Color Measurement of Strongly Scattering Media, with Particular Reference to Orange-Juice Beverages

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The color of a liquid of high turbidity, such as orangeade, is neither a transmission nor a reflection color. Color coefficients of a 5-cm i.d. cylindrical cell filled with orangeade measured with an all-sided illumination technique are in substantive agreement, however, with those of a colored card selected to visually match the color of the orangeade sample, the colored card being measured with standard diffuse reflection techniques. The design of such an illuminating hollow sphere, which has the sample in its center, is described. From independent goniophotometric studies on the 5-cm. i.d. cylindrical sample, it is deduced that pure transmission ($\theta = 180°$ if θ is defined as the angle between the direction of observation and the direction of the incident light) contributes very little to the total intensity of the emerging light under conditions of all-sided illumination; directions of maximum contribution vary from $\theta = 146°$ ($\lambda = 400$ nm) to $\theta = 166°$ ($\lambda = 700$ nm). Some applications of the color measuring technique are described; these include color control of commercial beverages, effects of coloring— or stabilization additives, the effect of bottle shape, and the direct measurement of spectral characteristics. In addition, some color coefficient data are given on the transmission color of carotene solutions and of orange juices and orangeades of various concentrations.

The analysis of carotenoids in citrus juices has been the subject of intensive investigations and has recently been reviewed by Davies (1965), Huslin (1962), Calvarano (1963), and Kock and Sajak (1965). Important as such analyses are, they do not give direct information on the visual color of the commercial product. There is no one-to-one correspondence between the concentration of carotenes and the color specification of, say, orangeade. The color measuring technique described in this paper provides a convenient means for the specification of the color of certain strongly scattering samples in terms of three color coefficients according to the CIE standard procedures. For nonscattering samples as well as for samples of low turbidity (optical density less than 0.1) standard transmission spectra provide satisfactory and meaningful color coefficients. An impressive arsenal of theories basically developed by Rayleigh (1881), Debye (1915), and Mie (1908), is available in this field. Of these, Mie scattering is the most general form since it does accommodate large particles and light absorbing particles. However, these theories of scattering all assume the absence of multiple scattering and are therefore of no use for the description of scattering in orangeades where multiple scattering is an important, if not dominating, feature. Standard diffuse reflection techniques can be used successfully for very strongly scattering samples. Such reflection spectra, like transmission spectra, can be directly converted into color coefficients. No microscopic theory exists for this case, but Kubelka and Munk (1931) and Kubelka (1948) have been able to express such diffuse reflection in terms of absorption and scattering coefficients. Their theory assumes, however, that the sample is a perfect diffuser. As our experiments show, orangeades and similar materials are very far from ideal diffusers and, therefore, Kubelka-Munk type analyses are not appropriate for such samples. Our transmission and diffuse reflection experiments on orangeade indicate that if any light intensity could be measured at all, the brightness was very low and nowhere near the brightness one would expect considering the appearance of a glass of orangeade. Apparently, orangeades and samples of similar turbidity fall in between the two classes of recognized and standardized theories and measuring techniques mentioned above. Our new method intends to fill this gap. This technique has the promise of providing a standardization of color specifications of such samples, but also shows how factors other than the amount of coloring agent affect the color. Our experiments also indicate that parameters like stability of color (against sedimentation, flotation, or oxidation), particle size and particle size distribution (both of noncolored and colored particles), the nature of the color dispersion (crystals, oily drops, or otherwise) and the shape of the container all contribute considerably and measurably to the observed color.

In the following text we will use the term "absorbance" (or transmittance) only in relation to molecular disperse, nonscattering solutions. The term "attenuance" will be used for samples where scattering, absorption, and multiple reflection all contribute to the total of transmitted light that reaches the detector under a narrow angle. Our experiments with an all-sided illuminated sample could be called "diffuse integral attenuance" in Shibata's nomenclature (Shibata, 1958); instead we propose to use the term "translucence."

