September 14-15, 2021

The 3<sup>rd</sup> European **Buckwheat Symposium** 

# **Book of** Abstracts







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Editor: Dagmar Janovská

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# **GENETIC RESOURCES AND BREEDING (GENETICS, OMICS)**

Chairperson: Galina N. Suvorova



### **ORIGIN AND DOMESTICATION OF TARTARY BUCKWHEAT**

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Keywords: Buckwheat, Genomic variation, Metabolite variation, Domestication, GWAS

Tartary buckwheat (*Fagopyrum tataricum*) is a nutritionally balanced and flavonoid-rich crop that has been in cultivation for 4000 years and is now grown globally. Despite its nutraceutical and agricultural value, the characterization of its genetics and its domestication history is limited. Here, we report a comprehensive database of Tartary buckwheat genomic variation based on whole-genome resequencing of 510 germplasms. Our analysis suggests that two independent domestication events occurred in southwestern and northern China, resulting in diverse characteristics of modern Tartary buckwheat varieties. Genome-wide association studies for important agricultural traits identify several candidate genes, including FtUFGT3 and FtAP2YT1 that significantly correlate with flavonoid accumulation and grain weight, respectively. Furthermore, by conducting a metabolic genome-wide association study on a panel of 200 Tartary buckwheat domestication. Interestingly, we characterized a glycoside hydrolase gene FtSAGH1, responsible for the release of active salicylic acid and a subsequent disease resistance. We describe the domestication history of Tartary buckwheat and provide a detailed resource of genomic variation to allow for genomic-assisted breeding in the improvement of elite cultivars.

#### Acknowledgement

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# ASSOCIATION GENETICS IN BUCKWHEAT USING GENOME-WIDE ALLELE-FREQUENCY FINGERPRINTS

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**Keywords**: buckwheat, genomics, genome-wide allele frequency fingerprints (GWAFFs), genome-wide association studies (GWAS), diversity

Common buckwheat (*Fagopyrum esculentum* Moench) is valued for its nutritional quality and ecosystem services. However, buckwheat is considered an orphan crop lacking the appreciable genetic gains realized in major crops through conventional breeding. Agronomic and biochemical traits in buckwheat are often highly variable between sites and years, and yields lag behind major crops. Buckwheat breeding is aggravated by the highly allogamous nature due to dimorphic, sporophytic self-incompatibility of the crop, as well as unsynchronized flowering (Joshi et al., 2019). Novel breeding methods such as marker-assisted or genomic selection have the potential to overcome these issues and increase genetic gains through accelerated breeding. However, the application of novel genotyping and variant calling techniques to buckwheat accessions is challenging, as these accessions are populations of genetically distinct and highly heterozygous individuals (Nay et al., 2020). Recently, a novel genotyping approach for outcrossing species making use of genome-wide allele frequency fingerprints (GWAFFs; Byrne et al., 2013) was implemented for buckwheat (Nay et al., 2020).

In this study, we demonstrate the applicability of GWAFFs to characterize the genetic diversity among buckwheat accessions and identify quantitative trait loci through genome-wide association studies (GWAS). Twenty buckwheat cultivars were phenotyped for agronomical and biochemical properties across two years and genotyped using GWAFFs. The GWAFFs proved to be accurate genomic descriptors of the accessions with a correlation between r = 0.7-1.0 within replicates. Principal component analysis revealed clear clusters of cultivars depending on origin and mode of reproduction (self-pollination vs. outcrossing). In the phenotypic data, a high intra-cultivar diversity was observed for the agronomical traits plant height, cluster number and the biochemical traits starch, protein, and antioxidant contents. The correlation of median values between the evaluation years was low (r = 0.00-0.27) for biochemical traits and higher for agronomical traits (r = 0.70-0.86). The GWAS revealed significant marker-trait associations for height, thousand kernel weight and yield but results differed depending on the year. Due to the small number of accessions, the GWAS results should be mainly regarded as proof of concept. Nevertheless, the results demonstrate that GWAFFs enable accurate genotyping of buckwheat accessions and facilitate the application of GWAS to identify quantitative trait loci.

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### **BUCKWHEAT OMICS**

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Keywords: Buckwheat, Barcoding, Markers, PAL gene, Nutrition

Buckwheat (*Fagopyrum* sp.) has immense nutritional and nutraceutical potential. *Fagopyrum esculentum* and *Fagopyrum tataricum* are the two commonly cultivated species. All the plant parts of buckwheat possess various metabolites. It is also rich in antioxidants and has antidiabetic, neuroprotective properties. Moreover, it helps in prevention of blood clot formation.

Northwestern Himalayas of Jammu and Kashmir, India were explored for collection of buckwheat germplasm and its characterization using molecular markers. A core diverse set of 132 genotypes of buckwheat was framed and further analysed for various biochemical/ nutritional and nutraceutical parameters (such as phenol, flavonoid, antioxidants, Fe, Zn, protein, amino acids and seed endosperm rutin content). Significant variations were observed in the seed endosperm rutin content between the species as well as among the genotypes of the species. In order to understand the variations in the concentrations of seed endosperm rutin content, allele mining of PAL (Phenylalanine Ammonia Lyase) gene, was performed and SNPs were identified that might be responsible for variations in the seed endosperm rutin content.

Further, DNA barcoding [Nuclear (ITS) and chloroplast (RBCL)] of nine morphologically distinct genotypes was carried out to eliminate ambiguity in species identification. DNA barcode sequences of these genotypes were submitted to NCBI database.

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## **BREEDING BUCKWHEAT FOR HIGH FLAVONOID CONTENT**

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Keywords: common buckwheat, Tartary buckwheat, flavonoids, breeding

Two cultivated species of buckwheat, common buckwheat (Fagopyrum esculentum Moench) and Tartary buckwheat (Fagopyrum tataricum (L.) Gaertn.) are an important sources of proteins with well-balanced amino acid composition, dietary fiber and phenolic substances (Luthar et al., 2020; Vollmannova et al., 2021). Among phenolic substances, special attention is given to flavonoids with antioxidative properties. Besides well-known antioxidative effects, it is recently suggested that buckwheat, as a source of the flavonoid guercetin, may prevent health problems in patients with diabetes, and even there are possibilities for potential antiviral effects of flavonoids (Luthar et al., 2020). It was established that phenolic substances are allocated in grain and other plant parts independently. These differences should be considered during breeding buckwheat for the high concentration of targeted phenolic substances and in the selection of cultivars for nutritional and pharmaceutical purposes (Vollmannova et al., 2021). As in buckwheat grain flavonoids are allocated mainly in the embryo (including the two cotyledons), it would be expected that breeding Tartary buckwheat and common buckwheat for larger embryo will enhance the amount of the grain flavonoids. Microscopic analyses of buckwheat grain are suggested for screening in buckwheat breeding for larger embryo, and thus also for higher flavonoid content in the grain. In buckwheat main flavonoid is rutin, but during harvesting, drying and further proceeding it could be transformed to quercetin, which is a bitter substance (Germ et al., 2019). Besides adapting the postharvest treatment of buckwheat grain, breeding for the low level of the enzyme, able to transform rutin to guercetin, maybe one of the options to preserve rutin content in buckwheat grain and to prevent bitterness.

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## SLOVENIAN BUCKWHEAT GENE BANK

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#### Keywords: accession, seed, storage, biodiversity, erosion

Collections of stored plant material *in situ* and *ex situ* are valuable genetic resources and have an important role in maintaining biodiversity and preventing genetic erosion. A narrowing of biodiversity can result in partial, or in the final stage, complete genetic erosion. In Europe, buckwheat is one of the plant species whose cultivation, especially after 1960, has led to narrowing and thus, in some areas, to genetic erosion. Buckwheat is one of the most endangered plant species not only in Europe, but also according to Bioversity International in 1993, which have a decisive advantage in being collected and stored (IPGRI, 1993). Five countries in Europe in its gene banks also stores buckwheat accessions. Two collections of buckwheat are stored in Germany and one each in France, Poland, Czech Republic and Slovenia. In the Slovenian plant gene bank 392 accessions of common buckwheat and 31 accessions of Tartary buckwheat are stored. Most of them originate from Slovenia (85%), about 5% from Balkan countries and 10% from other parts of the world. The latter were collected in different parts of the world. In our gene bank we have a collection of 57 lines of breeding material. They were the basis of Slovenian varieties.

