

Scientific Misconduct Revisited: Stories and Lessons Learned

Taipei Medical University
March 3, 2023



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有獎徵答 1 (10-dollar NTD)

Piero Anversa (Harvard): Cheating on heart stem cell. Consequence(s)?

(1) 5 years in prison

(2) Fined US\$5M

(3) Fined US\$10M

(4) Fined US\$20M

(5) Scot-free



有獎徵答 2 (10-dollar NTD)

Dong Pyou Han (formerly Iowa State)

Caught cheating on HIV vaccine

(1) 5 years in prison

(2) US\$2M

(3) US\$5M

(4) US\$7.5M

(5) Got out scot-free



有獎徵答 3 (10-dollar NTD)

German experimental physicist;
Done incredible experiments;
Always published in top
journals.



***How often did he publish
his papers?***

NEWEST

World Record

How many

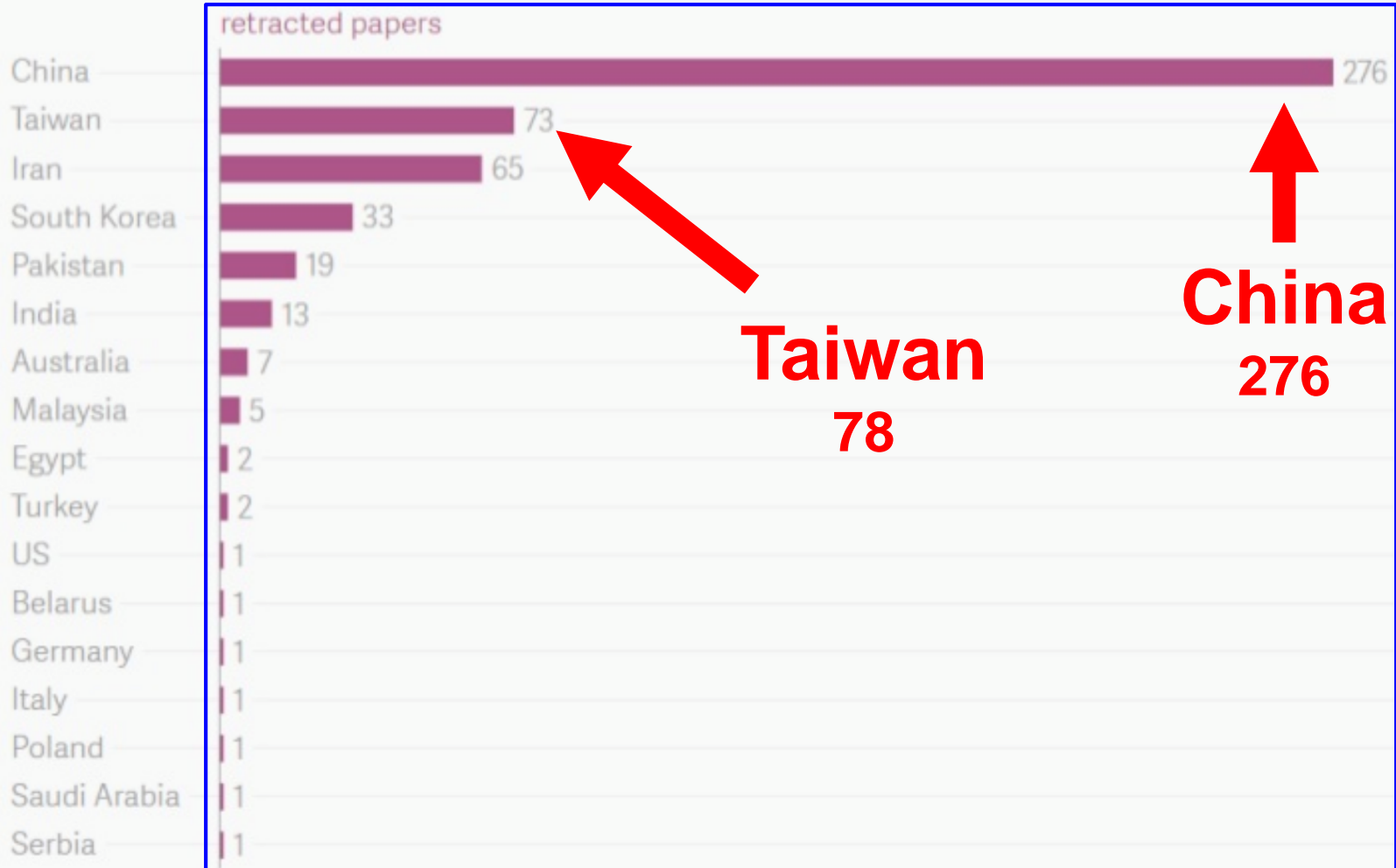
co-authors?

Once Upon a Time



Fake Peer Review

Retracted papers for fake peer review by country from 2012 to 2016





Altmetric: 21 Citations: 4

[More detail >>](#)

Article

G9a/RelB regulates self-renewal and function of colon-cancer-initiating cells by silencing Let-7b and activating the K-RAS/ β -catenin pathway

Shih-Ting Cha, Ching-Ting Tan, Cheng-Chang Ming-Tsan Lin & Min-Liang Kuo

Nature Cell Biology **18**, 993–1005 (2016)
doi:10.1038/ncb3395

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[Cancer](#) [Cancer stem cells](#)

[Cell signalling](#) [DNA methylation](#)



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RETRACTED ARTICLE

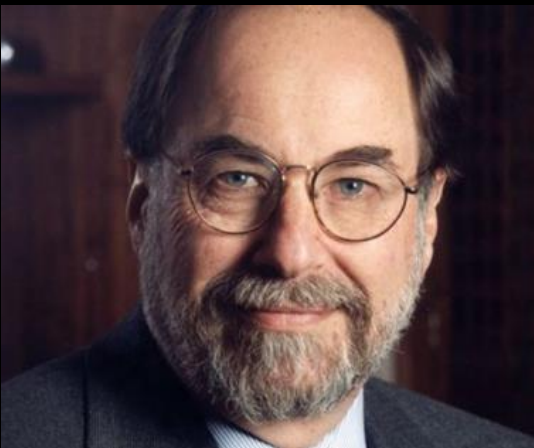
See: [Retraction Notice](#)

Nat Cell Biol. 2016 Sep;18(9):993-1005. doi: 10.1038/ncb3395. Epub 2016 Aug 15.

G9a/RelB regulates self-renewal and function of colon-cancer-initiating cells by silencing Let-7b and activating the K-RAS/ β -catenin pathway.

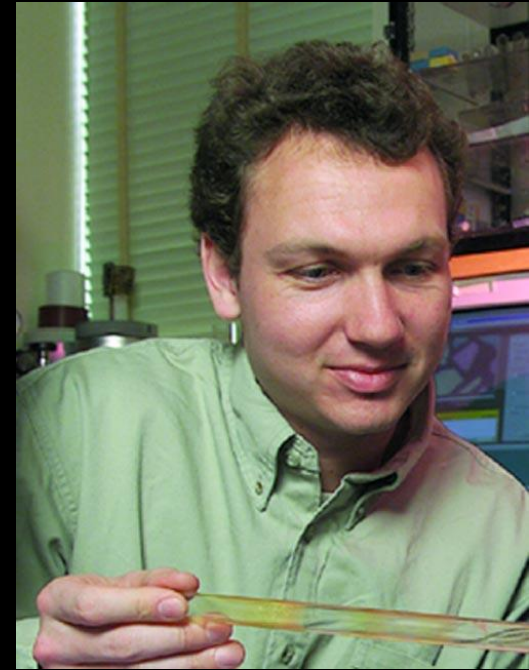
[Cha ST](#)^{1,2}, [Tan CT](#)³, [Chang CC](#)^{4,5}, [Chu CY](#)², [Lee WJ](#)⁶, [Lin BZ](#)⁷, [Lin MT](#)^{7,8}, [Kuo ML](#)^{9,10}.

