Annex 1. Prepared documents and materials for the NEGAT science camp

Safety protocol: It outlines the safety requirements needed in the laboratory. It ensures that everyone in the lab is aware of the hazards, risks, and protective measures associated with laboratory procedures.

Laboratory protocols: The laboratory protocols were developed for extracting essential oils from medicinal plants and assessing the antibacterial activity of plant extracts. These protocols describe the required materials and the specific procedures of the activities.

Probe questions: Pre-prepared questions for students to ask scientists during a question and answer session.

Questionnaires: Pre and post-questionnaires were developed to evaluate summer camp participants' attitudes towards science and test their knowledge.

Notebook: The notebook provides information about CDT Africa, NEGAT, and lists of websites that will help participants engage in scientific research in the future.

Photo/ Video release form: This is a consent paper for parents that allow CDT Africa to take photos and videos of their children and use them for promotional, educational, and advertising purposes.

Drop-off and Pick-up checklist: to record participants' attendance throughout the camping.

T-shirt: T-shirts bearing the motto "I'm Tomorrow's Scientist" along with their assigned plant names (አዝመሪኖ፣ ጤና አዳምና ዳማከሴ ቡድን) were prepared and worn by participants on the closing day.

Game: A magnetic dartboard and a collection of educational games were provided.

Annex 2. Pre and post evaluation questionnaires

Attitude test towards science

Please indicate the degree of your agreement and disagreement with each statement

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Africa can discover new drugs for global health challenges.		\cup			\cup	\cup

Knowledge test

ID______ Date____

TEST YOUR KNOWLEDGE

- 1. What percentage of the global biodiversity is found in Africa?
- A. 25%
- B. 5%
- C. 2%

- D. 10%
- 2. What percentage of Africa's essential medicines is imported?
- A. 94%
- B. 74%
- C. 54%

- D. 24%
- 3. What percentage of deaths in Africa is due to lack of essential drugs?
- A. 60%
- B. 80%
- C. 90%

- D. 50%
- 4. Most bacteria are harmful.
- A. True
- B. False

- 5. What can you do to help Africa make its own medicines?
- A. I will become a scientist that can make medicines
- B. I will build industries
- C. I will become a rich man and invest in making medicines
- D. I will help conserve Africa's biodiversity
- E. I will support initiatives like CDT-Africa that create scientists
- F. I will become a politician/a minister and make decisions to help scientists and industries

Annex 3. Safety and laboratory protocols

3.1. Safety protocol manual

- O Wear clothing, which does not leave large areas of skin exposed. Do not wear open toed shoes or sandals.
- O Always wear protective safety gloves, goggles, and lab coat in the lab.
- o Remove loose jewelry chains or bracelets while doing lab work.
- o Tie back long hair and loose clothing to keep them away from flames and equipment.
- o Read **all instructions** in the manuals before you begin a laboratory activity
- O **Ask** questions if you do not understand any part of the activity.
- Lab coats and gloves should not be worn outside the lab except your lab supervisors told you to do so.
- O Keep your hands away from your face while working in the laboratory.
- O Learn the location of the fire extinguisher, eyewash station and first aid kit.
- O Do not eat or drink while in the lab or store food in lab equipment or the lab refrigerator.
- O Work **ONLY** on activities assigned by your lab supervisor.
- O **Do not begin any activity until** directed to do so by your lab supervisor.
- O Do not handle any equipment without specific permission.
- O Do not take any materials or chemicals out of the laboratory.
- O NEVER work alone in laboratory.
- O **Do not substitute** other chemicals/substances for those listed in your activity.
- O Do not taste or smell or touch chemicals.
- O Wash your hands thoroughly with soap and water after each activity.
- O In case of any emergencies, immediately inform to your lab supervisor.

Annex 3.2 Protocol for essential oil extraction from medicinal plants

Objective: To extract essential oil from plants (Tenadam, Damakese, and Rosemery) to test their antibacterial effect.

