Fungal Genomics and Sustainable Development

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Outline

Genomics

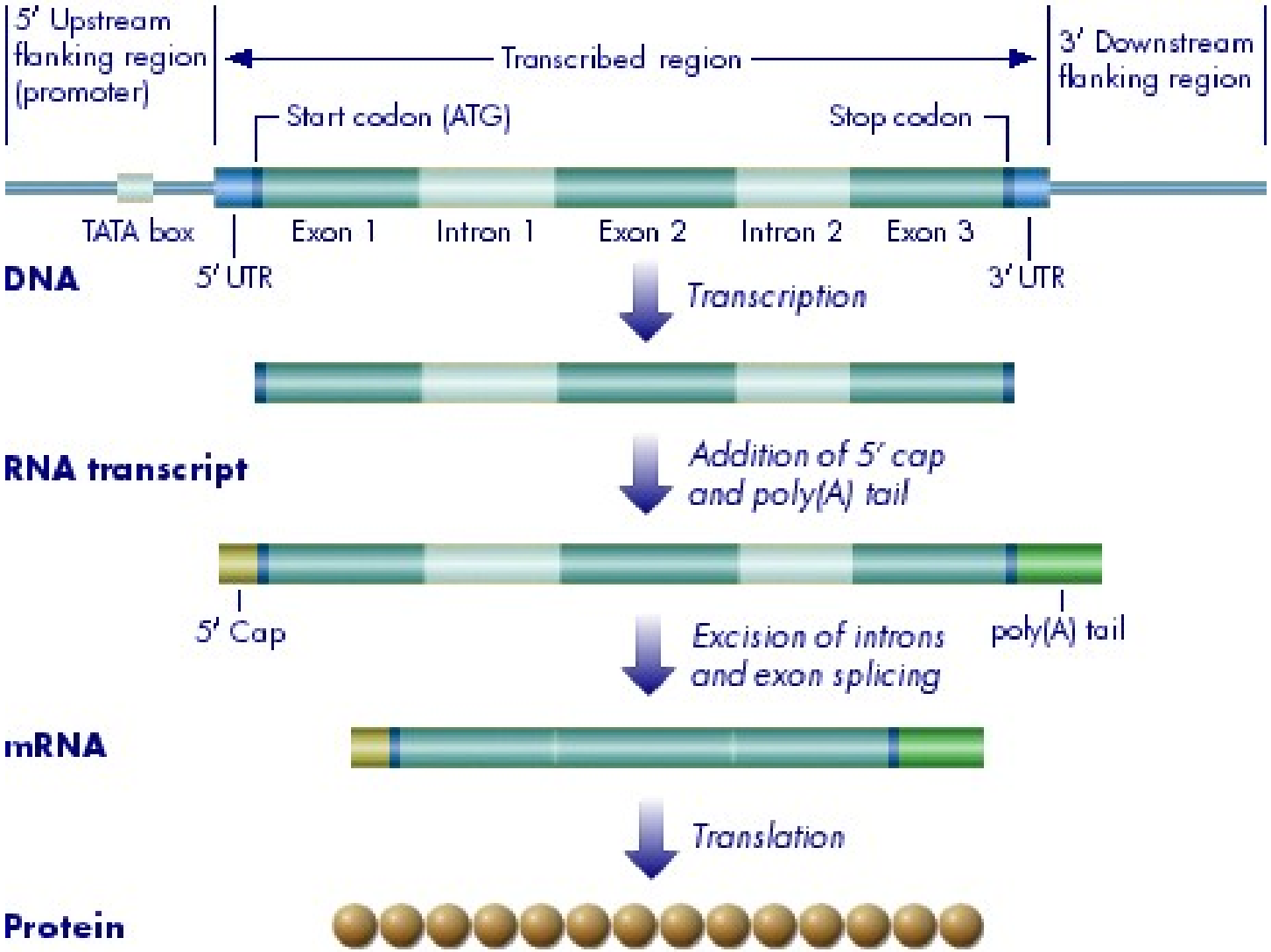
Enzymes and Bio-Processing for Sustainable Development

