Specialising in roses has allowed us to put a lot of time and effort into determining the precise requirements for growing on the crop and into sorting out some of the problems. It has also enabled us to give technical back-up to our customers to help them grow a quality crop. In fact we now even circulate a regular information sheet to all our customers to suggest what they should be doing each month. It also includes information about problems they might meet and those previously met by others—so hopefully they can avoid them. We hope that by giving this back-up we can help our customers produce quality plants which will establish well in the garden of the final consumer and thereby create further demand for micropropagated plants.

PROPAGATION SYSTEMS IN THE 1980s: A PERSPECTIVE ON THE BEST AVAILABLE

KEITH LOACH

Institute of Horticultural Research, Worthing Rd., Littlehampton, West Sussex, BN17 6LP, U.K.

The aftercare of newly micropropagated plant material and the rooting of conventional cuttings share similar environmental requirements. Specifically, these are:

1. The need to conserve water in the plant tissues, since cuttings, whether from *in vitro* or conventional sources, readily suffer water deficits because they have no roots. In microplants already rooted *in vitro*, the roots often function poorly; their leaves, having developed in high humidities, have thin cuticles (1), little surface wax deposition (4), and relatively few stomata (5) with imperfect stomatal control (3).

2. *It is nevertheless important to avoid excessive wetting of the* plant material. In the case of micropropagated plants, the weight of water droplets can be physically damaging. In conventional cuttings there is need to avoid waterlogging of the basal stem tissues, which occurs especially in winter conditions and results in rotting.

3. Irradiance conditions must allow for the gradual redevelopment of autotrophic nutrition in micropropagated plants which have relied on sugars in the medium while in culture. Conventional cuttings of many woody ornamentals apparently require only low irradiance until they develop roots and begin to grow actively (10).

4. Temperatures should be moderate (18 to 25°C) but not excessive, i.e. below 40°C.

THE IMPORTANCE OF WATER BALANCE

The requisite balance between maintaining turgor in the plant material and avoiding waterlogging changes with the season. For example, when softwood cuttings of 6 species were rooted in 3 different systems in summer (Figure 1a), best rooting occurred in the system which gave greatest gain in water content, i.e. enclosed-mist (mist operated within a clear, polyethylene tent). Poorest rooting was in open mist, where cuttings showed a net loss of water after insertion. In winter, with shorter days and lower irradiances, the water balance was more easily maintained, though the systems behaved in the same relative fashion in this respect (Figure 1b). However, rooting in winter was poorest in enclosed-mist because the stem bases were often waterlogged. In such conditions, alcohol rapidly accumulates in the basal tissues, suggesting that oxygen availability is reduced (R. S. Harrison-Murray, personal communication).

