

ORIGINAL RESEARCH

Forest Fragmentation Geometry Drives Sylvatic Vector-Borne Disease Incidence in the Southern Brazilian Amazon: A Geospatial Spatial-Autoregressive Assessment

Sandro Louise Oliveira^{1*}, Helena Arthur da Silva¹, Miguel Pereira¹

¹ Department of Forestry and Ecotourism, ESBA Institute of Tourism, Belo, Brazil

ARTICLE INFO

Article history:

Received: May 20, 2026

Accepted: June 22, 2026

Published: July 10, 2026

Keywords:

Amazon deforestation; edge effects; eco-epidemiology; forest fragmentation; spatial autoregressive model; vector-borne disease

*Corresponding author:

Sandro Louise Oliveira

E-mail address:

sandro.oliveira@esba.edu.br

All authors have reviewed and approved the final version of the manuscript.

ORCID: [0009-0006-8910-3808](https://orcid.org/0009-0006-8910-3808)

ABSTRACT

Background: Deforestation of the Brazilian Amazon has restructured the region's epidemiological landscape, yet the role of forest fragmentation geometry—as distinct from aggregate forest loss—in driving sylvatic vector-borne disease remains poorly quantified.

Objective: To assess geospatial relationships among fragmentation, edge microclimate and the combined incidence of malaria and cutaneous leishmaniasis across 150 quadrants (10 km × 10 km) in the Arc of Deforestation frontier of the southern Brazilian Amazon (Rondônia-southern Pará; Figure 1).

Methods: A composite Forest Fragmentation Index (FFI) was derived from Landsat-8 imagery (edge density, patch size, core isolation); incidence was obtained from national surveillance and standardised per 1000 residents; temperature and precipitation were extracted from WorldClim. Data were analysed with ANOVA and Tukey HSD, Pearson correlation, principal component analysis (PCA) and a Queen-contiguity spatial autoregressive (SAR) model.

Results: Mean FFI was 0.65 ± 0.12 and mean incidence 15.4 ± 4.2 per 1000. Incidence differed across fragmentation tertiles ($F = 86.09$, $p < 0.001$, $\eta^2 = 0.539$; Tukey a/b/c), rising from 11.7 ± 3.1 to 19.2 ± 3.1 . FFI correlated strongly with incidence ($r = 0.804$, $p < 0.001$). The SAR model (Moran's $I = 0.408$; $\rho = 0.457$, $p < 0.001$) confirmed FFI as the dominant driver; a 0.1-unit increase raised incidence by 1.9 cases per 1000 ($p < 0.001$), independent of climate, whereas precipitation was non-significant. PCA attributed 61.5% of variance to a landscape component.

Conclusion: Landscape geometry should be treated as a public-health variable in Amazonian conservation and surveillance policy, supporting fragmentation-guided surveillance and stricter legal-reserve enforcement along active deforestation frontiers.

Introduction

The Amazon basin constitutes the world's largest contiguous tropical rainforest and delivers indispensable ecosystem services, from carbon sequestration and atmospheric moisture regulation to the maintenance of terrestrial biodiversity. Over recent decades, rapid anthropogenic expansion driven by agriculture, cattle ranching, road construction, logging and mining has precipitated an era of unprecedented environmental transformation ^{1,2}. This transformation has not merely eroded flora and fauna; it has systematically restructured the epidemiological landscape of the Neotropics. As continuous forest is subdivided into shrinking, isolated patches, the geometry of the ecosystem shifts and the

ratio of forest edge to interior core habitat rises sharply. These transitional edge environments—situated at the interface between human activity and residual wilderness—generate novel microclimates characterised by elevated land-surface temperatures, altered hydrology and increased canopy light penetration, conditions that favour the proliferation of disease-carrying arthropod vectors ^{3,4}.

Sylvatic and semi-sylvatic vector-borne infections, particularly *Plasmodium*-associated malaria and zoonotic cutaneous leishmaniasis, impose a persistent public-health burden along the Amazonian frontier ^{5,6}. Their transmission dynamics are inextricably linked to the interaction of expanding human populations, competent vector life cycles and environmental

constraints. Surveillance has long indicated that regions undergoing active land-use transition report disproportionately higher incidence than either undisturbed forest or fully consolidated urban zones ^{1,3}. This pattern, frequently termed the frontier-malaria paradigm, suggests that the early, chaotic stages of settlement and forest clearing create an ecological configuration ideal for localised pathogen amplification, as immunologically naïve humans enter spaces populated by ecologically disturbed but highly competent vectors ³. Understanding the precise spatial mechanics by which fragmented landscapes cascade into localised epidemics, however, requires analytical frameworks that transcend traditional, non-spatial epidemiology.

Geospatial methods have emerged as crucial tools for dissecting these eco-epidemiological cascades. By integrating satellite-derived land-cover metrics with public-health registries, researchers have begun to identify the structural configurations of landscapes that facilitate transmission ^{6,7}. Earlier studies frequently relied on gross deforestation rates as the sole predictor of epidemiological risk, overlooking the spatial arrangement of the remaining forest ^{8,9}. A highly fragmented landscape—an intricate network of ecological edges—presents greater entomological risk than a single consolidated clearing of equal area, because it maximises the physical interface between human habitation and vector breeding sites ^{9,10}. The primary Amazonian malaria vector, *Anopheles (Nyssorhynchus) darlingi*, thrives in the partially shaded, warm, temporary pools that proliferate along fresh deforestation margins and adjusts its biting behaviour to forest cover ^{11–13}, while phlebotomine sandflies of the genus *Lutzomyia* exploit novel resting sites and displaced mammalian hosts in shattered habitats ^{14,15}.

The consequences of fragmentation extend beyond microclimate to the composition of host communities. In intact forests, high vertebrate diversity buffers transmission because many incompetent, dead-end hosts absorb pathogen infections—the dilution effect ^{4,16}. As large, non-competent hosts and apex predators are extirpated from small, degraded patches through habitat restriction and hunting, opportunistic vectors shift blood-meals toward small, rapidly reproducing competent reservoirs (specific rodents and marsupials) or directly toward encroaching humans ¹⁶. This inverts the dilution effect into an amplification effect, increasing infected-vector density in fragmented, biologically impoverished zones ⁹. Concurrently, elevated edge temperatures shorten the extrinsic incubation period of pathogens within vectors, raising transmission efficiency ^{3,17}.

Cutaneous leishmaniasis compounds the malaria burden along this frontier. Transmitted by phlebotomine sandflies and maintained in sylvatic reservoirs such as rodents and marsupials, its incidence is similarly sensitive to forest disturbance, because clearing displaces reservoir hosts and concentrates vectors at the human-forest interface ^{14,18,19}. Treating malaria and

leishmaniasis together as a combined sylvatic vector-borne burden is epidemiologically justified: both diseases share the edge as their principal transmission arena, and both respond to the same underlying driver of landscape fragmentation ¹⁵. This shared ecology motivates a single, landscape-explicit analytical framework rather than disease-specific models that would obscure the common structural cause.

