

## Technical Report: Sensory Properties of Whey Ingredients

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In the world of food and beverage manufacturing, no one factor holds more weight in the success of a new product than taste. Whey proteins are valuable in helping product developers achieve functional and nutritional properties in new and reformulated products. Ensuring proper handling and processing techniques can maximize whey's sensory properties.

This report summarizes recent research on improving the sensory properties of whey protein ingredients to help product developers utilize them more successfully in various food and beverage applications. Many different whey ingredients have been developed over time and are used widely in foods for their functional properties such as water binding, gelling, foaming, browning and solubility.<sup>1</sup> Whey protein ingredients also are desired for their nutritional properties, such as a high level of essential amino acids — in particular, branched chain amino acids. Their rich nutritional value has given whey protein ingredients more opportunities to be used as a food ingredient in protein-enhanced foods such as nutrition bars and beverages.

Just as research has been completed to improve the functional properties of whey protein ingredients, research has been conducted in the sensory field, as well. The Dairy Research Institute<sup>®</sup>, established under the leadership of America's dairy farmers through the dairy checkoff program, has supported research to help processors and end users of whey protein ingredients improve the flavor and aroma and reduce product barriers to increase utilization of whey protein ingredients.

# Characteristics of Whey Flavor

## **Whey Composition**

Whey is the liquid substance obtained by separating the coagulum from milk or cream in cheese making. Acid whey is obtained from a process in which either a significant amount of lactose is converted to lactic acid or from curd formation by direct acidification of milk. Sweet whey is obtained from a process with insignificant conversion of lactose to lactic acid.<sup>2</sup>

Most commercial whey ingredients are made from sweet whey, which is a coproduct of cheese varieties like Cheddar and mozzarella, among others. Based on USDA cheese production data, the U.S. Dairy Export Council® estimates that more than 95 billion pounds of sweet whey (liquid basis) would have been produced during 2011 compared with about 4 billion pounds of acid whey. Whey composition is affected by many factors, such as the original milk composition, type of cheese, cheese processing conditions, whey processing conditions and finally storage of the powder (Figure 1). Whey flavor is also influenced by all of these factors.

Liquid whey typically contains 93 percent water, 0.8 percent protein, 0.3 percent fat, 4.8 percent lactose and 0.5 percent ash.<sup>3</sup> Liquid whey is made into a variety of commercial ingredients from dried whey (13 percent protein) to whey protein concentrates (25 to 89 percent protein) and whey protein isolates (>90 percent protein). The protein, fat and lactose are all susceptible to chemical changes during production and storage that may influence the flavor of the final ingredient.



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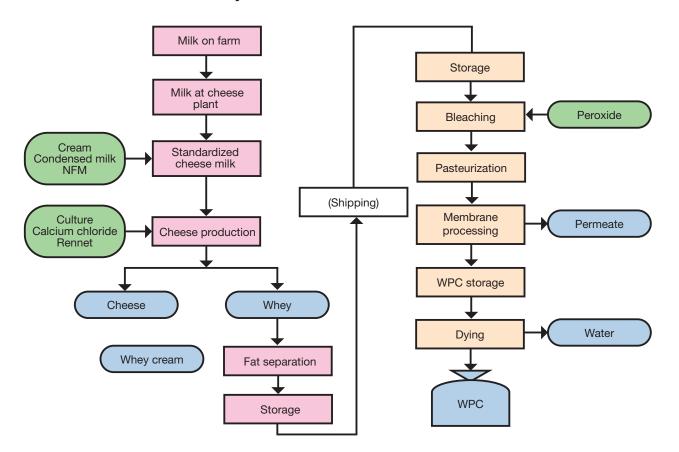
## **Flavor Characteristics of Whey Ingredients**

Food and beverage manufacturers are boosting the protein content of products to tap into the growing health and wellness trend. Many food and beverage manufacturers use whey proteins as the protein source of choice for product innovation and prefer to have whey protein with neutral flavor. Flavor and aroma of whey proteins can be a barrier for incorporating the product in food and beverage innovations, limiting the use of whey protein ingredients in various food formulations due to the presence of certain whey off-flavors.<sup>4</sup>

