

# The influence of heating on silage fermentation and quality

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## Summary

A description is given of a number of experiments on the influence of oxygen and of temperature on silage fermentation. Due to the influence of oxygen, carbohydrates are lost by respiration and as a result of this process the temperature in the silage may increase.

At a temperature above 40° C oxygen is responsible for the fixation of protein into an undigestible compound. Under farmscale conditions the highest percentages of butyric acid and of ammonia are found in silages with a maximum temperature of 40—50° C.

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## 1. Introduction

Opinions differ widely on the most desirable temperature in silage. Although a number of workers considers a low fermentation temperature advantageous for a good preservation (DIJKSTRA, 1958; MURDOCH, 1960), others prefer 35—45° C or even higher temperatures. In a previous paper (WIERINGA, 1960) it was already stated that temperature control by itself cannot be the base for an ensiling method, because unwanted conversions of the organic substances may occur at a low as well as at a high temperature. This is also demonstrated by the yearly survey of the silage quality in the Netherlands, as given by the "Laboratory for Soil and Croptesting".

From TABLE 1 it can be seen, that the quality of the silages made without additives is very poor in comparison to the quality of grass ensiled by other methods.

Since in this country the temperature in the so called warm silage generally exceeds

TABLE 1. Silage quality in 1958

| Method                        | Dry matter<br>% | pH  | Butyric acid<br>% | NH <sub>3</sub> -nitrogen<br>in % of total N |
|-------------------------------|-----------------|-----|-------------------|--|
| <i>Without additives :</i>    |                 |     |                   |  |
| Warm .....                    | 23,6            | 5,2 | 1,3               | 27   |
| Cold .....                    | 24,8            | 5,2 | 1,2               | 20   |
| <i>With additives :</i>       |                 |     |                   |  |
| A.I.V. ....                   | 24,4            | 4,4 | 0,7               | 14   |
| Molasses, mixed by hand ..... | 26,9            | 4,7 | 0,8               | 16   |
| "    "    " machine .....     | 25,1            | 4,3 | 0,2               | 11   |

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50° C, the question arose whether a moderate temperature of 35—45° C as recommended by WATSON would give better results than higher or lower fermentation temperatures.

**2. Experimental**

In order to investigate the influence of temperature on silage fermentation and on the digestibility of the crude protein in silage, a number of ensiling experiments were carried out on farm scale as well as on laboratory scale from 1958 to 1960. In the farm-scale experiments gunnysacks with grass were buried in the silo on different places.

Temperatures in the sacks were measured by means of thermocouples. Differences in silage temperature have been obtained by the use of different silo-types (clamps and pit silo's) and by differences in compaction of the grass. In the laboratory-scale experiments grass was ensiled in 1 l hermetically closed glass jars which were placed in incubators at different temperatures.

Chemical analyses were conducted by the Institute for Biological and Chemical Research of Field Crops (I.B.S.) at Wageningen.

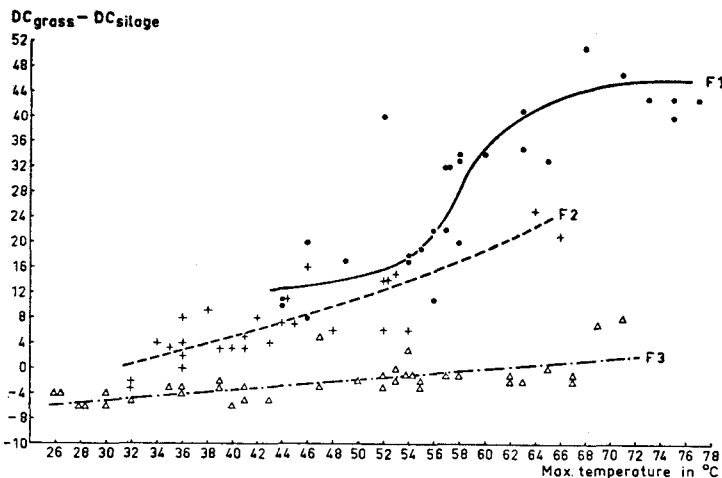
**3. Results**

*a. The influence of heating on the pepsin digestibility of the crude protein*

Since it is known that in warm silage the major part of the crude protein may become insoluble and undigestible, an attempt was made to establish the lower limit of the undesirable temperature range. In the first experiment on farm scale (F1 in FIG. 1) a temperature of 50° C or higher appeared to be detrimental to the pepsin digestibility of the protein. In two other experiments on farm scale (F2 and F3) the influence of temperature on the digestibility was less distinct.

