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Age Stages in the Ontogeny of Cultivated Panax ginseng C.A. Mey

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Abstract—The ontogeny of perennial polycarpic herb *Panax ginseng* C.A. Mey. (Araliaceae) under plantation conditions was described. Three periods (latent, pregenerative, and generative) and eight age stages have been identified in the ontogeny of cultivated *P. ginseng*. The generative period of this species is the longest ontogenetic period, which determines the timing of its cultivation in plantations.

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Ginseng *Panax ginseng* C. A. Mey. (Araliaceae) is a valuable medicinal plant. *P. ginseng* is a relic of the Tertiary flora; currently, it is identified as an endangered species of the 1st rank (Kharkevich and Kachura, 1981) and is listed in the Red Book of the Russian Federation (*Krasnaya kniga...*, 1988). *P. ginseng* is unique among medicinal plants by the diversity and breadth of curative effects and it attracts considerable attention over the years.

The world requirements in this plant are almost completely satisfied by the roots cultivated in plantations in different countries. In the Maritime Territory, P. ginseng is cultivated in large plantations such as the Zhen'shen' Farm in the Anuchinskii Region established in 1950s (Shreter, 1975) and small plantations scattered all over the area. Cultivated P. ginseng develops much faster compared to natural plants. Clearly, researchers demonstrated great interest to the growth and development of this species. The stages of individual development of P. ginseng have been published (Imamuri, 1936; Grushvitskii, 1961; Slepyan, 1968; Zhuravlev and Kolyada, 1996); however, no diagnoses of the age stages have been identified. The goal of this work was to identify the age stages in cultivated P. ginseng population. This plant is very rare in nature, which makes such study on wild plants impracticable; accordingly, plantation was used as a model.

MATERIALS AND METHODS

The material for the age stage analysis was taken in a population of cultivated plants (Spasskii Region, Maritime Territory) grown from seeds of wild *P. ginseng*. This population was cultivated for 15 years. The growth conditions simulate natural ones. For instance, forest soil is used without agrochemical treatment. The plantation has awnings with herb mats at a height of 1.2–1.5 m. The bed width and height was 1 and 0.3 m, respectively.

The ontogenetic periods were described using the Rabotnov–Uranov scheme (*Tsenopopulyatsii raste-nii...*, 1976). The age stage of individuals was identified with an account of a set of structural characters of their overground and underground parts. Each age stage was described using a least 5–7 plants; in total, more than 45 plants were analyzed. The plant morphology was described in terms accepted in current ontogenetic studies (*Ontogeneticheskii atlas...*, 1997).

RESULTS

The following age periods and stages have been identified in *P. ginseng* ontogeny.

Latent period. P. ginseng seeds (drupe) fall in autumn and germinate after at least 22 months (Gutnikova and Vorob'eva, 1959). The seed is flat, discoid, and is loose in the drupe. The seed is 5-5.5 mm long and 3.8-4.2 mm wide. It contains an endosperm. This species features embryonic underdevelopment (Grushvitskii, 1961). Seeds of this type require a long-term stratification. The following conditions provided for a high germination rate: stratification at +18...+20°C for 3 months; at $+9...+10^{\circ}$ C, for 1 month; and at $0...+3^{\circ}$ C, for 5 months. In this case, 78% of seeds germinated (Bogdanova, 1971). The seed germination rate in plantation was 72%. The fruit is a fleshy inferior syncarpous drupe. The fruits are bright red, their mean size is $1.5 \times$ 0.9×0.7 cm, contain yellow pulp, have flattened top and sides, and contain two flat yellow nuclei. The nucleus has a ridge along the upper edge and clearly pronounced ribs in parallel grooves. The nucleus sur-



Fig. 1. *Panax ginseng* plants at different age stages in plantation; pl, seedling; j, juvenile; im, immature; v_1 , virgin young; v_2 , virgin adult; g_1 , generative young; g_2 , generative medium; pr, principal root; h, hypocotyl; s, seed; c, caudex; lr, lateral root; ar, adventitious root; wb, winter bud in the cataphyll base; rl, rosette leaf; sl, stem leaf; cp, cataphyll (for Figs. 1–3).

face is covered with small tubercles and small pits and feels rough. The nucleus is 4–6 mm long and 4.9–5.1 mm wide and has dense coat.

