INSTITUTO NACIONAL DE PESQUISAS DA AMAZÔNIA - INPA

PROGRAMA DE PÓS-GRADUAÇÃO EM ECOLOGIA

Balanço nutricional em uma floresta na Amazônia Central: o papel dos estoques e fluxos na ciclagem de nutrientes

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Manaus

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Dissertação de mestrado apresentada ao Instituto Nacional de Pesquisas da Amazônia como parte dos requisitos para obtenção do título de Mestre em Biologia (Ecologia).

Manaus

Agosto, 2022

FICHA CATALOGRÁFICA

A848b Assis, Pâmella Leite de Sousa

Balanço nutricional em uma floresta na Amazônia Central: o papel dos estoques e fluxos na ciclagem de nutrientes / Pâmella Leite de Sousa Assis; orientador Carlos Alberto Nobre Quesada; coorientadora Laynara Figueiredo Lugli; - Manaus:[s. l.], 2022.

1.1 MB
62 p. : il. color.

Dissertação (Programa de Pós-Graduação em Ecologia) - Coordenação do Programa de Pós-Graduação, INPA, 2022.

1. Ciclos biogeoquímicos. 2. Dinâmica do carbono. 3. Dinâmica dos nutrientes. I. Quesada, Carlos Alberto Nobre. II. Lugli, Laynara Figueiredo. III. Título

CDD 634.975

Sinopse

Nesse estudo foi investigado, utilizando banco de dados, a dinâmica do carbono e nutrientes de uma floresta de *terra firme* na Amazônia Central, através da quantificação dos estoques, fluxos, retranslocação e eficiência no uso dos nutrientes, resultando no seu balanço.

Palavras-chave: Ciclos biogeoquímicos; Dinâmica do carbono; Dinâmica dos nutrientes; Florestas tropicais; Retranslocação; Eficiência no uso dos nutrientes; Demanda anual de nutrientes.

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ATA DA DEFESA PÚBLICA DA DISSERTAÇÃO DE MESTRADO DO PROGRAMA DE PÓS-GRADUAÇÃO EM ECOLOGIA DO INSTITUTO NACIONAL DE PESQUISAS DA AMAZÔNIA.

Aos 08 dias do mês de Julho do ano de 2022, às 08h30min, por videoconferência. Reuniu-se a Comissão Examinadora de Defesa Pública, composta pelos seguintes membros: o Dr. **David Montenegro Lapola**, da Universidade Estadual de Campinas – UNICAMP o Dr. **Demétrius Lira Martins**, da da Universidade Estadual de Campinas – UNICAMP e a Dra. **Flora Magdaline Benitez Romero**, do Instituto Nacional de Pesquisas da Amazônia – INPA,tendo como suplentes o Dr. Bruce Walker Nelson, do Instituto Nacional de Pesquisas da Amazônia – INPA e o Dr. José Luís Campana Camargo, do Instituto Nacional de Pesquisas da Amazônia – INPA, sob a presidência do orientador, a fim de proceder a arguição públicado trabalho de **DISSERTAÇÃO DE MESTRADO** de **PÂMELLA LEITE DE SOUSA ASSIS**,intitulado: **" BALANÇO NUTRICIONAL EM UMA FLORESTA NA AMAZÔNIA CENTRAL: O PAPEL DOS ESTOQUES E FLUXOS NA CICLAGEM DE NUTRIENTES"**, orientada
pelo Dr. Carlos Alberto Nobre Quesada e co-orientada pela Dra. Laynara Figueiredo Lugli,ambos do Instituto Nacional de Pesquisas da Amazônia – INPA.

Após a exposição, o discente foi arguido oralmente pelos membros da Comissão Examinadora, tendo recebido o conceito final:

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Nada mais havendo, foi lavrada a presente ata, que, após lida e aprovada,foi assinada pelos membros da Comissão Examinadora.

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AGRADECIMENTOS

Aos meus pais, que me apoiam a seguir os meus sonhos e que fazem o possível e o impossível para que eu possa me dedicar a minha formação. As minhas irmãs e sobrinhos, por compreenderem as minhas ausências e me incentivarem na minha carreira. Aos meus amigos Abdy da Silva e Bruna Teitelroit por acreditarem sempre em mim e no meu potencial mesmo quando eu não acredito.

Ao meu orientador Carlos Alberto Quesada pela confiança, pelas reuniões tranquilizadoras e por estar presente quando eu mais precisava. A minha orientadora Laynara Figueiredo Lugli por todo conhecimento transmitido e por estar ao meu lado durante todo o processo sempre extremamente atenciosa e solicita, espero me tornar uma cientista assertiva e gentil como você.

Aos meus colegas de turma que, apesar da distância, formaram uma rede de apoio, em especial a Giuliette Mano, minha companheira de aventuras que esteve ao meu lado fisicamente e virtualmente nos melhores e piores momentos do mestrado e ao Ícaro Lima por ser amigo e confidente nas noites no ASSINPA, no bar do cabelo ou na varanda do apartamento no Solmorar.

Ao Bruno Takeshi por todo carinho e apoio durante os momentos difíceis, além do suporte logístico que possibilitaram a execução desse trabalho.

Aos meus companheiros de república pelas horas que pareciam minutos de conversa na cozinha de casa.

A todos que disponibilizaram as informações essenciais para o desenvolvimento deste trabalho com destaque para Caroline Miron, Amanda Cordeiro, Sabrina Garcia, Julyane Pires, Juliane Menezes, Izabela Aleixo e Iokanam Pereira.

A todos os integrantes do laboratório de ciclos biogeoquímicos pelo apoio no levantamento de dados, nas idas a campo, na resolução de dúvidas e nas interações que muitas vezes foram à distância. Mesmo sendo um mestrado em meio a uma pandemia, o

grupo foi capaz de me acolher e tornar essa jornada mais alegre. Em especial gostaria de agradecer a Nathielly Martins, Flávia Santana, Luciana Bachega, Maria Pires, Yago Santos, Lara Siebert, Anna Moraes, Fernanda Luz, Gabriela Ushida e Alacimar Guedes.

A FAPEAM pela concessão da bolsa de pesquisa, ao Instituto Nacional de Pesquisas da Amazônia e ao Programa de Pós-Graduação em Ecologia pela oportunidade de cursar o mestrado e ao Comitê científico do AmazonFACE por todo suporte.

E a todos que de alguma forma contribuíram para que esse trabalho fosse possível.

RESUMO

Em geral, solos de florestas tropicais apresentam baixa fertilidade, e como resultado, a ciclagem dos nutrientes tornam-se de grande importância para o funcionamento do ecossistema, uma vez que esses elementos são essenciais nos processos metabólicos das plantas e na construção de tecidos. Compreender e quantificar os processos que envolvem a aquisição, armazenamento e ciclagem de nutrientes em diferentes compartimentos da planta, e a sua relação com a produtividade florestal e a acumulação de biomassa, é essencial para caracterizar a dinâmica dos nutrientes nos ecossistemas e projetar como as mudanças ambientais globais, tais como o aumento do CO_2 atmosférico, podem afetar tais processos. Nesse estudo investigamos a dinâmica dos nutrientes de uma floresta de terra firme na Amazônia Central, através da quantificação dos estoques, fluxos, retranslocação e eficiência no uso dos nutrientes, resultando no seu balanço nutricional. A produtividade primária líquida do ecossistema (PPL), os estoques na biomassa e as concentrações dos macronutrientes (N, P, Ca, Mg e K), e micronutrientes (Fe, Mn e Zn) em folhas, troncos, raízes finas, serapilheira, detritos de madeira grossa, e solos do banco de dados do projeto AmazonFACE (Free-Air CO_2 Enrichment), próximo à Manaus, Amazonas, Brasil, foram utilizados para calcular o balanço nutricional dessa floresta. O nosso objetivo foi testar a hipótese de que o macronutriente que cicla mais eficientemente no ecossistema potencialmente seria o elemento mais limitante para a produtividade. Nossos resultados mostraram que o fluxo de nutrientes do ecossistema foi mais rápido nas folhas > serapilheira > troncos > raízes finas, enquanto os estoques de nutrientes foram maiores nos solos > troncos > detritos de madeira grossa > folhas > serapilheira fina > raízes finas > serapilheira média. Entre os compartimentos estudados, o solo apresentou um maior armazenamento de nutrientes, desempenhando um papel importante como fonte de nutrientes para as plantas a longo prazo. No entanto, uma vez que muitos destes nutrientes são indisponíveis para a absorção pelas plantas, por outro lado, foram encontradas grandes quantidades de nutrientes nos troncos, esse compartimento funciona como um reservatório de nutrientes. Em resumo, a absorção de P e o seu retorno ao solo foi muito lento e a sua taxa de retranslocação e eficiência no uso foi elevada, o que sugere um ciclo fechado e um papel crucial como limitante da produção primária líquida. A eficiência no uso do N, por outro lado, foi baixa, enquanto o estoque foi o maior entre os nutrientes, assim como o fluxo que foi rápido, o que era esperado, uma vez que este elemento é abundante nas florestas da Amazônia Central e tem uma ciclagem aberta. Essas informações contribuem para a nossa compreensão sobre o estado atual dos nutrientes numa

floresta da Amazônia Central através da quantidade de carbono e nutrientes produzidos e armazenados e da dinâmica de cada nutriente e o quanto eles são reciclados, essas informações podem nos ajudar a prever melhor a resposta da floresta às alterações climáticas.

Palavras-chave: Ciclos biogeoquímicos; Dinâmica do carbono; Dinâmica dos nutrientes; Florestas tropicais; Retranslocação; Eficiência no uso dos nutrientes.

Nutrient budget in a Central Amazon forest: the role of stocks and flows in nutrient cycling

ABSTRACT

In general, soils of old growth tropical forests have low fertility, and as a result, nutrient cycling becomes of great importance for ecosystem functioning, once these elements are essential for plant metabolic processes and tissue construction. Understanding and quantifying the processes that involve nutrient acquisition, storage, and cycling in different plant tissues, and their relationship with forest productivity and biomass accumulation, is essential to characterize ecosystem nutrient dynamics and to project how global environmental changes, such as the increase in atmospheric CO_2 can affect such processes. Therefore, in this study we investigated the nutrient dynamics of a *terra firme* forest in Central Amazonia, through the quantification of carbon and nutrient stocks, flows, retranslocation rates and nutrient use efficiency. Ecosystem net primary productivity (NPP), biomass stocks and the concentration of macronutrients (N, P, Ca, Mg and K), and micronutrients (Fe, Mn and Zn) in canopy, stems, fine roots, litterfall, coarse wood debris, and soils from the AmazonFACE (Free-Air CO_2 Enrichment) project database, near Manaus, Amazonas, Brazil were used to calculate the nutrient budget of this forest. We aimed to test the hypothesis that the macronutrient cycling more efficiently in the ecosystem would potentially be the most limiting element for productivity. Our results showed that ecosystem nutrient flow was higher in canopy > fine litterfall > stems > fine roots > coarse wood debris > medium litter layer, while nutrient stocks were greater in soils > stems > coarse wood debris > fine litter layer > canopy > fine roots > medium litter layer. Among the studied compartments, soil was the one displaying higher nutrient storage, playing an important role as a nutrient source for plants in the long term. However, many of these nutrients are not immediately available for plant uptake. Large amounts of nutrients were also found stored in living stems, thus forming an important long-term nutrient reservoir. Phosphorus uptake and input into the soil were very slow and its retranslocation rate and use efficiency was high, suggesting a closed nutrient cycle of this element and a large potential to limit net primary production. Nitrogen use efficiency, on the other hand, was low, and both the stock and

fluxes were high. Knowing how much nutrient is needed to allow biomass production in a year, and how much of it can be recycled or relocated, sheds light on how nutrient limitation may constrain ecosystem level responses to elevated atmospheric CO_2, and help us to better predict the forest response to climate change.

Key words: Biogeochemical cycle; Carbon dynamic; Nutrient dynamics; Tropical forest; Retranlocation; Nutrient efficiency use.

