

The Unexpected Stability of Malaria Elimination: Research Needs The Feasibility of Malaria Elimination in Eastern Indonesia

Research Team

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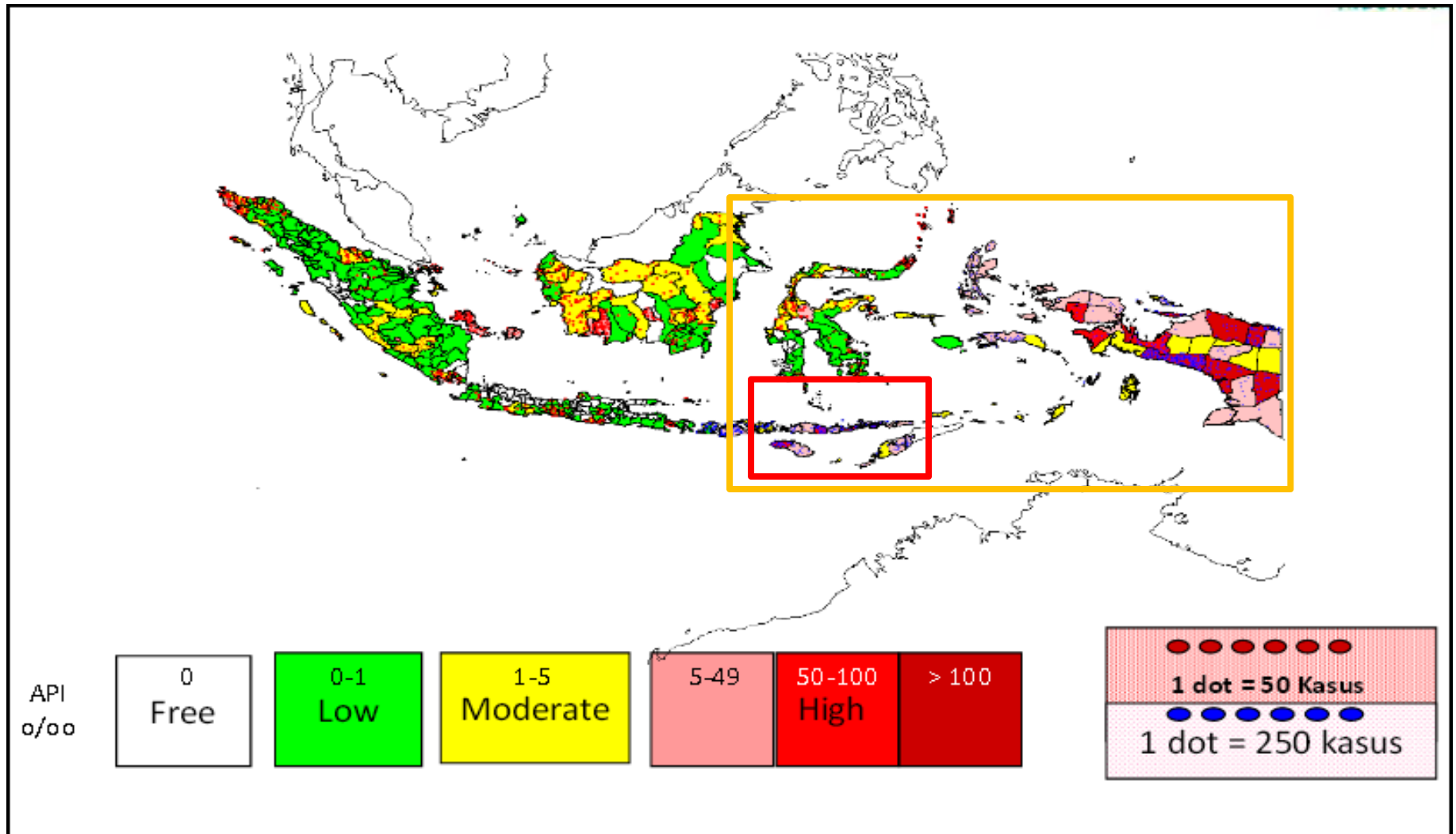


Indonesia will reached elimination Malaria in 2025

WHO Criteria	Current Situation Eastern Indonesia 2014-2016
1 case/1.000 Population	15-18 cases/1.000 Population
Be aware <i>P. vivax</i>	<i>P. Vivax</i> dan <i>P. falciparum</i> are Dominant
<i>Strong Surveillance system</i> ACD (active cases detection) & Mass Blood Survey (MBS)	<i>Surveillance</i> ????? Depend funding and political concern
Gold standard: Microscopic & Molecular	Gold standard: Microscopic Only (Error rate > 5 %)
<i>No local transmission</i>	Limited available data
G6PD Deficient	Limited available data

Source: WHO, 2013, Manual for Pre-elimination Malaria. P. 34-54

The map of malaria diseases in Indonesia 2010-2014



Source: MoH, Republic Indonesia & Eijkman Institute for



Research Purpose

To answer, whether Eastern Indonesia should proceed malaria pre-elimination?

