Cell line authentication to improve preclinical cancer research: Policies and training

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Background

Irreproducible research is a pervasive, expensive, and well-recognized problem that contributes to both delays and costs of drug discovery and development (1, 2). One common and preventable contributor to irreproducible cancer research is the widespread use of misidentified (e.g., cellular cross-contamination), mislabeled, or interspecies-contaminated cell lines (3). Immortalized cancer cell lines have been used for decades as preclinical models for drug target discovery and in multiple areas of biomedical research (4). At the same time, the problem of altered genotypic and phenotypic function (i.e., “drift”) as a result of over-passaging of cell lines also persists (5).

The Global Biological Standards Institute (GBSI), a Washington, D.C.-based nonprofit, is leading a multipartner effort to build awareness among cancer researchers regarding the importance of cell authentication and to actively change laboratory practices and policies to produce more credible, reproducible, and translatable preclinical data and studies. As a result of targeted training, effective policies, and use of standards and best practices for authentication and cell culturing, (1) knowledge of why and how to perform cell authentication will improve, (2) research reproducibility problems involving cell lines will decrease, (3) millions of dollars in wasted research expenditures will be saved, and (4) translation time from bench to clinic to bedside will be decreased. A companion paper, Cell line authentication to improve preclinical cancer research: Methods in cell line authentication, quality control, and annotation, is also available.

Discussion

Correct identification of the origin of a cell line (i.e., authentication) can be determined by comparing the genetic signature (profiling or fingerprinting) with established databases to confirm identity (6). Although an accredited standard for authentication is available based on short-tandem repeat (STR) profiling (7), there is little evidence that cell line monitoring and authentication is routinely used in the life sciences (8, 9)—particularly among academic researchers (10). Many scientists remain unaware or unconvinced of the need to obtain, establish, and carefully maintain cell cultures, or do not authenticate their cell lines often enough or at all.

Several journals, including Cancer Research and Nature (11), now recommend or require authentication for studies they publish, whereas a small but growing number of research funders...
such as the Prostate Cancer Foundation insist on a certificate of authenticity (or the equivalent) for the research they fund. NIH is also considering a variety of approaches to help catalyze improvements in identifying cell lines and maintaining their integrity (3). For example, NIH grant applicants may be required to provide information on how they intend to address concerns about the identity or contamination of their cell lines. A variety of organizations advocate best practices for handling biological resources (12), including cell lines and cell banking (13, 14), yet reports of misidentified or contaminated cell lines still appear in the literature (15, 16).

A search of NIH Reporter for projects using “cell line” or “cell culture” suggests that NIH currently spends about $3.7 billion on research using cell lines. If 15 percent to 36 percent of these research projects use misidentified or contaminated cell lines, per a widely cited review by Hughes, et al. (17) of the prevalence of this problem, potentially $550M to $1.33B in currently committed research dollars could be affected. Microbial contamination of cell lines by bacteria—most notoriously mycoplasma (18)—viruses, fungi, and other microorganisms is highly problematic in cell culture laboratories worldwide. A recent assessment of mycoplasma contamination found in the National Center for Biotechnology Information Sequence Read Archive conservatively found that 11 percent of projects were contaminated, such that hundreds of millions of dollars in NIH-funded research had been potentially affected (19).

Cell line misidentification and contamination are not new problems, and although several solutions involving authentication have been offered, and in some limited venues adopted (20), it has proven difficult to eradicate. Fixing the problem demands a systematic incremental approach with commitment by all stakeholders and that embraces the importance of targeted training and education.

Future Directions

As part of a broader, interconnected educational program aimed to improve the credibility, reproducibility, and translatability of life science research, GBSI is developing an exportable “active learning” training module for to reduce cell line misidentification, mislabeling, and contamination. It will be the first online multimedia training in cell authentication that can be used either as a free-standing, online, training module or be incorporated into Responsible Conduct of Research training, cell biology and cell culture courses, and other staff training mechanisms. As such, it will differ from more traditional “passive learning” approaches and resources (e.g., hosting webinars, lists of contaminated cell lines) that, to date, have not been systematically disseminated into the research communities that need them, nor have resulted in impactful changes to cell authentication research practices (2).

GBSI’s exportable training module will contain highly interactive training units that include “how to” videos and that will turn learning into practice by sending the trainees back into the laboratory to practice their skills. It will also incorporate a novel dissemination strategy featuring social media, partnerships with professional and research societies, and active engagement with alumni of the training module. Finally, it will evaluate the effectiveness of the module, both with respect to the aims and learning objectives (Fig. 1), and by tracking the level of implementation of cell authentication in the laboratories of the trainees. To advance this effort, GBSI is building partnerships and developing strategies to disseminate and promote this module to all
Aim 1: Create the content for an “active learning” cell authentication training module for postgraduate learners

a. Generate cell authentication content.
b. Support skill-building in cell authentication.
c. Pilot and evolve the curriculum using feedback from the first generation of trainees.
d. Identify the requirements for foundational knowledge.

Aim 2: Develop the exportable module

a. Load content for online dissemination.
b. Provide links to foundational knowledge.
c. Support use of the training module by individuals and classrooms.
d. Pilot individual and classroom uses of the training module.

Aim 3: Disseminate the training module to researchers across the United States and evaluate its effectiveness

a. Advertise and disseminate the module to NIH-funded academic institutions and programs.
b. Partner with societies to advertise and promote the module to their members.
c. Actively recruit the first class of postgraduate learners through a social media campaign, and make them part of subsequent dissemination.
d. Promote the use of this module within existing courses.
e. Evaluate the impact of the training module on the participants.

Overall Impact

As a result of this training, knowledge of why and how to perform cell authentication will improve and research reproducibility problems involving cell lines will decrease. This project will save millions of dollars in wasted research expenditures and decrease translation time from bench to clinic to bedside.

Fig. 1. Enhancing Data Reproducibility through Cell Authentication Training

postgraduate researchers who use cell lines, including graduate students, postdoctoral researchers, and emerging faculty.

Continued reliance upon a de facto honor system that assumes scientists have authenticated their cell lines is not working. Despite the existence of an STR standard for authenticating human cell lines and the availability of affordable fee-for-service options and commercial kits, universal adoption of authentication is far from becoming a reality. Solutions to this persistent
and expensive problem must include effective policies requiring and dedicated funding for cell line authentication. But to ultimately change the culture of cell authentication, the life science research community must also commit sufficient expertise, time, and dedicated resources to train and educate young researchers in good cell culturing techniques and practices.

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