Ferrography—a new method for isolation of particles from biological fluids

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Experienced microscopists examining synovial fluid have emphasised the cellular and crystalline material present.¹ There remain fibres² and amorphous particles³ whose nature and role in arthritis are yet to be clarified. Wear particles derived from contacting surfaces in machinery can be examined by processing lubricant using the technique of ferrography.⁴ This entails the separation of metallic wear particles from a sample of lubricant by virtue of their magnetism. A small sample of lubricant is pumped over a prepared glass trough or "substrate" (Fig. 1) situated on a permanent magnet. The metallic particles are precipitated according to their size and magnetic moment and can be identified by microscopy. Ferrography has been adapted for synovial fluid particles⁵ and it is envisaged that the method might be applied to other biological suspensions.

Fig. 1 Diagram of ferrograph analyser.

Material and methods

PREPARATION OF THE SAMPLE

Synovial fluid (2.5 ml) is collected into washed containers impregnated with 100 units of sodium heparin, and 5 ml of 0.9% saline added. After centrifugation (3000 rpm for 15 min) removal of the supernatant leaves a clot of protein, fibres and particulate material. This washing procedure is repeated with a further 5 ml 0.9% saline, and after the supernatant is discarded, the deposit is treated with 20 units of fungal hyaluronidase at 37°C for one hour. (This enzyme diminishes the quantity of coagulated material but does not destroy human articular cartilage under such conditions.) The

washing procedure is then repeated three times using 0.9% saline as above. The remaining deposit is treated with 1 ml of an aqueous solution of erbium chloride, which substitutes Er^{3+} for other cations on particle surfaces and allows particles to be deposited according to their size and magnetic moment in a magnetic field. The prepared suspensions can be analysed by ferrography. Isopropyl alcohol is used to fix biological particles to the substrate.

COST AND MATERIALS

Capital expenditure on the ferrograph analyser and

Fig. 2 Large particle isolated by ferrography. Surface markings suggest cartilaginous origin (incident light $\times 100$). (a) Sample prepared by suspension of cartilage scrapings in saline. (b) Similar particle isolated from synovial fluid.

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a suitable microscope would involve appreciable outlay, though a rheumatological unit will usually have adequate facilities for microscopy. The substrate and reagents can be purchased in kit form from the makers of the ferrography analyser (Foxboro Analytical, Milton Keynes, Bucks) at a cost for one examination comparable to that of a chest x-ray.

Discussion

The most obvious source of "wear particles" in human pathology must be the joints. Ferrography enables isolation of synovial fluid particles and an example of one believed to be cartilage is shown in Fig. 2. Particulate material was described by Ropes and Bauer in 1953,⁶ and since then the proposal that large numbers of fibrils in joint fluid signify extensive cartilage destruction⁷ has remained unchallenged. Lessons from metallurgy suggest that size and shape of particles may be at least as important as numbers. The use of joint replacements has increased the need to assess wear in human joints, both in terms of prosthesis lifespan and their effects on remaining human tissues. We would also suggest that ferrography may be used to prepare microscopic preparations of particulate material in bile or urine from

patients who form calculi, and sputum in patients exposed to occupational hazards.

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Letters to the Editor

Pulmonary haematoxyphil bodies

One of the interesting observations in the article "Effects of oxygen on the lungs after blast injury and burns"¹ was that of giant nuclear masses which the authors regard as being derived from endothelial cell nuclei similar to those reported by Brown *et al*² in experimental defibrination. I have found similar large nuclear masses in human lungs and in an experimental model of smoke inhalation in which acrolein was inhaled by rabbits³ (Fig. 1).

By electron microscopy these nuclear masses are found to have a rim of residual cytoplasm containing granules of the same type as those found in platelets (Fig. 2) which identifies them as megakaryocyte nuclei. Where many of these are present it is usually also possible to identify recognisable megakaryocytes even at light level.⁴ Brown *et al*² could distinguish giant endothelial nuclei from megakaryo-

Fig. 1 Numerous lobulated haematoxyphil masses in the lung of a rabbit which inhaled acrolein (arrows). Haematoxylin and eosin, original magnification $\times 300$.