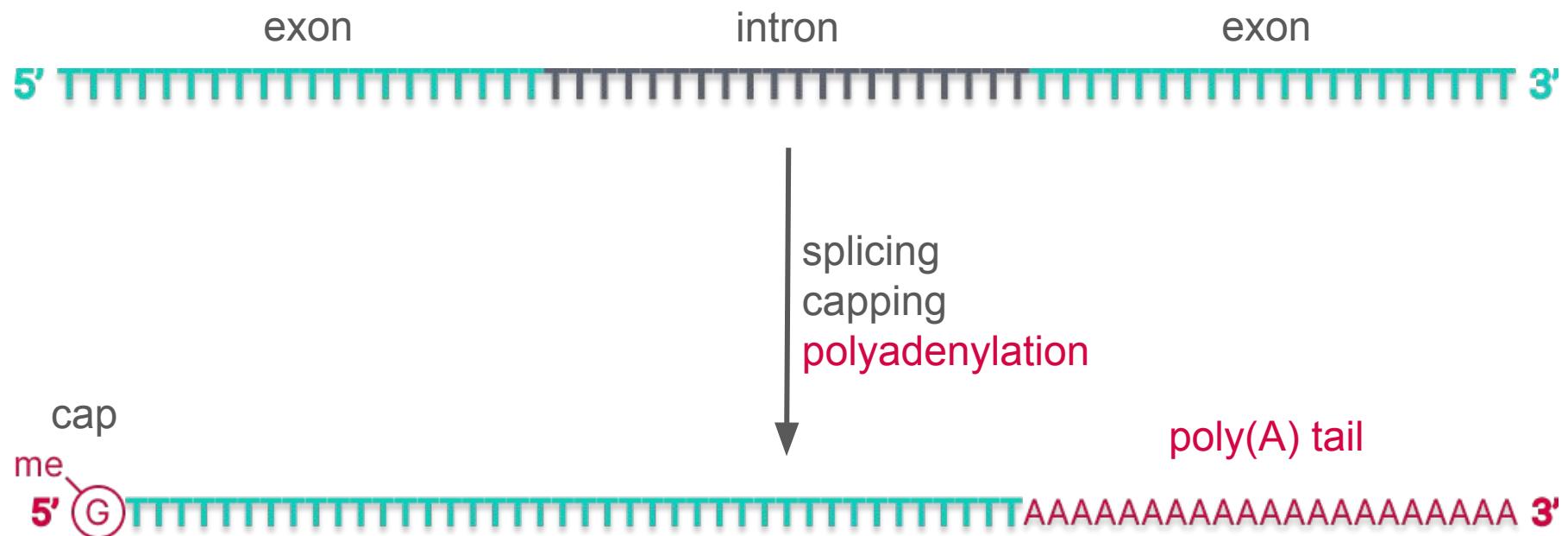


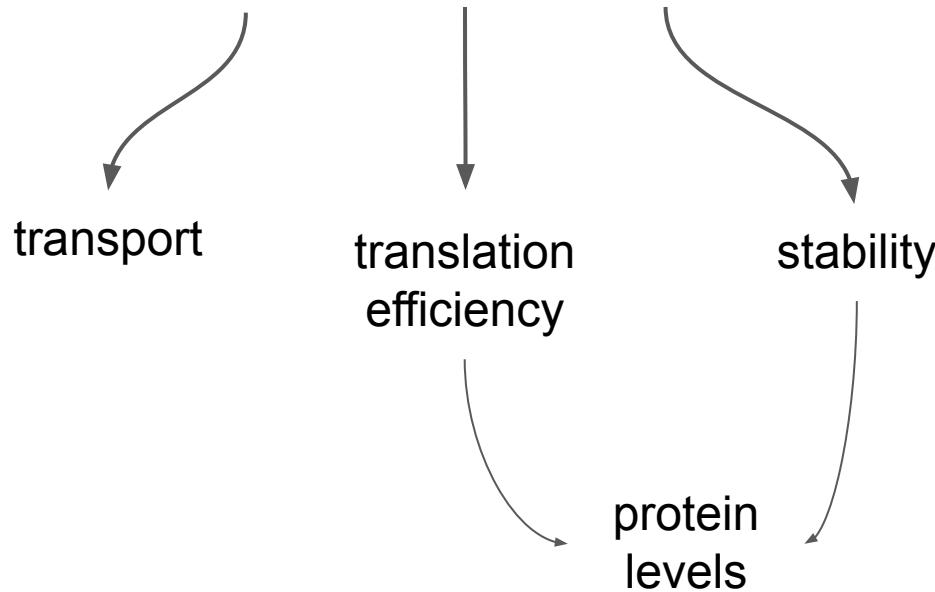
Analysis of poly(A) tails by sequencing

Maria Nizik

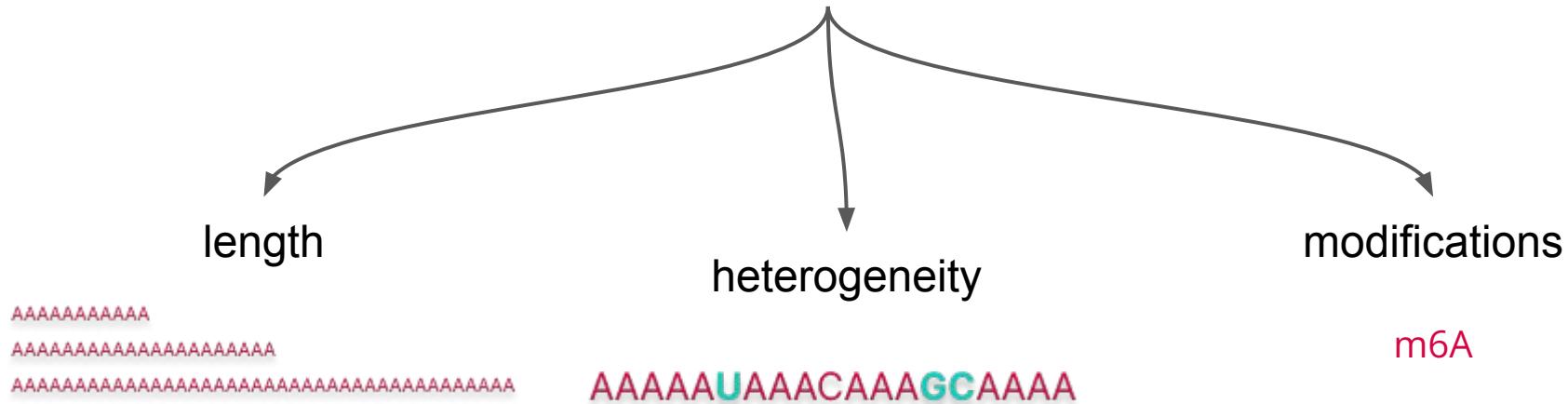
