THE MORPHOLOGY AND HISTOLOGY OF THE SEEDS OF STRYCHNOS NUX-VOMICA, LINN., AND ITS ADULTERANTS, STRYCHNOS NUX-BLANDA, HILL, AND STRYCHNOS POTATORUM, LINN.

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Received February 4, 1963

The morphology and histology of the seeds of Strychnos nux-vomica, Linn., Strychnos nux-blanda, Hill., and Strychnos potatorum, Linn., are described. The diagnostic features by which the two adulterants S. nux-blanda and S. potatorum can be distinguished from the pharmacopoeial drug S. nux-vomica, are noted.

VARIOUS authors have commented upon the presence of seeds of Strychnos nux-blanda, Hill, and Strychnos potatorum, Linn., in samples of Strychnos nux-vomica, Linn. Reports indicate that the amount of S. nux-vomica exported from India has more than doubled in the last 50 years, in the year 1913, for example, the quantity exported was 1,200 tons, in 1956, 2,700 tons (Chopra, 1949; Younken and Pratt, 1956). It is also apparent that adulterations of S. nux-vomica with one or both of the adulterants still persists, such samples having been investigated as recently as July, 1961 (Wallis, private communication).

With the exception of a description of the tree by Hill (1917) and a note on the histology of the seed by Small (1913) there is no record of a complete examination of the seed of S. *nux-blanda*, similarly the only accounts of the seed of S. *potatorum* are those of Basu and Kirtaker (1818) and Dymock (1885) and these do not include the histology.

It was decided that a complete examination of the two known adulterants should be made and the characters by which they may be distinguished from the seed of *S. nux-vomica*, investigated.

In the literature referring to the seed of *S. nux-vomica* there was a number of conflicting statements about the macroscopical and microscopical characters. In addition, a microscopical examination revealed certain structures which have not previously been described. A re-examination of *S. nux-vomica* seeds was therefore necessary.

Plant Materials

The seeds of *S. nux-vomica* were obtained from five commercial sources: those of *S. nux-blanda* were obtained direct from the Forest Research Institute and College, Dehra Dun, India, and the seeds of *S. potatorum* were obtained from the Conservator of Forests, Colombo, Ceylon, with a small sample from the Owens College Museum, Manchester.

Recorded Dimensions

The macroscopical measurements are recorded as four figures, those in bold type indicate the range of size between which approximately twothirds of the sample fall. These are obtained by a statistical analysis of

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not less than 500 measurements and represent the summation and difference of the standard deviation and the mean. The first and fourth figures are respectively the minimum and maximum values observed.

The microscopical measurements are recorded as three figures. The figure in bold type denotes the statistical mode of not less than 250 measurements whilst the first and third figures are respectively the minimum and maximum values observed.



FIG. 1. Strychnos nux vomica, Linn. seed. A, Surface views. B, Lateral views, of entire seeds. Strychnos nux blanda, Hill, seed. C, Surface and lateral views of seeds. Strychnos potatorum, Linn. seed. D, Surface and lateral views of entire seeds; All \times 1. m, micropyle; r, ridge; h, hilum; b, lateral ridge.

EXPERIMENTAL

Macroscopical Characters of the Dried Ripe Seed of Strychnos nux-vomica, Linn.

The macroscopical characters of the seed of S. nux-vomica are well known. The illustrations of the entire seeds in Fig. 1A and 1B serve as

a direct comparison with those of S. nux-blanda, Fig. 1C, and S. potatorum, Fig. 1D.

The dimensions of the samples examined were as follows:

Entire seed (Figs. 1A, 1B): diameter, 17.0—18.5 to 23.1—29.5 mm.; thickness, 2.2—3.5 to 5.3—7.0 mm.; weight, 0.8—1.3 to 2.1—2.7 g.

Horizontally split seed (Fig. 2A): disc shaped cavity, diameter, $13 \cdot 1$ — 17.5 to 20.0—25 mm.; thickness, $0 \cdot 1$ —0.9 to 1.5—1.9 mm.; Area of fusion of endosperm halves. Width 1.2—1.6 to 2.0—3.5 mm.

