







Features of Clonal Micropropagation and Anatomical Structure of Arctic Bramble (*Rubus arcticus* L.)

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ABSTRACT

The article presents the results of research on the clonal micropropagation of the arctic bramble (*Rubus arcticus* L.) at the stages of microshoot rooting *in vitro* and of regenerating plants' adaptation to non-sterile conditions (*ex vitro*). Regenerating plants of *R. arcticus* had the largest total root length in *in vitro* culture (on average 9.8 cm) when grown on Murashige & Skoog nutrient medium with the addition of 1.0mg L⁻¹ indole-3-butyric acid. The highest survival rate of regenerating plants of *R. arcticus* (90%) when adapting to non-sterile *ex vitro* conditions was noted when using a substrate from a mixture of peat with zeolite 3:1. An analysis of the anatomical structure of *R. arcticus* plants obtained by clonal micropropagation is presented for the first time. Significant anatomical and diagnostic traits of plants have been established, allowing for their species identification. The stem of *R. arcticus* is characterized by a fascicular type of structure in the upper part, while in the middle and basal part of the stem it is characterized by a transitional type of structure (from fascicular to non-fascicular). The leaves of *R. arcticus* are dorsoventral, hypostomatic, the stomatal apparatus is anomocytic. The petiole of the leaf of *R. arcticus* has a main parenchyma with 5 collateral bundles located there. The number of drupes in the fruit varied from 9 to 12; they are covered externally by a single-layer epidermis (exocarp), followed by a multi-layer storage tissue of the mesocarp. The inner part of the fruit consists of a woody endocarp (stone), inside of which is located one seed.

Keywords: Arctic bramble, *Rubus arcticus*, Clonal micropropagation, *In vitro*, *Ex vitro*, Morphological structure, Plant anatomy, Root system, Stem, Leaf, Fruit.

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INTRODUCTION

Currently, there is a close interest worldwide in the sparsely distributed berry crops of the genus *Rubus* L. due to their beneficial properties for human health caused by the content of various groups of biologically active compounds in them (George et al., 2017; D'Urso et al., 2018; Debnath & Ghosh, 2022). Arctic bramble (*Rubus arcticus* L.) is a perennial vegetatively mobile herbaceous plant of the *Rosaceae* family, reaching a height of about 25 cm. This is a Eurasian-North American Arctic-boreal species, which is mainly distributed in the temperate zone of the northern regions of the globe (Jiang et al., 2022). In nature, the plant is found in Scandinavian countries (Finland, Norway, Sweden) and North America (Canada,

U.S.A.), as well as in the northern regions of Russia. Arctic bramble grows mainly in damp places, outskirts of swamps, clearings and in the tundra. Wild plants of *R. arcticus* form fruits – juicy aggregate-accessory dark red, which contain various groups of biologically active substances and therefore have high economic value on the world market of fruit and berry products (Lindqvist-Kreuze et al., 2003; Gudovskikh et al., 2021; Tommila & Palonen, 2024). Arctic bramble fruits are sweet, fragrant, they contain about 7% sugars, 2% citric acid, tannins, and aromatic substances, anthocyanins, as well as vitamin C. Arctic bramble fruits are traditionally used by the population as food – they are eaten not only fresh, but compotes, jams are made from them, bitters and liqueurs are made (Kostamo et al., 2018).

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Today, increasing scientific attention is directed toward identifying groups of phenolic compounds in plants that act as natural antioxidants (Dai & Mumper, 2010; Wahle et al., 2010). Among these, arctic bramble fruits have emerged as a promising source of nutraceuticals and potential raw material for pharmaceutical applications due to their high content of ellagitannins and other phenolic constituents (Burlando et al., 2023). Ellagitannins, which are hexahydroxydiphenol esters of carbohydrates, represent the largest class of hydrolyzable tannins and are well recognized for their potent antioxidant properties (Newmark, 1996). The health-promoting effects of consuming arctic bramble fruits primarily result from the release of ellagic acid and the microbial metabolism of ellagitannins into urolithins in the human gut. These metabolites exhibit a wide spectrum of biological activities, including anti-inflammatory, antiviral, antibacterial, and anticancer effects, underscoring the therapeutic potential of arctic bramble as a functional food source (Sangiovanni et al., 2013).

