Robust In-Silico identification of sequenced Cancer Cell Lines

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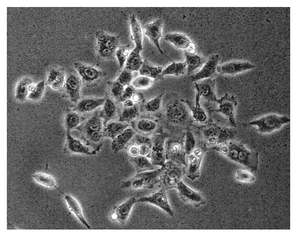
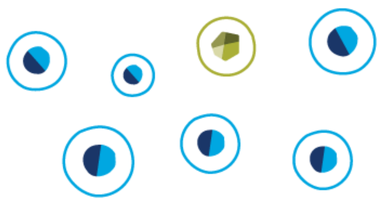
Cell Line Confusion



Cell Line Culture



1 in 7 mis-identified

NGS Identification limited or impossible

Consequence: Incorrect research

Background

Cancer cell lines (CCL) are biological samples of great importance to cancer researchers world-wide but highly susceptible for the critical errors **misidentification** and **cross-contamination** [1].

Identity crisis

See associated Correspondence: Masters, *Nature* **492**, 186 (December 2012)

It is time for all involved to tackle the chronic scandal of cell-line contamination. Funders first.

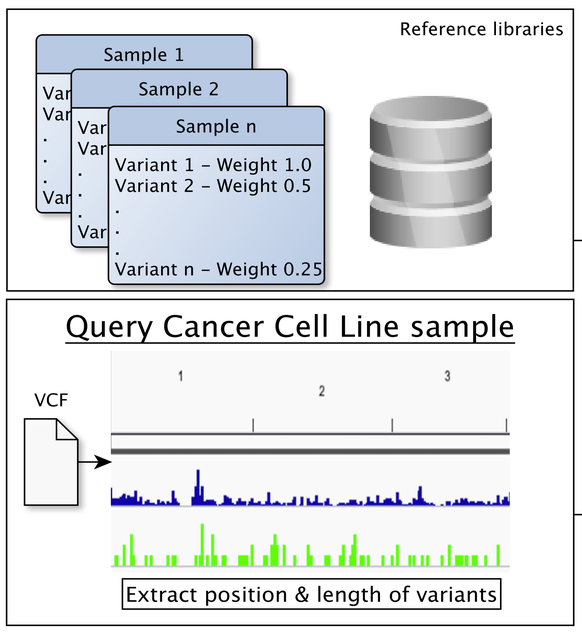
MDA-MB-435 cells are derived from M14 Melanoma cells––a loss for breast cancer, but a boon for melanoma research

James M. Rae, Chad J. Creighton, Jeanne M. Meck, Bassem R. Haddad, Michael D. Johnson

Optimal means to identify CCLs are crucial for cancer-researchers. Established methods (e.g. STR and SPIA) exist [2,3], but are generally not applicable for heterogeneous, incomplete and contradictive Next-Gen Sequencing data. Here, we present the Uniquorn method that reliably identifies CCLs based on their genotyped variation profiles (VCF-files), agnostic to the underlying genotyping-procedure. Uniquorn was evaluated by cross-identifying 1989 CCLs that were obtained with different technologies, laboratories, algorithms and sequencing-approaches. Our method achieves a sensitivity of ~92% and specificity of ~99%. Uniquorn and ~2000 reference CCL samples are freely available as R-BioConductor package.

Everything was going along fine until they discovered their HeLa cell line expressed Y chromosome markers.

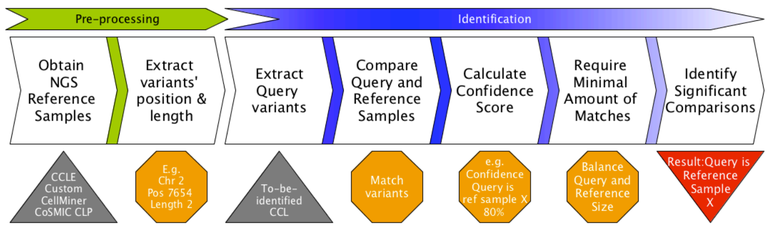
The Uniquorn Approach



Key concepts

1 Use optimal subset of variants for CCL identification (qualitative regulatization)

2 Penalize unspecific similarities (quantiative regularization)



- Uniquorn identifies CCLs by pair-wise comparison of the query to known reference samples
- If p-value and background-noise threshold are passed by one or many comparisons -> positive identification
- Reference variants weighted: weight represents a variant's association-strength to a specific CCL