mRNA processing



polyadenylation

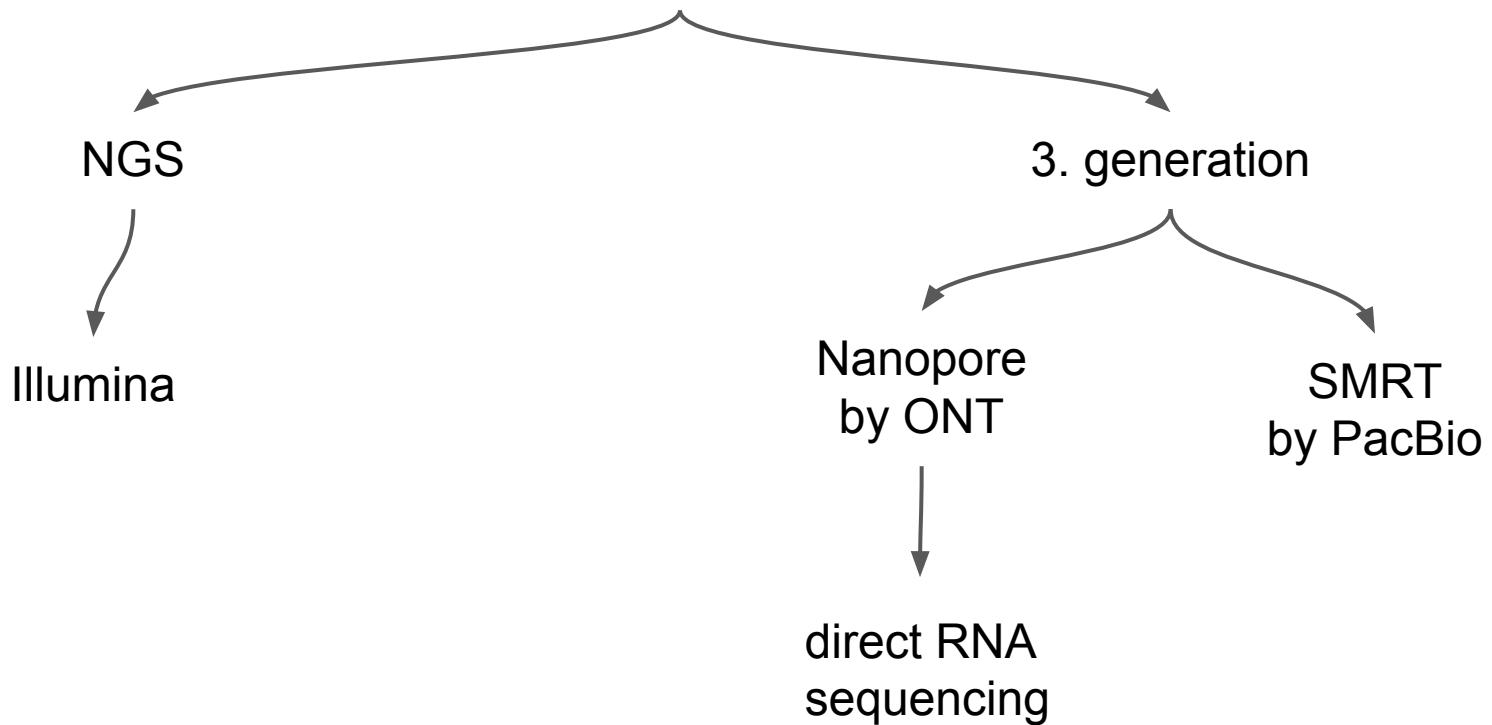


poly(A) tails diversity



Tails are dynamic!

High-throughput transcriptome sequencing



