



## Research paper

# Malaria, mental disorders, immunity and their inter-relationships - A cross sectional study in a household population in a health and demographic surveillance site in Kenya



Rachel Jenkins<sup>a,\*</sup>, Michael Ong'echa<sup>b</sup>, Caleb Othieno<sup>c</sup>, Linnet Ongeri<sup>b</sup>, Peter Sifuna<sup>d</sup>, Raymond Omollo<sup>b</sup>, Brian Leonard<sup>e</sup>, Bernhards Ogutu<sup>b</sup>

<sup>a</sup> Kings College London, UK

<sup>b</sup> Kenya Medical Research Institute, Kenya

<sup>c</sup> University of Nairobi, Kenya

<sup>d</sup> Kombewa West HDSS, Kenya

<sup>e</sup> National University of Ireland, Galway, Ireland

## ARTICLE INFO

## Article history:

Received 21 August 2018

Received in revised form 25 November 2018

Accepted 30 November 2018

Available online 11 December 2018

## Keywords:

Malaria

Immunity

Cytokines

Mental health

Common mental disorder

Psychotic symptoms

## Search terms:

Used were: malaria and immunity

Common mental disorders and immunity

Depression and immunity

Psychosis and immunity

Schizophrenia and immunity

Malaria, mental disorders and immunity

## ABSTRACT

**Background:** Both malaria and mental disorders are associated with immune changes. We have previously reported the associations between malaria and mental disorders. We now report associations between malaria, mental disorders and immunity.

**Methods:** A household survey of malaria, mental disorders and immunity was conducted in a health and demographic surveillance system's site of 70,000 population in an area endemic for malaria in western Kenya. A random sample of 1190 adults was selected and approached for consent, blood samples and structured interview.

**Findings:** We found marginally raised CD4/CD3 ratios of participants with malaria parasites, but no difference in CD4/CD3 ratios for participants with common mental disorder (CMD) or psychotic symptoms. People with psychotic symptoms had increased levels of IL-6, IL-8, and IL-10, and lower levels of IL-1beta. People with CMD had higher levels of IL-8 and IL-10. People with malaria had higher levels of IL-10 and lower levels of TNF-alpha. At the bivariate level, CMD was associated with log TNF-alpha levels using unadjusted odds ratios, but not after adjusting for malaria. Psychotic symptoms were associated with log IL-10 and log TNF-alpha levels at the bivariate level while in the adjusted analysis, log TNF-alpha levels remained highly significant.

**Interpretation:** This is the first population based study of immune markers in CMD and psychotic symptoms, and the first to examine the 3 way relationship with malaria. Our findings suggest that TNF-alpha may mediate the relationship between malaria and CMD.

**Fund:** The study was funded by UK Aid, Department for International Development, Kenya office.

© 2018 Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

## 1. Introduction

Health systems are unprepared for the transition from communicable to non-communicable disease burden [1], and for the complexity of their inter-relationships. There is long standing and extensive evidence of the two way association between mental and physical disorders, mostly focussed on non-communicable disease [2], but there has been rather less attention to the relationship of mental disorders and communicable disease, apart from HIV [3]. Malaria is endemic in Kenya around Lake Victoria, and we have previously reported adult population prevalence rates for malaria [4] and for mental disorders [5–10], as well

as the two way association of malaria and common mental disorder (CMD) in a health and demographic surveillance systems site in that region [11]. This paper reports their associations with inflammatory mediators.

Inflammation is a reflection of cell damage caused by infection, or physical injury or in the response of tissues to an antigenic challenge, and was first studied in relation to physical disease, but in recent years numerous clinical studies have established the presence of chronic low grade inflammation in major psychiatric disorders such as the affective and psychotic disorders and the anxiety disorders [10–15]. Thus, cytokines have been associated with the pathogenesis of not only physical disorders such as malaria [16–19], but also with depression [20–22] and schizophrenia [23]. It has been argued that the inflammasome is a central mediator by which psychological and

\* Corresponding author.

E-mail address: [rachel.1.jenkins@kcl.ac.uk](mailto:rachel.1.jenkins@kcl.ac.uk) (R. Jenkins).

## Research in Context

### *Evidence before this study*

Both malaria and mental disorders have been separately shown to be influenced by non-cellular immunity and several cytokines have been found in the pathogenesis of malaria, depression and schizophrenia.

Data bases searched include PubMed.

### *Added value of this study*

This is the first population based study of immune markers in the specific entities of CMD and Psychotic symptoms, as identified by the Clinical Interview Schedule – revised, and the Psychotic Symptoms Questionnaire, and the first study to examine the 3 way relationship between psychopathology, immunity and malaria. Our findings support the key role of TNF- $\alpha$  in the pathogenesis of both CMD and psychotic symptoms, and suggest that TNF- $\alpha$  may mediate the relationship between malaria and CMD. The study also indicates that the PSQ is measuring a biologically identifiable group or more likely a heterogeneous combination of sub-groups with excessive inflammation affecting the brain.

### *Implications of all the available evidence for policy and research*

Immunity is an important risk factor for both malaria and mental disorders in this adult household population in rural Kenya, and this information is relevant for public health measures, for training and supervision of health workers, and for health management information systems. It is possible that the relationship between malaria and CMD may be mediated by non-cellular immunity, specifically TNF- $\alpha$ , and this needs further exploration.

physical stressors can contribute to the development of depression, as well as a bridge to systemic diseases [12]. Pro-inflammatory cytokines are elevated in depressed patients, and can cause somatic symptoms such as fatigue and appetite loss [22]. Systemic administration of TNF in mice causes depressive behaviours such as reduced social interaction, increased despair behaviour, decreased food intake and weight loss [24].

While the 3 way relationship between inflammation, non-communicable diseases (eg cardiovascular disease, diabetes, Parkinson's and some cancers) and mental disorders has been extensively studied [25], there has been less research on the 3 way relationship between inflammation, communicable disease and mental disorders, apart from HIV where there is growing understanding of the 3 way relationship between inflammation, HIV and mental disorders [26]. The pro-inflammatory cytokines stimulated by HIV deplete tryptophan which is associated with depression, and such depression may reduce compliance with therapy. The 3 way inter-relationship between inflammation, malaria and mental disorders has not to our knowledge been previously explored. Therefore, the purpose of this community based survey, on an adult household population in a demographic surveillance site in an area of Kenya endemic for malaria, was to examine the associations between malaria infection, mental disorders and inflammation.. To our knowledge this is the first community based study to explore the possible relationship between malaria induced inflammation and mental ill health.

