How to Start A Molecular Laboratory

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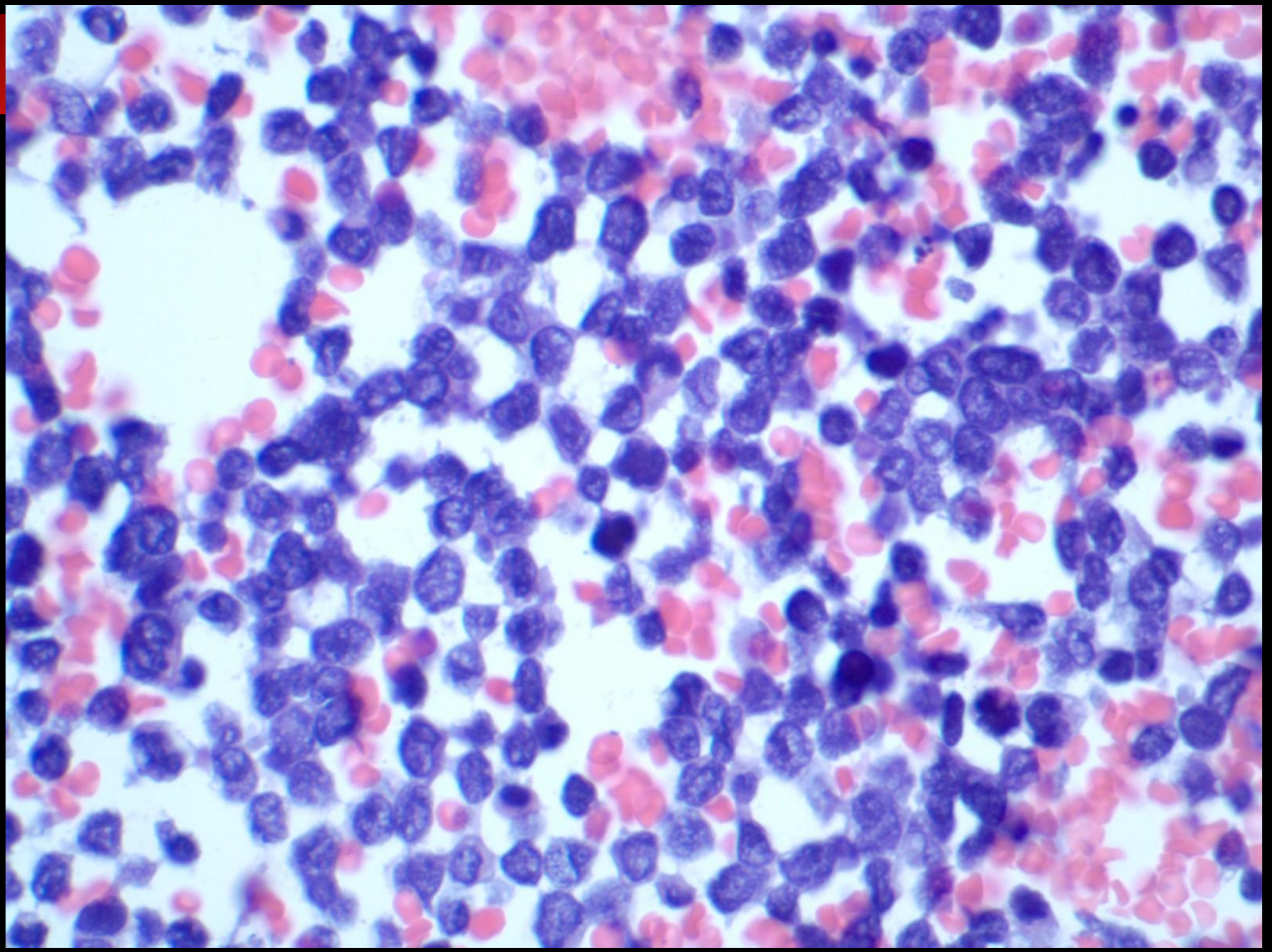
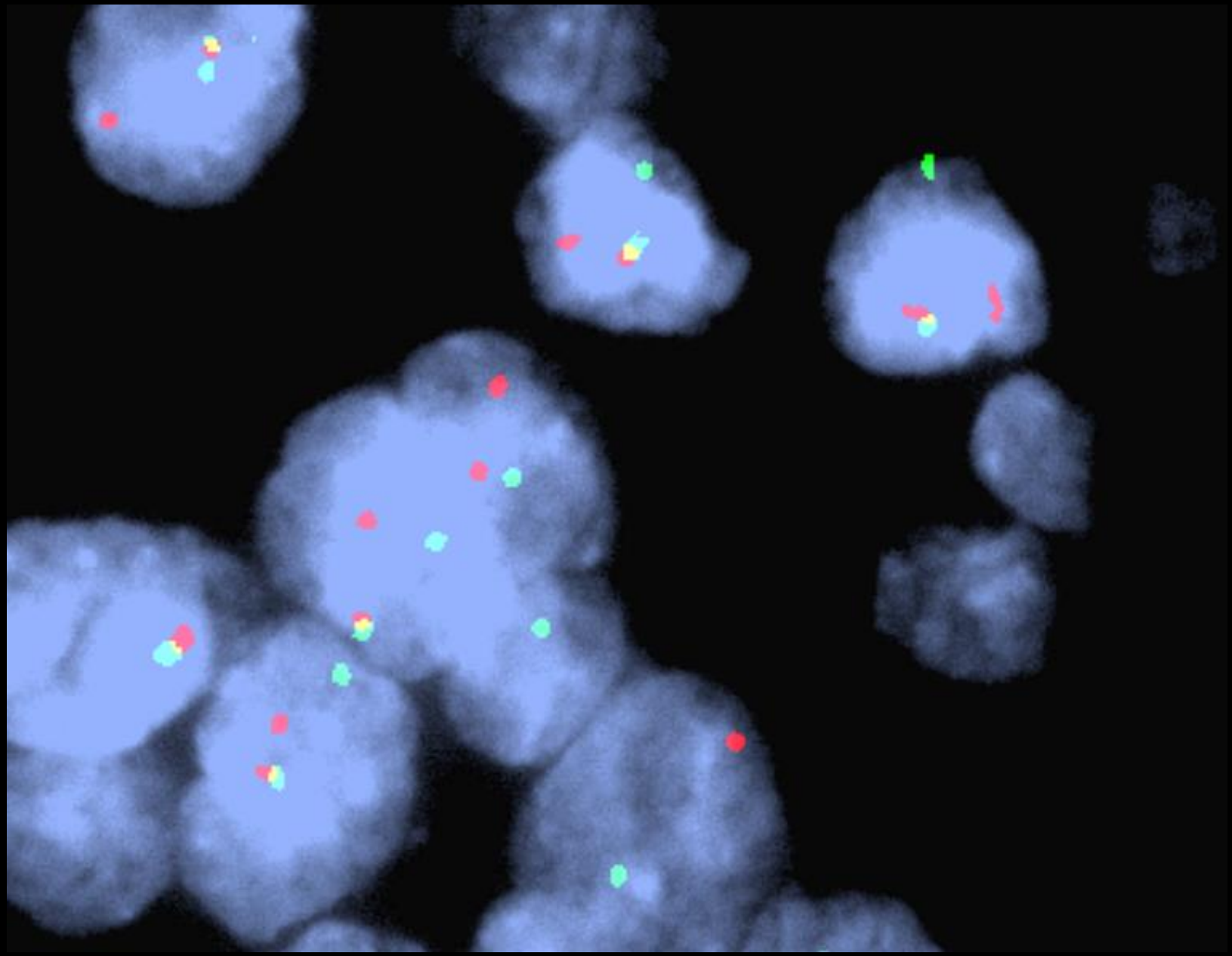
WHY?

