peripheral blood cell counts. With the method of preparing the nucleic acids used by these authors, degradation of the nucleic acids and heavy protein contamination was to be expected and thus transfer of intact templates may probably be excluded. Non-specific stimulation of the hæmopoiesis by thymidine residues and analogues was probably the mechanism of action in this case. This and similar works established the basis for the therapeutic use, now discontinued, of nucleotides in aplastic anæmia to stimulate hæmopoiesis. However, one problem is to stimulate hæmopoiesis and another is to redifferentiate anaplastic leukæmic stem cells. Further evidence for true redifferentiation of the leukæmic cells may have to be gained by the use of normal RNA on cultures of leukæmic cells in the absence of potentially hæmopoietic normal reticulum cells.

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CHANGE OF HUMAN CHROMOSOME COUNT DISTRIBUTIONS WITH AGE: EVIDENCE FOR A SEX DIFFERENCE

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In previous communications we reported that the proportion of aneuploid cells in cultures of human leucocytes increased with the age of the subject[1], and that, at any given age among adults, the proportions did not differ appreciably between persons with and without cancer[2]. We now have much more data, and also have evidence that there is a difference between the sexes both in the manner in which the proportions of aneuploid cells change with age, and also in regard to the chromosomes affected.

Altogether data are available for 8,380 cells from 247 subjects with normal karyotypes inclusive of those reported previously. These cells have been obtained from blood cultures prepared by the technique of Moorhead *et al.*[3]. For each aneuploid cell an attempt has been made to describe the missing or additional chromosomes. The distribution of chromosome counts is recorded in Table 1 for each age-group, and divided according to chromosomal sex. The proportion of aneuploid cells (expressed as a percentage of the diploid cells) increases with age in both sexes; but the rate of increase in the proportion of hypodiploid cells differs between the sexes. Comparison of the regression coefficients in the linear regression formulæ (Table 1) shows the coefficients for men and women to differ significantly ($P < 0\cdot01$). Furthermore the male data are satisfactorily explained by a linear regression, whereas inspection suggests that the female data would be better fitted by a cubic curve in which the rate of increase is low under the age of 45 years, high between the ages of 45 and 64 years, and low again at ages over 65 years. Under 45 years of age, the trend in the female data is similar to that for males, and can be fitted by the linear equation $y = 0\cdot056x + 2\cdot03$.

Table 1. DISTRIBUTION OF CHROMOSOME COUNTS BY AGE AND CHROMOSOMAL SEX

	Age group (yr.)	No. of subjects	Mean age*	No. of cells	No. of cells with counts of <46	46	>46	Expressed as % of diploid cells <46	>46
XY Chromosomal sex	0–4	10	1·10	300	11	287	2	3·83	0·70
	5–14	13	9·98	370	13	354	3	3·67	0·85
	15–24	22	20·89	989	36	948	5	3·80	0·53
	25–34	25	29·61	990	40	941	9	4·25	0·96
	35–44	8	37·72	256	13	239	4	5·44	1·67
	45–54	14	49·77	597	34	556	7	6·12	1·26
	55–64	10	58·50	300	19	275	6	6·91	2·18
	65–74	10	70·47	320	23	292	5	7·88	1·71
	75+	11	80·67	349	30	307	12	9·77	3·91
	All ages	123	36·15	4,471	219	4,199	53	5·22	1·26
XX Chromosomal sex	0–14	9	7·22	270	6	262	2	2·29	0·76
	15–24	33	20·08	968	33	930	5	3·55	0·54
	25–34	28	29·74	900	29	860	11	3·37	1·28
	35–44	13	39·08	452	19	427	6	4·45	1·41
	45–64	16	57·07	571	33	533	5	6·19	0·94
	65–74	11	68·09	330	37	291	2	12·71	0·69
	75+	14	81·72	418	46	356	16	12·92	4·49
	All ages	124	39·66	3,909	203	3,659	47	5·55	1·28

Regression on age of the percentage of: (i) hypodiploid cells (XY): $y = 0\cdot082x + 2\cdot26$; (ii) hypodiploid cells (XX): $y = 0\cdot161x - 0\cdot84$; (iii) hyperdiploid cells (XY): $y = 0\cdot360x - 0\cdot04$; (iv) hyperdiploid cells (XX): $y = 0\cdot031x + 0\cdot05$. Each of the regression coefficients differs significantly from zero. For (i)–(iii), $P < 0\cdot01$; for (iv) $P = 0\cdot04$.

* Weighted for the number of cells counted.

In examining the types of chromosomes involved in the formation of the aneuploid cells, an attempt has been made to assign the missing or additional chromosomes to one or other of the following six groups, based on the Denver classification. Where this has been possible a cell is said to have been fully analysed. In a number of cells a full analysis

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was not possible, but a partial analysis could be made. Such cells in males were included with those fully analysed even though the chromosome could only be ascribed to the M/L group, and in females where the chromosome could only be ascribed to the S group (S_1–S_4).

Chromosomes	Group
21, 22 and Y	S_1
16, 19 and 20	S_2
17 and 18	S_3
13, 14 and 15	S_4
6–12 and X	M
1–5	L

In group L, autosomes Nos. 1, 2 and 3 can of course be recognized as individual entities, but so few cells contained an abnormal number of L autosomes that this refinement was omitted. In the subsequent analyses of the aneuploid cells, those cells have been omitted which were of too poor a quality for full analysis, together with all cells carrying structural chromosomal abnormalities. Furthermore, cells with a chromosome count of 43 or less have also been discarded as many or all are likely to be artefacts due to rupture of the cell. The distributions of the counts in the remaining cells are given in Table 2; the results show that the proportion of hypodiploid cells still increased with age, and that the regression coefficients still differed significantly ($P < 0{\cdot}01$). In this and the subsequent analyses the numbers of hyperdiploid cells were found to be too small for any valid conclusions to be drawn.

