Reference intervals and discrimination values of the Lanthony desaturated D-15 panel test in young to middle-aged Japanese army officials: the Okubo Color Study Report 1

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Abstract

Purpose To better understand the reference values and adequate discrimination values of colour vision function with described quantitative systems for the Lanthony desaturated D-15 panel (D-15DS).

Methods A total of 1042 Japanese male officials were interviewed and underwent testing using Ishihara pseudoisochromatic plates, standard pseudoisochromatic plates part 2, and the D-15DS. The Farnsworth–Munsell (F–M) 100-hue test and the criteria of Verriest *et al* were used as definitive tests. Outcomes of the D-15DS were calculated using Bowman's Colour Confusion Index (CCI). The study design included two criteria. In criterion A, subjects with current or past ocular disease and a best-corrected visual acuity less than 0.7 on a decimal visual acuity chart were excluded. In criterion B, among subjects who satisfied criterion A, those who had a congenital colour sense anomaly were excluded.

Results Overall, the 90th percentile (95th percentile) CCI values for criteria A and B in the worse eye were 1.70 (1.95) and 1.59 (1.73), respectively. In subjects satisfying criterion B, the area under the receiver operating characteristic curve was 0.951 (95% confidence interval, 0.931–0.971). The CCI discrimination values of 1.52 or 1.63 showed 90.3% sensitivity and 90% specificity, or 71.5% sensitivity and 95% specificity, respectively, for discriminating acquired colour vision impairment (ACVI).

Conclusion We provided the 90th and 95th percentiles in a young to middle-aged healthy population. The CCI is in good agreement with the diagnosis of ACVI. Our results could be helpful for using D-15DS for screening purposes.

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Introduction

Although colour discrimination function declines with increasing age^{1–3} and some occupations such as artists, graphic designers, naval navigation officers, and soldiers require good colour discrimination to perform many job-related activities, colour vision testing is generally not part of a routine screening. Little research has been performed to determine the prevalence and risk factors for acquired colour vision impairment (ACVI) in individuals of working age. The desaturated Lanthony D-15 panel (D-15DS) is most commonly used in an occupational setting⁴ and provides fast, accurate testing of subjects exposed to solvents⁵ and many chemical toxins (ie, solvent mixtures, styrene, perchloroethylene, toluene, tetrachloroethylene, carbon disulphide, and alcohol) that are lethal to the visual system.^{6–8}

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However, the test yields only semiquantitative information if analyzed by the method recommended by the manufacturer. The Farnsworth–Munsell (F–M) 100-hue test⁹ is time-consuming; however, it assesses acquired colour discrimination abnormalities quantitatively and is suitable for clinical and scientific research.^{1,10}

Bowman¹¹ reported a method to sum up the colour differences between adjacent caps for the D-15DS calculated in the CIE L*a*b system. Bowman's Colour Confusion Index (CCI)¹¹ quantifies the total colour discrimination score as a ratio of the actual score to the ideal score.

Although some previous reports have proven that this quantitative scoring system using the D-15DS was useful for screening⁸ and correlated with the score of the 100-hue test,^{12,13} the reference intervals were investigated with only a small sample size^{3,14,15} and adequate discrimination values are still unclear. In general, the 90th or 95th percentile value in a normal population is important for colour vision testing. The gold standard for the past decades for diagnosing ACVI has been the age-specific 95th percentile score of Verriest *et al*^{1,10,13,16} on the F–M 100-hue test. When the reference intervals and discrimination values of D-15DS are clearly defined, they may become more useful to screen for ACVI. The purpose of this study was to survey the distribution and adequate discrimination values of the CCI of D-15DS in a population of working-age Japanese officials.