Overview of the Fungal Genomics Project

Bioinformatics for the Fungal Genomics Project

Conclusion

Biology — Transcription and Translation



Biology — Transcription Regulation

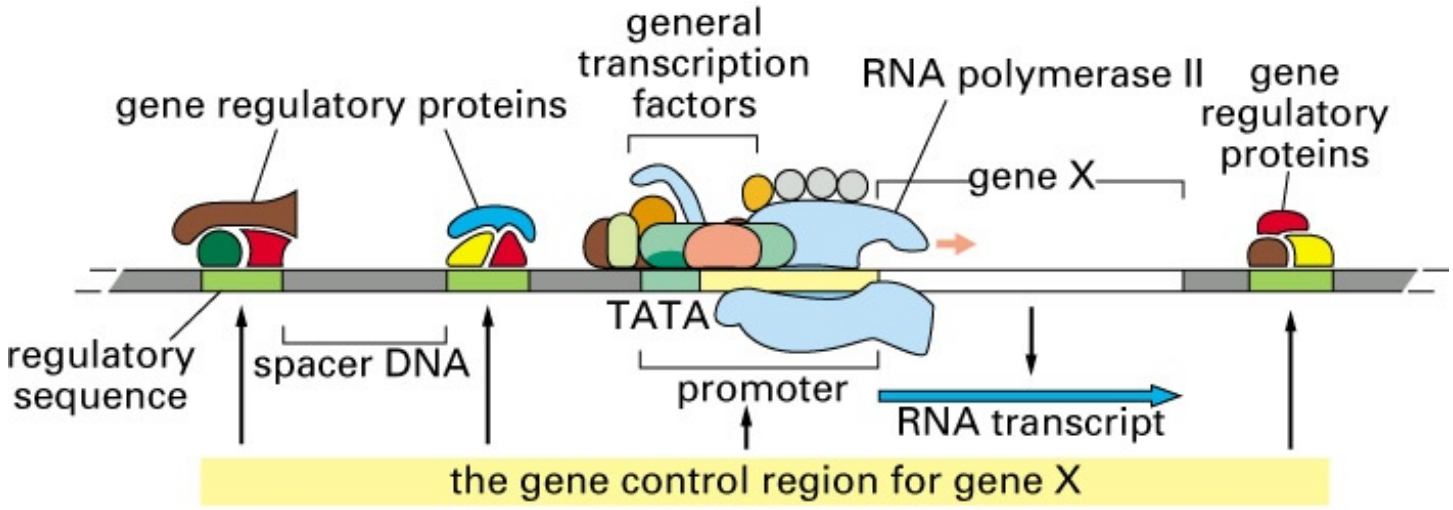
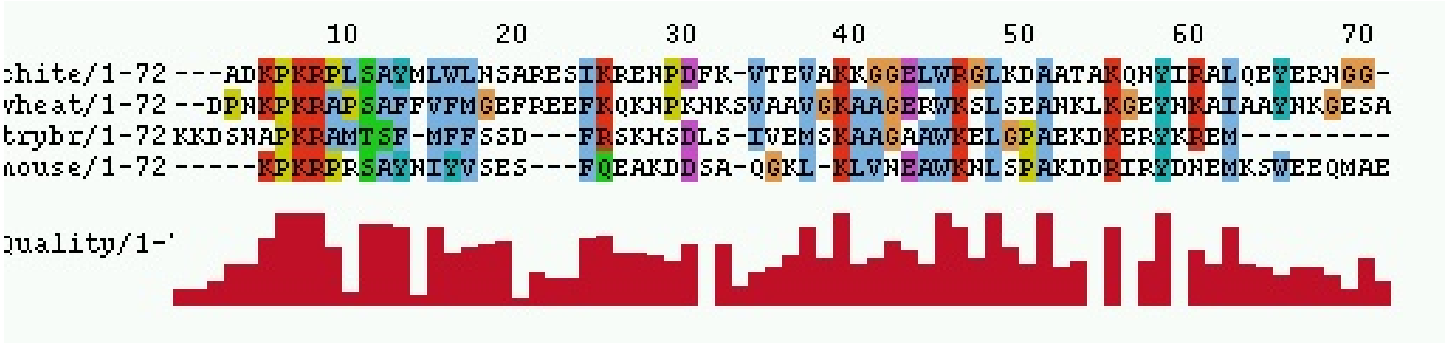


Figure 7–41. Molecular Biology of the Cell, 4th Edition.

