

A comparison of peripheral blood and buffy coat smear examination for the prediction of bone marrow relapse of acute lymphoblastic leukaemia in childhood

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SUMMARY In an attempt to see if buffy coat smear examination might be an alternative to bone marrow aspiration for predicting relapse, 98 consecutive bone marrow aspirates from 96 children with acute lymphoblastic leukaemia were examined blind with buffy coat and peripheral blood from the same patients. The 28 bone marrow aspirates from children no longer on treatment were all normal, and routine aspirates would appear unjustified in these patients. Eight of the remaining marrows showed relapse, but only three were not predicted from the peripheral blood and buffy coat. In no case was buffy coat superior to peripheral blood in the detection of bone marrow relapse. Routine bone marrow aspirates are an inefficient way of diagnosing relapse in acute lymphoblastic leukaemia in childhood, despite their precision, and a prospective study is needed to determine their value.

The repeated bone marrow aspirates used to confirm continuing remission during maintenance chemotherapy for acute lymphoblastic leukaemia are physically unpleasant and mentally distressing, especially for children. Recent work by Haworth *et al*¹ suggests that routine bone marrow examination does not alter prognosis, but it continues to have a place in planning therapy and may help reduce morbidity by detecting relapsed disease earlier. Watson *et al*² have proposed that routine bone marrow examination is not worthwhile and should be stopped.

Occasionally relapse is detected first in peripheral blood and the ability of this to predict relapse might be improved if more white cells could be examined, for instance by inspecting buffy coat smears. Such preparations are known to demonstrate more bone marrow elements than are usually seen in peripheral blood^{3,4} and this study was carried out to see if the examination of buffy coat smears might be of practical use in the detection of bone marrow relapse, and so enable the numbers of bone marrow aspirates to be reduced.

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Patients and methods

Ninety-eight consecutive, non-selected bone marrow aspirates from 96 children with acute lymphoblastic leukaemia attending the Leukaemia Clinic at the Hospital for Sick Children, Great Ormond Street, were studied. Twenty-eight were from children no longer on treatment, and 70 from 68 children on treatment.

Blood and bone marrow smears were prepared routinely. Because of the small volume of many of the blood samples, buffy coats were prepared by the centrifugation of peripheral blood in a capillary tube for three minutes using a microhaematocrit centrifuge. After breaking the capillary on the plasma side, the buffy layer was transferred to a glass slide, the cells resuspended in a drop of plasma, and the smear made as for a blood film.

All smears were collected, numbered and examined blind after all aspirates had been performed, which was over a six-week period. Bone marrow relapse was defined as more than 5% blasts, and blood and buffy coat smears were assessed in terms of requiring "no action," of necessitating bone marrow aspiration ("suspicious") or of showing the presence of leukaemia. The total haemoglobin, white cell count with differential, and platelet count were studied with each child's haematology flow chart to determine whether a bone marrow aspirate would be

indicated from these values. To act as controls, the peripheral blood and buffy coat smears of 18 non-leukaemic children were examined with the patients.

Results

None of the 18 control buffy coat and blood smears was considered to be suspicious or to show disease.

Of the 28 bone marrow aspirates from children off treatment, none showed disease. One suspicious blood, and one buffy coat smear was noted as false-positives. Both of these patients continue in remission.

There were eight relapsed bone marrow aspirates in the 70 samples from the 68 children on treatment for their leukaemia. Of the six florid relapses two were missed by blood and buffy coat smears, and peripheral blood count. Two were detected by both peripheral blood and buffy coat. In one case the peripheral blood was suspicious but the buffy coat preparation appeared normal, and in the sixth the blood count alone was abnormal. The two relapses missed had 12% and 65% blast cells in the marrow, and in those detected there were 61%, 95%, 76% and 55%, the latter two being detected by the peripheral blood alone. Two marrow aspirates had only 7% and 8% blasts respectively and one of these relapses (7%) was suggested by both peripheral blood and buffy coat. The other was not detected by either.

Thirteen marrows that were in remission were suspected of being in relapse from the examination of the other smears and blood counts. In nine cases the peripheral blood alone was suspicious, and in three only the buffy coat. In one child who had just successfully completed reinduction chemotherapy for recently relapsed disease both blood and buffy coat were suspicious. Of these 13 that may be considered false-positives for relapse, nine continue in complete remission over one year later. Only two relapses occurred within four months, both in the central nervous system, at one and four months respectively.

Discussion

Buffy coat smears did not provide any additional information to that obtained from peripheral blood. However, only three relapses were discovered that were not suggested by the peripheral blood. A reappraisal of routine marrow aspirates would appear warranted not only from the aspect of their contribution to outcome¹ but also with regard to the small number of unexpected positives.

In the 28 children who had completed their chemotherapy, no case of relapse was detected, and so routine bone marrow aspiration would appear inappropriate for this group of patients. If the policy

had been to inspect the peripheral blood and buffy coat before proceeding to the bone marrow aspiration two marrow aspirates would have been performed because of an incorrectly suspicious buffy coat or peripheral blood (one case each).

If the same policy had been in operation for the 70 aspirates from children on treatment the number of bone marrow aspirates performed would have been reduced to 16. Examining only the peripheral blood count and smear would have led to 14 aspirates of which five would have been in relapse. If the buffy coat had been examined first then six aspirates would have been performed with three relapses. In no case did buffy coat examination detect a relapse that was not suspected by the peripheral blood smear or count. Three cases of relapse would have been missed by both methods, these having 8%, 12% and 65% blast cells respectively. To detect these three cases it was necessary to do 56 aspirates not indicated by examination of the peripheral blood.

These data suggest that routine bone marrow aspiration in children on chemotherapy for acute leukaemia is of questionable value, particularly for children who have stopped treatment. Approximately half the relapses will be detected if a policy is adopted of careful study of peripheral blood films proceeding to bone marrow aspiration only if indicated. The detection of the remainder of relapses involves a much greater effort which may not be justified by the benefit achieved.

Haworth *et al*¹ have questioned the value of routine bone marrow aspirates in terms of their contribution to prognosis, and since most children are seen at regular intervals for chemotherapy, the likely delay in diagnosing relapsed disease from peripheral blood may not be great. The potential savings in terms of children and parents anxiety and medical and nursing time would be considerable if routine bone marrow aspirates were discontinued, and this would appear reasonable in the light of the results obtained. A prospective study to determine whether early diagnosis of relapse by routine marrow aspiration is of any benefit would be valuable but as has been noted by Haworth *et al*,¹ Watson *et al*,² and in this study, such events are relatively infrequent and a large group of patients would be needed.

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