Morphogenesis and vitality of seedlings of *Ginkgo biloba* in outdoor conditions

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Introduction

The negative effect of abiotic and biotic environmental factors, against the background of climate changes, in the conditions of Ukraine is inevitably reflected in the condition of the soil (Savosko et al., 2018), vegetation (Lykholat et al., 2018a, 2018b; Nazarenko et al., 2018), animals (Boyko & Brygadyrenko, 2016; Faly et al., 2017; Didur et al., 2019; Polkhynenko et al., 2019) and significantly affects the health of the population (Kotsarev et al., 2001; Pertseva et al., 2008, 2012; Lykholat et al., 2016). Preservation of the existing biological diversity and enlargement of its range in Ukraine is possible using relic species, including *Ginkgo biloba* L. This plant is a relic tree which has been preserved in natural conditions only in China. From China ginkgo was brought to Holland in 1730. Currently, due to the value of ginkgo as a source of medical substance it is being grown in many countries of the world. Further contributory factors to this are the plant’s decorative value and its undermanding requirements regarding growth conditions.

Due to the medical and decorative values of ginkgo, its biology, ecology, biochemistry and technologies of growth are described in several monographs and numerous articles (Tereshchuk, 2009; Crane, 2013; Fisher, 2016; Tereshchuk, 2009). Ginkgo plants are trees of 30–40 m height with trunk diameter measuring up to 3 m and crown diameter of 25 m. The lifespan of these trees can reach up to 2,000 years.

Ginkgo is a dioecious gymnosperm plant. Generative reproduction of ginkgo begins only at 20–30 years. On male trees, microstrobili – small cones with microspores – are formed in spring, and on female trees, seeds with three-layer seed cover form. The development of embryo occurs in ginkgo in non-ripened seeds which have already fallen from the tree, at low positive temperatures. The process of embryo development in ginkgo seeds lasts approximately 3–4 months after they have fallen. Leaves of ginkgo have a specific structure. They are two-lobed, 5–8 cm in width. The leading bundles in the leaf blades ramify dichotomously (Shchyrova et al., 2002). Ginkgo is characterized by heterophyllly. In spring, on the plants’ shortened stems, the first “early leaves”
emerge from wintering buds. In early summer and throughout the season, the “late leaves” form on the ginkgo stems. Particularly those two-lobed leaves are more characteristic for this plant (Crichfield, 1970). Leaves on shortened shoots are thinner, the network of leading bundles in them is more dense and has fewer stomata (Leigh et al., 2011).

Ginkgo is a deciduous plant and therefore in autumn thus leaves fall off. In China, the leaves of ginkgo have been used for medical purposes since ancient times. In European countries and the USA active research and medical use of the plant began in the middle of the XX century. Up to the present, research has revealed that the extracts of ginkgo leaves are effective against a broad group of diseases – memory dysfunctions, chronic diseases of vessels of the brain, Alzheimer’s disease, decline in hearing ability, impaired blood circulation, including capillary, atherosclerosis, hemorrhoid and a number of other conditions (Yoshikawa et al., 1999; Diamond et al., 2000; Mahadevan & Park, 2008; Kuznetsova & Shulzenko, 2015).

Moreover, overview research by Po-Chuen Chan with the co-authors (Po-Chuen et al., 2007) referring to 162 literature sources states that extracts from leaves of ginkgo have a general health-improving effect on the human organism, increase stability of the nervous system against stress impacts, and slow the ageing process.

Extract from ginkgo leaves has a complex chemical composition. It includes over 40 biologically active ingredients. Standardized extract from leaves of Ginkgo biloba L. used in modern medicine contains three main groups of substances that determine specific pharmacological activity. They are as follows:

a) terpene trilactones (bilobalide and ginkgolides), their content equals 5.4–12.0%.

b) various bioflavonoids, which account for 24–27%.

c) proanthocyanidins, organic acids, ginkgolic acids, nitrogen bases (thymine), microelements.