Sumário

Introdução

Solos de florestas tropicais geralmente apresentam baixa disponibilidade de nutrientes, com a vegetação adotando uma série de mecanismos de conservação a fim de evitar perdas, garantindo a disponibilidade desses recursos (Bruijnzeel, 1991). Outro aspecto recorrente na vegetação de ecossistemas pouco férteis é o crescimento lento, estratégia vantajosa uma vez que uma absorção lenta dos nutrientes, diminui a possibilidade de esgotamento (Chapin & Stuart, 1980). Florestas que apresentam baixas concentrações de nutrientes disponíveis possuem uma ciclagem de nutrientes "fechada", caracterizada pelo pouco escoamento de nutrientes para fora do sistema (Vitousek, 1984). Em decorrência disso, florestas tropicais não perturbadas se mantêm exuberantes com alto acúmulo de biomassa mesmo em solos pobres (Grau *et al.*, 2017).

A ciclagem de nutrientes é um processo de grande importância para os ecossistemas terrestres, principalmente para aqueles que apresentam solos com baixa fertilidade, como é o caso da Amazônia Central. Esse processo pode ser definido como a transferência de nutrientes entre compartimentos dentro de um sistema (Vitousek, 1982). Os nutrientes podem entrar no sistema, e ficar disponíveis para aquisição pelos organismos, através de diferentes meios como a partir da deposição atmosférica (Jordan, 1982) ou do aporte de material orgânico sobre o solo (Vitousek & Sanford, 1986). Estando disponíveis, esses nutrientes são absorvidos e alocados nos compartimentos (raízes, folhas ou troncos) de acordo com as necessidades metabólicas das plantas (Chapin *et al.*, 1987). Existem, no entanto, fatores que influenciam a ciclagem de nutrientes nos ecossistemas florestais como o clima, a composição de espécies, o estágio sucessional destas plantas e a fertilidade do solo (Vitousek & Sanford, 1986).

Florestas tropicais contribuem com 55% do total de carbono estocado na biosfera terrestre (Pan *et al.*, 2011). Essa alta alocação demonstra a importância das florestas como sumidouro de CO_2, base do metabolismo primário das plantas, e um dos gases intensificadores do efeito estufa, sendo assim a alocação de carbono (C) pelas plantas potencialmente serve como medida de mitigação da emissão antropogênica de carbono (Phillips *et al.*, 2017). Contudo, estudos recentes apontam uma tendência, a longo prazo, de diminuição da contribuição da floresta Amazônica na assimilação de CO_2 atmosférico

(Cernusak *et al.*, 2013; Brienen *et al.*, 2015; Fleischer *et al.*, 2019). Até o momento, somente o balanço de C na floresta Amazônica têm sido amplamente estudado (Malhi *et al.*, 2009; Aragão *et al.*, 2009), enquanto uma caracterização detalhada dos nutrientes só foi realizada para florestas temperadas (Whittaker *et al.*, 1979; Sollins *et al., 1980).* Por isso a importância em entender como a disponibilidades dos nutrientes mais abundantes como o nitrogênio (N), fósforo (P), potássio (K), cálcio (Ca) e magnésio (Mg) podem limitar a produtividade da floresta e, consequentemente, a incorporação de C na forma de matéria orgânica.

Nutrientes podem ser classificados de acordo com a sua demanda pelas plantas. Os macronutrientes são requeridos em maiores quantidades, uma vez que desempenham funções estruturais e metabólicas e podem ser classificados como primários, quando a sua disponibilidade limita o crescimento vegetal e engloba os elementos N, P e K ou macronutrientes secundários, elementos que geralmente não limitam o crescimento das plantas, como é o caso do Ca e Mg (Chapin *et al.*, 2011). Os micronutrientes, ferro (Fe), manganês (Mn) e zinco (Zn) por outro lado, são nutrientes usados em quantidades mais baixas pelas plantas, como parte dos processos de fotossíntese, respiração e como componentes de muitas proteínas nas plantas (Forieri and Hell, 2014; Kaspari *et al.*, 2008). Dessa forma, dependendo do ecossistema a relação de quais nutrientes irão limitar o crescimento irá variar de acordo com as características do ambiente (Quesada *et al.*, 2011; Vitousek et al., 2010).

Quando comparada com regiões subtropicais e temperadas, a grande maioria da bacia amazônica tem baixas concentrações de nutrientes derivados de rochas nos seus solos, tais como P que é necessário como componente de ATP, proteínas, açúcar e ácido nucleico (Yan *et al.*, 2015), a disponibilidade desse elemento limita a produtividade florestal na Bacia Amazônica (Quesada *et al.*, 2012) e poderia modular o futuro sequestro de C nestas florestas, de acordo com alguns modelos (Fleischer *et al.*, 2019). O fósforo é forncecido aos solos exclusivamente através da intemperização do mineral apatita, em decorrência disso a disponibilidade desse nutriente é geralmente baixa nas regiões da Amazônia Central e Oriental, uma vez que estas regiões tem solos altamente intemperizados e não têm atividade geológica recente, como o soerguimento de montanhas (Jordan & Herrera, 1981). Essas características geológicas da região proporcionam uma composição florística com predominância de grupos que apresentam estratégias de crescimento mais tolerantes ao

estresse nutricional e com maior densidade de madeira, tais como espécies das famílias Caesalpiniaceae, Lecythidaceae, Chrysobalanaceae e Sapotaceae (Quesada & Lloyd, 2016).

Outros nutrientes derivados de rochas que também são importantes para as atividades metabólicas das plantas são os cátions (K, Ca e Mg). Esses nutrientes desempenham um papel importante no transporte, produção e catalização de enzimas, regulação osmótica, clorofila e paredes celulares (Lira-Martins *et al.*, 2022; Campo *et al.*, 2000; Kumar *et al.*, 2015). Os cátions têm uma ciclagem semelhante à do P, uma vez que a origem destes elementos é principalmente através de atividades geológicas como o intemperismo de rochas, o que explica a baixa disponibilidade destes nutrientes na região central da bacia amazônica (Quesada *et al.*, 2012).

Com relação ao nitrogênio, existe uma grande demanda deste elemento pelas plantas devido à sua importância nos processos biológicos, por constituir enzimas, proteínas e, também, a molécula de clorofila, sendo que sua obtenção, para a maioria das plantas, se dá integralmente através da absorção dos íons inorgânicos de amônio e nitrato pelas raízes (Chapin *et al.*, 1987). Embora plantas tenham grande demanda desse nutriente, florestas tropicais, em geral, não apresentam escassez desse elemento a ponto de limitar o crescimento vegetal, uma vez que os processos de deposição atmosférica e fixação de nitrogênio por bactérias no solo é abundante (Vitousek, 1984; Martinelli *et al.*, 1999).

Por outro lado, a baixa disponibilidade de nutrientes nos solos da Amazônia Central pode ser parcialmente compensada por uma alta taxa de retranslocação, quando os nutrientes são movidos de um tecido em senescência para outro tecido, como ocorre em folhas e raízes finas (Pires-Santos *et al.*, 2020; Gordon e Jackson, 2000; Brant & Chen, 2015), influenciando não apenas a economia destes elementos, mas também a entrada de nutrientes no sistema pela decomposição de serapilheira (Walker e Syers, 1976; Schaap *et al.*, 2021). Além disso, outros processos de aquisição direta de nutrientes podem ter grande importancia na economia de nutrientes, entre eles a produção de raízes finas, a atividade de enzimas fosfatase, a exsudação de ácidos orgânicos e associação das raízes finas com micorrizas arbusculares (Schaap *et al.*, 2021; Lugli *et al.*, 2020; Reichert *et al.*, 2022). Em suma, ecossistemas em solos inférteis dependem da reciclagem de nutrientes de tecidos mortos e senescidos para se manter, portanto a biomassa das plantas desempenham um papel não só como sumidouro de carbono, mas também como reservatório de nutrientes que regressará ao sistema através da decomposição.

Devido à importância do carbono e dos nutrientes na biomassa como reservatório, da ciclagem de nutrientes através da absorção e retorno ao solo através da produção de tecidos vivos e mortos, e da retranslocação pelos tecidos em senescência, objetivamos quantificar os estoques e fluxos de carbono e nutrientes do dossel, troncos, raízes finas, serapilheira fina e média, detritos de madeira grossa e solos de uma floresta de *terra firme* na Amazônia Central, a fim de estimar o balanço e ciclagem de nutrientes (absorvidos, armazenados e liberados pelas plantas) e determinar quais os nutrientes mais limitantes para a produtividade primária líquida nesse ecossistema. Assim, nossa hipótese é de que o macronutriente que cicla mais eficientemente no ecossistema potencialmente será o elemento mais limitante para a produtividade primária líquida dessa floresta.

Objetivo geral

Este estudo visou quantificar os estoques e fluxos de nutrientes do dossel, troncos, serapilheira fina e média, raízes finas, detritos de madeira grossa e solo de uma floresta de *terra firme* na Amazônia Central, a fim de estimar a dinâmica da ciclagem de nutrientes e determinar quais são os nutrientes mais limitantes para a produtividade líquida primária além da demanda anual de nutrientes nesse ecossistema.

Objetivos específicos

- Determinar a produtividade primária líquida do ecossistema e os estoques de biomassa utilizando diferentes compartimentos florestais (dossel, troncos, serapilheira fina e média, detritos de madeira grossa, raízes finas e solo);
- Estimar a eficiência no uso dos nutrientes em diferentes compartimentos florestais e em todo o ecossistema;
- Quantificar a demanda nutricional anual do ecossistema para produzir biomassa e a quantidade de nutrientes que retorna ao sistema.

Capítulo Único

Assis, P. L. S.; Lugli, L. F.; Quesada, C. A. Nutrient budget in a Central Amazon forest: the role of stocks and fluxes in nutrient cycling. Manuscrito em preparação para *Ecosystems*

Introduction

Tropical forest soils generally have low nutrient availability, requiring a series of nutrient conservation mechanisms in order to avoid losses and sustain the availability of these resources (Bruijnzeel, 1991). Another recurrent aspect of forests growing in oligotrophic ecosystems is their slow growth, which can be seen as an advantageous strategy in this environment, with rates of growth being related to the rates of nutrient release, therefore decreasing the chances of nutrient losses (Chapin, 1980). Forests that have low concentrations of available nutrients often have a so-called "closed nutrient cycling", characterized by little nutrient losses out of the system (Vitousek, 1984). Nutrient cycling is a process of great importance for tropical ecosystems in soils with low fertility, as is the case of the Central Amazon, and as a result, undisturbed tropical forests remain lush with high biomass accumulation even in poor soils (Grau and others, 2017). Nutrient cycling can be defined as the transfer of nutrients between compartments within a system (Vitousek, 1982). Nutrients can enter the system, and become available for acquisition by organisms, through different means such as atmospheric deposition, fixation, organic material inputs to the soil and weathering of parent material (Jordan, 1982; Vitousek & Sanford, 1986). Once available, these nutrients are taken up and allocated to different plant compartments (roots, leaves or stems) according to their metabolic needs (Chapin and others, 1987). However, climate, tree species composition, successional stage and soil fertility could play important roles in controlling nutrient cycling in forest ecosystems (Vitousek & Sanford, 1986).

Tropical forests contribute 55% of the total carbon stored in the terrestrial biosphere (Pan and others, 2011). This high allocation demonstrates the importance of forests as a sink for atmospheric CO_2, which is the basis of primary plant metabolism, but also one of the intensifying greenhouse gasses, with plant carbon (C) allocation potentially serving as a mitigation measure for anthropogenic C emissions (Phillips and others, 2017). Despite that, elevated CO_2 (eCO_2) can promote a net increase in photosynthesis and increased productivity, with recent observations of the Amazon´s carbon balance suggesting a decreased contribution of the Amazon forest as a C sink (Cernusak and others, 2013; Brienen and others, 2015; Fleischer and others, 2019). To date, research has been focused on the C balance of Amazonian forests (Malhi and others, 2009; Aragão and others, 2009), but generally ignoring the links between C cycle and nutrient use, whereas detailed nutrient characterization has only occurred for temperate forests (Whittaker and others, 1979; Sollins and others, 1980).