Materials and methods

Study population

This study is a sub-study of The Okubo Color Study. The Okubo Color Study evaluates a relationship between colour vision function, especially ACVI, and ophthalmic disease, cardiovascular risk factors, and lifestyle factors. Subjects underwent annual health examinations in one community between April 2005 and March 2006 at the Okubo garrison. The authors collected physical information on current and previous diseases, use of medications, and lifestyle characteristics using structured questionnaires and personal physical records. All subjects underwent detailed interviews and measurement of visual acuity (VA), slit lamp biomicroscopy, and intraocular pressure with non-contact applanation tonometry, and also underwent a fundus examination. The VA was measured using a Landolt ring chart (with refractive correction, if any) at a distance of 5 m. Colour fundus photographs were obtained through undilated pupils using the fundus camera system (Canon, CR-45NM, Tokyo, Japan) with an angle of 30°. To assess colour sense, Ishihara plates

(Kanehara Shuppan Co. Ltd, Tokyo, Japan), standard pseudoisochromatic plates part 2 (SPP2; Igaku-Shoin, Tokyo, Japan), and D-15DS plates (Luneau, Paris, France) were used. According to previous reports,^{1,15} colour vision tests were carried out in the morning and under daylight illumination at a luminance level of 250 lux. To maintain a luminance level, light screen or artificial lighting, entitled Standard Illuminant C, was used if necessary. All subjects underwent the same sequence of colour testing as follows: Ishihara plates, SPP2, and D-15DS. Venous blood was drawn to measure fasting plasma glucose, total cholesterol, triglycerides, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, uric acid, and haemoglobin A1c after an overnight fast. All examiners and photograph readers were masked to other subjects' information including vision, referring diagnosis, systemic status, and lifestyle characteristics.

All studies were reviewed by the National Defense Medical College of Research Ethics to ensure that they met the ethical guidelines, and all subjects provided written informed consent before participating according to the Helsinki Declaration 2000.

Colour testing

Ishihara pseudoisochromatic plates (Ishihara plates)

Binocular inspection of Ishihara plates 1–21 (38-plate edition) was used to identify red–green colour sense abnormality. Subjects with more than four errors were classified as having failed.¹⁵

Lanthony D-15DS

The D-15DS was used to supplement standard D-15 testing to diagnose milder congenital colour anomaly or ACVI.^{5,8} Subjects with more than one major crossing were classified as subjects with failed results. The D-15DS was inspected monocularly, with the right eye tested before the left eye.

SPP2

SPP2 was especially designed to detect ACVI, and the test also can be used to identify blue–yellow colour sense abnormalities. Subjects with more than one blue–yellow error were classified as having failed. Missing the 2 on plate 3 was not considered a failure.^{16,17} SPP2 was inspected monocularly, with the right eye tested before the left eye.

F–M 100-hue test

If a subject was classified as an ACVI suspect, the F–M 100-hue test was performed according to the standard technique.¹⁶ Colour discrimination was tested monocularly at a distance of 30 cm with the subjective

near refraction placed in a trial frame. The subjects had abnormal hue discrimination; scores worse than the 95th percentile described by Verriest *et al* were diagnosed with ACVI.¹ The F–M 100-hue test was tested monocularly, with the right eye tested first.

Study design

The study design is summarized in Figure 1. There were two criteria for eligibility. For criterion A, all eligible patients had to have at least a 0.7 best corrected VA (BCVA) on the decimal VA chart to see the colour plates. Subjects who had undergone cataract or retinal surgery and had a corneal or retinal disease also were excluded. To exclude side effects of drugs on colour sense, subjects who were currently taking oral digitalis glycoside, antiepileptic drugs, antimalaria drugs, or antituberculosis drugs^{16–19} were excluded.

For criterion B, among patients who satisfied criteria A, those who failed testing with the Ishihara plates or had a typical pattern of congenital deutan or protan defect in SPP2 or D-15DS, or a past history of congenital colour discrimination anomaly by the Ishihara plates and

D-15DS at their enlistment examination were also excluded. Questionable cases underwent anomaloscopy. Subjects who satisfied criterion B and failed the SPP2 or D-15DS in either eye were classified as ACVI suspects and were tested using the F–M 100-hue test bilaterally. Subjects exceeding the age-specific limit score of Verriest *et al*¹ were diagnosed with ACVI.

