Heterogeneity of Human Serum Beta-lipoprotein

Abstract. Human serum beta-lipoprotein (specific gravity 1.063 to 1.007) has previously been shown to form two lines on immunoelectrophoresis. In the study of a large number of individual sera it appears that both are not always present, that they are present in varying amounts in normal individuals, that a third form sometimes exists, that they are not separable by sedimentation techniques, and that they cross-react immunologically. These can also be demonstrated on starch gels and starch gel immunoelectrophoresis, one form entering the gel and the other remaining at the origin.

Human serum lipoprotein of specific gravity 1.063 to 1.007 is generally considered to monodisperse in a vertical starch gel. Attention has been called to its immunochemical heterogeneity by quantitative precipitin techniques (*1*), gel diffusion techniques (*2*), and more recently to heterogeneous enzyme activities sequestered by this fraction (*3*). When starch gel immunoelectrophoresis is used in combination with other forms, a number of components can be demonstrated.

Vertical starch gels were prepared according to the method of Smithies (*4*). Immunoelectrophoresis was carried out as described by Scheidegger (*5*). A 2 percent agar concentration gave best definition of the beta-lipoprotein. Goat (*6*) and horse (*7*) anti-human sera were obtained from commercial sources. Rabbit and mule anti-human sera were prepared in our laboratory. Beta-lipoproteins were identified either by staining with Oil Red O or by staining for enzymatic activity as previously described (*8*). Both whole human serum and beta-lipoprotein purified by mepasulfate precipitation and ultracentrifugation as described by Florsheim (*9*) were used as antigens. In starch gel immunoelectrophoresis, strips from a vertical starch gel following electrophoresis were placed on a thin (2 mm) agar plaque poured in a flat plastic box. Parallel troughs were filled with antiserum. Lines developed in 2 days and were stained for lipoprotein.

Two immunoelectrophoretic lines were identified as beta-lipoprotein and were present either alone or together. The first (*A*, Fig. 1) was rarely seen alone. It began near the origin and extended anodically almost parallel to the antiserum trough for a short distance. The second (*B*, Fig. 1) was an arc which usually crossed the first when both were present (*C*, Fig. 1). Complete separation of the two components is shown in Fig. 1*D*. These are probably the same as the lines described by Heide *et al.* (*2*) although in our hands they were not completely separable by ultracentrifugal techniques.

Examination of more than 1000 patterns made from both whole serum and ultracentrifugally prepared beta-lipoprotein showed that only rarely was either line to be seen alone; dominance of one or the other as measured by intensity of staining did occur, but the two were equally present most often.

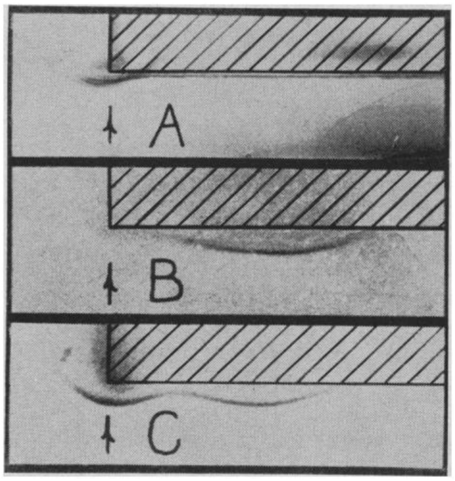

Fig. 2. Starch gel immunoelectrophoretic pattern showing various components. Blocking shows placement of starch on agar. Arrow indicates origin. *A*, fractions diffusing in agar which do not enter starch. *B*, fraction entering starch. *C*, two fractions in same serum. (Whole human serum vs. anti-human goat serum. Oil Red O stain.)

