with GM-CSF showed an absorbance of 50% of control at a concentration of 10 μM tiazofurin, which was 77% of the value observed for untreated cells. Dose–response studies are shown in Fig. 1; cells pretreated with GM-CSF were significantly ($P < 0.01$) more sensitive towards tiazofurin, particularly in the concentration range between 10 and 100 μM tiazofurin (concentrations which could be achieved in the plasma of leukaemia patients).

DISCUSSION

Tiazofurin, a novel chemotherapeutic agent, proved effective in the treatment of refractory chronic granulocytic leukaemia in blast crisis. The drug is metabolized to TAD, which is a potent and specific inhibitor of IMP dehydrogenase, the key enzyme of *de novo* guanylate biosynthesis. Treatment of malignant cells with tiazofurin therefore leads to a decrease in intracellular GTP and dGTP concentrations. An enhanced activity of the guanylate *de novo* pathway in logarithmically growing hepatoma cells compared to lag- or plateau-phase cells has been reported (Weber *et al*, 1988). While salvage activity seems to be sufficient to provide cells with guanylates in G0/G1- and G2-M phase of the cell cycle, the additional activity of the *de novo* pathway is required to meet an increased demand in S-phase (Weber *et al*, 1988). This also explains why tiazofurin, due to an increased IMP dehydrogenase activity in S-phase cells, is more effective in this cell cycle phase (Szekeres *et al*, 1992).

In addition to growth inhibition and cytotoxicity, tiazofurin has been reported to cause cell differentiation and down regulation of c-myc and Ha-ras oncogene expression *in vitro* and in patients, which might relate to the observed clinical potential of the substance (Knight *et al*, 1987; Weber *et al*, 1991).

Development of resistance through the emergence of quiescent leukaemia cell populations during chemotherapy continues to constitute a major problem in the treatment of leukaemia patients (Andreeff *et al*, 1987). Faluri demonstrated that incubation of fresh leukaemic cells as well as leukaemia cell lines with GM-CSF recruits quiescent leukaemia cells into the cell cycle (Faluri & Andreeff, 1990). These cell populations were more susceptible to subsequent killing by the cell cycle specific drug Ara-C (cytosine arabinoside).

Incubation of HL-60 cells with GM-CSF alone resulted in accelerated proliferation and increased IMP dehydrogenase activity. These data are in line with results from Weber *et al* (1988) who could show a proliferation- and transformation-linked increase of IMP dehydrogenase activity.

Novel aspects of this study include the following: (1) IMP dehydrogenase activity is increased in HL-60 cells pretreated with GM-CSF; (2) GM-CSF treatment enhances the biochemical (GTP depleting) and (3) cytotoxic effects of tiazofurin. As GM-CSF is not only able to enhance the biochemical and cytotoxic effects of tiazofurin *in vitro*, but also to recruit quiescent leukaemic blasts into the cell cycle, and to shorten chemotherapy induced neutropenia in patients, the sequential combination treatment with GM-CSF and tiazofurin might offer an important option in the treatment of refractory myeloid leukaemia.

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REFERENCES

Andreeff, M., Gaynor, J., Chapuis, D., Little, C., Gee, T. & Clarkson B.D. (1987) Prognostic factors in acute lymphoblastic leukemia in adults: the Memorial Hospital experience. *Haematology and Blood Transfusion*, Vol. 30 (ed. by T. Buechner *et al*), pp. 111–124. Springer, Berlin.

Bettelheim, P., Valent, P., Andreeff, M., Faluri, A., Hainz, J., Gorischek, C., Munn, M., Sillaber, Ch., Haas, O., Vinzer, E., Maurer, D., Schulz, G., Speiser, W., Geissler, K., Kier, P., Hinterberger, W. & Lechner, K. (1991) Recombinant human granulocyte macrophage colony stimulating factor in combination with standard induction chemotherapy in de novo acute myeloid leukemia. *Blood*, 77, 700–711.

Fritzer, M., Barabas, K., Szasz, V., Berczi, A., Szekeres, E., Faulk, W.P. & Goldenberg, H. (1992) Cytotoxicity of a transferrin–adriamycin conjugate to multidrug-resistant cells. *International Journal of Cancer*, 52, 619–623.

Jayaram, H.N., Dion, R.L., Glazer, R.I., Johns, D.G., Robins, R.K., Srivastava, P.C. & Cooney, D.A. (1982) Initial studies on the mechanism of action of a new oncolytic thiazole nucleoside, 2-β-D-ribofuranosyl thiazole-4-carboxamide (NSC 286193). *Biochemical Pharmacology*, 31, 2371–2380.

Knight, R.D., Mangion, J., Lucas, D.L., Cooney, D.A., Khan, E.C. & Wright, D.G. (1987) Inosine monophosphate dehydrogenase and myeloid cell maturation. *Blood*, 69, 634–639.