In natural growing conditions, the arctic bramble reproduces both by seed and vegetatively through the formation of root stalks during the growing season. In nurseries, arctic bramble is traditionally propagated by cuttings or by separating root stalks from the parent plant, which requires a lot of time for large-scale production of planting material. The seed method of propagation of arctic bramble is not economically effective, since it requires a longer time to produce standard seedlings. Currently, clonal micropropagation of representatives of the genus *Rubus* attracts researchers with a high potential for large-scale production of genetically homogeneous planting material of the plant (Zayova et al., 2016; Ghadakchiasl et al., 2017; Makarov et al., 2021; Turdiyev et al., 2023). Since today there is a shortage of arctic bramble planting material all over the world to lay the necessary number of plantations, this problem can be solved by developing and introducing scientifically sound methods of clonal micro-propagation of this valuable berry crop into production. Moreover, progress in the breeding of economically valuable plants is aimed at finding methods by which improved garden forms are preserved during long-term cultivation (Makarov et al., 2024a). Methods of vegetative propagation of berry crops have now very firmly occupied their niche in the general cycle of production of healthy planting material of the plants (Debnath, 2007). Since each somatic cell of plants contains all the genes necessary for the reproduction of genetically homogeneous material, the methods of clonal micropropagation are the most promising in this regard. It should also be said that there are always many different microorganisms (bacteria, fungi, etc.) on the surface of plants, and the cells of the shoot apices are freed from them, so only clonal micropropagation allows to get absolutely healthy plants – pathogen-free clones. In the tissue culture system (*in vitro*), plants produce hundreds of identical copies within a short period of time, which greatly increases the commercial production of clones of hybrid and parent lines (Debnath, 2014). Due to the above, the development of methods of clonal arctic bramble

micropropagation is an urgent task of modern fruit growing today.

The use of zeolites in fruit and vegetable growing helps improve the agrochemical and water-physical properties of the soil (zeolites retain moisture), and consequently, increase the vegetative mass, productivity of plants and products quality (Polat et al., 2004; Szymańska et al., 2004; Abdi et al., 2006; Eroglu et al., 2017; Jankauskienė et al., 2019; Prisa, 2023; Allegro et al., 2024; Makarov et al., 2025; Sangiorgio et al., 2025). A decrease in the infectious load on European blackberry (*Rubus fruticosus* L.) plants was found when growing in protected soil using a substrate with the addition of a zeolite-containing mineral complex (ZMC) (Maslova et al., 2024). When growing raspberry (*Rubus idaeus* L.) on an inert growing medium of peat moss and perlite acquire with the addition of different ground mineral rocks (mills) rich in nutrients (including zeolite), a significant increase in the content of useful minerals (Ca, Cu, K, Mg, P, S) and phenolic compounds in fruits, as well as the CAT enzymatic activity was noted (González-Fuentes et al., 2020). However, studies on the use of zeolites in *ex vitro* adaptation of micro-plants of the genus *Rubus* (including) have not been reported. *In vitro* establishment and multiplication plant germplasm is the first step to set up a backup bioresource (genetic) collection. Genetic classification and identification of plants of the genus *Rubus* is primarily based on differences in phenotypes and chromosomal composition. Molecular markers are an effective method for analyzing the genetic diversity of germplasm resources. The creation of a *Rubus* germplasm collection seems to be extremely important today for solving the issues of taxonomy and phylogenetic relationships of species of the genus *Rubus*, in particular determining the boundaries and studying the taxonomic status of subgenera, sections, and species. Moreover, today a deeper knowledge of the transcriptome and genomic coding sequences of *Rubus* representatives is needed to facilitate the use of simpler and more effective markers of single nucleotide polymorphisms (SNPs) and insertion-deletion (InDel) markers for molecular plant breeding (Yu et al., 2022). Currently, the United States National Plant Germplasm System (USDA-ARS) contains over 2 250 *Rubus* species collection accessions, and the USDA-ARS National Genetic Resources Preservation Laboratory has cryopreserved and maintains 200 accessions in liquid nitrogen (Jenderek et al., 2025). However, the most effective practice for maintaining the *Rubus* clone collection is long-term preservation of shoot tips in liquid nitrogen using cryopreservation. The *Rubus* germplasm collection consists of extremely diverse genotypes with equally diverse responses to *in vitro* culture and cryopreservation (Bruna et al., 2023; Lu et al., 2024; Zhou et al., 2024). The cryo longevity of *Rubus* clonal germplasm (including *R. arcticus*) is largely uncharacterized but needs to be systematically documented to guide the organized and routine conservation of plant collections. Therefore, conducting additional anatomical studies for *Rubus* representatives seems to be very important today.

