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THE INFLUENCE OF CERTAIN FACTORS UPON THE CHEMICAL COMPOSITION OF SAUERKRAUT.[1]

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The production of sauerkraut is an excellent illustration of the economic value of a fermentation process in the preservation of human food. It is an acid fermentation and in many respects is like that obtaining in the formation of silage, pickles, and dairy products. The art of making sauerkraut has been known and practiced for many years, but until recently little attention has been given to the different agents and factors involved in the process. Since the fermentation is a spontaneous one, it is evident that the type of flora which develops is the result of native and competing micro-organisms. Some of these organisms are desirable while others are objectionable. Although the lactic acid bacteria tend to dominate the fermentation, other bacteria or yeasts frequently develop and produce undesirable colors or flavors. The production of good kraut is, therefore, more or less a matter of chance and it is probable that inoculation with suitable bacteria will result in a better and more nearly uniform product.

The type of organisms present and the nature of the products formed in the production of sauerkraut were not studied until comparatively recent years. One of the earliest investigators in this field was Reichardt[2] who made an approximate analysis of sauerkraut and discussed its wholesomeness and food value.

In 1897 Conrad[3] made the first serious attempt to study the biochemical changes involved in the production of sauerkraut. He made a bacteriological examination of both cabbage and sauerkraut, and isolated two yeasts and an organism, *Bacterium brassicae acidae*, which he regarded as the chief agent responsible for the fermentation. With this bacterium he carried out a number of fermentations of cabbage and various carbohydrates to determine its fermentation characteristics, and its usefulness in the production of sauerkraut. Besides his work on the organisms of sauerkraut, he has supplied the most complete chemical analysis of this product that has been published. He reported the production of formic, acetic, butyric, and lactic acids, ethyl alcohol, carbon dioxide, methane, and hydrogen as a result of fermentation processes. The lactic acid was found to be inactive, and tests for acetone and mercaptans were negative. About the same time as Conrad's publication, a paper by Krause[4] appeared in which the acidity of sauerkraut was asserted to be due almost entirely to lactic acid with only a trace of acetic acid.

[1] Published with the permission of the Director of the Wisconsin Agricultural Experiment Station.

[2] Reichardt, *Z. Nahrungsm.*, **5**, 43 (1891).

[3] Conrad, *Arch. Hyg.*, **29**, 56 (1897).

[4] Krause, *Apoth. Ztg.*, **12**, 88 (1897).

In 1905 Wehmer[5] published two long papers on the types of organisms present in sauerkraut and the effect of inoculation, temperature, salts, and water on the quality of the kraut. He isolated and studied the fermentation characteristics of 2 bacteria and 5 yeasts. The organism which he considered as most important in the formation of the acid in sauerkraut was a short rod-form, *Bacterium brassicae*, which grew under both aerobic and anaerobic conditions and produced a high concentration of acid. The other type of bacterium was a long slender rod and produced only a small quantity of acid. The 5 yeasts were divided into 2 classes: bottom yeasts which produced gas and alcohol, and surface yeasts which destroyed the acids that had been formed earlier in the fermentation.

In 1905 Perekalin[6] isolated an organism from kraut, which he deemed typical of sauerkraut because of its growth and persistence in a highly acid medium.

In 1909 Gruber[7] isolated an organism *Pseudomonia brassicae acidae*, which when used in pure cultures gave a kraut of good flavor and shortened the period of fermentation.

Round[8] published a short note in 1916 calling attention to the large number of organisms present in the vats of commercial plants, and concluded that bacteria alone are concerned in a proper fermentation.

