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Appendix III

Field Methods for Processing Food Samples

C. M. HLADIK

Laboratoire d'Ecologie et Primatologie du CNRS, Muséum National d'Histoire Naturelle, Brunoy, France

1. Methods for Collecting Samples

Food samples and botanical specimens are generally collected simultaneously, the latter being necessary to identify different food samples. Methods for processing botanical specimens are generally well known. By contrast, food samples must be obtained in fairly large quantities (about 200 g, wet weight) to allow further analysis with standard techniques, and difficulties have been found in the collection and processing of such large samples. A few technical solutions adopted in our studies, especially on leaf monkeys and chimpanzees (Hladik, this volume), might be useful for future research about primate feeding behaviour.

Specimens located on trees were collected with the help of accessories, to avoid the risks and difficulties of climbing.

Extension poles

A tree pruner mounted at the top of a long pole was sufficient to collect all the specimens in the semi-deciduous forest of Sri Lanka where trees do not exceed 25 m in height. The pole was made of a series of five bamboos that can reach 15 m: this height is sufficient to collect specimens from the lower branches of most trees, but two persons were required to handle the pole which had a basic piece of 12-cm diameter. The bamboo pieces were held together with a wooden tenon and the end of the hollow parts reinforced with iron wire and araldite.

Tree climbing dress

In the rain forest, the lower branches of many tree species cannot be reached directly with a tree pruner. A special dress for climbing trees was designed and made of strong canvas with pieces of leather reinforcing the inside parts of knees and protecting the chest against the rough bark of the tree trunk. Direct climbing of lianas and trees is facilitated by such dress and collection of samples with the tree pruner can be done from a high position; but not without risk.

Tree climbing platform

The most useful accessory for collecting in the rain forest is the tree climbing stand sold by "Forestry Suppliers, Inc." (Jackson, Miss., USA). We used it in Gabon with the tree climbing dress (Fig. 1) to protect the chest. This platform is made of light plywood and allowed us to climb smooth vertical trunks up to 30 m without special training. From this position,[a] it was possible to collect food samples up to 40 m, with the help of the tree pruner mounted on its bamboo pole. Trees exceeding 40 cm in diameter cannot be held by the gripping bar of this stand but it is generally possible to collect samples from a nearby vertical tree trunk. Exceptionally, a 10-m collapsible aluminium ladder was used to start climbing with the platform from the point where the diameter of the trunk is small enough to mount the climbing tree stand.

2. Processing Food Samples

Drying

After collecting, the samples were carried in large polyvinyl bags to avoid desiccation before weighing. Samples of about 200 g were put in paper bags (about 50 g dry weight is enough to carry out the most important analysis with duplicates or triplicates: most of the specimens have 70 to 80% water content). Paper bags were made of non-glossy paper to allow moisture to escape (a cone of paper can be made with old newspapers).

Paper bags with food samples inside were dried in an electric oven when this was available. In the field, we used the heat of a kerosene lamp to make an "oven" with a big tin box penetrated by two tubes to allow air circulation (Fig. 2A). A cylinder of canvas maintained the

[a] A safety rope properly tied around the trunk and around the chest of the collector *must* be utilized.

samples. Paper bags with the dried samples can be kept in plastic bags carefully tightened or sealed.

Processing in alcohol

Fixation of the samples in boiling ethyl alcohol is one of the best methods of stopping all enzymatic reactions and preserving all components. In field conditions, samples of small size (about 20 g fresh weight) can be processed by this method. Thus, it complements the preceding method, and is necessary to allow detailed investigation on soluble sugars, amino acids and lipids (see Section 3).

Ethyl alcohol (96°) was heated to boiling in an Erlenmeyer flask and the food sample dropped into it after slicing in small pieces (less than 2 mm thickness). A condenser was adapted to a rubber cork on top of the flask. It was cooled with fresh water kept in a canvas water-carrier to prevent the alcohol from evaporating during processing. We utilized an alcohol lamp or a butane gas stove to heat the flask, with a shield to protect it as shown on Fig. 3.

The volume of alcohol in the flask must be about five times more than the volume of the food sample. It has to boil with the sample for 15 minutes. When the alcohol is no longer hot, the food sample with all the alcohol must be carefully taken out of the flask and can be stored in a plastic jar.

Freezing

Only deep freezing (below −30° C) adequately preserves food specimens. Several field stations in the tropics are now equipped with large freezers in which food samples can be kept but there are problems in long-distance transportation of such samples and the analytical operations must be started as soon as the samples are defrosted.