Baltimore Case: A 10-Years Ordeal

1986	Imanishi-Kari / Baltimore CELL paper; Question raised at MIT	
1986-89	Forensic analysis by Secret Service US Congress hearings	
1991	NIH OSI: “serious scientific misconduct”; CELL paper retracted	
Dec. 1991	Baltimore resigned from Rockefeller presidency	
1993	Scientists validated CELL paper’s findings	
1994	NIH ORI final report: data fabrication and cover-ups	
1996	Dept Health and Human Services overturns ORI’s verdict	
Aug. 1996	Tufts University re-instated Imanishi-Kari	

Hendrik Schön Scandal

- Ph.D. (1997) University of Konstanz
- Condensed matter physics and nanotechnology
- Bell Lab
- One paper every 8 days in 2001
- *Science* (9), *Nature* (7), *Physics Review* (6)



World Record: Fujii Yoshitaka (藤井 善隆)

- M.D., Anesthesiology
- Tokyo Medical and Dental U., Tsukuba U., and Toho U.
- **183!**



Shigeaki Kato (加藤 成亮)

- Ph.D., Endocrinology
- University of Tokyo
- *25 retracted; 43 suspected*
- *Cell, Nature, Science, G&D, NCB, EMBO J., MCB, etc.*



The Goodwin Case

- Rising star in RNA field
- Recruited to U. Wisconsin (2000), Associate Professor with tenure
- Falsify figures in grant proposals
- *NSMB*, *Mol. Cell*, *Dev. Biol.*
- Reported by students and postdocs
- Pleaded guilty; two-years probation; \$500 fine; pay back \$100,000.



Carlo Croce

- Superstar at Ohio State University
- Publish >1000 papers
- Empire builder
- OSU salary alone = NT\$2640 萬/year

SCIENCE

The New York Times

Years of Ethics Charges, but Star Cancer Researcher Gets a Pass

Dr. Carlo Croce was repeatedly cleared by Ohio State University, which reaped millions from his grants. Now, he faces new whistle-blower accusations.

By JAMES GLANZ and AGUSTIN ARMENDARIZ MARCH 8, 2017



What's the Point?



Old

Now



TW

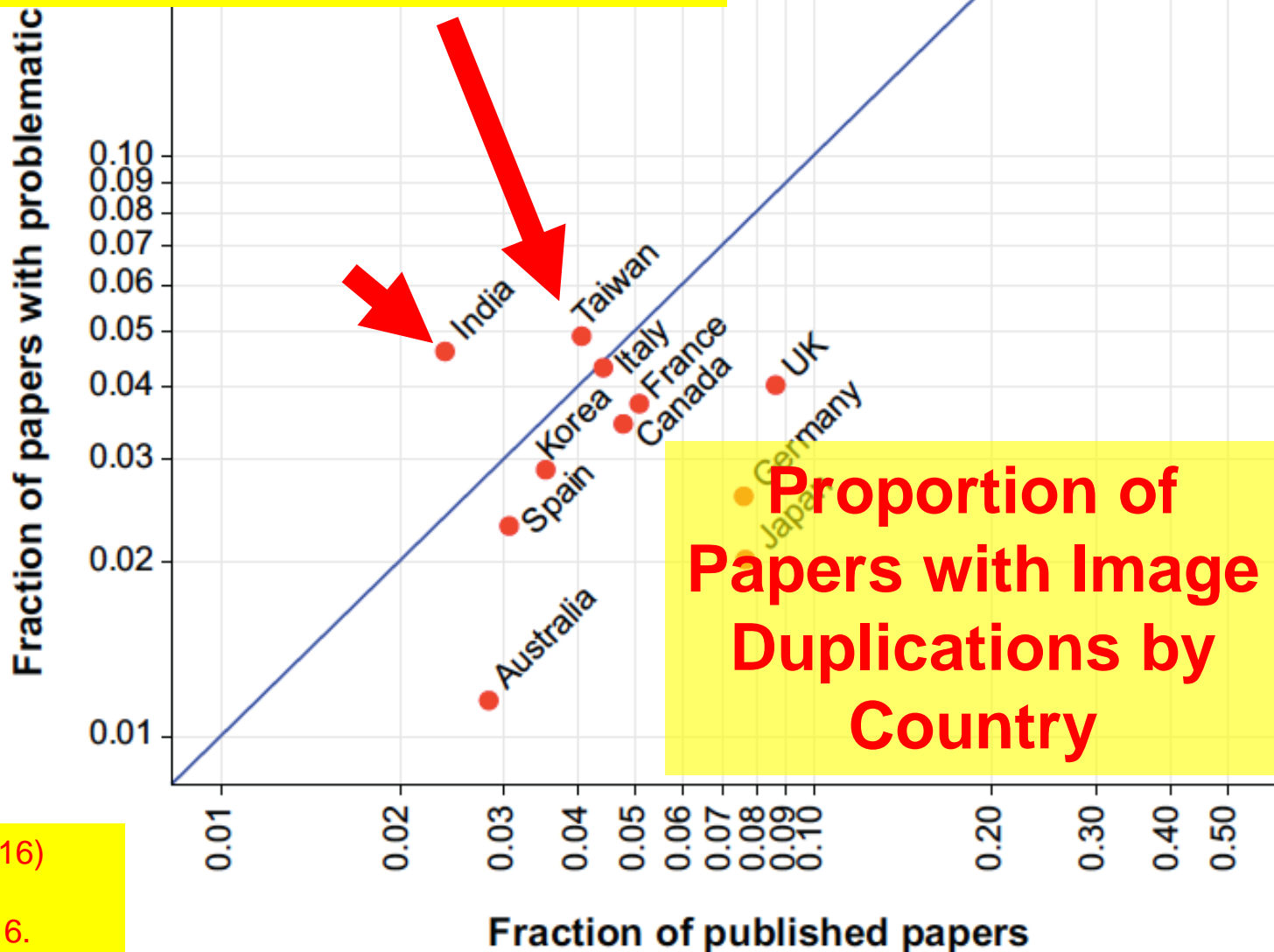


External
World

How Bad is It?

- **2%** scientists admitted misconduct (Fanelli, 2009)
- **90%** academic data cannot be reproduced in industry (Begley & Ellis, 2012; Prinz et al., 2011)
- Merck withdrew **Vioxx**: unreported heart failures
- **55,000** premature deaths estimated; settled with **\$4.8 billion** (Horton, 2004)

**We are NOT Faring
Too Well! ☹️**



Incentives: Why Do It?