## 2. Materials and methods

The methods of the survey have been described in detail elsewhere [4–6]. The survey was conducted in Kisumu County, specifically in Maseno area . (see Fig. 1), using the

Kombewa Health and Demographic Surveillance System (Kombewa HDSS) run by the KEMRI/Walter Reed Project [27]. Community engagement was undertaken prior to commencement of data collection, during which the communities via their community leaders received information about the study and an opportunity to discuss the project. A random sample of 7 households was drawn from each enumeration area to give a projected sample of 1190 households, which with an estimated response rate of 85% would give a total sample size of 1010. Households were identified using village maps, which were also used to guide the research assistants The Kombewa HDSS field guide identified the sampled households for data collection by the research assistants and study supervisors. A total of 1190 households were visited. One individual was chosen from each of the sampled households, using the Kish Grid Method. The research assistants recorded demographics and reasons for the refusal in notebooks.. The survey participants were approached in their own homes for informed consent (written and witnessed). The presence of malaria parasites was appraised by the collection of venous blood samples for preparation of thick and thin blood films for malaria parasites, and for immunological assays. The participants also received a comprehensive interview covering sociodemographic and mental health variables.

The field staff (2 team supervisors and 20 research assistants) were trained for 5 days in the KEMRI field centre in Kisumu, Kenya. The training included the procedures for selection of individuals using the Kish Grid Method, informed consent, recording of reasons for refusal, blood samples, and interviews, including the use of PDAs to collect the interview data... . The questionnaires were programmed on to the PDAs and tested before the actual field work. The sociodemographic and mental health data were recorded on the PDA held by each research assistant; and the data were then transferred at the end of each day to a personal computer for merging, and for subsequent transmission to the statistician for analysis.

The comprehensive interview received by each participant comprised a structured epidemiological assessment of common mental disorders, psychotic symptoms, alcohol and substance abuse, ADHD, PTSD, suicidal thoughts and attempts, accompanied by additional sections on socio-demographic data, life events, social networks, social supports, disability/activities of daily living, quality of life, use of health services, and service use. The questions had been adapted for Kenya from the adult psychiatric morbidity schedule [28] which was previously devised for the UK mental health survey programme. The adult psychiatric morbidity schedule includes assessments of CMD (the Clinical Interview Schedule-revised), psychotic symptoms (the Psychosis Screening Questionnaire), ADHD (WHO Adult ADHD Self-Report Scale Screener), PTSD (the Trauma Screening Questionnaire), suicidality, alcohol use disorders (the Alcohol Use Disorders Identification Test). For this analysis, the data on CMD and psychotic symptoms were used. To assess CMD, scores are calculated from an average of four questions across 14 symptom types.. A score of 12 or more across the 14 sections of the survey is considered an indication of any CMD. The past-year presence of psychotic symptoms is assessed by the PSQ which uses five questions to determine recent experience of mania, thought insertion, paranoia, strange experiences and hallucinations.

Demographic information collected included age, gender, ethnicity, marital status and household status (head, spouse or other). Socio-economic factors assessed included employment status, education attainment, economic assets and type of housing. An index of household assets was constructed as a proxy indicator for socio-economic status, in order to categorise households into different socio-economic levels. In developing the asset index, type of house, roofing & walling material, source of water, toilet facility and land have been used.

Microscopy was used to ascertain the presence of malaria parasites.. Each blood smear slide was dried, packed in the field, and sent for staining and microscopic examination by laboratory technologists trained by the KEMRI/Walter Reed Project's Malaria Diagnostics Centre, Kisumu, Kenya.

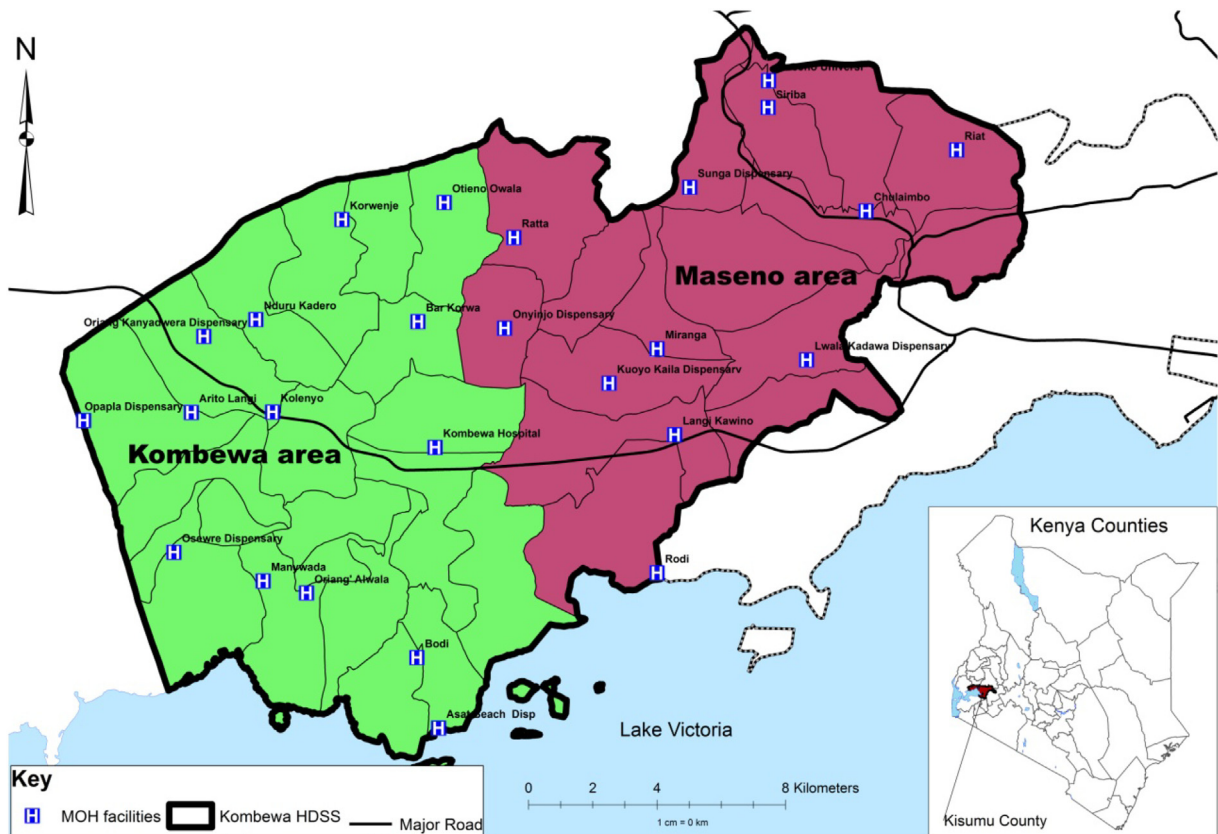


Fig. 1. Location of the study site.

For immunity measurements, about 6 mL of venipuncture blood was aseptically collected into EDTA containing vacutainers (BD Vacutainer®, Plymouth, UK) and transported to the KEMRI laboratories and processed within 2 h of collection. 100  $\mu$ L of blood was aliquoted for CD4 counts while the remaining blood was spun, plasma isolated and snap-frozen at  $-80^{\circ}\text{C}$  until use for circulating cytokine level determination. The counts for peripheral CD4 and CD3 T cells were determined using BD FACScount™ cytometer (BD Biosciences, San Jose, CA, USA) as per the manufacturer's instructions. Briefly, 50  $\mu$ L of blood was added to the reagent tube, vortexed and incubated in the dark at room temperature for 60 min. 50  $\mu$ L of fixative was then added, vortexed and the sample analysed on the BD FACScount™ cytometer to obtain a printout of the absolute CD4 and CD3 T cell values. CD4/CD3 ratios were calculated using the CD4 and CD3 T cell counts.

