PLANT SCIENCE TODAY
ISSN 2348-1900 (online)
Vol x(x): xx–xx
https://doi.org/10.14719/pst.3193

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RESEARCH ARTICLE

Development of technology for the manufacture of yogurt from combined cow and mare's milk enriched with plant extract in Kazakhstan

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OPEN ACCESS

ARTICLE HISTORY
Received: 12 December 2023
Accepted: 25 January 2024
Available online
Version 1.0 : 19 February 2024

Check for updates

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

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Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS, UGC Care, etc
See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

CITE THIS ARTICLE

Tleuova K Z, Shingisov A U, Vetokhin S S. Development of technology for the manufacture of yogurt from combined cow and mare's milk enriched with plant extract in Kazakhstan. Plant Science Today (Early Access). https://doi.org/10.14719/pst.3193

Abstract

In recent decades, more and more consumers are paying attention to a healthy diet and foods enriched with nutrients. In response to this demand, new production technologies are being actively developed in the food market. One of the interesting innovations in this area is the development of a technology for making yogurt from combined cow and mare milk enriched with plant extract in Kazakhstan. The purpose of this work is to determine the optimal technology for the manufacture of yogurt from combined milk, with the integration of advanced methods for processing the finished product and raw materials and to consider ways to obtain industrial strains of lactic acid bacteria that can potentially be used in the manufacture of such a product. To determine the list of bacteria, the data prescribed in the state standards and the results obtained in the publications of the reviewed studies were used. Also, the work uses theoretical methods of scientific knowledge, such as deduction, induction, abstraction, analysis and comparison of methodologies described in the scientific literature and synthesis based on their theoretical methodology for further testing. The state regulatory documents regulating various aspects of the manufacture of fermented milk products have been studied. The probable optimal sequence of processes for the manufacture of a fermented milk product has also been determined, starting from the preliminary preparation of raw materials of animal origin, to their combination and fermentation itself. At the same time, the number of manipulations carried out due to exposure to high temperatures is minimized. The main directions for checking the compliance of the finished product with state standards defined for similar products and how such a product will meet consumer expectations are outlined. The results of this work will form the theoretical basis for experimental studies to be carried out in the future.

Keywords

Fermentation; fermented milk product; lactic acid bacteria; plant extract

Introduction

The consumption of dairy products is widespread throughout the world and is estimated at more than 700 million tons/year. Every year this figure only grows (1). About 83% of world production is cow's milk. In the Republic of Kazakhstan, this figure exceeds 99% and milk consumption per capita is estimated at 160-220 kg. (2, 3). A significant share of total consumption is sour milk products. Thus, to meet the needs of the population in dairy products, it is necessary to constantly increase the productivity of milk yields and increase the number of livestock on farms and farms. Cow's milk is one of the most common foods in the diet, rich in protein, calcium and other important trace elements. Mare's milk, in turn, has unique properties such as a high content of vitamins and minerals and low cholesterol content. In addition, mare's milk is more easily absorbed by the body and has antioxidant properties (4).

By combining these 2 kinds of milk, a balanced combination of nutrients can be achieved and a product that combines the best qualities of both types of milk can be created. The most common such reasons are intolerance to lactose or other components of milk, religious reasons, as well as popular beliefs about the effect of drinking milk on an increase in the risk of cardiovascular disease and blood cholesterol levels, which, given the data of recent years, are beginning to be refuted. Thus, in a review study, data on the negative impact of the use of saturated fatty acids and other milk fats on the development of cardiovascular diseases are analyzed (5). The authors conclude that the data that indicated a significant positive correlation could not be entirely correct and are inclined to believe that milk consumption has a neutral effect on cholesterol levels. Nevertheless, it is suggested that reducing the content of saturated fatty acids may indeed be appropriate (5, 6). Later, data were compared from various studies on the composition of milk, which showed that mare's milk does indeed contain less dangerous saturated fatty acids.

Studies of the properties of mare's milk are devoted to the work of several domestic scientists (8-20) and the issues of using probiotic lactic acid cultures in the production of fermented milk products are considered in another work (21-24, 26-34).

Currently, a wide range of wild medicinal herbs are used as raw materials for the production of biologically active substances (30-32). Plant extracts can be used to give the product additional nutritional characteristics.

