





## **Enzyme Modified Cheese**



- **Enzyme-modified cheeses (EMCs) are concentrated cheese flavors produced enzymatically from cheeses of various ages and also from blends of casein, whey powder, skim milk powder, and butterfat.**
- **Used as a cheese flavor ingredient in a wide range of food products, providing a cost-effective alternative to natural cheese.**
- **Used as the sole source of cheese flavor in a product, to intensify an existing cheese taste, or to impart a specific cheese character to a more bland product.**
- **EMCs are available in a wide range of flavors, corresponding to natural cheese varieties (Cheddar, Swiss, Blue, and Parmesan).**



**EMCs are consumed  
exclusively as a  
flavor ingredient in a  
food product**



**Natural cheese is  
consumed for its flavor,  
texture, appearance,  
and functional  
properties**

# **Applications:**

## **Flavouring of....**

- ❖ **Processed cheese**
- ❖ **Analogue cheese**
- ❖ **Cheese spreads**
- ❖ **Snack foods**
- ❖ **Soups**
- ❖ **Sauces**
- ❖ **Biscuits**
- ❖ **Dips**
- ❖ **Pet foods**

**Advantages of EMCs over other sources of cheese flavor are high degree of flavor intensity, availability of a diverse range of flavors, reduced production costs, and extended shelf life.**

# Usage of Enzyme Modified Cheese

- Addition levels of EMC at 1% (w/w) have been reported for a number of applications (Romano EMC included in a tomato-based sauce, Swiss-type EMC added to frozen cheese sauce, or Cheddar EMC included in Cheddar flavored soup).
- Higher inclusion levels of 2.5% (w/w) have been reported for the application of Parmesan/Romano EMC in pizza sauce formulations.
- EMC powders have also been added to natural Cheddar cheese to accelerate ripening process.

# Technology of Enzyme-Modified Cheese Manufacture

Basis of EMC production involves addition of exogenous enzymes to cheese curd under controlled conditions to generate intense cheese flavors rapidly.

General production process involves incubating cheese with exogenous enzymes and/or microorganisms for short periods (24–72 h) at moderately high temperatures (30–45 C) and then terminating the reaction by heating (70–85 C) after the desired flavor intensity has been achieved.

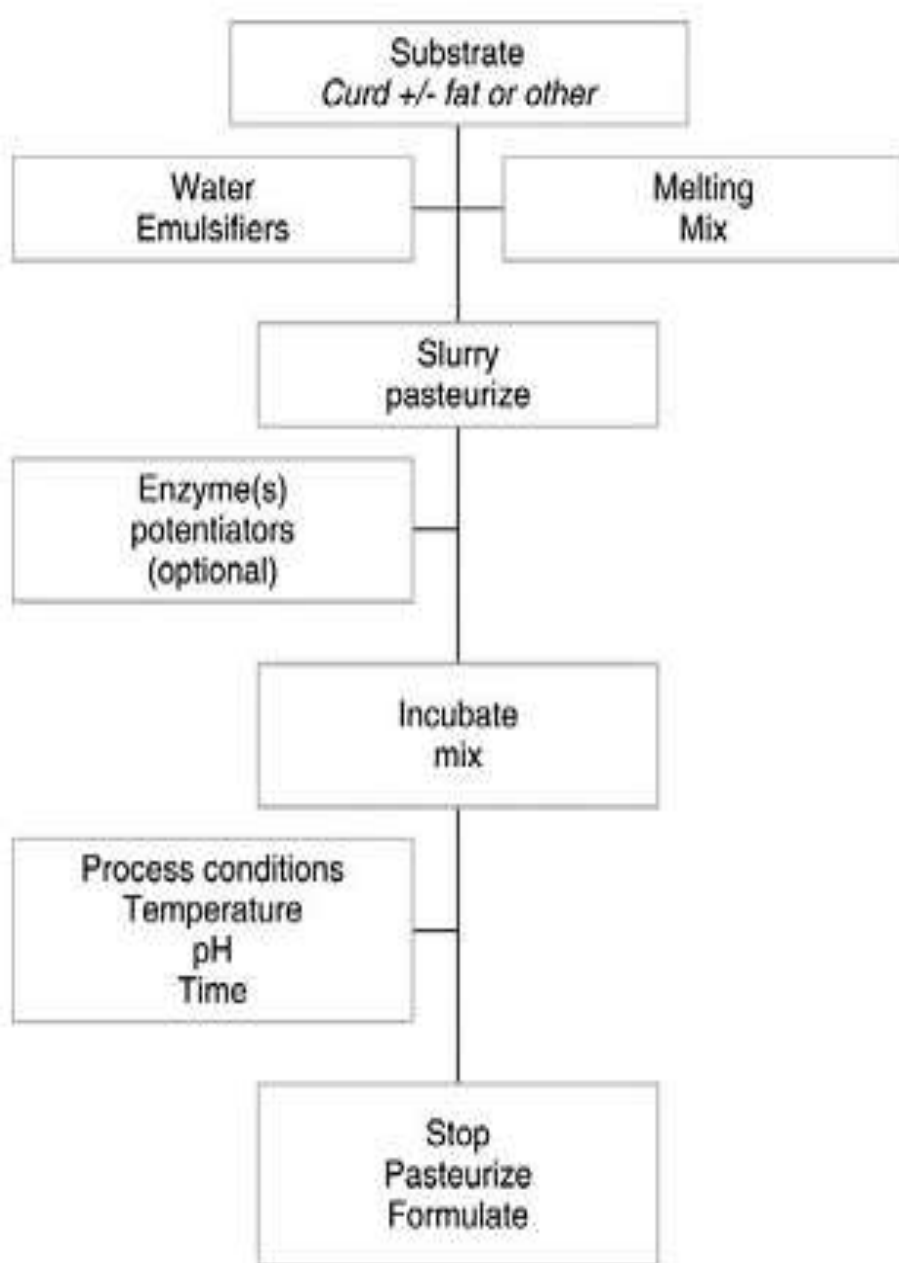
Produced by two methods: **An integrated single-step operation** in which lipolysis and proteolysis occur simultaneously and a **component approach** where several different flavor fractions are created separately and then blended to a final specifications.

