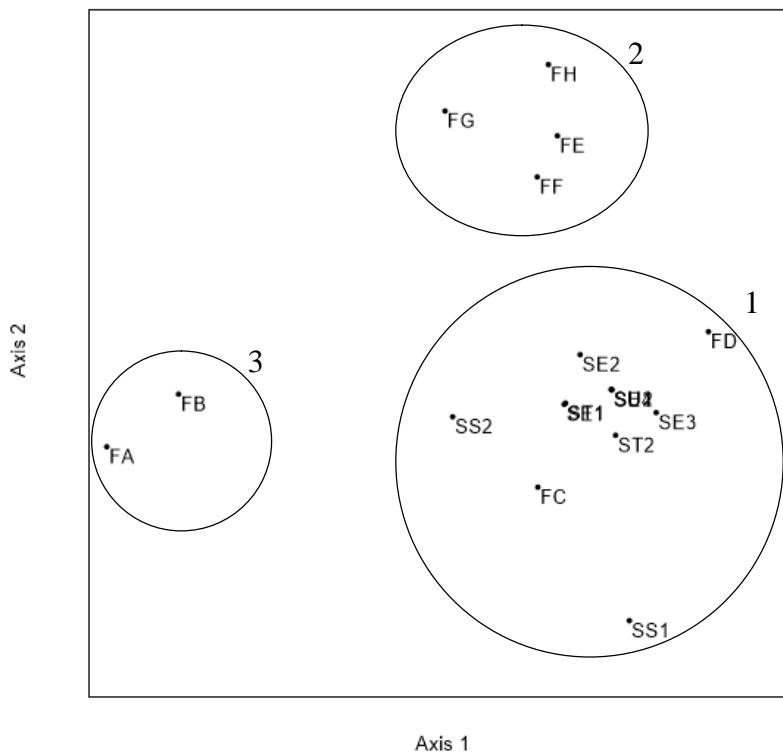


Figure 1 Non-Metric Multidimensional Scaling (NMDS) illustrating the divergence in community composition of soil nematodes between sugarcane soils (S) and in Atlantic Forest soils (F) in Pernambuco, Brazil. The plot was produced using Dice-Sorensen as a distance measure (Stress: 0.11).

Graphical representations reveal differences in the means and standard deviation of soil respiration at forest fragments and sugarcane areas, indicating differences in biological activity of the soils (Fig. 2).

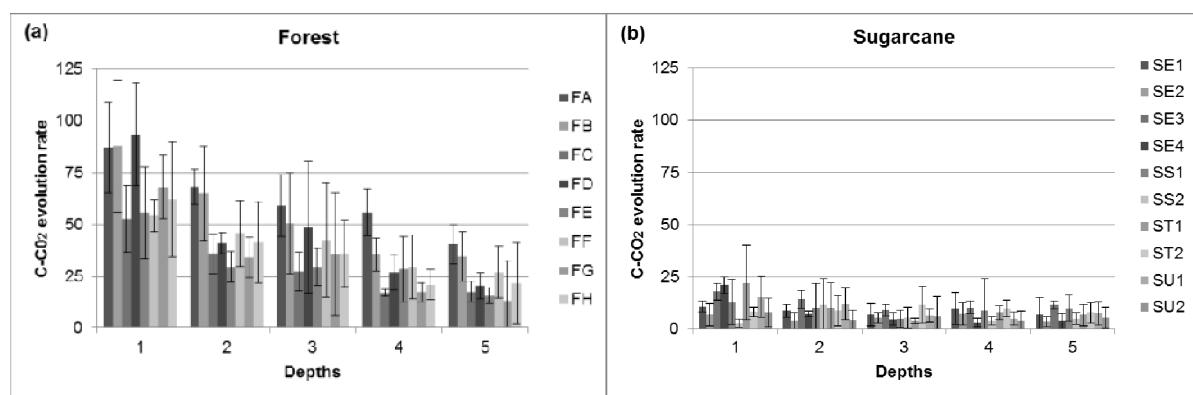


Figure 2 Soil respiration in forest fragments (a) and sugarcane areas (b). Bars represent the standard deviation. Depths: (1) 0-10-cm; (2) 10-20-cm; (3) 20-30- cm; (4) 30-40-cm; (5) 40-50-cm.

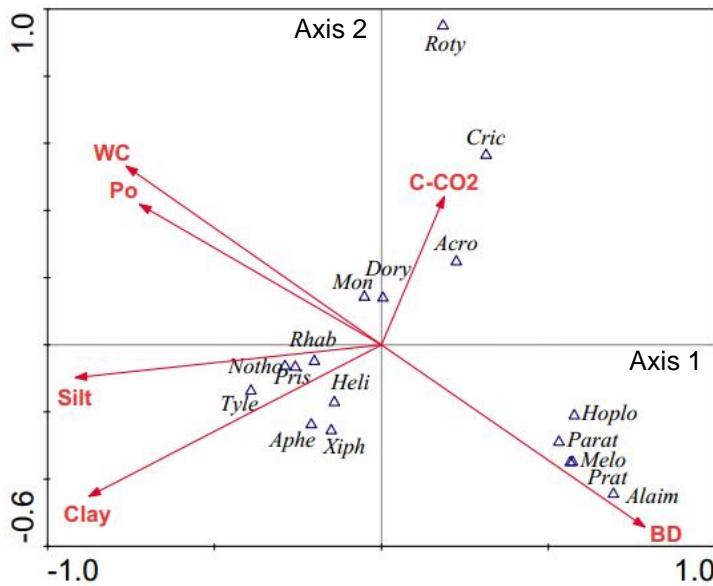
In forest soils, the C-CO₂ evolution rate decreased with depth (Fig. 2). Huang et al. (2012) reported similarly that soil microbial biomass carbon content and soil basal respiration decreased with increasing soil depth. However, the mean C-CO₂ rate was 42.01 mg in forest soils and 8.45 mg in sugarcane soils ($p < 0.05$; Standard Error: 1.04). In other words, the biological activity was higher in forest fragments soils than in sugarcane soils, corroborating observations of Stirling et al. (2010) in comparisons of pasture soils and sugarcane soils under conventional and non-conventional management.

We hypothesize that biological activity decreased under sugarcane soils due to limited rhizodeposition at greater depth and/or due to lower levels of soil carbon resulting either from reduced carbon input or increased carbon release. Grant and Rochette (1994) determined that optimum biological activity occurred between 60 and 70% of the soil total porosity, which corresponded to bulk densities between 1.06 and 0.8 Mg.m⁻³. In this study, these porosity levels occurred in the forest soils, which also had greater availability of decomposable organic substrates, with both factors supporting greater soil respiration.

We concur with other researchers that sugarcane management practices result in changes in bulk density and porosity, biological activity and nematode assemblages in sugarcane plantations compared with those in native forest (Bell et al. 2007; Stirling et al. 2010; Baquero et al. 2012). Moreover, conversion of forest soils to agricultural usage negatively impacts diverse soil quality parameters, reduces biodiversity and increases soil compaction (Rousseau et al. 2013; Gaji , 2013).

3.3 Relationships among soil physical properties, soil respiration, and composition of the nematode assemblage

Relationships between nematode assemblages and soil physical properties were examined by Canonical Correspondence Analysis (CCA) (Fig. 3). Soil physical properties (water content, total porosity, soil density, and sand and clay contents) and soil respiration (C-CO₂ evolution rate) are represented by straight lines. The CCA indicates eigenvalues: $\lambda_1 = 0.27$ ($p = 0.001$) and $\lambda_2 = 0.13$ ($p = 0.001$).



Po total porosity; *C-CO₂* soil respiration; *WC* water content; *BD* bulk density; *Melo* *Meloidogyne*; *Prat* *Pratylenchus*; *Cric* *Criconemella*; *Aphe* *Aphelenchidae*; *Noth* *Nothotylenchus*; *Pris* *Prismatolaimus*; *Acro* *Acrobeles*; *Hoplo* *Hoplolaimus*; *Rhab* *Rhabditida*; *Dory* *Dorylaimida*; *Heli* *Helicotylenchus*; *Mon* *Mononchida*; *Parat* *Paratylenchus*; *Roty* *Rotylenchus*; *Tyle* *Tylenchidae*; *Xiph* *Xiphinema*.

Figure 3 Ordination diagram of the nematode assemblages composition in Atlantic Forest soils by Canonical Correspondence Analysis (CCA).

Correlations between nematodes and environmental conditions were 0.78 on axis 1 and 0.64 on axis 2. The canonical axes explained 60% of the variation in the dataset. Thus, other factors, besides those measured, affect the structure and composition of nematode assemblages.

Plant-parasitic and fungivore nematodes were correlated with soil bulk density; Bacterivores, *Xiphinema* (plant-parasitic) and *Nothotylenchus* (fungivore) were related to soil sand content. *Prismatolaimus* (bacterivore), Tylenchidae (plant-parasitic), Dorylaimida (omnivore), and Mononchida (predator) were related to soil total porosity, biological activity and to forest soils. Most of these are large-bodied nematodes which need large pore spaces for their movement and access to food resources.

Ordination of nematode taxa by CCA suggests that *Pratylenchus*, *Hemicycliophora*, *Meloidogyne*, *Trichodorus*, *Helicotylenchus*, all plant-parasitic, and fungivores are correlated to sugarcane areas, which has soil characterized by higher bulk density, and finer soil particles, such as clay. Clay soils tend to have a higher total porosity than sandy soils. However, the relationship between texture and bulk density is tenuous and depends on a variety of factors such as organic matter content and depth in the soil profile. Therefore, in this study, soils with high clay content showed greater bulk density, and concomitant decrease

in the pore space favor the smaller-bodied nematodes, e.g. relevant plant-parasitic in sugarcane as *Pratylenchus* and *Meloidogyne*.

Similarly, Pattison et al. (2008) found that abundance of omnivore nematodes (e.g. Dorylaimida) was negatively correlated with increasing soil bulk density which suggests that in our study, unfavorable habitats may have occurred in soil with high soil bulk density. Activity and survival of large nematodes (e.g. family Dorylaimidae) may be restricted by smaller pores spaces which renders them sensitive to the changes in soil properties and are associated with intensive management (Goulart and Ferraz, 2003; Cardoso et al. 2012).

3.4 Effects of the depth in nematode assemblages

Relative abundances of plant-parasitic nematode and trophic groups decreased in depth in sugarcane soils, corroborating Caixeta et al. (2011) who investigated farming systems in the same geographic region as this study. Total nematode abundance also decreased with depths, similar to the observations of Biederman and Boutton (2010) that total nematode and family richness decreased with depth in forest clusters and grasslands. All these observations reflect the fact that important soil resources (organic carbon, microbial biomass, and root biomass) decline with increasing soil depth (Agnelli et al. 2004; Griffiths et al. 2006; Potthoff et al. 2006).

In forest soils, plant-parasitic nematodes were distributed almost uniformly across depths. This difference was probably associated with differences in root morphology and distribution; 65% the sugarcane root system is concentrated in the layer above 20-cm depth while in forest the root systems extend from the surface to 4-m (Otto et al. 2009; Stirling et al. 2011; Sternberg et al. 1998). Total predator nematodes, specialists (e.g. Mononchida) and generalists (e.g. Dorylaimida), were more abundant in the root zones and decreased in the deeper layers, in parallel with soil respiration and presumably with the presence of other organisms associated with the microbial biomass. Likewise, Cardoso el al. (2012) observed a negative correlation between depth and predator nematodes in forest soil.