Also, in this paper, "juice" will refer to the fruit juice in its natural concentration. The 6 to 10 times diluted, usually carbonated beverages derived from natural juices will be called orangeade, lemonade, limeade, etc. Some care in nomenclature is recommended here, since in many countries "lemonade" means a carbonated drink based on the natural juice of some fruit, not necessarily lemon. In such places

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Table I. Color Coefficients of β-Carotene Solutions (in Hexane, 1.0 cm Cell Length)

c × 10⁶ (Mole/l)	x	y	Y (%)	λ_d (nm)	P
185	0.573	0.427	43.1	590	1.000
74.0	0.535	0.465	58.9	583.8	1.000
37.0	0.512	0.486	69.1	580.0	1.000
18.5	0.494	0.501	76.6	577.3	1.000
7.40	0.476	0.513	83.2	575.4	1.000
3.70	0.454	0.514	88.8	573.4	0.944
1.48	0.412	0.466	92.3	572.5	0.784
.740	0.371	0.409	95.2	572.5	0.536
.370	0.338	0.363	97.5	572.5	0.268
.148	0.322	0.336	99.0	572.5	0.145

"orangeade" may even refer to a synthetic product, while the natural product is then sometimes called "orange-lemonade" (contains only juice of oranges!).

PRELIMINARY EXPERIMENTS

In Table I, the results are given on the transmission color of β-carotene solutions of varying concentrations (in hexane, 1 cm cell length).

Figure 1 (upper locus) shows these results in a graphical form. Such a locus is typical of Beer's plots of compounds that absorb only at one end of the visual spectrum. The dominant wavelength λ_d is constant at 572.5 nm for the dilute solutions. Its complementary color is at $\lambda = 468$ nm, close to the maxima of absorption at 452 and 478 nm. For the high concentrations the solutions act as cut-off filters, the resulting "edge-colors" (Bouma, 1947) following the locus of the saturated spectral colors.

It appears that for homogeneous β-carotene solutions, depending on concentration range, one can either influence the saturation (at constant λ_d and little change in Y) or the dominant wavelength (at constant saturation p and large changes in Y).

In Figure 2, the attenuance spectrum of an orangeade (2.0 cm cell length) is given, along with an absorbance spectrum of a β-carotene solution in hexane (0.2 cm cell) that had
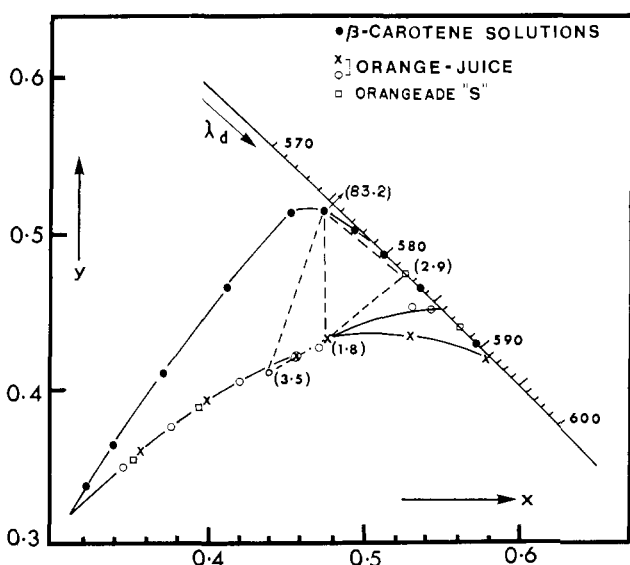

Figure 2. Attenuance spectrum of orangeade in a 2.0 cm cell (———), the absorbance spectrum of β-carotene solution in hexane in a 0.2 cm cell (- - - - - -), and the "translucence" spectrum of orangeade in a 5 cm cylindrical cell (·—·—·—·—). The β-carotene concentration in all three samples was approximately equal

the same β-carotene concentration as the orangeade. The difference is very striking, but there can be little doubt that the color of the orange is actually caused by the carotenes. For example, lemonade, which is physically and chemically (except for the carotenes) very similar to orangeade, has a greyish color; the same colorless hue can be reproduced by extracting the carotenes from orangeade. Also, using the opal glass technique of Shibata *et al.* (1954) an orangeade sample will give a "semi-integral attenuance" or "translucence" spectrum that is similar to that of a suspension of β-carotene crystals (Shibata, 1956).