Conserving and documenting plant genetic resources is time- and labor-intensive, yet germplasm preservation is essential for biodiversity and for plant breeding. To ensure the conservation and sustainable use of *Rubus arcticus* (arctic bramble), it is critical to characterize its genetic diversity and geographic distribution in natural populations. Beyond identifying genetic traits, integrative analyses of genetic diversity, phylogenetic relationships, and population structure can provide the foundation for conservation strategies, germplasm management, and efficient breeding. However, the paucity of studies on the genetic diversity and population structure of *R. arcticus* using DNA markers currently limits the application of modern breeding approaches for this valuable berry crop. Accordingly, the aim of this research is to develop and optimize clonal micropropagation methods for *R. arcticus* and to elucidate the anatomical features of vegetative organs in micropropagated plants.

MATERIALS & METHODS

Research on clonal micropropagation of plants was carried out in 2023-2024 in accordance with generally accepted methods (Butenko, 1999; Makarov et al., 2023a). The objects of the study were plants of *R. arcticus* of wild forms selected in places of natural growth (Verkhnetoyemsky district of the Arkhangelsk region, Ponazyrevsky district of the Kostroma region). At the stage of micro-shoots rooting in *in vitro* culture, regenerating plants were grown at an illumination intensity of 2500-3000lux, an air temperature of 23–25°C, a relative humidity of 75-80%, a photoperiod of 16h light / 8 h dark, on a culture medium according to Murashige & Skoog (1962), including in the variant with dilution of the mineral base by 2 times (the acidity level of the medium pH (H₂O) is 5.6–5.8). To regulate growth processes, indole-3-butyric acid (IBA) and indole-3-acetic acid (IAA) were added at concentrations of 0.5 and 1.0mg L⁻¹. Three replications (10 plants for each replication) were tested in the experiment. The significant differences between means were evaluated using a two-factorial analysis of variance consistent with the stated factors (A – concentration of growth-regulator; B – composition of culture medium) and the least significant difference for 5% of the significance level (LSD₀₅) (Dospikhov, 2011).

To adapt the micro-plants obtained *in vitro* to non-sterile conditions (*ex vitro*), they were removed from a test tube, the plant roots were washed in 1% KMnO₄ solution within 30sec. Further, the plants were transplanted into cassettes with substrates. High-moor peat (fraction size 20.0–40.0mm; pH_{KCl} 2.8–3.5; mass fraction of moisture – up to 60%; ash content <5%; degree of decomposition – up to 18%; main inorganic compounds: N – up to 1.5%, P+K+Ca (in total) – up to 0.6%) was used as substrates for rooting, as well as mixtures of peat + river sand (fraction size 0.4–0.8mm) in a ratio of 3:1, peat + vermiculite (fraction size 1.0–3.0 mm) in a ratio of 3:1, peat + agro perlite (fraction size 1.0–5.0 mm) in a ratio of 3:1, peat + natural volcanic zeolite (fraction size 3.0–5.0mm) in a ratio of 3:1. At the same time, the peat was pre-steamed at a temperature not lower than +90°C for 40 minutes using a

cover with a humidity of 80-90%. Previously, river sand was washed and calcined at a temperature of +180°C for 2 hours. Moisture of natural volcanic zeolite at planting is 70%. Conditions were maintained in the adaptation room: the lighting intensity is 8000 lx, using LED lamps OSRAM Fluora L36/77 T8 (OSRAM Licht AG, Germany) with color temperature 4000K, PPFD 165μmol m⁻² s⁻¹), in the ratio red (650–660 nm) and blue (440–450 nm) light 3:1, at the air temperature is +25°C, the relative humidity 80–90%. The plants were sprayed with water daily for 14 days. Survival rate of plants was considered on the 14th day. For each treatment (substrate), 10 plants in 3 replications were tested.

A detailed analysis of the anatomical structure of microclonally propagated plants of *R. arcticus* was also carried out in the work. For anatomical analysis of plants, colored water-glycerin micro-preparations of cross sections of roots, stems, leaves and fruits of *R. arcticus* were made. The processes of lignification of plant parts were detected using a qualitative reaction of phloroglucin with concentrated hydrochloric acid. The microscopic structure of plant organs was studied in 10-fold repetition using the Biolam M-1 research microscope (model NMM–820TRF) and the MS–HDMI 4K imaging complex (LOMO, Russia). Statistical processing of experimental data was carried out using Microsoft Office Excel 2021 software.

