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Shingare JD
P.G. Student, Department of Animal Husbandry and Dairy Science, V.N.M.K.V. College of Agriculture, Latur, India

Padghan PV
Associate Professor, Department of Animal Husbandry and Dairy Science, V.N.M.K.V. College of Agriculture, Latur, India

YN Patil
Assistant Professor, Department of Animal Husbandry & Dairy Science, MGM NKCA, Aurangabad

Correspondence
Shingare JD
P.G. Student, Department of Animal Husbandry and Dairy Science, V.N.M.K.V. College of Agriculture, Latur, India

Studies on standardization the process for *dahi* preparation by using the proper proportion of milk and wheatgrass extract

Shingare JD, Padghan PV and YN Patil

Abstract
The study was planned to prepared *dahi* by optimizing the proportion of milk and wheatgrass extract, stage of addition of wheatgrass extract during *dahi* preparation, incubation time and culture rate for setting *dahi*. The incubation period of *dahi* with 1 percent and 2 percent culture rate requires time were 11.20, 10.20, 10.05, 9.50, 10.90, 10.40, 9.90 hrs and 10.00, 9.30, 9.00, 8.65, 9.07, 9.10, 8.90 hrs for treatments T_1, T_2, T_3, T_4, T_5, T_6, and T_7, respectively. The wheatgrass extract was added in buffalo milk at different stages viz. before incubation, during incubation and after incubation. The parameters considered for the optimization stage of *wheat grass* addition were time of setting *dahi*. By comparing time required for incubation of *dahi* with 1 percent and 2 percent culture, addition of 2 percent culture were found less incubation period for each treatment by i.e. 1.20, 0.90, 1.05, 0.85, 1.83, 1.30 and 1.00 hrs for T_1, T_2, T_3, T_4, T_5, T_6, and T_7 respectively. It was observed use of wheat grass extract reduce the incubation period as compared to control *dahi*. Same trend but less effect was observed for one percent culture was used. Therefore, two percent culture rate and nine hours incubation period was found most suitable for the preparation of *dahi* form buffalo milk mixed with wheat grass extract. The stage of wheat grass extract addition was found suitable after 4 hrs of incubation period.

Keywords: *Dahi*, raw/pasteurized wheat grass extract, optimize culture rate

1. Introduction
In modern era consumers are very much aware about their health. So, the demand of functional foods is increasing day by day at a global (Singh *et al.*, 2012) [11]. Moreover, milk is consumed by people of all age can act as potent carrier for the herb which can add functional attribute to the product and consumer well being. Nowadays, more and more people are adopting herbal way of life for their health benefits.

There is also a need to find diverse technologies to add value to milk and its by-products (Pugazhenthi and Jothylingam, 2013) [8].

Dahi has been with associated health benefits since time immemorial. Fermented milk contains all the milk components modified through the process of fermentation by lactic acid bacteria (LAB). *Dahi* is a very good source of calcium, phosphorus, riboflavin vitamin B2 and iodine. *Dahi* also is a good source of vitamin B12, pantothenic acid- vitamin B5, zinc, potassium, protein and molybdenum. These ten nutrients alone would make *dahi* a health supportive food (Deb and Seth 2014) [3]. *Dahi* is prepared by using mix culture of *Streptococcus lactis, S. thermophillus, S. citrophilus, Lactobacillus bulgaricus, and L. plantrum etc.* According to FSSAI Rules (2011), "*Dahi* or Curd" means the product obtained from pasteurized or boiled milk by souring, natural are otherwise by a harmless lactic acids or other bacterial culture. *Dahi* may contain additional cane sugar. It should have the same percentage of fat and SNF as the milk from which it is prepared. Where *dahi*, other than skimmed milk *dahi*, is sold are offered for sale without any indication of the class of milk, the standard prescribed for *dahi* prepared from buffalo milk shall apply (Chowdhury and Bhattacharyya 2014) [1]. Different types of *dahi* are available such as sweet *dahi*, sour *dahi* and flavored *dahi*. Its demand is increasing day by day for its multipurpose health benefits such as improves intestinal health, which preventing constipation, diarrhea and dysentery and also prevent gastrointestinal infections (Munzur *et al*., 2004) [6].