Further it appeared to be impossible to cause even the slightest decrease in the digestibility in the laboratory-scale silage experiments, although temperatures up to

FIG. 1. Influence of the maximum temperature in the silage on the digestibility coefficient (D.C) of the crude protein



70° C were applied. This discrepancy between the results of the farm-scale and the laboratory-scale experiments led to the conclusion, that the small-scale method failed in imitating the warm fermentation process. The main difference between both methods is the way of heating of the silage. In practice the increase in temperature is the result of an aerobic biochemical process, whereas in the laboratory silages the temperature is raised artificially in a hermetically closed anaerobic system. Thus it was thought that the aerobic conditions play an important part in decreasing the digestibility of the protein in warm silage.

In order to test this hypothesis glass jars filled with grass were aerated for 0, 1 or 2 days, at temperatures of 55, 60 and 65° C. After aeration the jars were closed and placed for one week at the same temperature, after which the temperature was lowered by 5° C per week down to 20° C. No direct influence of the temperature on the digestibility coefficient could be detected in this experiment, as can be seen from TABLE 2. But the aeration caused a highly significant reduction of the digestibility at all the three temperature levels.

TABLE 2. Digestibility coefficients of the silage crude protein

| Aeration time<br>(days) | Number of<br>silages | Digestibility coefficients at maximum<br>temperature (°C) |      |      | Mean values |
|-------------------------|----------------------|---|------|------|-------------|
|                         |                      | 55°   | 60°  | 65°  |             |
| 0                       | 6                    | 75,5  | 77,5 | 74,5 | 75,8 ± 1,0  |
| 1                       | 6                    | 71,5  | 71,0 | 67,5 | 70,0 ± 1,3  |
| 2                       | 5                    | 56,0  | 62,5 | 62,0 | 61,0 ± 1,5  |

This result was the motive for a renewed study of the data given in FIGURE 1. Out of a number of samples of each of these three experiments the temperature was recorded over a period of 2—3 months. For these samples the influence of the time of heating on the decrease of the digestibility coefficient is recorded in FIG. 2. It can be seen that, if the decrease of the digestibility is plotted against the time of heating (FIG. 2) instead of the max. temperature (FIG. 1), the apparent contradiction between the results of the three trials disappears.

In FIG. 2 the time during which the temperature exceeded 35° C was used as an indication for (semi-)aerobic conditions, although it cannot be said whether this is the best criterion. Because the amount of oxygen available for reaction with proteins

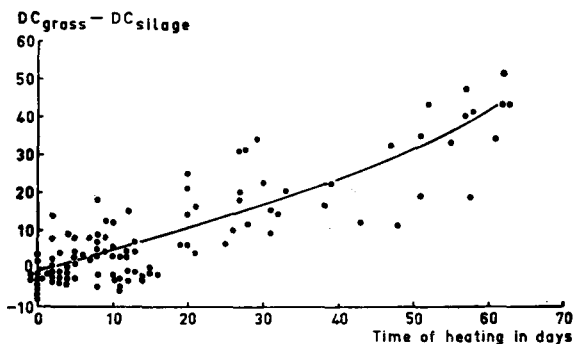


FIG. 2. Influence of the time of heating (temperature above 35° C) on the digestibility coefficient (D.C.) of the protein

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in the silage is correlated with the increase in temperature, and because a high temperature often goes hand in hand with a long time of heating, it is difficult to determine whether aeration at a low temperature is as disastrous as at a high temperature. The results of the farm-scale experiment 0.59 indicate that the speed of fixation of the protein increases with the temperature.

From FIG. 3 it can be seen that the loss of digestibility remains small if the temperature is 40° C or lower, and becomes very serious at temperatures above 50° C.

The FIG.'s 1, 2 and 3, in which the digestibility coefficient has been plotted against time or against temperature, may give a misleading picture of the actual protein fixation. During the fermentation a certain percentage of the protein is converted into ammonia. Due to the fact that silage crude protein figures are always recorded "free of ammonia", the relative percentage non-digestible protein will increase if the ammonia is produced exclusively from the digestible protein.