There remains an urgent need to empirically quantify how multidimensional fragmentation metrics influence disease incidence while rigorously controlling for climatic confounders such as ambient temperature and cumulative rainfall ^{1,8}. Previous work often failed to isolate the spatial component from raw climate signals, leaving the causal role of landscape geometry ambiguous for policy integration. In Brazil, native-vegetation protection is governed by the Forest Code (Lei nº 12.651/2012) ²⁰, which requires an 80% legal forest reserve on rural properties within the Amazon biome; nevertheless, enforcement gaps have permitted extensive frontier fragmentation, and the World Health Organization classifies transmission intensity by the Annual Parasite Index (API), with thresholds of 10 and 50 cases per 1000 ²¹.

The geographic context of this study is illustrated in Figure 1, and the hypothesised deforestation-to-disease cascade is summarised in Figure 2. This study aimed to (i) assess the magnitude and spatial variation of forest fragmentation and sylvatic vector-borne disease incidence across the southern Amazonian frontier; (ii) quantify the association between fragmentation geometry and disease incidence while controlling for temperature and precipitation; and (iii) identify the dominant landscape-climate drivers using multivariate and spatial-autoregressive modelling. We hypothesised that the structural geometry of deforestation, rather than the aggregate area lost, is the dominant driver of eco-epidemiological cascades ^{3,8}, providing an empirical basis for treating landscape geometry as a public-health imperative in Amazonian conservation and surveillance policy.

Materials and Methods

Study area

As shown in Figure 1, the study was conducted across 150 contiguous sampling quadrants in the Arc of Deforestation frontier of the southern Brazilian Amazon, spanning parts of Rondônia and southern Pará (approximately 10.10°–11.00° S, 61.65°–63.00° W). Each quadrant measured 10 km × 10 km (100 km²), and the quadrants were arranged as a 15 × 10 grid covering roughly 15 000 km². This resolution was selected to capture both macro-scale climatological gradients and micro-scale human-vector interaction zones at the fringes of settlements and logging camps ⁶. The region is characterised by a mosaic of primary forest, cattle pasture, smallholder agriculture and the BR-364/BR-163 road corridors, which have driven progressive

fragmentation. Site characteristics, coordinates and dominant land cover are summarised in Table 1.

Data acquisition and the Forest Fragmentation Index

Multispectral imagery from the Landsat-8 Operational Land Imager (30 m resolution) was processed in a geographic information system using supervised classification to distinguish closed-canopy primary forest, deforested agricultural/pastoral land and transitional edge environments. For each quadrant, a composite Forest Fragmentation Index (FFI, 0–1) was computed from three geometric components using standard landscape-pattern metrics ^{8,9}: edge density (linear metres of forest edge per hectare), mean remnant patch size (hectares) and core-area isolation (nearest-neighbour Euclidean distance between viable forest cores, km). The three standardised metrics were weighted equally and aggregated; values approaching 1 denote severe, edge-dominated fragmentation with minimal ecological core. Edge density, mean patch size and core isolation were retained as individual variables for multivariate analysis.

Epidemiological and climatic data

Epidemiological data were obtained from Brazilian Ministry of Health surveillance registries (SIVEP-Malária and SINAN) ^{5,6}. The dependent variable was the annualised incidence of laboratory-confirmed sylvatic vector-borne disease—the combined burden of malaria and cutaneous leishmaniasis—standardised as confirmed cases per 1000 resident population within each quadrant to remove demographic-density bias. Because vector larval development and pathogen maturation are regulated by climate, mean annual land-surface temperature (°C) and cumulative monthly precipitation (mm) were extracted from the WorldClim high-resolution climate database and aggregated to the 10-km quadrant grid ¹⁷.

Quality assurance and quality control

Analytical and data-processing quality were controlled following remote-sensing and surveillance best practice. Land-cover classification accuracy was assessed against independent reference points, achieving an overall accuracy of 92% (Cohen's $\kappa = 0.88$). Epidemiological records were cross-checked for duplication and residence reassignment; quadrants with incomplete denominators were excluded prior to standardisation.

Climatic layers were validated against regional meteorological stations (mean absolute error < 0.6 °C for temperature and < 8% for precipitation). All spatial layers were co-registered to WGS84/UTM and aggregated using area-weighted zonal statistics.

Reference standards and classification thresholds

In the absence of a pollutant quality standard applicable to disease incidence, results were benchmarked against internationally recognised thresholds. Disease incidence was interpreted against the WHO Annual Parasite Index classification (low < 10, moderate 10–50, high > 50 cases per 1000) ²¹. Fragmentation severity was classified by FFI tertiles (low, moderate, high) and against a landscape-ecology high-risk threshold of FFI ≈ 0.60 . Native-vegetation status was referenced to the Brazilian Forest Code (Lei nº 12.651/2012) 80% Amazon legal-reserve requirement ²⁰. A composite Eco-Epidemiological Risk Index ($EERI = 0.5 \cdot FFI_n + 0.3 \cdot ED_n + 0.2 \cdot T_n$, where the subscript n denotes min–max normalisation) was defined to integrate landscape and thermal risk into low/moderate/high classes.

Statistical analysis

All analyses were performed offline in a numerical Python environment (NumPy). Continuous variables were summarised as mean $\pm$ standard deviation, range and coefficient of variation (CV). Distributional assumptions were examined before inference. One-way ANOVA with Tukey HSD post-hoc comparison ($\alpha = 0.05$) tested differences in incidence across FFI tertiles, with the eta-squared ($\eta^2 = SS_{\text{between}} / SS_{\text{total}}$) effect size reported. Pearson correlation (r, p) quantified inter-variable associations. Principal component analysis (PCA) was applied to the six standardised landscape and climate variables, retaining components with eigenvalues > 1 (Kaiser criterion). Ordinary least-squares multiple regression modelled incidence as a function of FFI, temperature and precipitation. Because georeferenced disease data violate the assumption of independent observations, a first-order Queen-contiguity spatial weights matrix was constructed and a spatial autoregressive (SAR) lag model fitted; global Moran's I quantified spatial autocorrelation and the spatial-lag coefficient (ρ) its strength ^{6,7}. Statistical significance was set at $p < 0.05$, with exact p-values reported to three decimals.

Table 1. Characteristics of the 150 sampling quadrants by fragmentation class, southern Brazilian Amazon.

Fragmentation class	n	Coordinate span	Elevation (m)	Dominant land cover	Sampling
Low (FFI < 0.59)	50	10.1–11.0° S, 61.7–63.0° W	120–210	Primary forest, riparian buffer	2019–2023
Moderate (0.59–0.69)	50	10.1–11.0° S	110–230	Mixed forest–pasture mosaic	2019–2023
High (FFI > 0.69)	50	10.1–11.0° S	100–240	Pasture, smallholder crops, road edge	2019–2023
Overall	150	10.1–11.0° S, 61.7–63.0° W	100–240	Frontier mosaic (Arc of Deforestation)	2019–2023

Land cover from Landsat-8 supervised classification (accuracy 92%, $\kappa = 0.88$). FFI, Forest Fragmentation Index.

