



Editorial

Guidelines for preparing color figures for everyone including the colorblind



Color coding of objects is a useful and important way to convey information in the biological and physical sciences. Examples include the use of colors to denote variable gene expression or double staining in confocal micrographs to determine co-localization of various cellular components. Because about one in 12 males (8%) and one in 200 females (0.5%) are red-green colorblind [1], care must be taken in preparing colored figures to accurately convey the desired information. For effective communication, we must consider our readers, referees, grant reviewers, and the audience for a PowerPoint presentation during the formulation of color figures. In an audience of 100 men and women, about four people with be color blind. If your paper is sent to three male reviewers, the odds are about one in four that one reviewer will be color blind. If your study section is made up of six women and six men, the odds are one in two that one reviewer will be color blind.

So-called color blindness does not result in the loss of perception of all colors, or monochromacy; instead, such visual deficiencies make it difficult to distinguish between various colors. The primary colors include red, green, and blue while the secondary colors include purple, green, and orange (Fig. 1). Colors are sensed by three types of retinal cone cells which correspond to the primary colors (red, green, and blue). Defective red, or long wavelength, cone cells result in a color blindness called protanopia (from Greek *prot* for the “first” type of cone). Defective green, or medium wavelength, cone cells result in deuteranopia (from Greek *deuter* for the “second” type of cone). Both of these defects are transmitted by X-chromosome-linked inheritance and result in red-green

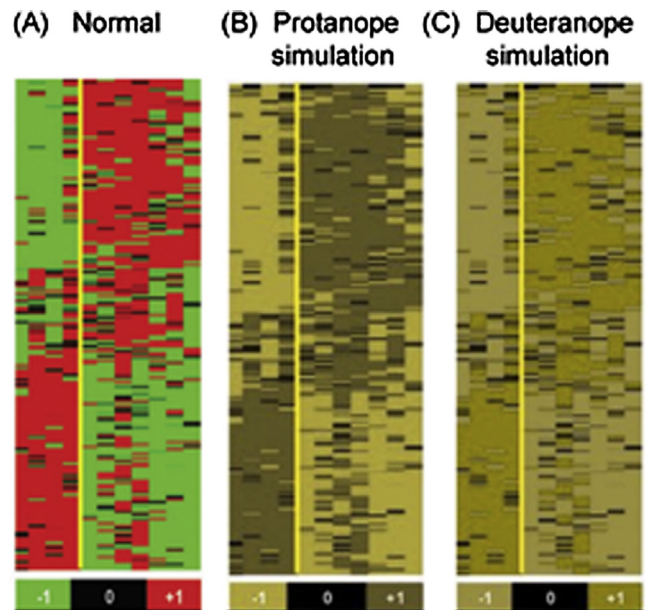


Fig. 2. A gene expression profile indicated by red, black, and green matrices (from Ref. [2]; part A is reprinted with permission of the American Association for Cancer Research).

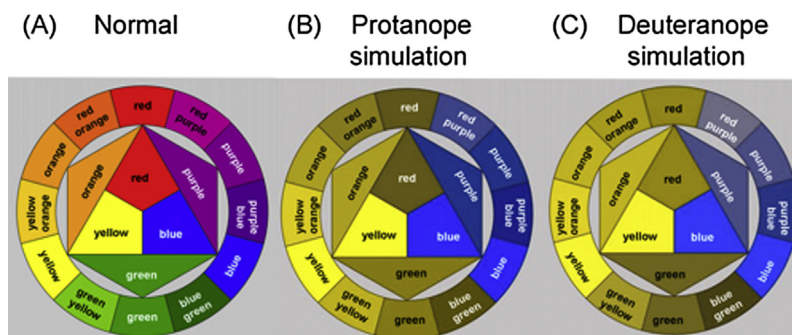


Fig. 1. A color wheel as seen by (A) a normal individual, (B) a protanope, or (C) a deuteranope.

	N	Pro	Deu
Red	-NH ₂	-NH ₂	-NH ₂
Green	-NH ₂	-NH ₂	-NH ₂
Light blue	-NH ₂	-NH ₂	-NH ₂
Dark blue	-NH ₂	-NH ₂	-NH ₂
Yellow	-NH ₂	-NH ₂	-NH ₂
Yellow	-NH ₂	-NH ₂	-NH ₂
Black	-NH ₂	-NH ₂	-NH ₂
Grey	-NH ₂	-NH ₂	-NH ₂

Fig. 3. Colored text and lines (the dashes) as seen by a person with normal color vision (N), a protanope (Pro), or a deuteranope (Deu).

colorblindness. About 2% of males are protanopes and 6% are deuteranopes. Defective blue cone cells are not associated with X-linked inheritance, but result in tritanopia (from Greek *trit* for the “third” type of cone); however, this condition is rare. Note in Fig. 1 that blue and yellow are perceived nearly identically by normal (euchromatopsic) and red-green colorblind individuals.

For both protanopic and deuteranopic people, distinguishing between red and green is more difficult than distinguishing between yellow and green (Fig. 1). Thus, having red and green lines cross each other or placing red and green objects close to each other represent depictions that should be avoided; note that for the protanope red is perceived as a dark color and for the deuteranope, red and green are nearly identical (Fig. 2). Although yellow has good color character for both the protanope and deuteranope, it may be too close to white for the normal eye to distinguish unless it has a non-white border or background (Fig. 3). However, distinguishing light yellow from green may be difficult for both protanopes and deuteranopes. Black, white, and gray are easily differentiated by nearly everyone. Of all of the hues, blue is perceived uniformly as a color by all individuals and should probably be used first when formulating colored objects. Red is perceived nearly as black by protanopes (Fig. 1); accordingly, red print over a black background may be invisible. Also the use of dark red text for emphasis when incorporated within a string of black text will not be perceived as different for protanopes. Moreover, dark blue text may be perceived as black by everyone, so it may be better to use sky blue or Carolina blue for highlighting.

When you have made your colored figures and want to check them for how they are perceived by the colorblind, go to the free-ware at <http://vischeck.com/>. You can upload your figures and see how they appear to a colorblind person, or you can download the program and make analyses and revisions as necessary. This was the procedure that was used for generating Figs. 1–3. For additional advice in preparing color figures, visit http://jfly.iam.u-tokyo.ac.jp/html/color_blind.

To summarize, blue is the best color to use followed by yellow depending upon the background. With care, the use of two different shades of blue may be distinguishable. Instead of using red, it is usually better to use an off-red such as magenta, vermilion, or orange. Instead of using green, it is usually better to use an off-green such as blue green. It helps to examine such figures generated on a color printer to ensure that different shades can be differentiated. If a figure is understandable when an object or line is red or black, one can safely use red for the many people for whom red is a favorite color. Realize, however, that the protanope will perceive it as black. White, gray, and black are non-colors, but they are readily distinguishable and useful. If a figure is worth a thousand words, a suitably made color figure may be worth more owing to the additional information that color coding conveys. However, the coloring should be made so that it can be perceived and differentiated by everyone including the readers of *Pharmacological Research*.

Conflict of interest

The author is not colorblind and there are no known conflicts of interest.

References

- [1] W.H. Swanson, J.M. Cohen, Color vision, *Ophthalmol. Clin. North Am.* 16 (2003) 179–203.
- [2] M.L. Ascierto, T.L. McMiller, A.E. Berger, L. Danilova, R.A. Anders, G.J. Netto, et al., The intratumoral balance between metabolic and immunologic gene expression is associated with anti-PD-1 response in patients with renal cell carcinoma, *Cancer Immunol. Res.* 4 (2016) 726–733.

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