Embryo (Fig. 2D): length, $5\cdot0-6\cdot5$ to $8\cdot0-9\cdot0$ mm.; width cotyledons at widest point, $2\cdot5-3\cdot0$ to $4\cdot0-4\cdot5$ mm.; length of radicle, $2\cdot0-3\cdot0$ to $3\cdot5-4\cdot5$ mm.



FIG. 2. Strychnos nux vomica, Linn. seed. A, Horizontally split seed; D, embryo. Strychnos nux blanda, Hill. seed. B, Horizontally split seed; E, embryo. Strychnos potatorum, Linn. seed. C, Horizontally split seed. F, embryo. Seeds \times 1. Embryos \times 5. ep, epidermis; f, area of fusion of two endosperm halves; c, area of central cavity; em, embryo; cot, cotyledons; r, radicle.

Macroscopical Characters of the Dried Ripe Seed of Strychnos nux-blanda (Hill)

The overall shape of the seed approximates to an ellipsoidal disc, $16\cdot 2$ — 17.2 to $20\cdot 0$ —23.0 mm. long and $11\cdot 0$ —13.3 to $15\cdot 8$ —17.0 mm. wide. A few of the seeds are almost flat on one or both sides but the majority are concave on both upper and lower surfaces.

The margin is keel-shaped, due to the presence of a wide ridge which extends laterally from the otherwise rounded edge. Figure 1C shows the surface and lateral views of the seed. The seed is $5 \cdot 6 - 6 \cdot 1 - 7 \cdot 2 - 7 \cdot 8$ mm. thick, and weighs $0 \cdot 4 - 0 \cdot 9$ to $1 \cdot 3 - 1 \cdot 4$ g.

Situated on the margin of the seed, at the subacute end, the lateral ridge has a marked labiate protuberance which indicates the position of the micropyle. A raised ridge runs from this marginal prominence to the hilum.

The surface is buff coloured with brown-black patches and is densely covered with appressed hairs arranged along the approximate radius with the hair tip directed toward the periphery.

In the horizontally split seed, the endosperm has the same shape as the seed and is hard and horny in texture, the colour is a translucent white or greyish white.

Lying centrally and in the horizontal plane of the endosperm is a disc shaped cavity which is $15\cdot0-17\cdot1$ to $18\cdot0-19\cdot5$ mm. long, on a line running through the micropyle and the centre of the seed, and $10\cdot0-12\cdot2$ to $14\cdot1-16\cdot0$ mm. wide, on a line running through the centre of the seed and at right angles to the first dimension. In the entire seed the cavity is $0\cdot1-0\cdot6$ to $0\cdot8-1\cdot0$ mm. wide at the centre.

(The centre of the seed in the above dimensions is taken as that point positioned immediately beneath the hilum and midway between the two endosperm discs.)

In the split seed (Fig. 2B) the cavity is surrounded by the area of fusion of the two endosperm halves this extends from the edge of the cavity to the testa and is $1\cdot 1-1\cdot 5$ to $2\cdot 0-2\cdot 2$ mm. wide.

Within the cavity lies the embryo, radially directed, with the radicle in a cylindrical channel leading from the central cavity to the micropyle.

The embryo is $5 \cdot 0 - 6 \cdot 0$ to $7 \cdot 0 - 7 \cdot 5$ mm. in length, greyish white and as shown in Fig. 2E consists of two superposed straight, broadly ovate, leafy cotyledons $2 \cdot 0 - 3 \cdot 0 - 4 \cdot 0 - 5 \cdot 0$ mm. wide at their widest point showing pinnate venation with from five to seven principal veins. The radicle is $1 \cdot 5 - 2 \cdot 0$ to $2 \cdot 5 - 3 \cdot 0$ mm. in length and terete in section with a conical apex.

The dried seeds are odourless, the soaked seeds develop a slight earthy odour after 3 days. The taste is bland and mucoid.

Macroscopical Characters of the Dried Ripe Seed of Strychnos potatorum, Linn.

The seed is sub-spherical 9.5-10.0 to 11.8-13.2 mm. in diameter and 3.0-6.4 to 8.2-9.0 mm. thick, the weight is 0.41-0.52 to 0.88-1.13 g.