Currently, various types of wild, aromatic and medicinal plants are widely used as biologically active additives in the food industry. Enrichment of the composition of food products with an extract of wild, aromatic and medicinal plants not only increases their nutritional value but also gives them a functional orientation. The southern regions of the Republic of Kazakhstan are rich in ecologically clean aromatic and medicinal plants with medicinal properties, which can be a raw material base for obtaining polyphytocomponents to enrich the composition of new domestic functional food products.

The choice of a specific herbal extract depends on the goals of the developer. For example, the addition of an extract is used in the form of coltsfoot leaves, in the form of an infusion, for diseases of the respiratory tract, as an expectorant, disinfectant and anti-inflammatory agent, which positively affects blood health. The extract can impart antioxidant properties to the product.

Considering the need to reduce the negative impact on cholesterol compared to pure cow's milk, to achieve the optimal combination of proteins and composition and at the same time not to lose the quality of sensory properties, it seems rational to make fermented milk products from combined milk. In addition, this is one of the ways to reduce the cost of dairy production, which will help reduce the shortage of fermented milk probiotic products in third-world countries (12, 13).

The concept of "combined milk" herein after is used as a mixture of animal milk and plant extract in optimal proportions. The purpose of this study is to develop a technology for making yogurt from combined milk, in particular, to determine the optimal ratio of plant and animal components, depending on the needs and asking consumers. Particularly consider the methodology of processing vegetable raw materials with the preservation of the greatest amount of nutrients and minimizing, as far as possible, undesirable compounds that make up fermented milk products.

Materials and Methods

This work is based on a study of scientific publications related to the feasibility and ways of using combined milk for the manufacture of yogurt, which would satisfy the needs of consumers as a source of probiotics and protein as well as its sensory properties. Several articles were selected in which they examined various technologies and methods for processing plant material and preparing mare's and cow's milk, their combination and further fermentation. The basis of this study was data obtained from 4 publications devoted to a review of the latest methods for the processing of vegetable raw materials and the processing of combined milk (14-17), which were the basis for drawing up a scheme for further manufacturing of a yogurt product.

Also, among the materials, state standards and requirements for the commissioning of own strains (20, 21) and requirements for dairy and sour-milk products certified in the Republic of Kazakhstan (22-24) were studied.

The main methods used in the work were the methods of theoretical knowledge. First of all, analysis - when studying the scientific literature on the topic, the data described in the articles are disassembled into its parts, each of which is considered separately. Thus, the presented methods, the mechanisms and nature of their work, the logic of the sequence of their application as well as the structure of such studies and the procedure for testing the resulting product are analyzed.

The study of publications and their components was the use of deductive and inductive (generalizing) methods, the abstraction method was also used to highlight the most important in publications and facilitate the classification of various elements of the technologies described in them, by abstracting from insignificant differences - identifying vegetable milk substitutes with milk, or various types of vegetable milk substitutes in the analysis of the actual production or testing of yogurt etc. The comparison method was used to compare various processing options for plant raw materials and the resulting fermented milk product from combined milk, described in the analyzed publications and to select individual processing methods aimed at obtaining the maximum amount of proteins and other nutrients from raw materials, increasing protein digestibility, neutralizing anti-nutritional factors and compounds, causing a specific taste and smell for mare's milk etc. Particular attention is paid to avoiding the choice of processing methods, including the use of exposure to ultrahigh temperatures and the overall minimization of heat treatment.

Similarly, work was carried out to analyze and compare various methods for the actual production of a yogurt product and its further verification, an assessment was made of data available in the public domain on the use of microorganisms for the fermentation of combined milk, and an analysis was made of the use of "wild" microorganisms, communities to obtain industrial strains stages of their selection and preparation.

As a result, the components of the developed technology are combined by modeling and elementary-theoretical synthesis, which is a combination of phenomena, in compliance with a strict pattern and cause-and-effect relationship.

Thus, a set of methods was selected that have the most reasonable theoretical justification and the optimal ratio of benefits and potential losses.

Results

Since yogurt is a fermented product, the first step in preparation for its production should be the selection of lactic acid microorganisms. In conditions where the raw material is combined with milk, it is important that the selected microorganisms can effectively ferment both substrates and that the possibility of obtaining undesirable by-products as a result of fermentation is excluded.

