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Important Aspects of the Evidence for TCDD Carcinogenicity in Man

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Most of the evidence for the carcinogenicity of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in humans has centered on whether TCDD causes soft-tissue sarcomas (STS) and malignant lymphomas (ML). Recently, reports from two of the largest occupational cohort studies have become available. A critical reappraisal of these and other recent reports indicates that it is unlikely that TCDD causes malignant lymphomas in humans. For STS, the evidence for an etiologic role for TCDD is not convincing. However, more data and further clarification are needed before a clear and objective evaluation can be made. Factors such as level of exposure, sex, and host susceptibility may be critical determinants of whether cancer occurs; there is evidence from both humans and animals that these factors play a role, and therefore these factors should be considered in future evaluations. There is a serious need to rule out the possibility that observed effects are due to other concomitant exposures. Consideration of the carcinogenic effects of TCDD in animals reveals consistency with the human data and points to other cancers such as those of the thyroid gland and lung, for example, which are more likely candidates for investigating the role of TCDD in their occurrence, while at the same time providing a basis for a better understanding and interpretation of the human data. There are now sufficient epidemiologic studies in place that will provide a better climate for a definitive evaluation in the near future.

Introduction

The issue of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and possible cancer effects in humans has very important implications for the research on mechanisms of action, and biologic effects of chemicals in general, across different species, as well as implications for how policy makers assess public risk from chemicals in the future. Whether or not TCDD is found to be a human carcinogen, the experience gained from the study of TCDD will have a profound effect on how scientists, policy makers, and the public react in the future to possible long-term health effects of toxic chemicals in the environment. It is important, therefore, that every piece of evidence be critically evaluated for an objective assessment of the carcinogenicity of TCDD in humans. If TCDD is carcinogenic in humans, it is important that the site(s) that are targets for its action be correctly identified. A recent editorial (*1*) has precipitated the need for further clarification and reappraisal of the literature. The present paper discusses certain issues of importance and examines in great detail those studies that have reported a statistically significant association between TCDD exposure and cancer.

The discussion of TCDD carcinogenicity in humans so far has centered primarily around whether or not it causes soft-tissue sarcomas (STS) and malignant lymphomas (ML), mainly because these were the first sites an association was suspected from case reports and case–control studies, and hence these were the sites most frequently studied. However, a review of a total of 17 case–control studies (*2*) that have investigated this putative association shows that only 6 have reported a statistically significant association between STS, ML, and exposure to phenoxyacetic acids and chlorophenols likely to be contaminated with TCDD in the study group as a whole or in any subgroup of it (*3–8*). These six studies therefore deserve close scrutiny because of their uniqueness. Five of the six positive studies (*3–7*) were conducted in Sweden by the same two principal investigators, Hardell and Eriksson, and the sixth study was conducted in Italy by Vineis and colleagues (*8*).

Hardell and Eriksson Case–Control Studies

There is evidence of possible interviewer bias and recall bias in the Hardell and Eriksson studies (*3–7*). In the Hardell series, the risks of STS and ML for exposure to phenoxy compounds were 2.6 and 3.0, respectively, using questionnaire data only (*9*). These risks increased to 5.3 (i.e., doubled) and 4.8 for STS and ML, respectively, when questionnaire data were supplemented by interview data, which was the formal analysis used by the authors (*3,6*). A similar picture was also seen in the Eriksson studies (*4,9*). Thus, use of telephone interview to verify *a*) all doubtful questionnaire responses in relation to exposure and *b*) all exposures reported in the questionnaire by a subset of the study group of agriculture and forestry workers only, resulted in disproportionately more controls being reclassified as unexposed than cases. A truly unbiased verification of exposure would have resulted in similar proportions reclassified in both cases and controls.

Similarly, in one of the more recent studies (*7*), the relative risk obtained for exposure to phenoxy acids was 3.3 using population controls, but was only 2.2 using cancer controls,

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prompting the authors to entertain the possibility of recall bias, in spite of the fact that a previous colon cancer study by the same authors did not show that recall bias was a potentially serious problem in earlier studies (9). It should be pointed out, however, that these biases, although possibly contributory, would not entirely explain all of the increased risk observed, as a reduced but significant risk remained even after taking the biases into account.

There is lack of consistency in the findings in an attempted replication of these studies by the authors. An earlier study by Eriksson reported a statistically significant 17-fold risk for exposure to 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and other phenoxy acids, and a 4-fold risk for non-TCDD-containing phenoxy compounds (*4*). In a later study, Eriksson reported a nonstatistically significant risk of only 1.8 for exposure to 2,4,5-T, a negative risk of 0.6 for non-TCDD-containing phenoxy compounds, and a statistically significant risk of 5.3 for exposure to "high grade" chlorophenols not containing TCDD (*5*). Similarly, Hardell and Sandstrom reported a statistically significant 6.6-fold risk for exposure to chlorophenols in one study (*6*), and on repeating the study, Hardell and Eriksson obtained a negative risk of between 0.4 and 0.5 for exposure to "high grade" chlorophenols (*7*). These inconsistencies are by no means trivial. Thus, the picture from these studies is rather confusing, and difficult to interpret and thus calls for caution, particularly since nearly all of the evidence from case–control studies for TCDD being a human carcinogen is based on these studies.

The risks reported were associated with unusually short periods of exposure and tissue samples from exposed cases did not show elevated levels of TCDD. The findings in the Swedish studies were remarkable in that risks of up to 6-fold were reported to be associated with very short duration of exposure. For example, in one of Hardell's studies, of the 11 cases exposed to 2,4,5-T, only 2 were exposed for more than a year (all 10 controls were exposed for less than a year) (*6*). Hardell also analyzed fat samples for TCDD from patients with STS and ML who had sprayed phenoxy compounds more than 10 years previously (*10*). Rather unexpectedly, TCDD levels in these STS and ML cases were found to be well within normal limits (background levels in the general population are usually not more than 20 ppt). The mean level of TCDD in adipose tissue of these Swedish patients was found to be only 2.7 ppt, which, even if one assumes that the samples were collected on average 20 years after exposure ceased (the authors said more than 10 years), the mean TCDD levels of these patients were on average probably never much above background levels at any time in their lives, if at all. In fact, the absence of elevated TCDD levels in these patients can be interpreted to mean that, indeed, as observed in the Swedish studies, exposure was brief, perhaps to the point of being insignificant for most of the cases.