The main reason for collecting domestic buckwheat samples in Slovenia is the abandonment of growing domestic populations. Slovenia began abandoning the cultivation of Tartary buckwheat already before the World War II. After 1960, the decline was significant, as it was believed to have been suitable only for fodder. Growers also abandoned domestic populations of common buckwheat and replaced them with imported domestic and foreign varieties and populations whose seeds were available in sufficient quantities. As the remainder of the domestic populations did not mate with the then-growing material, Prof Ivan Kreft after 1975 collected most of the Slovenia's preserved buckwheat genetic resources, which are now kept at the Biotechnical Faculty, University of Ljubljana – part of Slovenian gene bank. The accessions of common and Tartary buckwheat are kept *ex situ* in cold storage, packed in glass jars with lids or vacuumed in aluminium bags. Before storing, the seed is cleaned, dried to about 8% moisture and stored at 4 °C. Buckwheat in these conditions maintains good germination up to 30 years or longer. After this period, the accessions are renewed or reproduced, but it is recommendable to do this already earlier - depending on the preserved germination.

The basic tasks and work at the gene bank are focused on the appropriate medium-term storage of accessions, which allows maintaining proper germination, regeneration and multiplication of seeds, as well as on describing and evaluating the data collected according to the International descriptors (IPGRI, 1994).

The majority of common buckwheat accessions are diploid (2n), some of them are tetraploid (4n). Tetraploid accessions are different from diploid, they have larger leaves with wavy edges, slightly larger flowers with large rounded petals and darker seeds. Only one of the tetraploid accessions has grey seeds. The collected diploid accessions can be considering the previous descriptions, roughly divided into two groups. The accessions with grey seeds, which have a finer grey seed, from light to dark grey, often with darker streaks and predominantly white flowers, only in some accessions occur individual plants with slightly pink flowers. Some plants in these accessions have the recessive gene (*d*) for determinant growth habit. They are adapted to lowland and hilly soil and climate conditions, as well as to areas without frequent early autumn frosts and fogs. In the second group are the accessions with a slightly thicker dark seed - from light to dark brown, dark streaks are often present in the basic colour of seeds. The basic colour of flowers is pale to deep pink, some flowers can be also slightly red. They are suitable for elevated, hilly locations with a 7 to 10 day shorter growing season. Accessions of Tartary buckwheat are distinguished from each other by the colour and size of seeds, plant height, branching, leaf size and earliness.

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# **CULTIVATION AND MANAGEMENT**

Chairperson: Jana Pexová Kalinová



# AGROTECHNICAL FACTORS AND BUCKWHEAT YIELD. WHAT WE KNOW SO FAR

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Keywords: cultivar, sowing term, nitrogen, yield, yield components

Common buckwheat (*Fagopyrum esculentum*) is a crop belonging to the genus *Fagopyrum* of the family Polygonaceae that is grown mainly in Europe (Russian Federation, Ukraine, Poland). Buckwheat belongs to dicotyledonous plants, however, it is classified in a pseudocereal crops because of its similar cultivation, harvesting and use of raw material. It is cultivated mainly for production of food and pharmacological products for humans.

Average world yield is considered small and amounts to 0.97 t/ha. The highest yield of 3.7 t/ha is achieved by buckwheat producers in France, followed by the Czech Republic (2.5 t/ha). In Poland, the yields are almost twice as high as the world average, amounting to 1.45 t/ha but it can vary between years and regions of Poland. Over the years a large number of studies investigated agro-technical and technological factors on grain yield and its quality were determined. It was found that the effect of agricultural factors on the growth and development of buckwheat plants was significantly modified by weather conditions during vegetation period of buckwheat, in particular by the amount and distribution of precipitation.

Sowing time, mineral fertilization and sowing density, are the main factors influencing grain yield and its quality. An increase in density of plants caused a decrease in plant height and air-dry matter, as well as the number of branches, inflorescences, and flowers per plant were affected. The optimal sowing date leads to maximum yield compared to other dates. Interestingly, one of the most important factors in selecting sowing date, considering spring sowing of this plant, is temperature.

One of the major required elements for proper growth of buckwheat is nitrogen. Nitrogen fertilization stimulates the vegetative growth and flowering of buckwheat, leaf surface increase as well as number of branches. This element also increases seed protein content. The literature states that the amount of nitrogen should not exceed certain level, because the excess nitrogen provokes growth and suppresses seed performance. The reaction of *Fagopyrum* to nitrogen depends on primary nitrogen in the soil and the amount of added nitrogen and the time of supplementation. The results showed that nitrogen fertilizer output reaches its maximum through dividing in two parts, before sowing and at the beginning of flowering.

The seeding density, and the associated number of plants per unit area, is also an important factor influencing the yield level. The method of growing buckwheat in wide rows (45 cm) allows the use of mechanical treatment against weeds. Compared to narrow crop cultivation (11 cm), higher yields of grains are obtained with 45 cm spacing. Plants create a greater number of flowers and flower stands. They have more foliage and a stronger root system.

Finally yet importantly, buckwheat cultivar strongly influences the yield. Plants of all Polish cultivars are characterized by similar morphological characters and yield-forming traits. Furthermore, the response of buckwheat to sowing density and sowing term is similar among cultivars.

In order to obtain high yields of grains, buckwheat should be provided with optimal conditions for growth and development. It is primarily the proper sowing date, appropriate sowing density and fertilization adapted to the needs of the plant and the conditions of the habitat.

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## THE EFFECT OF SELENIUM ON PHYSIOLOGICAL AND ANATOMICAL CHARACTERISTICS OF COMMON BUCKWHEAT AND TARTARY BUCKWHEAT GROWN AT DIFFERENT ALTITUDES

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Keywords: common buckwheat, Tartary buckwheat, selenium, altitude

Radiation with different wavelengths and intensities can stimulate photosynthesis, activate specific photoreceptors and/or cause harmful photomodifications of macromolecules (Verdaguer et al., 2017). Selenium (Se) is known to be essential for animals, but there is currently no evidence of its essentiality for plants (Trippe III and Pilon-Smits, 2021). Two species of buckwheat, common buckwheat (Fagopyrum esculentum Moench) and Tartary buckwheat (Fagopyrum tataricum (L.) Gaertn.), were grown at three locations with different altitudes in Slovenia: in Ljubljana (300 m a.s.l.). Podbeže (600 m a.s.l.) and Javorje (1100 m a.s.l.). Experimental plants of both buckwheat species were divided into two groups. One group was foliarly treated with potassium selenate (10 mg Se L-1; Se treated group), and another only with distilled water (control group). Biochemical parameters (contents of chlorophyll a and b, carotenoids, UV-A and UV-B absorbing compounds, and anthocyanins), physiological parameters (potential and effective photochemical efficiency of photosystem II, stomatal conductance), and anatomical parameters (thicknesses of leaves and leaf tissues, number and size of calcium oxalate druses in leaves) were measured. The results did not show the clear connection between altitude and Se treatment and parameters measured. There were not any clear gradients of values measured in plants grown at different altitudes, nor consistent differences between Se treated and control group from the same location. High values of potential photochemical efficiency of photosystem II suggest the good physiological performance of plants in all experimental groups and imply that Se treatment and/or conditions at different altitudes did not pose stress to the plants.

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## YIELD AND REPRODUCTION QUALITY OF TARTARY BUCKWHEAT ACHENES CULTIVATED IN DIFFERENT AGRO-ECOLOGICAL CONDITIONS

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Keywords: Tartary buckwheat, yield, seed quality, cultivation in Poland

Tartary buckwheat is the second most important cultivated species of the genus *Fagopyrum*. Its valuable prohealth properties arouse more and more interest among consumers who are aware of good nutrition (Luthar et al. 2021). In Poland, this species, although known for hundreds of years, has never been cultivated in pure culture. For centuries, however, it has been perceived as a weed in the cultivation of common buckwheat (sometimes other spring cereals) (Komenda et al., 1994). This form of Tartary buckwheat is found in common buckwheat fields and is limited by regulations in certified seed. Earlier observations indicate that the "weedy" form of Tartary buckwheat shows many adaptive features to the development in common buckwheat fields and it is difficult to cultivate it in pure sowing due to many wild plant features (fast maturity, rapid shedding of seeds, creeping type of growth) in the second half of the growing season (Kwiatkowski & Kłodawska, 2015). The aim of the research was to evaluate the yield and sowing value of seeds of several cultivated forms of Tartary buckwheat against the background of the weedy form, under various agro-ecological conditions in

2019 and 2020.