Most valuable pharmacologically are terpene trilactones and bioflavonoids (Bikram et al., 2008; Singh et al., 2008).

Technologies of obtaining medicinal forms (tablets, capsules, etc.) from ginkgo leaves vary in different pharmaceutical companies. The quality of the output material also varies. In Germany a standard has been developed, according to which the extract of ginkgo leaves should contain 22–27% of flavonoid glycosides, 5–7% of terpene lactones and no less than 5 parts per million of ginkgolic acids (Jacobs & Browner, 2000).

During preparation of leaves, one must take into account that the highest amount of such substances as rutin, kaempferol and bilobetin is present in young leaves until the middle of summer, whereas the amount of bioflavonoids is the highest in the autumn leaves which begin to turn yellow and fall off (Lobstein et al., 1991). Maximum amount of polyphenolic compounds in the leaves was recorded during the ripening and falling off of the fruits (Yudina et al., 2010). Therefore, two-time preparation of ginkgo leaves as medical raw material can be recommended: the first – in the middle of the summer with collection of up to 30% of leaves from the trees, the second – at the very beginning of November. Traditional periods of collecting leaves for obtaining medical substances have established: July in the USA, August in China, September–October in France (Schmid & Bulz, 2003), in Lithuania the best period for preparation of leaves is believed to be July–August (Rimkeni et al., 2017). Leaves of ginkgo are collected manually or using special devices. The output of useful production (leaves) depends on the age of the tree and the region. In India, in pre-mountain areas of the Himalayas in some years 30 t/ha of leaves are collected (Gopichand & Meena, 2015).

In the conditions of the steppe Ukraine ginkgo grows well and survives winters under the open sky (Burdak & Koniaik, 2019). The plant is low-demanding for soil fertility. The plant is resistant to pests and diseases. However, in the conditions of Poltava oblast cases have been recorded of damage to leaves by diseases caused by Alternaria, and damage to seeds by Monilinia (Samorodov & Pospelova, 2016).

In North-East Ukraine the growing of ginkgo is only in the process of development, but in some places ginkgo trees grown for decorative purposes show high vitality. Therefore, a relevant and promising task is the creation of industrial plantations of ginkgo, necessary for preparation of leaves. Peculiarities of growing ginkgo in the conditions of North-East Ukraine have not been determined. The study was aimed at survey of morphological peculiarities of ginkgo plants and their vital condition depending on the region of seed origin, calendar age of the seedlings at the time they were transplanted to the open ground.

**Materials and methods**

For growing ginkgo plants in Sumy, a special comparative analysis was performed for seeds obtained in different climatic conditions, particularly the forest-steppe region (Kamianets-Podilskyi) and steppe region (Odessa): morphometric characteristics of ginkgo seeds were studied: length of seed, rim width, cross-section width of seed, mass (expressed as air dry weight). The amount of the selection was 100 for seeds obtained in Kamianets-Podilskyi, and 98 for seeds from Odessa.

In our studies, for planting ginkgo a combined two-stage scheme was approbated, according to which the seedlings were grown in greenhouses for 2–3 years, and then continued growing in open soil.

Earlier, similar technology of growing ginkgo was developed by A. O. Ostudimov for the conditions of Liviv Oblast (Ostudimov & Guz, 2010; Ostudimov, 2011). It includes growing 1–3 summer seedlings of ginkgo from seeds in greenhouses, followed by their growing up to the required sizes outdoors.

In our study, as planting material we used ginkgo seedlings from the plant nursery (greenhouse) of Sumy National Agrarian University planted in 2014–2015. Their characteristic is provided in the studies by Yaroshchuk (2016) and Kovalenko et al. (2018).

Transplantation of seedlings into the soil took place in early spring. The seedlings for growth completion were planted in rows with 0.8 m distance between the rows and 2.5–3.0 m between the rows.

