E3S Web of Conferences **395**, 03005 (2023) https://doi.org/10.1051/e3sconf/202339503005
ETSAIC2023

Developing the Adapted Scale of Microphenological Phases for the Controlled Sprouting of Grain Crops

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Abstract. As a result of the research and in view of the methodological approach to the controlled sprouting of grain crops, the term "sprouted grain" adapted to the conditions of controlled sprouting has been proposed. A unified scale of microphenological phases for controlled sprouting of different types of grain crops treated with low doses of gamma radiation has been developed. In order to take the reading for the quiescence state of the grains of the "Ekaterina" soft spring wheat and the "Remembrance of Chepelev" spring barley of 2019-2021 harvest years zoned in the Ural region, the treatment with low doses of ionizing gamma radiation (5Gy, 10Gy, and 15Gy) has been carried out. In the developed scale of microphenological phases for germination of grain crops, the dry grain phase has been introduced and the technological features of the sprouting and its effectiveness have been given. In the second phase of the sprouting, intense water absorption is observed in all groups of samples. Bioactivation of grain crops effects the increase in germination energy—up to 92-93% when treated with a dose of 10Gy. The germination rate of the 1st seed is on average 2-3 days. Low-dose gamma radiation of grains is distinguished by its industrial applicability when sprouted grains are used as an independent product or as a raw material component.

1 Introduction

One of the most important tasks, according to the *Strategy for Improving the Quality of Food Products in the Russian Federation until 2030* is the need to create conditions for the production of new generation food products with specified quality characteristics, including functional food products intended for systematic consumption in the diet of different age groups of the population.

Grain crops, primarily wheat and barley, are a strategic raw material resource of plant origin for various sectors of the food industry, in accordance with ITS (ITG) 44-2017 *Information and Technical Guide to the Best Available Technologies. Food Production*: flour-grinding, cereal and pasta production; brewing, alcohol, and bakery industries. The quality of the finished product depends on the quality of the feedstock and the innovative biotechnological methods used to increase the biological value of the product [1].

In the consumer market, the range of food products from germinated grains, both in fresh form—underground sprigs (sprouts) and microgreens for adding to salads and in

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processed one for enrichment with biologically active substances, dietary fiber, pectins, vitamins, macro and micronutrients in the development of recipes for cereals, yogurts, curd products, fermented milk products, drinks, smoothies, side dishes, breakfast cereals, flour and sugar confectionery products, minced meat products, giving them new organoleptic and functional properties, is expanding [2]. There is an expansion of the range of bakery products with a change in the recipe composition, which is a complex result of the impact of several factors, including the genotypes of different types of crops, agronomic treatments, environmental conditions, flour composition, and the technology used for the production and storage of the finished product [3]. During seed germinating, the content of enzymes increases significantly as a result of increased respiration intensity and subsequent complete and partial oxidation of carbohydrates, which contributes to the destruction of the structure of the seed endosperm. Rearrangement of the whole grain enzyme complex leads to a change in the activity of hydrolytic and amylolytic enzyme complexes [4]. Depending on the timing of malt sprouting in the process of malting under the influence of α, β, and γ-amylase, the possibility of using malt not only in brewing, but also in bakery expands [5-7]. Sprouted grains can reduce the production of phytic acid, which hinders the absorption of iron and zinc, and the formation of raffinose—a trisaccharide that causes gas and fermentation in the intestines. In sprouted grains, there are no enzyme inhibitors, due to which nutrients are absorbed in full [8].

The most important process factors in the sprouting of grains include air temperature, illumination intensity, availability of the substrate for sprouting (most often it is water), and the duration of the process. From a physiological standpoint, the sprouting of grains begins with the absorption of water by the grain and the development of two mutually exclusive biological processes—assimilation and dissimilation—in the process of metabolism.

At the same time, the bioactivation of grain in the quiescence state, following the so-called afterripening process, the importance of which is noted in [9], is characterized by two interrelated processes: the hydrolysis of substances in the endosperm and the synthesis of new substances in a grain kernel, which lead to a change in the chemical composition of the grain [10].

