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Effects of Spices on the Growth of Lactic Acid Bacteria

by

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香辛料가 乳酸菌의 增殖에 미치는 影響

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Abstract

This study was undertaken in order to examine the effect of ginger, garlic and red pepper, usually used as the ingredients of Kimchi, on the growth and acid production of *Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* during incubation in the medium containing different amount of each extract.

The results obtained are as follows:

1. The effect of ginger extract
   a. The growth of *Lact.plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* tended to be stimulated as the amount of extract added into the medium increased in a certain amount (3.64 mg soluble solid/ml).
   b. The pH of the culture medium *of Lact. plantarum ATCC 8014* became lower to some degree and acid production tended to be stimulated but acid production of *Lact. fermenti ATCC 9338* tended to be suppressed as the amount of extract added in a certain amount (3.64mg soluble solid/ml) increased.
2. The effect of garlic extract
   a. The growth of *Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* tended to be stim ulated as the amount of extract added in a certain amount (31.7 mg soluble solid/ml) increased.
   b. The acid production of *Lact plantarum ATCC 8014* was suppressed ($p<0.01$) and drop in pH was suppressed ($p<0.05$) as the amount of extract added in a certain amount increased. In case of *Lact. fermenti ATCC 9338*, the acid production t ended to be suppressed also.
3. The effect of red pepper extract
   a. The growth of *Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* tended to be stimulated as the amount of extract added in a certain amount (14.5 mg soluble solid/ml) increased.

b. The acid production of *Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* tended to be stimulated as the amount of extract added in a certain amount (14.5 mg soluble solid/ml) increased.

Introduction

Lactic acid bacteria are cocci or rods which are div ided into two groups, Homo- or Hetero-fermentative lactic acid bacteria according to their fermentation patterns and have high nutritional requirements.

Avery (1881) was the first person to produce lactic acid successfully on a commercial basis. Since then, a series of effort has been made to substitute calcium lactate for the tartarate and lactic acid production by fermentation has become a very important industry (Prescott et al. 1959).[1]

Since it was shown that addition of vegetable extract to synthetic medium for lactic acid bacteria stimulates the growth of them(Mickle at al. 1924),[2] there are so many studies to sound the occurence of growth stimulants of lactic acid bacteria from tomato (Mickle et al. 1925,[3] Kulp et al. 1927[4]), carrot (Garley et al. 1941)[5] and orange (Murdock et al. 1952).[6]

Kuiken et al. (1943)[7] reported the stimulant of lactic acid bacteria in tomato juice and made clear it is acid and heat stable substance.

Also, Metcalf et al. (1946),[8] Colio et al. (1948)[9] and Snell et al.(1948)[10] reported the lactic acid bacteria growth stimulants from cabbage, red pepper and spinach, which are alkali-labile, malt, and asparagus respectively.

Recent studies made clear the occurence of growth stimulating substances but could not turn out what substances they are, only leaving the possibility that they might be such vitamin group as thiamine, vitamin B_6 and vitamin B_{12} (Shord 1948,[11] Hoffman et al. 1949[12]) or their analogous substances and thymine, deoxyribose, purine, pyimidine(JRI Soska 1956[13]) and N-acetyl-D-glucosamine (Lambert et al. 1965[14]) or their analogous substances.

There are several reports on effects of inorganic ions such as Mn^{++}, Mg^{++}(Stamer et al. 1964,[15] Nowakowsk et al. 1965,[16] Macleod et al. 1947[17]).

Kim et al. (1968)[18] reported that addition of 1% cucumber as a component of Kimchi accelerates the fermentation and alcohol extract of cucumber stimulates the growth of *Lactobacillus plantarum* but this effect is disappeared by the treatment of dialysis or heating at 120°C for 10 minutes.

Nishiyama et al.(1965,[19] 1969, a[20], b[21]) carried out a series of study on stimulants monitoring the reference lactic acid bacteria,obtaining the result that growth stimulants in Kuko are amino acids and sub stances analogous to nucleic acids.

Yang Lin et al.(1966)[22] reported the growth factors of *Lact. bulgaricus* G.S. are widely distributed in natural materials from animal tissues to bacterial cells and the speics has two growth factors(PF,FF), of which PF is essential growth factor and FF is non-essential growth factor but increases the growth.

Weinman et al. (1963)[23] studied on the effect of liver extract, *Torula* yeast extract, RNA digest and DNA digest on the growth of lactic acid bacteria.

Auclair et al. (1957)[24] indicated as a growth factor formic acid which is considered to be generated by decomposition of lactose when autoclaving.

Zofia Tynecka et al. (1973)[25] stated that garlic extract inhibited the growth of *Staphylococcus aureus, Candida albicans* but has a little effect against *Escherichia coli*, corelating the inhibitory factors with allicin concentration.

Fletcher et al.(1974)[26] reported that garlic extract (1 mg soluble solid/ml) inhibited the coagulase activity and 1.4mg of soluble solid/ml decreased the growth of *Staph. aureus*, whereas 5.6 mg of soluble solid/ml completely inhibited the growth.