We studied the condition of seedlings in two variants: variant 1 – three-year seedlings grown in the greenhouse were transplanted in spring of 2018; variant 2 – two-year seedlings started in the greenhouse were transplanted in spring of 2017.

The comparative morphometric analysis and evaluation of vitality of these plants were performed in August of 2019. Both groups of seedlings were five years old at the period of examining their condition.

The typical seedlings at the end of vegetative period of 2019 looked as shown in Figure 1. For evaluation of the morphological structure of ginkgo seedlings and their vital conditions, randomized selections of up to 30 specimens from variants 1 and 2 were used. Analysis of plants was undertaken in the end of the vegetative period of 2019, when active growth of plants had stopped. The plants were analyzed for four main parameters: height, size of the annual increment, diameter of stem near the root neck and amount of leaves.

The vital condition of the seedlings was evaluated on the basis of algorithm of the vitality analysis (Zlobin, 2018). The method has been successfully used both for determining the vitality of populations of herbaceous plants (Zlobin & Klimenko, 2010; Tikhonova, 2011; Kyrchuk, 2014) and tree plantations (Skliar, 2013; Skliar & Zlobin, 2013). The vitality of each seedling was evaluated by vitality coefficient Q, the value of which ranged 0.0–1.0. Depending on the Q value, the seedlings were divided into five vitality classes: 0.0–0.2 – cc (specimens of the lowest vitality), 0.2–0.4 – c (low vitality), 0.4–0.6 – b (average vitality), 0.6–0.8 – a (high vitality), 0.8–1.0 – aa (specimens with highest vitality).

Vitality of population as group of specimens was determined according to the formula:

\[ Q = (a/a + n + a + b)/n, \]

where aa, a, and b – quantity of the specimens of the respective class of vitality selection, n – amount of the selection. This indicator allows evaluation of the entire group of seedlings.

Kamianets-Podilskyi (48°40′50″ N, 26°34′50″ E) is located in the West Ukrainian province of forest-steppe physical-geographical zone. The climate of Khmelnytsky region is moderately-continental with a short, mild winter, relatively dry spring (low amount of precipitation in April), rainy summer and relatively dry autumn. According to the distribution of thermal resources of climate, the territory of Khmelnytsky Transnistria, to which Kamianets-Podilsky belongs, is characterized by an average annual temperature of +7.5–7.9 °C, the norm of annual precipitations of 600 mm, the sum of temperatures over 10 °C in the

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interval of 2,650–2,750 °C. By the pattern of geographical zoning, the city of Odessa (47° N, 30° E) is located in the Prymorie plain area of the Prychornomorska-Pryazovska southern steppe (dry steppe) province of the steppe zone.

The climate in Odessa oblast is moderately continental with insufficient moisture, a short, mild winter and long hot summer. The distribution of thermal resources in Odessa is as follows: average annual temperature is +10.7 °C, the norm of annual precipitations – 451 mm, total of the temperatures over 10 °C in the interval of 3,200–3,400 °C. Sumy (50°54′24″ N, 34°47′57″ E) is located in South-East part of Ukraine in the forest-steppe physical-geographical zone. The climate of this region is continental: with a mild winter and warm summer. Annual relative air moisture on average equals 78%, is the lowest in May (64%) and highest in December (89%) (Gorun, 2013; Trygub, 2016).

The survey results were statistically analyzed using the generally accepted modern methods of mathematical statistics with use of dispersion and vitality analysis. We used the computer software pack Statistica 8.0. Using methods of multiple regression, mathematical models were developed, connecting mass of <i>Ginkgo biloba</i> L. seed with its length, rim width of seed and cross-section width of seed.

**Results**

Comparative analysis of the main morphometric characteristics of seeds of <i>Ginkgo</i> in different climatic conditions of growing revealed the following: among such morphometric characteristics of ginkgo seeds as length of seed, rim width of seed, cross-section width of seed, mass of seed depending on the climatic conditions of growing, a reliable difference between the average values was found for rim width of seed and mass of seed (Table 1). The obtained values were lower for seeds obtained in the conditions of Odessa oblast with insufficient level of moisture and long hot summer.