Research Methodology

1. Ethic Approval from Fac. Of Medicine UGM No. Ref: KE/FK/85/EC.

2. A survey (systematic random sampling), total 555 **HEALTHY PEOPLE**.

3. At 5 districts, Eastern Indonesia by three different Annual Parasite Incidence (API).

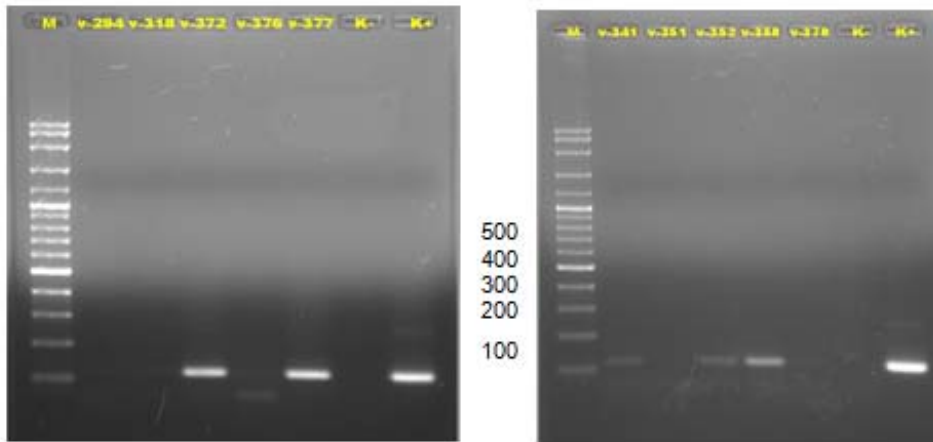
4. Inclusion Criteria:

- Hemoglobin (Hb > 10 gr/dL)
- Healthy People
- Thick and thin blood smears
- Whole blood samples (3 mL EDTA)
- Complete Questioner & Consent form

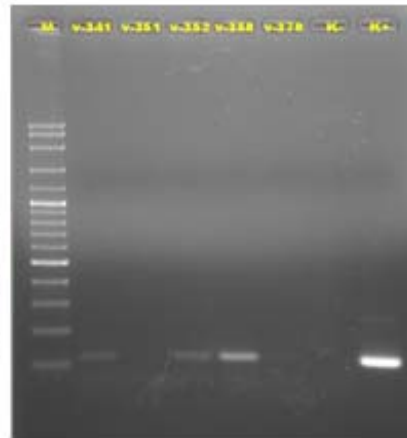
Molecular Analysis

Assessment	Protocol	Laboratories
Microscopy test	WHO, MoH & Eijkman Institute (Giemsa 3%)	<ul style="list-style-type: none"> Local Laboratory Parasitology Lab, University Gadjah Mada
DNA Isolation	Promega, Madison, USA. Cat. No. A-1123	Biochemistry Fac. of Medicine-UGM
Nested PCR <i>Plasmodium</i>	<ul style="list-style-type: none"> Promega, Madison, USA. Cat. No. M-7122. Primers: Snounou, 1993 	<ul style="list-style-type: none"> Parasitology Fac. Of Medicine-UGM MoH Parasitology Lab
PCR <i>G6PD</i> gene (Exon 5, 6, 9, 11 and 12)	<ul style="list-style-type: none"> Promega, Madison, USA. Cat. No. M-7122. Primers: Nguyen <i>et al.</i>, 2009 	<i>IVRCR&D, Ministry of Health, Salatiga, Central Java</i>
Sequencing product PCR <i>G6PD</i>	<ul style="list-style-type: none"> ABI Prism . 310 Genetic Sequencer. MEGA-5, Bio-Edit and Finch TV 	<i>Macrogen, Korea</i>
PCR-RFLP	KAPA HiFi, Boston, USA Cat. No.	Eijkman Institute for Molecular, Jakarta