Munekata et al.(1973)[27] reported the inhibition of lactic acid bacteria in beer and sake, Shankman et al.[28](1960) of *Pediococcus cerevisiae and Lact. plantarum* by lower peptides, Kulshrestha et al.(1974, a[29] b,[30] c,[31] d[32]) of *E. coli, Streptococcus lactis, Leuconostoc citrovorum and Str. thermophilus* by volatile and non-volatile compounds.

As many investigations have reported, there are

many growth inhibitors and stimulants of lactic acid bacteria in the nature and many reports have been compiled about the growth of lactic acid bacteria.

Ginger, garlic and red pepper are important ingredients of Korean Kimchi, which is fermented by lactic acid bacteria. Major lactic acid bacteria, associated with Kimchi fermentation, are *Lact. plantarum, Lact. fermenti, Lact. brevis, Str. faecalis, Leu. mesenteroides* (Kim et al. 1959[38]). Many reports on the growth of lactic acid bacteria were compiled but few of them covered on the influence of ginger, garlic and red pepper which might be related to the speed of fermentation of Kimchi in connection with the growth of lactic acid bacteria.

The purpose of the present study was to investigate the effect of different amount of each extract addition to media and whether these extacts stimulate the growth of lactic acid bacteria or inhibit.

Materials and Methods

1. Raw Materials

In this investigation, ginger, garlic and red peper were tested,which were purchased from market. Their moisture contents were 60.6%, 80.5% and 13.5% respectively. All extracts of these materials were prepared according to the technique described by Fletcher (1974)[26] with a little modification.

a) Ginger

Ginger (500 g) was blended with 500 ml distilled water for 5 minutes at high speed in a waring blender, the mixture filtered through 5 thicknesses of gauze, the filtrate heated for 5 minutes at 80°C to inactivate various enzymes, fitered with filter paper (Whatman No. 1, grade 17, size 12 cm, W& R Balston Ltd.) and centrifued with a high speed centrifuger(MSE type high speed 18) at the speed of 10,000 rpm for 30 minutes.

b) Garlic

Garlic extract was prepared by the same method as ginger extract.

c) Red pepper

Red pepper (200 g) was blended with 1 L distilled water, soaked at 65°C for 40 hours, filterd, heated and centrifuged as ginger.

All extracts after centrifugation were refiltered with membrane filter(Oxoid, Nuflow membrane filter, size 6 cm, pore size: 0.4mμ, U.K.) to get bacteria free extract. The filtered extract was bottled aseptically and refrigerated at 4°C.

d) Dry weight determination of extracts

Dry weight of the extract was determined by drying 5ml extract at 105°c.

e) Confirmation of bacteria-free in extract

In order to examine the living cell-free of the extract, this test was conducted before being used. Standard plate plate count method(Thatcher et al. 1968)[34] was introduced to inoculate 1 ml of the extract to duplicate petridishes, incubating at 35°C for 2 days. After incubation,each plate was examined whether the extract was contaminated or not. Tryptone glucose yeast agar (Oxoid) was used for this test.

2. Culture Strains

Lact. Plantarum ATCC 9338 were used as reference lactic acid bacteria, which were obtained from ATCC and preserved in Food Research Institute, Agriculture and Fishery Development Corporation.

3. Media

Lactobacilli MRS broth (DIFCO) (Deman et al. 1960)[35] was used in this study and its composition was as Table 1 and 2.

4. Extracts Addition and Incubation

For the determination of the changes in pH and

Table 1. The composition of medium A for control, 0.1 ml and 0.5ml extract addition lot(Lactobacilli MRS Broth)

Bacto proteose peptone #3	: 10 g
Bacto beef extract	: 10 g
Bacto yeast extract	: 5 g
Dextrose	: 20 g
Tween 80	: 1 g
Ammonium citrate	: 2 g
Sodium acetate	: 5 g
Magnesium sulfate	: 0.1 g
Manganese sulfate	: 0.05g
Disodium phosphate	: 2 g
Distilled water	: tol L
pH	: 6.0

Table 2. The composition of medium B for 2 ml extract addition lot

Bacto proteose peptone #3	: 10 g
Bacto beef extract	: 10 g
Bacto yeast extract	: 5 g
Dextrose	: 20 g
Tween80	: 1 g
Ammonium citrate	: 2 g
Sodium acetate	: 5 g
Magnesium sulfate	: 0.1g
Manganese sulfate	: 0.05g
Disodium phosphate	: 2 g
Distilled water	: to 800ml
pH	: 6.0

acidity during incubation period, 10 ml of medium A for control, 0.1 ml extract addition lot (T_1), 0.5 ml extract addition lot (T_2) and 8 ml of medium B for 2 ml extract addition lot(T_3) were dispensed into sterile cottonplugged test tubes and autoclaved at 121 °C for 15 minutes respectively. 0.1 ml or 0.5 ml of each extract was added into 10 ml of medium A except for the control lot and 2 ml of each extract was added into 8 ml of medium B.