Table II gives the coefficients of attenuance color (2 cm rectangular cell) of dilutions of two orange juice concentrates (concentration factor 6×); both samples had approximately the same carotene content.

These results have been included in Figure 1. The most conspicuous differences with the color of β-carotene solutions are the red shift of the dominant wavelength, the loss in

Figure 1. x,y, color plane with loci for the data of Tables I, II, and III. Dotted lines connect points representing samples of approximately equal carotene content; numbers in brackets are the brightness data Y(%)
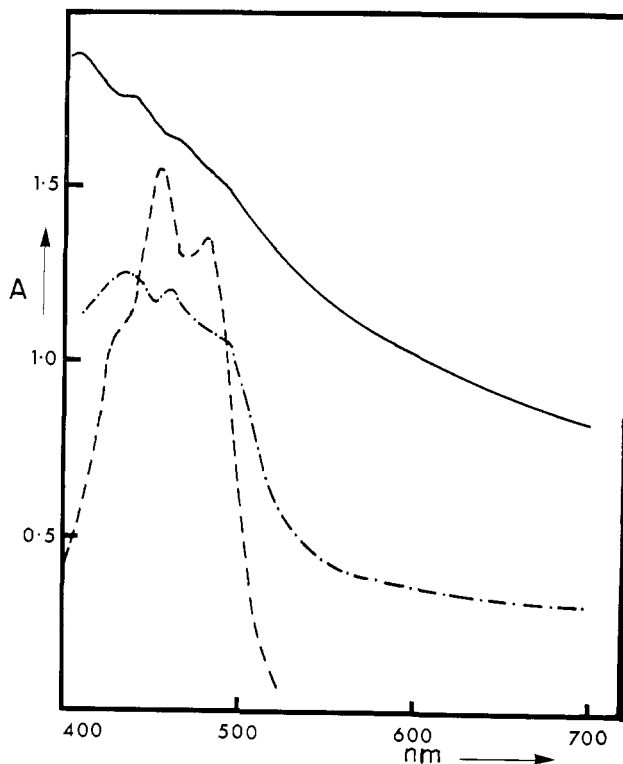
Table II. Attenuance Color Coefficients of Orange Juice Concentrates and Dilutions (2 cm Cell)

Concentration[a]	Sample "6" x	Sample "6" y	Sample "6" Y(%)	Sample "35" x	Sample "35" y	Sample "35" Y(%)
6.0	0.544	0.449	0.76	0.581	0.419	0.37
3.0	0.529	0.453	0.81			
2.0	0.471	0.426	1.16			
1.5	0.457	0.421	1.81	0.530	0.435	0.83
1.0	0.440	0.410	3.47	0.476	0.432	1.82
0.75	0.422	0.402	6.81	0.457	0.421	4.61
0.38	0.377	0.374	22.3	0.401	0.393	18.6
0.19	0.346	0.350	45.5	0.357	0.360	42.5

