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Why toxicologists resisted and radiation geneticists supported EPA'S adoption of LNT for cancer risk assessment

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ABSTRACT

The linear non-threshold (LNT) dose response model for cancer risk assessment has been a controversial concept since its initial proposal during the 1930s. It was long advocated by the radiation genetics community in the 1950s, some two decades prior to being generally adopted within the chemical toxicology community. This paper explores possible reasons for such major differences in the acceptance of LNT for cancer risk assessment by these two key groups of scientists.

1. Introduction

The US Congress passed, and President Richard Nixon signed into law the Safe Drinking Water Act in 1974. A significant provision of the Act involved engaging the US NAS to advise the EPA on multiple scientific and technical areas such as chemical and radiation risk assessment, including cancer risk assessment. To achieve these goals the NAS created the Safe Drinking Water Committee (SDWC) in 1975. In 1977 the SDWC published the 700 page *Drinking Water and Health* [1] report offering EPA widespread guidance, including cancer risk assessment and its underlying scientific foundations that supported the LNT. Within two years EPA would issue the first national drinking water standard for a chemical carcinogen using the LNT for total trihalomethanes (THM) [2]. This action would jump start an avalanche of other LNT based cancer risk assessments by EPA, not just for drinking water but for other environmental media as well. The decision to go linear by the SDWC for chemical carcinogens was therefore as highly significant as it was precedent setting, and led the way for future EPA cancer risk assessment actions.

The actions of the SDWC to recommend LNT for chemical carcinogens was more than two decades after a similar recommendation of the 1956 NAS BEAR Genetics Panel to switch from a threshold to LNT for radiation induced mutation. This action of the BEAR Genetics Panel was soon followed by a recommendation of the National Committee for Radiation Protection and Measurement (NCRPM) to generalize the LNT concept to somatic cells for cancer risk assessment. This two decade time gap in the decision to go linear for cancer risk assessment for ionizing radiation and chemical carcinogens suggests the possibility that chemical toxicologists and radiation geneticists/cancer researchers may have evolved considerably differently with respect to the concept of cancer risk assessment, prompting the present paper.

2. How radiation geneticists came to embrace LNT

While ionizing radiation and chemicals induce cancer, their historical research foundations concerning cancer risk assessment have some important differences. In the case of ionizing radiation, it was known as a human carcinogen within a decade of its discovery in 1895 as well as having a range of diagnostic and therapeutic applications [3]. The applications of X-rays lead to the development of substantial scientific, occupational health, and clinical data with important implications for cancer risk assessment. There was also considerable research assessing ionizing radiation induced mutations and their dose response relationships in multiple biological models.

During the initial two decades following Muller's report on X-ray induced gene mutations [4], the radiation genetics community became concerned with protecting humans from the harmful effects of X-rays on germ cells, developing embryos and fetuses. The protection of workers exposed to ionizing radiation also became a priority with some radiation geneticists becoming members of national (e.g. NCRPM) and international advisory committees (e.g. ICRP) starting in the 1930's concerned with health and safety [5–7].

These activities quickly drew the radiation genetics community into the domain of human risk assessment, led by Muller's Proportionality

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Rule and its LNT-single hit [8] dose response model for germ cell mutation and later application to cancer risk estimation [5,7,9–11]. This linear dose response assumption lead to the conclusion that there was no safe exposure to ionizing radiation, challenging a threshold model interpretation for some birth defects and most cancer endpoints.

What is striking during this time period, and perhaps little appreciated, is the paucity of experimental animal model studies concerning ionizing radiation induced cancer (See Stannard and Baalman [12] for a detailed history of experimental radiation cancer research in the US from the 1930s through the 1970s). Large-scale animal studies were initiated by the US Atomic Energy Commission (AEC) with extensive lifespan Beagle dog studies in the early 1950s and continued for several decades. These studies were undertaken following preliminary research principally with mice and rats during and after World War II.

