RESPONSE TO THE DOCUMENT ENTITLED "NEW ZEALAND NUCLEAR TEST VETERANS: A SUMMARY OF EXPERT REVIEWS".

The 2007 Cytogenetic Study by Dr Al Rowland and co-researchers at Massey University found unequivocally that those New Zealand veterans who participated in the British atomic bomb tests at Christmas Island and Malden Island in 1957/58, known as Operation Grapple, incurred long term genetic damage. This was found using a highly sensitive cytomolecular technique called mFISH. This technique detects chromosome translocations which are an accepted marker for genetic damage. The damage shown by the New Zealand nuclear test veterans was similar to that seen in Chernobyl clean-up workers.

Not surprisingly, these findings met with resistance from certain quarters. Unfortunately a myth has grown up around the study stating that the study was poorly designed, on the erroneous belief that the participants were selected by the veterans themselves. Nothing could be further from the truth. The study was very well designed and experts from all over the world have complimented the researchers on the thoroughness of their scientific approach.

I believe the myth arose from a letter which the Chairman of the Nuclear Test Veterans, Roy Sefton sent to all the veterans, before the study was even agreed upon, asking them if they would be willing to take part in a genetic study. Obviously, without their wide support no study could proceed.

Roy Sefton received a positive reply from 100% of the veterans on the NZNTVA database which was exactly the same list as held by the Department of Veterans' Affairs. Detractors of the study have used this letter as evidence that the veterans self-selected themselves. But if these same critics had ***actually read*** the Cytogenetics Report, they would see that the Department of Veterans Affairs under the direction of the Secretary of Veterans Affairs, Col. Jesse Gunn, sent a letter to all nuclear test veterans inviting them to participate in a genetic study to be conducted by Massey University. The names on the VANZ list were exactly the same as Roy Sefton's list and again the response was 100% positive.

All the responses were sent to the researchers at Massey University and ***they*** were the ones who embarked upon a rigorous process of selection as detailed in the Materials and Methods section of the Report (see Materials and Methods below). The nuclear test veterans had absolutely no say in the selection of participants nor did they have any input into the study. Ethics approval for the study had to pass through seven ethics committees including the Massey University Ethics Committee and six Hospital Board Ethics Committees. If even one of them had found any evidence of poor design it would have been detected and the study rejected.

Reasons for not including ex-naval personnel as Control subjects is explained in the Materials and Methods section below and was accepted by all the Ethics Committees. Selecting ex-military men or policemen of comparable age as control subjects, taking in to account the healthy soldier syndrome, was considered perfectly valid. It is clutching at straws for the writer of the Executive Summary to say that the Experimentals and Controls were not well matched on the basis that the nuclear test veterans on average "had a lower education and lower income".

This issue of supposedly poor study design has been addressed on numerous occasions with the Department of Veterans Affairs and with several past Ministers, by both myself and Roy Sefton, to no avail. The myths are still perpetuated in the Summary of Expert Reviews on the VANZ website. This is not a matter of scientists possessing alternative views about a debatable topic; the comments expressed in the Summary are plainly wrong and must be exposed as such. The Summary should be deleted because it is completely misleading, but I am at least grateful to the current Minister of Veterans Affairs', Hon. Ron Mark, for allowing me to include my comments on the website to correct the gross misinformation.

To clarify the method of selection of participants in the study and to remove all doubt it is necessary to include the Materials and Methods section of the Cytogenetics Report below.

MATERIALS AND METHODS

(1) Population and sampling procedure

Fifty male New Zealand naval nuclear test veterans (Experimentsl group) and 50 male matched controls who had also undergone military or police training when they were younger participated in the study....

Names of volunteer veterans were communicated to the researchers through the Office of Veterans' Affairs. A letter from the Veterans' Affairs Office was mailed out to all nuclear test veterans in the North Island listed on the Boards database. Also included with the letter was a Preliminary Inclusion Criteria Questionnaire (see Appendix I) along with an addressed FREEPOST envelope which was returned to the research team at Massey University with the completed questionnaire. Information furnished enabled the researchers to decide whether a veteran was excluded/included from the potential pool.