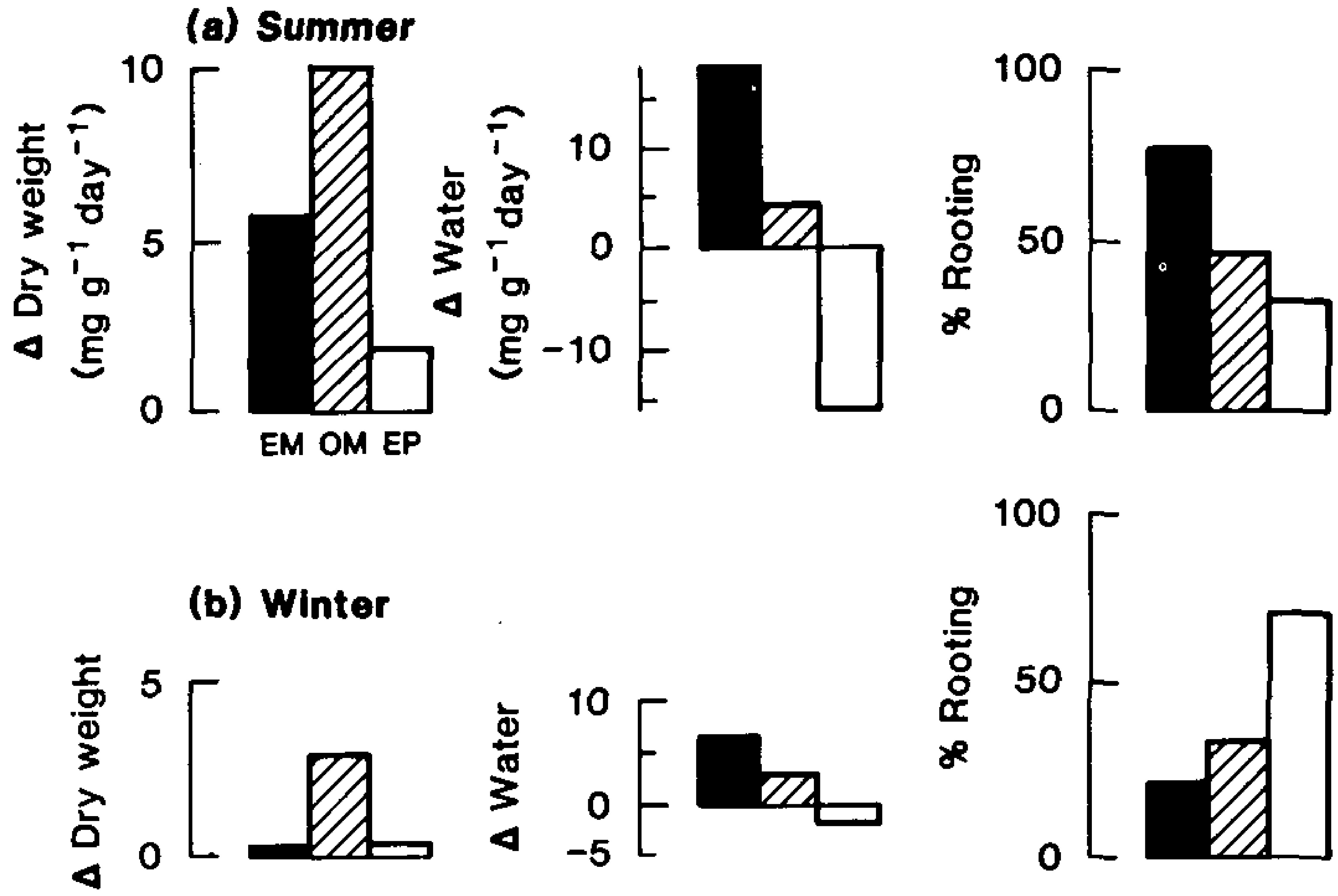

Figure 1. Changes in dry weight and water content of cuttings in open mist (OM), polyethylene-enclosed mist (EM), and a non-misted polyethylene enclosure (EP), measured over 2–4 weeks after insertion, and the eventual rooting percentages.

(a) Means for 6 species in summer.

(b) means for 4 species in winter.

Changes are expressed relative to water content and dry weight of cuttings at insertion.

These principles were further evident in experiments using a fog tunnel. Cuttings of 21 species were inserted in a clear polyethylene enclosure, 8.4m long × 1.7m wide × 0.9m high, with a single sonic fogging nozzle at one end. This allowed a simultaneous comparison of a range of propagation environments at 10 equally-spaced locations away from the nozzle. Nozzle operation was controlled by a psychrometric controller set at 97 per cent relative humidity, positioned 3.4m from the nozzle. Figure 2 shows that for 4 species rooted in July, when maintenance of turgor is the prime consideration, rooting was best close to the nozzle. For 4 species in winter conditions, rooting was best at location 7. Note, however, that different species were used in July and October and subsequent evidence suggests that the differences may have a species as well as a seasonal component (see Figure 4). The remarkable feature of these results is the very evident sensitivity of rooting to environment, considering that the locations are only 0.84m apart and mean daily relative humidities ranged from 100 per cent at the nozzle to around 90 per cent at the far end. It would be instructive to carry out similar studies using cuttings and plantlets from *in vitro* sources.