Molecular Pathology

- ✓ Costly
- ✓ Time consuming
- ✓ Technical expertise
- ✓ Takes valuable space
- ✓ Challenging specimens
- ✓ Small volume testing

✓ It is Our Future





Ewing's Sarcoma, EWS break-apart probe

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Molecular Testing

Agenda

- Planning for a Molecular Lab
  - What is the purpose of your lab?
  - What will be the focus for test development?

Planning

- What is your mission?
- Who will run your lab?
- What type of assays?

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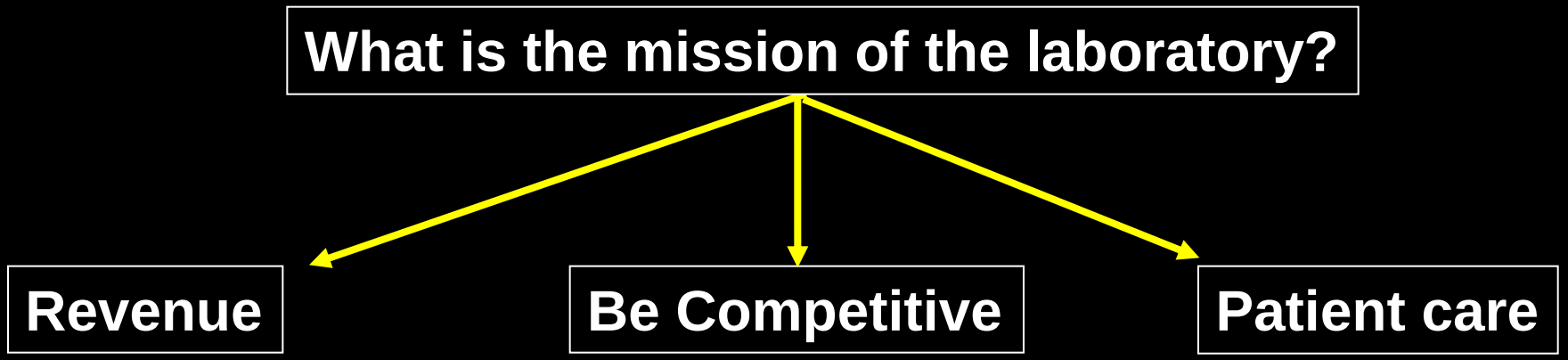
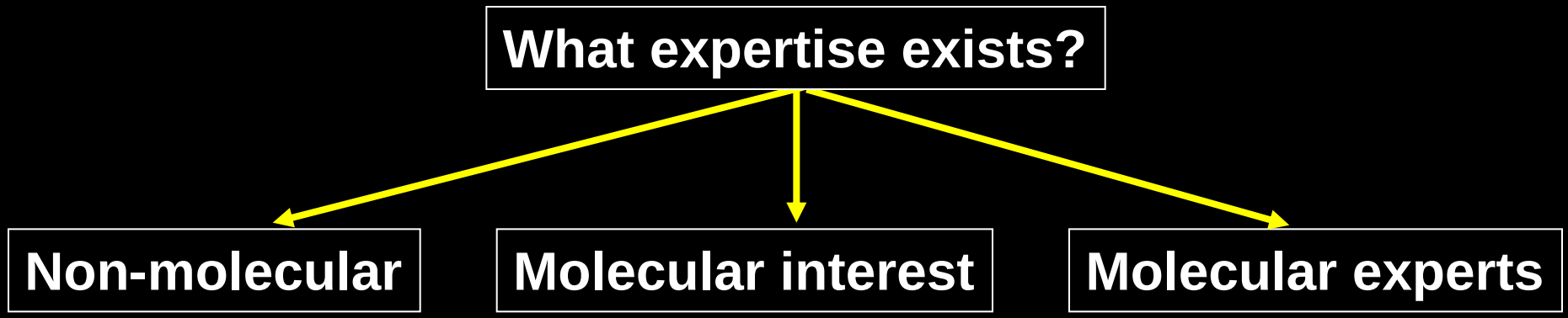
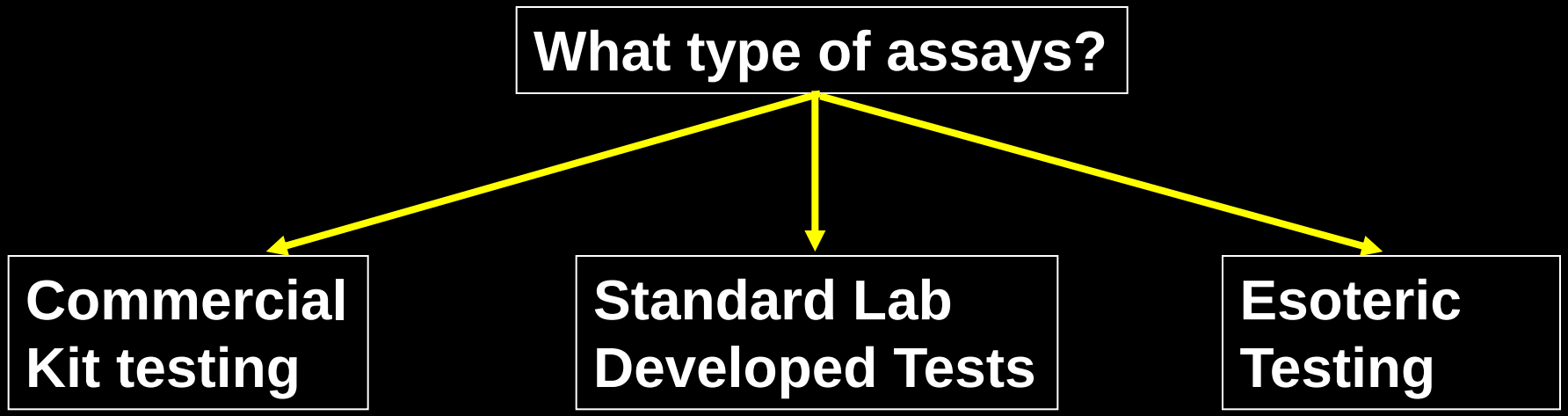
Mission


