

# Evaluation of Malaria Species in Haiti: Detection of Sub-Clinical Infection

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## Introduction

**Malaria is a significant public health concern in Haiti where 30,000 cases are reported annually, putting an estimated ten million Haitians at risk of acquiring an infection. Infections are believed to be entirely due to Plasmodium falciparum. There is little existing information on species present in Haiti, asymptomatic reservoirs and comparisons of testing methods.**

## Materials and Methods

**Study design:** In this study, we evaluated malaria species in patients presenting to ten acute care medical and dental clinics in seven separate geographic locations in Haiti from January 2011 through February 2013. Clinical diagnoses were corroborated in the field with Paramax-3(r) rapid test; results were later confirmed by polymerase chain reaction (PCR).



**Sample collection:** Samples were collected by student volunteers, adjacent figure, from the following sources: Symptomatic patients underwent finger prick sampling; samples from asymptomatic patients were collected through secondary sources including 115 tooth extractions and twelve finger prick glucose tests.

**PCR based species testing:** Based on the Rubio et al protocol, 5uL of DNA extracted from filter paper was amplified in a nested reaction as follows: Primer oligo (5'-3'): Universal reverse(GACGGTATCTGATCGTCTTC), Plasmodium forward (AGTGTGTATCCAATCGAGTTTC) and Human forward (GAGCCGCCTGGATACCGC). In the second reaction, 1uL of the product of the first PCR product was amplified using plasmodium forward, falciparum reverse (AGTCCCCTAGAATAGTTACA), malariae reverse (GCCCTCCAATTGCCTTCTG) and vivax reverse (AGGACTTCCAAGCCGAAGC).

**DNA sequencing confirmation:** Selected PCR bands were excised, cloned and sequenced in order to investigate the occurrence of spurious bands.

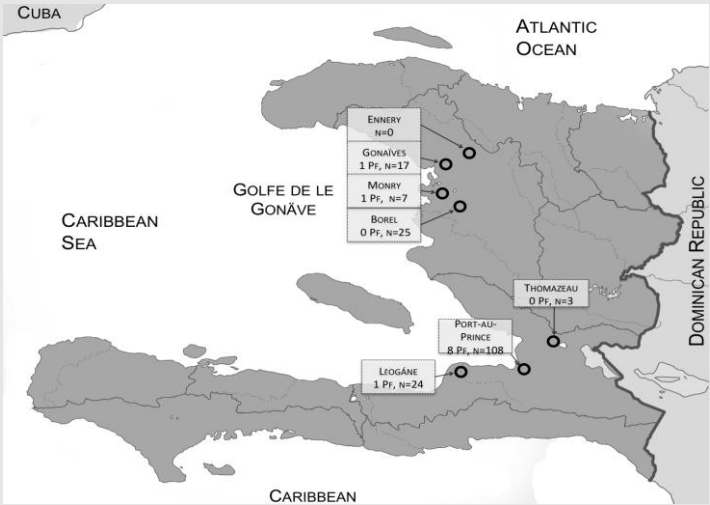
## References

Rubio JM, Benito A, Roche J, et al. Semi-nested, multiplex polymerase chain reaction for detection of human malaria parasites and evidence of Plasmodium vivax infection in Equatorial Guinea. Am J Trop Med Hyg 1999; 60(2): 183-7.  
 Raccurt CP, Ciceron M, Dossil R, Boncy J. Prevalence of Plasmodium falciparum during the rainy season (June-December) in the southeast district of Haiti. Med Sante Trop 2012; 22(4): 435-9.

## Results

- Out of 5775 patients presenting to acute care medical clinics, *P. falciparum* infection was suspected in 51 patients and confirmed in eight of these cases by PCR. Three *P. falciparum* infections were detected by PCR out of 127 tested afebrile patients undergoing tooth extraction or a finger-prick glucose check.
- No other malaria species were detected using PCR. The sequences from 10 spurious PCR products were found to be a result of a human sequence that was amplified by the Plasmodium forward and reverse oligos.

This figure illustrates the geographical distribution of samples collected and locations where *P. falciparum* was detected.



## Conclusions and Future Directions

### Conclusions:

- In accordance with other studies, *P. falciparum* continues to be an important cause of febrile illness in displaced persons following the devastating earthquake and subsequent flooding in 2010.
- Uniquely, results presented here demonstrate the presence of asymptomatic infection, which may act as a reservoir of continued *P. falciparum* transmission. It is important to take this into consideration during future efforts to control and potentially eradicate malaria in Haiti.
- It was also found that human DNA from individuals in this study produced PCR bands that were 50bp different from the targeted *P. vivax* sequence and should be recognized as a potential source of false positives.

### Future Direction:

- The presence of malaria in Haiti is a public health threat for Haiti. Additional information is needed about the species of malaria, drug resistance of strains of malaria, as well as information on the clinical manifestations, severity of disease and complications related to genetic characteristics of patients and malaria strains.
- This information will be critical to aid clinicians in therapy of malaria and the Haitian government in selecting strategies to use for detection and the most appropriate prophylaxis and treatment regimens and reach their malaria elimination goals.