RESULTS

As a result of research on clonal micropropagation of *R. arcticus* it was noted that the plants had the largest total root length in *in vitro* culture when grown on the full composition of the MS culture medium with a content of 1.0mg L⁻¹ IBA (on average 9.8 cm), which is 1.49 times more than when using IAA in the same concentration. At the same time, the total length of the roots of *R. arcticus* on the MS culture medium turned out to be on average 1.24 times longer than on the ½ MS medium. An increase in the IBA concentration in the composition of the culture medium from 0.5 to 1.0mg L⁻¹ contributed to an increase in the total root length of *R. arcticus* by an average of 1.24 times, whereas with the same increase in the IAA concentration, this indicator decreased slightly (by 1.08 times) (Table 1).

Table 1: Average values of the total root length of *Rubus arcticus* in *in vitro* culture, cm

Growth regulator (factor A)		Culture media composition (factor B)		Mean value
Auxin type	Concentration, mg L ⁻¹	MS	½ MS	
IBA	0.5	8.60±0.72	6.00±0.56	7.30
	1.0	9.80±0.85	8.30±0.76	9.10
IAA	0.5	7.20±0.65	6.00±0.60	6.60
	1.0	6.60±0.74	5.50±0.46	6.10
Mean value		8.10	6.50	-

LSD₀₅, cm: A = 0.93; B = 1.07; AB = 1.13.

The reliability of the obtained data on *R. arcticus* total root length is confirmed by the results of the ANOVA analysis (F statistic value > F critical value; p-value < 0.05) (Table 2).

As a result of studies conducted at the stage of adaptation to non-sterile *ex vitro* conditions, it was revealed that the maximum survival rates of *R. arcticus*

regenerating plants was found on a substrate of a mixture of peat with natural volcanic zeolite 3:1 (90%), while the minimum ones were found on a substrate of peat with river sand 3:1 (58%) (Table 3).

Table 2: ANOVA results for number of *Rubus arcticus* total root length in *in vitro* culture (n = 30; $\alpha = 0.05$)

Source	SS	df	MS	F	p-value	F critical
Factor A	30.63	3	10.21	54.63545	1.25E-08	3.238872
Factor B	15.36	1	15.36	82.19398	1.06E-07	4.493998
Factors A×B	2.13	3	0.71	3.799331	0.031264	3.238872
Inside	2.99	16	0.186875			
Total	51.11	23				

Table 3: Survival rate of regenerating plants of *Rubus arcticus* on the 14th day of adaptation to non-sterile *ex vitro* conditions, %

Substrate composition	Survival rate, %
High moor peat	62.31±4.24
High moor peat + vermiculite 3:1	65.17±4.12
High moor peat + perlite 3:1	68.01±3.87
High moor peat + river sand 3:1	58.07±4.35
High moor peat + natural volcanic zeolite 3:1	90.14±4.40
LSD ₀₅ , %	9.46

The reliability of the obtained data on *R. arcticus* survival rate *ex vitro* is confirmed by the results of the ANOVA analysis (F statistic value > F critical value; P<0.05) (Table 4).

Table 4: ANOVA results for number of survival rate of *Rubus arcticus* micro-plants in *ex vitro* conditions (n = 30; $\alpha = 0.05$)

Source	SS	df	MS	F	p-value	F critical
Between	1881,6	4	470,4	26,66316	2,58E-05	3,47805
Inside	176,4232	10	17,64232			
Total	2058,0232	14				

When studying *R. arcticus* plants obtained by clonal micropropagation, it was noted that the root on a cylindrical cross-section is characterized by a pronounced secondary anatomical structure, with clear differentiation into ground tissue, secondary bark, and central cylinder (Fig. 1).

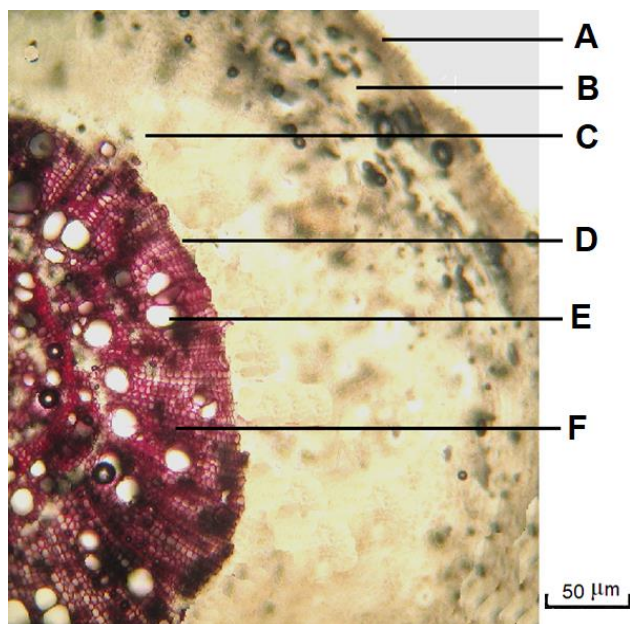


Fig. 1: Anatomical structure of the root of *Rubus arcticus* on the cross section (×200); A – cork; B – secondary bark parenchyma; C – phloem; D – cambium; E – secondary xylem vessels; F – woody parenchyma.