Sufficient plasma samples for cytokine measurement was obtained from over 700 participants, enabling 742 assays of IL-1 $\beta$ , 737 of IL-6, 739 of IL-8, 742 of IL-10 and 702 of TNF- $\alpha$ . Circulating cytokine levels were determined using standard capture and detection BD OptEIA™ ELISA sets (BD Biosciences Pharmingen, CA, USA; IL-1 $\beta$  - Cat #557953; IL-6 - Cat #555220; IL-8 - Cat #555244; IL-10 - Cat #555157; and TNF- $\alpha$  - Cat #55512) as per the manufacturer's recommendations. Briefly, 96-well Nunc MaxiSorp® ELISA plates (ThermoFisher Scientific, Waltham, MA, USA; Cat #442404) were coated overnight at  $4^{\circ}\text{C}$  with 100  $\mu$ L/well of 1:250 dilution of the capture antibody. The plates were then washed 3 times with wash buffer (PBS + 0.05% Tween 20 (Sigma-Aldrich; Cat #P1379)); PBS prepared using PBS tablets (Sigma-Aldrich Cat #P4417-100Tabs) and blocked for 1 h at room temperature with blocking buffer (PBS + 10% FBS). The plates were then washed 3 times and 100  $\mu$ L of plasma samples per well added in duplicates. Serially diluted standards together with negative controls (Assay diluent - PBS + 10% FBS (Hyclone Cat # SH30088) were also added in duplicates. The plates were then incubated at room temperature for 2 h before being washed 5 times. Biotinylated detection antibody at 1:250 (1:500

for IL-10) and streptavidin-horseradish peroxidase conjugate at 1:250 dilutions in assay diluent were then prepared and added to the plates at 100  $\mu$ L/well and incubated at room temperature for 1 h (IL-6, IL-8, IL-10, and TNF- $\alpha$ ). For IL-1 $\beta$ , the biotinylated antibody (1:500 dilution) was added to the plates and incubated at room temperature for 1 h, then washed 5 times before addition of the enzyme reagent (1:250 dilution) with a further incubation at room temperature for 30 min. After the 1 h (IL-6, IL-8, IL-10, and TNF- $\alpha$ ) and 30 min incubations (IL-1 $\beta$ ), the plates were washed 7 times before 100  $\mu$ L of the substrate solution (Sigma-Aldrich TMB substrate reagent set; Cat #T0440) was added to each well and incubated for 30 min at room temperature in the dark. 50  $\mu$ L of 1 M  $\text{H}_3\text{PO}_4$  (Sigma-Aldrich, Cat #345245) was then added to each well to stop the reaction and absorbance read using a Vmax® microplate reader (Molecular Devices, LLC, CA, USA) at 450 nm within 30 min and cytokine concentrations obtained using the SoftMax® Pro Software (Molecular Devices, LLC, CA, USA).

### 2.1. Statistical analysis

Our prevalence rate of malaria [4], prevalence rates of mental disorders [5–10], the influence of sociodemographic variables, and the association between malaria and mental disorders [11] have been previously reported.

In this paper, levels of peripheral CD4, CD3 T cells, and circulating cytokines are examined, and their variation with the presence of malaria parasites, common mental disorders and psychosis. Multivariate analyses using unadjusted and adjusted odds ratios were conducted for the following dependent variables in turn, namely malaria parasitaemia, CMD and psychotic symptoms. For the adjusted analyses, only variables considered significant at 10% level ( $p \leq 0.1$ ) in the univariate analyses were included.

Thus, we examined mental disorder and immunity risk factors for malaria (unadjusted and adjusted odds ratios using malaria as the

dependent variable); mental disorder and malaria risk factors for CD4 counts (unadjusted and adjusted odds ratios using CD4 counts as the dependent variable); mental disorder and malaria risk factors for non-cellular immunity (unadjusted and adjusted odds ratios using cytokines as the dependent variable); immunity and malaria risk factors for any CMD (unadjusted and adjusted odds ratios using any CMD as the dependent variable); immunity and malaria risk factors for psychotic symptoms (unadjusted and adjusted odds ratios using psychotic symptoms as the dependent variable). The cytokine levels that were undetectable were first assigned a value of 0.01 before being log transformed and analysed as normally distributed variables. Comparison between immunity levels ( $\log_{10}$ ) and malaria or any CMD or any psychotic symptom has been done using *t*-test for equality of means.

The data dictionary and deidentified data will be archived on publication through the World Wide Network for Anti-malarial Resistance (<http://www.wwarn.org/>) on the Infectious Diseases Data Observatory. <https://www.iddo.org/>, and will be accessible on application to Data Access Committee.

## 2.2. Ethics

Ethical approval was granted by the Kings College London and the Kenya Medical Research Institute Boards of Research Ethics respectively (PNM/11/12–54, SSC2374), and permission was obtained to conduct the study in households in Maseno area, which is part of the KEMRI/WRP Kombewa HDSS. Informed written and witnessed consent was asked of heads of sampled households, and then of sampled participants to take part in the study.

## 3. Results

### 3.1. Response rates

1158 participants consented to the study while 32 refused to participate in the study interviews, giving a response rate of 97.2% for the interviews. 149 participants refused to give a blood sample, thus giving an overall response rate of 91.4% for the blood tests. Due to financial constraints, CD4 and CD3 T cell counts were only assayed for the first 470 participants. Sufficient plasma samples to run the 5 cytokines concurrently were obtained from 742 participants, with the remaining samples being insufficient.

### 3.2. Sociodemographic characteristics

The sociodemographic characteristics of the population are given in Table 1. We checked to see whether there were significant differences between the sample with immunological markers and the whole sample, based on demographic characteristics, and found no significant difference (*p* values of 0.373, 0.086, 0.560 and 0.723 for sex, marital status, education and employment respectively. For age there was some significant difference (*p* value 0.028).

### 3.3. Mean levels of immune variables

The mean circulating levels of cytokines and CD4 counts are shown in Table 2. We found marginally raised CD4/CD3 ratios of people with malaria parasites, but no difference in CD4/CD3 ratios for people with CMD or psychotic symptoms. People with one or more psychotic symptoms had increased levels of IL-6, IL-8, and IL-10, and lower levels of IL-1 $\beta$  than non-cases of psychotic symptoms. People with CMD had higher levels of IL-8 and IL-10 than non-cases of CMD. People with malaria parasites had higher levels of IL-10 and lower levels of TNF- $\alpha$  than non-cases of malaria. When we divided people with malaria parasites into high and low parasitaemia using the median > 208 as the cut off, we found higher levels of IL-1beta in people with high parasitaemia, and marginally raised IL-6.

