AN INVESTIGATION INTO THE USE OF STARCH-GEL-UREA ELECTROPHORESIS AS A TECHNIQUE FOR STUDYING THE PROTEOLYSIS OCCURRING DURING CHEESE CURING

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A man would do nothing if he waited until he could do it so well that no one could find fault with what he has done -

Cardinal Newman.

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CONTENTS

Continued .....

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Supplementary Volume

There is a Supplementary Volume containing the original graphs that were obtained from individual gel strips and were used in building up the composite graphs appearing in the body of the main volume.

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INTRODUCTION

The protein of Cheddar Cheese makes up a quarter of its bulk, supplies its high biological value and is a major factor in regulating the characteristics of its body. Knowledge of the agents involved in converting milk casein into typical cheese protein must have value in indicating ways by which cheese quality can be improved, or alternatively indicate ways to accelerate or control the rather haphazard process of cheese curing.

Years of study into the subject of cheese protein degradation have shown the existence of a number of proteolytic agents present in cheese, viz:

1. The natural enzymes of milk.
2. The rennet enzymes.
3. Enzymes originating from the starter.
4. Enzymes originating from the adventitious flora of the cheese.

Enquiry as to the relative importance of each enzyme system has been a long and confusing process employing a variety of techniques.

Sherwood (1935) studied the changes in the various nitrogen fractions of cheeses in which bacterial numbers had been reduced by use of chloroform, but he was not able to completely eliminate the bacteria, neither distinguish between the activities of the various bacteria present in cheese, nor eliminate the effect of starter in the early period of manufacture.

Study of the characteristic enzyme systems of pure cultures of various organisms with comparison to the characteristics of enzyme systems of cheese was carried out by a number of workers (e.g. Peterson, 1948; Baribo & Foster, 1952; and Brandsaeter & Nelson, 1956), but this type of study did not yield reliable conclusions because of the variability of enzyme characteristics, even between strains of a bacterial species and further variability of results according to the conditions of the experiment. More recently, research into bitterness of cheese (Jago, 1962) has confirmed this inter-strain variability between starters and underlined the difficulty of approaching the problem, even through using pure cultures.

With the introduction of chromatographic techniques, the study of the order in which amino-acid release occurs during cheese ripening became possible. Results obtained by various workers (e.g. Dacre, 1953a; Kosikowski, 1951; Mabbitt, 1955, etc.) in different countries were not consistent with one another. However, it became apparent that different making and curing procedures were influencing the paths of proteolysis.

The tremendous complexity and variability of cheese as a medium for scientific study is apparent from the confusing and often conflicting results available in the literature. The desirability of simplifying and controlling the medium has become a necessity to basic study on this subject.

The complexity of casein itself indicates that a study of its components, rather than the protein as a whole, should provide a more fruitful approach to an understanding of the problems of proteolysis. Such an approach is possible, and has been demonstrated by Lindqvist & Storgårds (1959a) who used electrophoresis to follow the degradation of protein components during curing.

One aim of the present study is to extend the work of Lindqvist & Storgårds by the use of starch-gel-urea electrophoresis, which allows greater resolution of casein components than was achieved by these workers, Wake & Baldwin (1961).

Control of the cheese medium itself is also desirable and has been made possible by the introduction of methods of making cheeses under controlled bacteriolgical conditions by Mabbitt et al.(1959) and extended by McGillivray & Perry (1963). By application of this method, this study aims to manufacture cheeses with different but controlled bacteriological populations, and to compare the starch-gel-urea electrophoretic patterns obtained throughout their curing, in the hope that useful information may be obtained on this technique as a aid in the investigation on the role of the different proteolytic agents active in cheese curing.