Method used for extraction is Hydrodistillation. Blended plant is mixed with water and boiled to evaporate volatile components.

Safety warnings:

- O Always wear protective safety gloves, goggles, and lab coat in the lab.
- O Remove loose jewelry chains or bracelets while doing lab work.
- o Tie back long hair and loose clothing to keep them away from flames and equipment.
- o Read all instructions in the manuals before you begin a laboratory activity.
- O Ask questions if you do not understand any part of the activity.
- O Keep your hands away from your face while working in the laboratory.
- O Work ONLY on activities assigned by your lab supervisor.
- O Do not begin any activity until directed to do so by your lab supervisor.
- O Do not handle any equipment without specific permission.
- O Do not take any materials or chemicals out of the laboratory.
- O Wash your hands thoroughly with soap and water after each activity.
- O In case of any emergencies, immediately inform to your lab supervisor.

Materials and Equipment

- 1. Plant Leaves: Tena adam (Group 1), Damakese (Group 2) or Rosemary (Group 3)
- Distilled water
- 3. Clevenger's apparatus
- 4. Oil collector tube
- 5. Balancing Scale
- 6. Heating mantle
- 7. Blender/food processor

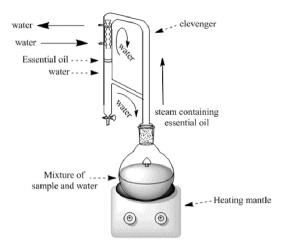
Procedures

Day 1: Mon Aug 14, 2023

- 1. Cut leaves of your assigned plant.
- 2. Wash the leaves with tap water.
- 3. Blend the leaves using food processer.
- 4. Weigh the blended plant material using balancing scale.
 - a. Weight=____g
- 5. Place the blended plant material in the fridge until it can be used the next day for extraction.

Day 2: Tue Aug 15, 2023

- 6. Take out the blended plant from the fridge.
- 7. Put the blended plant in the flask and add distilled water until it covers the blended plant.
- 8. Assemble the Clevenger's apparatus as indicated in the picture.



9. Boil the mixture for about 3 hours at 90° c using a heating mantle.

Note: do not stand too close to the Clevenger's apparatus during this step.

- 10. Collect the essential oil using oil collector tube.
- 11. Measure the volume of essential oil collected.
 - a. Volume= ml
- 12. Calculate yield
 - a. Yield = _____
- 13. Store the oil in fridge until use.

Annex 3.3. Protocol to test antibacterial activity of plant extracts

Materials:

- Test tubes
- Beaker
- Hot plate
- Sensitive balance
- Spectrophotometer
- Measuring cylinder
- Spatula
- Micropipette
- Sterile swab
- Cuvette
- Sterile Pipette tips
- Test tubes
- Mueller Hinton broth powder
- Distilled water
- Ampicillin (0.1g/ml stock solution)

Procedures

Tuesday August 15, 2023: Seed culture preparation

- 1. Prepare broth Muller Hinton medium by adding 7.35 g of Muller Hinton powder in to 350 ml of distilled water.
- 2. Boil and stir the mixture to fully dissolve the medium.
- 3. Let it cool and Divide the medium in to 10 test tubes (10 ml to each test tube).
 - a. Label 9 test tubes with each student's initial.
 - b. Label 1 test tube as negative control.
- 4. Divide the rest medium to 24 test tubes for each group (3ml to each test tube).
- 5. Sterilize by autoclaving at 121°C for 15 minutes.

- 6. Take oral swab sample from your mouth and inoculate to your assigned test tubes.
- 7. Leave one of the test tubes without inoculating as a negative control.
- 8. Put the rest test tubes at 4 °C for the next experiment.
- 9. Incubate the test tubes for 24h at 37 °C.