The covering tissue of the root is represented by several layers of cork (2-3 layers), consisting of light brown thin-walled cells. Behind the cork there is the secondary root bark parenchyma, which is composed of cells of the main parenchyma, which carry the function of a starch reserve. The cells of the main parenchyma of the bark are thin-walled, slightly tangentially elongated. The root phloem is represented by sieve-like elements and small-cell cells of the phloem parenchyma. The cambium ring is clearly expressed in the root. The root wood consists of tracheal elements (vessels, tracheids) and woody parenchyma. It should be emphasized that the root wood of *R. arcticus* does not have a pronounced radiant structure. It should also be noted that large wide-branched vessels of secondary xylem have a diffuse distribution pattern in the wood of the central cylinder of the root.

The cylindrical stem of *R. arcticus* is characterized by a transitional type of anatomical structure associated with the formation of additional conductive bundles in plant ontogenesis. It was found that the anatomical structure of the plant stem is not the same along its entire length: in the upper, younger part of it, a transition from a bundle structure to a non-bundle (solid) one was detected (Fig. 2).

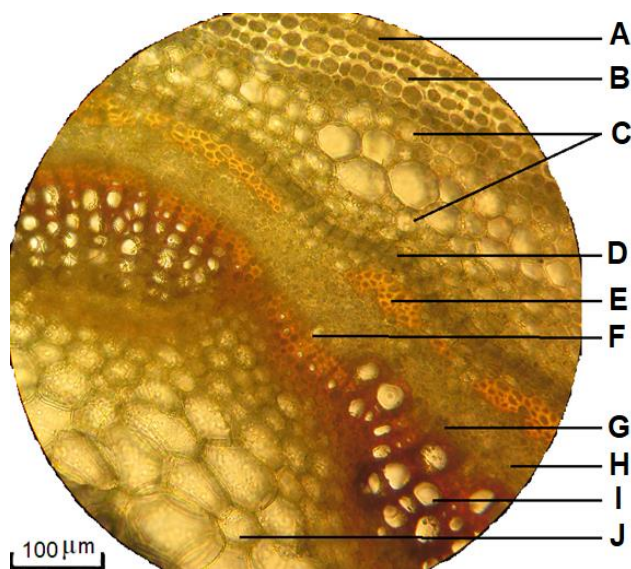


Fig. 2: Anatomical structure of the upper part of the stem of *Rubus arcticus* on the cross section (×200); A – epidermis; B – lamellar collenchyme; C – chlorenchyme; D – endoderm; E – sclerenchyma fibers; F – interstitial cambium; G – bundle cambium; H – phloem; I – xylem; J – core parenchyma.

In the stem basal part, due to the formation of numerous additional open collateral bundles from the interstitial cambium, the conductive tissues of the xylem and phloem were located on the cross section in a continuous closed ring (Fig. 3). Thus, it has been established that the true structure of the stem of *R. arcticus* is transitional. When considering a cross-section of a plant stem, the following anatomical and topographic zones can be distinguished: epidermis, primary bark, central cylinder, and core. Externally, the stem of *Rubus arcticus* is covered by a uniseriate epidermis. Beneath the epidermis lies a well-developed primary cortex comprising a subepidermal, 3–4-layered lamellar collenchyma, a multiseriate chlorenchyma, and an inner, uniseriate endodermis (starch

sheath). At the cortex–stele boundary, pericyclic sclerenchymatous fibers form a discontinuous sheath. The stele occupies much of the central cylinder and is dominated by the vascular system. In the upper internodes, the collateral vascular bundles are relatively large and clearly differentiated, each comprising cambium, primary and secondary phloem, and secondary xylem. The composition of additional open collateral bundles formed in plant ontogenesis from the interstitial cambium includes only secondary conductive elements of xylem and phloem.

