Myelodysplastic syndromes (MDS) are a group of hematopoietic neoplasms manifest as cytopenia, and which frequently progress to leukemia. Certain agents, primarily alkylating drugs used as chemotherapy for various malignancies, can induce cytogenetic and chromosomal damage, thereby causing the MDS/leukemia progression. Ethylene oxide (EtO), an alkylating agent and suspected human leukemogen, may cause leukemia via a similar mechanism, but epidemiological studies thus far have been inconclusive. EtO exposure in association with MDS has not been previously reported. The case of an MDS/leukemia patient with an occupational history of EtO exposure is presented, the syndrome of MDS/leukemia is discussed, and cytotoxic properties of EtO are reviewed. The possible impact of this case on the question of whether EtO is a human leukemogen is discussed.
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I. INTRODUCTION

Ethylene oxide (EtO), a gas at temperatures above 10.7 degrees C (51.3 degrees F), is a colorless, flammable, mucous membrane irritant, a highly reactive epoxide and alkylating agent. It is a known animal carcinogen suspected of being a human carcinogen as well. EtO is a potent biocide and is used in sterilizing medical equipment and supplies which cannot be steam sterilized without being damaged by the process. Workers are exposed to EtO in the gas sterilization setting as well as in the commercial production of EtO.

Three cohorts of Swedish workers exposed to EtO were found to have a greater than expected incidence of leukemia in reports published in 1979 and 1986 (20, 21, 22). However, subsequent studies have largely failed to confirm these findings and the question of whether such exposures result in human malignancy remains unresolved. Acute leukemia known to be caused by certain chemical exposures are frequently preceded by a myelodysplastic syndrome. The following case of an EtO-exposed worker who was diagnosed with myelodysplastic syndrome and who later developed acute nonlymphocytic leukemia has characteristics which may add to
the accumulation of evidence for a causative role of ME0.
II. CASE REPORT

A. Clinical History

The occupational medicine service at Duke University Medical Center was consulted in the case of a 56 year old American Indian male with a 17 year history of intermittent occupational exposure to EtO, who developed a myelodysplastic syndrome (MDS) and, later, acute nonlymphocytic leukemia (ANLL).

The patient's initial complaints began in January, 1992, with an appearance of white patches on his tongue and an onset of pruritus involving both forearms. There was no associated rash. He sought advice from his family physician on 1/31/92. Evaluation included a complete blood count (CBC) which showed leukopenia, thrombocytopenia with giant platelets, and a mild anemia (WBC count 2,200/cubic mm, platelets 73,000/cubic mm, hemoglobin 13.9 g/dl and hematocrit 40.8%). He had been taking phenytoin for 2 1/2 years for a stuttering condition; the phenytoin was discontinued and the presumed oral thrush treated with nystatin solution. The CBC was repeated on 2/17/92 and was essentially unchanged.
The patient was referred to a hematologist for further evaluation. An extensive workup on 2/26/92 showed an unchanged CBC and a hypercellular, nondiagnostic iliac crest bone marrow aspirate and biopsy. Results of cytogenetic studies on the bone marrow sample were returned several weeks later showing a partial chromosome deletion (5q-) in one metaphase out of 30 metaphases. This suggested an abnormal clone, but such a diagnosis requires at least two cells having the same abnormality. Repeat bone marrow aspirate and biopsy on 6/29/92 showed 4.3% blasts, megaloblastic and dysplastic erythrocytic maturation, impaired hemoglobinization and decreased granulopoiesis. This was interpreted as being consistent with MDS the refractory anemia (RA) classification. Apparently, the cytogenetic study was not repeated. He was restricted from work at that time because of concerns that his neutropenia might render him susceptible to hospital-acquired infection. He was placed on granulocyte colony stimulating factor (G-CSF) therapy, administered subcutaneously twice weekly. Granulopoiesis responded well, with a normal, sustained maintenance of a normal WBC count. A six week trial of erythropoietin resulted in no improvement in RBC count or hemoglobin.

Despite an improved WBC count on G-CSF, his anemia worsened and remained refractory, requiring several single
unit transfusions of packed red blood cells (PRBCs) through the summer and fall of 1992. Symptoms continued to increase in variety and intensity despite the response to G-CSM, and included weakness, malaise, fatigue, nausea, dizziness, headaches and occasional low-grade fevers. His activity became quite limited due to rapid fatigue marked by shortness of breath, diaphoresis and weakness. Other symptoms included decreased erectile function, mild weight loss, and orthostatic dizziness. Anxiety and worry became marked, with a short attention span, poor concentration and a mild decrease in short-term memory.

The patient was evaluated by the occupational medicine service at Duke University Medical Center on 10/15/92. Medications at that time were G-CSM and astemizole, a histamine H1-receptor antagonist. He reported allergies to cephalexin (pruritis, rash) and sulfa medications. He smoked 1/2 packages of cigarettes per day for 15 years and quit 10 years ago. He used alcohol "a lot" while in the Navy; felt he was overdoing it, and quit drinking 10 years ago. He denied any known family history of hematologic diseases or cancer. He denied a history of eye, nose, throat, or bronchial irritation associated with the workplace.

The physical examination showed a large-framed male
with a ruddy facial complexion, appropriate affect and normal mental status. No cervical, supraclavicular or inguinal lymph nodes were palpable. The thyroid, liver and spleen were not palpable. Pulmonary, cardiovascular and neurological examination were normal. No dysarthria or dysphasia was noted.