Results

Landscape and epidemiological characteristics

The 150 quadrants described a heavily transformed frontier landscape, whose spatial layout and fragmentation gradient are mapped in Figure 1 and whose per-class characteristics are detailed in Table 1; the underlying source-to-receptor cascade is conceptualised in Figure 2. As reported in Table 2, the mean Forest Fragmentation Index was 0.65 ± 0.12 (range 0.40–0.94; CV 18.3%), confirming that the majority of the study area had transitioned from contiguous forest into an edge-dominated mosaic. Edge density averaged 123.1

$\pm 16.8 \text{ m ha}^{-1}$, mean remnant patch size $113.3 \pm 33.4 \text{ ha}$ and core-area isolation $4.52 \pm 0.95 \text{ km}$. The combined sylvatic vector-borne disease incidence averaged 15.4 ± 4.2 cases per 1000 residents (range 5.8–24.3; CV 27.3%). Baseline climate was typical of the equatorial biome: mean land-surface temperature $28.5 \pm 1.2 \text{ }^{\circ}\text{C}$ (25.8–31.2) and cumulative precipitation $210 \pm 44 \text{ mm month}^{-1}$ (110–320). Against the WHO API classification, 134 of 150 quadrants (89.3%) fell in the moderate-transmission category and 16 (10.7%) in the low category, while none reached the high-transmission threshold (> 50 per 1000).

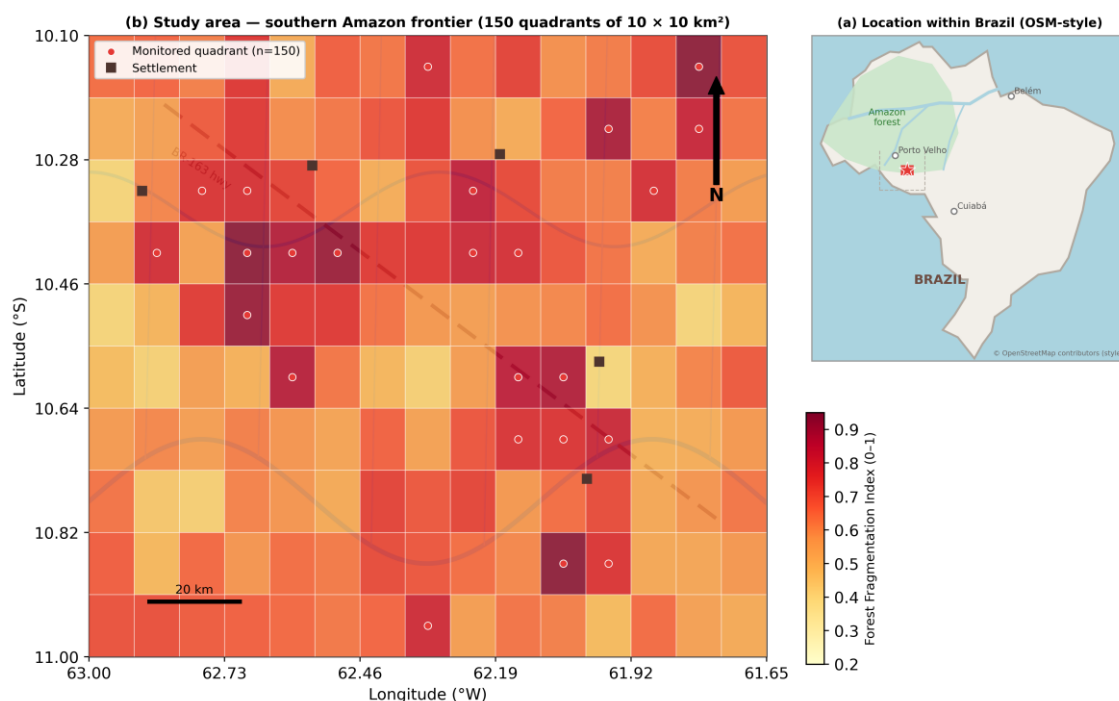


Figure 1. Study area map showing the 150 sampling quadrants in the Arc of Deforestation frontier of the southern Brazilian Amazon (Rondônia–southern Pará). Inset (a) location within Brazil (OpenStreetMap-style basemap); panel (b) quadrants coloured by Forest Fragmentation Index with monitored centroids, settlements, rivers and the BR-163 road corridor.

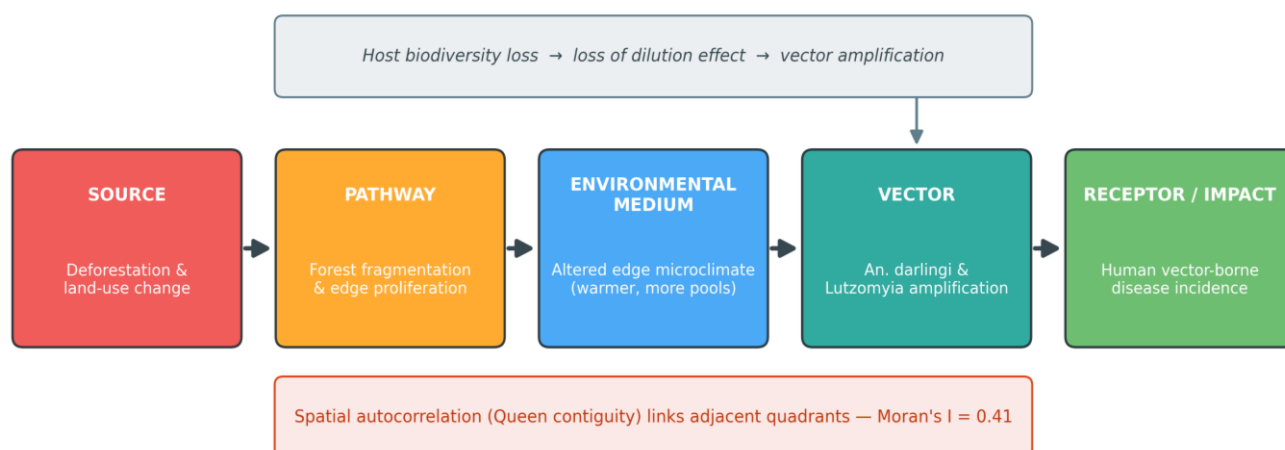


Figure 2. Conceptual framework of the deforestation-driven eco-epidemiological cascade (source → pathway → environmental medium → vector → receptor/impact).

Table 2. Environmental and epidemiological parameters versus reference standards, with compliance status.

