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ANATOMICAL STATUS AND ROOTING OF *EUONYMUS JAPONICA L.* CUTTINGS

B. BOGDANOV AND P. ALEXANDROV

Higher Institute of Forestry Engineering
Sofia, Bulgaria

The vegetative propagation of the evergreen shrub, *Euonymus japonica* Thunb., plays an important role in the production of decorative material. In Bulgaria this species is grown mainly along the Black Sea coast but it may also be grown in other regions of the country where there is a warmer climate. We are looking for other ways for successful propagation of the subject through rooting cuttings. These are usually taken from one-year-old shoots.

The main task of our study is to analyse the anatomical structure of the current (one-year-old) shoot of *E. japonica* since it has a definite importance for the emergence and forming of the root system of the cuttings with respect to rooting during different times of the year.

MATERIALS AND METHODS

Initial mother plants were 5 and 15-year-old groups of shrubs. The one year old shoots collected from them for anatomical analysis were fixed in FUS. The cuts were performed on Reichert's microtom. The colouring of the preparations is carried out with hematoxilin-eosin. The anatomical observations conducted with the aid of a light microscope on three cuts of the shoots — base, middle, and below the top bud.

Softwood cuttings were cut the middle of each month from a shoot with a length of 8 to 10 cm (three to four nodes) and reducing the leaf mass by about 70%. the cuttings were rooted in cold frames and heated greenhouses in a mixture of washed river sand and perlite in the proportions of 2:1.

RESULTS AND DISCUSSION

During the growing period *E. japonica* forms three flushes of growth. The first is from the middle of March (the bursting of buds) until the second half of May. The second one is from late May until the middle of July; the third one is from the beginning of August until the end of the growing period. The first and the second growth are almost equal but the third is more reduced. This is not strong growth and may be destroyed by frost during the winter. The total length of the shoots is about 40 cm.

The investigations show that at the end of the winter the epidermis is clearly outlined around the whole shoot (Figure 1) by a row of cells which have a thickened tangential wall.