Some specimens show a shallow concavity on the hilar side but most are smoothly curved. The margin is rounded except for a fine ridge which, as shown in Fig. 1D divides the seeds into two almost equal parts. At one point on the margin is a small depression which marks the position of the micropyle. The hilum is situated in the centre of one of the curved surfaces of the seed and occurs as a small circular cavity.

The surface is pale buff in colour, a few specimens exhibiting shiny black patches which, on examination, proved to be fragments of the pulp of the berry.

The seeds are covered with closely appressed hairs which are set radially with their tips directed towards the periphery. This trichomatous layer is enclosed by an adherent endocarpic layer which gives to the seeds a finely rugulose appearance.

The horizontally split seed shows that the endosperm has the same shape as the seed and is hard and horny in texture. The colour is a translucent white.

Lying centrally and in the horizontal plane of the endosperm is a disc shaped cavity less than 0.1 mm. wide at the centre and 8.0-9.0 to 11.1-12.0 mm. in diameter. In the split seed (Fig. 2C) this is surrounded by the area of fusion of the two endosperm halves this extends from the edge of the cavity to the testa and is 0.25-1.00-1.25 mm. wide.

Within the cavity lies the embryo, radially directed, with the radicle in a cylindrical channel leading from the central cavity to the micropyle.

The embryo is $3\cdot0-3\cdot5$ to $4\cdot0-4\cdot5$ mm. in length, greyish white in colour and (Fig. 2F) consists of two superposed straight, narrowly ovate, leafy cotyledons, $1\cdot5-2\cdot0-2\cdot5-3\cdot0$ mm. wide at their widest point, showing palmate venation with from three to five principal veins. The radicle is $0\cdot75-1\cdot0-1\cdot5$ mm. long, terete and shortly clavate.

The dried and soaked seeds are odourless, the taste is bland.

Histology of the Seed of S. nux-vomica

The principal histological characters of S. nux-vomica are well known; this re-examination revealed certain characters not previously reported. These are reported in detail with the microscopical dimensions of all the structures.

The testa consists of one integument, having two cell layers, an outer trichomatous epidermis and an inner layer of ground tissue Wallis (1951).

Trichomatous epidermis (Fig. 3A ep.). The dimensions of the base of the trichomes are R, 65-80-100 μ , T, 40-60-85 μ , L, 45-65-87 μ . In longitudinal sections the anticlinal walls are strongly thickened and are pierced by tubular pits which are sometimes branched. Transverse sections of the upper region of the base (Fig. 3B) and the central part of the base (Fig. 3C) are shown.

A transverse section of the basal periclinal wall (Fig. 3D) indicates a slight tapering of the trichome base and a decrease in the sinuosity of the anticlinal walls. The section has a retiform appearance caused by the irregularly protuberant nature of the cell wall.

The trichome limb is composed of 6-10-15 ribs, 3-10-12 μ in diameter (Fig. 3F), the entire limb is 18-24-45 μ in diameter and 600-950-1290 μ in length. The ribs exhibit a varying degree of anastamosis as illustrated in the partially disintegrated trichomes in Fig. 3E, Fig. 3G a-f shows the variation that exists in the anastamosis of the trichome tips.

On the *rounded acute margin* of the seed the trichomes are not appressed but are arranged vertically directed away from the seed centre.



FIG. 3. Strychnos nux vomica, Linn. seed. A, Longitudinal radial sections of testa. B, Transverse section of upper half of trichome base. C, Transverse section of lower half of trichome base. D, Transverse section of basal periclinal wall of trichome base; E, Trichome with partially disintegrated tip; F, Transverse section of trichome limb; All \times 100. G, a to f; trichome tips showing varying degrees of anastamosis, \times 200. ep, epidermis; nl, ground tissue; r, ribs on secondary wall; b, basal periclinal wall.



FIG. 4. Strychnos nux vomica, Linn. seed. A, Longitudinal tangential section of testa in region of ridge. B, Surface view, upper epidermis of cotyledon. C, Surface view, lower epidermis of cotyledon. D, Transverse section of midrib and lamina of cotyledon; All \times 100. E, Diagram to show distribution of tissue in hilar region. Strychnos potatorum, Linn. seed. F, Diagram to show distribution of tissue in hilar region. e, epidermis; se, subepidermal layer; nl, ground tissue; ue, upper epidermis; le, lower epidermis; v, vascular tissue; r, raphae; end, endosperm; en, endocarp.