Pregenerative (virgin) period. Seedlings appear in mid-May. Germination occurs on the ground, i.e., the cotyledons remain in the underground nucleus (Fig. 1). The cotyledons are thin, have absorption function, and deliver nutrients to the growing plant from the endosperm. The seminal root emerges first and grows deep into soil. Next, the only ternate leaf sandwiched between the cotyledons is released; first, it appears on the soil surface as a typical loop and then the leaf blade is unfolded in the daylight. The leaf blade is 3.4–3.6 cm long and 3.4–4.1 cm wide. The length and width of the leaflets are (3.1) 3.3-3.5 and 1.5-1.7 cm, respectively. The leaf blade margin is bicrenate, and the lower leaf surface has a typical indumentum of soft simple hairs. The butt length and diameter are 6.7-7.2 cm and 0.7–0.9 mm, respectively. The formed seedling has a rosette shoot with a single ternate leaf, hypocotyl (2-4 mm), and unbranched root 4.5-5.2 cm in length and 2-3 mm in diameter. The cotyledons remaining in the seed die after 3-4 weeks. Seed dying is the marker of the next age stage—juvenile (Fig. 1). The juvenile stage lasts from late September to early October. The juvenile plants demonstrate quantitative changes in the root system: the primary root grows 7.8-8.2 cm long and 5-9 mm in diameter and second-order lateral roots appear.

Immature stage (Fig. 1). In the second year of life in plantation, a shoot is formed by monopodial growth from the apical winter bud (Fig. 2). The plant has a single assimilating palmately compound leaf (pentamerous) on an elongated butt (2.5–3 cm). The largest of five leaflets is terminal (4–6.5 cm in length), while the smallest and often asymmetrical leaflets are lateral (up to 3.2–3.4 cm in length). The leaflet margins are serrulate with very rare hairs on the underside. The stem base harbors two cataphylls and winter buds are initiated in their bases (Fig. 2). Bud scales (from three to

four) are fleshy and white. Later, the apical bud restarts growth first, while the lower bud in the base of yet persistent bud scale remains dormant. The tap root system includes 3–6 lateral roots of two orders. The primary root is pronounced and light colored, 19–22 cm in length and 9–17 mm in diameter. Contractile root activity forms ring wrinkles, which are particularly pronounced on the primary root. Annually dying basis of the overgrown shoots start to form single-headed caudex, which remains vertical in the first years.

Virgin young stage (Fig. 1). In the third year of life in plantation, a shoot is formed by sympodial growth from the apical winter bud formed in early autumn of the previous year. Later in ontogeny, only elongated monocyclic shoots are formed, i.e., monopodial growth is replaced with sympodial one (Fig. 2). Plants 9-12 cm in height have two compound leaves, which can be pentamerous, tetramerous, and trimerous and occur in different combinations. However, nearly all observed plants contain one leaf with five leaflets. The terminal leaflet is the largest in pentamerous and tetramerous leaves, while all leaflets are similar in size in trimerous leaves. The leaf size varies from 6.9 to 7.8 cm in length and from 7.1 to 9.5 cm in width. The base of the overground stem harbors two cataphylls with winter bugs. In the first half of July 35-40 days after spring shoot growth started, the primordial future winter bud in the base of the upper cataphyll starts to grow and differentiate. In autumn, the winter bud is formed with primordial shoot and two lateral buds. This structure is often referred to as "bud in bud." The bud in the base of the lower cataphyll remains dormant. Caudex formation continues in the underground part; it reaches (1.8) 2.1–2.3 cm in length and 0.8–0.9 cm in thickness. Thick first-order adventitious roots branch from the caudex (Figs. 1, 2). The primary root grows to 24-26 cm in length and up to 2-2.2 cm in diameter. The number of lateral roots can sometimes reach nine. These include large roots (commonly two or three) growing horizontally or obliquely downward



Fig. 2. Ontogeny of cultivated *Panax ginseng* (diagram); *1*, seed; *2*, assimilating leaf; *3*, cataphyll; *4*, vegetative shoot; *5*, generative shoot; *6*, bud; *7*, root; *8*, annual gain, caudex top.

to reach 19–26 cm in length and (0.5) 0.8–1.2 cm in diameter.