On the basis of obtained morphometric data and their further statistical processing, by methods of multiple regression, the mathematical models were obtained, connecting the mass of seed with its length, rim width of seed and cross-section width of seed.

**Table 1**
The main characteristics of ginkgo seeds in different climatic conditions of growing

| Parameter | Selection | Mean arithmetic error | Median | Standard deviation | Variation coefficient, % | Asymmetry Excess |
|-----------|-----------|-----------------------|--------|--------------------|--------------------------|-----------------|
| Length of seed, mm (conditions of Kamianets-Podilsky) | 100 | 20.62 ± 0.28 | 21.71 | 2.80 | 0.14 | –1.61 | 3.11 |
| Length of seed, mm (conditions of Odessa) | 98 | 20.67 ± 0.07 | 20.73 | 0.74 | 3.60 | –0.47 | 0.12 |
| Rim width of seed, mm (conditions of Kamianets-Podilsky) | 100 | 15.61 ± 0.26 | 16.57 | 2.60 | 0.17 | –0.42 | –0.39 |
| Rim width of seed, mm (conditions of Odessa) | 98 | 14.83 ± 0.09 | 14.90 | 0.92 | 0.06 | –0.80 | 1.54 |
| cross-section width of seed, mm (conditions of Kamianets-Podilsky) | 100 | 12.37 ± 0.24 | 13.48 | 2.36 | 0.19 | –0.92 | –0.63 |
| cross-section width of seed, mm (conditions of Odessa) | 98 | 12.10 ± 0.05 | 12.16 | 0.54 | 0.04 | –0.58 | 0.31 |
| Mass of seed, g (conditions of Kamianets-Podilsky) | 100 | 2.40 ± 0.04 | 2.46 | 0.36 | 0.15 | 0.12 | 2.91 |
| Mass of seed, g (conditions of Odessa) | 98 | 1.68 ± 0.02 | 1.70 | 0.22 | 0.13 | 0.61 | 4.95 |

Note: statistically reliable difference with significance level (according to Student’s criterion): ** – 0.01–0.001; *** – <0.001.

For the conditions of forest-steppe zone the equation of regression looked as follows:

\[ M_{seed} = 0.1035 \cdot L_{seed} + 0.0972 \cdot W_{seed \ rim \ Kam} – 0.1031 \cdot W_{seed \ cross-section} (R^2 = 97.5\% ; corrected R^2 = 97.4\%), \]

where \( M_{seed} \) – mass of seed (g), \( L_{seed} \) – length of seed (mm), \( W_{seed \ rim} \) – rim width of seed, \( W_{seed \ cross-section} \) – cross-section width of seed (mm).

A peculiarity of the abovementioned equation of regression is that the free member of the equation is excluded from the model. Such an approach allowed us to obtain an equation with the best statistical characteristics, and also to quantitatively evaluate the effect of each factor (independent variable) on the extent of the manifestation of the parameter of optimization (dependent variable, Table 2).

**Table 2**
Statistical evaluation of independent variables in the mathematical model for seeds of ginkgo grown in the territory of forest-steppe zone (Kamianets-Podilsky)

| Parameter | Estimate | Standard Error | T Statistics | P-Value |
|-----------|----------|----------------|--------------|---------|
| L seed + | 0.1035 | 0.0214 | 4.84 | <0.0001 |
| W seed rim | 0.0972 | 0.0311 | 3.12 | 0.0024 |
| W seed cross-section | –0.1031 | 0.0393 | –2.62 | 0.0102 |

For the conditions of the steppe zone, the regression equation is as follows:

\[ M_{seed} = –0.0470 \cdot L_{seed} + 0.0668 \cdot W_{seed \ rim} + 0.1372 \cdot W_{seed \ cross-section} (R^2 = 99.0\% ; corrected R^2 = 98.9\%). \]

During the following statistical evaluation of the proposed mathematical model, we found that it has a high level of applicability (indicated by high coefficient of determination in the regression equation, equaling 97.5%) and reflects the orientation of changes of mass of the formed seed depending on the contribution of length and rim width of seed and cross-section width of seed. Furthermore, the developed mathematical model of predicting the mass of seed of <i>G. biloba</i> is adequate due to high level of significance of P-value (Table 3).

