

MICROBIOLOGY OF EGGS¹

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ABSTRACT

It is generally agreed that milk and eggs are nature's most perfect foods. Milk is extremely perishable while eggs in the shell are not. The reason that eggs are not as perishable as most foods is that they possess protective devices including the cuticle, shell, shell membranes, lysozyme, conalbumin, avidin, pH of the albumen, etc. Having these protective devices is nature's way of allowing reproduction of birds. Chicken eggs, for example, take 21 days to hatch under ideal conditions for growth of microorganisms. Without the protection, reproduction could not take place. Once eggs are broken out of the shell and mixed, they are as perishable as milk. Pathogenic bacteria of concern to humans found in eggs (particularly liquid eggs) are *Salmonella*, *Staphylococcus aureus*, and *Clostridium perfringens*. These pathogens are not common in shell eggs but can be found in liquid eggs primarily because of reinfection by humans.

It is generally agreed that milk and eggs are nature's most perfect foods. Milk, on one hand, is extremely perishable while eggs in the shell are not. The reason eggs are not as perishable as most nutritious foods is that they possess several protective devices which include the cuticle, shell, shell membranes, lysozyme, conalbumin, avidin, pH of the albumen, and so forth.

I will limit my discussion to eggs and bacteria because bacteria are more bothersome to eggs by far than other microbes. Molds will affect eggs on occasion but this is not common today. What I intend to do in this paper is to describe how eggs are infected with bacteria and hope that you have a better appreciation of bacteria and eggs when I finish.

When a hen lays an egg, she doesn't intend for us to eat it. This is not what she has in mind at all. She intends that the egg is for reproduction. For a chicken egg to hatch, it must remain under ideal conditions for microbial growth for 21 days. If an egg didn't have protective devices against bacteria or other microbes, there would be no possible way that reproduction could take place. Nature does a wonderful job in protecting the egg. Although the egg seems simple, it is very complicated and even though scientists have studied it for many years, there are many factors about eggs still not known, particularly in the area of protective mechanisms.

When you stop to think about it, the egg is re-

markable. You can put eggs out on the kitchen table and two months later, they are still likely to be edible. One cannot do this with other foods as high in nutrition and water as the egg. The same nutrition that we obtain from the egg as humans is also beneficial to bacteria.

THE CUTICLE

To understand how bacteria get into eggs, it is important that one knows the parts since the egg is extremely complicated. Bacteria try to go to the yolk of the egg, which is really where the nutrients are and thus it is the target for the organisms. If bacteria work from the outside in, the first barrier involved is the cuticle which is largely protein. If we were to dip an egg into a weak hydrochloric acid solution, the cuticle would swell up and one could gather as much as a full teaspoon. The purpose of the cuticle is to protect the egg when it is first layed. Eggs are layed in places where there are usually many bacteria, and they need protection at this time. When an egg is first layed, it is at 107 F and the temperature drops to ambient in a short time. This drop in temperature creates a tremendous force inward, enough to suck in bacteria. With the cuticle, however, this does not happen. Thus it is a way of protecting the egg when it is first layed.

We know the cuticle gives protection for at least 100 h because we have challenged eggs at time intervals for 100 h and the cuticle still gives protection. How long it protects beyond 100 h is not known. We feel that the cuticle eventually dries out and cracks and then bacteria can enter the pores of the shell. We have taken eggs from the hen before the cuticle is put on, and we find that these eggs do not resist the challenge nearly as long as eggs with cuticle. Hence, the cuticle is extremely important.

THE SHELL

The next layer that bacteria come to is the shell. The shell has thousands of pores. We have made many studies on pores of the egg shell because we are interested in learning where bacteria enter. We find most of the pores are in the equatorial region of the egg and in the big end. Our research has shown that most bacteria enter the big end of the egg in the area of the air cell. The shell of an egg is not a particularly good barrier; in fact without

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the cuticle the egg will become infected much sooner. The shell does give some protection but it's not extensive.

SHELL MEMBRANES

The next layer that bacteria encounter is the outer shell membrane. Perhaps you never knew that eggs have membranes because you haven't studied or observed them. The outer shell membrane is five times as thick as the inner shell membrane, but it does not give nearly as much protection as the inner shell membrane. The outer shell membrane does give some protection, but I will emphasize the inner shell membrane because it is much better.

The air cell is formed between the two membranes. Usually, it is formed at the big end because this is where the two membranes separate the easiest. There is no air cell when the egg is layed but during contraction, it is formed. As the egg dries out, the air cell gets larger. This does influence quality because air cell size is a quality factor. As the air cell gets larger, it has some influence on the sucking in of bacteria.

