

OBSERVATIONS ON ESTABLISHMENT AND EARLY SHOOT EMERGENCE OF *VISCUM MINIMUM* (VISCACEAE)

J. KUIJT

Department of Biological Sciences, University of Lethbridge, Lethbridge, Alberta T1K 3M4,
Canada

SUMMARY

Viscum minimum Harvey (Viscaceae), a parasite limited to very few succulent species of *Euphorbia*, has a germination pattern which closely resembles that of *Arceuthobium* of the same family, in that germination is cryptocotylar, the shoot apex eventually aborting. As in *Arceuthobium*, the great majority of aerial shoots are produced from the endophyte but, unlike that genus, aerial shoots may in *V. minimum* also be produced from the margin of the haustorial disk, or even from directly below it. *V. minimum* is heterophyllous.

1. INTRODUCTION

The seedlings of Viscaceae have received little careful attention in different parts of the world, perhaps in part because of their slow rate of growth and rather uniform general appearance. Germination is said to be of a phanerocotylar type except in the highly reduced, squamate genus *Arceuthobium* (KUIJT 1969). In other genera, seedlings are believed to withdraw the cotyledons eventually, these then becoming photosynthetically active even though they are very small. The hypocotyl-radicle complex is often bright green, the latter eventually making contact with the host surface and developing a haustorial disk. It is from this disk that haustorial penetration is achieved. In *Arceuthobium*, the epicotylar apex – indeed, all parts external to the host – eventually die without having been withdrawn from the endosperm, and all aerial shoots thus originate adventitiously from the endophytic system. The situation in the similarly reduced, squamate genus *Korthalsella* is not clear. STEVENSON (1934) describes and illustrates seedlings of two New Zealand species, but her account leaves a number of questions unanswered. This is especially true for the so-called ‘cotyledonary collar’ possibly indicating a nearly cryptocotylar condition while the shoot apex has apparently already started to develop. The shoot-emergence pattern of some other species of *Korthalsella* may well show similarities to that of *Arceuthobium* in that the endophyte almost certainly generates some shoots; the larger species, however (as those of Hawaii), are single-stemmed plants obviously developing in the standard fashion. Beyond these two genera, the seedlings of *Dendrophthora*, *Notothixos*, and *Ginallia* have not been described for any species, and in *Viscum* and *Phoradendron* illustrations are available for only very few species

(THODAY 1951, VISSER 1981, WEBER 1978, MAY 1971, GLIMCHER 1938, TUBEUF 1923).

The smallest member of the large Old World genus *Viscum* is *V. minimum* Harvey, said to be restricted to the succulent *Euphorbia polygona* Haw. and *E. horrida* Boiss. in the eastern Cape region (HORWOOD 1972, WIENS & TOELKEN 1979). Its similarity to the smaller species of *Arceuthobium* has been a matter of frequent comment and, in fact, recently led to the question of whether, as in that genus, phloem might be absent in the species. This has turned out to be not so, the phloem being comparable in this respect to that of the leafy *V. album* L. (OLSON & KUIJT 1985). A brief, recent study has also dealt with the origin of the endophyte in this plant (OLSON & KUIJT 1986).