The yield of buckwheat was significantly differentiated, both between locations and the years of research. Tartary buckwheat yielded significantly higher on the soil of the poor rye complex type as compared to the very good rye complex. The yield of Tartary buckwheat achenes in 2020 was almost twice as high as in 2019. In 2020 Tartary buckwheat, on average, formed more seeds on the plant with a higher TG weight than in the previous year.

The reproductive value of seeds of all Tartary buckwheat accessions was very high on both locations and years of research.

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### GERMINATION AND SEEDLING GROWTH BIOASSAYS TO EVALUATE THE ALLELOPATHIC POTENTIAL OF BUCKWHEAT GENOTYPES FOR WEED SUPPRESSION

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Keywords: Allelopathic crops, weed suppression, germination, growth bioassays, chlorophyll pigments

The use of chemical herbicides has negative effects on the environment, plants, animal and human health. In addition, the growing resistance acquired by weeds also demands alternate options such as the use of allelopathic crops for weed management in agro-ecosystems. Buckwheat (common buckwheat Fagopyrum esculentum Moench. and Tartary buckwheat Fagopyrum tataricum (L.) Gaertn.), are important allelopathic crops. Allelopathic prospects of four genotypes (common buckwheat Gema, Kora, and Eva and Tartary buckwheat Westwood Ican) were evaluated on monocot (Lolium rigidum L.) and dicot (Portulaca oleracea L.) weeds in a laboratory-based germination and seedling growth bioassays. Pre-germinated seedlings of individual buckwheat genotypes were grown for one week in plastic trays having perlite. Ten seeds of either P. oleracea or L. rigidum were planted in the same plastic tray in the 2nd half using Equal-Compartment (EC) method. Germination was measured and interference in photosynthetic pigments was evaluated through HPLC. Germination of P. oleracea was significantly reduced when grown with Kora (-73%) as compared to all other varieties. Total weight of P. oleracea was only reduced with Kora (-70%). An increase in leaf length was most significant with Westwood Ican (+81%), followed by Eva (+33%), As for root length, significant increase was obtained after co-culture with Gema (86%) and Westwood Ican (72%). With these data we can conclude that Kora is the most inhibitory variety against P. oleracea while Eva produces a significant stimulation in root and leaf length. Germination and growth attributes of L. rigidum was decreased following co-culture with Gema (-81%), Westwood Ican (-65%), followed by Eva (-52%) and Kora (42%). Highest reduction in total weight was 88% and 73% after co-culture of L. rigidum with Gema and Eva. No significant changes in leaf and root length of L. rigidum occurred after treatment of 10-days with either buckwheat variety. Photosynthetic pigment contents such as chlorophyll a, b,  $\beta$ -carotene, lutein, and violaxanthin were significantly higher in P. oleracea and L. rigidium leaves compared to control trays. The neoxanthin of L. rigidum was significantly decreased when grown with Kora (73%), Eva (75.6%), and Westwood Ican (77.2%). Results showed that buckwheat genotypes are allelopathic in nature and can be an alternate option to control L. rigidum and P. oleracea. However, buckwheat genotypes differ considerably in their allelopathic potential and their performance should be checked against weeds under natural field conditions.

#### Acknowledgement

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# TEMPERATURE RISE AND WATER STRESS EFFECTS ON TWO BUCKWHEAT SPECIES (FAGOPYRUM ESCULENTUM AND FAGOPYRUM TATARICUM)

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Keywords: common buckwheat, Tartary buckwheat, abiotic stress, plant physiology, reproduction

Buckwheat is a pseudocereal with high nutritional and nutraceutical properties. Although common buckwheat (*Fagopyrum esculentum*) is the main cultivated species, Tartary buckwheat (*Fagopyrum tataricum*) is gaining interest. In most western European countries, buckwheat production has declined during the 20th century with the development of more productive crops, but it is currently receiving renewed attention for its nutritional and environmentally friendly qualities. Both species were cultivated under field conditions in Belgium and showed good yield (2037 kg/ha - 3667 kg/ha) and good seed quality (Aubert et al., 2021). *Fagopyrum esculentum* flowered earlier, produced less nodes, less branches, less inflorescences, but more flowers per inflorescence than *F. tataricum*. The yield was higher in *F. tataricum*, while the thousand-grain weight was higher in *F. esculentum*. Both species differed by their reproduction: *F. esculentum* is a heterostylous and self-incompatible species mainly pollinated by insects while *F. tataricum* produces one type of flower that can self-fertilize. However, we observed that insects visited both species: *F. esculentum* was twice as visited a greater diversity of insects than *F. tataricum*, which generally attracted smaller insects.

In the context of ongoing climate change, expected temperature rise and increase of drought periods may significantly limit plant growth and productivity of crop species. We investigated the effects of a sub-optimal temperature ( $27^{\circ}$ C vs.  $21^{\circ}$ C) and of water stress (<20% vs. 40-50\% soil water content) on *F. esculentum* and *F. tataricum* under controlled conditions (Aubert et al., 2020a, b). High temperature increased leaf production mainly in *F. tataricum* but decreased leaf area in both species. Water and photosynthesis-related parameters were affected by high temperature but our results suggested that although transpiration rate was increased, adaptive mechanisms were developed to limit the negative impact on photosynthesis. High temperature mainly affected the reproductive stage. It delayed flowering time but boosted inflorescence and flower production. Nevertheless, flower and seed abortions were observed in both species at 27 °C. Regarding flower fertility, heat affected more the female stage than the male stage and reduced the stigma receptivity. Pollen production increased with temperature in *F. esculentum* while it decreased in *F. tataricum*. Both species increased their antioxidant production under high temperature to limit oxidative stress. Total flavonoid content was particularly increased in the leaves of *F. esculentum* and in the inflorescences of *F. tataricum*.

Regarding water stress, our results suggested *that F. tataricum* was more resistant to water stress than *F. esculentum* and that *F. esculentum* had characteristics of drought avoidance, while *F. tataricum* exhibited traits of drought tolerance. The vegetative growth was affected in *F. esculentum* but not in *F. tataricum* as water stress decreased leaf production, leaf fresh, and dry weight, stomatal conductance, transpiration rate, and photosynthesis rate in the former but not in the latter. However, chlorophyll fluorescence parameters were not affected by water stress, whatever the species, and the chlorophyll content increased in water-stressed plants in both species. Oxidative stress was observed in both species in response to water stress, and antioxidant content was increased in *F. tataricum*. The reproductive phase was affected by water stress in both species: the number of inflorescences and pollen production decreased, mainly in *F. esculentum*.

Altogether, our results showed that abiotic stress response differed between buckwheat species and that reproductive stage was more affected than vegetative stage in both species but antioxidant production was boosted by abiotic stress, which could be interesting from a nutraceutical point of view.

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### PHOSPHORUS CONTENTS IN SOILS IN RELATION TO BUCKWHEAT P-UPTAKE

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Keywords: Phosphorus, soil, availability, buckwheat, varieties

Buckwheat (*Fagopyrum esculentum* Moench.) is a crop with origin in southwest China. Buckwheat appeared in Central Europe after the year 1200, after that it spread to Western Europe. Buckwheat was popular mainly among poor people in mountainous and foothill areas, where it was a part of their diet. After World War II, there was a significant decline in buckwheat cultivation. Since 1990s the renaissance of cultivation and its consumption has begun mainly due to increasing interest in healthy nutrition and organic farming. Buckwheat is characterised by a higher nutritional value compared to cereals (Mühlbachová et al., 2020). Buckwheat grows quickly and is utilized also as a cover crop, for weed suppression, a nectar source for native pollinators, and for soil nutrient enrichment, primarily phosphorus (Boglaienko et al., 2014).

