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Title:

Nigerian propolis;

An immune booster with anti-sickling properties

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RESEARCH CARRIED OUT IN

- *University of Lagos, Dept of Pharmaceutical Chemistry,
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And in collaboration with;

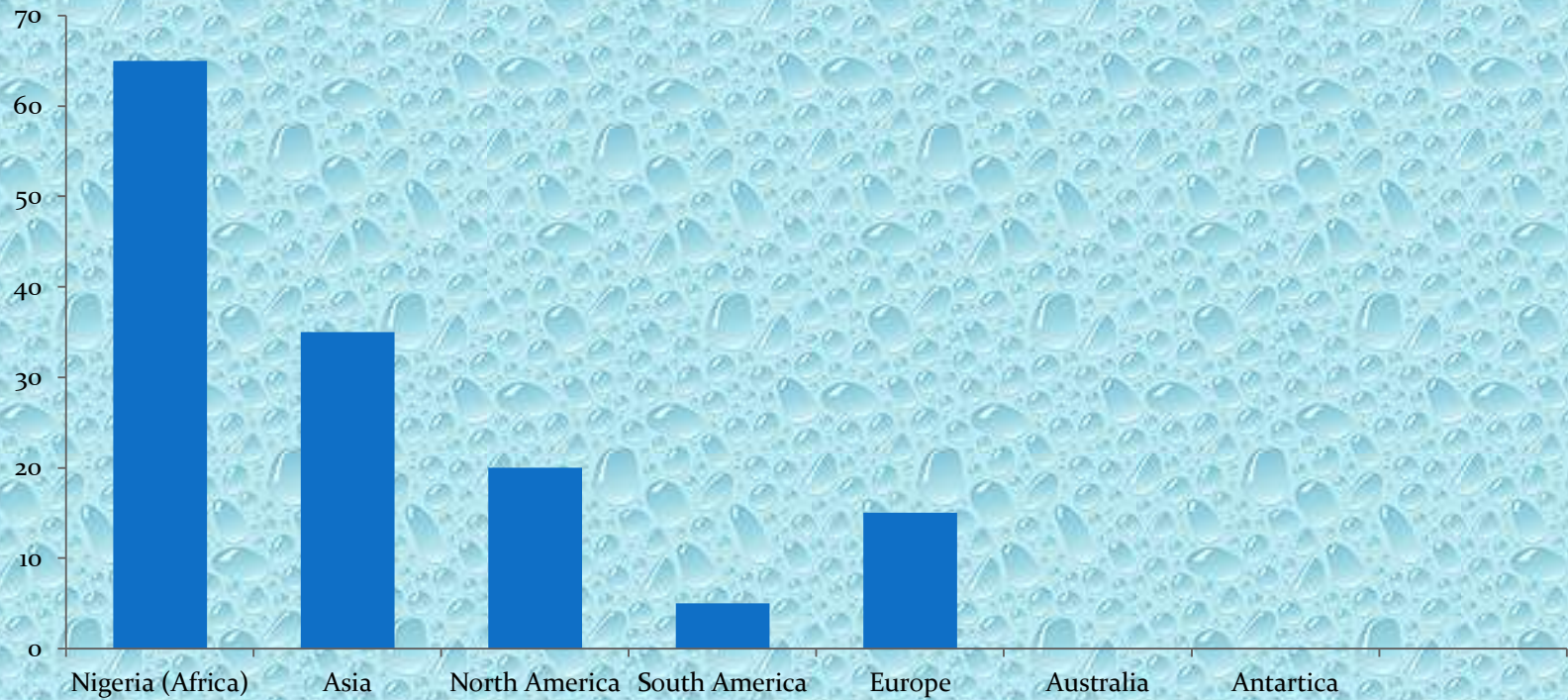
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INTRODUCTION