Current research has focused on the processing conditions (Figure 1) that influence off-flavor and aroma development in whey. This understanding has enabled manufacturers to improve flavor. Though most whey ingredients have a clean, bland flavor after drying, they are known to develop off-flavors resulting from lipid oxidation and Maillard browning.<sup>4</sup> Using sensory and chemical analysis, researchers have identified many different flavor attributes and their chemical compounds in whey ingredients.<sup>5-18</sup> Some of the sensory attributes associated with liquid whey include musty, metallic, sour, salty, diacetyl, astringency, cooked, sweet, milkfat and cardboard.<sup>5</sup> Attributes associated with whey protein concentrates (WPCs) and isolates include sweet aromatic, cardboard/wet paper, animal/wet dog, soapy, brothy, cucumber, cooked/milky flavors, bitter and astringent.<sup>6,7</sup>

Approximately 200 chemical compounds have been identified in liquid and dry whey products that may influence their final aroma and flavor.<sup>7,11</sup> Table 1 shows some of the sensory attributes and their corresponding chemical compounds in whey. Research has helped to determine the sources of these chemical compounds starting with the original milk that is used for cheese making, through whey processing and storage of whey after drying.<sup>5-18</sup>

#### Figure 1.



#### Cheese and whey manufacture. K. Smith, Wisconsin Center for Dairy Research

## Parameters That Contribute to Whey Flavor

Fresh whey has many flavor characteristics that are desirable, but typical processing and ingredient storage conditions can lead to the development of undesirable off-flavors. These off-flavors can carry through into the finished food application and negatively impact its flavor profile. Researchers are working to understand the variables, such as cheese type, starter culture, fat content in the whey, bleaching, handling and storage, that contribute to the development of off-flavors.

## **Cheese Type and Starter Cultures**

Lipid oxidation, the main cause of whey off-flavors, increases during the fermentation process of cheese making. The type of starter culture used to make mozzarella or Cheddar cheese ultimately influences the flavor profile of cheese whey. Cheddar whey is more prone to lipid oxidation than mozzarella whey, resulting in a higher concentration of rancid flavors such as sour aromatic and cardboard notes. Researchers believe that mesophilic starter cultures (used in Cheddar manufacturing) initiate the oxidation process and produce more rancid off-flavors compared with thermophilic starter cultures (used in mozzarella manufacturing).<sup>11,12</sup>

Whey protein concentrate manufactured from whey with starter cultures has more prevalent cardboard flavor and higher lipid oxidation products (hexanal and heptanal) compared with WPC manufactured from cheese without use of starter. Fluid whey made with starter cultures had higher intensities of aroma, sweet aromatic, buttery and sour aromatic flavors compared with fluid whey made with direct acidification. During storage, the sweet aromatic and cooked notes disappeared and the whey developed cardboard and oxidized notes, suggesting a link between starter culture and lipid oxidation compounds.<sup>11,12</sup> These findings help manufacturers select certain strains and cultures that produce the least amount of lipid oxidation compounds.

| Compound                                 | Odor Description | Compound                                 | Odor Description   |
|--|------------------|--|--------------------|
| Acetic acid                              | Vinegar          | 2-Methyl-3-furanthiol                    | Brothy/burnt       |
| Dimethyl sulfide                         | Garlic/rubbery   | 2-Methoxy phenol                         | Smoky              |
| Hexanal                                  | Green grass      | Nonanal                                  | Fatty/citrus       |
| Butanoic acid                            | Cheesy/rancid    | 3-Hydroxy-4,5-dimethyl-2(5H)<br>furanone | Maple/spicy        |
| Methional                                | Potato           | (E,Z)-2,6-Nonadienal                     | Cucumber           |
| 2-Acetyl-1-pyrroline                     | Popcorn          | (E)-2-Nonenal                            | Cucumber/old books |
| Dimethyl trisulfide                      | Cabbage          | Decanal                                  | Fatty              |
| Octanal                                  | Citrus/green     | O-Aminoacetophenone                      | Grape              |
| Hexanoic acid                            | Sweaty           | (E,E)-2,4-Decadienal                     | Fatty/oxidized     |
| 2,5-Dimethyl-4-hydroxy-3(2H)<br>furanone | Burnt sugar      | Γ-Nonalactone                            | Coconut            |

#### Table 1. Some aroma compounds and odor descriptions of Whey Protein Concentrate (WPC) and Whey Protein Isolate (WPI).