The papers of Henneberg[9] published in 1917, give a very extensive discussion of the manufacture of sauerkraut. In his bacteriological work on the flora of kraut he found 8 different yeasts, 7 strains of lactic acid bacteria, 2 colon types, 1 acetic acid type, and 5 molds. The bacteria which he considered the most important were *Bacterium lactis acidi*, and *Bacillus cucumeris fermentati*, a long rod related to Wehmer's *Bacterium brassicae*. The most important yeast was a small celled form, *Saccharomyces panis fermentati*. He studied the influence of inoculation with pure cultures of organisms upon the fermentation of sauerkraut, and obtained the best results by inoculating with a mixed culture of lactic acid bacteria and yeasts.

Nelson and Beck[10] published a short article in 1918 on the fermentation products found in sauerkraut. They reported the volatile acids to consist largely of acetic and a small proportion of formic and propionic acids. The alcohols were found to be mainly ethyl alcohol and a small amount of propyl alcohol.

In recent papers Le Fevre[11] has discussed the effect of temperature and the use of pure cultures on the production of sauerkraut. While inoculation aids in the fermentation of the cabbage, he considered temperature to be the most important factor in bringing about a rapid and satisfactory fermentation.

From the foregoing review of the literature it is clear that little is known concerning the kinds and amounts of fermentation products contained in sauerkraut.

Experimental.

The object of this work included the determination of the main products formed in sauerkraut during a normal fermentation and the influence of

[5] Wehmer, *Centr. Bakt. Parasitenk., II Abt.*, **14**, 682, 781 (1905).

[6] Perekalin, *ibid., II Abt.*, **14**, 225 (1905).

[7] Gruber, *ibid., II Abt.*, **22**, 555 (1909).

[8] Round, *J. Bact.*, **1**, 108 (1916).

[9] Henneberg, *Die Deut. Essigind.*, **20**, 133, 141, 152, 160, 166, 176, 184, 192, 199, 207, 215, 223 (1916).

[10] Nelson and Beck, THIS JOURNAL, **40**, 1001 (1918).

[11] Le Fevre, *The Canner*, **48**, 176 (1919); **50**, 161 (1920); **52**, 146 (1921).

inoculation on the products formed, and quality of the sauerkraut. The effect of salt concentration on the quality of the sauerkraut was also noted.

Most of the kraut was made under laboratory conditions, but two tanks were made on a commercial scale; one of these was inoculated with a pure culture of organisms and the other was left uninoculated in order to serve as a control.[12]

The kraut made under laboratory conditions consisted of 6 series of fermentations, comprising 61 separate samples. In each series 2 containers were always used as controls. In these the usual amount of salt (2–2.5%) was used and the cabbage allowed to ferment normally. To the other containers in the series pure cultures of organisms were added.

The fermentations were carried out in glass percolators of 2-liter capacity. The bottom of each percolator was fitted with a 1-hole rubber stopper, through which was passed a glass tube closed at the lower end with a piece of rubber tubing and a pinchcock. Some glass wool was placed over the end of the glass tube inside the percolator to prevent the cut cabbage from plugging the tube. This arrangement allowed the brine to be readily drawn from the bottom of the container without disturbing its contents. Fifteen hundred g. of cut cabbage, the weight usually taken, was prepared by removing the outer leaves and the cores of the cabbage and cutting with a hand cutter. The salt was mixed with the cabbage in a large pan and when cultures of organisms were used they were added at this time to insure uniform distribution. The whole was then transferred to the percolator. A large paraffined cork, slightly smaller than the inside diameter of the percolator, was placed over the cabbage, and this was held down by a bottle containing 1 kg. of mercury or sand.

The main chemical changes in the production of good sauerkraut involve the destruction of the sugars in the cabbage, resulting in the formation of volatile and non-volatile acids, alcohol, and mannitol. A decrease in the toughness of the cabbage and the development of a translucent appearance accompany this change.