**Table 1**  
Sociodemographic characteristics of the population.

	Mean (SD)	2.5 (2.05)
	Median (IQR)	2 (1 to 3)
Household size (N = 1158)		
Sex of respondent: n (%)	Male	1019 (49.4)
	Female	1043 (50.6)
Age of respondent (N = 1589)	Mean (SD)	40.3 (17.7)
	Median (range)	36 (25 to 53)
Marital status: n (%)	Single	370 (18.1)
	Married and living together	1313 (64.1)
	Married and separated	52 (2.5)
	Divorced	23 (1.1)
	Widowed	281 (13.7)
	Living with partner, not married	11 (0.5)
Highest Education level: n (%)	None	191 (9.3)
	Primary [1–8]	1101 (53.4)
	Secondary [1–4]	634 (30.8)
	College/university	118 (5.7)
	Other	18 (0.9)
Employment status: n (%)	Employed full time - salaried	91 (4.4)
	Employed full time - self employed	516 (25.0)
	Employed part time-salaried	97 (4.7)
	Employed part time- self employed	293 (14.2)
	Unemployed but looking for work	190 (9.2)
	Not working and not looking for work	426 (20.7)
	Disabled	24 (1.2)
	Housewife	353 (17.1)
	Student	72 (3.5)

SD = Standard Deviation; IQR = Inter-quartile Range.

### 3.4. Risk factors for malaria

Table 3 shows risk factors for malaria using bivariate analysis. As previously reported elsewhere, females had a higher risk of having malaria compared to males [2] while having any CMD was related to an increase in the risk of having malaria [3], but in this further analysis there was no significant relationship between the immune variables and the presence of malaria. Therefore, in the adjusted analysis as well, there was no effect of immune markers, and the risk of malaria remained 40% higher in females compared to males and 60% higher in those with any CMD.

### 3.5. Risk factors for common mental disorder

Tables 4 and 5 show the risk factors for common mental disorders, using bivariate and multivariate analyses respectively. Table 4 shows that the risk of having any CMD was significantly higher in females compared to males (odds ratio = 5.4), and in those who tested positive for malaria, where the risk of having any CMD was 70% higher than those with no malaria (*p* = .014), and was also related to levels of TNF- $\alpha$ . In the adjusted analysis, the relationship of CMD with TNF- $\alpha$  was no longer significant, after adjusting for presence of malaria, and IL-6.

### 3.6. Risk factors for psychotic symptoms

Tables 6 and 7 show the risk factors for the presence of psychotic symptoms using bivariate and multivariate analyses respectively. At the bivariate level, psychotic symptoms were associated with log IL-10 (OR 3.2, *p* = .029) and log TNF- $\alpha$  (OR 3.7, *p* = .001), levels using unadjusted odds ratios; and in the adjusted analysis, log IL-10 levels was only significant at the 10% level (OR 1.1 *p* = .107) while log TNF- $\alpha$  levels remains highly significant (OR 1.3, *p* = 0.003), after adjusting for IL-10 and TNF- $\alpha$ .

**Table 2**  
Immunity in relation to malaria parasitaemia, common mental disorders, and psychotic symptoms.

Immune variable		Overall	Malaria		Any CMD		Psychosis		High parasitemia (median > 208)	
			Negative	Positive	No	Yes	No	Yes	Low	High
logCD4CD3 ratio, n = 217	Mean (SD)	0.18 (0.04)	0.18 (0.04)	0.19 (0.04)	0.18 (0.04)	0.20 (0.02)	0.18 (0.04)	0.19 (0.04)	0.19 (0.04)	0.19 (0.02)
	Median (IQR)	0.19 (0.17 to 0.21)	0.19 (0.16 to 0.21)	0.20 (0.19 to 0.21)	0.19 (0.17 to 0.21)	0.20 (0.19 to 0.22)	0.19 (0.16 to 0.21)	0.19 (0.18 to 0.21)	0.21 (0.19 to 0.21)	0.19 (0.17 to 0.20)
	p-value**		0.079		0.316		0.189		0.184	
logIL-1beta, n = 742	Mean (SD)	0.96 (0.61)	1.08 (0.68)	0.88 (0.55)	0.99 (0.63)	0.94 (0.60)	1.03 (0.63)	0.60 (0.30)	0.63 (0.26)	1.04 (0.63)
	Median (IQR)	0.75 (0.53 to 1.16)	0.81 (0.56 to 1.04)	0.73 (0.52 to 1.12)	0.76 (0.54 to 1.26)	0.66 (0.53 to 1.12)	0.81 (0.54 to 1.30)	0.66 (0.36 to 0.76)	0.64 (0.48 to 0.73)	0.88 (0.68 to 1.30)
	p-value**		0.311		0.675		0.048		0.012	
logIL-6, n = 737	Mean (SD)	0.80 (0.52)	0.78 (0.53)	0.77 (0.40)	0.78 (0.51)	0.91 (0.55)	0.77 (0.52)	0.91 (0.54)	0.84 (0.41)	0.71 (0.39)
	Median (IQR)	0.71 (0.40 to 1.09)	0.67 (0.38 to 1.08)	0.73 (0.46 to 1.01)	0.68 (0.40 to 1.08)	0.87 (0.40 to 1.20)	0.67 (0.38 to 1.08)	0.82 (0.56 to 1.20)	0.90 (0.46 to 1.09)	0.63 (0.47 to 0.92)
	p-value**		0.474		0.136		0.066		0.065	
logIL-8, n = 739	Mean (SD)	0.93 (0.41)	0.93 (0.40)	0.89 (0.38)	0.92 (0.38)	1.02 (0.53)	0.92 (0.39)	1.01 (0.46)	0.87 (0.34)	0.89 (0.42)
	Median (IQR)	0.89 (0.67 to 1.10)	0.90 (0.69 to 1.10)	0.85 (0.61 to 1.10)	0.88 (0.69 to 1.09)	0.90 (0.63 to 1.25)	0.88 (0.67 to 1.10)	0.91 (0.72 to 1.25)	0.85 (0.58 to 1.10)	0.85 (0.62 to 1.08)
	p-value**		0.187		0.407		0.210		0.985	
logIL-10, n = 742	Mean (SD)	1.08 (0.52)	1.05 (0.53)	1.12 (0.44)	1.05 (0.50)	1.33 (0.64)	1.03 (0.51)	1.33 (0.51)	1.07 (0.39)	1.17 (0.48)
	Median (IQR)	0.99 (0.76 to 1.32)	0.97 (0.71 to 1.30)	1.09 (0.87 to 1.33)	0.97 (0.74 to 1.30)	1.36 (0.95 to 2.00)	0.95 (0.71 to 1.30)	1.22 (0.97 to 1.61)	1.02 (0.87 to 1.25)	1.14 (0.86 to 1.47)
	p-value**		0.097		0.008		0.001		0.308	
logTNF-alpha, n = 702	Mean (SD)	1.24 (0.63)	1.38 (0.70)	0.99 (0.36)	1.20 (0.61)	1.49 (0.72)	1.23 (0.63)	1.25 (0.64)	0.95 (0.25)	1.05 (0.48)
	Median (IQR)	1.00 (0.81 to 1.71)	1.16 (0.79 to 2.04)	0.95 (0.78 to 1.09)	0.99 (0.79 to 1.37)	1.31 (0.85 to 2.16)	0.99 (0.81 to 1.93)	0.99 (0.84 to 1.55)	0.96 (0.81 to 1.08)	0.92 (0.78 to 1.12)
	p-value**		0.035		0.251		0.715		0.979	
			0.002		0.101		0.879		0.398	

\*p-value from t-test for equality-of-means: \*\*p-value from Kruskal Wallis test for equality of population.