A good option is to use starter cultures with a selected strain or complex of strains of microorganisms because they give a reproducible and predictable result (23). However, there are other options - the so-called spontaneous and reverse fermentation. Spontaneous fermentation occurs at the expense of microorganisms found in unpasteurized milk. Reverse fermentation occurs after the transfer of the substrate, in which fermentation is already taking place and its spread to a new substrate (26, 27).

GOST lists only microorganisms that ferment animal milk with the formation of yogurt - these are bacteria of the genus Streptococcus and *Lactobacillus bulgaricus* (23).

In the Republic of Kazakhstan, only certain organizations are certified to store industrial strains, so ready-made starter cultures for making yogurt can only be obtained through them (20, 21). In laboratory conditions, it is possible to obtain fermented milk and probiotic microorganisms by isolating them from spontaneously fermented milk. For this, fresh untreated milk was incubated at 35±5 °C for one to 2 days. Or ready-made yogurt was used (28). Selected 1 mL sample and carried out 3-4 serial dilutions followed by inoculation of the last 2 by the lawn method on a Petri dish with MRS-agar. Selected colonies phenotypically corresponding to lactic acid organisms are isolated and left incubated at the same temperature until biomass is accumulated. To verify the purity of the culture, microscopy was performed, followed by a repetition of inoculation and isolation. The obtained pure strains were further identified to the species using biochemical as well as phylogenetic methods and further screenings of enzymatic and probiotic activity was carried out (26).

Thus, a strain that is potentially used in the industry must meet several characteristics such as matching metabolic properties to the characteristics of the product that is going to be produced, the absence of by-products that threaten health or degrade the quality of the product properties, resistance to existence in conditions that are best contribute to the production, such as a certain low-temperature value, ease of operation and affordability of industrial quantities of the substrate. When it comes to probiotic bacteria, they must remain alive for a long time after the actual fermentation, maturation and packaging of the product and coexist with the host in the gastrointestinal tract, as well as exhibit antibacterial and antioxidant properties (20, 24).

In the manufacture of the starter, a solid medium specific for the cultivation of lactobacilli. Pre-sterilized by autoclaving and refrigerated. Next, inoculation was carried out with a volume of liquid culture, one or a complex of strains, corresponding to the mass of the medium (mL/g). Incubation took place for at least 8-12 h at 37 °C. To extend the shelf life, 20 g of glycerol was added for every 100 mL. the initial culture volume. The resulting starter can be divided into aliquots and stored at –20 °C (13, 29).

It is known that the content of the complex of useful substances in the extract depends on the type and composition of plants, as well as the place of their growth, weather conditions and the time of collection of plants. Therefore, to produce an extract rich in a complex of useful substances, an extract of one plant material is mixed with an extract of another plant material, as a result of which the composition of the new combined extract is enriched with new useful substances C (19).

The extraction of vegetable raw materials was carried out in an experimental setup by the method of low-frequency vacuum-ultrasonic technology. To do this, plant raw materials crushed to a granulated composition of 1.5-2.0 mm were placed in a container poured with a 40% aqueous-alcohol solution and infused for 2.5-3.0 h. Then, the infused raw material at a temperature of 38-40 °C for 15-16 min was subjected to low-frequency ultrasonic treatment in a vacuum at a residual pressure of 10.1 kPa with an oscillation frequency of 32 kHz and an ultrasonic intensity of 70 W/cm^2. After reaching the predetermined time of sonication, turning off the vacuum of the pump, and opening the vacuum of the bypass valve, the studied raw material with the container was taken out. Next, the extract was filtered into a measuring cylinder through a press strainer and a glass funnel. The resulting extract was subjected to organoleptic evaluation.

Further processing methods are aimed at improving the nutritional and sensory properties of the product and preparing it for fermentation with lactic acid bacteria.

In addition to removing compounds that impair the nutritional value or sensory properties of combined milk, an important part of preparing it for consumption is the stage of quality improvement through enrichment with bioactive components and microelements.

