EFFECT OF TEMPERATURE, GIBBERELLIC ACID, AND INDOLYLACETIC ACID ON ROOT AND SHOOT GROWTH OF CUTTINGS FROM *PODOCARPUS LAWRENCEI* HOOK f.*

By A. G. Khan†

It has been shown (Khan 1967) that roots of *Podocarpus* produce nodules under sterile conditions, thus proving that nodule production is a normal feature of the root system and is not induced by the presence of any microoorganism.

During this study it was observed that the roots produced by cuttings became nodulated sooner than roots of seedlings grown under the same conditions. Many vegetative buds expanded on the shoot cuttings but root production and growth were poor when daily air and soil temperatures were high, whereas the converse was the case during a following period when temperatures were lower.

In an effort to find a reliable method for the rapid production of nodulated roots for anatomical and physiological studies, an experiment was set up to study the effects of temperature and growth substances on root and shoot production by cuttings.

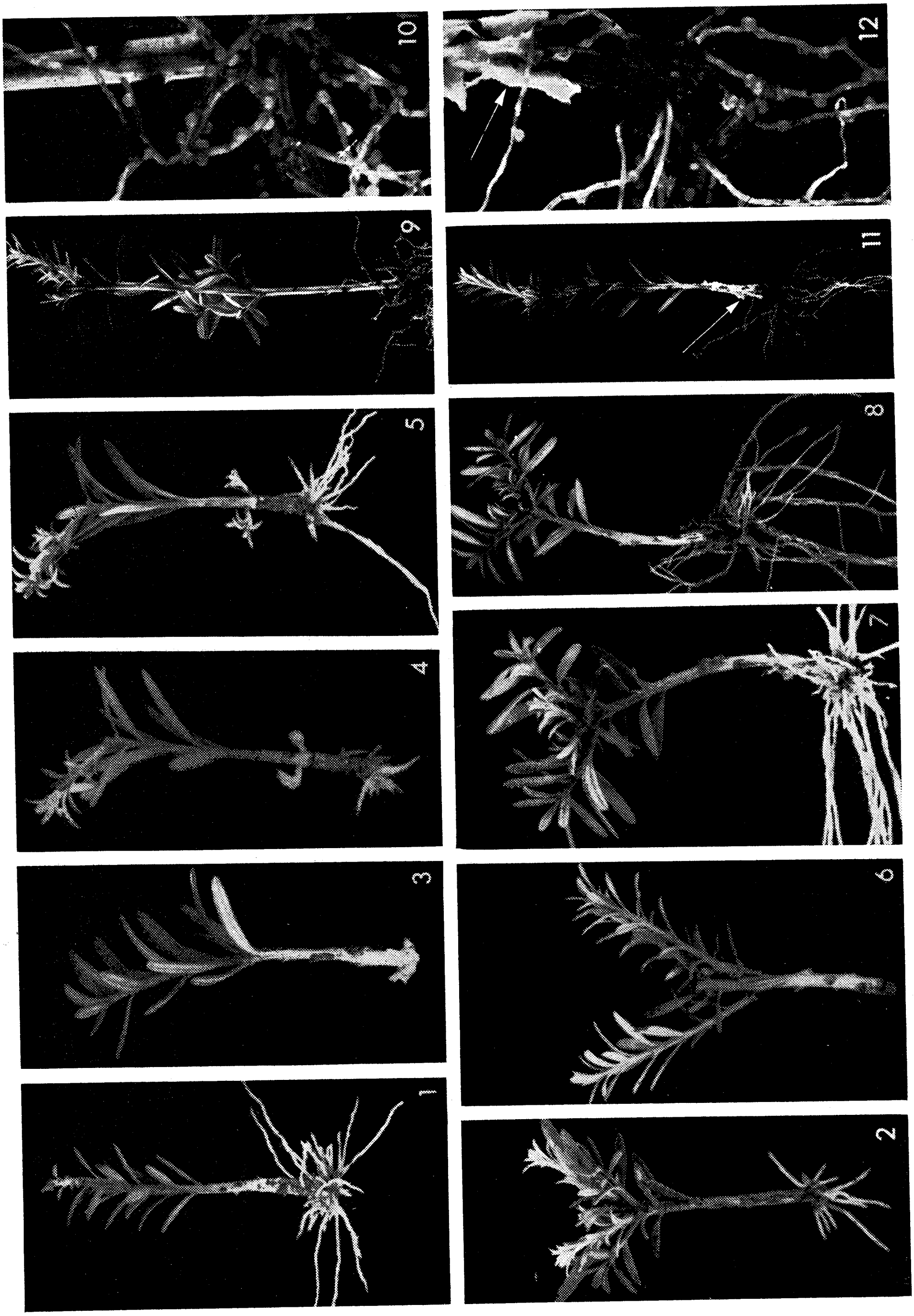
Materials and Methods

Vegetative material from the field was collected from Mt. Ginini, A.C.T. (the site from which material was obtained by Bergersen and Costin 1964) in February 1966. Shoot cuttings 6–7 cm long were surface-sterilized for 10 min with a 10% solution of sodium hypochlorite, washed six times with autoclaved distilled water, denuded of lower leaves, and planted in pots (five cuttings per pot) of autoclaved sand and peat (equal parts). The pots were completely enclosed in new polythene bags and were kept at "low" temperatures (day 21°C, night 15°C) or "high" temperatures (day 28°C, night 21°C) in Sherer environmental growth chambers with a 16-hr photoperiod. In these chambers the source of light was VHO Cool White Powertubes supplemented with Phillips 40-W incandescent lamps, giving a light intensity of 4000 f.c. at the level of the plant bed as measured with a selenium photocell light-meter. The light intensity, however, was reduced to 1000 f.c. by shading the pots with white paper.

The experiment lasted for 9 weeks. Cuttings were removed under aseptic conditions every 3 weeks for observation and recording. The pots were watered aseptically every 3 weeks with autoclaved distilled water.

* Manuscript received November 17, 1967.

† School of Biological Sciences, University of Sydney, N.S.W. 2006.



[*For explanation of Figures 1–12, see opposite page.*]

Observations and Results

