The Health Watch Case–Control Study of Leukemia and Benzene

The Story So Far

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ABSTRACT: A case–control study nested in the Health Watch cohort of petroleum industry workers, investigated whether the excess of lymphohematopoetic cancers, identified among male members of the Health Watch cohort, was associated with benzene exposure. Cases of non-Hodgkin's lymphoma ($n = 31$), multiple myeloma ($n = 15$), and leukemia ($n = 33$) were identified between 1981 and 1999. Cases were age-matched to five controls. Exposure was retrospectively estimated for each occupational history using an algorithm in a relational database. Benzene exposure measurements, supplied by Australian petroleum companies, were used to estimate exposure for specific tasks. The tasks carried out within the job, the products handled, and the technology used, were identified from interviews with contemporary colleagues. More than half of the subjects started work after 1965 and had an average exposure period of 20 years. Exposure was low, 85% of the cumulative exposure estimates were <10 ppm years. Matched analyses showed that non-Hodgkin's lymphoma and multiple myeloma were not associated with benzene exposure. Leukemia risk, however, was significantly increased for the subjects with greater than 16 ppm years cumulative exposure, odds ratio (OR) 51.9 (5.6–477) or with greater than 0.8 ppm intensity of highest exposed job. Cumulative exposures were similar to those found in comparable studies. The inclusion of occasional high exposures, for example, as a result of spillages, reduced the ORs, when the exposure was treated as either a continuous or a categorical variable. Our data demonstrate a strong association between leukemia and modest benzene exposure. The choice of cut-point and reference group has a marked effect on the ORs, but does not change the overall conclusions.

KEYWORDS: case–control study; exposure; benzene; leukemia; non-Hodgkin's lymphoma; multiple myeloma; petroleum industry

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Ann. N.Y. Acad. Sci. 1076: 80–89 (2006). **© 2006 New York Academy of Sciences.**
doi: 10.1196/annals.1371.024

HEALTH WATCH COHORT

Health Watch is a prospective cohort study of employees in the Australian petroleum industry who have worked for more than 5 years. It compares the mortality and cancer incidence of the cohort with that of the Australian population. The cohort includes employees from offices, upstream extraction and processing sites, refineries, terminals, and airports all over Australia. The cohort includes about 18,000 employees of which some 1300 are women.

Health Watch commenced in 1981 at Melbourne University, with a face-to-face survey and this survey was repeated in 1986, 1991, 1996, and 1999. At the surveys, subjects provided demographic details, health status information, smoking, and alcohol data and details of their work history. In 1999 the cohort was closed to further recruitment and transferred to Adelaide University. In 2005 it was moved to Monash University.

About 95% of eligible employees have participated.[1] Subjects were actively followed and matched with the Australian national death and cancer registries. The standardized mortality ratio (SMR) for men has remained stable over 25 years at about 0.7.[1] However, an excess incidence of lymphohematopoietic cancers (LH cancers) was identified among the male members of the cohort in 1997.[2] The LH cancers included leukemia, non-Hodgkin's lymphoma (NHL), and multiple myeloma (MM). The observed excess of these cancers has reduced over the years (TABLE 1), but a nested case–control study was set up in 1995 to investigate whether the excess was related to occupational benzene exposure.

Benzene has been designated a class 1 carcinogen by International Agency for Research or Cancer (IARC) because of its leukemogenic properties.[3] It is present as a small percentage of crude oil, gasoline, and at many stages in the refining process.[4] Historically much higher percentages of benzene were handled at some petroleum industry sites in Australia. One or two refineries used BTX (benzene/toluene/xylene mixture) a byproduct of coke operations, which contains 70% benzene. Some refineries and terminals also used benzene as a feedstock or drummed benzene itself. At most sites, the consequent exposures were brief, intermittent, applied to relatively few employees, and ceased around 1975. One refinery used benzene as a feedstock until relatively recently, taking delivery of benzene by tanker a few times a year.