The findings from animal studies during the period leading up to the NCRPM cancer linearity recommendation in late 1958 were therefore limited and those which were potentially relevant were generally viewed as not supportive of an LNT recommendation. In a 1958 article dealing with radiation-induced cancer in experimental models Glucksmann [13] noted that “none of the animal experiments have indicated a linear relationship between tumor incidence and dose”. On September 19, 1958 in the journal *Science* Finkel [14] claimed to have published “the best current information on the shape and origin of the dose-response curve” in lifetime experiments assessing the effects of Strontium 90 on tumor formation in mice. This study employed 12 treatment groups with up to 150 mice in the control and lowest exposure group. In general, there was no treatment related responses at the lowest four doses. The lowest dose tested was 100 fold greater than the level established for the general population. This most definitive study for that time-period also did not provide support for the LNT model. In further agreement, Upton [15], following a detailed review of the animal experimental cancer data, stated that “In no instance recorded to date, has a linear relationship between neoplasia and radiation dose been adequately demonstrated.” These consistent animal model study perspectives merged with the findings of Russell [16] on dose rate in mouse spermatogonia and oocytes which discredited the radiation geneticist LNT dogma that all ionizing radiation induced genetic damage was cumulative, not repairable and irreversible.

These developments just prior to the NCRPM's late December 1958 generalization of the 1956 BEAR Genetics Panel germ cell linearity recommendation to cancer risk assessment raised the question of how this NCRPM committee made its LNT cancer risk recommendation in light of the mounting scientific questions and uncertainties, if not open challenges.

In NCRPM Committee discussions in 1958, E.B. Lewis indicated that the LNT model predicted a 1×10^{-6} leukemia risk with 1 rad/year, which was translated into a 10% increase in leukemia incidence per year when exposure was assumed to be twice the background dose [6] (page 613). This risk at low doses was hard to reconcile with the emerging findings of Russell [16] and others that mutation thresholds occurred at many thousands of times greater than background doses and the non-supportive animal model cancer studies. The battle lines were therefore drawn in the radiation community between those siding with the extrapolative predictions of the LNT model and those supporting empirical animal studies that were consistent with a threshold conclusion. With both sides articulating their concerns with opposing views, a compromised position was adopted based on a Precautionary Principle driven LNT policy acknowledging it was not based on “sound” science (see Ref. [17]-page 5, left column, for a summary) but upon both a fear of ionizing radiation and possible limitations in available data sets such as sample size, the capacity of animal models to predict human responses, the capacity of epidemiology to detect very low risks, amongst other factors.

Such Committee decisions often are affected by the backgrounds, beliefs, and potential biases of those comprising the committee. In this case the NCRPM was comprised of highly prestigious individuals but only a few with relevant education, training and experience such as James Crow [18], Edwin B Lewis [19], and Clinton C. Powell [17,20] each of whom had a published record of support for the LNT, despite its limitations. Their LNT position was challenged by Austin Brues [21], director of research at Argonne National Labs, probably leading to the compromised position, yet one still favoring the adoption of LNT.

3. Why toxicologists were skeptical of LNT

With respect to chemical carcinogenesis risk assessment, tumors were first induced in 1918 by chemicals in tars rubbed on rabbit ears [22] and Ichikawa 1918]. By the early 1930s extensive experimental studies had established that numerous polycyclic aromatic hydrocarbons (PAHs) were carcinogenic in animal models [23–34]. Such findings were subsequently submitted to quantitative analyses via low dose biostatistical modeling which set the stage for similar regulatory cancer risk assessment estimates some four decades later by EPA [35,36].

During these early decades of the 20th century, the field of chemical carcinogenesis profited greatly from the collaboration of pathologists (e.g., Kennaway) [25–27] and synthetic organic chemists, such as John W. Cook who synthesized and tested many hundreds of compounds for their carcinogenic effects [28,29]. Cook was widely credited with using his organic chemistry synthesis skills to assess tumorigenicity with single compounds, thereby permitting the reproduction of tumors under experimental conditions. This research was also integrated with advances in fluorescent spectroscopy that lead to the identification of similar spectra of carcinogens [27], facilitating the development of structure activity frameworks for predicting carcinogenic hydrocarbons. By 1947 the number of papers published on the carcinogenicity of individual compounds had exceeded an astonishing 5000 [37]. While these developments revealed a striking relationship between chemical structure and biological effects and had a profound influence on the development of the field, it failed to provide a sound mechanistic foundation for tumor induction.