Biology implies ...

... use Multiple Sequence Alignment (MSA)

MSA Problem: Given a set of protein sequences, and an *objective function*, determine the *optimal* alignment of the sequences.



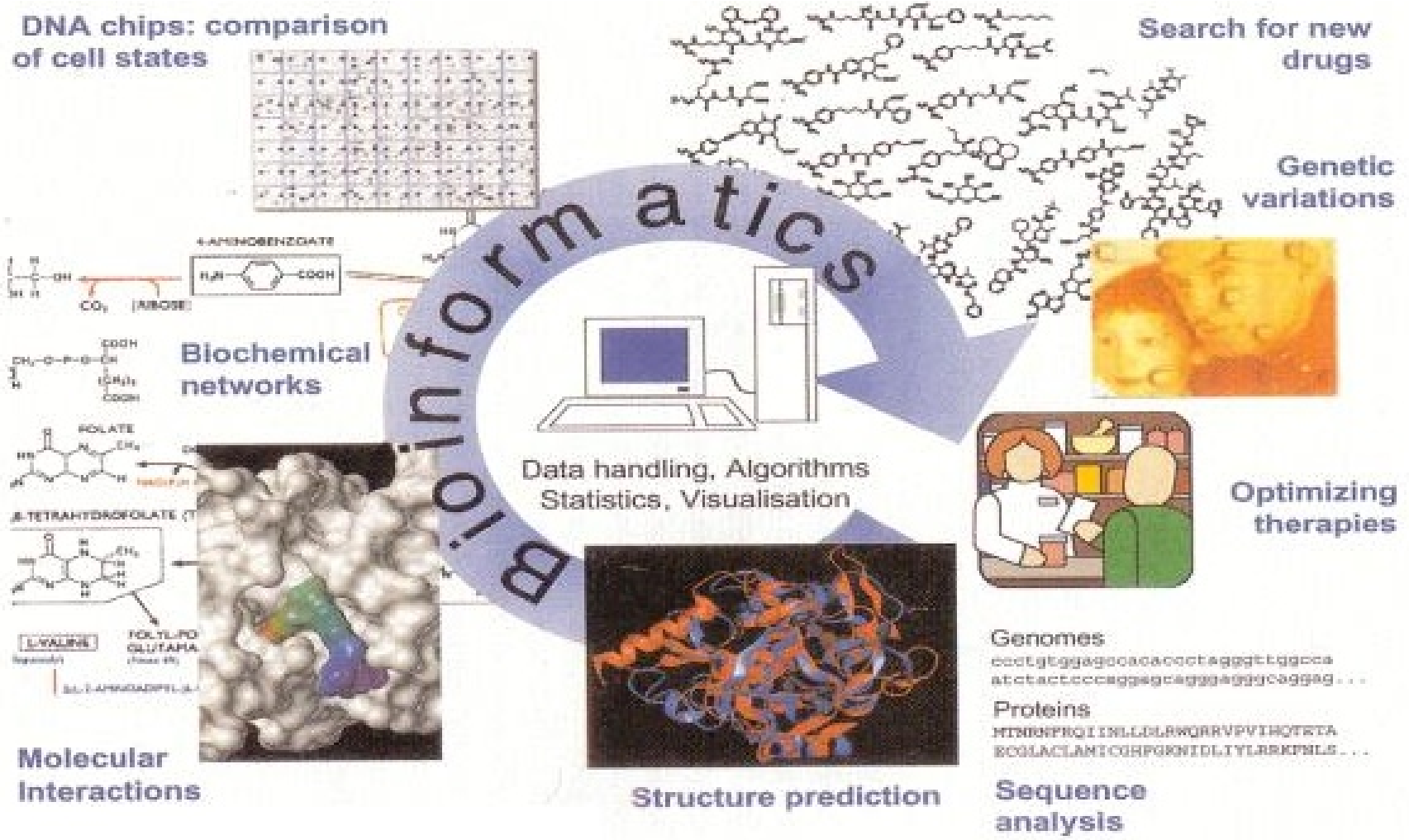
Why?