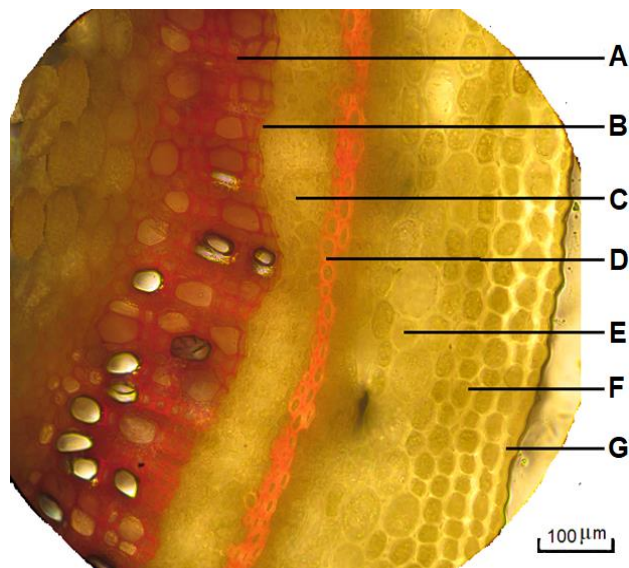


Fig. 3: Fragment of a cross-section of the basal part of the stem of *Rubus arcticus* ($\times 200$); A – xylem; B – cambium; C – phloem; D – sclerenchyma of pericycle; E – chlorenchyme of primary cortex; F – lamellar collenchyme; G – epidermis.

The central part of the plant stem consists of rather large, thin-walled cells of the core parenchyma, which carry a nutrient reserve function. It should be emphasized that the established type of anatomical structure of the stem of *R. arcticus* is a very important characteristic, which should be considered when choosing a cutting technique, as well as when developing a technological scheme and organizing work on clonal micro-reproduction of this valuable berry plant.

The leaves of *R. arcticus* are tricomponent, petiolate, with stipules, rarely pubescent with simple hairs. The edge of the leaf blades of a complex leaf is serrated. The leaf blade is herbaceous, covered on both sides with a single-layer epidermis with a cuticle (Fig. 4). The leaves of *R. arcticus* are dorsoventral, hypostomatic. The upper epidermis is underlain by two rows of palisade mesophyll; under the lower epidermis there is a multilayer spongy mesophyll, the number of rows of which varies from 5 to 6. The conductive system of the leaf blade is represented by numerous closed vascular-fibrous collateral conductive bundles.

Currently, the study of petiolar anatomy is of great importance in plant taxonomy, since the nature of the conducting system location in the petioles of leaves is species-specific (Cheryatova, 2023). In cross-section, the petiole of *R. arcticus* leaf is cylindrical, concave on the upper side, forming a groove (Fig. 5).



Fig. 4: Anatomical structure of the leaf blade cross section *Rubus arcticus* ($\times 200$); A – upper epidermis with cuticle; B – palisade mesophyll; C – collateral conductive bundle; D – spongy mesophyll; E – lower epidermis with cuticle.

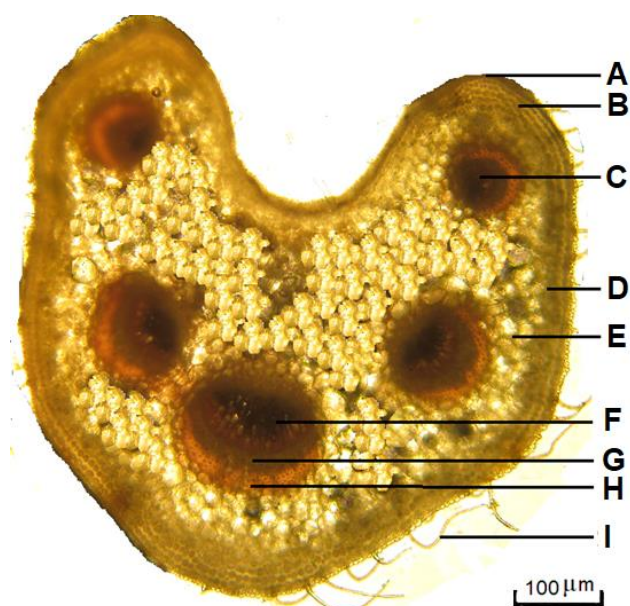


Fig. 5: Anatomical structure of the leaf petiole cross section *Rubus arcticus* ($\times 200$); A – upper epidermis with cuticle; B – lamellar collenchyme; C – collateral conductive beam; D – chlorenchyme; E – main bark parenchyma; F – xylem; G – phloem; H – sclerenchyma fibers; I – trichomes.