Table 2. DISTRIBUTION OF CHROMOSOME COUNTS BY SEX AND AGE AFTER EXCLUSION OF CERTAIN GROUPS OF ANEUPLOID CELLS*

Chromosomal sex	Age group (yr.)	No. of cells with chromosome counts <46	No. of cells with chromosome counts >46	Percentage of diploid cells: Cells with hypodiploid counts	Percentage of diploid cells: Cells with hyperdiploid counts
	0–14	17	3	2·65	0·25
	15–24	28	5	2·95	0·52
	25–44	45	3	3·81	0·57
XY	45–54	24	2	4·32	0·47
	55–74	21	1	3·70	0·34
	75+	20	10	6·52	2·81
	All ages	155	24	3·69	0·64
	0–24	26	4	2·18	0·62
	25–34	20	2	2·09	0·21
XX	35–54	18	10	3·39	0·85
	55–64	21	5	4·90	0·90
	65–74	20	5	6·87	0·88
	75+	32	7	8·99	2·28
	All ages	137	33	3·65	0·79

Regression on age of the percentage of:
selected hypodiploid cells, sex XY : $y = 0{\cdot}036x + 2{\cdot}39$
" " " " XX : $y = 0{\cdot}104x - 0{\cdot}40$
Both regression coefficients are significantly different from zero ($P < 0{\cdot}01$)

* See text for definition. The number of subjects, their mean ages and the numbers of diploid cells counted in each group are given in Table 1.

Inspection of the data suggested that in males an undue proportion of the hypodiploid cells was missing an S_1 chromosome, whereas in females an undue proportion was lacking an M chromosome. In fact the proportion of these cells not only increases with age (Table 3), but also these cells account for nearly all the regression on age previously observed. If these cells are excluded then the regression of the remaining hypodiploid cells on age becomes extremely small, and the coefficients do not differ significantly from zero in either sex (males, $y = 0{\cdot}006x + 2{\cdot}15$; females, $y = 0{\cdot}005x + 1{\cdot}41$).

Table 3. DISTRIBUTION OF HYPODIPLOID CELLS LACKING S_1 AND M CHROMOSOMES BY AGE AND CHROMOSOMAL SEX

Chr. sex	Age group (yr.)	Number	% of Diploid cells
		Cells lacking only S_1 chromosomes	
	0–14	4	0·62
	15–24	4	0·42
	25–44	6	0·51
XY	45–54	8	1·44
	55–74	8	1·41
	75+	10	3·26
	All ages	40	0·95
		Cells lacking only M chromosomes	
	0–24	9	0·76
	25–34	5	0·52
XX	35–54	10	1·88
	55–64	10	2·33
	65–74	15	5·16
	75+	27	7·58
	All ages	76	2·02

Regressions on age of the percentage of
(i) XY cells : $y = 0{\cdot}030x - 0{\cdot}120$
(ii) XX cells : $y = 0{\cdot}098x - 1{\cdot}822$
Both regression coefficients differ significantly from zero ($P < 0{\cdot}01$)

The results reported here are open to criticism on the grounds that the observations were not made on a representative sample of the general population. The subjects were comprised of healthy volunteers, hospital in-patients, parents of children with chromosome abnormalities, etc. To offset this difficulty a study is in progress of a group of subjects selected at random from the lists of general practitioners. This investigation will eventually include men and women of all ages from 15 years and upwards, but so far data only referable to those of 65 years and more are available. These, however, confirm the findings already noted for this group, and in this study every cell has been fully analysed. Examination of 1,050 cells from 35 males (mean age 71·69 years) shows 9·26 per cent to be aneuploid and that the proportions increase progressively for the age-groups 65–69, 70–74 and 75 years and over, being respectively 7·14, 10·09 and 12·15 per cent. 48 cells with 45 chromosomes were available for analysis, 27 of which were missing an S_1 chromosome (expected number on the basis of random loss, 5·22). In 12 of these cells the missing chromosome could confidently be said to be a Y chromosome, whereas only 1·04 such cells would be expected if all chromosomes had an equal chance of being lost. The findings from the analysis of 1,290 cells from 43 females (mean age 73·49 years) again confirmed the earlier findings. The proportion of aneuploid cells was 11·50 per cent and this proportion did not change progressively for the age-groups 65–69, 70–74, 75–79 and 80 years and over, the relevant proportions being 11·37, 12·68, 11·94 and 10·20 per cent. 95 cells with a count of 45 chromosomes were available for analysis, of which 81 were missing an M chromosome (expected number on the basis of random loss, 33·04). The substantial agreement between the earlier and later findings for ages of 65 years and more does indicate that it is unlikely that any serious bias has been introduced into the earlier findings due to the composition of the study population.

It seems reasonable to postulate that the observed increases in the proportion of aneuploid cells with age, which in males is mainly due to the loss of an S_1 chromosome and in females to the loss of an M chromosome, is in fact largely due to divisional errors involving the Y chromosome in men and the X chromosome in women. Direct evidence is available for the Y chromosome as in good preparations it is identifiable, and therefore its absence is recognizable. Direct evidence for the involvement of the X chromosome is not yet available, but it may become so from techniques using tritiated thymidine labelling. It may well also be relevant that the major change involving an M chromosome in women occurs at the time of a sharp decline in sexual activity. Both in males and females the findings could be explained: (1) if there was a special liability for cells to lose a sex chromosome, or (2) if all chromosomes were liable to be involved, but that the cells with an abnormal number of autosomes were at a disadvantage and failed to survive, or (3) if both possibilities occurred together.

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