Bioprospecting for antimalarials from medicinal plants

Jane Namukube
Why bioprospect for antimalarials

- Malaria is one of the most devastating diseases of the developing world
- Kills 1–3 million people annually, mostly affecting the children and pregnant women\(^1\)
- In every 30 seconds, a child dies of malaria
- In Uganda, malaria accounts 30–50% of outpatient visits
- 35% of hospital admissions
- 9–14% of hospital deaths\(^2\)

\(^1\) WHO, 2012; \(^2\) Yeka et al. 2012
Malaria distribution - affect many tropical and subtropical regions of the world\(^3\)

3. Gething et al., 2010
Plants as sources of anti-malarials

- Antimalarial compounds have been isolated from plants & developed into drugs e.g.

- Quinine isolated from Cinchona bark in 1820

• Artemisinine isolated from *Artemisia annua*

**Artemisinin Derivatives**

![Artemisinine and Derivatives](image)

- $R_1 = H, R_2 = OH$
- $R_1 = H, R_2 = OMe$
- $R_1 = H, R_2 = OEt$
- $R_1 = H, R_2 = OCH_3C_6H_5CO_2H$
- $R_2 = H, R_1 = O_2CCH_2CH_2CO_2H$

- Dihydroartemisinin
- Artemether
- Arteether
- Artelinic acid
- Artesunic acid

9. Meshnick et al., 1996
Standardised herbal medicines for treatment of malaria

- The use of many validated anti-malarial phytomedicines formulated from traditional medicines have been reported in recent years.\(^{10}\)
- Their use lifts the burden of drug pressure for development of resistance.
- Act as alternatives where there is unavailability of the recommended anti-malarial.

**Government approved**

- *Argemone mexicana* (Mali),
- *Artemisia annua* (China)
- *Cinchona bark*

10. Willcox, 2011
**Malaria treatment**

- WHO recommends artemisinin-based combination therapy (ACT), as the first-line treatment for malaria.
- Artemisinin-lumenfatrine (*Coartem*)
- Non artemisinin combinations like- atovaquone–proguanil (*Malarone™*).
- Sulphadoxine–pyrimethamine (*Fansidar™*).
- It is believed that combining two medicines with different mechanisms of action lowers the probability that a resistant parasite will emerge.
Prevention: Malaria vaccine

• There are control measures such as vector control, insecticide-treated bed-nets and anti malarial therapy.

• A vaccine esp to infants and pregnant mothers would greatly contribute on the malaria prevention. Unfortunately, there is no vaccine available yet.

• The most advanced malaria vaccine candidate coded as “RTS, S,” has reached phase III clinical trials\textsuperscript{10,11}

• GSK has applied for approval of RTS, S vaccine from the European medicine Agency.\textsuperscript{[www.reuters.com:July 24 ,2014]}

\textsuperscript{10}Casares et al., 2010; \textsuperscript{11}Moorthy et al., 2013; \textsuperscript{12}Aide et al., 2011.
Challenges of malaria treatment

• In Uganda, a proper treatment of malaria can be estimated at 10-15 USD & not affordable to most people

• Parasite resistance to the cheap and available antimalarials (Chloroquine, mefloquine)

• Recently resistance to artemisinins has been reported\(^\text{13}\)

• The need to search for more antimalarials is of utmost importance

The problem?

• Though plants are being used in medicine, Most of them are not documented.

• Efficacy, safety and active compounds in some of the plants are not known

• This limits their wider use, standardization and development into drugs/medicines
OBJECTIVES

• Document the indigenous knowledge on the use of the medicinal plants- Creating a basis for phytochemical investigation

• Extract and screen for antiplasmodial activity

• Isolate & characterize the active compounds which can be lead compounds to discovery of new drugs.

• Markers for herbal preparations
Materials and methods:

**Ethnobotanical survey**

- Efficacy – Antiplasmodial activity

- determination the chemical structures; chromatographic and spectroscopic means.

15. Tabuti, 2008
Materials and methods: Ethnobotanical survey: study site (Kibale National Park)
Isolation and Purification

fractions

Active fractions

PTLC
Prep HPLC
SFC

In vitro activity on Plasmodium

Cytotoxicity

Pure compounds

14. Smilkstein et al., 2004; 15. Desjardins et al., 1979
Materials and Methods

• 1D and 2D-NMR spectra recorded on Bruker 300-600 MHz spectrometer

• $^1$HNMR & $^{13}$CNMR
• DEPT- $^{\text{CH}_2}$, $^{\text{CH}_3}$,CH
• HSQC- C-H ↓
• COSY- H-H
• HMBC- H—C—C—C—
Results and discussions: Ethnobotanical survey

- Data Organized in table form: Plant name, local name, diseases treated, plant part; mode of preparation and administration;

- **131** plant species belonging to **121** genera and **55** families were documented to treat several illnesses.

<table>
<thead>
<tr>
<th>Medicinal plants</th>
<th>Disease</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Vernonia amygdalina</em> (Kibirizi, omululuza, Olubirizi)</td>
<td>Malaria</td>
</tr>
<tr>
<td><em>Albizia coriaria</em> (Omusisa, Mugavu, Omusita)</td>
<td>Cough</td>
</tr>
<tr>
<td><em>Neoboutonia macrocalyx</em> (Ekihora, Omweganza)</td>
<td>malaria</td>
</tr>
</tbody>
</table>
1. Neomacrolactone

\[ \text{IC}_{50} = 1.1 \mu g/ml \]

2. 22α-acetoxyneomacrolactone

\[ \text{IC}_{50} = 1.4 \mu g/ml \]

Criteria for in vitro anti plasmodial activity

- \( \text{IC}_{50} < 10 \mu g/mL \), good activity;
- \( \text{IC}_{50} \) of 10-50 µg/mL, moderate activity;
- \( \text{IC}_{50} \) of 50-100 µg/mL, low activity;
- \( \text{IC}_{50} > 100 \mu g/mL \), inactive

7. Neonthrene

IC$_{50}$ = 1.0 µg/ml

8. Neomacroin

IC$_{50}$ = 1.7 µg/ml

9. Neomacrotriol

10. Di-(2'-ethylhexyl) phthalate

11. 7-methoxy-2, 8-dimethyl-9, 10-dihydrophenantherene-3, 6 diol
7. Neonthrene

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Conclusion and recommendations

- Traditional medicinal plants contribute significantly in the treatment of several diseases esp malaria in this area – need for conservation sensitisation - (medicinal plant gardens)

- These new compounds could serve as leads to the development of new drugs for malaria; need to look at their synergy with available antimalarial.

- Compounds – marker in formulation of a herbal preparation from this plant

- The identification of antimalarial chemicals from *N. Neoboutonia* suggests that these compounds may play a role in the medicinal properties of this plant and therefore its use as a medicinal plant is supported.
Acknowledgement
Thank you