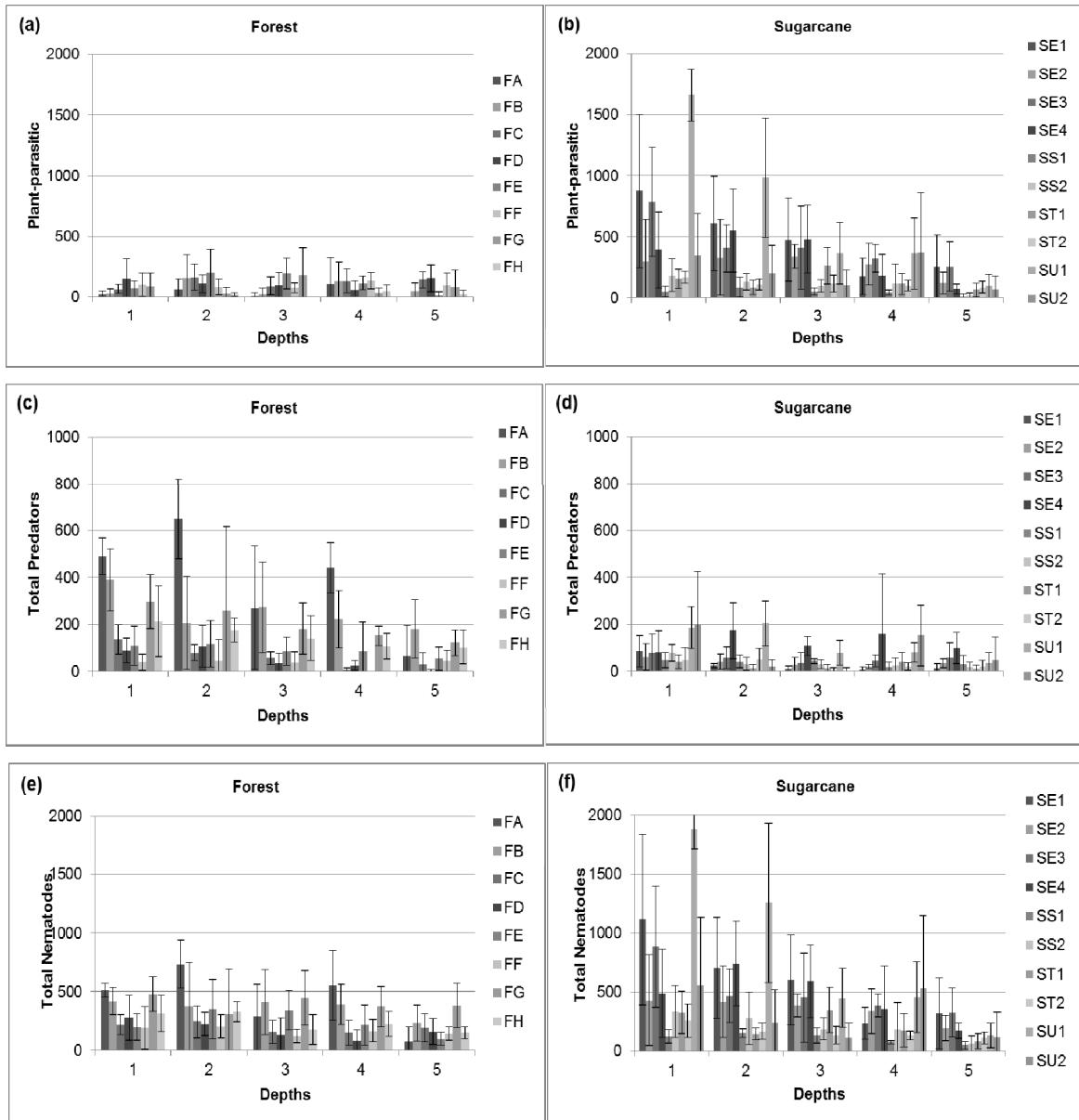


Figure 4 (a) plant-parasitic nematode in forest; (b) plant-parasitic nematode in sugarcane; (c) predator nematode forest; (d) predator nematode in sugarcane; (e) total nematodes forest; (f) total nematodes sugarcane. Depth: (1) 0-10-cm; (2) 10-20-cm; (3) 20-30- cm; (4) 30-40-cm; (5) 40-50-cm.

4. Conclusion

In summary, the present study shows difference between forest and sugarcane soils with regards to soil properties and the structure and composition of the nematode assemblages. Soil bulk density and abundances of all nematodes and of plant-parasitic nematodes tended to be higher in sugarcane soils. In contrast, the total porosity, soil respiration, predator nematodes were at lower levels in the sugarcane soils. We conclude that

sugarcane management practices result in changes in soil properties, and concomitantly alter the composition and structure of the nematode assemblages. Non-metric multidimensional scaling and Canonical correspondence analysis revealed differences in the nematode assemblage composition of study sites in relation to some of the measured parameters but also indicated the probable effects of other environmental factors on the nematofauna.

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Soil food web analyses and environmental condition: an assessment of the soil quality in sugarcane fields*

CAPÍTULO IV

Soil food web analyses and environmental condition: an assessment of the soil quality in sugarcane fields

SHORT COMMUNICATION

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Abstract

Understanding about occurrence, relevance and ecology of plant-parasitic nematodes in sugarcane fields is essential to its effective management. Soil food web indices based on the abundances of nematode functional guilds have been utilized to study the effect of pollution, management, and vegetation on agroecosystems. Thus, this study had as objective evaluating the condition of the soil food webs through faunal analysis and metabolic footprints. Soil samples were collected between 2010 and 2011 from four farms (Estreliana, União e Indústria, Trapiche, and Salgado) located in sugarcane zone of Pernambuco State, Brazil. At each site, soil samples were collected at five 10-cm depth increments from the four corners of a 10 x 10 m square. According to faunal analysis the studied sugarcane soils have complex and stable soil food webs, with higher connectance (dominated by larger species with longer life cycles); and predominant decomposition channel bacterial-mediated. In addition, Maturity Index and Modified Maturity Index indicated an intermediate stage of succession in the areas. Metabolic footprints indicated that direct bacterivore and fungivore activity were not consistent or important channels of resources into the soil food web in sugarcane soils; most resource assimilation was via the herbivore and predator channels.

Keywords: faunal analyses, metabolic footprints, decomposition channel, nematode, bioindicators.

Soils are complex and heterogeneous habitats for many animals. Trophic interactions between soil animals are depicted in soil food webs which form an essential component of terrestrial ecosystems. Soil food web contains wide populations of innumerable species and these populations are continually interacting with each other. These interactions become more complex as the diversity within the soil food web increases (Stirling et al., 2011).

However, food webs are dynamic energy flow systems without a fixed structure over time and both bacterial and fungal energy channels operate simultaneously in most soil food webs with temporal and spatial variation (Hassall et al., 2006). The dominance of fungal or bacterial energy channels are often determined by the quality and quantity of soil organic matter (Holtkamp et al., 2008). In addition, the soil food web can be measured by counting organism groups (bacteria, protozoa, arthropod, etc.) or subgroups (bacterivores, fungivores and predator nematodes) are counted and through calculations, can be converted to biomass, measuring activity levels (the amount of by-products, i.e., respiration, nitrification and decomposition rates) or measuring cellular constituents (biomass carbon, nitrogen, phosphorus, or enzymes).

Soil food web indices based on the abundances of nematode functional guilds (c-p scale integrating with food sources) have been utilized to study the effect of pollution, management, and vegetation on agroecosystems (Liang et al., 2005; Stirling & Logde, 2005; Wang et al., 2006). The knowledge about distribution of nematode assemblages and their interactions in the soil food web are scarce in Brazil, but necessary. Moreover, understanding about occurrence, relevance and ecology of nematodes in sugarcane fields is essential mainly to effective management of plant-parasitic nematode.

Hence nematode assemblages were investigated in sugarcane fields located in sugarcane zone of Pernambuco State, Brazil. We hypothesized that sugarcane soils have food webs enriched, but unstructured, and are conducive to bacterial decomposition. Thus, this study had as objective evaluating the condition of the soil food webs through faunal analysis and metabolic footprints.

This research was carried out in ten sugarcane areas. Soil samples were collected between 2010 and 2011 from four farms (Estreliana, União e Indústria, Trapiche and Salgado). At each site, soil samples were collected at five 10-cm depth increments from the four corners of a 10 x 10 m square. About 600g of soil were collected at each depth for nematode analyses and packaged in labeled plastic bags and immediately transported to the laboratory.

Nematodes were extracted by the sucrose centrifugation (Jenkins, 1964). The suspensions were stored in a refrigerator for no more than 3 days before counting and identification. Nematodes were counted at x20 magnification in two replicate aliquots on Peters glass counting slides (1 ml of capacity). All nematodes counted were identified to genus/family level at x40 and x100 magnification. For identification to the genus level, temporary slides were prepared. Nematode abundance was expressed per 300 cm³ of soil.

Nematodes were assigned to five trophic groups according to feeding habits: plant-parasitic, bacterivores, fungivores, predators, and omnivores (Yeates et al., 1993). Even though the trophic habits of the Tylenchidae are uncertain, all individuals in this family were considered plant-parasitic. Nematodes also were classified in functional guilds based on feeding habits and five colonizer-persister (c-p) groups which represent life history characteristics and sensitivity to environmental perturbation (Bongers, 1990; Bongers & Bongers, 1998). Plant-parasitic nematodes were identified to genus level using keys and

descriptions of Mai et al., (1996) and free-living nematodes to the family level according to keys of Tarjan et al., (1977).

Faunal profiles were constructed to indicate whether the soil communities are basal, enriched, or structured and stable. Indices of ecosystem condition: Enrichment (EI), Structure (SI), Basal (BI), and Channel (CI) were calculated following Ferris et al., (2001): enrichment index (EI) was calculated as $EI = 100 e / (b + e)$, structure index (SI) was calculated as $SI = 100s/(b + s)$, where $s = 1.8 \times (Ba_3 + Fu_3 + OP_3) + 3.2 \times (Ba_4 + Fu_4 + OP_4) + 5 \times (Ba_4 + Fu_5 + OP_5)$, $b = 0.8 \times (Ba_2 + Fu_2)$, $e = 3.2 \times (Ba_1) + 0.8 \times (Fu_2)$. The numbers from 1 to 5 represented the c-p value.

BI is an indicator of the prevalence of the general opportunistic nematodes that are tolerant of soil perturbation; CI provides a quantitative measure of the component flow of C and energy through the fungal and bacterial decomposition channels; EI is an indicator of enrichment opportunistic nematodes, both fungal and bacterial feeders, which respond rapidly to increase in food resources; and SI indicates soil food web length and food web resilience (Ferris et al., 2001).

In order to measure the magnitude of ecosystem functions and services provided by component organisms of the soil food web, the nematode metabolic footprints (NMF) was also calculated. NMF calculations are based on biomass and metabolic activity of components of the nematode assemblages calculated from published dimensions of each species and averaged across species for genera and families (Ferris, 2010). The ecophysiological attributes of nematodes, assembled at species, genus and family levels were obtained from Nemaplex (<http://plpnemweb.ucdavis.edu/nemaplex/>). The Maturity Index (MI) was calculated as the weighted mean of the cp values of the free-living nematodes (Bongers, 1990; Bongers & Bongers, 1998). MI was applied in order to evaluate the functional responses of soil nematodes to resource and environmental change.

Ecological indices were significantly different among sugarcane areas, but there was no significative difference among depths ($p < 0.05$) (data not shown).