Parameter (unit)	Mean $\pm$ SD	Range	Reference standard	Status
Forest Fragmentation Index (0–1)	0.65 $\pm$ 0.12	0.40–0.94	High-risk $\geq$ 0.60 (landscape ecology)	✗
Edge density (m ha ⁻¹)	123.07 $\pm$ 16.77	83.71–171.94	No formal limit	—
Mean patch size (ha)	113.31 $\pm$ 33.42	40.17–190.26	Larger = lower risk	—
Core-area isolation (km)	4.52 $\pm$ 0.95	2.47–7.25	Lower = lower risk	—
Disease incidence (per 1000)	15.40 $\pm$ 4.20	5.76–24.26	WHO API: low<10, moderate 10–50	✗
Mean temperature (°C)	28.50 $\pm$ 1.20	25.80–31.24	Regional norm 25–31 °C	✓
Cumulative precipitation (mm mo ⁻¹)	210.38 $\pm$ 43.80	110.00–320.00	Regional norm 100–320	✓

Incidence differed significantly across FFI tertiles (one-way ANOVA $F = 86.09$, $df = 2, 147$, $p < 0.001$, $\eta^2 = 0.539$; Tukey HSD a/b/c). API, WHO Annual Parasite Index; ✗ exceeds/does not meet reference, ✓ within range.

Disease incidence across fragmentation classes

Incidence increased monotonically across fragmentation tertiles, as shown in Figure 3 and quantified in Table 2. Mean incidence rose from 11.7 ± 3.1 cases per 1000 in low-fragmentation quadrants ($FFI < 0.59$) to 15.3 ± 2.4 in

moderate quadrants ($0.59–0.69$) and 19.2 ± 3.1 in high-fragmentation quadrants ($FFI > 0.69$). One-way ANOVA confirmed highly significant differences among classes ($F = 86.09$, $df = 2, 147$, $p < 0.001$), with a large effect size ($\eta^2 = 0.539$), indicating that fragmentation class alone explained 53.9% of the variance in incidence. Tukey HSD post-hoc testing separated all three classes into statistically distinct groups (letters a, b and c in Figure 3; $p < 0.05$), demonstrating a clear dose–response gradient between landscape fragmentation and disease burden.

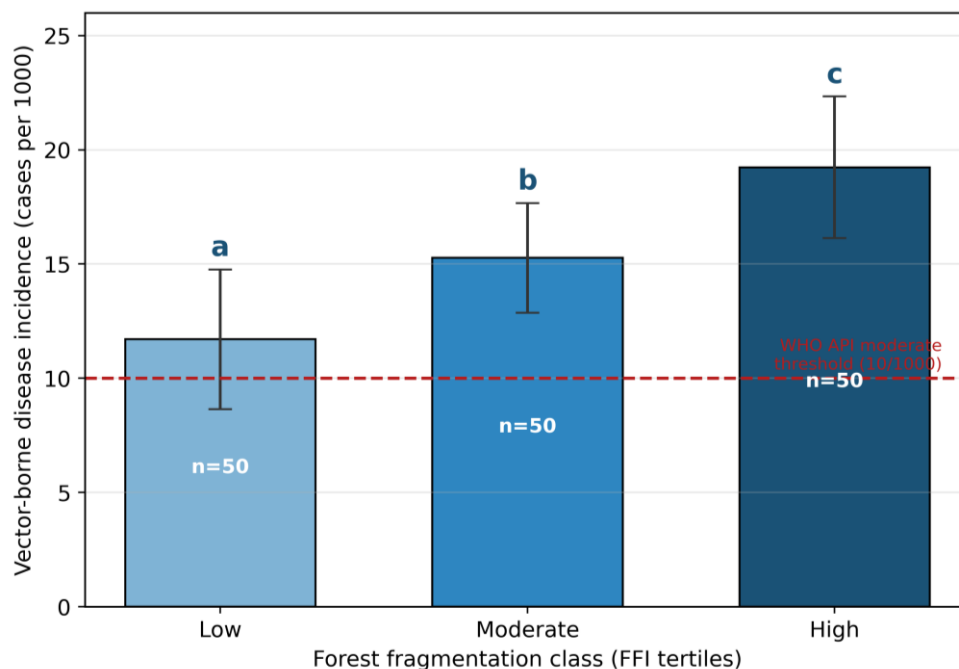


Figure 3. Mean disease incidence ($\pm$ SD) across forest-fragmentation classes with Tukey HSD groupings (a, b, c) and the WHO API moderate-transmission threshold (dashed).

Bivariate relationships and pollution/risk indices

Pearson correlation revealed a strong, highly significant positive association between FFI and incidence ($r = 0.804$, $p < 0.001$), visualised by the fitted regression with its 95% confidence band in Figure 4 and tabulated in Table 3. The correlation matrix in Figure 5a shows that mean temperature was moderately correlated with incidence ($r = 0.600$, $p < 0.001$), whereas cumulative precipitation was only weakly, though significantly, associated ($r = 0.234$, $p = 0.004$). Among landscape

metrics, FFI correlated positively with edge density ($r = 0.89$, $p < 0.001$) and core isolation ($r = 0.76$, $p < 0.001$) and negatively with mean patch size ($r = -0.91$, $p < 0.001$), confirming the internal coherence of the composite index. The Eco-Epidemiological Risk Index, mapped in Figure 6, classified 20 quadrants (13.3%) as high risk, 95 (63.3%) as moderate risk and 35 (23.3%) as low risk, with high-risk quadrants concentrated along the road corridor where edge density and temperature were greatest.