Observations of the color and texture of the cabbage, as well as titrations of the brine were made daily during the first 6 to 8 days of the fermentation period, after which they were made at longer intervals. Titrations were made on the brine from the top and bottom of several containers to determine whether there existed any difference in the acidity. In one series stained mounts were made daily, and at the end of the fermentation, of the brine from the control and of the brine inoculated with *B. lactis acidi*. In several experiments counts were made of the number of micro-organisms present in the brine. From the data secured from a study of commercial kraut, inoculation appeared to increase the number of bacteria present,

[12] The authors desire to express their appreciation to the Onalaska Canning Company, Onalaska, Wisconsin for their coöperation in carrying out this part of the work.

but this was not true of the kraut made in the laboratory. The number of micro-organisms present in the samples of kraut investigated varied from about 500,000 to 91,000,000 per cc. of brine.

The rate at which the fermentation proceeds can be measured by titrating portions of the brine from day to day, and can be influenced by the temperature under which the fermentation takes place and the flora which is present.

The curves in Fig. 1 represent typical titrations of the rate and amount of acid developed and the influence of inoculation with pure cultures of organisms. It will be seen that in the case of the inoculated kraut there is an increase in the development of acidity over the control during the first few days. A fall in the amount of acid on about the tenth or twelfth day seemed characteristic of both fermentations. This decline was

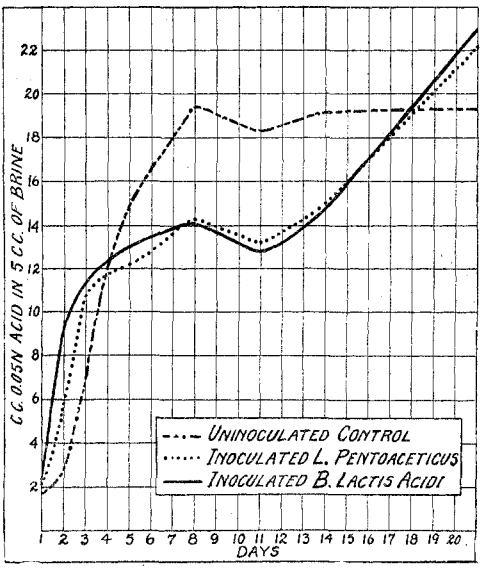

Fig. 1.—The influence of inoculation upon the development of acidity.

followed by another distinct rise in a few days. This second rise as well as the first one was invariably more pronounced in the inoculated fermentations than in the control. After the twenty-fifth day there was seldom much change in the amount of acid in the brine from the bottom of the container. However, there is always a marked decrease in the acidity of brine from the top, due to the destruction of the acid by *mycodermae*. The acidity at the top and bottom of the percolators was followed closely in several fermentations and it was found that during the first four days of the fermentation period there was practically no difference between the two. On the fifth day there was an increase in the acidity of the brine

from the top over that from the bottom except in one case where a great many *mycodermae* had developed. The acidities of the brine from the top exceeded those of the brine from the bottom until the fourteenth day when they generally fell below those of the bottom brines.

TABLE I.

THE INFLUENCE OF SALT ON THE FERMENTATION OF KRAUT.

Series.	Salt.	Temperature.	Inoculation.	Age when opened.	Titratable acidity in 5 cc. of brine expressed as 0.05 *N* acid. When opened.	Maximum.	Quality of Product.
	%	°C.		Days	Cc.	Cc.	
I[a]	3.5	20–23	none	31	17.0	17.1	Taste and odor good, texture tough.
I[a]	3.5	20–23	41–11	31	15.5	15.6	" " " " " "
II[b]	2.0	16–26	none	23	15.9	15.9	Taste, odor, and texture good.
II[b]	2.0	16–26	41–11	23	16.0	16.0	" " " " "
II[b]	0.0	16–26	none	20	14.3	14.3	Taste and odor disagreeable, texture too soft.
II[b]	0.0	16–25	41–11	23	12.8	19.6	Taste very sour and bitter, odor bad, texture too soft.
VII[c]	2.5	16–25	none	28	18.4	19.7	Taste, odor, and texture very good.
VII[c]	2.5	16–25	*B. lactis acidi*	28	17.5	19.0	Excellent taste, odor, and texture.

