Gelatin Filtration

Introduction

Gelatin, a high grade protein derived from animal collagen, is used in numerous products in food, beverage, pharmaceutical, photographic and industrial markets. Some edible uses for gelatin are gelatin desserts, gummed candies, marshmallow, wine fining or clarification and prepared meat products. Among the pharmaceutical uses are hard and soft capsules, stabilizers for oil emulsions and glycerinated gelatin for suppositories. In the photographic industry, gelatins are used for paper coating and as a component in silver halide emulsion coatings. Other technical applications would be for micro-encapsulation, bacteriological culture media and in emulsion polymerizations.

Commercial grade gelatin contains 84-90% protein, 8-12% water, and 2-3% minerals. The protein portion is comprised of 18 different amino acids, including 9 of the 10 essential amino acids required for human life. The amino acids are joined in polypeptide chains. The secondary structure if formed when three chains join together in a left-hand helix. In the tertiary structure, refolding into a right-hand triple helix results in rod-shaped molecule also called a proto-fibril.

The methods and extent of processing steps are dependent on the final use of the gelatin. Pharmaceutical grade gelatin typically requires further refinement to meet specific quality standards compared to industrial grade gelatin.

This Customer Application Brief will address the use of disposable filters to produce commercial grade gelatin, as well as the benefits derived from their use. These include:

- Enhanced gelatin clarification to meet product turbidity specifications and maintain high bloom values.
- Control of microbiological contaminants to increase stability.
- Reduction of raw material contaminants.

The Process

Two main processes are used to make gelatin, an acid process and an alkaline process. Raw materials can include bovine bones and hides, pig hides, and fish skins. Bovine bones are treated with acid, followed by an alkaline treatment, while bovine skins only receive an alkaline treatment. Pig hide and fish skin only receive an acid treatment.

The basic process begins with crushing and thorough degreasing stages to remove soft tissue and fat. This is followed by lengthy treatment with hydrochloric acid (or lime, as with bovine hides), and multiple extractions in hot water. The resulting
material is then purified via one or more filtration steps along with ion exchange separation step. The solution is then further concentrated using evaporation or other methods, sterilized, extruded into “noodles” dried, and milled into powder form. The schematic (figure 2) represents a generic process using any of the four common raw materials. As mentioned earlier, the number of filtration and refining steps can vary according to the final use of the gelatin.

The resulting gelatin molecule can carry either a negative or positive charge depending on the molecule isoelectric point and the pH of the gelatin application. The isoelectric point (the pH at which the gelatin solution is neutral) is determined by the manufacturing process used. With acid treatments, pig skin gelatin typically has an isoelectric point between pH 8.0-9.0. While in alkaline treatments, the resultant isoelectric point is typically in the 4.8 to 5.4 range.

**The Problems**

The end properties of commercial grade gelatin are determined in part by the amino acid sequence of the molecules, but also by the quality of the gelatin process itself, especially where purification and filtration steps are concerned.

Final application conditions, such as temperature, pH, and ionic strength among others, will ultimately determine the performance of the gelatin.

Gelatin quality is measured using a number of parameters. Chief among them are: the bloom value, viscosity, color, clarity, melting point, isoelectric point, ability to foam, particle size, and microbiological quality.

The bloom value, the most common specification for gelatin, is a measurement of the strength of a gel formed by a 6 2/3% solution of the gelatin that has been kept in a constant temperature bath at 10° C for 18 hours. High-bloom value gelatins are desirable for most applications since they are generally lighter in color, less gelatin is needed for a given task, they solidify more rapidly, have better odor and taste, and they have higher melting and solidification points. Viscosity is also measured under these conditions an disassociated with the bloom value for a given gelatin.

Various other analyses are performed on the final product to ensure purity including: microbiological content, inorganic ash, clarity, moisture content and heavy metal content.

Poor filtration will negatively affect many of these parameters, resulting in either down grading the end product, or costly reprocessing until the desired parameter is achieved.

Filtration is employed on two levels. The first is to reduce larger unwanted particles native to the raw material. These include remaining bone fragments, undissolved hide, and other foreign matter. The second is to reduce unwanted colloids, bacteria and other microorganisms, and turbidity-causing matter.
The 3M Solution
Zeta Plus™ filter sheets and cartridges are ideally suited to clarify and stabilize gelatin across a wide range of end products. Zeta Plus H series filters are composed of inorganic filter aids (diatomaceous earth, perlite) in a high wet-strength cellulose fiber matrix. The filter media is bound by resin that not only contributes to the durability of media under demanding process conditions, but also imparts a positive charge to the internal filter surfaces. This positive charge attracts and captures negatively charged debris (electrokinetic adsorption), such as bacteria, particles, contaminating colloids, allowing for more efficient removal of submicron particles than mechanical sieving alone would afford.