A respondent database was compiled from all those who posted their completed Preliminary Inclusion Criteria Questionnaire to Massey University. A *potential participant pool* was formed by excluding any respondents who failed to meet specific inclusion criteria. A *final participant pool* was formed by randomly selecting the specified number of participants from the potential pool database.

Matched controls were selected from a pool of volunteers according to criteria identical to the veterans, but with the essential difference that they did not participate in Operation Grapple. Ex-servicemen were selected as controls where possible, most from the army. Some ex-policemen were also chosen. Ex-naval servicemen were excluded as control subjects on the grounds of controversy as to whether the frigates involved were completely "clean" upon returning to New Zealand and subsequently manned by other crew who may have been theoretically exposed to contamination. Ex-airforce personnel, except for ground crew, were also excluded for reasons of possible increased past exposure to cosmic radiation. Vietnam veterans were also not included in either the Control or Experimental Group because there is a risk these people have been adversely affected by exposure to defoliants. Neither was any man selected, Control or Experimental, who had previously worked in the timber industry, received prolonged exposure to solvents, or was currently receiving chemotherapy or radiotherapy. Selection of both Experimentals and Controls was stratified across the North Island to achieve a random geographical distribution of participants.

Selected final participants were sent an Information Sheet (E = Experimental; C = Control), Consent Form and Detailed Questionnaire (Appendix II) that gathered information relating to their life events and general health. This was necessary in order for the researchers to take into account any other factors that may be causing chromosomal damage, if it appeared, other than participation in Operation Grapple. The participants were asked to sign the Consent Form, fill in the Detailed Questionnaire and return these to researchers at Massey University.

On receipt of the Detailed Questionnaire, a face-to-face interview was arranged and conducted by a psychologist skilled in eliciting memory recall. This was in order to clarify if necessary any incomplete details in their responses, and secure more information related to any substances that might potentially affect the blood sample that would be used for analysis. It was important in this study that we obtained the best recall data possible to validate our results, which is why a face-to-face interview with a trained reviewer was essential. A blood sample was collected at the same time as the interview, or else arrangements were made to collect a sample from the participant at a later convenient date. The whole study was conducted following strict ethical guidelines as specified by the World Medical Association Declaration of Helsinki. Ethics approval to conduct the study was given by the Massey University Human Ethics Committee (PN Protocol 01/61) and the following regional hospital ethics committees: the Manawatu/Whanganui Ethics Committee, the Taranaki Ethics Committee, the Hawkes Bay Ethics Committee, the Bay of Plenty Ethics Committee, the Wellington Ethics Committee and the Auckland Ethics Committee.

It is also important to note that not one geneticist was included amongst the ten members on the Ministerial Advisory Group established by the Minister of Veterans Affairs at that time. This was a staggering oversight. Furthermore, only one of the referees called upon by the panel to review the cytogenetic study was a geneticist. This sole referee from Britain, an eminent scientist by the name of Dr Yuri Dubrova, supported the findings of the study, whereas the other two who did not support the findings had absolutely no experience in this area and were totally out of their depth.

Notwithstanding this lack of expertise, the Advisory Group in their conclusion advised the Minister in December 2010 that the evidence shows the nuclear test veterans had incurred genetic damage as a consequence of their participation in Operation Grapple and that the likely cause of damage was radiation exposure. The only response the veterans have ever received from successive Ministers has been a wall of silence, except for a critical review of the literature be written for a lay audience, known as the Summary of Expert Reviews (May 2013). It is a puzzle that the Executive Summary should be at odds with the recommendation of the Advisory Group.