Dates: [a] October 9, 1920; [b] October 25, 1920; [c] March 21, 1921.

Influence of salt concentration.—It has been reported by some investigators that good kraut can be made without salt. With this idea in view 4 fermentations were carried out in the absence of salt. Two were inoculated with a pure culture of *Lactobacillus pentoaceticus* while the other two were left uninoculated. All 4 made a product having a bad odor, a sour and bitter taste, and a dark color. The data in Table I show that those inoculated produced higher acidity than the controls. This would be expected because Culture 41–11 is an organism forming a high degree of acidity.

In another series the salt content was increased from 2.0 to 3.5%. Although the flavor was good, a salty taste was predominant and the kraut was tough. In this experiment the control krauts developed a higher acidity than those which were inoculated with pure cultures. The reason for this is probably due to the sensitiveness of Culture 41–11 to a high salt concentration.

For the remaining series a concentration of 2 or 2.5 % of salt based on the weight of the cut cabbage was used and at either concentration very good results were secured with and without inoculation.

Influence of inoculation.—In Table II the results of a large number of inoculations with pure cultures are given; 50 cc. of a 48-hour yeast-water culture was used for each 1500 g. of cut cabbage, and to the controls 50 cc. of distilled water was added in place of the inoculating material.

Cultures 41–11 and 118–8 are the same type of organism and are named *Lactobacillus pentoaceticus*. They are essentially acid producers and ferment such sugars as xylose, arabinose, glucose, galactose, mannose, and fructose with the formation of lactic and acetic acids, ethyl alcohol, carbon dioxide, and mannitol. Cultures 52–7 and 124–1 were isolated from sauerkraut during this study; Culture 52–7 has the same characteristics as 41–11; 124–1 is a pentose-fermenter but does not belong to the same group as 41–11. Cultures 52–3 and 85 are yeasts and were isolated during this study from commercial sauerkraut that had turned red. These yeasts were found to produce red pigments when grown on agar plates. A report of red kraut and the factors influencing its formation is given in another paper now in press. The mixed culture was isolated from sauer-

TABLE II.

THE INFLUENCE OF INOCULATION ON THE FERMENTATION OF KRAUT.

2% of Salt used in Series IV and V, and 2.5% in Series VI.

Temperature 17–23° in Series IV; 21–23° in Series V; 16–25° in Series VI.

Series.	Inoculation.	Age when opened.	Titratable acidity in 5 cc. of brine expressed as 0.05 *N* acid.		Quality.
			When opened.	Maximum.	
IV	none	33	20.7	20.7	Flavor, odor and texture good.
	118–8	33	21.8	21.8	Texture good; flavor fair.
	B. lactis acidi	33	24.1	24.1	Excellent flavor and color; texture and odor good.
	B. bulgaricus	33	15.6	15.6	Developed red color; flavor fair.
	52–7	33	23.3	23.3	Taste, odor and color excellent.
	52–3	33	13.9	13.9	Developed red color; flavor poor.
	(52–3 +) (52–7)	33	15.0	15.0	Flavor, odor and texture, fair.
V	*B. lactis acidi*	31	19.1	19.1	Excellent flavor, color and texture.
	124–1	31	15.2	16.8	Developed red color; taste fair.
VI	none	28	18.4	19.7	Taste, color and texture good.
	124–1	28	12.3	15.2	Developed pink color; taste fair.
	B. lactis acidi	28	17.5	19.0	Flavor, odor and color excellent; texture good.
	B. lactis acidi	14	15.4	15.4	Taste, odor and color excellent; texture fair.

kraut but the characteristics of the organisms present were not determined. Strains of *B. lactis acidi*, and *B. bulgaricus* were also used.