[a] Relative to juice in natural concentration.
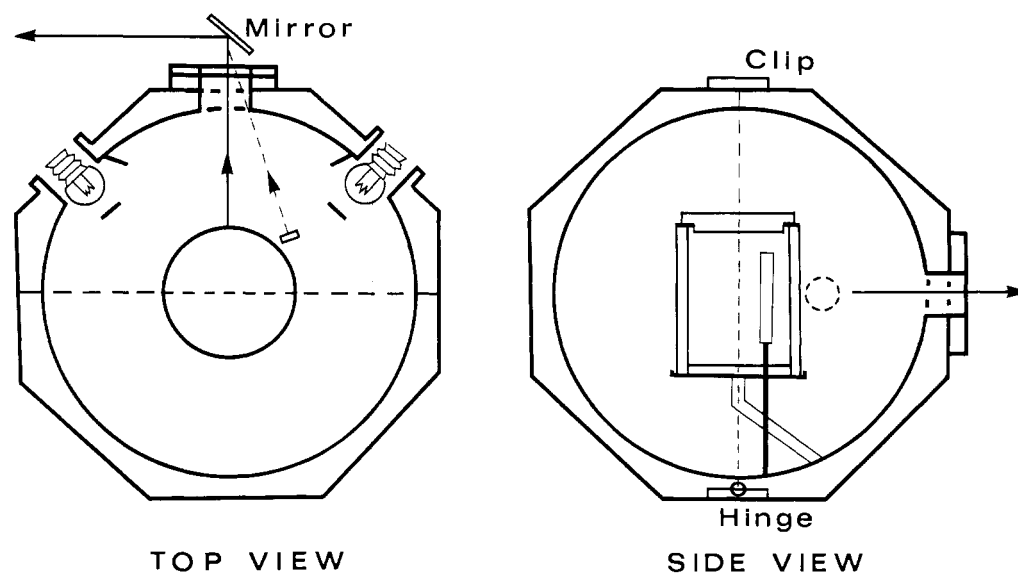
TOP VIEW   SIDE VIEW

Figure 3. Design of the illuminating sphere

Table III. Attenuance Color Coefficients of Orangeade "S" as a Function of Cell Length

Cell length (cm)	x	y	Y(%)
5.000	0.560	0.440	0.13
2.000	0.527	0.476	2.9
1.000	0.394	0.384	14.6
0.500	0.349	0.354	34.8

Table IV. Color Coefficients Indicating the Performance of the Diffusing Sphere with All-sided Illumination

Sample	Method of measurement	x	y	Y(%)
Munsell card 7.5Y/8/10	Zeiss RA-3 Diffuse illumination, perpendicular reflection	0.401	0.433	58.7
Orangeade "S" in 5.0 cm (i.d.) cylindrical cell	Diffuse all-sided illumination with diffusing sphere	0.411	0.447	42.1
Orangeade "S" in 5.0 cm rectangular cell	Transmission (Cary 14)	0.560	0.440	0.13

brightness, and the considerable loss in saturation. Edwards *et al.* (1966) have reported on the reflection color of orange juice concentrates; it seems likely indeed that reflection is the more useful technique for such samples. Also included in Figure 1 are the attenuance color prints of a commercial orangeade in cells of various lengths (Table III).

The very low brightness (Y = 0.13%) for the attenuance color of a 5-cm layer of orangeade means in effect that a glass of orangeade is virtually black (at least as far as transmitted light is concerned) in seeming contrast to everyday observation. Subsequent attempts to measure the diffuse reflection of orangeade failed; irrespective of the reflection technique used, no diffuse reflection could be detected at all. To account for the color of orangeade and in particular its brightness, only one alternative seems left—scattering and multiple reflection from all directions. Indeed, one usually views orangeade under conditions of all-sided illumination and this could possibly account for the observed brightness.

DESIGN AND PERFORMANCE OF AN ILLUMINATING HOLLOW SPHERE

To test the idea of a color formed by the integral contributions of an all-sided illumination, a diffusing sphere was constructed that holds the sample in its center. The design was made to fit the RA-3 reflection accessory to the Zeiss PMQII spectrophotometer. The RA-3 has a detachable Ulbricht sphere (diameter 7.5 cm) with the sample holder on a wall port. The optics are such as to image the plane of the sample on the monochromator slits. It, therefore, followed that for a sphere holding a sample in its center, the diameter had to be twice (15 cm) as large as the original sphere. No further modification of the optics is then required. The increased size of the sphere has the added advantage of allowing the use of a 5-cm (i.d.) cylindrical cell, thereby holding close to the dimensions of a common drinking glass. Figure 3 gives the essentials of the design.

