

## I. STRUCTURAL AND FUNCTIONAL DIVERSITY OF PLANT ORGANISMS

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### BREEDING WAY TO CREATE INTENSIVE VARIETIES OF JERUSALEM ARTICHOKE (TOPINAMBOUR) AND TOPINSA SUNFLOWER

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**Abstract:** The work substantiates the necessity for accelerated breeding of new intensive Topinambour (Jerusalem artichoke) and Topinsa sunflower cultivars endowed with permanent heterosis, suited to modern agriculture at continuous aridization of the European climate. Corresponding to the genetic algorithms of their creation and results of practical selection are adduced. Wild *Helianthus* spp. from the North American Gene Centre and topinsa sunflower can become genes' donors for tuber 'optimal cork layer' synthesis.

**Key words:** Jerusalem artichoke (topinambour), topinsa sunflower, intensive cultivars, optimal tuber cork layer, heterosis.

#### METODE DE AMELIORARE PENTRU CREAREA SOIURILOR INTENSIVE DE TOPINAMBUR ȘI FLOAREA SOARELUI TOPINSA

**Rezumat:** Lucrarea demonstrează necesitatea ameliorării accelerate a soiurilor de topinambur și floarea soarelui Topinsa, înzestrate cu heteroză permanentă, care sunt potrivite pentru agricultura modernă în condițiile aridizării continue a climei pe teritoriul Europei. Sunt prezentați algoritmi genetici corespunzători ai creării soiurilor și rezultatele practice ale selecției. Exemplarele de *Helianthus* spp. de la Centrul Genetic Nord American pot deveni donatori de gene pentru sinteza „stratului optim de plută” al tuberculilor.

**Cuvinte-cheie:** topinambur, floarea soarelui Topinsa, soiuri intensive, strat optim de plută al tuberculilor, heterozis.

#### INTRODUCTION

Topinambour (*Helianthus tuberosus* L.) or tuberous sunflower (TUS) – the plant called by the name of Indian tribes Tupinambás, coastal Brazilian natives who spoke Tupi [1]. TUS has synonyms – Jerusalem artichoke, sunchoke, sunroot. TUS is a tuberiferous perennial plant with large leaves and up to 4...5 m high stem. TUS belongs to the Compositae family and originates from the North American center of origin of cultivated plants. The plant has a polyploid genome (*viz* its haploid, base and hexaploid numbers of chromosomes are  $n = 51$ ,  $X = 17$  and  $2n = 102$ , respectively) [2, 3]. TUS was introduced into Europe by French sailors of the Marc Lescarbot's expedition in 1605. TUS, due to environmental 'plasticity' and relatively high yields, successfully naturalized in Europe, and later, thanks to its remarkable taste and healing properties, it quickly spread to Australia, as well as to countries in Asia, Africa, and South America [5].

Topinsa sunflower (TOS) is an experimental hybrid of sunflower (*H. annuus* L.,  $n = 34$ ) and sunchoke. TOS morphological and economically valuable characteristics

resemble those of TUS, but TOS has a tetraploid number of chromosomes (68), so haploid number of its chromosomes is 34, and the base number is  $X = 17$  [5].

To current moment specialists have been developing technologies to make from TUS/TOS above-ground biomass and tubers different products (inulin, pharm. preparations, and ethanol, dietary supplements [food additives, functional nutrition products, and biocorrectors] ...) that are in high market demand [6, 7]. This has prompted a number of countries to expand TUS/TOS sown area. So, in the world agriculture these crops now occupy more than 2.5 million ha [1, 5]. As for us in the Republic of Belarus (R.B.), sunchoke is industrially grown on an area of ~500 ha, it is also traditionally cultivated in the private sector (peasant estates, summer cottages and garden plots). Its tubers are used for the preparation of salads, including for the prevention and correction of blood sugar in diabetes mellitus; green mass is used in folk medicine, for example, for the prevention of joint diseases (arthrosis, bursitis, arthritis, etc.), as well as herbal baths to relieve fatigue, increase immunity and improve the overall health in the winter-spring period. The R.B. has scientific developments, patents and technical specifications for topinambour usage [5], e.g.: 1) Balneology therapeutic and health-improving sunchoke product [R.B. patent No. 17961], 2) Dietary supplement 'Kalfosil' [R.B. patent No. 17826], 3) Dietary product 'made of' topinambour [R.B. patent No. 17853], 4) Solid biofuel [TU BY 100050710.162-2012] and others.