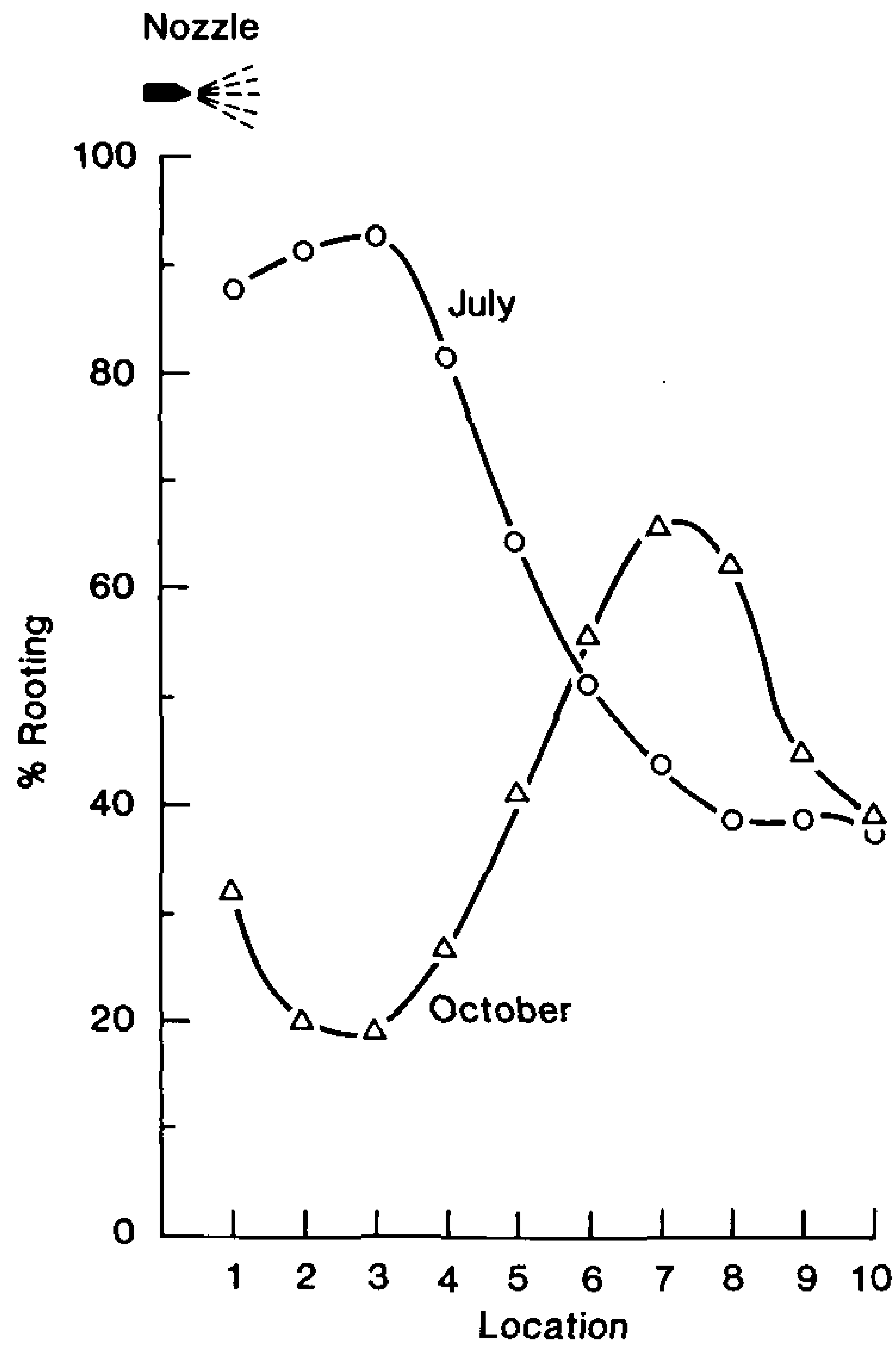

Figure 2. Mean rooting percentages for 4 species inserted in July and 4 in October, at 10 equidistant locations (0.85 m apart), from a single fogging nozzle in a clear, polyethylene enclosure.