Virgin adult stage (Fig. 1). The transition to this stage is accompanied by the appearance of the third leaf, formation of the proper leaf whorl, and increase in the caudex thickness and height of the overground shoot. On average, plants are 16-22 cm high. The leaves have the same shape as adult leaves but smaller (9.2–10.5 cm long and 10.5–12.5 cm wide). The leaflets are bare, obovate or oblanceolate, and with serrulate margins. Despite the pool of winter bugs in the basis of cataphylls, four buds are initiated annually (two of which become dormant) and shoot is initiated only from the apical bud containing differentiated primordial shoot. In this context, new shoot formation after apical bud damage occurs not earlier than next year. Plants have a tap root system. The root gradually becomes horizontal or obliquely vertical and occupies the upper soil layer. In plantations with high beds, the root remains vertical (Zhuravlev and Kolyada, 1996). The primary root is light colored and grows lengthwise (39-44 cm) and widthwise (2-2.8 cm in diameter). The second-order lateral roots are thick, up to 26-34 for cm in length and 0.9–1 cm in diameter. The caudex length and diameter are 2.3-2.5 and (0.9) 1.1-1.2 cm, respectively. Fourth-order lateral roots appear. First-order adventitious roots (commonly one or two) grow to 17-23 cm in length and up to 0.3-0.7 cm in diameter. The virgin adult state lasts 1-2 years in plantation.

Generative period. Generative young stage (Fig. 1). The generative annual shoot can be 40–48 cm high and its upper part carries three palmately compound leaves in a whorl. The shoot terminates with the umbel inflorescence. Flowering takes place in the first half of June. The flowers are actinomorphic, bisexual, greenish white, and about 4 mm in diameter. The number of set fruits varies from 9 to 14. The leaf butts are up to 5.9-6.5 cm long. The leaves can reach 16-18 cm in length and 18-20 cm in width. When the overground shoot dies, the winter bud in the base of the lower cataphyll loses the bud scale. A trace or scar is formed at the place of the fallen shoot and a dormant bud appears at the end of this scar. The bases of died annual shoots form the caudex, whose length and diameter can reach at the generative young stage 4.5-5 and 1-1.3 cm, respectively. The number of scars on the caudex allows the absolute plant age to be determined. The primary root is 35–42 cm in length and 2.7–3.3 cm in diameter. Third- and forth-order lateral roots are abundant. Second-order lateral roots grew to 23-24 cm in length and up to 2-2.3 cm in diameter. The cultivated plants are 5–6 years old.

Generative medium stage (Fig. 1). The transition to this stage is accompanied by the increase in the num-

BIOLOGY BULLETIN Vol. 34 No. 2 2007

ber of leaves in a whorl to four. The leaves are penta-, tetra-, and trimerous palmately compound. Plants reach the height of 50-53 cm. Leaf size varies from 17 to 20 cm in length and from 18 to 25 cm in width. The number of fruits increases to 17–31. Up to four winter buds are formed in August in the base of the cataphylls. Plants have a tap root system; the third- and forth-order lateral roots considerably elongate; and several second-order lateral roots clearly stand out by their large size. A considerable fraction of photoassimilates move to the first-order adventitious roots, which provides for their growth to reach 1.9–2.1 cm in diameter, which is similar to thick second-order lateral roots. The primary root is 55–58 cm in length and 3.3-3.6 cm in diameter. The caudex is 5.3-5.5 cm in length and 1.5-1.7 cm in diameter. The cultivated plants are 6-7 years old.

Generative mature stage (Fig. 3). Plants at the generative mature stage reach the maximum size; their height is 66–77 cm. The stem is cylindrical, bare, and commonly longitudinally grooved. The whorl includes 5-6 palmately compound leaves, most of which are pentamerous and less commonly tetramerous. The leaf butts are long, 9.5-10 cm; while the butts of the terminal and lateral leaflets reach 2.5-2.8 and 0.5-1.5 cm, respectively. The proper leaflets are obovate or oblanceolate and have serrulate margins. The leaf size varies from 19 to 22 cm in length and from 21 to 28 cm in width. The length of the terminal leaflets is 18.5-19.7 cm. The winter buds are initiated in *P. ginseng* two years before they form a shoot. The apical winter bud (closed) gives rise to a shoot next year, while the lower bud remains dormant. Dormant buds are open in P. ginseng; although initially they had a coat and then lost the bud scales. The underground part features a sharp increase in the thickness of the third-order lateral roots and second-order adventitious roots to 1.5-2.8 cm. As a result, the lateral and adventitious roots further separate sideways in many plants. The primary root continues to grow in length and diameter (57-63 and 4.7-5.1 cm, respectively); although its elongation is realized through the extension and branching of the lower part, while the thick upper part does not elongate. The caudex reaches 7.8–8.2 cm in length and 2.8–3.1 cm in thickness. The duration of this stage in plantation is 5-7 years.