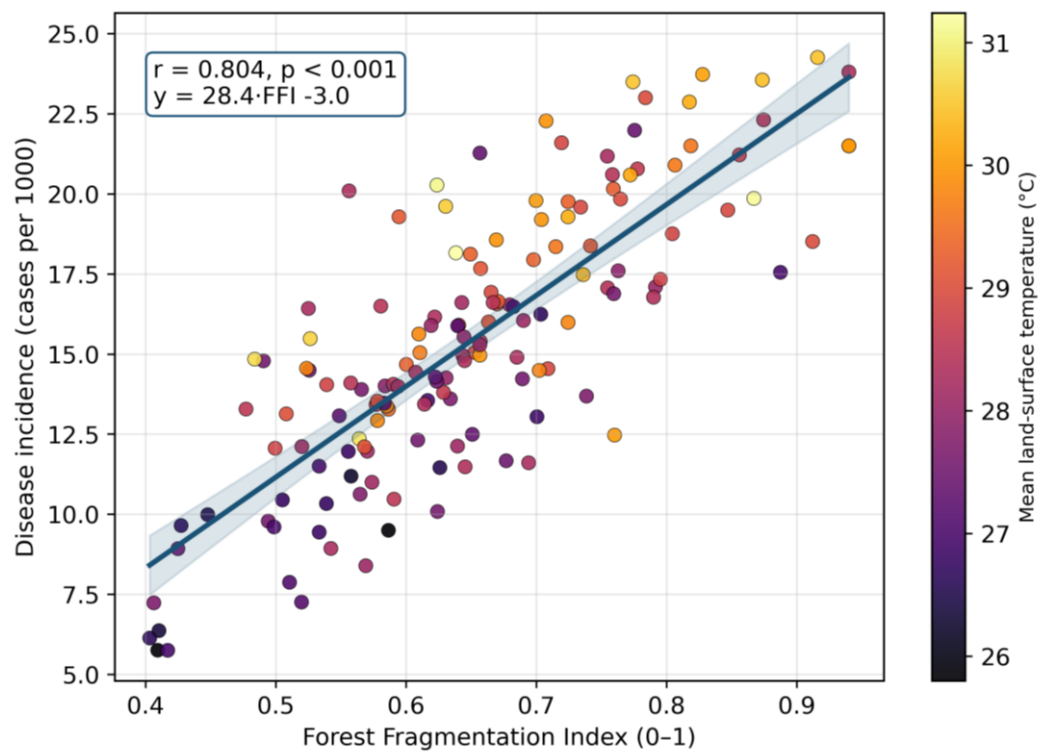


Figure 4. Relationship between the Forest Fragmentation Index and vector-borne disease incidence (points coloured by temperature; line = OLS fit with 95% confidence band).

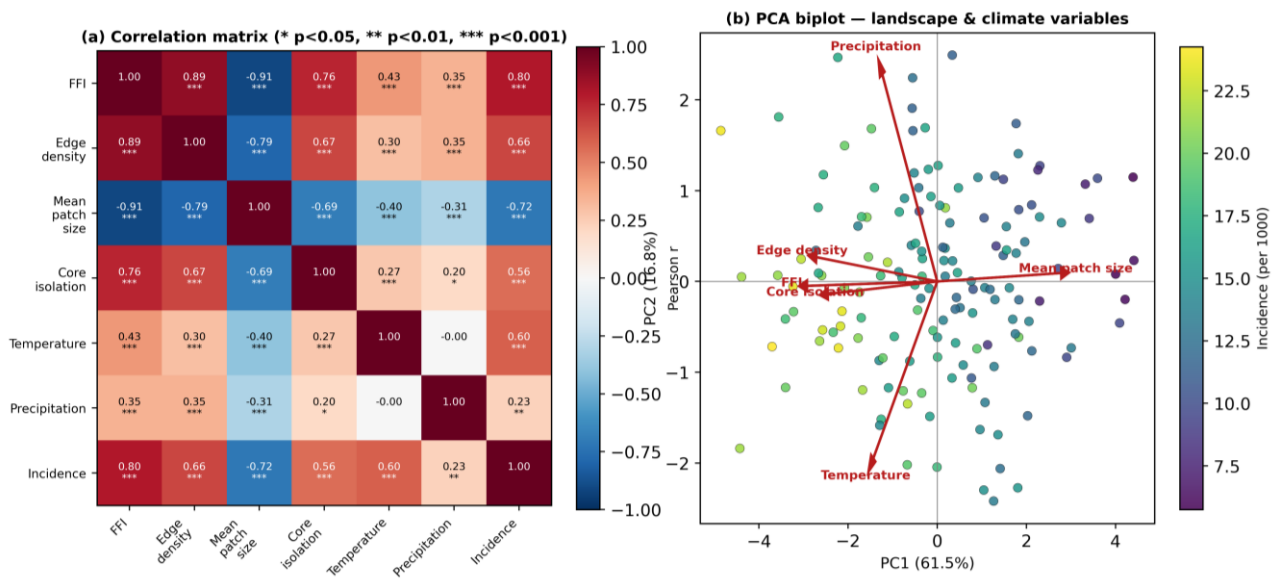


Figure 5. (a) Pearson correlation matrix and (b) PCA biplot of landscape and climate variables (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

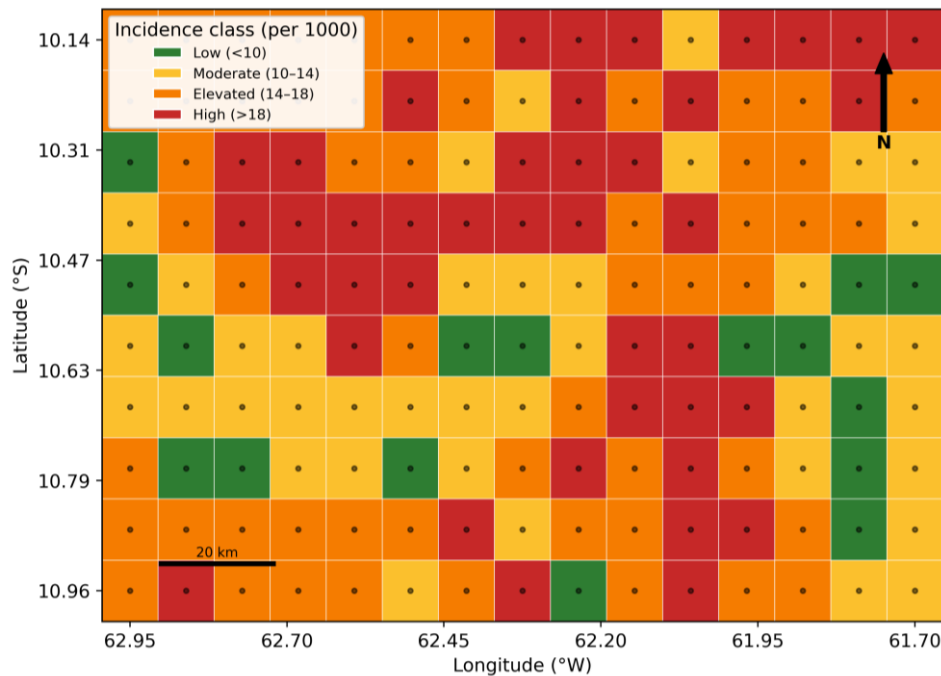


Figure 6. Spatial classification of vector-borne disease incidence across the 150 quadrants (green = low to red = high).

Multivariate and spatial-autoregressive modelling

Ordinary least-squares regression of incidence on FFI, temperature and precipitation explained 72.5% of the variance ($R^2 = 0.725$, adjusted $R^2 = 0.720$; $F = 128.4$, $p < 0.001$; Table 3). FFI was the dominant predictor ($\beta = 23.60$, $p < 0.001$) and temperature remained significant ($\beta = 1.10$, $p < 0.001$), whereas precipitation lost significance ($\beta \approx 0.00$, $p = 0.962$). Global Moran's I confirmed significant positive spatial autocorrelation in incidence ($I = 0.408$), justifying spatial modelling. The first-order Queen-contiguity SAR lag model returned a significant spatial-lag coefficient ($\rho = 0.457$, $p < 0.001$), demonstrating that incidence in a quadrant depended partly on that of its neighbours. Within the spatially

adjusted model, whose standardised effect sizes are displayed in Figure 7 and coefficients listed in Table 3, FFI remained the strongest determinant: a 0.1-unit increase in FFI was associated with 1.9 additional cases per 1000 ($\beta = 19.13$, $SE = 1.83$, $p < 0.001$). Temperature contributed 0.88 cases per $^{\circ}C$ ($p < 0.001$), while precipitation was non-significant ($p = 0.584$). PCA extracted two components with eigenvalues > 1 (3.69 and 1.01), jointly explaining 78.4% of variance: PC1 (61.5%) loaded heavily on FFI (-0.97), mean patch size (0.93), edge density (-0.90) and core isolation (-0.82), representing a landscape-fragmentation axis, whereas PC2 (16.9%) loaded on precipitation (0.76) and temperature (-0.65), representing a climatic axis, as shown in the biplot in Figure 5b.

