# Action Spectrum for Vitamin D Synthesis

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## Introduction

Exposing the skin to UV radiation initiates vitamin D synthesis: 7-dehydrocholesterol (located in the membranes of skin cells) absorbs UVB photons and converts into previtamin D, which then thermally isomerises into vitamin D over a period of 12 to 24 hours.

UV levels decrease with increasing latitude and the wavelength composition also varies by latitude, across the day, and across the year. Given the importance of vitamin D for our health, it is essential to understand how vitamin D-effective UV varies by region and by season.

To measure the level of vitamin D-effective UV, an action spectrum is used. An action spectrum for producing previtamin D (the initial photochemical reaction) is already in existence. This was obtained by MacLaughlin et al. (1982). The CIE recently adopted this as the standard action spectrum for previtamin D synthesis in human skin (CIE, 2006). The CIE action spectrum presents the effectiveness (in relative units) of the UV wavelengths at synthesising vitamin D (Figure 1). Vitamin D-effective UV irradiance ($W/m^2$) is determined by weighting the ambient UV spectrum with this action spectrum, and then integrating across the wavelength range.

The CIE action spectrum has some limitations. In particular, in the original 1982 experiments, the monochromatic UV source used to obtain the action spectrum had a relatively large bandwidth, ranging from 6 to 10 nm. This may have decreased the precision of the measurements that were conducted (at 1 nm intervals) to assess the previtamin D-producing potential of each wavelength.

Here we use an *in vitro* model to test the effectiveness of various UVB wavelengths at producing vitamin D, and then compare the findings with the CIE previtamin D action spectrum.

## Method

We used an *in vitro* model consisting of 7-DHC dissolved in high-grade ethanol (Olds et al., 2008). Individual samples were made by placing a small volume of this solution into a UV-transmissive quartz cuvette.

Samples were then exposed to monochromatic radiation from 290 to 320 nm, in 5 nm intervals. Three samples were exposed at each wavelength (total of 21 samples).

UV exposures were conducted with an irradiation monochromator (IM), which was calibrated to the NIST standard of irradiance. The bandwidth of the IM (1.7 nm) is considerably narrower than that used in the original 1982 study. A constant dose of 20 $J/m^2$ of unweighted UV was given at each wavelength.

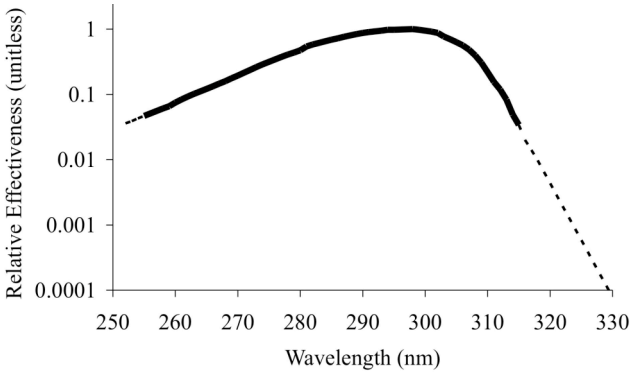


**Figure 1.** The CIE action spectrum for previtamin D synthesis. The solid region is the data that was obtained by MacLaughlin et al. in 1982. The dashed region is data extrapolated by the CIE (CIE, 2006).

In the *in vitro* model, vitamin D is measured as the endpoint (whereas previtamin D was the endpoint in the MacLaughlin et al. experiments). Hence, after exposing each sample, it was placed into a vial and incubated at 37ºC (physiologic temperature) for 24 hours. This allowed sufficient time for the majority of previtamin D in the sample to convert into vitamin D. Since the incubation time and temperature were standardised for every sample, these two endpoints (previtamin D and vitamin D) should be similar and allow us to compare our results with the CIE action spectrum.

After precisely 24 hours, the amount of vitamin D formed in every sample was analysed by high-performance liquid chromatography (HPLC), which is considered a very accurate measurement technique.

Finally, the mean vitamin D production was calculated for the three samples at each wavelength. These data were normalised to unity (1) at the peak (which occurred at 295 nm) and plotted as a function of wavelength.

## Results

Figure 2 shows the data we obtained, based on exposure of the *in vitro* samples to monochromatic UV radiation (1.7 nm bandwidth). This figure also shows the currently accepted CIE action spectra for comparison. It is apparent that the vitamin D production in the QUT data decays earlier (at a shorter wavelength) than in the CIE action spectrum. This cut-off is very important from a biological point of view, because it defines the range of wavelengths that will initiate vitamin D synthesis. Very little UVB radiation < 290 nm reaches ground-level on Earth. Therefore, a small shift in the cut-off would have a large impact on the measured level of vitamin D-effective UV.

