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May 22, 1974

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Dear Francis:

Thanks for your letter. I was glad that you were interested in the promoter sequence. I shall briefly bring you up-to-date with the different aspects of the work on the tyr tRNA gene and my other research interest as well.

The enclosed sheets essentially summarise the status of the work. (1) Synthesis: The DNA corresponding to the transcribed part of the gene (Smith-Altman precursor) has been synthesized. The focus is now on the synthesis of the terminator and promoter regions. (2) The promoter sequence (sheets 2 and 3) looks very beautiful to me. It could hardly be without significance. Transition from the "regular" DNA to the looped out form could be aided by the enzyme without loss of the essential recognition features and strand selection and site selection could both be accomplished in the process?? (3) The terminator sequence is on sheets 4 and 5. This too is interesting. Whatever the postulates at this stage, I am discontinuing sequencing just now and, instead, we are setting up precise systems (containing the known sequences for the signals and the adjacent parts) for studies of initiation and termination of transcription. After all, we have to prove the significance and lengths of sequences in the start and stop regions by actually carrying out transcription. If necessary, we shall go back to do more sequencing.

The ultimate goal is still to have a gene which is functional in in vitro transcription by virtue of its own signals. This should then provide a powerful approach to systematic alterations of the structural gene. Also, I would like to add our synthetic promoter and terminator to ends of "indifferent" DNA's to really prove what part does what.

Although this work takes up much of my effort just now, I hope that I shall be done with all this in the next year or so. In the last couple of years I have also become very deeply interested in the chemistry of membranes and have had a small group working in this field. Last fall I spent some time with Racker and his group at Cornell and this was most stimulating. I am still running a minor collaboration with him on reconstitution of membrane functions. At least this much I have definitely concluded that this sort of work is a reasonable starting point with my limitations and, especially, my deficient biological background.

I enjoyed your article in the 21st Anniversary issue of the DNA structure. If you have other writings in press, I would love to receive copies.

I hope all goes well in Cambridge researchwise and with your family. My warm regards to them.