2. MATERIALS AND METHODS

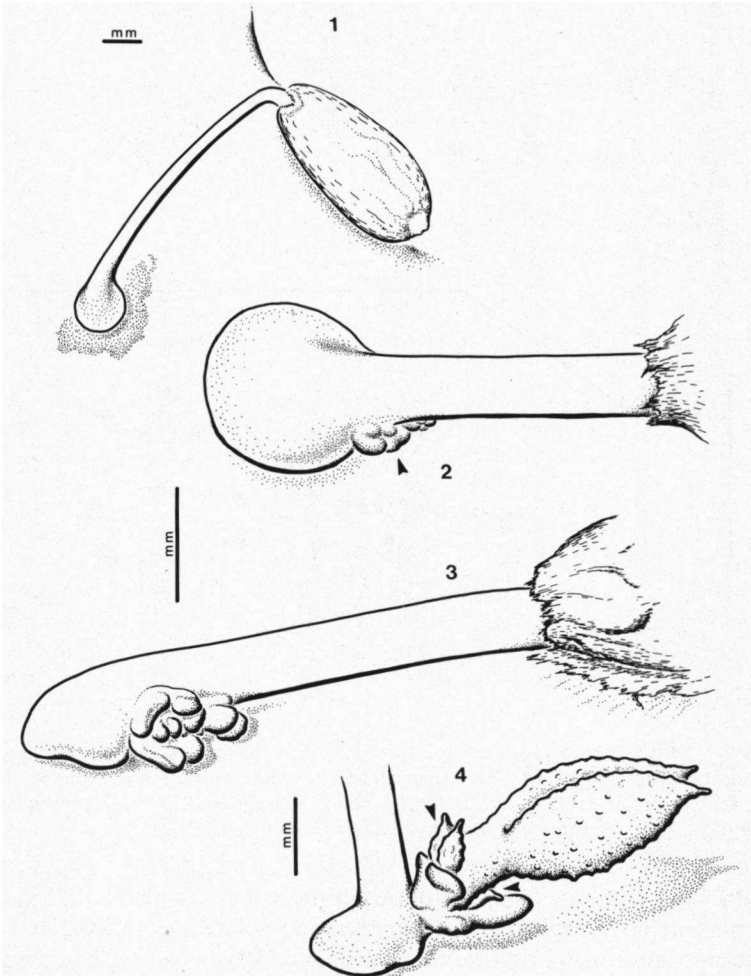
Ripe fruits were harvested from a mature plant originally obtained from the Botanical Garden at Copenhagen. Seeds were placed on several potted individuals of *Euphorbia meloformis* Ait. and *E. obesa* Hook. f. during the first week of October (1983) and kept on an east-facing window under standard room temperatures and natural illumination. No water was applied to the surface of the seed at any time.

3. OBSERVATIONS

Germination proceeded almost immediately, as evident from the beginning of elongation of the radicle within a couple of days of sowing. The initial growth rate was variable, but in some cases very rapid. In two seeds, slender radicles 8 and 10 mm long had formed in approximately three weeks (*fig. 1*). The blunt, clavate radicles tend to grow straight for a while, eventually forming a light green haustorial disk at the tip. When such a disk, now adhering to the host surface, is present the radicle frequently extends at an abrupt angle to that portion of the embryo which is still enclosed by the endosperm (*fig. 1*). This may indicate that the germinating radicle reacts to a certain, directional set of stimuli from the very beginning of its growth. The disk reaches 1.5 mm in diameter.

Even though early growth was rapid, it was not until early July 1984 that further change was observed. During the intervening 7–8 months (seeds were sown in October, 1983), the radicles remained bright green and turgid. The subsequent formation of the intrusive organ has been detailed by OLSON & KUIJT (1986).

The first sign of change was the appearance of irregular, callus-like cushions on the margin, or on the convex, upper surface of the haustorial disk of several seedlings (*figs. 2–7*). In each case, these protuberances did not undergo any further development, but remained fresh and turgid. In the fall of the same year



Figs. 1-4. Four consecutive stages in the germination and establishment of *Viscum minimum*. Fig. 1. A seedling 3 weeks after sowing; haustorial disk fully formed. Fig. 2. A seedling of 9 months old showing a callus cushion (arrow) being formed on the edge of the disk. Fig. 3. Same, at 11 months. A pair of leaf primordia is visible in the center of the larger callus cushion on the left. Fig. 4. Same, at nearly 15 months. The leaf primordia visible in *fig. 3* have now grown into a pair of young, apiculate leaves. Additionally, two primordial shoots are visible at the base of the first shoot (arrows).

(early September), a pair of minute leaf primordia had become evident in the center of each of two or three such irregular cushions, often flanked by one or more smaller, similar developments (*fig. 3*).

