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Reflectance spectra and colour of beef packed in different concentrations of oxygen.

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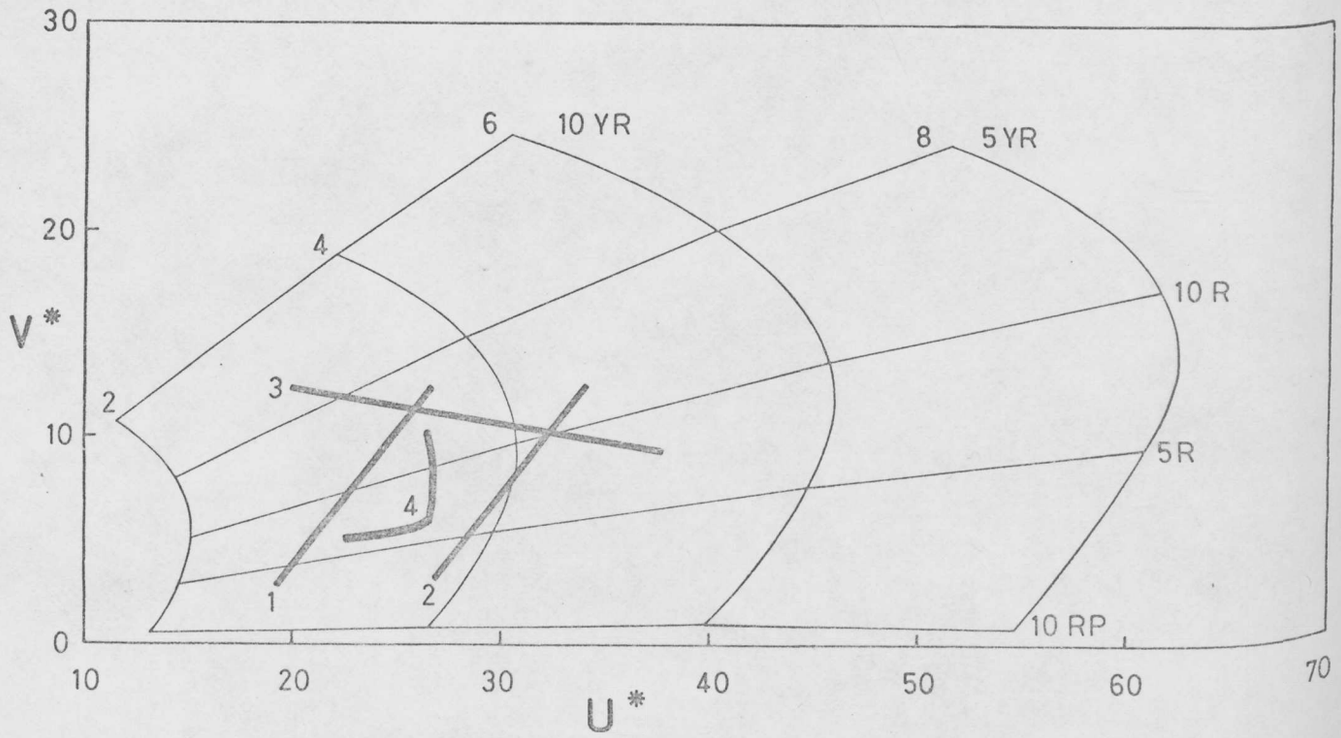
INTRODUCTION

The attractive red colour of fresh meat is produced by the oxygenation of the purple haem pigment myoglobin to oxymyoglobin, but continued exposure results in its oxidation to brown metmyoglobin. The rate of oxidation is maximal at low partial pressures of oxygen (George and Stratmann[1]) and for meat packed in the presence of oxygen there are effectively two separate oxidation zones, one at the boundary near the limit of oxygen penetration where the low partial pressure rapidly forms metmyoglobin displaying the typical brown-green subsurface band when cut, (Brooks[2]) and the other, within the surface layer in which the partial pressure is high and the rate of oxidation considerably slower. For meat packed in air where the surface oxymyoglobin layer is relatively thin, oxidation proceeds by the subsurface brown layer expanding upwards and diluting the surface red layer until it is displaced and unsightly brown areas develop (MacDougall and Rhodes[3]). Recent literature, for example Georgala and Davidson[4], has indicated that the shelf life of fresh meat can be extended if it is packed in the presence of high concentrations of oxygen to preserve redness and in the presence of carbon dioxide to retard microbiological spoilage. The relationships of gaseous environment to appearance are more fully discussed in the paper by Taylor[5] in this symposium.