Wheatgrass is a food prepared from the cotyledons of the common wheat plant (*Triticum aestivum*) belonging to family Gramineae. Wheat grass culms are simple, hollow, or pithy, glabrous, and the leaves are approximately 1.2 m tall, flat, narrow, 20-28 cm long and 1.3 cm

broad. The spikes are long, slender, dorsally compressed and somewhat flattened. Its use in acidity, colitis, kidney malformation, atherosclerosis and swelling has been shown to be beneficial (Sareen *et al*., 2014) [10]. Wheatgrass is cost efficient and a source to provide all kind as of nutrients like vitamins, proteins, minerals, antioxidants and medicinal benefits for a healthy and rejuvenating body. Wheatgrass has high concentrations of chlorophyll, minerals (calcium, potassium, sodium, iron, magnesium and sulphur), and 17 forms of amino acids, vitamins (A, B, C, E and K) and active enzymes (Murali *et al*., 2016) [7]. It has some curative properties like anti-cancer activity, anti-ulcer activity, antioxidant activity, anti-arthritic activity, and blood building activity in Thalassemia Major. It has been argued that wheat grass is diseased blood flow, digestion and general detoxification of the body (Runjala and Murthy, 2016) [9]. Hence, by keeping the nutro-functional importance of wheatgrass and *dahi* in mind the project was designed to decide the proportion of milk and wheatgrass extract for setting *dahi* and optimized the stage of addition of wheatgrass extract during *dahi* preparation, incubation time and culture rate for setting *dahi*.

2. Material and Methods

The materials used and methods employed during the course of present investigation on preparation of *dahi* from blends of buffalo milk with (raw/pasteurized) wheat grass extract were as under.

2.1 Collection of buffalo milk

Fresh and standardized buffalo milk for fat 6 percent and SNF 9 percent was procured from Natural Milk Pvt, Ltd, Latur.

2.2 Starter cultures

The standard mixed *dahi* culture i.e. Standard *dahi*, contained *streptococcus thermophilus and lactococcus lactis NCDC-167(BD4)* in this study was procured from the National collection of Dairy culture (NCDC), NDRI, Karnal.

2.3 Methods

Wheatgrass extract was prepared from the wheat grass cultivated in laboratory as shown in flow chart by two ways i.e. without pasteurization and after pasteurization of wheat grass following the steps sorting wheat grass, washing, pasteurization, grinding and filtration. (Kumar *et al.*, 2017) [4].

2.3.1 Preparation of wheat grass (*Triticum aestivum*) extract

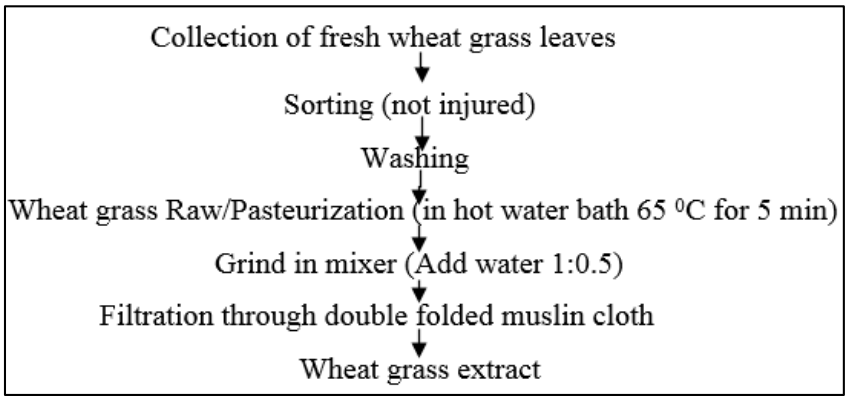

Fig 1: Preparation of wheat grass (*Triticum aestivum*) extract

2.3.2 Optimization of method for *dahi* preparation by using wheatgrass extract

The *dahi* was prepared from buffalo milk as per the method described De, 2009 [2] in his book Outline of Dairy Technology with slight modification for addition of wheat grass extract. The incubation period and culture rate were optimized on the basis of setting *dahi* as shown in Figure 2.