Liu, M.S., Laderian, M.A., Diepstraten, H.J., Natsumeda, Y., Olah, E., Jayaram, H.N. & Weber, G. (1984) Modulation of IMP dehydrogenase activity and guanylate metabolism by tiazofurin (2-β-D-ribofuranosylthiazole-4-carboxamide). *Journal of Biological Chemistry*, 259, 5078–5082.

Pillwein, K., Jayaram, H.N. & Weber, G. (1987) Effect of ischemia on nucleosides and bases in rat liver and hepatoma 3924A. *Cancer Research*, 47, 3092–3096.

Szekeres, E., Fritzer, M., Pillwein, K., Felzmann, T. & Chiba, P. (1992) Cell cycle dependent regulation of IMP dehydrogenase activity and effect of tiazofurin. *Life Sciences*, 51, 1309–1315.

Faluri, A. & Andreeff, M. (1990) Kinetic rationale for cytokine induced recruitment of myeloblastic leukemia followed by cycle specific chemotherapy in vitro. *Leukemia*, 4, 826–834.

Tricot, G.J., Jayaram, H.N., Lapis, E., Natsumeda, Y., Nichols, C.R., Kneebone, P., Heerema, N., Weber, G. & Hoffman, R. (1989) Biochemically directed therapy of leukemia with tiazofurin: a selective blocker of inosine 5′ phosphate dehydrogenase activity. *Cancer Research*, 49, 3696–3701.

Weber, G., Jayaram, H.N., Lapis, E., Natsumeda, Y., Yamada, Y., Yamaji, Y., Tricot, G.J. & Hoffman, R. (1988) Enzyme-pattern targeted chemotherapy with tiazofurin and allopurinol in human leukemia. *Advances in Enzyme Regulation*, 27, 405–433.

Weber, G., Nagal, M., Natsumeda, Y., Ible, J.N., Jayaram, H.N. & Parikh, I. (1991) Tiazofurin down-regulates expression of c-Ki-ras oncogene in a leukemic patient. *Cancer Communications*, 3, 61–66.

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SHORT REPORT

Haematological disorders and occupational hazards: a British Society for Haematology/Health and Safety Executive study

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Summary. During a 3-year period 229 patients in the U.K. with haematological disorders thought to be associated with occupational hazards were notified to the British Society for Haematology/Health and Safety Executive office. Most were suffering from malignant or premalignant states or aplastic anaemia. Benzene and ionizing radiation were the most common agents recorded. While no estimate can be made of the incidence of chemical or radiation induced disease at the present time, the data suggests that routine documentation of exposure at work is an important step in allowing more specifically focused studies.

The only specific haematological disorders recognized as occupational diseases under the Social Security (Prescribed Diseases) Regulations 1985 are poisoning by lead, arsenic, benzene and the nitro- or amino derivatives of benzene, and blood dyscrasias due to electromagnetic radiations or to ionizing particles. While statistics are available on the incidence of these conditions there is no certainty about their accuracy. Similarly, although the literature suggests that patients with haematological malignancies, including leukaemia, myelodysplasia and lymphoma, have an increased incidence of occupational chemical exposure compared to the rest of the population, no estimate of the number of distribution of occupationally related cases can be made. It is possible that in some cases the conditions mentioned above may be initially diagnosed as refractory anaemia or have other non-specific changes such as thrombocytopenia. In some instances only a persistent abnormality in the blood count may be observed which does not warrant a diagnosis of myelodysplasia.

A notification scheme for haematological disorders thought to be associated with exposure to an occupational hazard was proposed by the Health and Safety Executive (HSE) and agreed to by the British Society for Haematology (BSH) in order to gain an initial impression of the number and location of such cases and possibly to stimulate questions to be asked about the role of physical and chemical agents in the development of haematological disease. A pilot scheme was set up (A.J., C.G.) and ran from 1 January 1989 to 31 December 1991. All members of the BSH were circulated with a request to inform the co-ordinator (A.J.) of any case where they considered that occupational hazards may have played a part in the pathogenesis of the patient's disorder. Notification forms (Fig. 1), notes of guidance and reply paid envelopes were supplied to all BSH members for this purpose. During the 36 months of the study a total of 229 cases were entered into the database. An additional request for cytogenetic data was made for all cases of acute myeloid leukaemia or myelodysplastic syndrome. The results are summarized below. Full details of all reported cases have been circulated to BSH members and further copies can be obtained from C.G.

SUMMARY OF REPORTED CASES

Patients

201 men and 28 women, of whom 26 were over 75 years old and four were less than 22 years old. Most had first attended hospital since 1989 but a few had been diagnosed during the previous 10 years. 30 patients had been exposed to the

* Deceased.

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suspected haz … ad for less than a year before diagnosis, but many had se… …ral decades of exposure. Most patients were seen either … …thin a year of exposure ceasing or while exposure still … …sisted.

