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ABSTRACT

STABILITY AND SENSORY PROPERTIES
OF YOGURT DRINK

By

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The effect of variations in processing parameters and in the concentration of stabilizers on the stability and sensory properties of yogurt drink was investigated. Yogurt drink was composed of 25% plain low fat yogurt, 10% frozen concentrated orange juice, 9% sugar and 56% water.

Stability studies were carried out by observing sediment or syneresis volume of the samples during storage at $5^{o}C$ and by determining protein content on the supernatant fluid of the samples after storage. Subjective sensory analysis was conducted using a nine-point hedonic scaling method or ranking method.

Of four pH values (3.2, 3.6, 4.0 and 4.2), pH 4.0 was the best for product stability and flavor. Heat treatment variation ($65^{o}C$/30 min, $75^{o}C$/10 min, $88^{o}C$/2 min and $115^{o}C$/4 sec) and homogenization pressure variation (70 kg/cm^2, 105 kg/cm^2, 140 kg/cm^2 and 175 kg/cm^2) had little effect on the sensory properties of the product. High temperature pasteurization and low pressure homogenization favored stabilization of the product.

Among the stabilizers studied, carrageenan, propylene glycol alginate and pectin were not effective in stabilizing the product. Guar gum and a commercial proprietary stabilizer were very effective, stabilizing the product at the concentrations of 0.20% and 0.30% respectively. The use of guar gum in combination with alginate, pectin or carboxymethyl cellulose did not show the synergistic effects of the combined stabilizers.

The stabilized samples showed excellent acceptability in taste panel evaluation.

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INTRODUCTION

The tremendous increase in yogurt consumption in the United States and all over the world has led to the development of other yogurt based dairy products as an opportunity for product expansion. Yogurt drink has been introduced as a product with a refreshing taste of liquid drink combined with the natural ingredients of yogurt.

Yogurt drink is somewhat unstable because of its high solids content (usually higher than 15.0%) in liquid medium and the physical instability of the milk proteins at low pH. Therefore, the problem is to produce a finished product that is stable and has the necessary sensory characteristics to make it acceptable to the consumer.

A few general trade journal articles have described yogurt drink and suggested composition and manufacturing procedures. However, little research appears to be available on the effects of processing parameters and the functions of stabilizers on the stability and sensory properties of the product. In the present investigation, a step by step approach to the stabilization of the product is employed using four variations in pH, heat treatment and homogenization pressure respectively. Six stabilizers are investigated individually and in combinations at various concentrations. Subjective

sensory analysis is conducted in each step prior to the stability studies which include sediment or syneresis observation and protein determination on the supernatant fluid of the samples.

The present study was undertaken to establish optimum processing parameters and the concentration of suitable stabilizer(s) on the stability and sensory properties of yogurt drink. This approach to the problem offers the possibility of developing information applicable to the control of processing parameters and to the use of stabilizers to maximize the stability in yogurt based drinks.

LITERATURE REVIEW

Historical Aspects of Yogurt Products

Yogurt is a cultured milk product that is made by inoculating warm pasteurized and/or homogenized milk with a combined culture of S. thermophilus and L. bulgaricus (International Dairy Federation, 1969). The word yogurt (also spelled yoghurt, yourt or yoghourt) is of Turkish origin. The yogurt or yogurt-like products dates back to Biblical days--or to when man first began drinking the milk of cows, goats, sheep and camels, although there is no precise record when they were first made.

In the Bible, it is recorded that Abraham served sour and sweet milk along with a calf while entertaining three angels (Genesis 18:8). It is believed that every civilization on earth has known and consumed fermented milk of one type or another. Many of these foods can still be found throughout the world, such as buttermilk in Europe, acidophilus milk in the Soviet Union, yogurt in Turkey, dahi in India, leben in Egypt, yiaourti in Greece, cieddu in Italy, kefir in Balkan countries, and kumiss in Mongolia (Kosikowski, 1977; Rasic and Kurmann, 1978).

Originally yogurt was made from the milk of sheep, goats, and cows. The propagation was carried out by using a small quantity of the previously fermented milk to seed the next container of milk. Modern interest in yogurt owes

much to the work of Metchnikoff, a Russian born bacteriologist who worked at the Pasteur Institute in Paris (Tramer, 1975; Davis, 1974; Rasic and Kurmann, 1978). The characteristic organism of the traditional soured milk of the Balkans was described and named as Lactobacillus bulgaricus. The good health and longevity of the Balkan peasants were attributed to the effect of this organism in the intestinal tract through repressing the undersirable putrefactive bacteria. Although this theory exaggerated the value of yogurt, it significantly influenced the spread of the product to many countries of Europe and promoted extensive studies by subsequent workers.

Since World War II, an increased knowledge of the science of microbiology, better understanding of the factors affecting the sensory properties of yogurt and the application of modern equipment have resulted in the rapid advancement of yogurt technology. The introduction of fruit and the application of a wide range of flavors promoted further growth in the consumption of yogurt products.

The United States has commercially produced yogurt only since 1940. The first fruit flavored yogurt was introduced in the United States in the early 1960's and consumption has increased at a steady pace since then. According to the annual report of the Milk Industry Foundation (1979), yogurt consumption has increased from 0.11 lbs per capita in 1955 to 2.68 lbs per capita in 1978, an increase of 2,500%.

Yogurt Technology - Plain Yogurt

Yogurt is similar to other cultured dairy products in that high quality ingredients and strict attention to sanitary procedures are required for satisfactory manufacture of an acceptable product. There are certain basic principles to be followed regardless of the type of yogurt made. Standardization of milk, pasteurization temperature and time, homogenization, time and temperature of incubation and handling of the product after manufacture are important steps in the process.

Standardization and Treatment of Yogurt Mix

Standardization - There are no concrete regulatory standards of composition as to fat and SNF contents of yogurt (Duitschaever et al., 1972; Kroger and Weaver, 1973; Robinson and Tamime, 1976; Richmond et al., 1979), as shown in Table 1. It has been common practice for the dairies to experiment with fat and solids not fat (SNF) content of yogurt by the application of available knowledge and expertise, and to modify its composition as desired. Standardization, not only insures consistency of the end product but also enables control of the flavor and aroma, viscosity, stability and nutritive value of the finished product.

Kroger (1973, 1976) and Davis (1974) stated that milk fat in yogurt mix definitely contributes to the smoothness of mouthfeel and richness of the product if the mix is

Table 1. Suggested standards for the chemical composition of yogurt in terms of milk fat and SNF.

Country of origin	Types of yogurt based on % fat			% SNF
	Normal	Medium	Low	
Denmark	3.8	1.8-1.5	0.3	-----
France	3.0	-----	1.0	-----
Netherlands	3.2	-----	0.3	-----
United Kingdom	3.5	2.0-1.0	0.3	8.5
United States	3.8-3.0	2.8-2.0	1.0-0.5	8.25-8.3
FAO/WHO	3.0	3.0-0.5	0.5	8.2

Source: Robinson and Tamime, 1976.

homogenized. Nielsen (1975) stressed the importance of fat in relation to both body and texture of yogurt. It was indicated that even 1.0-2.0% fat in yogurt has a beneficial influence on body and texture of the product. Rasic and Kurmann (1978) observed that skimmilk yogurt is more acid, less mild and less aromatic than yogurt which contains milk fat. It was further stated that the fat content of 1.5% can counteract the above defects.

It is usual in practice to increase SNF content in milk for yogurt by concentration of the milk or by addition of nonfat dry milk (NFDM). Powell (1970) and Davis (1971) considered that NFDM added at the rate of 1 to 2% gives optimum solids content for yogurt mix, yielding a firmer gel and no syneresis in the finished product, and contributing to greater nutritional value. According to Cottenie (1978), greater improvements in quality can be obtained if the increase in solid concentration is brought about by partial concentration under vacuum, than if NFDM is added. A change in the calcium-phosphorus-caseinate complex induced by the removal of water is beneficial for building up an optimum micelle structure of proteins which reduces and eliminates the tendency for syneresis. Excessive addition of SNF gives rise to defects such as saltiness, powder flavor and coarseness in the end product.

Pasteurization - Heat treatment of yogurt mix is carried out with the purpose to kill pathogenic organisms

which may be present, to produce an improved medium for the growth of lactic acid bacteria, and to improve consistency and firmness of the product (Powell, 1970; Sellars, 1973; Nielsen, 1974, 1975; Davis, 1975; Cottenie, 1978; Rasic and Kurmann, 1978).

The improved medium for the growth of the lactic acid bacteria is attributed to a reduction in bacteriostatic substances and the formation of substance which act as stimuli to the growth of the culture. Davis (1975) suggested that heating lowers the oxygen tension and makes a more favorable medium for the growth of L. bulgaricus. Heat treatment may also liberate amino acids from the milk proteins which may stimulate the growth of L. bulgaricus. This organism in turn elaborates amino acids which stimulate growth of S. thermophilus (Pette and Lolkema, 1950 a and b; Nielsen, 1975).

Nielsen (1975) suggested that the improved consistency and firmness of yogurt made from heated milk is associated with the denaturation of whey proteins, the change of caseinate structure and the association of these two phenomena which increase the water-binding capacity of the system. Cottenie (1978) reported that accumulative heat treatment of yogurt mix at 190^{o}F for 2 min followed by an additional 192^{o}F for 3 min results in good consistency and firmness of yogurt. Galesloot and Hassing (1968) recommended heating of yogurt mix to 185^{o}F for 30 min for firm consistency. Nielsen

(1974) suggested that the best texture can be obtained with a heat treatment of 195°F for 5-30 min. Rasic and Kurmann (1978) explained that the differences in the data reported by many investigators may be due to the probable effects of other factors on the hydrophilic properties of porteins in addition to heat treatment of yogurt mix, e.g. the total solids content and particularly the protein content of milk, the solid content adjustment of milk, the properties of yogurt cultures, homogenization, etc. Nielsen (1975) claimed that a yogurt mix with high SNF content should be heated at 195°F for not less than 10 min and not more than 50 min.

Generally, yogurt mix is pasteurized at temperatures of 190°F, holding times being between 5 and 45 min. (Humphreys and Plunkett, 1969).

Homogenization - Pette and Lolkema (1951 b) maintained that homogenization is essential for the production of firm curd yogurt. Cottenie (1978) noted that homogenization, if carried out efficiently, is quite necessary because it prevents fat separation, increases the firmness of the gel by changing the physical properties of fat globules and casein particles, improves the taste through the formation of a soft, finely homogeneous curd and shortens the curdling time.

Pette and Lolkema (1951 b) recommended that pasteurization be carried out before homogenization to prevent

lipolysis caused by lipolytic enzymes. However, Rasic and Kurmann (1978) stated that this procedure has a limitation because of the risk of recontamination. Rasic and Kurmann (1978) recommended high pressure homogenization, claiming that high pressure prevents rising of fat by breaking up the fat globules to sufficient small size and increases viscosity of yogurt by denaturing more serum proteins. On the other hand, Humphreys and Plunkett (1969) maintained that homogenization at a low pressure significantly increases firmness and viscosity and reduces the rate of whey separation on storage. These investigators, quoting from personal communications from Barker (1965), also mentioned that good quality yogurt can be produced without homogenization. Stocklin (1969) reported that homogenization at 110^{o}F at a pressure of 1400 or 1500 psi produced a good yogurt.

Rasic and Kurmann (1978) presented the effect of temperature on homogenization of yogurt mix as shown in Table 2. The temperatures shown differ depending on the purpose of the homogenization.

Microbiological Aspects of Yogurt

There is considerable confusion in terminology in the literature as to the microorganisms that are used for the manufacture of yogurt. However, most workers agree that yogurt is basically cultured milk fermented by L. bulgaricus growing in association with S. thermophilus (Pette and Lolkema, 1950, 1951; Stocklin, 1969; Humphreys

Table 2. Effect of temperature on homogenization of yogurt mix.

Temperature	Effect on milk constituents
100°F	Minimum temperature that can be used.
122-140°F	Optimum condition.
140-158°F	Damaging the ability for fat rising.
Above 158°F	Serum protein begins to be denatured.
Above 212°F	Caseins begins to coagulate when temperature combined with a sufficient pressure.

Source: Rasic and Kurmann, 1978.

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and Plunkett, 1969; Powell, 1970; Angevine, 1972; Tramer, 1973; Sellars, 1973; Robinson and Tamime, 1975; Kosikowski, 1977; Cottenie, 1978; Lundstedt, 1978; Sharpe, 1979).

Yogurt cultures are best obtained from established culture manufacturers, from research institutes or other reputable sources and should be renewed at frequent intervals.

Handling of yogurt cultures requires skill and appreciation of several facts, namely, that they have different optimal growth temperatures, that their rate of growth is different and that in spite of all these variations it is essential to maintain a balance of the two organisms.

It is well established that L. bulgaricus and S. thermophilus stimulate each other during their associative growth.

Pette and Lolkema (1950 a and b) proved the stimulation of S. thermophilus by L. bulgaricus. The study suggested that L. bulgaricus liberates in milk certain amino acids which stimulate S. thermophilus. In their futher investigations, it was shown that valine was the most important amino acid among those which stimulate the growth of S. thermophilus.

Bautista et al. (1966) confirmed the stimulatory effect, but found that the active factors were glycine and histidine.

Galesloot et al. (1968) showed that S. thermophilus also stimulates the growth of L. bulgaricus by producing a factor that is equal to or can be replaced by formic acid and this stimulation can only be demonstrated in moderately heated milks. It was further pointed out that Pette and Lolkema (1950 a and b) and Bautista et al. (1966) had failed to observe this stimulation by S. thermophilus because they had used steamed milk or sterilized milk which contains formic acid as a consequence of intensive heat treatment.

Tramer (1973) showed that a mixed yogurt culture of both L. bulgaricus and S. thermophilus gave more acid development in milk heated to the usual time/temperature combination of 203^{o}F for 30 min than in milk heated severely (autoclaved).