The faunal indices from all sugarcane areas are depicted in Figure 1. BI and CI indices showed low values in all areas. In most sugarcane areas the EI values were low (<60), higher values were shown in three areas (ST1, SE1, and SE2). SI values were moderate in most areas, and SI values were high in four sugarcane areas (SE2, SE3, SE4, and SU1).

Maturity index (MI) shown highest values (ranging 3.2 to 3.6) in only three sugarcane areas (SE3, SE4, and SU1). No differences were observed in modified MI (MI) among areas ($p < 0.05$) (Fig. 2). Plant Parasitic Index also was no shown significantly different among sugarcane areas, except for SS1 and SU2 that obtained lowest values (Fig. 2, $p < 0.05$). As illustrated in Figure 3, the metabolic footprint assessments of the food web (Ferris, 2010) differed among areas ($p < 0.05$).

The herbivore footprint did not differ among areas (Fig. 3, $p < 0.05$). The NMF values of herbivore nematodes were high in most sugarcane areas. Conversely, the NMF values of fungivore nematodes in all sugarcane areas were low and that of bacterivores also was low in most areas (Fig. 3). The predator footprint was low in most areas and high in SE4, SU1, and SU2, likewise the structure footprint, reflecting the metabolic activity of higher trophic level nematodes, which include predators of opportunistic species. The enrichment footprints were correlated with bacterivore footprint, which represented the predominantly bacterivore resource available to predators. The fungivore footprint has lower values than other metabolic footprints.

In contrast to our expectation, all sugarcane areas showed lower values of BI, characterizing a nematode assemblage with low levels of perturbation-resistant nematodes (Fig. 1). Generally, the nematode assemblages from annual croplands have higher values reflecting greater disturbance (Culman et al., 2010). Possibly, these areas are characterized by

lower nutrients inputs and organic matter turnover, consequently reducing the bacterivore nematode abundance (Okada & Harada, 2007).

Low values of CI suggests that decomposition pathways in the detrital food web were predominantly mediated by bacterial (Fig. 1), corroborating Stirling et al., (2011) who also investigated nematode assemblage in three sugarcane crops in Australia. Ruess & Ferris, (2004) asserted that decomposition pathways of agricultural systems are predominantly bacterial, which provides rapid transfer of nutrients to plants.

In most sugarcane areas the EI values were low (<60) (Fig. 1). Higher EI values were occurred in three areas (ST1, SE1, and SE2) and suggest an enriched community structure. In general agricultural soil food webs are characterized by higher EI values relative to unmanaged natural areas (Ferris et al., 2001; Ferris & Bongers, 2006; Leroy et al., 2009).

Usually agricultural systems are characterized by low SI values (van Diepeningen et al., 2006; Sánchez-Moreno et al., 2009). However, in our studies, high SI values were found in four sugarcane areas (SE2, SE3, SE4, and SU1) (Fig. 1), similar to observations of Mondino et al., (2009) in pasture, bean and arrowroot crops. Higher SI values suggest a complex community structure with many linkages in the food web and potentially suppressive to opportunistic species due to higher proportions of omnivores and predators (Ferris et al., 2001). Six areas showed medium values, denoting soil food webs more simplified due to reductions in abundance of omnivores and predators nematodes.

The sensitivity of the SI is determined mainly by the omnivore and predator nematodes, which require more time to establish than fungivore and bacterivore nematodes (Liang et al., 2009). Moreover, omnivore and predator nematodes are susceptible to disturbance and can reestablish when the intensity of management is reduced (Briar et al., 2007).

MI, likewise SI, indicated lower disturbance in these areas (Fig. 2). The higher MI values result from the presence of larger nematodes with longer life-cycle; these nematodes are favored by soil stability and have high cp value, characterized as k-strategists (Bongers, 1990; Bongers & Bongers, 1998). Thus, both MI and SI indicated a low disturbance in the areas SE3, SE4, and SU1. Concerning to PPI, this index showed inverse behavior to MI, agreeing with Bongers et al. (1997).

The sugarcane areas which were characterized by low disturbance are located in the floodplain, where there is greater accumulation of sediment and water content. Additionally, the harvest in these areas is performed manually. Thus, these characteristics of the soil can be positively influenced nematode assemblages, which are sensitive to disturbances.

According to faunal analysis the studied sugarcane soils have complex and stable soil food webs, with higher connectance (dominated by larger species with longer life cycles); and predominant decomposition channel bacterial-mediated. In addition, MI and $\hat{U}MI$ indicated an intermediate stage of succession in the areas.

Based on the much smaller sizes of the bacterivore and fungivore than the herbivore and predator metabolic footprints, we conclude that direct bacterivore and fungivore activity were not consistent or important channels of resources into the soil food web in sugarcane soils; most resource assimilation was via the herbivore and predator channels.

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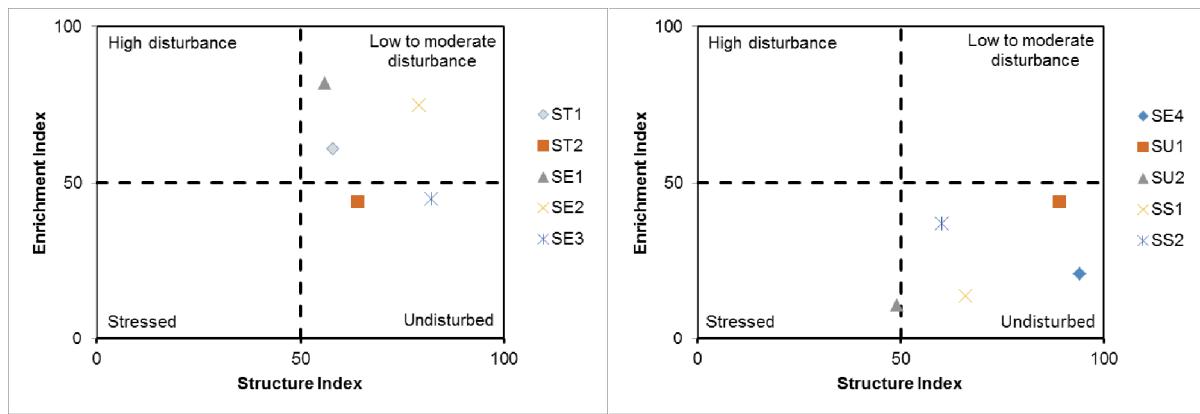


Figure 1. Faunal analysis of nematodes from 10 sugarcane areas in different farms. ST1 and ST2: Trapiche Farm; SE1, SE2, SE3, and SE4: Estreiana Farm; SU1 and SU2: União & Indústria Farm; and SS1 and SS2: Salgado Farm.

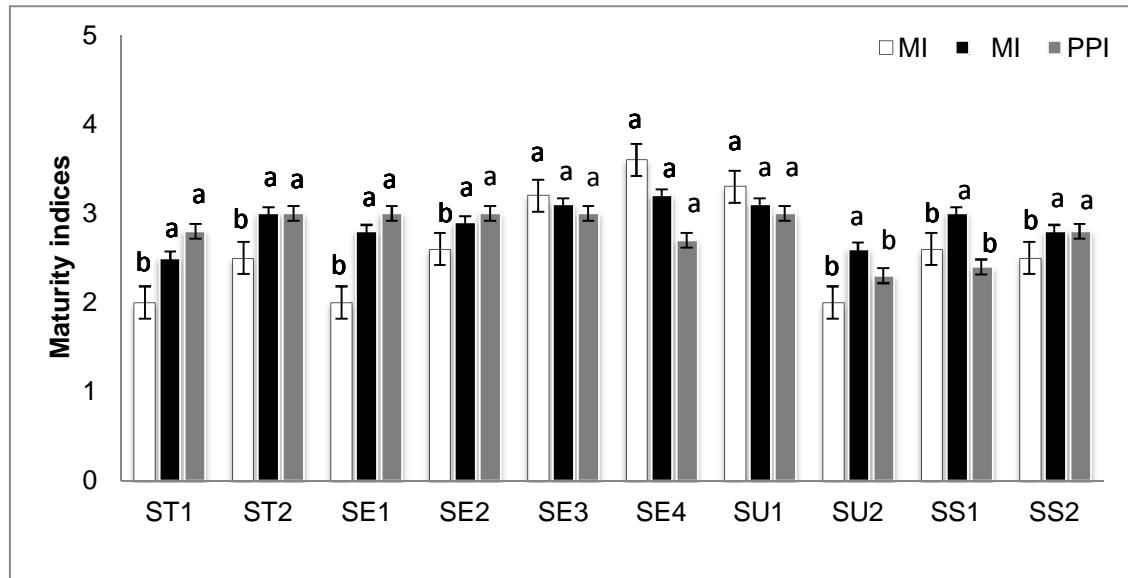


Figure 2. Maturity indices including MI Maturity index, MI Modified Maturity index, and PPI Plant-parasitic index in different sugarcane areas. Error bars represent standard error and are followed by a lowercase letter indicating a significant difference according to Scott-Knott test ($p < 0.05$). ST1 and ST2: Trapiche Farm; SE1, SE2, SE3, and SE4: Estreiana Farm; SU1 and SU2: União & Indústria Farm; and SS1 and SS2: Salgado Farm.

