SOME PROBLEMS IN CHEESE-CURING.

WHAT PRODUCES CASEIN-CHANGES?
F. H. HALL, L. L. VAN SLYKE, H. A. HARDING AND E. B. HART.

HOW CAN LOSS OF WEIGHT BE CONTROLLED?
F. H. HALL AND L. L. VAN SLYKE.

PUBLISHED BY THE STATION.
BOARD OF CONTROL.

GOVERNOR BENJAMIN B. ODELL, JR., Albany.
STEPHEN H. HAMMOND, Geneva.
AUSTIN C. CHASE, Syracuse.
FRANK O. CHAMBERLAIN, Canandaigua.
FREDERICK C. SCHRAUB, Lowville.
NICHOLAS HALLOCK, Queens.
LYMAN P. HAVILAND, Camden.
EDGAR G. DUENSBURY, Portville.
OSCAR H. HALE, North Stockholm.
MARTIN L. ALLEN, Fayette.

OFFICERS OF THE BOARD.

STEPHEN H. HAMMOND, 
President.

WILLIAM O’HANLON, 
Secretary and Treasurer.

EXECUTIVE COMMITTEE.

STEPHEN H. HAMMOND, 
MARTIN L. ALLEN,
FRANK O. CHAMBERLAIN,

FREDERICK C. SCHRAUB,
LYMAN P. HAVILAND,
NICHOLAS HALLOCK.

STATION STAFF.

WHITMAN H. JORDAN, SC. D., Director.

GEORGE W. CHURCHILL, 
Agriculturist and Superintendent of Labor.

WILLIAM P. WHEELER, 
First Assistant (Animal Industry).

FRED C. STEWART, M.S., 
Botanist.

HARRY J. EUSTACE, B.S., 
Student Assistant in Botany.

LUCIUS L. VANSLYKE, PH.D., 
Chemist.

CHRISTIAN G. JENTER, PH.C., 
* WILLIAM H. ANDREWS, B.S., 
* J. ARTHUR LECLERC, B.S., 
FREDERICK D. FULLER, B.S., 
EDWIN B. HART, B.S., 
* CHARLES W. MUDGE, B.S., 
ANDREW J. PATTON, B.S., 
Assistant Chemists.

HARRY A. HARDING, M.S., 
Dairy Bacteriologist.

LORE A. ROGERS, B.S., 
Assistant Bacteriologist.

GEORGE A. SMITH, 
Dairy Expert.

FRANK H. HALL, B.S., 
Editor and Librarian.

VICTOR H. LOWE, M.S., 
†F. ATWOOD SIRRINE, M.S., 
Entomologists.

PERCIVAL J. PARROTT, A.M., 
Assistant Entomologist.

SPENCER A. BEACH, M.S., 
Horticulturist.

NATHANIEL O. BOOTH, B.AGR., 
Assistant Horticulturist.

ORRIN M. TAYLOR, 
Foreman in Horticulture.

FRANK E. NEWTON, 
JENNIE TERRILL, Clerks and Stenographers.

ADIN H. HORTON, 
Computer.

Address all correspondence, not to individual members of the staff, but to the NEW YORK AGRICULTURAL EXPERIMENT STATION, GENEVA, N. Y. The Bulletins published by the Station will be sent free to any farmer applying for them.

* Connected with Fertilizer Control.
† At Second Judicial Department Branch Station, Jamaica, N. Y.
‡ Absent on leave.
WHAT PRODUCES CASEIN CHANGES IN CHEESE-RIpening?

F. H. HALL.

When Cheddar cheese comes from the press it Ripening of consists principally of coagulated casein. It is then tough, almost tasteless and odorless, mostly cheese necessary. insoluble and a wholly unattractive article of diet. By curing for sufficient time under proper conditions it changes from a tough, rubber-like substance to one which is mellow and friable; develops its characteristic fragrant, nutty odor and agreeable taste; and becomes soluble and digestible, a spur to appetite and a satisfying food.