The studies show a lack of corroboration by cohort studies of users. The risks reported by Hardell and Eriksson in these case–control studies stemmed from exposure mainly during the use of phenoxy acid herbicides and chlorophenols. Yet, with one exception (*11*), these findings have not been confirmed by 15 cohort studies of professional users (sprayers and applicators), even those conducted within Sweden (*12*), even considering only the few studies with sufficient power.

There is a possible lack of corroboration by the National Institute of Occupational Safety and Health (NIOSH) study. The recently published mortality study of a combined group of 12 plants in the United States producing 2,4,5-T, trichlorophenol (TCP), pentachlorophenol (PCP), and hexachlorophene by the NIOSH does not confirm the findings of the Swedish case–control studies as yet (*13*). The NIOSH cohort study reported that, contrary to the Swedish case–control studies, workers exposed to TCDD for less than a year with a possible mean serum TCDD level of 640 ppt at the time exposure ceased showed no excess of STS or ML, even allowing for a 20-year latency (*13*). This may be due to the fact that the NIOSH study does not yet have sufficient power to examine risk of these rare cancers in this particular subgroup. However, if these results hold later, they contradict the Swedish findings in which 5- to 6-fold risk of both STS and ML were observed in persons mainly with less than 1 year of exposure to TCDD, particularly as adipose tissue samples from exposed STS and ML cases in the Swedish studies did not show elevated levels of TCDD (*10*).

There is insufficient evidence of dose–response relationship. For STS, a dose–response analysis was either not done (*4,6,7*) or a relationship not evident (*5*) for exposure to phenoxy acids. For exposure to chlorophenols, no analysis was done in two studies (*4,6*), and in one, a negative risk was observed (*7*), while in another, no study subject was exposed to chlorophenols containing TCDD (*5*). For ML, no significant dose–response relationship could be demonstrated for exposure to phenoxy compounds, and a positive relationship was observed with chlorophenols (*2*).

An important point that has to be considered in any evaluation of putative TCDD association with cancer in humans is that occupational groups that have been exposed to TCDD-containing phenoxys/chlorophenols have also been exposed to other chemicals concomitantly, some of which may cause STS and ML in man or animals (e.g., arsenicals, ethylene dibromide, aniline, vinyl chloride). Moreover, case–control studies of STS and ML from the U.S. National Cancer Institute and other sources have given valuable insight into how significant risks of these diseases seem to be associated with several other chemicals to which TCDD workers were concomitantly exposed (*14–16*).

None of the Swedish case–control studies controlled for these other exposures while investigating phenoxy acid or chlorophenol exposure, hence the studies did not attempt to rule out these concomitant exposures as the possible cause of the risk observed in the study subjects. Moreover, information on many of these other chemicals was not collected. It is virtually certain that even if they were asked about these exposures, the respondents would have been unable to accurately identify and recall exposure to many of them. The reality of this problem was faced by one investigator who rather than settle for uncertainty and possibly unreliable data, gave up this method of inquiry and used occupation as a surrogate instead (*17*).

Arsenicals, which could also cause STS in humans, were widely used to preserve wood in Swedish homes (*18*), yet none of the Swedish case–control studies provided information on this exposure; neither did the rest of the Swedish studies report on creosote exposure, which was found in one Swedish study to be more significantly associated with ML than phenoxy acids (*19*).

Concomitant exposure is an intractable problem in all studies of occupational groups (*20*). Similarly, the NIOSH study by

Fingerhut et al. (*13*) reported that most of the 12 plants involved were large U.S. chemical manufacturing sites that produced thousands of chemicals, and that TCDD-exposed workers spent five times longer working in other areas of these plants. Furthermore, half of the STS cases that featured so prominently in the NIOSH study came from a plant in which it had previously been reported (*21*) that a substantial proportion of the workers had also been exposed to other chemicals such as *p*-aminobiphenyl, a known human bladder carcinogen that also causes hemangiosarcomas in mice (*22*). Unfortunately, the one study in which the issue of concomitant exposures is least likely to be problematic (that of exposed persons in the Seveso accident) does not yet have sufficent-latency or power to show consistent patterns (*23*).

The Italian Case–Control Study

In the Italian study (*8*), none of the male STS cases was definitely exposed to phenoxy herbicides, and only two men could possibly have had any phenoxy exposure. Thus, no increased risk of STS from phenoxy exposure was observed in males. Similarly in females, no excess risk of STS due to phenoxy exposure was observed among deceased subjects. The only evidence relevant to TCDD in this study centers around the fact that among living women, four were classified as definitely occupationally exposed to phenoxy acids and one as possibly exposed, giving a relative risk of 2.4, which was not statistically significant. On further restricting the analysis to women less than 75 years of age who were exposed between 1950 and 1955, a statistically significant relative risk of 15.5 was obtained. It is to be noted that for these women definite exposure meant only that they worked as rice weeders after 1950, since phenoxy compounds were used as chemical weeders in rice fields.

It should be noted that whereas there were ofiicial records available that documented the use of 2,4-dichlorophenoxyacetic acid (2,4-D) and 2-methyl-4-chlorophenoxyacetic acid (MCPA) in the study area in the 1950s, no such data were available for 2,4,5-T, thus it is not known whether 2,4,5-T was actually used in the 1950s. Also, during the period 1950–1955, manual weeding was in use, and chemical weeding was only experimental. This can be interpreted to mean that the experimental use of 2,4,5-T during the 1950s was so insignificant that records were not kept. Hence, the risk of 15.5 reported is for unvalidated exposure. Furthermore, of the total of five STS cases with definite or possible exposure, two were chondrosarcomas of the lower limb and thorax, not soft-tissue sarcomas (only chondrosarcomas of the eyelids and ear lobes are allowed to be coded as soft-tissue sarcomas in the International Classification of Diseases); for another STS, histology was uncertain; one was a Kaposi sarcoma, and one was a leiomyosarcoma. Thus, in this study, ascertainment of exposure and case definition were suspect, not taking into account the sparsity of the data and the absence of risk in males and deceased females.