Adapted from Carunchia Whetstine ME, et al., 2005.6

## Fat Content and Use of Antioxidants

The lipid concentration in liquid whey also has an impact on flavor and volatile lipid oxidation products in dried whey protein ingredients. The fat content of milk used to make cheese determines the lipid concentration of whey. The WPC80 from cheese made with whole milk has higher concentrations of lipids, stronger cardboard flavor and a greater abundance of lipid oxidation products compared with WPC80 from cheeses made with low-fat or skim milk. Separating fat from pasteurized liquid Cheddar whey prior to further processing decreases, but does not eliminate, lipid oxidation compounds that lead to off-flavors in whey.

Use of ascorbic acid (0.05 percent) or whey protein hydrolysate (0.05 percent) in pasteurized whey resulted in lower concentrations of the oxidation compounds such as pentanal, heptanal and nonanal than the control WPC and less off-flavors and aroma.

## Handling and Storage

Oxidation in both milk and whey increased with storage time.<sup>9</sup> In a related study, researchers evaluated the effect of holding time of liquid retentate (liquid whey concentrated by ultrafiltration and microfiltration) on the flavor of WPC80 and WPI.<sup>8</sup> Liquid WPC80 and WPI were stored at 3 C for up to 48 hours and then spray-dried. Powders were stored at 21 C and evaluated every four months for 12 months of total storage. Cardboard flavors increased in both spray-dried products with longer retentate storage time, while cabbage flavors increased in WPI. Hexanal, heptanal and octanal (lipid oxidation products) and dimethyl sulfide and dimethyl trisulfide (sulfur degradation products) increased in the dry products with increased retentate storage time, while diacetyl decreased. Flavor stability of the stored dry products also decreased with increases in retentate storage time. Researchers determined that liquid retentate should be held for less than 12 hours prior to spray-drying for maximum flavor quality and shelf stability.

In two other studies, pH history of liquid whey was found to affect Maillard browning in whey and whey protein powders.<sup>9,10</sup> Liquid whey contains active cultures that will continue to grow and produce lactic acid unless the whey is pasteurized and/or cooled soon after it is separated from the curd. Researchers found that the increase in acidity (pH 6.3 to 4.9 and 4.2) caused by warmer temperatures and longer holding times produced intermediates of the Maillard browning reaction (3-deoxyosone), which later resulted in accelerated browning of the whey or whey protein ingredient. Maillard products resulting in off-flavors predominate in whey powder and WPC34, containing a higher concentration of lactose. Lipid and protein-related off-flavors are prevalent in WPC80 and WPI.

## **Bleaching**

Another variable in whey processing is bleaching of colored Cheddar whey. Annatto is a yellowish-orange natural color that is used to make colored Cheddar cheese. Annatto contains the carotenoids bixin and norbixin. Studies suggest that norbixin (water-soluble) is able to bind with  $\beta$ -casein or  $\beta$ -lactoglobulin to form a stable complex that prevents easy removal of the annatto. This colorant is not all retained in the cheese; approximately 20 percent of annatto added to cheese milk passes into whey, which is highly undesirable from a visual standpoint.