Since the 80s of the last century, the use of physical impact methods, in particular ionizing radiation, has been one of the effective ways to increase the biological potential of grain seeds. Seed treatment with stimulating doses of gamma radiation is one of the approaches to study the molecular basis of high yield of agricultural crops and their resistance to biotic and abiotic stress [11]. Stimulation effects are observed when plants are irradiated at doses determined for each species and depend on their physiological state, for example, for wheat, the stimulating dose of gamma radiation for wheat ranges from 5Gy to 8Gy; for barley, it varies from 10Gy to 30Gy [12-14]. The manifestation of stimulating effects after radiation treatment occurs along molecular pathways similar to other stressors [15], when low doses of gamma radiation become plant growth stimulants [16].

However, deviations of the direct "dose-effect" dependence and the manifestation of diametrically opposite results are possible when using low doses of radiation, which may be due to the qualitative difference in cell responses to treatment with different doses of radiation [17, 18].

An analysis of open scientific studies by domestic and foreign authors shows that the issues of sprouting of grain crops, including those after treatment with ionizing radiation, are considered primarily in terms of crop production. In our opinion, in the context of the active use of sprouted grain in various sectors of the food industry, it is necessary to conduct comprehensive studies on the differentiation of the stages of sprouting of grain crops, which are subsequently used sprouted grains as independent food products or as food ingredients of high biological value, given the varietal characteristics of grain crops, which determines the purpose of the experiment—the development and study of a unified scale of controlled sprouting broken down into microphases for bioactivated crops treated with low doses of gamma radiation.

2 Materials and Methods

As objects of study, grain crops that occupy the 1st–2nd places in terms of gross harvest in the Russian Federation and are represented by varieties released in the territory of the Ural region have been selected: the "Ekaterina" soft spring wheat and the "Remembrance of Chepelev" spring barley of different harvest years (2019-2021), which are used for food, production, and feed purposes. The "Remembrance of Chepelev" spring barley grain is large, elliptical, aristulate, yellow in color, with a well-defined groove and a seed length of 9.6±0.5mm, a width of 4.1±0.3mm, and a thickness of 2.9±0.2mm. The "Ekaterina" soft spring wheat grain is medium, elongated, colored, yellow-creamy in color; its groove is shallow and narrow, with a seed length of 6.8±0.2mm, a width of 2.8±0.2mm, and a thickness of 2.6±0.2mm.Selected grains meet the requirements of TR CU 015/2011, GOST 28672-2019, GOST 27186-86, and GOST 9353-2016. For sprouting grains of each type of grain crop, 1 control group (not treated with radiation) and 3 experimental groups (treated with low doses of gamma radiation—5Gy, 10Gy, and 15Gy) have been formed. In each group, 500 seeds have been selected. Radiation treatmenthas been carried out on the RTU-3000 complex (gamma radiation source—^{60}Co) at Era RCIT (Regional Center for Irradiation Technologies). Thesprouting of grains has been carried out under the same experimental conditions: indoor air temperature of +(20±0.5)°C, mixed natural lighting (day/night—16h/8h), air humidity of 73±3%. The grains have been placed in the germinating chambers between layers of moistened filter paper with periodic irrigation with water at a temperature of +(20±0.2)°C, in accordance with the requirements of GOST 12038-84.Phenological observations were carried out 2 times a day. All studies were performed, according to generally accepted methods in five repetitions.

3 Results and Discussion

For a differentiated approach to the terminology of grain after sprouting, based on its subsequent use—for growing agricultural plants or for consumption in the diet in fresh or processed form, the definitions and/or concepts used in reference sources and research literature have been investigated (Table 1).

