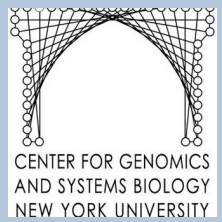




The dynamics of copy number variant evolution in fluctuating environments

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Abstract

Microbes live in dynamic environments which can pose ongoing challenges for survival. Unlike static conditions in which a selective pressure is continuous, fluctuating conditions cycle between selection and relaxation resulting in complex evolutionary dynamics. Copy number variants (CNVs) are a class of mutation in which a genomic locus varies in copy number. CNVs are widespread across all domains of life and have been implicated in diseases such as cancer. Despite the frequent occurrence of fluctuations in the natural environment, it is not well understood how they influence the dynamics of CNV generation and selection and the diversity of CNVs in a population.

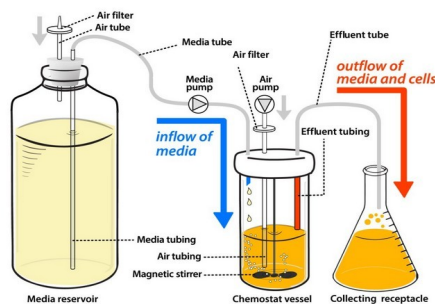
To understand the dynamics of CNVs in fluctuating environments, we track their frequency in budding yeast populations over hundreds of generations, while alternating between two conditions limited for nitrogen in the form of either glutamine and proline or glutamine and ammonium sulfate. Previously, we have found that transporters of glutamine (GAP1), proline (PUT4), and ammonium sulfate (MEP2) undergo gene duplication in chemostats limited for their respective nitrogen source. However, it remains unclear what the evolutionary outcome is when populations fluctuate between any two of these conditions. We developed dual-fluorescence CNV reporters for these transporters enabling inexpensive and rapid detection of duplications or deletions at two loci simultaneously. We find that the dynamics of CNV generation and selection are highly dependent on both condition and genetic locus. Overall, static conditions selected for greater copy numbers of a single transporter that is under selection, while fluctuating conditions selected for duplication at the two loci simultaneously but at lower copy numbers. This highlights the unique strategies required for survival in diverse conditions.

Questions

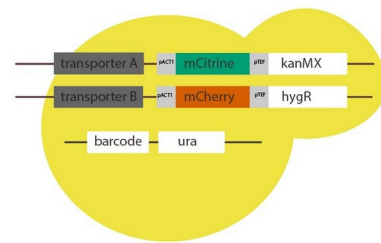
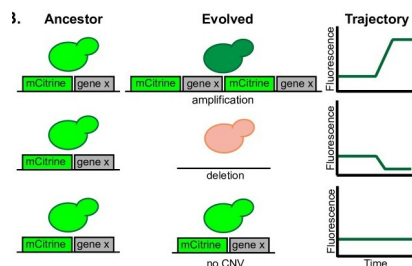
- What are the evolutionary dynamics of generation and selection of CNVs?
- What are the adaptive strategies co-opted? Generalists? Specialists?
- How does the environment affect population diversity?

Experimental Approach

Experimental evolution is performed in continuous culture



A dual-fluorescent reporter setup is used to track CNV dynamics



Adapted from Lauer et al 2018

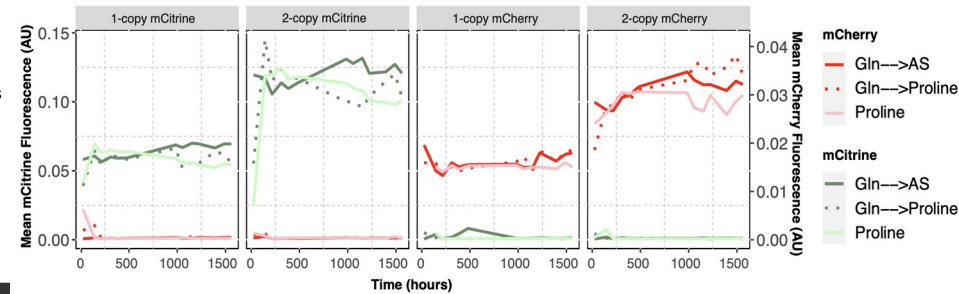
Evolve cells in static and fluctuating conditions for 200 generations

- Replicates of four.
- Generation time is six hours.
- Media switch every eight generations.

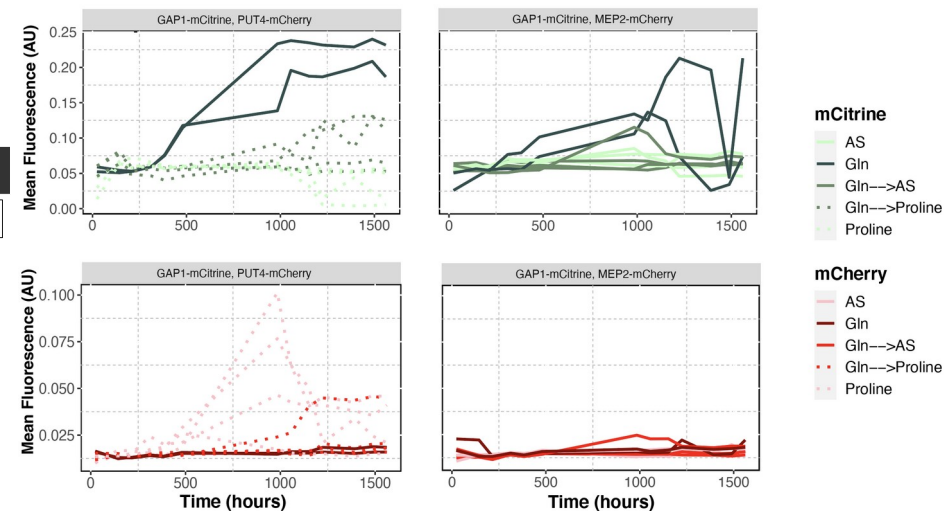
mCitrine-kanMX	mCherry-hygR	Nutrient-limitation
GAP1	MEP2	Amm-Sulfate Glutamine Amm-Sulfate → Glutamine
GAP1	PUT4	Proline Glutamine Proline → Glutamine

Results

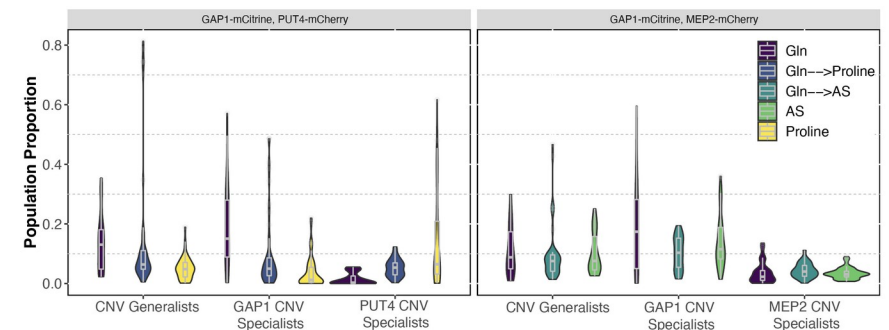
Fluorescence profile is a proxy measurement for gene copy number at two distinct loci



CNV evolutionary dynamics are condition- and genetic locus- dependent



CNV specialists arise in static conditions and CNV generalists arise in fluctuating conditions



Conclusions

Static environments show stronger selection for transporter CNVs.

No strong evidence that fluctuating environments select for greater heterogeneity in a population.

Fluctuating environments select for generalists with simultaneous CNVs at two distinct transporter loci.

Acknowledgements

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