Desperation

Ph.D. degree

Grant funding

Promotion & Tenure

Job loss

Money stress

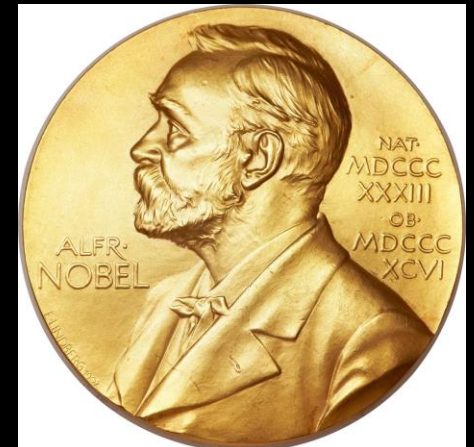


Prestige

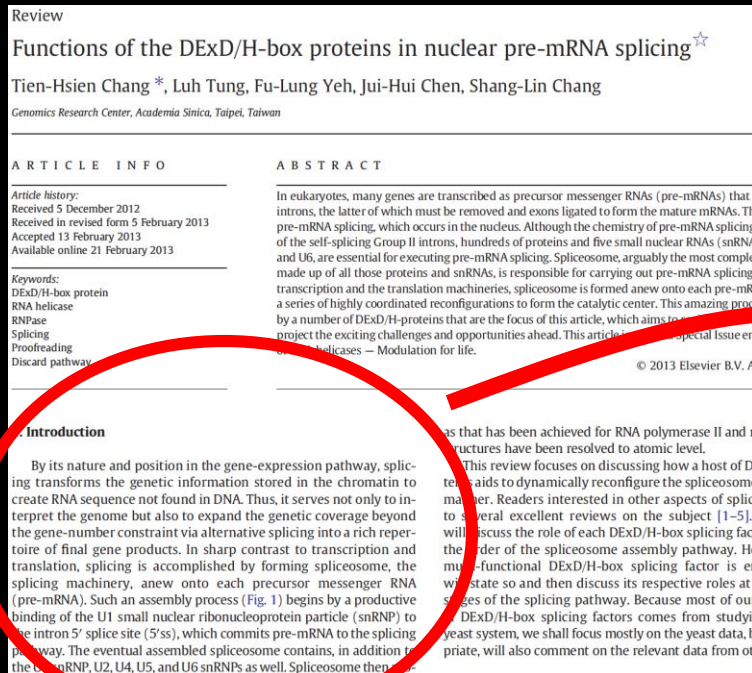
Award & Fame

Power

Big Money reward



(Self) Plagiarism: Copy-and-Paste Published Text and Data



RNA-RNA interaction compared to the number of U1

Nuclear precursor mRNA (pre-mRNA) splicing takes place in the spliceosome, a large dynamic complex consisting of over 100 proteins and five small nuclear RNAs (snRNAs) (32, 70). During spliceosome assembly, the U1 small nuclear ribonucleoprotein particle (snRNP) first contacts the pre-mRNA 5' splice site (5'ss), followed by binding of the U2 snRNP to the branch site and the joining of the U5-U4/U6 tri-snRNP (32, 64, 70). The step in which U1 snRNP binds to the 5'ss is arguably one of the most critical, because it probably commits pre-mRNA to the splicing pathway (38, 48, 49, 60, 74). In the budding yeast *Saccharomyces cerevisiae* in vitro system, two U1-snRNP-containing commitment complexes (CCs), CC1 and CC2, can be detected by native gel electrophoresis prior to the U2 snRNP's joining to form the prespliceosome (38, 60). CC1, whose formation is dependent on a functional 5'ss, appears to be a kinetic precursor to CC2, whose formation requires both a functional 5'ss and branch site and the participation of the branch-site-binding protein (BBP) and Mud2p, which are likely equivalent to SF1 and U2AF65, respectively, in the mammalian system (1–3, 75).

An Act of Stealing

Fabrication (Faking): Painting the White Mice Black

**William Summerlin:
Dermatologist (1974)**



Dr. Robert Good

Falsification: Doctoring Data to Make Believe

Published July 6, 2004

JCB
Feature

2004 JCB Guideline of Image Manipulation

What's in a picture? The temptation of image manipulation

Mike Rossner¹ and Kenneth M. Yamada²

¹Managing Editor, The Journal of Cell Biology

²Editor, The Journal of Cell Biology, and the National Institute of Dental and Craniofacial Research, National Institutes of Health

It's all so easy with Photoshop¹. In the days before imaging software became so widely available, making adjustments to image data in the darkroom required considerable effort and/or expertise. It is now very simple, and thus tempting, to adjust or modify digital image files. Many such manipulations, however, constitute inappropriate changes to your original data, and making such changes can be classified as scientific misconduct. Skilled editorial staff can spot such manipulations

ing or modifying a band in a polyacrylamide gel image) can represent falsification or fabrication.

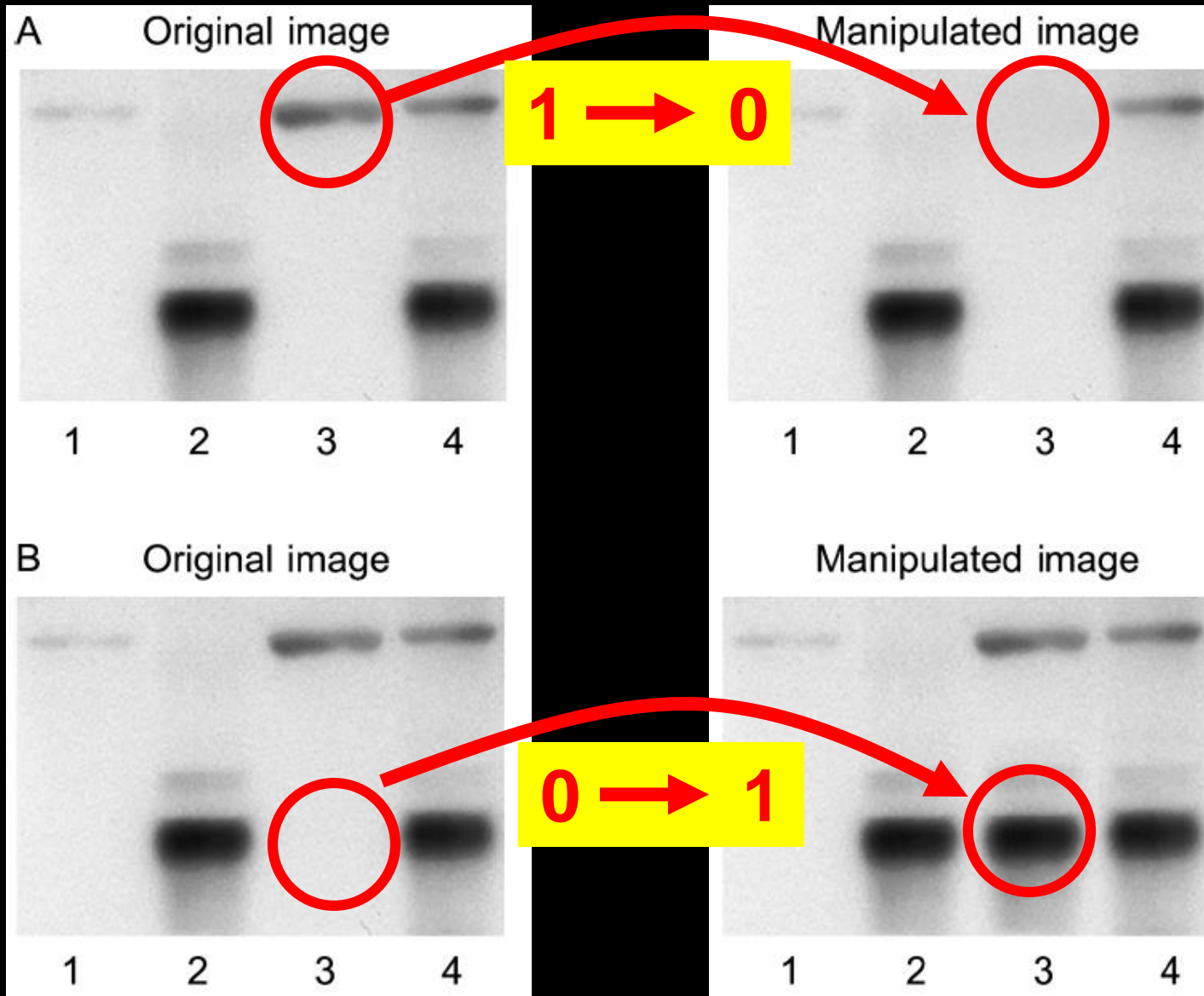
Being accused of misconduct initiates a painful process that can disrupt one's research and career. To avoid such a situation, it is important to understand where the ethical lines are drawn between acceptable and unacceptable image adjustment.