Wednesday August 16, 2023: Test antibacterial activity of your plant extract

- 1. Each group should prepare 24 test tubes. For the three groups we would need 72 test tubes.
 - a. Tena adam group (24 test tubes).
 - i. Untreated Student 1's bacteria (2 test tubes).
 - ii. Tena Adam treated Student 1's bacteria (2 test tubes).
 - iii. Ampicillin treated Student 1's bacteria (2 test tubes).
 - iv. Untreated Student 2's bacteria (2 test tubes).
 - v. Tena Adam treated Student 2's bacteria (2 test tubes).
 - vi. Ampicillin treated Student 2's bacteria (2 test tubes).
 - vii. Untreated Student 3's bacteria (2 test tubes).
 - viii. Tena Adam treated Student 3's bacteria (2 test tubes).
 - ix. Ampicillin treated Student 3's bacteria (2 test tubes).
 - x. Control Tena Adam treated media only (No bacteria) negative control (2 test tubes).
 - xi. Control ampicillin treated media only (No bacteria) (2 test tubes).
 - xii. Control untreated media only (No bacteria) (2 test tubes).
 - b. Damakese group (24 test tubes)
 - i. Untreated Student 1's bacteria (2 test tubes).
 - ii. Damakese treated Student 1's bacteria (2 test tubes).
 - iii. Ampicillin treated Student 1's bacteria (2 test tubes).
 - iv. Untreated Student 2's bacteria (2 test tubes).
 - v. Damakese treated Student 2's bacteria (2 test tubes).
 - vi. Ampicillin treated Student 2's bacteria (2 test tubes).
 - vii. Untreated Student 3's bacteria (2 test tubes).
 - viii. Damakese treated Student 3's bacteria (2 test tubes).

- ix. Ampicillin treated Student 3's bacteria (2 test tubes).
- x. Control Damakese treated media only (No bacteria) negative control (2 test tubes).
- xi. Control ampicillin treated media only (No bacteria) (2 test tubes).
- xii. Control untreated media only (No bacteria) (2 test tubes).
- c. Rosemary group (24 test tubes)
 - i. Untreated Student 1's bacteria (2 test tubes).
 - ii. Rosemary treated Student 1's bacteria (2 test tubes).
 - iii. Ampicillin treated Student 1's bacteria (2 test tubes).
 - iv. Untreated Student 2's bacteria (2 test tubes).
 - v. Rosemary treated Student 2's bacteria (2 test tubes).
 - vi. Ampicillin treated Student 2's bacteria (2 test tubes).
 - vii. Untreated Student 3's bacteria (2 test tubes).
 - viii. Rosemary treated Student 3's bacteria (2 test tubes).
 - ix. Ampicillin treated Student 3's bacteria (2 test tubes).
 - x. Control Rosemary treated media only (No bacteria) negative control (2 test tubes).
 - xi. Control ampicillin treated media only (No bacteria) (2 test tubes).
 - xii. Control untreated media only (No bacteria) (2 test tubes).
- 2. Take test tubes with 3 ml Muller Hinton medium from the refrigerator and incubate it for 10 min at 37 °C.
- 3. Add 30 µl of overnight seed culture to each of your test tubes.
- 4. Add 60 μl of the essential oils (tena adam, damakese, or rosemary) (Equally diluted with DMSO solution) on their respective test tubes.
- 5. Add 30 μl of Ampicillin solution.
- 6. Incubate the culture for 24h in 37 °C.

Thursday August 17, 2023: Measure the antibacterial activity of your plant extracts

- 1. After 24 hr, take out the tubes from the incubator.
- 2. Put the culture to cuvettes and measure using spectrophotometer at 600 nm.
- 3. Blank the optical density of the negative controls (treated and untreated controls).
- 4. Using spectrophotometer, read the optical density for the untreated bacteria, essential oil treated bacteria, and ampicillin treated bacteria.
- 5. Record the data.
- 6. Plot a graph for the drug concentration vs the optical density (OD).