TABLE 1. Cancer incidence for males reported over time in the Health Watch cohort

Male SIR	7th Report 1987[2]	8th Report 1988–89[21]	9th Report 1992[22]	10th Report 1998[23]	11th Report 2000[24]	12th Report 2005[1]
Leukemia	2.0(0.6–4.6)	3.4(1.7–5.9)	2.8(1.7–4.5)	2.0(1.3–2.9)	1.5(1.0–2.2)	1.1(0.7–1.5)
NHL	1.1(−)	1.7(0.8–3.1)	1.3(0.7–2.2)	0.9(0.6–1.3)	1.0(0.7–1.4)	0.9(0.7–1.2)
MM	1.1(−)	2.2(0.6–5.6)	1.8(0.7–3.9)	1.9(1.0–3.3)	1.7(0.9–2.8)	1.1(0.6–1.8)

Standardized by age, sex, and calendar period of follow-up. SIR: standardized incidence ratio.

CASE–CONTROL STUDY METHODS

A nested case–control study was carried out in which each case was individually matched on age and sex to five controls from the cohort, drawn with replacement.[5] Cases were defined as male members of the Health Watch cohort, who had:

- first diagnosis of LH cancer after entering the Health Watch cohort; and
- diagnosis confirmed by pathology report, cancer registration, letter from medical practitioner, or death certificate; and
- had reported LH cancer to Health Watch either by self or by family, unless they were lost to contact by Health Watch, or were deceased.

There were 31 NHL, 15 MM, and 33 leukemia cases meeting these criteria identified in the cohort between 1981 and 1999.

Exposure to benzene was estimated for individual subjects based on their job history, to which an algorithm was applied, which used Australian exposure data provided by participating oil companies.[5–7] A job history was prepared for each subject, based on data that had been collected, largely prospectively, from the four cohort interviews that took place between 1980 and 1999.[5] The job histories were complete for 98% of subjects ($n = 494$), and were then checked by the employing company against their records. We used each subject's job history to identify which of several standard structured job-specific questionnaires was appropriate for each job. A contemporary colleague or colleagues were then interviewed about each job using the appropriate job-specific questionnaire, identifying the time spent on each of the tasks carried out, the technology used, and the products handled, for example, petrol, diesel, benzene. The interviews were carried out by one of the two experienced occupational hygienists, blind as to case status.

The basic tasks in refineries and terminals have not changed greatly over time, although the technology used and the frequency with which they are carried out, have changed. We collected information about the task frequency and technology change at the site visits and allowed for this in the exposure assessment. We did this by, for example, allocating a longer time for tank farm operators doing dipping in the 1950s because of more frequent excise checks.

We collated Australian personal benzene exposure monitoring data provided by participating companies and calculated the arithmetic mean exposure for each task or job represented. This included more than 3870 individual benzene measurements. If more than one technology could have been used, for example, top and bottom loading for road tankers, separate base estimates (BEs) were calculated for each technology. The BE was then allocated to individuals carrying out that task in a relational database. Few or no local data were available for some short-term tasks and data for these were sought from the

literature. The BEs were validated against relevant exposure data identified from the literature.[8]

Cumulative exposure was estimated using a task-based algorithm in a relational database. The algorithm was similar to that used in other petroleum industry studies,[9,10] it took into account: the time spent on different tasks with a job, the proportion of time spent handling each product within the task, the percentage of benzene in the product handled, the technology used for the task, and the years spent on each job.[6] If the BE was applied to a task with different technology, a multiplier was used to scale the exposure, for example, the BE for top loading was attributed to a driver who used top splash loading and a factor of three was applied to increase the exposure estimate. Some of the multipliers were derived from the literature; others were agreed by a panel of local industry occupational hygienists. These expert-derived multipliers were used for fewer than 160 tasks from a total of 3457 tasks assessed.

