suggestive that the thioester bond is also the precursor of peptide bonds in a number of bacterial products that happen to be heterogeneously constructed from D as well as L α-amino acids, and sometimes other precursors, as must have been the multimers postulated by my model. Thus, the thioester bond, born in hot, acidic sulphurous waters, could have been the central prebiotic source of both catalysis and energy, allowing the spreading of a long and complex chain of metabolic events that eventually led to something that could be called an RNA world, except that it would have been richly endowed with peptides.

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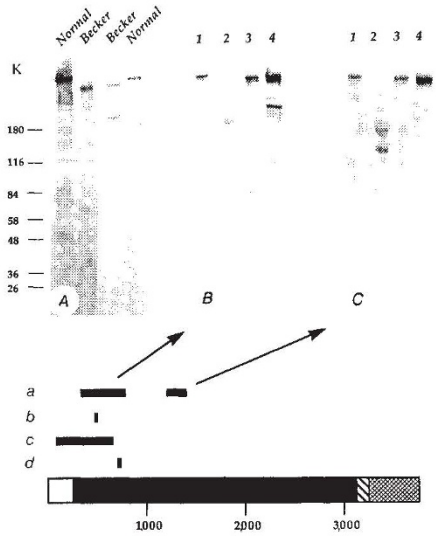
Proteolytic fragment or new gene product?

SIR—A recent report by Arahata *et al.*[1] has apparently created some confusion regarding the protein product of the Duchenne muscular dystrophy (DMD) gene. In an attempt to clear up this confusion, we would like to discuss some recent published data, and offer alternative explanations for the results presented by Arahata *et al.* which are more consistent with these data.

First, the DMD messenger RNA contains coding information for a protein of relative molecular mass (M_r) of 427,000 (427K)[2], a size in complete agreement with that of our previously reported product of the DMD gene, 'dystrophin'[3,5]. Contrary to this, Arahata *et al.* report a relative molecular mass of the DMD protein as 210K, without offering an explanation for how such a protein becomes half the predicted size.

Second, three independent laboratories have used peptide sequences derived from the same or similar messenger RNA sequences as those used by Arahata *et al.* to produce antisera which recognize the predicted 427K protein, called 'dystrophin' (see accompanying figure)[3-5]. Third, there is a large body of data which strongly correlates abnormalities of dystrophin (427K) with Duchenne and Becker muscular dystrophies at both the genetic and biochemical levels. Perhaps the most convincing of these are the dystrophin abnormalities associated with the milder 'Becker' alleles of DMD. Most Becker patients are predicted to possess 'in frame' exon deletions of the DMD gene which would result in a mRNA encoding a smaller DMD protein[6]. Indeed, the majority of Becker muscular dystrophy patients have a dystrophin of lower M_r in their muscle (ref. 7 and accompanying figure). Such results are impossible to explain unless dystrophin (427K) is the principal product of the DMD gene.



Bottom, a schematic drawing of the 427K dystrophin protein with its four distinct domains[2]. Above this are the regions of the dystrophin protein to which polyclonal antisera have been produced. (*a*, ref. 3; *b*, ref. 1; *c*, ref. 4; *d*, ref. 5). All antisera recognize a common 427K protein (dystrophin), except that characterized by Arahata, *et al.*[1]. Top, *A* shows two normal human muscle samples and two samples obtained from patients with Becker muscular dystrophy. Dystrophin (427K) is seen to be of abnormal (smaller) relative molecular mass in both Becker patients. The second patient exhibits a 200K degradation product of dystrophin which is characteristic of approximately 50% of Becker patients. *B* and *C* are duplicate immunoblots incubated with anti-dystrophin (427K) antisera directed against the indicated peptides[3]. Lane 1, normal human total muscle protein; lane 2, total muscle protein from a patient diagnosed as spinal muscular atrophy type III; lane 3, a second, fresh, biopsy obtained from this same patient; lane 4, homogenization and crude fractionation of the muscle biopsy shown in lane 3. The samples shown in lanes 2 and 4 exhibit non-specific degradation of dystrophin which resemble the immunoblot pattern of Arahata *et al.* There is also a slightly smaller smooth-muscle-specific isoform of dystrophin migrating immediately below the predominant striated muscle isoform[9]. Marker sizes are derived from prestained proteins (Sigma).

The immunofluorescent data provided by Arahata *et al.* for the 210K protein are in complete agreement with those obtained for dystrophin (427K) by two independent laboratories[4,5]. It is therefore likely that their antisera does detect dystrophin. However, the immunoblot results presented closely resemble those we have obtained when protein samples have been exposed to partial proteolysis (*B* and *C* of figure). Lane 2 contains total muscle protein from a previously frozen diagnostic muscle biopsy, and exhibits apparently 'abnormal' dystrophin of either 200K (*B*) or 200K and 150K (*C*), depending on the antibodies used. Because such an abnormal dystrophin pattern was unique in more than 200 patient biopsies that we have tested, a second, fresh, muscle biopsy was obtained from the same patient and similarly tested for dystrophin (lane 3). This new biopsy contained completely normal dystrophin. We conclude that the original biopsy had undergone partial, non-specific, proteolysis of dystrophin.

We believe that the extensive fractionation protocol employed by Arahata *et al.* resulted in a similar degradation of dystrophin. Although we find it impossible to replicate the fractionation conditions used by this group with the experimental detail given, we think it is significant that the homogenization of the fresh human biopsy (*B* and *C*, lane 3) followed by crude fractionation in the presence of protease inhibitors results in the appearance of proteolytic fragments of dystrophin of approximately 200K (lane 4). Such proteolytic fragments are of similar size to those reported by Arahata *et al.* They interpret the differing biochemical subcellular fractionation characteristics of the 210K protein as evidence for its non-homology with dystrophin, but it is difficult to equate the fractionation characteristics of a degradation product with that of its parent protein. We have previously reported that many Becker patients also exhibit a degradation product of dystrophin, similar in size to that shown by Arahata *et al.* (ref. 7, second Becker patient in accompanying figure.) We therefore suggest that the 210K protein is a proteolytic fragment of 427K 'dystrophin'.

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