Despite these limitations, Cook explored further mechanistic understandings when it was revealed that sterol hormones and related agents (e.g., vitamin D, ovarian hormones, bile acids) had the same chemical backbone as did the carcinogenic polycyclic hydrocarbons. Such structural similarities suggested functional relationships implying that sterols might be precursors of carcinogenic hydrocarbons. This lead to the hypothesis that endogenous sterols may affect the occurrence of spontaneous tumors via abnormal metabolic mechanisms [34]. The converging role of sterols and hydrocarbon carcinogens in cancer biology then lead to attempts to implicate hormones in cancer causation. These developments suggested that PAH carcinogens may modulate sterol metabolism, closing the gap between chemically induced cancer and normal hormonal effects. From these developments emerged the idea that chemically induced cancer was governed by pharmacological principles that there were well known to be mediated via threshold dose responses.

As suggested above, the research of Kennaway, Cook and others had a dominating impact on the field of cancer risk assessment. For example, by 1936 the British Medical Association annual meeting had a session entitled “Substances Promoting Normal and Abnormal Growth”, with Cook presenting his findings that linked sterols and hydrocarbon carcinogens. By 1939, Kennaway and Cook would receive the first Anna Fuller Memorial Prize for their research on carcinogenic aromatic hydrocarbons. Further reflecting the impact of his chemical carcinogen research, Kennaway would receive the King's Medal in 1941 and be knighted in 1947, an honor that would also be given to Cook. Kennaway was also nominated for the Nobel Prize in 1951 and 1953.

Despite extensive research on radiation induced mutation following the dramatic discovery of radiation-induced gene mutation by Muller (1927), chemical induced mutation evaded discovery, not being reported until 1946, some 20 years later, achieving this with mustard gas

Table 1
NAS SDWC (1977) low dose linearity guiding principles: no longer tenable four decades later [Source: 7].

Only one or two changes in a cell could transform it and this could lead to cancer.	Not tenable
Human population heterogeneity was a factor, and some people may be at greater risk. Such heterogeneity leads to the conclusion that there was no population-based threshold.	Impossible to practically study
A transformed cell will be irreversibly propagated.	Not tenable
If the mechanism involved mutation, there would be no threshold; in fact, if there were no information on mechanism and cancer occurred, mutation should be assumed.	Not tenable
It is necessary to assume that a single molecule or a few molecules can cause a mutation. Therefore, linearity at low dose can be assumed	Not tenable
There is also the assumption that the exposure would be directly additive to background, if acting via the same mechanism. This would also support the linearity conclusion.	Not tenable
Available mutagenicity data with radiation indicated that it was linear at relatively "low" doses.	Not tenable
Since chemical carcinogens act like ionizing radiation, low dose linearity should also be assumed to be the case for such chemicals.	Not tenable

[38]. Despite the delay in reporting that chemicals could induce mutations, chemical carcinogenesis researchers revealed tumor promotion with mouse skin [39–42]. Carcinogenesis was found to be a multistage process, with apparent dose-related thresholds at different stages. Given the above historical foundations, it is not surprising that a mutation-tumor initiation model would be slow to mature within the chemical carcinogenesis community relative to the field of radiation. It would eventually receive a major boost with the discovery in 1953 that DNA was the genetic material and the development of genotoxicity toward the end of the 1960s [43].