His hematology evaluation continued on 11/5/92 with a bone marrow aspirate and biopsy showing a progression of disease with 6% blasts, marked hypercellularity (cellularity to fat ratio of 100:0), markedly increased erythropoiesis with impaired hemoglobinization, decreased plasma cells and megakaryocytes, and abnormal, decreased granulopoiesis. The histologic features were considered consistent with an evolving erythroleukemia. Cytogenetic analysis on this date, and on all subsequent studies, showed no chromosomal abnormalities. CBC showed 2,100 WBCs, 51,000 platelets and hemoglobin 10.2. Repeat aspirate and biopsy on 1/21/93 showed a similar picture with 8% blasts and appearance of myelofibrosis, and was interpreted as meeting the criteria of ANLL, M6 type (erythroleukemia).

The patient was admitted to the hospital by his hematologist in February, 1993, for remission-induction chemotherapy consisting of a combination course of idarubicin (3 days) and cytarabine (7 days). His four week
hospital course was difficult, with persistent fevers and a deep perivascular, perifollicular hemorrhagic rash presumed to be secondary to one or more of the antibiotics used for broad spectrum antimicrobial coverage. He received several units of PRBCs and platelets. Bone marrow aspirate and biopsy on 2/25/93 confirmed a massive kill-off, with no cells seen. He was able to return home on 3/10/93. By late March he reported feeling the best he's felt "in several years." Repeat bone marrow aspirate and biopsy on 3/25/93 showed regenerative, hypercellular, left-shifted marrow with less than 5% blasts, normal amount of erythropoiesis with megaloblastoid erythrocytic maturation and impaired hemoglobinization, increased granulopoiesis with abnormal maturation, with normal plasma cells and megakaryocytes, and no myelofibrosis. This was interpreted as suggestive of persistent MDS. CBC showed 4,500 WBCs, 193,000 platelets and a hemoglobin of 11.5. Because of the difficult hospital course, the patient and his hematologist scheduled consolidation chemotherapy to follow induction by two months (late May, 1993) rather than the more typical one month. The patient continues to be asymptomatic.

B. Occupational and Exposure History

The patient joined the Marines following an incomplete first year of college in the mid 1950s. His duty title was
Aviation Electrician. Following a two year active duty tour, he spent a brief time as a civilian, then joined the Navy. He was an Aviation Electrician for the following twenty years until retirement. His naval duties included maintenance and repair of aircraft instruments and electrical systems. This included essentially all electrical systems on the airplanes, as well as weapons control and delivery systems. He did not work on radar or microwave source systems. He feels his radiation exposure on his two carrier tours was minimal, as restrictions on use of radar on the carrier deck was extremely strict. All planes had to be pointing out to sea prior to engaging radar systems.

In 1977, following his Navy tour, he joined a hospital equipment manufacturing company as a service representative, where he worked until the spring of 1992. Although he worked in several locations in the southeastern United States, his job description and exposure history was essentially uniform through the 17 years he held this position. The locations included Statesboro, Georgia, for the five years prior to this evaluation; Nashville, Tennessee, for six years; Virginia Beach, Virginia, for three years; and Savannah, Georgia, for two years. His job description included installation, repair and preventive maintenance of hospital equipment manufactured by his
company. This consisted, in his case, almost exclusively of autoclave units. Ninety-five percent of these units were of the steam sterilization design, while about five percent used EtO. The gas mixture was 88% freon, 12% EtO.

Nearly all of the units were of intermediate size, 20 x 20 x 36-48"., built in with the door and controls flush with the wall. The maintenance room was behind the wall, and contained the greater part of the autoclave units, as well as EtO bottles and the individual boiler units for the steam autoclaves. This was the area where nearly all of his maintenance and repair work was done. Ventilation in this room in all the hospitals he serviced was modified as needed in the mid 1980s to ensure 10 air changes per hour.

He described his potential sources of EtO exposure: 1) When disconnecting the EtO line at the autoclave to replace the filter; 2) unplanned gas leaks from the cylinder and pressurized line; 3) an unknown potential for exposure to EtO from possible undetected leaks (none were documented) of the pressurized EtO systems of other manufacturers while carrying out his autoclave maintenance work; and 4) lack of knowledge prior to 1981 that EtO exposure presented a possible risk.
The filter changing procedure was done on each EtO autoclave every three months. This worked out to approximately one filter changing procedure per week. He gave a brief description of the procedure, which involved turning off the EtO pressure at the gas cylinders main valve, followed by venting the pressure in the line to a dedicated exhaust system vented to the outside. After closing the venting valve to prevent backflow into the room, he would disconnect the line at the autoclave and leave the room for ten to twenty minutes to allow EtO from the line to disperse. The length of the gas line was generally 10 to 15 feet (ranged up to 40 feet) with an inside diameter of 3/8". He used a freon detector which he identified as a TIF 5500 pump-style automatic halogen leak detector (TIF Instruments, Inc., Miami, FL) to test room air before re-entering to complete the maintenance work, which generally took 1-1/2 to 2 hours. The freon gas detection threshold of the sniffer is 3 ppm, which would correspond to an EtO level of 0.41 ppm with the 88:12 freon:EtO mixture.