These data were used to estimate for each subject, the cumulative exposure to benzene in ppm years, and to identify the intensity of the most highly exposed job in ppm (cumulative exposure divided by years spent on that job). The subjects were categorized by cumulative exposure in ppm years and intensity of exposure in ppm.

We also identified jobs where there may have been infrequent high exposure events (HEEs) as a result of spillages or work practices that are no longer carried out and so would not be represented in the exposure data used for the BEs. We estimated the exposure that might have resulted from these HEEs and added them to the mean exposure for that subject.[7] Odds ratios (ORs) were then recalculated.

ORs for leukemia, NHL, and MM were calculated from conditional logistic regression using Stata.

CASE–CONTROL STUDY FINDINGS

The cases and controls were well matched. Subjects worked in the industry for an average of 20 years (range 4 years to 42 years), the majority started employment after 1965.[5] Exposure was low, the estimated lifetime cumulative benzene exposures ranged from 0.005 to 57.3 ppm years, with a mean of 4.9 ppm years. Eighty-five percent of the cumulative exposure estimates were <10 ppm years. For cases the mean exposure was 10.6 ppm years, for controls it was 3.86 ppm years.[5] The addition of occasional high exposures, increased exposure for 25% of subjects but for most, the increase was less than 5% of total exposure (FIG. 1).[7]

In matched analyses, NHL and MM were not associated with benzene exposure.[5] However, the OR for leukemia increased with cumulative exposure when exposure was treated as a continuous variable in a matched analysis, OR 1.10 (95% CI: 1.04–1.16) per ppm year. When the HEEs were added to the cumulative exposure, the continuous variable OR was 1.03 (1.01–1.05).[7]

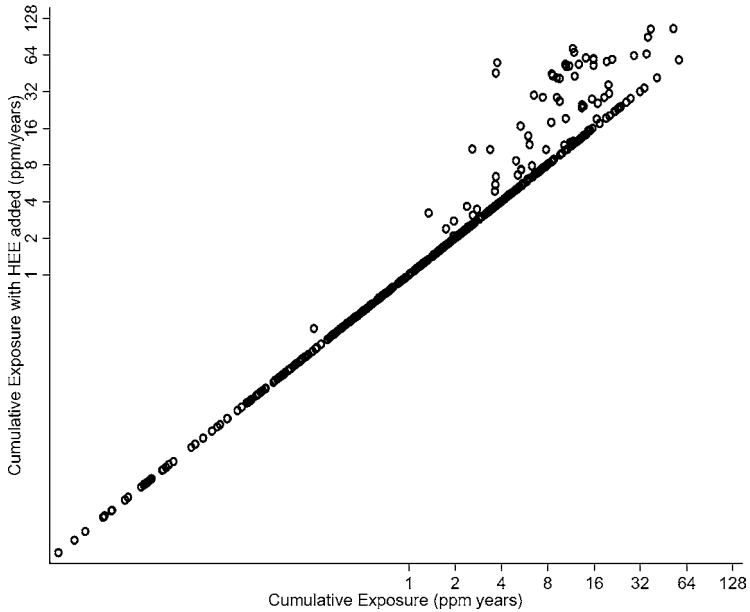

FIGURE 1. Comparison of benzene exposures with and without HEEs in a case–control study of petroleum workers.

The OR also increased with increasing cumulative exposure when exposure was treated as a categorical variable. The OR for the most highly exposed group, including the 7 cases with >16 ppm years cumulative exposure, was 98 (8.8–1090) when compared to those with less than 0.5 ppm years exposure to benzene.[5] A later reanalysis was carried out because it was recognized that the original reference group was small and because the precision of the exposure estimation process made it difficult to justify separating those with less than 0.5 ppm years from those with 0.5–1 ppm years exposure. Combining the two lowest exposure groups made a larger and more stable reference group containing the nine lowest exposed case sets. When the highest exposed group was compared to this new grouping the OR was 51.9 (5.6–477)[11] without HEEs and OR 7.79 (2.34–25.89) with HEEs included.[7] The reduction in ORs when the HEEs are added is probably because the leukemia risk is associated with higher exposures and hence the risk per ppm year was reduced.