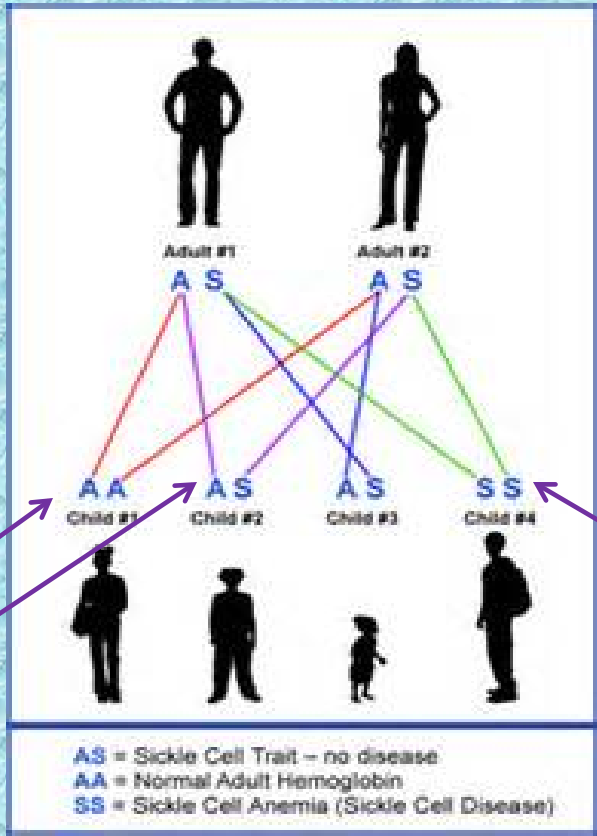
Sickle cell disease (SCD) also known as **drepanocytosis** is a disease that has become rather convincing to the medical doctors and scientists that there is yet no orthodox or herbal medication capable of providing total therapeutic cure for SCD in humans.

Graph comparing Nigeria and other countries.



➤ The body and blood

- HbAA } normal Hb
- HbAS } normal Hb
- HbSS } abnormal Hb
- HbSC } abnormal Hb
- HbS/β⁺ } abnormal Hb
- HbS/β^o } abnormal Hb



VAL-HIS-LEU-THR-PRO-GLU-LYS-SER-ALA

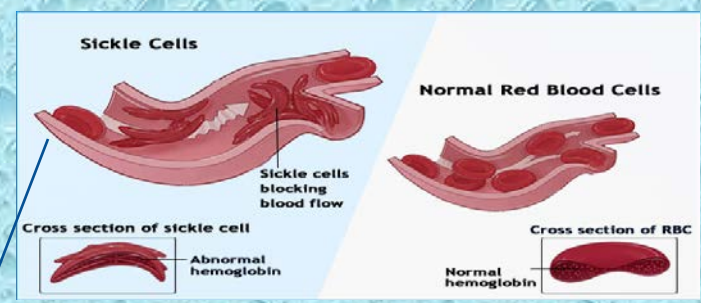


VAL-HIS-LEU-THR-PRO-VAL-LYS-SER-ALA



Origin: The inheritance of sickle hemoglobin gives rise to the pathological characteristics.

➤ SCD is the first genetic disorder to be characterized at the molecular level



Clinical manifestations

Constitutional

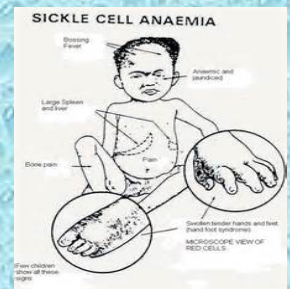
Haematological

vaso-occlusion

- impaired growth/dev
- increased risk of infection
- Meningitis
- osteomyelitis
- pneumonia
- skin ulcers
- among others

- haemolytic anaemia
- aplastic crises
- splenic sequestration

- cardiovascular complication
- gastrointestinal
- genitourinary
- neurologic, ocular



Factors that induce and favor sickle cell formation can be used to deduce some therapies for the management of SCD.

- Deoxygenation of HbS molecule
- Increased percentage of HbS in the red blood cell as a result of cell dehydration
- Low pH and increased temperature

Various managements of SCD

- Good hygiene practice / good ventilation
- Dietary and nutrients
- Bone marrow transplantation
- Gene replacement therapy
- Orthodox (anti-sickling agents/drugs)
 - Turning of genes that turn on HbF
- Herbal remedies

Erythropoietin
Butyrate
Clotrimazole
Hydroxyurea
Flucor
Clofibrate analogues
Niprasan
Cyclavit
To mention but a few

Side effects manifested are:

- Toxicity
- Addiction
- Alloimmunization
- Other complications

Dietary and Nutrients

- Good feeding habits and Rich nutrients that can be utilized for managing SCD.
- Food rich in anti-oxidants (vitamins and minerals) such as fruits and vegetables.
- Parsley (diuretic)
- Red clover (can serve as lung/blood tonic and blood cleansers
- Alfalfa
- Echinacea (king of blood purifiers)
- Garlics , gingers
- To mention but a few

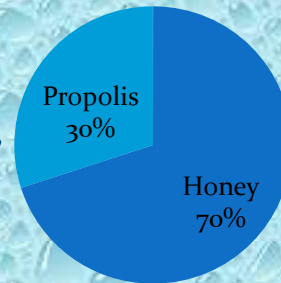
➤ HERBAL MEDICINE

- Management of disease conditions with the use of herbal medicine has gained prominence and generating success world wide (globally) with Asian countries leading in this aspect
- Natural products (NP) of diverse sources such as terrestrial plants, marine/aquatic ecosystems, animal /insects products continue to provide veritable sources of bioactive compound, crude or purified which have been used and are still been used for the management of different disease conditions , SCD inclusive.
- NP claimed to have potential anti-sickling activities include;
 - Cissus populnea L CPK
 - Xanthoxylum xanthoxyloids (Fagara)
 - cajanus cajan (cyclavit)
 - Niprasan

