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# How Avocado Fruits Soften During Ripening

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Fruit softening is one of the most obvious changes that occur during avocado ripening. Softening is easily detected by simply squeezing the fruit, thus providing the consumer with a convenient means of checking for ripeness.

While avocado softening is easy to detect, it is not so easy to understand. How does softening occur during avocado ripening? This question has been studied by plant physiologists for more than 20 years, and much has been learned about softening. Our knowledge of softening and other related changes during ripening is still far from complete, however, so much research continues to be carried out in these areas. The immediate goal of all of this research is to put together a detailed understanding of how softening and other changes occur during ripening. If these processes can be understood at the most basic level, then it might be possible to invent improved methods of controlling ripening. Control of avocado ripening could open the door for market expansion both in terms of the length of the season and in terms of geographic distribution.

The purpose of this article is to describe some of the knowledge that has been gained about avocado softening during the last 20 years. Most of this research has been carried out at a molecular level. This fact presents an immediate communications problem, however, because most of the readers of this article are not accustomed to thinking about avocado fruits at a molecular level. In an attempt to solve this communications problem, we will make use of an analogy. That is, we will compare the molecular events of avocado ripening to events that might occur in a farm barnyard. Since it is easy for all of us to think about what happens in a barnyard, it will also be easy to understand what happens during avocado softening.

To begin this description of avocado softening, it is first necessary to take a close look at an avocado fruit. Figure 1 provides such a look at a mature but unripe Hass fruit. The fruit has been broken open, and the broken surface examined with a scanning electron microscope. This figure illustrates several features that are important as regards avocado softening. Like most living things, the avocado fruit is made up of small compartments called cells. The area marked between the arrows in Figure 1 is a single cell, which is more-or-less round in shape. Cells are very small, since 500 of them lined up like beads on a string would cover a distance of only about one inch.

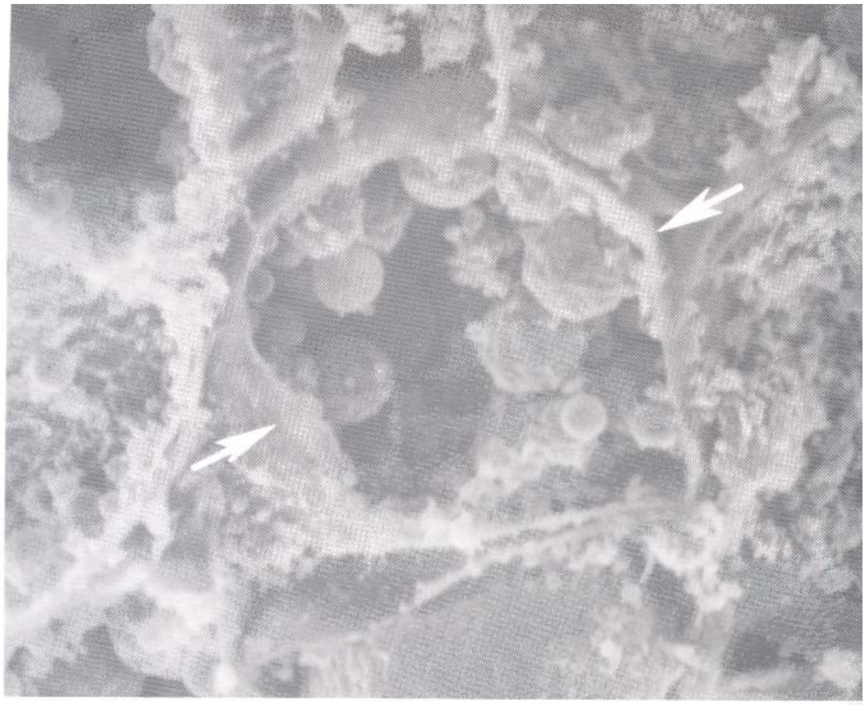

*Figure 1. Scanning electron micrograph of a mature but unripe Hass fruit. The fruit was broken open, and the broken surface examined. The arrows mark the cell wall at opposite ends of a cell that has broken open to reveal the cytoplasm. Approximately 500 such cells lined up like beads on a string would cover a distance of one inch.*

Although small, each cell is a complete functioning unit. The inside of the cell is called the cytoplasm and is made up of a variety of smaller parts called organelles. Some of these organelles are round and can be seen in the cell shown in Figure 1. As regards fruit softening, the most interesting part of the cell is the border, shown at the tips of the arrowheads in Figure 1. Most of this border is made up of the cell wall. Pressed against the inner face of the cell wall is the plasma membrane, a sheet-like organelle that serves as a sealant between the inside and the outside of the cell.

Cell walls give strength and rigidity to plant materials. In particular, cell walls cause an unripe avocado to be hard. While we may not realize it, cell walls are already familiar to all of us. Dead, dried wood is made up almost exclusively of cell walls. As a tree trunk dies, the organelles on the inside of the cells break down and are lost until only the cell walls remain. When an avocado fruit ripens, however, the opposite situation occurs. In this case, the cell walls are broken down while the organelles of the cytoplasm remain intact. As the cell walls are degraded, the avocado fruit gradually loses its rigidity and hardness. By the time that the fruit is ripe and soft, the cell walls are gone so only the plasma membrane and cytoplasm remain.