We also examined risk by intensity and found that the risk was increased for subjects whose highest intensity job was greater than 0.8 ppm (exposure group >0.8–1.6 ppm, OR 6.34 [1.54–26.18]; exposure group >1.6–3.2 ppm, OR 5.58 [1.00–31.04]). However, exposure intensity and cumulative exposure are highly correlated and neither goodness-of-fit statistics nor the stepwise conditional logistic regression algorithm allowed us to distinguish which was the better predictor of risk.[5]

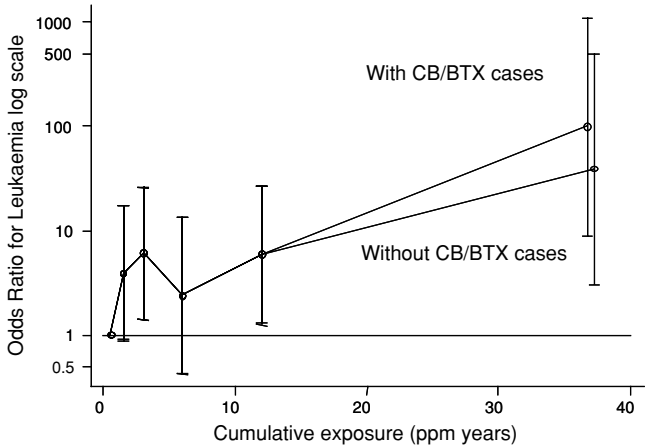

FIGURE 2. Conditional (fixed-effects) logistic regression, leukemia by cumulative exposure to benzene (ppm-years) with and without cases exposed to CB/BTX.

Risk of leukemia was strongly associated with exposure to 100% benzene (concentrated benzene or CB) or to BTX (FIG. 2). There was very little difference in the estimated cumulative exposures between the non-CB/BTX exposed group (mean 32.67 ppm years, median 29.85 ppm years) and the CB/BTX exposed group (mean 32.25 ppm years, median 28.08 ppm years). In fact, the mean and median exposure of the CB/BTX exposed group was slightly lower than that of the non-CB/BTX exposed group. The difference in OR for the highest exposure group with and without the CB/BTX exposed subjects is not of itself statistically significant. However, it is a large difference which, if real, cannot be explained in terms of differential exposures as estimated.

We showed that the risk of leukemia is associated with exposure within 15 years of diagnosis, the association with exposure prior to this period is weak.[12] There was no association between leukemia (with or without adjustment for cumulative benzene exposure) and date of starting work in industry, period of employment, or duration of work. After adjusting for cumulative benzene exposure, there was no association between leukemia and industry sector. We found no association between tobacco or alcohol consumption and leukemia. We consider it unlikely that there were other confounding exposures, for example, radiation or retroviruses.[5]

ORs were calculated for the leukemia subtypes acute nonlymphocytic leukemia (ANLL), chronic lymphocytic leukemia (CLL), and chronic myeloid leukemia (CML). Because there were relatively few cases of the leukemia subtypes, it was necessary to combine the three lowest exposure groups and the two highest exposure groups. The OR in the exposure group >8 ppm years were significantly raised only for ANLL OR 7.17 (1.27–40.44).[5] An association between ANLL and higher benzene exposures has been reported in several

papers previously.[13] The OR for CLL was high 4.52 (0.89–22.90) and this has not been reported previously. It should be noted that this is an incidence study, however, and of the 11 CLL cases in the study only 1 has died.[1] Most previous ORs in this field have been calculated on cancer deaths and an OR based on deaths might well have a different result for this disease.

