INTRODUCTION

Quark is an acid dairy product originating from Germany, resembles with yoghurt of thick consistency. It was introduced (in Holland) as a fat free dairy product and is exactly a very fresh kind of cheese. Quark is made by adding rennet or another souring agent (like Buttermilk) to milk. The milk then separates into whey and curds. The whey is poured off or drained. The remaining curds are stirred until a smooth consistency is reached: this is called quark. Dictionaries usually translate it as curd cheese. It is soft, white and un-aged, similar to Fromage frais, but with a higher fat content. Quark is cultured dairy products obtained by fermenting milk to pH of 4.3 to 4.8 with mixed mild cheese cultures, usually comprising Streptococcus cremoris and Streptococcus lactis and sometimes Streptococcus diacetylactis. This is followed by removal of whey and concentration of solids to obtain a stiff, white to off white, spreadable paste that should be homogenous, short textured and free from serum of graininess. The product should have clean lactic flavor and be mildly acidic. Rennet is usually added during fermentation to assist in curd formation and improve the separation of whey.

Quark is a member of the acid set cheese group, meaning it is traditionally made without the aid of rennet. Because quark is consumed without aging, in the United States the milk must first be pasteurized. Once the milk is ready, lactic acid bacteria are added in the form of mesophilic lactococcus starter culture. Continuing acidification until the pH reaches 4.6, which causes precipitation of the casein proteins. In Germany, the curd is continuously stirred to prevent it from getting hard, resulting in a thick, creamy texture.

To make the firmer eastern European version, a small amount of rennet may be added to make the curd firmer. Some or most of the whey is removed to standardize the quark to the desired thickness.

Amount of lactic acid production by L. bulgaricus is 1.7-1.8% while S. thermophilus is 0.6-0.8%. Acid contributes to the formation of gel by conversion of colloidal calcium-phosphate complex present in the casein micelle to soluble calcium phosphate fraction. At pH 4.6-4.7 this leads to loss of calcium from micelles and coagulation of casein.

Rennet consists of mainly rennin and pepsin, principally the former being responsible for milk clotting and the latter for proteolysis (although both possess the two characteristics). Rennin is liable to heat and alkali besides other physical and chemical agents and gets inactivated at pH 6.8 - 7.0 at 70°C in 14 minutes.

MATERIALS AND METHODS

Raw materials and sources
About 15 liters of fresh whole cow milk was taken for each lot of quark preparation. The temperature and acidity of the milk at the time of collection was 16°C and 0.14 % respectively. The milk was collected within the two hours of milking and the pH of the milk was 6.4

**Rennet powder**

Information from the label:

STAMINS 1150, NATUREN™
CHR. HANSEN, DENMARK

Instruction: to use approx. 3g/100lt milk
Store at -15°C

**Preparation of rennet solution**

Rennet 0.1 gm was mixed in 50 ml distilled water at normal temperature (25°C) and was agitated until completely solubilized. It was cooled in the freezer at 5°C. One ml of rennet solution was pipetted out for each lot (500 ml) of milk.

**Methodology**

Raw milk kept in refrigerator (at 4°C) for 2 hours was then pasteurized at 68°C for 30 minutes and allowed to cool at 20°C. The yoghurt made by Dairy Development corporation, was taken as starter culture.

The basic steps for Quark making procedure was employed from the procedure provided by ptc+ 2007 (Netherland). The details of the procedure are given below.

1. **Fresh milk (cow)**
2. Pasteurized the milk at 68°C for 30 min or 72°C for 15 sec.
3. Cool down to 20°C
4. Inoculate with 3% starters
5. Add 1 ml of rennet solution
6. Incubate 24 hrs at 20°C
7. Cut the coagulum into large sized cubes
8. Drain the whey
9. Collect the drain whey to calculate the Quark yield 75% whey + curd
10. Adding of sugar
11. Homogenizing
12. Packaging
13. Ready for consumption

**Fig 3.1 Process employed for making Quark.**

**Recipe formulations**

1. First optimization of rennet
   A = 500 ml milk + 1 ml of 0.234 % rennet + 2 % starter culture
   B = 500 ml milk + 2 ml of 0.234 % rennet + 2 % starter culture,
   C = 500 ml milk + 1 ml of 0.139 % rennet + 2 % starter culture,
   D = 500 ml milk + 1 ml of 0.139 % rennet + 2 % starter culture of rennet were used.

2. The products made from different concentrations of rennet were taken for sensory evaluation and the product having highest score was then taken for optimization of sugar. For the optimization of sugar, A₁ = 2 %, A₂ = 4 %, A₃ = 5 % were used. Again these 3 formulations were sensorily analysed

3. The best product was again taken for flavour optimization. The different flavors used were; vanilla, pineapple and orange and their concentrations were 125 ppm each.

Sensory evaluation was carried out using numeric scoring method described by American Dairy Science Association.