Besides the symbiotic action, Pette and Lolkema (1950 a) demonstrated an inhibitory action of L. bulgaricus upon S. thermophilus, caused by the lactic acid produced. S. thermophilus, which are stimulated in the first stage by L. bulgaricus, predominate when the acidity of the culture is still low, while L. bulgaricus increase in number mainly in the later stage of the fermentation process, because they are more acid tolerant.

According to Kroger (1973), Tramer (1973), Sellars (1973) and Davis (1975), S. thermophilus grew best at pH 6.5, with growth stopping at pH 5.2-4.4; L. bulgaricus grew best at pH 5.5, with growth stopping at pH 3.5-3.8.

Temperature of 110-115°F are ideal for optimal growth of L. bulgaricus, while S. thermophilus grow better at 90-100°F (Angevine, 1972; Sellars, 1973; Davis, 1975).

Pette and Lolkema (1951 a) found that during incubation the proportion of both kinds of bacteria is influenced by the incubation time, the acidity of the culture and by the incubation temperature, as the optimum growth temperature is somewhat lower for S. thermophilus than for L. bulgaricus. The concentration of S. thermophilus cells can be increased by lowering the incubation temperature, lowering the amount of inoculum or by incubating to a lower acidity. The converse of these conditions favors the increase of L. bulgaricus.

Most investigators (Pette and Lolkema, 1951 a: Stocklin, 1969; Powell, 1970; Davis et al., 1971, 1974; Kroger, 1973; Sellars, 1973; Nielsen, 1975; Kosikowski, 1977) agree that the optimum ratio of S. thermophilus and L. bulgaricus at the end of incubation should be about 1:1.

Propagation of cultures - The medium used for propagation of cultures should be clean and fresh, of low bacterial content, and free from inhibitory substances. Spray-dried NFDM reconstituted to 10-11% solids has been claimed to be an excellent medium for the propagation of cultures (Davis, 1956; Sellars, 1973).

Sellars (1973), Cottenie (1978), Rasic and Kurmann (1978) suggested that the reconstituted medium should be

heat treated in an autoclave for 15 min at 15 lbs steam pressure, or in a water bath at 190-220°F for 1 hr or in flowing steam for 1 hr since these heat treatments improve the medium as substrate for the bacteria by destruction of the initial organisms, destruction or inactivation of vegetative forms of spores, denaturation of whey proteins and by expelling dissolved oxygen.

The exact temperature of incubation depends on the particular culture employed and balance of organism desired. The incubation time depends on the amount of inoculum used and on culture activity.

According to Sellars (1973), most cultures when inoculated into the medium at the 1% concentration and incubated at 104°F cause coagulation in 6-8 hr. Lundstedt (1969) and Angevine (1972) reported that lower incubation temperature of 86-90°F results in better growth balance of L. bulgaricus and S. thermophilus. Cottenie (1978) advocated 110-113°F as an optimum temperature for cultures. Ashton (1963) advocated the use of separate culturing, claiming that this allows for a greater degree of flexibility and control. If the cultures are propagated spearately, the L. bulgaricus is usually inoculated at 110°F, while S. thermophilus is inoculated at 100°F (Stocklin, 1969).

To maintain optimum activity most cultures should be cooled to 40-45°F when they have developed a pH of 4.0-4.5.

Inoculation and Incubation - The pasteurized and homogenized yogurt mix is cooled to the inoculation temperature and then inoculated with thoroughly mixed culture.

Many investigators have reported and suggested different amount of inoculum, temperature/time of incubation and pH at the end of incubation. This reflects the differences in type and activity of the culture, composition of yogurt mix, condition of the culture at the time of incubation, rate of cooling and the acidity of yogurt desired.

The data in Table 3 indicate that the amount of culture used ranges from 1 to 5%, incubation temperature from 104 to 113°F, incubation time from 2 to 6 hr and final pH from 4.1 to 4.4.

Ashton (1963) advocated incubation of S. thermophilus at 98-100°F to a low acidity, and of L. bulgaricus at 108-110°F to higher acidities and for longer times. At inoculation, the culture conditions can be varied depending on the relative acidity of the two cultures. It was claimed that the separate culture method is the more flexible and easier to control and tends to enable better control of variable physical characteristics which are less controllable by the mixed culture method.

According to Lundstedt (1969, 1973) the best incubation time-temperature combination for obtaining top quality product in terms of body, texture, and flavor is 86°F, for

Table 3. Some methods currently employed for the inoculation and incubation of yogurt cultures.

Origin	Amount of Culture	Temperature	Time	Final acidity
Stocklin (1969)	1%	110°F	3-3 1/2 hr	pH 4.3
Powell (1970)	2-5%	108 - 113°F	2-6 hr	pH 4.1
Sellars (1973)	2%	104 - 113°F	Depends on desired final acidity	
Davis (1973)	1-2%	110°F	"	
Kroger (1973)	2%	110°F	3-3 1/2 hr	pH 4.2
Kosikowski (1977)	2-5%	113°F	3-6 hr	pH 4.4
Cottenie (1978)	2.2%	110°F	4 hr	pH 4.4

14 to 16 hr. The advantages of the low temperature, long set method were pointed out; a) the proper ratio of bacteria is obtained and as a result the symbiotic metabolic byproducts are balanced to give the ideal acid-acetaldehyde concentration, b) the body and texture are good and there is less tendency to wheying-off, c) cooling to prevent too much acid development can be achieved faster and it requires less energy input, and d) packaging could be scheduled more conveniently. Obert (1978) reported that prolonged incubation for 10-16 hr at 90-95^{o}F is the most effective and cheapest for improving yogurt consistency. However, Kosikowski (1977) pointed out that this low temperature, long set method requires markedly increased net time of processing and more critical control of temperature is necessary since this temperature range is near the point at which the culture organisms do not thrive.

After inoculation, the symbiotic growth of two microorganisms starts. S. thermophilus grows rapidly until a pH of about 5.5 is reached. Then, the growth of L. bulgaricus is progressively favored. If incubation is not halted at between pH 4.0 to 4.4, the acidity would go well below pH 4.0 since L. bulgaricus is capable of growing at this low pH (Kroger, 1976). Normally 20 to 30% of the lactose is fermented, although greater fermentation has been reported (Rasic and Kurmann, 1978). Coagulation commences at pH of about 5.3, depending on the milk and

the protein content (Davis, 1956). At pH 4.6-4.7 precipitation is essentially completed, with the caseins existing in a salt-free state (Humphreys and Plunkett, 1969; Rasic and Kurmann, 1978).

Acid development can be monitored by titration or measurement of pH.

Flavor Development in Yogurt - Development of yogurt flavor goes hand in hand with acid production through a symbiotic bacterial relationship. The characteristic flavor of yogurt is due to lactic acid, which is odorless, and to trace amounts of acetaldehyde, diacetyl and acetic acid. The original milk components and their concentrations also play a role, especially the fat and SNF (Kroger, 1973; Sandine et al., 1974; Kosikowski, 1977).

According to Pette and Lolkema (1950 c) the main flavor components of yogurt are lactic acid and an aroma substance produced by L. bulgaricus. The aroma substance from the cultures was distilled and acetaldehyde was found in the distillate. Greater amount of acetaldehyde was observed when the acidity was high. Keenan and Bills (1968) also emphasized the importance of this compound in yogurt flavor. They suggested that high concentration of acetaldehyde is necessary to produce a desirable flavor in yogurt.

Bottazzi and Vescovo (1969) correlated acetaldehyde content with yogurt flavor. A good flavored yogurt

resulted from L. bulgaricus strains producing greater than 8.0 ppm acetaldehyde and weak flavor resulted when less than 4.0 ppm of acetaldehyde was produced. These workers also suggested that acetone may be involved in typical yogurt flavor. A ratio of 2.8:1 (acetaldehyde:acetone) produced a strong yogurt flavor. Hamdan, et al. (1971) have ascertained levels of 23-26 ppm acetaldehyde in natural yogurt, while 15-20 ppm was indicated as a normal range by Bottazzi and Vescovo (1969). Kroger (1976) also stressed the importance of acetaldehyde for good yogurt flavor, indicating that acetaldehyde comprises about 90% (50 ppm) of the carbonyl compounds present in yogurt. Rasic and Kurmann (1978) considered that the optimum flavor of yogurt is obtained by an acetaldehyde content ranging between 23 and 41 ppm.

Bottazzi and Dellaglio (1967) considered that acetaldehyde and diacetyl are important flavor components of dairy products. The production of these two compounds by S. thermophilus and other lactic streptococci was investigated. It was shown that S. thermophilus formed more acetaldehyde and diacetyl than other homofermentive lactic streptococci studied. Lindsay and Day (1965) found that the concentration of acetaldehyde and diacetyl in lactic cultures determine the intensity of the flavor but that relative amounts are of great importance for the flavor quality. These investigators reported that an

acetaldehyde:diacetyl ratio of 1:4 existed in yogurt cultures of normal flavor. However, in a study by El-Sadec et al. (1972) on zabady (the Egyptian fermented milk similar to yogurt) cultures, it was indicated that acetaldehyde is an important flavor component in zabady while the role of diacetyl appeared to be negligible.

Although there have been various reports from many investigators on desirable acetaldehyde concentration in yogurt, on other carbonyl compounds and on their ratio, there has been increasing evidence that the presence of acetaldehyde is essential if the product is to have an acceptable flavor.

A principal role in acetaldehyde production is attributed to L. bulcaricus, although various strains of these species shown considerable differences in fermentation patterns.

Hamdan et al. (1971) examined the production of acetaldehyde by commercial yogurt cultures and their single strain components. A single strain of S. thermophilus, L. bulgaricus, and a 1:1 mixture of both were used separately to inoculate milk. L.bulgaricus produced more acetaldehyde (8 ppm) than S. thermophilus (4 ppm) during a period of five hr but the amount was still below that (25 ppm) formed when the organisms were grown together. From these results Hamdan et al. (1971) not only supported the symbiotic relationship that exists between the two

organisms as indicated by other workers (Pette and Lolkema, 1950 a and b; Bautista et al., 1966; Galesloot et al., 1968; Veringa et al., 1968), but also demonstrated that L. bulgaricus is the principal organism in acetaldehyde production in a yogurt culture.

Cooling and Storage

Cooling of yogurt serves to restrict further acid development. Mocquot and Hurel (1970) reported that cooling begins when the desirable pH of 4.5-4.7 is reached. Yeager (1975) suggested that cooling should start at pH 4.4-4.8 in order to obtain a final pH of 3.9-4.2. Kosikowski (1977) and Cottenie (1978) recommended pH 4.4 as a starting point for cooling. Rasic and Kurmann (1978) considered that pH 4.4 was too low. Stocklin (1969) claimed that the appearance of traces of whey on top of the coagulated yogurt is a better criterion to determine when to cool than pH or titratable acidity.

Sellars (1973) and Tamime and Greig (1979) suggested that at the end of incubation the yogurt should be cooled as quickly as possible. According to Rasic and Kurmann (1978) the rate of cooling should not proceed too slowly or too fast since too slow cooling may result in additional increase in acidity and cooling too rapidly causes the formation of condensate and contraction of the gel.

Kosikowski (1977) noted that a good forced-air blower system sufficient to chill the product within one hr or

less after entering into cold room is a major requirement in cooling. Cottenie (1978) described a cooling tunnel equipped with a conveyer and a cold-air blowing system at both sides.

Most investigators agree that yogurt should be stored at or below 41°F to prevent over-acidification and microbial or enzymatic spoilage of yogurt. Kosikowski (1977), Kroger (1973) and Davis (1971) advocated the storage of yogurt at 40°F or lower. Humphreys and Plunkett (1969) recommended a temperature range of 39-41°F for yogurt storage. Higher temperatures can lead to defects such as bitterness and low temperature to partial freezing and destabilization.

Yogurt Drink

The tremendous increase in yogurt consumption in the U.S. and all over the world has made food manufacturers look to other yogurt based dairy products as an opportunity for product expansion. Yogurt drink has been introduced as one of the yogurt based dairy varieties which also include yogurt salad dressing and yogurt mayonnaise.

Yogurt drink is not a new concept, but earlier efforts to introduce this new product were unsuccessful because the market was not ready (Morley, 1979). The recent trend to acceptance of more varied yogurt products and the existing vast beverage market suggest a bright future for this liquid yogurt product.

Morley (1979) reported that about 10 dairy plants in the U.S. were manufacturing liquid yogurt drinks. Another report (Griffin, 1979) listed only eight dairy companies marketing yogurt drinks. Some stabilizer companies (Anonymous, 1978 a and b; Anonymous, 1979) introduced processing methods of yogurt drinks using proprietary blended stabilizers.

Processing

Yogurt drinks currently produced can be classed into two somewhat arbitrary categories.

One is the viscous and semiliquid type which is a heavy fluid but still drinkable. Morley (1979) defined this type of yogurt drink as a sweetened, low fat milk which has been cultured with the usual yogurt producing bacteria S. thermophilus and L. bulgaricus and which is drinkable at refrigerator temperature.

The other is a less viscous liquid drink made by diluting plain yogurt with water or aqueous flavorings. Flavoring materials (usually fruit), sweeteners and stabilizers are used in either type of drink.

Duitschaever and Ketcheson (1974) studied the production of yogurt drink. Homogenized 2% fat milk was heated to 176^{o}F for 20 min, cooled to 110^{o}F and inoculated at this temperature with 3% yogurt culture and incubated to a pH 4.4-4.3. The coagulum was homogenized at 0-50 psi and heated to 128^{o}F. Sugar, 0.25% high

methoxyl pectin and 0.25% ascorbic acid were added at this stage. Sugar (7-10%) was used when the flavor extract was used at a concentration of 0.1-1%, or 3-5% sugar was employed when natural flavor concentrates were used at concentrations of 5-10%. The product was then cooled to 60-68°F, homogenized at 2500 psi, packaged and stored at 39°F. Data obtained in taste panel evaluation showed good acceptance of natural orange and cherry flavored yogurt drinks.

