PROCEEDINGS B

royalsocietypublishing.org/journal/rspb

Research

Cite this article: Aronoff JE *et al*. 2025 Inflammaging is minimal among forager-horticulturalists in the Bolivian Amazon. *Proc. R. Soc. B* **292**: 20251111.
https://doi.org/10.1098/rspb.2025.1111

Received: 28 April 2025
Accepted: 3 July 2025

Subject Category:
Evolution

Subject Areas:
health and disease and epidemiology, immunology

Keywords:
inflammaging, inflammation, cytokines, ageing, mismatch

Authors for correspondence:
Jacob E. Aronoff
e-mail: jarnoff@asu.edu
Benjamin C. Trumble
e-mail: btrumble@asu.edu

Electronic supplementary material is available online at https://doi.org/10.6084/m9.figshare.c.7921356.

Inflammaging is minimal among forager-horticulturalists in the Bolivian Amazon

Jacob E. Aronoff[1], Carrie L. Jenkins[1], Angela R. Garcia[2], Stephanie V. Koebele[1], Suhail Ghafoor[3], Kate L. Woolard[1], Mia Charifson[4], Ivan M. Suarez[5], Daniel Eid Rodriguez[5], Bret Beheim[6], Daniel K. Cummings[7], Paul L. Hooper[7], Thomas K. Kraft[8], Kenneth Buetow[2], Caleb E. Finch[9], Maximilien Franck[10], Alan A. Cohen[11], Jonathan Stieglitz[12], Michael Gurven[13], Hillard Kaplan[7] and Benjamin C. Trumble[1]

[1]School of Human Evolution and Social Change; Center for Evolution and Medicine; Institute for Human Origins, [2]School of Life Sciences; Center for Evolution and Medicine, and [3]Center for Evolution and Medicine, Arizona State University, Tempe, AZ, USA
[4]Department of Population Health, Grossman School of Medicine, New York University, New York, NY, USA
[5]Faculty of Medicine, Universidad Mayor de San Simon, Cochabamba, Cochabamba Department, Bolivia
[6]Department of Human Behavior, Ecology and Culture, Max-Planck-Institute for Evolutionary Anthropology, Leipzig, Germany
[7]Economic Science Institute, Chapman University, Orange, CA, USA
[8]Department of Anthropology, University of Utah, Salt Lake City, UT, USA
[9]Leonard Davis School of Gerontology, University of Southern California, Los Angeles, CA, USA
[10]Research Center on Aging, Faculty of Medicine and Health Sciences, University of Sherbrooke, Sherbrooke, Quebec, Canada
[11]Robert N. Butler Columbia Aging Center; Department of Environmental Health Sciences; Mailman School of Public Health, Columbia University, New York, NY, USA
[12]Department of Social and Behavioral Sciences, Toulouse School of Economics, Toulouse, Occitanie, France
[13]Department of Anthropology, University of California Santa Barbara, Santa Barbara, CA, USA

JEA, 0000-0002-1422-801X; CLJ, 0000-0002-2235-2360; SVK, 0000-0001-5502-2790; BB, 0000-0003-4653-3155; TKK, 0000-0002-0634-9233; MF, 0009-0007-0431-2772; AAC, 0000-0003-4113-3988; JS, 0000-0001-5985-9643; MG, 0000-0002-5661-527X; HK, 0000-0002-7398-7358; BCT, 0000-0003-3201-0628

An increase in chronic systemic inflammation in later life, termed inflammaging, is implicated in health risk. However, it is unclear whether inflammaging develops in all human populations, or if it is the product of environmental mismatch. We assessed inflammaging in Tsimane forager-horticulturalists of the Bolivian Amazon, using serum cytokines in a primarily cross-sectional sample (1134 samples from n = 714 individuals, age 39–94, 51.3% female). IL-6 was positively associated with age (β = 0.013, $p < 0.01$). However, other pro-inflammatory markers, including IL-1β and TNF-α, did not increase with age (β = −0.005 and β = −0.001, respectively). We then compared the Moseten, a neighbouring population that has experienced greater market integration (423 samples from n = 380 individuals, age 39–85, 48.2% female). The Moseten also showed a positive age association for IL-6 that attenuated at later ages (age β = 0.025, $p < 0.01$; age^2 β = −0.001, $p < 0.05$). Further, IL-1β and TNF-α were both positively associated with age (β = 0.021, $p < 0.05$ and β = 0.011, $p < 0.01$, respectively). Our results demonstrate minimal inflammaging in the Tsimane, highlighting variation across populations in this age-related process. They also suggest that inflammaging is exacerbated by lifestyle shifts.

THE ROYAL SOCIETY PUBLISHING

1. Introduction

The development of chronic systemic inflammation with age, termed 'inflammaging', is implicated in risk for numerous age-related diseases (e.g. cardiovascular, neurodegenerative, cancer) [1,2]. It can be caused by a variety of factors, including chronic antigenic load, accumulation of cellular waste and senescent cells, mitochondrial dysfunction and damage, gut permeability and excess adiposity [3–5]. However, while prior studies of inflammaging have been informative, they have primarily focused on Western industrialized populations [6,7]. These populations experience novel conditions in human evolutionary history, including reduced exposure to microbes and parasites, low fertility, diets high in saturated fats and processed sugars, sedentary behaviour and smoking [8,9]. As a result, it is unclear how inflammaging may have manifested in pre-industrial contexts or whether it is primarily a consequence of environmental mismatch.

Studies with non-industrialized, subsistence populations can help address both theoretical and public health discussions on inflammaging. Understanding commonalities versus variability across subsistence populations can inform theoretical discussions on the evolutionary origins and contextual determinants of inflammaging. Further, studies with these populations might also provide new insights into the evolved relationship between inflammation, health and ageing. This has been shown previously with the Tsimane forager-horticulturalists in the Bolivian Amazon. The Tsimane experience a high exposure to infections, resulting in frequent inflammation throughout life [10,11]. However, certain chronic diseases in which inflammation has been implicated as a contributing pathway are rare. For example, the prevalence of both cardiovascular disease and Alzheimer's disease and related dementias are among the lowest reported of any global population [12,13].

Recent studies with the Tsimane highlight the importance of considering both the causes and context of inflammation for understanding its relationship to health and ageing. Industrialized populations experience a high prevalence of chronic sterile inflammation owing to lifestyle factors (e.g. caloric excess and sedentary behaviour), which is a major contributor to ageing and disease [14,15]. This has been termed meta-inflammation, or metaflammation, because it is caused by metabolic dysfunction [16,17]. In contrast, the Tsimane are much more physically active, rarely obese, and they consume smaller quantites of processed sugars and saturated fats compared to industrialized populations [18]. Further, high infectious exposure among the Tsimane may be protective for metabolic health and downstream meta-inflammation. For example, mounting an inflammatory response against an infection increases metabolic expenditure, while parasites consume circulating glucose and lipids in their host [19,20]. Finally, parasitic and microbial exposures that are largely absent in industrialized populations might aid in appropriate immune regulation, consistent with the 'old friends' hypothesis [21].

It is currently unclear whether the Tsimane or similar subsistence populations develop inflammaging [22,23]. To address this gap, we assessed a collection of cytokines, which are key mediators of immune function and inflammation-related health risk [24,25]. We focused primarily on three of the most commonly used pro-inflammatory cytokines to assess inflammaging: IL-1β, IL-6 and TNF-α [26,27]. Based on their subsistence lifestyle and the minimal prevalence of chronic diseases with inflammatory aetiologies, we hypothesized that the Tsimane would have minimal or no inflammaging, reflected in weak or null associations with age [18,28]. We measured these cytokines as part of a multiplex assay that also included IL-1α, IL-2, IL-4, IL-5, IL-10, IL-12, IL-13, IL-15, IL-17, IL-23, IFN-γ and TNF-β. To utilize all available data, we also tested exploratory age associations for these additional biomarkers.

Comparisons of the Tsimane inflammaging profile with other populations might be complicated by methodological differences across studies or genetic differences across populations. Therefore, we also measured cytokine profiles in the Moseten, a neighbouring population that has experienced greater lifestyle shifts in recent years. The Moseten diverged from the Tsimane a few hundred years ago, owing to sociocultural changes following missionary contact [29]. Genetic analysis has revealed minimal distance between these populations [30]. While many Moseten still engage in a subsistence lifestyle, not all do, and there is greater average consumption of market-derived foods, including sugar and oil [18,29–31]. Moseten cytokine profiles were measured in the same laboratory with the Tsimane samples (randomized across batches). Based on the greater lifestyle shifts in the Moseten, we hypothesized that they would display more pronounced inflammaging compared with the Tsimane.