One valid query was raised in the Summary of Expert Reviews concerning smoking, that the two groups (Exp and Con) were not comparable. One should note that both the Experimentals and the Controls were virtually identical for current smoking consumption – nearly all have been non-smokers for several years. Our analyses showed no statistical difference in translocation frequencies between the Experimentals and the Controls for smoking. In fact, in both groups never-smokers had a marginally higher total stable translocation frequency than smokers. We conclude that smoking was not a confounding factor in explaining elevated translocation frequencies in the nuclear test veterans.

Furthermore, Whitehouse et al. (2005) are most often quoted in relation to smoking and translocation frequency. They state, "No lifestyle factor, other than age, has been identified as contributing significantly to translocation yield." This important fact is conveniently overlooked in the Summary, or maybe the writer was not aware.

The Cytogenetic Study was further reviewed independently for the British High Court by the world's foremost researcher on genetic damage in humans as a consequence of radiation exposure, Dr David Brenner at Columbia University, New York. Dr Brenner is Director of the Columbia University Center for Radiological Research and holds the Higgins Chair of Radiation Biophysics which is the oldest and largest radiological research laboratory, worldwide, being founded by a student of Marie Curie around a century ago. Dr Brenner fully endorses the study and one cannot receive a higher endorsement than that.

In his submission to the High Court Dr Brenner states, "In my opinion, the Rowland mFISH study (Wahab et al, 2008) provides extremely strong evidence that the nuclear test veterans have a statistically significantly increased burden of chromosome aberrations compared to the controls. The measured aberration rates in the matched control group were what one would expect for individuals of their age – indicating that the methodology, precision and accuracy of the 2008 Rowland mFISH study was appropriate."

It is also salient to note that Dr Brenner acknowledges our valid attempt at dose reconstruction that the veterans received, a point wilfully ignored in the Executive Summary.

The British High Court judge, Judge Foskett, studied all the evidence for over two years and ruled in the veterans favour.

Dr Brenner's letter of submission is included below. This is the review that should take precedence in deciding merits of the study.

The study was published in the foremost prestigious chromosomal journal in Europe, *Cytogenetics and Genome Research 121, 79 – 87 (2008).* Authors Wahab et al.

INCLUDE DR BRENNER'S LETTER HERE

Dr R E (Al) Rowland
Principal Researcher of the Massey University Cytogenetics Study of New Zealand nuclear test veterans who participated in Operation Grapple.

25 November 2019

COLUMBIA UNIVERSITY MEDICAL CENTER
CENTER FOR RADIOLOGICAL RESEARCH
630 WEST 168TH STREET, ROOM VC11-230
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To: The High Court of Justice, Queen's Bench Division November 6 2008

Re: The Atomic Veterans Litigation

Final Report by Dr. David J Brenner

Prepared on the instructions of Rosenblatt Solicitors

I have been asked by Rosenblatt Solicitors to comment on the case between AB & others and the Ministry of Defence (Claim Number HQ04X04168), in the High Court of Justice, Queen's Bench Division. I have specifically been instructed to prepare a report for the Court on current and previous cytogenetic data on relevant nuclear test veterans. I have been informed that, while these considerations are relevant to the preliminary hearing on limitation, the Court may not actually decide in the same way that it would at a hearing of the main case. I have been told that, because of this difference of approach, this report should address only the general issues and need not be as detailed as a report that would be expected for the main case.

My name is David J. Brenner, and I am the Higgins Professor of Radiation Biophysics at Columbia University in New York City, New York. I am the Director of the Columbia University Center for Radiological Research (CRR), which is both the oldest and the largest radiological research laboratory, worldwide, being founded by a student of Marie Curie almost a century ago. The CRR currently has over 45 research scientists.

I was trained at the Universities of Oxford, Surrey, and London, with a post-doctoral fellowship at Los Alamos National Laboratory in New Mexico, USA. My field is the biological effects of radiation, both at the cytogenetic and the human level, and I have over 200 peer-reviewed publications in this field (see www.columbia.edu/~djb3). I am a past recipient of the Radiation Research Society Annual Research Award, and a member of the National Council for Radiological Protection and Measurements (NCRP).