CRITICISMS OF THE STUDY

Three main criticisms that have been made of the study are:

- that there was recall bias in the exposure assessments[14];
- that the exposure estimates were too low; and
- that there is diagnostic bias, more highly exposed subjects had their blood screened and thus more cases were identified in this group.[15]

With respect to the first criticism, our exposure estimates were very detailed, based on verified job histories and very largely on exposure data collected by the companies occupational hygienists. The job history was checked against company records and the interviews were case blind and structured, so the potential for recall bias is limited.[5] In addition, the lack of association between benzene exposure and NHL or MM, for whom the exposure data were collected in exactly the same way, suggests that recall bias is unlikely to explain association with leukemia.

As regards the extent of exposure, most of the exposures in this study are similar to those estimated in previous comparable studies although they do not range as high. However, the cohort is more recent than the Canadian[16] and UK[17] studies, which have subjects whose job histories go back to before 1920. Thus the use by subjects of older technologies, resulting in higher exposure, will be less frequent in our study. There is, for example, only one top splash loader in this study. Most of the BE values were derived from local exposure monitoring data and have been validated.[8] One of the most highly exposed jobs is that of drum filler, however, drum fillers in Australia have always been required to have open-sided rather than fully enclosed drum sheds as needed in Canada. The increased ventilation is likely to have resulted in lower exposure and hence it is expected that an Australian study would have a lower drum filling BE than the Canadian study. The association between blood changes and very low exposure to benzene (in terms of cumulative and intensity of exposure) has been confirmed by a recent study from China.[18]

With respect to diagnostic bias, no cases in the study were identified from routine blood screening carried out by petroleum companies. We understand in fact, that no cases of leukemia have ever been diagnosed on the basis of such routine blood screening in Australia.

THE FUTURE

There were too few cases for much detailed analysis by leukemia subtype or to establish clearly what time window is pertinent to exposure assessment for benzene with respect to increased risk of leukemia. In 2002, we drew up a proposal with colleagues who had conducted similar case–control studies in the UK and Canada to combine the data in the three studies to provide a more powerful analysis of these questions. We may also be able to analyze the data using the new WHO or REAL[19] criteria for grouping leukemias and NHL.

In 2004, the Australian, Canadian, and UK petroleum industry case–control studies were audited by a team from two external research institutions. They considered that "all of the studies had been well performed, there were no issues of subject selection, methods, or general data quality that were likely to have distorted their internal comparisons." They also concluded that "the evidence of an increased risk at higher exposures in Australia was convincing."[20] The audit team saw no obstacles to the data pooling and recommended that this should happen.

CONCLUSIONS

Our data demonstrate a strong association between leukemia and modest benzene exposure measured as cumulative exposure in ppm years or as intensity of exposure of the highest exposed job in ppm. The study was carried out to high standards and is robust to external audit.

Combining the data with that of the UK and Canadian studies would provide more power to establish the nature and extent of the association between benzene exposure, leukemia, and its subtypes and to derive more precision on time frames associated with this increased risk.

REFERENCES

1. GUN, R.T., P. RYAN, D. RODER, *et al* 2005. Health Watch twelfth report 2005. Department of Public Health, Adelaide University. Adelaide, Australia.
2. CHRISTIE, D.K., I. ROBINSON, I. GORDON, *et al*. 1988. Health Watch seventh annual report 1987. The University of Melbourne, Department of Community Medicine. Melbourne, Australia.
3. IARC. 1982. IARC Monograph on the evaluation of the carcinogenic risk of chemicals: some industrial chemicals and dyestuffs. IARC WHO. Lyon.
4. WALLACE, A. 1996. Environmental exposure to benzene: an update. Environ. Health Perspect. **104:** 1129–1136.
5. GLASS, D.C., C.N. GRAY, D.J. JOLLEY, *et al*. 2003. Leukemia risk associated with low level benzene exposure. Epidemiology **15:** 569–577.

