BREAD FLAVORANTS OBTAINED BY FERMENTATION OF CERTAIN MILK PRODUCTS

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Abstract
The obtaining of fermented flavorants by biotechnological pathway is based on the action of microbial systems on natural optimized media, to produce or potentiation of flavoring substances. Flavor compounds may be synthesized in this way, because of the complex biochemical potential of microorganisms, the rapid multiplication and their adaptation to medium conditions and the possible use in single or mixed cultures.
Fermented flavorants that are the subject of most research in this field, are: cheese (Cheddar, Roquefort, Camembert), butter, bread, with fruit or mushrooms flavor.
Fermented dairy products used as such, but also as functional ingredients, flavoring default, can be grouped as follows: traditional fermented dairy products, mainly fresh cheese and yogurt; form-modified fermented dairy ingredients (spray dried yoghurt); flavor-modified dairy fermented ingredients (enzyme modified-cheese); fermented dairy ingredients with single or mixed microbial cultures.
Bread flavorants can be obtained as by an efficient fermentation process, in the form of fermented dairy ingredients.
Sweet whey, a by-product resulting from processing milk to obtain cheese using rennet as coagulation agent, is preferred as the substrate for fermentation, because it contains important flavor precursors, such as lactic acid, lactose and lipids.
This paper presents the obtaining by biotechnological pathway of some bread flavorants, where the substrate is sweet whey alone or mixed with milk and selected microbial cultures for fermentation are those used in technology of the fermented dairy products.
Fermented dairy ingredients for flavoring bread can be used as dry or liquid form, when it only want the enhance of bread flavor.

Keywords: fermentation, bread flavors, sweet whey, sensory characteristics.

1. INTRODUCTION

Decrease of the wheat bread flavor, the most kind of bread with the consumer demand, is due to intensive development of agriculture, involving cultivation of varieties productive wheat, as well as preference for white flours obtained by advanced processing of the grains.
Aroma substances from bread are found in small quantities in the flours used as raw materials, forming them mostly on dough fermentation and baking. (Main, A. [1], Bordei, D. [2], Banu, C. [3], Thiele, C. [4], Rothe, M., Rutloff, H. [5]).
Bread flavor is as a „key” factor that influence consumer opinion on the quality and consistency of the product.
The perception of flavor is full and complex and involves three characteristics: smell, taste and texture. Each characteristic is recorded as a sensation of the specific stimulus, that act on the analyzers.
The smell of bread is due to volatile components of aroma and are analyzed by the olfactory mucous membrane. The taste (sweet, salty, sour and bitter) is sensed by the taste buds of the tongue and the texture is a tactile sensation perceived inside the mouth, influenced by the chewing quality (Iordan, M. [6] Stone, H. [7]).
Fresh bread have a pleasant smell as a yeasty, „ripe wheat” and the taste is sweet, slightly salty, sourish. The texture of crumb is soft and slightly moist, while the crust is crisp and dry.
The different kind of bread have distinct flavor, more or less intense, depending on the type of ingredients and processing methods. (Bordei, D.[2], Banu, C. [3]).
The substances responsible for flavor are largely accumulated in the crust of bread. Golden-brown color of the crust obtained from
baking is correlated with a high content of flavor substances. They are results of sugars caramelize and of the compounds formed by Maillard reactions, the main pathway to develop bread flavor.

Maillard reactions take place in stages and show a special importance, due of the many end components, derived from a small number of reagents. Initial reaction involve group (-CHO) of specific bread reducing sugars (glucose, fructose, maltose, lactose) and amino (-NH$_2$) of an amino acid to form the unstable Schiff bases, which are transformed into intermediate colourless compounds Amadori. In the following stages, through condensation, isomerization, polymerization, cyclization reactions and degradation of Amadori compounds are obtained over 100 components, inclusive melanoidin pigments. Maillard reaction involves the Stecker degradation of $\alpha$-amino acids into aldehydes (Banu, C. [3], Bordei, D. [2], Miller, A.G., Gerrard, J. [7]). In the modern technology of breadmaking, the choice of microbial starter homofermentative cultures, (L.plantarum, L.acidophylus) and heterofermentative (L. brevis, L. fermenti), of lactic acid bacteria and yeasts is particularly important. (Brümmer, J. M. [8], Dellagio, F. [9]). Therefore, fermented dairy products were considered suitable for use as ingredients in processed bread, knowing that milk is the best medium for developing lactic bacteria. This paper presents important aspects for obtaining fermented dairy ingredients used as flavorants in bread, using sweet whey as substrate.