3. Different Types of Analysis Consistent with the Different Methods of Field Processing

Dried food samples can be used for the standard operations of analysis such as those described and referred to in Hladik *et al.*, 1971a. There was no significant difference in the titrations of minerals, nitrogen, lipids, cellulose and total glucids, between food samples boiled and preserved in alcohol and dried food samples of the same origin. By contrast, important differences between the two parts of the food sample (one processed by drying, the other one processed in alcohol) appeared when comparing the proportions of the different soluble sugars (Hladik *et al.*, in press). This was particularly obvious for samples requiring a long

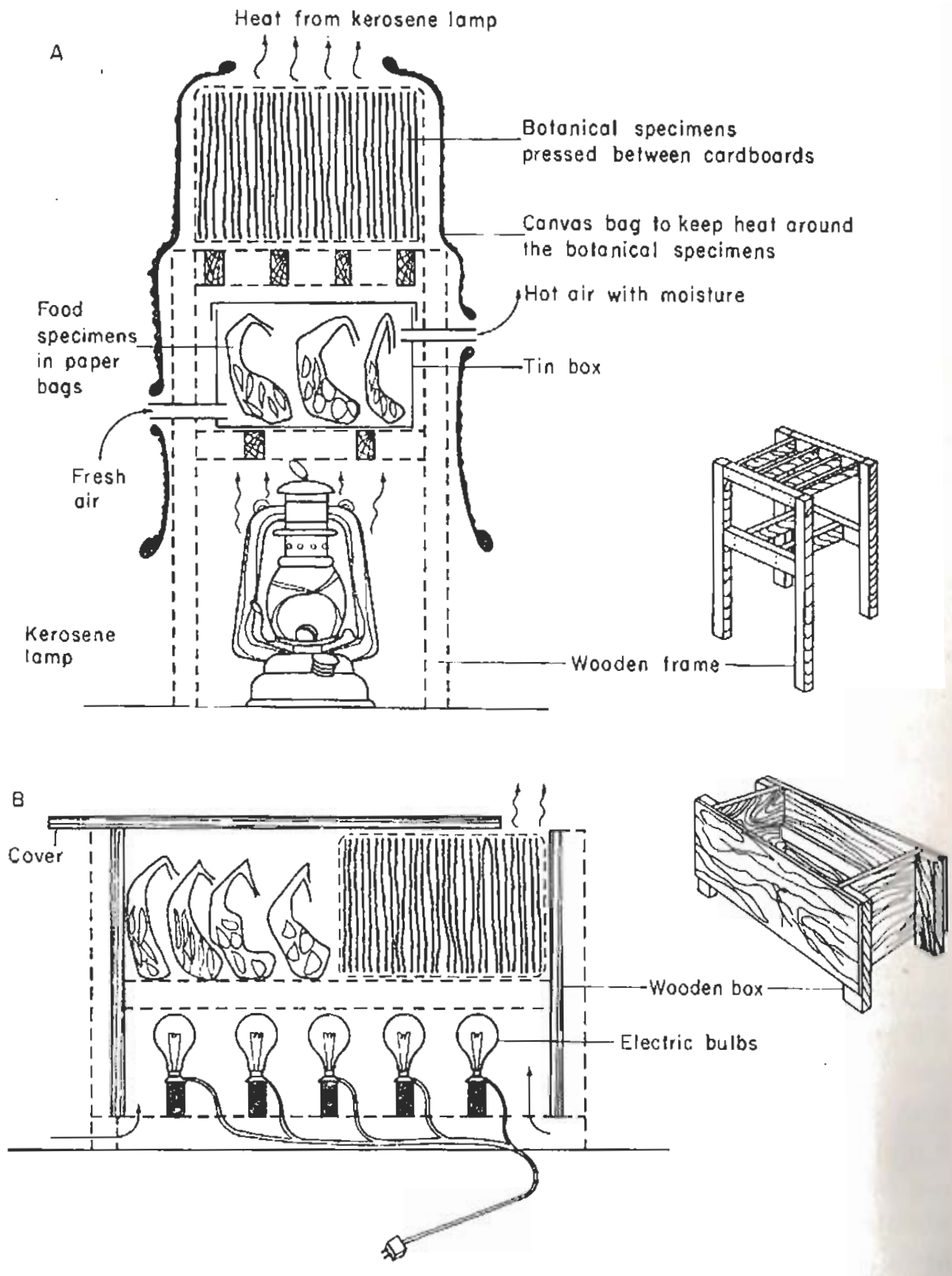

FIG. 2. Models of dryers for food samples and botanical specimens. A. To be used in the field. B. To be used at the field station.