Two 6V, 30 watt Tungsten lamps, fed by the spectrophotometer's power supply, served as the light sources. Baffles were placed such as to prevent the lamps shining directly on sample, reference plate, or exit port. The sphere was made from solid aluminum; the two halves were connected by means of a hinge and a fastening clip; aside from a constructional necessity, this also provided a convenient means of sample insertion. The insides of the sphere were coated with MgO in a coarse surface texture. A MgO coated plate, placed inside the sphere, acted as the reference; the mirror in the RA-3 set can be rotated over about 8° to alternately allow light from reference and sample into the monochromator.

It is difficult to establish whether the experimental setup of this diffusing sphere will measure the exact and correct color. The following experiment at least gives a fair indication. In a visual test in diffuse daylight, the color of the 5 cm cylindrical cell filled with orangeade was compared with colored paper strips from the "Munsell Colour Atlas" (Hilger & Watts, London, U.K.), and the best matching strip selected. The two samples were then measured, the colored paper strip with the original RA-3 accessory, the orangeade with the new diffusing sphere. Table IV shows the results (the attenuance color of the same orangeade is included for comparison). In Figure 2, the actual "translucence" spectrum is shown.

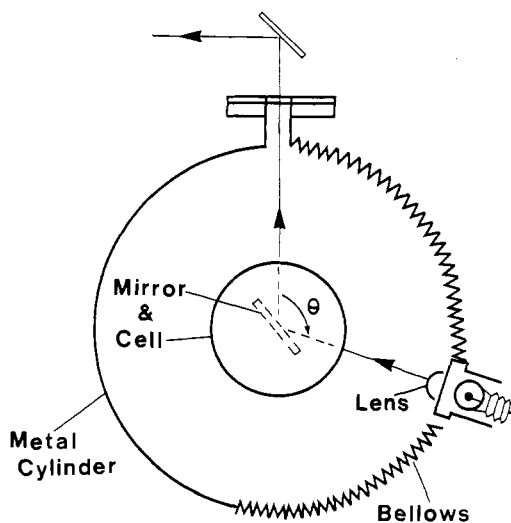The agreement between the data of the two upper lines of

Figure 4. Design of the gonio-photometer

Figure 5. Integrated intensities of scattered light as a function of scattering angle θ

Table V. Color Coefficients of Commercial Orangeades

Orangeade	x	y	Y(%)
"R"	0.415	0.441	42.1
"S"	0.411	0.447	42.1
"H"	0.423	0.440	40.3
"L"	0.446	0.457	41.7
"F"	0.426	0.443	42.7

Table VI. %T $\times$ 10^3 Values for a 5.0 cm (i.d.) Cylindrical Cell Filled with Orangeade, as a Function of Wavelength (nm) and of Observation Angle θ

λ(nm)	$\theta =$ 180°	$\theta =$ 150°	$\theta =$ 120°	$\theta =$ 90°	$\theta =$ 60°	$\theta =$ 30°
700	360	50	6.7	2.3	1.7	0.3
670	323	47	6.3	2.0	1.7	0.2
640	300	46	6.0	2.0	1.3	...
610	255	43	6.0	2.0	1.3	...
580	207	39	5.3	1.3	0.7	...
550	147	33	5.3	0.7	0.3	...
520	87	26	4.3	1.0	0.3	...
490	35	12	1.7	0.7	0.3	...
460	23	10.3	1.3	...	...	...
430	17	9.3	0.7	...	...	...
400	11	6.7	0.3	...	...	...

Table IV is as good as can be expected if only because of the limited number of color cards in the Munsell atlas. The extent of agreement is considered sufficient warranty for the use of the all-sided illuminating sphere in comparative, qualitative, and semi-quantitative color studies of highly turbid samples.

