The previous Meat Technology Update (Issue 5/06, October 2006) discussed the development of a brown colour in fresh meat, as a result of oxidation of the pigment myoglobin to metmyoglobin. Other undesirable colours and appearances are described in this issue.

Greening

Green colour may develop in meat through reaction of the meat pigment myoglobin with certain compounds. When myoglobin reacts with hydrogen sulphide (H₂S), the resultant pigment is sulphmyoglobin, and when myoglobin reacts with hydrogen peroxide (H₂O₂), a number of pigments are produced. These pigments are green. The compounds H₂S and H₂O₂ are produced by certain microorganisms under specific storage conditions.

Sulphmyoglobin greening is associated with growth of the bacterium Pseudomonas mephitica. This organism requires low oxygen levels and pH greater than 5.9 for production of hydrogen sulphide from sulphur-containing amino acids. Greening is associated with poor barrier films used for vacuum packaging, which allow a small amount of oxygen into the pack. The good barrier provided by most modern films has led to sulphmyoglobin greening being quite uncommon. The organism can produce greening even if the cells are less than 5% of the total microbial population. If oxygen is absent, or the level is high, or the pH is below 6, it cannot produce hydrogen sulphide and greening will not occur. To avoid green discolouration at low oxygen levels (e.g. in vacuum or low-oxygen MAP with some residual oxygen), high pH meats should not be used. When sulphmyoglobin green packs are opened, the green colour often disappears because the pigment is oxidised to a red form.

Other green pigments result from the interaction between myoglobin and hydrogen peroxide. Their formation is favoured between pH 4.5 and 6. The source of the hydrogen peroxide may be bacterial, may result from the interaction of ascorbic acid (vitamin C) with the oxygen molecule of

Figure 1: Two packs of lamb showing greening, compared with a normal pack above.
oxymyoglobin, or may be produced by the muscle itself. Hydrogen peroxide greening has been associated with cooked cured meats under aerobic conditions. In vacuum-packaged fresh meat, the meat enzyme catalase breaks down hydrogen peroxide, so production of the green pigments is limited, and greening due to sulphydmyoglobin is much more important.

**Brown and black spots**

Brown discolouration in the form of spots on fat surfaces has been attributed to the yeasts *Yarrowia lipolytica* (formerly *Saccharomyces lipolytica*) and *Candida zeylanoides*. Yeasts can survive and grow on chilled meat stored in air, and in vacuum-packed meat if the oxygen transmission rate of the packaging film is too high. Brown spot was a problem on vacuum-packaged beef in the 1970s, but is a rare occurrence today.

Yeasts and moulds require oxygen to grow, and can grow at temperatures from just below zero to up to 40°C, dependent on the species. Most tolerate reduced water activity, and prefer a slightly acidic environment of pH 4.4 to 5.5. Yeasts and moulds survive well in chillers, and incidents of brown spot on carcasses and corresponding packaged meat have been associated with contaminated condensate dripping onto the meat.

Black spots may develop on frozen meat stored at −5 °C for 40 days or more, and this has been associated with a number of yeasts and moulds including *Cladosporium cladosporioides*, *Cladosporium herbarum*, *Penicillium hirsutum* and *Aureobasidium pullulans*. These organisms penetrate into the meat surface, and must be trimmed off.

**Spots on cured and fermented meats**

Some moulds are considered desirable in cured and fermented meats because they impart certain flavours, and assist in the fermentation process; however, some are undesirable. The mould *Cladosporium* causes unsightly, deep-seated black spots on cured hams, which cannot be washed off. It has strong proteolytic activity, allowing it to break down the product surface and burrow into it. *Scopulariopsis* causes white spots on the skin of hams, and a number of other moulds cause whiskers on fermented meats. Lightly smoking the product during curing or before packaging can help to reduce mould growth.

A Gram-negative bacterium, *Carnomonas nigricans*, has also been found to cause black spots on cured meats. These spots begin as small rust-coloured areas which blacken over a few hours. Adding sodium nitrite to the cure prevented the formation of these spots; while adding dextrose, maltose or dextrin encouraged formation.

**Bones and marrow**

The marrow of bones contains a high proportion of blood, and when bones are cut, red blood cells may be ruptured, releasing haemoglobin which gets smeared onto the surface of the bone. Haemoglobin is the red pigment in blood, and it responds to oxidation in the same way as myoglobin. Initially, on exposure to oxygen, it is bright red, but brown methaemoglobin develops rapidly in response to low oxygen levels in packaged product. When packaged in a high oxygen-modified atmosphere (e.g. 80% O₂, 20% CO₂), bones discoulour significantly within 24 hours. Alternatively, if the oxygen in the gas mixture above is replaced with nitrogen, a stable purplish marrow colour will remain for 1–2 weeks at 4°C or less.

