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NEW DEVELOPMENTS REGARDING DNA MISMATCHES AND ITS RELATION TO SARS-COV-2 DETECTION

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DNA base-pair complementarity, cytosine binding to guanine (CG) and thymine to adenine (AT), is the essential mechanism of DNA replication. However, this complementarity rule is not written in stone. Mismatches, that is, anything that is neither CG nor AT, are a common occurrence and their properties are strongly influenced by the neighbouring base pairs. There is still much that we do not know about mismatches, and their thermodynamic properties are still poorly characterized. This lack of knowledge is a problem for the determination of DNA melting temperatures, which is a crucial parameter for the design of PCR primers. If mutations occur in PCR target regions, the primers will hybridize with mismatches and determining the changes in melting temperatures in those conditions can be problematic. Our group specialises in thermodynamic models that use melting temperatures as input parameters. We have recently used an extensive set of melting temperatures covering 4032 configurations of mismatches to calculate their thermal parameters, which now enables us to predict temperatures of sequences with mismatches, including those for PCR primers. In this presentation I will discuss how we used this temperature set, the challenges we faced, and the new understanding we gained regarding mismatches. Then I will discuss how we applied this to primers used in PCR and in loop-mediated isothermal amplification (LAMP) that are currently in use for SARS-CoV-2, particularly in regard to the latest omicron variants.

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