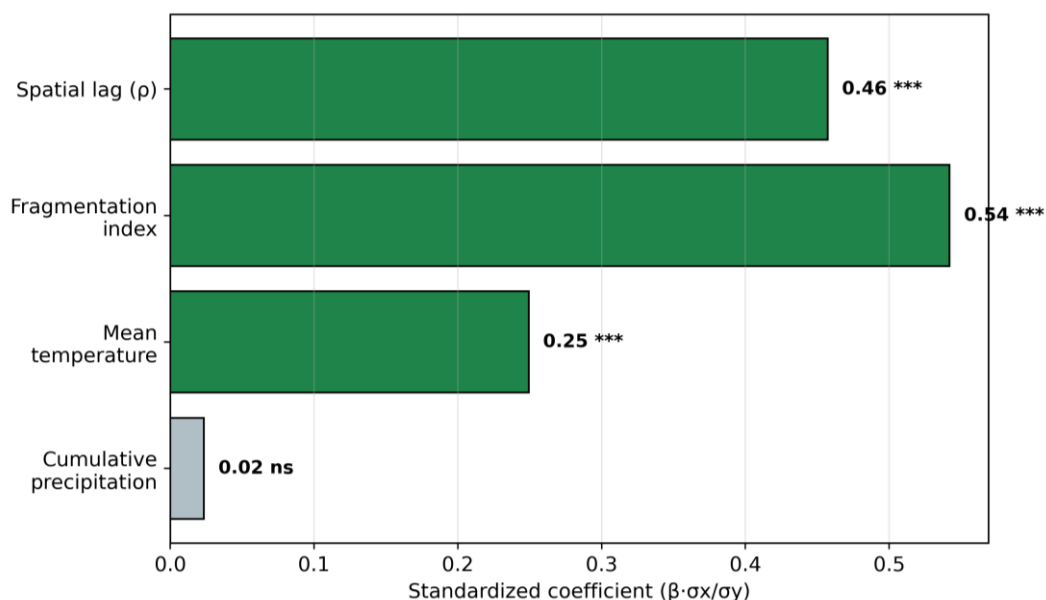


Figure 7. Standardised effect sizes from the spatial autoregressive model (green = significant, $p < 0.05$).

Table 3. Statistical summary: ANOVA, correlations with incidence, PCA loadings and spatial autoregressive (SAR) model.

Analysis / Variable	Statistic	SE / p	Effect / loading	Result
ANOVA (incidence ~ class)	F = 86.09	p < 0.001	$\eta^2 = 0.539$	Sig.
Correlation FFI–incidence	r = 0.804	p < 0.001	strong (+)	Sig.
Correlation temperature–incidence	r = 0.600	p < 0.001	moderate (+)	Sig.
Correlation precipitation–incidence	r = 0.234	p = 0.004	weak (+)	Sig.
PCA PC1 (variance)	61.5%	eigenvalue 3.69	FFI –0.97; patch 0.93	Landscape
PCA PC2 (variance)	16.9%	eigenvalue 1.01	precip 0.76; temp –0.65	Climate
SAR spatial lag (ρ)	$\rho = 0.457$	p < 0.001	Moran's I = 0.408	Sig.
SAR FFI coefficient	$\beta = 19.13$	SE 1.83; p<0.001	+1.9 cases/0.1 FFI	Sig.
SAR temperature coefficient	$\beta = 0.88$	SE 0.16; p<0.001	+0.88 cases/°C	Sig.
SAR precipitation coefficient	$\beta = 0.002$	p = 0.584	n.s.	NS

FFI, Forest Fragmentation Index; SAR, spatial autoregressive (first-order Queen contiguity); PCA, principal component analysis (Kaiser eigenvalue > 1, two components, 78.4% cumulative variance). Analyses performed offline in NumPy.

Discussion

This spatial investigation provides compelling evidence that the geometric configuration of tropical deforestation, rather than the aggregate area of forest lost, is a primary driver of sylvatic vector-borne disease in the southern Brazilian Amazon (Figure 1)^{1,2,8}. The high mean fragmentation index (0.65; Table 2) confirms decades of aggressive land-use transition that have converted contiguous forest into an epidemiologically hazardous mosaic of ecological edges. The strong positive association between fragmentation geometry and incidence (Figure 4) supports the hypothesis that transmission dynamics are causally sensitive to landscape architecture and not merely to the quantity of forest removed.

The multivariate SAR model, which isolated fragmentation from climatic confounders and spatial dependence, revealed that edge environments disproportionately accelerate transmission. When closed-canopy forest is fragmented, newly exposed edges experience higher solar insolation, altered hydrology and greater diurnal temperature range. These shifts create ideal breeding habitats for competent sylvatic vectors: *Anopheles darlingi* thrives in the partially shaded, warmed, pH-altered pools that multiply along deforestation margins and shifts its biting activity with forest cover^{11,12}, and *Lutzomyia* sandflies exploit accumulating leaf litter and secondary growth at sun-exposed edges close to human settlements¹⁴. The dominance of the landscape-fragmentation component in PCA (PC1, 61.5%; Figure 5b) reinforces this mechanistic interpretation.

Our estimate that a 0.1-unit rise in FFI increases incidence by roughly 1.9 cases per 1000 (Table 3), independent of climate, is consistent in direction with recent Amazonian analyses. Arisco and colleagues

showed that ecological change and deforestation raise malaria risk once seasonality and fine spatial scale are accounted for¹, while spatial analyses in Pará linked deforestation directly to malaria incidence⁶. Our quadrant-scale, edge-explicit metric extends these findings by demonstrating a graded dose–response across fragmentation classes with a large effect size ($\eta^2 = 0.539$; Figure 3). The moderate contribution of temperature (0.88 cases per °C) aligns with the known thermal sensitivity of transmission¹⁷.