The shoots thus produced were not, however, necessarily the first ones, nor the most rapid in their growth. Sometime before September 1984, a number of small, brownish necrotic spots became visible on the smooth, otherwise glaucous green host surface. The spots tended to form transverse series across the



Figs. 5 and 6. Seedlings and shoot emergence patterns of two plants of 11 months old. The dashed lines represent the ribs of the host. The blisters referred to in the text, most of which have developed fissures, are represented in black. At s, in fig. 6, a rather large cluster of shoots is developing. Both seedlings show small shoots originating from the disks (arrows). See also fig. 7.

host ribs (figs. 5 and 6) possibly following the main vascular bundles of the host, and were of various sizes. A rather remote, isolated spot occasionally also made its appearance, in one instance on precisely the opposite side of the globular host stem. Many of the blisters eventually cracked open and, from these fissures, irregular callus cushions emerged essentially like those described from the disks. Similarly, many of these cushions eventually generated regular aerial shoots, usually in small clusters (figs. 8 and 9). Aerial shoots are thus not, as often indicated for *Arceuthobium*, surrounded by a discrete, ring-like 'basal cup' but rather by irregular protuberances; nor do the shoots themselves break through the host surface. One or more of the shoots thus generated from the endophyte, in some instances, grew more rapidly than nearby disk shoots belonging to the same individual (fig. 6 s). Additionally, it was noted in several cases that a whorl of shoots emerged from directly below the disk, even when the latter bore a primordial shoot on its upper surface, as shown in fig. 7. Eventually, this resulted in a rather dense cluster of flowering shoots.

It is important to realize that the radicle during all these developments underwent no visible changes. The thin, dry viscin shell by now was partly empty,

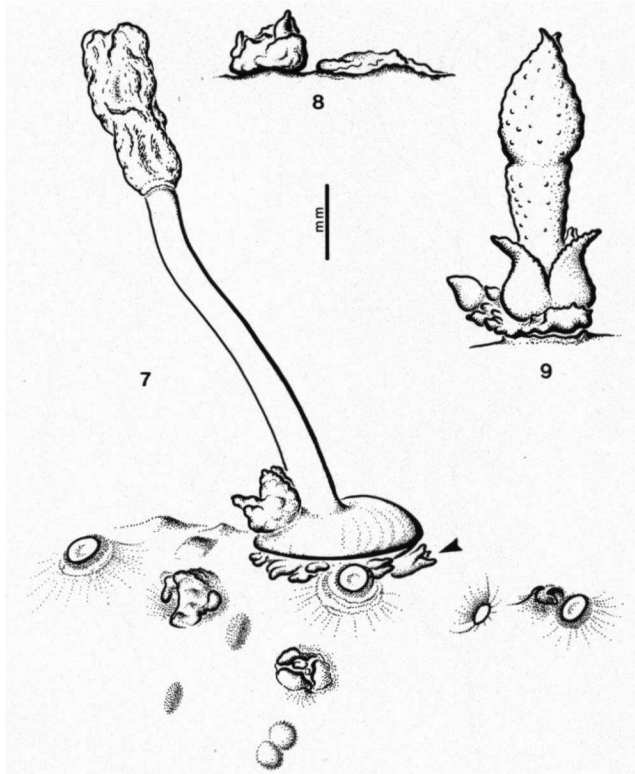
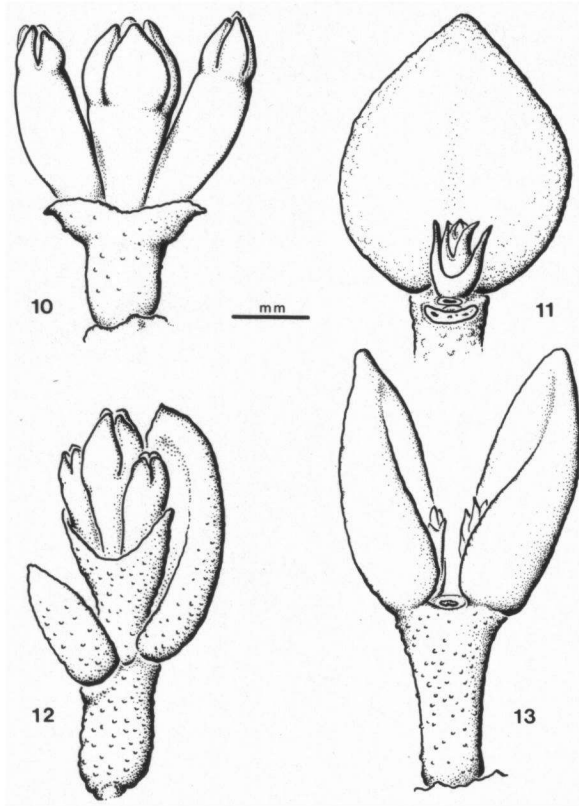


Fig. 7. Close up of the erect seedling illustrated in *fig. 6*, to show primordial shoots emerging below the haustorial disk. Fig. 8. Two emergent callus cushions. Fig. 9. Young emergent shoot comparable to the disk shoot in *fig. 4*, at the same age.

the endosperm within it being completely shrivelled up. Although in one or two instances the radicle became more or less erect, the cotyledons were never withdrawn as would normally be expected. I am assuming that this loss of the epicotylar pole also occurs under more natural conditions, since the development of the remainder of the plants in this period seemed healthy.