An important factor characterizing dairy products is the calcium content. For optimal absorption of this element, it is rational to add calcium carbonate along with vitamin D, a chelating agent - sodium citrate and carrageenone, which acts as a stabilizer. Along with iron, in turn, vitamin C was added.

From biologically active substances, 0.1% glutamate decarboxylase and phenolic compounds (34) and 2% plant sterol powder were added. The last two elements are especially important in the context of the further production of combined milk, since the first increases the digestibility of starch and lactose and the second prevents the absorption of cholesterol (15).

The stage of preparation of cow's milk is one of the shortest. It is recommended to use skimmed milk, which contains up to 1% fat, which meets the requirements of GOST for skimmed milk (22). When using skimmed milk powder, it is dissolved in distilled water with the calculation of 107 g/L. The ratio of components in obtaining combined milk depends on the priority of production: if it is more important to maintain high sensory properties, then the content of soy milk should not significantly exceed 1/4 if the main goal is to minimize the proportion of animal milk, then 1/2 will provide acceptable sensory properties and the proportion of vegetable substitute in the combined milk above 3/4 will lead to a noticeable deterioration in performance. Before introducing the soy product, cow's milk is heated to 90 °C or the components are combined in the desired proportion, thoroughly mixed and pasteurized, without boiling for 10-15 min. Next, the mixture is cooled to 40 °C and becomes ready for inoculation with sourdough (11, 19).

The volume of the starter should be approximately 3% of the total amount of raw materials. It is added to still-warm milk, stirred and left to incubate at 30-45 °C (depending on the bacterial strain used) until an acidic pH of 4.5 is reached. This takes approximately 8-16 h (18). The time also varies depending on the microorganisms used and the content of the soy product, which usually speeds up fermentation (10). It is important that probiotic strains can be used both as a starter and added to an already finished product. To do this, the inoculation was carried out again and the product was left to ferment overnight at 37 °C (25). Further, if necessary, flavors or sweeteners are added and mixed thoroughly and after an hour at room temperature, they are placed in a room with a temperature of 3-5 °C.

An equally important step is to check the resulting product for compliance with microbiological and sanitary standards and the quality of sensory properties.

Microbiological analysis should establish the qualitative and quantitative composition of statistically significant organisms. Statistically significant are fermented milk microorganisms, which should normally be found in yogurts and pathogenic or opportunistic microorganisms as well as those that negatively affect the quality or shelf life of the product. Predominantly, product samples are taken, serially diluted and plated on Petri dishes (medium selection depending on the purpose). The number of colony-forming units (CFU) of lactic acid microorganisms is determined, which in yogurt should be at least 106CFU/mL. to be considered probiotic (11, 18).

To determine the acidity, a pH meter was used and for titrated acidity, titration with a 0.1 N alkali solution was used; the content of lactic acids was determined separately (33). The acid content reflects the efficiency of the fermentation.

Sensory research is entirely based on the subjective feelings of the respondents. The test subjects were randomly selected among people of different ages and genders who did not have intolerance to any of the components of the combined milk. Prepared samples of the obtained combined yogurt and similarly prepared yogurt from pure animal milk in a volume sufficient for repeated sips. Provided participants with water to rinse their mouths between sample tastings. The study took place in 2 stages: in the 1st stage, participants tasted only the combined milk yogurt and rated it on a hedonic scale from 7 to 1, where 7 is very like and 1 is not at all like. And in the 2nd stage, yogurts were evaluated in comparison, without knowing the composition of the samples and a score was determined for each on the same scale (11).

To determine such sensory characteristics as a change in color and density, it is advisable to use instrumental methods, provided that the interpretation of the obtained measurement data is linked to the context of the results obtained on the hedonic scale. The color was analyzed using a chromometer and the interpretation of the result depended on the type of instrument. The density was measured using a rotary viscometer, or a special analyzer, the principle of which is to measure the rate of immersion of a probe of a certain mass. In addition, the rate and moment of the beginning of the recovery of the "equilibrium" initial state of the product surface were analyzed. Thus, using the functionality of the equipment software, numerical expressions for cohesion, consistency and viscosity were obtained (12, 33).

As a result of the work, a technology for the development of a fermented milk product from combined milk was compiled. The general scheme of the process looks like this (Fig. 1). After confirming that the resulting product meets all the criteria and norms, it will be possible to consider the transition to industrial production.