**Table 3**
Dispersion analysis of mathematical model for seeds of <i>G. biloba</i> which grow in the conditions of the forest-steppe (Kamianets-Podilsky)

| Source | Sum of Squares | Df | Mean Square | F-ratio | P-value |
|--------|---------------|----|-------------|---------|---------|
| Model  | 573.99        | 3  | 191.33      | 1282.99 | <0.0001 |
| Residual | 14.46        | 97 | 0.12        |         |         |
| Total  | 588.46        | 100|             |         |         |

For the conditions of forest-steppe zone, the equation of regression is as follows:

\[ M_{seed} = 0.1035 \cdot L_{seed} + 0.0972 \cdot W_{seed \ rim} + 0.1372 \cdot W_{seed \ cross-section} (R^2 = 98.9\%). \]
where \( M_{seed} \) – mass of seed (g), \( L_{seed} \) – length of seed (mm), \( W_{seed} \) – width of seed along the margin, \( W_{cross-section} \) – cross-section width of seed (mm).

In the abovementioned regression equation, similarly to the previous case, the free member is excluded from the model. This allowed us to obtain the equation with the best statistical characteristics and also quantitatively evaluate the effect of each factor (independent variable) on the extent of manifestation of the dependent variable (Table 4).

### Table 4

| Parameter       | Estimate | Standard Error | T statistic | P-value |
|-----------------|----------|----------------|-------------|---------|
| L seed          | -0.0470  | 0.0222         | -2.12       | 0.0370  |
| W see rim       | 0.0668   | 0.0251         | 2.66        | 0.0091  |
| W seed cross-section | 0.1372 | 0.0340         | 4.03        | 0.0001  |

Conducting the statistical evaluation of the proposed mathematical model we found that it has a high level of applicability (indicated by the high coefficient of determination of regression in the equation, equaling 99.0%) and reflects changes in the mass of the formed seeds depending on the contribution of length and rim width and cross-section width of seed. Furthermore, the developed mathematical model can be used for predicting mass of \( G. \) biloba seed and is adequate due to the high level of significance of \( P \)-value (Table 5).

### Table 5

| Source         | Sum of Squares | Df | Mean Square | F-Ratio | P-Value |
|----------------|----------------|----|-------------|---------|---------|
| Model          | 278.01         | 3  | 92.669      | 3147.48 | <0.0001 |
| Residual       | 2.79           | 95 | 0.029       |         |         |
| Total          | 280.80         | 98 |             |         |         |

The obtained mathematical models of evaluation of predicting mass of seeds from \( G. \) biloba which grew in different climatic zones conditioned the expedience of using seeds from forest-steppe conditions (Kamianets-Podilsky) in further studies.

Comparative morphometric analysis of two variants of planting ginkgo plants showed (Table 6) that planting two-year old seedlings (variant 2) led to better growth and development of the plants. They exceeded the plants which were transplanted at the age of three years (variant 1) in the annual increment of the main stem, diameter of the stem and number of leaves, being inferior to them only in height (Fig. 2). Plants that developed from the two-year seedlings exceeded the three-year old plants by annual increment only by 0.4 cm, by 0.15 cm by the stem diameter, and in number of leaves by 8. These differences were statistically reliable only at the level of \( P = 0.00–0.03 \).