Finally, an important caveat when studying inflammation in high-pathogen environments is the frequent fluctuations in biomarkers with the onset and clearing of infections. As a result, an elevation of inflammatory markers on one day might not reflect a stable profile over time that distinguishes individual differences [32,33]. These acute fluctuations might therefore confound associations with age, making it difficult to detect inflammaging. We assessed this possibility using intra-class correlations (ICCs), a commonly used method for assessing measure reliability. The ICC estimates the proportion of the total variation attributable to between-person variation [34]. A lower ICC indicates that a larger proportion of the total variation in a measure is attributable to within-person variation, which in this case would indicate a greater influence of acute infections on cytokine fluctuations [35]. While most serum samples were cross-sectional, we relied on a subsample of participants for whom we have multiple samples (average 2 years apart) to estimate ICCs. Owing to the frequent infections among the Tsimane, we hypothesized lower ICCs compared with published US/European estimates [34,36–38]. Further, to address this possible confound, we considered models testing age associations while adjusting for leukocytes, a proxy for acute infection.

2. Methods

(a) Study participants

The Tsimane have an estimated population size of approximately 17 000, with most families relying on horticulture and hunting/fishing for subsistence [10,29]. Most villages have little or no access to running water and sanitation [39]. The total

fertility rate is approximately nine children born per woman over their lifetime [40]. Infections are a leading cause of morbidity and mortality, particularly in early and late life, and they impact reproduction, impair child growth and elevate basal metabolic rates [20,41–44]. The neighbouring Moseten have an estimated population size of approximately 3000 and a fertility rate of approximately six children per woman [29]. They have experienced greater lifestyle shifts compared with the Tsimane owing to closer proximity to the market and a stronger historical influence from missionaries. The Moseten also have greater Spanish fluency and schooling, as well as greater access to medical and other services (e.g. electricity, running water) [18].

Individuals aged 40+ years were recruited from approximately 100 Tsimane villages in the Beni Department and 10 Moseten villages across the Beni and La Paz Departments [12,29]. Age was assigned to each individual by cross-referencing known ages from written records, photo comparisons of individuals with known ages, relative age rankings by multiple informants and dated events. In one region, estimated ages were compared against Catholic mission birth records dating back to 1952, and all estimates fell within a 3-year age range [44]. After recruitment, three individuals (two Moseten, one Tsimane) were found to be 39 years old but were included in the analysis as their ages closely matched the intended inclusion criteria.

(b) Cytokines

Cytokines were measured in serum samples from intravenous blood draws collected in the morning between 6:00 and 7:00 after an overnight fast and before any strenuous physical activity. Serum was allowed to clot, then immediately centrifuged, aliquoted and frozen in liquid nitrogen. Samples were shipped on dry ice to Arizona State University, where they were stored at −80°C until analysis using the Q-Plex Human Cytokine HS 15-Plex (Quansys) multiplex ELISA kit on the first freeze–thaw cycle. The panel included IL-1α, IL-1β, IL-2, IL-4, IL-5, IL-6, IL-10, IL-12, IL-13, IL-15, IL-17, IL-23, IFN-γ, TNF-α and TNF-β. Control samples were run in duplicate to assess intra- and inter-assay variability, as indicated by the coefficient of variation (CV). Across cytokines, the high control, between-plate median CV was 1.4% (range = 0.4–2.2), while the within-plate median CV was 2.4% (range = 1.0–5.9). The median for the low control, between-plate CV was 20.4% (range = 12.9–30.8), while the within-plate median CV was 6.7% (range = 3.9–42.9; see electronic supplementary material, table S1).

(c) Statistical analysis

All cytokines were log-transformed owing to positive skew. Values below the limit of detection were replaced by the mid-point between the lower limit and zero. The proportions of samples below the limit of detection for each cytokine are as follows: IL-1α = 0.05, IL-1β = 0.54, IL-2 = 0.02, IL-4 = 0.31, IL-5 = 0.22, IL-6 = 0.03, IL-10 = 0.03, IL-12 = 0.41, IL-13 = 0.44, IL-15 = 0.06, IL-17 = 0.60, IL-23 = 0.22, IFN-γ < 0.01, TNF-α < 0.01 and TNF-β = 0.30. The small number of samples with concentrations above the limit of detection (<1%) were replaced with the highest detected value. Cytokine distributions are presented in electronic supplementary material, figures S1–S15 and table S2. Pearson correlations were calculated among all cytokines and principal component analysis was used to further assess covariances. Age associations were tested using regression models (adjusted for sex) with clustered robust s.e. to account for multiple measures provided by some participants. Since biomarkers often show quadratic age trends, we considered both linear and quadratic terms in the models. ICCs were estimated from mixed effects models adjusted for age, sex and time between samples (mean two years) with bootstrapped 95% confidence intervals [34].

3. Results

In total, 714 Tsimane participants (51.3% female) provided 1134 serum samples and 380 Moseten participants (48.2% female) provided 423 serum samples (table 1). Of these individuals, 255 (36%) Tsimane and 42 (11%) Moseten provided repeat samples. Ages for Tsimane participants ranged between 39 and 94 years (mean = 61.2), while ages for the Moseten participants ranged between 39 and 85 years (mean = 58.3). As expected, based on greater lifestyle shifts in the Moseten, mean body mass index (BMI) and waist circumference were slightly higher than the Tsimane (25.4 versus 23.9 kg m^{-2} and 94.5 versus 90.5 cm, respectively).

The correlations among cytokines were all positive, varying in magnitude and were similar between the populations (electronic supplementary material, tables S3–S5). The strongest correlations were among IL-1β, IL-12, IL-15 and TNF-β (r range, 0.65–0.74), while the weakest correlations were between IFN-γ and IL-1α (r = 0.11), IL-5 (r = 0.09), IL-17 (r = 0.12) and IL-23 (r = 0.08), as well as between IL-23 and IL-2 (r = 0.12) and IL-10 (r = 0.12).

We also considered principal components (PCs) of the cytokines, based on a prior approach to measuring inflammaging as a composite of multiple cytokines [45]. In both populations, the first PC (PC1) explained a large proportion of the total variance (Tsimane = 34.2%, Moseten = 43.8%), with a large drop in explained variance for the other PCs (all < 10%) (electronic supplementary material, table S6). The loadings for PC1 were positive for all cytokines and nearly identical between the Tsimane and Moseten (electronic supplementary material, table S7). The strongest loadings were for IL-1β, IL-12, IL-15 and TNF-β, while the weakest loadings were for IL-2, IL-17, IL-23 and IFN-γ. We included PC1, calculated with populations combined, in further analyses. Since it had a normal distribution (electronic supplementary material, figure S16), we standardized values to improve interpretability (mean = 0, s.d. = 1).

Table 1. Sample characteristics.

	Tsimane	Moseten
n	714	380
% female	51.3	48.2
number of samples	1134	423
mean age in years (s.d.)	61.2 (8.9)	58.3 (10.4)
age range (years)	39–94	39–85
mean BMI in kg m^{-2} (s.d.)	23.9 (3.7)	25.4 (4.9)
BMI range in kg m^{-2}	13.5–47.5	12.7–43.5
mean waist circumference in cm (s.d.)	90.5 (9.4)	94.5 (10.5)
waist circumference range in cm	61.5–130.0	68.0–163.6

(a) Tsimane ICCs varied widely, with some lower but others approximating US/European samples

The ICCs ranged from 0.27 for IFN-gγ, indicating lower between-person variation relative to within-person, to >0.70 for IL-1β, IL-12, IL-15, IL-23 and PC1, indicating higher between-person variation (table 2). The ICC for IL-6 was 0.49, indicating approximately equal variation between- versus within-individuals, while the ICC for IL-10 was 0.42, indicating slightly greater within-person variation relative to between-person. We also include Moseten ICCs, although owing to the smaller sample size these should be interpreted with caution and considered exploratory. Moseten ICCs tended to be higher compared with the Tsimane, although there was considerable overlap in 95% confidence intervals, consistent with the small sample size.

There are four published studies among post-industrialized populations that provided approximately comparable ICC estimates for most cytokines, based on age range and time between samples. These include a study of US female participants aged 35–65 years who provided serum samples 1-year apart [37], a US study of both male and female participants, aged 67–90 years, who provided serum samples approximately 18–24 months apart [34], a US study of male and female participants aged 55–70 years, who provided serum samples at baseline, 1 year post-baseline and 5 years post-baseline [38] and a study of Swedish female participants, aged 42–62 years, who provided plasma samples an average of 2 years apart [36]. While the Tsimane ICCs for some cytokines were lower than the US/European estimates (IL-2, IL-4, IL-5, IL-6, IL-10, IL-13, IL-17, IFN-gγ, TNF-α), others approximated them (IL-1β and IL-12). Further, while comparable estimates for IL-1α, IL-15, IL-23 and TNF-β were unavailable, these Tsimane ICCs approximated the US/European ICCs for other cytokines, ranging between 0.66–0.75.