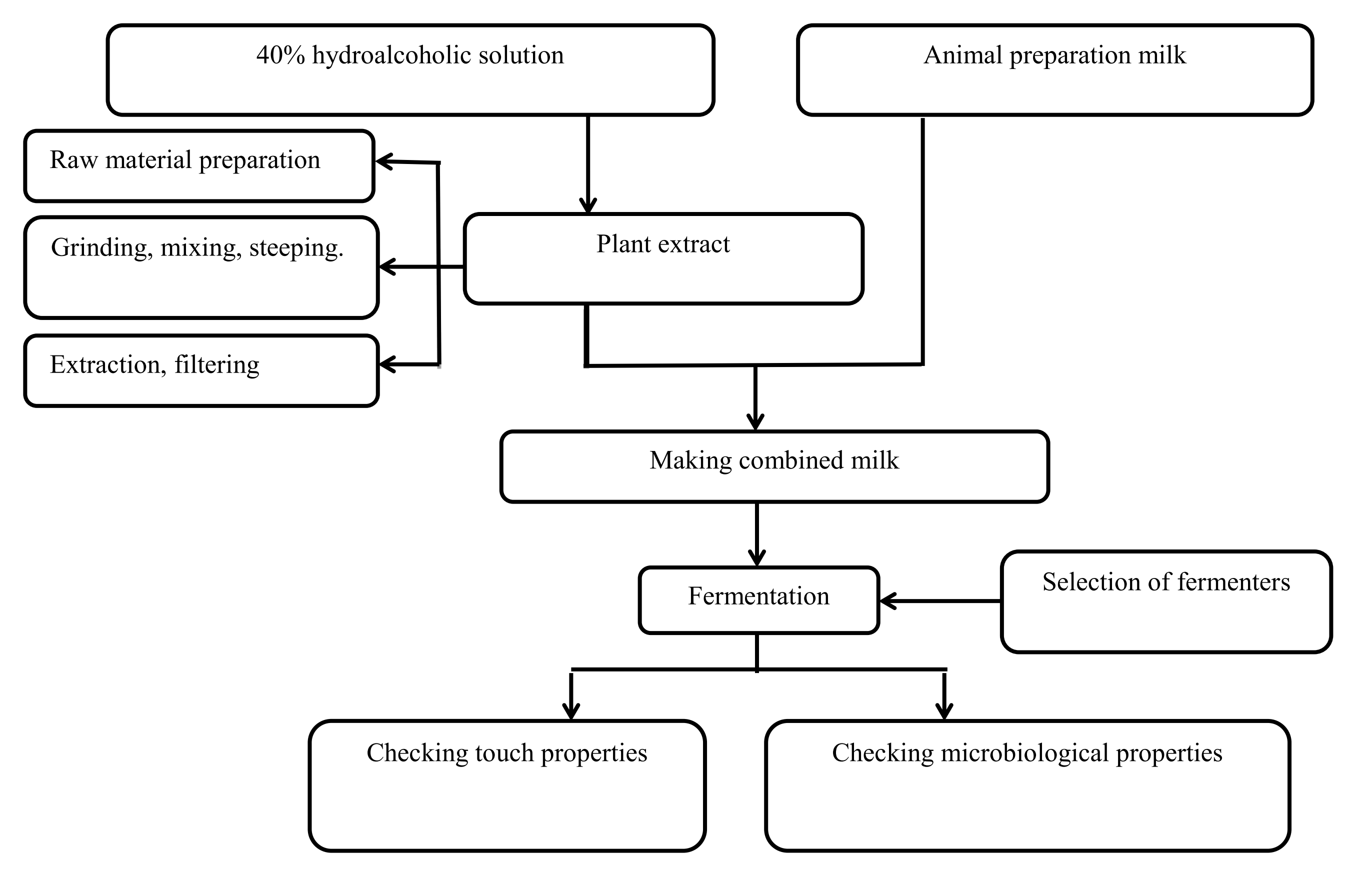

Fig. 1. Scheme of the developed technological process.