Outcome criteria and statistical analysis

Data were analyzed with SPSS software, version 5.0 (SAS, Cary, NC, USA), and $P < 0.05$ was considered significant. Individuals and not eyes were the unit of analysis because the right and left eyes were not examined independently. A subject was considered to have an ACVI when it occurred in either eye.

The area under the receiver operation characteristic (ROC) curve (AUC) analysis was used to assess the diagnostic values of variables for the determination of ACVI. A perfect test would have an AUC of 1, whereas a test without diagnostic value would have an AUC of 0.5. The sensitivity and specificity for detecting ACVI were determined by the F–M 100-hue test and the age-specific

Figure 1 Schematic diagram of The Okubo Color Study. Diagnostic schema of ACVI.

criteria of Verriest *et al.*¹ This analysis was applied to eligible subjects who satisfied criterion B; subjects who declined to undergo the F–M 100-hue test were excluded.

According to the guidelines,²⁰ the serum low-density lipoprotein cholesterol level was categorized as less than 130 mg/100 ml, equal to or exceeding 130 mg/100 ml but less than 160 mg/100 ml, or equal to or exceeding 160 mg/100 ml. Systemic hypertension was defined as a systolic/diastolic pressure of 140/90 mmHg or more at the time of examination, or a history of diagnosis of hypertension and current use of medication for hypertension.²⁰ Diabetes was defined as a previous history of diabetes treated with either insulin, oral hypoglycaemia agents, or diet. Newly diagnosed diabetes was defined as no previous medical history of diabetes in the presence of elevated glycated haemoglobin or a fasting blood glucose level of 126 mg/100 ml or more at the time of examination.²¹ Hyperuricemia was defined as a serum uric acid value of 8.0 mg/100 ml or more at the time of examination, or a history of gout and current use of medication for hyperuricemia.

Smoking status

Subjects were categorized as those who have never smoked, current, and prior smokers. Current and prior smokers were defined as current or past habitual smokers who had smoked for at least 1 year. Subjects who had been smoking for less than 1 year were not considered to be smokers. Cigarette smokers were classified into light and heavy smokers based on the cigarette pack-years smoked, which indicates the personal amount of smoking over a long period. Cigarette pack-years were calculated by multiplying the number of packs of cigarettes smoked per day by the number of years the person had smoked. For example, 1 pack-year is equal to smoking one pack per day for 1 year. In this analysis, subjects were considered to be light smokers if they had fewer than 19 pack-years of smoking (median pack-years of smoking) and heavy smokers if they had 19 or more pack-years of smoking. We checked the medical records and the subjects' response to questionnaires to confirm past and current diseases.

Results

The demographic data are shown in Table 1. A total of 959 and 884 subjects, respectively, were eligible for criteria A and B. Table 2 shows the number of subjects and the mean/median CCIs in each age group. The median CCIs in each age group were similar between criteria A and B. Figure 2 shows the distribution of CCI in

Table 1 Baseline characteristics of eligible subjects

	Criterion A	Criterion B
Number of subjects	959	884
Age (years)	38.0 ± 8.7	37.8 ± 8.7
Height (cm)	170.2 ± 5.9	170.2 ± 5.9
Weight (kg)	68.5 ± 9.1	68.6 ± 9.0
Body mass index (kg/m ²)	23.7 ± 2.7	23.6 ± 2.7
	Number of subjects (%)	Number of subjects (%)
Hypertension	121 (12.6)	110 (12.4)
LDL cholesterol, mg/100 ml		
< 130	614 (64.0)	561 (63.5)
130–159	231 (24.1)	215 (24.3)
≥ 160	114 (11.9)	108 (12.2)
Hyperuricemia	195 (20.3)	187 (21.2)
Diabetes	20 (2.1)	17 (1.9)
Smokers		
Never smoked	374 (39.0)	340 (38.5)
Light smoker	293 (30.6)	274 (31.0)
Heavy smoker	292 (30.4)	270 (30.5)

± values are means ± SD.