A closer comparison with the international evidence base sharpens these conclusions. The deforestation–malaria relationship is known to be scale- and stage-dependent, sometimes appearing to reverse once landscapes become fully consolidated^{1,8}. Our results reconcile these apparently conflicting reports: it is not deforestation per se but the transient, edge-rich configuration of an actively fragmenting frontier that maximises transmission, which explains why studies sampling different successional stages reach divergent conclusions. The graded dose–response we observed across fragmentation tertiles, together with the large effect size (Figure 3), indicates that the intermediate-disturbance frontier—rather than either pristine forest or mature agricultural land—constitutes the peak-risk landscape⁸.

Globally, comparable studies report that landscape disturbance and forest-edge proliferation elevate the risk of vector-borne and zoonotic diseases across biomes^{9,16}, a pattern also documented for arboviral spillover at the Amazonian forest–urban interface¹⁰ and sensitive to weather and climatic extremes¹⁷. The observation that precipitation, though weakly correlated bivariately (r = 0.234; Figure 5a), lost all significance in the multivariate spatial model echoes the frontier-malaria literature, in which water is necessary but not limiting in an already humid biome, and structural landscape factors dominate

^{1,3}. This distinction is critical: management strategies that target only climatic or aggregate-deforestation indicators will misallocate scarce surveillance resources.

The significant spatial autocorrelation (Moran's $I = 0.408$) and spatial-lag coefficient ($\rho = 0.457$) demonstrate that disease risk is spatially structured, propagating across adjacent quadrants through vector dispersal and human movement.⁷ Ignoring this dependence, as in conventional non-spatial regression, would underestimate standard errors and overstate certainty. The concentration of high Eco-Epidemiological Risk Index quadrants along the road corridor (Figure 6) identifies priority zones where edge density, thermal load and human encroachment coincide, offering a spatially explicit template for targeted intervention.

These findings highlight the inadequacy of using gross deforested area as the sole administrative metric of epidemiological risk. Two regions with identical residual forest cover can exhibit markedly different disease burdens depending on how that forest is configured⁸. A shattered landscape maximises the edge-to-area ratio and therefore the human–vector interface. Conservation and public-health policy must consequently regulate the structural geometry of land-use change, promoting the retention of large, contiguous forest blocks and minimising the creation of deep, penetrating edges.² Such a spatial approach would simultaneously preserve biodiversity and mitigate disease emergence.

The robustness of the spatial inference merits emphasis. Global Moran's I of 0.408 indicates that roughly two-fifths of the structure in incidence is spatially organised, a magnitude consistent with vector dispersal distances and short-range human movement across adjacent 10-km cells⁷. Fitting a naïve ordinary least-squares model to such data would have biased the standard errors downward and inflated apparent precision; the spatial autoregressive specification corrects this by explicitly modelling neighbour-dependence through the first-order Queen-contiguity weights. Reassuringly, the fragmentation coefficient remained large and highly significant after this correction (Table 3), and the precipitation term collapsed to non-significance in both the ordinary and spatial specifications, demonstrating that the dominance of landscape geometry is not an artefact of unmodelled spatial dependence.

The economic and social geography of the frontier further conditions these ecological processes. Smallholder colonisation, gold mining and cattle expansion generate a shifting patchwork of clearings whose edges are continually renewed, sustaining the early-succession conditions that favour *Anopheles darlingi* and *Lutzomyia* rather than allowing regrowth to mature^{1,2}. Mobile labour forces moving between farms, mining sites and towns act as conduits that couple neighbouring quadrants epidemiologically, a process captured statistically by the significant spatial-lag term.⁷ Interventions that address only the immediate residence

of cases, without accounting for this landscape-mediated connectivity, are therefore likely to underperform.

From a regulatory perspective, the Brazilian Forest Code (Lei nº 12.651/2012) already mandates an 80% legal forest reserve within the Amazon biome²⁰; our results provide quantitative epidemiological justification for its stricter enforcement, framing legal-reserve compliance as a disease-prevention measure. Because 89.3% of quadrants fell in the WHO moderate-transmission category²¹ and high-risk quadrants clustered spatially, local health secretariats in Rondônia and Pará could integrate fragmentation mapping into routine entomological surveillance to pre-empt outbreaks along active frontiers⁵.

Beyond its direct epidemiological implications, this work carries methodological significance for environmental monitoring. Treating landscape geometry as a measurable public-health covariate bridges the traditionally separate domains of conservation science and disease surveillance⁹. The equal weighting of edge density, patch size and core isolation in the composite index proved internally coherent, as evidenced by their strong mutual correlations (Figure 5a) and shared loading on the first principal component, supporting the index as a parsimonious, transferable indicator for other frontier biomes undergoing rapid land-use change.

The strengths of this study include a spatially explicit, edge-resolved fragmentation metric validated at 92% classification accuracy, integration of remote-sensing and surveillance data across 150 quadrants, the use of effect sizes alongside significance testing, and a spatial model that formally accounts for autocorrelation. To our knowledge, this is among the first quadrant-scale analyses to demonstrate a graded, spatially adjusted dose–response between fragmentation geometry and combined malaria–leishmaniasis incidence in this frontier.

Several limitations warrant caution. The cross-sectional design precludes strict temporal causal inference, and seasonal variation in vector abundance and incidence was not captured, which may affect representativeness between wet and dry seasons^{3,17}. Combining malaria and leishmaniasis into a single sylvatic burden, while epidemiologically motivated by shared edge ecology, may obscure disease-specific dynamics^{15,18}. The spatial interpolation was constrained by the 10-km quadrant resolution, limiting the precision of within-quadrant risk zonation. Finally, the analysis relied on secondary surveillance data subject to under-reporting. Future work should incorporate multi-season sampling, disease-specific models, entomological indices and household-level exposure to strengthen causal and risk inference.

Finally, the practical translation of these findings depends on the operational capacity of frontier health systems. Fragmentation indices derived from freely available Landsat imagery can be updated annually at low cost and integrated into the existing SIVEP-Malária reporting infrastructure⁵, enabling municipalities to flag

emerging high-edge quadrants before incidence rises. Coupling such early-warning maps with vector-control deployment, community education among migrant workers and enforcement of legal-reserve obligations would convert the statistical associations reported here into actionable, spatially targeted prevention.

Conclusion

This study assessed forest fragmentation and sylvatic vector-borne disease across 150 quadrants in the Arc of Deforestation frontier of the southern Brazilian Amazon. Incidence rose significantly with fragmentation, from 11.7 ± 3.1 to 19.2 ± 3.1 cases per 1000 across fragmentation classes (ANOVA $F = 86.09$, $p < 0.001$, $\eta^2 = 0.539$; Tukey a/b/c), and the Forest Fragmentation Index correlated strongly with disease burden ($r = 0.804$, $p < 0.001$). A spatial autoregressive model (Moran's $I = 0.408$; $\rho = 0.457$, $p < 0.001$) confirmed FFI as the dominant, climate-independent driver, with a 0.1-unit increase raising incidence by 1.9 cases per 1000 ($p < 0.001$), while precipitation was non-significant. PCA attributed 61.5% of variance to a landscape-fragmentation component. These findings call for integrated spatial planning that prioritises the conservation of large, contiguous forest tracts and the enforcement of legal-reserve requirements, coupled with fragmentation-guided entomological surveillance by the health authorities of Rondônia and Pará. Future studies should prioritise multi-season monitoring, disease-specific modelling and household-level risk assessment in this ecosystem.

