Importance of

Radiation Dosimetry standards in pre-clinical radiobiology studies

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NIH NATIONAL CANCER INSTITUTE

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OUTLINE

1. Pre-clinical radiobiology studies as basis for new tx technologies.

2. Current challenges on reproducible and translatable expt'al results.

3. Some examples on how we might proceed:

    i) Modern medical linac beam dosimetry
    ii) Modern dicentric assay for biodosimetry

1. Pre-clinical radiobiology studies as basis for new tx technologies.

Some examples of NEW radiation treatment technologies:

- *Targeted radionuclide therapy*
- *Radiation Immunotherapy*
- *Flash RT*
- *Proton and heavy-ion particle therapy*

frontiers in Oncology

REVIEW
published: xx April 2018
doi: 10.3389/fonc.2018.00130

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Radiation Oncology in the 21st Century: Prospective Randomized Trials That Changed Practice… or Didn't!

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OPEN ACCESS

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Chandan Guha,
Albert Einstein College of

In a two-part article published in 2009, we discussed the limitations of conventional radiation therapy, the challenges of studying new technologies in radiation oncology, and summarized the state-of-the science for various malignancies (1, 2). Here, we summarize some of the most important prospective, randomized trials that during the intervening years have attempted to improve the tumor control and/or decrease the adverse effects of radiation therapy. For consistency, we have focused here on the null and alternate hypotheses as articulated by the investigators at the onset of each trial, since the outcome of the investigational treatment should be considered clinically significant only if the null hypothesis was rejected. The readers (and patients) are of course free to make their own judgments about the clinical significance of the results when the null hypothesis was not rejected.

TABLE 1 | Current state of the science by anatomic site.

Type of cancer	Trial arms	Null hypothesis	Trial outcomes
Glioblastoma	Surgery, radiation, and chemotherapy with or without tumor-treating fields (TTF) (3)	Adding TTF would not prolong PFS	Median PFS 7.1 (HR = 0.62; $p = 0.001$), median survival 19.6 mont (HR = 0.64; $p = 0.004$) Death in 57% at 2 years Gr 3/4 nervous system toxicity 22% Gr 3/4 hematologic toxicity 12% No increase in Gr 3+ toxicity with TTF but increase in mild-to-moderate skin irritation
Anaplastic oligodendroglioma	Surgery and radiation with or without PCV chemotherapy (4)	PCV would not prolong overall survival (OS)	Median survival 4.6 vs 4.7 years Median survival was longer in codeleted tumors treated with PCV (14.7 vs 7.3 years; HR = 0.59; $p = 0.03$) Gr 3/4 toxicity in 65% (most common: hematologic, neurologic, and Fatal chemotherapy induced neutropenia in 1%
	Surgery and radiation with or without PCV chemotherapy (5)	PCV would not prolong OS by 12 months or longer	PCV prolonged median OS by 11.7 months: 42.3 vs 30.6 months; HR = 0.75; $p = 0.018$
Anaplastic glioma, non-codeleted	Surgery followed by 2 × 2 randomization to radiation with or without temozolomide and with or without adjuvant temozolomide (6)	Concurrent or adjuvant temozolomide would not prolong OS	Adjuvant temozolomide improved 5-year survival (55.9% vs 44.1%; HR = 0.65; $p = 0.0014$) Gr 3/4 toxicity in 8–12% with temozolomide
Low-grade glioma	Surgery and radiation with or without PCV chemotherapy (7)	OS would not be improved with PCV	Median survival 13.3 years (HR = 0.59; $p = 0.003$) Death in 28% at 5 years Any grade late events due to radiation in 22%
Brain metastases	Radiosurgery with or without WBRT (8)	Cognitive deterioration at 3 months would not be less after radiosurgery alone	Cognitive deterioration at 3 months improved with radiosurgery: 63.5 vs 91.7% ($p < 0.001$) No difference in survival (10.4 vs 7.4 months)

2. Current challenges on reproducible and translatable experimental results.

Open access, freely available online

Essay

Why Most Published Research Findings Are False

John P. A. Ioannidis

Summary

There is increasing concern that most current published research findings are false. The probability that a research claim is true may depend on study power and bias, the number of other studies on the same question, and, importantly, the ratio of true to no relationships among the relationships probed in each scientific field. In this framework, a research finding is less likely to be true when the studies conducted in a field are smaller; when effect sizes are smaller; when there is a greater number and lesser preselection of tested relationships; where there is greater flexibility in designs, definitions, outcomes, and analytical modes; when there is greater financial and other interest and prejudice; and when more teams are involved in a scientific field in chase of statistical significance. Simulations show that for most study designs and settings, it is more likely for a research claim to be false than true. Moreover, for many current scientific fields, claimed research findings may often be simply accurate measures of the prevailing bias. In this essay, I discuss the implications of these problems for the conduct and interpretation of research.

factors that influence this problem and some corollaries thereof.

Modeling the Framework for False Positive Findings

Several methodologists have pointed out [9–11] that the high rate of nonreplication (lack of confirmation) of research discoveries is a consequence of the convenient, yet ill-founded strategy of claiming conclusive research findings solely on the basis of a single study assessed by formal statistical significance, typically for a *p*-value less than 0.05. Research is not most appropriately represented and summarized by *p*-values, but, unfortunately, there is a widespread notion that medical research articles

It can be proven that most claimed research findings are false.

should be interpreted based only on *p*-values. Research findings are defined here as any relationship reaching formal statistical significance, e.g., effective interventions, informative predictors, risk factors, or associations. "Negative" research is also very useful.

is characteristic of the field and can vary a lot depending on whether the field targets highly likely relationships or searches for only one or a few true relationships among thousands and millions of hypotheses that may be postulated. Let us also consider, for computational simplicity, circumscribed fields where either there is only one true relationship (among many that can be hypothesized) or the power is similar to find any of the several existing true relationships. The pre-study probability of a relationship being true is $R/(R + 1)$. The probability of a study finding a true relationship reflects the power $1 - \beta$ (one minus the Type II error rate). The probability of claiming a relationship when none truly exists reflects the Type I error rate, α. Assuming that c relationships are being probed in the field, the expected values of the 2×2 table are given in Table 1. After a research finding has been claimed based on achieving formal statistical significance, the post-study probability that it is true is the positive predictive value, PPV. The PPV is also the complementary probability of what Wacholder et al. have called the false positive report probability [10]. According to the 2

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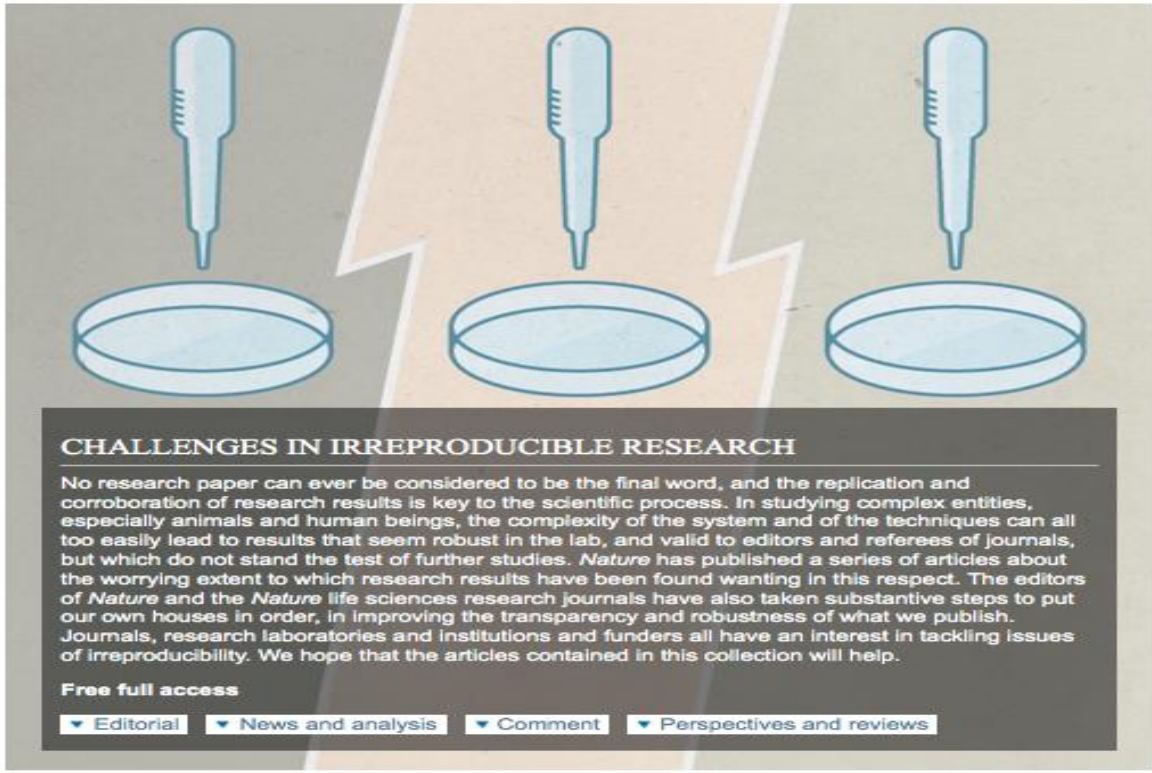