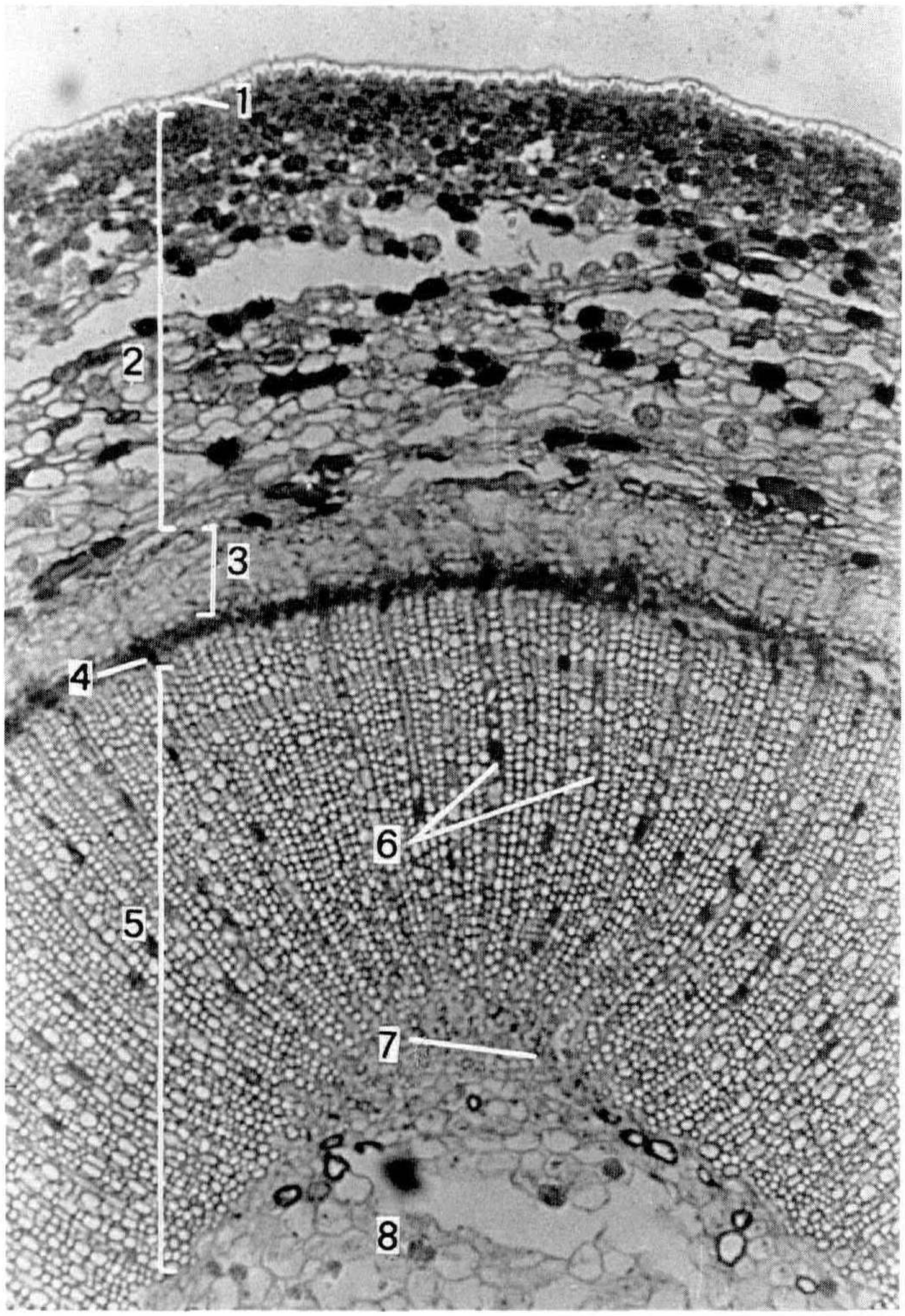

Figure 1. Cross section of *Euonymus* shoot before rooting — (1) epidermis; (2) primary bark; (3) phloem; (4) cambium; (5) xylem; (6) medullary rays; (7) perimeduler zone; (8) pith

The primary bark consists of colenchyma and a parenchyma tissue. Under the epidermis there are 4 to 5 rows of thin-walled colenchyma cells filled with chloroplasts. Many of the parenchyma cells contain tannin; others have big cavities with some of the cells empty. Small intercellular structures are observed. The parenchyma cells of the primary bark are clearly demarked by intercellular spaces. The phloem is without primary fibres, but contains fairly discernible medullary rays.

The cambial zone is clearly revealed due to the activity of the cambium at that time.

E. japonica wood is diffused, porous with vessels, wood fibres, and medullary rays. The vessels are round with five walls and have a mean diameter of about 20 microns. The medullary rays are clearly differentiated, being a single row about 190 microns, situated at intervals of 2 to 6 but most frequently at intervals of 4 rows of wood fibres.

The perimedular zone of the pith is slightly differentiated by 1 to 2 rows of cells. Localised secretory cells are found around the wood.

The pith is composed of large rounded or multiangular parenchyma cells with the presence of starch, mainly near the wood. Large multiangular spaces and a considerable number of large-sized cavities are formed at many places on it.

Table 1. Correlation between the different tissues in the base, the middle, and the top of the one-year-old shoots.

Location of tissue	Pith (microns)	Xylem (microns)	Phloem (microns)	Primary bark (microns)	Height of epidermal cells (microns)
Base	1788	699	151	342	88
Middle	1815	548	137	480	32
Top	1986	206	77	447	27

The data (Table 1) and the investigations show that the one-year-old shoot preserves its epidermal layer and does not develop epiderma. There are no sclerenchyma fibres in the primary bark. The conducting system of the base, middle, and top is not clearly differentiated. At the top the pith is thicker than at the base or in the middle — with bigger or smaller multiangular spaces.

After the appearance of the first 1 to 3 newly formed roots the anatomical status of the cutting (Figure 2) is characterized by an advanced stage of recovery of the tissues of the stem in the area of the wounding. On the periphery of the segment the initial epidermis before cutting is partially preserved but there are no epidermal cells near the place of cutting, i.e. they have degenerated.

The post-cutting phellogen is formed from some colenchyma cells of the primary bark in the sub-epidermal zone.