Effect of Temperature: Location Unchanged.—Two pots of cuttings were kept in the low-temperature cabinet for the whole 9-week period without treatment with any growth-promoting substances. The cuttings developed many roots (nodule-free), and no shoot growth occurred (Fig. 1). By contrast, the untreated cuttings of two pots kept in the high-temperature cabinet for the whole 9-week period developed fewer adventitious roots (nodule-free), and good shoot growth occurred with many buds expanding (Fig. 2).

Effect of Temperature: Location Interchanged.—The untreated cuttings of two pots which were kept in the low-temperature cabinet for the first 3 weeks developed few adventitious root primordia at their bases and no shoot growth occurred (Fig. 3). These cuttings were then transferred to the high-temperature cabinet where two to three vegetative buds per cutting expanded within the next 3 weeks (Fig. 4). At the end of the experiment the cuttings showed good shoot growth and poor (nodule-free) root growth (Fig. 5). The untreated cuttings of two pots, kept in the high-temperature cabinet for the first 3 weeks, had two to three vegetative buds per cutting expanded (Fig. 6). These cuttings were then transferred to the low-temperature cabinet, where there was an increase in root development and a reduction in the rate of shoot expansion during the next 3 weeks (Fig. 7). At the end of the experiment the roots were nodulated and much more extensive than those roots produced by cuttings held at the low temperature for the whole period (Fig. 8).

Effect of Application of Gibberellic Acid to Tops of Cuttings.—Cuttings in two pots in the low-temperature cabinet were treated weekly all over with a solution of gibberellic acid (100 μg/l) in the form of a fine spray. Vigorous root and shoot growth occurred and the roots were nodulated (Figs. 9 and 10). Shoot growth occurred here at the temperature range of 15–21°C at which vegetative buds did not expand on untreated cuttings.

Effect of Application of Indolylacetic Acid to Bases of Cuttings.—Cuttings in two pots in the high-temperature cabinet were treated, once before planting, with 1% indolylacetic acid in the form of lanoline paste over the surface of the basal denuded portion. Observations similar to those for cuttings sprayed with gibberellic acid in the low-temperature cabinet were recorded, i.e. both root and shoot showed good growth (Figs. 11 and 12). Root growth here occurred at the temperature range of 21–28°C at which root development was very poor on untreated cuttings.