Illumina

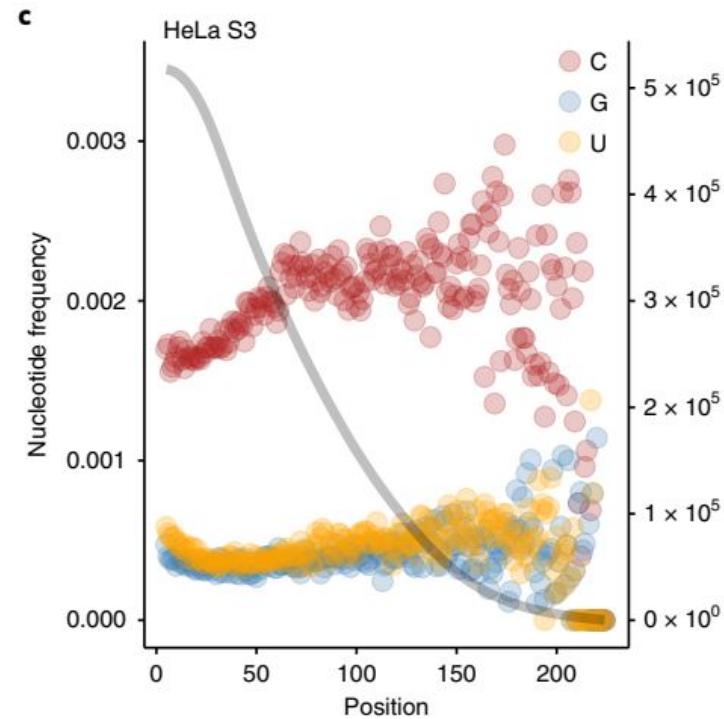
PAL-Seq: poly(A) tail length estimated from the fluorescence signal intensity normalized to cluster density