**Analytical methods**

The total solid content of the quark was determined by hot air oven method according to AOAC. Similarly, moisture content was determined by difference method according to AOAC. The fat content of the quark was determined by Gerber’s method for fat determination. The protein content was determined by formal titration by following the method described by K.C. and Rai. The milk pH was determined directly with a digital pH meter of CCT, Hattisar, Dharan. Acidity of the milk was determined according to Nepal Bureau of Standard. The ash content and lactose content of the quark was determined by Lane and Enyon method.

Total coliform of the quark was determined by pour plate technique on Violet Red Bile Agar (VRBA) medium. Total plate count (TPC) was determined by pour plate technique on plate count agar (PCA) medium. The total yeast and mould count was
determined by pour plate technique on potato dextrose agar (PDA) medium.

Statistical data analysis of The samples were analyzed by Genstat programming (Genstat version 5) at 5 % level of significance. The mean values were compared using LSD and the best treatment was selected.

RESULTS AND DISCUSSION

Quark was produced from cow milk having 4.3% of protein and 4% fat. The milk quality was in accordance with the actual regulation of the milk and dairy products. The quark was made of good quality. The final product was analysed and it was found the coliform count was nil, total plate count was 28 colonies at 10⁻³ dilution and yeast and mould count was 41 colonies for 10⁻³ dilution. This result is similar to the Milanovic et al

<table>
<thead>
<tr>
<th>Attribute Formulation</th>
<th>Flavor</th>
<th>Body texture</th>
<th>Acidity</th>
<th>Appearance</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>33^a</td>
<td>20.8^a</td>
<td>8.00^a</td>
<td>10.00^b (1.41)</td>
</tr>
<tr>
<td></td>
<td>(3.46)</td>
<td>(4.21)</td>
<td>(0)</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>28.4^a</td>
<td>16.40^b</td>
<td>5.20^b</td>
<td>7.20^a (2.28)</td>
</tr>
<tr>
<td></td>
<td>(5.02)</td>
<td>(4.39)</td>
<td>(044)</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>27.4^a</td>
<td>15.00^ab</td>
<td>6.00^c</td>
<td>6.20^d (1.30)</td>
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<td></td>
<td>(3.71)</td>
<td>(3.53)</td>
<td>(0.70)</td>
<td></td>
</tr>
<tr>
<td>D</td>
<td>25.4^a</td>
<td>11.60^a</td>
<td>7.40^d</td>
<td>7.60^a (1.81)</td>
</tr>
<tr>
<td></td>
<td>(2.88)</td>
<td>(5.89)</td>
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</table>

Optimization of rennet concentration for the preparation of quark

Table 2. Represents the mean sensory score and chemical score of quark made by using different rennet concentration for the optimization of rennet. The different rennet concentration such as 1 ml of 0.234 % rennet (A), 2 ml of 0.234% rennet (B), 1 ml of 0.139% rennet (C) and 2 ml of 0.139% rennet (D) was used.

Table 2 Mean sensory and chemical score of quark produced by different rennet formulations.

Values are the mean of three replicates and values in parentheses are S.D. values in the column having same superscript did not vary significantly (P > 0.01) with LSD.

The flavor of product A prepared by using 1 ml of 0.234 % rennet had significantly (P < 0.01) higher score than the others. The recommended rennet concentration for cheese given on the label was 0.003 %. Among them 0.00234 % (1 ml of 0.234 %) gave the best results in terms of acidity, flavor and texture and it was selected for further improvement by manipulating the sugar content.

Optimization of sugar concentration

Table 3 represents the sensory sugar formulation for optimization of sugar content. The different sugar concentration such as 2 % of sugar (A1), 4 % of sugar (A2) and 5 % sugar (A3) was used for optimization of sugar level in quark.

Table 3. Analysis of difference in attributes of the formulations (sugar)

Values are the mean three replicates and values in the parentheses are S. D. values in the column having same superscript did not vary significantly (p>0.01).

The flavor of product A2 prepared by using 4 % sugar concentration has significantly (p < 0.01) higher score than the others. This result was similar to the result obtained by Atal,
2006. She made soya yoghurt which was found to be the best in terms of taste when 4% sugar was used. The total solid content after adding 4% sugar was 17% which was in the range (16-18) used by Miloavic, et al to prepare probiotic quark. In our context also the dairy industries of Nepal (Dairy Development Corporation) use 4% sugar for the preparation of yoghurt. So the product A2 was selected for the further improvement by manipulating the color & flavor.