CHALLENGES IN IRREPRODUCIBLE RESEARCH

No research paper can ever be considered to be the final word, and the replication and corroboration of research results is key to the scientific process. In studying complex entities, especially animals and human beings, the complexity of the system and of the techniques can all too easily lead to results that seem robust in the lab, and valid to editors and referees of journals, but which do not stand the test of further studies. *Nature* has published a series of articles about the worrying extent to which research results have been found wanting in this respect. The editors of *Nature* and the *Nature* life sciences research journals have also taken substantive steps to put our own houses in order, in improving the transparency and robustness of what we publish. Journals, research laboratories and institutions and funders all have an interest in tackling issues of irreproducibility. We hope that the articles contained in this collection will help.

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EDITORIAL

Reducing our irreproducibility
Nature **496**, 398 (25 April 2013)

Further confirmation needed
A new mechanism for independently replicating research findings is one of several changes required to improve the quality of the biomedical literature.
Nature Biotechnology **30**, 806 (10 September 2012)

Error prone
Biologists must realize the pitfalls of work on massive amounts of data.
Nature **487**, 406 (26 July 2012)

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3rd Max Planck Freiburg Epigenetics Meeting
03 December 2014 — 05 December 2014
Stuebeweg 51, Freiburg im Breisgau, Germany

Upstream and Downstream of Hox Genes
14 December 2014 — 17 December 2014
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Nature | 17 September 2014

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Nature | 20 June 2012

Published in final edited form as:
Nature. 2014 January 30; 505(7485): 612–613.

NIH plans to enhance reproducibility

Francis S. Collins [director] and **Lawrence A. Tabak [principal deputy director]**
US National Institutes of Health, Bethesda, Maryland, USA

Lawrence A. Tabak: lawrence.tabak@nih.gov

Abstract

Francis S. Collins and Lawrence A. Tabak discuss initiatives that the US National Institutes of Health is exploring to restore the self-correcting nature of preclinical research.

A growing chorus of concern, from scientists and laypeople, contends that the complex system for ensuring the reproducibility of biomedical research is failing and is in need of

stages, utilizing grant mechanisms that allow more flexibility and a longer period than the current average of approximately four years of support per project.

"Efforts by the NIH alone will not be sufficient to effect real change in this unhealthy environment."

In addition, the NIH is examining ways to anonymize the peer-review process to reduce the effect of unconscious

NIH-PA Author Manuscript

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Then there is the problem of what is not published. There are few venues for researchers to publish negative data or papers that point out scientific flaws in previously published work. Further compounding the problem is the difficulty of accessing unpublished data — and the failure of funding agencies to establish or enforce policies that insist on data access.

PRECLINICAL PROBLEMS

Reproducibility is potentially a problem in all scientific disciplines. However, human clinical trials seem to be less at risk because they are already governed by various regulations that stipulate rigorous design and independent oversight — including randomization, blinding, power estimates, pre-registration of outcome measures in standardized, public databases such as ClinicalTrials.gov and oversight by institutional review boards and data safety monitoring boards. Furthermore, the clinical trials community has taken important steps towards adopting standard reporting elements[7].

Preclinical research, especially work that uses animal models[1], seems to be the area that is currently most susceptible to reproducibility issues. Many of these failures have simple and practical explanations: different animal strains, different lab environments or subtle changes in protocol. Some irreproducible reports are probably the result of coincidental findings that happen to reach statistical significance, coupled with publication bias. Another pitfall is overinterpretation of creative 'hypothesis-generating' experiments, which are designed to uncover new avenues of inquiry rather than to provide definitive proof for any single question. Still, there remains a troubling frequency of published reports that claim a significant result, but fail to be reproducible.

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Proposed Principles and Guidelines for Reporting Preclinical Research

- NIH held a workshop in June 2014 with Nature Publishing Group and Science on this issue and developed a consensus
  - A number of journals have endorsed the consensus developed in this workshop
- Rigorous statistical analysis – Information to authors
- Transparency in reporting – generous or no limit to length of methods sections
  - Use of standards, replicates, statistics, randomization, blinding of samples, sample-size estimation, Inclusion and exclusion criteria
- Data and material sharing
- Consideration of refutations of a paper
- Consider establishing best practice guidelines for reporting:
  - Image based data, antibodies, cell lines, animals

(http://www.nih.gov/about/reporting-preclinical-research.htm)

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Varian to equip New York proton consortium with ProBeam system
Location: New York City
Consortium: MSKCC , Mt Sinai, Montefiore , Opening date: end of 2018
Cost: $ 300 M Maintenance : $ 120 M/10 yrs