From the remarks in Table II on the quality it will be seen that there are certain organisms which uniformly produce good kraut when used in pure culture. In the commercial kraut it was thought that Culture 41–11 produced a grade of sauerkraut superior to the uninoculated, but this could not be duplicated in the laboratory fermentations. The one organism which was always found to produce an especially good kraut was a

strain of *B. lactis acidi*. This organism was used in 3 series and produced the best kraut of the series each time. The product has a desirable appearance, a mild, pleasant odor and taste, and good texture. Although it does not produce acid as rapidly as some of the other organisms, nevertheless, kraut inoculated with this organism was declared ready to can after fermenting for 14 days. In contrast to the favorable results obtained with *B. lactis acidi* was the production of pink kraut whenever Culture 124–1 was used for inoculating. Culture 124–1 is a high acid-forming organism and appears to favor the development of the yeasts which produce the red pigment. Aside from this the kraut was of fair quality.

From the brine of several inoculated and uninoculated fermentations microscopic mounts were made. In nearly every case where pure cultures were used it was found that these organisms predominated throughout the fermentation period. In the last series of fermentations the flora was watched closely in both the uninoculated and inoculated kraut. Stained microscopic mounts were made daily on the brine. It was found that in the brine from the control the organisms were yeasts and long rod, short rod, and coccus forms of bacteria. The rod forms were about equal in distribution and were less numerous than the coccus forms during the first 7 or 8 days of the fermentation. After this time the long rod forms increased in number and were predominant at the end of the fermentation period.

In several of the series of experiments wet microscopic mounts were made and examined at the time the sauerkraut was removed from the containers. A comparison of the number of yeasts with that of the bacteria could be roughly determined in this manner and whenever yeasts were in excess over bacteria or were predominant in the field the kraut was invariably poor. It was also true that whenever the kraut turned red large numbers of yeasts were found to be present.

Chemical Analyses.—The kraut was removed from the containers and the brine pressed out with a powerful hand press. A small sample of the kraut was dried at 105° to constant weight to obtain the moisture content. A sample of the brine was used for determining the total titratable acidity and the remainder was used for determining the volatile acid, non-volatile acid, and alcohol. The methods outlined by Fred, Peterson and Davenport[13] were used in determining these constituents. A portion of the brine was distilled with steam and the volatile acids were titrated with 0.1 *N* barium hydroxide solution. The residue from the steam distillation was placed in a Kutscher and Steudel[14] apparatus and extracted with ether to obtain the non-volatile acids. The alcohols were determined in another portion of the brine by saturating it with sodium chloride and

[13] Fred, Peterson and Davenport, *J. Biol. Chem.*, **39**, 347 (1919).

[14] Kutscher and Steudel, *Z. physiol. Chem.*, **39**, 473 (1903).

distilling the alcohol which was in turn oxidized to the corresponding acid with potassium dichromate. The acid formed was distilled off and titrated with 0.1 *N* barium hydroxide solution. In all of the determinations

TABLE III.

ANALYSIS OF SAUERKRAUT SHOWING THE CHIEF FERMENTATION PRODUCTS.

Sample number and treatment.	Moisture.	0.1 *N* acid in 100 cc. of brine.	Volatile acid as acetic in 100 g. of kraut.	Non-volatile acid as lactic in 100 g. of kraut.	Alcohol as ethyl in 100 g. of kraut.
	%.	Cc.	%.	%.	%.
Commercial sauerkraut					
Inoculated with 41–11	90.0	144.7	0.233	1.220	0.843
Uninoculated[a]	88.0	147.5	0.244	1.254	0.860
Uninoculated[b]	90.6	140.0	0.224	0.929	0.658
Laboratory sauerkraut					
Control	91.6	158.0	0.315	1.195	0.371
Inoculated with 52–7	91.7	147.0	0.222	0.986	0.408
B. lactis acidi	90.3	173.0	0.265	1.259	0.267
B. lactis acidi	92.6	181.0	0.255	1.304	0.249
41–11, no salt	92.9	129.0	0.442	0.607	0.267
41–11	93.4	166.5	0.272	0.970	0.436

[a] This kraut had developed a pink color.