Hence the importance of understanding how the availability of major nutrients such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) could limit forest productivity and, consequently, the incorporation of C in the form of organic matter.

Nutrients can be classified according to their demand by plants into macro and micronutrients. Macronutrients are required in higher amounts since they perform structural and metabolic functions, being subclassified as primary, when their availability limits plant growth which usually includes the elements N, P and K or secondary macronutrients, elements that generally do not limit plant growth, as is the case of Ca and Mg (Chapin and others, 2011). Micronutrients, iron (Fe), manganese (Mn) and zinc (Zn) on the other hand, are nutrients used in lower amounts by plants, as part of photosynthesis and respiration processes and components of many plant proteins (Forieri and Hell, 2014; Kaspari and others, 2008). Thus, depending on the ecosystem, which nutrients will limit growth will vary according to the characteristics of the environment (Quesada and others, 2011; Vitousek and others, 2010).

When compared to other subtropical and temperate regions, the vast majority of the Amazon basin has low concentrations of rock-derived nutrients in their soils, such as P that is necessary as a component of ATP, protein, sugar and nucleic acid (Yan and others, 2015). Many studies across the Amazon basin point to the fact that the availability of this element has great potential to limit forest wood productivity in Amazonia (Quesada and others, 2012) and could modulate C sequestration under eCO2 in these forests, according to some model projections (Fleischer and others, 2019). Phosphorus is supplied solely through the weathering of the apatite mineral in soils, and the availability of this nutrient is usually low in the Eastern and Central Amazon regions, since these regions are highly weathered and do not have recent geological activity such as mountain uplift (Jordan & Herrera, 1981). These geological characteristics and very low nutrient content of the soils of the region provide a floristic composition with dominance of groups that present growth strategies more tolerant to nutritional stress and with higher wood density, such as species of the families Caesalpiniaceae, Lecythidaceae, Chrysobalanaceae, and Sapotaceae (Quesada & Lloyd, 2016).

Other rock-derived nutrients that are also important for the metabolic activities of plant organisms are cations (K, Ca and Mg). These nutrients play a role in transport, enzyme production and catalyst, osmotic regulation, chlorophyll and cell walls (Lira-Martins and others, 2022; Campo and others, 2000; Kumar and others, 2015). Cations have a similar

cycling route as P, since the origin of these elements is mostly through the weathering of ancient rocks, which explains the low availability of these nutrients in the Central region of the Amazon basin (Quesada and others, 2012). On the other hand, with respect to N, there is a large demand for this element by plants due to its importance in biological processes, as it constitutes nucleic acid, proteins, and also the chlorophyll molecule, and it is taken up by most plants through the absorption of inorganic ammonium and nitrate ions by the roots (Chapin and others, 1987). Although plants have a large demand for this nutrient, tropical forests, in general, do not present a scarcity of this element to the point of limiting plant growth, since the environment favors the processes of atmospheric deposition and fixation by bacteria in the soil along millions of years of soil development (Vitousek, 1984).

The low nutrient availability in Central Amazon soils can be compensated by a series of efficient nutrient conservation mechanisms such as high nutrient retranslocation rates, when nutrients are moved from senesced tissues by leaves and fine roots (Pires-Santos and others, 2020; Gordon and Jackson, 2000; Brant & Chen, 2015), recycling of litterfall nutrient inputs and fast decomposition (Walker and Syers, 1976; Schaap and others, 2021), and efficient nutrient acquisition mechanisms such as high phosphatase enzyme activity, organic acid exudation and fine root association with arbuscular mycorrhizal (Schaap and others, 2021; Lugli and others, 2020; Reichert and others, 2022). In short, the ecosystem in infertile soils depend on dead and senesced tissue nutrient recycling to maintain itself, so the plant biomass plays a role not only as a C sink but also as a long term nutrient storage pool that will eventually come back to the system via decomposition.

Due to the importance of C and nutrient stored, cycled , retranslocated and recycled in plant biomass, we aimed to quantify the ecosystem-scale stocks and fluxes of C and nutrients, linking element concentrations in each tissues and their respective biomass and annual production in order to estimate ecosystem storage and annual demand. We estimated stocks and fluxes from canopy, stems, fine roots, medium litter, coarse wood debris, and soils of a *terra firme* forest in Central Amazonia, to estimate the balance of nutrient cycling (acquired, stored, and released by plants) and determine which nutrients are the most limiting for net primary productivity in this ecosystem. We hypothesized that the macronutrient that cycles most efficiently in the ecosystem would potentially be the most limiting element for the net primary productivity of this forest.

Material and methods

Study site description

This project was developed at the AmazonFACE (FACE - Free-Air CO_2 Enrichment) study site, a research program that aims to investigate the effects of increasing atmospheric CO_2 in the Amazon. The data used in this work has been collected since 2015 in the permanent AmazonFACE plots during the pre-experimental phase, without CO_2 fertilization. The eight plots of the program are located in the Cuieiras Biological Reserve - ZF2 (2° 35'40"S, 60° 12'28"W) at approximately 70 Km north of Manaus State of Amazonas, Brazil, in the Experimental Station of Tropical Silviculture (EEST/ZF-2), Central Amazon, managed by the National Institute of Amazonian Research (INPA). The plots are circular, with 30 m in diameter (706 m^2) and the vegetation is classified as dense Ombrophilous forest representative of the Amazonian *terra firme* forests (Pereira and others, 2019). The climate of the region is classified as rainy tropical (Peel and others, 2007) based on the Köppen-Geiger system, the average annual precipitation is 2,407 mm (Tanaka and others, 2014), mean annual temperature is 26.7ºC and the dry season with the lowest precipitation (<100 mm per month) corresponds to the period from July to September (Chambers and others, 2004). The botanical families with the greatest number of species are Sapotaceae, Lecythidaceae, Fabaceae, Caesalpiniaceae, Chrysobalanaceae and Euphorbiaceae. These six families contribute to 45% of the local richness (Oliveira & Amaral, 2004). The soil of the region is predominantly of the Ferralsol type, which is a soil representative of 30% of the Amazon basin and chemically similar to around 60% of soils spanning the Amazon basin (Quesada and others, 2011). These soils have low pH, high clay content (> 70%), are well drained, rich in nitrogen (0.2% to 0.3%), but with low concentration of rock-derived elements such as phosphorus (total concentration ranging from 50 to 130 mg/kg) and cations, due to the absence of recent geological activity in Central Amazonia (Quesada and others, 2011; Cordeiro and others, 2020).

Stocks and productivity estimates of the different forest compartments: fine roots, fine and medium litter, coarse woody debris and canopy were obtained through measurements in plots 1 and 2, where project monitoring is more intense, while stem stocks and productivity were measured in all 8 plots. We describe the data source and sample sizes for each ecosystem compartment bellow in Table 1.

Table 1. References, period of data collection and sample sizes of each ecosystem compartment.

Compartiment	Author	Sample size	Years
Nutrient concentration			
Leaves	Garcia and others, unpublished data	70 (mature leaves; 11 trees)	2017
Stems	Lira-Martins and others, 2022	41 (branches)	
Fine litterfall	Garcia and others, unpublished data	24 (composite samples)	2017-2018
Medium litter layer	Present study	11 (litter pieces)	2022
Coarse wood debris	Present study	12	2022
Fine root (0-15 cm <1 mm)	Valverde-Barrantes and others, 2016	15	2022
Soil (0-30 cm)	Quesada and others, 2010		
Stocks			
Leaves	Garcia and others, unpublished data	12 (months)	2017
Stems	Garcia and others, unpublished data	2,045 trees	2019
Fine litter layer	Present study	10 samples	2021
Medium litter layer	Present study	10 samples	2021
Coarse wood debris	Present study	12 samples	2021
Fine root (0-30 cm <2mm)	Miron and others, 2022	29 tubes and collection data	2016-2017
Soil (0-30 cm)	Quesada and others, 2020		
Production			
Leaves	Garcia and others, unpublished data	3 years	2015-2017
Stems	Garcia and others, unpublished data	2 years	2017-2019
Fine litterfall	Garcia and others, unpublished data	4 years	2016-2019
Medium litter layer	Present study	10 samples	2021-2022
Coarse wood debris	Present study	1 sample	2022
Fine root	Miron and others, 2022 and Cordeiro and others, 2020	28 tubes and collection data	2016-2017
Other information			
Ecosystem retranslocation	Santos-Pires and others, 2020	188 (mature leaves; 11 trees) 24 (composite samples – litterfall)	2017-2018
Root mortality	Miron and others, 2022	28 tubes and collection data	2016-2017

Stem stocks and productivity

Since 2015, annual inventories have been conducted in the eight plots (706 m^2 each) for trees with DBH (diameter at breast height) ≥ 10 cm. Approximately 2,045 individuals were measured annually, averaging 256 individuals per plot. The sampled trees were marked with identification plates and had their DBH and taxonomic classification recorded. Stems had their diameter measured once a year in order to determine the periodic increment of all trees that persisted between censuses and also all recruiting trees (trees that migrated from diametric class <10 cm to ≥10 cm). From these data, the stocks and the annual productivity between censuses (mean of years since 2016 until 2019) were estimated applying the allometric equation 1 proposed by Chave and others (2014) for when tree height is known and equation 2 for when height is unknown (we know the height of 1,565 tree), using an estimate factor called environmental stress, as described below.

Equation 1: AGB = 0.0673 * (p*D²H)^0.976

Where, AGB is the above ground biomass (kg), ρ is the wood density (g cm^3), D is the tree diameter at breast height (cm) and H is the height (m) of every tree measured.

Equation 2: AGB = exp(-1.803 - 0.976*E + 0.976*ln(p) + 2.673*ln(D) - 0.0299*(ln(D))^2)

Where, AGB is the above ground biomass (kg), E is the environmental stress factor (for FACE site is -0.1050176 according to Chave´s website), ρ is the wood density (g cm^3), D is the tree diameter at breast height (cm) of every tree measured.

Fine and medium litter standing crop stocks and litterfall productivity

Fine litter is defined here as the fraction of litter smaller than 2 cm in diameter (Luizão, 1995) that includes leaves, branches and reproductive parts. The medium litter layer, on the other hand, comprises the same type of material, but with a diameter ≥2 cm and <10 cm, being mostly twigs.

The stock of litter standing crop was estimated by first sampling the litter layer on the ground (July 2021) using five PVC squares (1 x 1 m tubes) randomly installed in plots 1 and 2 near the aboveground litterfall traps (Methodology adapted from Luizão, 1995). After collection, the material smaller than 2 cm in diameter was separated into leaves, reproductive material, twigs and fine fragmented material (corresponding to organic matter retained on the 3 mm sieve). Soil, fine roots, and organic matter that crossed the sieve mesh were discarded.

To estimate the medium litter layer stock, we sampled the material (≥2 cm and <10 cm) that were inside the PVC squares on the ground (the same square that we used to sample the fine litter layer) in July 2021. To estimate the productivity of the medium litter fractions, the sampling of the same emptied squares was repeated every two months, aiming to capture freshly fallen material resulting in four samples for the period from September 2021 to January 2022. To determine medium litter nutrient concentrations we analyzed the 11 samples collected between July 2021 and January 2022.

For the determination of average fine litterfall production, litterfall data collected during the period from 2016 to 2019 were used, where the contents that fell into the 24 litter traps of 0.5 x 0.5 m installed 1m above the ground in plots 1 and 2 (12 traps per plots) were separated into leaves, branches, reproductive material and others. Litterfall was sampled every two weeks and here we consider litterfall as not only the leaf fraction but as the sum of all plant material that fell inside the traps.

Canopy stocks and productivity

Canopy biomass stocks were estimated from LAI (leaf area index - m^2 m^{-2}) values measured monthly during the year 2017 with hemispherical photos and analyzed by CAN-EYE software. Together with LAI, we also used values of SLA (specific leaf area - m^2 kg^{-1}) obtained from 70 mature leaves around the towers installed in plot 1 and 2 (Menezes and others, 2021). To estimate leaf biomass, equation 4 was used (Malhado and others, 2009).