As a result, whey is often bleached to remove the yellow color. Hydrogen peroxide (HP) and benzoyl peroxide (BP) are legally permitted bleaching agents in the United States. More information on the bleaching process and conditions of use are available within industry publication and research.<sup>16,19</sup> A research study added HP at 250 and 500mg/kg and BP at 10 and 20mg/kg to liquid whey for 1.5 hours at 60 C and cooled to 5 C.<sup>20</sup> Researchers evaluated the flavors of the treated liquid wheys and a control whey with no added bleach. Of the two bleaching agents, BP is more effective in bleaching annatto and results in whey proteins with fewer off-flavors when compared with HP. The liquid whey and WPC with added HP had higher concentrations of heptanal, hexanal, octanal and nonanal compared with the unbleached or BP bleached liquid whey or WPC. Fat and cardboard flavors also were higher in the HP liquid whey and WPC. The addition of HP resulted in a whiter WPC with more off-flavors, but there was no difference in the norbixin recovery between HP and BP. Bleaching appears to impact the flavor of whey and the type of bleaching agent used also has an effect.

A similar study compared the effects of bleaching at cold or hot temperatures, bleaching before or after fat separation, and bleaching agent on whey flavor.<sup>21</sup> Hot bleaching was the main variable that was found to contribute to increased lipid oxidation in whey and subsequent off-flavors.

Other recently published research studied the partitioning of annatto in whey between the protein, milk fat globule membrane (MFGM) and the serum phase.<sup>22</sup> Researchers found that annatto in Cheddar WPC is distributed between the MFGM and serum phase. They used a novel method of removing the MFGM followed by filtration to provide an alternative to bleaching and prevent the off-flavor formation in the dried WPC caused by bleaching agents.

## Astringency

Astringency is not a flavor, but rather a drying sensation in the mouth that is typically associated with beverages like wine or tea. It is thought to be caused by compounds in foods that bind with and precipitate salivary proteins.<sup>23</sup> Astringency is an important consideration for whey protein ingredients as it is a sensory property of whey proteins, especially in acid conditions. Similar to the issues with off-flavor of whey protein ingredients, astringency of whey protein beverages has been noted as a barrier to mainstream acceptance of higher-protein acidic beverages.

Researchers have examined the relationship between protein concentration, pH, acidity, viscosity and charge interactions as possible mechanisms for the perception of astringency. Research sponsored by the Dairy Research Institute explored mechanistic causes of astringency and consumer perception of astringency in high-protein acidic beverages. In one study conducted at University of Minnesota, researchers examined the overall acidity of whey protein beverages as a cause of astringency.<sup>26</sup> Whey protein has a high-buffering capacity. The higher the protein concentration, the more acid is required to reduce the pH to levels in the high acid pH range of 3 to 4.5. This research hypothesized that solutions with equal acid concentration would be equally astringent, regardless of whey protein content or pH. Acid solutions were made with and without whey proteins with 1 and 6 percent whey protein isolate and phosphoric acid at a pH of 3.4. The acid-only solutions were made to match either the acidity or pH of whey protein solutions. Because the acid-only solutions equal in total acidity to the whey protein solution were more astringent, researchers believed that astringency of acidic whey proteins was caused by their high acidity and not directly by the whey proteins.

Researchers at North Carolina State University proposed that astringency was related to interactions between positively charged whey proteins and negatively charged saliva proteins. In sensory comparisons of whey protein beverages containing WPI at 6 percent protein at pH 6.8 and 3.4 showed that the beverage at pH 3.4 was more astringent.<sup>24</sup>

More recently, beverages containing 0.25 to 13 percent  $\beta$ -lactoglobulin and 0.017 percent sucralose at pH 2.6 to 4.2 were compared with phosphate buffer controls at similar pH and phosphoric acid concentration for their sensory properties.<sup>27</sup> Astringency significantly increased with protein concentration from 0.25 to 4 percent at pH 3.5, but it remained constant from 4 to 13 percent protein. Astringency decreased at pH 2.6 with an increase from 0.5 to 10 percent protein. These results suggest a complex relationship with pH and the buffering capacity of the beverages. Saliva flow rates also were measured and found to rise with increasing protein concentration, suggesting a physiological change also occurred in the mouth.