Here we present some general guidelines for the proper handling of digital image data and provide some specific

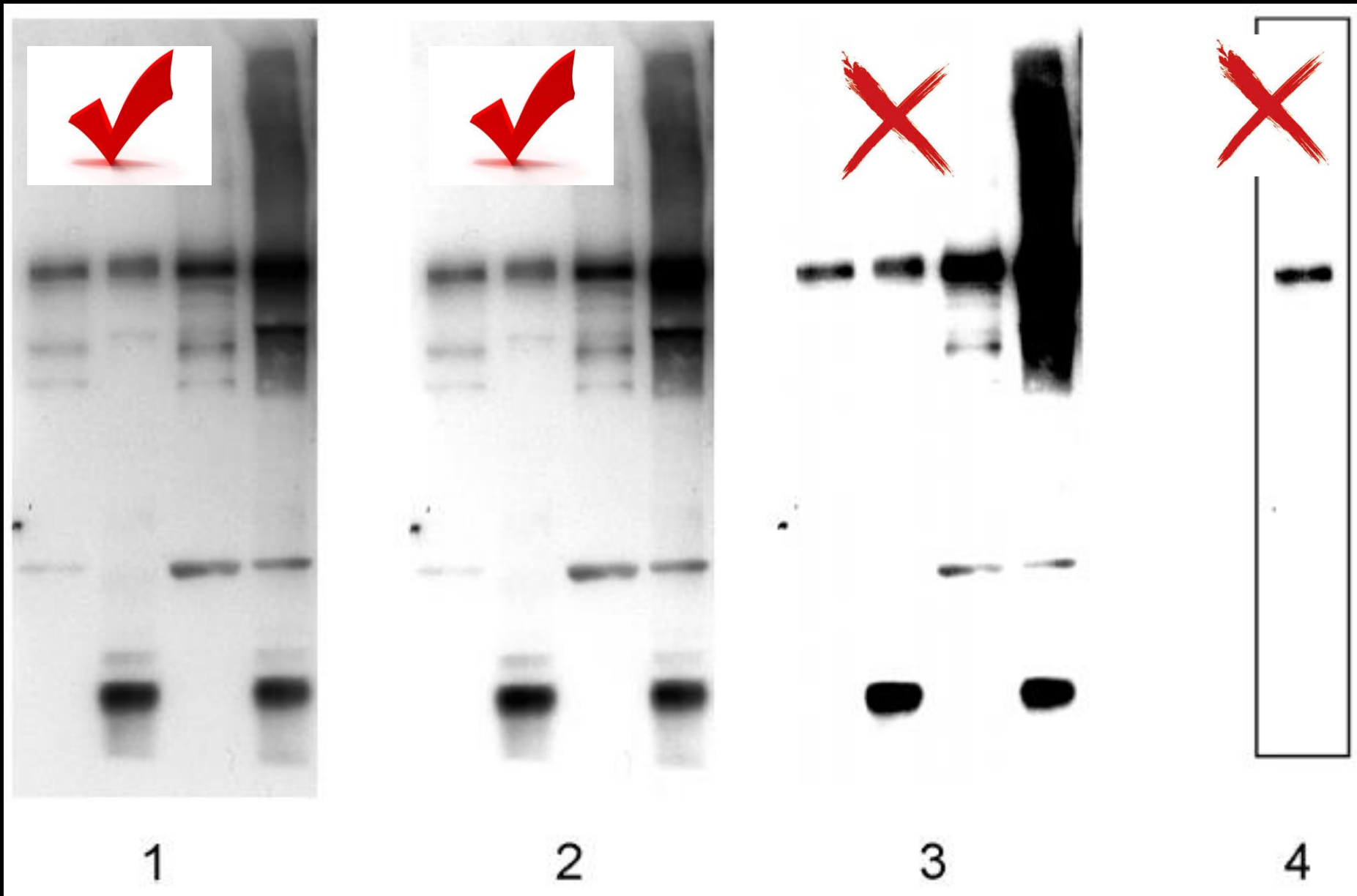
age usually carries information beyond the specific point being made. The quality of an image has implications about the care with which it was obtained, and a frequent assumption (though not necessarily true) is that in order to obtain a presentation-quality image, you had to carefully repeat an experiment multiple times.

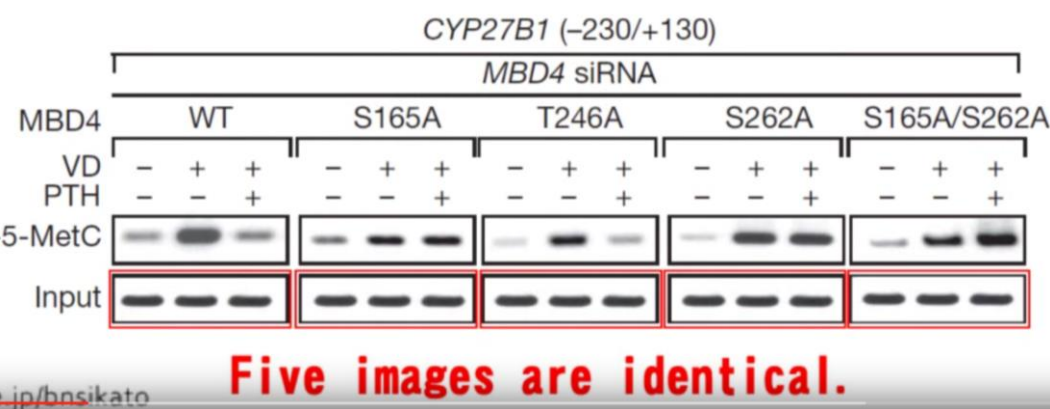
Manipulating images to make figures more simple and more convincing may also deprive you and your colleagues of seeing other information that is often

Gross Manipulation of Blot



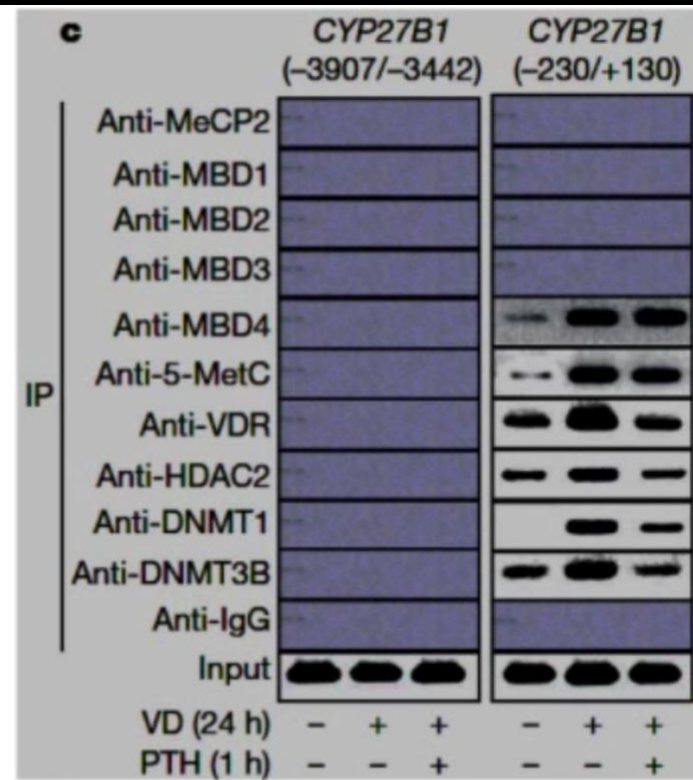
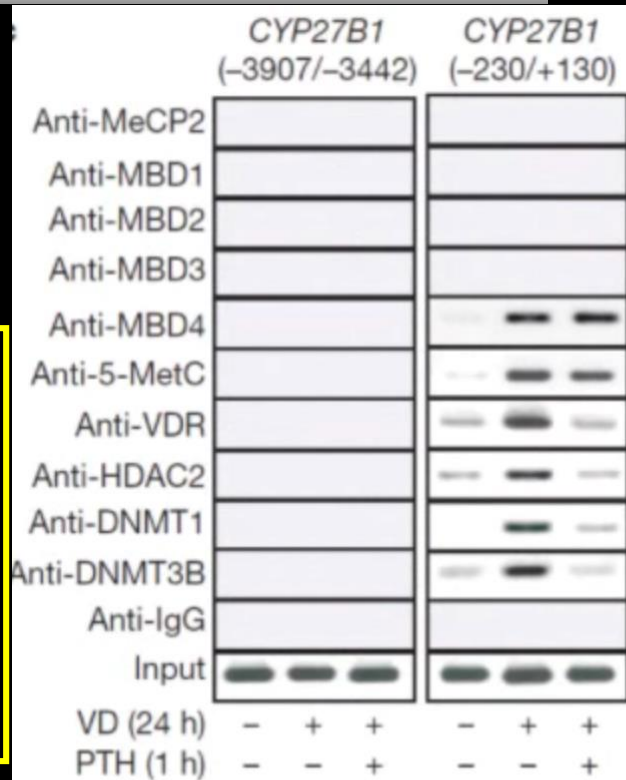
Contrast & Brightness: Excessive Manipulation