How have these changes been accomplished? How brought We do not know. We can state with some confidence many of the conditions necessary to secure proper ripening; though it has been only within a very few years that even such elementary, external facts as the degree of temperature and the amount of moisture in the air needed to secure best results, have been carefully studied. We know something, also, of the chemical transformations which take place in the cheese while on the curing shelves; but as to

*This is a brief review of Bulletin No. 203 of this Station, on A Study of Enzymes in Cheese, by L. L. Van Slyke, H. A. Harding and E. B. Hart. Anyone specially interested in the detailed account of the investigations will be furnished, on application, with a copy of the complete bulletin. The names of those who so request will be placed on the Station mailing list to receive future bulletins, popular or complete as desired. Bulletins are issued at irregular intervals as investigations are completed, not monthly.
the methods by which these changes occur and the agencies which produce them, investigators, so far, have given us only dim or contradictory notions.

For many years the changes in cheese were Agency of bacteria. regarded, like other fermentations and decompositions, as merely chemical reactions. With advancing knowledge of bacteriology, joined with the evident presence of these minute organisms in all cheese, bacteria were assumed to be the active agents both in producing flavor and in breaking-down casein. This view appeared to be proved correct by making cheeses with the addition of thymol or kreolin, which prevent bacterial action; under which conditions the cheeses did not appear to ripen, even when kept twice the usual time. It was held that if cheese failed to ripen when the bacteria were killed, the bacteria must be the cause of the ripening.

But bacteria are merely plants and, as such, are unable to assimilate solid food. Whatever serves for their nutrition must be in solution; so, if they live upon the casein and thus change it into the forms found in ripened cheese, they must themselves excrete some substance which can dissolve the coagulated curd or else the curd must be made soluble by some other agency.

There are such agents—certain chemical compounds, known as enzymes, which are the lifeless products of the activities of living animal or vegetable cells and which possess the peculiar property of causing chemical changes in other bodies with little or no change in themselves. It is known, also, that certain bacteria do excrete enzymes which can dissolve milk-casein and produce cheese-like odors. Consequently, one of the more recent beliefs has been—probably still is, with many—that these enzyme-secreting bacteria are the agents in cheese-ripening. But, again, careful bacteriological analyses, made within the last few years, have shown that such enzyme-producing bacteria are very few in number as compared with other kinds in cheese and that they cease their activities very early in the curing of the cheese, while lactic-acid-forming bacteria increase rapidly and remain in the cheese during the curing. This has led a group of scientists to uphold the view that the lactic-acid-formers are the cause of the ripening. This theory is opposed by other investigators.
In 1897 another factor was introduced into the problem when Babcock and Russell of the Wisconsin Station announced that milk itself contains an enzyme, galactase, which can digest casein. In 1900, these same authors and, almost at the same time, Jensen in Switzerland, noted that pepsin, an enzyme associated with rennet, possesses a similar power.

Thus it appears that Cheddar cheese, as ordinarily made, contains enzymes from three sources, bacteria, milk glands of cows, and rennet. The amount of work done by enzymes in cheese-curing, however, and the exact function of those from different sources are unsettled questions. The problem is a complex one; as, under ordinary conditions, the activities of germ life confuse the observations. The enzymes from the different sources are so thoroughly mingled, also, in making the cheese, that it is very difficult to study the action of one alone.

Yet study of these separate enzymes is demanded, for their action upon cheese-casein is apparently a fundamental factor in cheese-ripening. Such study necessarily involves coöperation of the expert cheese-maker, the bacteriologist and the chemist, working where conditions of making and curing the cheese can be held under close control. This coöperation and proper working conditions are available at this Station and have been devoted for a considerable time to work upon enzymes in cheese. Only a beginning has been made, but the investigations have already thrown light upon some phases of cheese-ripening.

In order to secure enzyme action free from the disturbing influences of germ life, it is necessary to destroy the bacteria or to prevent their growth. This can be done easily by heat; but heating the milk would not keep the cheese sterile during making and curing, and a temperature high enough to kill the bacteria would also destroy the chemical activity of the enzymes present. After trials with several disinfectants and anesthetics, chloroform was selected as the best agent to preserve the cheese from bacterial activity. This apparently had but little restraining effect upon enzyme action, when it was mixed with the milk, but destroyed the growing bacteria.
The chloroform was added to the milk in the vat and the usual cheese-making processes followed. The repeated stirrings kept the chloroform from settling and distributed it quite uniformly throughout the curd. The cheeses were kept under pressure from 18 to 24 hours and then transferred to a curing-room kept very closely to a temperature of 60° F. Each cheese was placed under a bell jar in an atmosphere of chloroform. Check cheeses were made under similar conditions except that the chloroform was omitted.