Propolis (Bee product)

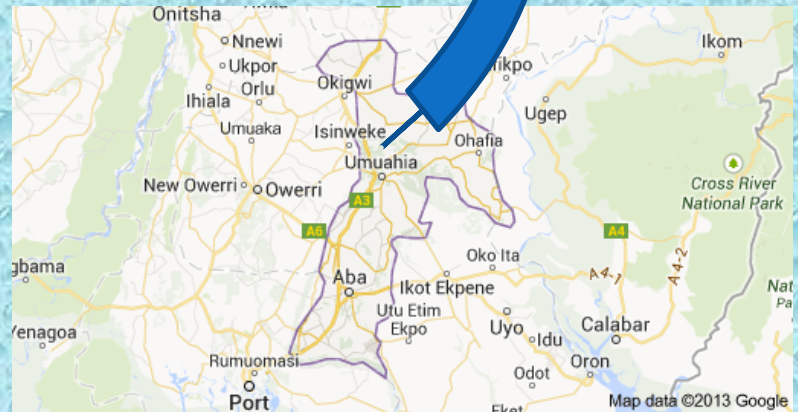


Nigerian propolis



Crude honey

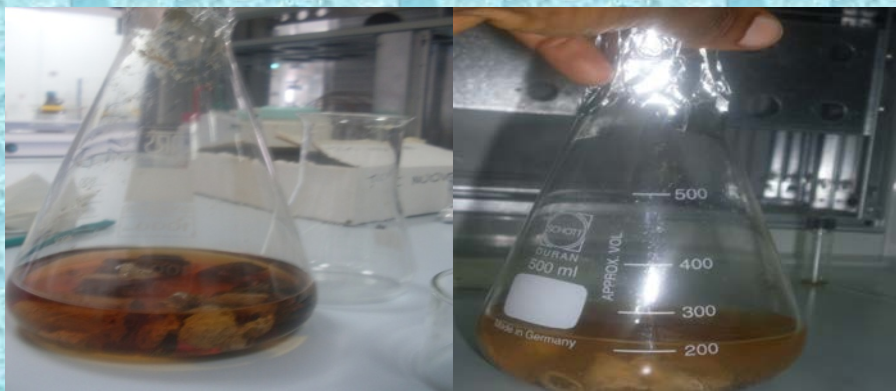
- **Propolis** has a long history of medicinal use, dating back to 350 B.C., the time of Aristotle.
- Propolis are;
 - Natural antibiotics
 - Natural anti-oxidants
 - Natural immune boosters, etc



Methodology

Extraction protocol

Aim of study: To extract the phytochemicals in propolis using the best solvent system.



Propolis soaked in methanol for 72 hrs



Isolation of crude methanolic extract of NG Propolis using sephadex LH-20 on column

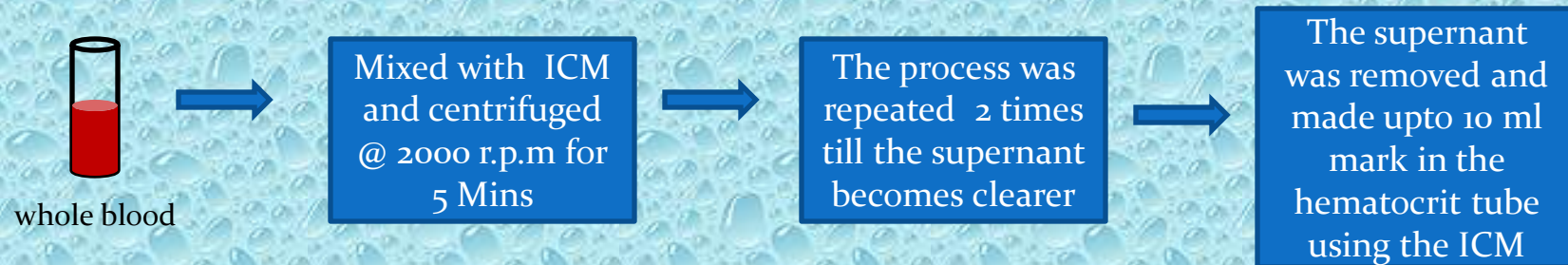
Anti-sickling (*in vitro* assay)

Inhibition assay

Aim: To evaluate the inhibitory properties of methanolic extracts of propolis against sodium metabisulphite induced sickle cell.