4. The radiation geneticist – toxicologist comparison

Researchers in chemical and radiation carcinogenesis therefore viewed the cancer risk assessment differently. The LNT model was adopted relatively early by many in the radiation community, based on assumed X-ray induced gene mutation and the LNT-single hit model. Linearity for mutation became more convincingly integrated with human radiation induced cancer risks following the 1957 paper of Lewis in *Science*, with this perspective receiving an influential endorsement by the editor in chief [44]. The adoption of LNT by the radiation geneticist community occurred without the assistance of animal model cancer research, in fact, it required ignoring the consistent non-supportive animal model study conclusions.

A linearity decision for chemically-induced cancer proved to be a more difficult task for the chemical toxicologists due, at least in part, to the prolonged delay in demonstrating mutagenicity via chemicals, the strong general tendency of threshold responses in most chemical cancer bioassays, the association of PAH carcinogen effects with sterols and hormones and their pharmacological threshold-like processes, that most toxicologists of the era were trained as pharmacologists, the need for prolonged exposure to high doses of tumor promotors to ensure cancer outcomes for subthreshold doses of carcinogens and the capacity to separate initiation from promotion via different doses, leading to thresholds. Thus, LNT was a much harder sell within the chemical toxicology community as compared with its far more immediate and widespread acceptance in the radiation genetics community.

When the EPA sought to reconcile and even harmonize cancer risk assessment for radiation and chemical carcinogens within the LNT framework in the mid-1970s, it required an administrative fiat according to Roy Albert [45], the chair of the EPA Carcinogen Assessment Group, failing to give appropriate weight to multiple types of adaptive mechanisms such as DNA repair, the consistent evidence of threshold-like, non-linear dose responses in animal bioassays, and also failing to adequately integrate the role of promotion and its threshold like dose dependency which occurs with ionizing radiation [46–51] and chemical carcinogens [52].

The EPA would also seek to obtain support for their actions from prestigious NAS committees such as the SDWC. With respect to the US NAS SDWC, once the LNT perspective was agreed upon by the radiation geneticists and chemical toxicologists, the SDWC then constructed a theoretical foundation of eight "principles" that guided/justified their newly acquired belief in the validity of the LNT model (Table 1). These eight principles, which were published in *Drinking Water and Health* (1977), have not withstood the test of time well, with most being found to be incorrect and others simply not possible to prove.

5. FINALZING the NAS SDWC LNT debate

Since the LNT debate was central to the future of EPA leadership, direction and policy, it could not end with an "undecided" conclusion or even one simply driven by a scientifically weak Precautionary Principle policy as was the in the case of the 1958 NCRPM recommendation and the 1960 BEAR Genetics and Medicine/Pathology Panels [53]. It needed the image of sound science, an endorsing recommendation by yet another prestigious NAS Committee, such as the SDWC.

The LNT debate was eventually won by the EPA and its LNT supporters with the assistance of an overpowering influence of low dose biostatistical modeling perspectives that swayed the quantitatively overwhelmed chemical toxicologists. This resulted in the LNT policy going forward, becoming broadly institutionalized across many governmental agencies and in multiple countries. The rest is history.

Declaration of interest

None.

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References

[1] National Academy of Sciences (NAS), S.D.W.C., Drinking Water and Health, National Academy of Sciences, Washington DC, 1977.
[2] U.E.P. Agency, National interim primary drinking water regulations: control of trihalomethanes in drinking water, Fed. Regist. 44 (231) (1979) 68624–68632.
[3] J.T. Godwin, Carcinogenic effects of ionizing radiations, Int. Rec. Med. Gen. Pract. Clin. 165 (6) (1952) 355–357.
[4] H.J. Muller, Artificial transmutation of the gene, Science (New York, N.Y.) 66 (1699) (1927) 84–87.
[5] E. Calabrese, The linear No-Threshold (LNT) dose response model: a comprehensive assessment of its historical and scientific foundations, Chem. Biol. Interact. 301 (2019) 6–25.
[6] G. Whittmore, The National Committee on Radiation Protection, 1928-1960: from Professional Guidelines to Government Regulation, Ph.D. Thesis (1986).
[7] E. Calabrese, The road to linearity: why linearity at low doses became the basis for

