Severity and mortality of severe *Plasmodium ovale* infection: A systematic review and meta-analysis

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Abstract

*Plasmodium ovale* can infect humans, causing malaria disease. We aimed to investigate the severity and mortality of severe *P. ovale* infection to increase the awareness of physicians regarding the prognosis of this severe disease and outcome-related deaths in countries in which this disease is endemic. Articles that were published in the PubMed, Scopus, and ISI Web of Science databases prior to January 5, 2020 and reported the prevalence of severe *P. ovale* infection were systematically searched and reviewed. Studies that mainly reported severe *P. ovale* infection according to the 2014 WHO criteria for the treatment of malaria were included. Two reviewers selected, identified, assessed, and extracted data from studies independently. The pooled prevalence of severe *P. ovale* mono-infections was estimated using the command “metaprop case population, random/fixed”, which yielded the pooled estimate, 95% confidence interval (CI) and the I² value, indicating the level of heterogeneity. Meta-analyses of the proportions were performed using a random-effects model to explore the different proportions of severity between patients with *P. ovale* and those with other *Plasmodium* species infections. Among the eight studies that were included and had a total of 1,365 ovale malaria cases, the pooled prevalence of severe *P. ovale* was 0.03 (95% CI = 0.03–0.05%, I² = 54.4%). Jaundice (1.1%), severe anemia (0.88%), and pulmonary impairments (0.59%) were the most common severe complications found in patients infected with *P. ovale*. The meta-analysis demonstrated that a smaller proportion of patients with *P. ovale* than of patients with *P. falciparum* had severe infections (P-value = 0.01, OR = 0.36, 95% CI = 0.16–0.81, I² = 72%). The mortality rate of severe *P. ovale* infections was 0.15% (2/1,365 cases). Although severe complications of *P. ovale* infections in patients are rare, it is very important to increase the awareness of physicians regarding the prognosis of severe *P. ovale* infections in patients, especially in a high-risk population.
Introduction

Severe malaria results in the dysfunction of one or more vital organs [1]. *Plasmodium ovale*, which causes tertian malaria, was first reported in 1922 as one of the five *Plasmodium* species that can infect humans [2]. *P. ovale* accounts for between 0.5 and 10.5% of all malaria cases, and it is geographically distributed in sub-Saharan Africa, the Western Pacific, Timor, and Indonesia [3]. The highest prevalence of *P. ovale* has been reported in Papua New Guinea (15%) [4] and Nigeria (15%) [5]. However, the most recent retrospective cohort study conducted in Papua, Indonesia during 2004–2013 demonstrated a low prevalence of *P. ovale* infections (0.06%) among 68,361 patients [6]. Other recent studies demonstrated that *P. ovale* infections accounted for 2.5% of malaria cases in Uganda [7] and 2.7% of malaria cases in China [8]. Infections due to *P. ovale* have been underestimated compared with those of other *Plasmodium* species, as *P. ovale* has been demonstrated to lead to low parasitemia and have morphologic similarities with *P. vivax* and mixed infections [9, 10]. Similar to *P. vivax*, *P. ovale* can cause relapsing infection due to the presence of latent parasites (hypnozoites) in the liver long after the first treatment is administered with anti-malarial drugs [3]. *P. ovale* is detected and identified with the standard microscopic method. Rapid diagnostic tests (RDTs) can also be used in cases where microscopic detection cannot be performed, such as in rural or remote areas. However, the sensitivity of RDTs to detect *P. ovale* is low (22.2%) [11]. The low sensitivity of RDTs can be attributed to the low parasitemia level [12, 13] or different targeted antigens [14]. Recently, a molecular method with nested polymerase chain reaction (nested PCR) has been used to identify *Plasmodium* species [15]. Although continuous efforts regarding PCR techniques have been made to develop new diagnostic techniques, such as *Plasmodium* species-specific PCR-restriction fragment length polymorphism (PCR-RFLP) [16], for identifying malaria parasites, nested PCR techniques with high sensitivity and specificity are needed.

According to the molecular technique, two subspecies of *P. ovale*, *curtisi* and *P. ovale wali-keri*, were identified in 2010 by a nested PCR detection assay of dimorphism in the gene encoding the *P. ovale* tryptophan-rich antigen (potra) in West Africa [17]. Currently, *P. falciparum* is still the leading cause of severe malaria [18]. *P. ovale* is usually associated with low morbidity and mortality. However, *P. ovale* can cause severe complications and death [19, 20]. Previous studies have reported that the severe complications of *P. ovale* infections include acute respiratory distress syndrome (ARDS) [21–26], renal impairment [24, 27], jaundice, and hypotension [26, 27]. The most recent study on severe *P. ovale* malaria in travelers and migrants demonstrated that 5.3% of patients with *P. ovale* developed severe complications according to the 2015 WHO criteria, including hyperbilirubinemia, pulmonary edema, shock, significant bleeding, and impaired consciousness [28]. There are a limited number of systematic reviews and meta-analyses on severe *P. ovale* malaria. A previous systematic review of 33 articles published between 1922 and 2015 demonstrated that a total of five out of 22 severe cases of *P. ovale* malaria were fatal, and two cases of congenital *P. ovale* malaria were fatal [29]. A more recent systematic review conducted by Yerlikaya et al. in 2018 demonstrated that a RDT had poor performance in detecting *P. ovale* because *P. ovale* infections usually occur at very low parasite densities, leading to missed detection by microscopy and RDTs [30]. Nevertheless, studies on the prevalence of severe *P. ovale* malaria provide more information on this neglected species and are urgently needed. This systematic review and meta-analysis aimed to investigate the severity and mortality rates of severe *P. ovale* infection to increase the awareness of physicians regarding the prognosis of this severe disease and outcome-related deaths in countries in which this disease is endemic and to identify the differences in the proportions of patients with severe *P. ovale* manifestations and with other severe *Plasmodium* spp. infections.
Methods

Study selection

This systematic review was designed on the basis of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (S1 Checklist). Two authors (MK and KUK) searched the Medline, Scopus, and ISI Web of Science databases independently for articles published in English prior to January 5, 2020. Additional articles from other databases were also selected. The search terms, which included the Boolean operators “OR” or “AND”, were as follows: “(severe OR complicated OR complication) AND (Plasmodium ovale)” (S1 Table). EndNote software version X7 (Thomson Reuters, New York, NY) was used to process all references in our study.