**4. Discussion**

This survey of malaria, mental disorders and immune markers in a representative sample of adults in a health and demographic surveillance site in rural western Kenya has demonstrated specific associations between them. We have separately reported the association of malaria and mental disorders, finding an association between malaria and CMD in the general adult population, but not between malaria and psychotic symptoms, the latter probably only being evident in acutely hospitalised populations [11]; and we now report the associations of malaria, CMD and psychotic symptoms with immune markers in a general adult population sample. To summarise, we found marginally raised CD4/CD3 ratios of people with malaria parasites, but no difference in CD4/CD3 ratios for people with CMD or psychotic symptoms. People with psychotic symptoms had increased levels of IL-6, IL-8, and IL-10, and lower levels of IL-1 $\beta$ . People with CMD had higher levels of IL-8 and IL-10 while those with malaria had higher levels of IL-10 and lower levels of TNF- $\alpha$ . At the bivariate level, CMD was associated with log TNF- $\alpha$  levels using unadjusted odds ratios, but not after adjusting for malaria. Psychotic symptoms were associated with log IL-10 and log TNF- $\alpha$  levels at the bivariate level while in the adjusted analysis, log TNF- $\alpha$  levels remained highly significant.

*4.1. Association of malaria and mental disorder*

To our knowledge, there have been no previous studies of malaria parasitaemia and CMD measured by standardised clinical interview. This association does not arise from shared method variance via measurement of symptoms of malaise and fatigue, because although CMD caseness was ascertained by presence of 14 different psychological symptoms including fatigue and excessive concern about bodily symptoms, malaria parasitaemia was ascertained by presence of actual malaria parasites rather than of symptoms per se. The lack of association between malaria and other mental disorders indicates that the association is specific to CMD, and has implications for the clinical assessments and management plans of health care workers. The fact that we did not find an association between malaria and psychotic symptoms is interesting but not surprising, since our sample were all ambulant adults living at home, so none would have had cerebral malaria which often presents with psychotic symptoms, as cerebral malaria necessitates urgent hospital admission.

As this is a cross sectional study, we are unable to attribute directional causality, but it is possible that both causal directions are relevant. It is possible that people with malaria have increased susceptibility to depression. Malaria as a debilitating physical illness may predispose to depression,

**Table 3**  
Risk of malaria by immunity, common mental disorders and psychotic symptoms (unadjusted).

Factors	N	Prevalence: n (%)	Unadjusted OR (95% C-I)	p-value
Malaria	1002	282 (28.1)		
Psychotic symptoms	940		1.1 (0.74–1.67)	0.601
Any CMD				
No	871	234 (26.9)	1	–
Yes	95	37 (39.0)	1.7 (1.12 to 2.69)	0.014
CD4/CD3 ratio	205		9.3 (0.23 to 376)	0.236
Immunity: CD4 counts				
≥500 cells/μL	377	42 (11.1)	1	–
<500 cells/μL	58	7 (12.2)	1.1 (0.47 to 2.57)	0.835
log <sub>10</sub> IL-6				
Low	332	102 (30.7)	1	0.216
High	18	3 (16.7)	0.5 (0.13 to 1.59)	
log <sub>10</sub> IL-8				
Low	235	64 (27.2)	1	0.830
High	318	84 (26.4)	1.0 (0.66 to 1.40)	
log <sub>10</sub> IL-10				
Low	309	86 (27.8)	1	0.450
High	20	4 (20.0)	0.6 (0.21 to 1.99)	
log <sub>10</sub> IL-beta				
Low	88	47 (53.4)	1	0.177
High	17	6 (35.3)	0.5 (0.16 to 1.40)	
log <sub>10</sub> TNF-alpha				
Low	109	44 (40.4)	1	0.097
High	13	2 (15.4)	0.3 (0.06 to 1.27)	

OR = odds ratio; C-I = confidence interval.

and pro-inflammatory cytokines promote tryptophan depletion which is associated with depressive symptoms. On the other hand, depression, by altering immunity and by altering behaviour, may predispose to malaria. Depression may hinder recovery from malaria, and vice versa. There is evidence that people with depression have worse outcomes from many communicable diseases such as HIV as well as from non-communicable diseases [29], but this has not yet been studied in people with malaria.

This association between malaria and common mental disorder has implications for clinical settings where accurate assessment and effective management are needed. In addition to the possible associations referred to above, clinicians are known to frequently misdiagnose complaints of fatigue and general malaise as malaria when in fact the person has no parasitaemia [30–34], but does have depression [35]. Such misdiagnosis may lead to erroneous prescriptions of antimalarials [36] which may clear protective low grade parasitaemia, thus making the person more vulnerable to subsequent malaria. Furthermore, clinicians frequently miss depression, thus leaving the depressed and untreated individual in a state of lowered immunity, which may predispose to malaria or, in those already with malaria, may worsen prognosis.

#### 4.2. Association of immunity and malaria

The changes in the immune response to *P.falciparum* are very complex [19]; Type 1 cytokines are important in controlling early parasitaemia but are counterbalanced later in the infection by a Type 2

**Table 4**  
Risk of CMD by malaria, cellular and non-cellular immunity (unadjusted).

Factors	N	Unadjusted OR (95% C-I)	p-value
Any CMD	1157		
Malaria			
No	695	1	0.014
Yes	271	1.7 (1.12 to 2.69)	
CD4/CD3 ratio	217	1.0 (0.99 to 1.00)	0.172
log <sub>10</sub> IL-1beta	742	2.8 (2.24 to 3.58)	0.905
log <sub>10</sub> IL-6	737	3.1 (2.65 to 3.84)	0.096
log <sub>10</sub> IL-8	739	2.9 (2.46 to 3.65)	0.402
log <sub>10</sub> IL-10	742	2.7 (2.34 to 3.17)	0.914
log <sub>10</sub> TNF-alpha	702	3.4 (2.77 to 4.26)	0.032

OR = odds ratio; C-I = confidence interval.

**Table 5**  
Risk of CMD by malaria, cellular and non-cellular immunity(adjusted).

Factors	Adjusted OR (95% C-I)	p-value
Malaria	1.3 (0.76 to 2.15)	0.346
log <sub>10</sub> IL-6	3.1 (2.58 to 3.94)	0.159
log <sub>10</sub> TNF-alpha	2.9 (2.36 to 3.77)	0.566

OR = odds ratio; C-I = confidence interval.

response which leads to antibody synthesis [37]. An early reaction of the host to infection with protozoan parasites is the secretion of TNF, interleukin-1 and IL-6, causing fever, leucocytosis and plasma proteins such as the C-protein. These early responses influence the course of Infection [38].