Available systems. Propagators are thoroughly familiar with the use of polyethylene tents and conventional open mist, which need not be described here. Enclosed-mist is used less frequently, probably less then it should be. In U.K. conditions it has the advantage of providing a useful temperature rise relative to open mist (8).

It also maintains higher humidities than occur in a non-misted tent and the mist-wetted leaf surfaces provide insurance against tissue water loss. As shown in Figure 1, enclosed-mist can be over-supportive in winter, particularly with conifers. Nevertheless, with appropriate shading and regulation of the misting frequency, it can be very effective at all times of year (7).

We have limited experience in the use of enclosed-mist for weaning micropropagated material but 100 per cent survival of unrooted lettuce microplants has been achieved in March. For this purpose, the enclosed-mist was shaded to reduce an average 11 $MJ/m^2 \cdot day$ outdoors (total short-wave) to 3 $MJ/m^2 \cdot day$ in the enclosure. Misting frequency was controlled by a radiometer during the day to provide a two sec burst for every 0.12 MJ/m^2 of radiation and by timer at night to produce a two sec. burst every 60 min.

Fog presents potentially the most effective means of regulating the propagation environment. Fog generators produce very fine water droplets ($<20\ \mu m$) which remain airborne for a long time, have a large surface to volume ratio, and so evaporate readily to humidify the air. Fog is favoured for delicate, micropropagated material because less foliar wetting occurs than with mist. It is also valuable to provide cooling of glasshouse air, again because fine droplets evaporate easily. However, because transpiration into a humid atmosphere is restricted, little of the energy absorbed by leaves is lost through evaporative cooling and leaf temperatures can be relatively high and even damaging. Shading is therefore essential to reduce the incoming irradiance and avoid water stress in the cuttings (2).

An extensive range of fogging equipment is now available and employs one of three basic principles:

1. **Centrifugal systems,** which use centrifugal force to atomize water. Outputs range from 1 to 190 litres per hour but the larger models produce a wide range of droplet sizes, resulting in fall-out close to the units, so making them less suitable for micropropagules.

2. **High pressure water nozzles,** which are similar to mist nozzles but with ultrafine orifices, and operated at high pressures up to 1000 psi (6.9 MPa). Typical outputs per nozzle are 5 to 8 litres per hour and arrays of these relatively inexpensive nozzles can be used to produce a large and evenly distributed fog output.

3. **Pneumatic nozzles,** which use compressed air and water. Recently developed "sonic" nozzles generate a field of high-frequency sound waves at the tip of the nozzle, which disrupts the water into uniformly fine droplets. They can produce up to 55 litres per hour of fog (more typically 15 to 20 litres per hour). The individual nozzles and associated air compressors are relatively expensive.

The merits of different fogging systems are compared in a subjective fashion in Table 1. These must be considered in relation to the individual application, e.g. for micropropagules that require minimal foliar wetting and a spatially uniform fog distribution, sonic nozzles, or a high pressure water system are probably better than most centrifugal generators. For effective glasshouse cooling, the large capacity and low cost of centrifugal foggers may be attractive.

Table 1. A subjective comparison of fogging systems (*** = highest rating).

	Sonic nozzles	Centrifugal generators	High-pressure water nozzles
Low price	*	***	*
Capacity/unit cost	*	**	***
Effectiveness for ventilated cooling	*	***	**
Spatially uniform distribution	**	*	***
Uniformly fine droplets (minimal fallout)	***	*	**
Uniform air flow	**	*	***
Low maintenance requirement	***	**	**

Uneven spatial distribution of fog is a common complaint in many propagation installations. The ill-effect on rooting can be judged from Figure 2. It is helpful to install ancilliary ventilation fans to gently move the fog around the glasshouse. If ventilation is too vigorous, droplets coalesce and may fall-out out of the air.