Quality of the included studies

The quality of the observational studies was assessed in accordance with the Newcastle-Ottawa Scale (NOS) [31]. A ‘star system’, in which a study is judged on the basis of the following three broad perspectives was used: the selection of the study groups; the comparability of the groups; and the ascertainment of either the exposure or outcome of interest.

Data extraction

All raw and available data from eight studies were extracted to explore the prevalence of severe complications in patients with P. ovale infections. Two authors (MK and KUK) extracted the data from the selected studies. If there were any discrepancies between the two reviewers, another reviewer (GM) determined whether the study should be included. For the prevalence outcome of severe P. ovale malaria, all prospective cohort, cross-sectional studies, and case-control studies that reported the number of patients with severe complications from P. ovale were included. The following articles were excluded from our study: book and book chapter reviews, conference papers, editorials, letters, correspondences, notes, reviews, animal studies, case reports, drug studies and clinical trials, entomological studies, experimental studies, knowledge assessments, studies written in local languages, studies on mixed infections of P. ovale and other Plasmodium spp., studies of P. ovale during pregnancy, prevalence studies of non-P. ovale infections, and prevalence studies of P. ovale that did not report data on severe complications.

Statistical analysis

The primary outcome of the present study was the pooled prevalence of severe P. ovale infections and mortality rates. The number of severe P. ovale infections and the total number of P. ovale infections were entered into an Excel data sheet (Microsoft Corporation, USA). The pooled prevalence of the severity of P. ovale mono-infections was estimated using the command “metaprop case population, random/fixed” available in STATA software version 15.0 (StataCorp LLC, USA). The results are presented as the pooled estimate and 95% confidence interval (CI). A meta-regression with the median age as a covariate was performed to evaluate whether the age of the participants was a confounder of the pooled prevalence of severe P. ovale.

The secondary outcome of the present study was the different proportions of severity between patients with P. ovale and those with other Plasmodium species. The meta-analyses of the proportions were performed using a random-effects model provided in Review Manager 5.3 software (Cochrane Community). The heterogeneity was assessed with the Mantel-Haenszel method, and the I² values were also calculated. The I² was considered low (≤25%),
moderate (25–50%), or high (>50%). A fixed-effects model was used when $I^2 < 50\%$, whereas a random-effects model was used when $I^2 > 50\%$. The publication bias was also assessed using funnel plots.

Results

Characteristics of included studies

A total of 1,504 articles were retrieved from three databases during the search, and eight of these articles, including retrospective studies, prospective studies, and case series, were included in this study (Fig 1). The characteristics of the eight studies are shown in Table 1. The studies were conducted in Belgium (2000–2005) [32], the Ivory Coast (2007–2008) [33], the US National Malaria Surveillance System (NMSS) (1985–2011) [19], Indonesia (2004–2013) [34], Ethiopia (2013–2014) [35], Italy (2014–2017) [36], Spain (2005–2011) [37], and Sweden (1995–2015) [38]. The ages of the participants were reported in seven of the included studies [32–38], as they were not reported in a study by Hwang et al. [19]. Five studies reported severe imported P. ovale infections in European countries, including Belgium [32], France [33], Italy [36], Spain [37], and Sweden [38], while other studies reported severe P. ovale infections in the United States of America [19], Indonesia [34], and Ethiopia [35]. Most of the included studies (7/8, 87.5%) used microscopy as the gold standard for malaria identification, except for a study conducted by Ramos et al. in 2016 [35]. The combination of microscopy and PCR was used in three of the included studies [19, 36, 37]. Most of the Plasmodium spp. infections that were reported among the included studies were caused by P. falciparum (116,898 cases, 51%), P. vivax (78,282 cases, 34.1%), mixed infection (26,049 cases, 11.4%), P. malariae (6,428 cases, 2.8%), and P. ovale (1,365 cases, 0.6%).

Prevalence of severe P. ovale infections

Among the eight included studies, which included a total of 1,365 ovale malaria cases, the pooled prevalence of severe P. ovale was 0.03 (95% CI = 0.03–0.05%, $I^2 = 54.4\%$). The highest proportions of severe P. ovale were found in the study by de Laval et al. [33] (OR = 0.17, 95% CI = 0.03–0.56) and the study by Bottieau et al. [32] (OR = 0.12, 95% CI = 0.05–0.27), whereas the lowest proportion was found in the study by Langford et al., 2015 [34] (Fig 2). According to the results of the meta-regression of six included studies, the age of the participants did not confound to the pooled prevalence of severe P. ovale infections (P-value = 0.3, 95% CI = -0.002–0.005). The complications most commonly found in patients with P. ovale infections were jaundice (1.1%), severe anemia (0.88%), and pulmonary impairments (0.59%) (Table 2).

Comparison of severity between the P. ovale and Plasmodium spp. infections

The meta-analysis of three included studies [19, 34, 38] demonstrated that a smaller proportion of patients with P. ovale than of patients with P. falciparum had severe infections (P-value = 0.01, OR = 0.36, 95% CI = 0.16–0.81, $I^2 = 72\%$) (Fig 3).