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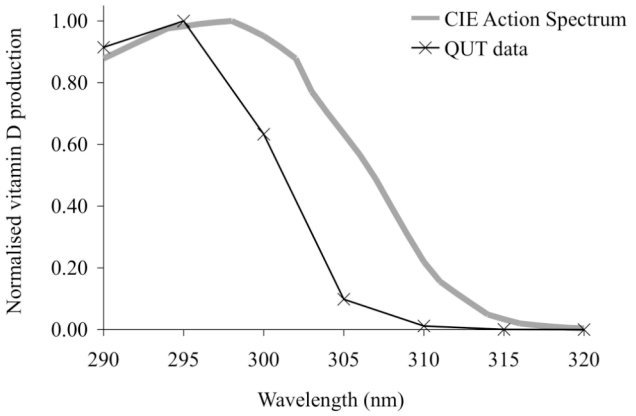


**Figure 2.** Normalised vitamin D production (QUT data), to allow comparison with the currently accepted CIE previtamin D action spectrum.

## Discussion

In this work, we used a narrow-bandwidth UV source to test the vitamin D-effectiveness of UVB wavelengths, for comparison with the currently accepted CIE action spectrum. The data we obtained, when plotted, showed vitamin D was produced by a narrower range of wavelengths than the CIE action spectrum suggests. Since the CIE action spectrum was obtained with a wider-bandwidth UV source, it seems that this may be the reason for its broader shape.

A limitation of the *in vitro* model is that it ignores the optical properties of skin. Skin is comprised of various layers that absorb and scatter UV. It also contains DNA, RNA, proteins and melanin that absorb UV before it can reach 7-DHC. The original MacLaughlin et al. action spectrum was obtained for skin; nonetheless our finding that the effective wavelength-range may be narrower should still be valid: there should be *more* vitamin D produced in ethanol, for a given dose of UV, than in skin because ethanol is more transparent to incoming UV. However, this was not the case – the CIE action spectrum is higher than the QUT data over the region from ~ 295 to 315 nm (Figure 2). Therefore, it seems that the narrower width of the QUT data, compared to the CIE action spectrum, can be attributed to the use of a narrower-bandwidth UV source in this work.

To highlight the impact of these results when measuring vitamin D-effective UV levels, a solar UV spectrum for Queenstown (NZ) was obtained using the TUV model[3] (Figure 3) (midday, wintertime, 300 DU ozone concentration, clear-sky day). We weighted it with the CIE action spectrum as well as the QUT data (to weight the solar spectrum, the QUT data were interpolated to 1 nm intervals using a simple linear interpolation between adjacent 5 nm datapoints, to create a simple action spectrum). As shown in Figure 3, the level of vitamin D-effective UV is approximately 5-fold higher when measured with the CIE action spectrum, compared with using the QUT data. This is a considerable difference, resulting from the narrower width of the QUT data compared with the CIE action spectrum.

[3] http://cprm.acd.ucar.edu/Models/TUV/Interactive_TUV/

## Conclusion

We believe that the data presented here suggest that the CIE action spectrum may need to be reviewed. The QUT data imply that a narrower range of wavelengths are responsible for synthesising vitamin D than the current CIE action spectrum would suggest (Figure 2). Therefore, using the CIE action spectrum would result in larger measurements of the vitamin D-effective irradiance. This may lead to overestimates in the available level of vitamin D-effective solar UV, in any given location. These matters are explored elsewhere in more detail (Olds et al., 2010).

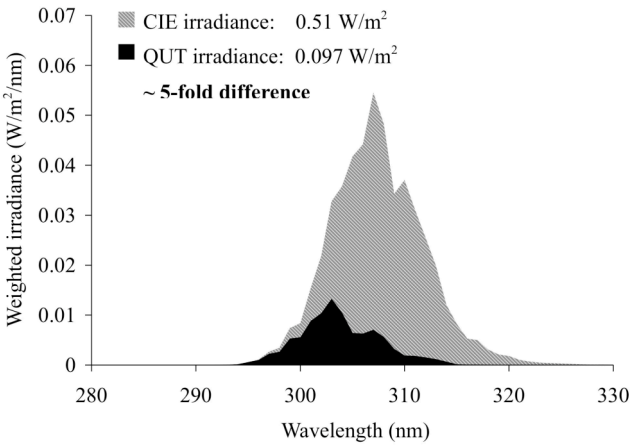


**Figure 3.** The solar spectrum for Queenstown after being weighted. Using the QUT data as a simple action spectrum, there would be approximately 5-fold less vitamin D effective UV than the CIE action spectrum would predict, in this location.

## References

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