- **First stage in EMC manufacture is the inactivation of enzymes in the substrate prior to enzyme reaction, generally achieved by heat treatment of curd.**
- **Consistency and quality of curd substrate are critical for flavor development.**
- **Most EMCs are produced from an immature cheese of the same type to give authentic flavor.**
- **Of particular importance in the production of EMC is the emulsification of curd substrate to semisolid paste by heating, shearing, and use of emulsifying salts. This provide heat inactivation of substrate and provide optimal conditions for enzyme–substrate interactions, thereby accelerating flavor formation.**
- **Stable emulsions for EMC manufacture are obtained by the addition of emulsifying salts, which solubilize the protein by chelating calcium.**

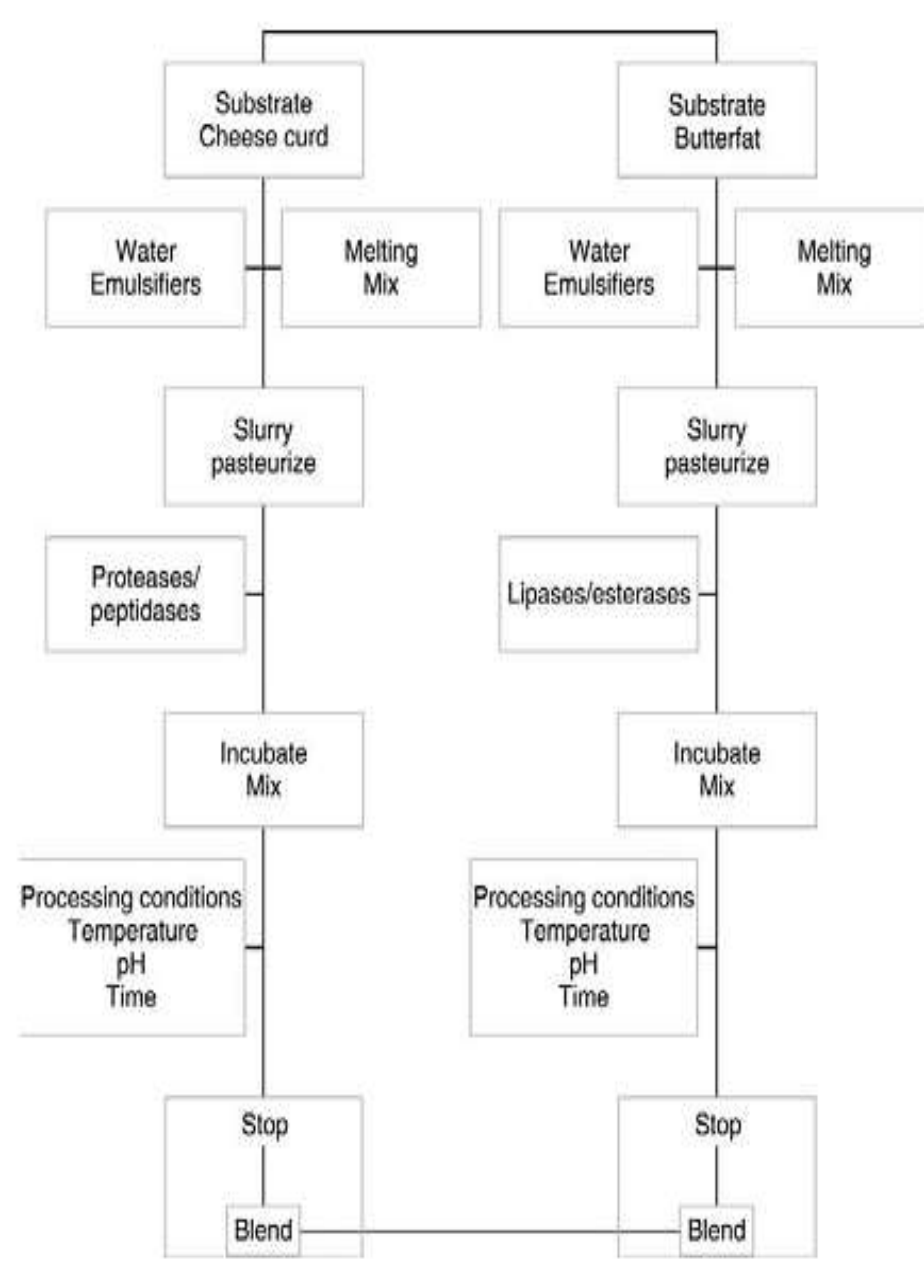
**Commonly available emulsifying salts are citrates, orthophosphates, pyrophosphates, and polyphosphates; these salts can also be used to control the pH, and some have additional bacteriostatic effects.**