The meta-analysis of four studies [19, 32, 34, 38] revealed there are no significant differences in the proportion of patients with severe P. ovale infections and that of patients with severe P. vivax infections (P-value = 0.75, OR = 0.91, 95% CI = 0.5–1.65, $I^2 = 47\%$) (Fig 4).

The meta-analysis of four studies [19, 32, 34, 38] demonstrated that there was no significant difference between the proportion of patients with severe P. ovale infections and the proportion of patients with severe P. malariae infections (P-value = 0.75, OR = 0.92, 95% CI = 0.56–1.52, $I^2 = 0\%$) (Fig 5).
Fig 1. Flow diagram. Flow chart of the study selection process.

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| No. | Reference | Study area (years of the survey) | Participants | Age range | Reference method for malaria identification | Plasmodium sp. | Severe malaria | Fetal | Impaired consciousness | Prostration | Severe anemia | Renal impairment | Hyperbilirubinemia | Pulmonary | Significant bleeding | Shock |
|-----|-----------|---------------------------------|--------------|-----------|---------------------------------------------|----------------|---------------|-------|------------------------|------------|--------------|----------------|---------------------|-----------|---------------------|-------|
| 1.  | Bottieau et al., 2006 | Belgium (2000–2005) | Travelers, expatriates, and foreign Visitors | 35 (11–77) | Microscopy | Pf = 48 | Pf = 8 | Po = 34 | Po = 4 | Pf = 16 | Pf = 3 | Pr = 8 | Po = 4 | Pm = 3 | Po = 1 |
| 2.  | de Laval et al., 2010 | France (2007–2008) | French soldiers | 32 (30–36) | Microscopy | Case series | Po = 1 | Po = 6 |
| 3.  | Hwang et al., 2014 | USA (1985–2011) | Travelers | NA | Microscopy | Pf = 15,272 | Pf = 1,416 | Pf = 122 | Pf = 514 | Pf = 140 | Pf = 303 | Pf = 176 | Pr = 35 | Po = 4 | Pm = 1 |
| 4.  | Langford et al., 2015 | Indonesia (2004–2013) | Patients presenting to the hospital | <1 | Microscopy | Pf = 100,078 | Pf = 4,038 | Pr = 65,306 | Pr = 2,118 | Po = 120 | Po = 1 | Pf = 2,521 | Pf = 415 | Po = 1,099 | Po = 81 | Po = 0 | Po = 1 | Po = 1 |
| 5.  | Ramos et al., 2016 | Ethiopia (2013–2014) | Patients with severe anemia | 15 (0.5–65) | PCR | Pf = 111 | Pf = 4 | Po = 4 |
| 6.  | Rojo-Marcos et al., 2018 | Italy (2014–2017) | Patients with imported P. ovale | 33 (22.2–53) | Microscopy | Pf = 79 | Po = 5 | Po = 4 |
| 7.  | Rojo-Marcos et al., 2014 | Spain (2005–2011) | Patients with imported P. ovale | 36.5 (11.8–52.7) | Microscopy | Pf = 35 | Po = 3 | Po = 2 |
| 8.  | Wangdahl et al., 2019 | Sweden (1995–2015) | Travelers and Migrants | All = 32.6 (0.2–83) | Microscopy | Pf = 1,548 | Pf = 146 | Pf = 3 | Pf = 28 | Pf = 9 | Pf = 16 | Pf = 31 | Pf = 66 | Pf = 15 | Pf = 17 | Pf = 33 |
|     |            |                                |              | Pf = 34.4 (0.2–83) | Pr = 776 | Pr = 60 | Pr = 0 | Pr = 1 | Pr = 1 | Pr = 15 | Pr = 1 | Pr = 28 | Pr = 4 | Pr = 5 | Pr = 8 |
|     |            |                                |              | Pr = 28.9 (1–79) | Po = 188 | Po = 10 | Po = 0 | Po = 1 | Po = 0 | Po = 0 | Po = 0 | Po = 2 | Po = 1 | Po = 2 |
|     |            |                                |              | Pr = 30.2 (0–65) | Po = 64 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 |
|     |            |                                |              | Po = 30.2 (0–65) | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 |
|     |            |                                |              | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 |

Total cases

Pr = 118,898 | Pf = 5,651 | Pf = 125 | Pf = 542 | Pf = 9 | Pf = 395 | Pf = 949 | Pf = 66 | Pf = 1,286 | Pf = 17 | Pf = 33 |
Pr = 78,282 | Pf = 2,353 | Pf = 10 | Pf = 38 | Pf = 1 | Pf = 1,134 | Pf = 130 | Pf = 36 | Pf = 977 | Pr = 5 | Pr = 8 |
Pr = 1,365 | Po = 46 | Po = 2 | Po = 5 | Po = 2 | Po = 13 | Po = 6 | Po = 15 | Po = 8 | Po = 1 | Po = 2 |
Pr = 6,428 | Po = 187 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 | Po = 2 |
Pr = 26,049 | Mixed = 1,217 | Mixed = 1 | Mixed = 1 | Mixed = 1 | Mixed = 786 | Mixed = 87 | Mixed = 4 | Mixed = 343 | Mixed = 1 | Mixed = 3 |

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Mortality rates of *P. ovale* and *Plasmodium* spp. infections

There was only one study that reported death cases resulting from *P. ovale* infections [19]. The mortality rate of severe *P. ovale* infections in their study was 0.22% (2/903). The mortality rate of severe *P. ovale* infections among the eight included studies was 0.15% (2/1,365).