**Optimization of flavor**

Table 4 represents the mean sensory and chemical score of quark made by using different formulation for the optimization of flavor the different formulation such as orange food color + no flavor (A2a), no food color + pineapple flavor (A2b), no food color + vanilla flavor. (A2c)

**Table 4: Mean sensory and chemical score of Quark produced by different flavor.**

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Formulation</th>
<th>Flavor</th>
<th>Body texture</th>
<th>Acidity</th>
<th>Appearance</th>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A2a</td>
<td></td>
<td>29.67a</td>
<td>20.17a</td>
<td>6.17a</td>
<td>8.00a (2.68)</td>
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<td></td>
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<td>35.83ab</td>
<td>23.33a</td>
<td>6.67a</td>
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<tr>
<td></td>
<td></td>
<td>(2.76)</td>
<td>(2.065)</td>
<td>(1.03)</td>
<td>(2.13)</td>
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<tr>
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<td></td>
<td>37.50bc</td>
<td>22.33a</td>
<td>8.00b</td>
<td>11.17a</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(4.18)</td>
<td>(3.50)</td>
<td>(1.41)</td>
<td>(2.13)</td>
</tr>
</tbody>
</table>

Values are the mean of three replicates and values in parentheses are S. D. values in the column having superscript did not vary significantly (P> 0.01) with LSD.

The flavor of product A2a prepared by incorporating orange food color (but without essence) did not significantly differ from others at 5% level of significance. The data shows that there is a considerable agreement between panelists. The panelists were familiar with the yogurt. The product A2c having vanilla flavor was well perceived by the sensory panelist. So the product A2c was selected for the final product development.

**Storage life of the product**

Figure 1 shows that the pH of quark stored at room temperature (33°C±2) decreased significantly faster rate. The corresponding acidity of the lowest pH (3.2) is 0.45% (as lactic acid). The product in this stage was already in an unacceptable state with respect to taste, smell, texture and appearance. This was natural because the activity of lactic acid bacteria and other spoilage bacteria was higher at room temperature compared to the lower storage temperatures counterpart. There was no change in the pH and acidity in the control (deep freeze temperature, -18°C). For a storage period of a sample up to 17 days (samples were not analyzed after 17 days), the acidity and pH showed a slight change. Quark stored at refrigerated temperature (Temp. 4°C) behaved erratically for the first week and then stabilized at a pH of about 3.4 (corresponding acidity = 1.78) up to the 15th day of storage. The product was still acceptable up to the 15th day. This implies that, quark can be typically stored at refrigerated condition for about two weeks without much loss in sensory quality. The shelf life obtained in this study was slightly lower than the result obtained by Miloavic, et al. The shelf life of probiotic quark made by Miloavic, et al was 30 days kept at lowerthan4°C

**Storage life of the product**

Figure 2 Changes in smell of quark with time under different storage conditions

The sample which is stored at room temperature (33°C±2) had slightly acceptable smell in the first day but in the second day it was totally unacceptable (score ≤ 5 has been arbitrarily ranked unacceptable in the present experiment). Refrigerated sample (~4°C) was same as control (acceptable) up to six day. It indicates that it has high quality life up to six day.
The quark sample which was stored at room temperature (33°C±2) had slightly acceptable taste in the first day but in the second day it was totally unacceptable. Refrigerated (4°C) sample was same as control (acceptable) up to six days (Fig. 4), thereafter the quality decreased slightly and remained stable for 15 days at a score of 7. It indicates that quark can be stored at refrigerated temperature for 15 days without much loss in taste. However, one also has to take into account the smell of the product (figure 3), in which case the overall acceptability will be about 6 days.

The sample which was stored at room temperature (33°C±2) had slightly acceptable texture in the first day but in the second day it was totally unacceptable. Refrigerated (4°C) sample was same as control (acceptable) up to six days, thereafter the quality decreased slightly and remained stable for 10 days at a score of 6-7. It indicates that quark can be stored at refrigerated temperature for 10 days without much loss in texture.

The sample which was stored at room temperature (33°C±2) had slightly acceptable appearance in the first day but in the second day it was totally unacceptable. Refrigerated (4°C) sample was same as control (acceptable) up to six day. Thereafter the quality decreased sharply and became no more acceptable. It indicates that quark cannot be stored at refrigerated temperature for more than a week without adversely affecting the appearance. Afterall, the refrigerated storage can hold quark for up to 6 days in perfectly acceptable condition.

**CONCLUSION**

Quark was made by using different formulations and they were optimized for different parameters like rennet concentration, sugar concentration and flavor. Use of 1ml of 0.23% rennet was found to be better than other concentration of rennet for flavor, texture and acidity. The 4% of sugar concentration was found to be the best in terms of taste, texture and acidity for the development of quark. The vanilla flavor was found to be the best among the other flavors used. The shelf-life of the best product made was studied under room temperature (33±2°C), refrigerated condition (-4°C) and deep freezed condition (-18°C). The product was acceptable up to 2 days kept in room temperature whereas in refrigerated condition the high quality life of the product was found to be 6 days and normal product life was found to be 15 days. In deep-freezed condition, the product remained unchanged up to 45 days.

**ACKNOWLEDGEMENT**

I would like to express my sincere thanks to the faculty of food technology, central campus of technology, Dharan and Sunsari technical college Dharan for their support and encouragements during the study.

**REFERENCES**
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Cite this article as: Yadav RS. Optimization of processing parameters and extension of shelf life of ‘quark’-a type of thick yoghurt. STCJ 2015,2(1): 38-43.