Sections were taken as follows:

 $\mathbf{R} = \mathbf{A}$ longitudinal section made in a radial direction perpendicular to the surface. $\mathbf{T} = \mathbf{A}$ transverse section made parallel to the surface. $\mathbf{L} = \mathbf{A}$ longitudinal section perpendicular to the surface at a right angle to the radius.

The ground tissue of the testa has functioned as a nutrient layer (Wallis, 1951) and is represented by a brown band of flattened parenchyma, R, 12–25–32 μ (Fig. 3A, nl.) (Fig. 4A, nl.). In transverse section they appear as ill defined polygonal cells T, 29–55–70 μ (Fig. 3D, nl.).

The narrow ridge which runs from the hilum to the micropyle is composed of an epidermal and a sub-epidermal layer. The trichomes of the epidermis in this region (Fig. 4A, ep.) are similar to those covering the remainder of the seed but the hairs are irregularly arranged with more conical bases and partially chordaceous limbs. The sub-epidermal cells (Fig. 4A, se.) are lignified and appear as extensions of the trichome bases. The anticlinal walls are ribbed and are pierced by longitudinal slit-like pits. In longitudinal section they appear to be reticulate in nature, the lumen being traversed by anastamosing and branching lignified rods. The height varies according to their position in the ridge. Those at the highest point of the ridge are $182-220-280 \mu$, decreasing gradually towards the lateral limits of the ridge.

The hilum is marked by the trichome limbs being raised from the appressed to the vertical position (Fig. 4E, ep.). The vertical trichomes surround the raphae (Fig. 4E, r) which is a narrow vascular strand composed of ruptured spiral vessels. This vascular strand traverses the epidermis and then extends into the ground tissue for a short distance on all sides.

The endosperm of the seed is found adjacent to the ground tissue of the testa, and is composed of simple parenchyma (Fig. 6A). The outermost layer consists of radially elongated polyhedral cells, R, $33-52-68 \mu$; T, $10-16-20 \mu$; L, $10-18-20 \mu$; the outer walls of which are thickened. Within this outer layer are two to three layers of cells which are almost isodiametric, R, $25-40-45 \mu$; T, $20-30-40 \mu$; L, $20-35-45 \mu$. The remainder of the endosperm is composed of large polyhedral cells, R, $42-60-100 \mu$; T, $35-55-75 \mu$; L, $35-60-75 \mu$. The cell walls are $20-25-30 \mu$ thick. The cells of the endosperm show fine perforations in the walls; these are described by Tschirch and Oesterle (1900) and other authors as containing the plasmodesmal strands which unite the protoplasts of neighbouring cells.

The cell content is an oil plasma the aleurone grains of which are irregularly shaped, show one or more globoids and are $15-30-50 \mu$ in diameter.

The embryo is composed almost entirely of small, thin walled parenchyma. In transverse section (Fig. 4D) the lamina is up to 180 μ thick and shows partial cellular differentiation into upper and lower epidermis and vascular tissue. In surface view the upper epidermal cells (Fig. 5B) have almost straight anticlinal walls and are 20–25–40 μ long and 8–

15—20 μ wide. The lower epidermal cells in surface view (Fig. 4C) are polygonal in outline and isodiametric, 5—10—17 μ . The cells of the embryo are filled with fixed oil and contain a few scattered aleurone grains.

Histology of the Seed of Strychnos nux-blanda, Hill

The testa of the seed consists of one integument having two layers, an outer trichomatous epidermis and an inner layer of ground tissue.

The trichomatous epidermis is composed of strongly lignified cells having a short prismatic base extended to form an appressed hair which is directed at an angle to the base and radiates towards the margin of the seed (Fig. 5A, ep.).

The dimensions of the base are R, 90–100–116 μ ; T, 52–63–80 μ ; L, 50–69–80 μ . In longitudinal section the anticlinal walls appear thickened and are pierced by simple ovoid pits and scattered groups of fine slit-like pits. The simple ovoid pits are arranged with the long axis in a vertical direction; the slit-like pits are irregularly arranged and appear on the cell wall as fine striations in surface view and as serrulations in section.