Visual acceptability limits of fresh beef colour in objective terms have appeared from time to time in the literature and are illustrated after transformation from their original colour systems to the uniform colour spacing of the 1964 C.I.E.[6] and Munsell[7] systems in Fig. 1[3, 4, 8].

Fig.1. 1964 C.I.E. uniform colour diagram showing Munsell hues 10 RP (Red Purple) to 10 YR (Yellow Red), chroma 2 to 8, and published limits of acceptability of fresh beef colour.



1. "Undesirable", (Hilbert et al[3])
2. "Desirable", (Hilbert et al[3])
3. "Unacceptable", (Georgala and Davidson[4])
4. "Definite Brownness", (MacDougall and Rhodes[5]).

The objects of this present study were to ascertain the acceptability limits of the various colours of fresh meat in objective terms when packed in different concentrations of oxygen, and to determine the effects of oxygen concentration on the reflectance spectra and development of metmyoglobin.

MATERIALS, METHOD AND PROCEDURE

Beef topsides (semimembranosus) from prime animals approximately 18 months old were purchased locally and packed 2 days after slaughter. The meat was held + 1°C during cutting and packaging.

Experiment 1. Circular slices, 20 mm thick and 55 mm in diameter were packed into cans in such a way that the entire surface was exposed to the gas mixture in the head-space. The cans were sealed, a small hole punched into the lid, mixtures of either 40% O_2 + 20% CO_2 + 40% N_2, 60% O_2 + 20% CO_2 + 20% N_2, 80% O_2 + 20% CO_2 or 100% O_2 flushed into the headspace and the hole immediately sealed with solder. The ratio of headspace gas volume to meat volume was 1.2. Slices were also packed in expanded polystyrene trays for exposure to air and overwrapped with a commercial clear oxygen permeable film with a permeability of >5000 ml (STP) m^{-2} 24 hr^{-1} atm^{-1}. The samples were stored at 0 ± 0.5°C and + 5 ± 0.5°C and the canned samples examined after 1, 4 and 14 days storage. The wrapped samples were examined after 1 and 4 days storage and then daily until beyond visual acceptance.

Experiment 2. Circular slices, 50 mm thick and 55 mm in diameter were packed into cans to give headspace gas volume to meat volume ratios of 0.9, 1.5 and 2.7. The gas mixtures used were 40% O_2 + 20% CO_2 + 40% N_2, 60% O_2 + 20% CO_2 + 20% N_2 and 80% O_2 + 20% CO_2 and the samples stored at 0 ± 0.5 °C. Cans were examined after 1, 2, 5, 8 and 12 days. Slices wrapped in film for exposure to air were also stored at 0 ± 0.5°C and examined daily for 9 days.

Headspace gas. Analysis of the headspace gas was carried out on a Fisher Gas Partitioner.

Oxygen penetration. For meat packed in cans, the depth of oxygen penetration was determined immediately upon removal from the can by cutting through the slice and measuring the thickness of the oxymyoglobin + metmyoglobin layer with a magnifying glass with an integral 0.1 mm scale. For meat wrapped and exposed to air, the depth was measured by a method based on that devised by Brooks.[2] Meat with one edge covered by the film was pressed between two Perspex plates, stored with the packages, and the thickness of the layer measured through the plates.

Objective colour measurement. Reflectance spectra were measured between 400 and 700 nm using an Optica CF4DR double beam recording spectrophotometer with integration sphere, specular light trap and barium sulphate as reference. The tristimulus values were determined for source C and the 1964 C.I.E. uniform chromaticness and lightness co-ordinates U*, V* and W* were calculated[3].