Table 2 Numbers of subjects and, median and mean CCI for each criterion by age group

Age (years)	Criterion A			Criterion B		
	No.	Median	Mean (95% CI)	No.	Median	Mean (95% CI)
20–29	228	1.22	1.28 (1.25–1.31)	209	1.21	1.26 (1.23–1.29)
30–39	225	1.19	1.27 (1.23–1.31)	210	1.18	1.23 (1.20–1.26)
40–49	451	1.28	1.38 (1.34–1.42)	416	1.24	1.29 (1.27–1.31)
50–59	55	1.28	1.38 (1.26–1.50)	49	1.20	1.28 (1.20–1.36)
Total	959	1.22	1.33 (1.31–1.35)	884	1.22	1.27 (1.25–1.30)

the worse eye, with worse defined as the personal worse score between the right and left eyes of one subject. The CCI in the worse eye did not have a normal distribution even after transformation ($P < 0.001$ Kolmogorov–Smirnov test).

Figure 3 shows the age-specific 90th and 95th percentile CCIs that met criteria A and B, respectively. In all subjects, the 90th percentile (95th percentile) scores in the worse eye were 1.70 (1.95) in criteria A and 1.59 (1.73) in criteria B, respectively. Figure 4 shows the ROC curve of the worse CCI values. AUC was 0.951 (95% confidence interval (CI), 0.931–0.971). For this algorithm, specificities of 80, 85, 90, and 95% were reached for sensitivities of 96.8, 93.6, 90.3, and 71.0%, respectively (Table 3).

Discussion

This study determined the age-specific normal range of CCI in working-age Japanese men and indicated that the CCI is in good agreement with the diagnosis of ACVI using the F-M 100-hue test.

Surveillance of reference intervals and discrimination values is important because previous reports have described that ACVI can precede deterioration of VA or be correlated with an ocular disease such as cataract,²² diabetic retinopathy,¹⁰ glaucoma, age-related macular degeneration, and lifestyle factors such as acute and

Figure 2 Distribution of CCI in the worse eye (eligible for criterion B).

Figure 4 ROC curve analysis. The AUC is 0.951 (95% CI, 0.931-0.971).

Figure 3 The 90th and 95th percentile limits of CCI by age.

Table 3 Sensitivity, decision threshold value of CCI, and positive predictive rate at a fixed specificity

Decision threshold value of CCI	Sensitivity (%)	Positive predictive rate (%)	Specificity (%)
1.40	96.8	15.2	80
1.44	93.6	18.8	85
1.52	90.3	25.7	90
1.63	71.0	30.0	95

chronic ethanol intake, and tobacco smoking.^{4,6,8,23,24} Moreover, extended exposure to numerous chemical agents and medicine also has been associated with alterations in colour discrimination.^{6–8,19} Although those case-control studies were well reported, multifactorial analysis of ACVI has been studied less. The Okubo Color Study will investigate these relationships in later reports.

Colour vision testing generally provides a wide range of reference intervals in clinical work.¹⁶ In previous reports by Ihrig *et al*,²⁵ the median CCI value was 1.00–1.17 in 238 normal German subjects (mean age, 27 years). Good *et al*¹⁴ reported that 54 normal men without ocular or systemic disease scored 1.10 (average) on the primary D-15DS test in 1400 lux illumination. Iregren *et al*¹⁵ reported a median CCI score of 1.13 in 199 healthy volunteers or 1.16 in 1000 lux illumination. Urban *et al*⁷ reported that 24 healthy subjects (mean age, 42 years) scored 1.04. These values were better than the values obtained in this study. The reason for these discrepancies is unknown, but several factors can be considered. First, Woo²⁶ reported that differences in macular pigmentation between Caucasians and Asians cause substantial differences in the results of the F–M 100-hue test. Our entire study population was Japanese, which might have contributed to the difference. Second, we analyzed the worse eye score, and individuals rather than eyes were the unit of analysis because the right and left eyes were not independent. Our methodology might have contributed to the discrepancy. Third, standardized conditions, especially the illumination level, might be important when administering the D-15DS. The differences in light luminance may partly explain the differences.