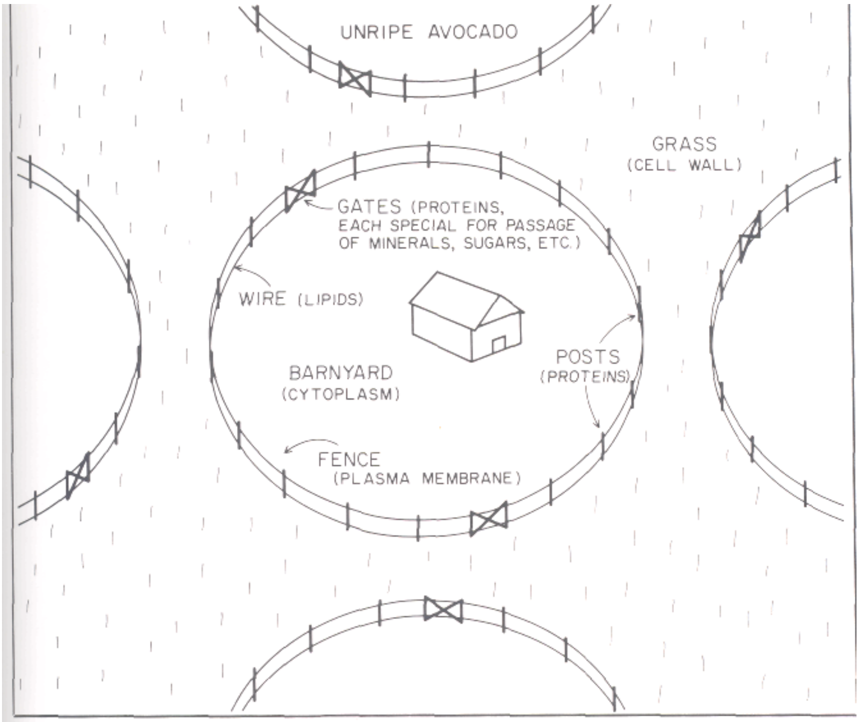


*Figure 2. Barnyard analogy of the image shown in Fig. 1. Each cell in Fig. 1 is like a barnyard in this figure. The fruit is mature but not yet ripening.*

To understand how cell walls are degraded during avocado ripening, we will now introduce the barnyard analogy. Compare Figures 1 and 2. A barnyard in Figure 2 is like a cell in Figure 1. Each barnyard is surrounded by a fence which acts as a barrier between the inside and outside of the barnyard. Thus, the fence is like the plasma membrane, the part of the cell that acts as a sealant between the inside and outside of the cell. Just outside of the fence, between the barnyards, is grass. The grass is like the cell wall. The barnyard itself is like the cytoplasm of the cell, which contains a variety of organelles. While only a barn is shown in the barnyard in Figure 2, we can imagine that a variety of other items such as watering stations and feeding troughs are also present in the barnyard.

If we examine the fence in Figure 2 more closely, we see that it is made up of wires, posts, and gates. In the same way, the plasma membrane of the cell is made up of lipids (fats) and various proteins. The wires of the fence are like the lipid molecules of the plasma membrane. The posts and gates of the fence are like the proteins of the plasma membrane. Some of the proteins act to form the structure of the plasma membrane. These proteins are like the posts that hold up the fence. Because the plasma membrane acts as a sealant, special provisions are required to allow minerals, sugars, and other needed molecules to pass in and out of the cells. Certain proteins in the plasma membrane act as channels that allow passage of these molecules. These channel proteins are like gates in the fence of the barnyard. Since a special channel protein is required for the passage of each type of molecule through the plasma membrane, we can think of the barnyard fence as containing several different kinds of gates, each of which allows passage of only a particular kind of animal or piece of equipment. Using this analogy between the avocado cells and the barnyards, we can now discuss softening during avocado ripening. As already indicated, fruit softening occurs when the cell walls of the fruit are broken down or degraded. Research during the past 20 years has shown that cell wall degradation is brought about by the action of a special class of proteins called enzymes. These particular enzymes have the ability to break down or degrade the cell wall. In the barnyard analogy, we can account for these enzymes by adding sheep to the picture, as shown in Figure 3. The sheep (enzymes) appear first in the barnyard (cytoplasm) and then go outside of the barnyard to eat the grass (cell wall). To get to the grass, however, the sheep must get through the fence (plasma membrane). The sheep require assistance to pass through the fence, and we can think of this assistance in the form of special sheep gates (proteins) which are new additions to the fence (plasma membrane) in a ripening avocado.