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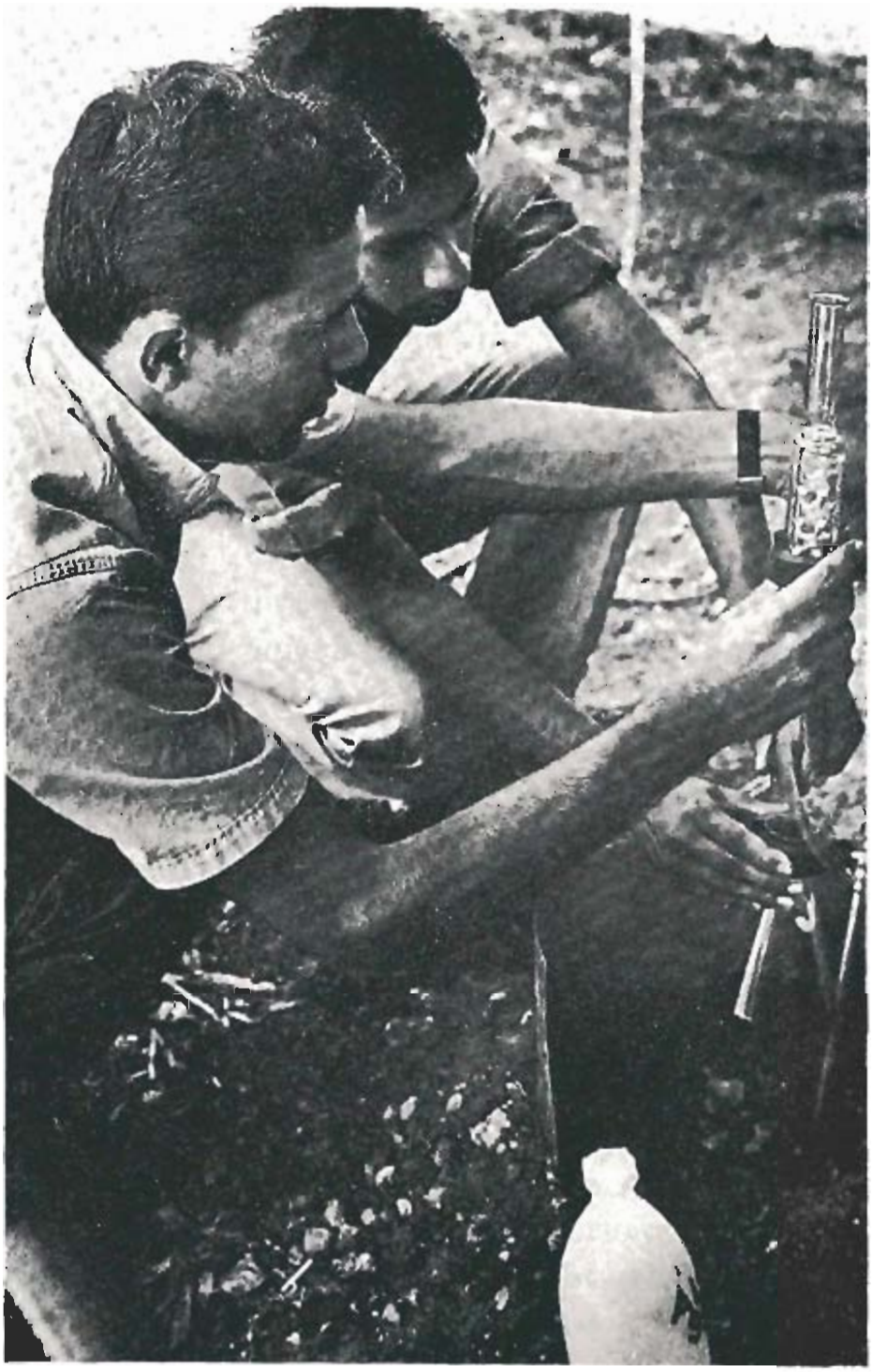

FIG. 3. Students of the University of Sri Lanka processing food samples in boiling alcohol, at the Polonnaruwa Field Station. Water for cooling the condenser is in a canvas water-carrier hanging above the apparatus.

time to dry such as large flowers and leaves. In these dried samples, sucrose was missing, probably because rapid fermentations occurred at the beginning of the drying process; glucose, fructose and other glucids of small molecular size were found in larger amounts.

Food specimens boiled and preserved in ethyl alcohol can be used for analysis and titration of soluble glucids and for research on the amino acids after lyophylization and grinding (Hladik and Viroben, 1974).

Research on fatty acids can also be carried out on food samples processed in alcohol or in 10% formalin (Hladik *et al.*, 1971b). We have no comparative result with other methods for these last two analyses: although oxidation might not affect most of the lipidic components, the only reliable process for detailed analysis would be deep freezing and lyophylization.

Tests for different secondary compounds (for instance, alkaloids) can be made on dried specimens but the results may differ slightly from those obtained on fresh specimens (see discussion in A. Hladik, 1977). Nevertheless, fractioning and further chromatographic analysis necessitates dried food specimens.