Primary diagn…	sis	Cases
Aplast…		22
Myelo…	anaemia	49
Myelo…	splastic syndrome	17
Acute…	obliterative disease	33
Acute…	ryeloid leukaemia	12
Chron…	mphocytic leukaemia	17
Chron…	granulocytic leukaemia	25
Myelo…	lymphocytic leukaemia	21
Hairy…	idiosyncratic macroglobulinaemia	5
Lymp…	ell leukaemia	23
	ama	5
Lead…	isoning	1
Acute…	haemolytic anaemia	

Occupation

It is difficu… …o provide a precise classification of industrie… and occup… …ns but those most commonly indicated wer… the chemi… industry (30 cases), the nuclear industry (1… cases), th… rmed forces (15 cases), the National Healt… Service (1… …ases), rubber manufacture (11 cases) and th… petrochei… …industry (10 cases). Other industries noted i…

small numbers of cases included some associated with can… and road transport and various specialized forms of chemical manufacture and engineering. It is, perhaps, of interest to note that nine radiologists or radiographers, five nurses and a medical laboratory scientific officer were identified. Amongst hazards reported having occurred in the armed forces, three patients had been present at nuclear weapons testing.

Benzene was the most commonly notified chemical thought to be implicated in the pathogenesis of the patient's disease (34 cases) and ionizing radiation was implicated in 11 cases. Other substances noted in more than occasional cases include paints and lacquers (16 cases), trichlorethylene and various other solvents.

Cytogenetics

An attempt was made to see whether the cytogenetic abnormalities found in patients with AML or MDS reported to the database differed significantly from those found in unselected cases. Only 31 patients with MDS and 25 patients with AML had reports available on satisfactory preparations. These were compared with 47 reports from consecutive newly diagnosed cases of MDS and 34 reports from newly diagnosed cases of AML at the University Hospital of Wales (A. D. White). The numbers are too small for adequate analysis but no difference between notified and control cases was seen with regard to the incidence of abnormal karyotypes, complex karyotypes or abnormalities affecting chromosomes 5 or 7.

HSE/BSH HAEMATOL… …CAL DISORDERS DATABANK

Ph… …se in CAPITALS

HOSPITAL

DATE ATTENDED

HAEMATOLOGIST

CASE RECORD NUMBER

5. DATE OF BIRTH

6. MALE (M) FEMALE (F)

7. DI… GN… S…

A.

B

8.1 OCCUPATION

8.2 INDUSTRY

8.3 AGENTS EXPOSED TO

A)

B)

C)

D)

8.4 DURATION OF … POSURE

Years … Months …

Weeks … Days …

TIME SINCE EXPOSURE

Years … Months …

Weeks … Days …

8.6 SUSPECT (S) DEFINITE (D)

9. ANY COMMENTS ?

Fig 1. Notification … …te … used by collaborating haematologists.

DISCUSSION

It is of some interest that almost all the disorders reported to the BSH/HSE database were of haematological malignancy, premalignancy or aplastic anaemia, the main exception being five cases of lead poisoning. It does not appear to be uncommon for a consultant haematologist to be of the opinion that occupational exposure to an injurious agent is a significant factor in causing the patient's disease. However, the data reported here cannot be considered definitive. There is considerable geographical variation in reporting. While 22 notifications came from two hospitals in Manchester and 10 from one hospital in Frimley, there was a striking absence of notifications from some areas. In addition, no attempt was made to validate diagnoses or to confirm the information provided by patients regarding their occupational background. Even with total reporting, consultants' opinions regarding the significance of particular hazards can only be subjective. A number of collaborating haematologists have wondered whether they are simply confirming known associations but this has in fact been a useful part of the exercise.

The value of this study has been to confirm that there is a sufficient area of concern to warrant more focused investigations of the importance of occupational factors in the aetiology of haematological malignancy. These are likely to be cohort studies such as previously described for benzene (Rinsky *et al*, 1987) or case control studies such as recently carried out for myelodysplasia (Farrow *et al*, 1989). The numerous enquiries, both by letter and by telephone, during the course of this study have highlighted the need for an easily accessible source of information about both household and industrial chemicals to help haematologists evaluate the importance of these in their patients' histories. The recording of lifetime work history and hobbies for all patients with malignant or premalignant bone marrow disorders would be a useful addition to our current routines and might identify injurious substances which only rarely cause haematological disease.

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The data described in this report were collected to further the understanding of the extent of work related ill health. The opinions expressed are those of the authors. They are not therefore a statement of HSE policy.

REFERENCES

Farrow, A., Jacobs, A. & West, R. (1989) Myelodysplasia: chemical exposure and other environmental factors. *Leukaemia*, **3**, 35–39.

Rinsky, R., Smith, A., Hornung, R., Filloon, T., Young, R., Okun, A. & Landrigan, P. (1987) Benzene and leukemia. An epidemiologic risk assessment. *New England Journal of Medicine*, **316**, 1044