The reproducibility of color coefficients for any sample, measured over several months, was found to be within ±0.001 for x and y and ±0.1% for Y. Color resolution seemed such that any difference noticeable by visual inspection was reflected in the coefficients obtained, but no exhaustive tests were carried out to confirm this. A qualitative indication of the color resolution was obtained by studying a number of commercial orangeades (Table V). These orangeades did exhibit visual color differences when viewed under laboratory conditions, while a more casual inspection under noncontrolled lighting conditions would usually pronounce them of
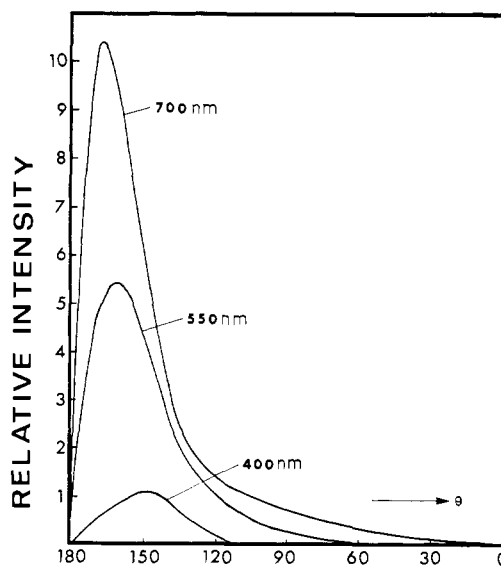
equal color (as was, no doubt, the intention of the various manufacturers).

ANGULAR WAVELENGTH AND INTENSITY DISTRIBUTION

Although it is gratifying that the illuminating sphere does work, there still remain questions of why and how. With no reflected light (less than 0.001%) and very little (about 0.1%) transmitted light, it seems difficult to account for the high brightness (around 40%) of the illuminating sphere experiments. A simple goniometer was constructed to measure the "transmitted" light as a function of the angle θ between direction of observation and the incident light beam. In Figure 4, the essentials of the design are sketched.

The goniometer consists of a metal cylinder of 15 cm diameter with a centered sample holder for a 5 cm cylindrical cell suspended from the top lid; the whole unit fits to the Zeiss RA-3 optics with an adapter. The inside walls of the cylinder were painted a matte black. A 6V, 30 Watt Tungsten lamp was mounted in the plane of the sample and the spectrometer optics; a slit and bellows construction allowed it to move in this plane. The angle θ could be varied between 30° and 180°. The sample holder and a small mirror mounted vertically on top of it could be rotated in any desired position. In addition, the sample with reference mirror unit could be moved along a vertical axis to either of two positions. In this way ratio measurements of reference and sample could be obtained. The moveable light source had a vertical diaphragm with a cylindrical lens in front of it, dimensions such as to give a sharp image both at the center of the cylinder and on the spectrometer slits.

A typical series of results obtained on an orangeade is given in Table VI.

The %T values for θ = 180° are practically identical to those for the straight transmission measurement mentioned before, corresponding to a brightness of Y = 0.14%. Table VI further shows that indeed light from all directions can find its way to the direction of observation (monochromator) although the %T values rapidly fall off with decreasing θ. It should be remembered, however, that these measurements were made with a constant light beam cross section. In order to obtain a better comparison with the illuminating

sphere experiments the data of Table VI were multiplied with a factor $\pi r^2(\Delta\theta)^2 \times (\cos\theta| + 2\sin\theta/\Delta\theta)$ this being the surface area element of a ring, cut out from a sphere between θ and $(\theta + \Delta\theta)$ and with radius r. For $\Delta\theta$ a value of 0.02 was assumed. This, together with a value of r = 75 mm, gives a surface element for $\theta = 2\pi$ of $\pi r^2 = 4.8$ mm². The diaphragm cross section of the light source was actually $2 \times 10 = 20$ mm², but allowance for the overfilling of the monochromator entrance slit and for the convergent action of the cylindrical lens in front of the light source diaphragm makes the above assumption reasonable.