Equation 4: LB = LAI/SLA*10

Where, LB is the leaf biomass (Mg ha^{-1}), LAI is the leaf area index (m^2 m^{-2}) and SLA is the specific leaf area (m^2 kg^{-1}).

To determine leaf production, in addition to leaf biomass stock data (2015-2017), the monthly mean leaf mortality was used, given from the monthly litterfall production of our litter traps using equation 5 (Malhado and others, 2009).

Equation 5: $LG = LB_t - LB_{t-1}/\Delta t + LM$

Where LG is the leaf growth (Mg ha^{-1} $month^{-1}$), LB is the leaf biomass in two successive measurements (Mg ha^{-1}), Δt is the time interval between these measurements (months) and LM is the leaf mortality, in other words, the litterfall production (Mg ha^{-1} $month^{-1}$).

Nutrient Retranslocation

Nutrient retranslocation is a process that happens before leaf abscission when some elements are moved to another leaf or different plant tissues. This process is an important strategy for nutrient use, since it avoids nutrient losses to the system. Here, we use nutrient retranslocation as a way to correct our estimates of nutrient uptake and cycling in the study area, since some nutrients are constantly recycled within plant biomass, relying less on external inputs from soil.

Retranslocation was previously investigated (Santos, 2020) by sampling green mature leaves from the canopy of 11 known species and newly fallen leaves in litterfall traps (ecosystem-level estimate) installed in plot 1 and 2 at the AmazonFACE area around each meteorological tower. Leaf litter was sampled every 15 days (January 2017 to December 2018), the material was separated and dried at 65°C for 48 h then a composite sample of each month was made for litter nutrient analysis. All nutrient retranslocation data used in this study was collected and analyzed by Pires-Santos, 2020, which consisted of comparing the concentration of nutrients in senesced leaves (litterfall) with the mean concentration of nutrients in mature green leaves (Killingbeck, 1996; equation 6).

Equation 6: RE = (([MDW]-[SDW]))/([MDW])*100

Where, RE is the retranslocation efficiency (%), [MDW] is the mean nutrient concentration in mature green leaves and [SDW] represents the mean nutrient concentration in senesced leaves.

Fine root stocks and productivity

Fine root biomass and productivity in the AmazonFACE plots were determined by minirhizotron analysis (Cordeiro and others, 2020 and Miron and others, 2022). This methodology consisted in the insertion of 10 acrylic tubes with 5 cm in diameter and 2 m in length, which generated an effective depth of 90 cm (in this study, we use only stock and productivity depth 0-30 cm in order to compare with the same soil layer). Five tubes in each plots 1 and 2 s were inserted into the soil at an inclination of 45º. Monthly images were recorded using a moving camera inserted in the tubes during the period from 2016 to 2019 Only fine roots < 2 mm in diameter were considered in the analyses. Biomass was estimated through the allometric relationship between root mass, diameter and length of the fine roots following the approach described by Iversen and others (2008) and Cordeiro and others (2020).

Mortality was estimated as equal fine root productivity (Mg ha^{-1} yr^{-1}) an approach representative of an ecosystem at steady state (Freschet and others, 2021, Miron and others, 2022).

Coarse wood debris stocks and productivity

Coarse wood debris (CWD), or coarse litterfall, was defined here as fallen branches and stems with diameter ≥10 cm and includes lianas, palms, and trees.

The stock of coarse wood debris was calculated as described in Lira-Martins and others (2015), using the line intersect sampling (LIS). The protocol consisted of the installation of two parallel 100 m lines in plots 1 and 2. After installation, debris with diameter ≥10 cm that crossed the line had its diameter measured in July 2021 (12 CWD crossed the 200 m line). Woody materials were classified according to their decomposition status (Chao and Baker, 2008) in order to adjust wood density in the stock calculation

(equation 7 and 8). The decomposition classes used were: Class 1 - Solid wood that has recently fallen, with intact bark and some branches present and no noticeable decay (assumption of net mass loss of less than 10%); Class 2 - Solid wood, but shows bark detachment and decomposition characteristics (assumption of 11 to 30% loss); Class 3 - Non-solid wood that breaks easily when stepped on or handled (+ 30% loss). Thus, of the 12 CWD 4 belonged to class 2 and 8 to class 3, and no class 1 material was found.

Equation 7: $VLIS = (\pi^2 \times \sum d^2)/(8*L)$

Where, VLIS is the volume of CWD that crossed the line intersect (m^3 ha^{-1}), d is the diameter (cm) of each CWD piece and L is the length of the transect line (m).

Equation 8: CWD_{stock} = VLIS x Density

Where, the CWD_{stock} (Mg ha^{-1}), VLIS is the CWD volume and mean density of each CWD class (g cm^3).

For each woody debris that crossed the line, we used a machete or chainsaw (depending on the degree of hardness of the CWD) and collected a section that contained bark, heartwood and sapwood fractions. CWD density was determined through the ratio between dry mass and displaced volume, estimated through the weight of water displacement, resulting from the volume of the CWD sample (Chave and others, 2005). The stock was determined for classes 2 and 3 while CWD production was determined by the ingress of new debris six months after the installation of the line, thus the only new CWD that entered this census had its diameter measured and we assumed that CWD production represents wood mortality.

Soil stocks

Information about the soil properties for the plots was obtained from Quesada and Lloyd, 2016 and Quesada and others (2010; 2011; 2020). Total nutrient concentration and soil bulk density up to 30 cm soil depth (three separate soil layers each 10 cm) were used here. The soil of the region was classified as Ferralsol, this soil type is characterized by strong weathering and has low P concentrations but a high presence of kaolinite and

aluminum, and a high capacity to accumulate organic matter. To calculate nutrient stocks in the soil the equation was used:

Equation 9: $S_N = S_C * 10000 * \rho * l$

Where, S_N is the soil nutrient stock (kg ha^{-1}), S_C is the soil nutrient concentration (g kg^{-1}), ρ is soil bulk density (g cm^{-1}), 10000 is a constant to determine nutrient stocks in kg ha e l is the layer depth (m).

Sampling for tissue and soil nutrient determinations

After sampling, all samples were dried at 65ºC to constant weight, weighed, ground and had the nutrient concentrations determined, except CWD samples that were dried at 105ºC. To determine mean stem nutrient concentration, coarse branch samples from 41 tree species growing in an adjacent location at the same study site were used from trees that had DBH ≥ 10 cm (Lira-Martins and others, 2022).

Fine litterfall nutrient concentration was estimated by 24 composite samples by litter trap, spanning the years 2017 and 2018, while leaves mean nutrient concentration was estimated from 70 mature leaves samples from 11 species (Menezes and others, 2021).

Fine roots samples used for nutrient analysis were collected using the ingrowth core methodology along the transect near the AmazonFACE plots. Fine roots ≤1 mm diameter from 15 ingrowth cores that covered the depth of 0-15 cm were measured and we used composite samples by ingrowth core, spanning two sampling dates (February and May 2016) to analyze for nutrient concentrations. Total soil nutrient concentrations were determined from samples collected from soil pits (Quesada and others, 2010), air dried and was sieved at 2 mm before laboratory analysis.

Plant tissue nutrient analyses

The nutrients analyzed for this study were the macronutrients: nitrogen, phosphorus and cations (Ca, Mg, K) and the micronutrients (Fe, Zn, Mn). For the analysis of phosphorus, cations and micronutrients, the nitric perchloric wet digestion protocol proposed by

Malavolta and others (1989) was applied. Subsequently, cations and micronutrients were determined by atomic absorption spectrometry (AAS, 1100 B, Perkin-Elmer, Ueberlingen, Germany) as described by Anderson and Ingram, (1993). Total phosphorus concentration was determined by colorimetry (Anderson & Ingram, 1993) and quantified by spectrometry (UV-120-01, Shimadzu, Kyoto, Japan). Nitrogen and carbon were determined using an automatic analyzer (VARIO MAX CHN Element Analyzer) as described in Nelson and Sommers, 1996. Most nutrient and carbon analyses (leaves, fine root, fine litterfall, medium litter layer and coarse wood debris) were made at LTSP (Laboratório Temático de Solos e Plantas) in INPA, Manaus, Amazonas. Carbon and nutrient concentrations used in this study are shown in Table 2.

Table 2. Mean element concentrations in plant tissues in Central Amazon forest (AmazonFACE site).

	C	N	P	K	Ca	Mg	Mn	Fe	Zn
				g kg^{-1}				mg kg^{-1}	
Leaf	495.75±3.09	21.42±0.68	0.49±0.02	6.41±0.26	3.24±0.38	1.92±0.09	81.34±7.01	230.79±19.25	42.49±2.68
Stem	472.83±0.92	5.75±0.34	0.16±0.01	1.48±0.14	1.10±0.12	0.55±0.07	n.a	n.a	n.a
Fine litterfall	497.03±3.34	15.28±0.24	0.18±0.01	1.02±0.09	5.02±0.17	1.80±0.03	121.68±4.42	60.06±3.25	8.29±1.02
Medium litter layer	466.80±6.88	9.01±1.09	0.13±0.02	0.21±0.03	3.74±0.54	0.51±0.13	89.15±37.41	789.8±333	16.61±3.27
Coarse Wood debris	468.22±8.59	6.58±0.97	0.09±0.02	0.12±0.02	1.62±0.42	0.23±0.05	39.74±9.94	518.8±214.4	5.27±1.43
Fine root	433.45±2.65	14.16±0.29	0.28±0.01	0.74±0.04	1.70±0.08	0.60±0.06	104.12±11.52	2623.60±94.92	64.63±6.49
Soil	19.26±5.04	1.39±0.38	0.06±0.01	154.39	421.98	32.01	n.a	n.a	n.a

Note: Mean stem nutrient concentration is from coarse branch samples (Lira-Martins et al. 2022) and Soil nutrient concentrations are total nutrients in the soil for the 0-30 cm layer (Quesada et al. 2020). Stem and soil micronutrients are not available (n.a).

Forest nutrients stocks, fluxes, use efficiency and ecosystem demand

Nutrient stocks for each compartment were calculated by multiplying the total stock of each compartment, in Mg ha^{-1}, by the concentration of each nutrient (Table 2), in g kg^{-1}, according to equation 10, while the total annual stock of each nutrient for the forest was determined by summing the mean stocks of each compartment whilst also propagating their respective errors and deviations (see below). Nutrient fluxes were also calculated by multiplying the total production of the compartments, in Mg ha^{-1} $year^{-1}$, by the nutrient concentration data (same as stock estimates). We use the annual fluxes estimates to determine nutrient ecosystem demand, defined here as the nutrient uptake into net primary production (canopy, stem and fine root) discounting leaf retranslocation of the canopy.

Equation 10: $C_N = C_B * C_C$

Where, C_N is the nutrient stock or flux (kg ha^{-1} or kg ha^{-1} yr^{-1}, respectively), C_B is the total stock or production of each compartment (Mg ha^{-1} or Mg ha^{-1} $year^{-1}$, respectively) and C_C is the nutrient concentration (g kg^{-1}).

The nutrient use efficiency (kg g^{-1}) was calculated as the ratio between biomass production (kg ha^{-1} yr^{-1}) and nutrient flux (kg ha^{-1} yr^{-1}), as shown in Vitousek, 1982 and Harrington and others, 2001 for each compartment (stem, fine root and fine litterfall) and also for the whole ecosystem (mean between compartments).

Ecosystem demand is the quantities of nutrients that need to be uptake to promote plant grow and was calculated by the difference between nutrient flux in canopy and retranslocation rate, after sum this value with nutrient flux by stem and fine root. On the other hand soil demand was calculated by difference between ecosystem demand and nutrient flux via dead material.