What is the mission of the laboratory?
Revenue
Be Competitive
Patient care

Expertise


What expertise exists?
Non-molecular
Molecular interest
Molecular experts

Assay Type


What type of assays?
Commercial Kit testing
Standard Lab Developed Tests
Esoteric Testing

Requirements

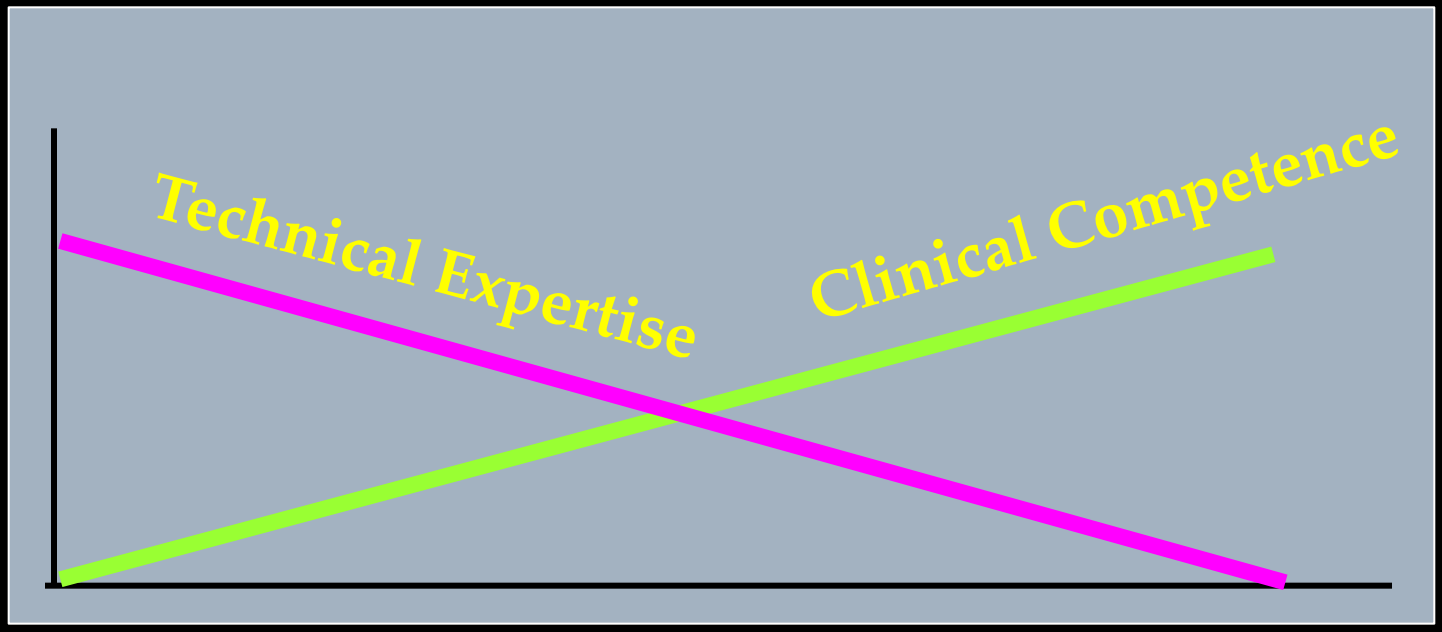
- What *personnel* do you need?
- What *space* do you need?
- What *resources* do you need?