Is There an Exposure Level below Which TCDD Does Not Cause Cancer?

The issue of the level of exposure, if any, that TCDD is carcinogenic in humans is of importance to policy makers, and great care must be taken that this judgment is made on a sound basis. The NIOSH study is without doubt one of the largest studies of occupational exposure to TCDD, hence it is of considerable importance. Serum collected from a sample of workers with at least 20 years latency from two of the plants showed that persons exposed 15–37 years ago for less than 1 year had a mean TCDD level of 78 ppt as compared with a mean of 462 ppt among those with more than 1 year of exposure (*13,24*). When these values were extrapolated to the dates when individuals were last employed in TCDD-contaminated jobs, the mean TCDD levels were 640 ppt and 3600 ppt, respectively. Thus, assuming TCDD exposure in the plants are roughly comparable, the NIOSH study indicates that persons exposed to TCDD for less than 1 year show as yet no statistically significant elevation of any cancer, even with mean serum TCDD levels of 640 ppt at the time exposure ceased. This finding so far is consistent with observations among individual cohort studies of persons who sprayed phenoxy herbicides professionally. For example, to date, no individual cohort study of sprayers or applicators (including those with sufficient power) has reported a statistically significant elevation of STS or ML or all cancers combined (*3*), except one (*11*) in which there was gross evidence of misclassiScation of STS and ML cases.

Civilian occupational sprayers and applicators can be assumed to have TCDD exposure somewhat similar to that of U.S. Ranch Hands and members of the U.S. Military Chemical Corps who sprayed phenoxy herbicides in Vietnam. The mean serum TCDD levels among ranch hands 15 and 20 years after exposure ceased were 115 ppt (range 16.9–423 ppt) and 49.4 ppt (range 3.2–313 ppt), respectively (*25*). From these results, the mean serum TCDD level for these groups at the time exposure ceased can be extrapolated to be probably less than 500 ppt, and for the individual with the highest exposure the level was less than 1700 ppt (*25*). As with civilian sprayers, no statistically significant increased risk of STS or ML or all cancers has been observed among ranch hands or members of the Chemical Corps (*26–28*).

A mean serum TCDD level of around 460 ppt in sprayers/applicators as a group is quite significant compared to background levels in the general population of not more than 20 ppt and is only low compared to mean levels of more than 3600 ppt seen in production workers and the exposed Seveso population (*24,29,35*). It is possible that failure of the NIOSH study to observe increased risk of any cancer in persons exposed for less than 1 year may be due to the fact that the study does not have sufficient power as yet. However, even if, for the sake of argument, group mean levels of 400–600 ppt are considered low, clearly, the NIOSH study does not yet show that this level of exposure is associated with any statistically significant increased risk of cancer. On the contrary, what the NIOSH study shows and what current knowledge supports is that there is as yet no evidence from cohort studies that exposure to TCDD at significant levels resulting in group mean serum TCDD concentrations of around 640 ppt, i.e., higher than that found even in professional sprayers and applicators, is causally associated with STS or ML, or any cancer, except possibly thyroid cancer (see below). If these results hold after further extension of follow-up of occupational cohorts, they provide strong evidence that if TCDD is eventually found to be carcinogenic in humans, there may be a dose that may vary for different cancer sites below which no cancer occurs. This is consistent with the recent consensus achieved in the scientific community about the mechanism of action of TCDD

at the molecular level (*31*) and also with the animal data (see below).

NIOSH and IARC/NIEHS Registry Studies

The NIOSH study was a carefully designed and well-conducted study. However, caution should be exercised in interpreting the findings. Overzealous interpretation of the findings may only add further confusion to an already complex issue and diminish the value of this important study. It is of interest to compare the findings of the NIOSH study with the recently published results of the larger and equally important International Register of Persons Occupationally Exposed to Phenoxy Acids and Contaminants by the International Agency for Research on Cancer (IARC) sponsored by the U.S. National Institute of Environmental Health Sciences (NIEHS) (*32,33*). This registry involves pooling not only cohorts derived from TCDD-containing and non-TCDD-containing phenoxys/chlorophenols manufacturing/formulating plants like the NIOSH registry, but also cohorts derived from professional sprayers/applicators of phenoxy herbicides from 10 participating countries, involving a total of 18,910 workers (of which about 20% were unexposed).

Two important points must be taken into account in comparing the findings of these two registries for workers in manufacturing plants. *a*) In the NIOSH study, by definition, every subject was exposed to TCDD; in the IARC study, exposed workers were classified only as having probable exposure to TCDD because, although they were known to have worked in phenoxy/chlorophenol plants in which TCDD exposure did occur, it was not known whether individual workers were actually exposed. Thus, because of the potential for misclassification of TCDD exposure for some production workers in the IARC study, a lower risk of a given cancer putatively associated with TCDD exposure in this study for these workers than in the NIOSH study would not be entirely unexpected. *b*) Although every subject was exposed to TCDD in the NIOSH study, everyone was also exposed to the parent phenoxys (2,4,5-T or 2,4-D) or chlorophenols. Thus, the NIOSH study cannot differentiate whether an effect is due to TCDD or due to the parent phenoxys/chlorophenols. The IARC study, on the other hand, was able to identify separately workers who worked in phenoxy/chlorophenol-producing plants in which no TCDD exposure occurred, and thus potentially has the capacity to distinguish a TCDD effect from a phenoxy/chlorophenol effect.

