Warner J R, Knopf P M & Rich A. A Multiple Ribosomal Structure in Protein Synthesis. Proceedings of the National Academy of Science **49:**122-9, 1963.

While it had been known for some years that the ribosome was the site of protein synthesis, this paper was the first demonstration that protein synthesis in rabbit reticulocytes takes place on a multiple-ribosomal structure, termed a poly-some. The ribosomes are held together by a strand of RNA, presumably messenger RNA. The authors proposed a general model for the simultaneous translation of a single messenger RNA by several ribosomes. [The SCI® indicates that this paper was cited 547 times in the period 1961-1975].

Professor Jonathan R. Warner
Department of Biochemistry
Albert Einstein College of Medicine
Bronx, New York 10461

June 9, 1977

"This paper is a prime example of serendipity. I was a graduate student at MIT in Alex Rich's lab, interested in protein synthesis. Shortly after Nirenberg and Matthei's report on poly U stimulated protein synthesis, I had been working with Jim Darnell, trying to get f. *coli* ribosomes to translate poliovirus RNA. Reasoning that eukaryotic ribosomes might be more effective, I proposed a collaboration to Paul Knopf (now at Brown University). He had just finished his Ph.D. doing the Dintzis' experiment in a cell free extract of reticulocytes, and had two months to kill before setting out on the then obligatory post-doc at the MRC in Cambridge. We soon forgot the polio RNA. for in a control experiment analyzing ribosomes from reticulocytes which had been briefly labeled with ^{3}H leucine to identify active ribosomes, we observed peaks of optical density and radioactivity representing structures larger than 80S ribosomes. These structures were enriched in active ribosomes. Two possibilities occurred to

us: Were these aggregates of ribosomes created by the high speed centrifugation used to prepare the ribosomes, or were they fragile structures partly destroyed by the homogenization needed to dissolve the ribosomal pellets?

"To resolve these two possibilities, we decided to sediment the reticulocyte ribosomes directly from a cell lysate on a sucrose gradient Within a month we had done the experiments necessary to show that all protein synthesis in reticulocytes takes place on structures containing several ribosomes, usually five. The structures were insensitive to detergents, to ionic strength and to DNAse. but highly sensitive to RNAse and to shearing forces. **While Paul and I** were convinced of the importance of the finding, it remained for Alex Rich to **develop** the conceptual foundation of a stream of ribosomes flowing down a molecule of messenger **RNA.** A collaboration with Henry Slayter and Cecil **Hall,** also at MIT, provided the electron microscopic data to support this concept of a "polysome." One could see five ribosomes in a group, and occasionally a densely staining strand between them. It soon became **apparent that protein** synthesis in all organisms occurs on polysomes, which are a naturally efficient way to translate messenger RNA.

"Undoubtedly the time was ripe for finding polysomes Messenger RNA had been clearly established, and the race was **on to break the** genetic code. Alfred Cierer in Tubingen, also using reticulocytes, and Hans **Noll in** Pittsburgh, using rat liver, independently came to the conclusion that protein synthesis takes place on multi-ribosome **structures that** are highly sensitive to RNAse. **It is noteworthy that** all the pioneering **work in this area was carried** out in mammalian cells. Polysomes are so sensitive to shear **that they were, and still** are, difficult to prepare from tough organisms like bacteria and fungi.

For me, the experience was much more than a thesis. It was a "coming-of-age" as a scientist, giving me a sense of the excitement of science. Finally, it destroyed my **illusion that** science is a wholly rational pursuit. The fun is the unexpected observation which **one follows to** a discovery, and the challenge is distinguishing between the promising observations **and** the trivial artefacts."