FUTURE DEVELOPMENTS

The experiments in the fog tunnel clearly show that rooting of cuttings is very sensitive to environmental variations. As already noted, this sensitivity derives from the simple fact that cuttings, whose water uptake is restricted through lack of roots, are especially prone to tissue water deficits. Conversely, if transpiration is too restricted by environmental conditions cuttings suffer basal waterlogging. These observations raise the interesting challenge of characterising the narrow optimal range of conditions for rooting and controlling the propagation environment within this range.

It has proved possible to characterise propagation environments either by using sensitive evaporimeters (6) or by calculating from measurements of temperature, humidity, and leaf wetness, a "water stress index" (WSI) for the environment. The WSI is a measure of the accumulated leaf-to-air vapour pressure difference during the day (i.e. the summed driving force for water loss from the leaves), adjusted to accommodate the effects of any surface wetness of the foliage.

Calculation of the WSIs at each location in the fog in Figure 2, explains the seasonal differences in rooting pattern (Figure 3); in both July and October, best rooting occurred at the same WSI.

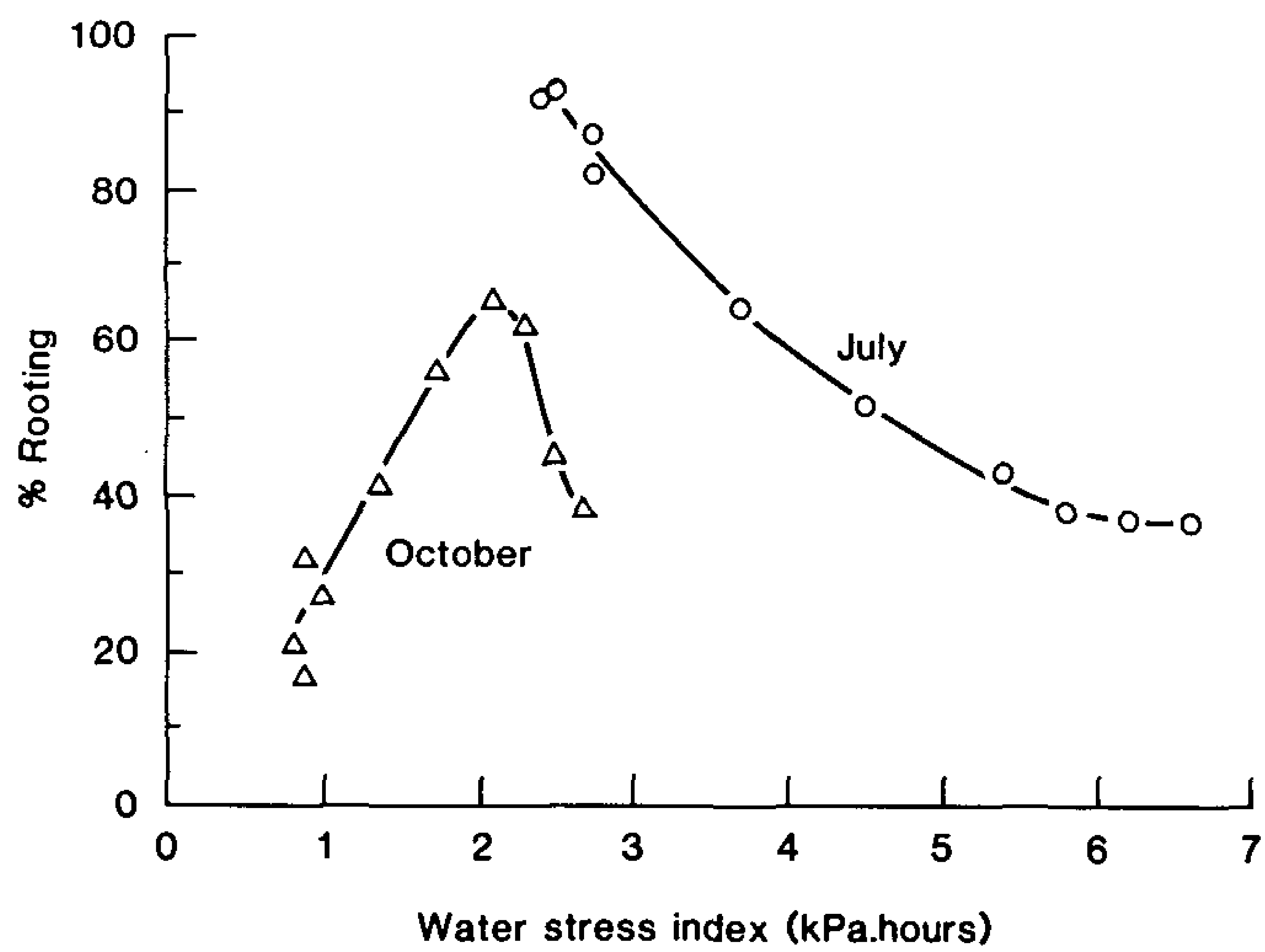

Figure 3. Percent rooting of fogged cuttings shown in Figure 2, in relation to the environmental water stress index (WSI), calculated for each location.