carcinogen risk assessment, Arch. Toxicol. 83 (3) (2009) 203–225.
[8] N.W. Timofeeff-Ressovsky, Ueber die Natur del- Genmutation und der Genstruktur, Nachr. von der Gesellschaft Wiss. Gottingen 1 (3) (1935) 189–245.
[9] E.J. Calabrese, On the origins of the linear no-threshold (LNT) dogma by means of untruths, artful dodges and blind faith, Environ. Res. 142 (2015) 432–442.
[10] E.J. Calabrese, From Muller to mechanism: how LNT became the default model for cancer risk assessment, Environ. Pollut. 241 (2018) 289–302.
[11] E. Calabrese, Toxicology rewrites its history and rethinks its future: giving equal focus to both harmful and beneficial effects, Environ. Toxicol. Chem. 30 (12) (2011) 2658–2673.
[12] J.N. Stannard, R.W. Baalman, Radioactivity and Health : a History, Pacific Northwest Laboratory, 1988.
[13] A. Glucksmann, L.F. Lamerton, W.V. Mayneord, Cancer, Butterworth London, (1958), p. 497.
[14] M.P. Finkel, Mice, men, and fallout, Science 128 (3325) (1958) 637–641.
[15] A.C. Upton, Dose-response relation in radiation-induced cancer, Cancer Res. 21 (6) (1961) 717-&.
[16] W.L. Russell, L. Russell, E.M. Kelly, Radiation dose rate and mutation frequency, Science 128 (3338) (1958) 1546–1550.
[17] C.C. Powell, Radiation hazards, Am. J. Public Health Nation's Health 49 (1) (1959) 1–9.
[18] J. Crow, Genetic effects of radiation, Bull. At. Sci. 14 (1) (1958) 19–22.
[19] E.B. Lewis, Leukemia and ionizing radiation, Science 125 (3255) (1957) 965–972.
[20] C.C. Powell, The government looks at radiation hazards, Radiology 72 (4) (1959) 489–&.
[21] A. Brues, Critique of the linear theory of carcinogenesis, Science 128 (3326) (1958) 693–699.
[22] K. Yamagiwa, K. Ichikawa, Experimental study of the pathogenesis of carcinoma, J. Cancer Res. 3 (1918) 1–21.
[23] E.L. Kennaway, On the cancer-producing factor in tar, Br. Med. J. 1924 (1924) 564–567.
[24] E.L. Kennaway, The formation of a cancer-producing substance from isoprene (2-methylbutadiene), J. Pathol. Bacteriol. 27 (3) (1924) 233–238.
[25] E.L. Kennaway, Experiments on cancer-producing substances, BMJ Br. Med. J. (Clin. Res. Ed.) 1925 (1925) 1–4.
[26] E.L. Kennaway, Further experiments on cancer-producing substances, Biochem. J. 24 (2) (1930) 497–504.
[27] E.L. Kennaway, I. Hieger, Carcinogenic substances and their fluorescence spectra, Br. Med. J. 1 (3622) (1930) 1044–1046.
[28] J.W. Cook, The production of cancer by pure hydrocarbons. Part II, Proc. R. Soc. Lond. - Ser. B Contain. Pap. a Biol. Character 111 (773) (1932) 485-+.
[29] J.W. Cook, et al., The production of cancer by pure hydrocarbons. Part I, Proc. R. Soc. Lond. - Ser. B Contain. Pap. a Biol. Character 111 (773) (1932) 455–+.
[30] J.W. Cook, C. Hewett, I. Hieger, Coal tar constituents and cancer, Nature 130 (1932) 926-926.
[31] J.W. Cook, C.L. Hewett, I. Hieger, The isolation of a cancer-producing hydrocarbon from coal tar Parts I, II, and III, J. Chem. Soc. (1933) 395–405.
[32] G. Barry, J. Cook, A comparison of the action of some polycyclic aromatic hydrocarbons in producing tumours of connective tissue, Am. J. Cancer 20 (1) (1934) 58–69.