Statistical analysis and propagation of uncertainties

All statistical analyses were made using R 3.4.4 (R Core Team). We determined if the data variability showed a normal distribution by measures of dispersion as confidence

intervals, standard error, standard deviation, media and tested the normality of data by p value in linear models. If p value was ≤ 0.05 this suggest a high significant level, so a high data confiability

Propagation of uncertainties was made to estimate errors when i) we sum or divide and ii) multiply or divide a number of variables (Hogan, 2006 and Malhi and others, 2009). In the first case, the absolute uncertainties are propagated in quadratura (equation 11). In the second case, the relative uncertainties are propagated in quadratura (equation 12). In this study, we used the first equation when we estimate ecosystem stocks and fluxes by the sum of each compartment. The second equation was used when we calculate stocks and fluxes by multiplying nutrient concentration by biomass or production.

Equation 11: $(\Delta y)^2 = \sum_{i=1}^{n} (\Delta x_i)^2$

Where, Δy is the resulting error (in the variables unit) and Δx is the variable error.

Equation 12:$(\Delta y/y)^2 = \sum_{i=1}^{n} (\Delta x_i/x_i)^2$

Where, Δy is the resulting error (in the variables unit), y is the variable value, Δx is the variable error and x is the variable value.

Results

Carbon fluxes

Inputs: Net primary productivity

Total ecosystem net primary productivity (NPP_{total}) estimated for our study area was 8.09±0.26 Mg C ha^{-1} yr^{-1}. When partitioning for the different plant compartments, from higher to lower, we found that canopy NPP was 4.65±0.14 Mg C ha^{-1} yr^{-1}, stem NPP was 1.73±0.09 Mg C ha^{-1} yr^{-1} and fine root NPP for the 0-30 cm soil layer was 1.71±0.20 Mg C ha^{-1} yr^{-1} (Figure 1).

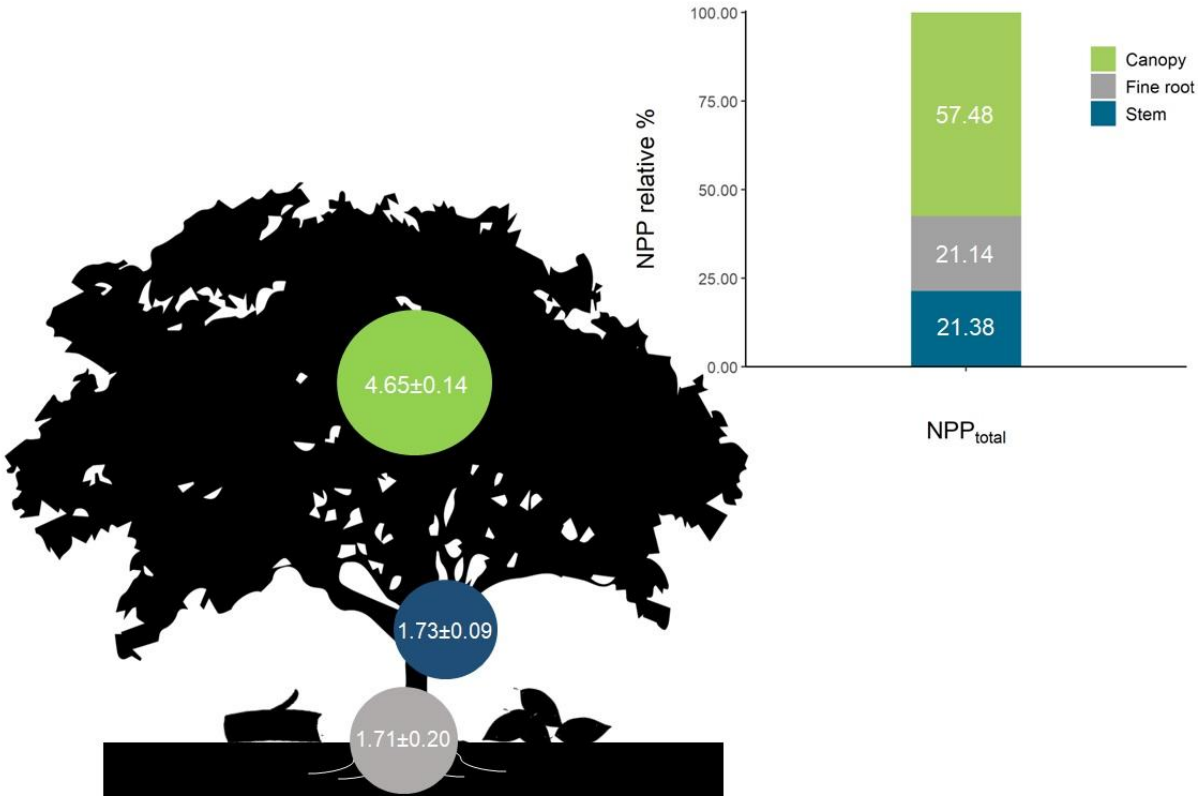

Fig. 1. Net primary productivity in a Central Amazon forest near Manaus, Brazil in Mg C ha^{-1} yr^{-1}. On the left, colored circles correspond to the NPP of each compartment: green represents canopy, blue for stems and gray for fine roots. On the right, bars correspond to total NPP (100%) and each color represents the percentage of compartments contribution to NPP$_{total}$, following the same color code described above.

Outputs: tissue mortality

Fine and medium litter annual production was 4.56±0.59 and 0.22±0.10 Mg C ha^{-1} yr^{-1} respectively (Fig. 2). Coarse wood debris was estimated at 0.35 Mg C ha^{-1} yr^{-1}, which could be seen as an underestimation since after six months of monitoring, only one new CWD piece entered in our LIS. For fine roots, since stocks are in equilibrium, we assumed fine root mortality being equal to NPP$_{fineroot}$ resulting in 1.71±0.20 Mg C ha^{-1} yr^{-1} (Fig. 2).

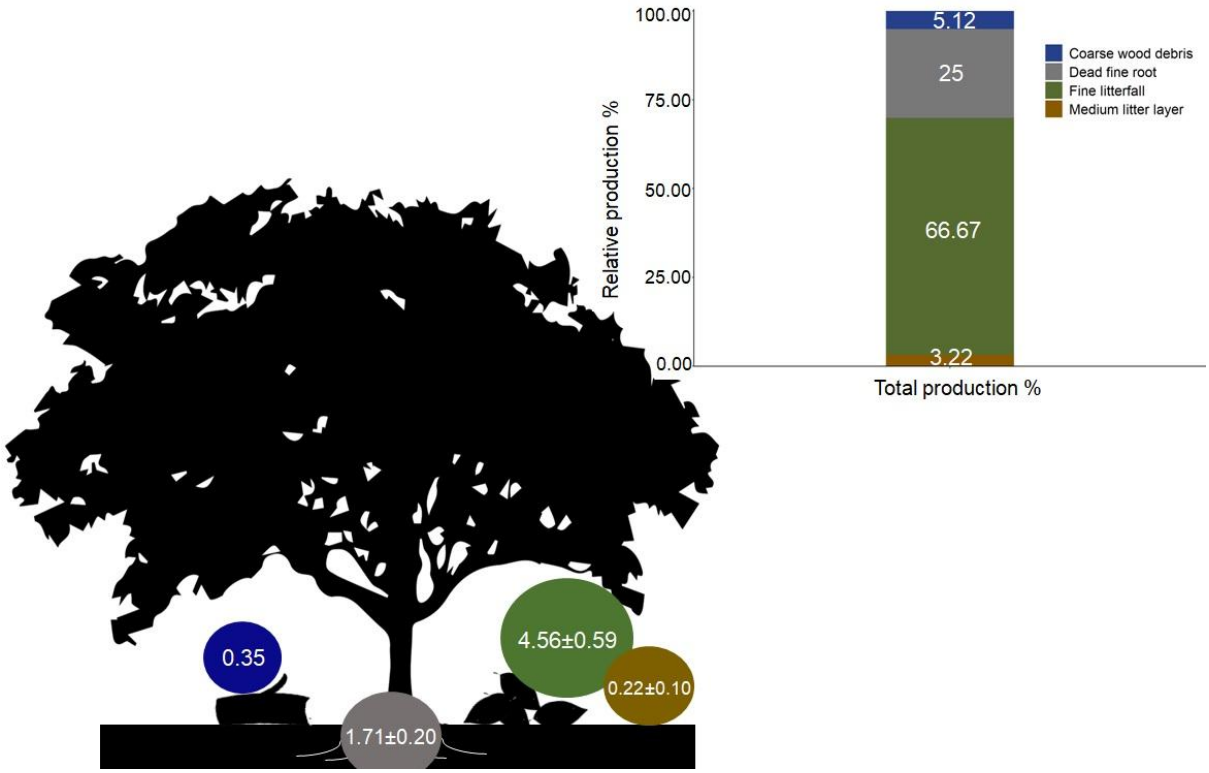

Fig. 2. Tissue mortality in a central Amazon forest near Manaus, Brazil in Mg C ha^{-1} yr^{-1}. On the left, colored circles correspond to the production of each dead compartments: Dark green is fine litterfall, dark blue is coarse wood debris, dark gray is dead fine root and dark orange is medium litter fraction. On the right, bars correspond to the total C production and each color represents the percentage of compartments contribution to ecosystem production, following the same color code described above.

Carbon stocks

As a result of C inputs and outputs in this forest, total C stocks, i.e. sum of all compartments, reached 264.71±15.47 Mg C ha^{-1}. Canopy corresponded to 2.46±0.06 Mg C ha^{-1} or 1.17% of total C stocks in our study site. Stems contributed with 188.81±14.04 Mg C ha^{-1} (89.47%). Fine root C stocks estimated were 2.13±0.11 Mg C ha^{-1} (1.01%) to the depth 0-30 cm. Fine and medium litter layers showed, respectively, 2.47±0.30 (1.17%) and 0.28±0.09 Mg C ha^{-1} (0.13%). For coarse wood debris, we found a total stock of 9.62±0.64 Mg C ha^{-1} (4.56%) and for soil to the depth 0-30 cm a stock of 58.94±15.47 Mg C ha^{-1} (2.49%).

Nutrient fluxes

Inputs

Mean nutrient concentrations used to calculate nutrient stocks and fluxes, are shown in material and methods (Table 2). We used nutrient concentrations in each compartment

(leaf, stem and fine root) and then extrapolated our measurements for nutrient inputs to the system via tissue productivity (Table 3).

Nitrogen showed greater input into the system compared to other nutrients, contributing with 279.41±11.09 kg ha^{-1} yr^{-1}, resulting in N being the most abundant element in all plant tissues after C (Table 3). Contrary to N, P was the macronutrient cycling in lower amounts, contributing to 6.32±0.27 kg ha^{-1} yr^{-1}. For the other macronutrients we observed the following pattern, from higher to lower cycling rates: K > Ca > Mg and in among micronutrients we observed the following order: Fe > Mn > Zn (Table 3; note that we do not have micronutrient concentrations for stems and therefore their inputs are underestimated and we use leaf nutrient concentrations to estimate canopy stocks and fluxes).

Table 3. Nutrient used in annual net primary production in a Central Amazon forest.

	C	N	P	K	Ca	Mg	Mn	Fe	Zn
	Mg ha^{-1} yr^{-1}				kg ha^{-1} yr^{-1}				
Canopy	4.65±0.14	202.63±8.65	4.64±0.23	60.64±3.01	21.19±3.65	18.16±0.99	0.77±0.07	2.18±0.19	0.40±0.03
Stem	1.73±0.09	20.99±1.62	0.58±0.05	5.40±0.58	4.02±0.48	2.00±0.27	n.a	n.a	n.a
Fine root	1.71±0.20	55.79±6.75	1.10±0.14	2.92±0.38	6.69±0.86	2.36±0.37	0.41±0.07	10.34±1.29	0.26±0.04
Total	**8.09±0.26**	**279.41±11.09**	**6.32±0.27**	**68.96±3.09**	**31.90±3.78**	**22.52±1.09**	**1.18±0.09**	**12.52±1.30**	**0.66±0.05**

Note: After ± symbol is the propagated standard error. We did not estimate micronutrient used by stems.