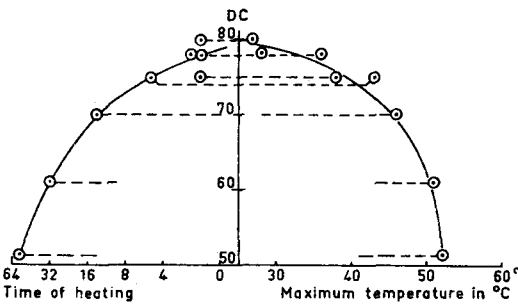


FIG. 3. Influence of temperature and time of heating in the digestibility of crude protein. Experiment 0.59

From FIG. 4 it can be concluded that the decrease in digestibility means a real quantitative increase in non-digestible protein (n.d.p.).

From these experiments it may be concluded that the actual amount of non-digestible protein may increase from above 60 % (in % from grass n.d.p.) to more than 300 %.

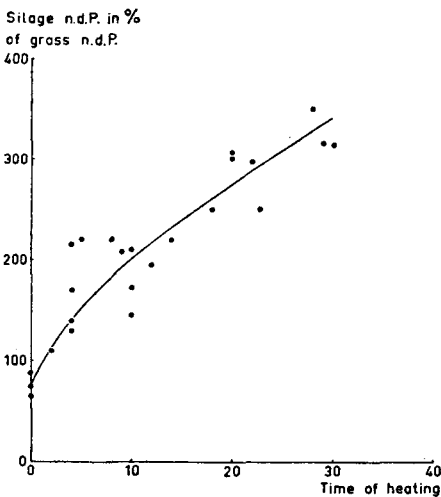


FIG. 4. Influence of time of heating (days) at a temperature above 50° C on the actual amount of non-digestible protein (n.d.p.). Experiment F and 0.59

b. *The influence of heating on the bacterial fermentation*

The first laboratory-scale experiments under strictly anaerobic conditions showed that the optimum temperature for butyric acid fermentation and for putrefaction were respectively 35° and 30° C (WIERINGA, 1960).

In the more complicated experiments with different aeration periods and with a descending storage temperature both optimums were higher.

From FIG. 5 it can be seen that the grass under anaerobic conditions gave a good quality silage at all temperatures used. But aeration during one or two days caused a decrease in lactic acid production and an increase in production of ammonia, butyric and acetic acid. The fact that the temperature optima for ammonia and butyric acid in this experiment were both about 45° C is probably due to the low soluble-sugar content of the grass (4,3 %). For it must be assumed that, due to aeration at higher

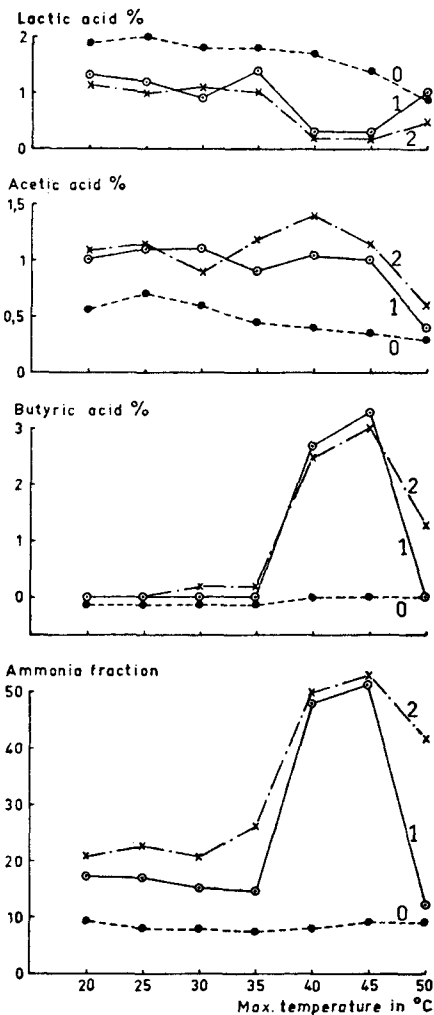


FIG. 5. Influence of aeration time and temperature on silage fermentation.

0 = no aeration  
 1 = aeration during one day  
 2 = aeration during two days  
 organic acids in percentages of wet material.