The field experiments were established on organic certified fields in the frame of the Horizon2020 project ECOBREED to evaluate the effect of phosphorus uptake by different varieties of buckwheat and to determine potentially and readily available phosphorus content in soils in the Czech Republic and in Slovenia. 10 different varieties of buckwheat originating from the Czech Republic (Zoe, Zita, Zamira), Austria (Bamby, Billy), Poland (Kora, Hruszowska, Panda), France (La Harpe) and Slovenia (Eva, Čebelica) were grown in 2019 and 2020. Zamira was grown only in the Czech Republic. Plant growth, P-content and P-uptake by buckwheat was determined together with P-content in soils (extractants Mehlich 3, deionized water and 0.5 M ammonium acetate).

The results showed the differences among buckwheat varieties in their ability to mobilise phosphorus and their uptake. The weather conditions, mainly temperature and precipitations played a role in obtained results in 2019 and 2020. The differences in the growth, P-content and P-uptake were observed among selected varieties grown in field trials. Good phosphorus content and P-uptake showed in the variety La Harpe in 2019 in the Czech Republic and Slovenia and in Slovenia also in 2020. Zamira showed the best growth, P-content and P-uptake under more regular precipitations in the Czech Republic in 2020. On contrary the variety Hruszowska showed one of the lowest P-contents, yields and P-uptake by plants. The weather conditions (distribution of precipitations) affected buckwheat growth and played an important role in the capacity of buckwheat varieties to mobilize phosphorus. No significant correlation was found between P-content in soils and P-uptake was visible in varieties with the best growth and P-uptake. From the point of view of the possibilities of phosphorus uptake by various varieties of buckwheat, La Harpe and Zamira varieties appear to be the best as they showed a good plant growth and grain yield. In addition post-harvest residues can enrich the soil with phosphorus.

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# STUDY OF THE EFFECT OF SELECTIVE MEDIA WITH HIGH ZINC DOSES ON THE SURVIVAL, GROWTH AND DEVELOPMENT OF COMMON BUCKWHEAT *IN VITRO*

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Keywords: common buckwheat, selective media, zinc, in vitro

The modern methods of biotechnology application are a promising direction for increasing the efficiency of the breeding process. A significant place in solving these problems is occupied by cell-tissue selection based on the selection of genotypes that are resistant to the selective factor. Plant breeding with the use of heavy metal ions as selective backgrounds in vitro can be an effective method for creating breeding material that is tolerant to abiotic stresses (Shupletsova & Shirokikh, 2015). In addition, heavy metals belong to the group of especially dangerous toxicants-supermutagens, which can increase the number of mutations in nucleic acid molecules, leading to the appearance of genetically modified mutant cells and the production of genotypes with new economically valuable traits that are of practical interest for breeding (Sresty et all., 1999). The purpose of this work was to study the effect of selective media with high zinc doses on the survival, growth, and development of common buckwheat in vitro. As the initial plant material, we used regeneranted plants of the lzumrud buckwheat variety and the Izumrud x Inzerskaya hybrid, which were obtained as a result of repeated multiplication on a hormone-free Murashige & Skoog nutrient medium (MS). To create selective conditions, zinc salt (ZnSO<sub>4</sub> x 7H<sub>2</sub>O) was added to the main MS nutrient medium containing 20 g/l sucrose and 6 g/l agar in the following amounts according to the experimental protocols: 808, 909, 1010, 1111, 1212, and 1313 mg/l. Aseptic single-node cuttings 0.7-1.5 mm long with an axillary bud, obtained by dividing the stem (2-3 lower internodes) of the obtained test-tube regenerant plants, were cultivated for 33 days on MS medium with a standard content of zinc sulfate (control) and selective media with zinc according to the variants of the experiment. Subsequently, the surviving genotypes were microcloned onto MS nutrient media. As a result of the study, a strong inhibitory effect of high concentrations of zinc salt (808-1313 mg/l) in the nutrient medium in vitro on buckwheat plants of the Izumrud variety and the Izumrud x Inzerskaya hybrid was revealed, which was expressed in a decrease in the values of morphological parameters and inhibition of rhizogenesis. The toxic effect of high zinc doses negatively affected 7% of the total number of plants of the Izumrud variety. The Izumrud x Inzerskaya hybrid proved to be less resistant to the stress factor; 91% of microplants were viable. Subsequent multiplication on conventional MS culture media allowed the surviving genotypes to recover and show a high degree of growth and development - morphological parameters and root growth were at the level of the control indicator. As a result, for further breeding work, including for obtaining genotypes with new economic traits, F. esculentum regenerant plants that are tolerant to the high ionic load of Zn<sup>2+</sup> were selected.

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# NUTRITION AND HEALTH (PROCESSING)

Chairperson: Krzysztof Dziedzic



### **BUCKWHEAT - TRENDS IN NUTRITION AND TECHNOLOGY**

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Keywords: buckwheat, Tartary buckwheat, flour, food products, gluten-free products

The nutritional quality and properties of buckwheat give its growing popularity in the diet. The market is dominated by common buckwheat, and the production of Tartary buckwheat is also growing. New varieties of Tartary buckwheat have been registered, and the technology of husking and grinding is being developed. The offer of Tartary buckwheat is becoming more accessible for consumers. In general the offer of buckwheat products and beverages is strongly increasing in the industry. Buckwheat groats can be produced raw, or precooked before husking. Such products are also mixtures of buckwheat groats with other cereals and crops, groats are also an ingredient of some instant soups. Recent trends suggest the production of various buckwheat flours (whole meal, white, dark buckwheat flour). Low-granulation white flour contains more starch, while darker bran and husk flour contains more fiber, protein and minerals. In the technological processes of grinding buckwheat flour fractions in larger mills depends on grinding technologies and is not sufficiently researched. Individual fractions could be used for different nutritional purposes and for different categories of the population. There is also buckwheat semolina on the market, crumbs and buckwheat flour, we also know Tartary buckwheat couscous.

On the market there are many different buckwheat breads (the part of buckwheat is mostly up to 30%), we also know buckwheat toast and rusks. The use of common buckwheat prevails, and bread made from Tartary buckwheat is scarce. Ready-made mixes for buckwheat breads, pancakes, etc. are also being developed. Fresh and frozen products from puff and leavened puff pastry are becoming a regular offer in the retail network. There are also various buckwheat biscuits on the shelves. Extruded buckwheat mixed with other cereals can be found in breakfast cereal blends. Buckwheat flakes, buckwheat corn flakes and buckwheat popcorn are used as snacks. Buckwheat is an ingredient in baby food of major industrial manufacturers. It is also produced as a non-dairy beverage, as buckwheat tea and buckwheat iced tea. Buckwheat beer, buckwheat vodka, whiskey, juices and other beverages with buckwheat as well as buckwheat, ice cream and chocolate products with buckwheat groats or roasted buckwheat flour. One can also find buckwheat energy drinks suitable for athletes. Buckwheat sprouts, green buckwheat tea, as well as green leaves.

The offer of buckwheat dishes in gastronomy is becoming more and more noticeable. Buckwheat festivals, days of buckwheat delicacies, weeks of buckwheat cuisine, competitions in the preparation of buckwheat dishes are organized. Traditional and modern media are contributing to the growing popularity of buckwheat. Gluten-free, sugar-free, lactose-free products (including buckwheat) are becoming a food hit (Vombergar et al., 2020). Buckwheat products with less salt are part of the offer of healthy foods. Due to the high content of flavonoids, buckwheat in particular can be added to other food products as a functional supplement. Even more, studies on gluten-free buckwheat products, the development of technologies for the production of buckwheat products without white sugar (with sweeteners), and research on high-value additives in buckwheat products are currently underway.