Discussion

One of the important features of this product is its uniqueness. The development was carried out using the most modern technologies and advanced scientific research. At the same time, it is based on national traditions, taking into account the needs and preferences of our compatriots. It is important to note that the use of a plant extract in the production process makes it possible to achieve especially valuable properties of the product.

In general, the results of this work, as well as the technology obtained, can be conditionally divided into 2 parts, one of which is devoted to the manufacture of vegetable raw materials and the second to the actual production of a yogurt product. Both are equally important for the manufacture of the combined yogurt product.

To start developing a technology for the manufacture of a fermented milk product, it is necessary to select lactic acid bacteria that can be used for fermentation. The list of microorganisms found in the literature was studied and approved by GOST. In addition, such starter cultures predominantly contain more than one strain of bacteria and there is an opinion that a mixture of fermenting strains has a positive effect on the taste of the final product (33).

Also, in the course of the work, regulatory documents were developed that regulate the introduction of the so-called "wild" strains into industrial use: non-pathogenic bacterial strains can be used for personal or scientific purposes, but the strain that is supposed to be used in industrial production and which is isolated is modified or described by an investigator, must be patented and certified as fit. In this case, the researcher must provide accurate information about the characteristics and properties of bacteria, they must be identified to the species and the strain itself must have an individual name (21). Now in Kazakhstan, there are no patented strains specific for combined milk.

A fermented milk product enriched with plant extract contains many useful substances that have a beneficial effect on the human body. It is a source of vitamins, minerals, amino acids and other elements that contribute to the overall strengthening of the immune system, improve digestion and maintain a healthy intestinal microflora. In addition, the plant extract included in the product has antioxidant properties that can protect the body from free radicals and slow down the aging process.

The invariable advantage of this fermented milk product is its taste. Thanks to the combination of a natural milky taste with a slight hint of herbal extract, it is pleasant to drink and will appeal to people of all ages. It also contains no artificial additives or preservatives, making it a safe choice for anyone who cares about their health.

However, although the plant-dominated combinations have different sensory properties than the more conventional product, they have all the basic attributes of yogurt and have beneficial probiotic properties. In addition, no correlation has been found between protein content and yogurt structure or texture (24). Possibly the low ratings were due in part to the unusual texture and taste, especially in direct comparison. Therefore, it will be rational in the following practical studies to evaluate the perception of such fermented milk products more independently. And their gradual integration into the food culture will not only reduce the consumption of animal milk and diversify the diet, but also make these products more familiar, which will also change the perception of their sensory properties. And, perhaps, after some time, combined milk with a content of a plant product above 50% will receive higher ratings from consumers. This will be facilitated by the improvement of processing technologies for both plant raw materials and the products obtained from them.

Conclusion

As can be seen from the description above, the fermented milk product enriched with plant extract is an original and innovative development that will be presented in the Kazakhstan market. It not only provides people with new choices in nutrition but also helps to strengthen and maintain their health. Let's hope that this product will become popular and find numerous followers who want to take care of themselves and monitor the quality of their nutrition.

In this paper, the technologies for the preparation of vegetable raw materials and the further processing of combined milk in scientific publications are considered. A unique technology has been developed that, at the stage of preparation of vegetable raw materials, almost eliminates the use of heat treatment and the use of ultra-high temperatures, and at the same time is aimed at maximizing the enrichment of combined milk and minimizing the loss of amino acids. Some innovative methods have been introduced to neutralize unwanted anti-nutritional, aromatic and flavor factors. The optimal ratios of cow's milk and mare's milk are 75:25 and 50:50 respectively. It has been determined that fermented milk microorganisms capable of fermenting animal milk also predominantly act in a plant product. Several microorganisms have been selected that can be used for the fermentation of combined milk, such as *Bifido bacterium, Lactobacillus bulgaricum, L. casei, L. acidophilus, Streptococcus thermophilus, L. rhamnosus* and *S. thermophilus*. Also considered is the option of obtaining "wild" strains through spontaneous fermentation of non-pasteurized milk and legislative restrictions governing the introduction of such strains into industrial production. This study can become a full-fledged theoretical basis for further experimental work in this direction.

Authors contributions

KZhT, AUS: designing of the experiments, contribution of experimental materials, execution of field/lab experiments and data collection; KZhT, AUS: analysis of data and interpretation; SSV; preparation of the manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None.

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