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To the NIGMS F32 study session:

I am writing in support of John Desmarais’ application for a Ruth L. Kirschstein National Research Service Award for an Individual Postdoctoral Fellowship (F32). Since 2018, I have collaborated with John on two projects and **SOMETHING HERE!!!!**. I recommend him for this fellowship with great enthusiasm.

During our collaborations John has displayed the skills and drive important to become a successful independent researcher. He has repeatedly demonstrated his ability at statistical methods, computational modeling, and science communication. Beyond this, John displays scientific integrity and collaborative spirit that will greatly aid his future endeavors.

As part of our response to the emergence of the SARS-COV2 pandemic, the Innovative Genomics Institute put together an interdisciplinary team of researchers to investigate methods of improving CRISPR based diagnostics to meet the challenges of the time. This team included researchers from across a wide variety of specialties and from multiple institutions including UC Berkeley, UCSF, the Gladstone institutes, and others. John joined the team to lend his expertise at modeling, statistics, and data analysis. As part of this effort, he collaborated closely with researchers across multiple projects and specialties analyze their experimental results, rapidly update our theoretical understanding of the system, and use that theory to guide new experiments to overcome limitations in our diagnostics. In the process demonstrating an ability to work quickly on fast moving projects and coordinate effort in large collaborations. All while maintaining his independent thesis research separately.

During this collaboration, John demonstrated his science communication skills repeatedly. In order to maintain coordination on a team of this size, we organized weekly meetings where all collaborators would present their findings from the week to the entire team. These meetings required communicating to a group of mixed experimentalists and theorists from across a variety of fields. John described complicated modeling and statistical results simply making his findings accessible to the entire team. This enabled rapid incorporation of model guided changes such using a chemically-modified uncleavable activator for csm6 after John’s modeling showed that stopping activator cleavage would dramatically improve detection characteristics.

John also showed his expertise in statistical methods and modeling. He developed data analysis and statistical protocols for identifying positive samples from one-off microfluidics time courses. In the process working out sampling strategies with experimental collaborators to maximize statistical power and minimize time to detection. He wrote software for parsing plate reader data and provided training on its use to others on the team. And he performed kinetic modeling of enzyme reactions that provided insight into issues such as low sensitivity, or high background in early diagnostic designs and helped to guide experiments to overcome these issues.

John is also a capable bench scientist, when my lab was characterizing CasX as a new RNA-guided CRISPR nuclease, we collected Cryo-EM data that revealed the presence of domain with potential zinc binding motif. In order to confirm that zinc was the bound metal, one of the graduate students in my lab at the time reached out to John who was able to quickly confirm the presence of zinc with an x-ray spectroscopy experiment.

In summary, John has been an outstanding collaborator

One example of this was when we were investigating the use of csm6 to amplify signal in a CRISPR diagnostic. John’s modeling suggested that blocking of activator degradation by csm6 could dramatically improve signal and

He also demonstrated considerable aptitude for modeling and statistics.

Two particularly relevant findings were that cas13 release of guide-target duplexes instead of sequential release of target and guide could produce early saturation behavior at low target levels and worsen limit of detection for diagnostics and that activator self-cleavage was

These results included using modeling to test mechanistic explanations of our experimental results

demonstration that cas13 dependent sequestration of guide and target through release of guide-target duplex instead of