The IARC/NIEHS registry reported a 2-fold risk of STS among workers exposed to phenoxy acids and chlorophenols, which, rather surprisingly, was the same both in workers with probable exposure to TCDD as well as in workers exposed to phenoxys/chlorophenols not containing TCDD. The increased risk was observed only among sprayers/applicators, but not among production workers who had much higher TCDD exposures. It is pertinent to note that at least one of the Swedish case–control studies also reported significant risks of STS in persons exposed to phenoxy herbicides containing TCDD (relative risk = 17.0) and in persons exposed to phenoxy herbicides not containing TCDD (relative risk = 4.2) (*4*); another reported a relative risk of 3.3 for exposure to all phenoxy acids, which did not change when only TCDD-containing phenoxys were considered (*7*). Still yet another study reported a 5-fold increased risk of STS for exposure to chlorophenols, which do not contain TCDD (*5*). The NCI studies showed no association of STS with phenoxys and either showed excess of ML in persons with 2,4-D exposure (*34*) or that exposure to 2,4,5-T or 2,4-D had similar risks of 1.6 and 1.5, respectively (*16*). These types of results, together with the fact that the majority of studies have been negative, do not provide evidence implicating TCDD specifically as the cause of STS or ML.

The importance of the statistically significant increased standardized mortality ratio (SMR) for all cancers observed in the NIOSH study should perhaps be tempered with some caution, for the following reasons: Of 23 distinct cancer sites studied, among persons with 20-year latency, the SMR for those exposed to TCDD for less than 1 year was elevated and actually higher than the corresponding SMR among persons with 1 or more years of exposure for six sites. On the other hand, the SMR for those exposed for 1 or more year was elevated and higher than the corresponding SMR among persons with less than 1 year of exposure for only eight sites. The remaining sites could not be similarly compared, as less than one death was expected in each case.

Also, in the NIOSH study, it should be noted that of 12 plants studied, only 2 of them reported deaths from STS, even though at least 1 was of comparable size and latency as 1 of the 2 which recorded STS deaths. Furthermore, two of the four deaths (one from each plant) reported as STS on death certificates were, in fact, found not to be STS on histological verification. It is noteworthy that in one of the two plants with STS, no excess deaths from all malignant neoplasms were observed, and in a subset of chloracne workers from which the two STS deaths originated, the SMR for all cancers was, in fact, much less than 100 (*35,36*). This inconsistency is vexing and undermines efforts to cleanly put both the excess risks of STS and all cancers combined under the same umbrella of being due to TCDD exposure.

It is interesting that in the Seveso study, the SMRs for all cancers in both males and females from zone A (the most highly contaminated area) were well below unity also, although the numbers involved were very small, and latency too short. Indeed, the risk from STS again seemed to be inversely related to risk from all cancers combined in less contaminated zones B and R (*23*).

The facts that in the NIOSH study not all persons who should be in the cohort showing the highest risk of STS were included (139 workers with chloracne and possible high TCDD exposure representing at least 24% of the cohort were excluded from the study, and no death from STS as underlying cause was reported among them), that person-years could not be counted for certain years for persons known not to have died from STS, and that 45% of the cohort who were alive were excluded from the dose-response analysis, means that the risk of death from STS given in the study has been overestimated, although by how much is difficult to tell. This problem is particularly of relevance considering that half of the STS cases were not STS. Hence, at this time, information on STS from the NIOSH study is only tentative, although a rough calculation indicates that substantial STS risk would remain, even allowing for the overestimation.

The findings in the IARC/NIEHS study with respect to all malignant neoplasms combined seem to corroborate those of the NIOSH study, and the failure of the elevated risk observed for this cause of death, to be statistically significant, may be related

to the lack of a more rigorous definition of TCDD exposure in the IARC study. Interestingly, both the NIOSH and IARC/NIEHS studies observed a slightly elevated but not statistically significant risk for lung cancer, and in the NIOSH study the risk for respiratory cancer was statistically significant among workers with more than 1 year of exposure to TCDD. The IARC/NIEHS study observed a statistically significant 4-fold risk of thyroid cancer based on four deaths, with the highest risk specifically associated with probable TCDD exposure and also excess of tumors of other endocrine organs. Unfortunately, the NIOSH study did not provide information on these sites for comparison.

Lack of Evidence for Malignant Lymphomas

Finally, it should be pointed out that results from a total of 28 of 29 individual cohort studies of production workers and sprayers, and also both the IARC/NIEHS and NIOSH registries, and 6 of 7 case–control studies that investigated TCDD specifically (*3*) now indicate that it is unlikely that malignant lymphoma is causally associated with TCDD exposure because none of the studies showed a statistically significant risk associated with possible TCDD exposure. The Danish cancer incidence study is the only cancer incidence study of production workers and also the only cohort study in which an attempt was made to separate workers engaged in phenoxy production from those engaged in the production of other chemicals. Importantly, a statistically significant excess of ML observed in the factory as a whole was not among workers in phenoxy acids production, but among workers engaged in the production of other substances, mainly pigments (*37*). This is consistent and reminiscent of the observation that the excess of ML reported in Vietnam veterans was ultimately found to be concentrated among veterans who served in ships, without possible exposure to Agent Orange (*38*). Interestingly, a statistically significant excess of lymphomas due to TCDD in male animals has been reported only when TCDD was given at high toxic doses during infancy resulting in the occurrence of mainly thymic lymphomas (*39*).

The editorial mentioned in the Introduction (*1*) closed by recommending that medical history of a patient with STS should include a search for dioxin exposure, and, conversely, the diagnostic workup of some problems reported by an exposed person should include a search for STS. In order to safeguard against future spurious associations that might result if emphasis is laid on dioxins only, it is recommended that, routinely for every case of STS, as part of the medical history, a detailed search for all known and unknown risk factors should be made, including detailed inquiry into all possible chemical and occupational exposures. Similarly, persons with well-documented high exposure to TCDD should be put under close, long-term surveillance for any significant medical outcome, such as is being done in cohort studies of cancer incidence or mortality. The IARC/NIEHS and NIOSH registeries are resource centers for these and other types of investigation.