[b] This kraut was of good quality.

the barium salts formed were evaporated to dryness and used for the identification of the acids and alcohols. The results of a number of analyses on commercial and laboratory kraut are given in Table III.

The data show a fair agreement in the case of the uninoculated krauts, both laboratory and commercial, in all the products except in the alcohols. It has been shown by Peterson and Fred[15] that alcohol is easily lost by evaporation from a fermenting solution. Since the containers were not covered, it may well be assumed that alcohol was lost during fermentation while gases were escaping from the brine. Cultures 52–7 and 41–11 are the same type of organism and produce the same products. They are essentially high acid- and alcohol-producing organisms and a glance at the table will show that these products were characteristic of the fermentation. In kraut inoculated with *B. lactis acidi* large amounts of lactic acid would be expected. The data in the table are in harmony with this view, although the presence of volatile acid and alcohol indicate the activity of other organisms.

The analysis of kraut containing no salt shows an unusually high content of volatile acid with a very low content of non-volatile, and very little difference in the amount of alcohol. The absence of salt has evidently favored the growth of volatile acid and alcohol-producing organisms. The acids and alcohol from this fermentation were not identified, but butyric acid was suggested by a very disagreeable taste and odor. Butyric acid has been reported by Wehmer[5] in kraut made without salt.

[15] Peterson and Fred, *J. Biol. Chem.*, **42**, 273 (1920).

Identification of Products.—In the identification of the volatile acids three methods were used: the fractional precipitation and analysis of the silver salts; the Duclaux analysis; and the determination of the barium content of the barium salts. The barium salts of the volatile acids were dissolved in water, filtered from any insoluble material, and the filtrate made up to a definite volume. The qualitative method of Orla-Jensen[16] was used to determine what acids were present. From the solution of the barium salts the acids were fractionally precipitated as the silver salts by the addition of 2 *N* silver nitrate. The silver salts of the acids of highest molecular weight come out first and contain the lowest percentage of silver. After precipitation, the several fractions of silver salts were dried, weighed, and ignited to determine the silver content. In each fraction

TABLE IV.

DISTILLING CONSTANTS OF VOLATILE ACIDS OBTAINED BY DUCLAUX METHOD.

		Fraction.									
Source of acid.	Cc.	10.	20.	30.	40.	50.	60.	70.	80.	90.	100.
Purified acetic acid		7.4	15.2	23.6	32.2	41.4	51.0	61.3	72.4	85.1	100.0
Alcohol from inoculated kraut		7.6	15.6	23.8	32.5	41.5	51.0	61.2	72.3	84.8	100.0
Alcohol from uninoculated kraut		7.4	15.2	23.4	32.0	40.9	50.5	60.9	71.9	84.4	100.0
Volatile acid from uninoculated kraut		7.7	15.8	24.3	33.0	42.2	51.7	64.0	73.2	85.5	100.0
Volatile acid from 50 cc. fraction of uninoculated volatile acid		7.6	15.6	23.9	32.6	41.7	51.2	61.4	72.6	85.0	100.0

TABLE V.

BARIUM CONTENT OF BARIUM SALTS.

Source of acid.	Weight of barium salts. G.	Weight of barium sulfate. Found. G.	Weight of barium sulfate. Calculated. G.	Salt assumed in calculation.
Alcohol distillate inoculated with 41–11	0.3472	0.3121	0.3172	Barium acetate
	0.2893	0.2590	0.2643	
Alcohol distillate uninoculated	0.4726	0.4308	0.4318	
Volatile acids uninoculated	0.5304	0.4780	0.4846	
Volatile acids from 50 cc. fraction	0.3217	0.2980	0.2939	
	0.3226	0.2934	0.2947	
Non-volatile acids uninoculated	0.3038	0.2194	0.2248	Barium lactate
	0.3022	0.2192	0.2236	
Non-volatile acids inoculated with 41–11	0.3814	0.2787	0.2822	
	0.3890	0.2846	0.2878	

except the first, which was slightly contaminated with chlorides, the percentage of silver was always in very good agreement with that required for pure silver acetate. This result led to the conclusion that only acetic acid was present, and the correctness of this conclusion was proven by a determination of the barium content of the barium salts, and by a Duclaux