Unfortunately, it should be noted that in conditions of Belarus intensive agriculture, the main limiting factors for the expansion of TUS and TOS are the absence of not only modern technological facilities for their cultivation and processing, but also the lack of their intensive varieties, since genetic and selection works on this crop were not carried out until 2014. With all this going on, the human world experience in crop production convincingly shows that only intensive varieties of crops cultivated in dense agrocenoses using modern technologies are able to form the maximum possible payback yield, as well as to ensure its profitability, quality and protection from adverse environmental factors, diseases and pests. At the same time both in modern agriculture and in the wild flora, the most productive dense homogeneous monocenoses consist of plants which possess xeromorphic morpho-physiological structure such as xerophytes, meso-xeromorphs, xeromorphs, succulent-xeromorphs [4, 5]. This is due, firstly, to that the 'compaction' of the monocenosis provides a greater output per area unit, and its eco-coenotic conditions act in the same direction as those of aridity and drought. Secondly, the ongoing aridization of the planet climate and the fact that this process is proceeding at a faster pace in Europe (than the world average) requires orientation of genetic selection into acceleration of the targeted creation of intensive xeromorphic plants varieties (with different types of development: spring-sown, autumn-sown, facultative, perennial), as well as to 'replenish' their cultivation technologies in different farming systems: classical, *Strip-Till*, *No-Till*, *Mix-Cropp*, *Rot-Mix* [4, 5, 7]. Consequently, research on Topinambour and Topinsa sunflower should also be aimed at more rapid breeding of their intensive varieties (endowed with above mentioned types of development), which, in turn, will significantly increase crop yields and ensure the extent of their 'industrial plantings'. The polyploidy of TUS and TOS gives an additional advantage because it make it possible to effectively use these crops in hybridization with intensive varieties of sunflower (*H. annuus* L.) [5], both to create perennial sunflower (*Helianthus perennials*) and a fundamentally new

culture of the interspecific hybrid *Helianthus solaris* (which can provide cost-effective yield of both seeds and tubers). For successful breeding work in appointed direction, a prerequisite is the presence of intensive variety models, according to which appropriate personnel can select the desired genotypes using both collected and selected materials.

## MATERIALS AND METHODS

In the 2014-2016 period, comprehensive (morpho-physiological, biochemical, immunological, agrotechnical) studies on collections of cultivated varieties (cultivars, cvs.) of topinambour and topinsa sunflower (58 samples) were carried out by us at the Central Botanical Garden of the National Academy of Sciences of Belarus (CBG NASB) and at field conditions of the partner farm 'Bortniki-agro' (R.B., near Minsk). The research based on the 'Methodology for testing the characteristics of distinctness, uniformity and stability of topinambour *H. tuberosus* L. (using generally accepted approaches for obtaining analytical information) [5, 9, 10].

## RESULTS AND DISCUSSION

It is well known [5] that the 'designed' *variety model* gives an indicative presentation of the 'object' characteristics ratio, which ensures maximum plant productivity, in the specified environmental conditions (at unlimited nutrition and moisture supply).

Based on the results of our own comparative agrotechnical, morpho-physiological, immunological and biochemical studies on the TUS and TOS collections, we developed the *model of the intensive cultivar* for topinambour tubers production (see Fig. 1), which is verbally described as follows. Plants of this model are low (70÷90 cm), have a compact habit. The optimal density of their agrocoenosis is 80÷90 thousand plants per 1 ha. The stalk is resistant to lodging, has a saturated anthocyanin (purple) color and strongly shortened monopodial and sympodial branches. The plant has dark green xeromorphic or xeromorphic-succulent leaves. The specific leaf weight (SLW) i.e. leaf mass per unit leaf area equals is 8...9 mg/cm<sup>2</sup> (congruent to Russian 'the specific surface density of the leaves'). Plants are able to quickly form an optimal surface of leaves for highly productive coenoses. The leaf area index (LAI) is 5...8 m<sup>2</sup> leaves/m<sup>2</sup> of field. Plants are tolerant to drought and resistant to a complex of economically significant fungal and viral diseases. The tuber nest is moderately compact; the tubers are easily separated from the stolons (it is important for mechanized harvesting of crops). Tubers are spherical or ovoid, 'aligned', and growing fast. The surface of the tubers is smooth (without outgrowths and 'bulbs'). The tuber lenticels (buds) are depressed and their depth is shallow. Tuber's cork layer (periderm) thickness ≥ 180 μm.

