Objective Measures of Meat Color

Donald H. Kropf*, Leader
Dennis G. Olson, Cooperator
Roger L. West, Recorder

To begin this reciprocation session, I have prepared what I consider some of the important considerations in making objective meat color measurements, including reasons for using objective measurements, decisions that have to be made and sources of measurement differences.

An important reason for using objective measurements is to support visual observations, provided that the visual observation is descriptive and not hedonic (acceptability or preference). I think visual observations have a great deal of value and we are not going to do away with them. Objective measurements reinforce visual observations. When you are writing a paper, it is surely nice to have visual observations and then have objective color measurements which support what the visual observations are saying. However, regardless of which evaluation is being made, you must be very careful to handle each muscle, each steak, each ground beef pattie or each ham slice exactly the same so as not to create handling artifacts. This is especially critical when live animal treatments are being compared for muscle color.

Another use for objective color measurements would be as a basis for acceptance or rejection of a product. Veal, in which light color is required, would be a good example of using an objective color specification. Don Raymond from Agriculture Canada says that they have veal standards based on objective color parameters and that some of their veal buyers are now buying by a reflectance number. Other marketing problems, such as dark cutting beef or PSE pork, could be handled in this manner. Also, objective measurements could be used to determine frozen color deterioration and display and acceptability.

Estimating the proportions of the various chemical states of myoglobin is an increasingly important use of objective color measurements. This gives clues about what is happening in the sample relative to the treatment of packaging material. An "estimate" is appropriate especially when reflectance is used, since it is not a really sensitive system. These measurements are within 6% to 7% of the actual value. Three helpful references on the estimation of oxymyoglobin, reduced myoglobin and metmyoglobin are Broumand et al. (1958), Dean and Ball (1960) and Stewart et al. (1965).

Once you have decided to make objective color measurements, you have some decisions to make. A major decision will be whether to use extraction of the pigments and measurement of transmission or absorbance, or the determination of reflectance on the intact sample. There are advantages and disadvantages for each, but I am strongly in favor of reflectance. The use of extraction procedures will give sharper peaks and better separation than reflectance. However, the primary problem with this technique involves the overestimation of oxymyoglobin and metmyoglobin and the underestimation of myoglobin because of changes occurring during extraction and measurement. Krzywicki (1982) of Poland maintains that, if temperature is kept low and pH is controlled, there is minimal change in metmyoglobin during extraction. However, I feel that there may be some oxygenation and change from reduced myoglobin to oxymyoglobin taking place.

With extraction, there is also a problem in sampling. Since we are primarily interested in surface color, how deep can we sample and still have the correct proportions of pigments? If we have a muscle that has been displayed for a while, on the outside of it there will be a layer of oxymyoglobin where the partial oxygen pressure is high enough to support that pigment, in the middle of the muscle will be the purple-red reduced myoglobin, and somewhere in between will be a line of metmyoglobin. Grinding the entire sample will not give an accurate evaluation of the surface. Some researchers scrape off one millimeter of the surface, or try to, but this is very difficult with a frozen steak.

Other problems with use of an extraction procedure are that oxymyoglobin and metmyoglobin differ in solubility and that sampling and extraction terminates usefulness of a sample. I feel that because of these problems the results of extraction procedures are usually invalid, except when measuring total heme or total myoglobin pigment.

Reflectance measurements more accurately reflect what the eye and brain see. With this technique, repeated measurements over time can be made on the same sample. It is rapid and easy, but peaks are not as sharp as with extraction procedures. However, reflectance measurements are affected by muscle structure, surface moisture, fat content and pigment concentrations. Many of these problems can be corrected by using ratios of reflectance at different

*D.H. Kropf, Kansas State University, Manhattan, KS 66502

D.G. Olson, Iowa State University, Ames, IA 50011

R.L. West, University of Florida, Gainesville, FL 32611

Reciprocal Meat Conference Proceedings, Volume 37, 1984.

wavelengths or by using differences between reflectance at different wavelengths. With reflectance techniques, a variety of measurements can be taken and the single or combination of measurements that zero in on the color change of interest can be selected.

Instrument selection for color measurements depends on your objectives. A spectrophotometer is not very portable but produces data that are convertible into various systems of nomenclature. When linked to a computer, these instruments can calculate parameters for different illuminants. After using a spectrophotometer, you can help decide what characteristics to build into a colorimeter or fiber-optic scanner. Other instruments, such as colorimeters, photometers, etc., may be more portable and better suited for specific purposes. At Kansas State University, we are presently using a Hunter Lab Spectrophotometer that will give us data at various wavelengths and will also give us L, a and b and X, Y and Z values. Dennis Olson at Iowa State has a Hunterlab Lab Scan 2 which has some features you might want to consider when you buy an instrument.

Various accessories can be used for reflectance measurement of color. A diffuse integrating sphere with 0° incident light is an important addition to a reflectance spectrophotometer. A K/S conversion using the Kubelka-Monk equation (Judd and Wyszecki, 1967) is very useful for producing linearity in reflectance measurements of percentage of pigments.