S. Kato's 2009 Nature Paper

Duplicated Control Panels & Images



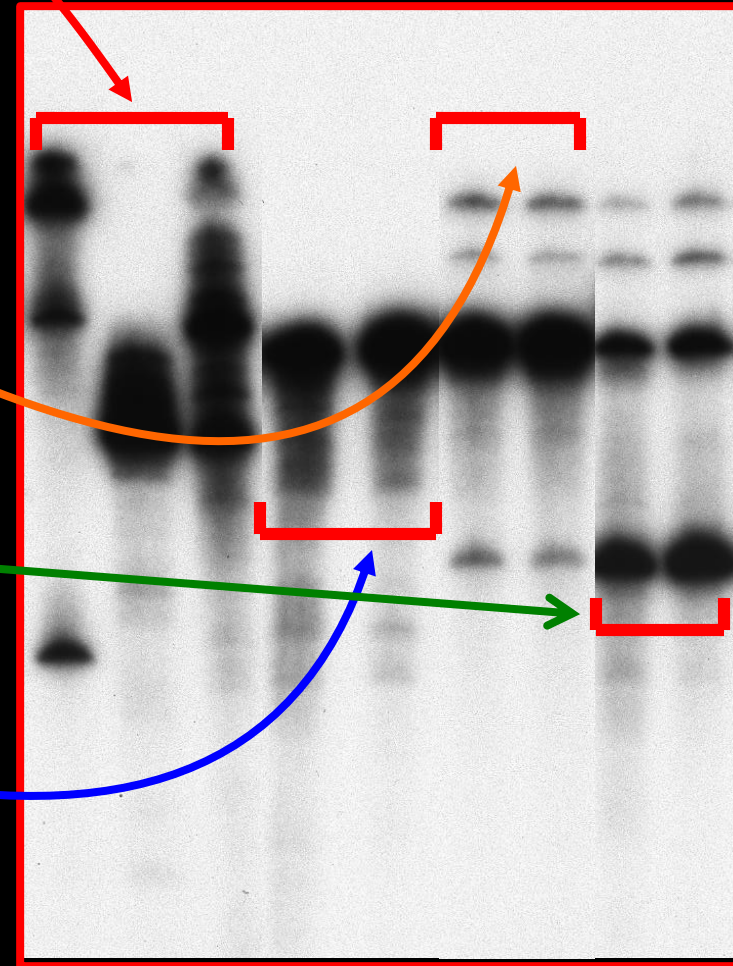
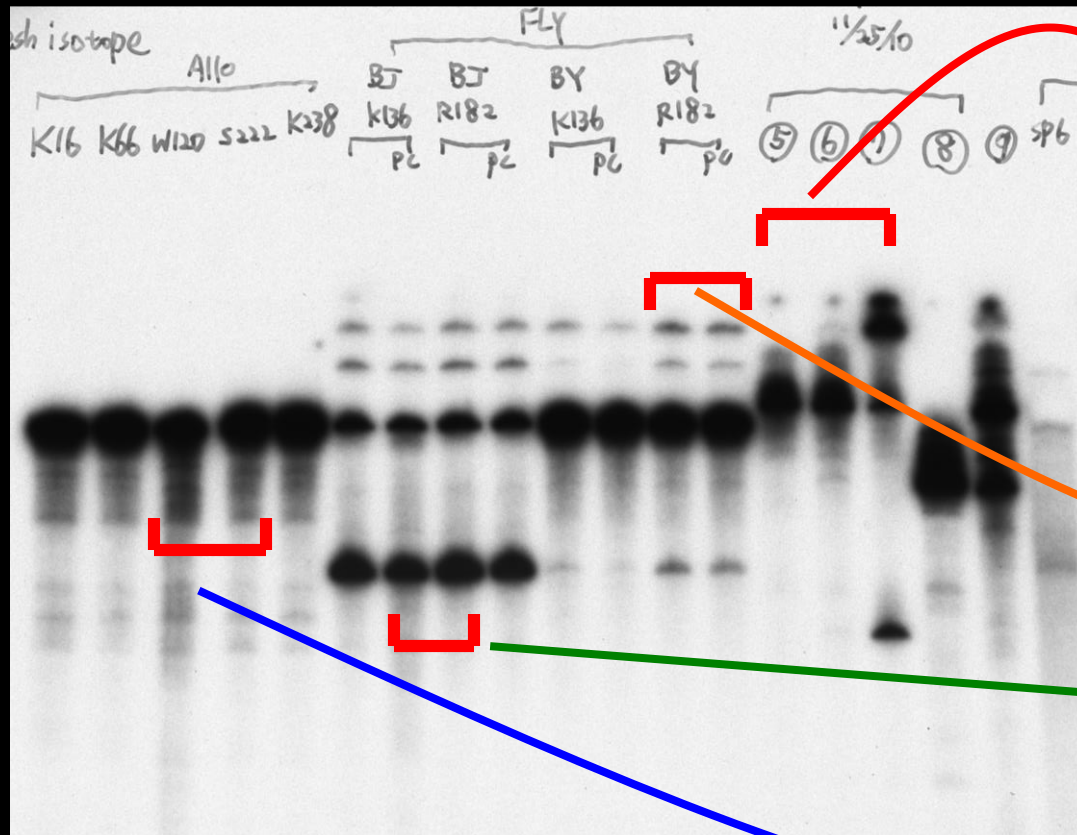
Brightness -35
Contrast +85

Sixteen images are identical.