The amount of enzyme action was measured by the amount of soluble nitrogen compounds formed at successive stages in the ripening of the cheese; that is, by the amount of insoluble casein made soluble.

Some results.

The chloroform checked bacterial action and prevented the formation of enzymes after its addition, but did not materially affect the action of enzymes already existing. Hence any breaking-down of casein in the chloroform cheese must be ascribed to galactase in the milk, pepsin in the rennet, or to some unknown factor.

Two cheeses, one with chloroform, the other without, but both made without salting the curd, show the results given below:

**Comparison of Normal Cheese with a Cheese Made and Cured with Chloroform.**

<table>
<thead>
<tr>
<th>Conditions of curing</th>
<th>Total water-soluble N. formed for 100 lbs. N. in cheese.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2 weeks.</td>
</tr>
<tr>
<td>11.50</td>
<td>18.50</td>
</tr>
<tr>
<td>Chloroform . . . . .</td>
<td>5.30</td>
</tr>
</tbody>
</table>

At the end of one month more than three times as much water-soluble nitrogen was found in the normal cheese as in the chloroform cheese; and the difference was not overcome in 15 months of curing. The breaking-down of casein in the chloroform cheese increased faster during the later months of curing than of that in
the normal cheese, so that at the end of 15 months the enzymes in the chloroform cheese had digested 72 per cent. as much casein as had been made soluble in the normal cheese. Thus the peculiar conditions of manufacture in the presence of chloroform were not such as to prevent the enzymes from rendering cheese-casein soluble.

The bacteria in normal cheese form a considerable amount of acid in the cheese while curing. But this acidity could not be produced in the chloroform cheese because the bacteria were killed. It was, therefore, thought necessary to make other chloroform cheeses, adding lactic acid during the manufacture.

These cheeses, as compared with others made with chloroform and without acid, showed much more rapid ripening, from the end of the first month up to the end of 12 months. The acid appears to favor enzyme action, at least of that associated with the rennet.

In none of these cheeses had salt been used, a condition contrary to all practice; therefore other cheeses were made from salted curd, with chloroform, both with and without lactic acid. The salt appears to exert a strongly repressive influence upon the action of the enzymes; for at the end of 12 months there had been formed for each 100 lbs. of nitrogen in the cheese, 22.6 lbs. and 31.65 lbs. of soluble nitrogen in the two unsalted chloroform cheeses, without and with lactic acid, respectively; and only 17.2 lbs. and 19.65 lbs. in the salted cheeses. Here, as in the preceding set, the acid favored enzyme action.

A chemical study of the soluble matter of the cheeses showed marked differences in the relative amounts of the separate nitrogenous compounds formed in the normal and chloroform cheese. In the normal cheese ammonia was formed; and the simpler, crystallizable nitrogen compounds were considerably more in quantity than the more complex, non-crystallizable bodies at the end of one month, with the disproportion increasing throughout the curing. In the chloroform cheese, the more complex bodies were the predominating ones; and no ammonia was produced for at least six months and only minute quantities after a year.
The results of our work up to this time appear to show. (1) That the use of chloroform excludes bacterial action in milk and cheese, and limits the work of ripening to those enzymes contained in milk when made into cheese; (2) that the presence of salt noticeably decreases the effect of such enzymes; (3) that the presence of two-tenths of one per cent. of lactic acid increases the ripening action, at least of rennet enzymes; (4) that the percentage of cheese-casein made soluble by the enzymes under consideration in nine months (which may be regarded as the extreme limit of the commercial life of Cheddar cheese, kept under usual conditions) is about 12 per cent., or one-third the amount of soluble nitrogen found in normal cheese; and (5) that the amount of ripening caused by enzymes present in the milk when made into cheese is apparently more limited than was previously supposed."

It also "appears that there is some agent at work in normal cheese which is not active in cheese made with chloroform. Just what this additional factor is, our present data do not explain; but our efforts are being directed to the task of identifying this agent."