Most previous cytokine studies in malaria are on clinical samples rather than on general population groups [39]. Furthermore, Kisumu is a high transmission area, where malaria is endemic. In high transmission areas, more frequent infections mean an almost chronic state of infection [40].

Our finding that people in the community with malaria parasitaemia had significantly higher mean scores of log IL-10, and lower mean scores of log TNF-α may be compared with that of Berg et al. 2014 [41]) who also found that log TNF-α was low in those patients co-morbid with HIV, but that otherwise malaria parasitaemia was associated with increased levels of log IL-8 and log IL-10 in hospitalised patients with falciparum malaria in Maputo. We did not measure the presence of HIV, but HIV infection rates are high in this part of Kenya compared to the national average. TNF-α is thought to be central to the immune response to falciparum infection because of its pyrogenic properties and its key role in triggering the cascade of pro-inflammatory cytokines that regulate immune cells. Genetic studies have suggested that TNF-α gene variants are associated with susceptibility to malaria [42]. Low levels of TNF-α are necessary for enhancing phagocytic activity and controlling parasite densities but high levels can trigger severe malaria, anaemia, fever and cerebral malaria [43]. Our finding that IL-1beta and IL-6 were associated with higher levels of parasitaemia may be compared with that of Mbengue et al. 2015 [44] who demonstrated high levels of TNFα and IL6 (proinflammatory cytokines) are indicators of malaria severity while antiinflammatory IL-10 does not differentiate mild and severe cases.

We did not find a bivariate association between community malaria parasitaemia and IL-10, but Ong'echa et al. [45] reported that IL-10 was a predictor of parasitaemia in children presenting with falciparum malaria to a hospital in western Kenya. The levels of IL-6 and IL-10 were lower in children with malaria with concurrent anaemia in Mali while severe malaria, cerebral malaria, was associated with raised IL-6 and IL-10<sup>17</sup>. Similar findings have been reported from Gabon [46] and Ghana<sup>39</sup>. It should be noted that in these studies of the immune changes in malaria, none of the authors mentioned the possible changes in psychopathology of the patients.

**Table 6**  
Risk of psychotic symptoms by malaria, cellular (CD4/CD3 ratio) and non-cellular immunity (cytokines) (unadjusted).

Factors	N	Unadjusted OR (95% C-I)	p-value
Psychotic symptoms	1121		
Malaria			
No	678	1	0.601
Yes	262	1.1 (0.74 to 1.67)	
CD4/CD3 ratio	217	1.0 (0.99 to 1.01)	0.562
log <sub>10</sub> IL-1beta	742	2.4 (1.97 to 3.03)	0.252
log <sub>10</sub> IL-6	737	3.0 (2.56 to 3.54)	0.257
log <sub>10</sub> IL-8	739	2.6 (2.28 to 3.10)	0.659
log <sub>10</sub> IL-10	742	3.2 (2.76 to 3.78)	0.029
log <sub>10</sub> TNF-alpha	702	3.7 (3.05 to 4.65)	0.001

OR = odds ratio; C-I = confidence interval.

**Table 7**  
Risk of psychotic symptoms by malaria, cellular and non-cellular immunity (cytokines) (adjusted).

Factors	Adjusted OR (95% C-I)	p-value
log <sub>10</sub> IL-10	1.1 (0.98 to 1.29)	0.107
log <sub>10</sub> TNF-alpha	1.3 (1.09 to 1.51)	0.003

OR = odds ratio; C-I = confidence interval.

#### 4.3. Association of immunity and mental disorders

We have been unable to find any studies of cytokines and CMD, rather than of either specific categories of depression or anxiety, although CMD is a more representative and less artificial diagnostic category in community populations (and indeed primary care clients) where symptoms of depression and anxiety usually co-exist. Indeed a recent review found that the most recent biological findings points towards a valid comorbid entity of mixed anxiety depression [47]. We found a bivariate association between CMD cases and TNF- $\alpha$  at the bivariate level, and this association between depressed mood and TNF- $\alpha$  has been found elsewhere [48] TNF- $\alpha$  may underlie the mechanism of depression by activation of the hypothalamo-pituitary-adrenocortical axis, activation of neuronal serotonin transporters and the stimulation of the indoleamine 2,3-dioxygenase which leads to tryptophan depletion [49]. We also found that IL-8 and IL-10 levels were elevated in CMD in this community population, whereas a recent meta-analysis of IL-6 and IL-10 between people with and without depression found that IL-6 was elevated in people with depression, and that the effect size was greatest for inpatients and outpatients than for depressed people in the general community, while IL-10 effect size was not significant [50].

We found increased levels of IL-6, IL-8 and IL-10 and lower levels of IL-1 $\beta$  in people with one or more psychotic symptoms, together with significant associations between people with one or more psychotic symptoms and IL-10 and TNF-alpha at the bivariate level remaining highly significant for TNF-alpha in the adjusted analysis.

Previous studies of cytokines and psychosis have generally been conducted on patients with diagnosed schizophrenia, and we have not found any previous studies of the relationships between cytokines and people with psychotic symptoms as measured by the PSQ. Maes et al. 2002 [51] and Kunz et al. 2011 [52] reported that patients with schizophrenia showed significantly increased IL-10 serum levels. A meta-analysis found that blood IL-10 levels were significantly decreased and IL-6, IL-8, TNF- $\alpha$ , IFN- $\gamma$ , TGF- $\beta$  and IL-1RA levels were significantly increased in acutely relapsed inpatients compared with control subjects, and the most replicated finding was for TNF- $\alpha$  [53]. (Manu et al. [54] have argued for the need to follow patients from the attenuated psychosis syndrome to full blown schizophrenia, and to measure cytokine levels in those who convert to psychosis and those who do not.

#### 4.4. Association of malaria, immunity and common mental disorders

Our multivariate analysis found that the relationship of CMD with TNF a was no longer significant after adjusting for the presence of malaria and for IL-6, suggesting that TNF a may be a mediating variable in the association of malaria and CMD. *Such mediation may arise because as described above, TNF a may underly the mechanism of depression, and TNF a is associated with the pathogenesis of malaria. Further longitudinal research is needed to elucidate the directions of causality, and the most appropriate management strategy.*

#### 4.5. Strengths and limitations

The key strength of this study is the use of a large representative sample of adults in a health and demographic surveillance site, with a

high response rate, which contributes to the external validity of the study and generalisability of the findings to similar populations in other areas endemic for malaria. This was a cross sectional rather than longitudinal study, and so conclusions on causal relationships cannot be drawn.