2008 JBC Paper (a Taiwanese Shame): “Reused” and “Duplicated” Everywhere

This article has been withdrawn by authors Ming-Tsan Lin, I-Hsin Kuo, Cheng-Chi Chang, Chia-Yu Chu, Been-Ren Lin, and Min-Liang Kuo. **The same images were used to represent different experimental conditions.** In Fig. 1A, lanes 2 and 4 of the HIF-1 α DNA gel were **duplicated**. The HIF-1 α DNA gel from Fig. 1A was **reused** in Fig. 1E in the HIF-1 α rCyr61 panel. The GAPDH DNA gel from Fig. 1A was **reused** in Fig. 1E as GAPDH rCyr61 and IGF-1 panels, Fig. 5A as GAPDH, and Fig. 6B as input, left panel. The HIF-1 β immunoblot from Fig. 1A was **reused** in Fig. 1B as HIF-1 β , AGS and TSGH panels, Fig. 1D as HIF-1 β , N87 panel, Fig. 1F as HIF-1 β , rCyr61 panel, and Fig. 3D as HIF-1 β . The tubulin immunoblot from Fig. 1A was **reused** in Fig. 5B as tubulin, lower panel, and **reused** in Fig. 5E as tubulin, left panel. In Fig. 1C, lanes 1 and 2 of the HIF-1 α immunoblot were **reused** in lanes 5 and 6. In Fig. 1D, the HIF-1 α immunoblot from the N87 panel was **reused** in Fig. 1F in the HIF-1 α IGF-1 panel. In Fig. 1E, lanes 2 and 3 of the HIF-1 α DNA gel from the rCyr61 panel were **duplicated** in lanes 5 and 6 of the same panel. Also in Fig. 1E, the HIF-1 α DNA gel from the CoCl₂ panel was **reused** in the IGF-1 panel as HIF-1 α . In Fig. 1F, lanes 4 and 5 were **duplicated** in the HIF-1 β immunoblot from the CoCl₂ panel. The HIF-1 β immunoblot from the IGF-1 panel in Fig. 1F was **reused** in Fig. 3A as tubulin. In Fig. 1G, lanes 1 and 2 of the tubulin immunoblot, left panel, was **reused** in lanes 3 and 4 of the same panel. In Fig. 2A, lanes 2 and 4 of the HIF-1 α immunoblot and lanes 3 and 4 of the HIF-1 β immunoblot from the CoCl₂ panel were **duplicated**. In Fig. 2C, lanes 1 and 2 of the HIF-1 β immunoblot were **duplicated** in lanes 4 and 5, lanes 7 and 8, lanes 9 and 10, and lanes 11 and 12. Also, in the same panel, lanes 3 and 6 were **duplicated**. In Fig. 3A, lanes 4 and 5 of the HIF-1 β immunoblot were **duplicated**. Also in the same figure, lane 1 of the p-AKT immunoblot was **duplicated** in lanes 3 and 5, and lane 2 of the AKT immunoblot was **duplicated** in lane 5. The AKT immunoblot from Fig. 3A was also **reused** in Fig. 3D as 4E-BP1. In Fig. 3B, lane 1 of the p-AKT immunoblot was **reused** in lanes 5 and 6, and lane 1 of the AKT immunoblot was **reused** in lane 6. In Fig. 3D, lane 1 of the HIF-1 α immunoblot was **reused** in lane 6, and lane 1 of the p-p70S6K immunoblot was **reused** in lane 5. The graphs in Fig. 4A were **duplicated**. In Fig. 5A, lane 1 of the c-MET DNA gel was **reused** in lanes 5 and 6, and lane 2 of the same gel was reused in lane 4. Also in Fig. 5A, lanes 1–3 of the AMF gel were **reused** in lanes 4–6. In Fig. 5C, lane 1 of the PAI-1 DNA gel was **reused** in lane 2, and lane 1 of the GAPDH DNA gel was **reused** in lane 2. In Fig. 6A, lanes 1 and 4 of the tubulin immunoblot were **duplicated**. Lane 2 of the PAI-1 DNA gel from Fig. 6B, left panel, was **reused** in lanes 2 and 3 of the PAI-1 DNA gel, right panel. In Fig. 6B, lanes 1 and 4 of the input DNA gel, right panel, were **duplicated**.

Cherry-Picking, Gel-Slicing-and-Dicing: A Recipe for Disaster



Ph.D.?

Doctor of **Ph**ilosophy

Vs.

Doctor of **Ph**otoshop

Falsification: Computational **Get-Loss Syndrome**

INSIGHTS | POLICY FORUM

REPRODUCIBILITY

Enhancing reproducibility for computational methods

Data, code, and workflows should be available and cited

By **Victoria Stodden**,¹ **Marcia McNutt**,² **David H. Bailey**,³ **Ewa Deelman**,⁴ **Yolanda Gil**,⁴ **Brooks Hanson**,⁵ **Michael A. Heroux**,⁶ **John P.A. Ioannidis**,⁷ **Michela Taufer**⁸

Over the past two decades, computational methods have radically changed the ability of researchers from all areas of scholarship to process and analyze data and to simulate complex systems. But with these advances come challenges that are contributing to broader concerns over irreproducibility in the scholarly literature, among them the lack of transparency in disclosure of computational methods. Current reporting methods are often uneven, incomplete, and still evolving. We present a novel set of Reproducibility Enhancement Principles (REP) targeting disclosure challenges involving computation. These recommendations, which build upon more general proposals from the Transparency and Openness Promotion (TOP) guidelines (1) and

to understanding how computational results were derived and to reconciling any differences that might arise between independent replications (4). We thus focus on the ability to rerun the same computational steps on the same data the original authors used as a minimum dissemination standard (5, 6), which includes workflow information that explains what raw data and intermediate results are input to which computations (7). Access to the data and code that underlie discoveries can also enable downstream scientific contributions, such as meta-analyses, reuse, and other efforts that include results from multiple studies.

RECOMMENDATIONS

Share data, software, workflows, and details of the computational environment that generate published findings in open trusted repositories. The minimal components that enable independent regeneration of computational results are the data, the computational steps

MUST DESCRIBE IN
DETAIL:

- Data sharing
- Software sharing
- Workflows
- Detailed comput. Environment
- etc.

Stodden et al. (2016) *Science* **254**, 1240-1241.

Falsification: Vaux's 10 Rules of Thumb for Statistical Presentation

Australian Biochemist Students' Page

Ten Rules of Thumb for the Presentation and Interpretation of Data in Scientific Publications

David Vaux's quest is to improve the quality of data in scientific publications. The Australian Biochemist asked him to provide a brief description of his 'Ten Rules'. Because of space limitations, and the need to avoid litigation, no genuine examples (or counter examples) have been included to illustrate the rules here, but Professor Vaux says he is willing to give the full presentation in person, including many humorous, shocking and libellous examples, whenever and wherever he is invited.

Science is new knowledge gained through repeated experiment or observation. This knowledge is communicated by publishing papers, which, by convention, include not only the conclusions, but also the data upon which the conclusions are based. The data should be sufficient, and presented in such a way, that those reading the paper are able to interpret them themselves, and come to the same conclusions as the authors. The discussion should include all conclusions that are consistent with the published data.

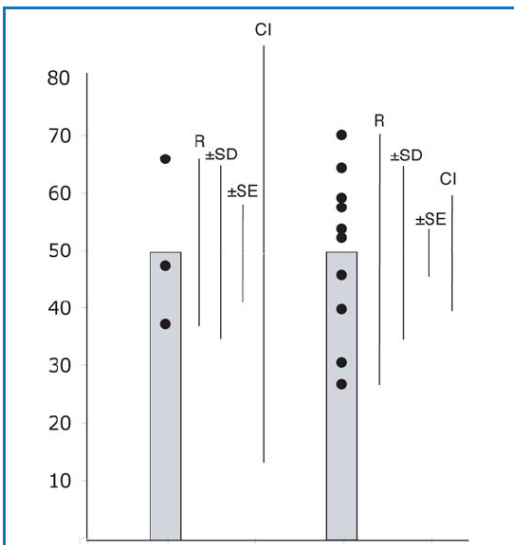
To convince readers of a paper that a new fact has been discovered, and that the results are not just a fluke or a statistical anomaly, the experiments or observations must be repeated often enough to make it highly probable that the conclusions are correct. Sometimes all of the data are shown, but more commonly they are combined and statistics are used to indicate how they are distributed and how much confidence one should have in any inferences about them.

To be communicated honestly and convincingly and in a way that can be comprehended by those reading a paper, the data must be presented properly. To be convinced that the authors' conclusions are correct, those reading a paper must know how to interpret the data.

This article provides ten rules of thumb for the presentation of data in publications, and each rule can also be considered from the point of view of the reader of a paper, when they are trying to decide whether the data are sound and the authors' conclusions are correct.

Multiple results are often summarised by showing means

was 35.51 with $CI_{95\%}$ of 0.64 would be less help to a tourist than knowing the SD, but might be of great interest to a climatologist who knew that the values between 1920 and 1960 were 34.32 with a $CI_{95\%}$ of 0.62.



What Kind of Error Bars?

- **Descriptive (range):** least & greatest & \pm standard deviation [SD]
- **Inferential:** standard error of the mean (SE or SEM) & confidence intervals (CI)

Co-Author's Responsibility: World Record of 5,154 Co-authors

Selected for a Viewpoint in *Physics*
PHYSICAL REVIEW LETTERS

week ending
15 MAY 2015

Combined Measurement of the Higgs Boson Mass in pp Collisions at $\sqrt{s} = 7$ and 8 TeV with the ATLAS and CMS Experiments

G. Aad *et al.*^{*}

(CMS Collaboration)[‡]

(Received 25 March 2015; published 14 May 2015)

A measurement of the Higgs boson mass is presented based on the combined data samples of the ATLAS and CMS experiments at the CERN LHC in the $H \rightarrow \gamma\gamma$ and $H \rightarrow ZZ \rightarrow 4\ell$ decay channels. The results are obtained from a simultaneous fit to the reconstructed invariant mass peaks in the two channels and for the two experiments. The measured masses from the individual channels and the two experiments are found to be consistent among themselves. The combined measured mass of the Higgs boson is $m_H = 125.09 \pm 0.21$ (stat) ± 0.11 (syst) GeV.