One commercial report (Anonymous, 1978 a) described the procedure for making lowfat yogurt drink. The formulation suggested for this product is 0.5-2.0% fat, 8.25-10.0% SNF, 0.2-0.3% stabilizer and 4.0-10.0% sugar. Fresh milk is standardized to the desired butterfat and SNF level with Grade A NFDM. Stabilizer is blended with sugar and added to the milk. The mixture is homogenized at 3,000 psi single stage after the temperature reaches 145°F, and heated to 180°F and held for 30 min. A frozen yogurt culture is added after the mix is cooled to 100°F. When the yogurt reaches pH 4.3, it is cooled with slow agitation to 90°F, flavored and then cooled rapidly to 40°F. The product is held in 38°F cooler overnight to develop the desired body, texture and mothfeel.

Morley (1978) proposed a procedure for manufacturing yogurt drink. Liquid milk products, NFDM, sugar and commercial yogurt drink stabilizer are blended. This mixture

is pasteurized at 180°F for 30 min, homogenized at the pasteurization temperature at 1300 psi, cooled to 108-112°F, and then inoculated with a yogurt starter and agitated for 30 min. Incubation is continued without agitation to a pH of 4.3 over about 4-6 hr. The product is then agitated again and cooled to 45-50°F. Flavors are added at about 10% level to the product with the recommended formula of 1.00% fat, 9.25% SNF, 5.50% sugar and 0.25% stabilizer.

Another commercial report (Anonymous, 1979) suggested a method of manufacturing yogurt drink using a special proprietary blended stabilizer. Liquid milk products, sugar and stabilizer are blended. This mixture is pasteurized at 180-190°F for 30 min, homogenized at 1500-2000 psi, cooled to 110-112°F and then inoculated with a yogurt starter. The product is incubated to a pH of 4.3 during 4-6 hr, cooled to 45°F and packaged. The proposed composition of the product is 1.0% fat, 9.0% SNF, 5.0-6.0% sugar, and 0.1-0.2% stabilizer.

Hekmati and Lame (1974) studied the production of a fermented milk product, known as "dough" which is widely consumed as a refreshing beverage in the Middle East areas. Yogurt was prepared from milk of 2.5% fat content and diluted with an equal volume of water. Salt and flavoring essences were added. The mixture was homogenized at 2500 psi, bottled and held at low temperature. Sediment was

retarded by homogenization and low storage temperature. Composition of the product was 1.22% fat, 1.52% protein, 5.60% total solids, 1.0% salt and 0.006% flavoring essence.

Another commercial report (Anonymous, 1978 b) described a method of producing fruit yogurt drink. A commercial proprietary stabilizer is dry blended with sugar and color. Another stabilizer solution (10% concentration) is stirred into yogurt. The dry blended mixture and flavor are added to the yogurt using a high speed stirrer. The pH is checked and adjusted to 4.0 to 4.1. The product is pasteurized at 185^{o}F, homogenized at 1400 psi at the same temperature and hot (185^{o}F) filled. The suggested composition of the product is 75-85% skim milk yogurt, 14-24% fruit preparation, sugar, water and 1% stabilizer.

Future of Yogurt Drink

Yogurt drink is the combination of the natural ingredients of yogurt with a refreshing taste of a liquid drink. Dairy companies currently producing yogurt drink report a favorable acceptance by consumers. Flavor and fruit suppliers have indicated that dairy processors and major beverage companies are moving quickly into this new product area (Griffin, 1979).

Drucker (1973), Steinitz (1975), Morley (1978, 1979), and Griffin (1979) indicate that the prospects for yogurt drink are promising. Their predictions are based upon the

strong appeal of yogurt drink to dairy processors due to its versatility and relative ease of production, good consumer acceptance of it as a viable alternative to regular milks and soft drinks, and upon the huge beverage market which appears to be receptive to yogurt drink. However, Morley (1979) stresses the importance of producing a consistently good product and of creating a demand through creative advertising for this new product to be successful in the beverage market.

MATERIALS AND METHODS[a]

Preparation of Samples

Preparation of Mother Culture

Culture - A European SKI yogurt culture with slime forming capabilities was obtained from Michigan State University dairy plant.

Culture Medium - Spray-dried NFDM (Michigan Milk Producers Association, Ovid, Michigan) was reconstituted to 10% (W/W) as a medium for the propagation of culture. This reconstituted skim milk solution was dispensed into glass bottles (300 ml and 100 ml), sterilized in the autoclave at 121°C for 10 min and stored in a refrigerator.

Devices for Transfers - Pipets were dry sterilized at 180°C for 2 hrs. A pipet case was used for protection during sterilizing and storing pipets.

Culture Room - A hood in the laboratory was used. The table inside the hood was wiped off thoroughly before use with 95% ethyl alcohol.

Incubation - One percent culture was transferred to skim milk (10% solids) medium and the inoculated medium was held 45°C bath for 3 hr until coagulated. The culture was then cooled to 3-5°C and held in a refrigerator until the next propagation or use. Culture transfers were made every 5-7 days to assure proper activity.

[a]Mention of certain products or equipment does not imply endorsement.

Preperation of Yogurt

Treatment of Milk - The milk used in this study was obtained from the Michigan State University dairy plant. Fat content of the milk was determined by Babcock method (AOAC, 1975). The milk was standardized to 1.5% fat and 12.0% NFS with spray-dried NFDM and water. The mix was pasteurized by a batch process at 88°C for 40 min in 10 gallon stainless steel milk cans, cooled rapidly to 60°C in an ice water bath and homogenized in a two stage Gaulin BW-40 homogenizer at pressures of 70 and 35 kg/cm^2 on the first and second stages respectively.

Incubation - The pasteurized and homogenized mix was cooled to 43-45°C and inoculated with 1.5% culture. The inoculated warm mixture was dispensed into glass beakers (1500 ml and 2000 ml) and incubated at 43-45°C until pH 4.4 was attained. The time required to reach this pH was usually about 3 hr.

Cooling and Cold Storage - The yogurt was then cooled to 3°C in a moving-air cold room and maintained at this temperature for subsequent use in the preparation of yogurt drink. The procedures for yogurt preparation are outlined schematically in Figure 1.

Preparation of Yogurt Drink

Composition - Yogurt drink consisted of 25% low fat yogurt previously prepared, 10% frozen concentrated orange juice (4+1), 9% sugar and 56% water. The frozen concentrated

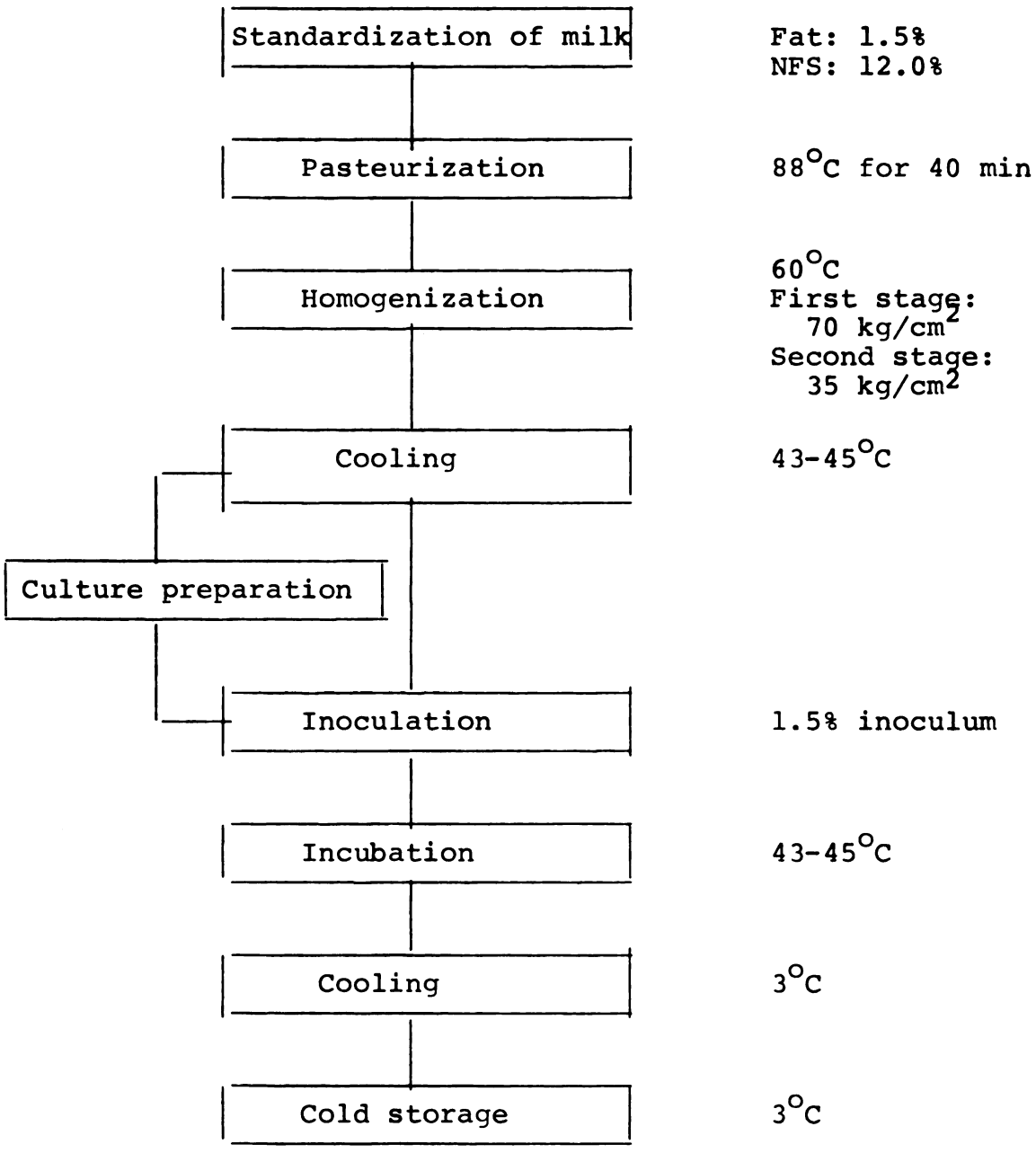

Figure 1. Flow diagram of yogurt manufacture.

orange juice was obtained from the Michigan State University food stores. Optimal sugar level was determined by sensory evaluation using ranking method (see sensory evaluation).

Processing - The ingredients were blended with a high speed mixer for 3 min in 5 or 10 gallon milk cans and dispensed in equal volume (5.0 liters) into large stainless beakers for processing to evaluate the experimental variables of pH, pasteurization temperature/time, homogenization pressure and stabilizers (see experimental variables). The pH of the mixture was adjusted using 50% food grade lactic acid (Monsanto Co., St. Louis, Missouri) and the stabilizers were added at this stage. The mixture was pasteurized either by a batch process or by means of a Cherry Burrell (UHT) heat exchanger (Spirotherm) equipped with a spiral holding tube, cooled to 60°C in an ice water bath and then homogenized in a two stage Gaulin BW-40 homogenizer.

Cooling and Cold Storage - The homogenized product was cooled rapidly to 5°C in an ice water bath and stored at 5°C. The procedures for yogurt drink preparation are outlined in Figure 2.

Determination of Fat and Total Solids in Yogurt and Yogurt Drink

Percentage of fat and total solids of yogurt and yogurt drink were determined by the Mojonnier modification of

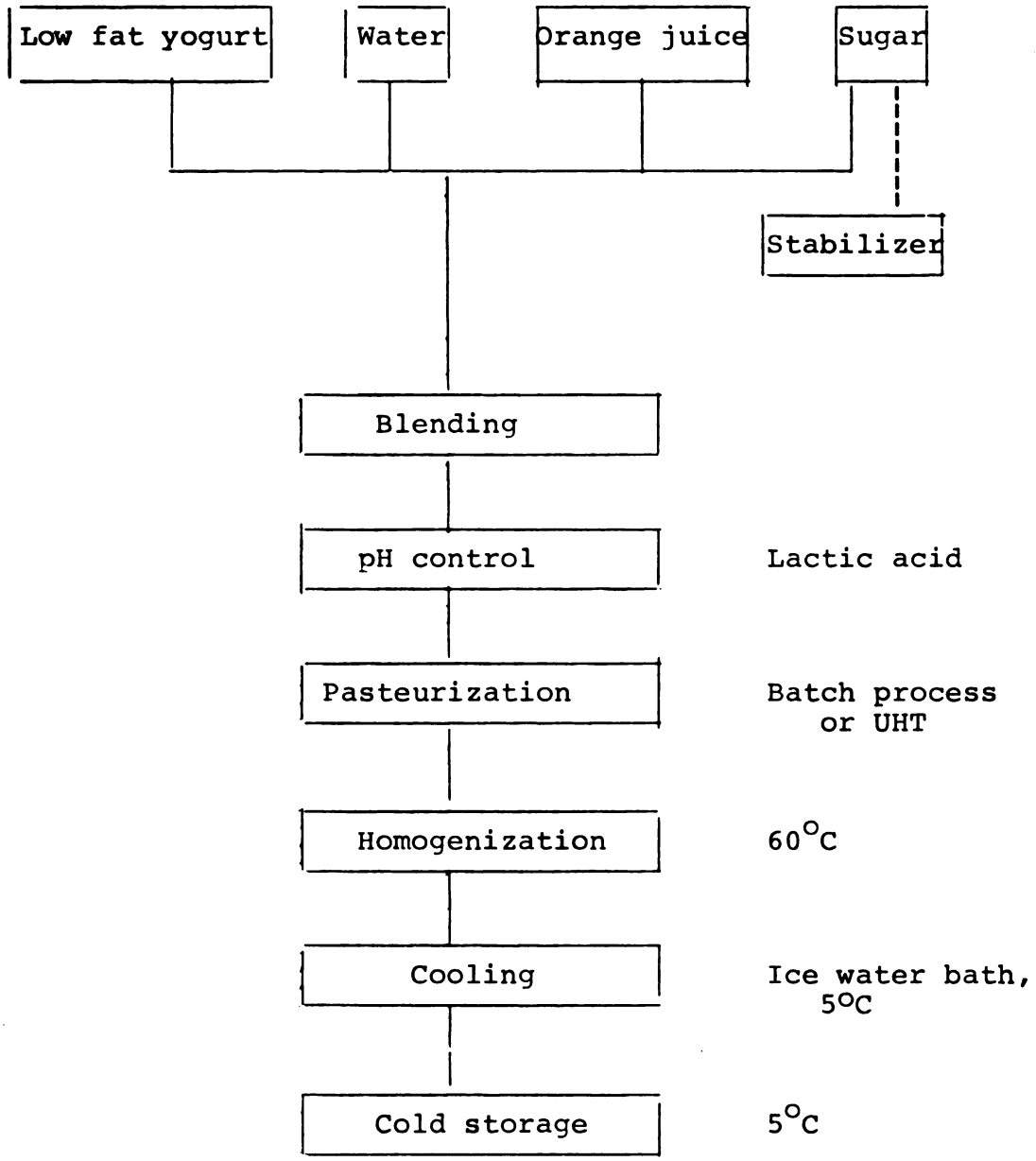

Figure 2. Flow diagram of yogurt drink manufacture.

the Roese-Gottlieb extraction procedure and the Mojonnier vacuum oven procedure (Mojonnier and Troy, 1972) respectively.