**Ground and comminuted meats**

The colour stability of meat is partly dependent on aerobic-reducing enzymes. The aerobic-reducing systems prevent browning by converting metmyoglobin back to myoglobin. When the myoglobin and reducing enzymes are contained within the muscle-cell membrane, the meat has relatively good colour stability.

When meat is minced, the cell structure is disrupted (see diagram below). This increases the exposure of myoglobin to oxygen, and destroys some of the muscle’s innate aerobic-reducing system. This partly explains the accelerated browning observed in minced versus whole muscle. In addition, the oxygenation of meat during mincing followed by restricted access to oxygen in the middle of a pack, creates low oxygen levels at the surface of particles. At low oxygen levels myoglobin is oxidised to metmyoglobin; but, whereas in intact meat the aerobic-reducing system is active, in minced meat the system is inactive and the meat turns brown from the inside of a pack.

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**Figure 2:** Brown spots on beef fat.

**Figure 3:** Destruction of muscle cells by mincing.
Researchers have found that antioxidants can inhibit myoglobin oxidation and improve colour stability, but are more useful in controlling other aspects of oxidation such as rancidity. For example, EDTA is an antioxidant and, although it decreases lipid oxidation and rancidity, it increases myoglobin oxidation and browning; vitamin C is an antioxidant, but it can sometimes react with meat and produce hydrogen sulphide, and cause greening. At present, such additives are not permitted in Australia.

Feeding vitamin E to cattle seems to decrease lipid oxidation and myoglobin oxidation and thereby improves fresh meat colour stability and shelf life. One study showed that while control packs of mince were okay up to 48 hours, those from cattle fed 500IU Vitamin E for 120 days were okay up to 67 hours, and those fed 2000IU for 120 days were okay up to 92 hours. This was in PVC overwrap displayed under fluorescent light.

Frozen meat

The colour of frozen meat is affected by: freezing rate; storage temperature and fluctuation in temperature during storage; intensity of light during display; and method of packaging. Very slowly frozen meat is excessively dark, while meat frozen in liquid nitrogen is unnaturally pale. Such extremes are unlikely to be seen under commercial conditions, but faster freezing regimes will give a paler product than slower freezing regimes. The large variation in lightness is a result of differences in the rate of ice-crystal growth. Small crystals formed by fast freezing scatter more light than large crystals formed by slow freezing; so fast-frozen meat is opaque and pale, while slow-frozen meat is translucent and dark. To optimise frozen-beef colour, fresh beef should be exposed to air for 30 minutes before freezing to allow optimum bloom to develop prior to freezing. This can result in frozen beef that is similar in appearance to fresh beef.

During storage of frozen meat, if it is exposed to air, slow thawing and refreezing of the exposed surface leads to dehydration of the meat. This leads to the development of freezer burn which appears as a grey-white area on the exposed surface where the fibres of the meat are visible. Freezer burn areas stay dry and pale when the meat is defrosted, and is tough and dry to eat.

The major colour problem during retail display of frozen meat is photodestruction. Frozen meat under direct illumination oxidises from the surface inwards (compared with fresh meat which oxidises from the subsurface outwards). Oxidation of frozen oxymyoglobin is temperature dependent—the rate increases from −5 to −12°C and then decreases to a minimum at −20°C. The rate of oxidation is affected also by muscle type—*Longissimus dorsi* (loin) will fade more slowly than *Gluteus medius* (rump). Frozen-beef colour remains attractive for at least 3 months in the dark, but only 3 days in the light.

Cooked meat

Thorough cooking causes denaturation (unfolding) of the globin part of myoglobin. This makes the pigment much more prone to oxidation. In the presence of air, the grey-brown cooked meat pigment (denatured globin haemachrome), is formed. Under anaerobic conditions (canned meats, vacuum bag in water), the pink denatured globin haemochromes can be formed. Owing to heat denaturation of the globin protein, these proteins coagulate and are not soluble in water, so meat juices don’t look red.

Variation in the colour of precooked meat products cooked to the same internal temperature has been a problem in the meat industry for over 30 years. The problem occurs sporadically and is characterised by variation in redness in highly pigmented muscles. There are two possibilities:

1. The myoglobin has been converted to a pink haemochrome during heating. This seems to occur at higher temperatures (>76°C).
2. The myoglobin has not been completely denatured. This seems to occur at lower temperatures (<76°C).

There seems to be some relationship between pH and cooked colour, but other factors are involved. High pH (above 6.0) stabilises myoglobin against the effects of heat, so decreases the percentage of myoglobin denatured during cooking, and this can be sufficient to produce obvious colour differences.

