C2D2 Centre for Chronic Diseases & Disorders

# Data and simulation for stem cell amplification studies in prostate disease development and control

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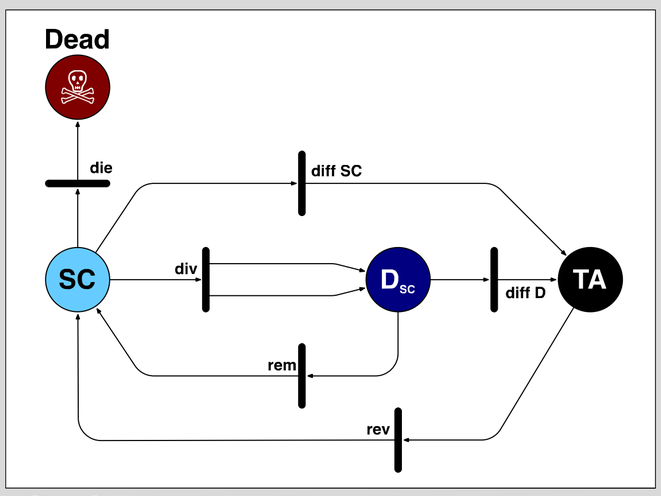
## Aims:

To produce a fit-for-purpose simulation of pathological changes in human prostate which will:
(i) Extend and adapt to look at the effects of rare events
(ii) Describe the cell dynamics of a range of different pathologies related to cell division and differentiation
(iii) Use the parameters in the model to predict the outcome of treatments which target different cell types

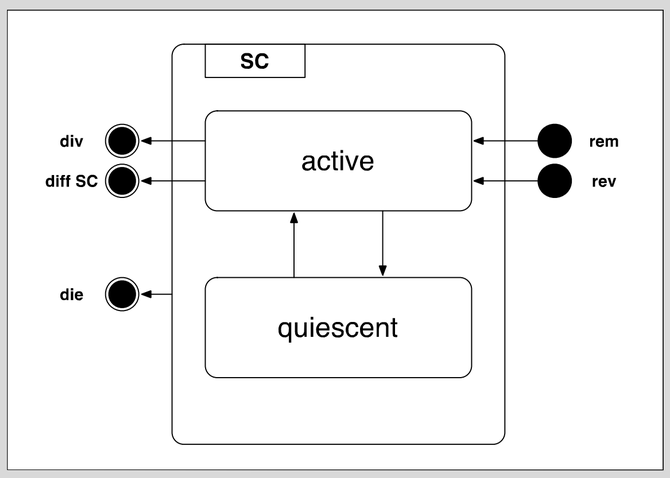
## Strategy

1. Produce a **Top level model** (Petri net) of cell division and differentiation, showing relevant outcomes for stem cells in prostate epithelium




**SC** = Stem Cell
**TA** = Transit Amplifying Cell (place is black because the model continues, with division and differentiation of TA cell towards terminally differentiated luminal cell type)
Transition labels containing:
**div** = division **diff** = differentiation **rev** = reversion **rem** = remain

2. Generate a **State diagram** for a single stem cell showing the lower level states that govern when transition might occur




Terminal blobs are black for entry, annular for exit.
Entry and exit labels refer to the transitions on the Petri net model that cause a cell to arrive in or leave the stem cell state.

3. Produce **a template** for converting the diagrams (Petri nets and associated state diagrams) to text, from which code can be generated.

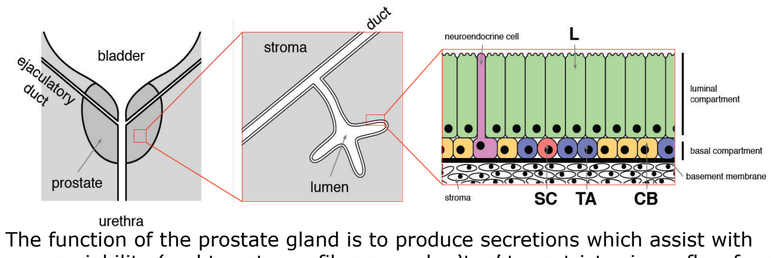
```
<model> ::= (<section>)*
<section> ::= "places:" (<place>)*
| "trans:" (<transition>)*
| "#" <commented-line>
<place> ::= <place-name> <start-count>
<transition> ::= <transition-name>
"in" <place-name>
"out" <place-name> ("," <place-name>)*
"rate" <number>
["mutability" <range>]
(<mutation>)*
<mutation> ::= "mutate" <transition-name> <range>
| "mutate_any" <range>
<range> ::= <number> "to" <number>
```

From this we can generate code in languages such as Python, Erlang, or, with some manipulation, Java.

## Biological data input

4. Generate cell proportion and cell fate data to provide numerical input for the model

### Glandular architecture and cellular structure of the human prostate




The function of the prostate gland is to produce secretions which assist with sperm viability (and to act as a fibromuscular 'tap' to restrict urinary flow from the bladder)
Secretory units are arranged as 'bunches of grapes' each draining via a collecting tubular duct into the urethra.
The bilayered organisation of the epithelial cells surrounding each lumen is shown.