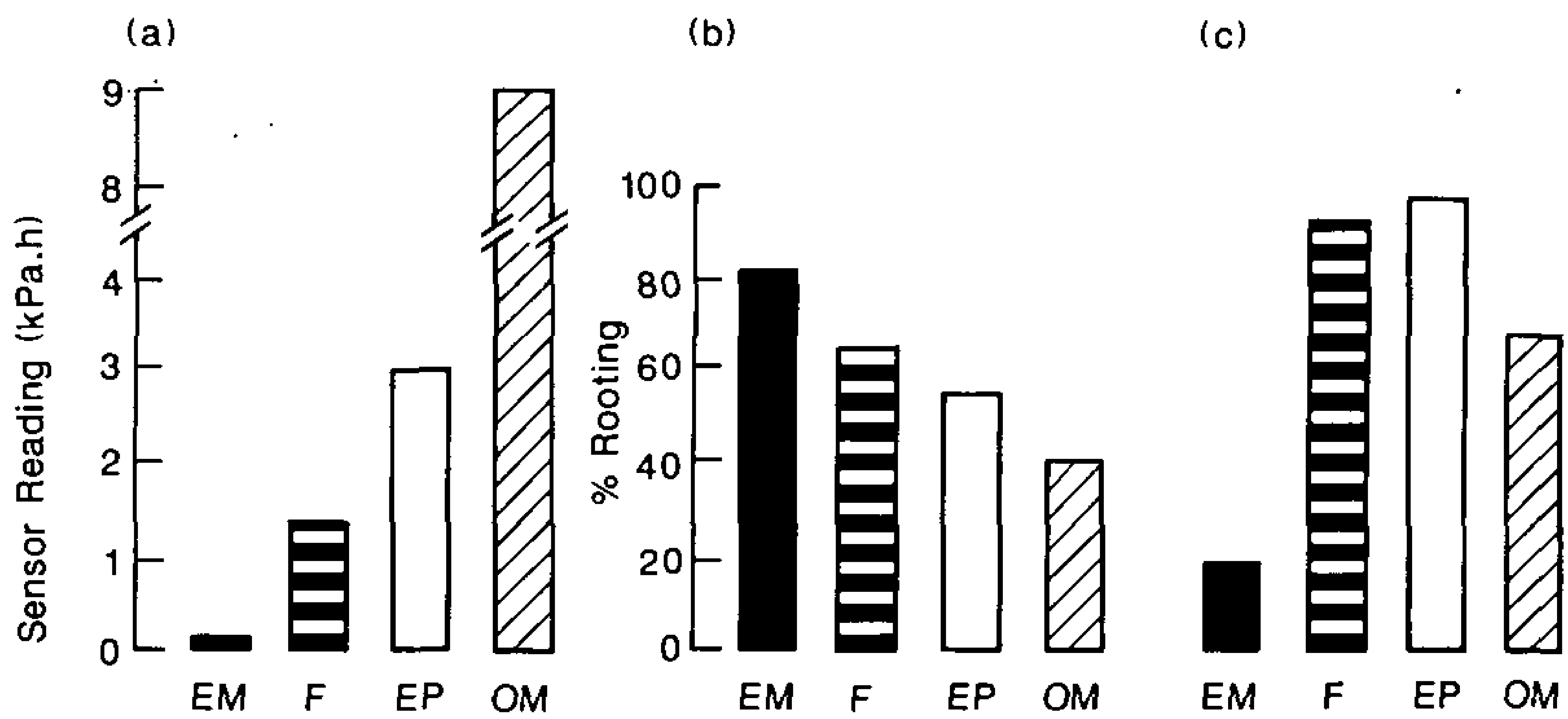

Figure 4. (a) Mean daily evapometric sensor readings in 4 propagation systems (EM, enclosed mist; F, fog; EP, non-misted polyethylene enclosure; OM, open mist), and % rooting of
(b) *Acer palmatum* 'Atropurpureum', *Photinia* × 'Red Robin' and *Viburnum* × *bodnantense* 'Dawn' (mean for the 3 species), and of
(c) *Cryptomeria japonica* 'Elegans'.

In collaboration with Dr. R. S. Harrison-Murray of East Malling, we have developed sensors which can provide continuous, quantitative monitoring of a propagation environment (details to be published). They can be used to compare different systems; for example in Figure 4a, open mist, enclosed-mist, a polyethylene tent, and fog were compared. In both the open and enclosed-mist the burst frequency was timer-controlled to give a 3 sec. burst every 20 min. from 04.30 to 20.30. The relative performance of each system could, of course, change if they were managed differently; one important use for the evapometric sensors is to compare, quantitatively, the effectiveness of alternative management procedures.

Preliminary rooting results showed that for May/June insertions of "soft" cuttings of broadleaf species, the percentage rooting achieved after three to five weeks improved with increasingly supportive environments (Figure 4b). However, the conifer, *Cryptomeria japonica* 'Elegans' rooted best in the "drier" polyethylene tent environment (Figure 4c), which suggests that different groups of species have different environmental optima.

CONCLUSIONS

Success in propagation is governed by many different factors but these studies show that the propagation environment plays a major role and that a narrow optimum range of conditions exists. Techniques for measuring the environment have been developed, fortunately at the same time as the methods for controlling that environment (through fog, computer control, automatic shading), have substantially improved. Combining these developments promises reduced failure rates both for weaning micropropagules and rooting conventional cuttings; this should provide the 'Best Available Systems' of the title, but in the 1990s rather than the 1980s. Experience gained from these technical improvements, should feed back into less sophisticated systems in the form of improved operational guidelines. The simpler systems will no doubt remain with us because they are often very cost-effective.