The second source of exposure occurred infrequently, three to four times per year, when a leak developed somewhere in the freon/EtO system and he was sent to find and repair it. He would enter the room, turn off the freon/EtO gas at the tank and leave the room if the sniffer detected freon. He would zero the sniffer and complete the
job only when he could re-enter the room without setting off the sniffer. On all jobs, except in unusual instances where he had no access to freon, he would pressurize the system with freon-12 (not freon/EtO mix) and use the sniffer to locate the leak and fix it. Leaks were usually detected by a built-in alarm on the gas sterilizer units, tripped by a computerized monitor. The sterilization cycle was under a 5-6 psi positive pressure, usually lasting about 2 hours. The automatic system would periodically draw more pressurized gas from the supply system to maintain this pressure, and an alarm would sound when this occurred above a set volume of gas. When the leak occurred, not in the sterilizer, but somewhere along the supply system itself, there was no such alarm, and a leak was generally detected when the supply system reached a low pressure faster than usual. Leaks were usually caused by a deteriorated seal on one of the automatic valves in the sterilizer unit or, occasionally, in the gas supply system.

The third exposure scenario is theoretical. During the last two years of his employment, two of his contract hospitals acquired autoclaves which used 100% EtO. He did no maintenance or repair work on these, as they were manufactured by another company. His concern was that, due to the high odor threshold of EtO, he would not have been able to detect a leak when he was in the same room working
on his own company's autoclaves. One of these two hospitals had a non-specific area hydrocarbon sensor set at a threshold of 10 ppm. This monitor never went off during his presence. Although hospital personnel working in the central supply areas were periodically monitored for EtO exposure, those maintaining the sterilizers were not, nor was the air in the maintenance rooms ever monitored specifically for EtO levels to his knowledge.

Finally, the patient took no precautions whatsoever against exposure to EtO prior to 1981. He did not use the freon detector, nor was he concerned about breathing the gas during maintenance operations or fixing system leaks. He recalls frequently noting an ether-like odor, particularly following the purge cycle of the sterilizers and while fixing leaks. There is no information of what his exposures were in those early years of his employment other than the presumption that he was indeed detecting the odor of EtO. EtO has an odor threshold of 700 ppm (38). However, he denied symptoms of mucous membrane or respiratory tract irritation, which appear at concentrations as low as 200 ppm (38).

During his 17 years with this company he never wore a respirator. This was apparently never discussed. He did place an EtO personal exposure detector on his clothing for
a single eight hour shift once in 1991. This monitor was
developed and marketed by his company, and he actually acted
as a sales representative for this instrument. The
monitored shift included a routine filter changing operation
on a gas autoclave. The personal monitor showed an
undetectable level of exposure. The lower limit sensitivity
of this instrument was 0.08 ppm for an 8-hour time-weighted
average, but its accuracy and reliability is unknown.

The patient did not operate the sterilizers, although
he was frequently present while gas sterilizers were in
operation. He was occasionally present at the end of a gas
sterilization cycle when the sterilizer door was opened by a
hospital employee. At that time, he usually noted an odor
of ethylene glycol emanating from the open unit.

His work week was 40 hours, of which, on the average,
28 hours were spent in hospitals doing maintenance work, and
approximately 12 hours travel time. Nearly all of the in-
hospital time was spent working on autoclaves. Rarely, he
would be called upon to work on an operating room table or
light fixture manufactured by his company.
III. MYELODYSPLASTIC SYNDROMES

A. Primary and Secondary Myelodysplastic Syndromes

Myelodysplastic syndromes refer to a group of clonal hematologic disorders characterized by cytopenia resulting from ineffective hemopoiesis. Recognized for several decades, they were thought to progress to leukemia 50-70% of the time and were referred to by such names as "pre-leukemia," "smoldering leukemia" and "preleukemic acute leukemia." However, as they came to be diagnosed at less advanced stages, it became apparent that progression occurred much less often, about 19% of the time (range 5% to 48% depending on the subtype) (40).

It became apparent that specific morphologic characteristics were predictive of different outcomes, and the "French-American-British group" proposed a new classification system which has been accepted and is referred to as the FAB classification of MDS. This has been expanded from two subtypes to five: refractory anemia (RA), refractory anemia with ringed sideroblasts (RARS), refractory anemia with excess of blasts (RAEB), refractory anemia with excess blasts in transformation (RAEB-t), and...
chronic myelomonocytic leukemia (CMML) (31). RAEB-t has the highest rate of leukemic transformation (mean 48%) and shortest median survival (6 months), while RARS has the most favorable prognosis (5% and 49 months). RA has a median survival of 37 months (range 19-64 mo) and an 11% progression rate (Table 1) (40).

Incidence data on the U.S. population as a whole is not available, as tumor registries do not have a code for MDS (10). The best data currently available is from a 5 year study, from 1982 through 1986, of incidence in whites and hispanics on the upper Texas Gulf Coast (26). From this data it appears that MDS is primarily a disease of the elderly, the median age being 65 years, and less than 10% of cases occurring in patients younger than 50 years. Age specific incidence: 0.06/100,000 (20-29 yrs), 7.4/100,000 (60-69 yrs), 19.6/100,000 (70-79 yrs). Individuals with a history of cytotoxic drug therapy or exposure to ionizing radiation or leukemogenic chemicals may be younger (17).