2. SUBSTANCES OF FLAVOR AND QUALITY OF THEIR REGISTRATION

Pleasant taste and smell of bread is due to complex organic substances (~ 200 of flavor components) consisting of organic acids, alcohols, esters, carbonyl compounds (aldehydes and ketones), organic substances with sulfur, the carbonyl compounds are determinants. In the fermentation stage of dough, the aroma substances are the primary and secondary compounds produced by alcoholic fermentation of fermentescible carbohydrates. Carbonyl compounds identified are: acetaldehyde (85-95%), acetoin, diacetyl, 2,3-butanediol and propionaldehyde, isobutyrate, valerianaldehyde, glyoxal etc. Some of the aroma substances formed by fermentation, may volatilize or change during baking.

In bread baking phase is formed most of carbonyl compounds, mainly by nonenzymatic browning (acetaldehyde, furfural and isobutanal, phenylacetaldehyde, 2-hidroxy-propional). The crust contains the most pleasant of aroma substances, which give the taste and odor of bread. Content, type and quality of aroma substances depends on the product assortment of bakery and optimal parameters chosen in stages of preparation, fermentation, and especially in baking. Also, it depends on the flavor index, defined as the degree of sensory perception of various flavor substances isolated from bread, correlated with the threshold of sensitivity (minimum concentration of substance that can be perceived by olfactory analyzer and / or taste analyzer), the flavor substances being recorded differently.

Table 1 shows the correlation between some factors affecting the recording quality of aroma substances presents in wheat bread (Bordei,D.[2])

<table>
<thead>
<tr>
<th>Flavor substances</th>
<th>Concentration (ppm)</th>
<th>Threshold of sensitivity (ppm)</th>
<th>Flavor index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol</td>
<td>3900</td>
<td>10,0</td>
<td>390</td>
</tr>
<tr>
<td>3-Methylbutanol</td>
<td>14,1</td>
<td>0,3</td>
<td>47</td>
</tr>
<tr>
<td>Hydroxymethyl 1-furfural</td>
<td>10,0</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Isobutanol</td>
<td>1,6</td>
<td>0,8</td>
<td>2</td>
</tr>
<tr>
<td>Acetaldehyde</td>
<td>3,6</td>
<td>0,9</td>
<td>4</td>
</tr>
<tr>
<td>Furfural</td>
<td>0,8</td>
<td>0,04</td>
<td>2</td>
</tr>
<tr>
<td>Acetoin</td>
<td>0,75</td>
<td>0,75</td>
<td>1</td>
</tr>
<tr>
<td>Diacetyl</td>
<td>0,2</td>
<td>0,04</td>
<td>5</td>
</tr>
</tbody>
</table>
Thus, although the acetaldehyde, isobutanol, hidroxymetilfurfural are present in large amounts in wheat bread, due to high sensitivity threshold, they are less perceived in the flavor complex, than diacetyl and isobutanal substances founding in small quantities, but with a low threshold of sensitivity.

3. DAIRY INGREDIENTS USED IN BREAD PROCESSING

Documentary information presents the researches on utilization of some unfermented and fermented dairy ingredients to bread processing with a view to accumulating and stimulating of flavor substances. Skimmed milk used to obtain of the bread improves quality by: increased capacity to absorb water, decrease the duration of fermentation, forming a golden-brown color of the crisp and bread flavor enhance (Vetter, J.L. [10], Pyler, E.J. [11]).

Yoghurt, milk product with certainly health benefits used in the loaf formulation, emphasize the positive effect on the flavor, but in the same time, decrease the product volume (Hill, L.G. [12]). The incorporation of acid whey by-product from obtaining Cottage cheese in bread dough rise to the change flavor. The finished product has a pronounced taste and smell of fermented dough. (Shenkenberg, D.R., et al [13]). Fermented dairy ingredients with single or mixed microbial cultures used as flavorings have positively influence in the process of obtaining bread, through shorten duration of processing, respectively of the length of fermentation (Main, A. [1], Marshall, V.M.E. [14]).