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WEANING AND AFTERCARE OF MICROPROPAGATED NURSERY STOCK

MARGARET A. SCOTT

Agricultural Development and Advisory Service
Efford Experimental Horticulture Station
Lymington, Hampshire

While there was initial enthusiasm from nurserymen to wean micropropagated propagules received directly from the laboratory, their success rate was variable. As a result most laboratories now have their own specialised weaning units to produce the rooted plantlet/liners for sale to growers, which can be handled in a similar manner to conventionally propagated material.

A specialist ADAS micropropagation unit at Brogdale Experimental Horticulture Station in Kent is, in collaboration with Efford EHS, investigating factors involved in successful weaning-off and growing-on of micropropagated material.

Work so far has concentrated on relatively high value crops, particularly those in the Ericaceae group, most of which are suited to growing under protection thus capitalising on the potential for growth from micropropagated material, e.g. *Rhododendron*, deciduous *Azalea*, *Pieris*, *Camellia*, *Kalmia*, and *Magnolia*.

This paper reviews the larger scale weaning and growing-on work in progress at Efford E.H.S.

Weaning has been defined as the acclimatization of the micropropagated material (propagules) from the precise laboratory medium and environment (*in vitro*) to typical horticultural growing environments and composts (*in vivo*). There are two distinct stages:

1. Transfer of propagules from culture media to compost and rooting under high humidity environments.

2. Hardening off and acclimatization of rooted material from high humidities to "normal" horticultural environments (under protection).

Laboratory treatments can have a marked influence on success