An Efficient Methodology for Processing of Herbarium Specimens of Cultivated Plants

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An efficient method of processing has been developed for depicting diverse range of material represented in the National Herbarium of Cultivated Plants, National Bureau of Plant Genetic Resources (NBPGR), New Delhi. This method is especially suitable for drying herbarium specimens and economic products that are bulky, fleshy, change quality drastically (colour) or separate into parts on processing. In addition, fungal/ insect infection manifested during processing was also minimized using this method. Thus this method proves to be an efficient one, less time and labour consuming as compared to traditional processing techniques.

Key words: Herbarium methodology, Preservation, Efficient methodology

The National Herbarium of Cultivated Plants (NHCP) holds diversity of plant specimens of cultivated taxa and their wild/weedy relatives, wild economic types and the economic products referred for studies on plant genetic resources and various research and teaching programmes. The variability included in different crop-groups has been represented as cereals and pseudo-cereals, legumes, fruits, vegetables (including rhizomes, bulbs, tubers), spices and condiments, etc. The herbarium specimens are prepared by processing the plant material by placing between absorbent sheets, pressing and drying by periodically changing the sheets until it dries (Lawrence, 1951; Jain and Rao, 1976; Nayar, 1998). The processing of difficult material is generally done by use of artificial heat, slicing/ sectioning and removal of extra tissue, use of hot-water. Besides, complementary collections as photographs/ digital images and illustrations/ line-diagrams and wet preservation are also included to cover wide range of diversity.

Specimens of difficult to preserve plant groups are generally represented as pickled material (wet preservation) or bulky herbarium. Traditional drying techniques are prolonged, and sometimes materials do not retain original quality and features and thus have least significance for use in taxonomic identification. Getting a perfect specimen is difficult in case of fleshy materials like succulent fruits and vegetables, tubers, rhizomes, rooted materials, fleshy flowers, etc.

Techniques using high frequency waves have been frequently used in processing of living material (Brown and Gordon, 1967; Trappe, 1982; Hu, 1992). Microwaves are one of the most commonly used radiations that have demonstrated their inertness to cellulose (Brandt and Berteaud 1987). They have been frequently used to disinfect cereals and liquid produce in food and agriculture industry (Delaney *et al.*, 1968). Use of microwave radiation for few minutes has been proved to be useful to cause mortality of fungal spores, eggs, larvae, nymphs and adults of many storage fungi/ insects (Flieder *et al.*, 1995). The microwave technique has been advantageous as it is safe to use and available as compact equipment ideal for routine handling.

Use of microwaves for preserving the plant specimens under long-term conditions is still uncommon. In the present investigation selected species that are difficult to process for inclusion as herbarium specimens have been standardized for microwave treatment. This methodology has proved to be an efficient one and may be extended to achieve good quality of specimens as well to prolong their self life by reducing bio-deterioration caused by storage fungi, insects, that infect/infest in succulents and tubers, rhizomatous material etc.

Meterials and Methods

The sample specimens of the test material was cut in to appropriate size and exposed to different doses of irradiation using microwave oven (BPL-Sanyo, model no BMC-900T; 40 litre capacity; 230 vol. 50 Hz). The plant specimen was placed on the glass turntable of the microwave oven and subjected to different degree of treatment from higher to lower dosage (in alternate days). Power (from 100-750 watts) and processing time (10-120 seconds) were selected according to the type and texture of the material (Table 1). After treatment with microwave, conventional drying procedure was followed. This process was repeated till specimen was completely dried. A control set of the test material was subjected to