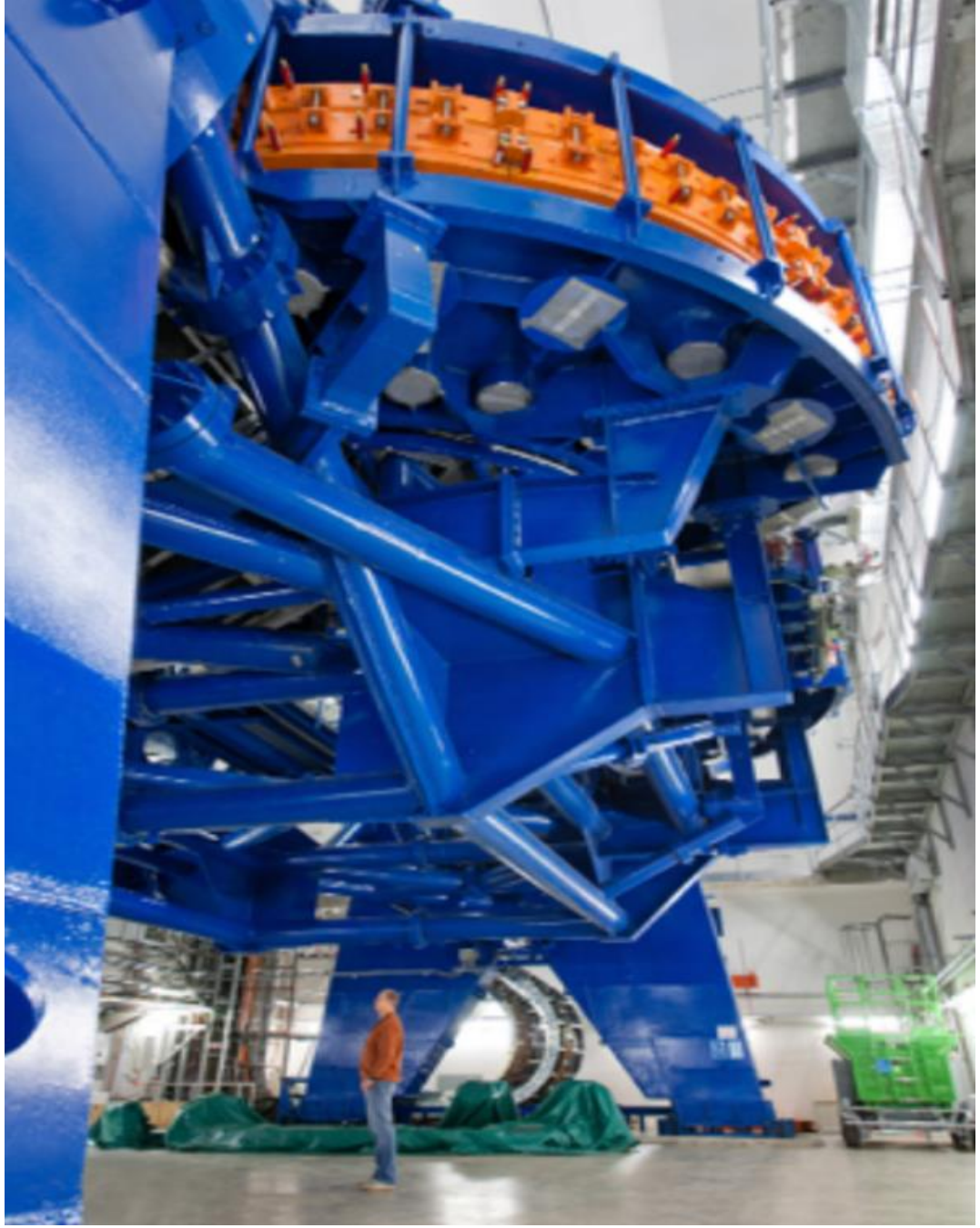
JOURNAL OF CLINICAL ONCOLOGY | EDITORIAL

February 2018

What Happens When Proton Meets Randomization: Is There a Future for Proton Therapy?

In contrast to the largest retrospective study of patients from the National Cancer Database,[10] this prospective randomized study failed to prove superiority of proton therapy. Instead, the PSPT arm had 10.5% grade ≥ 3 RP compared with only 6.5% in the IMRT arm, despite a significant reduction in low-dose volume in the dosimetric histograms for the PSPT arm. Significant dosimetric sparing of the heart and esophagus in the proton arm was found. The primary study


PROTON THERAPY CENTERS
= In Operation
= Under Construction or in Development
= Expanding
About Our Members



Carbon-ion Therapy Center in Heidelberg (H.I.T)

Characterization of the Physical Parameters of Particle Beams for Biological Research

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*manuscript submitted for publication

Table 4. A summary of recommended parameters, methods, reporting and desired accuracy

Parameter	Method/Source	Reporting	*Desired Accuracy	References
Irradiation technique	Facility report	For each experiment. (examples: scanned, scattered, micro-beam)	N/A	
Fluence Calibration	NIST traceable Ion Chamber + Monte Carlo, plastic scintillator for low dose experiments	At beginning of experiment, ion species change or machine down. For experiments with <100mGy exposure, consider plastic scintillator	+/-2% of total tracks	1
Physical Dose	Ion Chamber	Relative dosimetry including off axis dose. Chamber should be inter-comparable between centers. Comparisons between EGG at field center for absolute dose and traceable ion chamber to obtain calibration.	+/-2%	2-4
Time structure of the beam	Facility's Accelerator report	Once per data taking period	Only down to biological time scales relevant to experiments	5
Fluence on multiple time scales (#particles/area	**Ion Chamber + MC**	**For each relevant sample and all time scales relevant to biology/chemistry of experiment. MC may be used to determine values in the experimental setup**	**+/-5% of total tracks in the area and time unit**	**1**

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Seminars in
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Current Instrumentation and Technologies in Modern Radiobiology Research—Opportunities and Challenges

Eric Ford, PhD, FAAPM,* and Jim Deye, PhD†

There is a growing awareness of the gaps in the technical methods employed in radiation biology experiments. These quality gaps can have a substantial effect on the reliability and reproducibility of results as outlined in several recent meta-studies. This is especially true in the context of the newer laboratory irradiation technologies. These technologies allow for delivery of highly localized dose distributions and increased spatial accuracy but also present increased challenges of their own. In this article, we highlight some of the features of the new technologies and the experiments they support; this includes image-guided localized radiation systems, microirradiator systems using carbon nanotubes and physical radiation modifiers like gold nanoparticles. We discuss the key technical issues related to the consistency and quality of modern radiation biology experiments including dosimetry protocols that are essential to all experiments, quality assurance approaches, methods to validate physical radiation targeting including immunohistochemical assays and other biovalidation approaches. We highlight the future needs in terms of education and training and the creation of tools for cross-institutional benchmarking quality in preclinical studies. The demands for increased experimental rigor are challenging but can be met with an awareness and a systematic approach which ensures quality.

Semin Radiat Oncol 26:349-355

Table 1: Percent of sampled articles reporting items of information per NIST guidelines.

Category	Item	% of articles including item
Absolute Dosimetry/ Calibration	Published Standards Used	6.9
	Detector Type Used	3.4
Determination of Dose	Published Standards Used	10.3
	Specification of Medium	6.9
Radiation Source Specification	Detector Type Used	27.6
	Radioisotope	86.2
	kV, Filtration, HVL	50.0
Details of the Irradiation	Animal/Cell Type	100
	Dose Details	100
	Field Size and Shape	0
	Geometry of Fields	24.1

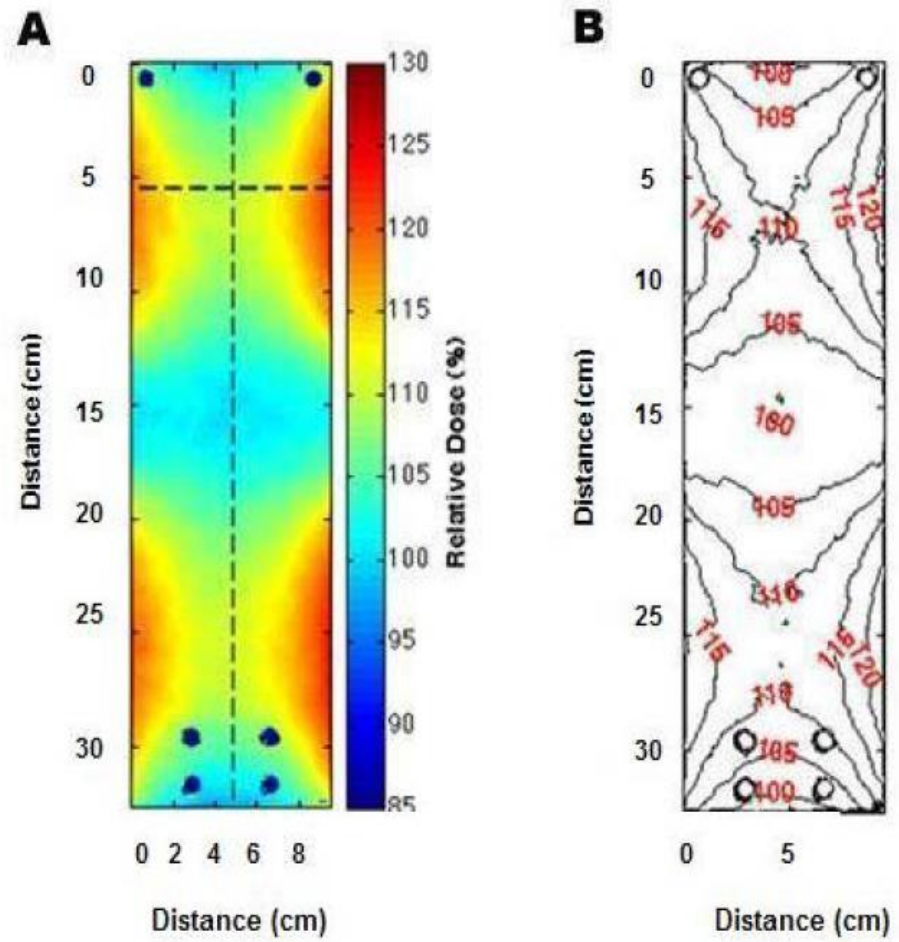


Fig. 4. Dose variation in a Cs-137 irradiator shown with a) dose color wash and b) isodose mapping of the +25 % and −15 % variation in dose throughout the irradiation volume [2].

1) observed that "the x-ray energy spectrum produced at a peak voltage of 50 kV and with added Al filters readily undergoes attenuation by the plastic tissue-culture Petri-dish covers or the culture media. For example, using a beam hardened with 0.18 mm of Al, the attenuation due to the medium can be as high as 60 % and the plastic cover will reduce the beam an additional 15 %." Manufacturer-supplied calibrations for a number of commercially-available irradiators have been found to differ by +5 % to −13 % from their true values with variations in dose rate over irradiation volumes from 70 % to 180 % of the stated value. (Ref. [8] and also Fig. 4.)