Cases which arise without apparent cause (de novo, or primary, MDS) represent the majority of cases, and are thought to reflect a genetic predisposition (32). MDS in association with mutagenic agents felt to be causative it is referred to as therapy-related MDS, or secondary MDS (7).
B. Secondary MDS and Acute Leukemia

The pathogenesis of secondary MDS and acute leukemias (AL) occurring after treatment of a variety of malignancies with alkylating agents or radiation, and those associated with environmental exposures, has been extensively reviewed by Levine and Bloomfield (32). Median latencies from exposure to development of MDS/AL ranged from 4-5 years but the range included some as long as 25 years. In contrast to primary MDS, 55-84% of secondary MDS progresses to AL (32). The risk of developing secondary MDS/AL after chemotherapy is 10-17% at 4 to 6 years for nitrogen mustard-treated myeloma and 2-10% at 7 to 10 years for Hodgkin’s Disease (15).

The mechanism of cytotoxic agents in the genesis of secondary MDS/AL appears to involve alkylation of DNA. There is little disagreement that treatment with alkylating agents plays a causative role in MDS/AL. The alkylating cytotoxins implicated include the nitrogen mustards, chlorambucil, cyclophosphamides and melphalan. On the other hand, MDS/AL is not associated with use of the non-alkylating cytotoxic drugs: 5-fluorouracil, cytarabine, methotrexate, vincristine, vinblastine, dactinomycin, mitomycin, bleomycin, daunorubicin and doxorubicin (32).
Secondary MDS/AL appears to be a heterogenous disorder with a subset of cases having a latency between 30 and 60 months. These cases appear to have biological differences from those with longer and shorter latency periods. The 30 to 60 month latency subset is more likely to present with MDS prior to AL (88% vs 47%), to have an abnormal karyotype (100% vs 33%), and to be resistant to treatment. Most cases of secondary AL are ANLL, with a very small percentage being acute lymphatic leukemia (ALL) or chronic myelogenous leukemia (CML) (32).

C. The Clonal Nature of MDS/AL

Although the clone is not always apparent in cytogenetic studies, it is clear that essentially all MDS/AL are clonal in nature (37). Neoplastic transformation of a single hematopoietic precursor cell is followed after a variable time period by autonomous proliferation, interfering with normal hematopoiesis (34). The abnormal clone also appears to be the same from MDS through progression to AL, despite the occasional appearance of further chromosomal abnormalities (24). The mechanism is a primary defect of the DNA of one or more chromosomes of a hematopoietic cell, probably at the pluripotential stem cell level (7). That MDS and leukemia represent a progression of the same disease process is now better understood through
molecular genetic studies (42).

Cytogenetic findings have been reported in approximately 50% of patients with non-therapy related MDS (7). About one-half of these consist of 5q- (a loss of the long arm of chromosome 5), -5 (monosomy 5) and -7. In contrast, clonal chromosome abnormalities are found in 76-97% of cases of secondary MDS/AL, with 50-90% involving all or part of chromosomes 5 and/or 7 (7, 39).

The presence of clonal abnormalities visible on karyotype is closely associated with a worse prognosis. Survival with a normal karyotype is longer than with a single chromosomal abnormality, and survival with a single aberration is longer than with complex karyotypic abnormalities.
IV. ETHYLENE OXIDE

A. Characteristics and Uses

In the United States, EtO is produced by oxidation of ethylene by air or oxygen in the presence of silver oxide (a catalyst). An older method using ethylene chlorohydrin, was discontinued by 1980 (11). Its reactivity makes it an ideal intermediate in the production of a variety of commercially valuable chemicals such as ethylene glycol, glycol ethers and glycols, ethanolamines and surfactants. Though over 99% of commercially produced EtO is used in this setting, its reactive, explosive nature means most processes take place in tightly contained, automated systems where, barring malfunctions, worker exposure is minimal (30). As a biocide it is used in sterilization of medical equipment and materials, spices and cosmetics.

B. Occupational Exposures to Ethylene Oxide

The Occupational Safety and Health Administration’s (OSHA) permissible exposure limit (PEL) is 1 ppm as an 8 hour time-weighted average exposure (TWA). This represents a 1984 revision from the prior standard of 50 ppm. OSHA
also set an action level (AL) of 0.5 ppm as an 8 hour TWA and a 15 minute short term exposure limit (STEL) of 5 ppm (38). The National Institute of Occupational Safety and Health (NIOSH) recommends that exposures not exceed 0.1 ppm as an 8 hour TWA and 5 ppm as a 10 minute STEL (36). The American Conference of Governmental Industrial Hygienists (ACGIH) recommend a threshold limit value (TLV) of 1 ppm as an 8 hour TWA, and makes no recommendation regarding a STEL (1).

Workers in the sterilization process rooms of hospitals may be routinely exposed to high air concentrations of EtO. Industrial hygiene measurements of such exposures in recent years have varied from non-detectable to 7.8 ppm with short term exposures from 103 to 795 ppm (5, 6, 10 16).

C. Genotoxicity of Ethylene Oxide

EtO is a strong alkylating agent and an established mutagen. Mutagenic tests of EtO have shown gene mutations and chromosomal damage in a wide variety of plants, bacteria, fungi, insects, mammals, and humans (43). Adducts are formed via irreversible covalent binding to DNA bases and heme proteins. The central role of mutagenic DNA adducts in the genesis of various human malignancies, including leukemia, hepatocellular carcinoma, tobacco smoke...
related lung cancer and others, is well-established (15). The DNA alkylation reaction caused by EtO in vivo, causing mutations via base-pair substitution, yields N7-(2-hydroxyethyl)guanine (7-HCG) (9). It forms hydroxyethyl adducts to N-terminal valine (HOrtVal) in hemoglobin (16). EtO acts as a direct-acting mutagen via base-pair substitution in the Ames Salmonella assay, cultured mammalian cell assays (mouse lymphoma and V79 Chinese hamster lung cells), and in vivo (Drosophila melanogaster, via oral or inhalation administration) (2).