Following the above sequence of events, more blisters appeared and produced shoots, now in apparently random positions. In older plants, such as the ones illustrated in HORWOOD (1972), a pattern of shoot emergence can no longer be identified.

While the plant is in most cases aphyllous (*fig. 10*), as stated in the literature, it is also frequently heterophyllous. In the latter case, leaves which are not at all scale-like are formed on larger shoots. They are ovate, fleshy, and about 3×4 mm (*figs. 11-13*). They are slightly constricted at the base, along which constriction they are presumedly dropped eventually. In the axils of some of these normal leaves secondary inflorescences may be formed (*figs. 11 and 13*), but it is not known whether this is a regular phenomenon.



Figs. 10–13. Shoots showing details of heterophylly. Fig. 10. The most common, squamate shoot (inflorescence). The central flower is male, the two lateral ones female; the entire shoot consists of a single internode. Fig. 11. A larger shoot with two internodes, the upper node squamate, the lower one bearing one normal leaf and one intermediate one. Fig. 12. A similar shoot as that in fig. 11, the upper internode and flowers having fallen away. Two lateral shoots (inflorescences) are developing in the leaf axils. Fig. 13. The same shoot as that in fig. 12, with one leaf and inflorescence removed.

4. DISCUSSION

The most tempting comparison to be made with the life cycle of *V. minimum* is undoubtedly with that of *Arceuthobium*, the main elements of which have already been sketched in the Introduction. The same, highly advanced pattern of germination and establishment is also seen in the Chilean mistletoe *Tristerix aphyllus* (DC.) Barlow & Wiens (Loranthaceae), as first described by REICHE (1904) and more recently by MAUSETH et al. (1984). The latter species, even though belonging to a different family, represents a remarkable convergence to *V. minimum*, being both aphyllous and limited to leafless succulents, in this case various large, columnar cacti (KUYT 1969). The *Arceuthobium* pattern is

diametrically opposite to what is here, for the sake of brevity, called the normal pattern.

Germination and establishment in *V. minimum* thus represents an intermediate situation between the *Arceuthobium* and normal patterns. The epicotylar pole survives for a year or more after haustorial penetration takes place, but undergoes no further differentiation, and eventually perishes. *V. minimum* thus has evolved the *Arceuthobium* pattern independently in the sense that all aerial shoots are adventitious, the shoot apex having apparently lost its potential to differentiate. Beyond *Arceuthobium*, *V. minimum* is the first known member of Viscaceae with cryptocotylar germination. However, the evolution of *V. minimum* has not gone quite as far as *Arceuthobium* in that, while most shoots are derived from the endophyte, some are also generated from both the top of the haustorial disk and from what appears to be the upper shaft of the intrusive organ. In other words, all haustorial tissues still possess a potential for shoot production. It is perhaps not going too far to suggest that *Arceuthobium* may have passed through a similar, transitional stage, but has subsequently restricted its adventitious shoots to the endophytic tissues.

It is interesting, in this context, that a single instance has recently been discovered of shoot production from the margin of the haustorial disk in the American Loranthaceous genus *Ixocactus* (KUIJT, personal observation). This monotypic genus is undoubtedly the most highly advanced one among the small-flowered neotropical group of that family. In this case there is no vegetative reproduction from the endophyte. Shoot production from the disk seems to be common, if not standard, and its effect can be clearly seen in the branching pattern of the mature plant.