The cell lumen is sometimes traversed by lignified rods which have their origin in the basal periclinal or anticlinal walls and terminate in the trichome limb. The apex of the base is irregularly tapered and the basal periclinal wall is coarsely papillate, the protuberances extending into the ground tissue.

A transverse section of the upper region of the base (Fig. 5B) shows the anticlinal walls to be slightly sinuous in outline with small circular structures and protuberances on the secondary cell wall, these latter structures are the basal portions of the trichome ribs (described later). The central region of the trichome base shows, in transverse section (Fig. 5C), a marked decrease in the number of protuberances on the secondary wall. The basal periclinal wall has a fine retiform appearance in transverse section (Fig. 5D) caused by the coarsely papillose nature of the wall.

The circular structures and protuberances found in the transverse section of the upper region of the base are extended longitudinally as lignified ribs which are drawn into a fascicle to form the trichome limb. Each limb is composed of 6-10-12 ribs, $4-8-16 \mu$ in diameter, with a crenulate outer margin surrounding a small lumen (Fig. 5G).

The entire trichome limb is $16-20-40 \mu$ in diameter and $700-1100-1,380 \mu$ in length.

The ribs show a degree of anastamosis and branching similar to that for *S. nux-vomica*. Each fascicle is surrounded by a thin cellulose membrane which retains the ribs in close proximity, in many trichomes this membrane is ruptured and the ribs separate along their length (cf. Small, 1913), recombining only at the trichome tip (Fig. 5A, ep.). This results in the trichome limbs having a loosely interwoven appearance and exhibiting a variation in apparent diameter proportional to the degree of separation.



FIG. 5. Strychnos nux blanda, Hill, seed. A, Longitudinal radial section of testa. B, Transverse section of upper half of trichome base. C, Transverse section of lower half of trichome base. D, Transverse section of basal periclinal wall of trichome base. E, Transverse section of ground tissue of testa. F, Longitudinal radial section of epidermis in region of ridge. All \times 200. ep, epidermis; nl, ground tissue.



FIG. 6. Strychnos nux vomica, Linn. seed. A. Longitudinal radial section of endosperm. Strychnos nux blanda, Hill, seed. B, Longitudinal radial section of endosperm. Strychnos potatorum, Linn; C, Longitudinal radial section of endosperm. D, Surface view of endocarpic layer. E, Transverse sections of trichome limbs. F, Longitudinal radial section of seed coats. G, Transverse section of basal periclinal walls of trichome bases and adjacent ground tissue. H, Longitudinal radial section through seed coats in hilar region. All \times 200. nl., ground tissue; a, aleurone; en, endocarpic layer; ep, epidermis.

The angle which the trichome limb makes with the base is not constant, thus trichomes of the same length may have their tips at varying heights above the seed surface. This factor, in conjunction with the separation of the limbs, described above, imparts the somewhat rough texture to the epidermal surface noted in the macroscopical description.

The trichome limbs in the region of the keel-shaped ridge on the margin of the seed are not appressed but rise perpendicularly from the base and are directed away from the seed centre.

The narrow ridge which runs from the micropyle to the hilum (Fig. 1C, r) is composed of trichomes, with different characters to those previously The trichome bases in this region are T, 56–76–98 μ and described. R, 240-290-340 μ . The lower half of the base is typical of the epidermal cells on the rest of the seed, with the exception that the lumen is traversed by a greater number of anastamosing and branching rod-like elements (Fig. 5F). This lower part of the base is surmounted by a compact formation of curving, recurving, anastamosing and branching rods which are finally drawn into a fascicle to form the trichome limb. The trichome limb is not appressed but is gently curved with a recurved chordaceous apex. The entire trichome is 950–1,100–1,150 μ in length at the thickest point of the ridge. As the lateral boundaries of the ridge are approached the area of recurving and curving becomes smaller, the longitudinal dimension of the base is decreased, and the degree of recurving of the trichome tip increases until the trichome reverts to the normal epidermal cell type.

The ground tissue of the testa is composed of a brown band of flattened parenchyma. R, 8–14–23 μ (Fig. 5A, nl.). In transverse section the tissue appears as ill defined isodiametric polygonal cells, T, 25–53–63 μ (Fig. 5E). In the region of the keel-shaped ridge on the margin of the seed the ground tissue is less flattened and is composed of six to eight layers of irregularly shaped but distinct cells.