Other limitations included the practical difficulties of collecting blood samples in the field, and getting them safely to the laboratory in difficult terrain and climatic conditions, contributing to the reduced sample size for immune variables. However we checked for potential bias and found no significant difference across sex, marital status, education and employment. We did find a significant difference for age but do not consider this would materially change our conclusions. This analysis used multiple comparisons and so there is a risk of potential chance findings, but again our relatively large sample size minimises this risk. The study used microscopy to assess parasitaemia but not PCR which would have been more sensitive. No assessment was made of the presence of HIV/AIDS in the study population, which is also associated with neuroinflammation and mental ill health, although all the participants had good health based on their CD4 counts. We also did not check for anaemia. As this is the first study of its kind, it was important to choose immune parameters which have been widely shown to be elevated in depression, anxiety and comorbid conditions in other settings. For this reason pro-inflammatory markers were selected. Future studies should consider more detailed examination of cellular and non-cellular markers of the immune response.

#### 5. Conclusions

This is the first population based study of immune markers in CMD and psychotic symptoms, and the first to examine the 3 way relationship with malaria. Our findings suggest that TNF-alpha may mediate the previously reported relationship between malaria and CMD (9). Our findings support the key role of TNF- $\alpha$  in the pathogenesis of both CMD and psychotic symptoms, and suggest that TNF- $\alpha$  may mediate the association between malaria and CMD. Further community studies should be longitudinal, use PCR, measure anaemia and extend the range of immune markers used.

Immunity is an important risk factor for both malaria and mental disorders in this adult household population in rural Kenya, and this information is relevant for public health measures to strengthen immunity, prevent malaria and comorbid mental disorder, for training and supervision of health workers in multiaxial assessment, diagnosis and management, and for health management information systems to be able to record multiaxial diagnoses and management plans, so that the health system is equipped to routinely address mental and physical comorbidity.

Finally, the study also indicates that the PSQ is measuring a biologically identifiable group or more likely a heterogeneous combination of subgroups with excessive inflammation affecting the brain. This finding adds to the known validity of the screening instrument in large scale community studies of psychosis.

#### Funding sources

The study was funded by UK Aid, Department for International Development, Kenya office. We are also grateful to the Nuffield Foundation for a timely travel grant (Grant 41346) to enable LO to contribute to local field supervision. Neither DFID nor Nuffield Foundation had any role in the study design, data collection, data analysis, interpretation or writing of the report.

#### Declaration of interests

Dr. Jenkins reports grants from Department of International Development during the conduct of the study. The remaining authors have no conflicts of interest to declare.

## Authors' contributions

R. J. conceived the study and had overall responsibility for the project; R. J., B. O., and C. O. designed the overall study while BL and MO advised on immune markers; P. S. drew the sample within Kombewa DHSS; B. O., C. O. and latterly L. O. provided local field supervision; R. O. analysed the data, R. J. wrote the first draft of the paper, all authors commented on successive drafts, interpretation of results and approved the final version.

## Acknowledgements

The authors are grateful to David Kiima, Ministry of Health, Kenya, for initial input into the conception of the study; the late Howard Meltzer, University of Leicester for advice on the survey sampling and interviews; Stuart Lancashire of Kings College London for liaison support between KCL and DFID; James Mboroki Kingora, project manager at Kenya Medical Training College, to the research assistants who conducted the clinical interviews and took the blood samples, to Kombewa Health and Demographic Surveillance Systems (Kombewa HDSS) site which hosted the survey, and last but not least, to the community of Kisumu West.