It is a striking fact that each of *V. minimum*, *Ixocactus*, and *Arceuthobium* is aphyllous. It must not be thought, however, that a direct relationship always exists between the production of adventitious shoots and the squamate condition. There are, indeed, additional squamate species where this is so, as in *Phoradendron juniperinum* Engelm. of western North America, and in *Dendrophthora cupressoides* (Macf.) Eichler of the Caribbean. In both these species, and certain others, many shoots originate from the endophyte. However, there are numerous, other squamate species where only the primary shoot develops; examples which may be mentioned are *Dendrophthora squamigera* (Benth.) Kuntze, *D. lueri* Kuijt, many squamate *Viscum* species, and at least the larger species of *Korthalsella*. Conversely, vegetative reproduction from the endophyte is a well known phenomenon in some leafy species of *Phoradendron* (e.g., *P. villosum* Engelm.) and occasionally in *Dendrophthora* (e.g., *D. costaricensis* Urban). In *Viscum* itself, such regeneration is known to take place in the leafy *V. album* L., where even shoot production from the haustorial disk was described and illustrated more than 60 years ago (TUBEUF 1923, *figs.* 122–123).

A final point about the germination of *V. minimum* is, in retrospect, a rather obvious one: surface water is not necessary for germination. As stated above, care was taken not to have water touch the seedling. The importance of this point lies in the fact that there are many conflicting ideas in the literature on

the possible functions of viscin, one suggestion of which is the absorption of water to be passed on to the seedling. While I cannot comment on the potential of viscin to do so (except that, during much of the development sketched above, the dried viscin is not in contact with living tissues) this function is clearly not important. The same may well be true for other mistletoes with similarly xeric habitats, such as *Phoradendron californicum* Nutt. in the deserts of the south-western United States and adjacent Mexico, where precipitation is very rare and irregular.

ACKNOWLEDGEMENTS

I am obliged to Dr. R. Dahlgren of the Botanical Garden at Copenhagen for supplying me with a plant of *V. minimum*. The Natural Sciences and Engineering Research Council of Canada supported the work financially.

REFERENCES

- GLIMCHER, J. (1938): The germination of *Viscum cruciatum* Sieb. *Pol. J. Bot. Jer. Ser. Jerusalem* **1**: 103–105.
- HORWOOD, F. K. (1972): Exotic xerophytes – 8. Two parasites of Euphorbia: *Viscum minimum* and *Hydnora africana*. *Nat. Cact. & Succ. J.* **27**: 96–98.
- KUYT, J. (1969): *The biology of parasitic flowering plants*. University of California Press, Berkeley and Los Angeles.
- MAUSETH, J. D., G. MONTENEGRO & A. M. WALCKOWIAK (1984): Studies of the holoparasite *Tristerix aphyllus* (Loranthaceae) infecting *Trichocereus chilensis* (Cactaceae). *Can. J. Bot.* **62**: 847–857.
- MAY, D. S. (1971): The role of populational differentiation in experimental infection of *Prosopis* by *Phoradendron*. *Amer. J. Bot.* **58**: 921–931.
- OLSON, A. R. & J. KUYT (1985): Sieve elements in the morphologically reduced mistletoe, *Viscum minimum* Harvey (Viscaceae). *Amer. J. Bot.* **72**: 1220–1224.
- & — (1986): The origin of the endophyte in *Viscum minimum* Harvey (Viscaceae). *Can. J. Bot.* (in press)
- REICHE, K. (1904): Bau und Leben der chilenischen Loranthacee *Phrygilanthus aphyllus*. *Flora* **93**: 271–297.
- STEVENSON, G. B. (1934): The life history of the New Zealand species of the parasitic genus *Korthalsella*. *Trans. & Proc. Roy. Soc. N. Z.* **64**: 175–190, Pl. 24–31.
- THODAY, D. (1951): The haustorial system of *Viscum album*. *J. Exp. Bot.* **2**: 1–19.
- TUBEUF, C. VON (1923): *Monographie der Mistel*. R. Oldenbourg, Munich & Berlin.
- VISSER, J. (1981): *South African parasitic flowering plants*. Juta, Cape Town.
- WEBER, H. C. (1978): *Schmarotzer – Pflanzen die von anderen leben*. Belser Verlag, Stuttgart.
- WIENS, D. & H. R. TOELKEN (1979): Viscaceae. In: O. A. LEISTNER (ed.); *Flora of Southern Africa* **1**: 43–59.