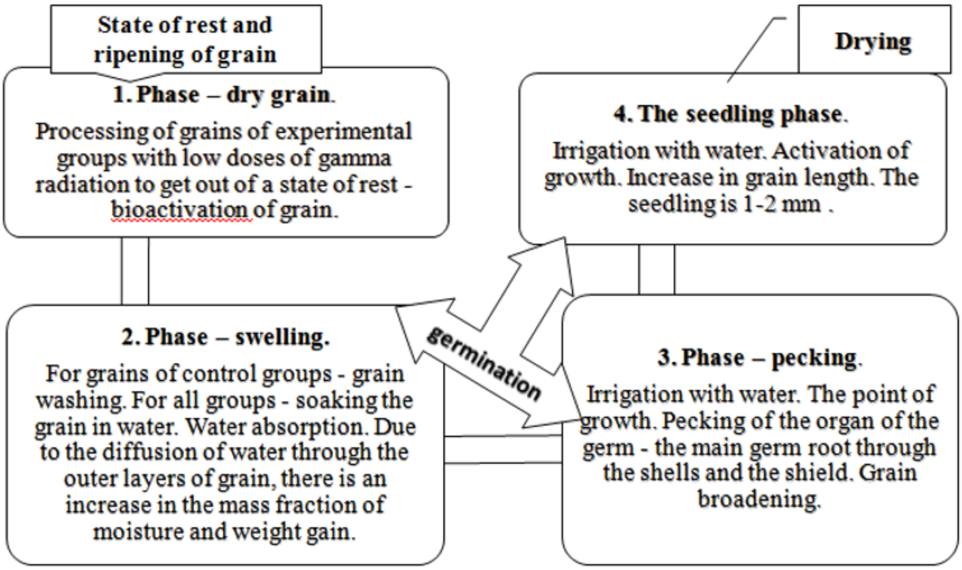
Table 1. Assessment of the conceptual apparatus.

Source	Term	Features
GOST 27186-86	germinated grain	radicles or sprouts that have come out of the tegmen
GOST 5060-2021	germinated grain	outgrown radicles or sprouts
GOST 12038-84	germinated seeds of wheat, rye, barley, oats, triticale	at least 2 normally developed radicles larger than the length of the seed and a sprig measuring no less than half of its length with visible primary leaflets, occupying at least half the length of the coleoptile
[19]	grain after sprouting	sprig length (1.2±0.2) mm
[10]	bioactivated grain	sprout length up to 2.5mm
[20]	germinated grain	sprig (sprout) does not exceed the grain length
[21]	germinated grain	visible changes that distinguish germinated grain from non-germinated

Author's approach	sprouted grain	sprouts of radicles or sprigs 1-2mm long
	germinated grain	at least 2 normally developed radicles larger than the seed length, a sprig—no less than half of its length

In our opinion, it is necessary to take into account the area of subsequent use of grain after controlled sprouting (the author's approach): for growing agricultural plants, the presence and length of radicles and sprouts, as well as the root system development are important; when used in the food industry—the presence of hatched sprouts up to 1-2mm; when sold fresh—the presence and length of the sprig itself.

At the next stage, taking into account the results of our own research and a number of scholars [10, 22-25], a unified scale of microphenological phases (MPP) of sprouted grain crops under controlled conditions has been developed (Figure 1). For samples of the control group, the first microphenophase shallnot be applied.



1. Phase – dry grain.
Processing of grains of experimental groups with low doses of gamma radiation to get out of a state of rest - bioactivation of grain.

2. Phase – swelling.
For grains of control groups - grain washing. For all groups - soaking the grain in water. Water absorption. Due to the diffusion of water through the outer layers of grain, there is an increase in the mass fraction of moisture and weight gain.

3. Phase – pecking.
Irrigation with water. The point of growth. Pecking of the organ of the germ - the main germ root through the shells and the shield. Grain broadening.

4. The seedling phase.
Irrigation with water. Activation of growth. Increase in grain length. The seedling is 1-2 mm .

Fig. 1. Scale of microphenological phases of sprouted grains of different types of crops.

In the *"dry grain"* phase, the samples of the experimental groups of grain have been treated with low doses of ionizing gamma radiation (5Gy, 10Gy, 15Gy), which can cause the effect of hormesis, in order to remove the state of metabolic quiescence, and to start physical and chemical processes against the background of weakening regulatory mechanisms, limiting the functional activity of grains [26, 27].