Figure 5 shows some of these integrated intensities as a function of θ. It is seen that the maximum amount of light is not coming from the 180° direction but from directions ranging from $\theta = 166°$ ($\lambda = 700$ nm) to $\theta = 146°$ ($\lambda = 400$ nm). It is also seen that these integrated intensities are much larger than the "unit area" intensities of Table VI. By a second integration over θ itself (the areas under the curves in Figure 5), it is even possible to find the total amount of light of any particular wavelength from all directions on the sphere, and to calculate the color coefficients for this total. The results were: x = 0.55, y = 0.43, and Y = 15%. Although these results do not quite match those obtained with the illuminating sphere of the preceding paragraph, it is gratifying to note that the calculated brightness is about 100 $\times$ higher than for the transmission ($\theta = 180°$) measurements and only about a factor of 3 short of the correct value. In view of the somewhat loosely defined optics of the variable θ experiment, of the inherent problems in measuring the very low intensities of Table VI and of the multiple manipulations with these data, better agreement could hardly be expected.

APPLICATIONS

One important application for the illuminating sphere is already indicated by the data of Table V. The technique gives color coefficients close to those expected from visual inspection and has sufficient color resolution to distinguish between samples that are visually different. Therefore, quality control of orangeade and similar products now could include the measurement and control of color.

As a further illustration of color control, Table VII gives color coefficients of a series of orangeades which were manufactured from the same basic concentrate, but with different amounts of "water soluble" β-carotene (Hoffmann La Roche "Biochem 100") added. The natural "β-carotene" concentration was 14 mg per liter of natural juice.

Typically an increase in λ_d is obtained at a rather insignificant loss in brightness. It also seems indicated that the effect per mg of added carotene is far less than that of the natural carotene. Mechanical factors such as particle size (distribution) as well as the nature of the particles (oily drops $vs.$ crystallites) undoubtedly play a very important role here. Quantitative understanding of these factors is impeded, however, by the varied and very complex structure of the carotene chromoplasts (Strauss, 1953).

Another application is the study of stability against sedimentation or flotation or of chemical stability. In Table VIII some results are given on oxidation stability. A commercial orangeade was measured immediately after opening of the bottle and then again after 24 hours of exposure to air. The experiment was carried out on two samples, one containing no added carotene, and the second identical to the first except for the addition of an extra 15 mg/l β-carotene. The two samples also contained an antioxidant and an emulsifier. Table VIII, column 2, does indicate the effectiveness of the

Table VII. Color as a Function of Added β-Carotene

| | β-Carotene (mg/l) natural plus added | | | |
	14 + 0	14 + 5	14 + 10	14 + 15
x	0.430	0.441	0.451	0.457
y	0.444	0.446	0.447	0.447
Y(%)	39.6	38.1	37.1	36.2
λ_d	576.6	577.7	578.5	579.3

Table VIII. Effect of 24 Hours Exposure to Air (b) in Conjunction with the Effects of Added β-Carotene

| | | β-Carotene (mg/l) | |
		14 + 0	14 + 15
x	(a)	0.420	0.452
	(b)	0.416	0.452
y	(a)	0.438	0.444
	(b)	0.434	0.443
Y	(a)	40.4	36.8
	(b)	40.8	36.8

Table IX. Effects of Container Shape on the Color of Orangeade

Shape	x	y	Y
△ ↑ [a]	0.375	0.384	68.7
△ ↑	0.437	0.446	54.6
▢ ↑	0.461	0.462	45.9
○ 4 cm ID	0.448	0.457	48.2
○ 5 cm ID	0.420	0.420	34.6

[a] Arrow indicates direction of exit port.

latter two additions; there is only a small change in color after 24 hours of exposure to air. Column 3 of Table VIII shows that the addition of this β-carotene preparation, apart from a color improvement, had also the rather unexpected by-result of further stabilizing the orangeade.