(b) Minimal inflammaging in the Tsimane, greater inflammaging in the Moseten

Associations between age and cytokines in the Tsimane are shown in table 3. Owing to space limitations, we present models unadjusted for leukocytes in electronic supplementary tables s8–9. Age was not associated with TNF-α ($\beta = -0.001$, $p = 0.793$), while there was a non-significant negative association for IL-1β ($\beta = -0.012$, $p = 0.096$). There was a positive quadratic age association for IL-6 ($\beta = 0.001$ $p < 0.01$) and a positive linear association for IL-10 ($\beta = 0.009$, $p < 0.01$). Adjusting for leukocytes in the models strengthened the association between age and IL-6, which became linear (table 3). In these adjusted models, a one-year increase in age was associated with a 1.3% increase in IL-6 and 1.2% increase in IL-10 (both $p < 0.01$; figure 1). We also present results for the pro-inflammatory cytokines involved in cellular immunity (IL-2, IL-12, IL-15, IFN-γ) in table 3 and figure 2. Age was positively associated with IL-2 ($p < 0.05$) as well as IFN-γ after adjusting for leukocytes ($p < 0.05$). For both cytokines, a 1-year increase in age was associated with a 0.8% increase.

Similar to the Tsimane, both IL-6 and IL-10 were positively associated with age in the Moseten (table 4 and figure 1). A 1-year increase in age was associated with a 1.9% increase in IL-10 ($p < 0.01$). There was evidence for a quadratic association for IL-6, which increased and then attenuated at later ages (age $\beta = 0.025$, $p < 0.01$, age^2 $\beta = -0.001$, $p < 0.05$). In addition, a 1-year increase in age was associated with a 0.8% increase in IFN-γ ($p < 0.05$). However, in contrast to the Tsimane, age was positively associated with IL-1β and TNF-α in the Moseten (table 4 and figure 1), where a 1-year increase in age was associated with a 2.1% increase in IL-1β ($p < 0.05$) and a 1.1% increase in TNF-α ($p < 0.01$). In further contrast to the Tsimane, IL-2 was not associated with age ($\beta = 0.005$, $p = 0.288$), while both IL-12 and IL-15 were positively associated ($p < 0.05$; figure 2). A 1-year increase in age was associated with a 2.2% increase in IL-12 and a 1.7% increase in IL-15. Age associations for the Moseten were similar before versus after adjusting for leukocytes; however, they were slightly attenuated owing to the limited subsample size (electronic supplementary material, tables S10 and S11).

Age associations for the remaining cytokines and PC1 are shown in electronic supplementary material, tables S12–S15. These additional biomarkers were largely unassociated with age in either population, although there were some negative quadratic associations with TNF-β ($p < 0.01$) and PC1 ($p < 0.05$) in the Tsimane. In addition, PC1 was positively associated with age in the Moseten ($p < 0.05$), with a one-year increase being associated with a 0.014 higher s.d. score.

Table 2. Intra-class correlations.[a]

	Tsimane[b]		Moseten[c]		estimates from US/European samples[d]
	ICC	(95% CI)	ICC	(95% CI)	ICCs
pro-inflammatory markers centrally involved in inflammaging					
IL-1β	0.74	(0.67, 0.80)	0.86	(0.71, 0.93)	0.73[a]; 0.86[b]
IL-6	0.48	(0.41, 0.59)	0.44	(0.00, 0.69)	0.81[a]; 0.92[b]; 0.84[d]
TNF-α	0.45	(0.36, 0.56)	0.69	(0.45, 0.83)	0.69[a]; 0.88[b]; 0.28[c]; 0.86[d]
marker centrally involved in anti-inflammatory signalling					
IL-10	0.41	(0.30, 0.50)	0.60	(0.33, 0.76)	0.75[a]; 0.75[b]; 0.60[d]
pro-inflammatory markers of cell-mediated immunity (e.g., viral and tumour defence)					
IL-2	0.63	(0.53, 0.69)	0.70	(0.40, 0.84)	0.80[a]; 0.81[b]
IL-12	0.71	(0.64, 0.76)	0.87	(0.68, 0.93)	0.77[a]; 0.83[b]
IL-15	0.73	(0.65, 0.78)	0.81	(0.62, 0.92)	—
IFN-γ	0.27	(0.16, 0.38)	0.51	(0.15, 0.74)	0.72[a]; 0.80[c]
other pro-inflammatory markers (presented in electronic supplementary material)					
IL-1α	0.64	(0.53, 0.72)	0.66	(0.42, 0.86)	—
IL-17	0.43	(0.33, 0.53)	0.29	(0.00, 0.61)	0.78[c]
IL-23	0.76	(0.69, 0.83)	0.86	(0.73, 0.93)	—
TNF-β	0.67	(0.60, 0.73)	0.74	(0.50, 0.87)	—
other anti-inflammatory markers (presented in electronic supplementary material)					
IL-4	0.45	(0.33, 0.52)	0.69	(0.41, 0.86)	0.70[a]; 0.92[b]; 0.76[c]
IL-5	0.52	(0.37, 0.63)	0.63	(0.38, 0.82)	0.73[a]; 0.89[b]
IL-13	0.60	(0.51, 0.69)	0.75	(0.57, 0.88)	0.81[a]; 0.73[d]
principal component reflecting a composite of all cytokines (presented in electronic supplementary material)					
PC1	0.75	(0.69, 0.80)	0.85	(0.61, 0.93)	

[a]ICCs obtained from mixed effects models adjusting for age, sex and time between samples (mean time between measures = 2 years).
[b]$n = 138$ Tsimane provided 2 samples; $n = 74$ provided 3 samples; $n = 38$ provided 4 samples; $n = 5$ provided 5 samples.
[c]$n = 41$ Moseten provided 2 samples; $n = 1$ provided 3 samples
[d]US/European study ICC estimates come from Clendenen *et al*. [36][a], Gu *et al*. [37][b], Guo *et al*. [34][c], and Hofmann *et al*. [38][d].

(c) Female participants had higher cytokines across both populations

Female sex was associated with higher cytokines in both Tsimane and Moseten populations, although this difference was more pronounced in the Moseten. In the Tsimane, female sex versus male was associated with higher average concentrations for the following cytokines: IL−1β (35.9%), IL−5 (46.3%), IL−10 (15%), IL−12 (37.1%) and IL−15 (34%). Female sex versus male in the Tsimane was associated with a 0.203 higher s.d. score for PC1 (table 3 and electronic supplementary material, tables S12 and S13). In the Moseten, female sex versus male was associated with higher average concentrations for the following cytokines: IL−1β (56.9%), IL−2 (23.1%), IL−4 (47.3%), IL−5 (60.5%), IL−6 (30.6%), IL−10 (24.3%), IL−12 (53.4%), IL−13 (45.1%), IL−15 (49.7%), IL−17 (44.7%), TNF-α (18.3%) and TNF-β (52.7%). Female sex versus male in the Moseten was associated with a 0.411 higher s.d. score for PC1 (table 4 and electronic supplementary material, tables S14 and S15).

(d) Additional results and sensitivity analyses

To assess whether the cytokine values below the limit of detection were biasing the ICCs, we re-estimated models excluding those samples. These models showed similar results, suggesting that the replaced values were not systematically biasing toward higher ICCs (electronic supplementary material, table S16). We also ran additional Tobit regression models for censored data with the cytokines that had a large proportion of values below the limit of the detection. Using the lower limit of detection as the censoring threshold, these models provided very similar results compared with the OLS models presented in the main text (electronic supplementary material, tables S17–S20). In addition, since excess adiposity—particularly visceral fat deposition—might be a pathway linking lifestyle shifts to inflammaging, we considered additional models with BMI and waist circumference. BMI was not associated with the cytokines in either population (electronic supplementary material, tables S21 and S22). Waist circumference was not associated with the cytokines in the Moseten, whereas in the Tsimane it was positively associated with TNF-α ($p < 0.05$; electronic supplementary material, tables S23 and S24). Relatedly, we also considered interactions between age and distance from the nearest town of San Borja in the Tsimane to further assess the link between lifestyle shifts and inflammaging. These interaction terms were null; however, greater market distance was generally associated with higher cytokines (electronic supplementary material, table S25). Finally, a small number of cytokine values were above the limit of detection (<1%), which was mostly attributable to IL-23 and IFN-γ. We ran additional models excluding values above the limit of detection for these cytokines, which provided similar results (electronic supplementary material, tables S26 and S27).