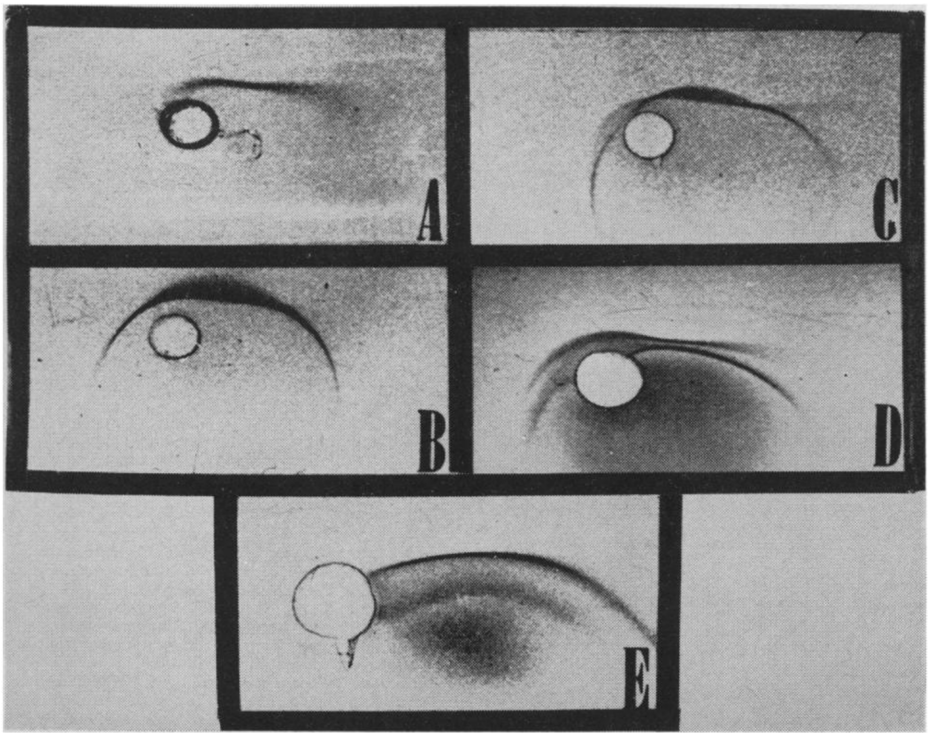

Fig. 1. Various configurations of beta-lipoproteins on immunoelectrophoretic slides. (Whole human serum vs. anti-human goat serum. Oil Red O stain.)

Another line which stained with Oil Red O but not with enzyme stains was sometimes seen inside the other two and could be eliminated by separating out lipoproteins of density less than 1.007 (*E*, Fig. 1). This line was more readily discernible in 1 percent agar.

Figure 2 shows the two components on starch gel immunoelectrophoresis. One component (*A*) does not enter the starch but will migrate in agar. The other (*B*) migrates a short distance into the starch. Both fractions in the same serum are shown in Fig. 2*C*.

Two serum samples were obtained 3 months apart from each of several individuals; the patterns of the two proteins were found to be identical in the paired samples.

Genetic control of at least four of the serum proteins has been proposed (*10*). It has been demonstrated that the beta-lipoproteins exist in more than one form which may be present singly or in combination in one individual which leads to the speculation about their genetic control (*11*).

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Gases in Tektite Bubbles

Abstract. Spectroscopic analysis of light produced by electrodeless discharge in a tektite bubble showed the main gases in the bubble to be neon, helium, and oxygen. The neon and helium have probably diffused in from the atmosphere, while the oxygen may be atmospheric gas incorporated in the tektite during its formation.

Gases in tektites have been reported by Döring and Stutzer (*1*), H. E. Suess (*2*), J. H. Reynolds (*3*), and Gentner and Zähringer (*4*). Suess also referred to earlier work by A. Brun. Döring and Stutzer, Brun, and Suess found CO, CO_2, and H_2. Suess found, in addition, water, which he considered to be absorbed in the outer layers of the tektites, and he estimated the gas pressure in a bubble to be less than 10^{-3} atms. Small quantities of argon and neon were detected by Reynolds, and of argon by Gentner and Zähringer. Reynolds measured the diffusion coefficients for australite glass at high temperatures, and showed that if the rates so obtained can be extrapolated to room temperature, the diffusion half-life for neon in a spherical tektite with 1 cm radius would be 1.1 million years and for helium 5.6 years.