Color measurements are a great way to pile up a lot of numbers in a hurry. I think that a very important accessory would be to interface some kind of data acquisition system with the spectrophotometer. The data could be placed directly on a floppy disk, a hard disk or stored in the computer. Most spectrophotometers now have systems for recording data as well as calculating alternative notations such as X, Y and Z, Hunter L, a and b or others.

In making color measurements, one must decide on what color scale or measurement to use. Lester Jeremiah and his colleagues (Jeremiah et al., 1972) should receive a lot of credit for looking at many different objective color scales and relating them to visual color scores in meat. Another reference that I find helpful is that of Hunt (1980), presented at the Purdue Reciprocal Meat Conference.

Data alteration is another thing you have to deal with in making objective color measurements. Reflectance measurements at various wavelengths can be made more linear by use of K/S ratios. Data can also be corrected to the same absorbance at 525 nm (isobestric point of myoglobin pigments). I am very strong on data alteration where we calculate ratios or differences of percent reflectance at different wavelengths to correct for effects of fat on the meat surface, differing amounts of fat or free moisture that can have an effect on pigment concentration. These calculations will also correct partially for differences in muscle structure. These corrections tend to reduce the coefficient of variation for the measurements.

Corrections can be made for the illuminant used. Newer spectrophotometers will allow you to make readings using illimunant A (tungsten light), B (sunlight at noon), C (daylight) or others. We have found that we get higher correlations between objective values and visual color if measurements are made using illuminate A, because it has a higher proportion of red than other illuminants.

Sources of measurement differences can be found both in the instrument used and in sample presentation. Recently, Mary Ellen Zuyus of Hunter Labs was on our campus and indicated that instruments differ in spectral narrowness (band width), which can be of some importance in making reflectance measurements. In addition, you must know whether you are measuring diffuse or specular reflectance. Another thing one should do with a new instrument, at least initially, is to check the repeatabiity of the reading at the same exact location. Finally, the standards used to calibrate the machine must be clean.

With reflectance measurements, sample presentation causes more differences than the instrument. The sample should be thick enough to be opaque. If not, an opaque backing should be used. Mary Ellen Zuyus recommended a white backing rather than a black one. Orientation of muscle fibers in the sample can also affect the readings. To correct for this problem, the people in our Food Nutrition Lab will take a reading, turn the sample 90 degrees and take another reading. Dennis Olson's instrument does sample averaging, which should reduce this problem.

One of the toughest problems with sample presentation involves uneven or variable discoloration. Often on the surface of a sample, some areas may be very discolored and others are bright. Where do you take the reading? The best averaging of color can be obtained by having a large aperture on the reflective sphere or by taking numerous readings. Recently, we have been doing what we call "worst point color". We feel that by visually and objectively measuring the worst point on a sample, a more realistic evaluation is made since this is the part of the sample that will prevent the product from selling in a display situation.

Melvin Hunt and Pam Sleeper (KSU) have found that the type of film on the sample affects the reading. You need to standardize the instrument for each type of film, which makes it difficult to make a comparison with several different films.

The pressure applied to the sample on the aperture can also affect the reading. You want the sample to be flat, not pillowing into the port. A cover glass over the port prevents this. Similar to the uneven surfaces of fresh states, frozen states create measurement problems because their surfaces will not be even. This unevenness can be corrected by freezing them in a pan; however, this may affect oxygen availability and color.

With these comments, I will open this session for questions and comments.

References

Billmeyer, F.W., Jr.; Saltzman, M. 1967. Principles of Color Technology. Interscience Publishers, John Wiley & Sons, New York.

Broumand, H.; Ball, C.O.; Stier, E.F. 1958. Factors affecting the quality of prepackaged meat. II E. Determining the proportion of heme derivatives in fresh meat. Food Technol. 12:65.

Dean, R.W.; Ball, C.O. 1960. Analysis of the myoglobin fractions on the surfaces of beef cuts. Food Technol. 14:271.

Erdman, A.M.; Watts, B.M. 1957. Spectrophotometric determination of color change in cured meat. J. Agr. Food Chem. 5:453.