Documented Errors seen using Mouse Phantoms

Cs137	
Institution	% Error
A	10.6
B	8.4
C	3.8
D	12.6
E	1.6
F	3.0
Avg	6.7
% Std Dev	67.3

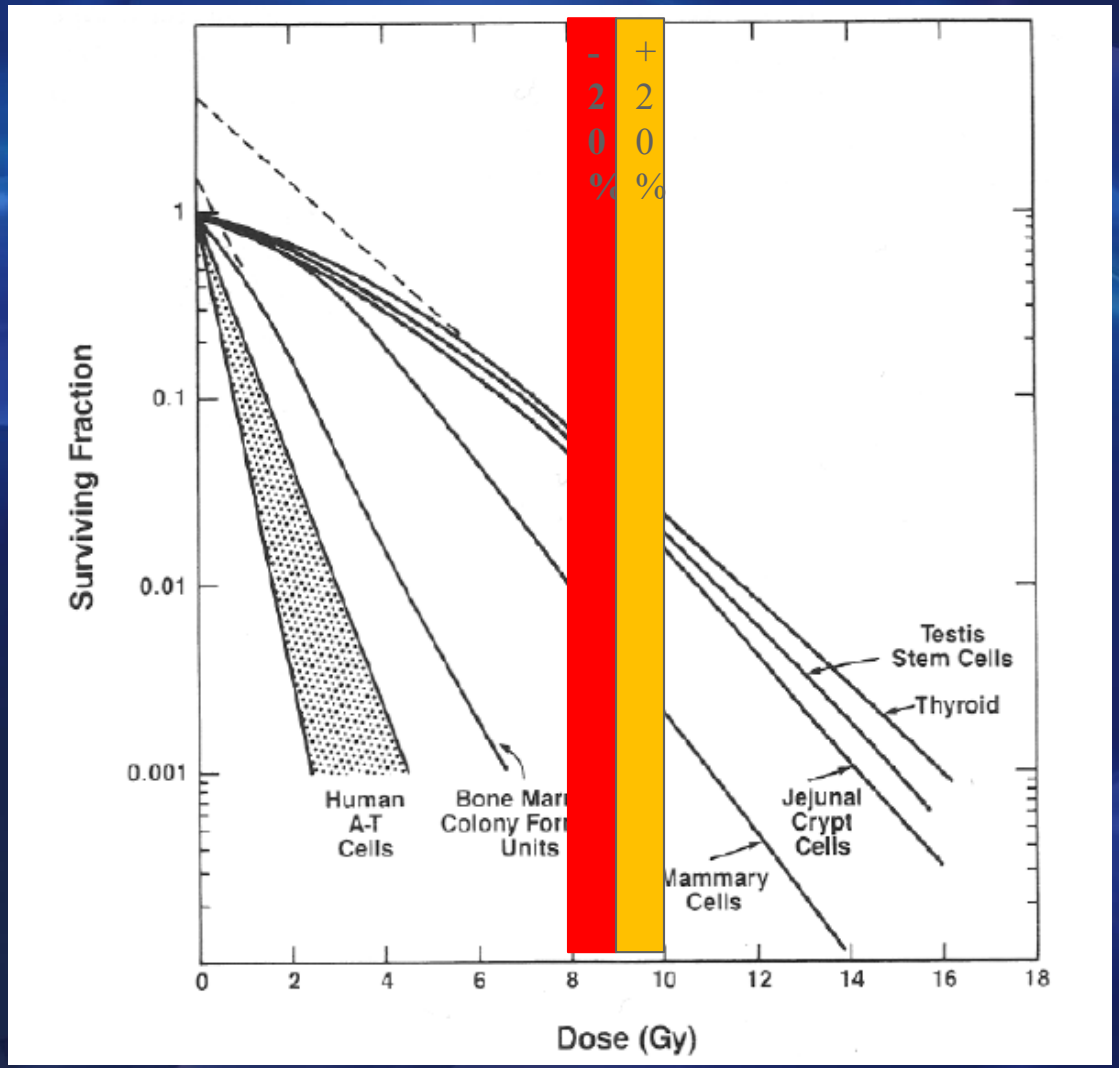
Xray	
Institution	% Error
G	-17.0
H	-53.6
I	-0.9
J	-17.1
K	-24.1
Avg	-22.5
% Std Dev	-85.8

Co60	
Institution	% Error
L	1.8
Avg	1.8
% Std Dev	0.00

Each Institution is given a total of 6 Mouse Phantoms. 3 Phantoms are to be given an Absorbed Dose to Water of 1Gy and 3 Phantoms are to be given an ADW of 4Gy. The "% Error" reported above is an average of the percent difference between the target dose (1 & 4 Gy) and the measured dose for all 6 Phantoms from each individual Institution.

Courtesy: Jim Dye

Dosimetry



Courtesy: Jim Dye

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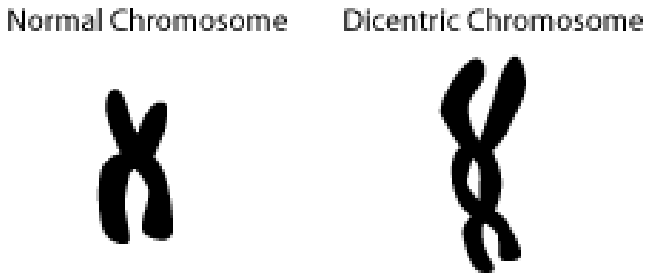
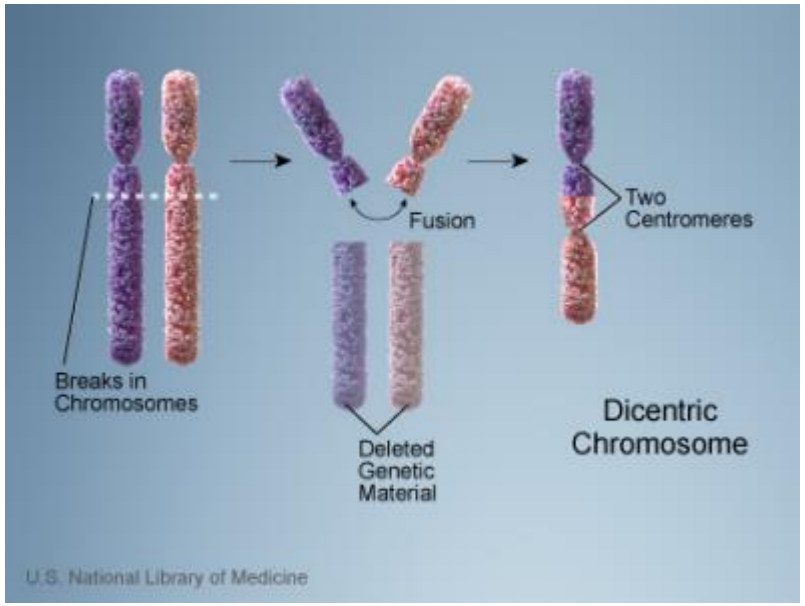
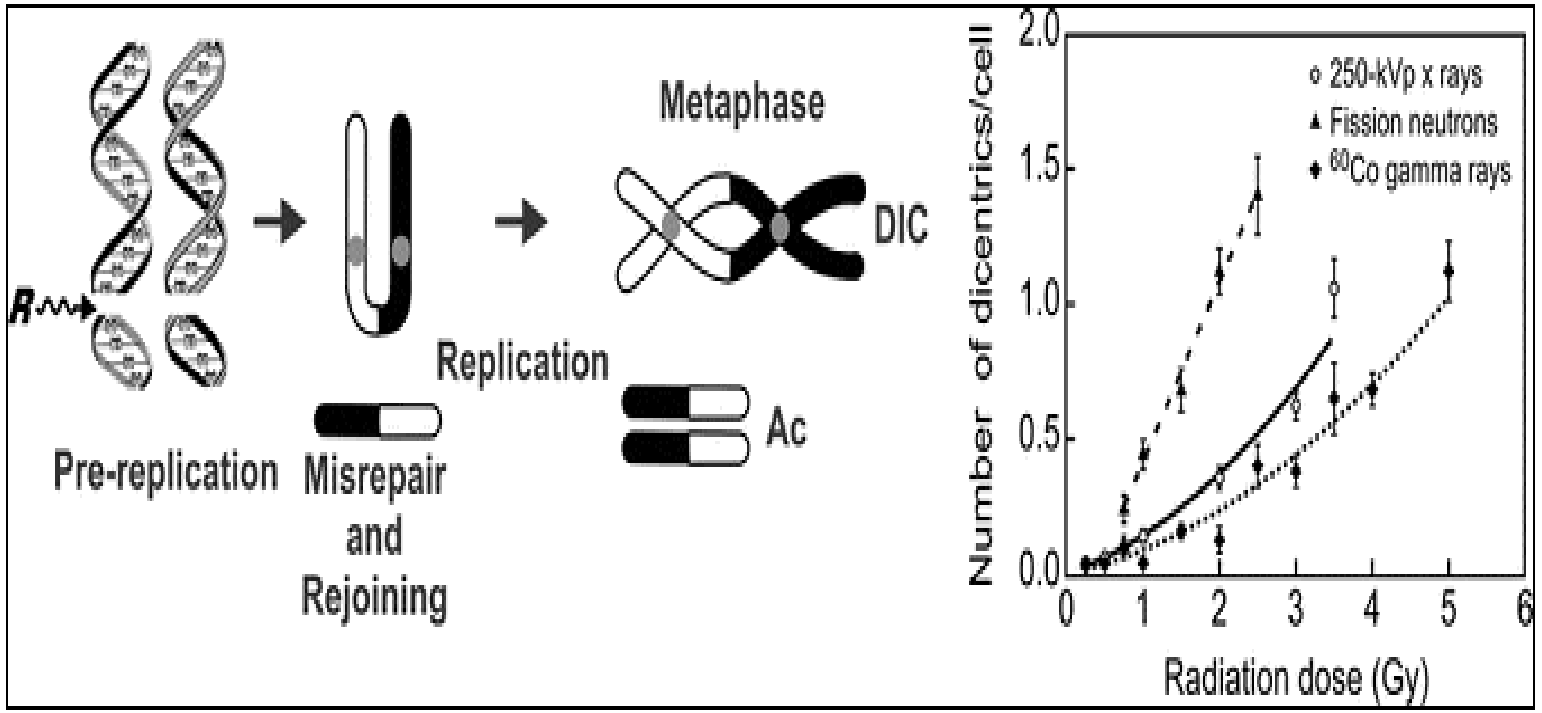
3. Some examples on how we might proceed:

   i) Modern dicentric assay for biodosimetry
   ii)Modern medical linac beam dosimetry



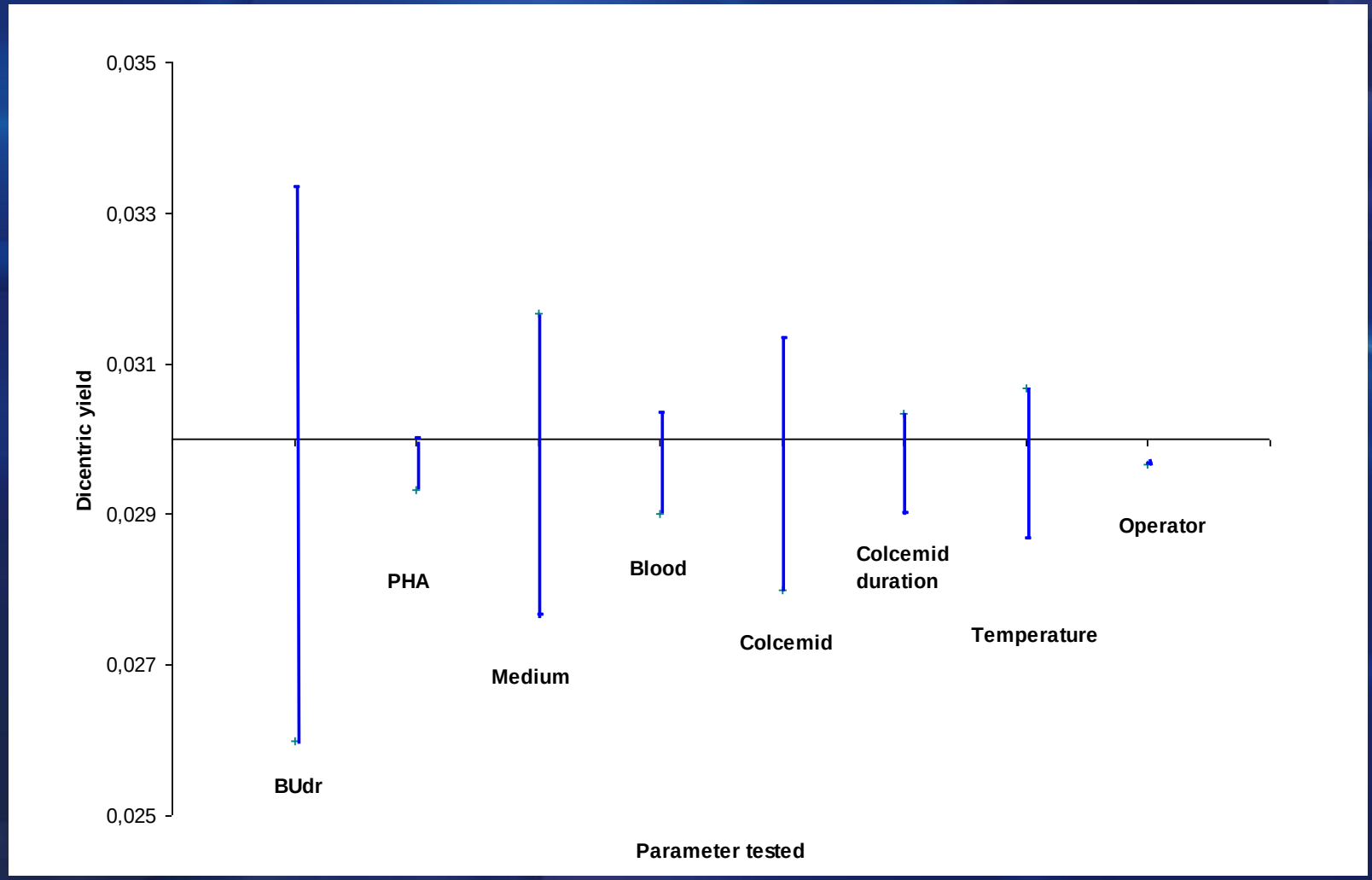




Notes on the graph above:

•**Left panel:** A schematic illustrating radiation-induced DNA damage (see left side of image) in an interphase cell and the resulting formation of radiation-induced dicentric (DIC) and accompanying acentric fragment (Ac) chromosome aberration in lymphocytes arrested in metaphase mitosis.

What Contributes to Variability?