TAIL-Seq and **m-TAIL-Seq**: identification of the transcript from read 1 (51 nt) and examination of the poly(A) tail with read 2 (231 nt)

SMRT by PacBio

FLAM-Seq (full-length mRNA sequencing)

PAlso-Seq (poly(A) inclusive RNA isoform sequencing)

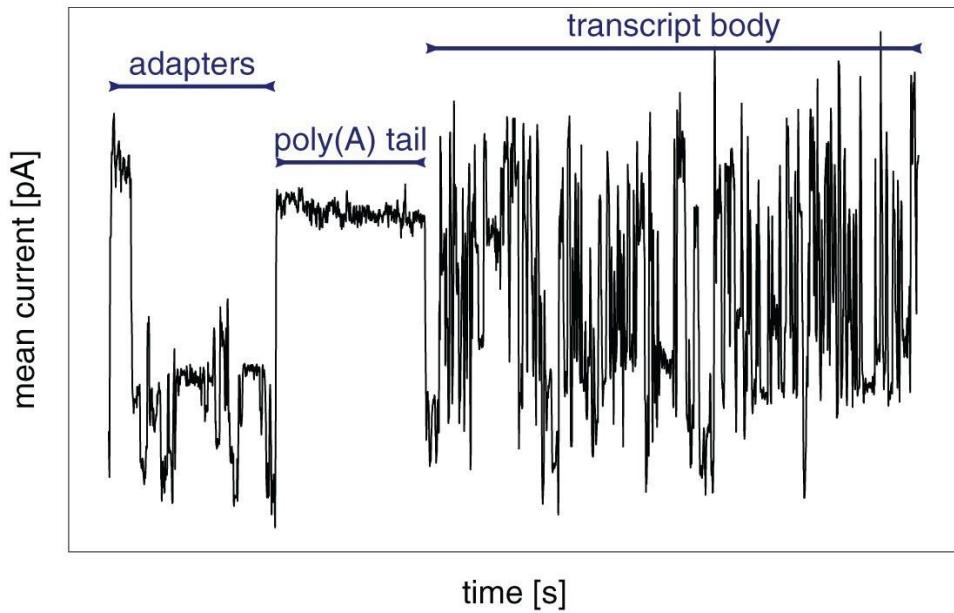


Legгинини, et al., 2019

Nanopore - direct RNA sequencing

Captures modifications and
enables precise length
measurements.

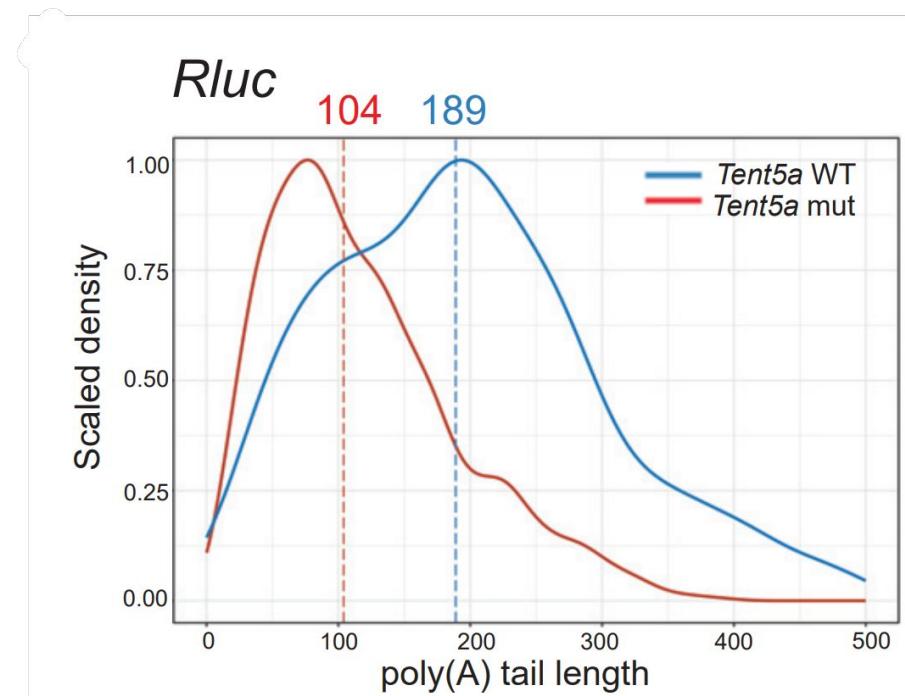
Nanopolish polyA, TailfindR -
poly(A) tail length measurement