In male mice, heritable translocations were induced by intraperitoneal injection of 150 mg/kg EtO, and dose-related dominant lethal mutations resulted from EtO inhalation at levels of 225 ppm, 6 hours per day, 5 days per week, for either 2 or 11 weeks (13, 14).

Sister chromatid exchanges (SCE) in human and other primate peripheral leukocytes has been shown in several studies to correlate with occupational exposure to EtO, and that this relationship is dose-dependent (dose based on breathing zone exposure) (27, 44). A study of 34 hospital sterilization unit workers exposed to concentrations of EtO at or below the current OSHA time weighted average (TWA) of 1.0 ppm showed an increase in SCE (adjusted for smoking). Chromosomal abnormalities and micronuclei did not at appear.
EtO-hemoglobin adducts correlated highly \((p < 0.02)\) with frequency of SCE (33).

D. Carcinogenicity of Ethylene Oxide

EtO is classified 2A by the International Agency for Research on Cancer (IARC), a probable human carcinogen with "almost sufficient" evidence for human carcinogenicity (25).

E. Other Risk Factors

This patient has a relatively minimal 7 1/2 packyear smoking history, ceasing about 10 years ago. Smoking as a risk factor for the development of leukemia is plausible, as cigarette smoke contains benzene and radioactive compounds, both of which cause leukemia in humans (3). However, doses of both are low. The delivered dose of the main radioactive substance, polonium-210 is much lower than from food consumption, and contributes only a small percentage to marrow irradiation from background sources. The delivered dose of benzene from cigarette smoking, 10-100 ug per cigarette is low in comparison to the benzene PEL (10 ppm or 32 mg/m³)(1, 29). Epidemiological evidence of smoking as a cause of leukemia is weak (24, 29).
This patient was exposed to an unknown, but probably minimal, amount of microwave radiation (radar) while in the Navy. This has not been associated with an increase in leukemia among radar-exposed Navy personnel, nor is there an apparent mechanism of secondary MDS/AL as there is with ionizing radiation.
V. EPIDEMIOLOGY OF ETHYLENE OXIDE AND LEUKEMIA

In 1979, Hogstedt et al. reported two cases of leukemia among 240 Swedish factory workers exposed to EtO from an EtO/methyl formate sterilizing process (21). 77 of these workers were exposed during their entire work shift, while 163 were exposed while passing through a hallway where newly sterilized boxes were stored and where EtO concentration ranged from 2 to 70 ppm. The TWA EtO concentration for all workers was 20 ± 10 ppm. Only 0.2 cases were expected based on population norms (RR = 10.0).

Hogstedt et al. also, in 1979, reported an excess of neoplastic disease among 89 Swedish workers in an EtO production plant which used the older ethylene chlorohydrin method (22). Two cases of leukemia were diagnosed with 0.14 expected based on Swedish national average rates (RR = 14.3). Three cases of stomach cancer occurred with an expected 0.4 (RR = 7.5). The average TWA EtO concentration was 25 ppm but the odor threshold of 700 ppm was occasionally reached. These workers were also exposed to a number of other chemicals, including ethylene chlorohydrin, ethylene dichloride (1,2-dichloroethylene), chloroform and DDT. They were also reported to have monitored the EtO production process by tasting the chemical reaction product.
In 1986, Hogstedt reported updated incidence and mortality data from the two previous cohorts and added a third cohort, a group of 355 Swedish production process workers in an EtO production facility which used the newer, direct ethylene oxidation process (20). The third group had estimated 8 hour TWA EtO exposure ranging from 1 to 8 ppm during 1963-1976, and from 0.4 to 2 ppm during 1977-1982, with short term (several minute) exposures of 333 to 1000 ppm in some individuals. A total of 733 EtO-exposed workers was included in this report, with data starting in 1968, 1961 and 1964 for the three cohorts, respectively. A total of eight cases of leukemia occurred with an expected rate of 0.83 (RR = 9.6), and 6 cases of stomach cancer occurred with 0.65 expected RR = 9.2). Five of the six stomach cancer cases were from the second cohort, those EtO production workers using the ethylene chlorohydrin method. Of the leukemia cases, three were from the first cohort (0.14 expected; RR = 21.4), four from the second (0.52 expected; RR = 7.7) and one from the third (0.16 expected; RR = 6.2). Five were chronic leukemias, three acute (Table 2).

Morgan et al. studied 767 men with "potential" exposure to EtO (35). This retrospective cohort consisted of workers in an EtO-producing chemical plant between January, 1955 and
December, 1977, who had been employed there for at least five years. Apparently, EtO was the plant’s only product. There were 46 deaths with 80 expected; there were no deaths from leukemia. Standard mortality ratios were elevated, but not significant at the 5% level, for deaths from other cancers: pancreas (3; 4 expected; SMR = 4.1), bladder (1; SMR = 3.5), brain (2; SMR = 3.1) and Hodgkin’s disease (2; SMR = 6.3), and fewer than expected from all malignant neoplasms (11; SMR = 0.8). The small size of the cohort meant that only a large excess in leukemia would have been detected. The authors calculated that, based on the cohort sample size (13,969 person years of observation) a 10.5 fold or greater excess would have been detected at a 95% confidence level with a power of 80%. Estimated EtO exposures were extrapolated from a single industrial hygiene survey. The EtO reaction systems were out of doors, and enclosed in a sealed, automated system. Air sampling measurements in the reaction area detected "virtually no" EtO. The text does mention some questionable sources, indicating that concentrations at these sources were "less than 10 ppm." Readings at the tank car loading operations area were approximately 6,000 ppm, caused by a leak (which was subsequently stopped) around a slip-tube used to gauge the level of EtO in the tank.