[16] Orla-Jensen, *Landw. Jahrb. d. Schweiz*, **18**, 314 (1904).

determination on the free acid. It will be seen from Table IV that the Duclaux constants are in very good agreement with those obtained with our apparatus on pure acetic acid. Table V shows the barium content to be very close to the calculated value for pure barium acetate. From these results it is believed that acetic acid was the only volatile acid present.

This was further substantiated by liberating 50 cc. of 0.1 *N* acid from the barium salt by the addition of an equal amount of sulfuric acid. The solution was distilled with steam and the acids in the distillate subjected to a Duclaux determination. Since the acids of higher molecular weight are more volatile than acetic, they should have been collected by this method in excess over acetic. However, it will be seen from Table IV that the results of the Duclaux determination are almost identical with those of the first analysis and are in good agreement with those for pure acetic acid. The barium content was determined on the barium salts of this fraction and it was found to agree closely with the barium content in the first analysis and the theoretical value for pure barium acetate. From these results apparently acetic acid was the only volatile acid present.

The identification of the volatile acid formed from the oxidation of the alcohol was carried out in the same manner as the identification of the volatile acid in the kraut. The results of the Duclaux determination appear in Table IV, and the barium content of the barium salt is given in Table V. Since these are in such good agreement with pure acetic acid it is concluded that only acetic acid was present and, therefore, ethyl alcohol was the only alcohol present.

In identifying the non-volatile acids the barium salts were dissolved in water and filtered from any insoluble material. After the filtrate was evaporated to about 30 cc. it was made up to 300 cc. with absolute alcohol. If any barium succinate were present, it would be precipitated at this point because it is insoluble in 90% alcohol. The only precipitate which formed was a sticky wax-like substance which was readily soluble in small amounts of water. The alcohol solution of the barium salt was then filtered, made

TABLE VI.

WATER OF CRYSTALLIZATION IN ZINC LACTATE.

Source of acid.	Wt. of zinc lactate used.	Water lost.		Water in $Zn(C_3H_5O_3)_2 + 3H_2O$.
	G.	G.	%.	%.
Uninoculated kraut (1)	0.8780	0.1597	18.26	18.17
(2)	2.3730	0.4334	18.19	18.17
Kraut inoculated with 41–11 (1)	0.9568	0.1726	18.04	18.17
(2)	0.7485	0.1356	18.11	18.17

up to volume and aliquot portions were taken for determining the barium content of the barium salts. The data are given in Table V and will be found to agree very well with the barium content of pure barium lactate. The remainder of the alcoholic solution of the barium salts was used for

making the zinc salts. This was done by distilling the alcohol and taking up the barium salts in water. The zinc salts were then formed by the addition of 0.5 *N* zinc sulfate solution. After the barium sulfate was filtered off the filtrate was concentrated to a small volume (20–30 cc.) from which the zinc lactate was allowed to crystallize. The kind of lactic acid present was determined by obtaining the water content of the zinc lactate. From Table VI it will be seen that these results are in very good agreement with those of the zinc salts of inactive lactic acid. From these data it appears that inactive lactic acid is the form of acid produced in the fermentation.