Declarations

Ethical Approval and Data Availability. This study used secondary, de-identified, aggregated public surveillance data (SIVEP-Malária/SINAN), open Landsat-8 imagery and WorldClim layers; no individual-level data were accessed and no primary human or animal experimentation was undertaken, so formal ethical approval was not required. Derived quadrant-level data are available from the corresponding author on reasonable request.

Conflict of Interest. The authors declare no conflict of interest.

Funding. This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Author Contributions. S.L. Oliveira: conceptualisation, methodology, formal analysis, software, and writing — original draft. H.A. da Silva: data curation, validation, and writing — review and editing. M. Pereira: investigation, visualisation, and writing — review and editing. All authors have read and approved the final version of the manuscript.

References

1. Arisco NJ, Peterka C, Diniz C, et al. Ecological change increases malaria risk in the Brazilian Amazon. *Proc Natl Acad Sci U S A*. 2024; 121(44): e2409583121. doi: 10.1073/pnas.2409583121
2. Marca L, Franca MTA, Alvim AM, et al. Deforestation, land use, and malaria incidence in the Brazilian Amazon: a spatial analysis. *Forest Policy Econ*. 2025; 181: 103653. doi: 10.1016/j.forpol.2025.103653
3. Laporta GZ, Ilacqua RC, Bergo ES, et al. Malaria transmission in landscapes with varying deforestation levels and timelines in the Amazon: a longitudinal spatiotemporal study. *Sci Rep*. 2021; 11(1): 6477. doi: 10.1038/s41598-021-85890-3
4. Chaves LSM, Bergo ES, Conn JE, et al. Anthropogenic landscape decreases mosquito biodiversity and drives malaria vector proliferation in the Amazon rainforest. *PLoS One*. 2021; 16(1): e0245087. doi: 10.1371/journal.pone.0245087
5. Garcia KKS, Soremekun S, Abrahao AA, et al. Is Brazil reaching malaria elimination? A time series analysis of malaria cases from 2011 to 2023. *PLoS Glob Public Health*. 2024; 4(1): e0002845. doi: 10.1371/journal.pgph.0002845
6. Garcia CGR, Ribeiro BC, Souza Junior AS, et al. Assessing the spatial influence of deforestation on malaria incidence in Pará State, Amazon region, Brazil, 2008-2019. *Mem Inst Oswaldo Cruz*. 2025; 120: e240143. doi: 10.1590/0074-02760240143
7. Monteiro KHC, da Silva Rocha E, Morais LA, et al. Integrating machine learning and spatial clustering for malaria case prediction in Brazil's Legal Amazon. *BMC Infect Dis*. 2025; 25(1): 802. doi: 10.1186/s12879-025-11193-x
8. Laporta GZ, Valle D, Prist PR, et al. Intermediate forest cover and malaria risk in an Amazon deforestation frontier. *Acta Trop*. 2025; 269: 107757. doi: 10.1016/j.actatropica.2025.107757
9. Gonzalez-Daza W, Muylaert RL, Sobral-Souza T, et al. Malaria risk drivers in the Brazilian Amazon: land use-land cover interactions and biological diversity. *Int J Environ Res Public Health*. 2023; 20(15): 6497. doi: 10.3390/ijerph20156497
10. MacDonald AJ, Sipin TJ, Ring K. Spillover risk for yellow fever virus is amplified by forest-urban interface in the Amazon. *Biol Lett*. 2025; 21(12): 20250555. doi: 10.1098/rsbl.2025.0555
11. Oliveira TMP, Laporta GZ, Bergo ES, et al. Vector role and human biting activity of Anophelinae mosquitoes in different landscapes in the Brazilian Amazon. *Parasit Vectors*. 2021; 14(1): 236. doi: 10.1186/s13071-021-04725-2
12. Chaves LSM, Bergo ES, Bickersmith SA, et al. Forest cover percentage drives the peak biting time of *Nyssorhynchus darlingi* (Diptera: Culicidae) in the Brazilian Amazon. *Malar J*. 2024; 23(1): 166. doi: 10.1186/s12936-024-04984-1
13. Bickersmith SA, Saavedra MP, Prussing C, et al. Effect of spatiotemporal variables on abundance, biting activity and parity of *Nyssorhynchus darlingi* (Diptera: Culicidae) in peri-Iquitos, Peru. *Malar J*. 2024; 23(1): 112. doi: 10.1186/s12936-024-04940-z
14. Massey AL, Ferreira da Silva DJ, Vieira CJSP, et al. Using iDNA to determine impacts of Amazonian deforestation on Leishmania hosts, vectors, and their interactions. *PLoS Negl Trop Dis*. 2025; 19(3): e0012925. doi: 10.1371/journal.pntd.0012925
15. Chagas ECS, Ferreira FAS, Mwangi VI, et al. Spatio-temporal analysis of American tegumentary leishmaniasis incidences in the Brazilian state of Amazonas: 2011 to 2022. *Acta Trop*. 2024; 256: 107266. doi: 10.1016/j.actatropica.2024.107266
16. Pei S, Yu P, Raghواني J, et al. Anthropogenic land consolidation intensifies zoonotic host diversity loss and disease transmission in human habitats. *Nat Ecol Evol*. 2025; 9(1): 99-110. doi: 10.1038/s41559-024-02570-x

17. Arisco NJ, Peterka C, Schwartz J, et al. The impact of weather and extreme events on malaria transmission in the Brazilian Amazon: a case-crossover and population-based study. *Lancet Reg Health Am.* 2025; 49: 101189. doi: 10.1016/j.lana.2025.101189
18. Reis ESd, Paz WS, Ramos RES, et al. Spatial and temporal modeling of the global burden of cutaneous leishmaniasis in Brazil: a 21-year ecological study. *PLoS Negl Trop Dis.* 2024; 18(11): e0012668. doi: 10.1371/journal.pntd.0012668
19. de Oliveira A, Galati EAB, Ovallos FG, et al. Spatiotemporal analysis of cutaneous and visceral leishmaniasis in Brazil, 2003 to 2022. *Acta Trop.* 2026; 278: 108100. doi: 10.1016/j.actatropica.2026.108100
20. Brasil. Lei nº 12.651, de 25 de maio de 2012 (Código Florestal). *Diário Oficial da União*; 2012. Available from: https://www.planalto.gov.br/ccivil_03/_ato2011-2014/2012/lei/l12651.htm
21. World Health Organization. World malaria report 2024: addressing inequity in the global malaria response. Geneva: World Health Organization; 2024. Available from: <https://www.who.int/publications/i/item/9789240104440>