[33] A.F. Watson, E. Mellanby, Tar cancer in mice II the condition of the skin when modified by external treatment or diet, as a factor in influencing the cancerous reaction, Br. J. Exp. Pathol. 11 (5) (1930) 311–322.
[34] G. Badger, The carcinogenic hydrocarbons - chemical constitution and carcinogenic activity, Br. J. Canc. 2 (4) (1948) 309–350.
[35] W. Bryan, M. Shimkin, Quantitative analysis of dose-response data obtained with carcinogenic hydrocarbons, J. Natl. Cancer Inst. 1 (1940) 807–833.
[36] W. Bryan, M. Shimkin, Quantitative analysis of dose-response data obtained with three carcinogenic hydrocarbons in strain C3H male mice, J. Natl. Cancer Inst. 3 (5) (1943) 503–531.
[37] I. Hieger, a.B, GM, Ernest laurence Kennaway, 23rd may 1881-1st january 1958, J. Pathol. Bacteriol. 78 (1959) 593–606.
[38] C. Auerbach, J. Robson, Chemical production of mutations, Nature 157 (3984) (1946) 302-302.
[39] I. Berenblum, The cocarcinogenic action of croton resin, Cancer Res. 1 (1) (1941) 44–48.
[40] I. Berenblum, The mechanism of carcinogenesis - a study of the significance of cocarcinogenic action and related phenomena, Cancer Res. 1 (10) (1941) 807–814.
[41] I. Berenblum, P. Shubik, The role of croton oil applications, associated with a single painting of a carcinogen, in tumour induction of the mouses skin, Br. J. Canc. 1 (4) (1947) 379–382.
[42] I. Berenblum, P. Shubik, A new, quantitative, approach to the study of the stages of chemical carcinogenesis in the mouses skin, Br. J. Canc. 1 (4) (1947) 383–391.
[43] B. Ames, W. Durston, Y.e. al, Carcinogens are mutagens - simple test system combining liver homogenates for activation and bacteria for detection, Proc. Natl. Acad. Sci. Unit. States Am. 70 (8) (1973) 2281–2285.
[44] G. Dushane, Loaded dice, Science 125 (3255) (1957) 963-963.
[45] R. Albert, Carcinogen risk assessment in the US environmental protection agency, Crit. Rev. Toxicol. 24 (1) (1994) 75–85.
[46] A. Ootsuyama, H. Tanooka, The tumor-initiating and tumor-promoting effects of ionizing radiations in mouse skin, Jpn. J. Cancer Res. 78 (11) (1987) 1203–1206.
[47] A. Ootsuyama, H. Tanooka, 100-percent tumor induction in mouse skin after repeated beta-irradiation in a limited dose range, Radiat. Res. 115 (3) (1988) 488–494.
[48] A. Ootsuyama, H. Tanooka, Zero tumor incidence in mice after repeated lifetime exposures to 0.5 Gy of beta-radiation, Radiat. Res. 134 (2) (1993) 244–246.
[49] H. Fujiki, M. Mori, H. Tanooka, Delayed induction of ornithine decarboxylase in mouse skin after irradiation with beta-rays, Cancer Lett. 15 (1) (1982) 15–17.
[50] H. Tanooka, Threshold dose-response in radiation carcinogenesis: an approach from chronic alpha-irradiation experiments and a review of non-tumour doses, Int. J. Radiat. Biol. 77 (5) (2001) 541–551.
[51] R.E.J. Mitchell, A. Trivedi, Chronic exposure to ionizing radiation as a tumor promoter in mouse skin, Radiat. Res. 129 (2) (1992) 192–201.
[52] A.L. Reddy, P.J. Fialkow, Influence of dose of initiator on 2-stage skin carcinogenesis in BALB/C mice with cellular mosaicism, Carcinogenesis 9 (5) (1988) 751–754.
[53] E. Calabrese, EPA adopts LNT: new historical perspectives, Chem. Biol. Interact. 308 (2019) 110–112.