Optical Communication: Heterodyne Detection Scheme

I should like to add a small contribution to R. Kompfner's discussion of optical communications (*1*):

With respect to the heterodyne detection scheme, Kompfner says that the practical realization depends on precise alignment of two independent light beams. Actually, a greater difficulty than this exists. An optical maser, or laser, has a frequency when free-running that is critically dependent on the distance between the two mirrors used to form the optical cavity. For a laser operating at 0.6328-μ wavelength (He–Ne) (approximately 5×10^{14} cy/sec), for example, and having a mirror separation of 1 m, a change in length of 10^{-7} cm would cause a frequency shift of about 0.5 Mcy. Such changes in length are easily caused by thermal variations or vibrations, and if not tracked exactly by a local oscillator result in prohibitive noise in the detected signal. These problems have both been recently solved (*2*).

The common type of gas laser operates with Brewster angle windows, which eliminate oscillation in one polarization plane of the laser. Thus, laser light is commonly plane polarized.

It has been demonstrated that a laser can operate in two different modes in the same space simultaneously, at different frequencies (*3*), if the laser is caused to move in such a way that the light in the two modes takes different times to make a circuit through the laser. I have since suggested use of two modes which are basically the two directions of polarization, separating them over part of the path, passing one of them through a plasma, and then from the beats (heterodyne detection) deriving the plasma density (*4*). Such a laser interferometer would be more sensitive than the previously described instruments (*5*) used for plasma diagnostics.

In the present case, special separation of the two beams is unnecessary. Instead, an electroactive crystal is put into the laser cavity (flat rather than Brewster windows are used), and the information signal is imposed on the crystal. The signal (a varying electric field) causes the crystal to have different optical lengths to the two light beams having different polarizations, and this difference in effective path length causes oscillation of the differently polarized beams at different frequencies, the frequency difference being dependent only on the field strength imposed on the crystal. The light beams occupy at all times the same space, and may be focused and transmitted together. Since the mirrors and other parts of the system are common, noise frequency changes track perfectly in the "local oscillator" beam, which is no longer generated locally at the receiver but at the transmitter. Perhaps it should be renamed; "reference" oscillator seems appropriate. The beams may be separated if desired by polarization film, a Glan-Thomson prism, or other means, but for communications this is not likely to be desirable. The system operates, of course, by frequency modulation.

I have written at length on this scheme because it is such an elegant solution to two of the important problems of laser communications and is not nearly so well known as it ought to be.

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Single Point Mutation or Chromosomal Rearrangement

Titani, Whitley, Avogardo, and Putnam (*1*) summarize their own findings and those of Hilschmann and Craig (*2*) on the amino acid sequences of three type I Bence Jones proteins. They discuss the origin of the many differences between the three Bence Jones proteins, which with one exception are all located in the NH_2-terminal half of the molecule. In an apparent attempt to explain the multiple differences as arising from a single event, they express the opinion that "The multiple structural differences . . . are incompatible with the concept of single point mutations now accepted for the abnormal hemoglobins." Instead they favor some sort of larger chromosomal rearrangement such as that postulated by Smithies (*3*).

It seemed desirable to check this conclusion by analyzing the amino acid interchanges in the light of recent information concerning the triplet code (*4*). The point mutations found in hemoglobin have all been explainable as substitutions of single nucleotide base pairs. If the variability in Bence Jones proteins is the result of larger chromosomal rearrangements, then the preference for single nucleotide shifts should not be present.

Of 12 probable interchanges indicated by Titani *et al.* in the NH_2-terminal half of the Bence Jones proteins, eight were based on definite sequence information. These were Asp and Glu at position 1, Ilu and Leu at 46, Glu and Ala at 55, Thr and Lys at 72, Leu and Val at 102, Glu and Asp at 103, Ilu and Phe at 104, and Lys and Arg at 105. All eight of these amino acid interchanges can be explained as single nucleotide substitutions. For example, according to Nirenberg's table the codons for aspartic acid are GAU and GAC, which can change in one step to the codons for glutamic acid, GAA and GAG. From the data in Nirenberg's table, slightly less than 40 percent of all possible amino acid interchanges can be explained as being due to a single nucleotide substitution. The probability that eight interchanges would be so explicable is thus 0.4^8 or 0.0006.

It thus appears that the best explanation of the amino acid interchanges in Bence Jones protein, like those in hemoglobin, is that they are point mutations due to the substitution of a single nucleotide base pair. If this is the case, then the multiple changes must represent an accumulation of point mutations.

