4 Neem seed oil: Bangladesh

General information

- **Implementing institution**
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- **Implementation period**
  July 2002 to June 2005
- **Costs**
  Total cost is approximately US$5,000 received from the BCSIR research and development fund.

Summary

The neem tree (*Azadirachta indica*) is found throughout the Indian subcontinent. For generations, its seeds, leaves and bark — and the oils extracted from these parts — have been used to treat a variety of ailments. Neem oils have also found uses in agriculture to control various insects and nematodes (parasitic worms). However, little is known about the make-up of neem oil and which parts have the most biological activity. This project was designed to analyse the fatty acids present in neem seed oil. To this end, novel extraction and analytical procedures were developed with a view to developing both more efficient purification processes and commercial products based on these components.

Background and justification

The neem tree, which grows throughout the Indian subcontinent, is one of the most important medicinal plants in Bangladesh. The evergreen tree is large, reaching 12 to 18 metres in height with a girth of up to 1.8 to 2.4 metres.

Despite almost every part of the tree having a bitter taste, neem has many diverse applications in indigenous medicine. Extracts have been used successfully to treat stomach worms, pyorrhoea (discharges of pus) and ulcers, and they have been reported to possess antidiabetic, antibacterial and antiviral properties.

The essential oil extracted from the leaves possesses marked antiseptic and antibacterial properties and, in laboratory tests, inhibits the growth of *Mycobacterium tuberculosis*, the causal agent of tuberculosis. The leaves are also used as an insect repellent.

The fatty oil extracted from neem seeds also has many therapeutic uses. It possesses antiseptic and antifungal activity and is found to be active against both gram-positive and gram-negative bacteria. Considerable quantities of the seed oil are employed in cosmetic preparations such as creams, soaps, tooth powders and toothpastes. Tender twigs are used to clean teeth.

Given the wide range of uses associated with extracts from the neem tree, there is a need to define which component oils are responsible for which biological activities. Only in this way can the indigenous uses of the neem tree be scientifically validated and commercial products developed that will conform to rigorous international standards.

In particular, the project aimed to:

- develop an efficient technique for extracting fixed and essential oils from neem seed so that maximum amounts of component fatty acids, including essential fatty acids, can be isolated;
- determine whether the fixed seed oil can be used as an edible oil by eliminating bitter-tasting and other components; and

- develop medicines and hair or body oils by incorporating neem-derived components into a benign base such as linseed oil, mustard oil, palmarosa oil, turmeric oil or a combination of these ingredients.

DESCRIPTION

Neem seeds (fig. 1), collected from April to August from mature trees growing on the BCSIR Dhaka campus, were thoroughly cleaned and carefully dried in the sun. Dried, hulled seeds were then ground and the resulting powder repeatedly extracted with petroleum ether at 40° to 60°C. After evaporating the solvent, a pale green oil with a pungent, but agreeable odour (equivalent to 38 per cent of the weight of the powder) was obtained. (In contrast, when leaves were extracted, a pale yellow oil with the characteristic odour of neem was obtained). The neem seed oil was then analysed for its different physical and chemical properties, including its melting point (23°C) and moisture content (4 per cent).

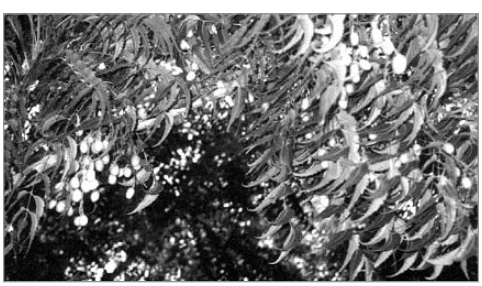

Figure 1 | Fruit of the neem tree.

Preparation of methyl esters

To analyse the fatty acid composition of neem seed oil, fatty acid esters were prepared by methyl esterification.

This was done by adding sodium hydroxide to the oil and boiling for 10 minutes. Boron trifluoride-methanol complex was then added and the mixture boiled for 2 minutes, followed by another minute after the addition of hexane. The hot mixture was then poured into a separating funnel with more hexane solvent plus water. After allowing the mixture to settle into layers, the hexane fraction was separated, dried over anhydrous sodium sulphate and filtered. Finally, the solvent was evaporated under reduced pressure to leave the purified methyl esters.

Gas liquid chromatography of methyl esters

The methylated neem seed oil was then subject to gas liquid chromatography to determine the fatty acid composition of the methyl esters. Samples were dissolved in chloroform and passed through a glycol succinate column maintained at between 230° and 250°C. Nitrogen was used as the carrier gas and the flow rate was adjusted to 30 millilitres per minute. A dual flame ionization detector was used to determine when the methyl esters exited from the column (i.e., their retention time), and internal and external standards were used to identify individual fatty acids. More than half of the oil was found to be oleic acid, with linoleic and palmitic acids making up the

bulk of the remainder (see table). Oleic acid has been shown to reduce levels of cholesterol in the blood and it is used in the food industry to make margarines and to flavour cakes, ice cream and soft drinks. Linoleic acid also has commercial potential as it is an essential component of the human diet.

Fatty acid composition of methyl esters from gas liquid chromatography analysis.

Components obtained	Fatty acid (per cent)	Retention time (seconds)
Palmitic acid	18.45	18.7
Stearic acid	0.20	19.4
Oleic acid	58.50	20.7
Linoleic acid	21.00	20.9
Arachidic acid	1.50	22.5

Patenting and commercialization

Considerable quantities of neem seed oil are employed in cosmetic preparations such as creams, soaps and toothpaste.

In particular, BCSIR has developed a neem toothpaste and a high-quality face cream based on crude neem seed oil. The neem toothpaste is currently undergoing trials to determine its shelf life.

Agreements based on these two products have been made with local entrepreneurs and they are both expected to be in commercial production in the near future.

Replicability

Based on such successful commercial ventures as the neem-based cream and toothpaste developed in Bangladesh, it has been estimated that, by harvesting and selling various parts of the tree, a farmer could earn up to US$1,500 a year from a single tree. Neem, therefore, has the potential to improve the income of even the poorest people in any developing nation where the species grows well.

Policy implications

It is now clear that a number of other indigenous plants, particularly those that bear fruit, should undergo a systematic process of investigation and utilization as

a means of furthering economic growth in Bangladesh.

Impact

Different categories of people, from scientists and technologists to policy-makers and end-users, are now aware that the indigenous flora of Bangladesh provides an as-yet-untapped — and poorly investigated — reservoir of chemical compounds that could be developed into commercial products.

Future plans

To further characterize the composition of neem oil from seeds, leaves and bark, analyses are being performed using thin layer chromatography; infrared, ultraviolet and nuclear magnetic resonance spectroscopy; spectrophotometry; refractometry; and other quantitative methods.

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