Francis, F.J.; Clydesdale, F.M. 1975. Food Colorimetry: Theory and Applications. AVI Publishing Co., Westport, CT.
Hunt, M.C. 1980. Meat color measurements. Proc. 33rd Recip. Meat Conf., Nat'l. Live Stock and Meat Board, Chicago.
Jeremiah, L.E.; Carpenter, A.L.; Smith, G.C. 1972. Beef color as related to consumer acceptance and palatability. J. Food Sci. 37:476.
Judd, D.B.; Wyszecki, G. 1967. Color in Business Science and Industry. 3rd edition. John Wiley and Sons, New York.
Kropf, D.H.; Tuma, H.J.; Allen, C.C.; Hunt, M.C.; Sandberg, M.L.; Schafer, D.E. 1976. Evaluation of color and other properties of frozen beef. In Objective Methods for Food Evaluation, Nat'l. Academy of Sciences, Washington, DC.
Krzywicki, K. 1976. Assessment of relative content of myoglobin, oxymyoglobin and metmyoglobin at the surface of beef. Meat Sci. 3:1.
Krzywicki, K. 1982. The Determination of Haem Pigments in Meat. Meat Sci. 7:29.
Stewart, M.R.; Zipser, M.W.; Watts, B.M. 1965. The use of reflectance spectrophotometry for the assay of raw meat pigments. J. Food Sci. 30:464.
Strange, E.D.; Benedict, R.C.; Guggaer, R.E.; Metzger, V.G.; Swift, C.E. 1974. Simplified methodology for measuring meat color. J. Food Sci. 39:988.

Discussion

J.L. Secrist: Do you have trouble with frost buildup on the surface of steaks?

D.H. Kropf: Not if we are very fast in putting them on the spectrophotometer and getting them off. Our instrument will scan the visible light spectrum in seven seconds, so we have not had that problem. Our spectrophotometer is in a different room from the display cases. We have some refrigerated, open-topped cases on wheels that get a lot of use in our laboratory. We put the steaks in there and wheel them over to the spectrophotometer. The other frost problem that I have encountered is that if you are doing display studies and have cases like ours, you run into the fact that twice in every twenty-four hours the retail case is going to have a defrost cycle that takes about a half hour. My advice is simply to wait until the defrost cycle is over and the frost goes off the steaks. You can wipe it off. So those are the frost problems I have encountered. I think we can work with them.

A.M. Booren: Some of the older instruments have a longer scan time. What scan time would you recommend?

Kropf: I really like our scan time of seven seconds. In that period of time, our Hunter Lab D54 stores the data and then we can, with a computer, calculate whatever color parameters we like. That time is especially important if you are doing something like Melvin Hunt and Pam Sleeper have done, where they are taking frequent readings to estimate the blooming speed, or oxygenation speed, of a steak. I think that is a critical measurement where readings must be made rapidly. But remember, if you have a fast reading, then probably you are getting a wide band width and a less sharp instrument reading. I think there is some possibility of that. We can live with our seven-second reading time. Now, my old instrument had several reading speeds, and one of them was 10 nm per minute. With the old B and L that I had, you could not do that kind of work, at the fastest scan speed of 250 nm per minute. It took 1.2 minutes to scan from 400 to 700 nm, which is pretty long. You get some changes. That scan started at the violet end of the visible spectrum and went to the red, and the red was always the last thing read when I put it on the instrument. That was an additional problem.

R.L. Henrickson: It seems that your standards would be really important. What do you recommend for beef and pork?

Kropf: The instrument we are using goes back to a white standard and a grey tile standard and then takes off from that. I used to fuss around with a magnesium carbonate block. Have any of the rest of you done that for 100% reflectance? They were always crumbling on me. I like the tile standard a great deal, as long as I keep them clean. Does your question imply what is a good color for beef and pork?

Henrickson: No, I was wondering if you would be better off to have a red standard than a white standard? Something closer to the exact color of the meat?

Kropf: It depends on which instrument you have. No, I am convinced that with this instrument I am getting good calibrations. That is my feel for it. Denny, do you have anything to expand on that?

D.G. Olson: We use a white standard also and are very happy with the reproducibility of it.

Henrickson: Will the data apply to the Munsell color standard?

Kropf: Let us take a look at interconversions. My first reaction was to tell you no. Figure 1 shows the interconversions that are possible with many instruments.

I am starting with a spectrophotometer and presumably a tristimulus reflectometer would do the same thing. From that, you can go to XYZ values and you will notice that we can calculate Munsell notations from that. I am not real sold on the Munsell system, I suppose. My feeling is that I think it is a good color description system, but it is a little bit awkward. I think many of you are familiar with the Munsell system and how it was set up. You have a three-dimensional system that involves something called hue (relative placement in the color order), chroma and value and you end up with three numbers and a letter. I think you can handle that on a computer. This is hue, and would be five red or could be yellow, red, blue or green. Also, you end up with a value number and a chroma number. It is a good sensitive system but I have trouble with interpretation. I like to use spectrophotometric data and that is my bias. They are interconvertible and the people from Hunter Labs tell me that they experience no serious error problems.

Henrickson: With the Hunter system, do you have any trouble relating what the eye sees to what the Hunter tells you?

Kropf: I am going to answer that no. I like the Hunter system pretty well for spectrophotometric possibilities. I have a picture of the Hunter system here for those of you who are not familiar with it. This still has a flaw in it, as near as I can tell. I think most of you have seen the Hunter system which is three dimensional also. For "a" value, plus is red and negative is green, blue to yellow is "b" value and it also has a lightness and darkness value. By the way, most fresh meats will be in this quadrant (plus values for both "a" and "b").