Experimental Variables in the Preparation of Yogurt Drink Samples

The effect of pH, pasteurization temperature/time, homogenization pressure and stabilizers on the stability and sensory properties of yogurt drink was studied.

Processing Parameters

pH - Acidity of yogurt drink samples was adjusted to four pH levels using 50% food grade lactic acid; 3.2, 3.6, 4.0, and 4.2.

Pasteurization Temperature/Time - Yogurt drink samples were pasteurized at four pasteurization temperature/time combinations: 65^{o}C for 30 min, 75^{o}C for 10 min, 88^{o}C for 2 min (batch process), and 115^{o}C for 4 sec (UHT).

Homogenization Pressure - Yogurt drink samples were homogenized at four different pressures at 60^{o}C; 70 kg/cm^{2}. 105 kg/cm^{2}, 140 kg/cm^{2} and 175 kg/cm^{2}, with second stage constant at 35 kg/cm^{2} and first stage increasing by 35 kg/cm^{2} from 35 kg/cm^{2} to 140 kg/cm^{2}.

Stabilizers

Guar gum carrageenan, pectin, propylene glycol alginate were used individually at four concentration levels: 0.05, 0.10, 0.15 and 0.20%.

Carboxymethyl cellulose gum (CMC) was used in combination with guar gum as follows:

1) Guar gum 0.20% + CMC 0.05%
2) Guar gum 0.15% + CMC 0.05%
3) Guar gum 0.15% + CMC 0.10%
4) Guar gum 0.00% + CMC 0.15%

Guar gum was also evaluated in combination with propylene glycol alginate and pectin:

1) Guar gum 0.10% + propylene glycol alginate 0.10%
2) Guar gum 0.10% + pectin 0.10%

One commercial proprietary yogurt drink stabilizer composed of gelatin, locust bean gum and carrageenan was evaluated at six concentration levels: 0.05, 0.10, 0.15, 0.20, 0.25 and 0.30%.

Stability Studies

Sediment

Sedimentation was observed in the yogurt drink samples (see determination of optimum processing parameters) to study the effect of processing parameters on the stability of the product.

The samples after cooling were dispensed into 250 ml graduated cylinders and stored in a refrigerator at 5°C. The amount of sediment was measured after storage for 5 and 8 days.

Syneresis

If syneresis was observed in the yogurt drink samples, it was noted and recorded to indicate the effect of stabilizers on the stability of the product.

The samples after cooling were dispensed into 250 ml graduated cylinders and stored in a refrigerator at $5^{o}C$. The amount of syneresis was checked 3, 5, 8 and 10 days after storage.

Protein determination

The supernatant fluid of yogurt drink samples was decanted into small beakers immediately after final checking of sediment or syneresis and protein content was determined on them. Total nitrogen was determined using a semimicro Kjeldahl method (Swaisgood, 1963; Harte, 1978). Samples containing approximately 15 mg of nitrogen were digested with 5 ml of digestion mixture on an electric burner for 90 min. The digestion mixture consisted of 5 g SeO_2 and 5 g $CuSO_4 \cdot 5H_2O$ in 500 ml of concentrated H_2SO_4. After the initial digestion, the samples were cooled, 2 ml of 30% H_2O_2 was added and digestion continued for another 90 min. After cooling, the flasks were rinsed down with 10 ml of redistilled water. Immediately after the flasks were attached to a steam distillation apparatus, approximately 25 ml of 40% NaOH was added. The released ammonia was steam distilled and collected into 15 ml of 4% boric acid solution containing 5 drops of mixed indicator. The

indicator consisted of 400 mg bromocresol green and 40 mg methyl red in 100 ml of 95% ethanol. Standard 0.0205 N HCl was used to titrate the sample. The nitrogen determination was performed in duplicate and total nitrogen values were converted to crude protein using a factor of 6.38. The recovery of nitrogen from standards was 98.11 ± 1.26% using L-tryptophan.

Sensory Evaluation

In this study sensory subjective analysis was carried out to determine the sugar content of yogurt drink and to determine the effect of processing parameters and stabilizers on the sensory properties of yogurt drink. Two types of preference test were employed for this purpose: ranking method and hedonic scaling method (Amerine et al., 1965; Lavery, 1972; Larmond, 1977).

Ranking Method

Ranking method was used to determine the sugar content of yogurt drink and the effect of homogenization pressure on the sensory properties of the product. In this method the panelists were presented with randomly coded samples to rank in order of preference. To determine if the difference between the sample is significant at the 5% level of significance, the rank totals were compared with the values in Table 23 of the Appendix. When one or more rank sums were lower than the upper left

value in the block and higher than the upper left value, statistical significance was indicated.

Hedonic Scaling Method

A nine-point hedonic scaling method were used to determine the effect of pH, pasteurization temperature/time and stabilizers on the sensory properties of the product. In this method the panelist were presented with randomly coded samples to express their degree of liking or disliking. The ratings for each sample were given numerical values ranging from like extremely (9) to dislike extremely (1). The results were anlyzed by analysis of variance, for which the correction factor (CF), the sum of squares (ss), the degrees of freedom (df), the mean square (ms), and the variance ratio (F) were calculated. To determine if the difference between the samples is significant at the 5% level of significance, the calculated F value was checked on Table 24 of the Appendix. In this method, the variance ratio (F) must exceed tabular value to be significant. When there was a significant difference between the samples, those which were different were determined using Tukey's test (Snedecor, 1956). Least significant difference was calculated and any two sample means (score/number of judgements) that differ by the value of least significant difference or more were considered to be different significantly.

Determination of Optimum Processing Parameters

Yogurt drink mix was subjected to various treatments (combinations of processing parameters) in order to determine optimum pH, pasteurization temperature/time and homogenization pressure on the stability and sensory properties of the product. All runs were made in duplicate.

pH

The experimental runs are shown in Table 4. The runs from No. 1 to No. 6 were used for sensory evaluation and the runs from No. 7 to No. 14 for sediment observation and protein determination.

Pasteurization Temperature/Time

The runs from No. 7 to No. 14 in Table 4 were used for sediment observation and protein determination. The runs made for sensory evaluation are shown in Table 5.

Homogenization Pressure

The runs made are shown in Table 6. All runs were used for sediment observation and protein determination, and runs from No. 1 to No. 4 for sensory evaluation.

Table 4. Treatments of yogurt drink to determine optimum pH on the stability and sensory properties of the product.

Run No.	pH	Pasteurization temperature/ time	Homogenization pressure	Remarks
1	3.6	65°C/30 min	140 kg/cm^2	Sensory evaluation
2	4.0	65°C/30 min	140 kg/cm^2	Sensory evaluation
3	4.2	65°C/30 min	140 kg/cm^2	Sensory evaluation
4	3.6	115°C/4 sec	140 kg/cm^2	Sensory evaluation
5	4.0	115°C/4 sec	140 kg/cm^2	Sensory evaluation
6	4.2	115°C/4 sec	140 kg/cm^2	Sensory evaluation
[a]7(2)	4.0	65°C/30 min	140 kg/cm^2	Sensory evaluation
[a]8(3)	4.2	65°C/30 min	140 kg/cm^2	Sediment, protein determination
9	4.0	75°C/10 min	140 kg/cm^2	Sediment, protein determination
10	4.2	75°C/10 min	140 kg/cm^2	Sediment, protein determination
11	4.0	88°C/2 min	140 kg/cm^2	Sediment, protein determination
12	4.2	88°C/2 min	140 kg/cm^2	Sediment, protein determination
[a]13(5)	4.0	115°C/4 sec	140 kg/cm^2	Sediment, protein determination
[a]14(6)	4.2	115°C/4 sec	140 kg/cm^2	Sediment, protein determination

[a]Two runs are same treatments.

Table 5. Treatments[a] of yogurt drink to determine optimum pasteurization temperature/time on the stability and sensory properties of the product.

Run No.	pH	Pasteurization temperature time	Homogenization pressure	Remarks
1	4.0	65°C/30 min	140 kg/cm^2	Sensory evaluation
2	4.0	75°C/10 min	140 kg/cm^2	Sensory evaluation
3	4.0	88°C/ 2 min	140 kg/cm^2	Sensory evaluation
4	4.0	115°C/ 4 sec	140 kg/cm^2	Sensory evaluation

[a]The treatments from No. 7 to No. 14 in Table 4 were used for sediment observation and protein determination.

Table 6. Treatments of yogurt drink to determine optimum homogenization pressure on the stability and sensory properties of the product.

Run No.	pH	Pasteurization temperature/ time	Homogenization pressure	Remarks
1	4.0	88°C/ 2 min	70 kg/cm^2	Sensory evaluation, sediment, protein determination
2	4.0	88°C/ 2 min	105 kg/cm^2	Sensory evaluation sediment, protein determination
3	4.0	88°C/ 2 min	140 kg/cm^2	Sensory evaluation, sediment, protein determination
4	4.0	88°C/ 2 min	175 kg/cm^2	Sensory evaluation, sediment, protein determiation
5	4.0	115°C/ 4 sec	70 kg/cm^2	Sediment, protein determination
6	4.0	115°C/ 4 sec	105 kg/cm^2	Sediment, protein determination
7	4.0	115°C/ 4 sec	140 kg/cm^2	Sediment, protein determination
8	4.0	115°C/ 4 sec	175 kg/cm^2	Sediment, protein determination

Determination of the Most Suitable Stabilizers and their Concentrations in Yogurt Drink

Various stabilizers at different concentrations (see experimental variables) were tested for their stabilizing effects at the predetermined processing parameters. Stabilizers were premixed with sugar and then the dry mixture was added to the mixture of yogurt and water under vigorous agitation until a uniform suspension was obtained. Stability studies were conducted on every sample made by observation of syneresis and protein determination. Sensory evaluation was carried out only on the two samples finally stabilized.

RESULTS AND DISCUSSION

Composition of Yogurt and Yogurt Drink

Mean values plus or minus standard deviation for the content of protein (Kjeldahl), fat (Mojonnier) and total solids (Mojonnier) of yogurt and yogurt drink are shown in Table 7. The composition of low fat yogurt made for this study was carefully controlled by precise standardization of ingredients since commercial yogurts, as previously mentioned, are subject to compositional variables which lead to undesirable complications regarding the analysis of experimental data. The approximate ratio of one (yogurt) to two (water) in yogurt drink was chosen for optimal viscosity and mouthfeel. Frozen concentrated orange juice was selected as natural flavor ingredient and was used at a concentration of 10%. At concentrations of 20 and 15%, the orange flavor was too strong and the unique flavor of yogurt was masked.

In Table 7, the values of protein and fat content in yogurt drink are higher than those of the same constituents divided by four (25% of yogurt was used in yogurt drink) in yogurt, due to the composition of the orange juice. According to data of the U.S. Department of Agriculture (1975), frozen concentrated orange juice contains 2.5% protein, 0.2% fat and 44.8% total solids.

The desired sugar level of yogurt drink was determined by subjective sensory evaluation using ranking method.

Table 7. Chemical composition of yogurt and yogurt drink.[a]

Product	Protein[b]	Fat[b]	Total solids[b]
Yogurt	4.67±0.02	1.54±0.03	13.63±0.04
Yogurt drink	1.43±0.02	0.41±0.02	17.12±0.06

[a]Variation indicated by standard deviation of duplicate analysis from two runs.
[b]% , w/w

Sugar concentrations of 7%, 8%, 9% and 10% were tested. Sugar concentrations of 4%, 5% and 6% were eliminated during preliminary evaluations because the yogurt drink was too sour. The panelists were presented with four randomly coded samples (each containing different sugar levels) to rank in order of preference. The rank scores given to the samples by the judges are shown in Table 8. A low score indicates greater acceptability (preference) in this method. The rank totals were compared with the values in Table 23 of the Appendix. Since there were four treatments and eight judges, the tabular entries are 13-27. The lowest insignificant rank sum is 13 and the highest insignificant rank sum is 27. One rank sum (11) was lower than 13, three other rank sums were between the range of 13-27 and no rank sum was higher than 27. To show significant difference statistically, one or more rank sums should be lower than 13 and higher than 27. There was no significant difference in preference for the samples at 5% level of significance. However, the lowest

Table 8. Ranked scores of yogurt drink at the sugar concentrations of 7%, 8%, 9% and 10%.