Pinking and premature browning when cooking

**Pinking in cooked meat**

Although cooked meats are typically grey-brown, pinking is not uncommon. Several conditions other than undercooking may result in pink or red colour of cooked meat. Some of these are:

- incomplete denaturation of myoglobin, as described above. This is associated with high pH and high pigment concentration;
- contamination with nitrite/ nitrate/ ammonia, or exposure to carbon monoxide or nitric oxide gases can lead to red pigments;
- the grey-brown pigment is not 100% stable: it can slowly be reduced to the pink haemochrome, and the interior of large roasts may slowly turn pink during refrigerated storage due to this effect.

Pink colour after cooking patties is due to incomplete denaturation of myoglobin. It is much more likely to occur in patties that have been frozen. Most frozen patties will brown when cooked to 71°C, but a few may retain a red to pink colour. A higher internal temperature (around 81–84°C) will be needed to completely remove the pink/red colour from these patties. Red internal colour at 71°C seems to be more frequent in products containing less than 20% fat. One study showed that the internal colour of patties cooked to 71°C within 12 hours of thawing at 7°C remained red-pink. Only after 18 hours or more of thawing did cooking to 71°C result in a well-done appearance. The cooked colour of patties which were thawed while vacuum-packed, was redder than patties thawed non-vacuum packed. The conclusions were that the best ways to prevent pinking was (i) to produce patties immediately prior to cooking, and not frozen, or (ii) if frozen, patties should be thawed in air for 18 hours prior to cooking. Thawing in air allows oxygenation of the myoglobin, and thus the grey-tan denatured globin haemachrome forms. When the patties are cooked from frozen, and are not fully oxygenated, the pink denatured globin haemochromes can form, so the interior of the patty looks pink. It is not, however, possible to apply the findings of the above study to a commercial situation.

Pink cured colour may result if meat is exposed to nitrite in ingredients, or nitrogen dioxide (NO₂) during cooking (surface-pinking). The nitrogen dioxide may be present in the combustion gases in gas ovens. Meat cooked in a gas oven, or heavily smoked, frequently develops surface pinking. Upon slicing, a pink ring is observed to a depth of...
~8–10mm from the surface. Pink ring is a traditional and desirable attribute in some products, e.g. ‘Texas BBQ’ beef roasts, but in most cases, the surface pinking is undesirable since consumers may associate pinking with undercooking. It was thought that carbon monoxide (CO) or nitric oxide (NO) produced by gas combustion was the cause of the pinking, but research has shown that nitrogen dioxide (NO₂) is probably the culprit. During cooking, the presence of up to 149ppm CO or 5ppm NO did not cause pinking, but as little as 0.4ppm NO₂ caused pinking of turkey rolls, and 2.5ppm caused pinking of beef roasts. Nitrogen dioxide has a much greater reactivity than nitric oxide with moisture at meat surfaces.

Premature browning

This is the other extreme, where ground beef appears thoroughly cooked at internal temperatures as low as 55°C. It is particularly associated with unaged ground beef that is stored in air. Premature browning appears to be related to frozen storage, but this is not the primary cause. It seems that if the meat is more oxidised, it is more likely to show premature browning, so any factor that promotes oxidation of myoglobin, or limits the reducing capacity of the meat, can result in premature browning. For example, aged meat has less reducing capacity than fresh meat. There have been some studies on the effect of the animal or diet (young animal or old animal, Vitamin E supplemented or not), but it seems that while it affects display life (see above), it has little effect on the internal colour of cooked patties.

Premature browning is particularly associated with patties that are frozen and thawed before cooking. Some researchers have managed to manipulate the thawing process to prevent premature browning—by thawing patties in vacuum packs, then giving them 4.5 hours anaerobic reduction by holding at 22°C before bringing them back to chill temperature (3°C) before cooking. If the patties were cooked from frozen, without thawing, premature browning did not occur.

When cooking patties, the juice produced becomes less red and more yellow as the endpoint temperature increases, but it doesn’t tend to run clear, as suggested in a number of cooking guidelines. Visual evaluation of patty colour is not an accurate indicator of doneness, and a more appropriate guideline would be ‘cook until the juices lack redness’. Cooking to a recommended internal temperature (e.g. 71°C) to achieve destruction of live pathogens remains the most reliable indicator, however.

Uneven surface browning of roasts

The Maillard reaction is a non-enzymatic reaction between amino acids and reducing sugars, which occurs during cooking. This reaction is responsible for the production of many of the flavours and odours associated with cooked meat. It also results in browning of the meat through production of pigments called melanoidins. These contribute to the dark brown colour on the surface of roasted meats, which is a key factor in consumer acceptance of the product. The actual products formed from the Maillard reaction depends on the duration and temperature of cooking, moisture content and pH, as well as the nature and concentration of the sugars and amino acids involved. There is a significant correlation between the temperature of cooking and increasing production of melanoidins. It also proceeds more rapidly at low moisture levels (optimum aw 0.65-0.75), which is why browning is greater at the surface, which has become dehydrated during cooking. Where the roast is in contact with the pan, or in contact with wrapping, the surface remains moist and there is less browning.