Nutrient return

We also estimated nutrient returned back to the system using each tissue mortality rate (fine litterfall, medium litter layer, coarse wood debris and dead fine root) and their respective nutrient concentrations (Table 4). We observed total fluxes of nutrients in the following order, from higher to lower: fine litterfall > fine root litter > medium litter layer > coarse wood debris. Among the studied macronutrients, N was the element with higher cycling rates through plant litter (204.72±19.78 kg ha^{-1} yr^{-1}) and P had the lowest (2.88±0.08 kg ha^{-1} yr^{-1}). The other macronutrients showed the following cycling pattern: Ca > Mg > K. When comparing the cycling rates derived from tissue mortality from different plant compartments, micronutrient fluxes showed the same pattern observed for macronutrients, with higher fluxes from fine litterfall and smaller from coarse wood debris, and among micronutrients, followed the order Fe > Zn > Mn.

Table 5. Nutrient returned to the system via plant tissue mortality in a Central Amazon forest.

	C	N	P	K	Ca	Mg	Mn	Fe	Zn
	Mg ha^{-1} yr^{-1}				kg ha^{-1} yr^{-1}				
Fine litterfall	4.56±0.59	140.12±18.48	1.65±0.24	9.35±1.48	46.03±6.22	16.51±2.18	1.12±0.15	0.55±0.08	0.08±0.01
Medium litter layer	0.22±0.10	4.33±2.05	0.06±0.03	0.10±0.05	1.79±0.86	0.25±0.13	0.04±0.03	0.38±0.24	0.01±0.004
Coarse wood debris	0.35	4.48	0.07	0.09	1.19	0.17	0.03	0.38	0.004
Dead fine root	1.71±0.20	55.79±6.75	1.10±0.14	2.92±0.38	6.69±0.86	2.36±0.37	0.41±0.07	10.34±1.29	0.26±0.04
Total	**6.84±0.63**	**204.72±19.78**	**2.88±0.08**	**12.46±1.53**	**55.70±6.34**	**19.52±5.01**	**1.60±0.03**	**11.65±1.32**	**0.35±0.04**

Note: After ± symbol is the standard error. Coarse wood debris do not have a standard error because we estimate this compartment using only one individual piece of CWD collected during our sampling interval.

Nutrient recycling: Retranslocation and annual NPP nutrient demand

We estimated leaf ecosystem retranslocation for C and macronutrients as the proportion of elements that were re-absorbed prior to leaf senescence (Santos, 2020). Carbon and Ca showed negative values of -0.46±% and -124.56%, respectively, which represents element accumulation in the tissue during leaf senescence. For the other macronutrients, mean retranslocation, from higher to lower was: K 84.06% > P 63.19% > N 28.52% > Mg 6.06%. For micronutrients, Zn and Fe showed 80.45% and 73.92%, with Mn accumulation (-49.89%). We then compared mature green leaf and litter nutrient concentrations in relative terms, with the difference being the fraction of nutrients in green leaves that were recycled (via retranslocation), with the remaining representing, therefore, nutrient that was newly incorporated by the system (Figure 5).

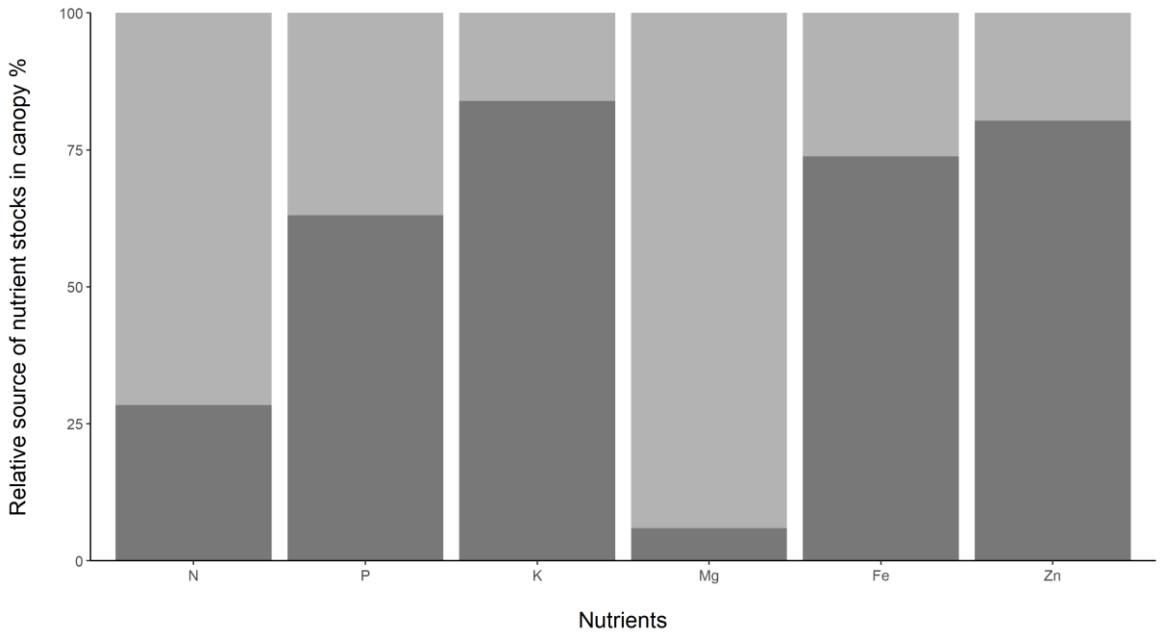

Fig. 3. Nutrient retranslocation in %. Bars correspond to the percentage of nutrient retranslocated before leaf senescence (dark grey) and the percentage of nutrient stocks that were newly incorporated in leaves (light grey) in a Central Amazon forest.

We also estimated the ecosystem nutrient demand to grow during one year (Table 4). Since, according our results, Ca and Mn were not retranslocated by leaves, all of the ecosystem nutrient demand was supplied by the soil. P showed the lower ecosystem demand between macronutrients, since almost half of P used by the leaves came from retranslocation, while N showed the higher ecosystem demand value. Among micronutrients Zn was the least supplied by the soil, while Fe showed the higher demand.

Table 4. Ecosystem nutrient demand in a Central Amazon forest.

	N	P	K	Ca	Mg	Mn	Fe	Zn
				kg ha^{-1} yr^{-1}				
NPP_{total}	279.41±11.09	6.32±0.27	68.96±3.09	31.90±3.78	22.52±1.09	1.18±0.09	12.52±1.30	0.66±0.05
Retranslocated by leaves (%)	28.52±3.69	63.19±8.95	84.06±13.35	0	6.06±0.81	0	73.92±11.49	80.45±19.89
Stock soil 0-30 cm (kg ha^{-1})	4,253.40±1.166.48	183.6±30.76	472,427.8	1,291,256	97,940.15	n.a	n.a	n.a
Dead material nutrient flux	195.91±19.67	2.75±0.28	12.27±1.53	39.34±6.28	43.94±3.25	1.53±0.17	10.89±1.29	0.34±0.04
Ecosystem demand	221.62±30.06	3.39±0.50	17.99±2.97	31.90±3.78	21.42±3.06	1.18±0.09	10.91±2.04	0.34±0.09
Soil demand	25.71±35.92	0.64±0.57	5.72±3.34	-7.44±7.33	-22.52±4.46	-0.35±0.19	0.02±2.41	0±0.09

Note: Ca and Mg were not retranslocated from leaves during senescence, so it retranslocation amounts was 0%. After ± symbol is the standard error. Negative values in soil demand represents nutrient recycling via dead tissue sustain ecosystem demand.

Nutrient stocks

Nutrient stocks were also estimated using total nutrients concentrations (Table 2) and plant biomass stocks of each ecosystem compartment, as well as soil stocks. Total ecosystem stocks of macronutrients followed the order, from higher to lower: Ca > K > Mg > N > P (Table 6). If we exclude soil stocks, total nutrients stored in plant compartments follow a different order: N > K > Ca > Mg > P (Table 6). For micronutrients, we found the following order of stocks for the whole ecosystem: Fe > Mn > Zn.

Table 6. Element stocks in different above and belowground ecosystem compartments in a Central Amazon forest.

	C	N	P	K	Ca	Mg	Mn	Fe	Zn
	Mg ha^{-1}				kg ha^{-1}				
Leaf	2.46±0.06	106.24±4.11	2.43±0.11	31.79±1.47	11.11±1.90	9.52±0.49	0.40±0.04	1.15±0.09	0.21±0.01
Stem	188.81±14.04	2,296.94±218.08	63.89±6.21	590.98±71.09	439.24±57.98	219.62±32.37	n.a	n.a	n.a
Fine litter layer	2.47±0.30	75.94±9.40	0.89±0.12	5.07±0.77	24.95±3.18	8.95±1.11	0.61±0.08	0.29±0.04	0.04±0.01
Medium litter layer	0.28±0.09	5.41±1.83	0.08±0.03	0.13±0.04	2.24±0.78	0.31±0.12	0.05±0.03	0.47±0.25	0.01±0.004
Coarse wood debris	9.62±0.64	121.36±17.15	1.83±0.42	2.44±0.43	32.90±8.69	4.67±1.04	0.81±0.21	10.54±4.39	0.11±0.03
Fine root	2.13±0.11	69.67±3.82	1.38±0.09	3.64±0.27	8.36±0.58	2.95±0.33	0.51±0.06	12.91±0.81	0.32±0.04
Soil	58.94±15.47	4,253.40±1.166.48	183.6±30.76	472,427.8	1,291.256	97.940.15	n.a	n.a	n.a
Total	**264.71±20.90**	**6,928.96±1.186.87**	**254.10±77.48**	**473,061.85±71.11**	**1,291,774.80±58.75**	**98,186.17±32.41**	**2.38±0.24**	**25.36±4.47**	**0.69±0.05**

Note: After ± symbol is the standard error. Nutrient concentration not available (n.a) to stems and soil.

Nutrient use efficiency

Mean ecosystem macronutrients use efficiency followed the order, from higher to lower efficiency: P > Mg > K > Ca > N (Fig. 4). When looking at different plant compartments, nutrient use efficiency varied greatly (Fig. 4). Phosphorus was used more efficiently in all plant compartments separately, with Mg as the second most efficiently used nutrient in leaf, stem and fine root, but K showing high efficiency in litterfall.

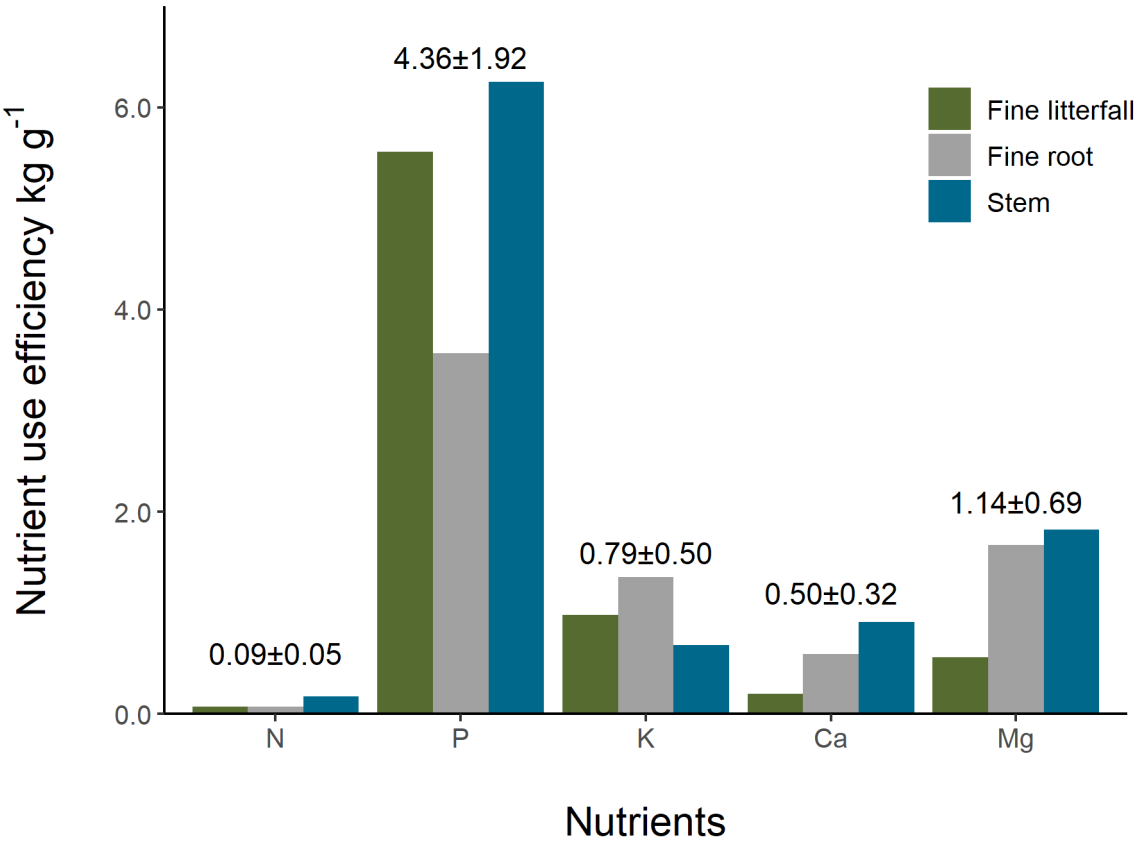

Fig. 4. Nutrient use efficiency in g kg^{-1}. Bars correspond to nutrient use efficiency in each plant compartment: blue is stem, gray is fine root and dark green is fine litterfall. The values above bars are mean ecosystem nutrient use efficiency estimates.