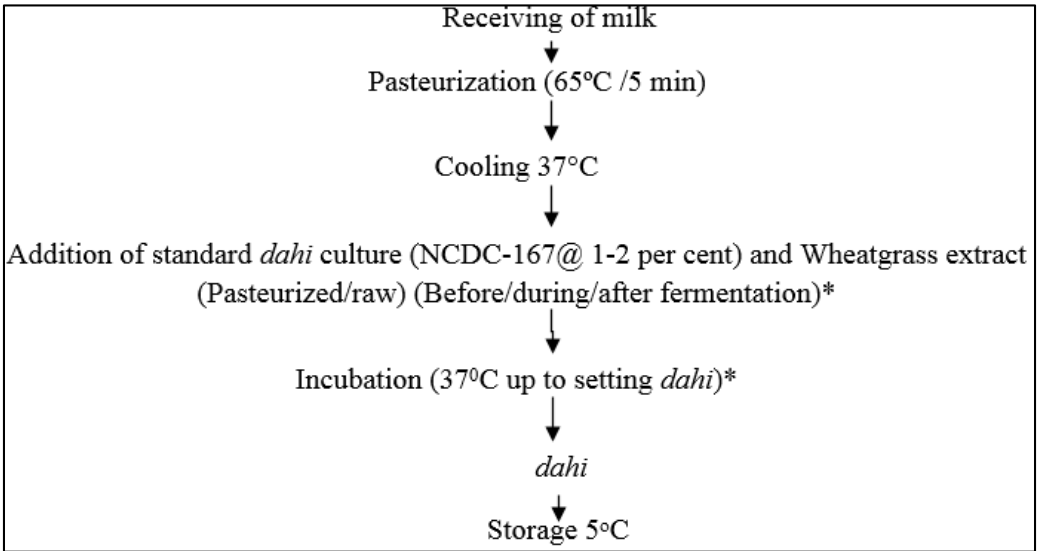

Fig 2: Optimization method for *dahi* preparation by using wheatgrass extract

3. Result and discussion

As the *wheat grass* extract tried in *dahi* preparation in different proportion it was necessary to optimized the culture rate and incubation period on the basis of time required for *dahi* setting, data presented in this respect as follows.

3.3.1 Optimization of culture rate and incubation period (hr.) for setting *dahi*

As the *wheat grass* extract tried in *dahi* preparation in different proportion it was necessary to optimized the culture rate and incubation period on the basis of time required for *dahi* setting, data presented in this respect as follows.

The selected *dahi* culture NCDC-167 procured from National Dairy Research Institute, Karnal (Haryana) was tried for preparation of *dahi* for different treatments and recorded the time required to set *dahi*. The incubation temperature for fermentation was kept 37^0C. For each treatment @1 and 2 percent culture rate was used and results presented in Table No. 1.

Table 1: Optimization of culture rate and incubation period (hr.) for setting *dahi* (in hrs)

Replication/Treatments	R – I		R – II		R – III		R – IV		Mean	
	Time required for incubation at 1 and 2% culture rate									
	1%	2%	1%	2%	1%	2%	1%	2%	1%	2%
T_1	11.30	10.00	11.00	9.60	11.30	9.90	11.50	10.50	11.20^{a}	10.00^{i}
T_2	10.30	9.00	10.00	9.90	10.20	9.30	10.40	9.30	10.20^{b}	9.30^{j}
T_3	10.20	9.10	10.00	9.20	10.10	9.00	9.90	8.90	10.05^{b}	9.05^{jk}
T_4	9.50	8.70	9.00	8.50	10.00	8.90	9.50	8.50	9.50^{c}	8.65^{lk}
T_5	11.30	9.00	11.00	9.40	10.30	8.90	11.00	9.00	10.90^{ad}	9.07^{jk}
T_6	10.00	8.90	10.50	9.00	11.00	9.30	10.30	9.20	10.40^{be}	9.10^{jkl}
T_7	10.00	8.90	10.00	9.00	9.90	8.90	10.00	9.00	9.90^{c}	8.90^{jkl}
For 1% S.E. ± 0.14 C.D. at 5% 0.43										
For 2% S.E. ± 0.12 C.D. at 5% 0.49										