The petiole is covered by a uniseriate epidermis bearing sparsely distributed stomata and simple, non-glandular trichomes. Epidermal cells have smooth anticlinal walls and are elongated along the petiole's longitudinal axis. Subepidermally, a 2–3-layered lamellar collenchyma provides mechanical support, followed by a two-layer chlorenchyma forming the cortical photosynthetic tissue. The remaining internal region is occupied by large-celled ground (fundamental) parenchyma which, together with the cortical chlorenchyma and collenchyma, contributes to tissue support and—where chloroplast-bearing—photosynthetic activity. The leaves of *R. arcticus* had five collateral bundles formed in the petiole. It should be noted that the largest collateral conductive bundle was located in the central part of the plant petiole, and the other four were symmetrically distributed along its lateral parts. The bundles located near the groove of the petiole were characterized by the smallest size.

Carpological analysis showed that the fruit of *R. arcticus* is a juicy, dark red polydrupe (Fig. 6A, B).

The sepals and elements of the sub-calyx of the flowers are preserved during the ripening of the fruits of plants. The individual fruits of the aggregate fruit, drupes (Fig. 6D) are located on a dry cone-shaped receptacle (Fig. 6C). The number of drupes in the fruit varied from 9 to 12. The drupes are covered externally by a single-layer epidermis (exocarp), followed by a multi-layer storage tissue of the mesocarp, which accumulates the main group of biologically active compounds of *R. arcticus* (Fig. 6E). The inner part of the fruit consists of a woody endocarp (stone), inside of which is located one seed. (Fig. 6F).

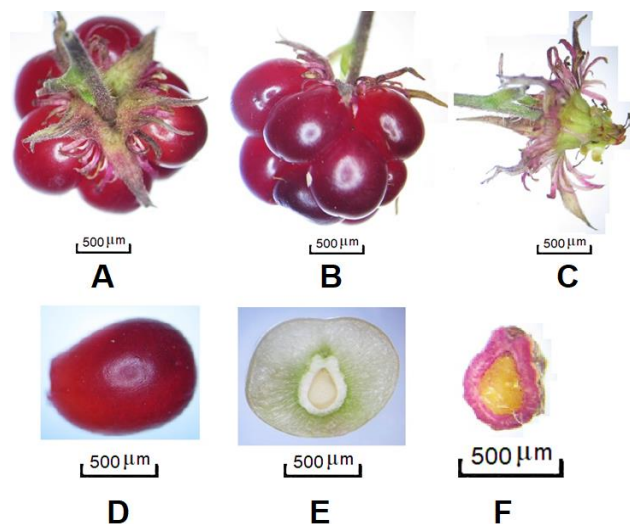


Fig. 6: Morphological and anatomical structure of fruits *Rubus arcticus* (×50); A – polydrupe (top view); B – polydrupe (side view); C – dry receptacle; D – drupe; E – cross section of drupe ovary; F – endocarp of a drupe with a seed.

DISCUSSION

To date, there have been very few studies in the field of clonal propagation of *R. arcticus*. At the same time, our results are quite consistent with the positive results of other researchers, including those using the MS culture medium (Konstantinov et al., 2012; Zontikov et al., 2020; Petrova & Sivtsev, 2022; Zontikov et al., 2022; Makarov et al., 2024b; Raeva-Bogoslovskaya et al., 2024). At the same time, the maximum rooting of regenerated plants of *R. arcticus* (82%) was revealed in one study (Raeva-Bogoslovskaya et al., 2024) when using the MS culture medium with the addition of 0.5mg L⁻¹ IAA, whereas the maximum rooting and the best morphometric parameters of plants in our study were noted with the addition of 1.0mg L⁻¹ IBA to the same culture medium.

Various studies on *in vitro* cultivation of red raspberry (*Rubus idaeus* L.) and blackberry (*Rubus* spp.) demonstrate better rooting results on nutrient media (including MS) supplemented with 0.1 to 2.0mg L⁻¹ IBA (Stoevska et al., 1995; Bobrowski et al., 1996; Gupta & Mahalaxmi, 2009; Najaf-Abadi & Hamidoghli, 2009; Vujović et al., 2010; Poncetta et al., 2012; Isac et al., 2014; Gomes et al., 2017; Ivanova-Khanina, 2018; Kefayeti et al., 2019; Raeva-Bogoslovskaya et al., 2021; Tashmatova et al., 2021; Ahmed