For the turbidimetrical test of the changes in growth, 5 ml of medium A for control, 0.1 ml, 0.5 ml extract addition lot and 4 ml of medium B for the 2 ml addition lot were dispensed into sterile cotton-plugged cell of spectronic 20 and autoclaved at 121°C for 15 minutes respectively. 0.05 ml or 0.25 ml of each extract was added into 5 ml of medium A except for the control and 1 ml of each extract was added into 4 ml of medium B.

All media after autoclaving were filled up to orignal volume with sterile distilled water to correct the loss during sterilization.

Inoculum was prepared by inoculating one loop of each strain from stab culture into test tube of 15 ml lactobacilli MRS broth(Medium A) which was cotton-plugged and autoclaved at 121°Cfor 15 minutes and then incubated at 25°C for2 days. Two loops of this culture were diuted with 10ml of 0.1% peptone dilution fluid and inoculated 0,4ml of diluted culture into each tube. Incubation was conducted at 25°C for 3 days.

5. Turbidity Determination

Turbity of culture was checked after shaking the precipitated cell by using Beckman spectronic 20 spectrophotometer and designated as optical density at the wavelength of 520nm.

6. Others

a) Titratable acidity

Acidity was determined by titrating 2ml of liquid culture with 0.1 N-NaOH solution to pH 8.1 using Fisher Titrimeter and expressed as the volume of 0.1 N-NaOH solution required.

b) pH

pH was checked with Beckman expandomatic SS-2 pH meter.

Results

1. The effect of ginger extract on the growth of *Lact. plantarum* ATCC 8014

As shown in Fig. 1, no difference was found in growth among each lot during 4 hours after inoculation. The growth was stimulated as the extract addition increased with 0.5 ml lot showing no difference from 2 ml lot at the early stage of incubation period from 4 hours to 54 hours, whereas 0.1 ml extract addition lot showed no difference from control.

Statistical analysis showed high significant difference at 1% level during the incubation period from 30 hours to 54 hours as shown in Table 3. Significant difference was found between control-0.1 ml extract addition lot, control-0.5 ml extract addition lot, con-

Table 3. Analysis of varience and Duncan multiple range test

Source	D.F.	S.S.	M.S.	F
Total	22	0.1889	—	—
Treatment	3	0.1096	0.0365	11.0606**
Error	24	0.0793	0.0033	

**Significant at 1% level.

Treatment	Control	T_1	T_2	T_3
Average	1.7929	1.8629	1.8786	1.9686
Comment				

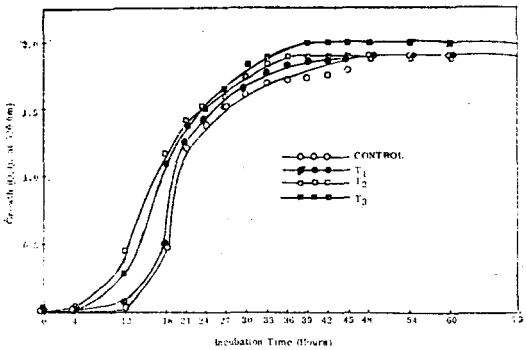

Fig. 1 Effect of ginger extract on the growth of *Lact. plantarum ATCC* 8014

trol. 2 ml extract addition lot, 0.5 ml extract addition lot-2 ml extract addition lot but no significant difference was appeared between 0.1 ml extractd adition lot-0.5 ml extract addition lot.

2. The effect of garlic extract on the growth of *Lact. plantarum ATCC* 8014

The results of the effect of garlic extract on the growth of the strain were shown in Fig. 2. No diffe rence in growth among each lot was appeared during 12 hours after inoculation. During the incubation period from 12 hours to 30 hours, 0.5 ml extract addition lot and 2 ml extract addition lot showed the inhibition in growth, whereas 0.1 ml extract addition lot had slight stimulation in growth. Extract addition lots gradually became stimulated in growth with 2 ml extract addition lot showing superiority in growth at the final stage of incubation period but 0.1 ml extract addition lot and 0.5 ml extract addition lot showed slight difference from control. As the result of statistical analysis,no significant difference was found among each lot at 5% level.

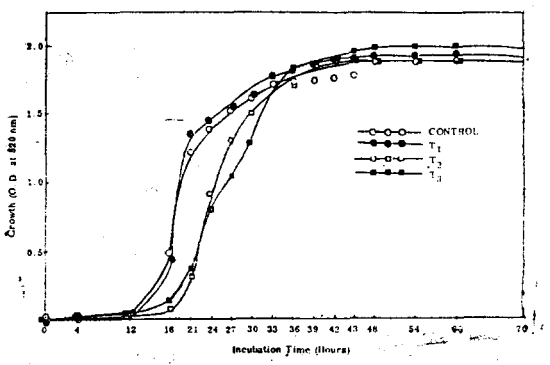

Fig. 2. Effect of garlic extract on the growth of *Lact. plantarum ATCC* 8014

3. The effect of red-pepper extract on the growth of *Lact. plantarum ATCC* 8014

The results of the effct of red pepper extract on the growth of the strain were shown in Fig. 3.