Subjective colour assessment. Each sample at the time of objective measurement was visually placed into one of three grades, very attractive-grade 1, attractive enough to purchase-grade 2 and unacceptable-grade 3. The allocation of a sample to a particular grade at time of examination was confirmed later by subjective judgement of projected photographic transparencies taken under standardized conditions.

Pigment analyses. Myoglobin plus residual haemoglobin was determined by the technique of Wierbicki et al[9]. The pH of the tissue extract was recorded. The relative proportions of myoglobin, oxymyoglobin and metmyoglobin were assessed from the reflectance spectrum by the K/S ratios at 474/525 nm and 571/525 nm as described by Stewart, Zipser and Watts[10], and Snyder and Armstrong[11].

RESULTS

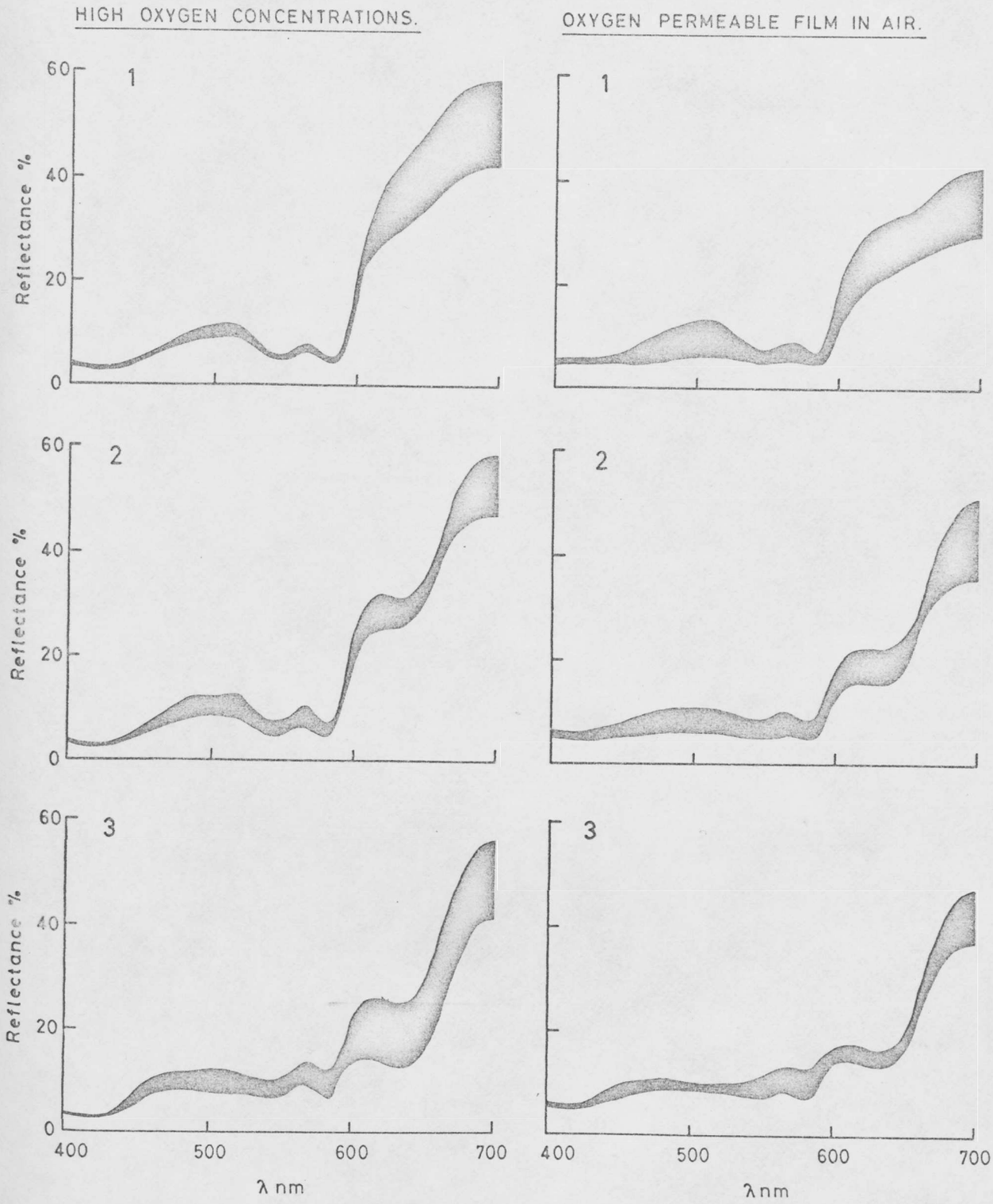
Pigment concentration and pH. The concentrations of myoglobin + residual haemoglobin, determined on each sample, were similar for both experiments; the mean ± standard deviation for experiment 1 was 0.51 ± 0.06% and for experiment 2 was 0.52 ± 0.08%. The pH of the meat in both experiments was 5.5.