& Abd Elaziem, 2022; Gusev & Plaksina, 2022; Sabooni et al., 2022; Topçu, 2022; Ricci et al., 2024) and 1.0 mg L⁻¹ IAA (Georgieva et al., 2016; Damiano et al., 2007; Raeva-Bogoslovskaya et al., 2021). As a result of studies on clonal micropropagation of cloudberry (*R. chamaemorus*), it was noted that the highest values of the number and total length of roots of micro-plants *in vitro* were on the MS medium (compared to ½ MS and ¼ MS), while an increase in the concentration of both IBA and IAA in the medium by 0.5 to 1.0mg L⁻¹ contributed to an increase in the number of roots by 1.4–1.8 times, but a partial increase or decrease (depending on the form) in the length of the roots by 1.2–1.7 times (depending on the sex of the plants) (Antonov et al., 2023, 2024; Kulikova et al., 2023; Makarov et al., 2023b). These results are partially consistent with our results obtained using IBA and IAA in the MS medium during the rooting of *R. arcticus* in *in vitro* culture, which allows to identify some features of clonal micropropagation of species of the genus *Rubus*.

According to our studies (Makarov et al., 2025), the highest survival rate of lowbush blueberry (*Vaccinium angustifolium* Ait.) micro-plants in the adaptation *ex vitro* was found on a peat + zeolite (3:1) substrate and amounted to 82-89%, while in the present study, the survival rate of *R. arcticus* micro-plants on the same substrate averaged 90%, which indicates the high efficiency of using natural volcanic zeolite as a substrate component in the adaptation of berry plants compared even to vermiculite, perlite and river sand.

To our knowledge, the studies of the anatomical and morphological structure of the vegetative (roots, stems, leaves) and generative (fruits) organs of arctic bramble were not conducted. As a result of the comparative morphological and anatomical analysis, it was revealed that in the presence of common features of *R. arcticus*, characteristic of representatives of the genus *Rubus* L. (*R. caesius* L., *R. idaeus* L., *R. chamaemorus* L.), there are also signs of differences that can be used in the future when drawing up regulatory documentation for medicinal plant materials and identifying planting material by micro-morphological features. In this regard, it is important to note the following feature in the structure of the stem of *R. arcticus*, which not described in the literature for other species of the genus *Rubus* (Fell & Rowson, 2011; Korobko & Stepanov, 2016; Petrova, 2019; Gulyaev et al., 2022): the established transitional type of anatomical structure of the stem of *R. arcticus*, since the upper part of the stem was characterized by a fascicular type of structure, and its middle and basal part were non-fascicular. It should also be noted that in the upper younger part of the stem of *R. arcticus* there is a multi-row large-cell primary cortex, which differs from the structure of the cortex of other representatives of genus *Rubus* by the diffuse distribution of large-cell chlorenchyma cells. The ecological conditions of plant growth affect the structure and functioning of the stem, as a rule, indirectly, through the influence on the functioning of the roots and leaves. The root system of introduced plants, therefore, turns out to be to some extent a buffer between the stem and the external environment. Like leaves, the stems of flowering plants,

due to changes in environmental conditions of growth, are characterized by certain changes not only in the external, but also in the internal structure. The diagnostic distinguishing marker features also include the nature of the arrangement of the conductive system of the petiole of a compound leaf: in the central part of the petiole there was the largest closed collateral bundle, and four collateral bundles were symmetrically distributed along its lateral parts, forming a semicircle. The obtained results in the form of identified anatomical and morphological diagnostic features of *R. arcticus* plants obtained by the method of clonal micropropagation can serve as a basis for assessing the adaptive potential of plants to atypical growing conditions.

Conclusion

Thus, as a result of the study on clonal micropropagation of *Rubus arcticus* plants, it was noted that regenerating plants had the greatest total root length in *in vitro* culture when grown on MS nutrient medium with the addition of 1.0 mg L⁻¹ IBA. The highest survival rate of regenerating plants of *R. arcticus* (90%) when adapting to non-sterile *ex vitro* conditions was noted when using a substrate from a mixture of peat with natural volcanic zeolite 3:1. As a result of the conducted research on the anatomical features of the roots, stems, leaves and fruits of *R. arcticus*, significant anatomical and diagnostic traits of plants were established, allowing for their species identification. The obtained results on the identification of anatomical and morphological diagnostic features of *R. arcticus* can be used to assess the adaptive potential of plants to atypical growing conditions. In addition, the materials of the work will be useful for interspecific identification of representatives of the genus *Rubus* and, in particular, can be recommended for compiling anatomical atlases of fruit and berry crops. The obtained data on the anatomical structure of *R. arcticus* can also be used in matters of systematics and taxonomy of the *Rosaceae* family.

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