The *hilum* has a similar structure to that described for S. *nux-vomica* (Fig. 4E).

The *endosperm* of the seed is found adjacent to the ground tissue of the testa and is composed of a parenchymatous tissue possessing small intercellular spaces, and with a clearly discernible middle lamella (Fig. 6B).

The outermost layer consists of radially elongated polyhedral cells, R, 24–44–60 μ ; T, 12–14–16 μ ; L, 12–15–16 μ , the outer walls of which are thickened and the cell contents are as described for the remainder of the endosperm. Within this layer are three or four layers of cells, R, 20–38–45 μ ; T, 14–25–40 μ ; L, 14–25–40 μ . The remainder of the endosperm is composed of large polyhedral cells, R, 40–60–95 μ ; T, 38–50–60 μ ; L, 35–50–60 μ .

The cell walls exhibit fine perforations and show little variation in thickness being $3-5-8 \mu$.

The *cell content* is an oil plasma the aleurone grains of which are irregularly shaped, show one or more globoids, are $4-20-30 \mu$ in diameter.

The embryo is composed almost entirely of small thin walled parenchyma. In transverse section the lamina is up to 150 μ thick and shows partial cellular differentiation into upper and lower epidermis and vascular tissue. In surface view the cells of the upper epidermis have almost straight anticlinal walls and are 18-23-29 μ long and 8-15-20 μ wide. The cells of the lower epidermis are polygonal and isodiametric, T, 5-9-15 μ .

The cells of the embryo are filled with fixed oil and contain a few scattered amorphous aleurone grains.

Histology of the seed of Strychnos potatorum, Linn.

Strychnos potatorum has three cell layers surrounding the seed; a discontinuous trichomatous layer bounded on the outer surface by a thin endocarp and on the inner surface by a layer of ground tissue. The trichomatous epidermis and the ground tissue comprise the testa of the seed (Fig. 6F, ep., nl.).

The trichomatous epidermis is composed of strongly lignified cells having a short conical base extended to form an appressed hair which is directed at an angle to the base and radiates towards the margin of the seed (Fig. 6F, ep.).

The base has the dimensions, R, $10-17-20 \mu$; T, $20-45-70 \mu$. In longitudinal section the anticlinal walls are seen to consist of a series of rod-like elements which arise directly from the basal periclinal wall and extend for a short distance to form the base before being drawn into a fascicle to form the trichome limb. The anticlinal walls are pierced by large ovoid pits which are situated between the constituent rods. The basal periclinal wall is irregular and heavily pitted. In transverse section (Fig. 6G) the trichome bases cannot be differentiated as single cells but appear as isolated patches of lignified tissue irregularly distributed and pierced by slit-like pits.

The trichome limb is strongly appressed and consists of 3-4-6 lignified ribs, enclosed by a thin cellulosic membrane and surrounding a small lumen, the ribs sometimes exhibit a crenulate outer margin, but the majority are smooth (Fig. 6E). The individual ribs are $4-7-12 \mu$ in diameter, the entire trichome limb is $12-16-32 \mu$ in diameter and $160-320-480 \mu$ in length. The ribs anastamose and branch along the length of the trichome limb but not to the same degree as those of *S. nux vomica*. The trichome tip is blunt, rarely pointed and frequently penetrates a short distance into the endocarpic layer.

The endocarp is a layer of collapsed parenchyma, R, $10-20-30 \mu$, which lies outside the trichomatous layer and is entire over the whole seed surface (Fig. 6F, en.). In transverse section (Fig. 6D) the cells are parenchymatous with some intercellular pits. They can be divided into two size groups: those polygonal cells having the dimension T, $15-30-50 \mu$, and a smaller group circular in outline and T, $10-12-17 \mu$. The smaller cells are those which receive those trichome tips that penetrate into the endocarpic layer. This penetration results in the finely rugulose appearance of the seed surface referred to under macroscopical characters.