The color of the tuber peel is red-pink or white; the tuber flesh has cream, light yellow or white colors. The mass of the tuber is in a range of 60...120 g. Tubers content protein (2...3 %), inulin (16... 20 %) and other ingredients. The cork layer of the tuber is optimally developed and ensures their high shelf life (8 ... 10 months or more). The vegetation period of the sunroot from planting to ripening takes 150...160 days. Tubers and green mass are suitable for industrial processing (to obtain inulin, fructose, ethanol, food and feed products, pharmaceutical preparations, etc.). The potential yield of tubers is 70...80 t/ha (green mass up to ~140 t/ha).



Figure 1. Natural model of intensive topinambour cultivar for tuber production (a), topinambour tuber nest (b) and tuber (c)

The TUS and TOS collection material studies showed that the available gene pool contains almost all the necessary characteristics and properties for intensive varieties breeding (which are fully consistent with the model). The only key trait (characteristics) that is 'absent' in our collection is the '*optimally developed cork layer*' (*OCL*) of tubers. Accordingly, this trait needs its experimental 'synthesis' by intraspecific and interspecific hybridization. However, the algorithm for topinambour *OCL* genetic selection synthesis yet was unknown. Luckily, patterns of changes in the genetic structure of traits were already revealed during domestication and selection of traditional agricultural plants. Therefore, we propose to use these patterns as a theoretical basis for the purposeful synthesis of the topinambour tuber trait *OCL*. To our knowledge, during the process of domestication many species genotypes (e.g. of wheat, rice, corn, sunflower, barley, lupine, cotton, etc.) accumulate mutant genes (responsible for various traits [mainly recessive]), many of which exhibit non-additive character of inheritance (pleiotropy, complementarity, epistasis) [4, 5, 11]. In addition, it was shown – in case of economic necessity, the successful selection (for the maximal and stable expression of a cultural trait) occurs if specific recombination between two or more non-allelic mutant genes took place. So, for the 'synthesis' of the cork layer in the skin of Jerusalem artichoke tubers, it is logical to involve in hybridization his ecologically and geographically distant wild forms and varieties (because the genetic systems that control particular traits in different ecological niches can differ from each other). Moreover, in the case of 'by elementary signs' non-allelic genes, the cleavage will have a clearly expressed transgressive character, and the traits distribution in  $F_2$  and subsequent generations will significantly go beyond the traits expression in  $P$  (parents' forms). This should give a

real opportunity to select recombinant genotypes with new values of characters among segregated traits of progeny, including the ‘optimal cork layer’ thickness. In addition, the use of interspecific hybrids of TUS, as well as TOS, with intensive varieties of sunflower may also turn out to be a promising way. Wild forms of topinambour can have a more developed tuber cork layer (60÷85 µm) than cultural samples and varieties of the plant (30÷50 µm) [2, 3, 12]. Some topinsa sunflower (TOS-forms), for instance, cultivar ‘Novost VIRA’, also have more developed cork layers (>50 µm). So, wild *Helianthus spp.* from the North American Gene Centre and topinsa sunflower can become genes’ donors for *OCL* synthesis. That is why we elaborated the algorithm for topinambour *OCL* genetic selection synthesis (please see the scheme in figure 2).

It should be noted that the top of the selection of all crops and most of other agricultural plants are heterotic varieties. Previous achievements (hexaploid TUS and tetraploid TOS) implies future principal increase in yield and adaptability only in line with the fundamental direction of breeding – targeted creation of new sunchoke hybrid varieties endowed with permanent (continuous) heterosis.