Amino acid *sequence*

determines protein *structure*

determines enzyme *function*

Genomics and Bioinformatics — Health Perspective



Genomics

high-throughput, collecting or using all genes

Genomics project: determine full chromosome sequence, predict genes

EST Project: mine mRNA, assemble into "unigenes"

Microarray Project: genome-on-a-chip, study gene activity

Expression Project: splice gene into host, produce enzyme

Enzyme Assay and Characterization Project: chemistry and biochemistry

Proteomics, ...

GenomeQuébec

The Bio-Economy

Renewable Bioresource Feedstock

- Plants
  - crops
  - trees
  - algae
- Animals, fish
- Microorganisms
- Organic residues
  - municipal
  - industrial
  - agricultural
  - forestry
  - aquaculture

Bioprocess Technology

Biocatalysis (Enzymes)

Fermentation (Microorganisms)

Physical – Chemical Process Technology

Extraction

Pyrolysis

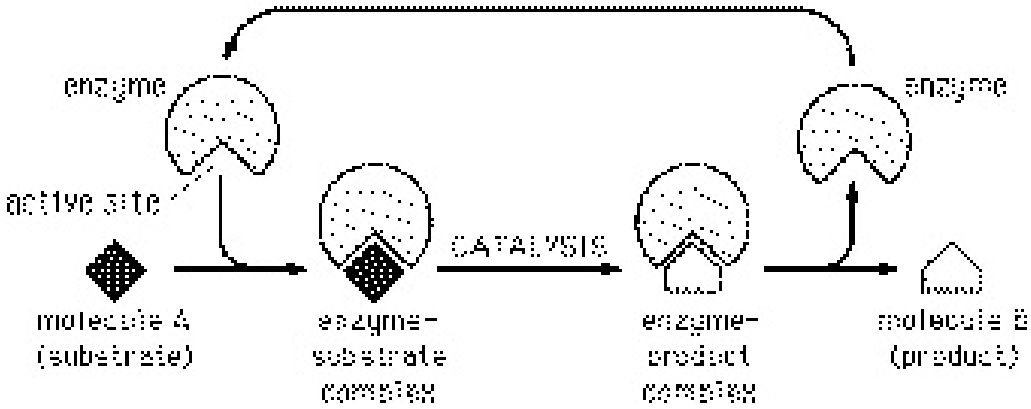
Gasification

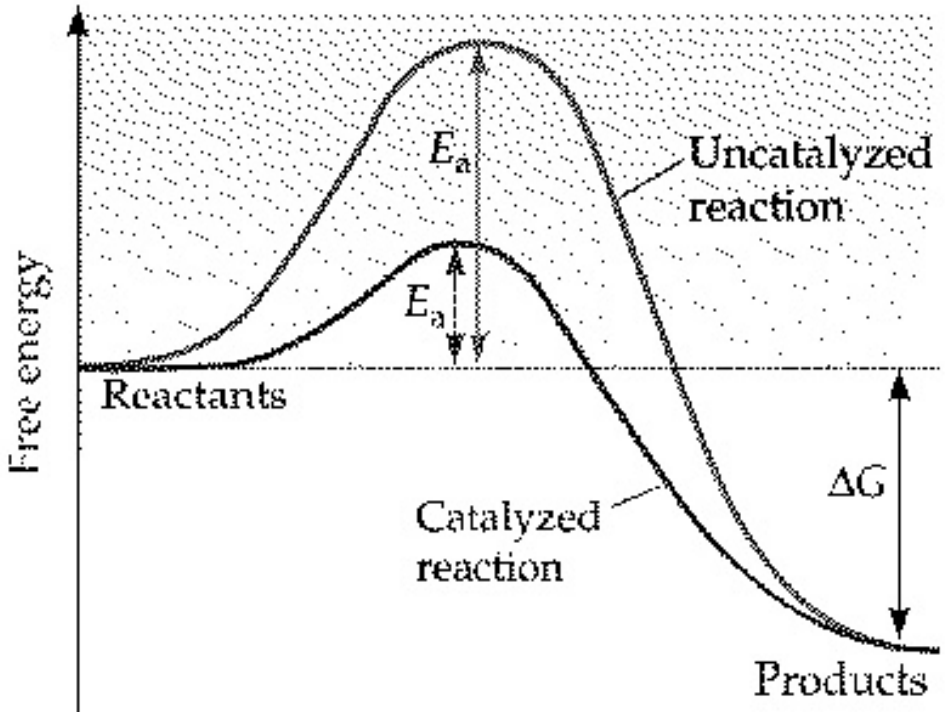
Industrial Bioproducts

- Bioenergy and Biofuels
- Manufactured products:
  - biochemicals
  - biosolvents
  - bioplastics
  - 'smart' biomaterials
  - biolubricants
  - biosurfactants
  - bioadhesives
  - biocatalysts
  - biosensors

What is an Enzyme?

Enzyme is a *protein* that *catalyses* a reaction.





What is an Enzyme?

Enzymes are very *specific*.

Enzymes are very *efficient catalysts*.

Some Rate Enhancements Produced by Enzymes

Cyclophilin	10^5
Carbonic anhydrase	10^7
Triose phosphate isomerase	10^9
Carboxypeptidase A	10^{11}
Phosphoglucomutase	10^{12}
Succinyl-CoA transferase	10^{13}
Urease	10^{14}
Orotidine monophosphate decarboxylase	10^{17}

Applications of Enzymes in Industrial and Environmental Processes

Industrial/Environmental Processes	Enzymes	Applications/Substrates
pulp and paper manufacturing	lignin peroxidases, manganese peroxidases, laccases, cellulases, pectinases, xylanases, mannases, esterases, lipases	lignin, hemicellulose, cellulose, pitch