As shown in Fig. 3, extract addition lots tended to be stimulated in growth comparing with control to have considerable growth stimulation with the lapse of incubation time but much difference was not found among 0.1 ml extract addition lot, 0.5 ml extract addition lot and 2 ml extract addition lot.

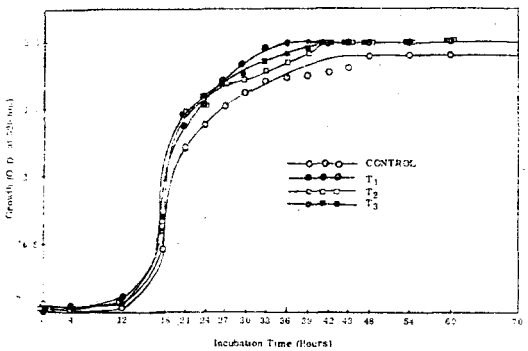

Fig. 3. Effect of red pepper extract on the growth of *Lact. plantarum ATCC* 8014

Statistical analysis showed high significant difference during the incudation period from 30 hours to 48 hours at 1% level as shown in Tadle 4. There was sibnificant difference getween 0.1 ml extract addition lot-control, 0.5 ml extract addition lot-control, 2 ml extract addition lot-control but no significant difference was found among 0.1 ml extract addition lot, 0.5 ml extract addition lot and 2 ml extract addition lot.

Table 4. Analysis of varience and Duncan multiple rangetest

Source	D.F.	S.S.	M. S.	F
Total	23	0.2408	—	—
Treatment	3	0.1606	0.0535	13.3416**
Error	20	0.0902	0.0040	

** Significant at 1% level.

Treatment	Control	T_2	T_3	T_1
Average	1.7750	1.9383	1.9483	1.9900
Comment				

4. The effect of ginger extract on the acid production and pH of *Lact. plantarum ATCC* 8014

The results of the effect of ginger extract on the acid production and pH of the strain were shown in Fig. 4.

In pH, no difference among each lot during 6 hours after inoculation was found. Comparing with control, 0.5 ml extract addition lot and 2 ml extract addition lot showed a drop in pH but 0.1 ml extract addition lot had higher pH value than control.

In the aspect of acid production, slight deviaton among each lot was appeared even at the early phase after inoculation. 2 ml extraet addition lot tended to be stimulated in acid production but during incubati-

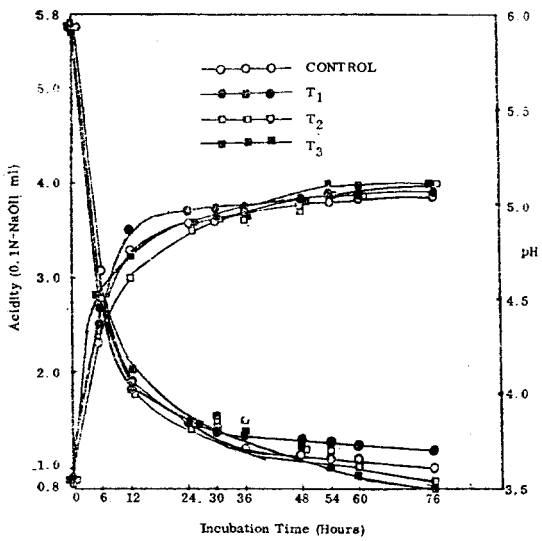

Fig. 4. Effect of ginger extract on the acid production and pH of media inoculated with *Lact. plantarum ATCC* 8014

on period from 10 hours to 40 hours, 0.1 ml extract addition lot showed higher acid production with 0.5 ml extract addition lot and 2ml extract addition lot having lower acid production than control on the contrary. Final acid production tended to be stimulated as the extract addition increased.

Statistical analysis explained there was no significant difference among each lot at 5% level.

5. The effect of garlic extract on the acid production and pH of *Lact. plantarum ATCC* 8014

The results of the effect of garlic extract on the acid production and pH of the strain were expressed in Fig. 5.

In pH, no difference was found among each lot at the early phase after inoculation as shown in Fig. 5. Extract addition lots gradually showed difference from control. 0.1 ml extract addition lot and 0.5 ml extract addition lot had higher pH value than control with 2 ml extract addition lot inhibited in drop of pH exceptionally. F-test showed there was significant difference at 5% level as indicated in Table 5 but no significant difference was recognized among control, 0.1 ml extract addition lot and 0.5 ml extract addit-