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# FATTY ACIDS IN COMMON AND TARTARY BUCKWHEAT GRAINS AND FLOURS

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Keywords: whole grain, hulls, bran, light flour, polyunsaturated fatty acids

Buckwheat (Fagopyrum sp.) has been proven to be a source of many nutritive and bioactive compounds including high level of polyunsaturated essential fatty acids (Wijngaard & Arendt, 2006; Gulpinar et al., 2012). Lipids represent a small part of cereals and pseudocereals but have an important physiological role and creating the food quality. The lipid content in buckwheat grain depends on the genotype. However, the lipid content in common and Tartary buckwheat grains are rather similar and ranges from 1 to 5% (Qin et al., 2010). The composition of fatty acids in the whole grain and milling fractions, *i.e.* hulls, bran and light flour of two buckwheat species used for human consumption were studied. The common (Fagopyrum esculentum Moench.) and Tartary (Fagopyrum tataricum (L.) Gaertn.) buckwheat samples were cultivated and harvested in 2012 and 2013. Fatty acid composition for different milling fractions was determined as methyl esters (FAMEs) using gas chromatography (GC). Nine fatty acids were determined: lauric (12:0), myristic (14:0), palmitic (16:0), palmitoleic (16:1), stearic (18:0), oleic (C18:1; *n*-9), linoleic (C18:2; *n*-6), α-linolenic (C18:3; n-3) and arachidic (20:0) acid. The highest relative content was determined for linoleic acid (from 35.54 to 47.57%), followed by oleic acid (from 20.96 to 40.76%) and palmitic acid (from 13.86 to 26.42%). The total fatty acid content was the highest in bran (up to 62.64 g/kg), followed by whole grains (up to 22.93 g/kg), light flour (up to 9.69 g/kg) and hulls (up to 5.87 g/kg). Saturated fatty acid content was the highest in the hulls and the lowest in the bran. Polyunsaturated fatty acid content was the highest in light flour. High positive correlations were found between saturated fatty acids with 18 carbon atoms or less. Overall, total fatty acid content and individual fatty acid contents of common and Tartary buckwheat fractions were comparable. Both buckwheat species showed optimal nutritionally indices, i.e. atherogenic index and thrombogenic index. Beside light flour and whole grains, which are already used in wholegrain flour, bran and hulls were proven to have as well nutritionally adequate fatty acid composition. Therefore, bran addition to the flour could enrich and improve the quality of bread, while hulls as by-product could be used for animal feed.

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### EFFECTS OF VARIETY AND LOCALITY ON SELECTED MORPHOLOGICAL TRAITS AND CONTENT OF SELECTED METABOLITES IN BUCKWHEAT

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Keywords: morphological traits, bioactive compounds, international buckwheat descriptors, breeding

Based on recent knowledge buckwheat seeds are the most valuable source of various bioactive compounds e.g. rutin, hyperoside, vitexin, isovitexin, orientin, isoorientin, catechin, epicatechin and quercetin. The content of these nutritive compounds varies depending on the genotype and environment. The aim of this study was to provide the complementary research analysis of selected phenotypic and morphological traits and selected metabolites on a set of 54 common buckwheat accessions grown in 2019–2020 at two localities of Central Europe, Czech Republic and Austria. For evaluation of pheno-morphological traits (crop height, plant length, number of flowers cluster per cyme, number of seeds per cyme) international buckwheat descriptors were used. UHPLC-ESI- MS/MS was used to analyse a wide spectrum of 20 phenolic compounds in buckwheat seeds, including four flavanols, three phenolic acids, seven flavonols, four flavones, and two flavanones. In addition, crude protein content, total phenolic content and antioxidant activity was analyzed.

Constructed heatmap based on obtained data confirmed significant differences among years and genotypes in pheno-morphological traits. 1000-seed weight (TSW) were very similar at both localities (26.25 g for Austrian and 26.35 g for Czech locality) in 2019. But in 2020 there were less favourable conditions for grain formation and yield production and TSW was significantly lower with mean value 22.32 g obtained in Austria (A) and with 15.99 g obtained in the Czech Republic (CZ). Variety 'Emka' reached the maximum values of TSW in both years and at both localities. In the set of buckwheat genotypes a great variability in the most targeted bioactive compounds at both localities and years was confirmed. Protein content and total phenolic content was stable across years and localities. Antioxidant activity (AA) significantly increased in 2020 than in 2019 and mean values of AA were similar for samples grown in Prague (CZ) and Gleisdorf (AT). The sum of target phenolic compounds was observed higher for buckwheat samples in 2020 than 2019 at both localities with the highest mean value (668.65 µg/g dw) recorded for samples grown in the Czech Republic. The most abundant phenols in buckwheat grains were rutin, epicatechin, catechin, vitexin, isovitexin, orientin, and isoorientin. These results support the idea that plants in 2020 probably responded strongly to environmental conditions by synthesizing secondary metabolites as defence. Based on individual ability to adapt to changes in the environmental conditions, buckwheat accessions responded individually. The obtained results provided valuable information about buckwheat accession useful for future breeding of new buckwheat varieties suitable to the conditions of Central Europe.

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# POSTERS

Chairperson: Lovro Sinkovič



# DISLOCATION OF PHENOLIC IN BUCKWHEAT PLANT PARTS

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#### Keywords: common buckwheat, anatomical parts, polyphenolic substances, phenolic acids

Common buckwheat (*Fagopyrum esculentum* Moench.) is a source of nutrients and vitamins playing an important role in a well-balanced diet. By its consumption, we take care of proper input of vitamins, minerals and other biologically significant substances, such as phenolic compounds, which are important in disease prevention. It is the most important traditional food source in China, Japan, Korea, India, and Brazil, where it is a popular basis for buckwheat pasta production, but it is also popular in Slovenia, Czechia, Poland and Ukraine. The most frequently grown species is common buckwheat (*Fagopyrum esculentum* Moench.) and less common is Tartary buckwheat (*Fagopyrum tataricum* (L.) Gaertn.). Common buckwheat is one of the best sources of polyphenol compounds. The total polyphenolic content is discussed in many works.

Grain but also other anatomical parts of buckwheat (flowers, leaves, stems) have been considered to be the most useful sources of polyphenols. According to scientific studies, the content of total polyphenolic compounds in individual anatomic parts of common buckwheat is as follows: flowers > leaves > achenes > stems. Based on our results, we can state that the highest total polyphenol content was recorded in flowers and leaves. The buckwheat flowers are useful in infusion mixture preparation and contribute to improving the quality of honey, due to content of biologically valuable compounds.

Besides phenolic compounds, buckwheat leaves and stems accumulate a remarkable amount of leucine (Sytar et al., 2018), which could be used to develop new functional foods with a positive effect on the human health.

Of the polyphenolic compounds, common buckwheat contains phenolic acids, as well as flavonoids, which are present in the free and conjugated forms (Balasundram et al., 2006). The major flavonoids in buckwheat are rutin, quercetin, orientin, homoorientin, vitexin, and isovitexin. Among these compounds, rutin, a flavonol glycoside, has been recognized as a major antioxidant component that accounts for about 85-90% of the total antioxidant activity. Rutin has interesting effect on the human organism, it reduces high blood pressure, reduces the risk of arteriosclerosis and has anticancer activity. Of the phenolic acids, common buckwheat leaves contained mainly chlorogenic acid, neochlorogenic acid, trans-caffeic acid, trans-coumaric acid, trans-sinapic acid and trans-ferulic acid.

The production of phenolic compounds in individual anatomical parts of the buckwheat is affected by various factors, such as variety, production conditions and phenological growth phase. It was found that several phenolic substances are synthesised and allocated in flowers, leaves and grains independently. These differences should be considered when breeding buckwheat for the high concentration of the selected phenolics and when selecting cultivars for diverse nutritional and pharmaceutical purposes.

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# HOMOSTYLOUS SELF-FERTILE COMMON BUCKWHEAT BREEDING IN CHINA

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**Keywords**: *Fagopyrum esculentum* var. *homotropicum, F. zuogongense*, common buckwheat, crossing breeding, hybrid vigour.