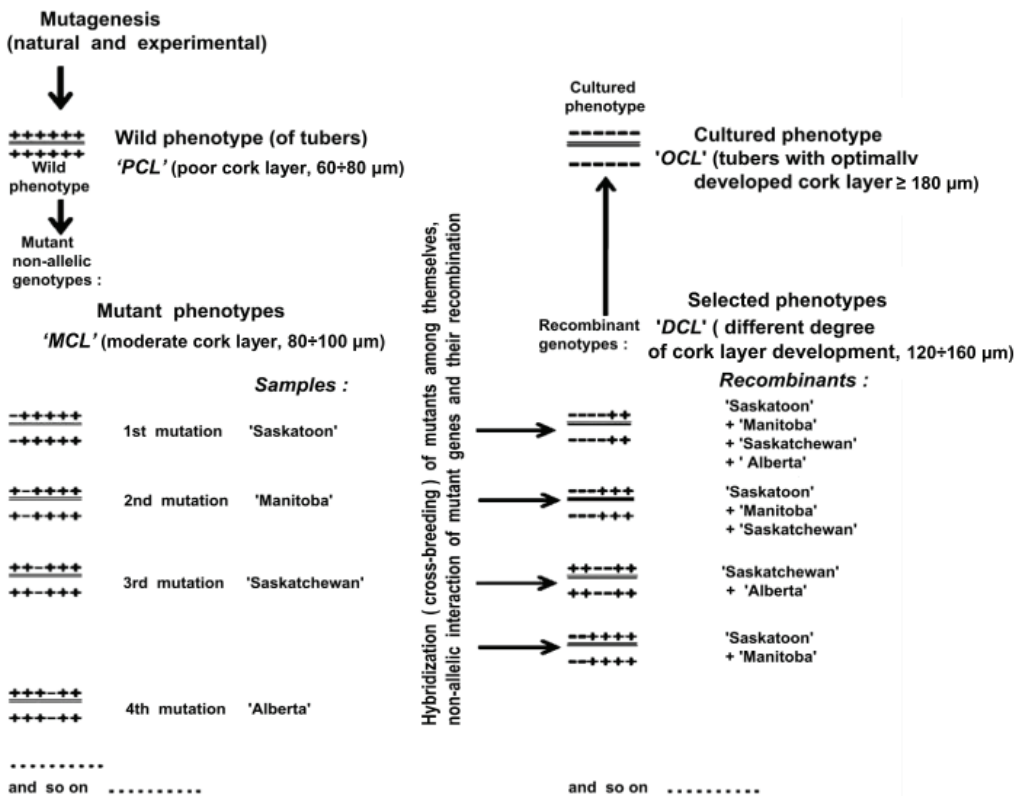


Figure 2. The algorithm for topinambour *OCL* genetic selection synthesis  
 Designation for the elementary tuber traits: *DCL* – ‘different degree of cork layer’,  
*MCL* – ‘moderate cork layer’, *OCL* – ‘optimal cork layer’, *PCL* – ‘poor cork layer’

According to the published data [11], natural and experimental polyploids (especially resulted from ‘ecological’ and ‘geographical’ crosses), form mainly heterozygous populations (representing hybrid offspring). Moreover, this is natural since polyploid organism has more chances for a favorable combination of genes (and alleles), including those causing heterosis. In accordance with this, the ability of polyploids to provide a high level of heterozygosity for a population means the basis for maintaining heterosis during lives of several hybrid generations. With this in mind, we developed an approach to create TUS- and TOS- cvs endowed with permanent heterosis (please see the scheme in Figure 3). As one can see, our breeding technology is based on the experimental synthesis of maximum hexa-allelic TUS-heterozygosity and four-allelic TOS-heterozygosity according to the desired elementary characteristics.

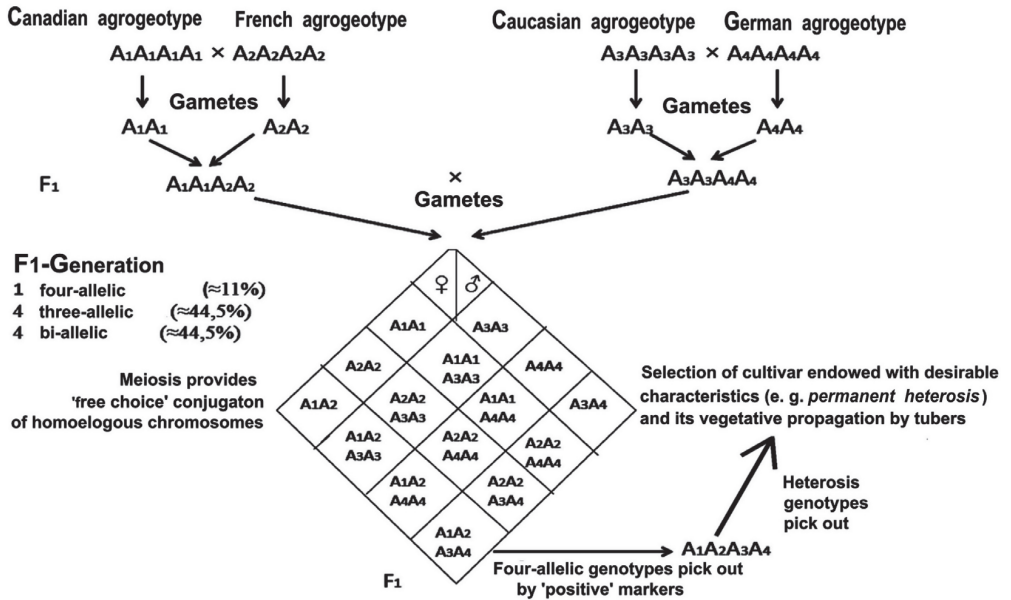


Figure 3. Technology for creating Topinsa sunflower cvs ( $2n = 68$ ) endowed with permanent heterosis

The predominant vegetative propagation of the appropriate culture varieties is able to provide multiple autorepeats of the heterosis effect on a large scale, which must be taken into account and used in the industrial TUS- and TOS- seed (sowing tubers) production.