The time required to set *dahi* were observed periodically after 4 hrs. of incubation period at every 30 minutes interval and incubation period was finalized at the time of *dahi* set in hrs. It is observed from Table no.1 that the time required for setting of *dahi* with 1 percent and 2 percent culture rate were ranges from 9.50 to 11.20 hrs and 8.65 to10.00 hrs, respectively. The incubation period of *dahi* with 1 percent and 2 percent culture rate requires time were 11.20, 10.20, 10.05, 9.50, 10.90, 10.40, 9.90 hrs and 10.00, 9.30, 9.00, 8.65, 9.07, 9.10, 8.90 hrs for treatments T_1, T_2, T_3, T_4, T_5, T_6, and T_7, respectively. It seems from the table that as the rate of addition of wheat extract and culture rate increased the incubation period was decreased. For 1 percent culture the minimum time was observed in T_4 (9.50) and maximum in T_1 (11.20), whereas for 2 percent rate the respective observation goes to T_4 (8.65) and maximum in T_1 (10.00). By comparing time required for incubation of *dahi* with 1 percent and 2 percent culture, addition of 2 percent culture were found less incubation period for each treatment by i.e. 1.20, 0.90, 1.05, 0.85, 1.83, 1.30 and 1.00 hrs for T_1, T_2, T_3, T_4, T_5, T_6, and T_7 respectively.

For one and two percent culture rate the wheat grass extract added treatments were statistically different from the control treatment. This might be due to the interaction effect of wheat and milk protein, easy denaturation of milk protein due to the protease enzymes of wheat extract, microbial synthesized enzymes and water holding capacity of wheat gluten (Mujoriya and Bodla, 2011) [5]. *Dahi* samples prepared by using two percent culture rate and wheat grass extract were found at par with each other at 5 percent level of significant. It means that use of wheat grass extract reduce the incubation period as compared to control *dahi*. Same trend but less effect was observed for one percent culture was used. Therefore, two percent culture rate and nine hours incubation period was found most suitable for the preparation of *dahi* form buffalo milk mixed with wheat grass extract. Hence the rate for culturing was optimized as two percent and means value for its incubation period was considered as 9.00 hrs and was followed during experiment for preparation of *dahi*.

3.3.2 Optimization the addition stage of *wheat grass* extract for setting *dahi*

The *dahi* was prepared as per method of De (2009) [2] with slight modification by using culture @ 2 percent and incubated for 9 hrs at 37^0C as optimized in section 3.3.1. The wheatgrass extract was added in buffalo milk at different stages viz. before incubation, during incubation and after incubation as per the treatment shown below. The parameters considered for the optimization stage of *wheat grass* addition were time of setting *dahi*, whey drainage and sensory score for flavor obtained for *dahi* and result of it is presented in table no. 2.

From the table no. 2 it is observed that all treatments having flavor score more than 7.00 ranges between like very much to like moderately. But most of the extract added treatments secured score less than control treatments except treatment T_2 (5 percent raw wheatgrass extract added) for wheatgrass extract added during and after incubation and T_5 (5 percent pasteurized wheatgrass extract added) for wheatgrass extract added during and after incubation were 8.20, 8.10 and 8.40. It is revived from table no. 2 that *dahi* prepared by added extract during fermentation secured more score as compared to the stage of extract addition before and after fermentation. Then it is also observed that the flavor score of pasteurized extract more than the raw extract.

Table 2: Optimization the stage of addition of *wheat grass* extract for setting *dahi*

Treatment	Before Incubation			During Incubation After 4 hrs			After Incubation		
	Time	Whey drainage (%)	Flavor Score	Time	Whey drainage (%)	Flavor Score	Time	Whey drainage (%)	Flavor score
T_1	11.00	0.42	8.00	11.00	0.42	8.00	11.00	0.42	8.00
T_2	10.20	0.40	7.40	9.15	0.26	8.20	11.25	0.45	8.10
T_3	10.00	0.43	7.20	10.30	0.30	7.80	10.45	0.48	7.80
T_4	9.40	0.45	7.00	10.00	0.35	7.50	11.00	0.50	7.40
T_5	10.00	0.39	7.50	9.30	0.23	8.40	11.20	0.44	7.50
T_6	9.50	0.43	7.30	10.00	0.31	8.00	11.00	0.47	8.00
T_7	9.20	0.44	7.10	10.00	0.36	7.60	11.20	0.51	7.50