In the *"swelling"* phase during the soaking process, the most intensive water absorption occurs, that is, the grain is impregnated with moisture.The moisture content of barley grain has increased from 11.3-13.2% to 30.7-34.3% and wheat grain—from 11.5-12.2% to 31.9-34.8%, while in this case, leaching of minerals from the husk occurs. As a result of the launch of hydrolytic processes in the endosperm and the transition to a soluble state of amino acids, peptides, sugars, diffusion of nutrient-saturated water occurs through the epithelium layer and scutellum to rudimentary processes and radicles with an increase in the moisture content of the kernel. At the same time, the hydrolytic process is enhanced under the influence of low doses of gamma radiation as a result of the activation of the free radical process with the formation of hydroperoxides and specific changes at the level of gene expression due to increased oxidative processes.

At the *"hatching"* phase, despite the decrease in the rate of water absorption, the moisture content in barley grain increases by 4.1-7.7% and in wheat grain—by 3.8-5.8%; a growth point occurs with the sprouting of the germinal root through the shells and shield as a result of the ongoing hydration of water, which promotes the hydrolysis of macromolecular compounds and the accumulation of osmotically active substances. Visually, we can determine the change in the grain—its broadening and the appearance of a white sprout from under the fruit and seed coats.

In the *"sprout"* phase, the moisture content reaches a relatively equilibrium state, when the hydrostatic pressure in the cells is comparable to the osmotic pressure created by the cell sap. Humidity of wheat grain reaches 41.1±0.9% and for barley grain—43.2±0.8%. The sprout length is 1-2mm with an increase in the length of the grain itself; enzymatic activity increases under the influence of amylases, proteases, lipases, peptidase, and gluconase.As a result of hydrolytic processes, starch, protein, and lipids are reduced with the formation of saccharides and simple sugars, peptides and free amino acids, glycerol and free fatty acids.

For the use of sprouted grain in the food industry, it is advisable to limit ourselves to the "sprout" phase, according to the proposed scale of microphenological phases for the sprouting of grain crops.

The effectiveness of radiostimulation with different doses of radiation for grains of the "Ekaterina" wheat and the “Remembrance of Chepelev” barley has been assessed (Table 2).

Table 2. Germination energy after 3 days, %.

Indicators	Radiation dose, Gy			
	0	5	10	15
Wheat the "Ekaterina"	83±2	89±2	93±1	86±1
Barley the "Remembrance of Chepelev"	74±3	86±1	92±3	82±3

The germination energy in samples of the control groups of wheat and barley grains is 83% and 74%, respectively. In experimental samples of wheat, the growth energy after 3 days is 89%, 93%, and 86% when treated with doses of 5Gy, 10Gy, and 15Gy, in experimental samples of barley—86%, 92%, and 82%, respectively. Treatment with low-dose gamma radiation leads to an increase in the energy of seed germination, while most of the grains hatched after 2-3 days, regardless of the dose of radiation, which may be due to the effect of free radicals when water and oxygen intake, which are catalysts for starting the process of grain germination. The results obtained are comparable with the conclusions of a number of authors [28, 29], when exposure to ionizing radiation in stimulating doses, provided that the irradiation process is controlled, leads to an increase in the sprout due to the germination energy.

4. Conclusion

As a result of the research and developed unified scale of microphenological phases of sprouting of the grains of the "Ekaterina" soft spring wheat and the “Remembrance of Chepelev” spring barley, untreated and treated with low doses of gamma radiation, the term "sprouted grain" has been formulated: "sprouted grain is a grain with sprouts of radicles or sprigs 1-2mm long obtained as a result of controlled sprouting conditions". In the adapted scale of microphenological phases of grain crop sprouting, in contrast to the previously proposed scales, a phase of dry grain has been introduced; at each phase, clarifying signs of the phase, the mechanism of development, and the result have been implemented. It has been experimentally proven that the biostimulation for grains of different types of crops effects the intensity of growth due to the germination energy. The optimal dose of radiation is in the range of 5-10Gy, with no significant differences depending on the year of harvest. Based on the study of the results of empirical investigations, it can be noted that low-dose gamma radiation of grain crop seeds is distinguished by its industrial applicability under conditions of controlled sprouting for use as a food product and a raw material component in the production of functional food products.

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