Courtesy: Voisin, IRSN, France

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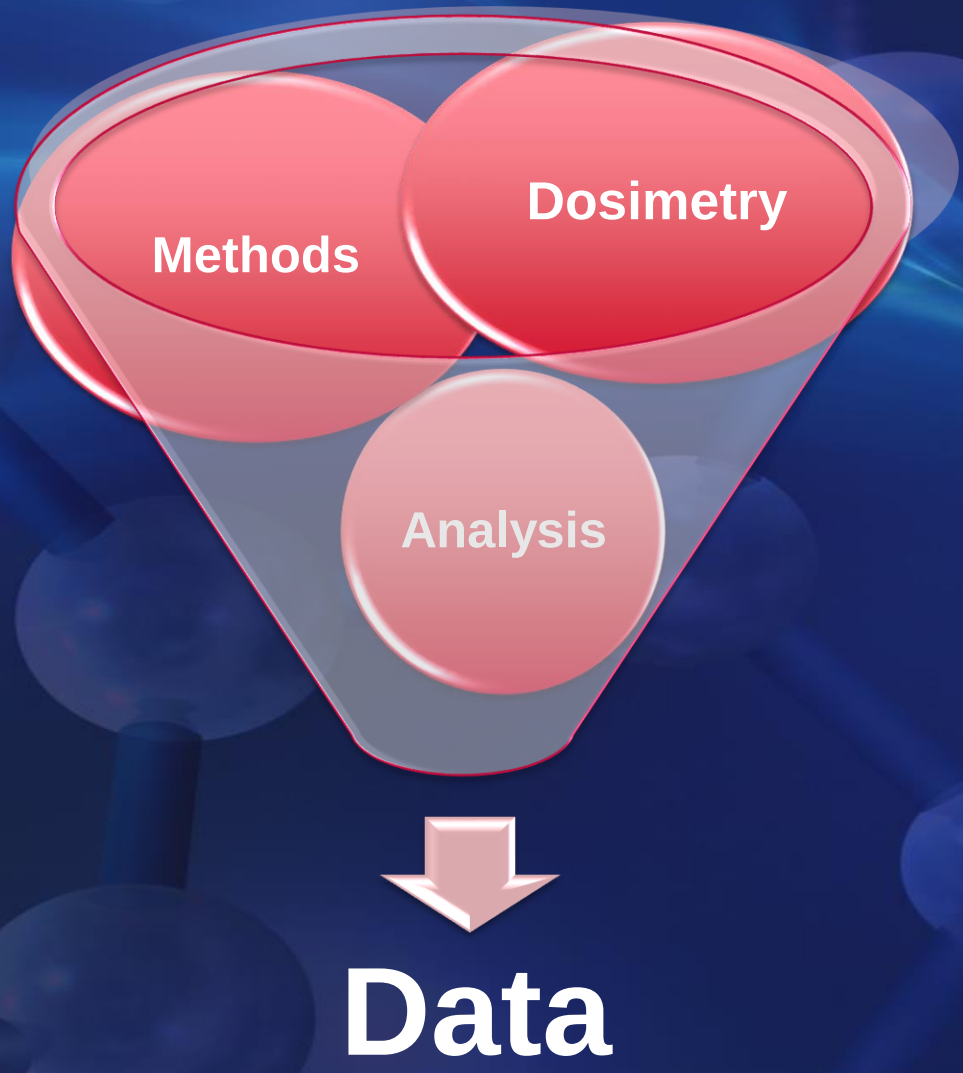
Important Variables That Influence Data



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Inter-Laboratory Comparison Studies

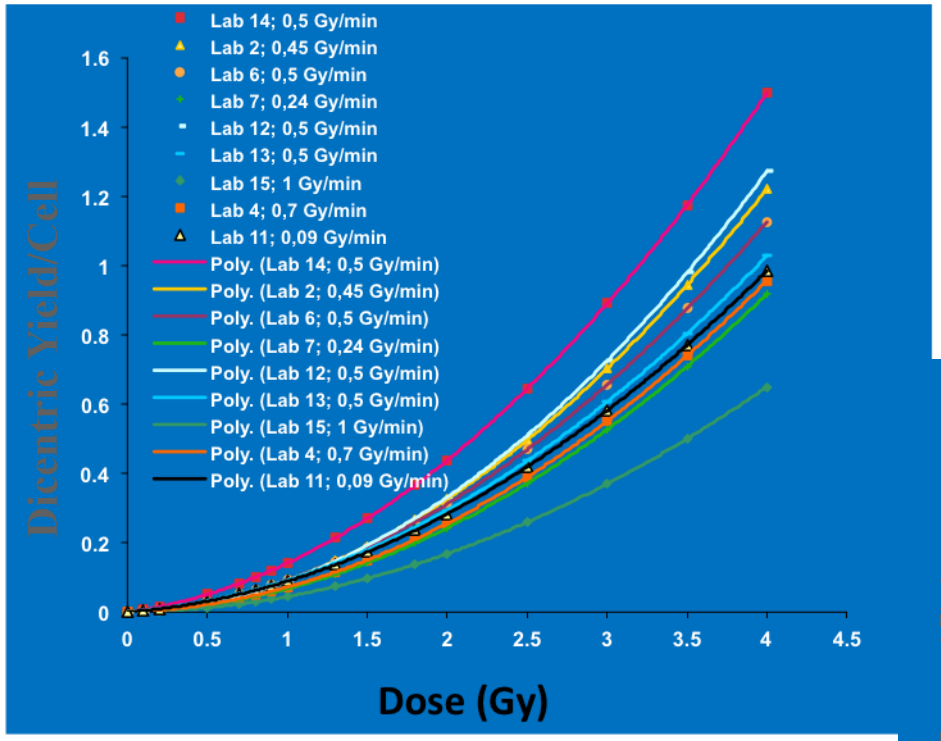
RADIATION RESEARCH 169, 551–560 (2008)
0033-7587/08 $15.00

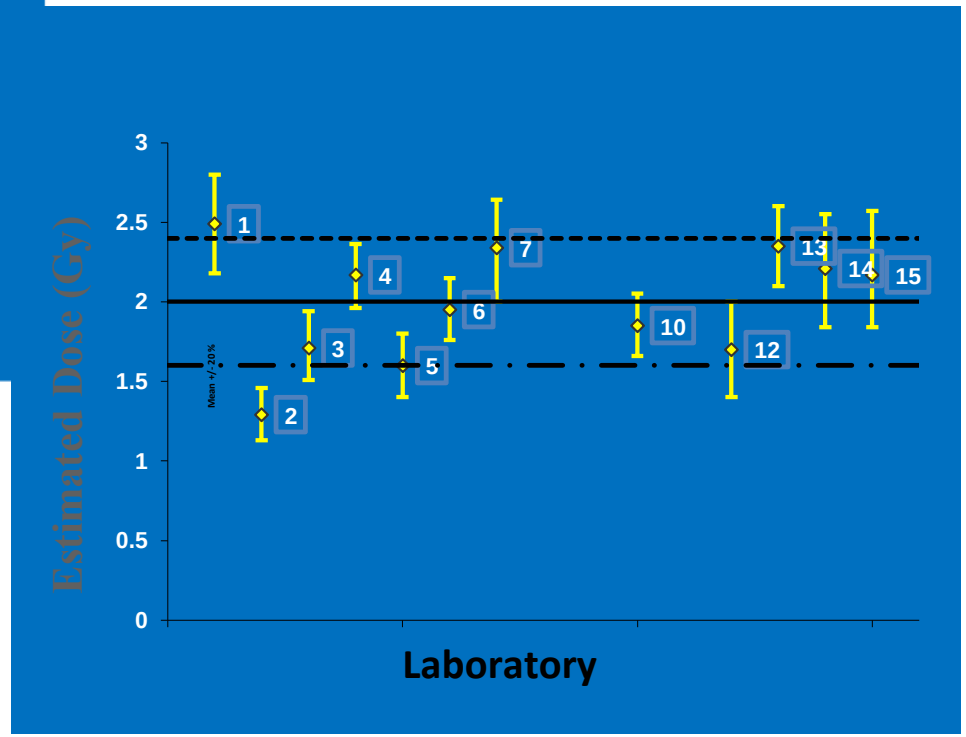
Interlaboratory Comparison of the Dicentric Chromosome Assay for Radiation Biodosimetry in Mass Casualty Events

Ruth C. Wilkins,[a] Horst Romm,[b] Tzu-Cheg Kao,[c] Akio A. Awa,[d] Mitsuaki A. Yoshida,[e] Gordon K. Livingston,[d] Mark S. Jenkins,[d] Ursula Oestreicher,[b] Terry C. Pellmar[f] and Pataje G. S. Prasanna[f,1]

[a] Health Canada, Consumer and Clinical Radiation Protection Bureau, Ottawa, ON K1A 1C1, Canada; [b] Bundesamt für Strahlenschutz, 38226 Salzgitter, Germany; [c] Uniformed Services University of the Health Sciences, Department of Preventive Medicine and Biometrics, Division of Epidemiology and Biostatistics, Bethesda, Maryland 20814-4799; [d] Oak Ridge Associated Universities, REAC/TS, Oak Ridge, Tennessee 37831; [e] National Institute of Radiological Sciences, Research Center for Radiation Emergency Medicine, 4-9-1 Anagawa, Inage-Ku, Chiba, Japan 263-8555; and [f] Uniformed Services University of the Health Sciences, Armed Forces Radiobiology Research Institute, Bethesda, Maryland 20889-5603