Common buckwheat is the important buckwheat species cultivated in the world. It has heterostylous selfincompatibility type of flowers with nectary and needs insect pollination for seeding. Naturally it has a seed rate of 10-20% and has a lower unstable yield than Tartary buckwheat with homostylous self-fertile flowers by about 50%. In order to increase the seed set and yield of common buckwheat, there are many attempt to change the type of flowers. Southwest China is the origin center of common buckwheat and Tartary buckwheat, where there are the relative species and variation including homostylous self-fertile wild type of common buckwheat. Since Ohnishi (1998) discovered the common buckwheat with homostylous self-fertile type of flowers (we called it as Homo (F.esculentum var. homotropicum)), many breeders have start to cross the homostylous variation with common buckwheat varieties with heterostylous self-incompatibility type of flowers, in order to develop homostylous varieties of common buckwheat. Most of them are not successful because of unlawful pollination or the effects of wild traits such as shattering and bad plant type etc., and obviously inbreeding depression caused by self-pollination. High seed setting rate and high yield are not necessarily associated with self-crossing fertility, unless the influence of harmful gene homozygocity is removed in common buckwheat. We discovered two types of homostylous accessions in Tibet, Yunnan, and Sichuan, that is, Homo (F.esculentum var. homotropicum, diploid) and F.zuogongense (tetraploid wild species, with strong reproduction isolation with diploid and tetraploid common buckwheat) (Chen, 1999). Among them, the former is easy to cross with common buckwheat varieties with long style as female parent and its progenies showed the single dominant gene model controlling Homo trait, two dominant complementary gene model controlling shattering, and obviously inbreeding depression. After many crosses and progeny selection, we developed and registered some homostylous self-fertile varieties such as Gui Tianqiao No.1, Bai Qiao No.1, Gui Tianqiao No.2, and Su Qiao No.2 registered in 2015 by Crop Variety Examination and Approval Committee in Jilin Province, Guizhou Province, and Jiangsu Province respectively. These varieties have used in production field as a total of 100,000 hectares or more. Besides, there are three varieties such as Gui Tiangiao No.2 (white flower), Gui Tiangiao No.11 (pink flower) and Gui Tiangiao No.12 (red flower) that have applied for protection of new plant varieties in China in 2018. The homostylous self-compatibility varieties have not only a higher seed set and yield but also can be used in hybrid seed production (see Chen QF, 2007: A production method of hybrid buckwheat, ZL200710201104.9, China National Intellectural Property Administration).

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# POLYMORPHISM OF THE FLOWER STRUCTURE AND POLLEN PRODUCTIVITY IN THE GENUS FAGOPYRUM MILL.

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Keywords: Fagopyrum, flower, embryological processes, pollen productivity, pollen fertility

Despite the active use of these species in scientific research and human activities, many aspects of their morphology and reproductive biology remain unexplored up to the present moment. Our research aimed to evaluate the polymorphism of the flower structure and the development of the male reproductive sphere in buckwheat species: *Fagopyrum esculentum* Moench, *F. tataricum* (L.) Gaertn., *F. cymosum* (Trevir.) Meisn., *F. homotropicum* Ohhishi, *F. giganteum* Krotov, *F. gracilipes* (Hemsl.) Dammer, *F. rubifolium* Ohsako & Ohnishi, *F. callianthum* Ohnishi and *F. capillatum* Ohnishi.

Buckwheat samples were sown in the Laishevsky district of the Republic of Tatarstan (Russia) from 2012 to 2020. Flower morphology was studied in the field using a 10x magnifying glass. We analysed three hundred flowers of each sample. When studying the embryological characteristics of inflorescences, the inflorescences were in Chamberlain's fixative. Therefore, permanent preparations were made using the standard technique; pollen fertility was determined by using the iodine method (Barykina, 2004). The pollen productivity of the flower was estimated by calculation the average value of the number of microspore mother cells in the anther nest (Kadyrova & Mukhametshina, 2015).

As a result of these studies, it was revealed that flowers with the formula  $P_5A_8G_{(3)}$ , typical for the genus *Fagopyrum*, are found in the studied species with a frequency of 61-96%. In addition to this typical variant, variants  $P_5A_7G_{(3)}$ ,  $P_6A_8G_{(3)}$ ,  $P_6A_3G_{(3)}$ ,  $P_5A_9G_{(3)}$ ,  $P_5A_8G_{(2)}$  and  $P_6A_9G_{(3)}$  are also often found. Among the studied species, the flower was found to be the most variable in *F. esculentum*, and the least variable in *F. capillatum* and *F. gracilipes*. In *F. tataricum* anomalous flowers most often had underdeveloped anthers of the stamens of the outer circle of androecium. The most variable part of the flower in all studied buckwheat species is androecium: 47% of atypical flowers had an altered number of stamens. With a frequency of about 7%, there are atypical flowers that have an altered number of carpels that make up the gynoecium, or an atypical number of tepals and stamens simultaneously. A study of the embryological features of the species of the Cymosum group buckwheat showed that (Kadyrova et al., 2019), in general, the processes of microsporogenesis and microspore mother cells. *F. esculentum* significantly exceeded the other types of buckwheat in this indicator. Pollen productivity ranged from 665 (*F. homotropicum*) to 1397 (*F. esculentum*) pollen grains per flower.

The most frequent deviation in the development of the male reproductive sphere is the suspension of the development of sporogenic tissue cells and their subsequent degeneration. In all species of the studied buckwheat species, pollen fertility was found to be reduced, which is explained by disturbances during embryological processes during the formation of pollen. In *F. esculentum* and *F. cymosum*, pollen fertility decreased in response to drought. In *F. tataricum* in a diploid sample, pollen fertility was significantly higher than in its tetraploid analogue. *F. homotropicum* showed high pollen fertility regardless of weather conditions. The revealed polymorphism in the flower structure in the buckwheat species is partly associated with the variability of the volume of the floral meristem and partly with disturbances during embryological processes. Frequently encountered abnormal flowers with underdeveloped stamens are an external manifestation of the suspension of sporogenic tissue development, regularly accompanied by the destruction of the anther wall. The fact, that the revealed polymorphism of the flower structure is repeated in all studied species and species forms of buckwheat, both cultural and wild, suggests that this phenomenon is a characteristic of the whole genus *Fagopyrum* as a whole.

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# BIOTECHNOLOGICAL APPROACHES IN BUCKWHEAT BREEDING

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Keywords: tissue cultures, haploids, genetic transformation, molecular markers, genomics

Buckwheat is a minor pseudocereal crop from the *Fagopyrum* genus that consists of more than 25 species. Among them are common buckwheat (*F. esculentum* Moench) and Tartary buckwheat (*F. tataricum* (L.) Gaertn.), which are cultivated and used for human consumption around the world. In recent years, there has been a renewed interest in buckwheat due to its many health benefits, high nutritional value and ecological adaptability.

Genetic improvement of buckwheat by means of conventional breeding is time-consuming and the results are poor, mostly because of heteromorphic self-incompatibility (common buckwheat), indeterminant growth, seed shattering and lodging. This is why there is an increasing interest in modern biotechnological approaches in buckwheat breeding. Here we will cover the methods that have been used to some extent in buckwheat breeding, or at least in its research. Plant tissue cultures are useful for quick *in vitro* propagation of plants, for physiological and genetic studies, which can accelerate breeding, and are the basis for genetic transformations and genome editing. In buckwheat, different protocols have been optimised and *in vitro* plant regeneration from various explants, such as hypocotyls, cotyledons, immature inflorescence and anthers has been achieved. The main remaining problem in buckwheat tissue cultures is the low rate of regeneration, which is also highly determined by genotype. Through interspecific hybridization, new traits can be introduced into elite varieties. Interspecific incompatibility in the *Fagopyrum* genus can be overcome with the help of immature embryo rescue. Some studies of haploid induction by androgenesis and growth regulators were tested, haploid induction and plant regeneration were very low. If haploid induction and regeneration was more successful, perhaps by using a microspore culture, it would enable the breeding of hybrid varieties (Luthar et al., 2021).

Plant transformations are a very powerful tool in plant breeding and can be used to rapidly produce new varieties with modified traits. In buckwheat, protocols for *in vitro* and *in planta Agrobacterium*-mediated transformations and for transient gene expression have been developed, but sadly no transgenic varieties have been produced so far (Suvorova, 2016).

Different molecular markers were developed for buckwheat, though most were only used in research and not for practical breeding. PCR markers, such as random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), sequence characterized amplified regions (SCAR) and microsatellites (SSR), were used to construct linkage maps for phylogenetic studies and for the genetic characterisation of buckwheat. Some markers linked to useful agronomical traits were also developed (Yasui, 2020).