With best wishes,

Gobind

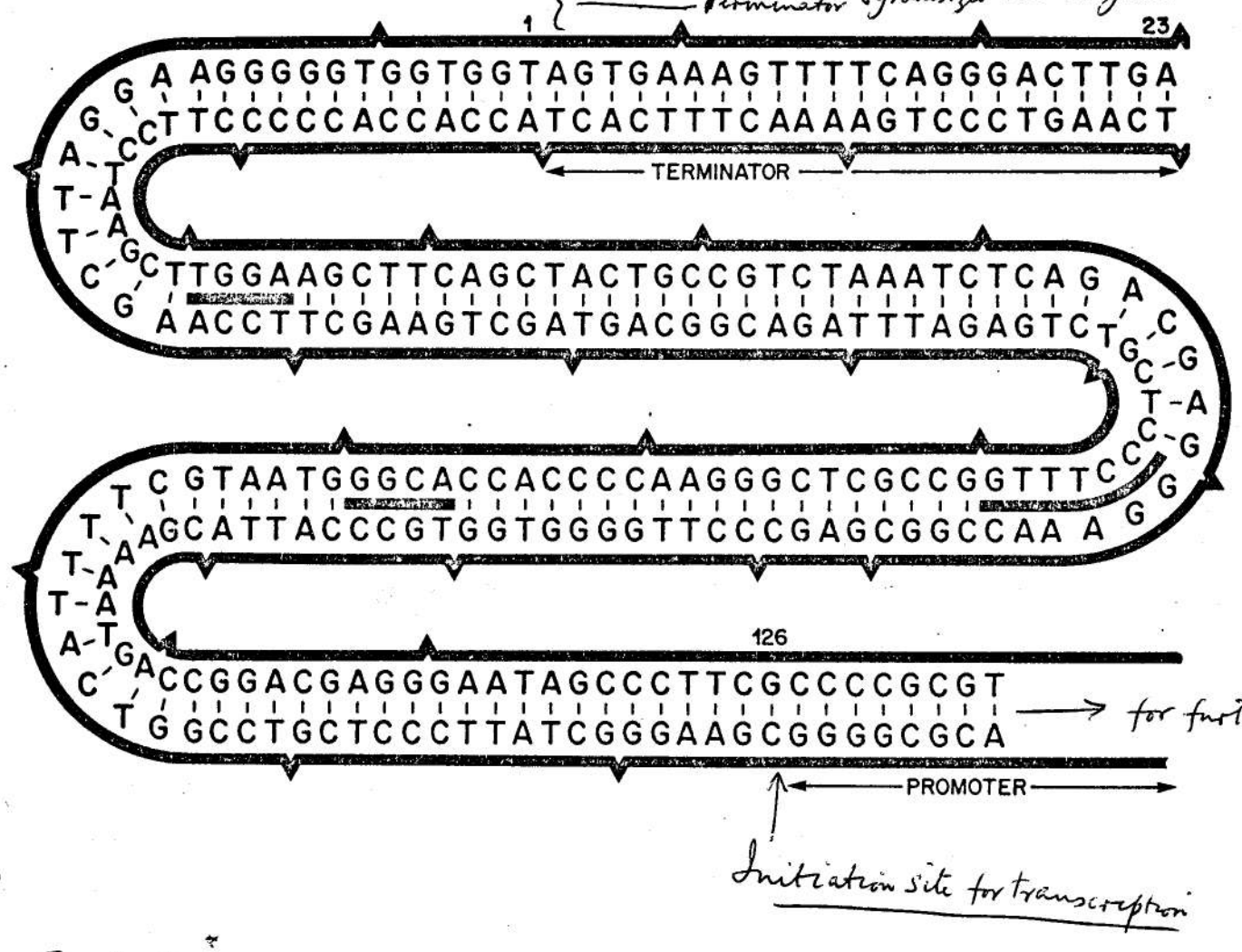
HGK/lb

H. Gobind Khorana

P.S. If you had any comments or thoughts on my above DNA work, I should of course be very happy to hear about them.

Synthetic Tyrosine tRNA Gene

{ ——— Terminator synthesized but not joined to the rest of the gene)