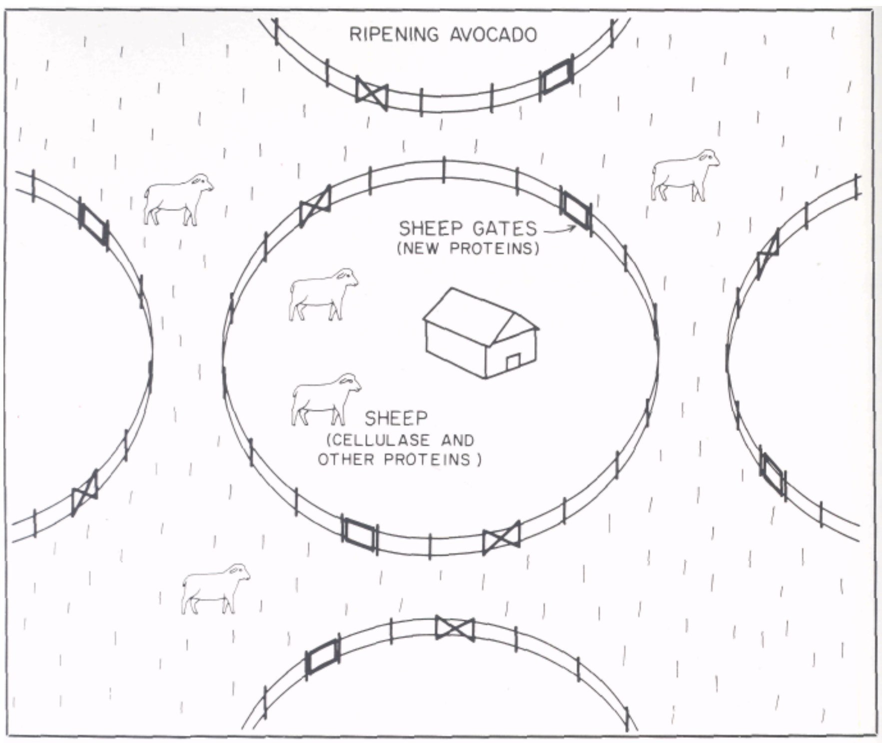


*Figure 3. Barnyard analogy of a ripening fruit. The sheep represent the enzymes that move into the spaces between the cells to degrade the cell walls and soften the fruit. Passage of the sheep through the fence may require the addition of special sheep gates.*

Research has shown that the actual situation in a ripening avocado is somewhat more complicated than indicated in Figure 3. The cell wall is actually a complex structure made up of several kinds of polysaccharides, such as cellulose and pectin, as well as one or more proteins, such as extensin. In the barnyard analogy, we can think of this situation in terms of several kinds of grass growing in the spaces between the barnyards. One or more special enzymes are required for the degradation of each of the different parts of the cell wall. Research so far has identified at least two of these enzymes in ripening avocado fruits. One enzyme, cellulase, degrades the cellulose portion of the cell wall. Another enzyme, polygalacturonase, degrades the pectin portion of the cell wall. These various enzymes correspond to separate breeds of sheep in the barnyard analogy. Each breed of sheep eats only one of the various kinds of grass that grow outside of the barnyard.

Much of the research on avocado softening has concentrated on the cellulase enzyme (sheep). This enzyme is essentially absent from the fruit until the start of ripening, but then it begins to be produced in large amounts. Once the cellulase is produced, it undergoes some alterations before it starts to work in the cell wall to soften the fruit. First, one end of the cellulase molecule is cut off. In the barnyard analogy, this step would be like cutting the horns off of the sheep. Some carbohydrates are then attached to the cellulase (the sheep grows some wool). Finally, a portion of the added carbohydrate is trimmed off of the cellulase (some of the wool is sheared off of the sheep). The purpose of these alterations to the cellulase molecule is not yet fully understood, but it is thought that they may be important steps in getting the cellulase through the plasma membrane to the cell wall. In the barnyard analogy, the dehorning and partial wool shearing may be important steps in getting the sheep to go through the gate to get to the grass.

The emphasis of avocado research in my own laboratory is on the plasma membrane, the fence surrounding the barnyard. In this work, we try to isolate the fence and study its various parts. When ripening starts, the buoyant density of the plasma membrane increases. This change indicates that the fence gets slightly less wire and/or slightly more posts and gates during ripening. In addition, changes occur in the relative abundances of some of the plasma membrane proteins. As regards the barnyard analogy, we are not yet certain whether this change in protein abundance is a change in the number of posts or in the number of gates. Some plasma membrane proteins first appear only with the start of ripening. These new proteins are at the focus of our current research effort because they might be pieces of the sheep gates shown in Figure 3.

The barnyard analogy used here is not exactly correct as regards all of the technical details of avocado fruit softening. Through the use of this analogy, however, it is hoped that at least some of the recent research results can be better understood by the nonscientist. In particular, it is hoped that the barnyard analogy has helped to clarify the relevance of this type of research to the long term goal of controlling avocado ripening. If we can learn how the sheep are produced, altered, and passed through the fence, then we may eventually be able to control this and other aspects of the ripening process.


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