Nested PCR Species *Plasmodium* Identification



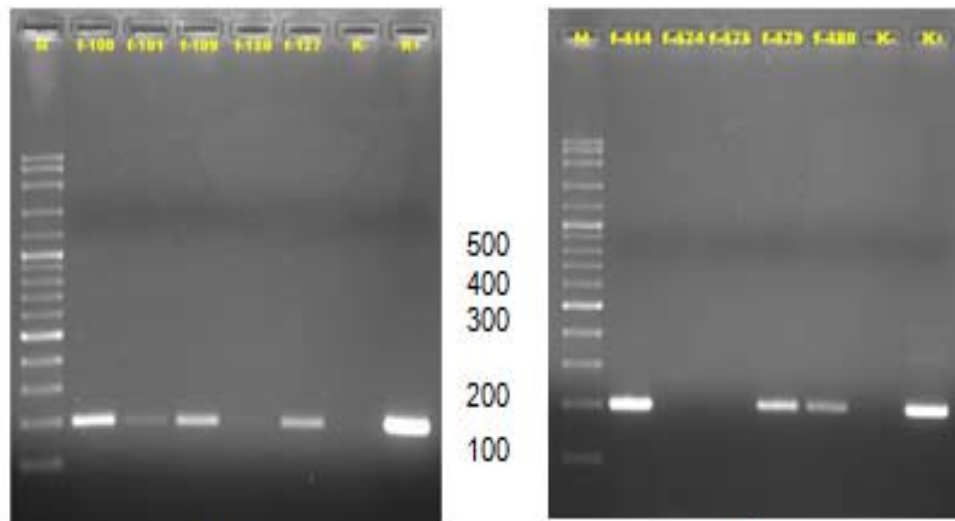
a



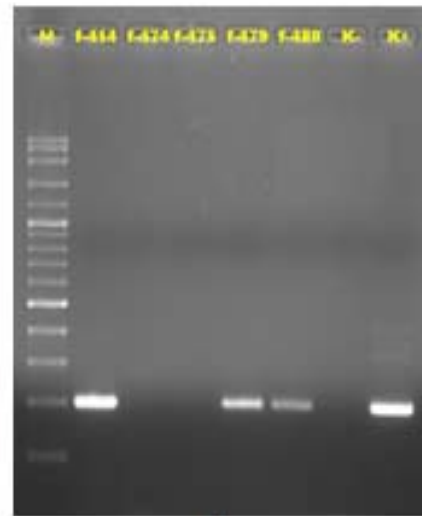
b



Figure 1. n-PCR *P. vivax* (120 bp), protocol by Promega, Madison, USA. Cat. No. M-7122, USA