Brouze et al., 2023

DRS-based pol(A) analysis in action

Role of cytoplasmic polyadenylation in bone formation.



References

- Brouze, A., Krawczyk, P. S., Dziembowski, A., & Mroczek, S. (2023). Measuring the tail: Methods for poly(A) tail profiling. Wiley Interdisciplinary Reviews: RNA, 14(1), e1737.
- Subtelny, A. O., Eichhorn, S. W., Chen, G. R., Sive, H., & Bartel, D. P. (2014). Poly(A)-tail profiling reveals an embryonic switch in translational control. *Nature*, 508(7494), 66–71. <https://doi.org/10.1038/nature13007>
- Chang, H., Lim, J., Ha, M., & Kim, V. N. (2014). TAIL-seq: Genome-wide Determination of Poly(A) Tail Length and 3' End Modifications. *Molecular Cell*, 53(6), 1044–1052. <https://doi.org/10.1016/j.molcel.2014.02.007>
- Lim, J., Lee, M., Son, A., Chang, H., & Kim, V. N. (2016). mTAIL-seq reveals dynamic poly(A) tail regulation in oocyte-to-embryo development. *Genes & Development*, 30(14), 1671–1682. <https://doi.org/10.1101/gad.284802.116>
- Legnini, I., Alles, J., Karaiskos, N., Ayoub, S., & Rajewsky, N. (2019). FLAM-seq: full-length mRNA sequencing reveals principles of poly(A) tail length control. *Nature Methods*, 16(9), 879–886. <https://doi.org/10.1038/s41592-019-0503-y>
- Liu, Y., Nie, H., Liu, H., & Lu, F. (2019). Poly(A) inclusive RNA isoform sequencing (PAIso-seq) reveals wide-spread non-adenosine residues within RNA poly(A) tails. *Nature Communications*, 10(1), 5292. <https://doi.org/10.1038/s41467-019-13228-9>
- <https://github.com/jts/nanopolish>
- <https://github.com/adnaniazi/tailfindr>

References

- Krause, M., Niazi, A. M., Labun, K., Torres Cleuren, Y. N., Müller, F. S., & Valen, E. (2019). *tailfindr*: alignment-free poly(A) length measurement for Oxford Nanopore RNA and DNA sequencing. *RNA*, 25(10), 1229–1241. <https://doi.org/10.1261/rna.071332.119>
- Workman, R. E., Tang, A. D., Tang, P. S., Jain, M., Tyson, J. R., Razaghi, R., Zuzarte, P. C., Gilpatrick, T., Payne, A., Quick, J., Sadowski, N., Holmes, N., de Jesus, J. G., Jones, K. L., Soulette, C. M., Snutch, T. P., Loman, N., Paten, B., Loose, M., ... Timp, W. (2019). Nanopore native RNA sequencing of a human poly(A) transcriptome. *Nature Methods*, 16(12), 1297–1305. <https://doi.org/10.1038/s41592-019-0617-2>
- Gewartowska, O., Aranaz-Novaliches, G., Krawczyk, P. S., Mroczek, S., Kusio-Kobiałka, M., Tarkowski, B., Spoutil, F., Benada, O., Kofroňová, O., Szwedziak, P., Cysewski, D., Gruchota, J., Szpila, M., Chlebowski, A., Sedlacek, R., Prochazka, J., & Dziembowski, A. (2021). Cytoplasmic polyadenylation by TENT5A is required for proper bone formation. *Cell Reports*, 35(3), 109015. <https://doi.org/10.1016/j.celrep.2021.109015>