It was found creditable about whey drainage, the maximum whey drainage was observed in *dahi* in which extract was added before and after incubation, whereas minimum whey drainage observed during incubation in which whey was added after 4 hrs of incubation period it might be due to the over fermentation and incomplete fermentation occurred respectively during these two condition. The addition of extract during fermentation also supportive for minimum whey drainage. The ideal condition for addition of wheat grass extract for *dahi* preparation by using buffalo milk and wheat grass extract was found during fermentation at the mid time of fermentation in which more flavor score, less whey drainage and optimum incubation period (9hr) were observed. The stage of addition of *wheat grass* extract optimized in present study agreed and supported by the (Thammana *et al.* 2016) [12]. In their review paper, it or mentioned that to get the beneficial effect of wheatgrass extract it should be consumed within 15 minutes of preparation, oxidized rapidly soon after squeezing and supplement only 2 percent benefits in dried powder form as compared to fresh one.

The process of *dahi* preparation after optimization of culture rate, incubation period and stage for wheat grass extract addition was finalized as shown below in figure no. 3.

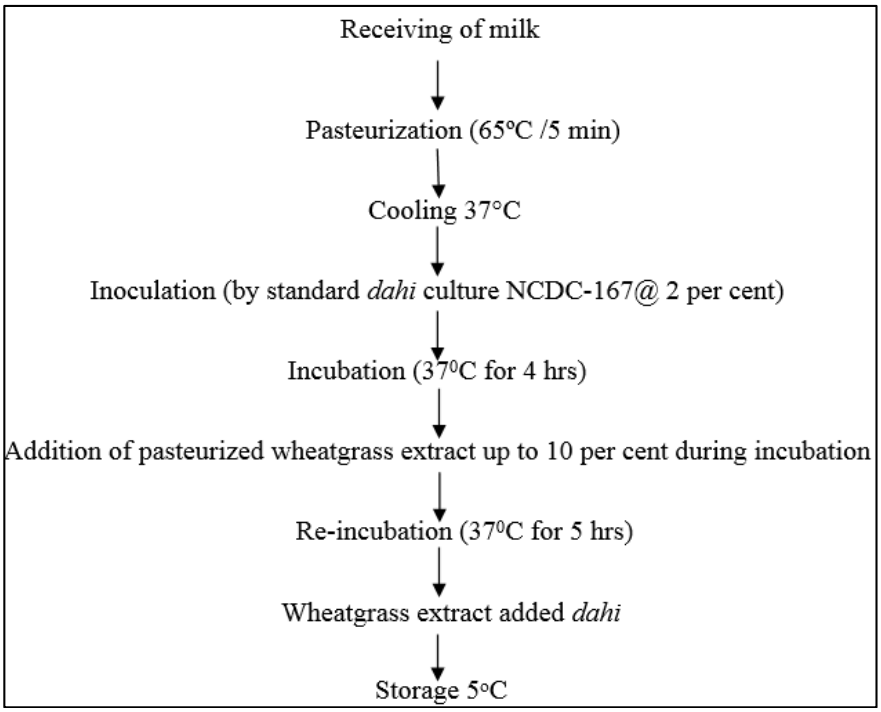

Fig 3: Preparation of wheat grass (*Triticum aestivum*) extract added *dahi*

4. Conclusion

Wheatgrass extract added *dahi* could prepared by adding extract during fermentation more beneficial as compared to other types. It would like to mention here that the addition stage of wheat grass extract in *dahi* preparation need to modify again to reserve the functional benefit of wheatgrass. Wheat grass extract added before and after fermentation increased whey drainage and alter its textural as well as physico-chemical properties. For the present investigation the wheat grass extract added during fermentation after 4 hrs old incubated samples for getting the optimum physical structure and sensory test of *dahi*.

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