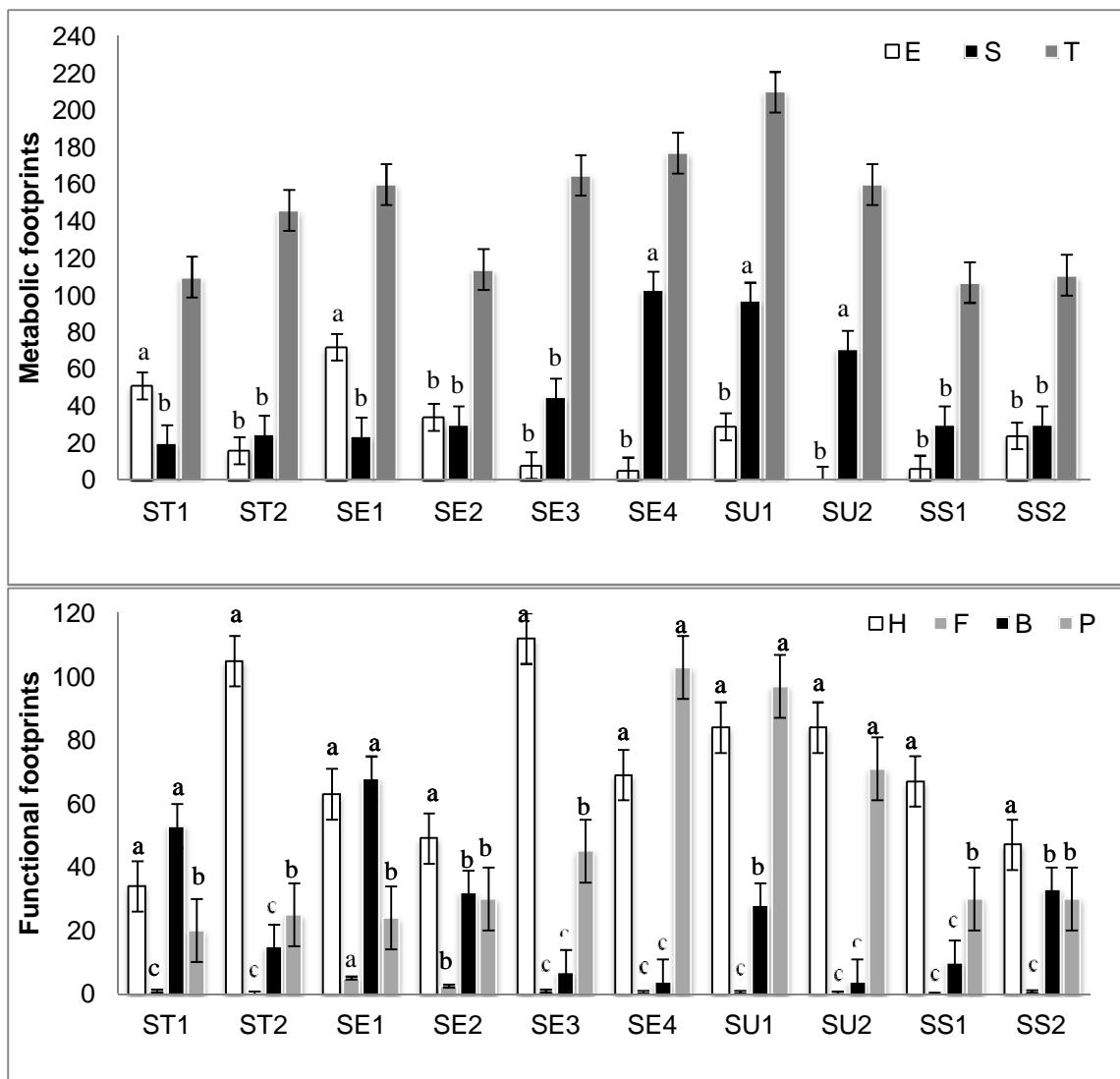


Figure 3. Nematode Metabolic Footprint, including E enrichment footprint, S structure footprint, T total footprint, H herbivore footprint, F fungivore footprint, B bacterivores footprint, and P predator footprint in different sugarcane areas. Error bars represent standard error and are followed by a lowercase letter indicating a significant difference according to Scott-Knott test ($p < 0.05$). ST1 and ST2: Trapiche Farm; SE1, SE2, SE3, and SE4: Estreiana Farm; SU1 and SU2: União & Indústria Farm; and SS1 and SS2: Salgado Farm.

*Spatiotemporal distribution of nematodes and soil food web interactions in
Atlantic Forest**

CAPÍTULO V

Spatiotemporal distribution of nematodes and soil food web interactions in Atlantic Forest

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Highlights

- Structure and composition of the nematode assemblages were quite different between areas and across time.
- The scale of sampling was too great for detection of spatial distribution patterns of the nematodes; alternatively, their aggregated spatial patterns were described by a Taylor's power law model.
- The soils of the studied forests have complex soil food webs, stable with high connectance and decomposition channels dominated by bacteria.
- The predator-prey relationships were not affected by soil properties that fluctuate with time.

Abstract

Studies of nematode assemblages in Atlantic Forest are scarce. They could contribute to better understanding of the occurrence, relevance and ecology of plant-parasitic and other soil nematodes. Nematode assemblages and environmental parameters (organic matter, water content, bulk density, total porosity, C-CO₂ evolution rate, soil texture) were investigated in two seasons (rainy and dry) in two forest areas of the Zona da Mata de Pernambuco. The aim of our research was: to describe the structure and composition of the nematode assemblages, observing their spatiotemporal distribution and relationships with soil attributes; to evaluate the condition of the soil food web, through faunal analysis and metabolic footprints; and to study predator-prey interactions. Structure and composition of the nematode assemblages were quite different between areas and across time. The scale of sampling was too great for detection of spatial distribution patterns of the nematodes; alternatively, their aggregated spatial patterns of the nematodes were described by a Taylor's power law model, with b-values ranged from 1.80 to 2.41. The forest soils have complex, stable soil food webs with high connectance and decomposition channels dominated by bacteria. Maturity indices did not differ between areas and time, and were >2, indicating an intermediate stage of succession. The predator-prey relationships were not affected by changes in soil properties that fluctuate with time.

Keywords: soil food web, metabolic footprints, ecological indices, predator-prey, soil physical properties.

1 Introduction

Spatial variability is a key component of the structure and function of soil biodiversity. Previous studies have shown that soil organisms are usually not randomly distributed but exhibit spatially aggregated patterns over scales ranging from square millimeters to hectares (Ettema and Wardle, 2002). Patterns of dispersion of the soil biological, physical and chemical attributes may vary in space (randomized, uniform and aggregated) and/or time, e.g. soil nitrogen (Yang et al. 2013); soil microorganisms (Wu et al. 2006) and soil water content (Vieira et al. 2010). Of the variables studied, nematodes have a natural tendency to develop in aggregates or patches due to their reproductive characteristics, behavioral activity and food availability (Ferris, 1985; McSorley, 1987), e.g. bacterivore nematodes which concentrate close to bacteria communities and plant-parasitic nematodes around roots (Blanc et al. 2006).

In order to describe the spatial distribution of soil nematodes, geostatistical analyses have been applied (Liang et al. 2005; Shi et al. 2008; Dinardo-Miranda and Fracasso, 2010; Cardoso et al. 2012). However, in such studies, the aggregated pattern of nematode dispersion has been considered to be a pure nugget effect (Dinardo-Miranda and Fracasso, 2010; Cardoso et al. 2012). As an alternative, the aggregated spatial patterns of nematodes may be assessed Taylor's power law model (Taylor, 1961; Ghaderi et al. 2012; Park et al. 2013).

Few studies of the spatial distribution of nematodes and soil physical properties have been conducted in the sugarcane zone of Pernambuco, but should be relevant to the development of sampling plans that will allow implementation of integrated pest management programs in the region. Considering that reservoirs and refugia of soil nematodes exist in remnant native vegetation areas, the investigation of nematode assemblages in such natural ecosystems would contribute to better understanding of the occurrence, relevance and ecology of soil nematodes in both natural ecosystems and agroecosystems.

Soil properties influence biological activity and play a critical role in the functioning of soil food webs (Jordan et al. 2003); however, their specific effects on functioning and nematode diversity remain relevant questions. In this study, some soil properties were selected to infer soil functioning and to relate it to soil nematode diversity: organic matter, water content, bulk density, total porosity, C-CO₂ evolution rate, and soil texture.

On the other hand, soil food web indices, based on the abundances of nematode functional guilds (c-p scale integrating with food sources) have been utilized to examine the effect of pollution, management, and vegetation on agroecosystems (Liang et al. 2005; Stirling and Logde, 2005; Wang et al. 2006; Culman et al. 2010). Thus, faunal analysis and

metabolic footprints analysis were applied in this survey in order to profile the soil condition in the studied forest areas based on soil nematodes. In addition, the predator-prey interactions also were investigated since understanding of these relationships can be explored more thoroughly in undisturbed systems.

We hypothesized that: (a) nematodes taxa have spatiotemporal distribution characteristics that are influenced by soil attributes; (b) forest soils have food webs with high structure and low enrichment, and are conducive to fungal decomposition; and (c) low prey-abundance is relate to high predator abundance. Thus, this study had following objectives: (a) to describe the structure and composition of the nematode assemblages, observing their spatiotemporal distribution and relationships with soil attributes; (b) to evaluate the condition of the soil food webs through faunal analysis and metabolic footprints; c) to evaluate predator-prey interactions.

2 Material and Methods

2.1 Study sites

This research was carried out in two forest areas (Pau Amarelo and Camucim), located in Zona da Mata Norte of Pernambuco State, Brazil (Fig. 1). Soil samples were collected in 2012 in the rainy and dry seasons. In Pau Amarelo forest, the rainy and dry seasons were designated F1R and F1D, respectively. Likewise, in Camucim forest, the rainy and dry seasons were denominated F2R and F2D, respectively. The local climate is Asø (humid tropical with hot summers), according to the Köppen Climate Classification (Köppen, 1948), with high and low temperatures happen in December and July, respectively.

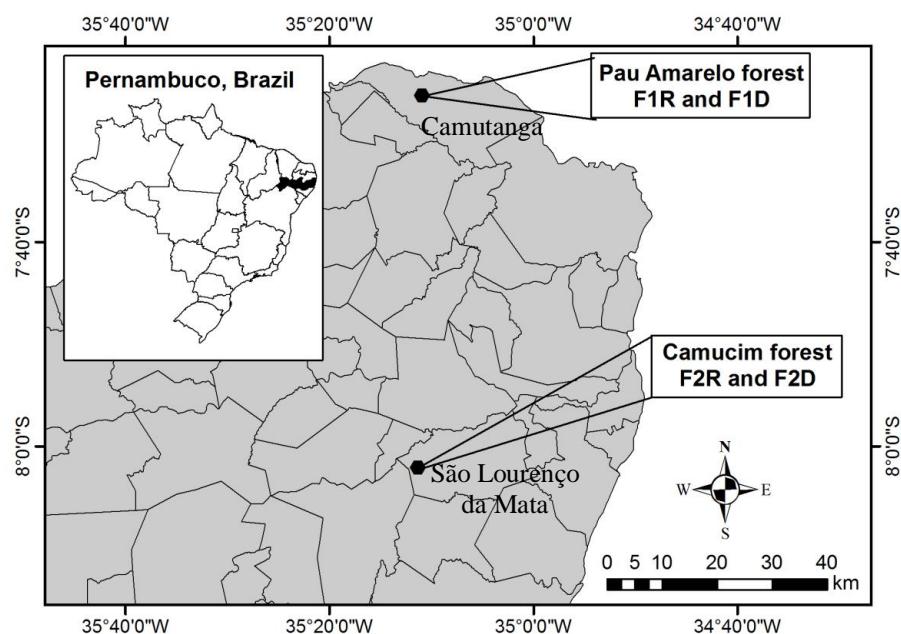


Figure 1 Map of areas localization including the forest areas sampled in rainy and dry seasons.

2.2 Soil sampling

Two transects were established in the form of a cross at each site. Each was 200 m long with sampling points spaced 10-m apart, that there were forty-two sampling points at each site. Soil samples were taken at 20-30-cm depth by a modified TORSOL® sampler for assessment of bulk density, water content, and soil respiration and stored in parafilm-sealed plastic containers. About 600g of soil was collected for nematode, organic carbon and texture

analyses. The samples were packaged in labeled plastic bags and immediately transported to the Phytonematology Laboratory, Agronomy Department, UFRPE.

2.3 Nematode analyses

2.3.1 Nematode extraction

Nematodes were extracted by the sucrose centrifugation method (Jenkins, 1964). The suspensions were stored in a refrigerator for no more than three days before counting and identification. Nematodes were counted at x20 magnification in two replicate aliquots on Peters glass counting slides (1 ml capacity). Temporary slides were prepared and all nematodes counted were identified to genus/family level at x40 and x100 magnification. Nematode abundance was expressed per 300 cm³ of soil.

2.3.2 Trophic and functional structure

Nematodes were assigned to five trophic groups according to feeding habits (plant parasites, bacterivores, fungivores, predators, and omnivores), based on the morphology of the stoma and esophagus (Yeates et al. 1993). Although the trophic habits of the Tylenchidae are uncertain, all individuals in this family were considered plant-parasitic. It is common to classify the order Dorylaimida as omnivores, but in this research we also considered them to be generalist predators as well as Mononchida as specialist predators. Plant-parasitic nematodes were identified to genus level using keys and descriptions of Mai et al. (1996) and free-living nematodes to the family level according to keys of Tarjan et al. (1977).