Mannitol.—One quantitative determination of mannitol was made in the following manner. The residue in the extraction apparatus after the non-volatile acids had been removed was freed from ether and neutralized with barium hydroxide. The precipitate of barium sulfate was filtered off and the filtrate evaporated to dryness on asbestos, from which the mannitol was extracted, by continuous extraction with boiling absolute alcohol. This extraction was continued until a 12-hour extraction recovered no trace of mannitol. The mannitol is insoluble in cold absolute alcohol and crystallizes out readily. Absolute alcohol is an improvement over 90 or 95% alcohol because it extracts the mannitol practically free from salt. It is difficult to separate mannitol from sodium chloride by crystallization if they are extracted together.

Feder[17] used 90% alcohol for extracting the mannitol and states that his product contained about $^1/_3$ salt. The results obtained are much lower than those reported by other investigators. In four determinations Feder found from 0.8 to 1.16%, while Nelson and Beck[10] found from 1.9 to 2.5% of mannitol. In a natural fermentation such as sauerkraut the amount of mannitol present is largely a matter of chance. It is formed by certain bacetria which ferment a part of the fructose to acids, etc., and at the same time reduce another part of the fructose to mannitol. The mannitol thus formed may subsequently be destroyed by the bacteria that produced it or by many other bacteria which use mannitol as a source of carbon. Cabbage contains approximately 4% of sugar, and it is probable that not more than half of this is fructose. Since the highest conversion of fructose to mannitol reported by Gayon and Dubourg,[18] was 66%, the above results appear very high. By extracting with absolute alcohol only 0.27% of mannitol was found. This had a melting point of 165°.

Summary.

From the results of 61 experiments it appears that inoculation with certain organisms produces a better grade of sauerkraut than is produced

[17] Feder, *Z. Nahr. Genussm.*, **22**, 295 (1911).

[18] Gayon and Dubourg, *Ann. inst. Pasteur*, **15**, 527 (1901).

by a natural fermentation. Although an improved product was obtained with several different organisms the only one which was consistently better than the control was *B. lactis acidi*. The data are not extensive enough to recommend the use of inoculation on a commercial scale. Further experiments are needed.

That the organisms used for inoculation dominated the fermentation was indicated by microscopic mounts made from the brine at intervals throughout the fermentation period. A chemical analysis also showed the characteristic products normally formed by these organisms.

The presence of large numbers of yeasts may cause red kraut, and undesirable flavors.

The best kraut was obtained when approximately 2.0% of salt was used. With concentration above 3.0% the kraut was tough and too salty.

The chief products formed in the fermentation of kraut are lactic acid, acetic acid, and ethyl alcohol. Mannitol in varying amounts may also be formed, depending upon the type of organisms present. These same products occur in a natural fermentation, but the relative amounts can be influenced by inoculation.

MADISON, WISCONSIN.

[CONTRIBUTIONS FROM THE CHEMICAL LABORATORY OF THE UNIVERSITY OF ILLINOIS.]

2-PHENYLQUINOLINE-4-CARBOXYLIC ACID-6-ARSONIC ACID.[1]

BY J. R. JOHNSON WITH ROGER ADAMS.[2]

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In connection with the investigations being carried on in this laboratory on arsenic compounds of possible therapeutic value, 2-phenylquinoline-4-carboxylic acid-6-arsonic acid has been prepared. It is formed by the action of pyruvic acid on benzaldehyde and arsanilic acid, and is the simplest of a large number of compounds which may be made in a similar manner. This communication is merely a preliminary notice of a rather extensive research which has been carried out in this laboratory during the past 2 years upon various quinoline arsonic acids, their reduction products and other derivatives.

When aniline[3] and some of the simplest aniline derivatives are treated with pyruvic acid and an aromatic aldehyde, it is generally possible to obtain both a phenyl-cinchoninic acid derivative and a phenyl-diketo-

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[2] This is a description of a portion of the laboratory work submitted by J. R. Johnson in partial fulfilment of the requirements for the degree of Doctor of Philosophy in Chemistry at the University of Illinois.

[3] *Ber.*, **41**, 3884 (1908).