2002 International Inter-Laboratory Inter-Comparison Study





Courtesy: Voisin, IRSN, France

Inter-Laboratory Comparison Studies

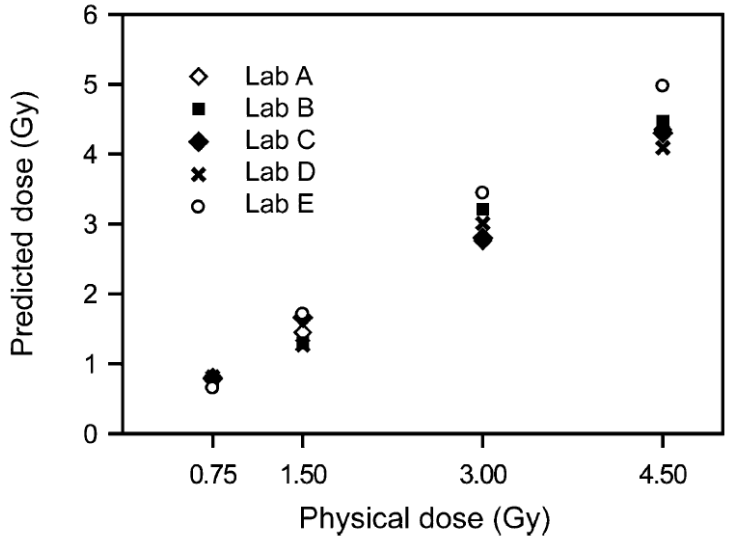


FIG. 2. Distribution of predicted biological doses to dose-blinded samples for actual physical doses in all laboratories.

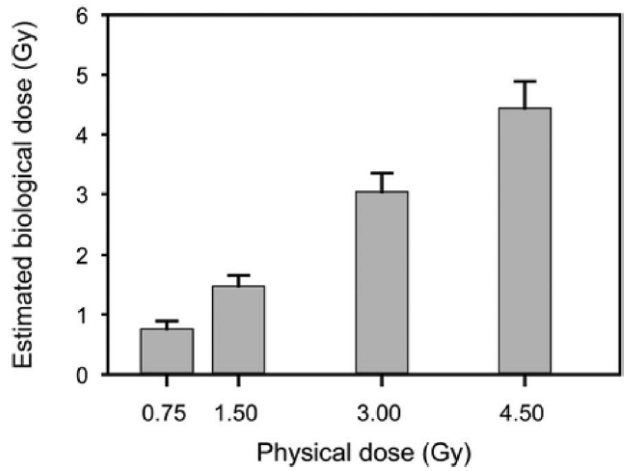
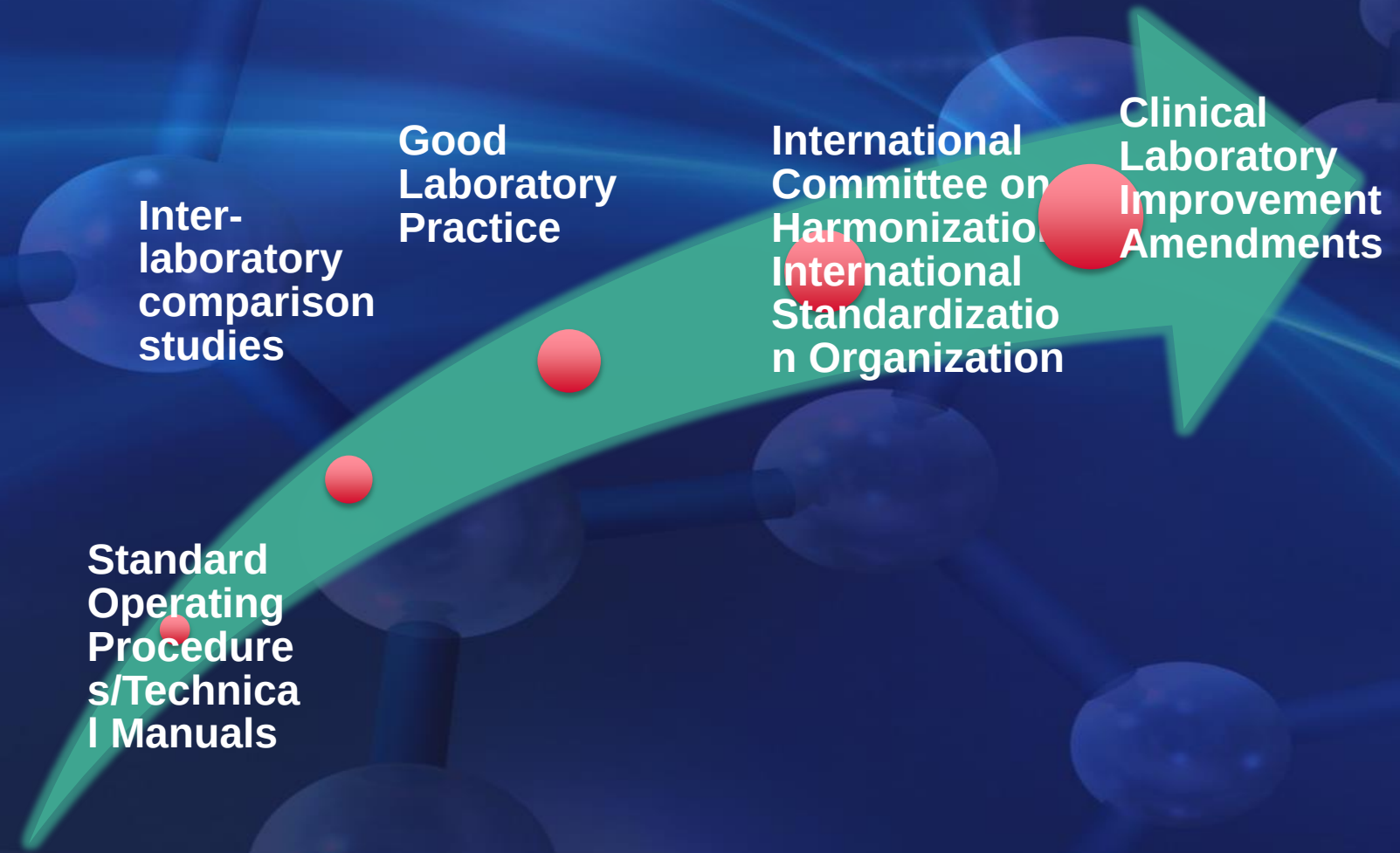


FIG. 3. Dose prediction accuracy of the dicentric chromosome assay. Means of predicted biological doses (Gy) for dose-blinded samples in laboratories A–E for actual physical doses (Gy). The error bars represent the standard deviations (SD).

Wilkins et al. 2008

Avenues for Improving Quality Control and Quality Assurance



Note: Stringent implementation of regulations will hinder assay/protocol development and innovation. Therefore, optimum balance between innovation and quality is key to success

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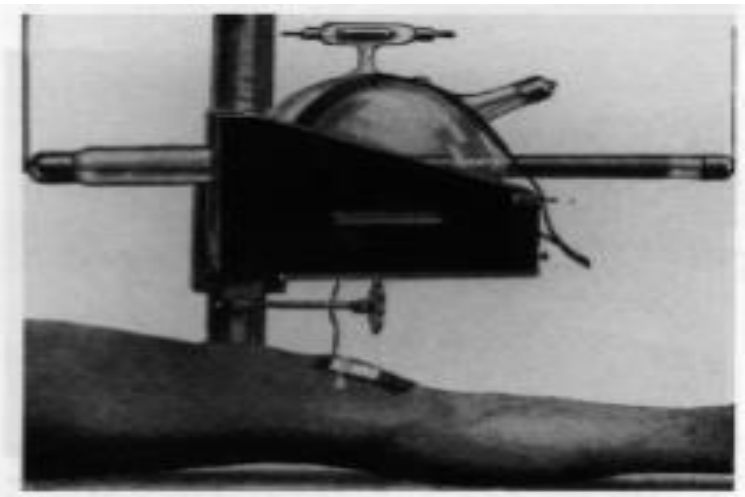
History



- First attempt at establishing a radiation dose limit – Skin Erythema Dose – 1920
- 200 mR/day dose limit established in 1931
- 25,000 mrem/year established during WWII.

Today's radiation dose limits:
5 R/yr, 0.1R/yr to non-radiation workers.

