

# Through-grid wicking enables high-speed cryoEM specimen preparation

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Blotting times for conventional cryoEM specimen preparation complicate time-resolved studies and lead to some specimens adopting preferred orientations or denaturing at the air-water interface. We show that solution sprayed onto one side of a holey cryoEM grid can be wicked through the grid by a glass fiber filter held against the opposite side, often called the 'back' of the grid, producing a film suitable for vitrification. This process can be completed in tens of milliseconds. We combined ultrasonic specimen application and through-grid wicking in a high-speed specimen preparation device that we name "Back-it-up", or BIU. The high liquid-absorption capacity of the glass fiber compared to self-wicking grids appears to make the method relatively insensitive to the amount of sample applied. Consequently, through-grid wicking produces large areas of ice suitable for cryoEM, while the device's speed reduces adoption of a preferred orientation in a test specimen. (Preprint available: <https://www.biorxiv.org/content/10.1101/2020.05.03.075366v1.full>)

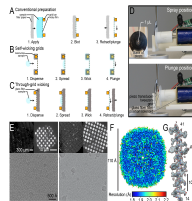


Figure 1

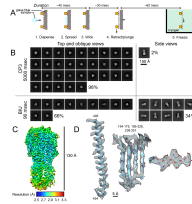


Figure 2