waste treatment and decontamination	lignin peroxidases, manganese peroxidases, laccases, cytochrome P450, monoxygenases, esterases, lipases	chloroaniline, lindane, chloro-dibenzo-p-dioxines, chlorobiphenyls, chlorophenols, DDT, polyaromatic hydrocarbons (anthracene, fluoranthene, benzopyrene), azo and heterocyclic dyes, tropaeolin, azure B, nitrotoluene, creosote, diesel oil, plastics
coal liquefaction	peroxidases, laccases, esterases	coal
ecotoxicology assessments	lignin peroxidases, manganese peroxidases, cytochrome P-450 monoxygenases, glutathione-transferases	biosensors, biomarkers, model of mammalian xenobiotic metabolism
household and industrial detergents	proteases, lipases, amylases, cellulases	cleaning laundry and dishes at a wide range of temperatures

Applications of Enzymes in Industrial and Environmental Processes

Industrial/Environmental Processes	Enzymes	Applications/Substrates
food processing and functionalities	proteases, invertases, alpha-amylases, pectinases, glucomylases, cellulases, xylanases, lactases, glucose isomerases	clarification and extraction of juice, flavour enhancement, lactose modification, meat extraction, arome production, production of sugar, production of syrup, production of alcohol, texture of pasta and noodles, production of baby formula etc.
fuel alcohol	cellulases, amylases, glucoamylases, xylanases, proteases	starch or sugar based raw plant materials into alcohol
baking	alpha-amylases, glucose oxidasess, lipases, lipoxygenases, xylanases, proteases	maximize fermentation, oxidize sulfydrl groups, dough conditioning, bleaching and strengthening dough
brewing	alpha-acetolactate decarboxylases, beta-glucanases, cellulases, xylanases, proteases	reduce beer maturation time, improve yield and filterability, extract protein to give desirable nitrogen level
wine making	pectinases, glucosidases, cellulases	release colour and aroma compounds, clarification, improve wine stabilization and filtration
animal feed	phytases, xylanases, beta-glucanasess, alpha-amylasess, proteases, endo-xylanases	improve digestibility; substrates include phytate, glucan, starch, xylan, raffinose, stachyose, pectin-like polysaccharides