**Table 2. Complications associated with *P. ovale*.

| Major complication (WHO, 2014) | Total number of patients with a severe case | Proportion of complications (%) among the total number of *P. ovale* cases (1,365 cases) |
|-------------------------------|-------------------------------------------|------------------------------------------------------------------------------------------|
| Pulmonary impairment          | 8                                         | 0.59                                                                                     |
| Cerebral malaria              | 5                                         | 0.37                                                                                     |
| Renal impairment              | 6                                         | 0.44                                                                                     |
| Prostration                   | 2                                         | 0.15                                                                                     |
| Hypotension/shock             | 2                                         | 0.15                                                                                     |
| Jaundice                      | 15                                        | 1.10                                                                                     |
| Severe anemia                 | 13                                        | 0.95                                                                                     |
| Bleeding/DIC                  | 1                                         | 0.07                                                                                     |

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The mortality rate of other *Plasmodium* infections reported in the eight included studies was 0.11% for *P. falciparum* infections, 0.013% for *P. vivax* infections, 0.03% for *P. malariae* infections, and 0.004% for mixed infections.

**Quality of included studies**

Four studies were rated as having 9 stars (good quality), three studies were rated as having 7 stars (adequate quality), and one study was rated as having 6 stars (adequate quality) according to the NOS concerning the selection process for the cases and controls included (Table 3).

| Study or Subgroup | P. ovale Events Total | P. falciparum Events Total | Odds Ratio M-H, Random, 95% CI | Odds Ratio M-H, Random, 95% CI | Risk of Bias A B C D E F G |
|-------------------|----------------------|-----------------------------|--------------------------------|--------------------------------|-----------------------------|
| Hwang et al., 2014 | 22 903               | 1416 15272                  | 46.0%                          | 0.24 [0.16, 0.37]               |                             |
| Langford et al., 2015 | 1 120           | 4031 100078                 | 13.2%                          | 0.20 [0.03, 1.43]               |                             |
| Wangdahi et al., 2019 | 12 188          | 146 1548                    | 40.8%                          | 0.65 [0.36, 1.20]               |                             |
| Total (95% CI)    | 1211 116898        | 100.0%                      | 0.36 [0.16, 0.81]              |                                |                             |
| Total events      | 35 5593            |                             |                                |                                |                             |

Heterogeneity: Tau² = 0.34; Chi² = 7.26, df = 2 (P = 0.03); I² = 72%

Test for overall effect: Z = 2.44 (P = 0.01)

Risk of bias legend

(A) Random sequence generation (selection bias)
(B) Allocation concealment (selection bias)
(C) Blinding of participants and personnel (performance bias)
(D) Blinding of outcome assessment (detection bias)
(E) Incomplete outcome data (attrition bias)
(F) Selective reporting (reporting bias)
(G) Other bias

**Fig 3. Forest plot comparing severe *P. ovale* cases and *P. falciparum* cases.** The forest plot compared the proportions of patients with severe *P. ovale* infections with that of patients with *P. falciparum* infections.

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**Fig 4. Forest plot comparing severe *P. ovale* cases and *P. vivax* cases.** The forest plot compared the proportions of severe *P. ovale* cases and *P. vivax* cases.

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Publication bias

A funnel plot analysis of the included studies could not be performed to assess the publication bias among the included studies, as a minimum of 10 studies were required for the analysis [39].

Table 3. Quality of the included studies.

| No. | Reference                   | Is the Case Definition Adequate? | Representativeness of the Cases | Selection of Controls | Definition of Controls | Ascertainment of Exposure | Same method of ascertainment for cases and controls | Non-Response Rate |
|-----|-----------------------------|---------------------------------|--------------------------------|-----------------------|------------------------|--------------------------|-----------------------------------------------------|------------------|
| 1.  | Bottieau et al., 2006       | ✴ ✴ ✴ ✴ ✴                        | ✴                              | ✴                     | ✴                       | *                        | *                                                   | ✴                |
| 2.  | de Laval et al., 2010       | ✴                                | ✴                              | ✴                     | ✴                       | *                        | *                                                   | ✴                |
| 3.  | Hwang et al., 2014          | ✴                                | ✴                              | ✴                     | ✴                       | **                       | *                                                   | ✴                |
| 4.  | Langford et al., 2015       | ✴                                | ✴                              | ✴                     | **                      | *                        | *                                                   | ✴                |
| 5.  | Ramos et al., 2016          | ✴                                | ✴                              | ✴                     | **                      | *                        | *                                                   | ✴                |
| 6.  | Rojo-Marcos et al., 2018    | ✴                                | ✴                              | ✴                     | **                      | *                        | *                                                   | ✴                |
| 7.  | Rojo-Marcos et al., 2014    | ✴                                | ✴                              | ✴                     | **                      | *                        | *                                                   | ✴                |
| 8.  | Wangdahl et al., 2019       | ✴                                | ✴                              | ✴                     | **                      | *                        | *                                                   | ✴                |
Discussion

The present systematic review and meta-analysis aimed to explore the prevalence of severe *P. ovale* infections and to summarize the mortality caused by *P. ovale*. The results demonstrated that 3% of patients infected with *P. ovale* developed severe complications according to the WHO 2014 guidelines [1]. Therefore, the term “benign malaria” might not apply to patients tested positive with *P. ovale*, especially travelers after returning from countries in which *P. ovale* is endemic. In addition, international and national healthcare providers should be recommending the use of chemoprophylaxis for travelers, as the present study revealed that *P. ovale*, long-recognized as benign malaria, can cause severe and fatal clinical manifestations. In the present study, a meta-regression with age as a covariate was performed, and the pooled prevalence of severe *P. ovale* infections was investigated. The results showed that the age of the participants in six of the included studies did not confound the pooled prevalence of severe *P. ovale* infections. A previous study demonstrated that the highest incidence of *P. ovale* was observed among children 0–7 years old, who had an 8-fold higher risk of *P. ovale* infections than did adults; nevertheless, clinical attacks were observed in all age groups [40].