Bisanti et al., in 1988, reported mortality figures for
a retrospective cohort of 1,971 male Italian chemical workers, covering the time period of 1938 to 1984 (19,000 person years) (4). No actual industrial hygiene or personal monitoring records were used. Italian law requires workers handling toxic gas to be licensed for each specific gas. The subjects were all chemical workers in two regions in northern Italy who had held a license to handle EtO for at least one year between 1938 and 1984. Some exposure of these workers to EtO was presumed; exposure levels were not estimated. The 76 total deaths were not significantly different from expected (82.6 expected, SMR = 0.9), but deaths from all cancers were (43 deaths; 23.9 expected; SMR = 1.8; p < 0.05). Deaths from lymphoma were increased (4; 0.5 expected; SMR = 8.4; p < 0.01). There were two leukemia deaths, more than expected, but not statistically significant (0.9 expected; SMR = 2.1; p > 0.05). There was no upward trend based on length of time the licenses were held.

A larger study was published by Gardner et al. in 1989 (12) of 2876 factory and hospital workers with direct exposure to EtO. No excess leukemia or stomach cancer were found. The period covered was from the early 1960s (depending upon the location) through 1987. Industrial hygiene records, kept since 1977, were used for exposure data. TWAs after 1977 were less than 5 ppm except for
surges up to several hundred ppm during the unloading of sterilizers (routine) and occasional operations problems at the manufacturing facilities. Prior to 1977 monitoring requirements, exposures were thought to have been much higher, and peak exposures were over the odor threshold. There were three leukemia deaths (2.09 expected; SMR = 1.4; ns) and 5 stomach cancers (5.95 expected; SMR = 0.8). All three leukemia deaths were within the factory subcohort, where 1.33 were expected (SMR = 2.2; ns). Total deaths were less than expected. There was a slight excess of all several malignancies but none were significant at the 5% level. This study was felt by the authors to suggest that then-current levels of EtO exposure posed no significant cancer risk.

A similar study was done in Germany by Kiesselbach et al. with 2658 male chemical plant workers, covering the period from 1928 to 1981, except that an internal matched control group was selected (28). No exposure measurements were taken, but subject selection criteria included having a record of working predominantly and exclusively with EtO apparatus for a minimum of 12 months. There were no statistically significant differences between the control and exposed groups in total mortality or mortality from malignancy (the trend was higher in the exposed group). There were no deaths from leukemia in the exposed group, and
Greenberg et al. reported in 1990 a large retrospective mortality study of 2174 male workers at a large U.S. chemical company from 1940 to 1978 (18). The subjects worked in a department which used or produced EtO. Comparison groups were the general U.S. population, the regional population, and a group of 26,965 unexposed males from the same plant. Three exposure groups were defined, high, medium and low. Large scale industrial hygiene monitoring was begun in 1976. EtO based chemical production areas averaged less than 1 ppm EtO for 8 hour TWAs, although the TWA was sometimes as high as 66 ppm. Since no data was available for the pre-1976 period, assignment to the relative exposure groups was based on estimated exposures based on production processes and changes in these over time. All cause mortality were less than expected for the exposed cohort (SMR = 0.8). There were seven leukemia deaths with 3.0 expected (SMR = 2.3; ns) and seven pancreatic cancer deaths with 4.1 expected (SMR = 1.7; ns). In the highest EtO exposure subcohort there were no leukemia deaths with 0.7 expected, and one pancreatic cancer death with 0.9 expected. Four of the leukemia deaths (0.7 expected; SMR = 5.7; p < 0.05) and six of the pancreatic carcinoma deaths were in men who worked in the ethylene chlorohydrin production department, where exposure to EtO
was estimated to have been low. Compared to the control group (no EtO exposure), the RR among the chlorohydrin workers for pancreatic carcinoma was 14.0, and for leukemia 8.7. Among those EtO workers not in the ethylene chlorohydrin subcohort, the leukemia SMR was 1.4 (3 observed/2.1 expected; ns). The authors noted from their review of the literature that ethylene chlorohydrin has not been found to be carcinogenic in rats or mice with gavage or dermal application and only inconsistently with subcutaneous injection.

Steenland et al. reported on the largest retrospective mortality study to date of EtO exposed workers. The cohort consisted of 18,254 EtO exposed workers at 14 different medical supply and spice manufacturing companies in the United States. Subjects averaged 4.9 years of exposure to EtO with an average of 16 years of follow-up. Time period studied varied with the plants with the first year ranging from 1943 to 1969, with personnel records review through 1987 for all subjects. Exposure monitoring began in the late 1970s and early 1980s; TWA EtO exposures averaged 4.3 ppm for sterilizer operators and 2.0 ppm for other workers. Mortality from all causes was decreased in the cohort as a whole. There was no increase in deaths from leukemia (13; SMR = 1.0), Hodgkin's disease (4; SMR = 1.1), all hematopoietic cancers (36; SMR = 1.1) stomach cancer (11;
SMR = 0.9) or pancreatic cancer (16; SMR = 0.9). Among men, however, there was more than expected mortality from all hematopoietic cancers (27; SMR = 1.6; p < 0.05). There was a significant trend among both men and women of increased risk of death from hematopoietic cancer with increasing time since the first exposure to EtO.
VI. DISCUSSION

While it is clear that patients treated with alkylating cytotoxic drugs for certain malignancies are at greatly increased risk of developing secondary MDS/AL, the role of occupational exposure to EtO, an alkylating agent which has been clearly shown to be carcinogenic in animals, and mutagenic and genotoxic in animals and humans, is not clear.