Nematodes also were classified into functional guilds based on feeding habits and the five colonizer-persister (c-p) groups, which represent life history characteristics and sensitivity to environmental perturbation (Bongers, 1990; Bongers and Bongers, 1998). In order to measure environmental disturbance, the Maturity Index (MI) was calculated as the weighted mean of the individual c-p values ($MI = \sum v_i \cdot f_i$, where v_i is the c-p value of i-taxon, f_i is the frequency of i-taxon). C-p values ranging from 1 for colonizers (r-strategists) to 5 for persisters (K-strategists) are assigned to nematode families to illustrate their life strategies and, thus, conditions of the surrounding environment. \bar{UMI} and Plant Parasite Index (PPI) also were determined similarly for all nematodes and for plant-parasitic genera, respectively (Bongers, 1990).

2.3.3 Environmental quality and soil food web structure

Faunal profiles were constructed to indicate whether the soil communities in forest areas are basal, enriched, or structured and stable. Indices of ecosystem condition: Enrichment (EI), Structure (SI), Basal (BI), and Channel (CI) were calculated following Ferris et al. (2001): enrichment index (EI) was calculated as $EI = 100e / (b + e)$, structure index (SI) was calculated as $SI = 100s / (b + s)$, where $s = 1.8 \times (Ba_3 + Fu_3 + OP_3) + 3.2 \times (Ba_4 + Fu_4 + OP_4) + 5 \times (Ba_5 + Fu_5 + OP_5)$, $b = 0.8 \times (Ba_2 + Fu_2)$, $e = 3.2 \times (Ba_1) + 0.8 \times (Fu_2)$. The numbers from 1 to 5 represented the c-p value.

In order to measure the magnitude of ecosystem functions and services provided by component organisms of the soil food web, the nematode metabolic footprints (NMF) was also calculated. NMF calculations are based on biomass and metabolic activity of components of the nematode assemblages calculated from published dimensions of each species and averaged across species for genera and families (Ferris, 2010). The ecophysiological attributes of nematodes, assembled at species, genus and family levels were obtained from Nemaplex (<http://plpnemweb.ucdavis.edu/nemaplex/>).

The connectance among predators and their prey was calculated as the total sum of all possible interactions among them, assuming that all taxa in each channel may interact (in this case any organism in the resource group may be eaten by any organism in the consumer group) (Sánchez-Moreno et al. 2011).

2.4 Soil respiration

Microbial activity was estimated from soil respiration as indicated by the C-CO₂ evolution rate (Grisi, 1978). Soil samples (100 g) and container with 10 ml 0.5N KOH were placed into sealed glass chambers and incubated at 25 ± 2°C for 15 days. The CO₂ absorbed by the KOH was determined by titration with HCL 0.1 N, using phenolphthalein and methyl orange as indicators.

2.5 Soil chemical analysis

Organic matter was determined indirectly by organic carbon content, according to methodology of Yeomans and Bremner (1988), which is based on the oxidative action of potassium dichromate. Then, the excess of potassium dichromate resulting from the oxidation of organic matter was titrated with ammonium ferrous sulfate solution, using ferroin as

indicator. This analysis was carried out at the Laboratory of Soil Chemistry, Department of Agronomy, UFRPE.

2.6 Soil physical analyses

Soil physical analyses were performed at the Laboratory of Soil Mechanics and Waste Utilization, Department of Agricultural Engineer, UFRPE, using methods detailed in EMBRAPA (1997).

- i. Soil texture was determined by the hydrometer method using sodium hydroxide as dispersant.
- ii. Soil bulk density (BD) was determined for intact soil cores 5 cm in diameter, 2.5 cm in length, and 50 cm³ in volume. To determine water content (WC), soil samples were dried at 105-110°C for 24 hours and weighed before and after to determine their weight loss. BD was estimated by division between dry weight (DW) and core volume ($BD = DW/50 \text{ cm}^3$).
- iii. Soil particle density (PD) was determined in a 50-mL volumetric flask using 20 g of air-dried soil and alcohol as fluid to determine the volume occupied by the particles ($PD = 20g/(50 \text{ ml ó alcohol volume})$).
- iv. Total porosity (Po) calculated from the values of PD and BD ($Po = (1 - (BD/PD)) * 100$).

2.7 Statistical analyses of data

Two-way ANOVA was applied to all datasets in order to assess the significance of differences between areas and sampling seasons. Tukey's multiple comparison test was used when significant differences were detected ($p < 0.05$).

Graphics with means values and errors bars were developed to visualize distribution of data in each area over time. Pearson's correlation coefficients between soil attributes and nematodes taxa were also determined. Classical statistical analyses and analysis of variance (ANOVA) were performed in Statistica 7.0 (Statsoft Inc. 2004).

In order to describe the spatial distribution of the nematodes and soil properties, geostatistical analysis was applied to the datasets using GS+ version 7 (Gamma Design Software, 2004). In the analyses, the classical variogram estimator (Journel and Huijbregts,

1978) was adopted, where σ^2 is the value of estimated semivariance of the experimental data and $N(h)$ is the number of observations pairs $Z(x_i)$ and $Z(x_i+h)$, separated by lag distance h .

$$\sigma^2(h) = \frac{1}{2 N(h)} \sum_{i=1}^{N(h)} [Z(x_i) - Z(x_i + h)]^2$$

Taylor's power law model (Taylor, 1961) was also applied to the data in order to describe the spatial distribution of nematodes. The power law is the observed variance of count data (S^2) expressed as a power function of the observed mean (Y) of the data ($S^2 = a^*Y^b$). This model has two parameters: a^* and b which are descriptive of the power relationship between variance and mean, such that b is considered an aggregation parameter and a^* is a scaling factor (Ferris, 1985; Allsopp, 1990; Ghaderi et al. 2012; Park et al. 2013). Thus, when $b > 1$ the organism has an aggregated pattern of dispersion, when $b = 1$, the dispersion is uniform and $b < 1$ the pattern of dispersion is random (Elliot, 1977). The b dispersion index was estimated by regression.

Rarefaction curves were fitted with 95% confidence and 1,000 randomizations in order to determine the sampling intensity necessary to detect taxa richness in the studied forest areas. Mao Tau and the first-order jackknife (SJack1) were chosen as estimators, Mao Tau is the species richness observed and SJack1 is a statistical technique for reducing the bias of an estimator by removing subsets of the data and recalculating the estimator with the reduced sample (Colwell, 2009). Rarefaction curve was determined by EstimateS version 8.2.0 available in <http://purl.oclc.org/estimates> (Colwell, 2009).

3 Results and Discussion

3.1 Structure and composition of nematode assemblages in rainy and dry seasons

Thirty-five nematode taxa were identified; twenty-four were common to both areas (Tab. 1). *Bunonema*, *Cryptonchus*, *Diploscapter*, *Wilsonema*, and *Hoplolaimus* were detected only in F1. *Isolaimium*, *Ditylenchus*, *Prodorylaimus*, *Seinura*, *Paratrichodorus*, *Trichodorus* and *Rotylenchus* were present only in F2.

Among the trophic groups, nematodes of the order Rhabditida were the most abundant of the bacterivores in both areas and *Aphelenchoides* and *Nothotylenchus* was most abundant among fungivores. Of the plant-parasitic nematodes Tylenchidae, Criconematidae (*Criconemoides*) and Longidoridae (*Xiphinema*) were dominant in F1 while Pratylenchidae

(*Pratylenchus*), Hoplolaimidae (*Helicotylenchus*) and Criconematidae (*Criconemella*) were dominant in F2. In F1R, the bacterivore nematodes were approximately 53% of the total nematode abundance. Omnivores predominated in F1D and F2R with 48% and 30% of total nematode abundance, respectively. Plant-parasitic nematodes represented 38% of total nematode abundance in F2D. The abundance, trophic diversity and taxa richness were significantly higher ($p < 0.05$) in the rainy season than in the dry season (Tab. 1). Therefore, the structure and composition of the nematode assemblages were quite different between areas and time.

Table 1 Abundance, average and dominance of the nematode assemblages in two Atlantic Forest, Zona da Mata Norte of Pernambuco.

Nematodes		Forest 1 (rainy)				Forest 1 (dry)				Forest 2 (rainy)				Forest 2 (dry)			
Trophic groups	Family	G	A	Means ± SD	D (%)	A	Means ± SD	D (%)	A	Means ± SD	D(%)	A	Means ± SD	D(%)	A	Means ± SD	D(%)
Bacterivores																	
<i>Acrobelus</i>	Cephalobidae	b2	606	14.42 ± 4.27	3.17	139	3.31 ± 1.89	2.35	32	16 ± 0.22	0.28	19	0.44 ± 0.35	0.33			
<i>Bunonema</i>	Bunonematidae	b1	0	0.00 ± 0.00	0.00	45	1.07 ± 1.08	0.76	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00			
<i>Cryptonchus</i>	Bathyodontidae	b4	87	2.07 ± 2.09	0.46	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00			
<i>Diploscapter</i>	Diploscapteridae	b1	1,277	30.40 ± 20.51	6.70	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00			
<i>Isolaimium</i>	Isolaimidae	b3	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	225	34.14 ± 3.02	1.99	0	0.00 ± 0.00	0.00			
<i>Prismatolaimus</i>	Prismatolaimidae	b3	2,201	52.40 ± 10.23	11.54	391	9.31 ± 4.13	6.60	345	24.64 ± 2.57	3.05	307	7.29 ± 1.73	5.28			
Rhabditida	Rhabditida	b1	5,945	141.54 ± 22.74	31.19	836	19.90 ± 6.58	14.12	2,578	80.56 ± 10.43	22.76	1,448	34.47 ± 7.57	24.92			
<i>Wilsonema</i>	Plectidae	b2	29	0.69 ± 0.69	0.14	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00			
Fungivores																	
<i>Aphelenchus</i>	Aphelenchidae	f2	58	1.38 ± 1.39	0.30	0	0.00 ± 0.00	0.00	157	39.25 ± 3.40	1.39	29	0.69 ± 0.42	0.49			
<i>Aphelenchoïdes</i>	Aphelenchoididae	f2	275	6.54 ± 2.41	1.44	0	0.00 ± 0.00	0.00	552	46.00 ± 6.99	4.87	155	0.00 ± 0.00	2.67			
<i>Ditylenchus</i>	Anguinidae	f2	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	31	15.50 ± 0.11	0.27	0	0.00 ± 0.00	0.00			
<i>Dorylillum</i>	Leptonchidae	f4	85	2.02 ± 1.55	0.45	0	0.00 ± 0.00	0.00	29	29.00 ± 0.00	0.26	50	1.19 ± 0.85	0.86			
<i>Nothotylenchus</i>	Anguinidae	f2	509	12.12 ± 6.27	2.67	0	0.00 ± 0.00	0.00	24	24.00 ± 0.00	0.21	95	2.26 ± 1.48	1.63			
<i>Tylencholaimellus</i>	Leptonchidae	f4	127	3.02 ± 2.41	0.67	0	0.00 ± 0.00	0.00	29	29.00 ± 0.00	0.26	0	0.00 ± 0.00	0.00			
<i>Tylencholaimus</i>	Leptonchidae	f4	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	17	17.00 ± 0.00	0.15	0	0.00 ± 0.00	0.00			
Omnivores																	
<i>Dorylaimus</i>	Dorylaimidae	o4	352	8.38 ± 3.11	1.85	0	0.00 ± 0.00	0.00	480	70.27 ± 8.59	4.24	0	0.00 ± 0.00	0.00			
<i>Eudorylaimus</i>	Qusianematidae	o4	861	20.50 ± 5.28	4.52	0	0.00 ± 0.00	0.00	209	23.22 ± 1.71	1.84	0	0.00 ± 0.00	0.00			