With the rapid advancement of genome sequencing methods came their reduced price, which warranted their application even in minor crops, such as buckwheat. In 2016 and 2017, the first genomes of common and Tartary buckwheat, respectively, were sequenced. Every year the number of genomic, transcriptomic and proteomic studies increases and new markers, genes, QTLs etc. are discovered. These "omics" methods will pave the way for precise genome editing of genes and will greatly facilitate the development of buckwheat varieties with improved agronomic traits (Luthar et al., 2021).

Until recently, there has not been much interest and funding of biotechnological studies of buckwheat because it is a minor crop. This is why these new biotechnological methods are mostly in the research phase and we have yet to fully exploit their great potential to accelerate the genetic improvement of buckwheat.

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### UNLOCKING GENETIC RESOURCES OF BUCKWHEAT TO DIVERSIFY SWISS AGRICULTURAL AND FOOD SYSTEMS

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**Keywords**: Buckwheat, orphan crops, genotyping by sequencing (GBS), genome-wide association study (GWAS), population genetics

The excellent nutritional quality of buckwheat (*Fagopyrum esculentum*) as well as its high ecological value makes it an extremely valuable crop for the future of agriculture (Joshi et al., 2019). However, the lack of efforts to genetically improve buckwheat – combined with the tremendous progress achieved in breeding programs of other staple crops – led to the nearly complete disappearance of buckwheat cultivation in Switzerland. Consequently, most of the domestic genetic resources of buckwheat were lost over the last centuries.

This project aims at reviving a large and diverse buckwheat collection with over 150 accessions of worldwide origin. The material will be multiplied to produce the seeds needed for conducting the planned trials and analyses. Field trials will be conducted over multiple locations and years to accurately describe the phenotypic diversity of the accessions and to identify agronomically and nutritionally interesting traits. In addition, the genetic diversity within and between the accessions will be characterized by means of genome-wide allele frequency fingerprints (GWAFFs) derived from genotyping by sequencing (GBS) (Byrne et al., 2013; Nay et al., 2020). Combining the phenotypic and genotypic information in genome-wide association studies (GWAS) will allow to link plant traits to genomic markers.

To ensure that the buckwheat varieties meet the consumers' taste and find their way into farmer's fields, this project is carried out in collaboration with ProSpecieRara – a Swiss Foundation dedicated to the conservation and utilization of cultivated plants and farm animals. In the setting of this collaboration stakeholder workshops will be organized to identify the needs of different actors in the buckwheat value chain and to actively involve them in the selection of suitable accessions.

With this project we anticipate identifying buckwheat varieties adapted to Swiss production conditions and to the market's needs. Furthermore, we aim to prepare the ground for buckwheat improvement through breeding by establishing genomic tools which will facilitate the process of combining beneficial traits of multiple accessions. The buckwheat varieties will be made available through the Swiss National Gene Bank (PGREL/RPGAA) and the outcomes of the project will be shared through stakeholder meetings and scientific publications.

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# NEW BUCKWHEAT VARIETY ROSE

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#### Keywords: breeding, phenolic compounds

Rose is a new, carnation-red flowering buckwheat variety developed in the Czech Republic. Legal protection of the variety entered into the force in April 2021. Rose variety was created by cross-breeding of varieties Rubra (originated in Russian Federation) and Victoria (Ukraine) with subsequent selection at Grassland Research Station in Zubří during 2008-2017. Cross-breeding was carried out in technical isolation (cage, pollination by bumblebees). In following years the harvested seed was sown into space isolation and during the vegetation the negative selections were conducted. During the harvest time, positive selection - choice of most suitable plants was performed. The same procedure was repeated in the following years. The selection traits were flower and stem colour, balance of maturation, weight of thousands achenes etc. In 2018, new breeding material was submitted to the state variety trials and in 2021 new variety Rose was registered. Variety Rose is characterized by middle height of stalk (90-115 cm), cordate leaves with distinct veins and especially from carnation to red coloured blossom. Time of flowering is medium. Stalk coloration is distinctly from red to purple. Maturing period it sorts among medium ripening varieties. Achene colour is light brown, thousand achene weight is 24-25 g. When compared to variety Zita, Rose has slightly higher content of nitrogen substances, but lower antioxidant activity. As well content of rutin is lower and similar to variety Zita. However, Rose has higher content of quercetin - the thermostable polyphenolic substance (Germ et al., 2019). The content of apigenin is 2-2.5 times higher in Rose compared to Zita. Rose is suitable for food industry and especially for use in seed mixtures for pollinators, bio-strips and gardens.

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# BUCKWHEAT PRODUCTION IN THE RUSSIAN FEDERATION Galina SUVOROVA, Vladimir ZOTIKOV, Vladimir SIDORENKO, Andrey POLUKHIN

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#### Keywords: buckwheat, yield, variety, determinate growth

The world buckwheat production varied from 1.5 to 2.0 million tons per year for the last two decades, although the production was twice higher than in the 70s and 90s (FAOSTAT, 2021). The record buckwheat harvest of 4.975 million tons occurred in 1992. The main producers of buckwheat in the world are Russia and China. The share of these two countries in the buckwheat production was 75% in 2019. Due to the fact that China has reduced the volume of buckwheat production, Russia has taken the first position in the world. It was harvested 898 thousand tons buckwheat grain from the sowing area of 873 thousand hectares in the Russian Federation in 2020. Buckwheat was known in Russia about 1000 years ago and has become a popular product. The main dish from buckwheat is a porridge, which is a dehulled boiled grains. Buckwheat occupies 1.1% of sowing areas of the Russian Federation. The main areas under the culture in 2020 according to the data of the Federal State Statistic Service were concentrated in the Altai Territory (55.9%), the Republic of Bashkortostan (6.7%), and Orel (6.1%), Orenburg (4.8%) and Novosibirsk (2.6%) regions (Polukhin & Panarina, 2021). The crop is also grown in other regions. Average buckwheat yield in Russia was 1.00 t/ha in 2019, that is 0.42 t/ha higher than in the world. The highest yield was achieved in the Krasnodar Territory - 2.51 t/ha, Belgorod region -1.83 t/ha, Orel region – 1.77 t/ha (Polukhin & Panarina, 2021). Fifty seven buckwheat varieties are registered in the Russian Federation in 2021. The varieties were bred by different institutions over the long time period. The oldest variety was registered in 1938 and is still grown in some regions of Bogatyr. All varieties, with the exception of Kitawasesoba were created in Russia and 18 of them were bred by the Federal Scientific Center of Legumes and Groat Crops (Orel region). The largest area is occupied by varieties bred in Orel region (Devyatka, Dicul, Design, Temp, Dialog, Druzhina) and in the Republic of Bashkortostan (Inzerskaya, Zemlyachka, Svetlana). Modern varieties created by the Federal Scientific Center of Legumes and Groat Crops are characterized by the determinate growth habit based on d (det) gene. Increase in the buckwheat yield in Russia coincides with introduction of determinant varieties. The share of determinant varieties in the total area under buckwheat in Russia is 56.7% (Fesenko & Fesenko, 2019). Therefore the Russian Federation occupies a leading position in the world buckwheat production and in the development of breeding technologies.

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# ACCUMULATION OF MYCOTOXINS IN BUCKWHEAT VARIETIES AFTER INOCULATION WITH TWO *FUSARIUM* SPECIES

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Keywords: buckwheat, Fusarium, inoculation, mycotoxins

According to European Food Safety Authority (EFSA, 2021), mycotoxins are toxic compounds naturally produced by various fungal species as a result of infection of crops before or after harvest. They can cause adverse health effects in humans and animals and include aflatoxins, ochratoxin A and Fusarium toxins such as deoxynivalenol. One of the ways to evaluate the potential of mycotoxin accumulation and better understand fungal disease infection is artificial inoculation. It is especially important in seasons when natural occurrence of fungal pathogens is low due to weather conditions. Field trials with 9 common buckwheat varieties (Eva, Bamby, Čebelica, Panda, Billy, Kora, Hruzsowska, La Harpe, Zita) and one Tartary buckwheat variety (KIS Doris) were established and evaluated using the same methodology in Prague in 2019, Czech Republic. Selected *Fusarium* genera were used for artificial inoculation to evaluate resistance differences between cultivars and to verify the methodology of artificial infection of buckwheat (suitability of inoculation timing, suitable species for infection).