The ground tissue of the testa is composed of a brown band of flattened parenchyma, R, 8–12–16 μ (Fig. 6F, nl.) and contains a few unidentified prismatic crystals. In transverse section (Fig. 6G) the tissue appears as ill-defined polygonal cells, T, 20–35–50 μ . In the region of the fine ridge on the margin of the seed, the tissue is less flattened and consists of two to three layers of irregularly shaped but distinct cells.

As reported under macroscopical characters *the hilum* is found as a small circular depression in the centre of one curved seed surface. A longitudinal section through the hilar region shows that the depression is caused by a change in the trichome formation. As the trichomes approach the hilum they exhibit a lesser degree of appression and become slightly recurved (Fig. 6H, ep.). Nearer the hilum the trichomes lose their curvature, exhibit a greater number of ovoid pits and become progressively shorter, finally, those cells in close proximity to the raphe are short conical cells with thick pitted walls devoid of the typical rod-like elements described previously. The endocarp in the hilar region is less flattened and as the trichomatous layer grows smaller the endocarp shows gradual differentiation into simple parenchyma (Fig. 6H, en.), (Fig. 4F, en.).

The *raphe* is a thin vascular strand composed of small spirally thickened vessels and traverses the endocarp to extend a short distance on all sides into the ground tissue of the testa (Fig. 4F, r.).

The endosperm of the seed is found adjacent to the ground tissue of the testa and consists of parenchyma with a clearly discernible middle lamella (Fig. 6C). The outermost layers of cells are radially elongated and polyhedral, R, $21-34-45 \mu$; T, $13-20-30 \mu$; $13-20-30 \mu$, the outer walls are thickened, and the cell content as described below. Within this layer is a single layer having the dimensions R, $18-30-45 \mu$; T, $16-26-33 \mu$; L, $16-26-33 \mu$. The remainder of the endosperm is composed of large polyhedral cells, R, $43-58-89 \mu$; T, $32-46-85 \mu$; L, $32-43-83 \mu$. The walls are $10-26-33 \mu$ thick and show a few large funnel-shaped pits which connect with pits in neighbouring cells.

The *cell content* is an oil plasma the aleurone grains of which are irregularly shaped, show one or more globoids and are $3-12-20 \mu$ in diameter.

The embryo is composed of small thin walled parenchyma. The lamina is up to 130 μ thick in transverse section and shows partial differentiation into upper and lower epidermis and vascular tissue. In surface view the cells of the upper epidermis have straight anticlinal walls and are R, 14-20-24 μ ; L, 9-18-22 μ . The cells of the lower epidermis are polygonal and isodiametric, 4-8-12 μ . The cells of the embryo are filled with fixed oil and contain a few scattered aleurone grains.

DISCUSSION

Macroscopical Characters

The examination of the three seeds shows certain characters which will distinguish S. nux-vomica from S. nux-blanda and S. potatorum.

Seed shape. The discoid shape of S. nux-vomica contrasts with the ellipsoidal disc of S. nux-blanda and the subspherical seed of S. potatorum. Both the adulterants show little divergence in general shape or marginal form, but S. nux-vomica varies from flat to concavo-convex and exhibits a margin which may be acute or smoothly curved. The slightly keel shaped ridge on the margin of some of the seeds is only slightly elevated in S. nux-vomica and contrasts with the broad laterally extended keel-shaped ridge of S. nux-blanda and the fine ridge of S. potatorum.

Surface characters. The seed surface of S. nux-vomica exhibits a sericeous lustre and differs markedly from the scabrous surface of S. nux-blanda and the finely rugose appearance of S. potatorum.

Dimensions. Various authors report different dimensions for the seed of S. nux-vomica; these may be accounted for by the different habitats of the samples measured. Pereira (1850) stated the maximum diameter as 25.4 mm., Moll and Janssonius (1923) gave the maximum as 28.0 mm.The average diameter was given as 22.09 mm. by Bentley and Trimen (1880) and the size range has been given as 13.5-22.0 mm. by Dunstan and Short (1883), 20-25 mm. by Wallis (1951) and 10-30 mm. by Trease (1957). Whilst most of these dimensions fall within the limits given for the sample examined (17.0-18.5 to 23.1-29.5 mm.) they do not give a true indication of the diameter of the seeds in the current commercial samples. The dimensions recorded here show that two-thirds of a sample will have a diameter of between 18.5 and 23.1 mm.

