Communications to the Editor

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Studies on Synovial Fluid by Infrared Spectra and their Application to the Diagnosis of Gout

Synovial fluid is usually a serum component mixed with hyaluronic acid, chondroitinsulfuric acid, cell components, and other substances. Infrared spectra of synovial fluid were measured with those obtained from patients suffering from rheumatism, gout, and other diseases. Spectra were measured with synovial fluid dried *per se* and with synovial fluid components obtained by deproteinization with methanol or other means under specific conditions.

The infrared spectrum of the dried whole fluid exhibited almost entirely the absorption of proteins, its chief component, and there was no marked difference among various articular diseases. However, infrared spectrum was found to be an effective means in the case of synovial fluid from gout patients. Gout is a disease in which uric acid and urates, which should physiologically be excreted easily from the kidneys, accumulate in the body in large quantities as a result of abnormal metabolism. In majority of cases, this accumulation occurs in the joint, causing inflammation and pain. In the initial stage, gout is difficult to distinguish from rheumatism, acute arthritis, and many other diseases.

The infrared spectrum of synovial fluid from patients suffering from gout, even in initial stages, exhibits the absorptions of sodium hydrogen-urate[1] which appear at 1664 (v. s.), 1600 (s), 1521 (m), 1430 (m), 1386 (s), 1353 (m), 1258 (m), 1195 (vw), 1132 (w), 1005 (s), 886 (m), 826 (m), 794 (s), 766 (s), 737 (m), and 717 (s) cm^{-1}. These absorptions appear overlapped with absorptions of proteins and polysaccharides. These absorptions become somewhat indistinct in the region of strong absorptions of proteins at 1700~1500 cm^{-1} and that of polysaccharides at 1000~1100 cm^{-1}, but are distinct in other regions. It had been known that synovial fluid in gout patients contained uric acid or urate but nothing was known in what form uric acid was present. In the range of spectra measured in the present series of work, the acid was invariably present in the form of its monosodium salt. It had also been believed that gout is a rather rare disease but the present spectral studies revealed that the disease is not so rare. The reason for it seems to be due to the difficulty of distinguishing gout from rheumatism and other diseases in most of the cases.

The specific absorptions become more distinct when infrared spectrum is measured with dried filtrate obtained after deproteinization of synovial fluid by addition of methanol. In this case, the absorption of sodium hydrogen-urate appears overlapped with sharp absorptions of mucopolysaccharides such as hyaluronic acid.

In many of articular diseases, absorption of sodium hydrogen-urate generally does not appear. The infrared spectrum of dried, deproteinized synovial fluid principally shows the absorptions of hyaluronic acid[2] with some changes in some of their intensity. Studies along this line are being followed.

The infrared spectrum of dried synovial fluid should be measured by the potassium bromide disk. Addition of procaine hydrochloride at the time of collection of the fluid should be avoided as the presence of this substance complicates the absorptions and makes it inconvenient for detection of uric acid.

1) G. Chihara, *et al.* : This Bulletin, **7**, 622(1959).
2) S. F. D. Orr : Biochim. et Biophys. Acta, **14**, 173(1954).

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Infrared Analysis of Bile

Analysis of gallstones by infrared absorption spectrum was established and this method was shown to be an effective and suitable means in the analyses of gallstones.[1~2] This work was further extended to the analysis of bile by infrared absorption spectra.

Bile contains bile acids, bile pigments, lipids, inorganic components, and many other substances, and detailed studies have been made on each of these components. The routine clinical examination of bile chiefly concerns the color, turbidity, and presence of bacteria, and analysis of chemical components is hardly carried out because the analysis requires complicated and difficult procedures.

There have never been any reports to date on the application of infrared absorption spectrum for analysis of bile and it was found from the present series of work that the infrared spectrum is also an excellent method for the analysis of bile, as was the case in that of gallstones.

1) Infrared Spectrum of Dried Whole Bile and its Interpretation :

Infrared absorption spectrum of dried whole bile gives informations on the components of bile, any changes thereof, and relative changes of the components. The infrared spectrum of the bile from the gallbladder shows approximately constant absorptions in healthy persons and, in spite of the bile being a mixture of numerous substances, a spectrum with many, fairly sharp absorptions can be obtained. The positions of main absorptions are at 796, 846, 852, 898, 913, 952, 982, 1001, 1017, 1052, 1079, 1111, 1171, 1201, 1229, 1311, 1405, 1465, 1551, 1655, and 1738 cm^{-1} and, although there are some shifts, these are overlapped absorptions of sodium glycocholate and taurocholate, cholesterol, lecithine, triglycerides, polysaccharides, and proteins. This fact was confirmed through separation of various bile components and comparison of absorptions of standard specimens. There are instances where absorption of sodium glycocholate is abnormally strong, bile acids are few in quantity, or cholesterol is abnormally great in quantity, and various changes in the components appear as the changes in spectral intensity.

1) G. Chihara, S. Yamamoto, H. Kameda : This Bulletin, **6**, 50(1958).
2) To be published.