DOI: [10.1103/PhysRevLett.114.191803](https://doi.org/10.1103/PhysRevLett.114.191803)

PACS numbers: 14.80.Bn, 13.85.Ok



- **Author list:**
24 pages
- **Paper *per se*:**
9 pages

Etc., etc.

PLA 114, 191803 (2015) PHYSICAL REVIEW LETTERS 13 MAY 2015

P. Barua,^{1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,35,36,37,38,39,40,41,42,43,44,45,46,47,48,49,50,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,66,67,68,69,70,71,72,73,74,75,76,77,78,79,80,81,82,83,84,85,86,87,88,89,90,91,92,93,94,95,96,97,98,99,100,101,102,103,104,105,106,107,108,109,110,111,112,113,114,115,116,117,118,119,120,121,122,123,124,125,126,127,128,129,130,131,132,133,134,135,136,137,138,139,140,141,142,143,144,145,146,147,148,149,150,151,152,153,154,155,156,157,158,159,160,161,162,163,164,165,166,167,168,169,170,171,172,173,174,175,176,177,178,179,180,181,182,183,184,185,186,187,188,189,190,191,192,193,194,195,196,197,198,199,200,201,202,203,204,205,206,207,208,209,210,211,212,213,214,215,216,217,218,219,220,221,222,223,224,225,226,227,228,229,230,231,232,233,234,235,236,237,238,239,240,241,242,243,244,245,246,247,248,249,250,251,252,253,254,255,256,257,258,259,260,261,262,263,264,265,266,267,268,269,270,271,272,273,274,275,276,277,278,279,280,281,282,283,284,285,286,287,288,289,290,291,292,293,294,295,296,297,298,299,300,301,302,303,304,305,306,307,308,309,310,311,312,313,314,315,316,317,318,319,320,321,322,323,324,325,326,327,328,329,330,331,332,333,334,335,336,337,338,339,340,341,342,343,344,345,346,347,348,349,350,351,352,353,354,355,356,357,358,359,360,361,362,363,364,365,366,367,368,369,370,371,372,373,374,375,376,377,378,379,380,381,382,383,384,385,386,387,388,389,390,391,392,393,394,395,396,397,398,399,400,401,402,403,404,405,406,407,408,409,410,411,412,413,414,415,416,417,418,419,420,421,422,423,424,425,426,427,428,429,430,431,432,433,434,435,436,437,438,439,440,441,442,443,444,445,446,447,448,449,450,451,452,453,454,455,456,457,458,459,460,461,462,463,464,465,466,467,468,469,470,471,472,473,474,475,476,477,478,479,480,481,482,483,484,485,486,487,488,489,490,491,492,493,494,495,496,497,498,499,500,501,502,503,504,505,506,507,508,509,510,511,512,513,514,515,516,517,518,519,520,521,522,523,524,525,526,527,528,529,530,531,532,533,534,535,536,537,538,539,540,541,542,543,544,545,546,547,548,549,550,551,552,553,554,555,556,557,558,559,560,561,562,563,564,565,566,567,568,569,570,571,572,573,574,575,576,577,578,579,580,581,582,583,584,585,586,587,588,589,590,591,592,593,594,595,596,597,598,599,600,601,602,603,604,605,606,607,608,609,610,611,612,613,614,615,616,617,618,619,620,621,622,623,624,625,626,627,628,629,630,631,632,633,634,635,636,637,638,639,640,641,642,643,644,645,646,647,648,649,650,651,652,653,654,655,656,657,658,659,660,661,662,663,664,665,666,667,668,669,670,671,672,673,674,675,676,677,678,679,680,681,682,683,684,685,686,687,688,689,690,691,692,693,694,695,696,697,698,699,700,701,702,703,704,705,706,707,708,709,710,711,712,713,714,715,716,717,718,719,720,721,722,723,724,725,726,727,728,729,730,731,732,733,734,735,736,737,738,739,740,741,742,743,744,745,746,747,748,749,750,751,752,753,754,755,756,757,758,759,760,761,762,763,764,765,766,767,768,769,770,771,772,773,774,775,776,777,778,779,780,781,782,783,784,785,786,787,788,789,790,791,792,793,794,795,796,797,798,799,800,801,802,803,804,805,806,807,808,809,810,811,812,813,814,815,816,817,818,819,820,821,822,823,824,825,826,827,828,829,830,831,832,833,834,835,836,837,838,839,840,841,842,843,844,845,846,847,848,849,850,851,852,853,854,855,856,857,858,859,860,861,862,863,864,865,866,867,868,869,870,871,872,873,874,875,876,877,878,879,880,881,882,883,884,885,886,887,888,889,890,891,892,893,894,895,896,897,898,899,900,901,902,903,904,905,906,907,908,909,910,911,912,913,914,915,916,917,918,919,920,921,922,923,924,925,926,927,928,929,930,931,932,933,934,935,936,937,938,939,940,941,942,943,944,945,946,947,948,949,950,951,952,953,954,955,956,957,958,959,960,961,962,963,964,965,966,967,968,969,970,971,972,973,974,975,976,977,978,979,980,981,982,983,984,985,986,987,988,989,990,991,992,993,994,995,996,997,998,999,1000,1001,1002,1003,1004,1005,1006,1007,1008,1009,1010,1011,1012,1013,1014,1015,1016,1017,1018,1019,1020,1021,1022,1023,1024,1025,1026,1027,1028,1029,1030,1031,1}

NEWEST Record 15,025 Co-authors

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Advance Access Publication Date: 24 March 2021

Original Article

SARS-CoV-2 vaccination modelling for safe surgery to save lives: data from an international prospective cohort study

COVIDSurg Collaborative, GlobalSurg Collaborative*

Members of the COVIDSurg Collaborative and GlobalSurg Collaborative are co-authors of this study and are listed under the heading Collaborators.

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Co-Author's Responsibility

ICMJE (Interntl. Commit. of Med. J. Editors; *NEJM, JAMA, Lancets* [13])

Every1 is Equally Responsible

- ***Substantial Contributions:*** conception or design; acquisition, analysis, or interpretation of data; **AND**
- Drafting the work and critically revising for ***important intellectual content; AND***
- Final approval of revision to be published; **AND**
- Agreement to be accountable for ***ALL ASPECTS***

PNAS Co-Author's Responsibility

- **Substantial Contributions:** conception or design, or software creation
- (OR) Draft the work
- (OR) Substantively revise it
- (AND) Approve submitted versions
- (AND) Agree to be accountable for author's own contribution
- (AND) Ensure questions related to any part of work...investigated, resolved, and documented in literature

Who Should **NOT** be a Co-Author?

- **Ghost Writers:** paid or not paid (Big-pharma hiding behind: Vioxx)
- **Honorary:** “*big cheeses*” *for authority enhancement*
- Financier
- Non-involvement lab head and higher-ups
- Providing published reagents
- Australia: misconduct
- US ORI: not misconduct

At minimum: you should have carefully read the damn paper!

(Vaux, D. [2008] *Australian Biochemist* 39, 37-39.)