Embryo. The embryos of *S. nux-vomica* and *S. nux-blanda* are similar in shape and size but the apex of the radical is clavate in *S. nux-vomica* and conical in *S. nux-blanda*. The embryo in *S. potatorum* is smaller and exhibits only 3-5 primary veins against the 5-7 found in the other cotyledons.

Histological Characters

The microscopical examination revealed certain characters which can be used to distinguish the three seeds.

The testa. In S. nux-vomica the trichomes were first described by Berg (1865). In his description of the basal-cell of the trichome he stated that it showed "spiral like pitting" in the anticlinal wall; later, Greenish (1920) modified this to "oblique pits", and Wallis (1951) described the pits as "sometimes more or less spirally twisted". Other authors have varied their description from "slit like" (Trease, 1957) to "branched slit like" (Moll and Janssonius, 1923) without any mention of obliqueness. In the material examined, the anticlinal walls of the trichome bases were pierced by tubular pits but the pits showed no spiral arrangement and were not oblique. The dimensions of the base have been stated to be R, 75 μ ; T, 75 μ (Wallis, 1951) and R, 35 μ ; T, 30–35 μ (Moll and Janssonius, 1923), whereas they were observed to be R, 65-80-100 μ ; T, 40-60-85 μ and L, 45-65-87 μ . A further anomaly was found in the descriptions of the trichome limb, all authors agreed on its general structure but many disagreed on its linear measurements. These varied from a maximum length of 1,500 μ (Greenish, 1923) to a range of size

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600–800–1,000 (Wallis, 1951) and 800–1,000 μ (Moll and Janssonius, 1923). In the samples examined, no trichome was observed to be greater than 1,290 μ in length, the mode was 950 μ and the minimum 600 μ .

That part of the epidermis in S. nux-vomica and S. nux-blanda shows the most divergent structure, is the region of the ridge joining the micropyle and the hilum.

The ridge of S. nux-vomica was at one time described as the raphe (Berg, 1865) and more recently as an area in which the trichome limbs are "irregularly directed" (Trease, 1957). The latter statement is correct in that the trichome limbs show a certain degree of sinuosity and are not closely appressed, but the height of the ridge is also increased by the subepidermal layer in S. nux-vomica and in S. nux-blanda by the extended trichome bases.

The epidermis of S. potatorum differs considerably from that of S. nux-vomica and S. nux-blanda. The trichomes form a discontinuous layer and have a base which bears little resemblance to the prismatic base of the other two seeds. The limb is about one-third as long (160–320–480 μ) and there is no variation in the epidermal cell structure excepting those truncated forms found in the hilar region. In addition, S. potatorum is the only seed in the group which shows an adherent endocarpic layer.

Endosperm tissue. In all three endosperms there are three cell types varying in size and radiating from the periphery towards the centre. In S. nux-vomica and S. nux-blanda the layer of cells at the periphery has a palisade-like structure which is radially directed. Small (1913) stated that in S. nux-vomica these cells had "more or less triangular ends" whilst those of S. nux-blanda were "more usually square ended". This was investigated as a possible means of differentiating between the two endosperms, but the variation within different seeds rendered this character of little value.

In S. potatorum the cells of the outer layer are smaller than those of S. nux-vomica and S. nux-blanda and exhibit a lesser degree of radial extension.

Within this outer layer there is a region where the cell dimension is less in the radial (R) direction than the outer layer but greater in transverse (T) and longitudinal (L) direction. In S. nux-vomica this region is two to three cells thick, in S. nux-blanda three to four cells thick and in S. potatorum consists of only one cell layer.

The remainder of the endosperm in each of the seeds is composed of large slightly elongated polyhedral cells.

The diagnostic feature of the endosperm which is of most value is the variation in the thickness of the cell walls. Those of S. nux-vomica are $20-25-30 \mu$ and are easily distinguished from those of S. nux-blanda which are only $3-5-8 \mu$ thick; in S. potatorum the walls have a thickness of $10-26-33 \mu$, which is similar to the dimension of S. nux-vomica but, in addition, S. potatorum exhibits large funnel-shaped pits and the cells appear almost cuboid in section.

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