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DAVIS EXPEDITION FUND REPORT

PAPUA NEW GUINEA ETHNOBOTANICAL SURVEY

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Abstract

Previously a highly portable antimicrobial assay was developed for the specific purpose of testing, whilst in the field, medicinal plants used by remote rainforest communities. During January and February of 2002 this assay kit was used to test a variety of traditional medicines used by several different communities living in the interior of New Britain, Papua New Guinea's largest island. Plants were collected that are used to treat bacterial infections such as tropical ulcers and conjunctivitis. Over 50 extracts made from traditional remedies were tested, 6 of which showed a clear antibacterial effect. The most potent extract was obtained from a species of insect whose defensive secretions sprayed from glands behind its head, are used by local people to treat tropical ulcers.

Project authorisation

Permission to carry out this project was granted by the National Research Institute of Papua New Guinea and the Provincial Government of West New Britain. A scientific research visa valid until 2002 was obtained from The Department of Foreign Affairs in Port Moresby. The project is affiliated to the Forest Research Institute of Papua New Guinea who are currently issuing export licences and helping with identification of plant species.

Location

Fieldwork was carried out at separate locations in the Whiteman and Nakanai Ranges of New Britain. Both are highly mountainous areas with rainfall exceeding 6m a year. The Nakanai area was selected as its inhabitants live at a higher elevation than anywhere else in New Britain and therefore may use different species in their traditional remedies. Of particular interest is the virtually unexplored Nakanai plateau which extends for 20km on each side and maintains an elevation of nearly 2000m.

Project aim

To search for plants with antimicrobial activity in the tropical forests of New Britain.



The Nakanai plateau as seen from the airstrip at Bili. The plateau is 20km along each side and maintains an altitude of nearly 2000m. It is apparently uninhabited. Forest in the foreground at 1000m was used for collecting plant and insect specimens.

Methods

Collecting a range of plants for testing

Lists of plant names used to treat key conditions associated with bacterial infection such as the phagedenic or tropical ulcer (caused by a combination of Spirochaete and Fusiform bacteria), bacterial conjunctivitis and respiratory tract infections were obtained by interviewing at least three different people from the same language group. For each plant the local name, part of the plant used, mode of preparation as well as the type of infection treated were recorded. Once an exhaustive list had been obtained, excursions were made into the surrounding forest and small representative samples of each plant were collected and prepared according to the ethnobotanical data.

Making extracts of the plants

Once a range of plant samples had been obtained, extracts were made by grinding each plant in a mortar and pestle with a small volume of 50% ethanol. This solvent ratio was selected as it is sufficiently rich in ethanol to keep the extracts fresh in such a warm climate but contains enough water to allow easy loading into the wells of the assay plate.

Testing the plants

The assay works by the simple well diffusion method where samples are injected into wells cut into agar on which a confluent lawn of bacteria is allowed to grow. A positive result is indicated by the presence of circular zones of inhibition around the well.

The assay kit is specially developed for testing samples in the field and consists of a purpose built lightweight pressure vessel used to sterilise glassware and vials of freeze-dried bacteria for making the cell cultures. Freeze-dried bacteria have the advantage that they can remain static for long periods of time at room temperature. *E. Coli* and *S. Epidermidus* are used, representing a Gram negative and Gram positive species respectively. When a fresh cell culture is required a single disk containing one of the two species is simply placed into sterile culture medium made using the pressure vessel and allowed to grow at ambient temperature (20°C- 30°C) which is normally sufficient to produce log phase cells within 15 hours.

The cell cultures are then used to seed sterile agar plates (made using the pressure vessel) which have 6mm diameter wells cut into them. Into each well is injected 50µl plant extract using a micro-pipette. The result is a sensitive antibacterial assay where cell cultures are pure despite the septic conditions of the surrounding forest.

A new inclusion to the testing equipment this year was an elicitor spray made with a 5g/L concentration of yeast extract which can be sprayed onto leaves to stimulate a defensive response by the plant. This may help to enrich the plant in secondary metabolites and other active molecules that might otherwise be at too low a concentration to cause a noticeable zone of inhibition.



Figure 1. Camp in the Whiteman Range. Experiments were carried out underneath the canvas.



Figure 2. The assay kit including specially designed pressure vessel as well as microbiological media, cultures and other equipment.



Figure 3. A species of polymorphic insect known locally as Kain-tagho which literally means, "eats Pandaunus sp." in the local dialect. Secretions sprayed from behind its head are used to treat tropical ulcers.

Further work

To back up these findings I intend to try and identify the active molecule(s) from the insect. A quantity of about 100 of the dead insects preserved in alcohol has been collected and using a combination of High Performance Liquid Chromatography (HPLC) and Nuclear Magnetic Resonance (NMR) it may be possible to identify the structure of the active principle(s) provided they are not very high molecular weight. It will first be necessary to identify the species name so any help or advice the Davis Fund Committee could offer would be greatly appreciated.