I am currently the Principal Investigator for over $6M (per year) in US government grants in this field, including grants from both the US National Institutes of Health (NIH) and the US Department of Energy (DOE) to study the use of chromosome aberration measurements to retrospectively estimate radiation doses in potentially exposed individuals. For example the DOE-funded study involves the use of the mFISH technique (and others) to estimate radiation dose in a cohort of nuclear weapons workers who were exposed to radiation in the 1960s, in the former Soviet Union (Hande *et al* 2003).

The issue I have focused on here is the validity and utility of Rowland's mFISH study of 49 NZ nuclear test veterans *vs.* 50 matched controls (Wahab *et al.* 2008)). It is pertinent to note that this work has now been peer-reviewed and published in a well regarded scientific journal (Cytogenetics and Genomics Research).

FISH (Fluorescent In-Situ Hybridization) is a mature biophysical technique in which different chromosomes are "painted" different colors, and can be visualized using microscopy. Painting individual chromosomes allows chromosome breaks and subsequent inter-chromosomal rearrangements to be visualized; for example seeing two colors (as opposed to one) within a single chromosome is incontrovertible evidence of chromosome breakage followed by an inter-chromosomal rearrangement. The number of these color junctions can then be related to the radiation dose that produced them.

Standard FISH methodologies, as used since the 1980s, typically involves measurements of only 3 chromosomes in each examined cell, and uses that information to extrapolate the damage to all the other chromosomes in each cell. Thus, typically, only effects in about 25% of the genome are actually measured with standard FISH, which means that the effects in the other chromosomes have to be estimated (essentially guesstimated) or extrapolated, in order to produce a dose estimate. By contrast, multi-colored (some called multi-fluor) FISH, commercially known as mFISH (or its equivalent, SKY), the technique used by Rowland and his colleagues (Wahab *et al.* 2008), analyzes at all the chromosomes in each cell. The result is both improved statistical power and elimination of the need to extrapolate / guesstimate the damage to all the other chromosomes that are not measured with standard FISH.

In my opinion, the Rowland mFISH study (Wahab *et al.* 2008) provides extremely strong evidence that the nuclear test veterans have a statistically significantly increased burden of chromosome aberrations, compared to the controls. The measured aberration rates in the matched control group were what one would expect for individuals of their age -indicating that the methodology, precision and accuracy of the 2008 Rowland mFISH study was appropriate.

It is pertinent to note that, while the Rowland mFISH study (Wahab *et al.* 2008) does provide very strong evidence that the studied test veterans have increased levels of chromosome aberrations compared to the matched controls, it does not necessarily follow that the all or even part of this increase was the result of radiation exposure. Thus, on the basis of the available evidence, one cannot exclude the possibility that the all or part of the excess chromosome aberrations were produced by exposure to clastogens (agents that produce chromosomal damage) other than ionizing radiation. In particular, there is evidence that seamen are sometimes exposed to increased levels of organic chemicals (e.g. Williams *et al* 2005), which in turn have been linked to increased levels of chromosome aberrations (e.g. Lazutka *et al* 1999).

This having been said, there is no direct evidence that I know of that the nuclear veterans were, in fact, exposed to increased level of organics. This being the case, the most likely source of the increased chromosome aberration levels is radiation exposure and, if this is the case, the radiation dose estimates produced in the Rowland study (Wahab *et al* 2008) may be considered "state-of-the-art" best estimates.