6. GLASS, D.C., G.G. ADAMS, R.W. MANUELL, *et al*. 2000. Retrospective exposure assessment for benzene in the Australian Petroleum Industry. Ann. Occup. Hyg. **44:** 301–320.
7. GLASS, D.C., C.N. GRAY, D.J. JOLLEY, *et al*. 2005. Health Watch exposure estimates: do they underestimate benzene exposure? Chem. Biol. Interact. **154/155:** 23–32.
8. GLASS, D.C., C.N. GRAY, G.G. ADAMS, *et al*. 2001. Validation of exposure estimation for benzene in the Australian petroleum industry. Toxicol. Ind. Health **17:** 113–127.
9. ARMSTRONG, T.W., E.D. PEARLMAN, R.A. SCHNATTER, *et al*. 1996. Retrospective benzene and total hydrocarbon exposure assessment for a petroleum marketing and distribution worker epidemiology study. Am. Ind. Hyg. Assoc. J. **57:** 333–343.
10. LEWIS, S.J., G.M. BELL, N. CORDINGLEY, *et al*. 1997. Retrospective estimation of exposure to benzene in a leukaemia case-control study of petroleum marketing and distribution workers in the United Kingdom. Occup. Environ. Med. **54:** 167–175.
11. GLASS, D., C. GRAY, D. JOLLEY, *et al*. 2002. The Health Watch case control study. *In* Proceedings of a Conference Institute of Petroleum 19th October 2002. London.
12. GLASS, D., M.R. SIM, L. FRITSCHI, *et al*. 2004. Leukaemia risk and reevant benzene exposure period-Re: follow-up time an risk estimates. Am. J. Ind. Med. **47:** 481–489, 2002 [comment]. Am. J. Ind. Med. **45(2):** 222–???.
13. BERGSAGEL, D.E., O. WONG, P.L. BERGSAGEL, *et al*. 1999. Benzene and multiple myeloma: appraisal of the scientific evidence. Blood **94:** 1174–1182.
14. SCHNATTER, R.A. 2003. Benzene exposure and leukemia. Epidemiology **15:** 509.
15. GOLDSTEIN, B.D. 2003. Benzene exposure and leukemia. Epidemiology **15:** 509–510.
16. SCHNATTER, R.A., T.W. ARMSTRONG, M.J. NICOLICH, *et al*. 1996. Lymphohaemotopoietic malignancies and quantitative estimates of exposure to benzene in Canadian petroleum distribution workers. Occup. Environ. Med. **53:** 773–781.
17. RUSHTON, L. & H. ROMANIUK. 1997. A case-control study to investigate the risk of leukaemia associated with exposure to benzene in petroleum marketing and distribution workers in the United Kingdom. Occup. Environ. Med. **54:** 152–166.
18. LAN, Q., L. ZHANG, G. LI, *et al*. 2004. Hematoxicity in workers exposed to low levels of benzene. Science **306:** 1774–1776.
19. HARRIS, N., E. JAFFE, H. STEIN, *et al*. 1994. A revised European-American classification of lymphoid neoplasms: a proposal from the International Lymphoma Study Group. Blood **84:** 1361–1392.
20. MILLER, B., W. FRANSMAN, D. HEEDERIK, *et al*. 2005. A review of the data quality and comparability of case-control studies of low-level exposure to benzene in the Petroleum Industry. Institute of Occupational Medicine. Edinburgh. Report No.: TM/05/04.
21. CHRISTIE, D., K. ROBINSON, I. GORDON, *et al*. 1990. Health Watch eighth annual report 1988-9. The University of Melbourne, Department of Community Medicine. Melbourne, Australia.
22. BISBY, J.A. & G.G. ADAMS. 1993. Health Watch ninth report 1992. The University of Melbourne, Department of Public Health and Community Medicine. Melbourne, Australia.

23. BISBY, J.A. & G.G. ADAMS. 1999. Health Watch tenth report 1998. The University of Melbourne, Department of Public Health and Community Medicine. Melbourne, Australia.
24. GUN, R.T., L. PILOTTO, P. RYAN, *et al*. 2000. Health Watch eleventh report 2000. Department of Public Health, Adelaide University. Adelaide, Australia.

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