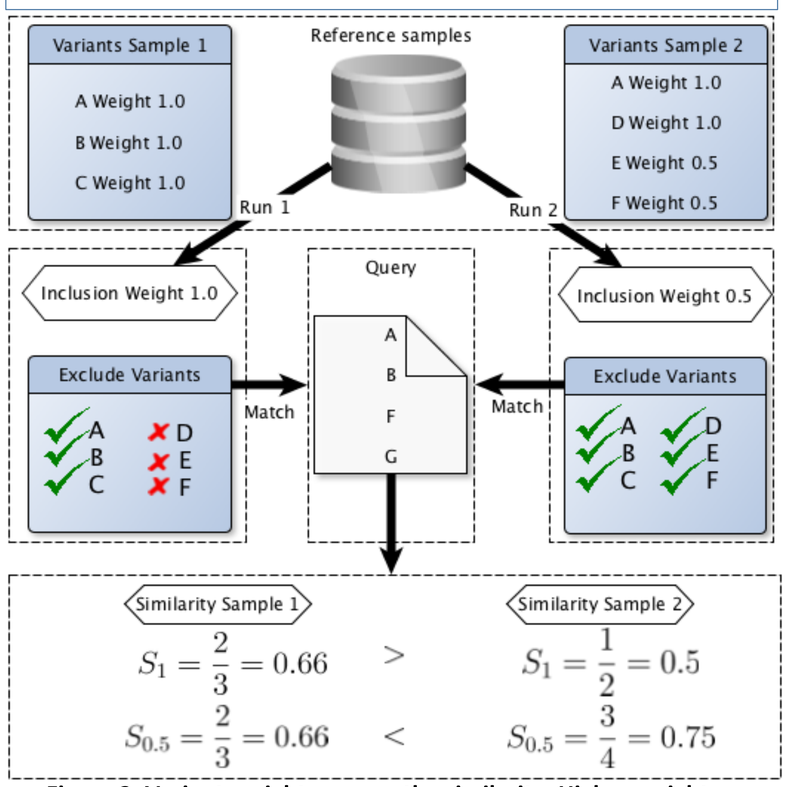


Figure 2: Variant-weights govern the similarity. Higher weights are prefereable but not always available.

COSMIC Cell Lines Project — Catalogue of somatic mutations in cancer
CCLE Cancer Cell Line Encyclopedia
CellMiner™

Sources for reference CCLs

Evaluation

Weight	1.0	0.5	0.25	0.0
Possible TP	3555			
TP	3255	3456	3456	2508
FN	300	99	99	1047
FP	18	38	55	1722
TN	~4 mil.			
Sens. %	92	97	97	71
Spec. %	99			
F1 %	95	98	98	64
PPV %	99			59

Table 1: Uniquorn's capacity to identify heterogeneous Cancer Cell Lines. 3555 CCL relationships of 1989 CCLs had to be found. Uniquorn found ~92% with the highest variant weights. Lower weights are useful for sub-optimal CCL reference libraries.

Summary

- Uniquorn is a novel algorithm to identify CCLs
- Optimized for Next-Generation sequenced CCLs in VCF-format
- Shows an identification-sensitivity of ~92% and specificity of 99%
- Works on all NGS DNA data
  - ✓ Whole-genome/exome, hybrid capture
  - ✓ Different algorithms, e.g.
    - Variant caller
    - Mapper
  - ✓ Sequencing technologies
- Requires reference samples, however, ~2000 can be freely obtained from
  - CCLE [4]
  - CellMiner project [5]
  - CoSMIC Cancer Cell Line Project [6]
- Available as R-BioConductor package *Uniquorn*
- Future development: RNA & Panel-seq

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References

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