I will comment briefly on the report of the noted epidemiologist, Professor John Kaldor, with regard to his comments (reproduced here) on cytogenetics-based radiation biodosimetry:

47 *A recent study (Wahab et al, 2008, that followed a preliminary report from the same group in 2005, led by Rowland as first author) found evidence that a group of people who were involved in the atomic bomb testing in the Pacific had higher levels of certain chromosomal changes than comparison people who had not been involved. As the comparison group was matched to the atomic bomb test participants in regard to age and place of residence, the authors concluded that the higher levels of chromosomal changes should be attributed to exposure to radiation. The authors also used the results of the chromosomal study to undertake a retrospective dose reconstruction and concluded that the median exposure was around 150 mSv, with the highest level being 431 mSv. I note the comments in the scientific literature that the use of these methods to reconstruct past levels of exposure to ionising radiation are still under development, and can not yet be seen as providing reliable estimates of exposure levels (Edwards et al 2005).*

48. *The Wahab study was based on a small group (50) of atomic bomb test participants, and used dose reconstruction methods that are not yet considered to be fully validated, and are unlikely to be so for the foreseeable future. Nevertheless, the results of the study raise a question about the validity of the dose levels that were measured and recorded for the participants at the time of the Pacific tests.*

I do not find these comments consistent with current developments in the field of radiation biodosimetry. I do not believe that there are credible comments in the contemporary scientific literature suggesting that the use of chromosome aberrations as a biodosimeter is "*still under development, and can not yet be seen as providing reliable estimates of exposure levels*". In particular Edwards *et al* (2005) do not make this statement in the cited reference, and in fact Edwards *et al* conclude in the quoted paper: "*In conclusion, the measurement of translocations using FISH is able to estimate the average doses to the bone marrow of an individual*"

Nor is it reasonable to suggest, as does Professor Kaldor, that the "*dose reconstruction methods ...are not yet considered to be fully validated, and are unlikely to be so for the foreseeable future*". One might point out here that both the IAEA (International Atomic Energy Agency, 2001) and ISO (International Organization for Standardization, 2004) have published and endorsed detailed calibration and validation protocols for dose reconstruction based on FISH measurements of chromosome aberrations. The approach is in fact routinely used whenever a radiation overexposure is suspected: As Dr. William Blakely, chief of Biodosimetry Research at AFRRI (Armed Forces Radiobiological Research Institute), stated in reviewing a 2001 International Conference on Low-Level Radiation Injury and Medical Countermeasures: "*Scientists at the conference agreed that chromosome analysis in metaphase spreads of mitogen-stimulated peripheral blood lymphocytes remains the radiation biomarker gold standard*" (Blakely *et al*. 2002).

In fact, the use of chromosome aberration measurements for retrospective dose reconstruction is an extremely well established technique. For example, Professor HJ Evans, then head of the MRC Clinical and Population Cytogenetics Unit in Edinburgh, published a paper in Nature in 1979 using this technique to study the past radiation exposure of nuclear dockworkers (Evans *et al* 1979). To quote from this 1979 paper "*it has become standard practice to use lymphocyte aberration frequencies as a biological dosimeter in cases of accidental in vivo exposure to radiation*". What has changed since 1979 is that the techniques to measure chromosome aberrations have become progressively more sensitive: first with the advent of (conventional) 3-chromosome FISH, as used, for example by Edwards and by Blakely (see above), and subsequently with the use of mFISH as used, for example, by Rowland *et al.*, in which all the chromosomes are measured. These progressive improvements in aberration measurement techniques allow corresponding improvements in statistical power, in terms of a decrease in the minimum detectable radiation dose.

I will comment here on the study by Phelps-Brown *et al* (1997) on cataract and stable chromosome aberrations in participants in the UK nuclear weapons testing program. This study is discussed by Thomas Lindahl in his report to the MoD (fourth bulleted point at paragraph 11; page 19). Lindahl uses the Phelps-Brown study as evidence that FISH studies have inadequate power to retrospectively assess doses in participants of nuclear weapons testing programs:

1) The Phelps-Brown study is not directly pertinent to the issue of the potential for dose reconstruction of nuclear weapons veterans, because all the participants were part of a case control study of cataract induction – in other words all the ~40 participants of the Phelps-Brown study, both the cases and the controls, were exposed nuclear veterans – the cases having cataracts and the controls not having cataracts. Thus, contrary to the suggestion of Lindahl, the Phelps-Brown study is not pertinent to the question of whether a potentially exposed population can be distinguished from an unexposed control population. Essentially the goal of the Phelps-Brown study was to investigate whether any of the study participants received doses greater than 1,300 mSv, which was considered the threshold dose for cataractogenesis – doses that are far higher than estimated in the recent mFISH study by Wahab *et al* 2008.
2) The cytogenetic methodology in the Phelps-Brown study was the older 3-chromosome FISH technique, rather than the state-of-the art mFISH technique used in the Wahab *et al* (2008) study– as discussed above, the mFISH technique has higher sensitivity to detect lower doses.

I will comment here on the links between excess chromosome aberrations and human health. Chromosome aberrations are relevant to the current issue in two related ways:

A) Measured excess chromosome aberrations are used by Rowland and colleagues (and many others, see above) as biomarkers of past exposure to radiation. Thus the link from the Rowland results to conclusions about human heath has two steps:

1. The excess chromosome aberrations measured by Rowland and colleagues provide evidence that the individuals have, in the past, been exposed to ionizing radiation, over and above natural background (in particular, a median estimated dose of ~150 mSv, with the highest dose estimate being 431 mSv)
2. There is independent evidence from large-scale epidemiological studies (in particular Japanese Atomic Bomb survivors, but also nuclear workers (Cardis *et al.* 2007)) that individuals exposed to radiation doses in this dose range have an increased lifetime risk of both cancer incidence and cancer mortality. For example, atomic bomb survivors exposed in 1945 in the dose range from 5 to 150 mSv (and followed up for many decades) show statistically-significant increased risks of both cancer incidence and cancer mortality (Preston *et al.* 2003, 2004, 2007). Atomic bomb survivors who received higher doses have proportionately higher lifetime cancer risks (Preston *et al.* 2003, 2004, 2007).

B) In addition to the relevance of chromosome aberrations as biomarkers of past exposure to radiation, there is a well established mechanistic link between chromosome aberrations and cancer. In particular, the majority of all human cancers contain one or more of the same chromosomal aberrations in virtually all the tumor cells, implying that this / these chromosome aberrations must have been present in the original damaged cell(s) from which the tumor originated. This link between chromosome aberrations and cancer has been extensively catalogued, for example by Mitelman *et al* (1997, 2008). It is important to emphasize here that there is no claim that the actual chromosome aberrations measured by Rowland and colleagues are themselves likely to be the originator(s) of a tumor – they are not, as they are measured in human lymphocytes, as opposed to stem cells which are likely to be the parental cells for a malignancy. Rather the chromosomal aberrations measured by Rowland and colleagues are biomarkers of past radiation exposure, as discussed above.

Finally, I will comment on the issue of the latency period between radiation exposure and the appearance of an associated cancer. A great deal is known about this issue, largely from the detailed follow-up studies of the Japanese atomic bomb survivors, whose cancer risk have been carefully monitored for more than half a century (Preston *et al.* 2003, 2007). What is well established from these studies is that, for solid tumors (as opposed to haematological cancers), the latency period is long, ranging from about 10 to at least 50 years. More precisely, the increased relative risk of cancer produced by a radiation exposure is generally maintained throughout the lifetime of the exposed individual. Whilst a complete mechanistic understanding of radiation-induced cancer is not yet established, the reasons for this prolonged period of increased radiation-associated risk are qualitatively understood: radiation-induced cancers originate with radiation-induced damage to stem cells, which can be passed on to progeny stem cells when they divide. Thus the radiation-induced damage can remain latent in stem cells for many years until the damaged stem cell or one of its progeny starts to divide inappropriately as a result of the damage.

I confirm that, insofar as the facts stated in my report are within my own knowledge, I have made clear which they are, and I believe them to be true, and that the opinions I have expressed represent my true and complete professional opinion.

Yours sincerely,

D. Brenner

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