To the best of our knowledge, few studies have been published on reference intervals and discrimination CCI scores. Regarding the 90th percentile score, Iregren *et al*¹⁵ tested 199 subjects, after excluding congenital colour vision deficits, and reported that the overall 90th percentile CCI scores were 1.54 in right eyes and 1.56 in left eyes. In this study, the overall 90th percentile score in the subjects who met criteria B was 1.50 in the right eyes and 1.47 in the left eyes (Figure 3), which are close to the

results reported by Iregren *et al*.¹⁵ Regarding the 95th percentile score, Ihrig *et al*²⁵ tested patients binocularly and reported that the CCI values in a healthy group ranged from 1.21 to 1.61 calculated by the mean and SD. Unfortunately, unlike the F–M 100-hue test scores, the 90th and the 95th percentile CCI scores cannot be calculated using the mean and SD, because the CCI using D-15DS does not have a Gaussian distribution even after transformation³ (Figure 2). Therefore, a large sample size is required to assume the accuracy of the 90th or 95th percentile score. Significantly better results with the patients who satisfied criterion B compared with those who satisfied criterion A implied that a small number of subjects with impaired congenital colour discrimination strongly affect the limit value rather than the median value (Table 2).

Bassi *et al*¹³ reported that in 35 patients with glaucoma included in an ACVI model, there is a significant correlation between the total error scores in the F–M 100-hue test and the C-index, which is another quantitative system, in the D-15DS. However, the investigators excluded participants with normal colour function and did not describe the discrimination value. In this study, when the discrimination value of the CCI was set to 1.52, there was good agreement with the diagnosis of ACVI using the longer F–M 100-hue test in an epidemiologic study (sensitivity, 90.2%; specificity, 90%). This score may be the best discrimination value.

In the current results, the reference value of CCI in the right eye was significantly higher than that in the left eye in only the youngest age group (Figure 3). The reason is unclear, but our methodology, in which the right eye was tested before the left eye, might have affected the results. In general, colour vision testing is affected by the learning curve or fatigue but it has been studied less. We plan to investigate these effects in later studies. Nevertheless, the differences affected our results less, because individuals were the unit of analysis not eyes.

A limitation of this study was that the subjects were all male Japanese officials on active duty. Research among women, individuals in other occupations, or individuals of other races may be necessary.

In addition, the illumination level might be important when administering the D-15DS. Iregren *et al*⁶ and Gobba⁸ reported that they applied the D-15DS in the morning using daylight fluorescent lamp providing 1200 lux. Baird *et al*²⁷ reported that the D-15DS required a 500-lux Macbeth lamp light. Melamud reported that 100 lux should be the minimal level of illumination for screening.¹⁶ However, we do not know whether or how the level of illumination used during testing might have affected the test results.²⁷ In our results, the median CCI was higher than the results reported by Iregren, but the 90th percentile CCI was similar.¹⁵ Thus, the difference in

the illumination level may be less important when discriminating between ACVI and healthy subjects.

Another limitation was related to positive prevalence rate, which is the chance of having a disease if one screens positive, and it is prevalence dependent. Even with high sensitivity and specificity values, the positive predictive rate is low if the prevalence is low. Because of the low prevalence of ACVI in this study, the CCI with the D-15DS also shows a low positive predictive rate (Table 3). Thus, the F-M 100-hue test cannot be replaced by the D-15DS completely and will remain the definitive test.

In conclusion, our study provided the normal healthy distribution in a large number of working-aged men on active duty using the D-15DS test with the CCI scoring system. Our results could be helpful for clinicians and patients when the D-15DS test is performed for screening purposes.

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