Table 3. Tsimane age–cytokine (natural log-transformed) associations (full sample: 1134 measures from $n = 714$ individuals, subsample with leukocytes: 849 measures from $n = 536$).

cytokine	IL–1β	IL–1β	IL–6	IL–6	IL–10	IL–10	TNF-α	TNF-α
age[a] β (s.e.)	−0.009 (0.008)	−0.005 (0.009)	0.013** (0.005)	0.011* (0.005)	0.012** (0.003)	0.012** (0.004)	−0.00001 (0.003)	0.0002 (0.003)
age^2 β (s.e.)		−0.001 (0.001)		0.0004 (0.0004)		0.00001 (0.0003)		−0.00005 (0.0003)
female[b] β (s.e.)	0.359* (0.163)	0.354* (0.163)	0.061 (0.099)	0.063 (0.099)	0.150* (0.065)	0.150* (0.065)	0.074 (0.057)	0.074 (0.057)
leukocytes[c] β (s.e.)	0.217 (0.240)	0.238 (0.238)	0.605** (0.159)	0.595** (0.158)	0.399** (0.113)	0.399** (0.113)	0.071 (0.081)	0.072 (0.081)
intercept β (s.e.)	1.034 (0.573)	1.041 (0.568)	0.557 (0.369)	0.554 (0.370)	2.453** (0.277)	2.453** (0.278)	2.856** (0.188)	2.857** (0.189)

cytokine	IL–2	IL–2	IL–12	IL–12	IL–15	IL–15	IFN-γ	IFN-γ
age[a] β (s.e.)	0.008 (0.004)	0.009* (0.004)	−0.0003 (0.007)	0.006 (0.008)	0.008 (0.006)	0.013 (0.007)	0.008* (0.003)	0.011** (0.003)
age^2 β (s.e.)		−0.0002 (0.0004)		−0.001* (0.001)		−0.001 (0.0005)		−0.001 (0.0003)
female[b] β (s.e.)	0.007 (0.085)	0.005 (0.084)	0.371* (0.152)	0.363* (0.152)	0.340** (0.119)	0.334** (0.119)	−0.001 (0.061)	−0.005 (0.060)
leukocytes[c] β (s.e.)	0.223 (0.163)	0.229 (0.161)	−0.209 (0.220)	−0.177 (0.221)	0.253 (0.185)	0.277 (0.185)	0.129 (0.097)	0.143 (0.095)
intercept β (s.e.)	0.290 (0.385)	0.292 (0.385)	1.894** (0.527)	1.904** (0.526)	2.866** (0.441)	2.874** (0.441)	2.544** (0.224)	2.549** (0.219)

Clustered robust s.e. were applied to all models to account for repeated measures.
Models unadjusted for leukocytes are shown in electronic supplementary material, tables S8–S9
*$p < 0.05$; **$p < 0.01$.
[a]Mean-centred at 60 years.
[b]Coded 1 = female, 0 = male.
[c]Natural log-transformed.

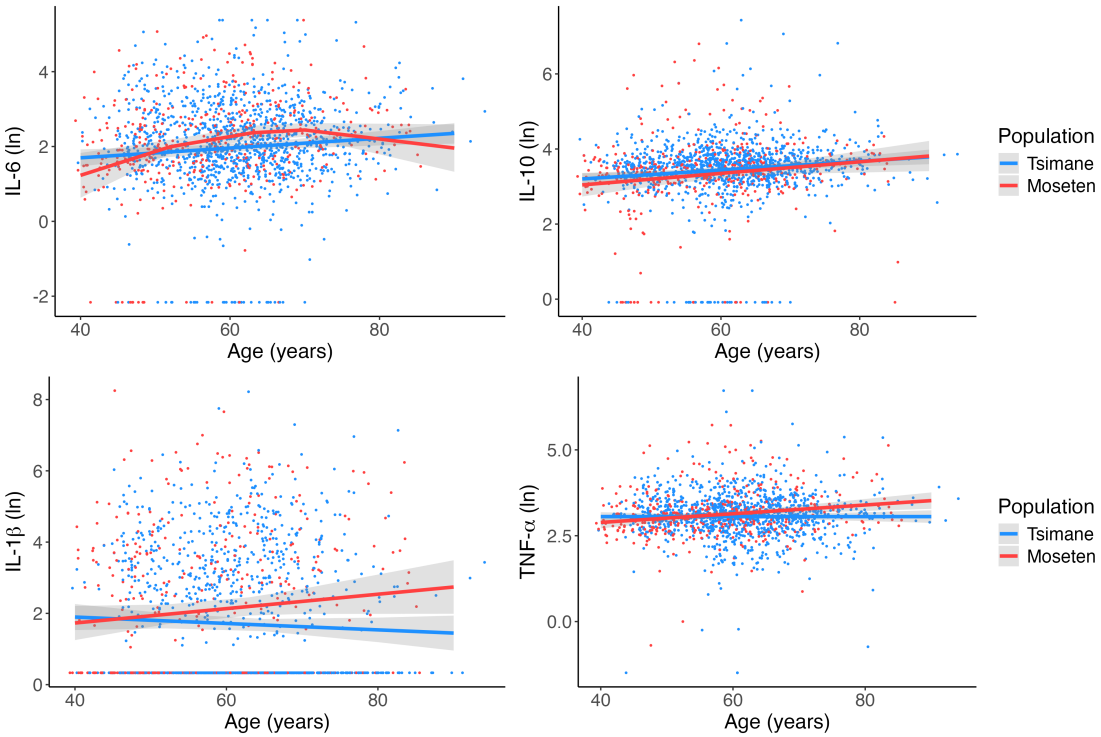

Figure 1. Age trends for Tsimane and Moseten cytokines (part 1). Lines reflect regression models shown in tables 3 and 4, with shaded 95% confidence intervals.

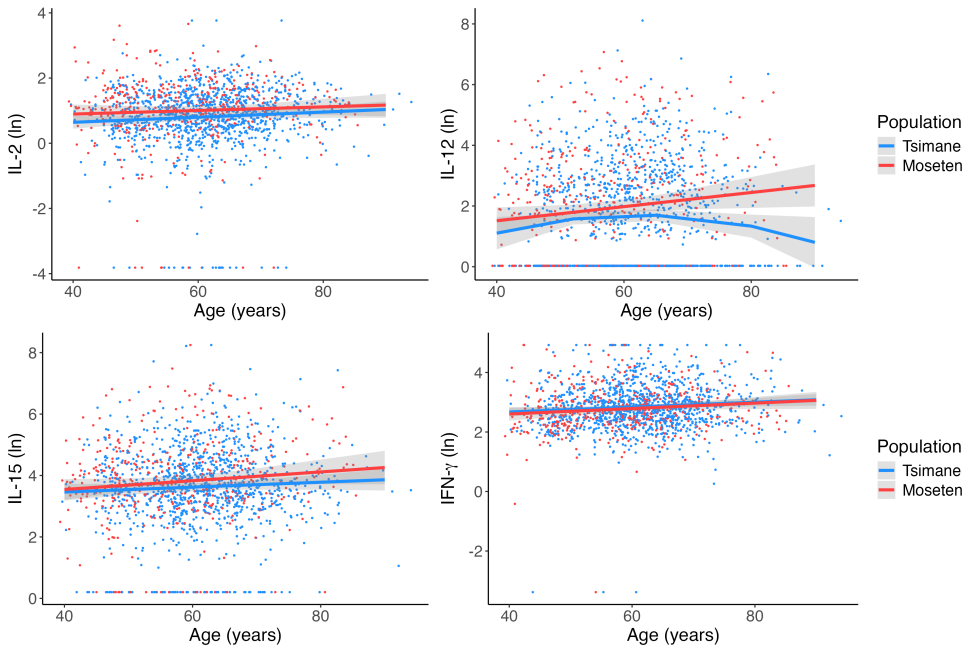

Figure 2. Age trends for Tsimane and Moseten cytokines (part 2). Lines reflect regression models shown in tables 3 and 4 with shaded 95% confidence intervals.