### Table 6

| Parameters                      | Mean arithmetic value and its error | Minimum value | Maximum value | Variation coefficient |
|---------------------------------|-------------------------------------|---------------|---------------|-----------------------|
| Height of plants                | 44.7 ± 2.32                         | 20.5          | 68.0          | 28.5                  |
| Annual increment                | 13.7 ± 1.46                         | 1.0           | 28.4          | 58.2                  |
| Number of leaves                | 21.0 ± 2.13                         | 8.0           | 58.0          | 55.4                  |
| Stem diameter                   | 1.43 ± 0.01                         | 1.25          | 1.52          | 5.1                   |
| Diameter of stem                | 1.58 ± 0.004                        | 1.60          | 1.64          | 1.63                  |

Difference by the size of annual increment of the main stem between the plants of the first and the second variants was statistically unreliable (Table 7). Obviously this is related to the fact that annual increments reflect the conditions of the current vegetative period the best. Earlier the specialists determined that over the first three years after being planted the seedlings develop a root system, having minimum increments of the stems during that period (Crane, 2013). Morphostructural uniformity of the total of seedlings of the variants 1 and 2 was approximately similarly low. Variation coefficient (Table 6) for the size of annual increment of shoots and the number of leaves was within the range of 44–58%, height of plants – 28–35%. The lowest variation parameter was the stem diameter ranging 1.6–5.0%.

### Table 7

| Parameters                      | Mean square | Fisher criterion | Reliability level, \( P \) |
|---------------------------------|-------------|------------------|----------------------------|
| Height of plants                | 1272        | 7.99             | 0.006*                     |
| Annual increment                | 1.7         | 0.03             | 0.856                      |
| Number of leaves                | 960         | 4.92             | 0.030*                     |
| Diameter of stem                | 0.301       | 99.06            | 0.000*                     |

Note: Sign * marks parameters by which the seedlings of variants 1 and 2 statistically reliably differed from one another.

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Fig. 2. Main morphometric features of \( G. \) biloba seedlings according to variants of planting: numbers of the variants are indicated by figures.
For the integral evaluation of the vital condition of the seedlings we used vitality analysis. The evaluation of vitality of some seedlings by a combination of four recorded parameters revealed that in both variants 1 and 2 the plants when arranged according to vitality level from lowest to highest develop an unbroken sequence. On the basis of division of the seedlings by level of their vitality into five groups we determined that in this variant, in the selection, out of 30 plants 2 seedlings had c vitality (6.7%), 13 b (43.3%), 12 a (40.0%) and 3 had the highest vitality class aa (10.0%).

In the variant 2 selection the vitality of some plants was in the amplitude from 0.41 to 0.89 (Fig. 4). In this variant the seedlings of the lower classes of vitality cc and c were absent, 16 (53.3%) specimens had class b, and 12 (40.0%) class a and 2 (6.7%) aa class. Thus, by the scale of vital condition of the plants, the seedlings of variant 2 on the graph are “shifted to the right”, that is more of them had high vitality. The basis of this group of seedlings is composed of specimens of average and high vitality (Fig. 5). In general, in this variant, as the representative selection showed, specimens of low vitality were absent.

Integral evaluation of the vitality of seedlings using formula \( Q = \frac{(aa/n + a/n + b/n)}{n} \) in two variants of their planting into the open soil revealed that plants of variant 1 had \( Q \) coefficient equaling \( Q = 0.93 \). For variant № 2 \( Q = 1.00 \). Thus, in the outdoor conditions, two-year old seedlings had better vital condition and higher morphostructural characteristics compared with the ginkgo plants of three-year seedlings. This can be explained by two circumstances. First of all, younger two-year seedlings adapted better to the environmental changes after they had been moved from greenhouse to soil compared with the seedlings transplanted at the age of three years. Secondly, in variant 2 the seedlings were in the field for three vegetative periods, whereas in variant 1 – only two. Seedlings of variant 2 had more time for adaptation to the outdoor conditions.