The present study also demonstrated a significant difference between the proportion of patients with severe *P. ovale* infections and severe *P. falciparum* infections. A lower proportion of patients with severe *P. ovale* infections than of patients with *P. falciparum* infections had severe cases. This finding indicated that although *P. ovale* can cause severe malaria, the chance of patients developing severe complications is lower with *P. ovale* than with *P. falciparum*, which is the *Plasmodium* species widely known to be the leading cause of severe malaria in humans. This study also demonstrated there are no significant differences in the proportions of patients with severe *P. ovale* and with *P. vivax/P. malariae*. This finding demonstrated that there are relatively few severe *P. ovale* cases, including *P. vivax* and *P. ovale* cases. In contrast to *P. vivax*, which is endemic in Asia, Central America, and South America, *P. ovale* is only endemic in some African countries [3]. However, the study conducted by Langford et al. in 2015 [34] indicated that there were 120 cases of *P. ovale* infection in Indonesia, whereas other included studies reported that travelers who returned to their home countries had imported *P. ovale* malaria cases [19, 32, 35–38].

Although microscopy is still considered to be the gold standard for the identification of *Plasmodium* species, the morphological characteristics of *P. ovale* are similar to those of *P. vivax* and are readily missed by less experienced microscopists [41, 42]. This problem may have caused certain *P. ovale* cases to have been missed in the included studies conducted by Bottieau et al. [32], de Laval et al. [33], Langford et al. [34], and Wangdahl et al. [38], as only microscopy was used to identify malaria parasites. Missed *P. ovale* cases have been reported when only microscopy was used [43]. In addition, missed diagnoses of *P. ovale* infections leading to relapses with a low parasitemia level is a problem related to this *Plasmodium* species [44]. Previous studies have indicated that the sensitivity of routine microscopy in detecting *P. ovale* in imported cases is very low and related to low parasitemia levels [32, 45]. Moreover, the rapid diagnosis tests (RDTs) that have been developed and are available lack the sensitivity required to detect low amounts of the circulating antigen of *P. ovale* [46, 47]. Moreover, a previous study indicated that routine microscopic examinations with thick and thin blood smears should be repeated three times in cases of suspected imported *P. ovale* malaria [48]. Ideally, molecular detection using polymerase chain reaction (PCR) to detect and confirm *P. ovale* infections in cases of low parasitemia or negative blood film is needed, although it is not routinely available [32, 43]. Therefore, the limitations of routine microscopic and RDT tests might affect the management/treatment of patients and lead to an increase in the morbidity associated with *P. ovale* infections [49]. The mortality caused by severe *P. ovale* infections among
travelers who returned to the USA was previously reported by Hwang et al. in 2014 [19]. The authors recommended the use of suppressive prophylaxis and antirelapse treatment with primaquine drugs for patients who returned from ovale-endemic countries [19]. Nevertheless, a previous case report showed that severe complications led to fatality in patients who had received anti-malarial prophylaxis treatment during their trip to Nigeria [20]. This finding may have been caused by the improper usage of prophylactic drugs or the survival of hypnozoites during anti-malaria chemoprophylaxis against the *P. ovale* infection [50].

The mechanism by which *P. ovale* infections lead to death is still unknown, but acute renal failure and acute respiratory distress syndrome might act as secondary contributing factors to death [51]. People who previously acquired *P. falciparum* malaria and those staying in malaria-endemic regions are known to show less severe symptoms during subsequent malaria infections than are people who are from non-malaria endemic regions. A previous study investigated two cases of *P. ovale* in patients, and it was suggested that one patient with a history of malaria was protected against developing severe complications, while the other patient, who was experiencing malaria infection for the first time, suffered severe complications [51]. Therefore, severe *P. ovale* infections can occur in nonimmune individuals in particular.

This systematic review indicated that jaundice, severe anemia, and pulmonary impairment are the severe complications most commonly found in patients infected with *P. ovale*. For jaundice, a previous study indicated that mild hyperbilirubinemia can be found in approximately 50% of patients infected with *P. ovale* [52]. Jaundice concomitant with *P. ovale* infection might be due to liver dysfunction to conjugate bilirubin in cases of severe red blood cell destruction during malaria infection. Severe anemia concomitant with *P. ovale* infection might occur in patients with underlying diseases such as hemoglobinopathies, causing the severe destruction of abnormal red blood cells. The mechanism of pulmonary impairment in patients with *P. ovale* malaria is not clear. In patients with *P. falciparum* infection, cytoadherence, mechanical obstruction, and inflammation of the microvascular endothelium at the pulmonary vasculature due to infected red blood cells that cytoadhere to the microvascular endothelium causes the mechanical obstruction of pulmonary vasculature; thus, alveolar capillary permeability increases, and intravascular fluid is spread into the lungs, leading to pulmonary failure [53].

**Summary of the evidence**

The present systematic review and meta-analysis presented cases of severe manifestations and demonstrated the severity and mortality of severe *P. ovale* infection to increase the awareness of physicians regarding the prognosis of this disease. It is of the utmost importance to test individuals for malaria after they return from malaria-endemic areas, as severe *P. ovale* complications could occur, especially in nonimmune travelers. Last, the knowledge on appropriate management strategies and the necessary interventions concerning malaria diagnosis affects the severity of malaria manifestations and patient outcomes. Clinicians' and technicians' familiarity with the endemicity of malaria in an area or a region therefore plays an important role in the initial stages of laboratory assessments and subsequent diagnoses.