A noteworthy effect is that of container shape. Table IX gives color coefficients of one and the same orangeade in prismatic or cylindrical cells of constant cross-sectional area but of different cross-sectional shapes (except for the 5-cm i.d. cylindrical cell).

Table IX shows that color of orangeades is very much affected by the container shape. The effect on λ_d is slight but it seems that saturation can be traded for brightness and $vice\ versa$ by varying the container shape. The effect is parallel to that of varying the concentration of fruit juice in the orangeade (Table II).

Finally, it may be pointed out that the new technique restores much of the spectral detail of β-carotene that is lost in a standard transmission spectrum of orangeade (Figure 1). The amount of detail seems sufficient to use "translucence" spectra for the identification of pigments. The similarity between the spectrum of a β-carotene crystal suspension (Shibata, 1956) and our "translucence" spectrum of orangeade is striking indeed. Little is known, however, on the processes by which these spectra differ from that of a homogeneous β-carotene solution in the first place. Lothian and Lewis (1956) in their study on the spectra of blood corpuscles developed a useful theoretical approach, but much more additional work is required in this field.

INSTRUMENTAL

A Zeiss PMQII spectrophotometer with the MM12 double monochromator equipped with glass prisms was used for all the measurements with the illuminating sphere and the goniophotometer. Also the mirror and lens optics of the RA-3

reflection unit were employed throughout. Reference 100% point measurements with the gonio-photometer required an approximate attenuation of 300×; this was obtained by changing the gain by a factor of 10× and the photomultiplier sensitivity by an approximate factor of 30× between the reference and the sample measurement.

Absorbance and attenuance measurements were made on a Cary 14 spectrophotometer (with the cells in the center of the cell compartment). Attempts to measure the diffuse reflection of orangeade were made first with a J-20 Hilger and Watts Tristimulus Colorimeter (perpendicular illumination, measuring diffuse reflection at 5° with the normal), and then with the Zeiss RA-2 reflection set (perpendicular observation, illuminating under an angle of 45°).

All color coefficients were measured using the method of 30 selected wavelengths as described by Hardy (1936). All colors are based on illuminant "C."

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LITERATURE CITED

Bouma, P. J., "Physical Aspects of Colour," published by the Philips Industries, Eindhoven (the Netherlands), 1947.
Calvarano, J., Essenze Deriv. Agrumari 33(4), 260–71 (1965).
Davies, B. H., Chap. 18 in "Chemistry and Biochemistry of Plant Pigments," T. W. Goodwin (Ed.), Academic Press (1965).
Debye, P., Ann. Physik 46, 809 (1915).
Edwards, G. J., Wenzel, F. W., Huggart, R. L., Barron, R. W., Proc. Florida State Hort. Soc. 79, 321–5 (1966).
Hardy, A. C., "Handbook of Colorimetry," (M.I.T. Cambridge, Mass.) 1936.
Huslin, F. E., Proc. Roy. Australian Chem. Inst. 29, 196–204 (1962).
Kock, J., Sajak, E., Z. Lebensm. Unters. Forsch. 126(4), 260–71 (1965).
Kubelka, P., Munk, F., Z. Tech. Phys., 11a, 593–603 (1931).
Kubelka, P., J. Optical Soc. Amer. 38, 448–57 (1948).
Lothian, G. F., Lewis, P. C., Nature 178, 1342 (1956).
Mie, G., Ann. Physik 25, 377 (1908).
Rayleigh, Lord, Phil. Mag. 12, 81 (1881).
Shibata, K., Benson, A. A., Calvin, M., Biochem. Biophys. Acta 15, 461–70 (1954).
Shibata, K., Biochem. Biophys. Acta 22, 398 (1956).
Shibata, K., J. Biochem. 45, 599–623 (1958).
Strauss, W., "Chromoplasts—Development of Crystalline Forms, Structures and State of the Pigment," Botanical Review 19, 147–86 (1953).