<i>Labronema</i>	Qusianematidae	o4	856	20.38 ± 5.07	4.49	0	0.00 ± 0.00	0.00	734	33.36 ± 4.02	6.48	0	0.00 ± 0.00	0.00
<i>Laimydorus</i>	Thornematidae	o5	299	7.11 ± 2.75	1.57	0	0.00 ± 0.00	0.00	1,048	65.50 ± 9.34	9.25	0	0.00 ± 0.00	0.00
<i>Mesodorylaimus</i>	Dorylaimidae	o5	1,183	28.17 ± 8.10	6.21	0	0.00 ± 0.00	0.00	167	20.87 ± 1.77	1.47	0	0.00 ± 0.00	0.00
<i>Prodorylaimus</i>	Thornematidae	o4	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	163	32.60 ± 2.44	1.44	0	0.00 ± 0.00	0.00
<i>Thornia</i>	Nordiidae	o4	200	4.76 ± 2.17	1.05	0	0.00 ± 0.00	0.00	706	35.30 ± 3.06	6.23	0	0.00 ± 0.00	0.00
Predators														
Mononchida	Mononchida	p4	891	21.21 ± 5.96	4.67	258	6.14 ± 2.91	4.35	197	24.62 ± 1.95	1.74	0	0.00 ± 0.00	0.00
<i>Seinura</i>	Aphelenchoididae	p2	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	85	42.5 ± 6.73	0.75	0	0.00 ± 0.00	0.00
Plant-parasitic														
<i>Criconemella</i>	Criconematidae	h3	135	3.21 ± 1.71	0.71	0	0.00 ± 0.00	0.00	195	65.00 ± 11.55	1.72	78	1.85 ± 0.86	1.34
<i>Criconemoïdes</i>	Criconematidae	h3	747	17.78 ± 6.48	3.92	142	3.38 ± 1.65	2.40	0	0.00 ± 0.00	0.00	173	4.11 ± 3.91	2.98
<i>Helicotylenchus</i>	Hoplolaimidae	h3	525	12.50 ± 8.04	2.75	497	11.83 ± 4.44	8.39	967	74.38 ± 10.98	8.54	767	18.25 ± 6.05	13.19
<i>Hoplolaimus</i>	Hoplolaimidae	h3	0	0.00 ± 0.00	0.00	108	2.57 ± 2.60	1.82	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00
<i>Paratrichedorus</i>	Trichodoridae	h4	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	21	0.50 ± 0.38	0.36
<i>Paratylenchus</i>	Paratylenchidae	h2	90	2.14 ± 2.16	0.47	0	0.00 ± 0.00	0.00	266	88.67 ± 18.86	2.35	0	0.00 ± 0.00	0.00
<i>Pratylenchus</i>	Pratylenchidae	h3	0	0.00 ± 0.00	0.00	467	11.11 ± 4.08	7.88	422	70.33 ± 7.85	3.73	121	2.86 ± 2.18	2.08
<i>Rotylenchus</i>	Hoplolaimidae	h3	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	77	38.50 ± 1.87	0.68	0	0.00 ± 0.00	0.00
Tylenchidae	Tylenchidae	h2	899	21.40 ± 8.51	4.72	0	0.00 ± 0.00	0.00	0	34.00 ± 2.68	0.00	790	18.81 ± 4.33	13.59
<i>Trichodorus</i>	Trichodoridae	h4	0	0.00 ± 0.00	0.00	0	0.00 ± 0.00	0.00	754	41.88 ± 4.56	6.66	170	4.04 ± 1.47	2.92
<i>Xiphinema</i>	Longidoridae	h5	748	17.81 ± 5.74	3.92	191	4.54 ± 1.99	3.22	457	38.08 ± 5.14	4.03	121	2.88 ± 0.81	2.08
Total Nematodes			19,060	453.81 ± 52.92		5,922	141.00 ± 19.50		11,325	269.64 ± 29.84		5,811	138.36 ± 18.22	

3.2 Spatial distribution and relationships between nematodes and soil attributes

3.2.1 Spatial distribution of nematodes and soil attributes

Geostatistical analysis did not allow assessment of the spatial distribution of nematodes and the soil attributes had a low range of spatial parameters. We conclude that the scale of sampling was too great for detection of spatial distribution patterns. Thus, all studied variables were considered to represent a pure nugget effect (data not shown). According to Carrasco (2010), the magnitude of the pure nugget effect is dependent on sample size, sampling density, sampling quality, assaying procedures, and of the nature of the phenomenon under study. Therefore, understanding the nature of the pure nugget effect is essential.

Of the variable studied, nematode assemblages have a natural tendency to develop in aggregates or patches due to their reproductive characteristics, behavioral activity and food availability (Ferris, 1985; McSorley, 1987). In geostatistical studies of nematode spatial patterns, the aggregated spatial patterns have been considered the basis of the pure nugget effect (Dinardo-Miranda and Fracasso 2010; Cardoso et al. 2012). Alternatively, the aggregated spatial pattern of the nematodes may be described by a Taylor's power law model (Taylor, 1961; Ferris, 1985; Allsopp, 1990; Ghaderi et al. 2012; Park et al. 2013). The b-values were estimated by regression for nematode taxa in this study (Tab. 2) and ranged from 1.80 to 2.41, confirmed the aggregated pattern of nematode distribution in the soil.

Table 2 Parameter of Taylor's power law model estimated by regression for nematode taxa associated to two forest areas.

Trophic groups	Nematodes		Forest 1		Forest 2	
	r ²	b	SE	r ²	b	SE
Bacterivore						
<i>Acrobeles</i>	0.97	2.11	0.06			
<i>Diploscapter</i>	0.99	2.16	0.02			
<i>Isolaimium</i>				0.99	2.26	0.03
<i>Prismatolaimus</i>	0.91	1.85	0.10	0.96	2.08	0.07
<i>Rhabditida</i>	0.90	1.80	0.10	0.56	1.81	0.30
Fungivore						
<i>Aphelenchus</i>				0.98	2.25	0.04
<i>Aphelenchoides</i>	0.99	2.28	0.03	0.98	2.20	0.04
<i>Ditylenchus</i>				0.99	2.41	0.0001
<i>Nothotylenchus</i>	0.99	2.25	0.02			
Omnivore						
<i>Dorylaimus</i>	0.99	2.19	0.04	0.99	2.18	0.03
<i>Eudorylaimus</i>	0.96	2.04	0.07	0.98	2.17	0.05
<i>Labronema</i>	0.95	2.03	0.04	0.96	1.96	0.07
<i>Laimydorus</i>	0.98	2.20	0.05	0.99	2.18	0.03
<i>Mesodorylaimus</i>	0.94	2.05	0.09	0.99	2.41	0.004
<i>Prodorylaimus</i>				0.99	2.39	0.004
<i>Thornia</i>	0.99	2.28	0.03	0.94	1.91	0.09
Predator						
<i>Mononchida</i>	0.94	2.04	0.09	0.99	2.25	0.04
<i>Seinura</i>				0.99	2.33	0.006
Plant-parasitic						
<i>Criconematidae</i>	0.96	2.08	0.07	0.99	2.28	0.008
<i>Helicotylenchus</i>	0.99	2.25	0.02	0.98	2.14	0.04
<i>Paratylenchus</i>				0.99	2.27	0.01
<i>Pratylenchus</i>				0.99	2.22	0.02
<i>Rotylenchus</i>				0.99	2.36	0.003
<i>Tylenchidae</i>	0.99	2.22	0.02	0.99	2.32	0.01
<i>Trichodorus</i>				0.90	1.89	0.11
<i>Xiphinema</i>	0.98	2.18	0.05	0.98	2.21	0.04

SE = Standard error of $\pm b$ -value. Number of regression data points = 30.

Rarefaction curves (RC) were calculated to determine the sampling intensity necessary to detect species richness with a known confidence level (95%) (Fig. 2). RC stabilized for all studied sites, indicating that 42 sampling points were more than sufficient to detect most of the taxa present (as represented by the limits of this dataset) in the studied areas. RC estimates of the number of taxa using Jackknife Estimator 1 (SJack1) (Fig. 2a) showed that the estimated number of taxa during the F1 rainy season was 27.9 (F1R) and 12.95 (F1D), which are values close to Mao Tau (observed values)

of 24 and 11, respectively. RC estimates of the number of taxa using SJack1 in F2 (Fig. 2b) were 31.9 (F2R) and 10.61 (F2D), values close to Mao Tau (observed values) of 28 and 10, respectively. Therefore, SJack1 estimated satisfactorily the total taxa richness in both areas.

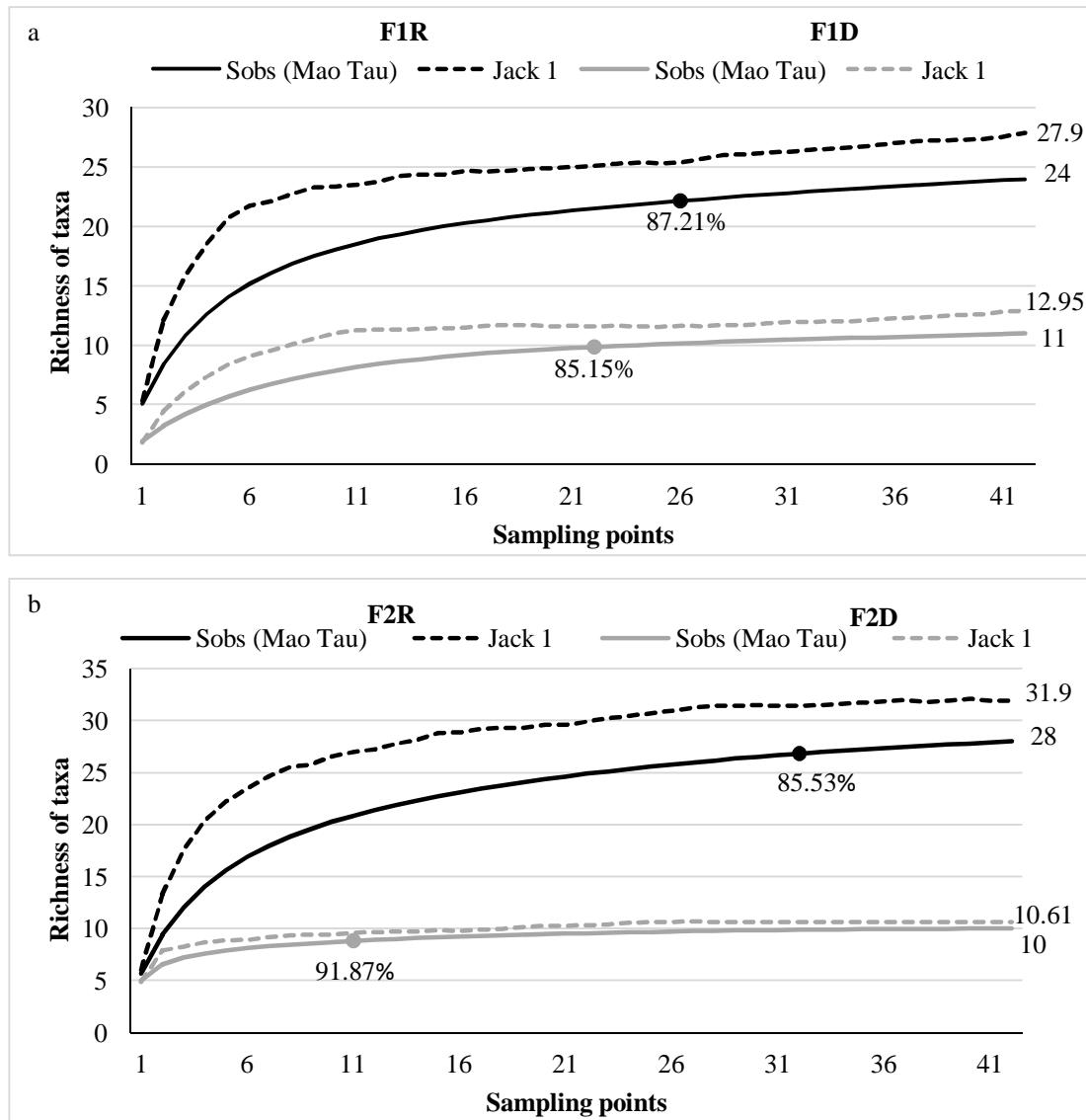


Figure 2 Rarefaction curves showing taxa richness observed (Mao Tau) and estimated (Jack 1); (a) forest 1: *F1R* rainy season and *F1D* dry season; (b) forest 2: *F2R* rainy season and *F2D* dry season. Observed and estimate values were based on 42 samples. Curves were estimated from 1,000 randomizations.