## References

- Atun R. Transitioning health systems for multimorbidity. *Lancet* 2015;386:721–2.
- Evans D, Charney DS, Lewis L, et al. Mood disorders in the medically ill: scientific review and recommendations. *Biol Psychiatry* 2005;58:175–89.
- Rabkin RG. HIV and Depression. 2008 review and update. *Curr HIV/AIDS Rep* 2008; 5:163–71.
- Jenkins R, Omollo R, Ongecha M, et al. Prevalence of malaria parasites in adults and its determinants in malaria endemic area of Kisumu County. *Kenya Malaria J* 2015; 14:263.
- Jenkins R, Othieno C, Ongeri L, et al. Common mental disorder in Nyanza province, Kenya in 2013 and its associated risk factors, including gender – an assessment of change since 2004, using a repeat household survey in a demographic surveillance site. *BMC Psychiatry* 2015;15(1):309.
- Jenkins R, Othieno C, Ongeri L, et al. Adult Psychotic symptoms , their associated risk factors, and changes in prevalence in men and women over a decade in a poor rural district of Kenya. *Int J Environ Res Public Health* 2015;12:5310–28.
- Jenkins R, Othieno C, Omollo R, et al. Probable post traumatic stress disorder in Kenya and its associated risk factors—a cross sectional household survey. *Int J Environ Res Public Health* 2015;12:13494–509.
- Jenkins R, Othieno C, Ongeri L, et al. Attention deficit hyperactivity disorder symptom self-report in adults in Kenya and its associated risk factors, an analysis from a household survey in a demographic surveillance site. *Global Mental Health* 2015;2(e14):1–11.
- Jenkins R, Othieno C, Omollo R, et al. Tedium vitae, death wishes, suicidal ideation and attempts in Kenya—prevalence and risk factors. *BMC Public Health* 2015;15:759.
- Jenkins R, Othieno C, Ongeri L, et al. Alcohol consumption and hazardous drinking in western Kenya –a household survey in a health and demographic surveillance site. *BMC Psychiatr* 2015;15:230.
- Jenkins R, Othieno C, Ongeri L, Ongecha M, Sifuna P, Omollo R, et al. Malaria and mental disorder: a population study in an area endemic for malaria in Kenya. *World Psychiatry* 2017;16(3):324–5.
- Iwata M, Ota KT, Duman RS. The inflammasome: pathways linking psychological stress, depression and systemic illnesses. *Brain Behav Immun* 2013;31:105–14.
- Smith RS, Maes M. The macrophage-T lymphocyte theory of schizophrenia: additional evidence. *Med Hypoth* 1995;45:135–41.
- Kirpatrick B, Miller BJ. Inflammation and schizophrenia. *Schizophren Bull* 2013;39: 1174–9.
- Leonard BE. Inflammation and depression: a causal or coincidental link to the pathophysiology. *Acta Neuropsychiat* 2017. <https://doi.org/10.1017/neu.2016.69>.
- Dodoo D, Omer FM, Todd J, et al. Role of proinflammatory and anti-inflammatory cytokines in the immune response to *Plasmodium falciparum* malaria. *J Infect Dis*; 185: 971–979.
- Lyke K, Burges R, Cissoko Y, et al. Serum levels of proinflammatory cytokines Interleukin-1 beta,IL-1L-8IL-10,TNF-alpha and IL-12 in Malian children with severe *Plasmodium falciparum* malaria and unmatched uncomplicated malaria or health controls. *Infect Immun* 2004;72:5630–7.
- Angulo I, Fresno M. Cytokines in the pathogenesis of and protection against malaria. *Clin Vaccine Immunol* 2002;9:1145–52.
- Jason J, Archibald LK , Nwanyanwu OC, et al . Cytokines and malaria parasitaemia , *Clin Immunol* 2001; 100: 208–218.
- Miller AH, Maletic V, Raison CL. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. *Biol Psychiatry* 2009;65(9): 732–41.
- Raison CL, Capuron L, Miller AH. Cytokines sing the blues: inflammation and the pathogenesis of depression. *Trends Immunol* 2006;27(1):24–31.
- Dowlati Y, Herrmann N, Swardfager W, et al. A meta-analysis of cytokines in major depression. *Biol Psychiatry* 2010;67(5):446–57.
- Fineberg A, Ellman L. Inflammatory cytokines and neurological and neurocognitive alterations in the course of schizophrenia. *Biol Psychiatry* 2013;73. <https://doi.org/10.1016/j.biopsych.2013.01.001>.
- Bluthe RM, Pawlowski M, Suarez S, et al. Synergy between Tumour necrosis factor alpha and interleukin-1 in the induction of sickness behaviour in mice. *Psychoneuroendocrinology* 1994;19:197–207.
- Anisman H, Hayley S. Illness comorbidity as a biomarker? *J Psych Neurosci* 2012;37 (4):221–3.
- Norcini Pala. A HIV, depression and compliance: the mediating role of cytokines. A review of the international literature. *Riv Psichiatr* 2013;48(6):432–40.
- Sifuna P, Oyugi M, Ogutu B, et al. Health & demographic surveillance system profile: the Kombewa Health and Demographic Surveillance System (Kombewa HDSS). *Int J Epidemiol* 2014. <https://doi.org/10.1093/ije/dyu139>.
- McManus S, Meltzer T, Bebbington P, Jenkins R. Adult Psychiatric Morbidity in England 2007. Results of a Household Survey; 2009 Available online <http://www.hscic.gov.uk/pubs/psychiatricmorbidity07>, Accessed date: 12 May 2018.
- Prince M, Patel V, Saxena S, Maj M, Maselko J, Phillips MR, et al. No health without mental health. *Lancet* 2007;370:859–77.
- Afrane YA, Zhou G, Yan G, Githeko AK, Yan G. Utility of health facility-based malaria data for malaria surveillance. *Plos One* 2013. <https://doi.org/10.1371/journal.pone.0054305>.
- Font F, Alonso González M, Nathan R, et al Diagnostic accuracy and case management of clinical malaria in the primary health services of a rural area in South-Eastern Tanzania. *Trop Med Int Health* 2001;6(6):423–8.
- Nankabirwa J, Zurovac D, Njogu JN, et al. Malaria misdiagnosis in Uganda – implications for policy change. *Malar J* 2009;8:66. <https://doi.org/10.1186/1475-2875-8-66>.
- Rowe AK, Rowe SY, Snow RW, et al. The burden of malaria mortality among African children in the year 2000. *Int J Epidemiol* 2000;35(3):691–704.
- Ye Y, Madise N, Ndugwa R, et al. Fever treatment in the absence of malaria transmission in an urban informal settlement in Nairobi. *Kenya Malaria Journal* 2009;8:160. <https://doi.org/10.1186/1475-2875-8-160>.
- Kiima D. Prevalence of mental disorders in Kariobangi clinic. MMed thesis University of Nairobi; 1987.
- Zurovac D, Njogu J, Akhwale W, et al. 2008 Effects of revised diagnostic recommendations on malaria treatment practices across age groups in Kenya. *Trop Med Int Health* 2008;13:784–7.
- Langhorne J, Ndungu FM, Sponaas A-M, Marsh K. Immunity to malaria: more questions than answers. *Nat Immunol* 2008;9(7):725–32. <https://doi.org/10.1038/nif.2051>.
- Titus RG, Sherry B, Cerami A. The involvement of TNF, IL-1 and IL-6 in the immune response to protozoan parasites. *Trends Immunol* 1991;12:13–6.
- Ademolue T, Aniwel Y, Asamoah K, et al. Patterns of inflammatory responses and parasite tolerance vary with malaria transmission intensity. *Malar J* 2017;20(17): 16. <https://doi.org/10.1186/s12936-017-1796-x>.
- Galatas B, Bassat Q, Mayor A. Malaria parasites in the asymptomatic: looking for the hay in the haystack. *Trends Parasitol* 2016;32:296–308.
- Berg A, Patel S, Gonca M, et al. Cytokine network in adults with falciparum malaria and HIV-1. Increased IL-8 and IL-10 levels are associated with disease severity. *PLoS ONE* 2014;9(12):e114480.
- Wanjiku NG, Melsa-Boonstra A, Feskens EJ, Demir AY, Veenemans J. Tumour necrosis factor allele variants and their association with the occurrence and severity of malaria in African children in a longitudinal study. *Malar J* 2015;14:249.
- Nasr A, Allam G, Hamid O, Al-Ghamid A. IFN-gamma and TNF associated with severe falciparum malaria infection in Saudi pregnant women. *Malar J* 2014;13:314.
- Mbengue B, Niang B, Niang MS, et al. Inflammatory cytokine and humoral responses to *Plasmodium falciparum* glycosylphosphatidylinositols correlates with malaria immunity and pathogenesis. *Immunity Inflamm Dis* 2016;4(1):24–34.
- Ong'echa JM, Remo AM, Kristoff J, et al. Increased circulating interleukin (IL)-23 in children with malarial anemia: in vivo and in vitro relationship with co-regulatory cytokines IL-12 and IL-10. *Clin Immunol* 2008;126(2):211–21.
- Oyegue-Liabgu SL, Bouopda-Tuedom AG, Kouna LC, et al. Pro and anti-inflammatory cytokines in children with malaria in Franceville. *Gabon Am J Clin Immunol* 2017;6: 9–20.
- Andreescu C, Lenze E. Comorbid anxiety and depression: bete noire of quick fix. *Br J Psychiatry* 2000;2012:179–81.
- Zunsain PA, Heppgul N, Pariante CM. Inflammation and depression. *Curr Top Behav Neurosci* 2013;14:135–51. [https://doi.org/10.1007/7854\\_2012\\_211](https://doi.org/10.1007/7854_2012_211).
- Krishnadas R, Cavanagh J. Depression: an inflammatory illness? *J Neurol Neurosurg Psychiatry* 2012;83:495–502. <https://doi.org/10.1136/jnnp-2011-301779>.
- Hiles SA, Baker AL, De Malmanche T, Attia J. A meta analysis of differences in IL-6 and IL-10 between people with and without depression: Exploring the causes of heterogeneity. *Brain Behav Immun* 2012;26:1180–8.
- Maes M. The depressogenic effects of cytokines: implications for the psychological and organic aetiology and treatment of depression. *Int J Neuropsychopharmacol* 2002;5(4):329–31.
- Kunz M, Ceresér KM, Goi PD, et al. Serum levels of IL-6, IL-10 and TNF- $\alpha$  in patients with bipolar disorder and schizophrenia: differences in pro- and anti-inflammatory balance. *Rev Bras Psiquiatr* 2011;33(3):268–74.
- Miller B, Buckley P, Wesley S, Melor A, Kirkpatrick B. Meta –analysis of cytokine alterations in schizophrenia: Clinical Status and antipsychotic effects. *Biol Psychiatry* 2011;70:663–71.
- Manu P, Correll C, Wampera M, et al. Markers of inflammation in schizophrenia: association vs causation. *World Psychiatry* 2014;13:189–92.