Postgenerative (senile) period. Senile stage. We observed no plants at this stage. In conditions of commercial plantation, plants with roots of tradable maturity are dug out. Accordingly, plants had no opportunity to reach the senile stage. However, some authors and ginseng collectors reported that *P. ginseng* can live 400 years in nature. Individual plants were described with the caudex 16–17 cm in length and up to 140 scars from the basis of died shoots, which suggests that these plants are at least 140–150 years old (Grushvitskii,



Fig. 3. Generative mature *Panax ginseng* and (g_3) ; bs, base of elongated shoots of previous years; ar, assimilating root; rw, ring winkles resulting from root contraction.

1961). According to singular reports of root collectors, old *P. ginseng* plants have short and thick dark green or brown stem (bluish black near the surface), stiff wrinkled leaves, and short flower stalk with not numerous flowers.

DISCUSSION

Differentiation between age stages of *P. ginseng* is facilitated by its specific developmental rhythm. In particular, specific structure of the overground shoot is determined at the bud stage and does not change until autumn dying. This applies to both the vegetative and generative periods of *P. ginseng*. This is confirmed by the studies on *P. ginseng* tissue culture: an isolated dormant bud could give rise to a small shoot composed of a leaf, stem, and primordial fluorescence in vitro (Zhuravlev et al., 1993). This explains rapid shoot growth in spring: the stem and leaf growth is over within the first 20-25 days of vegetation and the assimilation system is also formed within this period. The rapid shoot growth and ontogenetic variation of leaves provides for a highly reliable identification of age groups under plantation conditions. Thus, three age periods and eight age stages have been identified in cultivated P. ginseng. The age stages during this species ontogeny are largely similar to the age stages identified in some perennial herbs. However, the generative period is the longest of all ontogenetic periods. New organs are formed throughout this period, while dying processes are not pronounced except for the seasonal replacement of monocyclic shoots. This feature was noted by Voroshilov (1960) in his description of the rhythmologic group of species (ginseng type). According to this concept, some plant species of the ginseng type first flower being very small relative to the maximum size reached many years later. Likewise, cultivation of this species in plantation requires long periods-the roots reach tradable maturity only after 6 years. The expediency of digging out the roots of P. ginseng not younger than 6 years has been confirmed by the analysis of root chemical composition. The biological activity of P. ginseng roots is primarily attributed to the low molecular weight bioregulators, mainly, dammarane- and oleanol-type triterpene glycosides (Shibata, 1986). The qualitative and quantitative composition of dammarane-type glycosides in P. ginseng roots depends on their age. Their concentration in the root is relatively low within the first three years and starts to increase only in the fourth year. Later, the root biomass increases almost directly with the concentration of ginsenosides (Soldati and Tanaka, 1984; Kim et al. 1987).

The cultivation in plantations is not only a long-term but also a risky process. The *P. ginseng* roots entirely lack mechanical tissues; hence, they have no efficient barriers protecting them from infections and are susceptible to fungal and bacterial diseases (Zhuravlev and Kolyada, 1996). Moreover, phenological growth stages were identified in a closely related species *P. quinquefolius* when agrochemical prevention of diseases is recommended (Proctor et al., 2003). Many ginseng growers noted that mass diseases coincides with the arrest of differentiation of the overground shoot, i.e., when the plant whorl contains 5–6 leaves. This factor is also important for the optimum time of root digging out, which is sometimes as early as 5 years in commercial plantations.

The problem of *P. ginseng* life span in plantation can be solved a priori only and without considering its high susceptibility to diseases after the generative mature stage starts. Considering that cultured plants grew faster, the senile stage should also start earlier than under natural conditions. The descending period of *P. ginseng* ontogeny should be as long as the ascending one (about 15–20 years) considering that the vegetative and generative vigor decreases gradually similar to its increase in the virgin period. Given this assumption, the life span of *P. ginseng* in plantation should be at least 30–45 years.

CONCLUSIONS

P. ginseng is a perennial polycarpic herb with a tap root system and whorl shoots during early ontogeny

and elongated monocyclic monocarpic sympodially replaced shoots in the adult period. Specific structure of the overground shoot and its structural pattern are determined at the bud stage and realized during vegetation. The degree of the overground shoot differentiation is the major marker of the age stage. Under plantation conditions, P. ginseng ontogeny can be described as incomplete with a long generative period. P. ginseng development peaks at the generative mature stage, which governs the timing of its cultivation in plantation. Vegetative are production is not typical for the species. The main reproduction type involves seeds. By the time of falling, the seeds have underdeveloped embryo and require further development. P. ginseng features an almost two-year-old period of embryo development in the seed, which is the first of two-year-old cycles of primordial shoot formation in winter buds.

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