Subjective assessment of colour. Little difficulty was experienced in giving samples a definite visual grade. A few were difficult to classify but after the colour space for each group was established, it was found that those few, objectively as well as subjectively, lay on the border-line between groups and would therefore be expected to produce disagreement among observers.

Typical reflectance spectra. The reflectance spectra for meat packed in oxygen concentrations greater than air in experiment 1 were allocated to grades 1, 2 or 3, any borderline case being rejected. The spectra were then superimposed upon one another and the range in reflectivity plotted (Fig.2). The minimum number of spectra used for each grade was 6. In grade 1, the most attractive of all, the range was greatest between 600 and 700 nm (14% at 650 nm) and much less at wavelengths < 600 nm (2% at 500 nm). The changes in the spectra from grades 1 to 3 were caused by the progressive formation of metmyoglobin as evidenced by the characteristic increase in absorbance (decrease in reflectance) at 630 nm and overall loss of red reflection. The mean reflectivities at 630 nm were 34, 28 and 20% respectively for grades 1, 2 and 3 and the mean concentrations of metmyoglobin calculated from the reflectance spectra were 13, 23 and 60%. At high concentrations of oxygen, oxidation of oxymyoglobin to metmyoglobin occurred uniformly or nearly uniformly throughout the oxygenated layer. The metmyoglobin band, if present, was far enough removed from the surface to be of little consequence in the rate of colour deterioration, with the definite exception of 40% oxygen at 5°C.

The samples wrapped in film and stored in air were allocated to the three grades and the reflectance spectra plotted (Fig. 2). The minimum number of spectra used for each grade was 8. The presence of the film increased the reflectivity by 1 to 2% in the Soret region (400 to 440 nm) but this increase made only a minor contribution to the differences in the colour between high oxygen concentrations and air when compared with the effects on the colour produced by the substantially lower reflectivity between 600 and 700 nm. The typical spectra in grade 1 were chosen from individual spectra recorded within the first 2 days of storage. At 0°C and high oxygen concentrations, for example 80%, the 20 mm thick slice was completely oxygenated throughout its thickness and myoglobin was eliminated as a contributor to the spectrum. However, for meat packed in air, the oxymyoglobin surface layer was only 2.0 to 2.5 mm thick and the subsurface layer of myoglobin with its already forming boundary of metmyoglobin considerably reduced the reflectivity at > 600 nm; at 650 nm the reduction in reflectivity from the fully oxygenated spectra was approximately 12%. The pattern of oxidation from grades 1 to 3 for meat in air was by dilution and rapid displacement of the upper oxymyoglobin layer by the subsurface metmyoglobin layer. The reflectivity at 630 nm for grades 1 to 3 were 25, 19 and 13% respectively and the metmyoglobin concentrations were 17, 37 and 71%. Myoglobin was not detected in either of the mean spectra from group 1 by the K/S ratio technique[10, 11] although the differences > 600 nm would indicate its possible presence in the samples packed in air.