### Immunofluorescence antibodies

| Gene/antigen | Purpose |
|---|---|
| P63 | Marks basal cell nuclei |
| NKX3.1 | Marks luminal cell nuclei |
| Cytokeratin 18 | Marks luminal cells |
| Cytokeratin 5 | Marks basal cells |
| Ki67 | Marks replicating cells |
| PCNA | Marks replicating cells |
| Caspase 8 | Marks apoptotic (dying) cells |
| TUNEL | Marks apoptotic (dying) cells |

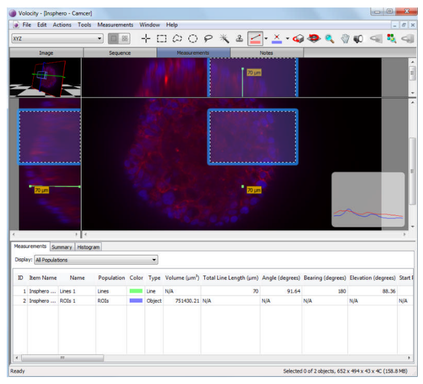
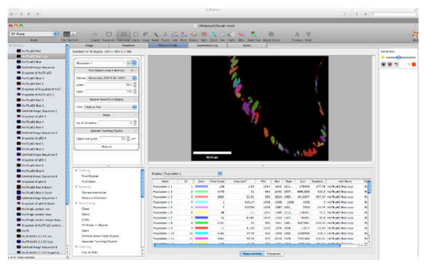
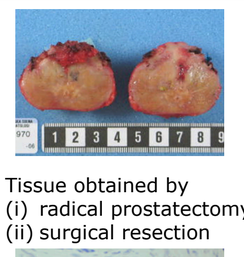


**Image acquisition and recording in Volocity**

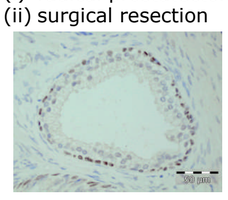


**Quantification of positive epithelial cell nuclei**
False colouring of positive basal cell population shown

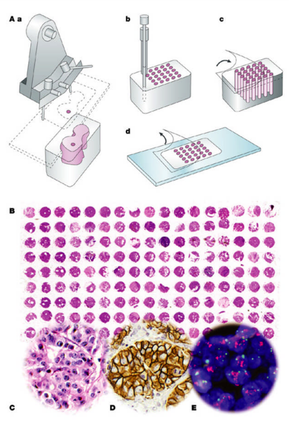
**Automated data recording/quantification of nuclei using digitised fluorescent images and Volocity**



Tissue obtained by
(i) radical prostatectomy
(ii) surgical resection

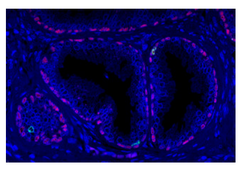


Identity of cells in glands from single tissue blocks confirmed by IHC

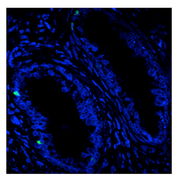


Biopsies from multiple human tissue samples arrayed as ***tissue microarrays***

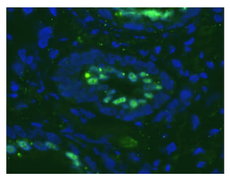
**Glandular architecture of benign prostate**
Basal layer marked by fluorescent immuno-histochemical detection of p63 (red) with nuclei counterstained in DAPI (blue)



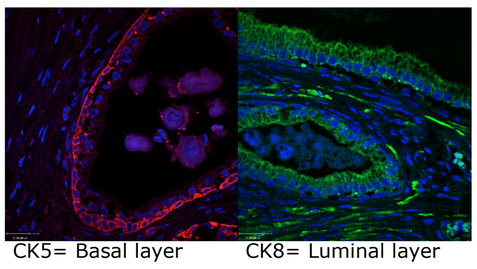
**Replicating cells in benign prostate**
Replicating cells in the basal layer marked by fluorescent immuno-histochemical detection of Ki67 (cyan) with nuclei counterstained in DAPI (blue)



**Glandular architecture of benign prostate**
Luminal layer marked by fluorescent immuno-histochemical detection of NKX3.1 (green) with nuclei couterstained in DAPI (blue)



**Cell phenotyping with cytokeratin antibodies**



CK5= Basal layer CK8= Luminal layer

CK antibodies *can* identify cell types but irregular cell shapes limit quantification:
We use antibodies against NUCLEAR proteins

**Combined basal:proliferation image of Benign prostatic hyperplasia gland**

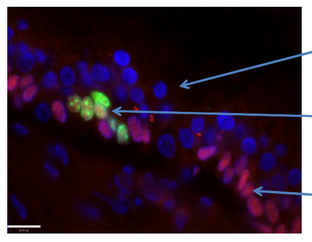



Image indicates that BPH is driven by excessive basal cell proliferation: but we currently treat it by inhibiting the luminal cells!

Fluorescent images digitised to quantify
(i) cell type proportions
(ii) Dying and dividing cell numbers in each cell compartment
Quantified from multiple glands and tissue samples

Data used to feed model

Iterative process of model building and refinement initiated

**Further Reading:**
Maitland N J (2013) Stem cells in the normal and malignant prostate. D J Tindall (Editor), Prostate Cancer: Biochemistry, Molecular Biology and Genetics, Protein Reviews 16, DOI 10.10007/978-1-4614-6828-8_1 Mayo Clinic

**C2D2 publication:**
Choosing and adapting design notations in the principled development of complex systems simulations for research. Fiona A. C. Polack, In Modelling the Physical World Workshop, at Models 2012, Innsbruck, Austria, October 2012. ACM Digital Library

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THE UNIVERSITY of York

yorkshire cancer research

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