Experimenter's Responsibility: Keep Detailed Raw Data

8/27/08

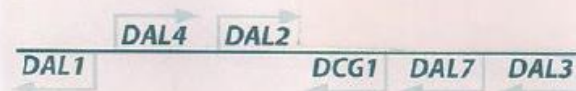
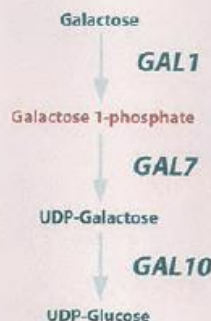
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Exp. Title

OPERON-LIKE ORGANIZATION OF THE GAL GENES

Although eukaryotes lack true operons, there are examples of operon-like gene clusters. Three examples of galactose utilization genes in *S. cerevisiae* (GAL1, GAL7, GAL10), the allantoin degradation genes in *S. cerevisiae* (DAL1, DAL2, DAL3, DAL4, DCG1, DAL7), and the thalianol synthesis genes in *Arabidopsis* (THAS, THAH, THAD):

Thoughts



I want to show that disruption of the operon-like organization of the GAL genes:

- (1) leads to less co-ordinated expression
- (2) reduces fitness
- (3) leads to a buildup of pathway intermediates

The strategy for (1) & (2) is to tag the GAL genes with fluorescent proteins or delete the GAL genes both in cis and in trans.

I have put a lot of thought into how to do the strain construction. Some of the issues:

- strain bkg: W303 or S288c
- auxotrophs or prototrophs
- HAP1+?
- delete all 3 genes (GAL1, 10, 7) in each strain? or just pairs (i.e. GAL1 & 10)
- leave drug markers in place or popout? (Report req. gal induction of (re))
- Fitness assays: indirect (FACS) or direct (sequencing)

8/28/08

NOTES ON STRAINS

I have decided upon a strain construction strategy. I will use prototrophic Hap1+ strains of S288c. FACS-based fitness assays will be more difficult since S288c has bkg fluorescence - also I can not use my standard (W303) reference. I would like

Thoughts

Exp. Planning

8/28/08

Date

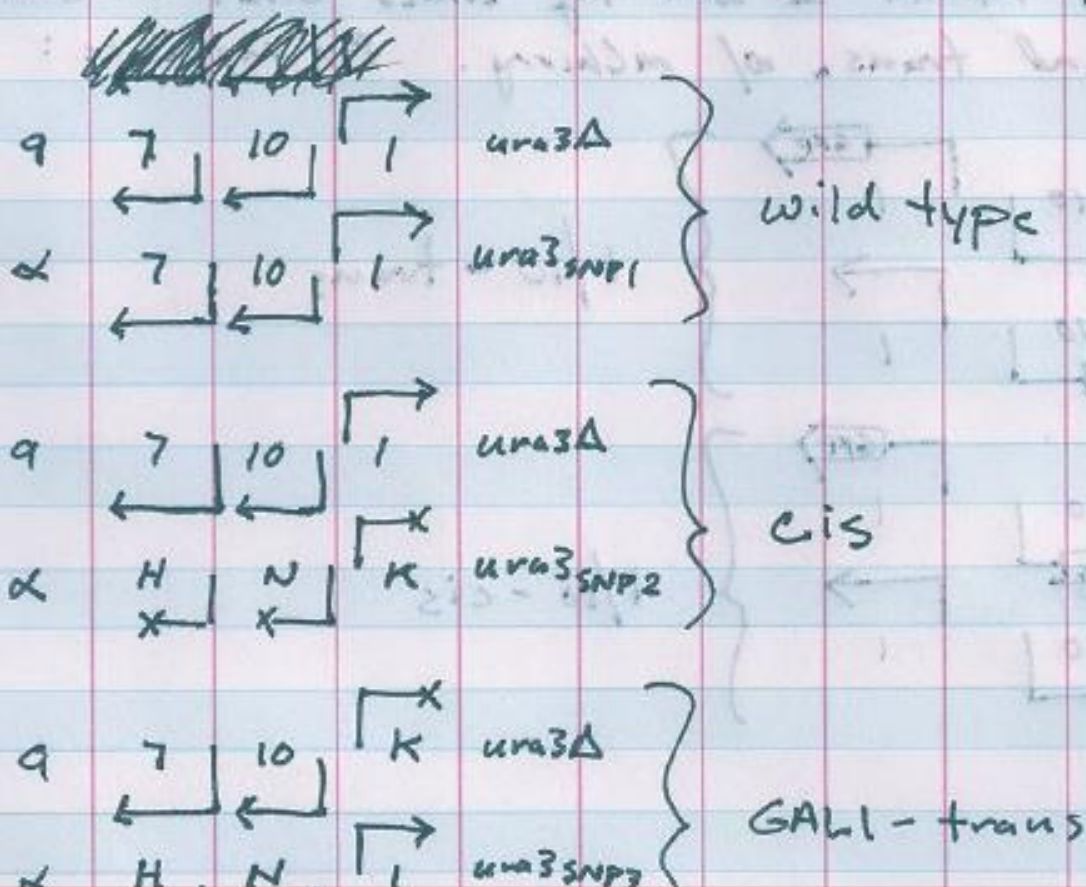
STRAINS TO BE CONSTRUCTED - FOR GENE DELETION/FITNESS

1 - GAL1 K - KanMX

7 - GAL7 H - HygMX

10 - GAL10 N - NatMX

Exp. Planning



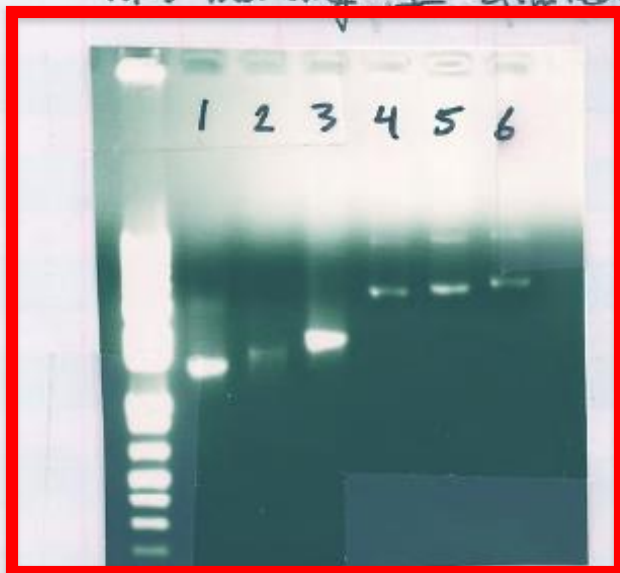
10/10/08

TRANSFORMATION OF ~~FWWNA~~ FY4 & FY5 w/ gal7A::HphMX

On Tuesday (10/7) I streaked FY4 & FY5 from frozen stock

Last night I inoculated 3mL YPD w/ single colony of FY4 & FY5

This morning I diluted each o/n 1:100 (100 μ L into 10mL) ~ 9:15



LANE

- 1 gal11A::Nat
 - 2 gal11A::Kan
 - 3 gal7A::Hph
 - 4 pAG25
 - 5 pAG32
- } primers on p. 5

RAW data

PCR prep → elute 30 μ L EB

Coulter count ~ 3:00 pm

Bkg	2.724E6	
FY4	4.192E7	4.222E7
FY5	3.947E7	3.968E7
Post Bkg	3.742E6	3.362E6

RAW data

10/21/08

SOME DISCUSSION

Soon I will have the strains I need to start doing experiments. First I will need to cross the strains to produce the diploids described on p. 3&4 (note that I ended up using Nat for GAC1 and Kan for GAC10).
↳ For the deletion strains I can build two indep. sets since I built everything in both mating types

- Possible control exp - mating types I can sure that there are

- The 1st expt. will prove that I can get a good idea of what Glucose/Galactose concentrations to use

- I will do all expts in ~~mix~~ glucose, galactose, and a mixture of glucose & galactose

Discussion of Exp. Outcome, Thoughts, and Planning

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PI's Responsibilities:

Keep the Records for 20 Years

- Regular discussion of and insist on research integrity
- Insist on seeing raw data
- Cross-check finalized figures with raw data in great detail
- Avoid creating an “oppressive” lab culture
- Do ***NOT*** serve as honorary co-author
- Mis-management: You are responsible!



**Secret Service: You *DON'T*
Want to See Them, God Forbid!**

Thou Shalt Not Cheat (in Science)



***Thou
Shalt Not
Even
Think About
It!***



Thanks!

**Wish you a great
success in your
HONORABLE
research!**