Fig. 2. Typical reflectance spectra showing range for each group (1, 2 and 3) of meat samples packed in high concentrations of oxygen (40 - 100%) and in air.



group 1 - Very attractive
group 2 - Attractive enough to purchase
group 3 - Unacceptable

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Uniform colour space. The mean lightness and colour co-ordinates for the typical spectra are shown in Fig.3 for high oxygen concentrations and Fig.4 for meat exposed to air. The limits in each group are based on the subjective judgement of all samples examined in the two experiments.

Fig.3. 1964 C.I.E. uniform colour diagram of typical spectra and subjective limits for groups 1, 2 and 3 for meat packed in high concentrations of oxygen (40-100). The mean lightness values (W*) are given in parenthesis.

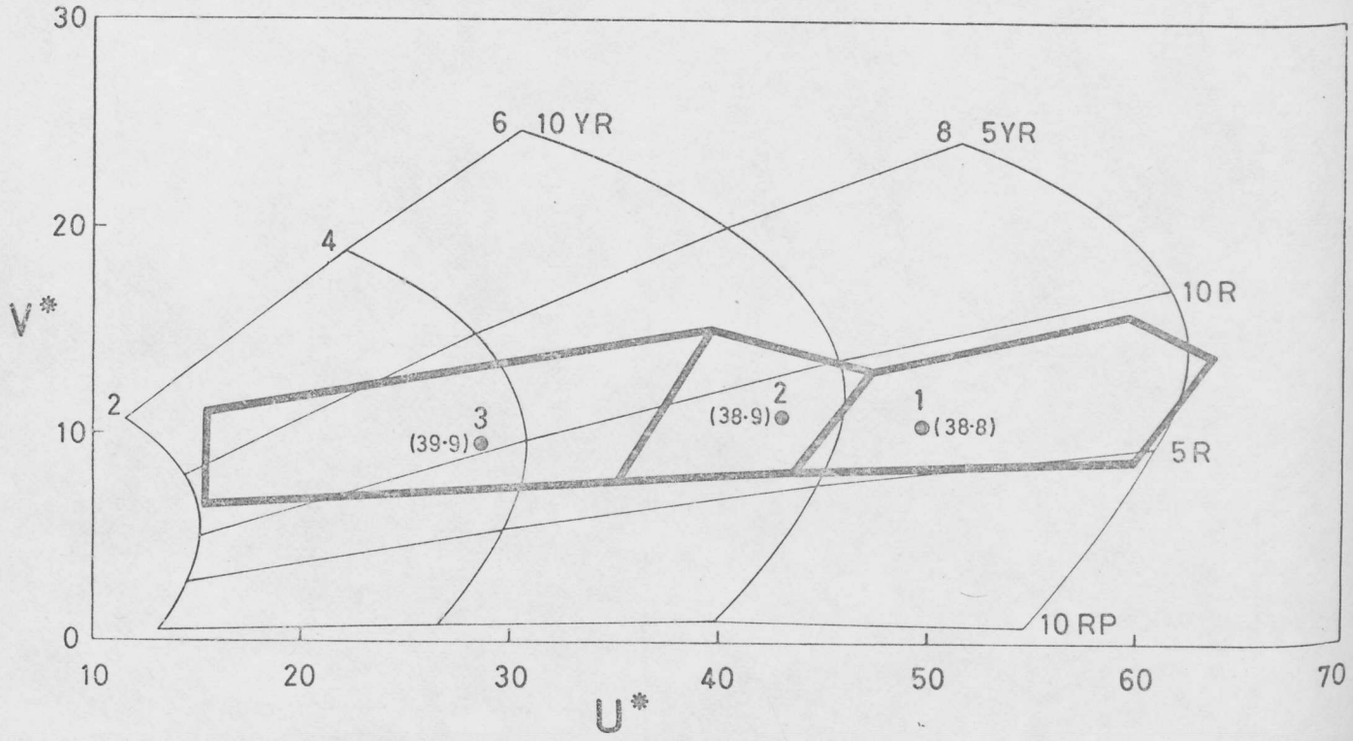
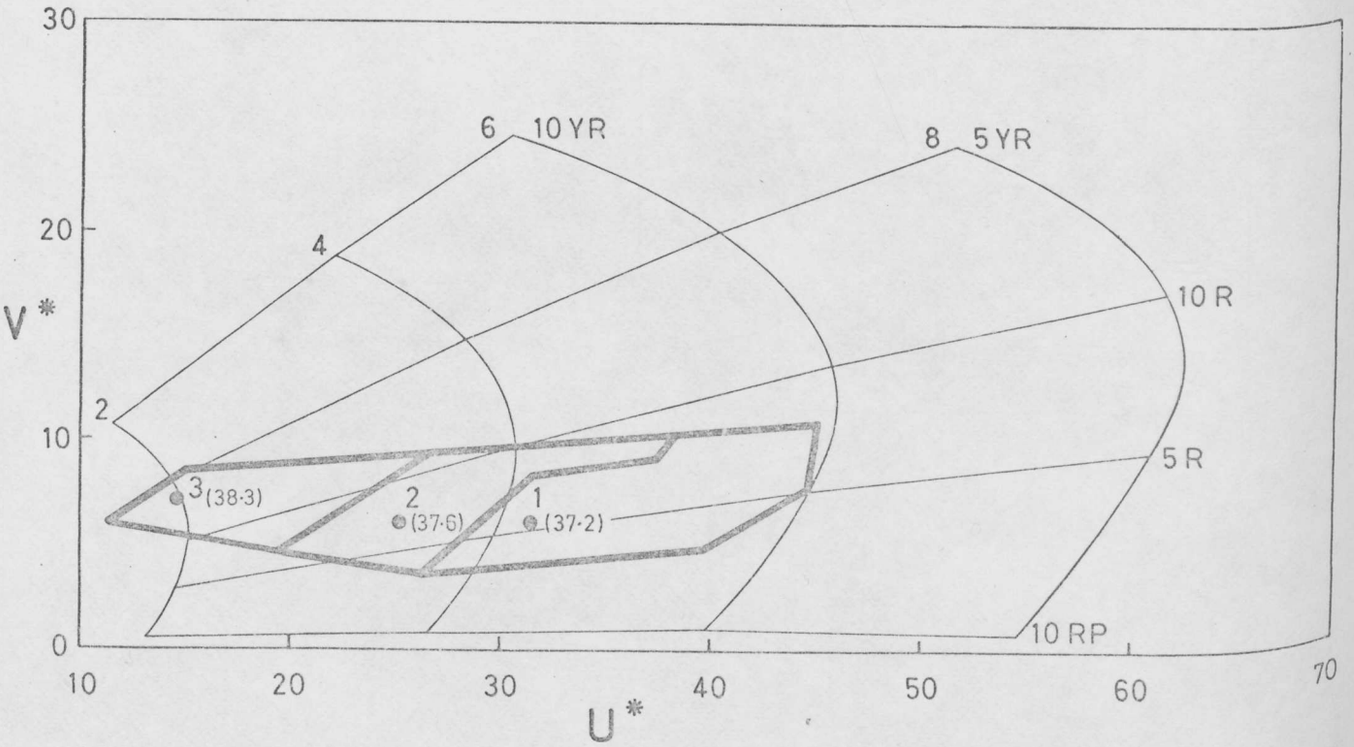


Fig.4. 1964 C.I.E. uniform colour diagram of typical spectra and subjective limits for groups 1, 2 and 3 for meat packed in air. The mean lightness values (W*) are given in parenthesis.



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Grade 1 samples packed in high oxygen concentrations had almost the same hue as those exposed to air, but were much more saturated. The size of the saturation difference was approximately 2 Munsell chroma steps or 10 C.I.E. units of colour differences, equivalent to about 10% of the visual distance from black to white. Grade 2 material from high oxygen concentrations was also more saturated than either grade 1 or 2 packed in air, and its hue was less red by about 2 Munsell hue steps. In addition to the differences in saturation and hue between groups 1 and 2 for both sets of material, meat which was packed in high oxygen concentrations was also objectively lighter by approximately 1.5 units of uniform lightness W*, which was caused entirely by the increased red reflection from the thicker oxygenated layer.