For mean ecosystem micronutrient use efficiency (Fig. 5) we found the following pattern, from higher to lower: Zn > Mn > Fe. The same pattern was found when analyzing micronutrients use efficiency for each plant compartment (Fig. 5).

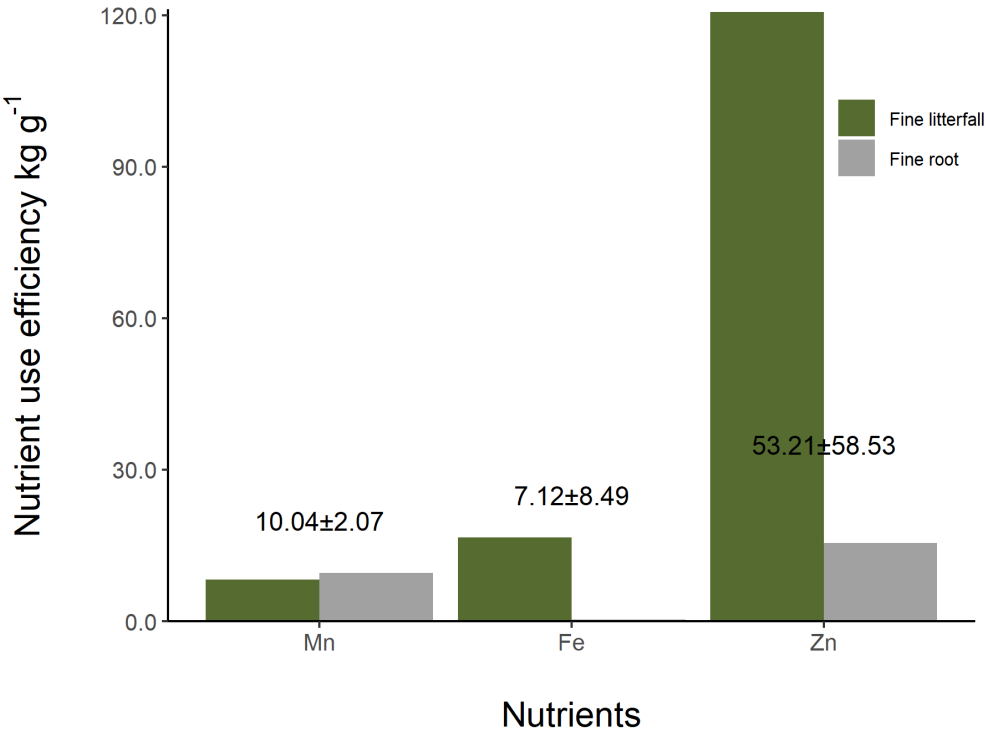

Fig. 5. Nutrient use efficiency in g kg^{-1}. Bars correspond to nutrient use efficiency in each plant compartment: gray is fine root and dark green is fine litterfall. The values above bars are mean ecosystem nutrient use efficiency estimates.

Discussion

Carbon balance between inputs and outputs

In our study area, total ecosystem NPP estimated (8.09±0.26 Mg C ha^{-1} yr^{-1}) was lower than estimates previously found in a nearby site in Manaus (10.1±1.4 Mg C ha^{-1} yr^{-1} by Malhi and others, 2009) and other parts of the Brazilian Amazon (Tapajós 14.4±1.3 and Caxiuaña 10±1.2 Mg C ha^{-1} yr^{-1} by Aragão and others, 2009). Such differences were to be expected since when compared to such above-mentioned studies, we did not have data on branches, coarse roots and volatile organic compounds to add to the total NPP calculations at our site. Another point is that in Malhi´s study, root productivity was taken as an average of Caxiuanã and Tapajós sites. As a result of the inputs and outputs of plant biomass in our study area, total C stock estimated for this ecosystem was 205.77±14.06 Mg C ha^{-1} (sum of all plant compartments except soil), close to previously reported by Malhi and others, 2009 (around 208±10.63 Mg C ha^{-1}), although the contribution of each compartment for total C stocks varied slightly due to methodological discrepancies discussed above.

In this study we assumed that this *terra firme* forest is at steady state. Such assumption comes from the fact that high leaf senescence rates that result in leaf litterfall production is usually accompanied by high new leaf production, with peaks of both processes in the dry season (Wu and others, 2016). Following this same pattern, NPP_{canopy} estimated in this study was 4.65±0.14 Mg C ha^{-1} yr^{-1}, with similar values for fine litterfall production of 4.56±0.59 Mg C ha^{-1} yr^{-1}. NPP_{canopy} along Amazon tropical forest was estimated between 0.48±0.17 Mg C ha^{-1} $month^{-1}$ (5.76 Mg C ha^{-1} yr^{-1}) and 0.20±0.07 Mg C ha^{-1} $month^{-1}$ (2.4 Mg C ha^{-1} yr^{-1}) according to Girardin and others, 2016 and similar to Malhi and others, 2009 estimate of 3.6±0.7 Mg C ha^{-1} yr^{-1}. Although NPP_{canopy} was higher than any other compartment, canopy stock was low (2.46±0.06 Mg C ha^{-1}) indicating the fast turnover of this compartment. Fine litterfall production was estimated as in equilibrium with leaf production and was higher than Malhi and others, 2009 estimates for Manaus site (3.6±0.7 Mg C ha^{-1} yr^{-1}) and Chave and others, 2010 (7.13±2.53 Mg C ha^{-1} yr^{-1}) for forest growing in similar soil type, but lower than another *terra firme* forest in southern Amazon, estimated in 5.7 Mg C ha^{-1} yr^{-1} (Selva and others, 2007). C stocks in the fine litter layer in our study were 2.47±0.30 Mg C ha^{-1}, while in a second-growth Central Amazon Forest was higher, around 3.21 Mg C ha^{-1} (Tapia-Coral and others 2005). Medium litter layer values were not reported in other studies, making it difficult to put the contribution of such compartments into the context of annual ecosystem component production and storage.

For NPP_{stem}, we estimated 1.73±0.09 Mg C ha^{-1} yr^{-1}. Pantropical databases showed an average NPP_{stem} of 2.5±0.7 Mg C ha^{-1} yr^{-1} in mature forests (Anderson-Teixeira and others. 2016), this represents around 32% of NPP_{total}, which in comparison to our results yields in about 10% higher values. C stocks are more expressive in stems (188.81±14.04 Mg C ha^{-1}) than in any other compartment and together with stocks of coarse woody debris (9.62±0.64 Mg C ha^{-1}), contributed to around 75% of total C stock, confirming their important roles in carbon balance and C sink and source.

Coarse wood debris are also hard to be compared in terms of their productivity since studies on this topic are scarce. Two studies in tropical forests were conducted by Chambers and others, 2000 and Chao and others, 2009, where CWD mortality mass input was estimated for a *terra firme* Amazon forest, where they found a higher mortality mass input (3.6 -5.2±0.3 Mg ha^{-1} yr^{-1} when compared to our estimates (0.35 Mg ha^{-1} yr^{-1}). In regards to its stocks, CWD showed the second higher C stock above ground, and even though CWD decomposition is usually slow and depends on climate, decomposers community diversity and chemical and structural wood properties, CWD can be an important component in the C

cycle (Palace and others, 2012). CWD stocks and production are stochastic processes. (i.e.. events of random character that depend on specific circumstances such as the emergence of clearings; Davis and others, 2015), thus great variation is expected, even when comparing nearby sites. Due the very short time frame of our CWD sampling and relatively small covered area, we recognize that our values reported here may be underestimated, when compared to other studies across the Amazon Basin. For instance, a mean CWD stock of 14±2 Mg C ha^{-1} was reported for Manaus, Brazil (Malhi and others, 2009), while in Lira-Martins and others, 2015 CWD stocks ranging from 5.73±0.94 to 14.11±3.15 Mg C ha^{-1} were estimated for a nearby site, at Ducke Reserve near Manaus, Brazil. For our study and Lira-Martins and others, 2015 CWD stocks were estimated based on fallen coarse wood debris using the line intercept sampling method (Van Wagner, 1968). Whereas in Malhi and others, 2009 the results are from Chambers and others, 2001 who performed random fallen tree sampling. Another explanation for our low values (stock of 9.62±0.64 Mg C ha^{-1} and flux of 0.35 Mg C ha^{-1} yr^{-1}) was a low CWD input rate or even high CWD decomposition rates (Baker and others, 2007).

$NPP_{fineroot}$ estimated in our study area was 1.71±0.20 Mg C ha^{-1} yr^{-1} for the 0-30 cm soil depth, close to values estimated for pantropical $NPP_{fineroot}$ of 2.43±0.23 Mg C ha^{-1} yr^{-1} (Huaraca Huasco and others, 2021). Comparisons among $NPP_{fineroot}$ have to be done carefully due to methodological differences (Huaraca Huasco and others, 2021). Moreover, a large part of root biomass and production can be neglected when using only 30 cm depth. For instance, using minirhizotrons capturing root images up to 90 cm soil depth, around 46.1% of root biomass socks and 40.6% of production was found in the layer 30-90cm in our study site (Cordeiro and others, 2020). Following the steady state assumption that canopy and litter production are similar, we also assumed that the fine root mortality rate was equal to $NPP_{fineroot}$ 1.71±0.20 Mg C ha^{-1} yr^{-1}. Such assumption was recently corroborated by a study at the same study site which showed similar values for both fine root productivity and mortality but also for canopy and litterfall production (Miron, 2022).

Although in our study site the vast majority of C is stored in plants (around 75% of total C), we estimated C stocks in soils of 58.94±15.47 Mg C ha^{-1} to a depth of 0-30 cm. Soil C stocks along the Amazon basin vary from 41±0.44 Mg C ha^{-1} in Plinthosols to 115±0.63 Mg C ha^{-1} in Leptosols, with our sites showing intermediate values, but slightly higher for the mean found for the same soil type (Ferralsols) of 47.3±0.26 Mg C ha^{-1} (Quesada and others, 2020).

In summary, plant biomass and soil play an important role in C sink, with wood tissues and soils displaying the largest stocks although with slow turnover rates, on the other hand stems and coarse wood debris showed lower production rates, while the more dynamics compartments (canopy, litter and fine root) had lower stocks and higher production. C stock and flux are components in C dynamics and its balance is limited by many resources available like light, water and nutrients, which we will discuss in more detail in the following sections.