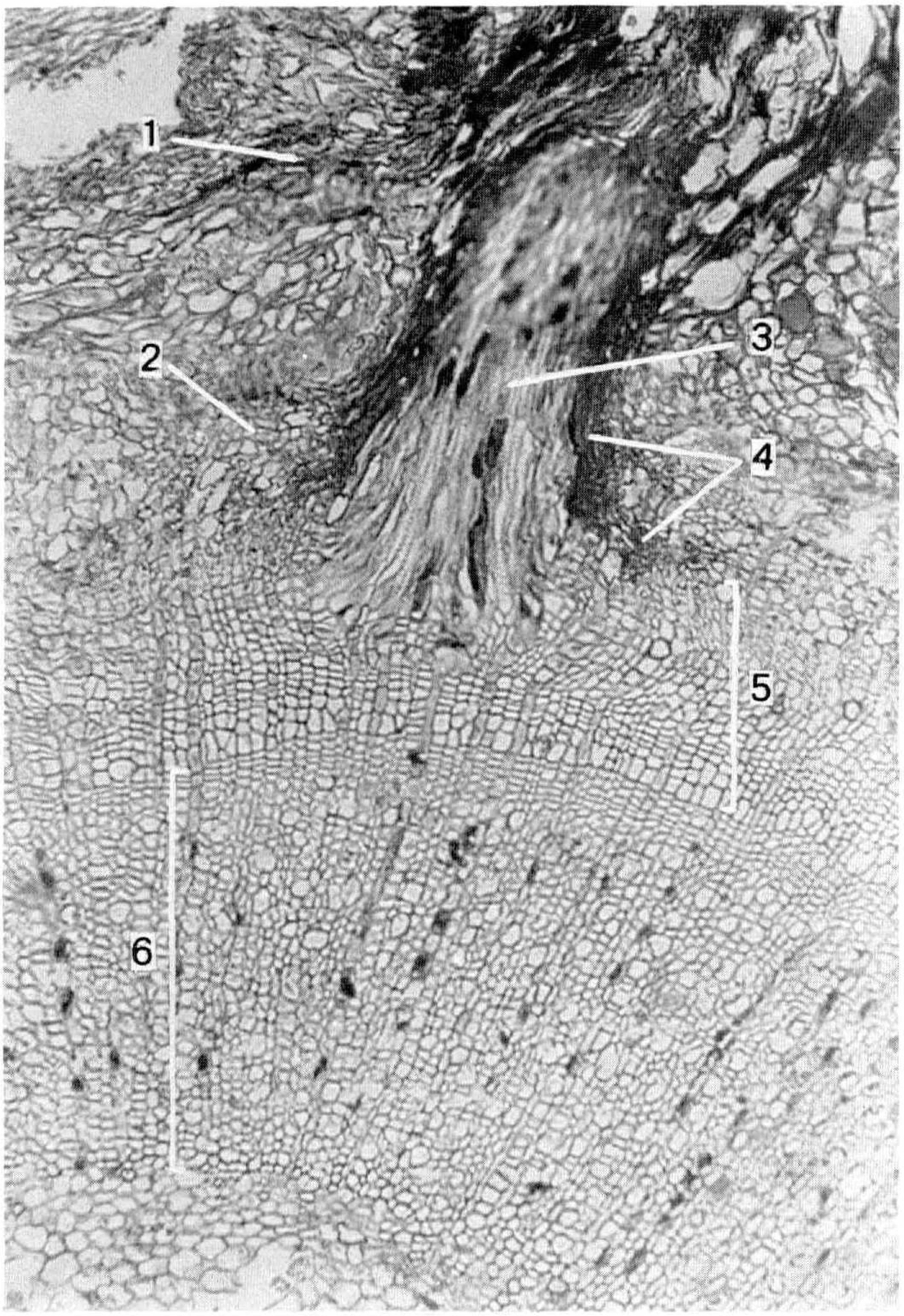

Figure 2. Cross section of euonymus shoot after an adventitious root has formed — (1) primary bark; (2) phloem; (3) newly-formed root; (4) cambium; (5) xylem formed after rooting; (6) primary xylem.

There are 2 to 3 rows of phloem cells. The remaining part of the colenchyma cells degenerate and drop off. The wood, after cutting characterised by a pathological structure is located very near to the peridermal zone. Its cells are tangentially lengthened and convoluted. The medullary rays are rarely formed and are very short. Initial pith rays are found which look twisted and inserted in the pathological wood. The initial pith is bent; its cells are pressed and have a dark brown content. They seem to be connected to the initial wood and are actually dropping off. A regeneration of the cutting's development is taking place at this stage which is a bit chaotic in the zone of the callus, but this secures a primary physiological and structural connection which prevents penetration of air in the inner tissues.

The newly-formed roots appear in the cambial zone in the region where phloem and xylem elements are not completely differentiated as well as the increase of the woody tissues which, after planting, is 253 microns. Small bundles of conducting elements are formed at the beginning which gradually group together to form the apex of growth.

The newly-formed roots grow through the primary bark or the callus and tear the covering tissues. It is assumed that the diffused-porous type of the wood of *E.japonica* favours the formation of roots since the cambium cells possess the ability to divide during the whole period of vegetative growth.

The observations show that visible signs for the initial stage of the rooting process appear after the callus formation, about 20 days after planting, root formation (about the 40th day, bud-growing, 50th to 60th day). The rooting proceeds most quickly with cuttings obtained during the months 2, 3, 4, 5, and 6, then at a slower rate — with those cut in the 7, 8, and 9 months, and in an intermediate time — with October and November cuttings.

At an optimal regime for rooting, e.g. a temperature of 20 to 25° C and a relative humidity above 80%, all cuttings develop a good root system and there is a high rooting percentage. The best results are obtained with cuttings taken in March, with 100% rooting: 42 plants have first class roots with 375 cm total length of the root system of one cutting. The most unsatisfactory are the cuttings taken in September with 90% rooting, only 17 plants with first class roots, and 186 cm total length of the root system for one cuttings.

CONCLUSION

E. japonica cuttings manifest high regeneration capacity during the whole period of their annual growth. The rooting proceeds mainly in the zone of the xylem and cambial layer and form after rooting around the cut and in the part of the cutting covered by the substrate. Under the experimental conditions the best results were obtained with cuttings prepared in March.