A similar analysis with the triplet code makes possible a prediction of the probable order of the amino acid residues in one of the proteins (Roy) at positions 75 and 76, where it appears there is a dipeptide interchange. The report of Titani *et al.* indicates that in the protein Cu the sequence of this dipeptide is Arg-Val. Available information indicates that in Roy these positions are occupied by Asp and Leu without any indication of order. Since the interchange of Asp and Arg requires a two-step mutation, and the other possible interchanges are one-step, the dipep-

tide Leu-Asp is more probable than Asp-Leu.

Because of the accumulation of a large number of mutations in the NH_2-terminal half of the Bence Jones proteins, there is a reasonable probability that two-step mutations will be found. However, if a predominance of one-step mutations continues to be found as more data develop, this might be considered as strong evidence against Smithies' hypothesis of major chromosomal rearrangements.

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In our report we concluded that "The multiple structural differences reported . . . for Bence Jones proteins are incompatible with the concept of single point mutations now accepted for the abnormal hemoglobins" because the latter differ in but a single amino acid. Talmage's interesting analysis of the amino acid interchanges in Bence Jones proteins does give strong support to the hypothesis of an accumulation of point mutations, provided one weights equally all conceivable amino acid substitutions without regard to the unequal frequency of occurrence of the amino acids in proteins. However, of the eight probable sequence interchanges cited by Talmage, the three involving glutamic and aspartic acids are not suitable for the test, since Hilschmann and Craig did not report whether these amino acids were present in the amide form or not. Aspartic acid cannot go to glutamine by a one-step mutation in the codon, nor can asparagine go to glutamic acid. This would change the probability calculated from the known interchanges to 0.4^5 or 0.01. We now have additional data for three other interchanges—Asp and Lys at 53, Tyr and Arg at 89, and Asp and Leu at 90. Only the first of these is compatible with a one-step mutation.

In theory, multiple structural differences could arise from accumulation of point mutations, somatic chromosomal rearrangements, intracodon crossover (recombinational pseudomutation), or independent synthesis of the NH_2-terminal and COOH-terminal halves of the molecule. The last, however, is only a special case of the first three. An accumulation of point mutations as suggested by Talmage could occur either as a rapid flux of somatic mutations induced during plasma-cell division by the antigen (for antibodies) or carcinogen (for myeloma proteins) or through the presence of many modified antibody genes resulting from successive duplication and mutation over many generations. Multiple mutations within a somatic cell are unlikely, but with repeated genes there is much more chance for nonlethal mutation, since the unaffected genes can carry out the same function. If there is some linkage among the controls of these genes such that once one gene is active all the others are repressed, then the product of one gene would fail to repress that gene but might repress all others in the linked system. There are systems in which the activity of one gene represses the activity of all others (see T. M. Sonneborn and G. H. Beale, *Hereditas*, suppl. vol. 1949). Thus, the presence of many modified genes could provide the mechanism for specific antibody synthesis, as well as for the selective synthesis by plasmocyte tumor cells of a single myeloma protein or Bence Jones protein to the exclusion of the heterogenous system of normal immunoglobulins.

A second possibility to explain multiple structural differences in the NH_2-terminal portion of Bence Jones proteins would be independent synthesis of the NH_2-terminal half (residues 1 to 105) under the control of many genes and of the COOH-terminal half (residues 106 to 212) under the control of a single gene. A linking enzyme would be needed to unite the two halves at the "switch peptide." In principle, however, some such explanation as we have given would be needed to explain the origin of the many modified genes determining the NH_2-terminal portion.

Because of the high probability for one-step mutations indicated by Talmage's calculation, we were led to reexamine critically the Smithies hypothesis of intrachromatid inversion as the basis of γ-globulin variability [*Nature* **199**, 1231 (1963)]. Further study of the sequence data with reference to Nirenberg's table of codons has led us to conclude that it is unlikely that the sequence differences in Bence Jones proteins are ascribable to frequent, short-range, inverted duplications. We have not found any area of sequence in antigenic type I Bence Jones proteins where the amino acid interchange is compatible with the reversed complementary form of the codons as required by the Smithies hypothesis. Indeed, in none of the eight probable exchanges is the reverse complement of the codon for one amino acid of a substitution pair equivalent to the codon for the other of the pair. Hence, in none of these cases was there a crossing-over with inversion for a single codon. Of course, the critical regions are not yet testable; these are the large blocks where the composition differs markedly but the sequence is not yet known in two proteins, for example positions 2 to 32 and 75 to 85. Clearly, as we pointed out in our report, complete amino acid sequence analysis of a number of Bence Jones proteins of each antigenic type will be needed for an experimental test of the various theories of γ-globulin variability. Both chromosomal rearrangements and an accumulation of point mutations may yet prove to be involved.

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