The inner shell membrane is much thinner and weighs less than the outer shell membrane but it effectively protects against bacteria. We have done considerable work at Cornell University on the inner shell membrane and in many respects, it is still a mystery to us as far as bacterial penetration is concerned. The inner shell membrane is very high in lysozyme. There is also some lysozyme in the outer shell membrane but not nearly as much as in the inner shell membrane. Lysozyme is an anti-bacterial agent and is effective against gram-positive bacteria, but not particularly against most gram-negatives. This means when a bacteriologist examines an egg, he may find gram-negatives but not many. This is a major reason why eggs don't spoil as fast as other foods similar in nutrition. Many of the bacteria are destroyed by the lysozyme in the inner shell membrane. In addition to the lysozyme, no pores can be found in the inner shell membrane. We have not been able to find pores even with the electron microscope. This is a mystery because without pores or holes, how do bacteria get through? Reasoning would say that the only way bacteria could possibly go through would be to create an enzyme system. We thought this was an easy solution but we have used every proteolytic and lipolytic enzyme known and we have not been able to influence the inner shell membrane. Our theory at this point is that the inner shell membrane is made up of a matrix of many fibers and these fibers are so interwoven that there are no holes that go straight through the membrane.

A bacterium, however, if it is active, can wiggle its way among the fibers until it gets through. We think we have proven this to a certain extent because if we cause bacteria to be inactive by using sulfa drugs, these bacteria will not penetrate. If the bacteria do get through the inner shell membrane, normally one cannot find them for sometime in the albumen. That was a mystery to us for many years but we finally discovered that they congregate on the inside of the inner shell membrane until they accumulate in numbers great enough to invade the albumen.

THE OUTER THIN WHITE

The next layer is the outer thin white. Although we haven't done a great deal of research on the outer thin white, we can compare it to the outer shell membrane as far as protection is concerned. There is some anti-bacterial activity due to pH since the pH is not ideal for bacterial growth, but beyond this we don't think there is much anti-bacterial activity.

THE THICK WHITE

The next part or layer is the thick white. Most people think that the thick white is continuous to the yolk but this is not true. It is just a very thin envelope that completely surrounds the yolk. If one looks at the top of the yolk, one will see the exact thickness of the thick albumen. The thick albumen contains lysozyme which again will destroy any gram-positive bacteria that may have penetrated to this point. We also find in the thick white a protein and a polypeptide that are important in resisting bacterial penetration. The protein is conalbumin which chelates iron. Bacteria need iron and if it is not available, they don't do well. In regions of our country where there is a great deal of iron in the water, bacteria may carry enough iron in their cells to supply their needs, and in those areas we have more trouble with egg spoilage. The polypeptide is avidin, which chelates biotin. Many bacteria need biotin and thus they don't do well.

INNER THIN WHITE

The next area bacteria find is the inner thin white. This is a very watery albumen and there is a large amount of it in every egg. As far as we know, the inner thin white doesn't have much anti-bacterial activity except the pH is not ideal for bacterial growth.

THE CHALAZIFEROUS LAYER

Before the bacteria get to the yolk, they have to penetrate the chalaziferous layer. This layer is very viscous albumen and has everything that the thick white has but in higher concentrations. The chalaziferous layer protects the yolk. The ends of the chalaziferous layer are milky white cords that are known as chalaza cords. These chalaza cords anchor the yolk in the center of the egg and they are important because they hold the yolk so it is protected by the thick white. If it wasn't for the chalaza cords, the yolk would float to the shell and, of course, eggs would decay much faster. Without chalaza cords, eggs wouldn't hatch and thus reproduction of the species would be inhibited.

VITELLINE MEMBRANE

The last layer and probably not a very important one as far as bacterial penetration is concerned is the vitelline membrane, which surrounds the yolk. The main purpose of this membrane is its impermeability to water, hydrogen sulfide and other elements going into the yolk and such elements as iron coming out of the yolk.

BACTERIA IN EGGS

As a general statement I can say that most any bacteria can get into an egg. We can find most bacteria but, as stated earlier, the gram-negatives predominate.

The pathogenic bacteria in eggs are probably of greatest interest and so I will devote the rest of this paper to *Salmonella*, *Staphylococcus aureus*, and *Clostridium perfringens*.

Salmonellae are of great importance and have received much publicity in conjunction with eggs. Much of this publicity goes back to the day when eggs were infected heavily with one species of salmonellae, known as *Salmonella pullorum*. Not too many years ago all chickens had Pullorum disease. The bacteria were found in the ovary and thus most eggs were infected. As far as we know, this is the only bacterium that can be transmitted through the egg and can be found in the egg when it is laid. Today, to find *Salmonella pullorum* would be quite a feat indeed. We haven't found salmonellae in eggs in several years. The egg industry, however, is still paying for its sins of many years ago.

Many species of *Salmonella* can be found on the shells of eggs because they are common in the intestinal tracts of chickens. Salmonellae are not particularly active in penetrating unless eggs are improperly handled. One of the problems in infection

of eggs is the contraction. If there should be contaminated water on the shell when the egg is cooling off, then bacteria can be sucked through. It is always important to remember that if eggs are washed, cold eggs should be placed in warm water so the eggs are expanding rather than contracting. To do the opposite is very dangerous because bacteria can be sucked in.

I think I can say with accuracy that the incidence of salmonellae in shelled eggs is not very great. Many people worry about using cracked eggs but, again, remember that I said the shell gives little protection.