In our work we use the developed sunchoke models: so we made the first intensive short-stemmed tuberous TUS-variety ‘*Anastas*’ (Figure 4, first bred in CBG NASB); he was introduced in 2018 to the State Register of R.B. The *Anastas* is not tall (in a dense monocenosis up to 60÷80 cm), he has a long vegetative and short generative development phases. The variety was created in conditions of severe infectious pressure by individual selection from ‘matching’ collection samples of TUS-population (resistant to complex of plant diseases). The stem of *Anastas* defies to lodging and has a weak anthocyanin (purple) color. Monopodial branching is well developed, while sympodial branching is poorly developed. Monopodial branches of *Anastas* are moderately shortened, while



Figure 4. View of the cv 'Anastas' in comparison with the standard cv 'Dessert' (on the right)

sympodial branches are strongly shortened; the foliage is abundant (the proportion of leaves from the mass of plants is ~55 %). His dark green leaves are xeromorphic. The leaf mass per unit leaf area, i.e. specific leaf weight (SLW) ~8.6 mg/cm<sup>2</sup>. The leaf area index (LAI) ~5.6 m<sup>2</sup> leaves/m<sup>2</sup> of field (at density ~9×10<sup>4</sup> plants/ha). The *Anastas* tuber nest is moderately compact (3.3 dm<sup>3</sup>); his tubers have an elongated-oval shape and are easily separated from stolons (Figure 1c). The surface of the *Anastas* tubers is moderately smooth (no outgrowths and 'bulbs'). The tuber buds are depressed and their depth is shallow. The color of the tuber peel is dark-red; the tuber flesh is white; their mass varies from 40 g to 90 g. *Anastas* tubers have a moderately developed cork layer. *Anastas* vegetation period from planting (in middle April) to harvesting tubers (in middle October) is 170...180 days (R.B., Minsk). As mentioned before *Anastas* cv is highly resistant to extreme environmental factors (to the main economically significant disease of *Sclerotinia*, to drought, to lodging). The cultivation and harvesting technology of *Anastas* is similar to that of potatoes. The yield of tubers at optimal agrocoenosis density reached 45÷55 t/ha. *Anastas* tubers are well suited for the industrial processing to obtain food, inulin, fructose, pharmpreparations, ethanol etc. The ripened crushed leaf-stem mass can be used as mulch.

In comparison with the 'intensive cv TUS-model' *Anastas* has the following shortcomings: height of the plant is 10 cm lower, the period of vegetation is 20 days longer, the tuber's cork layer is non-optimal, the yield of tubers is 25 t/ha lower.

For future breeding of intensive sunchoke varieties *Anastas* is recommended as a source of a complex of economically valuable traits (short stems; compact habit; good LAI and SLW; resistance to drought, lodging resistance and immunity to *Sclerotinia*); also as a model object for studying the 'compaction of monocenosis' and its effect on yield and quality of green mass and tubers.

Currently, we continue to work on creating intensive sunchoke cultivars using TUS/TOS collections at CBG NASB and at partner farm 'Bortniki-agro'. As a result of these efforts, two new varieties of the plant ('*Dominica*' and '*Bortnikovsky*') were submitted for State trials.

## CONCLUSIONS

1. In conditions of intensive agriculture, the main limiting factors for the expansion of cultivated areas under topinambour are the lack of his intensive varieties and improved technologies fitted for it.

2. Based on the results of 58 collection samples comprehensive study, the model of an intensive topinambour cultivar was developed; his characteristic sand parameters include xeromorphism, short stalk and compact habit.

3. In accordance with the designed parameters of the model, first intensive variety '*Anastas*' was created, which in 2018 was entered in the State Register of the Republic of Belarus.

4. The breeding-selection algorithm for creating topinambour and topinsa sunflower varieties with the characteristics '*OCL*' (optimal tuber cork layer) has been developed and is being used.

5. The genetic technology has been elaborated for the creation of topinambour and topinsa sunflower varieties endowed with permanent heterosis; this technology is based on: **a)** the principle of synthesis of maximum polyallelic heterozygosity; **b)** the ability of this plants to exclusively vegetative propagation.

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