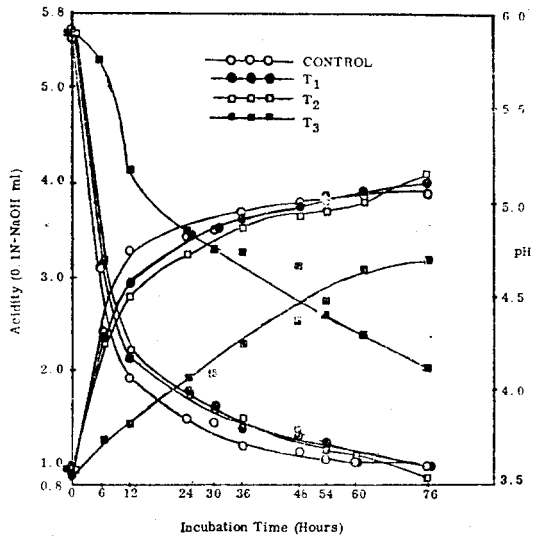

Fig. 5. Effect of garlic extract on the acid production and pH of media inoculated with *Lact. plantarum ATCC* 8014

Table 5. Analysis of varience and Duncan multiple range test

Source	D.F.	S.S.	M.S.	F
Total	39	21.5847	—	—
Treatment	3	4.7008	1.5669	3.3400*
Error	36	16.8839	0.4690	

* Significant at 5% level.

Treatment	Contol	T_2	T_1	T_3
Average	4.055	4.102	4.106	4.878
Comment				

Table 6. Analysis of varience and Duncan multiple range test

Source	D.F.	S.S.	M.S.	F
Total	39	17.3902	—	—
Treatment	3	8.1047	2.7016	10.4741**
Error	36	9.2855	0.2579	

** Significant at 1% level.

Treatment	Control	T_2	T_1	T_3
Average	2.166	3.121	3.209	3.266
Comment				

ion lot.

In acid production, there was slight difference among each lot from the early phase of incubation period with extract addition lots inhibited as the extract addition increased. After this period, 0.1 ml extract addition lot and 0.5 ml extract addition lot had slight stimulation but 2 ml extract addition lot was inhibited outstandingly comparing with control.

As shown in Table 6, there was significant difference at 1% level with Duncan's multiple range test showing no significant difference among control, 0.1 ml extract addition lot and 0.5 ml extract addition lot.

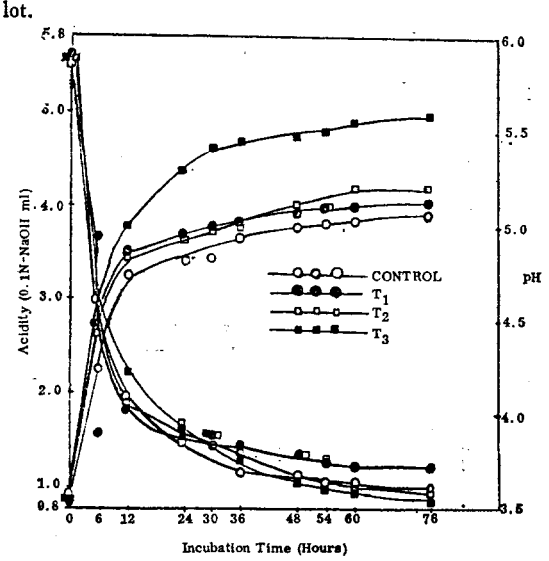

Fig. 6. Effect of red pepper extract on the acid production and pH of media inoculated with *Lact. plantarum ATCC* 8014

6. The effect of red pepper extract on the acid production and pH of *Lact. plantarum ATCC* 8014

The results of the effect of red pepper extract on the acid production and pH of the strain were shown in Fig. 6. Each lot did not have outstandingly different pH value during incubation period, but in the aspect of final pH, 0.5 ml extract addition lot and 2 ml extract addition lot had lower pH value than control. In acid production, much difference was not appeared among each lot at the early phase after inoculation. After the incubation period of 12 hours, difference was gradually increased as the extract addi tion became higher, Significant difference at 5% level was not recognized both in pH and acid production.

7. The effect of ginger extract on the growth of *Lact. fermenti ATCC* 9338

Fig. 7 showed the effect of ginger extract on the growth of the strain. No difference in growth among each lot was found during 12 hours after inoculation. After this incubation period, difference was appeared among each lot with 2 ml extract addition lot showing considerable stimulation in growth but much diff erence was not found among control, 0.1 ml extract addition lot and 0.5 ml extract addition lot. Generally growth tended to be increased as the extract addition became higher.

Statistical analysis showed there was significant difference at 1% level after the incubation period of 30 hours, as shown in Table 7, with showing no significant difference among control, 0.1 ml extract addition lot and 0.5 ml extract additon lot.

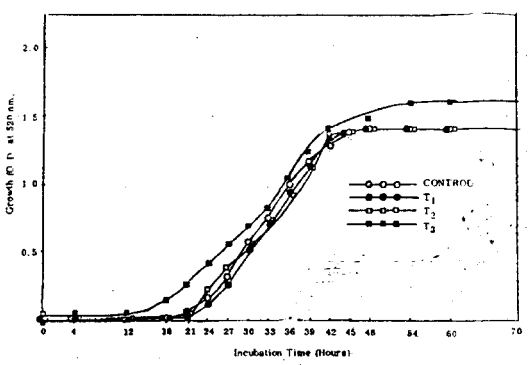

Fig. 7. Effect of ginger extract on the growth of *Lact. fermenti ATCC* 9338

Tbale 7. Analysis of varience and Duncan multiple range test

Source	D.F.	S.S.	M.S.	F
Total	35	0.1631	—	—
Treatment	3	0.0774	0.0258	9.5556**
Error	32	0.0857	—	—