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temperatures, the loss of sugar was caused by an insufficient lowering of the pH in these silages, which resulted in putrefaction and butyric acid fermentation during the lowering of the temperature. Some examples of sugar losses during the first period of heating in silages are given in TABLE 3.

TABLE 3. Influence of heating on the sugar content of grass

| Treatment         | Maximum temperature | Soluble sugars in % of |           |           |
|-------------------|---------------------|------------------------|-----------|-----------|
|                   |                     | Exp. 4.20              | Exp. 8.16 | Exp. 9.26 |
| Fresh grass ..... | —                   | 18,5                   | 6,5       | 2,9       |
| after 24 h .....  | 35° C               | —                      | —         | 6,7       |
|                   | 45° C               | —                      | 1,7       | 2,0       |
| after 48 h .....  | 20° C               | 14,0                   | —         | —         |
|                   | 35° C               | 11,0                   | —         | —         |
|                   | 40° C               | —                      | —         | 1,7       |
|                   | 45° C               | —                      | 1,1       | —         |
|                   | 50° C               | —                      | —         | 1,0       |

TABLE 4. Quality distribution of 112 farmscale silage samples

| Maximum temperature | Number of samples | Samples in % of total number | % of sample with a butyric acid |         |       | % of samples with ammonia fraction |       |      |
|---------------------|-------------------|------------------------------|---------------------------------|---------|-------|------------------------------------|-------|------|
|                     |                   |                              | < 0,2                           | 0,2-0,5 | > 0,5 | < 10                               | 11-20 | > 20 |
| < 30° C ..          | 8                 | 7                            | 50                              | 50      | 0     | 100                                | 0     | 0    |
| 31—40° C ..         | 27                | 24                           | 56                              | 22      | 22    | 56                                 | 30    | 14   |
| 41—50° C ..         | 28                | 25                           | 11                              | 14      | 75    | 25                                 | 32    | 43   |
| 51—60° C ..         | 30                | 27                           | 27                              | 16      | 57    | 57                                 | 23    | 20   |
| 61—70° C ..         | 13                | 12                           | 31                              | 7       | 62    | 77                                 | 8     | 15   |
| > 70° C ..          | 6                 | 5                            | 100                             | 0       | 0     | 50                                 | 50*   | 0    |

\* Mean amm. fraction value of these 3 samples is 11,0.

The optimum of 45° C found in the laboratory-scale experiments as well as for ammonia production is in good agreement with the results obtained from the farm-scale experiments. In TABLE 4 the distribution of butyric acid in % of wet material and ammonia (as NH<sub>3</sub> - nitrogen in percentages of total N) for the different temperature levels is recorded. From these figures it can be seen that a maximum temperature of 40—50° C gives the lowest percentage of good preserved silages. Further it can be noted that silages "free" of butyric acid (< 0,2 %) were only obtained at a temperature below 30° C or higher than 70° C.

#### 4. Discussion and conclusions

The influence of heating on the preservation of protein in silage is of two kinds. Firstly, under aerobic conditions, protein is converted to an undigestible compound. Secondly the temperature is of great importance for the development of the different kinds of micro organisms, thus ruling proteolysis and ammonia production.

According to the work of HARBERTS and IMMINK (1959) on the digestibility of protein in heated hay, the fixation of protein is a (bio)chemical reaction between amino groups and aldehyde groups. From our work on the heated silages it appears that oxygen also plays a role in this chemical process. High temperatures, and especially a long heating time, are favouring the protein fixation.

A second effect of the oxygen on silage is that the respiration goes on, and soluble sugars are lost. Here too the losses will increase with temperature and time of heating.

The respiration will result in a retardation of the production of lactic acid or even, in the case of grass low in soluble sugars, in an insufficient lactic acid production. Thus the preservation is hindered by aerobic conditions and a high temperature.

Although the optimum temperatures for putrefaction and for butyric fermentation are 30° and 35° C resp., in practice maximum butyric acid and ammonia production occur in silages with maximum temperatures between 40° and 50° C.

In practice 30 to 50 % of the original protein may be lost by ammonia production in this temperature range. Although these losses will decrease with a further rise in temperature, at the same time in the range from 40—80° C the non digestible protein will increase from 20 % to 60 % of the original crude protein.

The main conclusion is that the temperature of the silage must be kept below 30° C to prevent putrefaction and/or fixation of protein.

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