Differing effects of rapamycin and mTOR kinase inhibitors on protein synthesis
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The mammalian target of rapamycin, mTOR, forms two distinct complexes. mTOR complex 1 (mTORC1) regulates several proteins involved in mRNA translation, including the eukaryotic initiation factor 4E (eIF4E)-binding proteins (4E-BPs). Rapamycin inhibits some functions of mTORC1 but not all; inhibitors of its kinase activity such as PP242 inhibit additional functions of this complex. Short-term treatment (<3h) of HeLa cells with rapamycin caused a small (10-20%) inhibition of protein synthesis, while PP242 had a greater effect (30-40% inhibition). PP242 also blocked the phosphorylation of 4E-BP1, increased its binding to eIF4E and inhibited the formation of initiation factor complexes containing eIF4E and the scaffold protein eIF4G. In contrast, rapamycin did not exert these effects.

Given these contrasting effects of rapamycin and PP242 on the translation initiation machinery and overall protein synthesis, we studied their effects on the synthesis of specific proteins. To do this, we employed a newly-developed pulsed stable isotope-labelling method (pSILAC). This revealed differing effects of rapamycin vs. mTOR kinase inhibitors on a range of proteins. For example, the latter strongly inhibit synthesis of proteins encoded by 5’-tract of pyrimidine (5’-TOP) mRNAs much more strongly than rapamycin does. Synthesis of most other proteins was less prone to inhibition by mTOR kinase inhibitors or rapamycin. These data are important for understanding the effects of mTOR kinase inhibitors (potential anti-cancer agents) on gene expression and cell function.