Effect of storage condition on colour change. The rates of change in oxygen pressure within the cans are reported by Taylor[5] in this symposium. In no case did the oxygen pressure drop below 0.25 atms. The effects of the composition of the gases in the headspace on the growth of bacteria are reported by Kitchell[12].

All samples during the first two days of storage either at 0°C or 5°C and irrespective of oxygen concentration were grade 1. By 5 days at 0°C, all the canned samples between 40 to 100% oxygen were either grade 1 or 2, but the meat wrapped in film was grade 3. After 8 days at 0°C, only oxygen concentrations of 60% or more were grade 2, and by 12 days, all were definitely grade 3 or near its boundary. Increasing the temperature from 0° to 5°C markedly affected the rate of colour deterioration, for example in air and at 40% O_2 there was a grade change from 1 to 2 after 1 day, and at 80% O_2 there was a grade change from 1 to 2 after 4 days.

Discussion

Fresh meat is translucent. Its reflectance spectrum is controlled by the relative quantities of the light absorbing pigments, myoglobin, oxymyoglobin and metmyoglobin and the light scattering properties of structural components, the myofilaments and connective tissue proteins etc. Meat colour is determined by the first 8 mm of tissue since absorption and scatter beneath this do not contribute to the reflectance spectrum[13]. In the case of meat packed in air within a few days of slaughter[3], the thickness of the surface oxymyoglobin layer is much less than 8 mm and it does not obscure the subsurface layer which contains myoglobin and/or metmyoglobin. Differences in pigment absorption between the layers have a large effect on the reflectance spectrum, especially at >600nm where oxymyoglobin is less absorbing than either myoglobin or metmyoglobin (Bowen[14]). In the case of meat packed in high concentrations of oxygen, the surface layer of oxymyoglobin extends beyond 8 mm and therefore its spectrum is more highly red reflecting and the colour is visually more saturated (brighter). During storage at high oxygen concentrations, oxidation of the thick oxygenated layer to metmyoglobin proceeds uniformly throughout the layer and the high level of colour saturation persists for longer than that of meat packed in air where the subsurface band of metmyoglobin rapidly lowers the saturation level as it approaches and displaces the oxymyoglobin surface.

The direction or colour change during oxidation is essentially parallel to the U* axis of the 1964 C.I.E. diagram. A decrease in U* at a constant value of V* is equivalent to both a loss in saturation and a change in hue towards yellow. For meat packed in air, (Fig.4) the acceptability limits observed in this study confirm the limits found by Rikert et al[9], that is lines 1 and 2 in Fig. 1 are almost identical to the boundaries between groups 3 and 2, and 2 and 1. The limits of saturation and hue previously suggested by MacDougall and Rhodes[3] for the start of definite brownness meet on top of the typical colour of group 2, equal to a calculated concentration of 37% metmyoglobin. The unacceptable limit of 34% purity (Georgala and Davidson[4]) is applicable only to the high oxygen situation (Fig.3) since red acceptable air packed meat was often less than 34% in purity. The differences in the positions of the acceptability limits between Figs.3 and 4 illustrate the

visually compensating effect of saturation upon hue. In Fig.4 the boundaries are redder in hue but less saturated than the same acceptance boundaries in Fig. 3 where the increased saturation compensates for the distinct hue change towards yellow for grade 2.

The extension of the storage life of fresh meat by high concentrations of oxygen to preserve acceptable colour in the presence of carbon dioxide to retard the growth of bacteria would appear from this investigation to be of considerable value provided efficient temperature control was maintained up to and during display. Such techniques if brought into commercial practice would require stringent quality control at point of display, for although packs of meat stored for some time at high oxygen concentrations might be acceptable in the grade 2 condition if displayed on their own, their appeal would be lost if grade 1 material were present.

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