Comparison of Animal Bioassays and Epidemiologic Findings

It is necessary to compare the results of long-term carcinogenicity studies of TCDD in mice, rats, and hamsters (*39–48*) with the findings in humans so far. Because the human data available are for males, this comparison will be restricted to this sex only. Tumors observed to be statistically significantly in excess include neoplastic nodule of the liver/hepatocellular carcinoma (*39,40,43*), thyroid follicular cell adenoma/carcinoma (*40*), adrenal cortical adenoma (*40,42*), lung adenoma/carcinoma (*40,43*), fibroma of subcutaneous tissue (*40*), hard palate/nasal turbinates (*42*), squamous carcinoma of the tongue (*42*), skin carcinoma (*45*), and thymic lymphomas (from exposure during infancy) (*39*). Tumors observed in male animals receiving the higher doses and not in those in the lowest dose group, or controls, but not statistically significant, include neoplastic nodule of the liver/hepatocellular carcinoma (*44*), cholangiocarcinomas, retroperitoneal histiocytomas, glioblastoma, astrocytoma, Leydig cell adenoma/sclerosing seminoma of testes, adenocarcinoma of kidney, ear duet carcinoma, leukemia, fibroma (*43*), angioma/angiosarcoma (*39,43*), fibrosarcoma (*42,43*), thyroid follicular cell adenoma/carcinoma (*42*), mammary adenocarcinoma (*40*), fibroma of subcutaneous tissue (*43*), intra-abdominal schwannoma/sarcoma, leiomyosarcoma (*42*), skin carcinoma (*43,46*), and nonthymic lymphomas and squamous cell carcinoma of stomach (*39*).

It is significant that in the feeding/gavage studies (*39,40,42–44*), apart from thyroid follicular cell adenoma/carcinoma, no other tumor was statistically significantly in excess at a dose of 0.001 μg/kg/day or 0.01 μg/kg/week. Animals receiving these doses had mean adipose tissue levels of 540 ppt at the time of sacrifice (*42*). This is remarkably consistent with the human data, in which none of the individual cohort studies of professional sprayers (who have mean serum levels of TCDD probably not exceeding 460–500 ppt as a group at the time exposure ceased) except one (*11*) shows any significant excess of any cancer. It is also consistent with the NIOSH finding of as yet (power considerations notwithstanding) no significant excess of any cancer in workers with less than 1 year of exposure to TCDD with a possible mean serum TCDD level of 640 ppt at time of termination of exposure. The fact that the thyroid gland was the organ in which the carcinogenic action of TCDD was most readily evident in male animals is also consistent with the fact that thyroid cancer is the only cancer for which an excess was observed to be statistically significant in the IARC/NIEHS study in both sprayers and production workers, and specifically associated with TCDD exposure. This finding needs confirmation and further investigation, particularly to exclude the role of other chemical exposures that are known to cause thyroid cancer in animals. Unfortunately, the NIOSH study did not provide information on thyroid cancer.

It is of note that liver tumors were not observed to be significantly in excess in male animals given TCDD at levels of less than 0.5 μg/kg/week or 0.1 μg/kg/day (mean adipose levels of less than 8100 ppt). This is consistent with the absence of excess risk of liver cancer in the NIOSH and IARC/NIEHS registry studies and also in all other cohort studies, since even in the NIOSH study with the highest occupational exposure to TCDD, mean serum TCDD levels in workers exposed for more than a year (allowing for a 20-year latency) was only 3600 ppt (*24*).

It should be noted that, apart from thyroid tumors, all other tumors observed to be statistically significantly in excess in male

animals were associated with mean adipose tissue levels of TCDD much above 540 ppt, which in humans may be encountered in production workers but not usually in professional sprayers as a group. Thus, further comparison of the human data with animal data for these sites should concentrate on examination of risk of TCDD exposure at high levels. Because an excess of respiratory cancer at high levels of exposure was reported in the NIOSH study, and also other endocrine tumors in addition to thyroid were in excess in the IARC/NIEHS study, these sites warrant further investigation as they are causally associated with TCDD exposure in animals. It is noteworthy that a statistically significant excess of STS has not been reported in male animals, although a statistically significant excess of subcutaneous fibrosarcoma in female rats and mice has been observed (*40,41*). Whether the fact that these tumors, which are uncommon, have been analyzed by histologic type in these studies rather than being grouped together contributes to the failure to observe an excess of STS in male animals is not known.

The occurrence of one case each of six different tumor types, but none in the control animals, has been reported in rats fed 5 ppt TCDD in diet (*43*) [which is roughly estimated to be compatible with adipose tissue TCDD levels of possibly less than 200 ppt because adipose tissue TCDD levels of 540 ppt in animals mentioned above were recorded in rats fed 22 ppt TCDD in diet (*42*)]. The dose of 5 ppt TCDD in diet is the lowest dose at which any tumor has been recorded in TCDD-exposed animals. Since no tumor has been reported in animals fed 1 ppt TCDD in diet (*43*), the bioassay data are also consistent with the proposition that there may be a level of exposure below which TCDD does not cause cancer. A rough guess is that this dose of 1 ppt TCDD in diet in animals approaches background levels of TCDD exposure in the general human population.