Nutrient balance between inputs and outputs

Despite the importance of understanding C allocation and balance, nutrients stored in soils and plant biomass or necromass also play a significant role controlling NPP, with potential to further limit C assimilation and, consequently, carbon sink in future climate (Chapin and others, 1987). Total ecosystem N stocks (Table 6), inputs via NPP and outputs via tissue mortality (Table 3 and 5) were the highest among all the nutrients analyzed in this study, suggesting that plants indeed need N in great amounts to satisfy their metabolism, since N is an important component of plant protein, nucleic acids and phospholipids (Novoa and Loomis, 1981). Due to the high availability of N in soils of our study site, high amounts of N can also be lost via leaching, resulting, therefore, in the lowest nutrient use efficiency among the elements studied here. Such findings confirm that N has an open cycle and is potentially not among the nutrients that might limit ecosystem productivity in this lowland tropical forest (Vitousek, 1984; Vitousek and Sanford, 1986). Further evidence comes from $\delta^{15}N$ stable isotopes, with soils around our study site showing quite an open N cycling, ranging from a $\delta^{15}N$ of 8.2 to 11.1 % (Quesada and others, 2010), with undisturbed *terra firme* forests of the Amazon usually taken as not being limited by N (Martinelli and others, 1999). However, although N availability is high, it does not exclude the possibility that it could regulate some specific process in different seasons (Townsend and others, 2011) or exclude N limitation in other tropical forest (Wright, 2019).

Phosphorus, like N, is essential for plant metabolism, being a component of ATP, protein, sugar and nucleic acid (Yan and others, 2016). However, forests in the Central portion of Amazon, including in our study site, grow in soils with very low P availability (Table 6) (Quesada and others, 2010; 2012). Soil P availability in young terrestrial ecosystems rely on the weathering of parent material (Walker and Syers, 1976). However, in

old and already highly weathered soils, organic P in soil, which comes from litterfall inputs and decomposition, becomes the main source of P for these forests (Walker and Syers, 1976; Quesada and others, 2010; Schaap and others, 2021). To supply the demand of plant P, this element is usually recycled in high amounts (Fig. 3) via retranslocation prior to leaf and root senescence (Gordon and Jackson, 2000). Although nutrient retranslocation in roots is hardly measured in tropical forests and was not included in our study, root P reabsorption showed greater rates with increasing latitudinal, temperature and precipitation gradients (Brant and others, 2015), suggesting a potentially high importance of this process in tropical forests. As a result of low soil P availability and high leaf restranslocation, P showed the highest nutrient use efficiency among the elements studied here (Fig. 4), pointing to a closed cycle and the great potential for P to be the most limiting nutrient affecting net primary production in our ecosystem.

Potassium is also an important element in photosynthesis, ion transport, enzyme catalyst and osmotic regulation, playing an important role in plant water storage (Lira-Martins and others, 2022). Potassium showed the lowest return to the soil among the cations and higher retranslocation rates, however K is a very mobile nutrient in plant tissues (Chapin, 1980), and for this reason, high rates of K leaching out of the leaves could have resulted in overestimation of our retranslocation values. Contrary, Ca showed a high return to the soil and negative leaf retranslocation (accumulation in litter), probably because Ca is the least mobile element in plant tissues among our studied nutrients, not being able to move through cell walls during leaf senescence (Kumar and others, 2015). As a result, Ca stored in soil was higher than Ca inputs via NPP, especially when compared to NPP input of N and K. On the other hand, Mg, which plays an important role in chlorophyll and some enzymes production (Campo and others, 2000), showed a high use efficiency (after P) but had a low retranslocation value. Magnesium returned to the soil reaching similar values when compared to Mg uptake, being 19.52±5.01 and 22.52±1.09 kg ha^{-1} yr^{-1} respectively, which suggest an equilibrium between input and output for this nutrient.

Among micronutrients, Fe, which is an important nutrient controlling some steps during photosynthesis and respiration processes (Forieri & Hell, 2014), was the one used less efficiently and showed higher stocks and fluxes, which could be explained by the high abundance of this element in the soil (Quesada and others, 2010). Manganese plays a role in decomposition process and microbial activities while Zn shows the same role plus an important component in many plant proteins (Kaspari and others, 2008 and Sayer and others,

2020). In most compartments, Mn showed an intermediate use efficiency, while Zn showed the highest efficiency among micronutrients.

In canopy, some nutrients such as P and K tend to decrease concentrations with leaf longevity, when photosynthetic rates start to decline after mature age, leading to leaf senescence when nutrient retranslocation occurs (Menezes and others, 2021 and Pires-Santos and others, 2020). For micronutrients Fe and Zn, on the other hand, concentrations tended to increase with leaf age (Menezes and others, 2021 and Pires-Santos and others, 2020). Nonetheless, due to its fast turnover, canopy showed the highest nutrient fluxes among all plant tissue compartments studied here.

In this study, stems stocks and fluxes were calculated using coarse branch nutrient concentrations. In general, stems have lower concentrations than branches and depending on the portion of the stems sampled, nutrient concentrations might also change (Martinelli and others, 2006). For instance, higher concentrations of P and K were found in the heartwood of tree species in tropical forests (Bauters and others, 2022). In our estimates, stem nutrient concentrations were lower than more dynamic tissues (leaf and fine root) and P showed the lower concentration, while K had higher concentration among the rock-derived nutrients studied. Contrary to nutrient fluxes, stems stored nutrients in greater amounts when compared to other live tissues. Among nutrients, P showed the lowest stocks in stems, while N showed the highest stocks followed by K, Ca and Mg respectively. Such great storage capacity could point to the importance of stems as a long-term storage of nutrients, or can be taken as an important sink when forest biomass is aggrading, as has been noted across Amazonia (Brienen and others, 2015). It has been suggested that tree species growing in soils with low availability of rock-derived nutrients generally have a high stock of such nutrients in stems, which results in a high contribution of this compartment to the total nutrient stock in the ecosystem (Bauters and others, 2022). Due to the importance of stems in storing macronutrients in this Amazon forest, CWD followed a similar pattern, with high N stock of 121.36±17.15 kg N ha^{-1} and P stock of 1.83±0.42 kg P ha^{-1}. Such values were slightly higher than the reported for another forest in Central Amazon (85±10 kg ha^{-1} and 0.9±0.1 kg ha^{-1}. respectively; Buscardo and others, 2016), pointing to the potential importance of CWD as a sink and eventually source of nutrients along the wood decay process.

Fine roots are responsible for nutrient uptake and transfer from soils to plant, but due to its fast turnover, fine roots can also become an important source of nutrients being recycled through organic matter decomposition (Joslin and Henderson, 1987). Moreover, the presence of fine roots growing in the leaf litter layer, a common feature of these Central Amazon

forests, could also stimulate P and cations release, acting directly and indirectly during nutrient cycling (Martins and others, 2021). Since higher fine root stocks were commonly found when P and/or K soil availability was low in tropical forests, nutrients stored in fine roots and released via root decay could also become crucial sources of nutrients in such environments (Huaraca Huasco and others, 2021).

Soil nutrient stocks shown in this study represent total nutrient concentration in soil, being a proxy for the potential nutrients that can be used by the plants. However, due to nutrient immobilization in microbial biomass, adsorption to clay surface, organic matter, or occlusion into mineral matrix, which are common processes in Central Amazon soils (Buscardo and others, 2016), soil nutrient availability is usually low, especially for rock-derived elements (Quesada and others, 2011). For instance, in Ferralsols usually around 50 to 80% of total P is made by residual or occluded P, which is not likely to be available to plants, and most of the remaining P is thought to be of slow turnover (Quesada and others, 2010). Available cations are also only a small fraction of the total concentrations (Quesada, 2008). Therefore, multiple microbial and plant strategies, such as nutrient retranslocation, phosphatase enzyme activity, organic acid exudation and fine root association with arbuscular mycorrhizal are strategies often adopted to better take up and use nutrients in soil-poor environments and reduce the chance of nutrient losses (Schaap and others, 2021; Lugli and others, 2020; Reichert and others, 2022). Moreover, the abovementioned relative importance of each plant compartment acting as both sink and source of nutrients might act towards the maintenance of this forest ecosystem functioning.

With CO_2 increase emissions and, consequently, forest fertilization by CO_2, limitations in net primary production may be due nutrient availability, mainly P and cations. In this way we estimated that the nutrient used in annual net primary production that in part was taken up from soil, and another expressive fraction being restranslocated in leaves, resulting in what we call here ecosystem nutrient demand, or the amount of nutrients used by this forest coming from new (not-reused) sources (soils and dead tissues decomposition) (Quesada and others, 2010; Schaap and others, 2021). Our estimates suggest that although soil presented a high reservoir of total nutrient (nutrients that potentially can be used) a great part of P and cations is unavailable and will become more along the time (Walker and Syers, 1976), so this soil forest appears limited by P and the improve of ecosystem net primary production is only possible if plants increase cycling rates, nutrient use efficiency and invest in strategies of nutrient release and uptake. Other ways to improve nutrient availability refers to plants plasticity in retranslocation rates and natural selection of individuals with high

retranslocation capacity, what makes these species potentially more responsive to eCO_2. Different species showed different retranslocation rates for all nutrients, with P retranslocation, for instance, ranging from 10% to 70% in our study area (Santos, 2020).

In short, nutrients play an essential role in C assimilation by plant process and its availability modulates the net primary production, with nutrients, and particularly P limiting this Central Amazon forest. Wood compartments showed a high nutrient stock, although it also showed a slow turnover. Dynamic tissues, on the other hand showed low stocks and larger fluxes, with N being the nutrient with higher storage and faster turnover, while P showed an inverse pattern, with cations being intermediate between N and P. Adding information about retranslocation rates and nutrient use efficiency estimates, P performs a high retranslocation rate being the most efficient nutrient used in this forest. The importance of P as the most limiting factor in Central Amazon forests is also corroborated by a recent nutrient manipulation experiment, which report strong NPP responses to P (Cunha and others, accepted) with significant increases in leaf and fine root NPP only two years after P additions, with no response found for N or cations (Ca, Mg, K). With regards to micronutrients, Zn seems to be a crucial nutrient in NPP having low stock, low flux and high retranslocation rates and nutrient use efficiency. A better understanding of ecosystem nutrient balance is necessary to improve our predictions of ecosystem response to global change, with Fleischer and others, 2019 showing the importance of constraining ecosystem models representing the carbon balance of tropical forests whilst including the P cycle. In their simulations, CNP models predicted around 40% less NPP in response to eCO_2. Our results suggest that there is room to continued response to eCO_2 in our forest, depending on mechanisms of uptake and efficient use that may cycle the small P pools faster. This understanding underpins the likely responses of the recently funded AmazonFACE experiment.

Conclusion

Although little is known about nutrient stocks and fluxes in the Central Amazon tropical forest, these processes play an important role in nutrient dynamics via the balance between nutrient assimilated into net primary productivity, nutrient storage in plants tissues (dead and alive) and their return to the soil via tissue mortality and decomposition. Here, we

analyzed mean estimates of canopy, stem, fine root, litter, coarse wood debris and soil stocks and fluxes to determine the amount of nutrients used and stored in this forest, as well as nutrient use efficiency. After soil, all macronutrients were mainly stored in stems, while canopy showed higher rates of nutrient input into net primary production and fine litterfall higher rates of nutrient return back to the system. Among nutrients, N showed a higher stock and flux, with cations being intermediate and P showing distinct low stock and flux, with high P retranslocation and use efficiency. We found evidence that among all nutrients analyzed here, P has the potential to be the most limiting in net primary productivity in our study site, suggesting that forest functioning in future climate might be even more regulated and dependent of an efficient nutrient uptake and use by plants. We also found that a great part of nutrient used in net primary production was originated by leaf retranslocation, in particular, P and cations and by decomposition of dead material, suggesting that soil cannot provide the nutrient demand in the next years with CO_2 elevation scenarios. However, to sustain their nutrient demand in future climate, many important plant strategies might need to be implemented or become even more effective, such as nutrient use efficiency, nutrient retranslocation, phosphatase enzyme activity, organic acid exudation and fine root association with mycorrhizal.

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