a

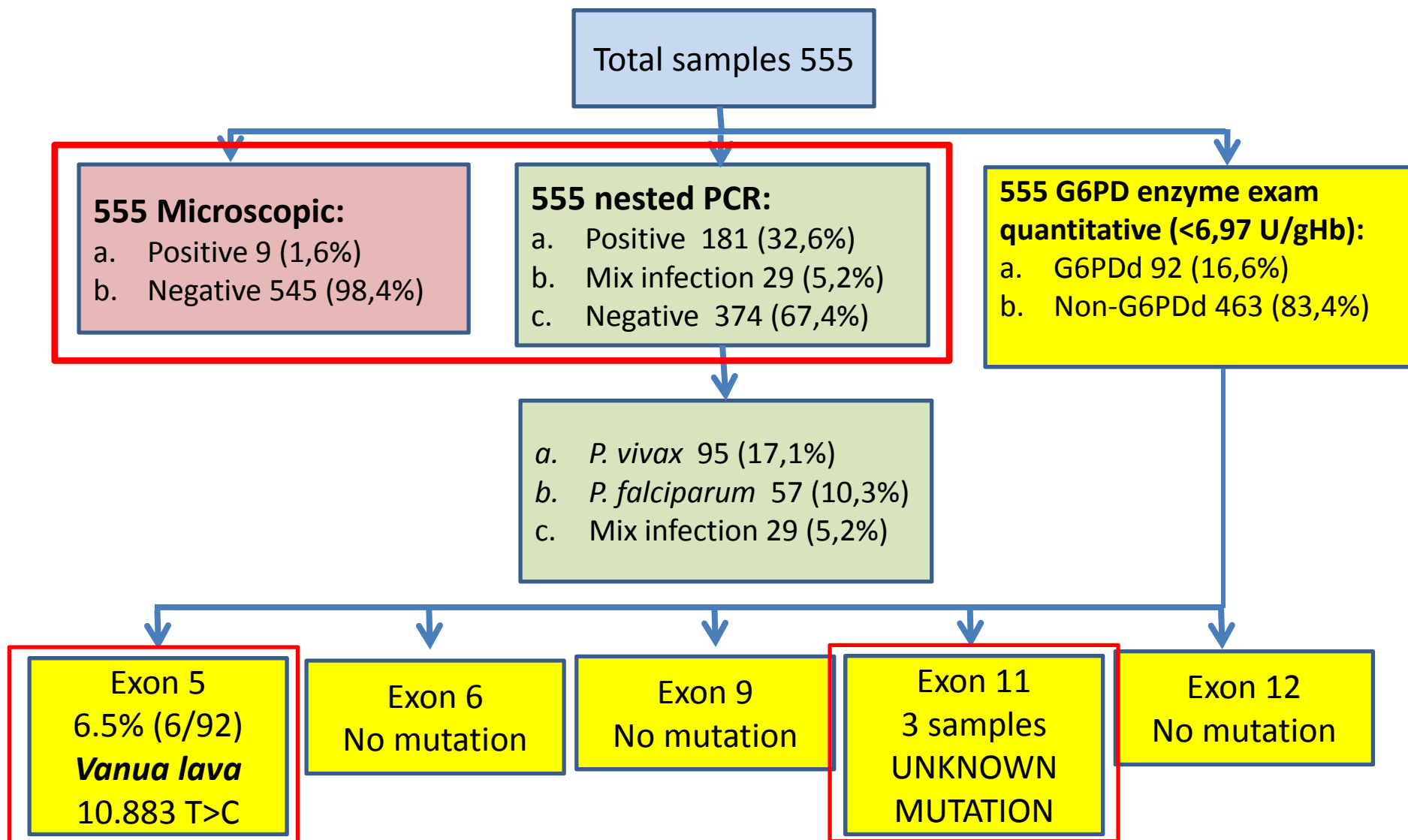


b



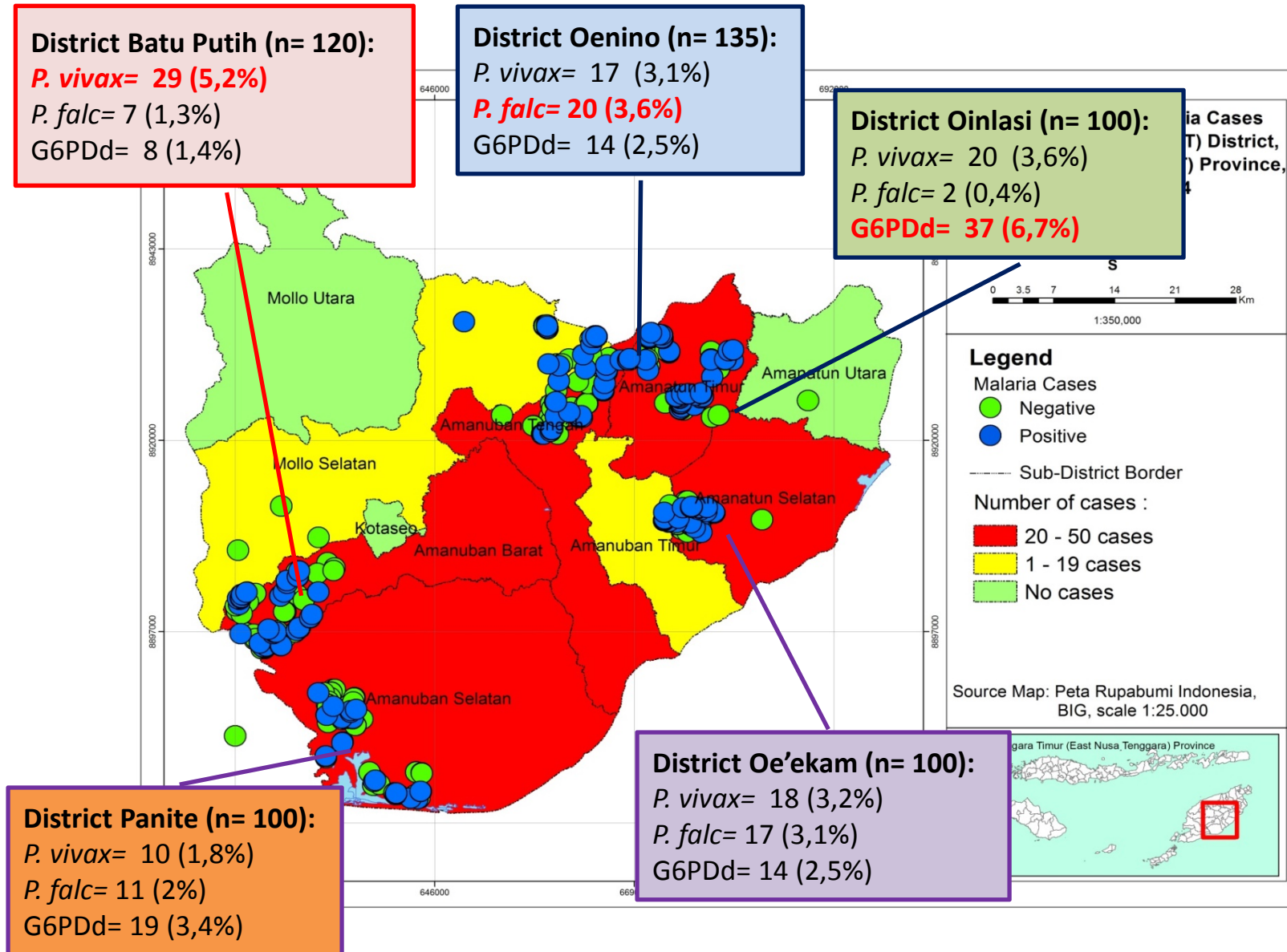
Figure 2. n-PCR *P. falciparum* (205 bp), protocol by Promega, Madison, USA. Cat. No. M-7122, USA