Table 1. Preparing herbarium specimens using different treatment with microwave

Species (Family)	Plant / part(s)	Treatments (sec.)	Days for complete drying		Observations
			Microwave	Control	
Daucus carota L. (Apiaceae)	Root, full plant	80, 40, 10	8	15	Time reduced, no shrinkage
Allium (*A. sativum*, *A. cepa*, *A. tuberosum*, *A. clarkei*) (Alliaceae)	Plant, bulb	70, 50, 20, 10	12	20	Time reduced, no infection
Brassica oleracea var. *gongylodes* L. (Brassicaceae)	Plant	90, 60, 20, 10	10	25	Shape retained
Ribes nigrum L. (Grossulariaceae)	Plant	60, 40, 10	6	10	Berries infection free, berry drop was low
Sansevieria hahnii (Liliaceae)	Plant	90, 40, 10	5	12	Retained colour and quality
Aloe barbadense Mill. (Liliaceae)	Plant, inflorescence	70,40,10	7	20	Retained colour and quality
Cyclanthera pedata L. (Cucurbitaceae)	Fruit, plant	60, 50, 20, 10	8	15	Fruit dried without losing shape
Beta vulgaris L. (Chenopodiaceae)	Plant	70, 60, 40, 10	12	25	Colour and plant shape was sustained
Chlophytum borivilianum Santapau (Liliaceae)	Plant with tubers	60, 50, 30,10	12	20	Fsat drying with good quality tubers
Musa acuminata Colla (Musaceae)	Inflorescence/ fruits	120, 60, 30, 10	14	25	Fast drying with good quality fruit/inflorescence
Momordica dioca Roxb. ex Willd. (Cucurbitaceae)	Fruit	60, 40, 20	7	15	Retention of colour and shape of plant
Nymphaea nouchali Burm. f. (*Nymphaeaceae*)	Torus/edible seed	110, 50, 20	15	25	Colour retention and good quality
Musa bulbisiana Coll. (Musaceae)	Fruit	120, 60, 30, 10	10	20	Colour retention and good quality
Trapa natans L. var. *bispinosa* Roxb. (Trapaceae)	Plant with fruit	90, 60, 20	7	20	Colour retention and good quality
Brassica juncea (L.) Czern & Cess. (Brassicaceae)	Plant	40, 20, 10	5	10	Colour retention
Spinacea oleracea L. (Chenopodiaceae)	Plant	40, 10	10	12	Colour retention
Amaranthus hypochondriacus L. (Amaranthaceae)	Plant	40, 10	7	10	Colour retention
Bassela rubra L. (Basellaceae)	Aerial parts	60, 20, 10	7	15	Colour retention; no defoliation in treated samples
Raphanus sativus L. (Brassicaceae)	Plant	120, 60, 40, 10	10	22	Colour retention, root shape retained
Brassica oleracea var. *botrytis* L. (Brassicaceae)	Head (sectioned)	120, 60, 20, 10	12	26	Retention of shape, no infection
Ipomoea aquatica Forsk. (Convolvulaceae)	Leafy twig	40, 10	5	10	No leaf detachment
Ficus palmata Forsk. (Moraceae)	Inflorescence	60, 40	7	15	No leaf detachment
Punica granatum L. (Punicaceae)	Fruit twig	90, 50, 20	8	20	No detachment of parts
Citrus aurantifolia Sw., *C. chinensis* (Christm.) Sw. (Rutaceae)	Flowering twig	60, 40, 10	5	10	No leaf detachment
Portulaca oleracea L. (Portulacaceae)	Aerial parts	30, 20, 10	6	12	Colour retention; no leaf detachment
Bryophyllum pinnatum (Lam.) Kurz. (*Crassulaceae*)	Flowering twig	50, 30, 10	6	12	Colour retained
Rhoeo discolour (*Commelinaceae*)	Leafy twig	60, 30, 10	7	14	Colour retained

(* Microwave treatment on alternate days)

traditional drying using recommended methods. Result of the treated material was compared with that of the control in terms of time needed for full processing, quality of specimen and labour involved in processing.

Result and Discussion

The microwave processing was applied for processing of 30 species specimens belonging to 25 genera and 18 families under different crop-groups of cultivated plants

(vegetables, roots, tubers, leafy types; fruits, flowers, etc.). The processing time was observed to be reduced by 40-60 per cent (Table 1). Besides, quality parameters such as low infection, retention of colour of parts, less shrinkage in tissue in case of tubers/ roots, optimal breaking of plant parts (leaves, flowers in ornamentals) were also taken care. This method proved to be relatively efficient in terms of time and labour and retention of quality. High frequency microwaves disrupt the soft tissue, soften and expel extra water to the organ surface and fix plant pigment(s) in tissue. The expelled moisture is soaked by the blotter and easily removed during processing/ pressing. During this process, secondary infection caused by the microorganisms or insect, was reduced and thus the shelf- life of the specimens was increased.

During the drying process using traditional methods, materials like succulents, fleshy types, ornamentals, etc. get infested by microorganisms and thus lost or their quality gets affected. Specimens of other types such as of leafy taxa (chenopods, brassicas, amaranths), deteriorated due drastic colour change (Pandey *et al.*, 1995; Pandey and Nayar, 1996). Using treatments by microwave radiations satisfactory results were achieved. The observations in treated and control material were presented in Table 1.

In case of fleshy material like *Musa* spp., *Daucus carota*, *Portulaca oleracea*, *Cyclanthera pedata*, *Beta vulgaris*, etc. there was reduced processing time followed by retention of the original shape of fleshy succulent tissue. In others, organ detachment was minimized by application of microwave treatment- *Ribes nigrum, Ficus* spp., *Ipomoea aquatica*, *Punica granatum*, etc. Organ discolouration was reduced in majority of specimens particularly in *Brassica* spp., *Spinacea oleracea*, *Amaranthus hypochondriacus* and *Rhoeo.*

Conclusion

This technique appears to be more efficient as compared to traditional drying method used for difficult taxa and can be extended for processing and inclusion of large number of species in the herbaria. However, long-term effects on the material use are under investigation.

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