**Prevention of bacterial or fungal contamination is important in producing a consistent EMC product, and potassium sorbate, nitrates and nisin are widely included in formulation to prevent growth of yeasts, molds, and bacteria.**





**Figure 1** Enzyme-modified cheese – one-step manufacturing approach.



**Figure 2** Enzyme-modified cheese – component manufacturing approach.

# Flavour Generation in Enzyme-Modified Cheese

- Flavor of EMC is principally derived from **proteolysis and lipolysis**, and degree of hydrolysis is much more extensive than what occurs during ripening of natural cheese.
- Extensive hydrolysis of protein and fat is primarily achieved through the addition of exogenous enzymes to the cheese curd.
- Proteolysis in EMC manufacture generates high levels of peptides and FAAs, which contribute to the overall background savory flavor of cheese.
- Extent of proteolysis in commercial Cheddar EMC can be up to 4 times that found in typical mature natural Cheddar cheese.
- Bitterness in EMCs is due to accumulation of hydrophobic peptides derived from  $\beta$ -casein by the action of proteinases. Bitter peptides contain 3–15 amino acids (leucine, isoleucine, proline, valine, phenylalanine, tyrosine, and tryptophan); bitterness can be alleviated by their further degradation to smaller peptides and FAAs. Removal of proline from peptide reduces bitterness by altering its three-dimensional structure, changing its solubility, and increasing its susceptibility to hydrolysis.

# Enzymes in Enzyme-Modified Cheese Manufacture

- Commercial enzymes used in EMC manufacture include proteinases, peptidases, and lipases derived from animal and microbial sources.
- Plant enzymes are not widely used in EMC manufacture due to high cost and lack of purity.
- Most microbial and animal enzymes contain other substances in addition to the active enzyme; the actual protein content may only constitute 1–5%, whereas sugars, salts, and preservatives make up the remainder of the preparation.
- Commercial enzymes are supplied as **liquid preparations**, which contain the active enzyme in a dilute solution, or as **powdered preparations with inert fillers**.

# Proteinases

- Many proteinases involved in EMC production are exclusively microbial, and most are derived from *Bacillus* or *Aspergillus* sp.
- Proteinases have different pH optima, which are used to classify them as neutral, acidic, or alkaline; however, this should not be accepted as absolute.
- Optimum temperature is a function of the reaction time used; longer the reaction time, lower the optimum temperature.



# Peptidases

- Commercially available peptidase preparations that enhance flavor and/or reduce bitterness are derived from *Aspergillus*, *Rhizomucor*, *Lactococcus* sp. and contain key endopeptidase, aminopeptidase, and carboxypeptidase activity.
- *Aspergillus* sp. tend to contain higher peptidase levels, which may minimize bitterness, a common problem in EMC production.
- EMC manufacture include endopeptidases (PepO and PepF), that cleave large casein fragments into smaller peptides.
- Most commercial peptidase preparations contain aminopeptidases (PepN and PepC) important in debittering and flavor production, as they remove amino acids from the N-terminus of peptides.

# Lipases

- **Animal lipases act almost exclusively on the sn-1 and sn-3 positions of triglycerides and thus produce butyric acid, which is an important volatile FFA associated with cheese flavor and aroma which is preferentially esterified at the sn-3 position.**
- **The most significant animal lipases are pregastric esterases (PGEs) isolated from pregastric tissues of kid goats, lambs, and calves. PGEs have an optimum pH of 5–6 and a temperature optimum of 45 C.**
- **Different PGEs produce characteristic flavor profiles: calf PGE generates a ‘buttery’ and slightly ‘peppery’ flavor; kid PGE generates a sharp ‘peppery’ flavor and lamb PGE generates a ‘dirty sock’ flavor, often called ‘peccorino’ flavor**

- **Microbial lipases tend to be cheaper than animal.**
- **The specificity of microbial lipases varies but most are sn-1 or sn-3 specific and most display a low activity toward the sn-2 position.**
- **They usually have an optimum pH of 6–9 and have temperature optimum of 45 C.**

#### Major Steps in the Manufacture of Lipolyzed Products

1. Prepare the substrate.
2. Mix the enzyme into an aqueous solution and standardize its activity.
3. Add enzyme to substrate.
4. Homogenize the mixture to promote the formation of an emulsion, which is needed to increase enzyme activity.
5. Incubate the emulsified substrate to develop desired flavor in endproducts.
6. Inactivate the enzyme.
7. Standardize the final product.
8. Package.