4. Discussion

While prior studies of inflammaging have been informative, a primary focus on post-industrialized populations precludes determination of whether inflammaging is a universal feature of ageing across populations, or if it is the product of environmental mismatch. To address this gap, we explored whether Tsimane forager-horticulturalists show evidence of inflammaging, reflected in the pro-inflammatory cytokines IL-1β, IL-6 and TNF-α. Inflammaging in the Tsimane appears to be minimal, with only IL-6 showing a weak positive association with age and no corresponding increase in IL-1β or TNF-α. In contrast, all three cytokines were positively associated with age in the neighbouring Moseten population, who are genetically similar but have experienced greater shifts towards a more industrialized lifestyle compared with the Tsimane. Our results indicate modest age-related increases in pro-inflammatory signalling within a subsistence population but also highlight inter-population variation in the breadth and magnitude of inflammaging. They are also consistent with the role of lifestyle shifts associated with market integration in growing health risk, including more pronounced inflammaging [46].

Our results are consistent with a previous Tsimane study finding a positive association between age and C-Reactive Protein (CRP), which is regulated upstream by IL−6 [11]. The positive association between age and IL−6 might reflect the accumulation of senescent cells with advancing age, which are known to produce this cytokine as part of the senescence-associated secretory phenotype (SASP) [47]. In contrast, the positive associations between age and IL−1β and TNF-α in the Moseten might partly reflect dysfunction and damage of the mitochondria. These organelles are essential for cellular metabolism and are one potential pathway linking caloric excess and sedentary behaviour to ageing and chronic disease [48,49]. Damaged and dysfunctional mitochondria increase reactive oxygen species (ROS) production, which activates the NF-κB inflammatory pathway and results in the production of IL−1β and TNF-α [27]. Further, mitochondrial function has been highlighted as a potential mechanism explaining the similarities in biomarker profiles between centenarians, who show limited inflammaging, and individuals undergoing caloric restriction [14]. These findings highlight the importance of future studies exploring mitochondrial damage and dysfunction in later life and its potential contribution to inflammaging in these populations.

The anti-inflammatory IL-10 was positively associated with age in both populations, with similar magnitudes. This is consistent with prior studies in other populations suggesting inflammaging does not simply reflect increases in pro-inflammatory signalling [45]. The increase in anti-inflammatory signalling might function to counterbalance pro-inflammatory signalling (anti-inflammaging) [26]. Alternatively, increases in IL-10 might reflect immunosenescence. For example, there is experimental evidence in mice that IL-10 plays a mechanistic role impairing later life immune responses [50]. The Tsimane show other evidence of immunosenescence, such as declining naive CD4+ T cells, which occurs at an accelerated rate compared with US data [11]. Future studies of immune response to antigenic challenge, including *in vivo* and *in vitro* assessments, may help clarify these possibilities.

Related to immunosenescence, the positive associations in both populations between age and cytokines involved in cell-mediated immunity might reflect the re-activation of latent viruses in later life with impaired immune surveillance. The cytokines IL−2, IL−12, IL−15 and IFN-γ are involved in anti-viral defence, and previous research suggests that latent viruses can be additional contributors to inflammaging [6,25]. Alternatively, these increases could reflect age-related immune remodelling, as these cytokines are also involved in augmenting programmed cell death and preventing tumour development [25,51]. Future studies on viral and cancer prevalence can help clarify these possibilities.

There is likely a complex relationship between infectious burden and inflammaging among the Tsimane. Most adults are infected with parasitic helminths that consume circulating glucose and lipids, potentially exerting protective cardiometabolic effects [19]. Further, helminths induce anti-inflammatory signalling in the host to inhibit immune clearance [21]. Their role in anti-inflammaging has been proposed previously [52], and it could mitigate chronic elevations of pro-inflammatory cytokines in the Tsimane. Therefore, while pathogens like viruses could potentially induce inflammaging, parasitic helminths might have the opposite effect. In addition, early-life microbial exposures are important immunomodulators, with potentially lasting effects into adulthood [7]. This might be another factor protecting the Tsimane from inflammaging. Future studies comparing generations, in which the older generation received more diverse microbial exposures in early life, can help decipher this possibility.

Owing to the more frequent infections experienced by the Tsimane and the resulting within-person cytokine variability, we expected lower ICCs compared with US/European samples. While this was the case for some cytokines, such as IL-6, others unexpectedly showed comparable estimates. These results suggest stable individual differences in immune function over time, which is likely owing to a combination of factors. First, given the large sample size spanning multiple communities, there could be differences in infectious and microbial exposures by community. Consistent with this, greater distance from the nearest town of San Borja was generally associated with higher cytokine concentrations. Second, there is evidence that genetic variation within the population contributes to individual differences in the immune response to these exposures [30,53].

The higher average BMI and waist circumference in the Moseten are consistent with their greater lifestyle shifts and rising health risk, including more pronounced inflammaging. However, these measures were not associated with cytokine variation. In addition, BMI was not associated with cytokine concentrations in the Tsimane, while waist circumference was only positively associated with TNF-α. We caution overinterpretation of these findings, given that BMI and waist circumference are not perfect proxies for visceral adiposity—the primary driver of chronic inflammation. They are also complicated by fat-free body mass, which is particularly salient given the high activity level of these populations, particularly the Tsimane. The relationship between adiposity and inflammation is also complicated by age, which could further explain the largely null results between waist circumference and markers of inflammaging. In early and middle adulthood, greater visceral adiposity drives meta-inflammation. However, adiposity tends to decline in late life, while inflammaging continues to develop and might in fact play a role in fat mass loss [54]. Therefore, while meta-inflammation and inflammaging are related, meta-inflammation precedes and augments inflammaging. Consistent with this, Moseten waist circumference declined with age (electronic supplementary material, figure S17). In addition, when we split the Moseten sample by median age, the younger group showed a marginally significant association between waist circumference and IL-6 ($\beta = 0.016$, $p < 0.10$), which was not present in the older subsample (electronic supplementary material, table S28). While the relationship with adiposity is complex, genetic data showing minimal differences between the Tsimane and Moseten [30], as well as dietary data showing greater market integration in the Moseten [18], suggest that lifestyle shifts likely explain their more pronounced inflammaging. Finally, town distance—a proxy for market integration—did not modify the age associations in the Tsimane. However, this is likely explained by the minimal overall age associations and resulting limited variability across communities.

While sex differences in cytokine concentrations were not a main focus of our study, adjusting for participant sex in regression models revealed that female participants across both populations tended to have higher cytokine concentrations compared with male participants. Stronger immune responses in females have been reported across species, and female sex is associated with greater risk for autoimmunity in post-industrialized populations [55,56]. While the mechanisms are complex and not fully understood, there is evidence for the roles of sex chromosomes and steroid hormones (oestrogen, progesterone,

Table 4. Moseten age–cytokine (natural log-transformed) associations (423 samples, $n = 380$).

cytokine	IL-1β	IL-1β	IL-6	IL-6	IL-10	IL-10	TNF-α	TNF-α
age[a]	0.021* (0.010)	0.021* (0.010)	0.025** (0.005)	0.025** (0.005)	0.019** (0.004)	0.018** (0.004)	0.011** (0.003)	0.011** (0.003)
β (s.e.)								
age^2		0.001 (0.001)		−0.001* (0.0004)		−0.0004 (0.0003)		−0.0003 (0.0003)
β (s.e.)								
female[b]	0.569** (0.213)	0.565** (0.213)	0.306* (0.121)	0.313** (0.121)	0.243* (0.102)	0.246* (0.102)	0.183* (0.071)	0.184** (0.071)
β (s.e.)								
intercept	1.845** (0.136)	1.777** (0.167)	2.027** (0.077)	2.130** (0.090)	3.286** (0.071)	3.331** (0.085)	3.090** (0.042)	3.119** (0.055)
β (s.e.)								

cytokine	IL-2	IL-2	IL-12	IL-12	IL-15	IL-15	IFN-γ	IFN-γ
age[a]	0.005 (0.004)	0.005 (0.004)	0.022* (0.009)	0.022* (0.009)	0.017* (0.007)	0.017* (0.007)	0.008* (0.004)	0.009* (0.004)
β (s.e.)								
age^2		−0.0001 (0.0004)		0.0003 (0.001)		0.0003 (0.001)		−0.0003 (0.0003)
β (s.e.)								
female[b]	0.231* (0.104)	0.231* (0.104)	0.534** (0.196)	0.531** (0.196)	0.497** (0.150)	0.495** (0.150)	0.115 (0.080)	0.117 (0.080)
β (s.e.)								
intercept	0.938** (0.086)	0.946** (0.105)	1.792** (0.125)	1.755** (0.159)	3.623** (0.104)	3.591** (0.129)	2.717** (0.059)	2.750** (0.073)
β (s.e.)								

Clustered robust standard errors were applied to all models to account for repeated measures.

*$p < 0.05$; **$p < 0.01$.

[a]Mean-centred at 60 years.