Judges	Sugar level			
	7%	8%	9%	10%
1	4	3	1	2
2	4	3	1	2
3	2	4	1	3
4	3	1	2	4
5	4	3	1	2
6	1	2	3	4
7	4	2	1	3
8	3	2	1	4
Total	25	20	11	24

rank sum (11) of the sample treated with 9% sugar indicated apparent preference for this sample to the other three samples. The judges who showed less preference for the sample treated with 10% sugar indicated that the product was too sweet and those who showed less preference for the samples with 7% and 8% sugar commented that both samples were too sour. Basing on this analysis a sugar concentration of 9% was selected for use in the yogurt drinks.

Chemical and Physical Properties of Yogurt Drink

In order to study the effects of various processing parameters and stabilizers on the stability of yogurt drink, it is reasonable to define the chemical and physical nature of this particular product. The yogurt drink used in this study was composed of 25% yogurt, 10% concentrated orange juice (4 + 1), 9% sugar and 56% water. The constituents of each component which contribute to the characteristics of the yogurt drink are milk proteins (caseins and whey), milk fat, lactose and salts from the yogurt; small amounts of proteins, lipids (orange oil), carbohydrates and trace amounts of salts from orange juice; the sucrose solids.

Milk proteins do exist not in their original form, but in changed form due to the treatments during the manufacture of yogurt. The casein originally is present in milk in the form of small micelles or in smaller

polymeric units and monomeric forms, comprising from 5-10% of the total casein. The micelles referred to as a calcium caseinate-phosphate-complex contain, in addition to casein, inorganic calcium and phosphate, a little magnesium and citrate. Casein is not a heat-coagulable protein in the strictest sense. In normal fluid milk casein is very stable to heat. Parry (1974) stated that casein resists coagulation for as long as 14 hr at boiling temperature and 1 hr at 130°C. During the manufacture of yogurt, however, there is a gradual removal of calcium and phosphorus from the colloidal complex to soluble state due to the acid produced by the yogurt culture. At pH 5.2-5.3 the casein particles are destabilized initiating precipitation. Complete precipitation occurs at pH 4.6-4.7, the isoelectric point, with the casein existing in a salt-free state. In contrast to casein, whey proteins in the native state are not sensitive to changes in hydrogen ion concentration. They are affected considerably by heat treatment of the milk. Above the normal pasteurization temperature of milk, the whey proteins are denatured and their specific structures are disrupted and unfolded. Larson and Rolleri (1955) reported that almost complete denaturation of whey protein occurred by heating milk at 77.5°C for 1 hr, 80°C for 30 min or 90°C for 5 min. In the present study, the yogurt mix was pasteurized at 88°C for 40 min. It is apparent that essentially all whey proteins in the yogurt used for yogurt drink had been denatured.

During the fermentation of yogurt, usually 20-30% of the lactose is fermented into lactic acid as previously mentioned. Calcium removed from the casein micelles mostly combines with lactic acid as calcium lactate.

Fat globules in milk are finely divided and mechanically incorporated within the coagulum structure by homogenization during the manufacture of yogurt. Thus, the semi-solid gel structure of yogurt represent a co-precipitate of destabilized casein and denatured whey proteins entrapping the fat globules and serum with dissolved constituents.

When all ingredients are blended using a high speed mixer for the manufacture of yogurt drink, the gel structure of yogurt is completely broken and consequently the agglomeration of casein micelles associated with denatured whey proteins may exist in the form of a micro-dispersion, while a part of the casein and undenatured proteose-peptone fraction are in true colloidal dispersion and the non-protein nitrogen compounds such as amino acids and urea, lactose and free salts are in true solubilized solution. The milk fat globules would exist as an oil-in-water emulsion in the mixture, and the sucrose would be completely dissolved in the system. The proteins of the orange juice are likewise dispersed colloidally, the orange lipids as an emulsion and the carbohydrates of the orange juice will be in true solution in the aqueous phase of the yogurt drink.

Effect of Processing Parameters on the Stability and Sensory Properties of Yogurt Drink

In this study, the first priority was given to the results of sensory evaluation for choosing optimum processing parameters since food products should be appealing and palatable. Therefore, sensory tests were performed prior to stability studies and when sensory evaluation data indicated that a significant difference existed between (among) the samples, only those treatments which gave satisfactory results were employed in subsequent experiments.

Regarding the stability of this system, the milk proteins are the constituents most sensitive to pH, heat treatment and homogenization pressure. It should be noted here that the caseins of yogurt and the denatured whey proteins which adsorb or associate with them, are of primary importance to the body and rheological properties of the yogurt drink. Therefore, protein determination on the supernatant fluid of yogurt drink as well as measurement of sediment were employed to measure stability.

pH

Sensory evaluation was performed to find out the effect of pH on the sensory properties of yogurt drink. The processing parameters employed for this sensory analysis are shown in Table 4. pH 3.2 was excluded in this sensory

evaluation since this concentration of H+ imparted an extremely sour flavor to the product in the preliminary evaluation by a small group of panelists. One heat treatment ($65^{o}C/30$ min) was chosen from the three conventional methods employed in this study and the other was UHT ($115^{o}C/4$ sec). A nine point hedonic scaling method which is most commonly used for preference testing was used. The results are shown in Table 9 and an analysis of variance was conducted on the data of Table 9, as shown in Table 10. The calculated variance ratio F value (5.73) was checked on Table 24 of the Appendix with 5 df in the numerator and 105 df in the denominator. The variance ratio F value (5.73) exceeded the tabular value (2.29), thus the difference between samples was indicated as significant. To determine the samples that are significantly different, least significant difference (1.35) was calculated by Tukey's test. The differences of mean values are presented in Table 11. For any two samples to be significantly different, the difference of their mean values should be greater than least significant difference (1.35). As shown in Table 11, the differences between the samples that are greater than 1.35 are from B-(A or D) and E-(A or D). Therefore, the samples B and E are significantly preferred to the samples A or D. It can be also found that the samples B and E are preferred much more if not significant to the samples C or F. The

Table 9. Scores of the samples used to study the effects of pH on the sensory properties of yogurt drink.

Judges	Treatments						
	A	B	C	D	E	F	
	[a] 3.6 [b] 65°C/30' [c] 140kg/cm^2	4.0 65°C/30' 140kg/cm^2	4.2 65°C/30' 140kg/cm^2	3.6 115°C/4" 140kg/cm^2	4.0 115°C/4" 140kg/cm^2	4.2 115°C/4" 140kg/cm^2	Total
1	5	8	7	5	7	8	40
2	7	8	8	7	7	8	45
3	6	7	3	5	7	3	31
4	6	6	5	5	3	4	28
5	2	7	8	3	6	8	34
6	4	7	5	4	7	7	34
7	6	7	7	6	8	8	42
8	6	7	9	6	6	8	42
9	3	6	7	4	6	6	32
10	5	7	4	6	4	4	30
11	3	8	3	4	8	5	31
12	6	6	7	6	6	6	37
13	3	5	5	3	7	5	28
14	7	6	7	6	7	5	38
15	3	7	7	3	4	4	28
16	6	6	7	6	7	2	34
17	2	7	4	3	7	3	26
18	7	8	3	7	9	6	41
19	7	7	2	6	9	7	38
20	2	7	4	2	8	5	28
21	7	6	1	6	5	2	27
22	6	6	7	6	6	6	37
Total	109	149	120	109	144	120	751
[d] Sample means	4.95	6.77	5.45	4.95	6.55	5.45	

[a] pH
[b] Pasteurization temperature/time
[c] Homogenization pressure
[d] Score/number of judgements

Table 10. Analysis of variance from the data of Table 9.

Source of variation	df	ss	ms	F
Samples	5	68.13	13.63	[a]5.73
Judges	21	112.10	5.34	2.24
Error	105	250.04	2.38	

[a]significant at 0.05

Table 11. Difference of sample means from data of Table 9.

Treatments	Mean values	Difference of mean values
B-[a](A or D)	6.77 - 4.95	1.82
B- (C or F)	6.77 - 5.45	1.32
B-E	6.77 - 6.55	0.22
E- (A or D)	6.55 - 4.95	1.60
E- (C or F)	6.55 - 5.45	1.10
(C or F) - (A or D)	5.45 - 4.95	0.50

[a]( ) - Mean values of both samples are same.

conclusion derived from the Tables 9, 10 and 11 is that the samples adjusted to pH 4.0 were significantly preferred to the samples adjusted to pH 3.6 and much more preferred to the samples adjusted to pH 4.2 at the 5% level of significance. So pH 4.0 was taken as the best for the product and pH 3.6 was excluded in the subsequent experiments. The samples adjusted to pH 3.6 were described as too sour. As for the samples adjusted to pH 4.2, many panelists evaluated them as a little too sweet. This also supports the result of preliminary evaluation with a smaller group of panelists. In the preliminary evaluation, products with a pH of 3.2 had been rated as very sour flavor, so this pH was not used in the sensory evaluations.

The influence of pH on the sediment volume of the samples 5 and 8 days after storage are shown in Table 12. The corresponding Kjeldahl protein data on the supernatant fluid of the samples 8 days after storage are shown in Table 13. In this experiment, pH variation was narrowed from pH 3.2, 3.6, 4.0 and 4.2 to pH 4.0 and 4.2 based on the results of previous sensory evaluation. Four different heat treatments were used (the data from this trial were also to be used for studying the effect of heat treatments on the product stability) at a constant homogenization pressure, 140 kg/cm^2. From Table 12 and 13, it is evident that the increase in sediment volume with increase in pH of the product at all heat treatments

Table 12. Sediment volume of the samples used to study the effect of pH on the stability of yogurt drink.

pH	Treatments		[a]Sediment (ml)	
	Pasteurization temperature/time	Homogenization pressure	5 days	8 days
4.0	65°C/30 min	140 kg/cm^2	219	196
4.2	65°C/30 min	140 kg/cm^2	222	200
4.0	75°C/10 min	140 kg/cm^2	215	192
4.2	75°C/10 min	140 kg/cm^2	218	197
4.0	88°C/2 min	140 kg/cm^2	160	126
4.2	88°C/2 min	140 kg/cm^2	171	129
4.0	115°C/4 sec	140 kg/cm^2	35	34
4.2	115°C/4 sec	140 kg/cm^2	37	35

[a]Average of duplicate runs.

Table 13. Protein content of supernatant fluid of the samples used to study the effect of pH on the stability of yogurt drink.

pH	Treatments		[a]Protein (%)
	Pasteurization temperature/time	Homogenization pressure	
4.0	65°C/30 min	140 kg/cm^2	0.37
4.2	65°C/30 min	140 kg/cm^2	0.33
4.0	75°C/10 min	140 kg/cm^2	0.41
4.2	75°C/10 min	140 kg/cm^2	0.35
4.0	88°C/2 min	140 kg/cm^2	0.44
4.2	88°C/2 min	140 kg/cm^2	0.41
4.0	115°C/4 sec	140 kg/cm^2	0.91
4.2	115°C/4 sec	140 kg/cm^2	0.59

[a]Average of duplicates from two runs.

used is responsible for the decrease of protein content in the supernatant fluid of the product. The decrease of sediment volume in each sample during prolonged storage is viewed as resulting mainly from the contraction of sediment portion. A slight reduction in the pH of yogurt drink resulted in a noticable decrease of sedimentation and increase of the stable protein in the supernatant. From these results, it appears that some control of sediment formation can be achieved by adjusting pH of the product to 4.0. Based on the fact that the isoelectric point of casein is 4.6, it seems possible that pH 4.2, being closer to the isoelectric point of casein than pH 4.0 is, caused more sedimentation and less stable protein in the supernatant. The solubility of casein is minimum at the isoelectric point and increases rapidly on either side.

Pasteurization Temperature/Time

Sensory evaluation was conducted to determine the effect of pasteurization temperature/time on the sensory properties of yogurt drink. The processing parameters used in this sensory analysis are shown in Table 5. pH 4.0 was used since it was proven in the previous investigations to be the best for the stability and sensory properties of the product. All of the four heat treatments were used with a homogenization pressure constant at 140 kg/cm^2.

Table 14. Scores of the samples used to study the effects of pasteurization temperature/time on the sensory properties of yogurt drink.

Judges	Treatments				Total
	[a]4.0 [b]65°C/30' [c]140 kg/cm^2	4.0 75°C/10' 140 kg/cm^2	4.0 88°C/2' 140 kg/cm^2	4.0 115°C/4" 140 kg/cm^2	
1	6	7	6	9	28
2	6	9	8	8	31
3	8	5	8	9	30
4	4	6	5	7	22
5	7	7	7	6	27
6	6	7	7	6	26
7	5	5	6	4	20
8	4	6	4	8	22
9	3	6	4	7	20
10	7	7	8	7	29
11	7	8	5	7	27
12	7	7	7	3	24
13	8	7	6	6	27
14	3	6	4	7	20
15	7	6	7	8	28
16	7	7	6	6	26
17	8	8	8	7	31
18	7	7	7	7	28
19	7	8	4	4	23
20	6	6	7	7	26
21	6	6	7	5	24
22	6	7	7	6	26
Total	135	148	138	144	565

[a]pH
[b]Pasteurization temperature/time
[c]Homogenization pressure

Table 15. Analysis of variance from the data of Table 14.

Source of variation	df	ss	ms	F
Samples	3	4.44	2.22	[a]1.46
Judges	21	61.19	2.91	1.91
Error	63	95.81	1.52	

[a]Significant at 5% level.

A nine point hedonic scaling method was also used in this sensory evaluation. The scores of each sample given by 22 panelists are presented in Table 14. Analysis of variance was conducted on the data of Table 14, as shown in Table 15. The calculated variance ratio F value was checked on Table 24 of the Appendix with 3 df in the numerator and 63 df in the denominator. The variance ratio (1.46) was smaller than the tabular value (2.76). So there was no signficant difference in preference for the samples at the 5% level of significance. This indicates that heat treatment variation had negligible effect on the sensory properties of the product.