In a related study, the role of protein charge was evaluated for its effect on astringency. Beta-lactoglobulin ( $\beta$ -lg )(4 percent solutions) and phosphate buffer solutions at pH 3, 4 and 6 were compared. The research concluded that the protein-containing solutions were more astringent and that proteins, not acid alone, contributed to astringency.<sup>28</sup> In the same study, 4 percent lactoferrin (positively charged at pH 7.0) and WPI solutions at pH 3.5, 4.5 and 7.0 were compared. The astringency of lactoferrin was higher at pH 7.0, while the WPI solutions decreased in astringency at pH 7.0. These results were explained by the positive charge on lactoferrin at pH 7.0 interacting with the negatively charged saliva proteins — the negatively charged whey proteins did not interact. Further evidence to support the charge interaction theory was evident when solutions of lactoferrin,  $\beta$ -lg and salivary proteins precipitated when they were mixed at the same conditions.

These proteins did not precipitate individually. In sensory comparisons of whey protein, beverages containing WPI at 6 percent protein at pH 6.8 and 3.4 showed that the beverage at pH 3.4 was more astringent.<sup>24</sup> Previous whey protein research showed astringency increased with protein concentration. Researchers proposed that this concentration caused astringency through aggregation and precipitation of protein molecules in the mouth.<sup>25</sup>

Another study evaluated the consumer acceptance of acidic whey protein beverages. Consumers were asked to assess fruit-flavored whey protein drinks with 6 percent protein (WPI), citric acid and sucrose at pH 3.4.<sup>29</sup> Consumers were able to identify astringency in the drinks, but it was not directly linked to dislike. Overall acceptance of the beverages was lower when the consumers were not wearing noseclips compared with acceptance scores for the beverages when consumers were wearing noseclips. These results suggested that the flavor/aroma combination of the WPI influenced the overall drink acceptability more than the astringency alone.

# Techniques to Formulate With Typical Whey Flavors — Applications

Research has helped identify the flavors, their sources and how processing conditions can reduce the off-flavors and aromas in whey ingredients. The whey processing industry can improve the flavor and aroma of their ingredients by incorporating these changes in how they process their whey. Many processors have already improved the flavor of their whey and whey protein ingredients. Increasing acceptance of whey proteins in a greater number of mainstream products is evidence of these improvements. Research efforts will continue to address the sensory properties of whey protein as finished products are fortified with increasing levels of whey protein.

Currently, beverages pose the greatest challenge to minimize whey flavor and aroma. Because of the popularity of thirst-quenching drinks such as sports drinks and juices, companies will continue to want to use whey proteins for their heat stability at pH less than 3.5. Because most drinks in this category don't include protein, whey aroma and flavor of WPC and WPI are not typical characteristics of these drinks. The added astringency at low pH also is unwelcomed by some consumers. Formulation tips to minimize these sensory attributes of whey protein ingredients include:

• Flavor selection: Because many of these drinks are fruit-flavored, it is important to note that the tropical flavors, such as mango, pineapple and coconut as well as other flavors like citrus, peach and apple work well with whey protein ingredients and will mask whey flavor and aroma. The berry flavors (strawberry, raspberry, blueberry, etc.) are some of the most difficult to use with whey protein ingredients because they don't mask whey flavor and aroma as well.



- **pH level:** When it comes to masking astringency, increasing the pH above pH 3.5 decreases astringency, but then heat stability becomes more challenging and clarity decreases.
- **Complementary ingredients:** Adding larger carbohydrates such as soluble fiber also may decrease astringency, but no formal studies have been conducted. Flavor companies also have developed masking flavors to help reduce whey flavor, aroma and astringency.

The Dairy Research Institute is focused on finding ways to minimize lipid oxidation of whey and researching alternatives to bleaching because it was identified as one of the primary contributing factors to off-flavor development in whey protein. These research efforts will continue to provide technical solutions so that food formulators can develop innovative foods and beverages using whey protein ingredients. Product developers and consumers can look forward to further improvements in the flavor, aroma and astringency of whey ingredients as science-based research from the Dairy Research Institute continues to offer commercial solutions.

For more information regarding the sensory characteristics of whey ingredients and applications, visit: InnovateWithDairy.com, www.usdec.org or USDairy.com/DairyResearchInstitute. For assistance with new or improved products using dairy ingredients, contact Dairy Technical Support at techsupport@InnovateWithDairy.com.



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