

**P020** Structural studies of stalled ribosomes undergoing translocation

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In our previously published studies, we showed that the IBV pseudoknot produces tension in the mRNA and A-P site hybrid tRNA, bending the tRNA, and we suggested a mechanism for -1 frameshifting. In this work we selected cryo-EM images showing the ribosome in either an EF2-tRNA-PK (pseudoknot) or apo state. Our current work points to a third population of ribosomes in a tRNA-PK bound state. We separated these states by supervised classification and analysed the structural differences between these two states. The EF2-tRNA-PK ribosome shows the tRNA with its anticodon loop bent towards the A-site and with its elbow compressed against the 60S subunit. The tRNA-PK ribosome again shows the tRNA with its anticodon loop bent towards the A-site, but more drastically so, illustrating that EF2 opposes PK action. In contrast to the EF2-tRNA-PK ribosome, the tRNA-PK ribosome's A-P site tRNA is more relaxed and its elbow is not compressed against the 60S subunit hinting that EF2-driven tRNA compression might be a consequence of normal translocation. These data suggest that the bending observed in our previous work is a function of two forces, the elbow compression resulting from EF2 action and anticodon bending resulting from PK action. Our latest data support our proposed -1 frameshifting mechanism, and suggest a novel model for translocation.