readily than microscopical examination of processed tissue," but this has not been systematically tested.

These five cases of microsporidia represent 6.5% of the African HIV positive patients with intestinal symptoms that we saw. Three of the patients also had cryptosporidiosis (diagnosed on faecal smears, though not seen on histological examination). Irrespective of the degree of inflammation, no microsporidia have been identified in 36 small bowel biopsy specimens from HIV negative Ugandan and Zambian controls; no microsporidia have been seen in over 50 rectal biopsy specimens from HIV negative or HIV positive patients.

More intestinal biopsy specimens from patients with AIDS should be examined for microsporidia. This group of parasites infects most phyla of invertebrates and all classes of vertebrates; yet *E. bieuensi* has only been described in human enterocytes in association with HIV-1 infection. It is unresolved whether they are genuine pathogens rather than mere passers. The enterocytes containing microsporidia in our material and that of other workers do not show obvious damage by light or electron microscopy. The aetiology of the diarrhoea in AIDS is multifactorial, and microsporidia may be involved, possibly by affecting secretion.

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References


Visual aid for quick assessment of coronary artery stenosis at necropsy

A chart was designed to give a quick visual assessment of the degree of coronary artery stenosis at necropsy. It was intended for routine cases in which coronary artery angiography and formal morphometric analysis of processed segments of vessel were not to be undertaken. The method described here is not the most accurate, but it is preferable to simple subjective categorisation into simple, moderate, and severe degrees of artery stenosis.

Ischaemic heart disease is one of the leading causes of death in Great Britain. Quantification of coronary artery stenosis as part of the necropsy is necessary to estimate the functional importance of any atherosclerotic disease present. The cross sectional area of the vessel lumen, compared with that contained within the elastic lamina, expressed as a percentage, is a widely used method of estimating the degree of luminal area narrowing within a vessel. A narrowing by 75% reduces coronary blood flow at times of stress and exertion; narrowing by 90% means that coronary blood flow is severely reduced at rest. Even in the absence of coronary thrombosis, cardiac death may occur if there is one segment of luminal area which has narrowed by more than 85%, although most of these patients have multiple foci of stenosis.

Coronary angiography can be used routinely at necropsy to show the luminal diameter and it can also show all branches of the coronary vessels. Formal morphometry with the use of either point counting, or planimetry of multiple cross sections of vessels that have been fixed, decalcified, and stained for elastin, give a good estimation of luminal area narrowing. Vessels that have been perfused and fixed at physiological pressures before dissection give the most meaningful results. An alternative, but the most routinely used method, is simple subjective assessment of a serially sliced vessel at the time of necropsy into categories of mild, moderate, and severe stenosis.

The first two methods are obviously more accurate, and the merits and demerits of each have been discussed elsewhere. The aim of our method is not to replace or compete with these methods but to make simple, immediate, subjective assessment more accurate and scientific.

Examination of coronary arteries by serial transverse slicing at 3 mm intervals is widely practised. Most pathologists open the heart first and then slice the vessels while gripping the artery, with forefinger outside the heart and thumb inside. A sharp brain knife allows a “clean slice” to be made even with moderate vessel calcification.

Once the vessels have been serially sliced, the cut surface is compared with the chart (figure). The vessels have been drawn with concentric round, eccentric round, and slit shaped lumina representing the three main patterns seen at necropsy. The diagrams have been photoreduced following the calculation of the areas on graph paper. Arteries that are distended at physiological pressure are almost circular in shape.

Figure Schematic diagram representing a percentage reduction in cross sectional area of coronary artery vessels. The outer circle represents vessel exterior, inner circle represents the elastic lamina, and black area represents the lumen.
slit shapes of some lumina are artefactual due to collapse: the degree of stenosis at necropsy is therefore greater than in life, and the degree of collapse will vary according to the proportion of the wall which is free of disease; this chart can only be used as a rough guide in these cases. Pressure fixation would be required to circumvent this problem.

Requests for a chart on A4 size paper can be made to Dr Champ.

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References

Elimination of parallax error in haematocrit readings using reflexion haematocritometry

The precision of haematocrit readings can be improved by using a magnifying glass. Unfortunately, the precision resulting from magnification may be undermined by the parallax error. The consequence of the parallax error is easily recognised if the eye or the lens is moved along the axis of the microhematocrit tube; the readings of the erythrocytes, buffy layer and plasma concentration will be completely changed. The respective error of the haematocrit increases with decreasing haematocrit and cytocrit.

To avoid the parallax error, a reflexion haematocritometer was developed according to the following principle. A transparent scale was mounted on a surface mirror and covered by a glass layer. This reflecting unit was provided with a screw driven fixation device for the microhematocrit tube so that the bottom of the red cell column could be gently adjusted to the zero line of the scale (figure). When observed by a magnifying glass, the real and reflected cell and plasma layers can be adjusted to a common level, indicating the reading position of the eye and the lens, thus avoiding the parallax error. Coefficients of variation were assessed with whole blood anticoagulated with edetic acid or leucocyte poor erythrocyte suspensions in anticoagulated plasma or in buffer.

Ten microhaematocrit capillaries of 75 mm in length were filled to a 60 mm mark and sealed. Between each filling procedure the blood tube was closed and mixed by hand. The 10 capillaries were centrifuged together for five minutes (12500 × g at the bottom of the tubes). After centrifugation they were kept in vertical position until the reading. In experiments 8–17 repeated readings of single capillaries were performed. In experiments 1–16 haematocrit measurements were performed with the instrument placed horizontally; experiment 17 was done with the instrument in vertical position. A 10 × eye piece of a microscope was used for all the readings because this is available in most haematological laboratories. The table shows the coefficients of variation of the haematocrit of whole blood and of erythrocyte suspensions in plasma or buffer. At high concentrations of haematocrit all the coefficients of variation were around or below 0.5%. For those around 0.07, the coefficients of variation averaged 2.5%. In experiment 1 the buffy layer was read in addition to the plasma and red cell layers and the cytocrit (sum of the red plus white cell columns divided by the length of the whole column) was also calculated and was 0.478 ± 0.001 and the coefficient of variation 0.21%. In experiments 8–17, 10 repeated readings of single capillaries were performed. In experiments 8–13 coefficients of variation were obtained similar to those in experiments 1–7. This was due to the fact that manipulation between readings led to some crowding of the uppermost erythrocyte layer after four to six readings (table). In experiments where manipulation was reduced to the necessary minimum, the coefficient of variation was zero for whole blood and erythrocytes suspended in buffer at a low haematocrit (experiments 14, 15). At a higher concentration of erythrocytes in buffer, however, some crowding also occurred (experiment 16) but could almost be suppressed when the instrument was in vertical position (experiment 17).

The high precision of the readings obtained with this instrument compared with those obtained with a non-reflecting scale was due to (1) the elimination of the parallax error and (2) the mechanical adjustment of capillaries to the instrument scale.

Figure Reflexion haematocritometer with haematocrit capillary and eye piece in position and screw (a) for driving the capillary holder (b) and light source (c).
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