Monthly Tests

TG40 → TG142

TG40

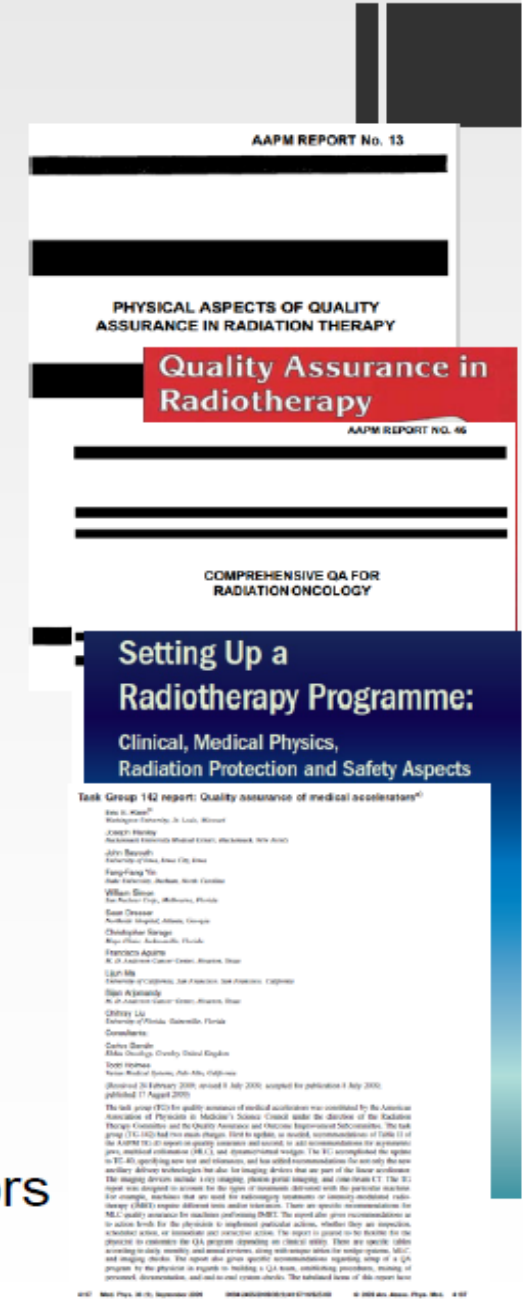
- X-Ray Output
- Electron Output
- Backup Monitor Chamber
- Electron Energy
- Xray Energy
- Xray Beam Flatness
- Electron Beam Flatness
- Xray Beam Symmetry
- Electron Beam Symmetry
- Light/Rad Field
- Gantry/Collimator Indicators
- Wedge position
- Tray position
- Applicator Position
- Field Size Indicators
- Jaw Symmetry
- Cross Hair Centering
- Treatment Couch Position Indicators
- Latching of Wedges, Blocking tray
- Emergency Off Switches
- Wedge, Cone Interlocks
- Field Light Intensity

TG142

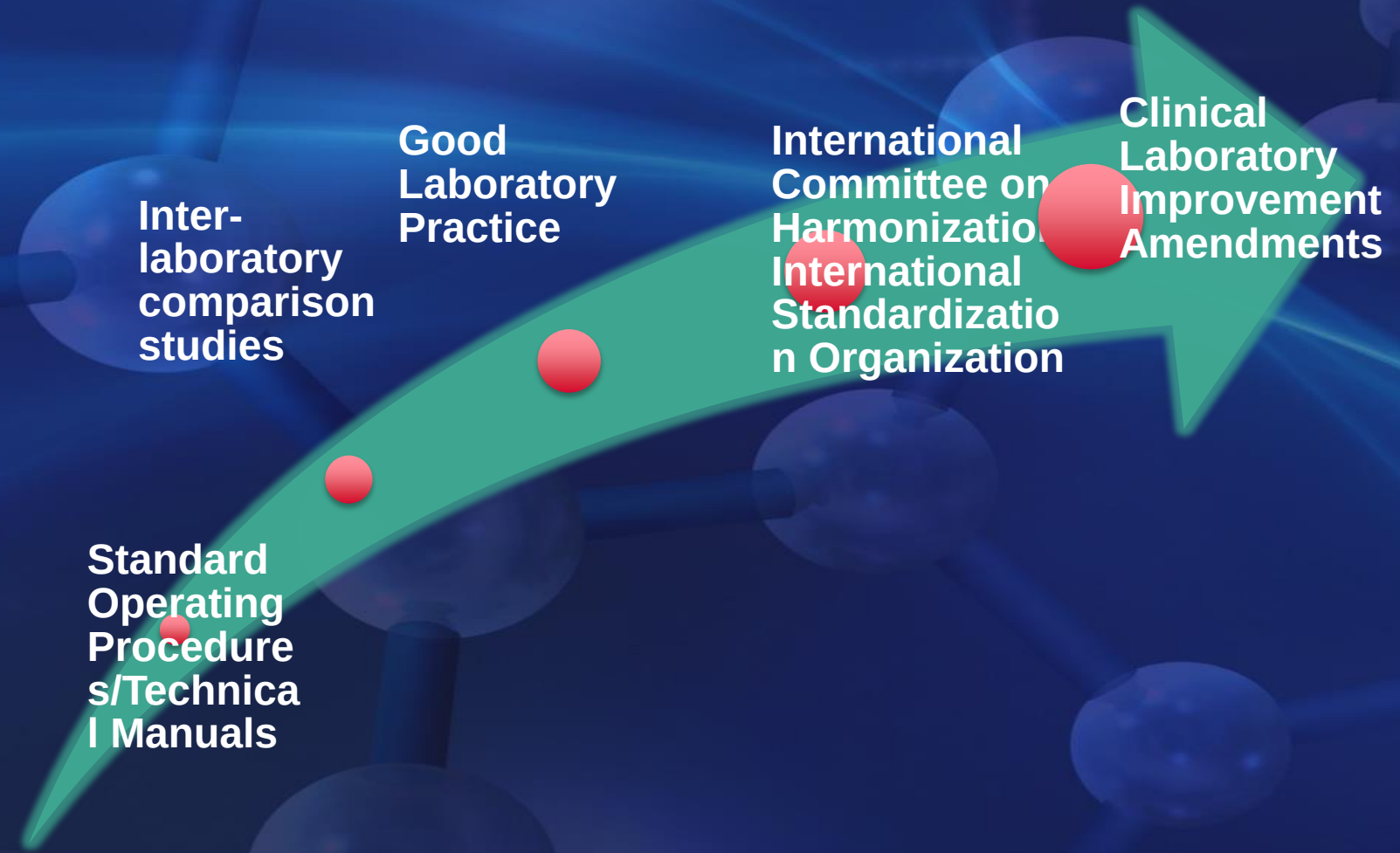
- X-Ray Output
- Backup Monitor Chamber
- Electron Energy
- Xray Profile Constancy
- Electron Profile Constancy
- Light/Rad Field (Sym)
- Light/Rad Field (Asym)
- Gantry/Collimator Indicators
- Wedge Placement
- Accessory Trays
- Jaw Position Indicators (Sym)
- Jaw Position Indicators (Asym)
- Cross Hair Centering
- Treatment Couch Position Indicators
- Latching of Wedges, Blocking Trays
- Lasers/ODI w/ Front Pointer
- Lasers
- Laser Guard Interlock Test
- Wedge Factor for All Energies
- [MLC] Setting vs Radiation Field
- [MLC] Backup Diaphragms (Elekta)
- [MLC] Travel Speed
- [MLC] Leaf Position Accuracy
- Compensatory Placement
- [Respiratory Gating] Beam Output
- [Respiratory Gating] Phase, Amplitude
- [Respiratory Gating] In Room Respiratory Monitoring
- [Respiratory Gating] Gating Interlock

Numerous Publications on Quality Assurance Tests for Linear Accelerators

- AAPM TG24, Physical Aspects of Quality Assurance in Radiotherapy (1984)
- World Health Organization, Quality Assurance in Radiotherapy (1988)
- AAPM TG40, Comprehensive QA for Radiation Oncology (1994)
- IAEA, Setting Up a Radiotherapy Program (2008)
- AAPM TG142, Quality Assurance of Medical Accelerators (2009)



Avenues for Improving Quality Control and Quality Assurance



Note: Stringent implementation of regulations will hinder assay/protocol development and innovation. Therefore, optimum balance between innovation and quality is key to success

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Acknowledgement:

Many thanks to my colleagues Pat Prasanna and Jim Deye for discussions, slides and references.