Data presented in Table 12 and 13 were also used to elucidate the effect of heat treatment variation on the stability of yogurt drink. Sediment volume decreased considerably with increasing temperature at the both pH 4.0 and 4.2 and corresponding increases of protein content in

the supernatant also occurred although there was little difference between the heat treatments of 65°C/30 min and 75°C/10 min. This difference may be due partially to the result of differing extents of β-lactoglobulin-k-casein interactions occurring during heating of the samples. The extent of this interaction is held to play an important role in the heat stability of milk system. The heat induced interaction of β-lactoglobulin and k-casein is believed to proceed through the mechanism of disulfide interchange involving the exposed thiol groups of β-lactoglobulin and the disulfides of k-casein. In yogurt drink, some interaction may have been already present as a result of heat treatment given during the manufacture of yogurt. More interactions may have been induced during the heat treatment of yogurt dirnk, the higher temperature resulting in the greater extent of interaction. Brunner (1977) stated that the association of β-lactoglobulin with k-casein increases the heat stability of colloidal casein.

Changes in the ionic calcium content of the samples induced by heat treatment may be partially responsible for the different stability of the product. It has been reported that ionic calcium plays a role in the stability of milk system, higher calcium ion causing lowered heat stability and that heat treatment results in a transfer of calcium from the ionic to the colloidal state at the higher temperature heat treatment, resulting in the increased stability.

This result may be also attributable partially to the increased hydration of proteins in the system at the higher temperature. When in solution, proteins apparently are hydrated in the sense that a substantial amount of water is bound or trapped by the protein. Increased hydration if not excessive favors the stabilization of proteins.

Homogenization Pressure

Sensory evaluation was conducted to determine the effect of homogenization pressure on the sensory properties of yogurt drink. The processing parameters employed in this sensory analysis are shown in Table 6. A pH 4.0 was maintained for these experiments as this pH had proven best for the stability and sensory properties of the product. Only one heat treatment at 88^{o}C/2 min was used since this temperature/time was optimal for the stability of product among those three conventional heat treatments tried in this study. Variation in heat treatment also apparently has negligible effect on the sensory properties of the product. Ranking method was used in this sensory evaluation. The panelists were presented with four randomly coded samples treated at four different homogenization pressures respectively with the same pH and heat treatment and asked to rank in order of preference. The ranks given to the samples by the panelists are

Table 16. Ranked scores of the samples used to study the effects of homogenization pressure on the sensory properties of yogurt drink.

Judges	Treatments			
	[a]4.0 [b]88°C/2' [c]70 kg/cm^2	4.0 88°C/2' 105 kg/cm^2	4.0 88°C/2' 140 kg/cm^2	4.0 88°C/2' 175 kg/cm^2
1	4	2	3	1
2	3	4	1	2
3	1	2	3	4
4	4	3	2	1
5	2	4	3	1
6	1	4	2	3
7	1	4	3	2
8	1	4	2	3
9	3	2	4	1
10	3	4	1	2
11	4	1	2	3
12	4	2	1	3
13	1	3	4	2
14	4	3	2	1
15	2	1	4	3
16	1	4	3	2
17	3	4	2	1
18	1	2	3	4
Total	43	53	45	39

[a]pH
[b]Pasteurization temperature/time
[c]Homogenization pressure

presented in Table 16. As mentioned previously, low score indicates greater acceptability (preference) in this method. The rank totals were compared with the values in Table 23 of the Appendix. Since there were four treatments and eighteen judges, the tabular entries are 34-56. The lowest insignificant rank sum is 34 and the highest insignificant rank sum is 56. To show significant difference statistically, one or more rank sums whould be lower than 34 or higher than 56. All ranks sums in Table 16 are between the range of 34-56. The conclusion is that there was no significant difference in preference for the samples evaluated. This result indicates that homogenization pressure variation had little effect on the sensory properties of the product.

The processing parameters used for the stability study are also shown in Table 6. pH was held constant at 4.0. Two heat treatments, 88^{o}C/2 min from the three conventional methods and 115^{o}C/4 sec (UHT), were used as heat treatment variation. Sediment volume 5 and 8 days after storage of the samples are shown in Table 17. The corresponding Kjeldahl protein data on the supernatant fluid of the samples 8 days after storage are presented in Table 18. Data in Table 17 reveal that a considerable increase in sediment volume occurred with increasing homogenization pressure at either heat treatment, indicating that homogenization definitely had much effect on the

Table 17. Sediment volume of the samples used to study the effect of homogenization pressure on the stability of yogurt drink.

Treatments			[a]Sediment (ml)	
pH	Pasteurization temperature/time	Homogenization pressure	5 days	8 days
4.0	88°C/2 min	70 kg/cm^2	125	94
4.0	88°C/2 min	105 kg/cm^2	143	112
4.0	88°C/2 min	140 kg/cm^2	156	123
4.0	88°C/2 min	175 kg/cm^2	159	128
4.0	115°C/4 sec	70 kg/cm^2	30	30
4.0	115°C/4 sec	105 kg/cm^2	34	33
4.0	115°C/4 sec	140 kg/cm^2	35	34
4.0	115°C/4 sec	175 kg/cm^2	37	36

[a]Average of duplicate runs

Table 18. Protein content of supernatant fluid of the samples used to study the effect of homogenization pressure on the stability of yogurt drink.

Treatments			[a]Protein (%)
pH	Pasteurization temperature/time	Homogenization pressure	
4.0	88°C/2 min	70 kg/cm^2	0.66
4.0	88°C/2 min	105 kg/cm^2	0.51
4.0	88°C/2 min	140 kg/cm^2	0.44
4.0	88°C/2 min	175 kg/cm^2	0.30
4.0	115°C/4 sec	70 kg/cm^2	1.05
4.0	115°C/4 sec	105 kg/cm^2	0.92
4.0	115°C/4 sec	140 kg/cm^2	0.89
4.0	115°C/4 sec	175 kg/cm^2	0.79

[a]Average of duplicates from two runs.

stability of the product. A corresponding decrease in protein contents of the supernatant fluid of the product also occurred. Parry (1974) stated that nonionic and inert materials such as fats, sugars and fruit pulps appear to adsorb protein in milk systems, finer dispersion tending to concentrate the protein with increasing effectiveness and this accumulation on the interface tends to lower protein stability by encouraging local coagulation, which quickly destabilizes the entire system. Homogenization increases the dispersion of these materials, resulting in more surface area for adsorption. In the present study, the lowered stability of the product at the higher homogenization pressure is believed due to the finer dispersion of the fat and the pulp (from orange juice) and their increased surfaces for adsorption of proteins. The system of yogurt drink used in this study can be closely compared to the system of evaporated milk in the sense that proteins in both are heat treated previously and that both systems have milk fat. Hunziker (1949) and Newstead et al. (1978) stated that homogenization tends to lower the stability of evaporated milk and this tendency increases with increasing homogenization pressure due to the increased formation of fat-protein complex which decreases the stability of the system.

Effect of Stabilizers on the Stability and Sensory Properties of Yogurt Drink

It has been shown in the previous investigations of this study that the optimum processing parameters for yogurt drink are pH 4.0, heat treatment 115°C/4 sec (UHT) and homogenization pressure 70 kg/cm^2. In this study of stabilizers, however, 88°C/2 min rather than UHT was used for heat treatment considering that not all dairy plants have UHT facilities and 175 kg/cm^2 rather than 70 kg/cm^2 was used as homogenization pressure for better distribution of the stabilizers in the product.

The stabilizers used in this study were expected to fulfill the following requirements: bland flavor (no masking of the flavorings), easy to incorporate, readily hydrated, stable, and imparting the desired stabilizing effects (no syneresis) at low concentration without imparting excessive viscosity (heavy mouthfeel) to the product.

Complete dissolution (no clumping) of the stabilizers was achieved successfully by premixing with sugar and adding the mix to the product under vigorous agitation. Concentration of each stabilizer was tried in the sequential order from a low concentration (0.05%) to high, increasing by increments of 0.05%. When certain level of a stabilizer gave too much viscosity to the product,

higher concentrations of the stabilizer were not tried even if the product was still not stabilized.

Syneresis volume of yogurt drink samples after storage for 3, 5, 8 and 10 days as a function of individual stabilizer at different concentrations are shown in Table 19-20 and plotted in Figure 3-7. The results of protein determination on the syneresis after storage for 10 days are also shown in Table 19 and 20.

The stability characteristics of yogurt drink system are of course related to the suspension of hydrated colloids in a aqueous medium. Stabilizers in this type of system are expected to maintain particles in maximum state of suspension and take up free water as water of hydration through intra- and intermolecular interactions. Syneresis of the samples on standing was interpreted as evidence of instability.

Carrageenan (Table 19, Figure 3) proved to be ineffective in stabilizing this system. During initial storage, the stability of the yogurt drink containing higher levels of carrageenan was improved. However, stability diminished markedly with time. After 10 days, almost identical volumes of syneresed fluid were observed in all samples. Increase of the concentration from 0.20% to 0.25% (the broken line) not only caused an adverse effect on the stability, but also imparted apparent viscosity to the product.

Table 19. [a]Syneresis volume (ml) of yogurt drink samples after storage as a function of stabilizers at different concentrations. pH:4.0, heat treatment; 88°C/2 min, homogenization pressure: 175 kg/cm^2.

Concentration (%)	Storage (Days at 5°C) 3	5	8	10	Protein	Remarks (mouthfeel)
	Carrageenan					
0.05	108	120	135	141	0.31	
0.10	86	114	135	141	0.31	
0.15	77	108	133	140	0.31	
0.20	60	88	123	132	0.28	
0.25	76	108	125	130	0.28	Heavy
	Guar gum					
0.05	24	41	76	81	0.31	
0.10	9	13	21	27	0.29	
0.15	3	5	8	9	0.28	
0.20	1	2	3	3	[b]---	
	Commercial proprietary yogurt drink stabilizer					
0.05	77	100	121	129	0.37	
0.10	66	87	107	116	0.42	
0.15	42	66	86	98	0.45	
0.20	0	31	65	70	0.85	
0.25	0	21	28	29	1.10	
0.30	0	0	0	0	[c]---	

[a]Average of duplicate runs

[b]Sample was so stable that essentially no syneresis occurred

[c]No syneresis

Table 20. [a]Syneresis volume (ml) of yogurt drink samples after storage as a function of stabilizers at different concentrations. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

Concentration	Storage (Days at 5°C) 3	5	8	10	Proteins(%)	Remarks (mouthfeel)
	Propylene glycol alginate					
0.05	70	85	91	96	0.38	
0.10	48	61	70	79	0.35	
0.15	30	42	60	73	0.32	
0.20	35	46	61	72	0.31	Heavy
	Pectin					
0.05	68	92	101	104	0.39	
0.10	51	65	76	81	0.35	
0.15	21	28	40	64	0.33	
0.20	34	44	60	75	0.32	Heavy

[a]Average of duplicate runs.

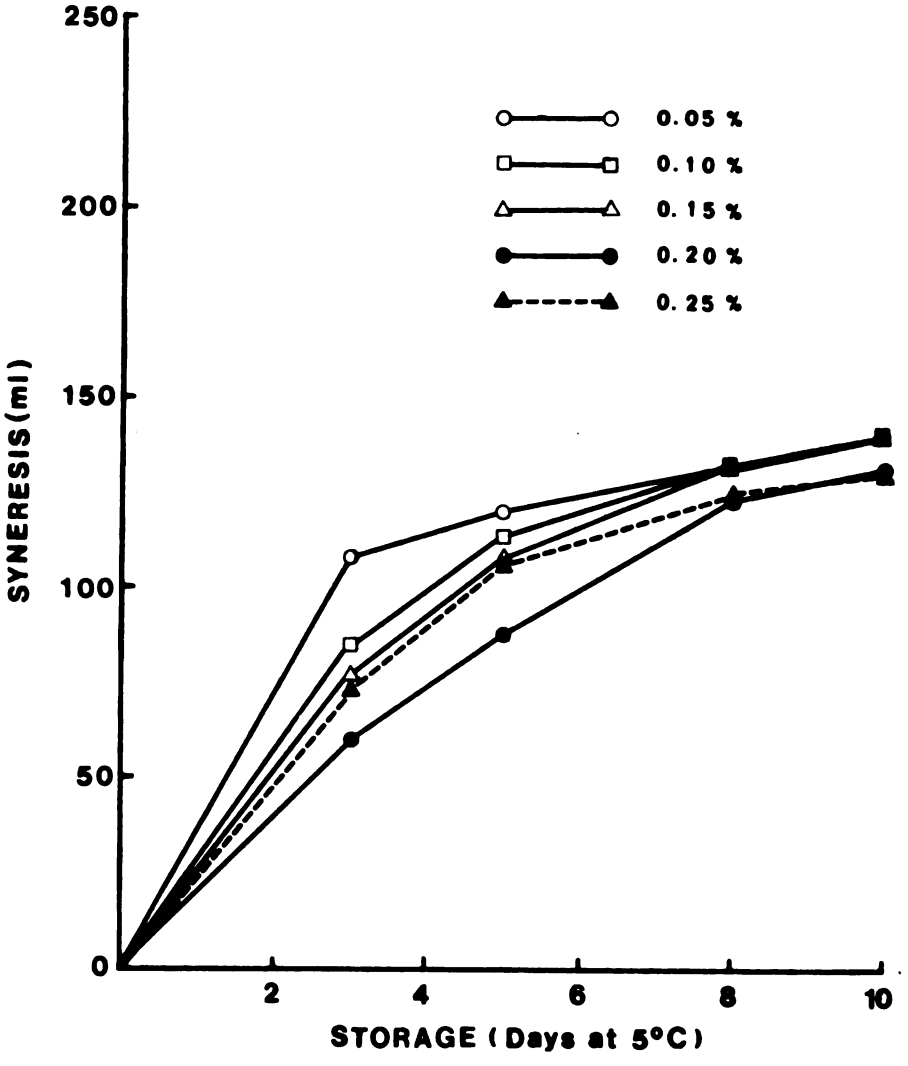

Figure 3. Syneresis volume of yogurt drink samples after storage as a function of different concentrations of carrageenan. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