4. BREAD FLAVORANTS PRODUCED BY BIOTECHNOLOGICAL PATHWAY FROM THE SWEET WHEY

Sweet whey is a by-product from processing milk to obtain cheeses, using rennet as coagulation agent. Sweet whey prior to recovery as a flavor concentrate is subjected to some treatments (centrifugal separation to recover fat, pasteurization before cultivating with bacteria and/or yeast, deproteinized to protein remove), for avoid the formation of unpleasant flavors. Microbial cultures selected for whey fermentation technology are used in fermented dairy products (Banu, C., et al [15], Gilliland, S.E. [16]) and include:
- Mesophilic lactic acid bacteria for the manufacture of cheese and butter obtaining, producing good flavor and less acidity (Leuconostoc cremoris, Streptococcus lactis subsp. Lactis, S. lactis subsp. diacetylactis, S. lactis subsp. cremoris);
- Thermophilic lactic acid bacteria with special purpose for yoghurt (L. bulgaricus, S. thermophilus) or to other fermented dairy products. Depending on the ability to produce lactic acid, thermophilic cultures contain optional heterofermentative lactic acid bacteria species (L.casei subsp. rhamnosus) or homofermentative (L. helveticus, S. thermophilus). These bacteria are good producing lactic acid and develop the flavor components such as acetaldehyde and diacetyl.

Bread flavorants obtained by the sweet whey fermentation, may be used in concentrated dry form or liquid form at low concentrations, when are used only as a flavor enhancer. Sweet whey is preferred as the substrate for fermentation, because it contains important as flavor precursors, such as, lactic acid, lactose and lipids. Fermentation conditions (temperature, duration, mode of agitation) affect the quality of flavor concentrates. Conditions of technological process are determinative to stimulating the develop of aroma compounds and concerns to: the consistency of semi-finished, duration and temperature of fermentation, baking time and temperature.

Thus, the conditions favorable to accumulation of aroma substances are (Bordei, D. [2] Marshall, V.M.E. [14]):
- Use of the semi-finished with a reduced consistency, and particularly, of the fluid leaven;
- Choosing a higher fermentation times (using three-phase process) and a higher temperature than ~ 35°C;
- Establishing a longer baking time (for it prefers bread with greater mass) and a sufficiently high temperature, which promotes
an increased rate of Maillard, respectively nonenzymatic browning reaction.
A bread flavored concentrate was obtained by inoculating whole sweet whey with a culture of lactic acid bacteria, containing S. lactis subsp. determination of volatile compounds compared to a control sample was observed a significant increase of benzaldehyde (650%), of the acetic and propionic acids and aliphatic alcohols. The lactones identified in crust and crumb have not been notable changes (Mulders, E.J. [17]).
Another bread flavored concentrate in powder form was obtained by a double fermentation of deproteinized sweet whey (a high-protein content affect negative bread volume). Whey was inoculated with single or mixed lactic bacteria cultures that include: S. lactis, L. helveticus, L. leichmanni, S. citrovorum, S. cremoris.
Duration of fermentation was 12-14 hours at a temperature of 35°C, under aerobic conditions. This fermented medium was then inoculated with yeasts Saccharomyces cerevisiae or S. unisporum, which grow well on medium containing lactic acid, under aerobic conditions and under strong stirring. After the final fermentation it was added a thickening agent (starch) and was dried by spray (Banu, C., et. at. [18]).
Obtaining of a bread flavoring from sweet whey was the objectiv of the research of Gellinas, P. and Lachance, O. ([19], [20]).
The dairy ingredients used were skimmed milk (100%), milk-whey mixture in different proportions (75%/25%; 50%/50%; 25%/75%) or sweet whey (100%), supplemented with sodium citrate (1%).
For medium fermentation it was selected a singular (L. casei subsp. rhamnosus) or mixed starter culture (L. casei subsp. rhamnosus + L. helveticus / S. thermophilus).
The fermentation was conducted in aerobic conditions, under shaking (140 rpm) for 24 hours at a temperature of 38°C, respectively, 42°C.
Fermented dairy ingredients were added in 26% proportion of bread dough, prepared after established recipe.
Determination of volatile components of dairy ingredients and bread, correlated with the assessment flavor intensity and pleasant „type cheese” showed that the best results were diacetylactis and S. lactis subsp. cremoris). The medium was fermented at a temperature of 28°C, for 18-24 hours, under aerobic conditions. The product has been added at a rate of 18-20% in bread dough. Following obtained under the following conditions:
- Medium for fermentation: 50%milk/50%whey;
- Optimal culture: mixed culture (L. casei + L. helveticus / S. thermophilus);
- Fermentation temperature: 38°C;
- Substances responsible for flavor in bread obtained with fermented dairy ingredients: lactic acid, ethanol, diacetyl.
Bread prepared with fermented dairy ingredient obtained from equal mixture by whey and skimmed milk, under experimental conditions, has a slightly lower volume than the control bread.
Bread flavor concentrate can be dried and used at a dose up to 10% compared with the dough, when we desire the flavoring of bakery product or 1-2% for the enhance of bread flavor.

5. REFERENCES