** Significant at 1% level.

Treatment	Control	T_1	T_2	T_3
Average	1.2456	1.2356	1.2356	1.3456
Comment				

8. The effect of garlic extract on the growth of *Lact. fermenti ATCC* 9338

Fig. 8 showed the effect of garlic extract on the growth of the strain. At the early phase of incubation, there was no difference in growth among each lot. After the incubation time of 12 hours, the growth of 0.5 ml extract addition lot and 2 ml addition lot tended to de stimulated as the extract addition increased with 0.1 ml extract having the similar growth to control. After the incubation period of 45 hours, 0.5 ml extract addition lot had similar growth to control also. There was no significant difference at 5% level according to the statistical analysis.

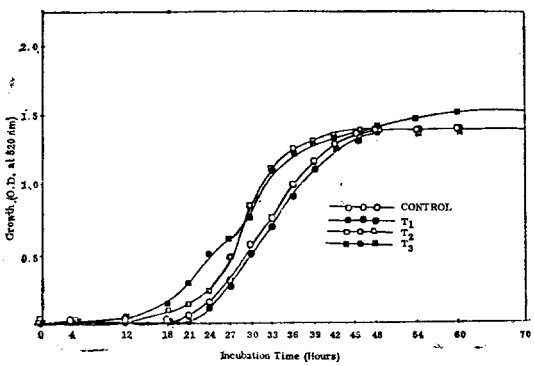

Fig. 8. Effect of garlic extract on the growth of *Lact. fermenti ATCC* 9338

9. The effect of red pepper extract on the growth of *Lact. fermenti ATCC* 9338

The results of the effect of red pepper extract on the growth of the strain were show in Fig. 9. Deviation in growth among each lot was appeared even at the early phase of incubation with 2 ml extract addition lot having the most remarkable growth. At the incubation period from 21 hours to 42 hours, 0.5 ml extract addition lot had the highest growth among 4 treatments but 2 ml extract addition lot had superior stimulation of growth to other lots again after this time.

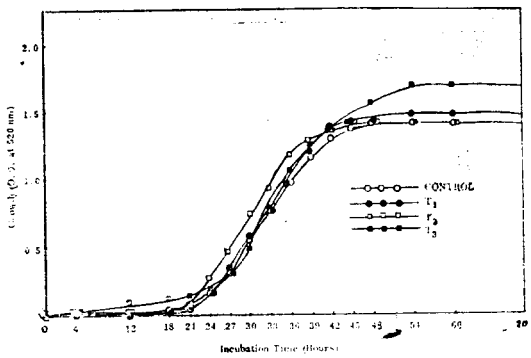

Fig. 9. Effect of red pepper extract on the growth of *Lact. fermenti ATCC* 9338

Statistical analysis showed significant difference from inoculation to 18 hours at 5% level as shown in Table 8, but no significant difference was recognized among each lot.

Table 8. Analysis of varience and Duncan multiple range test

Sourde	D.F.	S.S.	M.S.	F
Total	15	0.0272	—	—
Treatment	3	0.0131	0.0044	3.6667*
Error	12	0.0141	0.0012	

* Significant at 5% level.

Treatment	T_1	T_2	Control	T_3
Average	0.0100	0.0150	0.0125	0.0550
Comment				

10. The effect of ginger extract on the acid production and pH of *Lact. fermenti ATCC* 9338

The results of the effect of ginger extract on the acid production and pH of the strain were shown in Fig. 10.

In pH, there was difference between control and extract addition lot, with extract addition lots showing

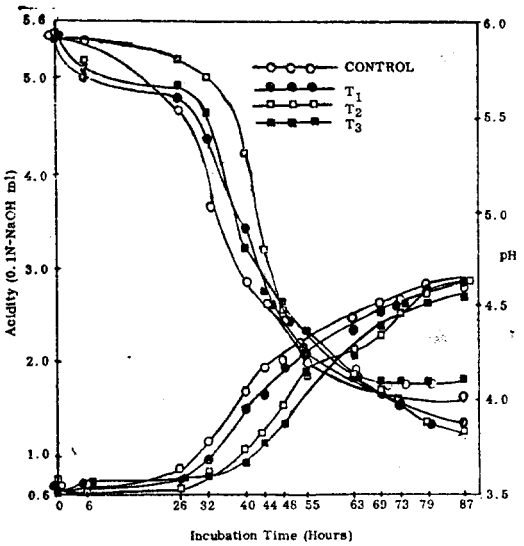

Fig. 10. Effect of ginger extract on the acid production and pH of media inoculated with *Lact. fermenti ATCC* 9338