Two species (*F. oxysporum, F. verticillioides*) were used for inoculation on two different dates (1st inoculation - July 25, 2nd inoculation - August 22). The inoculum (conidial suspension 0.8x10<sup>7</sup>/ml) was sprayed on the racemes of flowering buckwheat. To minimise year/site effects on results, it appeared necessary under these conditions to support disease development (if necessary) by irrigating the plots. At full maturity, seeds were harvested and analysed for the content of DON (deoxynivalenol), HT2 toxin, T2 toxin, fumonisin B1 and fumonisin B2, determined by LC - MS / MS.

The detected mycotoxin content in buckwheat varieties was relatively low. T2 and HT2 toxin were present in the greatest amount. The common buckwheat varieties Zita and La Harpe did not accumulate mycotoxins in this field trial. Very low levels of mycotoxins were also found in Tartary buckwheat KIS Doris and common buckwheat variety Hruzsowska. Common buckwheat varieties Čebelica, Bamby and Eva showed the highest accumulation of mycotoxins within the group of tested varieties. July date seems to be more suitable for accumulation. However, mycotoxins were detected only on the outer layer of buckwheat husks, which must be removed before food production, therefore mycotoxin contamination in the produced food is close to zero. These field trials were repeated in 2020 and 2021, but the data will be available after harvest 2021.

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# RESEARCHES ON STARCH PHYSICOCHEMICAL PROPERTIES OF SIX NEW PERENNIAL BUCKWHEAT HYDRIDS (*F. TATARI-CYMOSUM* Q.F. CHEN)

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#### Keywords: buckwheat, starch fine structure, thermal property

Six new types of allotetraploid Tartary buckwheat hydrids were obtained by using the annual Tartary buckwheat "Daku No.1" as the female parent and the perennial golden buckwheat "Hongxin Jingiao" as the male parent. In China Tartary buckwheat is an important crop species as it has very high nutritional and economic value. Starch is the main nutrient component of Tartary buckwheat, accounting for 40% to 60% of seeds total weight. Starch is mainly composed of amylose and amylopectin and their fine structure has an important influence on the physicochemical properties of Tartary buckwheat starch and also affects the nutritional and processing quality of the final product. In this study, 6 new perennial buckwheat hydrids (F. tatari-cymosum Q.F. Chen) were used to analyze their physicochemical properties. We have applied a series of techniques including: size exclusion chromatography (SEC) and fluorophore-assisted capillary electrophoresis (FACE) to analyze the molecular structure of debranched starch. Chain length distribution (CLDs) and differential calorimetry scanner (DSC) were used to analyze the thermodynamic properties of the sample. The FACE results showed that XX-37 is quite different from the other 5 samples, with the fewest amylopectin long chains and the most amylopectin short chains; XX-38 has the most amylopectin medium chains and the least amylopectin short chains; XX-12 has the most amylopectin long chains. The whole starch distribution results can divide the samples into three groups, XX-12 and XX-13 are a group (A), XX-6 and XX-9 are a group (B), XX-37 and XX-38 is a group (C). The amylose content of 6 varieties is higher than that of amylopectin, and the amylose content of group A is the least. The DSC results showed that six hybrid samples have very different thermal properties. XX-37 has the lowest gelatinization temperature because it has the highest amylopectin short-chain content, as the amylopectin short-chain composition is relatively low in crystallinity. XX-37, XX-38 and XX-12 have higher initial gelatinization temperature, indicating that these three starches have a more uniform and tighter crystalline structure. The starch fine structure and thermodynamic properties of the six hybrids samples are significantly different from each other. The results show that XX-12 and XX-37 have relatively unique physicochemical properties. This could benefit the molecular breeding of Tartary buckwheat in the future.

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### TARTARY BUCKWHEAT PRODUCTS

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Keywords: mill Rangus, Tartary buckwheat, Tartary buckwheat products

Mill Rangus is located in the village Dolenje Vrhpolje, it is a part of local community Šentjernej in Dolenjska region of Slovenia. Rangus traditional mill with milling stones, based on a hundred years tradition and ownership by Rangus family, have products which are very popular among bakers and other consumers. Rangus mill company is known for high quality milling products, based especially on common and Tartary buckwheat. Flour, groats, couscous and pasta are the main and special products from Tartary buckwheat. Tartary buckwheat flour, groats and couscous are special product, gluten free, rich in natural flavonoid antioxidants (rutin, quercetin).

Because of a high content of flavonoids it has special, pleasant, slightly bitter taste. From Tartary buckwheat flour we can make similar dishes and by similar process and recipes as from flour of common buckwheat. Buckwheat groats, prepared by traditional Slovenian technology are famous for having resistant (retrograded) starch. Tartary buckwheat groats husking is the technology, in Europe developed up to now only by Mr. Rangus. After cooking groats, we don't throw water away, but we may drink it as a refreshing healthy herb-tea, rich in natural antioxidants and mineral substances. Boiled groats are nice with diverse sauces, or with vegetables (for example pumpkins). Boiled Tartary buckwheat groats you can put into yogurt, cottage cheese, spread, milk or cheese cream. Tartary buckwheat waved pasta is a novel product, made from 34% of ecological Tartary buckwheat flour and 66% of ecological wheat durum grits. Waved shape of noodles has two important effects. In comparison to flat shaped buckwheat pasta, waved noodles do not stick together after cooking. Second one is that waved pasta has better connection with sauces. So Tartary buckwheat waved pasta is a nice savoury dish, with the sauce able to be well attached to pasta.

# BIOLOGICAL PROTECTION OF BUCKWHEAT FROM STRESS Lilia KLIMOVA, Fanusa KADYROVA

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Keywords: biological products, rhizospheric bacteria, buckwheat

It was revealed that inoculation of seeds by endophytes increases the adaptive potential of plants to biotic and abiotic stresses due to positive biochemical and physiological effects on plants. Our studies have shown that the treatment of seeds with a suspension containing the strain *Stroptomyces sp.* helped to preserve the activity of plant growth and laying of metamers in arid conditions.

The experiment was delivered in 2019 in the Pre-Kama climate zone of the Republic of Tatarstan. The area of the plots was  $32.2 \text{ m}^2$ , the repetition was fourfold, the placement of experience options was systematic. The technology of tillage and sowing is generally accepted for the Republic of Tatarstan. The seeding rate was 2.0 million of germinating seeds per hectare. The objective of the study was to isolate following strains from plant substrates: strain RECB – 50 B (gram-positive endophytic bacteria belonging to the species *Bacillus subtilis*); strain RECB – 95 B (gram-positive endophytic bacteria belonging to *Streptomyces spp*). Adaptogens are the products of the vital activity of endophytes isolated from the seeds of the Tatar Red millet variety.

The study of the effectiveness of the isolation protocols was carried out on the buckwheat variety Batyr using different options: when preparing seeds for sowing (RECB – 31 B with a rate of 1.5 l/t and RECB – 95 B 2.0 l/ton of seeds); leaf fertilizing at the beginning of flowering (RECB – 95 in its pure form and with the addition of an adaptogen with a rate of 1.0 l/ha); leaf fertilizing in the interphase period "fruit formation – the beginning of grain filling" with the composition RECB – 50 B with a rate of 0.5 l/ha in pure and with the addition of an adaptogen.

The analysis of biological schemes of buckwheat protection from stress showed that the most effective scheme favourably affecting plants was the RECB-31B + RECB-95B + RECB-50B variant with additional adaptogen addition (98%, 85% against the control variant of biosecurity with Rhizoplan and 83% against the variant without treatment). The variant with the use of Rhizoplan produced the largest number of fruits on the plant, which significantly distinguished this variant in terms of yield over the control without treatment (3.59 t / ha). From among the experimental variants, the yield values close to the control with biosecurity were obtained when applied according to the RECB-31B + RECB-95B + RECB-50B scheme with the inclusion of an adaptogen (3.47 t/ha or +26% to the control without treatment). The determining value in this increase was associated with a higher ecological stability of plants. In the years with the manifestation of spring-summer drought, the scheme of applying RECB-31B + RECB-95B + RECB-50B + biologics in combination with an adaptogen is most effective. And one of the main parameters determining the yield in conditions of soil-air drought is the ecological stability of plants.

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#### Notes

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