We have investigated the gases in a tektite bubble by subjecting it to an electrodeless discharge. The tektite was bediasite No. 1876 of the U.S. National Museum collection, kindly placed at our disposal by E. P. Henderson. The cavity cannot be seen from the outside; it was discovered by Henderson through its density deficiency, which indicates a bubble whose volume is approximately 0.98 cm^3.

The tektite was placed in the tank coil of a radio-frequency oscillator at the Naval Research Laboratory. The oscillator operates at 96 Mcy/sec; the electric field intensity was about 1.5 kv/cm. The tektite lit up after a few seconds, emitting a soft, orange glow. It remained cold, however, even after long exposures. There was no detectable change in the intensity or character of the light after repeated excitation.

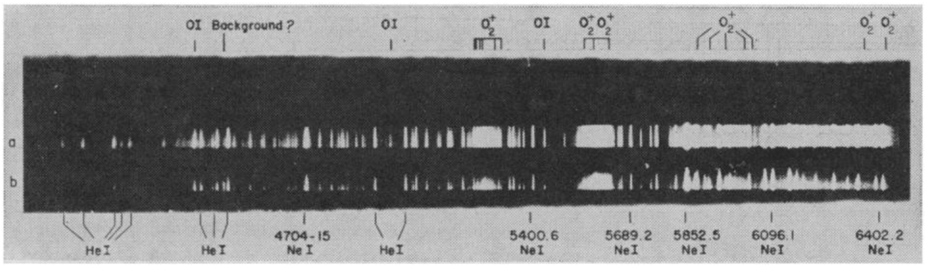

Fig. 1. Enlargements of spectrograms of bediasite No. 1876, film No. Z II. Spectrograms *a* and *b* were taken with different focusing conditions.

The light was focused on the entrance slit of a Meinel airglow spectrograph, kindly loaned by the Rocket Spectroscopy Branch, Atmosphere and Astrophysics Division of the Naval Research Laboratory. After a series of calibration and comparison exposures, the spectrograms shown in Fig. 1 were taken with exposures of 5 and 10 minutes on Agfa Isopan Record 35-mm roll film, which was developed for 8 minutes in D-76 at 70°C. The lines were measured with a D. W. Mann comparator and were identified from the M.I.T. wavelength tables compiled by Harrison (*5*) or from tables of molecular spectra compiled by Pearse and Gaydon (*6*). The dispersion of the spectrograph was found to be approximately 150 A/mm. We constructed dispersion curves by successive approximations after the strongest lines in the tektite spectrum had been identified by comparison with the spectrum of a neon bulb. Comparison exposures eliminated the possibility that the neon bulb used as a standard was responsible for any lines in the tektite spectrum.

The strongest atomic lines are those of neon (92 found) and helium (12 found). The helium lines found included all but three or four of the He I lines given in the *Handbook of Chemistry and Physics* (*7*) for the region between 3960 and 6400 A.

In addition, five lines in the spectrum, the strongest of which is at 4367 A, were identified with lines of atomic oxygen. All oxygen lines whose combined intensity in the tables of the *Handbook of Chemistry and Physics* (based on the M.I.T. tables) exceeded 150 were either found in the spectrum or explained as lost owing to blending with neon lines of intensity 1000 or more.

Broad bands, degraded to the violet, were found with heads near 5300 and 5630 A, as well as a band in the red, obscured by the strong neon lines, with head near 6020 A. We identify these as contributed by the First Negative system of O_2 (*6*). No lines or bands attributable to argon, hydrogen, or nitrogen were found.

The neon and helium have probably diffused into the bubble from the atmosphere because of the high permeability of tektite glass for these gases (*3*). This explanation is probably not possible for the oxygen; if so, it may be atmospheric oxygen incorporated into the tektite during its formation (*8*).

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