[b]Coded 1 = female, 0 = male.

testosterone) in the modulation of both innate and adaptive arms of the immune system [56]. However, sex differences in cytokine profiles and function are understudied, particularly in high-fertility populations [40,55,57,58]. A study of Italians reported higher IL-1β, IL-6 and TNF-α in men [59], while other studies have shown mixed results. For example, a sex difference in IL-1β was not found in the InCHIANTI study [60]. Men had higher average IL-6 in the US Health and Retirement Study (HRS) [61] but not in the Baltimore Longitudinal Study on Aging (BLSA) or Midlife in the United States (MIDUS) [62,63]. In contrast, women had higher average IL−6 in the Multi-Ethnic Study of Atherosclerosis (MESA) [64]. Multiple studies in Brazil, including a nationally representative study, have reported either mixed, weak, or no evidence for sex differences in cytokines [65–68]. While reasons for these discrepancies are not clear, our consistent results of higher average cytokine concentrations among female participants might reflect stronger immune responses to the highly infectious environment, although more research is needed.

Our study has important limitations. Our samples were largely cross-sectional, limiting our ability to assess changes within individuals over time. In addition, while the cytokines assessed are centrally involved in inflammaging, they are not an exhaustive list. The addition of other measures in future studies can further clarify inflammaging, such as high-throughput gene expression and proteomics. We were also limited by the detection range of the assay, which resulted in a large number of values falling below detection for some cytokines. However, our sensitivity analyses, including re-estimating ICCs without these values as well as Tobit regression models for censored data, suggest no bias owing to limits of detection. Another limitation is our sample size. With a larger sample, we might have found greater associations with age in the Tsimane. However, if a very large sample is required to detect more significant age trends, we think this reinforces our conclusion of minimal inflammaging. In comparison, despite the much smaller Moseten sample, we found more consistent associations between age and pro-inflammatory cytokines. Our cytokine measures were also influenced by technical variation, which we quantified using CVs in electronic supplementary material, table S1. Since this variation is random, it should not influence the population comparison. However, it could contribute to reduced statistical power and therefore possibly an underestimation of inflammaging. Finally, we did not use a multiple testing correction. This was because of our interest in assessing broad patterns across the cytokines, which would be obscured by a conservative significance threshold. Further, the number of significant age associations in the Moseten is much higher than would be expected by chance.

Here, we report minimal inflammaging among Tsimane forager-horticulturalists. These findings contribute to a growing body of work documenting variation in immune ageing trajectories across human populations, challenging the assumption of a universal inflammaging profile. While we find some increase in pro-inflammatory signalling, reflected in IL-6, we did not find increases in other notable pro-inflammatory markers including IL−1β and TNF-α. The more pronounced inflammaging observed in the Moseten, who are genetically similar to the Tsimane and whose samples were analysed simultaneously, points to lifestyle and environmental shifts as primary drivers, rather than genetic differences or batch effects. Future research can help clarify the relationship between lifestyle shifts and inflammaging, for example by incorporating direct measures of visceral adiposity. Studies are also needed to assess the relationship between infectious and early-life microbial exposures and inflammaging. Finally, future studies are needed to understand the potential health implications of these profiles, such as their relationship with immunosenescence, cardiometabolic health, brain ageing, frailty and all-cause mortality.

Ethics. Informed consent was given by the Tsimane Government (Gran Consejo Tsimane), village leaders and all study participants directly. Methods and procedures were approved by the Institutional Review Boards of the University of New Mexico (HRRC #07-157; #15-133; #17-230), University of California Santa Barbara (HRRC #28-21-0788) and the Universidad San Simon Cochabamba, Bolivia.

Data accessibility. Individual-level data are stored in the Tsimane Health and Life History Project (THLHP) Data Repository and are available through restricted access for ethical reasons. THLHP's highest priority is the safeguarding of human subjects and minimization of risk to study participants. The THLHP adheres to the 'CARE Principles for Indigenous Data Governance' (Collective Benefit, Authority to Control, Responsibility and Ethics), which assure that the Tsimane (i) have sovereignty over how data are shared, (ii) are the primary gatekeepers determining ethical use, (iii) are actively engaged in the data generation, and (iv) derive benefit from data generated and shared for use whenever possible. The THLHP is also committed to the 'FAIR Guiding Principles for scientific data management and stewardship' (Findable, Accessible, Interoperable, Reusable). Requests for individual-level data should take the form of an application that details the exact uses of the data and the research questions to be addressed, procedures that will be employed for data security and individual privacy, potential benefits to the study communities and procedures for assessing and minimizing stigmatizing interpretations of the research results (see the following webpage for links to the data sharing policy and data request forms: https://tsimane.anth.ucsb.edu/data.html). Requests for individual-level data will require institutional IRB approval (even if exempt) and will be reviewed by an Advisory Council composed of Tsimane community leaders, community members, Bolivian scientists and the THLHP leadership. The study authors and the THLHP leadership are committed to open science and are available to assist interested investigators in preparing data access requests. Data from the tables and figures, as well as the analysis code, can be found in a Dryad repository [69].

Supplementary material is available online [70].

Declaration of AI use. We have not used AI-assisted technologies in creating this article.

Authors' contributions. J.E.A.: conceptualization, formal analysis, visualization, writing—original draft; C.L.J.: data curation, methodology, writing—review and editing; A.R.G.: data curation, methodology, writing—review and editing; S.V.K.: data curation, methodology, writing—review and editing; S.G.: data curation, project administration; K.L.W.: data curation, methodology; M.C.: data curation, methodology; I.M.S.: data curation, methodology, project administration; D.E.R.: data curation, methodology, project administration, writing—review and editing; B.B.: data curation, project administration, writing—review and editing; D.K.C.: data curation, project administration, writing—review and editing; P.L.H.: data curation, project administration, writing—review and editing; T.K.K.: data curation, writing—review and editing; K.B.: funding acquisition, project administration, writing—review and editing; C.E.F.: funding acquisition, writing—review and editing; M.F.: writing—review and editing; A.A.C.: writing—review and editing; J.S.: funding acquisition, project administration, writing—review and editing; M.G.: funding acquisition, project administration, writing—review and editing; H.K.: conceptualization, funding acquisition, project administration, supervision, writing—review and editing; B.C.T.: conceptualization, data curation, funding acquisition, methodology, project administration, supervision, writing—review and editing.

All authors gave final approval for publication and agreed to be held accountable for the work performed therein.

Conflict of interest declaration. We declare we have no competing interests.

Funding. Funding was provided by the NIH/National Institute on Aging (R01AG054442 and RF1AG054442) and the NSF (1748282). J.S. acknowledges IAST funding from the French National Research Agency (ANR) under the Investments for the Future (Investissements d'Avenir) program, grant ANR-17-EURE-0010. Funding sources had no role in research conduct, study design or article preparation.

Acknowledgements. We thank the Tsimane and Moseten greatly for participating and Tsimane Health and Life History Project personnel for collecting and coding data. We also thank the tribal leadership organizations that helped guide this research: Gran Consejo Tsimane, Consejo Regional Tsimane Moseten and the Organization of the Moseten Indigenous People.