The issue of cancer in humans as a result of exposure to TCDD has been a complex one. However, careful examination and interpretation of the data reveal remarkable consistency both within published human studies and between human and animal bioassay studies. Available evidence does not support a causal role for TCDD in the occurrence of malignant lymphomas in men, and for soft-tissue sarcomas, the weight of evidence on balance seems to run against a causal role also (*2,20*). However, further clarification of the picture is required for a definite evaluation to be made for STS, since *a*) in spite of the large size of the NIOSH and IARC/NIEHS registries, cohort studies still do not have sufficient power to assess STS risk adequately and *b*) it is possible that perhaps only persons with a predisposition such as chloracne patients or persons exposed at extremely high doses of TCDD develop the disease. There is evidence of significant human interindividual variation in enzyme induction by compounds belonging to the TCDD-related family—as much as 500-fold difference between individuals in one study (*49*). Similarly, experiments in rats show that increasing doses of TCDD do not result in formation of preneoplastic lesions (liver foci) until after the dose which results in saturation of the Ah receptor (binding to which is necessary for any biologic effect of TCCD to occur) is exceeded, i.e., at TCDD doses of around 30 ng/kg/day, suggesting that TCDD may cause cancer only at higher doses (*49*). Nested case–control studies of STS or other candidate cancers within the cohorts assembled in the NIOSH and IARC/NIEHS registries may shed light on whether it is TCDD or concomitant exposures which accounts for any, or all observed risk of STS or other cancers in these occupational groups. It is imperative that the role of concomitant exposures and the level of TCDD exposure be seriously considered in further reports of epidemiologic studies.

It is interesting to recall that vinyl chloride (VC) causes angiosarcoma of the liver in humans at a frequency that is possibly at least 100 times less than that for STS (*50,51*). In addition, the carcinogenic potency for vinyl chloride in animals is estimated to be 50 million times less than that of TCDD (*52*). Yet, within 6 months of the report of the first three cases of angiosarcoma of the liver in a vinyl chloride plant, a total of 13 cases were identified over a 10-year period in four U.S. plants, and a further 6 in Europe, making it possible for vinyl chloride to be declared a human carcinogen within that same year (*50,53*). A clear excess of angiosarcoma of the liver was observed in a plant in which there were only a total of 24 deaths. Furthermore, 9 of the first 11 occupational studies evaluated by the IARC showed clear excess of angiosarcoma of the liver (*54*). Within less than 4 years of the initial cases, a total of 64 cases among VC workers had been identified worldwide (*55*). Even taking into consideration that plant workers were exposed to vinyl chloride at much higher concentrations than those found for TCDD (*54*), this should be compared to only a total of eight STS deaths that feature in mortality analyses of both the NIOSH and IARC/NIEHS registries involving most of the plants in which TCDD exposure occurs in the Western world, covering study periods of more than 40 years dating back from the 1940s to the present. In fact, in both registeries, since the 1940s, less than 20 soft-tissue sarcoma cases have been reported so far worldwide among workers exposed both in manufacturing plants and during spraying.

It is hoped that the above discussion will contribute toward keeping the issue in proper perspective. It is strongly advised that evaluations on whether TCDD is carcinogenic in humans should be based on ample or at least consistent and unambiguous data. There are now sufficient epidemiologic studies in place that may provide this framework in the near future. For the moment, focusing on neither STS nor ML makes a particularly strong case for TCDD being a human carcinogen. Focus should now also extend to serious consideration of other target sites identified through occupational cohort and animal studies as potential candidates for its carcinogenic action, particularly lung cancer, for which, in addition to all cancers combined, another recent major study of TCDD-exposed production workers reported a statistically significant increased risk (*56*). Now that increased risk of all cancers combined is being reported in important TCDD-exposed cohorts (*13,33,56*), it should not be difficult in the near future with increased statistical power to observe excess of specific sites to be consistently statistically significant also, if indeed it is TCDD that is partly or wholly responsible for these excesses.

REFERENCES

1. Bailar, J. C., III. How dangerous is dioxin? N. Engl. J. Med. 324: 260–262 (1990).
2. Johnson, E. S. Human exposure to 2,3,7,8-TCDD and risk of cancer. CRC Crit. Rev. Toxicol. 21: 451–464 (1992).
3. Hardell, L., Eriksson, M., Lenner, P., and Lundgren, E. Malignant lymphoma and exposure to chemicals, especially organic solvents, chlorophenols