The reports of greatly increased cases of leukemia and stomach cancer in EtO workers in Sweden, the suggestive animal and human data, and the model of human carcinogenicity by DNA-alkylating agents stimulated not only a number of retrospective mortality studies by other investigators, but also regulatory changes decreasing EtO exposure limits and an upgrading of the IARC human carcinogenicity classification. However, the question of human carcinogenicity is still uncertain (28).

The subsequent studies of EtO-exposed workers have not verified a human cancer risk. Limitations of some of these studies include exposures to other chemicals, some of which are themselves suspected human carcinogens (Hogstedt, et al., 1979 studies and Greenberg, et al.) (Table 3). The excess of stomach cancer in the second Hogstedt, et al. cohort may have been related to oral intake of a reaction
mixture containing, among other chemicals, the suspected carcinogen ethylene dichloride (Table 3) (18). One of these studies (Greenberg et al.) showed no statistically significant increase in leukemia for the entire cohort but did show an increase in leukemia among workers exposed to ethylene chlorohydrin, a group with the lowest estimated EtO exposures. The author noted, however, that there is little evidence for carcinogenicity of ethylene chlorohydrin, and exposure to other chemicals was quite limited. Other study limitations include possible insufficient length of exposure (Steenland, et al.) and inadequate to nonexistent exposure measurements or estimates (particularly Bisanti, et al. and Kiellelbach, et al.). Table 4 summarizes the results of these studies.

The Hogstadt, et al. cohorts may have been exposed to EtO air concentrations one to two orders of magnitude higher than in some of the negative and inconclusive studies, most of which appeared to have exposures, at least in recent years, well below 10 ppm. Hertz-Picciotto and Neutra extrapolated dose-response data from animal studies to "predict" leukemia deaths in some of these human cohorts (19). Despite some limitations which the authors discuss, the predictions were similar to, and consistent with, the EtO-related leukemia incidence and mortality data from two of the studies (20, 35).
The subject of this case study presented with MDS/AL which is quite suggestive of a secondary process, similar to that seen in secondary MDS/AL following treatment with cytotoxic drugs. The presence of a preleukemic stage, the trilineage cell involvement on the bone marrow biopsy, and the aggressive transformation over less than a year from a normal bone marrow biopsy to frank acute leukemia are strongly suggestive of a secondary, rather than a primary, MDS/AL process. The early leukopenia preceding anemia is also characteristic of secondary MDS/AL.
VII. CONCLUSIONS

This patient had prior high exposures to EtO, an alkylating agent which is a known animal carcinogen, during a time frame which corresponds to the known range of latency periods associated with secondary MDS/AL. His lack of awareness, prior to the early and mid 1980s that EtO was a potential health hazard, resulted in a lack of respiratory exposure precautions prior to that period. Exposures during that 1977 to mid 1980s period were frequently above the odor threshold (700 ppm) of the EtO, certainly in the same order of magnitude as the workers in the Hogstedt, et al. cohorts which did have extremely high relative risk for development of leukemia.

EtO is known to cause, in humans, gene mutations and chromosomal damage by a mechanism (DNA adducts) similar to the known carcinogenic mechanism of cytotoxic alkylating agents. Such agents are known to cause an MDS/AL progression indistinguishable from the clinical course taken by this patient. His limited smoking history is considered an unlikely factor in the etiology of his MDS/AL. Nor is his occupational exposure to microwave (radar) radiation considered a factor. Although it is not be possible establish certain causality, it appears highly probable that
this patient's malignant hematopoietic transformation was caused by occupational exposure to EtO.

This gives support to the hypothesis that the leukemogenic risk of occupational EtO exposure is related to the cytotoxic drug model of human leukemogenesis, and that a dose-response relationship, rather than a cluster of leukemia with an unrelated cause, would explain the negative and inconclusive studies which followed the original Swedish cohorts published by Hogstedt, et al.

The impending phasing out of freon production and use will soon require a change from the current EtO:freon gaseous mixture used for most current hospital equipment sterilization procedures. A new EtO:inert gas mixture has been tested at Duke University Medical Center, as well as at several other centers (8) and will be available on the market as early as the summer or fall of 1993. EtO will continue to be the only acceptable alternative to steam sterilization for the foreseeable future. It does, in all probability, pose a significant carcinogen hazard at exposure levels above the current exposure limits. Attention to personnel practices, equipment conditions, exposure monitoring and ventilation specifications should continue to be strongly emphasized to minimize workplace exposure and the risk of secondary hematopoietic
malignancies.
### APPENDIX

**TABLE 1. Myelodysplastic Syndromes.**

<table>
<thead>
<tr>
<th>Subtype</th>
<th>%</th>
<th>Median survival (mo) / Range</th>
<th>% Progression / Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>RA</td>
<td>25</td>
<td>37 / 19-64</td>
<td>11 / 0-20</td>
</tr>
<tr>
<td>RARS</td>
<td>18</td>
<td>49 / 21-76</td>
<td>5 / 2-15</td>
</tr>
<tr>
<td>RAEB</td>
<td>28</td>
<td>9 / 7-15</td>
<td>23 / 11-50</td>
</tr>
<tr>
<td>RAEB-t</td>
<td>12</td>
<td>6 / 5-12</td>
<td>48 / 11-75</td>
</tr>
<tr>
<td>CMML</td>
<td>17</td>
<td>22 / 8-60+</td>
<td>20 / 3-55</td>
</tr>
</tbody>
</table>