References

1. Franceschi C, Bonafè M, Valensin S, Olivieri F, De Luca M, Ottaviani E, De Benedictis G. 2000 Inflamm-aging: an evolutionary perspective on immunosenescence. *Ann. NY Acad. Sci.* **908**, 244–254. (doi:10.1111/j.1749-6632.2000.tb06651.x)
2. Franceschi C, Campisi J. 2014 Chronic inflammation (inflammaging) and its potential contribution to age-associated diseases. *J. Gerontol. Ser. A* **69**, S4–S9. (doi:10.1093/gerona/glu057)
3. Escrig-Larena JI, Delgado-Pulido S, Mittelbrunn M. 2023 Mitochondria during T cell aging. *Semin. Immunol*. **69**, 101808. (doi:10.1016/j.smim.2023.101808)
4. Franceschi C, Garagnani P, Vitale G, Capri M, Salvioli S. 2017 Inflammaging and 'Garb-aging'. *Trends Endocrinol. Metab.* **28**, 199–212. (doi:10.1016/j.tem.2016.09.005)
5. Fulop T, Larbi A, Pawelec G, Khalil A, Cohen AA, Hirokawa K, Witkowski JM, Franceschi C. 2021 Immunology of aging: the birth of inflammaging. *Clin. Rev. Allergy Immunol*. **64**, 109–122. (doi:10.1007/s12016-021-08899-6)
6. Batista MA *et al*. 2020 Inflammaging in endemic areas for infectious diseases. *Front. Immunol*. **11**, 579972. (doi:10.3389/fimmu.2020.579972)
7. McDade TW. 2023 Three common assumptions about inflammation, aging, and health that are probably wrong. *Proc. Natl Acad. Sci. USA* **120**, e2317232120. (doi:10.1073/pnas.2317232120)
8. Trumble BC, Finch CE. 2019 The exposome in human evolution: from dust to diesel. *Q. Rev. Biol*. **94**, 333–394. (doi:10.1086/706768)
9. Gurven MD, Lieberman DE. 2020 WEIRD bodies: mismatch, medicine and missing diversity. *Evol. Hum. Behav*. **41**, 330–340. (doi:10.1016/j.evolhumbehav.2020.04.001)
10. Gurven M, Stieglitz J, Trumble B, Blackwell AD, Beheim B, Davis H, Hooper P, Kaplan H. 2017 The Tsimane Health and Life History Project: Integrating anthropology and biomedicine. *Evol. Anthropol*. **26**, 54–73. (doi:10.1002/evan.21515)
11. Blackwell AD, Trumble BC, Maldonado Suarez I, Stieglitz J, Beheim B, Snodgrass JJ, Kaplan H, Gurven M. 2016 Immune function in Amazonian horticulturalists. *Ann. Hum. Biol*. **43**, 382–396. (doi:10.1080/03014460.2016.1189963)
12. Kaplan H *et al*. 2017 Coronary atherosclerosis in indigenous South American Tsimane: a cross-sectional cohort study. *Lancet* **389**, 1730–1739. (doi:10.1016/s0140-6736(17)30752-3)
13. Gatz M *et al*. 2023 Prevalence of dementia and mild cognitive impairment in indigenous Bolivian forager-horticulturalists. *Alzheimer's Dement*. **19**, 44–55. (doi:10.1002/alz.12626)
14. Franceschi C, Ostan R, Santoro A. 2018 Nutrition and inflammation: are centenarians similar to individuals on calorie-restricted diets? *Annu. Rev. Nutr*. **38**, 329–356. (doi:10.1146/annurev-nutr-082117-051637)
15. Aronoff JE, Trumble BC. 2025 An evolutionary medicine and life history perspective on aging and disease: trade-offs, hyperfunction, and mismatch. *Evol. Med. Public Health* **13**, 111–124. (doi:10.1093/emph/eoaf010)
16. Chen G, Yung R. 2019 Meta-inflammaging at the crossroad of geroscience. *AGING Med*. **2**, 157–161. (doi:10.1002/agm2.12078)
17. Hotamisligil GS. 2017 Inflammation, metaflammation and immunometabolic disorders. *Nature* **542**, 177–185. (doi:10.1038/nature21363)
18. Kraft TS, Stieglitz J, Trumble BC, Martin M, Kaplan H, Gurven M. 2018 Nutrition transition in 2 lowland Bolivian subsistence populations. *Am. J. Clin. Nutr*. **108**, 1183–1195. (doi:10.1093/ajcn/nqy250)
19. Gurven MD, Trumble BC, Stieglitz J, Blackwell AD, Michalik DE, Finch CE, Kaplan HS. 2016 Cardiovascular disease and type 2 diabetes in evolutionary perspective: a critical role for helminths? *Evol. Med. Public Health* **2016**, 338–357. (doi:10.1093/emph/eow028)
20. Gurven MD, Trumble BC, Stieglitz J, Yetish G, Cummings D, Blackwell AD, Beheim B, Kaplan HS, Pontzer H. 2016 High resting metabolic rate among Amazonian forager-horticulturalists experiencing high pathogen burden. *Am. J. Phys. Anthropol*. **161**, 414–425. (doi:10.1002/ajpa.23040)
21. Blackwell AD. 2022 The ecoimmunology of health and disease: the hygiene hypothesis and plasticity in human immune function. *Annu. Rev. Anthropol*. **51**, 401–418. (doi:10.1146/annurev-anthro-101819-110236)
22. Gurven M, Kaplan H, Winking J, Finch C, Crimmins EM. 2008 Aging and inflammation in two epidemiological worlds. *Journals Gerontol. Ser*. **63**, 196–199. (doi:10.1093/gerona/63.2.196)
23. Vasunilashorn S, Crimmins EM, Kim JK, Winking J, Gurven M, Kaplan H, Finch CE. 2010 Blood lipids, infection, and inflammatory markers in the Tsimane of Bolivia. *Am. J. Hum. Biol*. **22**, 731–740. (doi:10.1002/ajhb.21074)
24. Liu C, Chu D, Kalantar-Zadeh K, George J, Young HA, Liu G. 2021 Cytokines: from clinical significance to quantification. *Adv. Sci*. **8**, e2004433. (doi:10.1002/advs.202004433)
25. Akdis M *et al*. 2011 Interleukins, from 1 to 37, and interferon-γ: receptors, functions, and roles in diseases. *J. Allergy Clin. Immunol*. **127**, 701–721.(doi:10.1016/j.jaci.2010.11.050)
26. Minciullo PL *et al*. 2016 Inflammaging and anti-inflammaging: the role of cytokines in extreme longevity. *Arch. Immunol. Et Ther. Exp*. **64**, 111–126. (doi:10.1007/s00005-015-0377-3)
27. Rea IM, Gibson DS, McGilligan V, McNerlan SE, Alexander HD, Ross OA. 2018 Age and age-related diseases: role of inflammation triggers and cytokines. *Front. Immunol*. **9**, 334076. (doi:10.3389/fimmu.2018.00586)
28. Gurven M, Jaeggi AV, Kaplan H, Cummings D. 2013 Physical activity and modernization among Bolivian Amerindians. *PLoS One* **8**, e55679. (doi:10.1371/journal.pone.0055679)
29. Kaplan H *et al*. 2023 Brain volume, energy balance, and cardiovascular health in two nonindustrial South American populations. *Proc. Natl Acad. Sci. USA* **120**, e2205448120. (doi:10.1073/pnas.2205448120)
30. Lea AJ *et al*. 2023 Natural selection of immune and metabolic genes associated with health in two lowland Bolivian populations. *Proc. Natl Acad. Sci. USA* **120**, e2207544120. (doi:10.1073/pnas.2207544120)
31. Sakel J. 2009 *Mosetén y chimane (tsimane')*. La Paz, Bolivia: Plurales Editores.
32. Mcdade TW, Tallman PS, Madimenos FC, Liebert MA, Cepon TJ, Sugiyama LS, Snodgrass JJ. 2012 Analysis of variability of high sensitivity C-reactive protein in lowland Ecuador reveals no evidence of chronic low-grade inflammation. *Am. J. Hum. Biol*. **24**, 675–681. (doi:10.1002/ajhb.22296)