and phenoxy acids: a case-control study. Br. J. Cancer 43: 169–176 (1981).
4. Eriksson, M., Hardell, L., Berg, N. O., Moller, T., and Axelson O. Soft tissue sarcomas and exposure to chemical substances: a case-referent study. Br. J. Ind. Med. 38: 27–33 (1981).
5. Eriksson, M., Hardell, L., and Adami, H. Exposure to dioxins as a risk factor for soft tissue sarcoma: a population-based case-control study. J. Natl. Cancer Inst. 82: 486–490 (1990).
6. Hardell, L., and Sandstrom, A. Case-control study: soft-tissue sarcoma and exposure to phenoxyacetic acids or chlorophenols. Br. J. Cancer 39: 711–717 (1979).
7. Hardell, L., and Eriksson, M. The association between soft tissue sarcomas and exposure to phenoxyacetic acids. A new case-referent study. Cancer 62: 652–656 (1988).
8. Vineis, P., Terracini, B., Ciccone, G., Cignetti, A., Colombo, E., Donna, A., Maffi, L., Pisa, R., Ricci, P., Zanini, E., and Comba, P. Phenoxy herbicides and soft-tissue sarcomas in female rice-weeders: a population-based case-control study. Scand. J. Work Environ. Health 13: 9–17 (1987).
9. Hardell, L. Relation of soft-tissue sarcoma, malignant lymphoma and colon cancer to phenoxy acids, chlorophenols and other agents. Scand. J. Work Environ. Health 7: 119–130 (1981).
10. Nygren, M., Hansson, M., Rappe, C., Dommellof, L., and Hardell, L. Analysis of polychlorinated dibenzo-p-dioxins and dibenzofurans in adipose tissue from soft-tissue sarcoma patients and controls. Presented at the Division of Environmental Chemistry, American Chemical Society, Miami, FL, April 1985.
11. Corrao, G., Calleri, M., Carle, F., Russo, R., Besia, S., and Piccioni, P. Cancer risk in a cohort of licensed pesticide users. Scand. J. Work Environ. Health 15: 203–209 (1989).
12. Wiklund, K., Dich, J., Holm, L.-E., and Eklund, G. Risk of cancer in pesticide applicators in Swedish agriculture. Br. J. Ind. Med. 46: 809–814 (1989).
13. Fingerhut, M., Halperin, W. E., Marlow, D. A., Piacitelli, L. A., Honchar, P. A., Sweeney, M. H., Griefe, A. L., Dill, P. A., Kyle Steenland, A. B., and Suruda, A. J. Cancer mortality in workers exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin. N. Engl. J. Med. 324: 212–218 (1991).
14. Cantor, K., Everett, G., Blair, A., Gibson, R., Schuman, L., and Isacson, Farming and non-Hodgkin's lymphoma (Abstract). Am. J. Epidemiol. 122: 535 (1985).
15. Hoar Zahm, S., Weisenburger, D. D., Babbitt, P. A., Saal, R. C., Cantor, K. P., and Blair, A. A case-control study of non-Hodgkin's lymphoma and agricultural factors in Eastern Nebraska (Abstract). Am. J. Epidemiol. 128: 901 (1988).
16. Hoar Zahm, S., Weisenburger, D. D., Babbitt, P. A., Saal, R. C., Vaught, J. B., Cantor, K. P., and Blair, A. A case-control study of non-Hodgkin's lymphoma and the herbicide 2,4-dichlorophenoxyacetic acid (2,4-D) in Eastern Nebraska. Epidemiology 1: 349–356 (1990).
17. Wingren, G., Fredrikson, M., Noorlind Brage, H., Nordenskjold, B., and Axelson, O. Soft tissue sarcomas and occupational exposures. Cancer 66: 806–811 (1990).
18. Lindau, L. Emissions of arsenic in Sweden and their reduction. Environ. Health Perspect. 19: 25–29 (1977).
19. Persson, B., Dahlander, A., Fredriksson, M., Brage, H. N., Ohlson, C.-G., and Axelson, O. Malignant lymphomas and occupational exposures. Br. J. Ind. Med. 46: 516–520 (1989).
20. Johnson, E. S. Association between soft-tissue sarcomas, malignant lymphomas, and phenoxy herbicides/chlorophenols: evidence from occupational cohort studies. Fundam. Appl. Toxicol. 14: 219–234 (1990).
21. Suskind, R. R., and Hertzberg, V. S. Human health effects of 2,4,5-T and its toxic contaminants. J. Am. Med. Assoc. 251: 2372–2380 (1984).
22. IARC. Monographs on the Evaluation of Carcinogenic Risks to Humans. Overall Evaluations of Carcinogenicity: An Updating of IARC Monographs 1 to 42, Supplement 7. International Agency for Research on Cancer, Lyon, 1987, pp. 91–92.
23. Bertazzi, P. A., Zocchetti, C., Pesatori, A. C., Guercilena, S., Sanarico, M., and Radice, L. Ten-year study of the population involved in the Seveso incident in 1976. Am. J. Epidemiol. 129: 1187–1200 (1989).
24. Fingerhut, M., Halperin, W. E., Marlow, D. A., Piacitelli, L. A., Honchar, P. A., Sweeney, M. H., Griefe, A. L., Dill, P. A., Kyle Steenland, A. B., and Suruda, A. J. Mortality among U.S. Workers Employed in the Production of Chemicals Contaminated with 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD). Final Report. NTIS No. PB 91-125971. National Technical Information Service, Springfield, VA, 1991.
25. Patterson, D. G., Fingerhut, M. A., Roberts, D. W., Needham, L. L., Haring Sweeney, M., Marlow, D. A., Andrews, Jr., J. S., and Halperin, W. E. Levels of polychlorinated dibenzo-p-dioxins and dibenzofurans in workers exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin. Am. J. Ind. Med. 16: 135–146 (1989).
26. Wolfe, W. H., Michalek, J. E., Miner, J. C., Rahe, A., Silva, J., Thomas, W. F., Grubbs, W. D., Lustik, M. B., Karrison, T. G., Roegner, R. H., and Williams, D. E. Health status of Air Force veterans occupationally exposed to herbicides in Vietnam. I. Physical health. J. Am. Med. Assoc. 264: 1824–1831 (1990).
27. Michalek, J. E., Wolfe, W. H., and Miner, J. C. Health status of Air Force veterans occupationally exposed to herbicides in Vietnam. II. Mortality. J. Am. Med. Assoc. 264: 1832–1836 (1990).
28. Thomas, T. L., and Kang, H. K. Mortality and morbidity among Army Chemical Corps Vietnam veterans: a preliminary report. Am. J. Ind. Med. 18: 665–673 (1990).
29. Mocarelli, P., Pocchiari, F., and Nelson, N. Preliminary report: 2,3,7,8-tetrachlorodibenzo-p-dioxin exposure to humans—Seveso, Italy. MMWR 37: 733–736 (1988).
30. Mocarelli, P., Patterson, D. G., Jr., Marocchi, A., and Needham, L. L. Pilot study (Phase II) for determining polychlorinated dibenzo-p-dioxin (PCDD) and polychlorinated dibenzofuran (PCDF) levels in serum of Seveso, Italy residents collected at the time of exposure: future plans. Chemosphere 20: 967–974 (1990).
31. Roberts, L. Dioxin risks revisited. Science 251: 624–626 (1991).
32. Johnson, E. S., Winkelmann, R., L'Abbe, K. A., Kogevinas, M., Saracci, R., Bertazzi, P. A., Bueno De Mesquita, H. B., Coggon, D., Green, L. M., Johnson, E. S., Kauppinen, T., K. A., Littorin, M., Lynge, E., Mathews, J. D., Neuberger, M., Osman, J., Pearce, N., and Winkleman, R. Phenoxy acid herbicides and contaminants: description of the IARC International Register of Workers. IARC Working Group. Am. J. Ind. Med. 18: 39–45 (1990).
33. Saracci, R., Kogevinas, M., Bertazzi, P. A., Bueno De Mesquita, H. B., Coggon, D., Green, L. M., Kauppinen, T., L'Abbe, K. A., Littorin, M., Lynge, E., Mathews, J. D., Neuberger, M., Osman, J., Pearce, N., and Winkleman, R. Cancer mortality in workers exposed to chlorophenoxy herbicides and chlorophenols. Lancet 338: 1027–1032 (1991).
34. Hoar, S. K., Blair, A., Holmes, F. F., Boysen, C. D., Robel, R. J., Hoover, R., and Fraumeni, J. J., Jr. Agricultural herbicide use and risk of lymphoma and soft-tissue sarcoma. J. Am. Med. Assoc. 256: 1141–1147 (1986).
35. Bond, G. G, McLaren, E. A., Lipps, T. E., and Cook, R. R. Effect of reclassification of chloracne cases. J. Occup. Med. 32: 423 (1990).
36. Bond, G. G., Mclaren, E. A., Lipps, T. E., and Cook, R. R. Update of mortality among chemical workers with potential exposure to the higher chlorinated dioxins. J. Occup. Med. 31: 121–123 (1989).
37. Lynge, E. A follow-up of cancer incidence among workers in manufacture of phenoxy herbicides in Denmark. Br. J. Cancer 52: 259–270 (1985).
38. The Selected Cancers Cooperative Study Group. The association of selected cancers with service in the US military in Vietnam. I. Non-Hodgkin's lymphoma. Arch. Intern. Med. 150: 2473–2483 (1990).
39. Della Porta, G., Dragani, T. A., and Sozzi, G. Carcinogenic effects of infantile and long-term 2,3,7,8-tetrachlorodibenzo-p-dioxin treatment in the mouse. Tumori 73: 99–107 (1987).
40. NTP. Carcinogenesis Bioassay of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (CAS No. 1746-01-6) in Osborne-Mendel Rats and B6C3F, Mice (Gavage Study). NTP-80-31. Technical Report Series No. 209. National Toxicology Program, Research Triangle Park, NC, 1982.
41. NTP. Carcinogenesis Bioassay of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (CAS No. 1746-01-6) in Swiss-Webster Mice (Dermal Study). NTP-80-32. Technical Report Series No. 201. National Toxicology Program, Research Triangle Park, NC, 1982.
42. Kociba, R. J., Keyes, D. G., Beyer, J. E., Carreon, R. M., Wade, C. E., Dittenber, D. A., Kainins, R. P., Frauson, L. E., Park, C. N., Barnard, S. D., Hummel, R. A., and Humiston, C. G. Results of a two-year chronic toxicity and oncogenicity study of 2,3,7,8-tetrachlorodibenzo-p-dioxin in rats. Toxicol. Appl. Pharmacol. 46: 279–303 (1978).
43. Van Miller, J. P., Lalich, J. J., and Allen, J. R. Increased incidence of neoplasms in rats exposed to low levels of 2,3,7,8-tetrachlorodibenzo-p-dioxin. Chemosphere 9: 537–544 (1977).
44. Toth, K., Somfai-Relle, S., Sugar, J., and Bence, J. Carcinogenicity testing of herbicide 2,4,5-trichlorophenoxyethanol containing dioxin and of pure dioxin in Swiss mice. Nature 278: 548–549 (1979).
45. Rao, M. S., Subbarão, V., Prasad, J. D., and Scarpelli, D. G. Carcinogenicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin in the Syrian golden hamster. Carcinogenesis 9: 1677–1679 (1988).