Zeta Plus H series filter media are available in a wide range of grades to match individual process goals. Mechanical straining alone, as measured by mean-flow-pore analysis, is indicated in Figure 3. Particles smaller than the rated pore size can be removed through electrokinetic adsorption. Process conditions, such as pH, particle charge, and flux, play a role in how effective this mechanism is. The wide range of reduction ratings available allow processors to select the appropriate grade for a given application. Technical support with grade selection and application optimization can be obtained from the 3M Purification Inc. Scientific Applications Support Services (SASS).

Zeta Plus filter media demonstrate excellent microbial reduction as noted in Table 1. In these experiments, three organisms were used to evaluate the ability of various Zeta Plus H series grade filter media to remove microorganisms of different sizes. Saccharomyces cerevisiae, common yeast, Leuconostoc oenos, medium size lactic acid bacteria, and Brevundimonas diminuta, very small bacteria, were used in these studies. As the data demonstrate, Zeta Plus filter media are very effective in removing contaminating microorganisms. These data can also be interpreted to infer that contaminating particles of similar size (bone dust, undissolved colloids, and other turbidity-causing contaminants) can also be successfully reduced with Zeta Plus filter media.

![Figure 3 — Mechanical Straining](image)

**Table 1 — Zeta Plus™ Filter Media Microbial Reduction**

<table>
<thead>
<tr>
<th>Zeta Plus H Series Media Grade</th>
<th>Microorganism</th>
<th>Reduction (CFU/cm² of filter media)</th>
<th>Organisms in Filtrate</th>
</tr>
</thead>
<tbody>
<tr>
<td>30H</td>
<td><em>Saccharomyces cerevisae</em></td>
<td>4.1 x 10⁶</td>
<td>0</td>
</tr>
<tr>
<td>50H</td>
<td><em>Saccharomyces cerevisae</em></td>
<td>6.8 x 10⁶</td>
<td>0</td>
</tr>
<tr>
<td>60H</td>
<td><em>Saccharomyces cerevisae</em></td>
<td>6.0 x 10⁶</td>
<td>0</td>
</tr>
<tr>
<td>60H</td>
<td><em>Leuconostoc oenos</em></td>
<td>5.5 x 10⁵</td>
<td>0</td>
</tr>
<tr>
<td>90H</td>
<td><em>Leuconostoc oenos</em></td>
<td>7.2 x 10⁵</td>
<td>0</td>
</tr>
<tr>
<td>60H</td>
<td><em>Brevundimonas diminuta</em></td>
<td>9.7 x 10⁵</td>
<td>0</td>
</tr>
<tr>
<td>90H</td>
<td><em>Brevundimonas diminuta</em></td>
<td>1.3 x 10⁶</td>
<td>0</td>
</tr>
</tbody>
</table>

Challenge conditions: 10⁶ – 10⁷ CFU/ml in challenge solution; flux: 0.25 GPM/ft²
Conclusion

The quality of commercial grade gelatin is determined by various technical attributes including the bloom value, turbidity, viscosity, and purity. Contaminating materials such as bone fragments, hair/hide particles, undissolved colloids and microorganisms, diminish gelatin quality resulting in down grading of the final product, or time consuming reprocessing until the desired quality standard is achieved.

Whether deployed in a standard filter sheet, or an easy to use filter cartridge, Zeta Plus™ H series filter media provide excellent retention of contaminating particles and microorganisms in various grades of gelatin. This Customer Application Brief addressed the use of Zeta Plus series filters to produce commercial grade gelatin, as well as the benefits derived from their use. These include:

- Enhanced gelatin clarification to meet product turbidity specifications in maintain high bloom values.
- Control of microbiological contaminants to increase stability.
- Reduction of raw material contaminants.

Scientific Applications Support Services

The cornerstone of our philosophy is service to customers, not only in product quality and prompt service, but also in problem solving, application support and in the sharing of scientific information. The Scientific Applications Support Services (SASS) group is a market-oriented group of scientists and engineers who work closely with customers to solve difficult separation problems and aid in the selection of the most effective and economical filtration systems. 3M Purification Inc. offers specialized support to the pharmaceutical and biotechnology industry through our Validation Support Services Program. SASS routinely provides end-users with:

- Validation And Regulatory Support
- Extractable and Compatibility Analysis
- Filter System Optimization Studies

For more information regarding our Validation Support Services, please contact 3M Purification Inc. Technical Services or your local Distributor.