33. Urlacher SS, Ellison PT, Sugiyama LS, Pontzer H, Eick G, Liebert MA, Cepon-Robins TJ, Gildner TE, Snodgrass JJ. 2018 Tradeoffs between immune function and childhood growth among Amazonian forager-horticulturalists. *Proc. Natl Acad. Sci. USA* **115**, E3914–E3921. (doi:10.1073/pnas.1717522115)
34. Guo J, Schupf N, Mayeux RP, Gu Y. 2020 Reproducibility of serum cytokines in an elderly population. *Immun. Age.* **17**, 1–10. (doi:10.1186/s12979-020-00201-0)
35. Koo TK, Li MY. 2016 A guideline of selecting and reporting intraclass correlation coefficients for reliability research. *J. Chiropr. Med*. **15**, 155–163. (doi:10.1016/j.jcm.2016.02.012)
36. Clendenen TV *et al*. 2010 Temporal reliability of cytokines and growth factors in EDTA plasma. *BMC Res. Notes* **3**, 302. (doi:10.1186/1756-0500-3-302)
37. Gu Y *et al*. 2009 Reproducibility of serum cytokines and growth factors. *Cytokine* **45**, 44–49. (doi:10.1016/j.cyto.2008.10.014)
38. Hofmann JN, Yu K, Bagni RK, Lan Q, Rothman N, Purdue MP. 2011 Intra-individual variability over time in serum cytokine levels among participants in the prostate, lung, colorectal, and ovarian cancer screening trial. *Cytokine* **56**, 145–148. (doi:10.1016/j.cyto.2011.06.012)
39. Dinkel KA, Costa ME, Kraft TS, Stieglitz J, Cummings DK, Gurven M, Kaplan H, Trumble BC. 2020 Relationship of sanitation, water boiling, and mosquito nets to health biomarkers in a rural subsistence population. *Am. J. Hum. Biol*. **32**, e23356. (doi:10.1002/ajhb.23356)
40. Trumble BC *et al*. 2023 *Apolipoprotein- ε 4* is associated with higher fecundity in a natural fertility population. *Sci. Adv*. **9**, eade9797. (doi:10.1126/sciadv.ade9797)
41. Blackwell AD, Snodgrass JJ, Madimenos FC, Sugiyama LS. 2010 Life history, immune function, and intestinal helminths: trade-offs among immunoglobulin E, C-reactive protein, and growth in an Amazonian population. *Am. J. Hum. Biol*. **22**, 836–848. (doi:10.1002/ajhb.21092)
42. Blackwell AD, Tamayo MA, Beheim B, Trumble BC, Stieglitz J, Hooper PL, Martin M, Kaplan H, Gurven M. 2015 Helminth infection, fecundity, and age of first pregnancy in women. *Science* **350**, 970–972. (doi:10.1126/science.aac7902)
43. Garcia AR, Blackwell AD, Trumble BC, Stieglitz J, Kaplan H, Gurven MD. 2020 Evidence for height and immune function trade-offs among preadolescents in a high pathogen population. *Evol. Med. Public Health* **2020**, 86–99. (doi:10.1093/emph/eoaa017)
44. Gurven MD, Kaplan H, Supa AZ. 2007 Mortality experience of Tsimane Amerindians of Bolivia: Regional variation and temporal trends. *Am. J. Hum. Biol.* **19**, 376–398. (doi:10.1002/ajhb.20600)
45. Morrisette-Thomas V, Cohen AA, Fülöp T, Riesco É, Legault V, Li Q, Milot E, Dusseault-Bélanger F, Ferrucci L. 2014 Inflamm-aging does not simply reflect increases in pro-inflammatory markers. *Mech. Ageing Dev*. **139**, 49–57. (doi:10.1016/j.mad.2014.06.005)
46. Wong GCL *et al*. 2021 Horticultural therapy reduces biomarkers of immunosenescence and inflammaging in community-dwelling older adults: a feasibility pilot randomized controlled trial. *J. Gerontol*. **76**, 307–317. (doi:10.1093/gerona/glaa271)
47. Mosteiro L *et al*. 2016 Tissue damage and senescence provide critical signals for cellular reprogramming *in vivo*. *Science* **354**, f4445. (doi:10.1126/science.aaf4445)
48. Amorim JA, Coppotelli G, Rolo AP, Palmeira CM, Ross JM, Sinclair DA. 2022 Mitochondrial and metabolic dysfunction in ageing and age-related diseases. *Nat. Rev. Endocrinol*. **18**, 243–258. (doi:10.1038/s41574-021-00626-7)
49. Diaz-Vegas A, Sanchez-Aguilera P, Krycer JR, Morales PE, Monsalves-Alvarez M, Cifuentes M, Rothermel BA, Lavandero S. 2020 Is mitochondrial dysfunction a common root of noncommunicable chronic diseases? *Endocr. Rev*. **41**, a005. (doi:10.1210/endrev/bnaa005)
50. Almanan M *et al*. 2020 IL-10–producing Tfh cells accumulate with age and link inflammation with age-related immune suppression. *Sci. Adv*. **6**, eabb0806. (doi:10.1126/sciadv.abb0806)
51. Castro F, Cardoso AP, Gonçalves RM, Serre K, Oliveira MJ. 2018 Interferon-gamma at the crossroads of tumor immune surveillance or evasion. *Front. Immunol*. **9**, 847. (doi:10.3389/fimmu.2018.00847)
52. Zhang B, Gems D. 2021 Gross ways to live long: parasitic worms as an anti-inflammaging therapy? *eLife* **10**, e65180. (doi:10.7554/elife.65180)
53. Vasunilashorn S, Finch CE, Crimmins EM, Vikman SA, Stieglitz J, Gurven M, Kaplan H, Allayee H. 2011 Inflammatory gene variants in the Tsimane, an indigenous Bolivian population with a high infectious load. *Biodemography Soc. Biol*. **57**, 33–52. (doi:10.1080/19485565.2011.564475)
54. Shaulson ED, Cohen AA, Picard M. 2024 The brain–body energy conservation model of aging. *Nat. Aging* **4**, 1354–1371. (doi:10.1038/s43587-024-00716-x)
55. Natri H, Garcia AR, Buetow KH, Trumble BC, Wilson MA. 2019 The pregnancy pickle: evolved immune compensation due to pregnancy underlies sex differences in human diseases. *Trends Genet*. **35**, 478–488. (doi:10.1016/j.tig.2019.04.008)
56. Dunn SE, Perry WA, Klein SL. 2024 Mechanisms and consequences of sex differences in immune responses. *Nat. Rev. Nephrol*. **20**, 37–55. (doi:10.1038/s41581-023-00787-w)
57. Stieglitz J, Trumble BC, Finch CE, Li D, Budoff MJ, Kaplan H, Gurven MD. 2019 Computed tomography shows high fracture prevalence among physically active forager-horticulturalists with high fertility. *eLife* **8**, e48607. (doi:10.7554/elife.48607)
58. Gurven M, Costa M, Stieglitz J, Beheim B, Eid Rodriguez D, Hooper PL, Kaplan H. 2016 Health costs of reproduction are minimal despite high fertility, mortality and subsistence lifestyle. *Sci. Rep*. **6**, 30056. (doi:10.1038/srep30056)
59. Bernardi S *et al*. 2020 Sex differences in proatherogenic cytokine levels. *Int. J. Mol. Sci*. **21**, 3861. (doi:10.3390/ijms21113861)
60. Di Iorio A *et al*. 2003 Serum IL-1β levels in health and disease: a population-based study. 'The InCHIANTI study.' *Cytokine* **22**, 198–205. (doi:10.1016/s1043-4666(03)00152-2)
61. Kalair A, Pavan M, Alpert N, Ghaffari S, Taioli E. 2023 Blood inflammatory markers and mortality in the US population: a Health and Retirement Survey (HRS) analysis. *PLoS One* **18**, e0293027. (doi:10.1371/journal.pone.0293027)
62. Lustig A, Liu HB, Metter EJ, An Y, Swaby MA, Elango P, Ferrucci L, Hodes RJ, Weng N-p. 2017 Telomere shortening, inflammatory cytokines, and anti-Cytomegalovirus antibody follow distinct age-associated trajectories in humans. *Front. Immunol*. **8**, 1027. (doi:10.3389/fimmu.2017.01027)
63. Hartanto A, Majeed NM, Ng WQ, Chai CKN, Lua VYQ. 2021 Subjective age and inflammation risk in midlife adults: findings from the midlife in the United States (MIDUS) studies. *Comprehensive Psychoneuroendocrinol.* **7**, 100072. (doi:10.1016/j.cpnec.2021.100072)
64. Cainzos-Achirica M *et al*. 2018 The prognostic value of interleukin 6 in multiple chronic diseases and all-cause death: the multi-ethnic study of atherosclerosis (MESA). *Atherosclerosis* **278**, 217–225. (doi:10.1016/j.atherosclerosis.2018.09.034)
65. Valmorbida A, Longo GZ, Nascimento GM, de Oliveira LL, de Moraes Trindade EBS. 2023 Association between cytokine levels and anthropometric measurements: a population-based study. *Br. J. Nutr*. **129**, 1119–1126. (doi:10.1017/s0007114522002148)
66. Torres KCL *et al*. 2018 Immune senescence and biomarkers profile of Bambuí aged population-based cohort. *Exp. Gerontol*. **103**, 47–56. (doi:10.1016/j.exger.2017.12.006)
67. Milan-Mattos JC *et al*. 2019 Effects of natural aging and gender on pro-inflammatory markers. *Braz. J. Med. Biol. Res*. **52**, e8392. (doi:10.1590/1414-431x20198392)
68. Lima-Silva ML, Torres KCL, Mambrini JVdM, Brot NC, Santos SO, Martins-Filho OA, Teixeira-Carvalho A, Lima-Costa MF, Peixoto SV. 2024 A nationwide study on immunosenescence biomarkers profile in older adults: ELSI-Brazil. *Exp. Gerontol*. **191**, 112433. (doi:10.1016/j.exger.2024.112433)
69. Aronoff JE *et al*. 2025 Data for: Inflammaging is minimal among forager-horticulturalists in the Bolivian Amazon. Dryad Digital Repository. (doi:10.5061/dryad.2v6wwq00q)
70. Aronoff JE, Jenkins CL, Garcia AR, Koebele SV, Ghafoor S, Woolard KL *et al*. 2025 Supplementary material from: Inflammaging is minimal among forager-horticulturalists in the Bolivian Amazon. Figshare (doi:10.6084/m9.figshare.c.7921356)