Profile result schematic work flow

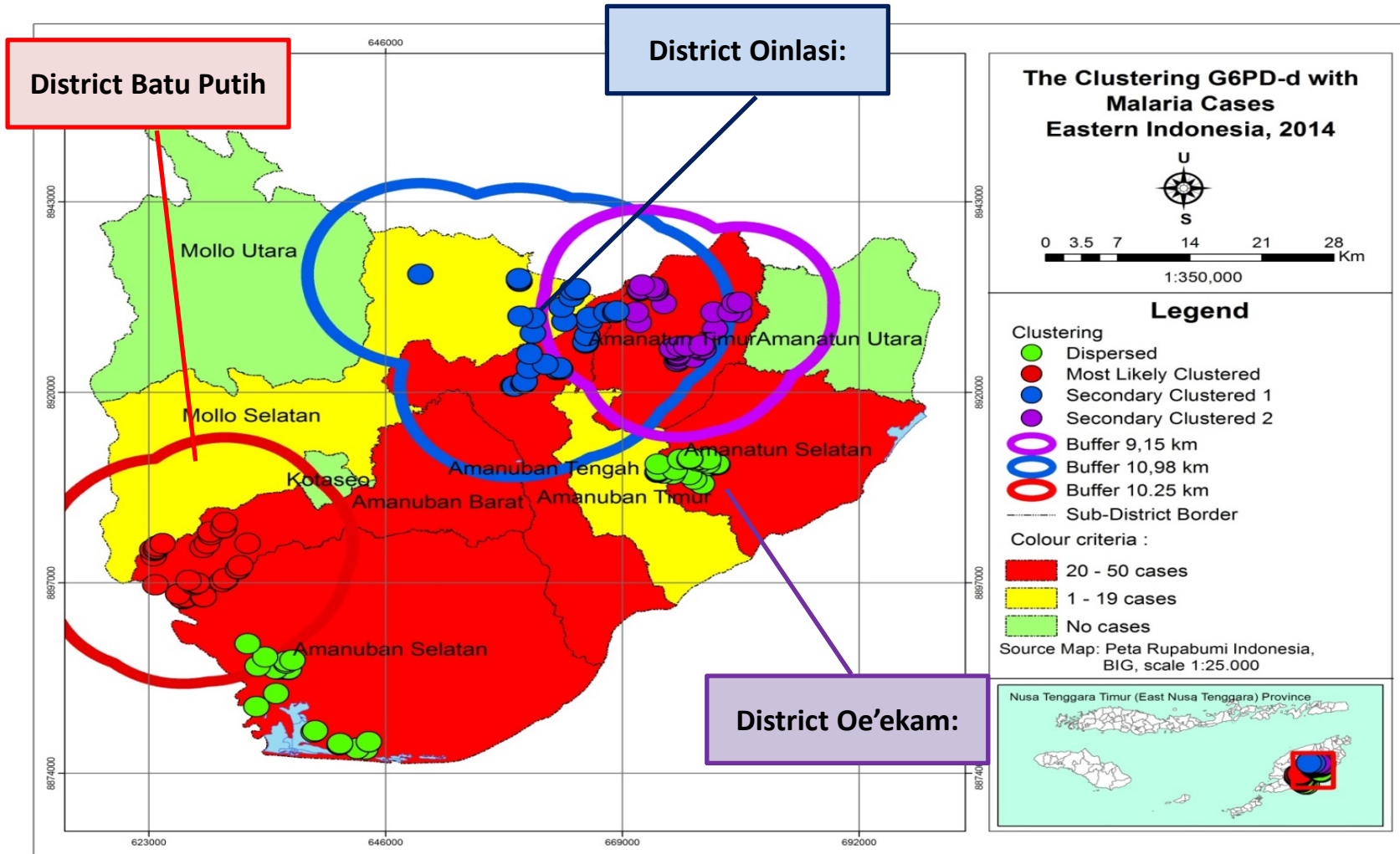


Reference sequence= Accession reference no. X-554481 (Minucci et al., 2012. Elsevier; Blood Cells, Molecules and Diseases)

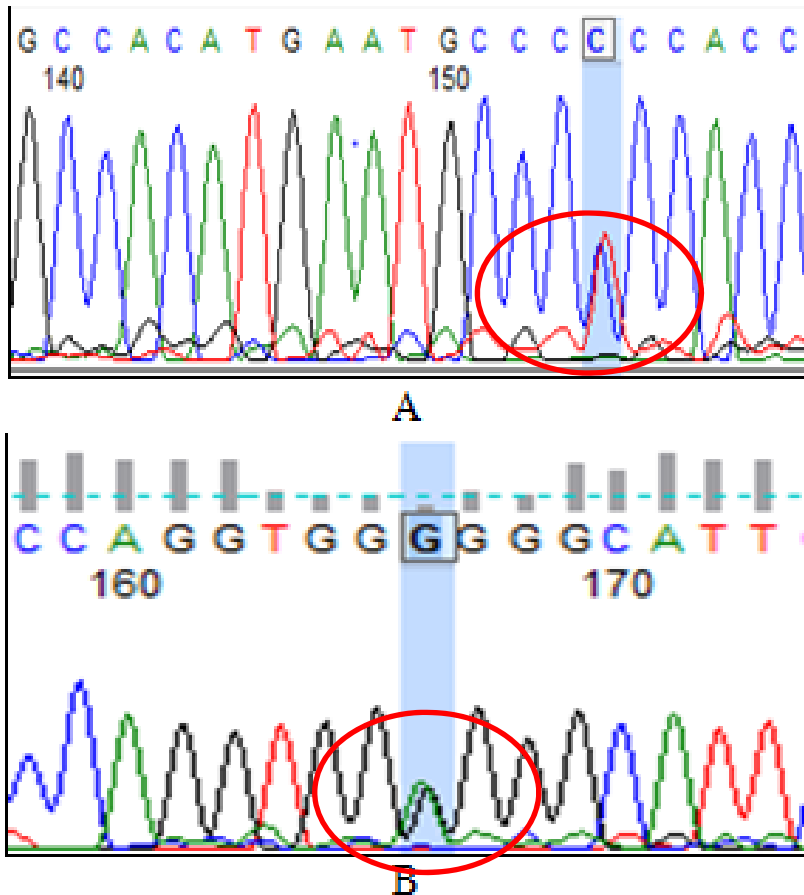
Five Study Sites (Nusa Tenggara Timur Province)



Three Clustering Malaria Cases (SatScan V.9.1.1, Skala 1: 25.000 with 10 km radius)



Sequence G6PD Variant Identification



- a) Age: 48 years/Female heterozygous.
- b) nPCR result: Negative malaria.
- c) Nucleotide substitution 10.884 C>T (Vanua Lava) at codon 128 exon-5.
- d) Amino Acid substitution Leu>Pro.
- e) 2nd Abortion history

Figure. (A) forwards primer, and (B) reverse primer.

Conclusions and Recommendations

1. Malaria *Pre-elimination* in eastern Indonesia *should be delay*, Other wise:
 - Routine treatment (silent transmission)
 - Outbreak control
 - Sensitive laboratories support
 - Surveillance (ACD and MBS)
2. *P. Vivax* dominant (*relapsing and new transmitted concern*)
3. Increasing G6PDd mutation 16.4% → BE AWARE PRIMAQUINE *Therapy*



All study respondent



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(Eijkman Institute)**

Smile From Eastern Indonesia