**Discussion**

Trees of *G. biloba* are cultivated artificially in a number of countries due to the fact that their leaves have a complex of unique medical properties (Mohanta et al., 2014; Torchik et al., 2018). Extract from the leaves is used for different diseases related to the problems of the blood vessels, hypertension (Mohanta et al., 2014; Heinonen & Gaus, 2015; Badore et al., 2017). Moreover, the extract from ginkgo leaves contributes to improvement of memory, boosts mental and physical abilities, has anti-stress properties and strengthens immunity, etc. (Qiu et al., 2017; Zhang & Cai, 2018). Another advantage of the medical preparations from ginkgo leaves is their complex effect on the human organism (Krauss et al., 2016; Fan et al., 2018).

For creation of industrial plantations of ginkgo, more accessible and economically effective technology is needed. One of the indicators of predicting the high vitality of plants is quality of seeds, which to a high degree is determined by their morphometric parameters. Through comparison of the obtained regression equations which allow prediction of the mass of seeds of *G. biloba*, one can note that all independent variables (length of seed, rim width of seed, cross-section width of seed) have statistically reliable levels of significance and cannot be excluded from the models. However, differences can be seen in the extent of their contribution. Thus, the extent of contributions of length of seed and rim width of seed to the mass of seeds collected from ginkgo plants in the conditions of forest-steppe was practically the same, and the negative value of the coefficient indicating cross-section width of seed shows that with increase in its size the seed mass decreases. And vice-versa, the amount of contributions of length and rim width of seed and cross-section width of seed to the mass of seeds collected from ginkgo plants in the conditions of steppe were not the same either by extent or by pattern (Fig. 6). At the same time, the coefficient indicating cross-section width of seed had the value which exceeded the contribution of seed length almost by three times and the value was negative. That is with increase in the length of seeds of *G. biloba* growing in steppe conditions (Odessa) decrease in the seed mass of this tree plant will be observed.

Therefore, the studies revealed that climatic resources of the studied region condition the statistically reliable difference in the average values of *G. biloba* plants' seed mass and rim width of seed. In the drier and
hotter conditions of the South Ukraine (Odessa oblast) longer seeds will have lower mass, whereas in the more moderate and humid conditions of the forest-steppe zone such values will be in the seeds wider in cross-section.

![Diagram](image)

**Fig. 6.** Prediction of the mass of seed of *G. biloba* growing in different climatic areas

Methods of planting ginkgo plants vary (Ivaniuk & Zavadska, 2015). Sometimes the seedlings are grown in special containers and then transplanted into a permanent place of growth after opening the side walls of the container, ensuring the integrity of the root system. However, this is an expensive method and is used only for growing singular trees. Vegetative reproduction of ginkgo by stem cutting can also be used. The propagules produce new roots in water or in a nutrient substrate, and then are planted into soil (Glukhov et al., 2008).

The optimum age of planting seedlings of ginkgo to a permanent site is within the amplitude 4–7 years with height of 60–90 cm. However, for better adaptation, many authors recommend planting seedlings to a permanent site at the age of no less than 10 years, because at a younger age ginkgo plants are more susceptible to winter frosts and often require additional shelter.

Our studies of plantations of *G. biloba* revealed that in both variants 1 and 2 the seedlings had high vital condition in the amplitude of 0.97–1.0 according to the scale 0.0–1.0. The difference is that transplanting seedlings outdoors at the age of three years led to the result that 6.7% of the total number had low vital condition (vitality class c), whereas after transplanting seedlings at the age of two years plants with this level of vitality were absent.

In our study, during transplantation of seedlings, maximum preservation of the root system was provided. For seedlings a pit of required size was prepared (on average equaling 0.8 x 0.5 x 0.5 m). A drainage layer was put at the bottom. After planting, the plants were intensely watered, and then soil was covered with straw mulch. Some authors recommend adding liquid manure when planting (Gopichand & Meena, 2015). In China, for growing ginkgo, use of fertilizers is recommended in the dose of nitrogen – 400 g per tree, phosphorus – 200 g per tree and potassium – 100 g per tree (Zhan Fan et al., 2016).  

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