**Limitations**

A limitation of this systematic review and meta-analysis was that there were a limited number of available publications on severe complications and death related to *P. ovale* infection. Another limitation was that results from case series and case-control studies were used for prevalence estimations. However, previous studies XXX have suggested and used case reports/series in meta-analyses to facilitate the decision making process when the evidence was limited
or a relatively rare or neglected disease was assessed [54–59]. In addition, the most recently published meta-analysis of 44 case series reported the 25-year pooled survival of hip replacements [60]. Considering these limitations, the meta-analysis results and conclusions on the prevalence of severity and mortality associated with *P. ovale* infection should be considered with caution in combination with the results of newly published reports.

**Conclusion**

This systematic review demonstrated that although *P. ovale* infections have long been considered cases of benign malaria, severe complications in patients have been reported on rare occasions. The possible reasons for the small number of reports on the severity of the malaria infections linked or caused directly by *P. ovale* should be studied further by the scientific community. Clinicians and technicians need to recognize that patients who return from malaria-endemic areas may develop severe *P. ovale* infections. Additional studies need to consider the potential underreported presence of *P. ovale* malaria infections in the clinical setting so that appropriate management strategies and interventions can be administered to infected individuals and the knowledge on malaria epidemiology can be expanded further.

**Supporting information**

**S1 Checklist.** PRISMA checklist. PRISMA statement for reporting systematic reviews and meta-analyses.

(DOC)

**S1 Table.** Search term.

(DOCX)

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1. World Health Organization. Guidelines for the treatment of malaria. 3rd ed. Geneva: WHO; 2015.
2. Stephens JWW. A new malaria parasite of man. Ann Trop Med Parasitol. 1922; 16:383–8.
3. Collins WE, Jeffery GM. Plasmodium ovale: parasite and disease. Clin Microbiol Rev. 2005; 18(3):570–81. https://doi.org/10.1128/CMR.18.3.570-581.2005 PMID: 16020691
4. Mehlotra RK, Lorry K, Kastens W, Miller SM, Alpers MP, Bockarie M, et al. Random distribution of mixed species malaria infections in Papua New Guinea. The American journal of tropical medicine and hygiene. 2000; 62(2):225–31. https://doi.org/10.4269/ajtmh.2000.62.225 PMID: 10813477
5. May J, Mockenhaupt FP, Ademowo AG, Falusui PE, Bienzle U, et al. High rate of mixed and subpatent malarial infections in south-west Nigeria. The American journal of tropical medicine and hygiene. 1999; 61(2):339–43. https://doi.org/10.4269/ajtmh.1999.61.339 PMID: 10463691
6. Dini S, Douglas NM, Poespoprodjo JR, Kenangalem E, Sugarto P, Plumb ID, et al. The risk of morbidity and mortality following recurrent malaria in Papua, Indonesia: a retrospective cohort study. BMC Med. 2020; 18(1):28. https://doi.org/10.1186/s12916-020-1497-0 PMID: 32075649
7. Mehlotra RK, Lorry K, Kastens W, Miller SM, Alpers MP, Bockarie M, et al. Random distribution of mixed species malaria infections in Papua New Guinea. The American journal of tropical medicine and hygiene. 2000; 62(2):225–31. https://doi.org/10.4269/ajtmh.2000.62.225 PMID: 10813477
8. Yu T, Fu Y, Kong X, Liu X, Yan G, Wang Y. Epidemiological characteristics of imported malaria in Shandong Province, China, from 2012 to 2017. Sci Rep. 2020; 10(1):7568. https://doi.org/10.1038/s41598-020-64593-1 PMID: 32371895
9. Lysenko AJ, Beljaev AE. An analysis of the geographical distribution of Plasmodium ovale. Bull World Health Organ. 1969; 40(3):383–94. PMID: 5306622
10. Smith AD, Bradley DJ, Smith V, Blaze M, Behrens RH, Chiodini PL, et al. Imported malaria and high risk groups: observational study using UK surveillance data 1987–2006. BMJ (Clinical research ed). 2008; 337:a120.
11. Tanizaki R, Kato Y, Iwagami M, Kutsuna S, Ujie M, Takeshita N, et al. Performance of rapid diagnostic tests for Plasmodium ovale malaria in Japanese travellers. Trop Med Health. 2014; 42(4):149–53. https://doi.org/10.2149/thm.2014-07 PMID: 25473374
12. Moody A. Rapid diagnostic tests for malaria parasites. Clin Microbiol Rev. 2002; 15(1):66–78. https://doi.org/10.1128/cmr.15.1.66-78.2002 PMID: 11781267
13. McMorow ML, Aidoo M, Kachur SP. Malaria rapid diagnostic tests in elimination settings—can they find the last parasite? Clin Microbiol Infect. 2011; 17(11):1624–31. https://doi.org/10.1111/j.1469-0691.2011.03639.x PMID: 21910780
14. Mason DP, Kawamoto F, Lin K, Laoboonchai A, Wongsrichanalai C. A comparison of two rapid field immunochromatographic tests to expert microscopy in the diagnosis of malaria. Acta Trop. 2002; 82(1):51–9. https://doi.org/10.1016/s0001-706x(02)00031-1 PMID: 11904103
15. Snounou G, Viriyakosol S, Jarra W, Thaithong S, Brown KN. Identification of the four human malaria parasite species in field samples by the polymerase chain reaction and detection of a high prevalence of mixed infections. Mol Biochem Parasitol. 1993; 58(2):283–92. https://doi.org/10.1016/0166-6851(93)90050-8 PMID: 8479452
16. Sharma S, Kaitikola K, Bharti RS, Singh MP, Mishra N. Novel molecular diagnostic technique for detecting the different species of Plasmodium. Infect Genet Evol. 2020; 78:104122. https://doi.org/10.1016/j.meegid.2019.104122 PMID: 31751755
17. Sutherland CJ, Tanomsing N, Nolder D, Oguike M, Jennison C, Pukrittayakamee S, et al. Two non-combining sympatric forms of the human malaria parasite Plasmodium ovale occur globally. J Infect Dis. 2010; 201(10):1544–50. https://doi.org/10.1086/652240 PMID: 20380562
18. WHO. World malaria report 2018. https://www.who.int/malaria/publications/world-malaria-report-2018/en/
22. Hachimi MA, Hatim EA, Moudden MK, Elkartouti A, Errami M, Louzi L, et al. The acute respiratory distress syndrome in malaria: Is it always the prerogative of Plasmodium falciparum? Rev Pneumol Clin. 2013; 69(5):283–6. https://doi.org/10.1016/j.pneumol.2013.03.001 PMID: 23688721