The RC analyses indicated that in F1, 25 (rainy season) and 22 (dry season) sampling points are sufficient to sample 87% and 85% of the taxa present in this area. In F2, 32 (rainy season) and 11 (dry season) sampling points are sufficient to sample 85%

and 92% of the taxa present, respectively. We conclude that in the rainy season the sampling to detect more nematode taxa should be more intensive due to higher taxa richness in this season.

3.2.2 Relationships between nematodes and soil attributes

Pearson ρ s correlations were calculated to assess the relationships between nematode populations and soil properties, (data not shown).

Predator nematodes (Mononchida and Dorylaimida) were positively correlated with water content ($r = 0.32; p < 0.05$), consistent with the aquatic nature of soil nematodes and the need of predator nematodes to move through the soil system to feed on other nematodes (Griffiths, 1994; Briar et al. 2012).

Contrary to our expectation, soil respiration rate was negatively correlated with bacterivore nematodes and enrichment metabolic footprint ($r = -0.35; p < 0.05$). Soil respiration rate is a measurement that is often used to estimate soil microbial activity (Dilly, 2006); therefore we expected that increase in the microbial activity would result in an increase in population levels of bacterivore nematodes. Also, a positive interaction between bacterial biomass and bacterivores was reported by Fu et al. (2005). Considering that this study was carried out in forest soils, where there are different microhabitats, other factors that were not measured may be influencing the distribution and abundance of this trophic group.

Predator nematodes (Mononchida and Dorylaimida) were negatively correlated with soil organic matter (SOM) ($r = -0.36; p < 0.05$). Forest soils have high plant density and diversity, which favors opportunistic nematode groups, such as bacterivores, through high inputs of organic matter (Biedermeier et al. 2008). Therefore, the negative correlation between these factors is contrary to our expectation because the high SOM should indirectly favor predator nematodes.

Enrichment index (EI) was negatively correlated with bulk density (BD) ($r = -0.33; p < 0.05$). The decrease in EI when BD increases is possibly because at higher BD the pore space is reduced (Klein and Libardi, 2002), which influences microbial activity for example by reducing acid phosphatase (Jordan et al. 2003) or by inhibiting growth of the total and nitrifying bacteria populations (Pupin et al. 2009). That would indirectly affect EI, which indicates the predominance of enrichment opportunist

bacterivore nematodes (e.g. the order Rhabditida, and the families Diploscapteridae and Bunonematidae) within the nematode assemblage (Ferris et al. 2001).

3.3 Soil food web: structure and condition

In the faunal analysis (Figure 3), the hypothesis that forest soils have food webs with greater structure and low enrichment was accepted at F1D. In F1R and in both seasons in F2 the structure and enrichment of the food web were high. According to Ferris et al. (2001), the soil food web in F1 is enriched and structured in the rainy season, and structured but resource-limited in the dry season. And in the F2, the soil food web is enriched and structured in both seasons.

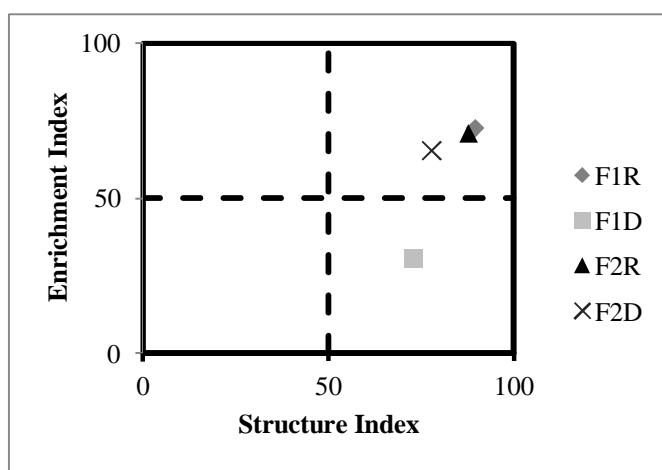


Figure 3 Faunal analysis from *F1R* Pau Amarelo forest in rainy season; *F1D* Pau Amarelo forest in dry season; *F2R* Camucim forest in rainy season; *F2D* Camucim forest in dry season.

In the F1, EI showed significant difference between time, highest values in the rainy season, but no significant differences between seasons in F2 ($p < 0.05$). Possibly, this fact occurred in F1 due to the dry season being characterized by lower soil water content, reduced microbial activity, concomitantly affects negatively the food availability for bacterivore and fungivore nematodes. McGuire et al. (2012) reported that the precipitation may be a more important factor than plant diversity or soil nutrient status in structuring soil fungal communities in tropical forest. Leff et al. (2012) suggest that soil bacterial communities depend of the nature of the organic matter to be decomposed. Concerning F2, the soil water content was not a determining factor, and therefore there must be other factors supporting bacterivore and fungivore nematodes.

Cesarz et al. (2013) reported that litter and root quality positively affect the abundance of specific nematode groups, e.g. bacterivore nematodes.

The CI indicates the predominance of fungivore nematodes in decomposition pathways (Ferris et al. 2001), with higher values suggesting that fungivore nematodes are important in organic matter decomposition and lower values indicating dominance of bacterivore nematodes. In this study CI differed significantly through time ($p < 0.05$), but kept low values, characterizing decomposition channel in the food web was bacterial-mediated in both forest soils. This was contrary to our expectation because fungi are usually considered to be the major decomposers of forest soils (Rayner and Boddy, 1988; Berg and McClaugherty, 2008; Ferris et al. 2004). Moreover, there is differences in the growth forms of fungi and bacteria, which exhibit hyphal (low growth rate) and individual cells (high growth rate), respectively (Hendrix et al. 1986). However, quality and quantity of soil organic matter must also be taken into account in regulating dominance of fungal or bacterial energy channels (Holtkamp et al. 2008).

As expected, there were no differences in BI between sites and time ($p < 0.05$), indicating no change in the proportional abundance of general opportunistic nematodes (e.g. *Acrobelus*), which are tolerant to soil perturbation.

SI values were high in all areas and seasons. The values of MI and $\hat{U}MI$ did not differ between areas and time ($p < 0.05$), and were >2 , indicating an intermediate stage of succession (Fig. 4A). Thus, consistent with general understanding, the forest areas have complex and stable soil food webs, with nematode assemblages dominated by larger species with longer life cycles, such as omnivores (K-strategists), which are sensitive to perturbation and need more time to establish compared to opportunistics fast growing bacterivore and fungivore nematodes (Bongers and Bongers, 1998; Ferris et al. 2001). Similarly, Eisenhauer et al. (2011), after a long-term experiment, reported that increase in plant diversity resulted in significant increase in the abundance of predator nematodes and greater soil food web complexity.

In F1, PPI differed significantly through time with highest values in the rainy season. However, there were no temporal differences in F2 ($p < 0.05$). Probably, a decrease in root production during the dry season decreased the abundance of plant-parasitic nematodes (Bongers, 1990; Rossouw, 2008; Teillet et al. 2013), contributing to lower values of PPI in the F1D compared to F1R.

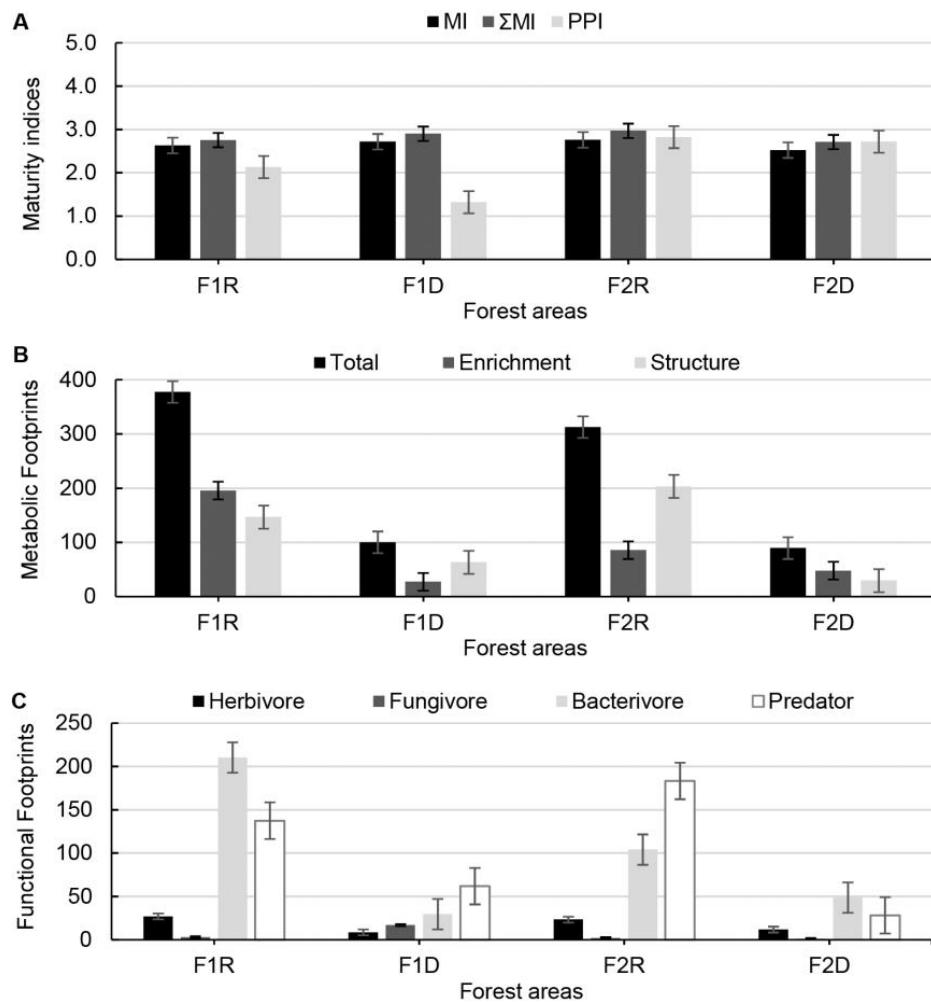


Figure 4 Maturity indices, Metabolic and Functional Footprints in different forest areas. *A* Maturity indices; *B* Metabolic Footprints; and *C* Functional Footprints. *F1R* Pau Amarelo in rainy season; *F1D* Pau Amarelo in dry season; *F2R* Camucim in rainy season; *F2D* Camucim in dry season.