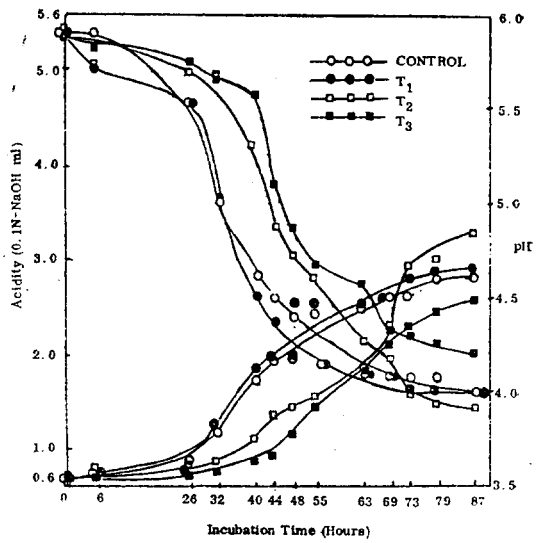

Fig. 11. Effect of garlic extract on the acid production and pH of media inoculated with *Lact. fermenti ATCC* 933

inhibition of drop in pH comparing with control. 0.1 ml extract addition lot and 2 ml extract addition lot showed similar inhibition of drop in pH each other with 0.5 ml extract addition lot showing highest inhibition. From the incubation period of 48 hours, 0.1 ml extract addition lot and 2 ml extract addition lot showed a sudden drop in pH and final pH became lower than control.

In acid production the inhibition was increased as the extract addition became higher. 2 ml extract addition lot had the least amount of acid production but each lots had similar acid production at the end phase of incubation period.

Statistical analysis showed no significant difference at 5% level.

11. The effect of garlic extract on the acid production and pH of *Lact. fermenti ATCC* 9338

The results of the effect of garlic extract on the acid production and pH of the strain were shown in Fig. 11. There was difference in pH among each lot from the early phase of the incubation period. In general, the addition of extract tended to inhibit the fall in pH but 0.1 ml extract addition lot had similar tendency to control.

Acid production tended to be inhibited considerably also as the extract addition increased. 2 ml extract addition lot had considerable inhibition comparing with control. 0.5 ml extract addition lot had similar cid production to 2 ml extract addition lot with a sudden stimulation after 69 hours.

Statistical analysis showed no significant difference at 5% level.

12. The effect of red pepper extract on the acid production and pH of *Lact. fermenti ATCC* 9338

The results of the effect of red pepper extract on the acid production and pH of the strain were shown in Fig. 12. As shown in Fig. 12, 0.5 ml extract addition lot and 2 ml extract addition lot tended to be stimulated in drop of pH comparing with control but no difference was found between control and 0.1 ml extract addition lot. In acid production, 0.1 ml extract addition lot, 0.5 ml extract addition lot and 2 ml extract addition lot showed no difference with 2 ml extract addition lot considerably stimulated. 0.1 ml extract addition lot was appeared to have severe inhibition during the incudation period from 30 hours to 44 hours with having similar final acid production to control.

Statistical analysis showed no significant difference at 5% level.

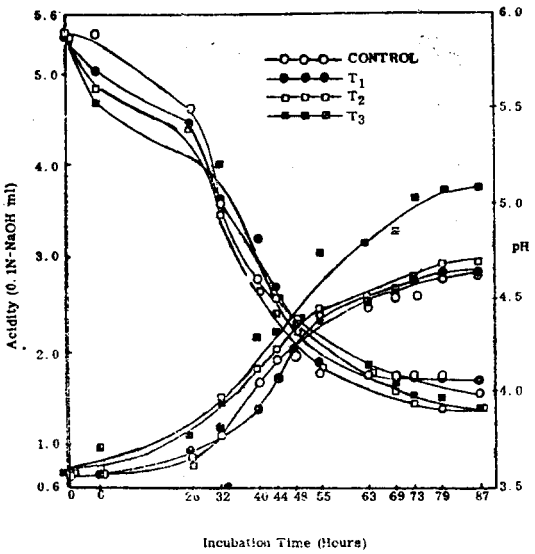

Fig. 12. Effect of red pepper extract on the acid production and pH of media inoculated with *Lact. fermenti ATCC* 9338

Discussion

Metcalf et al. (1946)[8] indicated that there are so many growth stmulants of lactic acid bacteria in nature including cabbage, red pepper andspinach.

Kim et al. (1968)[18] reported that cucumber stimulates Kimchi fermentation.

On the contrary, Zofia et al.(1973),[25] Fletcher et al.(1974)[26] informed that garlic extract inhibits the growth of *Stap aureus*, *E. coli*, *C. albicans* and coagulase activity.

As found out in the present research, in case of ginger extract, 2 ml extract addition lot (3.64 mg soluble solid/ml) and 0.5 ml extract addition lot(0.87 mg soluble solid/ml) showed marked growth stimulating effect as to *Lact. plantarum ATCC* 8014 and *Lact. fermenti ATCC* 9338 with 0.1 ml extract addition lot showing slight stimulating effect comparing with control.