23. Haydoura S, Mazboudi O, Charafeddine K, Bouakl I, Babak TA, Taher AT, et al. Transfusion-related Plasmodium ovale malaria complicated by acute respiratory distress syndrome (ARDS) in a non-endemic country. Parasitol Int. 2011; 60(1):114–6. https://doi.org/10.1016/j.parint.2010.10.005 PMID: 20971212

24. Lau YL, Lee WC, Tan LH, Kamaruzaman A, Omar SFS, Fong MY, et al. Acute respiratory distress syndrome and acute renal failure from Plasmodium ovale infection with fatal outcome. Malar J. 2013; 12:8. https://doi.org/10.1186/1475-2875-12-8

25. Rojo-Marcos G, Cuadros-Gonzalez J, Mesa-Latorre JM, Culebras-Lopez AM, de Pablo-Sanchez R. Acute respiratory distress syndrome in a case of Plasmodium ovale malaria. The American journal of tropical medicine and hygiene. 2008; 79(3):391–3. PMID: 18784231

26. Strydom KA, Ismail F, Frean J. Plasmodium ovale: A case of not-so-benign tertian malaria. Malar J. 2014; 13(1):85.

27. Tomar LR, Giri S, Baudhh NK, Jhamb R. Complicated malaria: A rare presentation of Plasmodium ovale. Trop Doct. 2015; 45(2):140–2. https://doi.org/10.1177/0049475515571989 PMID: 25672340

28. Wangdahl A, Wyss K, Saduddin D, Botta M, Ydring E, Vikerfors T, et al. Severity of Plasmodium falciparum and non-falciparum malaria in travelers and migrants: A nationwide observational study over 2 decades in Sweden. J Infect Dis. 2019; 220(8):1335–45. https://doi.org/10.1093/infdis/jjz292 PMID: 31175365

29. Groger M, Fischer HS, Veletzky L, Laiemruuta A, Ramharter M. A systematic review of the clinical presentation, treatment and relapse characteristics of human Plasmodium ovale malaria. Malaria journal. 2017; 16(1):112. https://doi.org/10.1186/s12936-017-1599-2 PMID: 28284211

30. Yerlikaya S, Campillo A, Gonzalez IJ. A systematic review: Performance of rapid diagnostic tests for the detection of Plasmodium knowlesi, Plasmodium malariae, and Plasmodium ovale mono-infections in human blood. J Infect Dis. 2018; 218(2):265–76. https://doi.org/10.1093/infdis/jjy150 PMID: 29554284

31. Lo CK, Mertz D, Loeb M. Newcastle-Ottawa Scale: comparing reviewers’ to authors’ assessments. BMC Med Res Methodol. 2014; 14:45. https://doi.org/10.1186/1471-2288-14-45 PMID: 24690082

32. Bottieau E, Clerinx J, Van Den Enden E, Van Esbroeck M, Colebunders R, Van Gompel A, et al. Imported non-Plasmodium falciparum malaria: A five-year prospective study in a European referral center. Am J Trop Med Hyg. 2006; 75(1):133–8. PMID: 16837719

33. de Laval F, Oliver M, Rapp C, Pommier de Santis V, Mendibil A, Deparis X, et al. The challenge of diagnosing Plasmodium ovale malaria in travellers: report of six clustered cases in French soldiers returning from West Africa. Malaria journal. 2010; 9:358. https://doi.org/10.1186/1475-2875-9-358 PMID: 21143962

34. Langford S, Douglas NM, Lampah DA, Simpson JA, Kenangalem E, Sugiiarto P, et al. Plasmodium malariae infection associated with a high burden of anaemia: A hospital-based surveillance study. PLoS Negl Trop Dis. 2015; 9(12):e0004195. https://doi.org/10.1371/journal.pntd.0004195 PMID: 26720002

35. Ramos JM, Tissiano G, Gosa A, Alegria I, Berzosa P. Study of the prevalence of Plasmodium infections by the polymerase chain reaction (PCR) among patients with severe anaemia treated at a rural hospital in southern Ethiopia. Bol Malaria Salud Ambient. 2011; 50(1):114–6. https://doi.org/10.1016/j.parint.2010.10.005 PMID: 20971212

36. Rojo-Marcos G, Rubio-Munoz JM, Angeheben A, Jaureguiberry S, Garcia-Bujalance S, Tomasoni LR, et al. Prospective comparative multi-centre study on imported Plasmodium ovale wallikeri and Plasmodium ovale curtisi infections. Malar J. 2018; 17:11. https://doi.org/10.1186/s12936-017-2153-9