Personnel You Need

- Medical Director
  - MD, MD/PhD, or PhD
- Technical Director
  - Masters or PhD
- Technicians & technologists
  - Formal medical technologist trained
  - Bachelors degree trained
- Secretarial support

Personnel Training

- Research Technicians
- Clinical laboratory technicians



Personnel Training

- Training: *Set your guidelines*
  - Observe technique
  - Perform technique under observation
  - Perform technique in parallel
  - Independent activity

- Document each step

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Space Requirements

- Concept: “clean” & “dirty”

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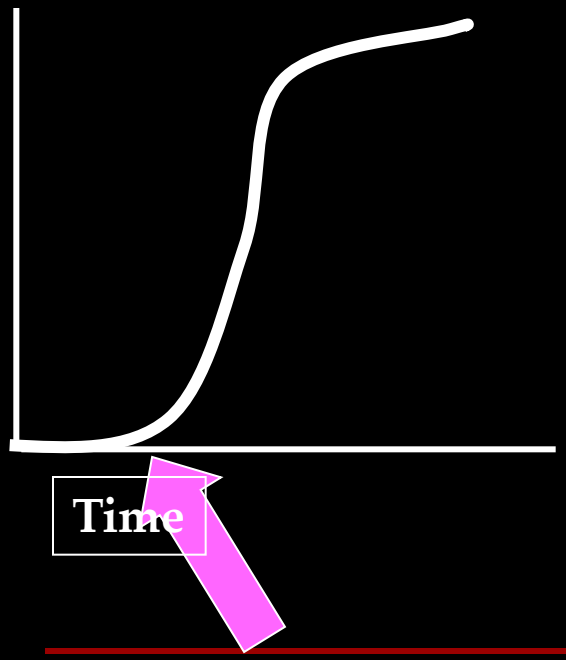
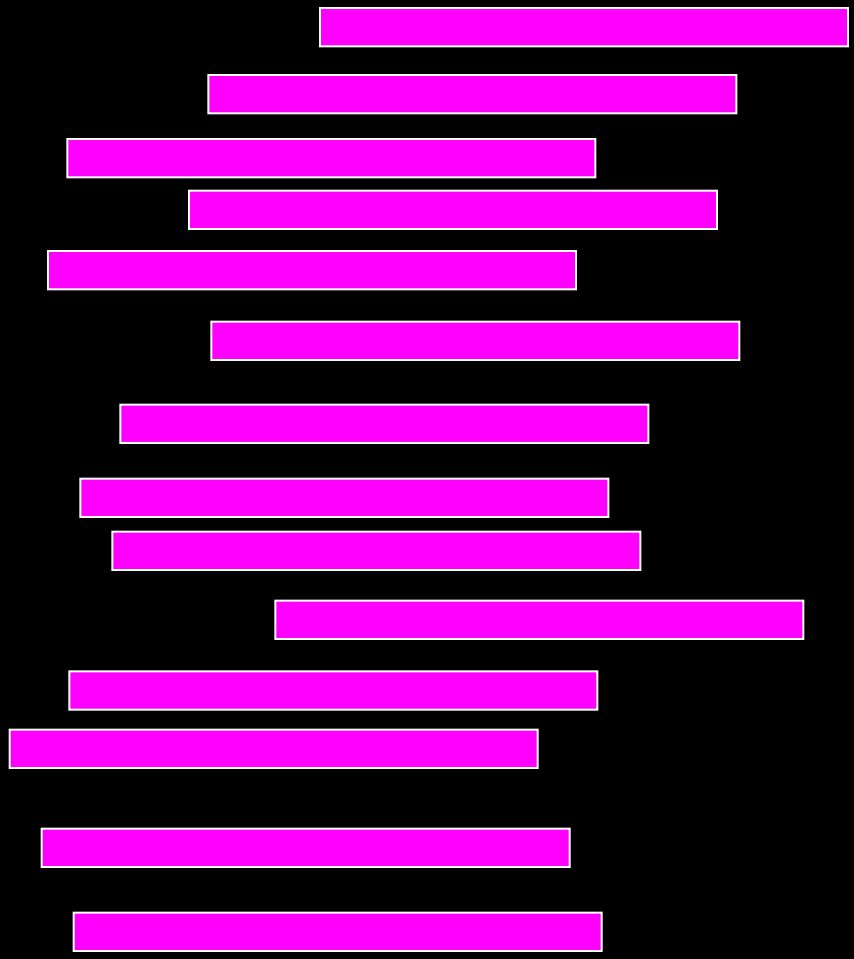
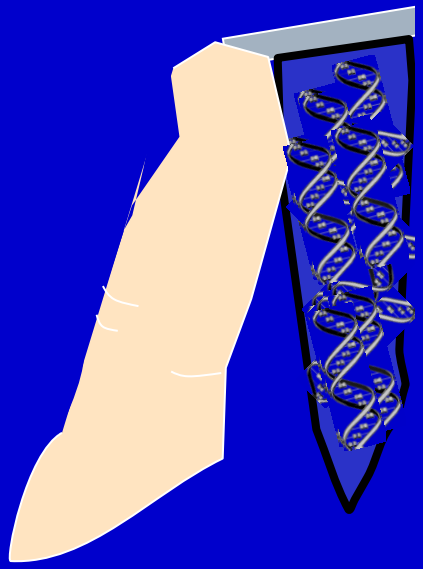
Amplification

