

July 29, 1965

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Dear Bob:

Sankar Mitra finds that the ϕ X sample you sent us primes as well as his M13 DNA. The enclosed graphs describe two experiments with differing amounts of DNA polymerase. Here are some pertinent facts about this system. The DNA polymerase appears to free the endonuclease under these conditions for an 80-minute incubation. There are no single strand breaks as judged by release of tagged nucleotides (with ample opportunity for exonuclease II to act upon liberated 3' hydroxyl ends). The M13 DNA primer (tritium H^3 -labeled) remained resistant to exonuclease I after replication, whereas the P^{32} -labeled product was almost completely degraded (the exo I action was imposed after alkaline denaturation). Also, in an alkaline sucrose gradient the M13 DNA primer was not covalently linked to the product and sedimented with an S that suggested it had remained intact.

In view of the positive result with the ϕ X DNA, I think it would be worth checking to see whether it remained intact after serving as primer in the system. Tritium-labeled material with a specific activity in the neighborhood of 5×10^5 cpm/ μ m P would be suitable. Please let me know if you have any comments or suggestions that we could respond to.

As ever,

Arthur Kornberg

AK:es

Enc.