All patients 19
APPENDIX

TABLE 2. Types of Leukemia Observed.
(Hogstedt et al, Reference R)

<table>
<thead>
<tr>
<th>Cohort</th>
<th>Type of Leukemia</th>
<th>Year of Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Chronic myelogenous</td>
<td>1972</td>
</tr>
<tr>
<td></td>
<td>Acute myelogenous</td>
<td>1977</td>
</tr>
<tr>
<td></td>
<td>Acute blast cell leukemia*</td>
<td>1979</td>
</tr>
<tr>
<td>2</td>
<td>Chronic lymphocytic</td>
<td>1961</td>
</tr>
<tr>
<td></td>
<td>Acute myelogenous</td>
<td>1971</td>
</tr>
<tr>
<td></td>
<td>Chronic lymphocytic</td>
<td>1972</td>
</tr>
<tr>
<td></td>
<td>Chronic myelogenous</td>
<td>1979</td>
</tr>
<tr>
<td>3</td>
<td>Chronic myelogenous</td>
<td>1976</td>
</tr>
</tbody>
</table>

*Unable to classify as myelogenous or lymphocytic
APPENDIX

TABLE 3. Classification of Carcinogenicity

<table>
<thead>
<tr>
<th>Compound</th>
<th>Animal</th>
<th>Human</th>
<th>In Vitro</th>
<th>IARC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethylene oxide</td>
<td>Adequate</td>
<td>Inadequate</td>
<td>Sufficient</td>
<td>2A</td>
</tr>
<tr>
<td>Formaldehyde</td>
<td>Adequate</td>
<td>Inadequate</td>
<td>Sufficient</td>
<td>2B</td>
</tr>
<tr>
<td>DDT</td>
<td>Adequate</td>
<td>Inadequate</td>
<td>Inadequate</td>
<td>2B</td>
</tr>
<tr>
<td>Ethylene dichloride</td>
<td>Adequate</td>
<td>Inadequate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chloroform</td>
<td>Adequate</td>
<td>Inadequate</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
APPENDIX: TABLE 4. Summary of Ethylene Oxide Cohort Studies.

<table>
<thead>
<tr>
<th>Author</th>
<th>Exposure (ppm)</th>
<th># Leukemia Cases</th>
<th>Other Ca</th>
<th>Other Exposure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hogstedt 1979a</td>
<td>20 ± 10 Peaks to 70</td>
<td>2 (10)</td>
<td>Methyl formate</td>
<td></td>
</tr>
<tr>
<td>Hogstedt 1979b</td>
<td>25 Peaks &gt; 700</td>
<td>2 (14)</td>
<td>Stomach Eth. dichloride</td>
<td>Eth chlorohydrin chloroform, DDT</td>
</tr>
<tr>
<td>Hogstedt 1986; 1+</td>
<td>20</td>
<td>3 (21.4)</td>
<td>Stomach</td>
<td></td>
</tr>
<tr>
<td></td>
<td>25</td>
<td>4 (7.7)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>0.4 - 8</td>
<td>1 (6.2)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Peaks 333 - 1000</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Morgan 1981</td>
<td>&lt; 10**</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bisanti 1988</td>
<td>?</td>
<td>2 (ns)</td>
<td>Lymphoma Others -- ?</td>
<td></td>
</tr>
<tr>
<td>Gardner 1989</td>
<td>&lt; 5</td>
<td>3 (ns)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kieselbach 1990</td>
<td>?</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Greenberg 1990</td>
<td>&lt; 1 Peaks to 66</td>
<td>7 (ns)**</td>
<td>Pancreatic Eth. chlorohy.</td>
<td></td>
</tr>
<tr>
<td>Steenland 1990</td>
<td>2.0 - 4.3</td>
<td>13 (ns)**</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

+1 and 2 are updates of the 1979a and 1979b cohorts, respectively; 3 represents a new cohort (see text).

++Out of doors, enclosed system, most measurements nondetectable; spot measurement of 6000 ppm at tank car loading area.

+++Four leukemia deaths occurred in a subcohort with ethylene chlorohydrin exposure and relatively low EtO exposure, RR = 8.7, SMR = 5.7, p < 0.05.

*In the ethylene chlorohydrin subcohort, RR = 14.0.

**Increase in all hematological cancers, men (27, SMR = 1.6, p < 0.05).


8. Dennis, B., Personal Communications, Materials Management Department, Duke University Medical Center, Durham, North Carolina, April 26, 1993.


11. EPA, "Health Assessment Document for Ethylene Oxide," EPA-600/8-84-009F, U. S. Environmental Protection Agency,


22. __________, Rohleu O., Berndtsson B. S., "A Cohort Study of Mortality and Cancer Incidence in Ethylene Oxide


32. Levine, E. G., Bloomfield, C. D., "Leukemias and Myelodysplastic Syndromes Secondary to Drug, Radiation, and


44. Yager, J. W., Hines, C. J., Spear, R. C., "Exposure to Ethylene Oxide at Work Increases Sister Chromatid Exchanges