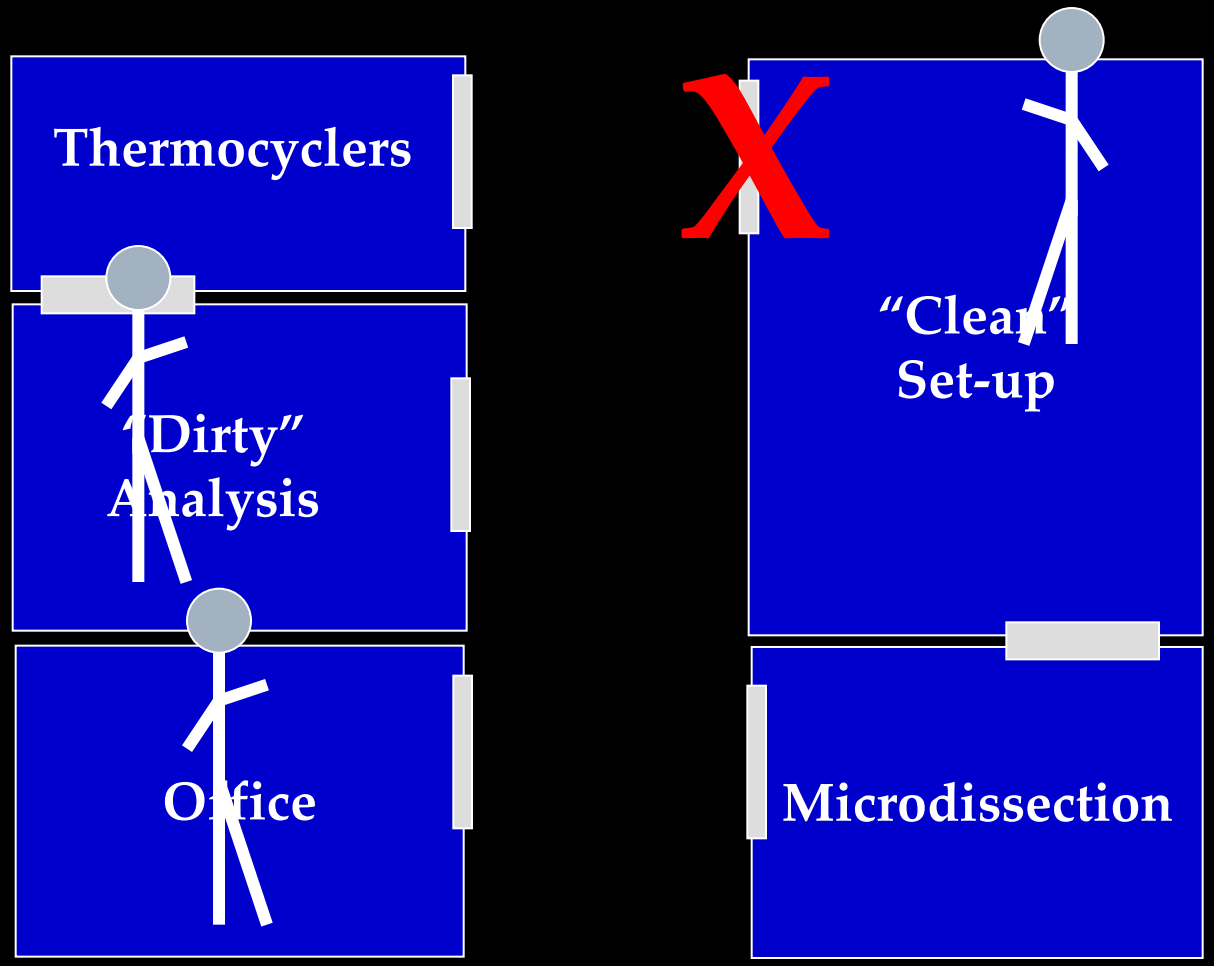




Contamination



Space


Thermocyclers
"Dirty" Analysis
Office
X
"Clean" Set-up
Microdissection

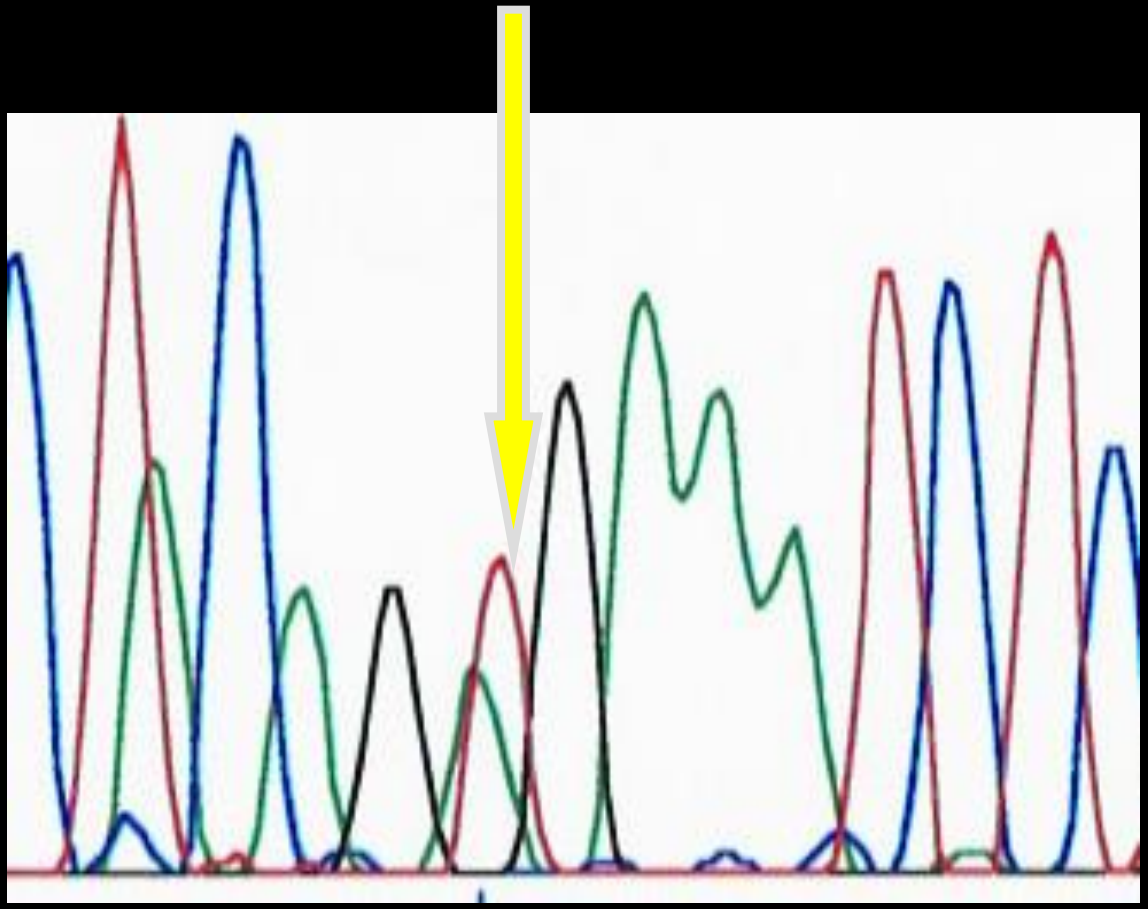
Resources

- Capital items
  - Large machines
- Small instrumentation
  - Start-up costs
- Reagents and consumables
  - Kits, tubes, pipettes

BRAF mutation

- Chromosome 7q34
- Serine-threonine kinase
  - RAS-RAF-MEK kinase pathway
- Mutation: V600E
  - Papillary thyroid carcinomas
  - Melanoma
  - Colon cancer

BRAF Gene Mutation



Choice, Choice, Choices!

- BRAF Gene mutation testing
  - Traditional Sanger sequencing
  - Pyrosequencing
  - Allele specific PCR
  - Quantitative PCR
  - Kit based testing
    - Commercial vendors
  - New immunohistochemical stain

BRAF Companion Diagnostic

Drug Link in Drugs@FDA	Therapeutic Area	Biomarker	Label Sections with Pharmacogenomic Information
Vemurafenib[1]	Oncology	BRAF	Indications and Usage Warning and Precautions Clinical Pharmacology Clinical Studies Patient Counseling Information

Resources

- Equipment needs
- What exists already
- Back-up machines

Your New Laboratory

- Set-up
  - At least 2-4 months
- Technical training
  - At least 3-6 months
- Assay development
  - At least 3-12 months

Laboratory Timeline

3 6 9 12 18

Set-up of space

Training of Technicians

Assay Development

Challenges

- Technical training
- Writing procedures
- Trouble shooting
- Validating new assays
- Integrating molecular into sign-out

Validation: ACCE*

- Analytical accuracy
- Clinical validation
- Clinical utility
- Ethical and social

Analytic Accuracy

- How well does the test detect the mutation?
- What is the sensitivity of the assay?

- *Performance on samples with known mutation status*
  - *Cell lines*
  - *Sample exchange*
  - *Tested with alternate method*

Clinical Validity

- How well does the mutation predict disease?

- *Use assay on samples with and without the disease*
  - *Positive predictive value*
  - *Negative predictive value*
  - *False positive rate*

Question

How many samples will you test?

- ✓ Disease prevalence
- ✓ Access to samples
- ✓ Expected positive rate

Clinical Utility

- When will you use the assay?

- *Guidelines*
  - *Sample requirements*
  - *Clinical reasons for getting the test*
  - *Clinical interpretation of positive and negative results*

Ethical Issues

- How will the assay affect the patient?

Validation Summary

- Description of disease and mutation
- Description of validation procedure used
- Results from validation studies
- Guidelines for specimen type and selection process
- Laboratory Director signature

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