by the free acid of glutamic acid and by acid glutamate. These conclusions are supported by the work of LANDUA, FUERST AND AWAPARA[2]. The spot of R_F 0.35 is therefore not given by an ionic species of glutamic acid.

TABLE I

VARIATIONS IN R_F VALUES OF GLUTAMIC ACID WITH CHANGES IN CONCENTRATION AND pH

Experiment number	*Amino acid(s) dissolved in*	*Glutamic concentration* $M \times 10^{-2}$	*Glycine concentration* $M \times 10^{-2}$	R_F *of spots*
1	water	1.25	1.33	0.22, 0.27, 0.40
2		1.25	6.67	0.22, 0.27, 0.40
3		6.25	1.33	0.22, 0.27, 0.35, 0.40
4		0.25	0.266	0.22, 0.40
5		1.25	0.266	0.22, 0.27, 0.40
6		6.25		0.22, 0.27, 0.35
7		0.25		0.22
8	buffer pH 1	6.25		0.27, 0.35
9	buffer pH 3			0.22, 0.27, 0.35
10	buffer pH 7			0.22
11	buffer pH 12			0.22
12	buffer pH 1	0.25		0.27
13	buffer pH 3			0.27
14	buffer pH 7			0.22
15	buffer pH 12			0.22

Aspartic acid gives similar results to glutamic acid which indicates that the spot of R_F 0.35 is not due to the internal lactam of glutamic acid (*i.e.* the pyrrolidone carboxylic acid). This was confirmed by the failure to obtain a spot of R_F 0.35 after 0.01 *M* aqueous glutamic acid had been boiled for 5 hours. Similarly 0.0625 *M* glutamic acid in 6 *N* hydrochloric acid after being boiled for 5 hours still gave a spot of R_F 0.35 (see FOREMAN[3]).

It is clear from the experiments described above that the results obtained by BECK AND ÉBREY are caused by the high concentrations of glutamic acid used and that there is no interaction between the two amino acids. Two of the spots (R_F 0.22 and 0.27) given by high concentrations of glutamic acid are due to ionic species. The intensely coloured spot of R_F 0.35 does not represent an ionic species of glutamic acid neither does it represent the pyrrolidone carboxylic acid. An explanation for this spot is that there is interaction between the glutamic acid and the phenol. This interaction produces an essentially new compound possessing its own partition coefficient and moving independently. The interaction only occurs with high concentrations of glutamic acid and it only occurs with some solvents. Butanol-acetic acid gives results similar to those given by phenol but in aqueous acetone both high and low concentrations of glutamic acid give single spots of the same R_F value. Usually for paper chromatography the concentrations of glutamic acid used are such that this interaction with solvents does not occur. Full details of this work will be published later.

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Infra-red dichroism of fibrous proteins

The dichroic ratio of the NH(ν) absorption band at 3300 cm^{-1} is surprisingly low for most naturally occurring fibrous proteins[1,2]. Similar measurements of our own show that feather keratin from goose quill rachis has a perpendicular ratio of about 1.8:1, while α-keratin from porcupine quill tip has a parallel ratio of 1.5:1. The particular specimens on which these measurements were made were shown by their X-ray diffraction photographs to have good orientation as regards their more crystalline regions. By contrast, a well oriented specimen of poly-benzyl-L-glutamate prepared from *m*-cresol has been shown to give a parallel ratio of 15:1[3]. It seems reasonable to suppose that

the structures of polypeptides and proteins are at least basically similar, and if this is so, we still have to explain the discrepancy in the dichroisms.

We have already suggested that the reason for the lower value in natural proteins is the presence of a relatively large component which absorbs from 3600–2700 cm^{-1} and which is non-dichroic[4]. This conclusion was first based on the shape of the absorption envelope and later supported by early infra-red studies involving partial deuteration of proteins. The latter work was carried out in collaboration with R. BIRLEY, of the Department of Textile Industries of this University. However, the method used had the drawback that the specimen had to be transferred from a deuterating compartment to the infra-red cell, where it was dried over P_2O_5. Both during transference and drying, regain of hydrogen could take place, so that the full effects of deuteration were not observed. A similar difficulty has been encountered by HVIDT AND LINDERSTRØM-LANG[5]. It has now been avoided in more recent experiments, and some of the results are given here.

Curve A, Fig. 1, shows the spectrum of a feather keratin section that had been enclosed in a cell with a drop of D_2O for 14 days, and curve B shows the spectrum of the same section after regain of hydrogen followed by drying over P_2O_5. The difference between the two curves gives the absorption due to the hydrogen atoms replaced by deuterium, and it is seen to consist of a broad region stretching from 3600 – 2700 cm^{-1}. Polarisation measurements show that this absorption is not dichroic.

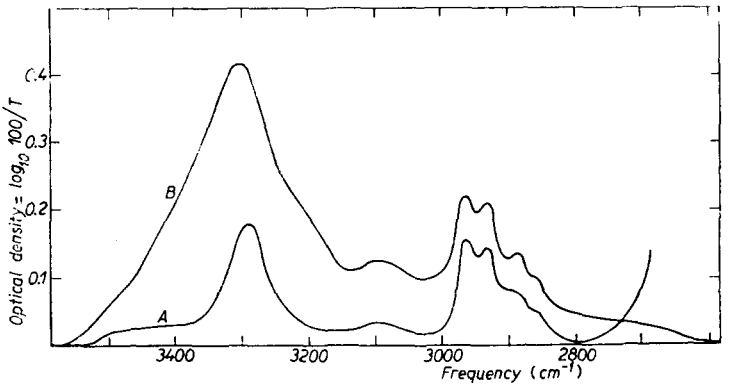

Fig. 1. Infra-red absorption of feather keratin. (A) In contact with D_2O; after 14 days. (B) After regain of hydrogen and dried over P_2O_5.

The following kinds of hydrogen may be involved:

(1) In chemically bound water still remaining in the keratin after drying over P_2O_5.

(2) Attached to oxygen or nitrogen atoms in side-chains. These hydrogen atoms are roughly equal in number to the peptide hydrogens, and owing to their diversity would be expected to absorb over the whole spectral region in question.

(3) Peptide hydrogen atoms which can be replaced by deuterium through contact with D_2O.

Probably all these types are replaced to some extent, but separation of their effects is not easy, and will not be discussed here.

The important point is that the observed dichroism of the sharp residual NH (ν) band is always considerably higher in the deuterated than in the original specimens. For porcupine quill a parallel value of 4.5:1 has been obtained, and for feather keratin a perpendicular value of 4.8:1. Since only a small number of sections have been examined, from one specimen of each material, it is likely that this method might eventually yield higher values still.

These results tend to discount one of the outstanding apparent differences between synthetic polypeptides and natural proteins and thus to support current theories of protein structure. Other materials, including collagen and chitin, have also given interesting results by this procedure. A fuller account of the investigation is in preparation.

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