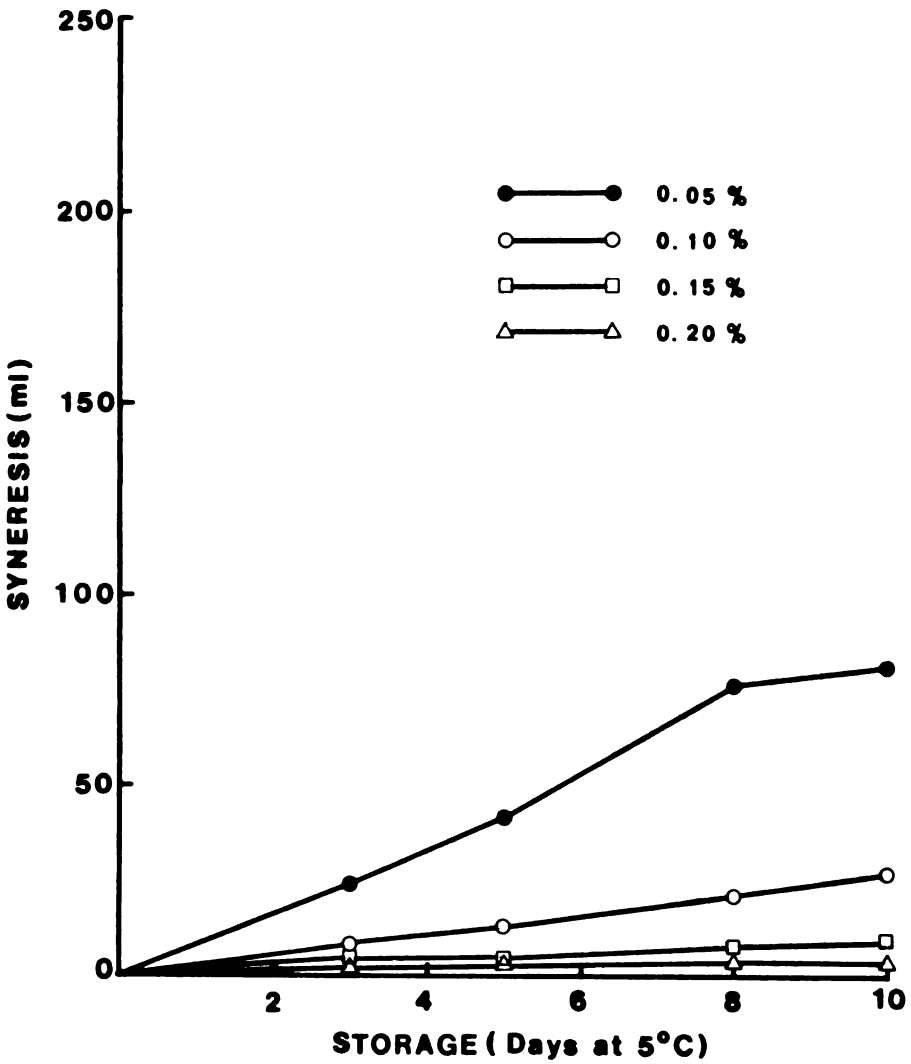

Figure 4. Syneresis volume of yogurt drink samples after storage as a function of different concentrations of guar gum. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

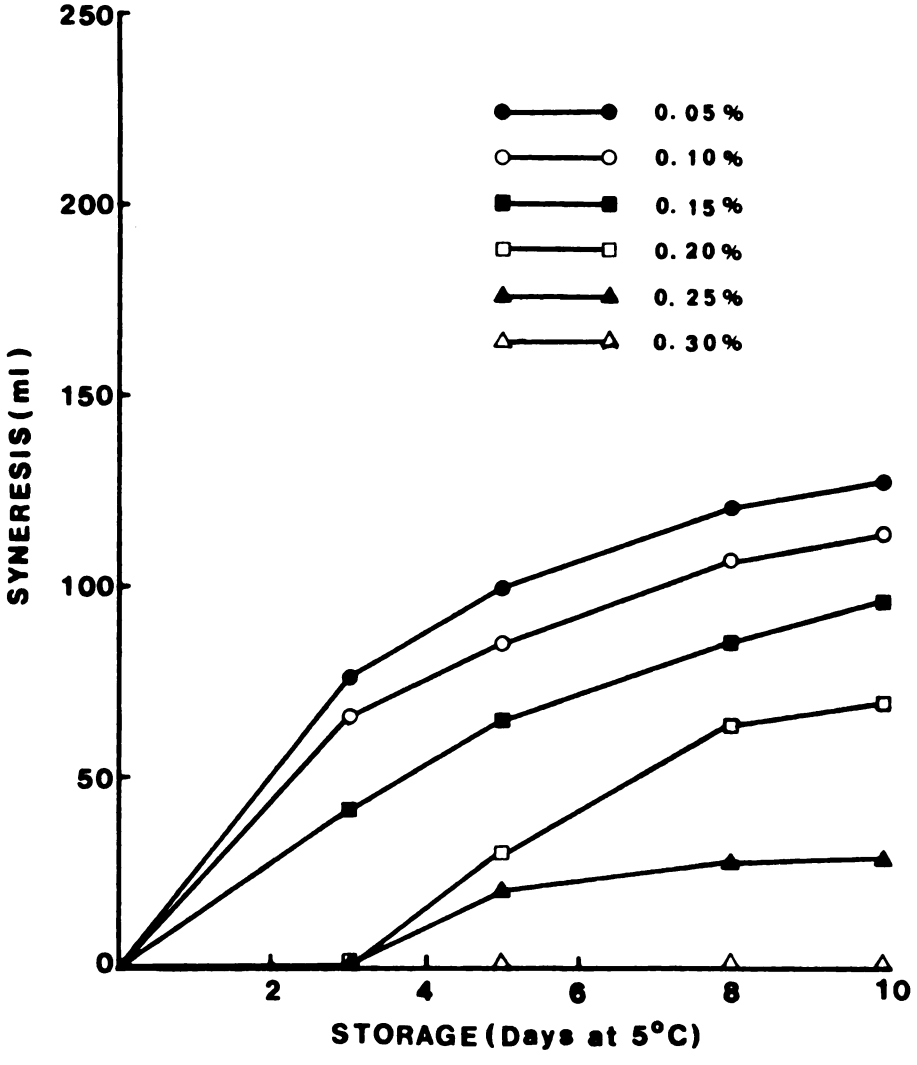

Figure 5. Syneresis volume of yogurt drink samples after storage as a function of different concentrations of commercial proprietary blended stabilizer. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

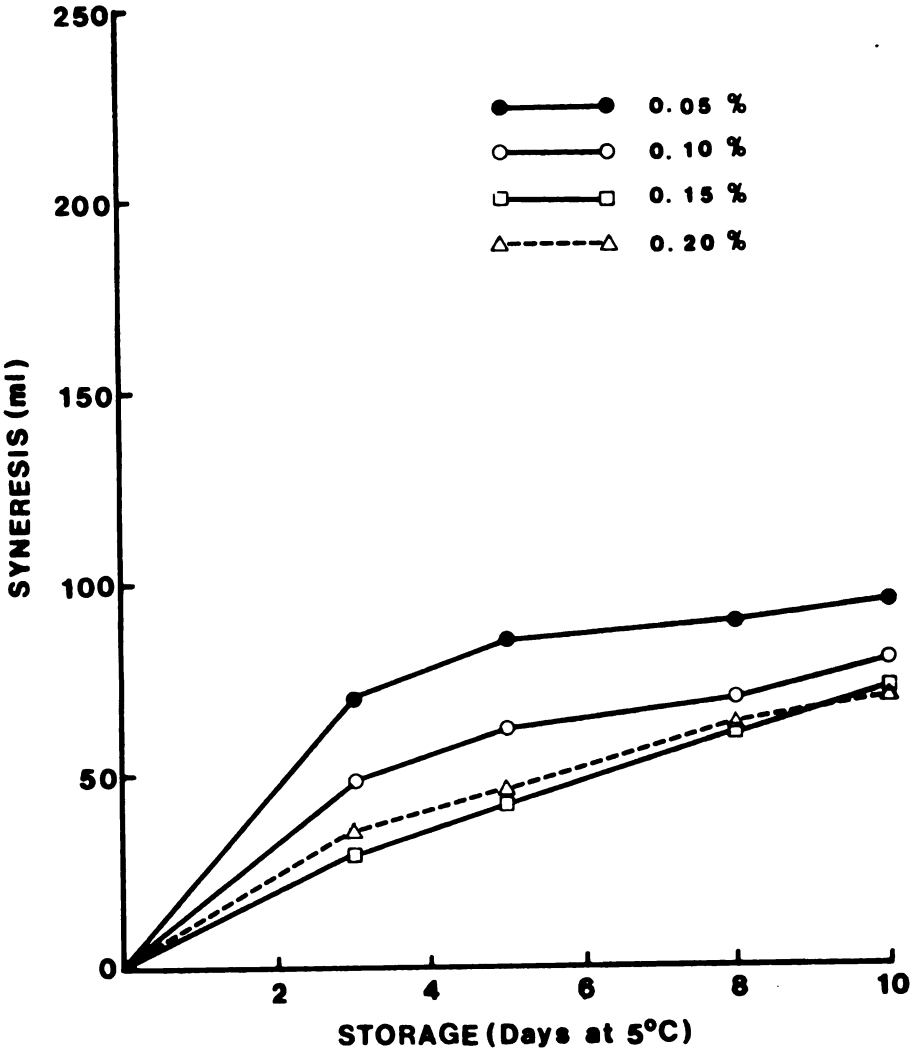

Figure 6. Syneresis volume of yogurt drink samples after storage as a function of different concentrations of propylene glycol alginate. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

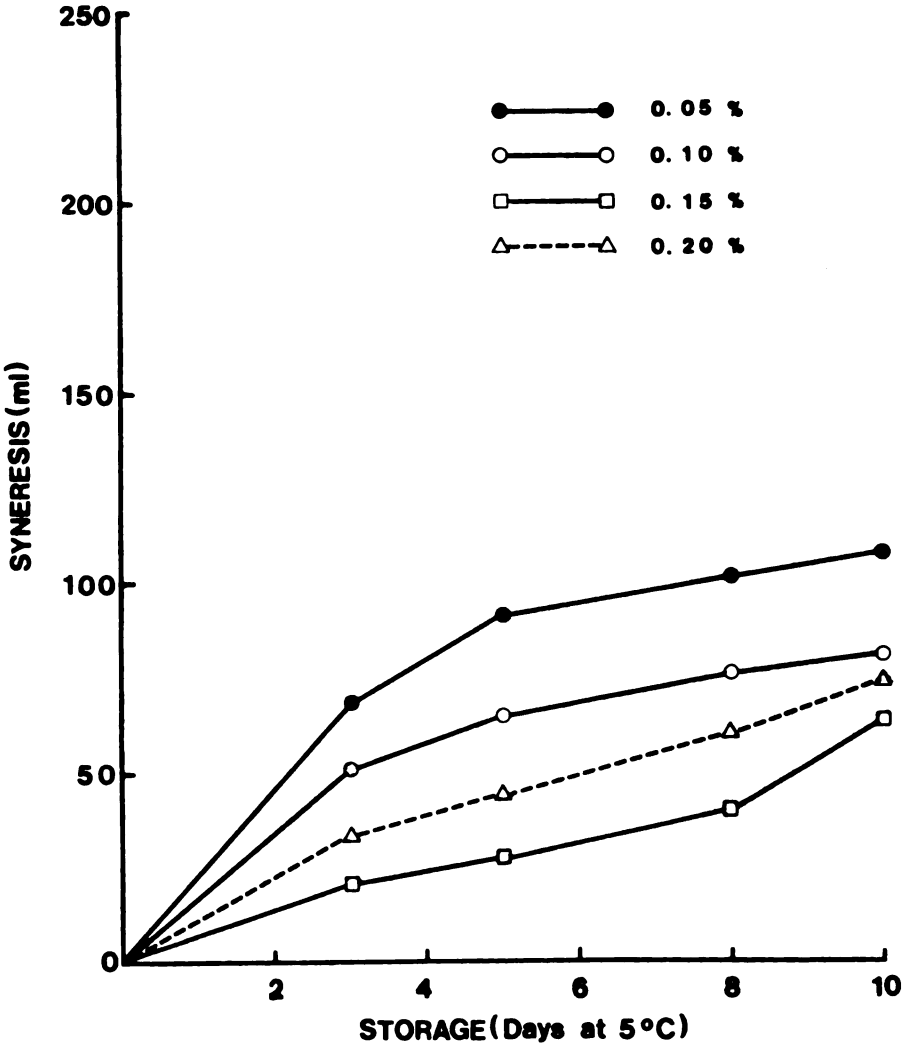

Figure 7. Syneresis volume of yogurt drink samples after storage as a function of different concnetrations of pectin. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

Guar gum (Table 19, Figure 4) was found very effective in stabilizing the system. Stability of the product increased with increase in its concentration until the product was essentially stabilized at the concentration of 0.20%. Notable stabilizing effect was observed when its concentration was raised from 0.05% to 0.10%.

As can be seen from Table 19 and Figure 5, the commercial proprietary blended stabilizer exerted an excellent stabilizing effect on the system. Stability of the product was enhanced increasingly with increase in its concentration until the product was finally stabilized at the concentration of 0.30%.

