Response to Reply by Ibrahim et al. in regarding photosystem II intermediate structures Jimin Wang^{1,*}, William H. Armstrong^{2,†}, Victor S. Batista³ ¹Department of Molecular Biophysics and Biochemistry, Yale University, 266 Whitney Avenue, New Haven, CT 06520-8114. ²Department of Chemistry, Boston College, 2609 Beacon Street, Chestnut Hill, MA 02467-3860. ³Department of Chemistry, Yale University, 225 Prospect Street, New Haven, CT06520-8499. *Corresponding author (<u>jimin.wang@yale.edu</u>). †Retired.

14 In our letter to the editor, we have demonstrated that the crystallographic data reported by Ibrahim et 15 al. do not contain sufficient information to support the hypothesis that a water molecule adds to the 16 Oxygen-Evolving Complex (OEC) of Photosystem II (PSII) during the 1F-to-2F transition (1, 2). We showed 17 that a single-conformation model fits their data very well whereas in their response Ibrahim et al. insist 18 that a two-conformation model is correct. However, they rely heavily on other experimental evidence to 19 justify their approach. Our critique, on the other hand, is based solely on a straightforward analysis of 20 their crystallographic data. We showed that there is no crystallographic evidence for the displaced 21 conformation of D1-E189 and that such a displacement is responsible for the positive peak that they 22 interpreted as Ox, which is proposed to be coordinated to both Mn4 and Ca (2). It is not surprising that a 23 model with additional parameters can fit the data but here such a model is clearly not warranted. In 24 fact, in an earlier paper (4), the same research group concluded that the 2F OEC structure does not 25 contain Ox, in seeming contradiction to the conclusion in their article under discussion here. Thus, we 26 stand by our position that there is no solid crystallographic evidence in support of the appearance of Ox 27 after two flashes of light.

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