With liquid eggs, the problem of salmonellae is different. A few years ago, it was not legal to use a sanitizer-detergent on eggs that were to be broken out. Today industry people wash eggs with sanitizer-detergents and leave them on the shells, which maintains sanitized shells for a long time. In the days when the industry did wash eggs but without a sanitizer-detergent, there were salmonellae on eggs when they were broken. The egg albumen would run to the liquid product. At that time, salmonellae down over the shell and carry salmonellae with it in were detected in some liquid eggs and this gave eggs a bad name. Today, most salmonellae infections in liquid eggs come from reinfection of the egg and, in most instances, from humans. Although liquid eggs are pasteurized they can be reinfected after pasteurization.

The next pathogen I would like to consider is *Staphylococcus aureus*. *S. aureus* can cause illness in humans in a short time. The incubation period is much less than that of salmonellae, although the two are confused from time to time. In eggs, the chance of *S. aureus* getting into a shell egg is very remote because it is a gram-positive organism. In egg products, however, such as liquid egg, there is a chance of the organism getting in, and again it comes mainly from humans since *S. aureus* is found in large numbers in sores and cuts on hands of employees.

The last pathogen of real importance in eggs is *Clostridium perfringens*, which is sometimes known as *Clostridium welchii*. It has only been of concern recently, and the concern really came from England where they have less refrigeration than in this country. Again, the chance of *C. perfringens* being found in shell eggs is very remote because it is a gram-positive organism. However, it can be found in liquid eggs because it is a widely distributed organism. We've found that *C. perfringens* can be found almost any place. Fortunately, only a few strains of the organism are pathogenic and even then the pathogenicity is not very high. It is my personal belief that many times when illness is diagnosed as salmonellosis, the real cause is the toxin from *C. perfringens*. We will

hear much more about *C. perfringens* in the future.

I could discuss the many bacteria that are found in eggs but those already mentioned are the ones of importance as far as illness to humans is concerned. Many of the gram-negatives can be found (sometimes in high numbers) and one of the primary causes of spoilage is *Pseudomonas*. One of the testing methods for bacterial infection in eggs is the ultra-violet light. For those *Pseudomonas* that fluoresce in ultra-violet light, the internal part of the egg will give a green color.

IN CONCLUSION

In closing, I have emphasized that shell eggs have many barriers against bacteria. Even with all these barriers, eggs can become infected because some people tend to grossly misuse shell eggs. The number of spoiled eggs that come into our laboratory today,

however, is only a fraction of what it was 20 years ago.

I would like to emphasize, however, that with liquid eggs (we are moving into the liquid egg business very rapidly), the situation is entirely different. Once yolk is mixed with albumen, all of the anti-bacterial properties of the egg are removed. First of all, we don't have the cuticle, shell, and the shell membranes for protection because they are thrown away. Because of the blending of yolk with the white, we are losing any protection we might get from the albumen; thus, mixed whole egg is an ideal medium for growth of bacteria. Liquid eggs have a shelf life very similar to that of milk. If one starts with shell eggs with low numbers of bacteria, breaks them out under sanitary conditions, and pasteurizes them, one can get up to about 20 days of shelf life. On the other hand, if one starts with shell eggs that are very high in bacteria counts, even with pasteurization, one can only get 2 to 3 days shelf life.

EDUCATION AND INFORMATION PROGRAM ABOUT FOOD PROCESSING INDUSTRY BOWS AT NATIONAL EXPOSITION FOR FOOD PROCESSORS

An information and education program designed to emphasize the contributions made by the Food Processing Industry to the American life-style has been launched here during the industry's annual trade show and convention.

The program, sponsored by the Food Processing Machinery and Supplies Association (FPM&SA), was presented during the association's National Exposition for Food Processors (NEFP), the largest annual trade show in the industry. The NEFP and the concurrent National Canners Association convention attract thousands of representatives from every aspect of the Food Processing Industry whose annual production is valued at more than 10 billion dollars.

The major thrust of the program is a series of posters which depicts some of the benefits Americans enjoy as a result of processed and manufactured foods. FPM&SA also has assembled a digest of statistics which highlight the industry's technological advances and its role in the Nation's economy. Initially, the program is being directed at the thousands of employees who produce, prepare, process, and package the canned, frozen, dehydrated, baked, and otherwise

manufactured food products. The posters are being supplied free of charge to food processors and their suppliers for display in about 4,000 operating plants and sales offices throughout the United States.

The posters also are suitable for use in print advertising, according to FPM&SA, which is urging industry firms to disseminate the material in their localities. "Recent inflationary trends in the economy have tended to overshadow the real contributions the Food Processing Industry has been making and will continue to make to American life-style," the association stated. "Our program simply seeks to re-emphasize the positive impressions consumers have about processed foods, and to disseminate some little-known facts about the Industry."

FPM&SA choose the NEFP and Canners Convention as the vehicle for introducing the campaign since the displays and meetings at this annual four-day gathering reflect much of the technology and innovations which have allowed the Food Processing Industry to contribute significantly to the American life-style and economy.