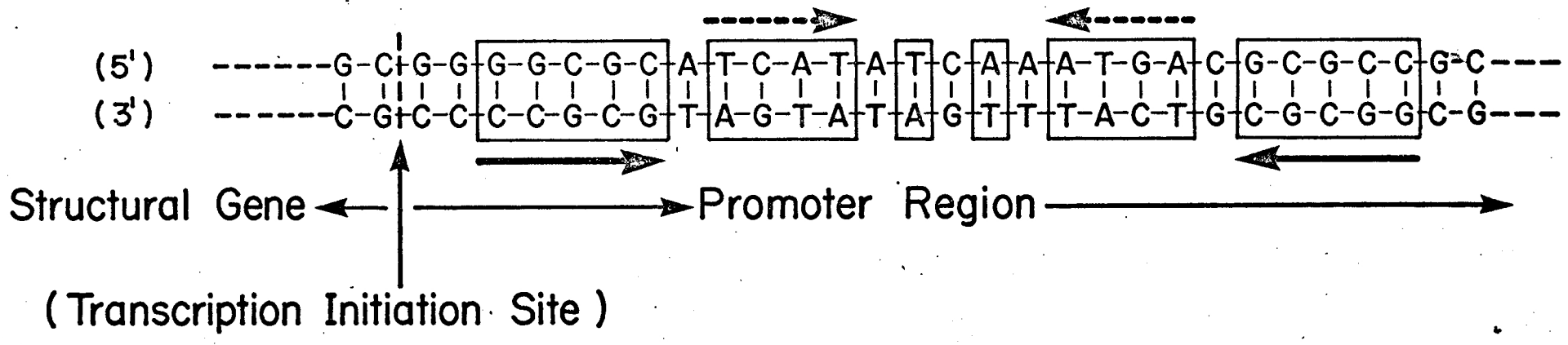


→ for further sequence see following sheets

Initiation site for transcription

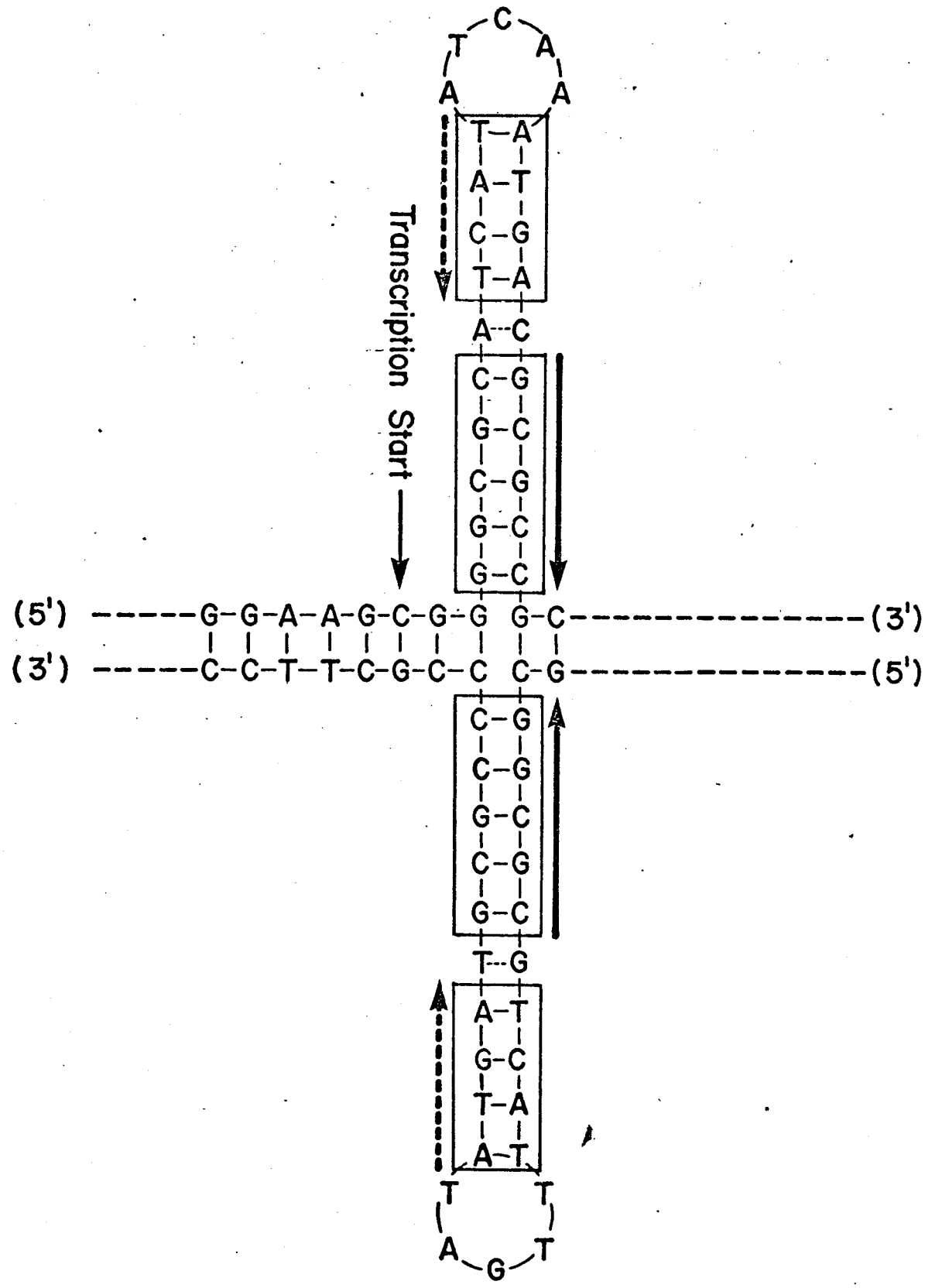
The Synthesis of the DNA corresponding to the total precursor (Altman + Smith) is complete. The above shows the scheme. The carrots show the points of enzymatic joining of the chemically synthesized segments (distances between carrots). The strips between the duplex are the sites of final joining

SEQUENCE IN THE PROMOTER REGION OF THE tRNA GENE (SYMMETRY AND SECONDARY STRUCTURE)



(3)

POSSIBLE SECONDARY STRUCTURE OF THE PROMOTER REGION



Possible Secondary Structure of Terminator Region