The nematode metabolic footprint analyses were applied as an indicator of responsiveness of the nematode assemblage to resources and their likely effect on the magnitude of the functions and services provided by the different functional guilds (Fig. 4B and 4C) (Ferris et al. 2010; Ferris et al. 2012).

In the F1, enrichment, herbivore and bacterivore metabolic footprints differed significantly across time; their values were greater in the rainy season. There were no temporal difference in F2 ($p < 0.05$). Thus, in the rainy season diverse groups of nematodes were favored by greater availability of nutrients and food.

The fungivore metabolic footprint (FF) differed in the F1, where it was greatest in the dry season, but there were no differences in F2 ($p < 0.05$). Similarly, Bakonyi and Nagy (2000) reported the preference of fungivore nematodes for dry sites. However,

Briar et al. (2012) suggest that all taxa within a trophic group may not respond to edaphic factors in the same way.

The structure metabolic footprint (SF) differed across between time and between areas ($p < 0.05$). SF is an indicator of higher trophic level nematodes, including predators of opportunistic taxa (Ferris et al. 2010). Thus, the decline in food availability in the dry season affected the prey availability (herbivore, fungivore and bacterivore nematodes) resulting in decrease of abundance of predator nematodes (Ferris et al. 2012).

3.4 Soil food web: functional connectance

Functional connectance is represented by spatial co-location between predators and their prey. If the predation function is to occur, predators and prey must be in the same place at the same time. Prey are defined as amplifiable if they can be increased by adding resources or by other environmental modification; target prey are defined as those that are, subjectively, the desired target to be suppressed or regulated by the predation (Sánchez-Moreno et al. 2011; Ferris et al. 2012). Due to this research to be carried out in forest areas, we tested if low prey-abundance was related to high predator-abundance, what is expected in a stable system. Confirming this hypothesis, the correlations were positive in both areas and time: predator vs. prey ($r = 0.31$; $p < 0.05$), predators vs. target prey ($r = 0.24$; $p < 0.05$) and predators vs. amplifiable prey ($r = 0.25$; $p < 0.05$). Eisenhauer et al. (2011) and Chung et al. (2007) assert that plant diversity benefits microbial communities and the ecosystem services of soils. Thus, positive correlations between predator and prey nematodes are expected in forest soils due to more complex soil food web which favors both prey and predators.

The interactions between predator and prey did not differ between areas and time ($p < 0.05$). In other words, the predator-prey relationship was not affected by changes in soil properties that fluctuate with time (e.g. soil water content, soil respiration).

4 Conclusions

- Structure and composition of the nematode assemblages were quite different between areas and time; there was higher abundance, trophic diversity and taxa richness in the rainy season; rarefaction curves suggest that sampling to detect more nematode taxa should be more intensive in the rainy season.
- The scale of sampling was too great for detection of spatial distribution patterns of the nematodes; alternatively, their aggregated spatial patterns were described by a Taylorøs power law model, with b-values ranged from 1.80 to 2.41, confirming the aggregated pattern of nematode distribution in the soil.
- According to faunal analysis the studied forest soils have complex and stable soil food webs, with higher connectance (dominated by larger species with longer life cycles); and predominant decomposition channel bacterial-mediated in both forest soils. In addition, MI and ÛMI indicated an intermediate stage of succession in the areas.
- The predator-prey relationships did not differ across areas and time, thus they were not affected by changes in soil properties that fluctuate with time (e.g. soil water content, soil respiration).

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Considerações finais e Perspectivas

CAPÍTULO VI

Considerações finais e Perspectivas

A composição e a estrutura das comunidades de nematoides diferiram entre os ambientes estudados (cana-de-açúcar sob diferentes manejos e remanescente de floresta) (Capítulo II). *Pratylenchus* predominou em áreas cultivadas intensivamente com cana-de-açúcar, *Dorylaimida* em solos de floresta e *Hoplolaimus* em áreas de cana-de-açúcar sob manejo não-intensivo. Assim, as condições edáficas inerentes a cada área dão suporte à diferentes *taxa* que compõem as comunidades de nematoides.

A ordenação dos *taxa* de nematoides pela Análise de Correspondência Canônica (CCA) sugere que os gêneros parasitas de plantas *Pratylenchus*, *Hemicycliophora*, *Helicotylenchus*, *Meloidogyne* e *Criconemella* estão relacionados à área sob manejo intensivo da cana-de-açúcar, a qual foi caracterizada por maiores valores de resistência mecânica do solo à penetração, densidade do solo e teor de argila. Enquanto que na área de cana-de-açúcar sob cultivo não-intensivo, os gêneros *Hoplolaimus* e *Acrobeles* (parasita de planta e bacteriófago, respectivamente) foram os gêneros dominantes e não se correlacionaram com os atributos físicos estudados. Do mesmo modo, em solo de floresta, *Prismatolaimus*, *Tylenchidae*, *Paratrichodorus*, *Rotylenchulus* e *Xiphinema* não se correlacionaram com as propriedades físicas do solo, indicando que outros fatores edafoclimáticos ou vegetacionais estão determinando a presença destes.

A presença do gênero bacteriófago *Prismatolaimus* e do parasita de planta *Xiphinema* em solo de floresta é bastante coerente, devido à sensibilidade destes gêneros à perturbações no solo, apresentando geralmente baixa abundância em áreas cultivadas. Desse modo, pode-se inferir que diferentes práticas de manejo e sistemas de cultivo afetam as propriedades do solo e, consequentemente, determinam a abundância e dominância de nematoides.

A densidade do solo, a abundância dos nematoides totais e dos parasitas de plantas apresentaram maiores valores em solos cultivados com cana-de-açúcar quando comparados com áreas de floresta. De modo contrário, a porosidade total, a respiração do solo e a abundância de nematoides predadores apresentaram os menores valores (Capítulo III). Assim, as práticas de manejo convencionais aplicadas em áreas de cana-de-açúcar resultaram em alterações nas propriedades do solo e, concomitantemente, alterou a estrutura e composição das comunidades de nematoides.

A ordenação pela Escala Não Métrica Multidimensional (NMDS) separou claramente as comunidades de nematoides das áreas cultivadas com cana-de-açúcar daquelas de floresta, formando três grupos distintos (Grupo 1: 10 áreas de cana-de-açúcar e 2 áreas de floresta; Grupo 2: 6 áreas de floresta; Grupo 3: 2 áreas de floresta). A Análise de Similaridade (ANOSIM) apontou diferença significativa entre os grupos 1 e 2/3, enquanto os grupos 2 e 3 não diferiram entre si. Portanto, as comunidades de nematoides associadas às áreas de cana-de-açúcar foram significativamente diferentes daquelas associadas às áreas de floresta; exceto duas áreas de floresta que se agruparam juntamente com as áreas de cana-de-açúcar.

A composição e estrutura das comunidades de nematoides e as características do solo em áreas de floresta e cultivadas com cana-de-açúcar foram distintas, mesmo quando analisadas em mesma usina e tipo de solo (ex.: Quatro fragmentos de floresta situados na Usina Estreliana sob Latossolo Amarelo). As áreas de floresta que não diferiram das áreas cultivadas com cana-de-açúcar foram caracterizadas por elevada densidade do solo e baixa porosidade total, relevo íngreme e presença de nematoides parasitas de plantas, os quais podem ter sido carreados pela água da chuva ou se estabelecido devido as características edáficas intrínsecas destas áreas.

A compactação do solo, por exemplo, é um fator resultante do uso de maquinários pesados durante as operações de colheita que, além de ocasionar queda da produtividade da cana-de-açúcar, altera a distribuição dos nematoides nos níveis tróficos, aumentando a abundância de parasitas de plantas e reduzindo a abundância dos demais grupos tróficos. O aumento da densidade do solo, frequentemente, favorece a dominância de um único gênero parasita de planta. Por isso, pesquisas recentes apontam que a rotação de cultura e pousio incrementam a produtividade da cana-de-açúcar, devido a supressão de organismos fitopatogênicos.

Em relação a *faunal analysis*, tanto os solos de floresta quanto os cultivados foram caracterizados por cadeia alimentar complexa e estável, com alta conectância (dominada por nematoides maiores com longo ciclo de vida); e o canal de decomposição da matéria orgânica foi mediado por bactérias. Os índices de maturidade indicaram um estágio intermediário de sucessão nas áreas de estudo (Capítulos III e IV).

Os canais de energia dominados por fungos tendem a se desenvolver quando os solos não são manejados por muito tempo. Apesar disso, a dominância dos canais de energia bacterianos e fúngicos são, frequentemente, determinados pela qualidade e quantidade da matéria orgânica no solo.

As relações entre nematoides predadores e presas não diferiram entre áreas e tempo. Logo, as relações não foram afetadas pelas alterações nas propriedades do solo que variam no tempo (umidade e respiração do solo) (Capítulo V). As correlações entre nematoides predadores e presas foram positivas, mesmo em áreas e estações distintas. Comumente, as áreas de floresta são caracterizadas por cadeia trófica do solo complexa e propicia uma melhor expressão das relações entre os nematoides predadores e àqueles que devem ser regulados ou suprimidos.

No que diz respeito à variabilidade espacial dos nematoides, não foi possível detectar um padrão de distribuição (Capítulo V). Alternativamente, o padrão de distribuição espacial dos nematoides tido como agregado foi confirmado pelo *Taylor's power law model*. Apesar de não ter sido possível esta detecção, estudos a cerca deste assunto são relevantes, pois informações acerca dos fatores que determinam a distribuição espacial dos nematoides são incipientes, com a maioria dos estudos de heterogeneidade espacial geralmente enfatizando apenas o padrão vertical. Além disso, a maioria dos estudos em padrões horizontais dos nematoides tem sido realizados em áreas agrícolas e estão focados apenas em espécies parasitas de plantas. A heterogeneidade do solo faz com que os nematoides apresentem um padrão de variação espacial reagindo diferentemente às condições do solo e à disponibilidade de recursos, e sua abundância e atividade variam horizontalmente e verticalmente através do perfil do solo.

Este estudo ressaltou a capacidade dos nematoides serem indicadores ecológicos do solo. Além disso, mostrou que a diversidade vegetal e as propriedades do solo, tais como densidade do solo, porosidade, textura, umidade e respiração do solo são fatores que, até certo ponto, determinam a estrutura, composição e abundância das comunidades de nematoides. Porém, este estudo reconhece que o solo é um ambiente heterogêneo e que outros fatores, os quais não foram mensurados, estão influenciando a ocorrência destes organismos. Contudo, a temperatura do solo, pH e a relação C:N estão entre os parâmetros a ser incluídos em futuras análises, pois exercem influência relevante na atividade microbiana, a qual interage diretamente com as comunidades de nematoides. Por outro lado, a diversidade e a abundância de nematoides parasitas de plantas e de vida livre têm influência sazonal, portanto, a amostragem em diferentes estações climáticas deve ser considerada, a depender do objeto de estudo. Por fim, a interação entre predadores e presas é um aspecto que também deve ser levado em consideração em futuros estudos, nos quais enfatizem os fatores ambientais que de-

alguma forma influenciam esta interação. Contudo, prévios estudos apontam que a complexidade das comunidades vegetais favorecem a predominância de nematoides bacteriófagos, micófagos e predadores, desse modo, estudos *in situ* em remanescentes de Floresta Atlântica vão auxiliar na busca de predadores generalistas e especialistas de nematoides parasitas de plantas de importância econômica para o sistema de produção da cana-de-açúcar, tais como: *Meloidogyne*, *Pratylenchus* e *Helicotylenchus*.