Figs. 1–12.—Cuttings and root systems of *P. lawrencei* showing the effects of low and high temperatures and growth substances on root and shoot growth. Cuttings illustrated are "average" samples from their treatment programme. Figs. 1 and 2, untreated cuttings after 9 weeks at the low (Fig. 1) and the high (Fig. 2) temperature. $\times 0 \cdot 45$. Figs. 3–5, untreated cuttings after 3 weeks at the low temperature (Fig. 3) and after transfer to the high temperature for 3 (Fig. 4) and 6 (Fig. 5) weeks. $\times 0 \cdot 9$. Figs. 6–8, untreated cuttings after 3 weeks at the high temperature (Fig. 6) and after transfer to the low temperature for 3 (Fig. 7) and 6 (Fig. 8) weeks. $\times 0 \cdot 9$. Figs. 9 and 10, whole cutting (Fig. 9) and central portion of nodulated root system (Fig. 10) after 9 weeks at the low temperature. Cutting sprayed weekly with gibberellic acid solution. $\times 0 \cdot 45$ and $3 \cdot 75$ respectively. Figs. 11 and 12, whole cutting (Fig. 11) and central portion of nodulated root system (Fig. 12) after 9 weeks at high temperature. Cuttings treated once before planting with indolylacetic acid–lanoline paste (arrow). $\times 0 \cdot 45$ and $3 \cdot 75$ respectively.

Conclusion

While the factors influencing root and shoot development are undoubtedly complex and imperfectly understood, it is generally considered that specific substances are concerned with the initiation of adventitious root primordia and that the major sources of these substances are the leaves, from which the substances are translocated to the bases of the cuttings (Sinnott 1960). The observations presented here show that, in spite of vigorous shoot growth at the high temperature, root growth was poor; but that when cuttings devoid of root primordia at the end of the first 3 weeks at high temperature (Fig. 6) were transferred to the low-temperature cabinet, in spite of little further shoot growth, there was vigorous development of nodulated roots. Possible explanations for this behaviour of *P. lawrencei* cuttings under the higher temperatures are either that an adequate supply of the root-inducing substance or substances was not formed in the leaves, or that the substance or substances were destroyed or inhibited. The production of roots at higher temperatures by cuttings treated with indolylacetic acid–lanoline paste could be interpreted as evidence in support of these suggestions. Indolylacetic acid here extends upward the temperature range for root development, perhaps by replacing the root-inducing substance or substances inhibited in production or destroyed at higher temperatures.

The present study also shows that *P. lawrencei* buds, dormant at low temperatures, expanded at high temperatures. However, the buds were induced to expand at the low temperature by spraying them with a solution of gibberellic acid, which here extended downward the temperature range for bud-break. The subsequent increase in root development might have resulted from some effect of gibberellic acid on the growth of the plant as a whole, possibly as a result of increased production of the root-inducing substance or substances by the resulting larger shoot. These results are consistent with the findings of earlier workers such as Morgan and Mees (1956, 1958), Wittwer and Bukovac (1957), Leben, Alder, and Chichunk (1959), and Biddiscombe, Arnold, and Scurfield (1962), who showed that gibberellic acid spray induced some plants to grow at temperatures lower than those usually required for the growth of untreated plants.

The appearance of nodules under the conditions for good root growth (Figs. 9 and 11) is in harmony with the conclusion of the earlier study (Khan 1967) that nodules are a normal morphological feature of the podocarp root system. The present study suggests that root development depends on the accumulation of sufficient root-inducing substance or substances, and that the time required varies with the conditions. This is borne out by the fact that on removal from the chambers, at the end of 4 months, all cuttings bore nodulated roots, even those kept under conditions apparently unfavourable for root development. It is intended to study this problem further from this point of view.

I am much indebted to several colleagues and especially to Dr. I. V. Newman, Dr. P. G. Valder, Professor M. G. Pitman, and Mr. P. Martin, all of this Department, and Dr. J. H. Palmer, School of Biological Sciences, University of New South Wales, for helpful discussion and advice, and to Professor S. Smith-White for the facilities provided. I would also like to thank Mr. B. Lester for his assistance in the preparation of photographic negatives for the illustrations used.

References

Bergersen, F. J., and Costin, A. B. (1964).—*Aust. J. biol. Sci.* **17**, 44–8.
Biddiscombe, E. F., Arnold, G. W., and Scurfield, G. (1962).—*Aust. J. agric. Res.* **13**, 400–13.
Khan, A. G. (1967).—*Nature, Lond.* **215**, 1170.
Leben, C., Alder, E. F., and Chichunk, A. (1959).—*Agron. J.* **51**, 116–17.
Morgan, D. G., and Mess, G. C. (1956).—*Nature, Lond.* **178**, 1356–7.
Morgan, D. G., and Mess, G. C. (1958).—*J. agric. Sci.* **50**, 49–59.
Sinnott, E. W. (1960).—In "Plant Morphogenesis". p. 391. (McGraw-Hill Book Co.: New York.)
Wittwer, S. H., and Bukovac, M. J. (1957).—*Q. Bull. Mich. St. Univ. agric. Exp. Stn* **39**, 682–6.