46. DiGiovanni, J., Viaje, A., Berry, D. L., Slaga, T. J., and Juchau, M. R. Tumor-initiating ability of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and arochlor 1254 in the two-stage system of mouse skin carcinigenesis. Bull. Environ. Contam. Toxicol. 18: 552–557 (1977).
47. Innes, J. R. M., Ulland, B. M., Valerio, M. G., Petrucelli, L., Fishbein, L., Hart, E. R., Pallotta, A. J., Bates, R. R., Falk, H. L., Gart, J. J., Klein, M., Mitchell, I., and Peters, J. Bioassay of pesticides and industrial chemicals for tumorigenicity in mice: a preliminary note. J. Natl. Cancer Inst. 42: 1101–1114 (1969).
48. Huff, J. E., Salmon, A. G., Hooper, N. K., and Zeise, L. Long-term carcinogenesis studies on 2,3,7,8-tetrachlorodibenzo-p-dioxin and hexachlorodibenzo-p-dioxins. Cell Biol. Toxicol. 7: 67–93 (1991).
49. Lucier, G. Humans are a sensitive species to some of the biochemical effects of structural analogs of dioxin. Environ. Toxicol. Chem. 10: 727–735 (1991).
50. IARC. Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man, Vol. 7. Some Anti-thyroid and Related Substances, Nitrofurans, and Industrial Chemicals. International Agency for Research on Cancer, Lyon, 1974, pp. 291–318.
51. NCI. Third National Cancer Survey: Incidence Data. National Cancer Institute Monograph 41. DHEW Publication No. (NIH) 75-787. National Cancer Institute, Bethesda, MD, 1975.
52. EPA. Health Assessment Document for Polychlorinated Dibenzo-p-dioxins. Final Report. EPA/600/8-84/014F. U.S. Environmental Protection Agency, Cincinnati, OH, 1985, pp. II-125–II-140.
53. Lee, F. I., and Harry, D. S. Angiosarcoma of the liver in a vinyl-chloride worker. Lancet i: 1316–1318 (1974).
54. IARC. Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans, Vol. 19. Some Monomers, Plastics, and Synthetic Elastomers, and Acrolein. International Agency for Research on Cancer, Lyon, 1979, pp. 377–438.
55. Spiritas, R., and Kaminski, R. Angiosarcoma of the liver in vinyl chloride/polyvinyl chloride workers. 1977 Update of the NIOSH Register. J. Occup. Med. 20: 427–429 (1977).
56. Manz, A., Berger, J., Dwyer, J. H., Flesch-Janys, D., Nagel, S., and Waltsgott, H. Cancer mortality among workers in chemical plant contaminated with dioxin. Lancet 338: 959–964 (1991).