37. Rojo-Marcos G, Rubio-Muñoz JM, Ramírez-Olivencia G, García-Bujalance S, Elcuaz-Romano R, Díaz-Menéndez M, et al. Comparison of imported Plasmodium ovale curtisi and P. ovale wallikeri infections among patients in Spain, 2005–2011. Emerg Infect Dis. 2014; 20(3):409–16. https://doi.org/10.3201/eid2003.130745 PMID: 24572501

38. Wångdahl A, Wyss K, Saduddin D, Botта M, Ydring E, Vikerfors T, et al. Severity of Plasmodium falciparum and non-falciparum malaria in travelers and migrants: A Nationwide Observational Study over 2 Decades in Sweden. J Infect Dis. 2019; 220(8):1335–45. https://doi.org/10.1093/infdis/jjz292 PMID: 31175365

39. Cochrane. Cochrane Handbook for Systematic Reviews of Interventions version 6.0 (updated July 2019). 2019 [Available from: https://training.cochrane.org/handbook/current.

40. Faye FB, Spiegel A, Tall A, Sodhna C, Fontenille D, Rogier C, et al. Diagnostic criteria and risk factors for Plasmodium ovale malaria. J Infect Dis. 2002; 186(5):690–5. https://doi.org/10.1086/342395 PMID: 12195397
41. Mueller I, Zimmerman PA, Reeder JC. Plasmodium malariae and Plasmodium ovale—the "bashful" malaria parasites. Trends Parasitol. 2007; 23(6):278–83. https://doi.org/10.1016/j.pt.2007.04.009 PMID: 17459775

42. Obare P, Ogutu B, Adams M, Odera JS, Lilley K, Dosoo D, et al. Misclassification of Plasmodium infections by conventional microscopy and the impact of remedial training on the proficiency of laboratory technicians in species identification. Malaria journal. 2013; 12:113. https://doi.org/10.1186/1475-2875-12-113 PMID: 23537145

43. Calderaro A, Piccolo G, Perandini F, Gorrini C, Peruzzi S, Zuell C, et al. Genetic polymorphisms influence Plasmodium ovale PCR detection accuracy. J Clin Microbiol. 2007; 45(5):1624–7. https://doi.org/10.1128/JCM.02316-06 PMID: 17360843

44. Leder K, Black J, O’Brien D, Greenwood Z, Kain KC, Schwartz E, et al. Malaria in travelers: a review of the GeoSentinel surveillance network. Clin Infect Dis. 2004; 39(8):1104–12. https://doi.org/10.1086/424510 PMID: 15486832

45. Milne LM, Kyi MS, Chiodini PL, Wardhouse DC. Accuracy of routine laboratory diagnosis of malaria in the United Kingdom. J Clin Pathol. 1994; 47(8):740–2. https://doi.org/10.1136/jcp.47.8.740 PMID: 7962629

46. Bigailllon C, Fontan E, Cavall JD, Hernandez E, Spiegel A. Ineffectiveness of the Binax NOW malaria test for diagnosis of Plasmodium ovale malaria. J Clin Microbiol. 2005; 43(2):1011. https://doi.org/10.1128/JCM.43.2.1011-2005 PMID: 15695736

47. Marx A, Pewsner D, Egger M, Nuesch R, Bucher HC, Genton B, et al. Meta-analysis: accuracy of rapid tests for malaria in travelers returning from endemic areas. Ann Intern Med. 2005; 142(10):836–46. https://doi.org/10.7326/0003-4819-142-10-20050517-00009 PMID: 15897534

48. Brent AJ, Angus BJ. Not all that is malaria is falciparum. Lancet Infect Dis. 2008; 8(3):208. https://doi.org/10.1016/S1473-3099(08)70044-6 PMID: 18291342

49. Taylor T, Agbenyega T. Malaria. In: Magill AJ, Hill DR, Solomon T, Ryan ET, editors. Hunter’s Tropical Medicine and Emerging Infectious Disease. 9th ed. Saunders: Elsevier B.V.; 2012.

50. Taylor WR, White NJ. Malaria and the lung. Clin Chest Med. 2002; 23(2):457–68. https://doi.org/10.1016/S0272-5231(02)00004-7 PMID: 12092039

51. Lau YL, Lee WC, Tan LH, Kamaruzaman A, Syed Omar SF, Fong MY, et al. Acute respiratory distress syndrome and acute renal failure from Plasmodium ovale infection with fatal outcome. Malar J. 2013; 12(1):389.

52. Nissen T, Wynn R. The clinical case report: a review of its merits and limitations. BMC Res Notes. 2014; 7:264. https://doi.org/10.1186/1756-0500-7-264 PMID: 24758689

53. Nakamura T, Igarashi H, Ito T, Jensen RT. Important of case-reports/series, in rare diseases: Using neuroendocrine tumors as an example. World J Clin Cases. 2014; 2(11):608–13. https://doi.org/10.1299/wjcc.v2.i11.608 PMID: 25405184

54. Jackso D, Daly J, Saltman DC. Not only randomized controlled trials, but also case series should be considered in systematic reviews of rapidly developing technologies. J Clin Epidemiol. 2009; 62(12):1202-6.e4. https://doi.org/10.1016/j.jclinepi.2008.12.010 PMID: 19349144

55. Evans JT, Evans JP, Walker RW, Blom AW, Whitehouse MR, Sayers A. How long does a hip replacement last? A systematic review and meta-analysis of case series and national registry reports with more than 15 years of follow-up. Lancet. 2019; 393(10172):647–54. https://doi.org/10.1016/S0140-6736(18)31665-9 PMID: 30782340