Acid production of *Lact.plantarum ATCC* 8014 tended to be stimulated with the increase in growth, comparing with control, whereas *Lact, fermenti ATCC* 9338 was inhibited in acid production with final acidity similar to control, meaning a sudden rise in acidity after 55 hours of incubation time. These results led to the conclusion that there could occur any growh stimulant in ginger just as the report compiled by Kuiken et al.(1943),[7] Garley et al.(1941),[5] Murdock et al.(1952),[6] Mickle et al.(1924)[2] and Kulp et al. (1927)[4] that growth factors exist in various vegetable extracts, leaving the possibility that certain inhibitors of acid production about *Lact. fermenti ATCC 9338* might exist.

When garlic extract was added,2ml extract addition lot (31.7mg soluble solid/ml) and 0.5 ml extract addition lot(7.55 mg soluble solid/ml) of *Lact. plantarum ATCC 8014* showed growth inhibition during incubation period from 12 hours to 30 hours of incubation period but growth was sped up as the extract was added, on the whole. It was turned out that acid production was decreased during the incubation period of growth inhibition. In case of Lact. fermenti ATCC 9338, garlic extract addition increased the growth. 2 ml extract addition lot (31.7 mg soluble solid/ml) and 0.5ml extract addition lot (7.55 mg soluble solid/ml) showed striking growth stimulation with 0.1 ml extract addition lot (1.57 mg soluble solid/ml) having similar growth to control. During the incubation period of growth stimulation from 18 hours to 42 hours, acid production was inhibited markedly. These results presumed that garlic extract might not inhibit the growth of *Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* at least but stimulate with holding different views elucidated by Fletcher et al.(1974)[26] and Zofia et al.(1973)[25] that garlic extract inhibited the growth of *Stap. aureus.*

In case of red pepper extract addition, 2 ml extract addition lot (14.5 mg soluble solid/ml) 0.5 ml extract addition lot (3.46 mg soluble solid/ml) and 0.1 ml extract addition lot (0.72 mg soluble solid/ml) showed salient growth stimulation of *Lact. Plantarum ATCC 8014* comparing with control. The increase of the extract addition was thought to promote acid production continuously, whereas growth was not stim ulated above a certain limit even if the extract addition became higher. Growth and acid production of *Lact. fermenti ATCC 9338* was accelerated as the extract addition increased. These results suggested

that there might be any growth factor in red pepper, just like the opinion described by Metcalf et al. (1946),[8] It was thought that there was no distinct difference in growth stimulating effect among ginger, garlic and red pepper, as shown in above results. In this experiment, ginger, garlic and red pepper, used in Kimchi processing, were thought to inhibit the growth of *Stap. aureus* and E. coli, but stimulate the growh *of Lact. plantarum ATCC 8014* and *Lact. fermenti ATCC 9338* without inhibiting at least. It is the problem to further study that what components or what factors stimulate the growth.

要 約

本 實驗은 김치製造에 利用되는 생강, 마늘, 고추가 乳酸菌의 增殖 및 酸生成에 미치는 影響을 檢討하기위하여 培地에 이들의 물抽出液을 添加量을 달리하여 添加하고 *Lact. plantarvm ATCC 8014, Lact. fermenti ATCC 9338*菌株를 接種 培養하여 이때 일어나는 各種 變化를 調査하여 다음과 같은 結果를 얻었다,

1. 생강抽出物의 影響

1) 一定한 添加量의 限度(3.64 mg soluble solid/ml) 內에서는 抽出物의 添加量이 增加할수록 *Lact. plantarum ATCC 8014, Lact. fermenti A TCC 9338*菌株의 增殖을 促進하는 傾向을 보였다.

2) *Lact. plantarum ATCC 8014*菌株에 대해서는 抽出物이 添加됨에 따라 若干의 pH低下와 酸生成이 促進되었으나 *Lact. fermenti ATCC 9338*菌株에 대해서는 酸生成을 抑制하는 傾向을 보였다.

2. 마늘抽出物의 影響

1) 一定한 添加量의 限度(31.7mg soluble solid/ml) 內에서 抽出物 添加量이 增加할수록 *Lact. plantarum ATCC 8014, Lact. fermenti ATCC 9338*菌株의 增殖을 促進하는 傾向을 보였다.

2) *Lact. plantarum ATCC8014*菌株에 대해서는 抽出物의 添加量이 增加할수록 酸生成을 抑制하고($p<0.01$) pH의 下降도 抑制했다($p<0.05$). 또, *Lact. fermenti ATCC 9338*菌株에 대해서 抽出液添加量이 增加할수록 酸生成은 抑制되는 傾向을 보였다.

3. 고추抽出物의 影響

1) 一定한 添加量의 限度(14.5mg soluble solid/ml) 內에서 抑出液의 添加는 *Lact. plantarum ATCC 8014 Lact. fermenti ATCC 9338*菌株의 增殖을 促進하는 傾向을 보였다.

2) 一定한 添加量의 限度(14.5mg soluble solid/ml) 內에서 添加量이 增加할수록 *Lact. plantarum ATCC 8014, Lact. fermenti ATCC 9338*菌株의 酸生成을 促進하는 傾向을 보였다.

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