Propylene glycol alginate (Table 20, Figure 6) and pectin (Table 20, Figure 7) showed almost identical stabilizing pattern with that of carrageenan except that in this case the adverse effect on the product stability was observed at even lower concentration, 0.20%. At this concentration, they also imparted apparent viscosity to the product.

In summary, guar gum and a commercial proprietary blended stabilizer proved to be very effective in stabilizing yogurt drink, while carrageenan, propylene glycol alginate and pectin were less effective. The adverse effect on the stability and increased viscosity of the product caused by the higher conentrations (0.20%

or 0.25%) of those three stabilizers mentioned above indicate that these concentration were excessive for this particular product. Rasic and Kurmann (1978) stated that over-use of a stabilizer in dairy products not only has adverse effect on the stability of the product, but also imparts undesired viscosity to the product. The viscosity cuased may be due to the thickening effect of those stabilizers through their ionic interactions with mono-, di- or trivalent salts in the product. Carrageenan, alginate and pectin are known to have polyanionic nature with a negative surface charge and to form complexes with cationic salts. Guar gum is known to be virtually neutral and non-ionic. Schachat and Raymond (1960) stated that carrageenan forms complexes with potassium and ammonium ions, alginate with calcium and acids and pectin with calcium and sugar, all giving thickening (viscosity promoting) effect. Behavior of the commercial proprietary blended stabilizer can not be fully explained since detailed information on the conentration of each ingredient stabilizer was not available.

Experience has shown that when two or more stabilizers are combined, the stabilizing characteristics of that combination are not predictable. So a trial and error method has been suggested to find a suitable formulation for the stability of a certain food product. Guar gum, being proven to be suitable stabilizer for yogurt drink,

Table 21. [a]Syneresis volume (ml) of yogurt drink samples after storage as a function of stabilizer blends at different concentrations. pH; 4.0, heat treatment; 88°C/2 min, homogenization pressure; 175 kg/cm^2.

Concentration (%) of stabilizers				Storage (Days at 5°C)					
[b]GG	[c]CMC	[d]PGA	Pectin	3	5	8	10	Protein(%)	Remarks (mouthfeel)
0.20	0.05	----	----	4	6	10	12	0.31	Heavy
0.15	0.05	----	----	7	11	18	21	0.31	Heavy
0.15	0.10	----	----	10	15	26	31	0.33	Heavy
----	0.15	----	----	59	71	95	103	0.34	
0.10	----	0.10	----	8	14	17	20	0.36	Heavy
0.10	----	----	0.10	7	11	16	20	0.34	Heavy

[a]Average of duplicate runs

[b]Guar gum

[c]Carboxymethyl cellulose gum

[d]Propylene glycol alginate

was chosen to be tried in combination with CMC, alginate and pectin. The concentration of each stabilizer, syneresis volume of each sample and protein content of the syneresed fluid after storage of 10 days are shown in Table 21. The comparison between the syneresis volumes appeared in Table 21 and those in Table 19 (guar gum only) indicates that the synergistic effect expected was not achieved by these combinations. Same concentration of guar gum caused more syneresis in all combinations tried when it was used with CMC than when it was used alone. And these combinations imparted apparent viscosity to the product. It is likely that the total concentration of these two stabilizers was excessive for this product due to the specific properties contributed by CMC which is also known to form complexes with cationic salts, thereby giving thickening effect. Stability of the product was improved slightly when guar gum was used in combination with alginate and pectin. However, these combinations also imparted viscosity to the product. This result may be due to the specific properties of alginate and pectin discussed previously.

Data presented in Table 19-21 show that there is no correlation between the syneresis volume of the samples and its percentage protein content. The protein contents of all samples are in the range of 0.34±0.06% except those of the samples for which commercial proprietary

blended stabilizer was used. It appears that much larger amounts of proteins were held in the suspension by the interaction with stabilizers even at the lower concentrations and that there were only smaller amount of proteins (which would be interacted by the higher concentrations of stabilizers) and smaller molecular nitrogen compounds such as amino acids and urea in the syneresed fluid. Thus the slightly decreased percentage protein contents at the higher concentrations of stabilizers may be attributable to the interactions of increased stabilizers with the small amounts of exudate proteins not interacted by stabilizers at the lower concentrations. Commercial blended stabilizer resulted in marked increase of exudate protein contents of the samples with increase of its concentration. It is most likely that all the proteins capable of being interacted by this stabilizer were interacted even at the lowest concentration (0.05%) of the stabilizer and the increased concentrations only enhanced its function of maintaining suspension and absorbing free water, thus making the exudate more and more concentrated due to its decreasing volume.

With the two samples which were finally stabilized by the use of guar gum (0.20%) and commercial stabilizer blend respectively, preference (acceptability) test was performed using a nine point hedonic scaling method. It was evident that there was no significant difference in

Table 22. Scores of the two samples finally stabilized by the use of stabilizers at the designated processing parameters[a].

Judges	Stabilizers		Total
	Guar gum (0.20%)	Commercial blended stabilizer (0.30%)	
1	8	8	16
2	9	9	18
3	8	7	15
4	8	8	16
5	9	8	17
6	9	8	17
7	8	8	16
8	7	9	16
9	7	8	15
10	9	8	17
11	8	7	15
12	8	6	14
13	7	7	14
14	9	8	17
15	7	8	15
16	8	8	16
17	9	8	17
18	8	9	17
19	9	6	15
20	4	8	12
21	8	8	16
22	4	7	11
23	8	8	16
24	7	8	15
25	8	8	16
26	6	8	14
Total	200	203	403

[a] pH 4.0
Heat treatment 88°C/2 min
Homogenization pressure 175 kg/cm^2

preference for these two samples since the two total scores given by panelists were almost identical (Table 22). Therefor an analysis of variance was not conducted on these data. The mean values (total score/number of judges) calculated were 7.7 and 7.8 respectively. These values are close to "like very much (8)" on the hedonic scale. Both of the stabilized samples showed excellent acceptability.

SUMMARY AND CONCLUSIONS

The objectives of this research were to establish optimum processing parameters and the concentration of suitable stabilizer(s) for the stability and sensory properties of yogurt drink. Yogurt drink was composed of 25% plain low fat yogurt, 10% frozen concentrated orange juice (4+1), 9% sugar and 56% water. Sedimentation of syneresis in the samples on storage was observed and protein content was determined on the sample supernatant fluid. Subjective assessment of sensory properties was conducted, being given the first priority for choosing optimum processing parameters. Subject to the conditions of this study, the following conclusions were derived:

1. Of the range of pH evaluated, pH 4.0 was the best in regard to flavor and stability of the product. Lower pH (3.2 and 3.6) resulted in excessive sourness. The sample with pH of 4.2 was acceptable, but a little too sweet. The sample with pH of 4.0 was more stable (less sedimentation and more proteins in the supernatant fluid) than the yogurt drink with pH of 4.2.

2. Heat treatment (65^{o}C/30 min, 75^{o}C/10 min, 88^{o}C/2 min and 115^{o}C/4 sec) had negligible effect on the sensory properties of the product. High temperature pasteurization improved stability of the product, which was considerably increased with increasing temperature of heat

treatment although there was little difference between the heat treatments of 65°C/30 min and 75°C/10 min.

3. Homogenization pressure (70 kg/cm^2, 105 kg/cm^2 and 175 kg/cm^2) had little effect on the sensory properties of the product. Low pressure homogenization favored stabilization of the product. Product stability was significantly decreased with increasing homogenization pressure.

4. Among the stabilizers used, carrageenan, alginate and pectin were ineffective in stabilizing the product under the conditions used in this project. When these stabilizers were used, the stability of the system dimished markedly with time and high concentrations (0.20% or 0.25%) imparted too much viscosity to the product.

Guar gum and a commercial proprietary blended stabilizer were very effective in stabilizing the system. Stability of the product increased with increasing concentration of these stabilizers until the product was finally stabilized at the concentrations of 0.20% and 0.30% respectively.

The use of guar gum in combination with alginate, pectin and CMC was not effective in stabilizing this system. The synergistic effect of the stabilizers was not observed in any combinations studied.

5. Both samples which were finally stabilized using guar gum (0.20%) and a commercial blended stabilizer (0.30%) respectively showed excellent acceptability in taste panel evaluation, the mean values of both samples being close to 8 (like very much) on the nine point hedonic scale.

APPENDIX

APPENDIX

Statistical Charts

Table 23. Rank totals required for significance at the 5% level ($P < .05$)

No. of Reps.	Number of treatments						
	2	3	4	5	6	7	8
2	-----	-----	-----	-----	-----	-----	-----
	-----	-----	-----	3-8	3-11	3-13	4-14
3	-----	-----	-----	4-14	4-17	4-20	4-23
	-----	4-8	4-11	5-13	8-15	8-18	7-20
4	-----	5-11	5-15	6-18	6-22	7-25	7-29
	-----	5-11	6-14	7-17	8-20	9-29	10-26
5	-----	6-14	7-18	8-22	9-26	9-31	10-35
	6-9	7-13	8-17	10-20	11-24	13-27	14-31
6	7-11	8-16	9-21	10-26	11-31	12-36	13-41
	7-11	9-15	11-19	12-24	14-28	16-32	18-36
7	8-13	10-18	11-24	12-30	14-35	15-41	17-46
	8-13	10-18	13-22	15-27	17-32	19-37	22-41
8	9-15	11-21	13-27	15-33	17-39	18-46	20-52
	10-14	12-20	15-25	17-31	20-36	23-41	25-47
9	11-18	13-23	15-30	17-37	19-44	22-50	24-57
	11-16	14-22	17-28	20-34	23-40	28-46	29-52
10	12-18	15-25	17-33	20-40	22-48	25-55	27-63
	12-18	16-24	19-31	23-37	26-44	30-50	33-57

Table 23 (cont'd.)

No. of Reps.	Number of treatments						
	2	3	4	5	6	7	8
11	13-20	16-28	19-36	22-44	25-52	28-60	31-68
	14-18	18-26	21-34	25-41	29-48	33-55	37-82
12	15-21	18-30	21-39	25-47	28-56	31-65	34-74
	15-21	18-29	24-36	28-44	32-52	37-59	41-67
13	16-23	20-32	24-41	27-51	31-60	35-69	38-79
	17-22	21-31	26-39	31-47	35-56	40-64	45-72
14	17-25	22-34	26-44	30-54	34-64	38-74	42-84
	18-24	23-33	28-42	33-51	38-60	44-68	49-77
15	19-26	23-37	38-47	32-58	37-68	41-79	46-89
	19-26	25-35	30-45	36-54	42-83	47-73	53-82
16	20-28	25-39	30-50	35-81	40-72	45-83	49-95
	29-27	27-37	33-47	39-57	45-67	51-77	57-87
17	22-29	27-41	32-53	38-64	43-76	48-88	53-100
	22-29	28-40	35-50	41-61	48-71	54-82	61-92
18	23-31	29-43	34-56	40-88	46-80	51-93	57-105
	24-30	30-42	37-53	44-64	51-75	58-86	65-97

Table 24. Variance ratio-5 percent points for distribution of F

b_{n_2} \ a_{n_1}	1	2	3	4	5	6	8
1	161.4	199.5	215.7	224.6	230.2	234.0	238.9
2	18.51	19.00	18.25	19.25	19.30	19.33	19.37
3	10.13	9.55	9.28	9.12	9.01	8.94	8.84
4	7.71	6.94	6.59	6.39	6.26	6.16	6.04
5	6.61	5.79	5.41	5.19	5.05	9.95	4.82
6	5.99	5.14	4.76	4.53	4.39	4.28	4.15
7	5.59	4.74	4.35	4.12	3.97	3.87	3.73
8	5.32	4.46	4.07	3.84	3.69	3.58	3.44
9	5.12	4.26	3.86	3.83	3.48	3.37	3.23
10	4.96	4.10	3.71	3.48	3.33	3.22	3.07
11	4.84	3.98	3.59	3.36	3.20	3.09	2.95
12	4.75	3.88	3.49	3.28	3.11	3.00	2.85
13	4.67	3.80	3.41	3.18	3.02	2.92	2.77
14	4.60	3.74	3.34	3.11	2.96	2.85	2.70
15	4.54	3.68	3.29	3.06	2.90	2.79	2.64
16	4.49	3.63	3.24	3.01	2.85	2.74	2.59
17	4.45	3.59	3.20	2.96	2.81	2.70	2.55
18	4.41	3.55	3.16	2.93	2.77	2.68	2.51
19	4.38	3.52	3.13	2.90	2.74	2.63	2.48
20	4.35	3.49	3.10	2.87	2.71	2.60	2.45
21	4.32	3.47	3.07	2.84	2.68	2.57	2.42
22	4.30	3.44	3.05	2.82	2.66	2.55	2.40
23	4.28	3.42	3.03	2.80	2.64	2.53	2.38
24	4.26	3.40	3.01	2.78	2.62	2.51	2.36
25	4.24	3.38	2.99	2.76	2.60	2.49	2.34
26	4.22	3.37	2.98	2.74	2.59	2.47	2.32
27	4.21	3.35	2.96	2.73	2.57	2.46	2.30
28	4.20	3.34	2.95	2.71	2.56	2.44	2.29
29	4.18	3.33	2.93	2.70	2.54	2.43	2.28
30	4.17	3.32	2.92	2.69	2.53	2.42	2.27

Table 24 (cont'd.)

b_{n_2} \ a_{n_1}	1	2	3	4	5	6	8
40	4.08	3.23	2.84	2.61	2.45	2.34	2.18
60	4.00	3.15	2.76	2.52	2.37	2.25	2.10
120	3.92	3.07	2.68	2.45	2.29	2.17	2.02
∞	3.84	2.99	2.60	2.37	2.21	2.09	1.94

[a]Degrees of freedom for numerator

[b]Degrees of freedom for denominator

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