Reagents/chemicals/ apparatus used: Incubation medium(ICM)Tris and normal saline, centrifuge, micropipette, hematocrit tubes, HbSS blood, propolis extract ($1-5\text{mgml}^{-1}$), microslides, cover slips, paraben, paraffin wax, microscope, timer

Procedure 1: preparation of 10 % erythrocytes suspension



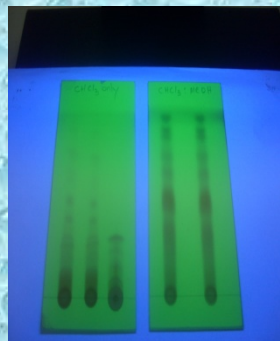
Procedure 2 : *In-vitro* inhibition assay



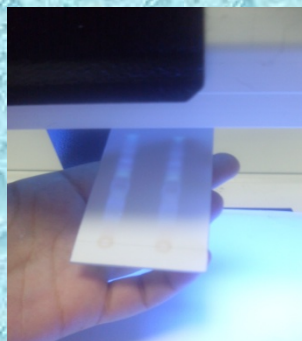
Findings – Inhibitory assay results

S/N	Extract Concentration	Total no of cells	Number of sickled cell at time		% tage sickled cell
			0min No of sickled cell at time 0mins	20 mins	
1.	methyl paraben (+ ve control)	602	-	5	0.83%
2.	sodium metabisulphite (- ve control)	400	-	240	60%
3.	5mg/ml	1000	-	4	0.4%

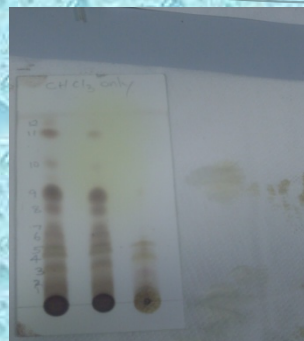
Findings - phytochemical assay



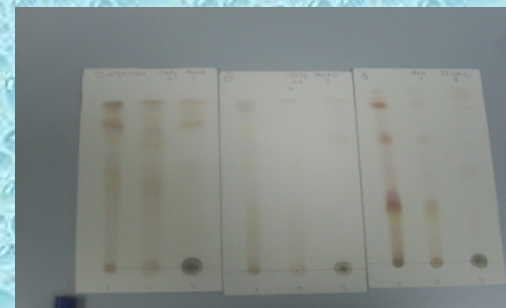
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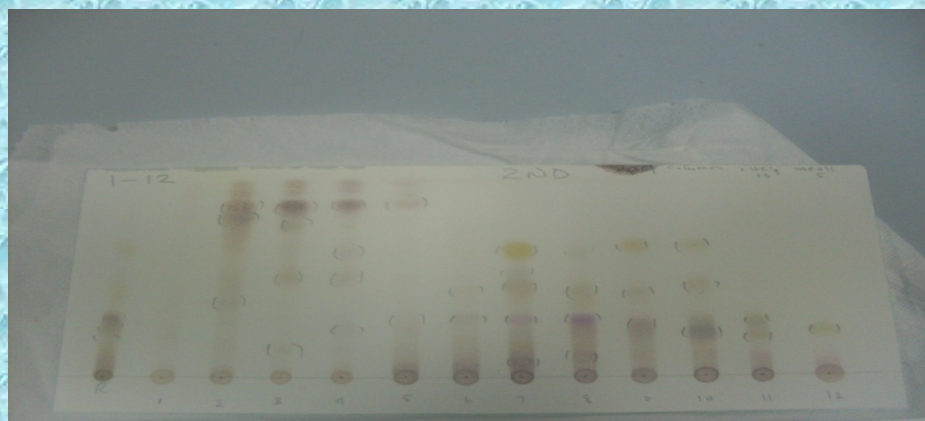
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iii



iv



v

(i - iv): TLC stripes showing spots of phytochemicals contained in Nigerian propolis
(v) : Fractions 1- 12 of NG pro separated using sephadex LH-20

Acknowledgements

- Mr Michael Ukattah of Amachi Farms, Ikwuano, Umuahia for providing the propolis

Useful References

- Bunn, HF; Forget, BG: Haemoglobin: molecular, genetic and clinical aspects; Philadelphia: WB Saunders, 1986
- Eaton, W.A., and Hofrichter, J (1990). Sick cell haemoglobin polymerisation[review].Advances in protein Chemistry 40,63-279
- Robert, F.W., and Philip, W.H., (1997). Genetics. WCB,Mcgraw – Hill Companies I.
- Ingram ,V.M. (1956). A specific chemical difference between globins of normal and sickle cell anemia haemoglobin. Nature 178,792 – 794



*Thank
you*