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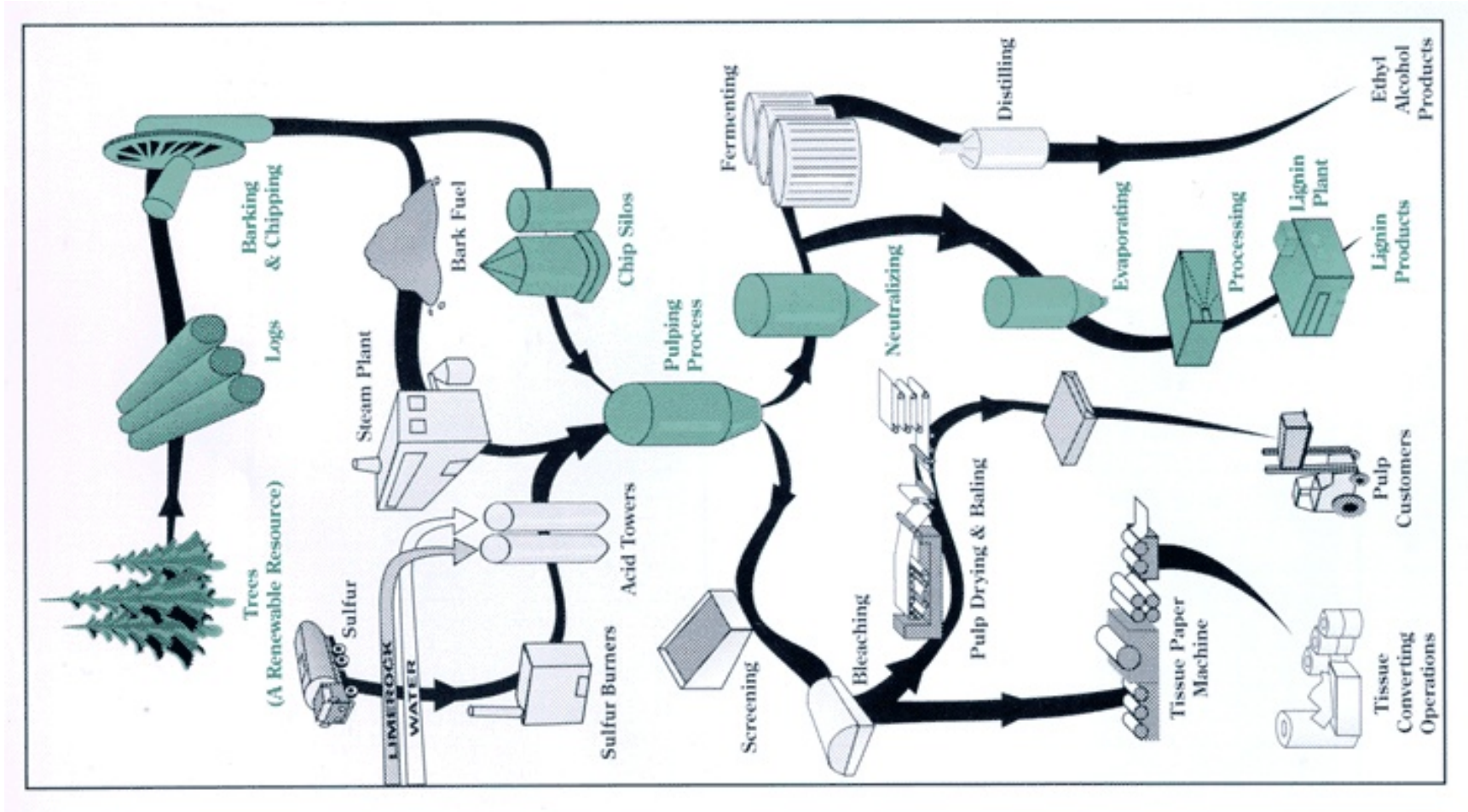
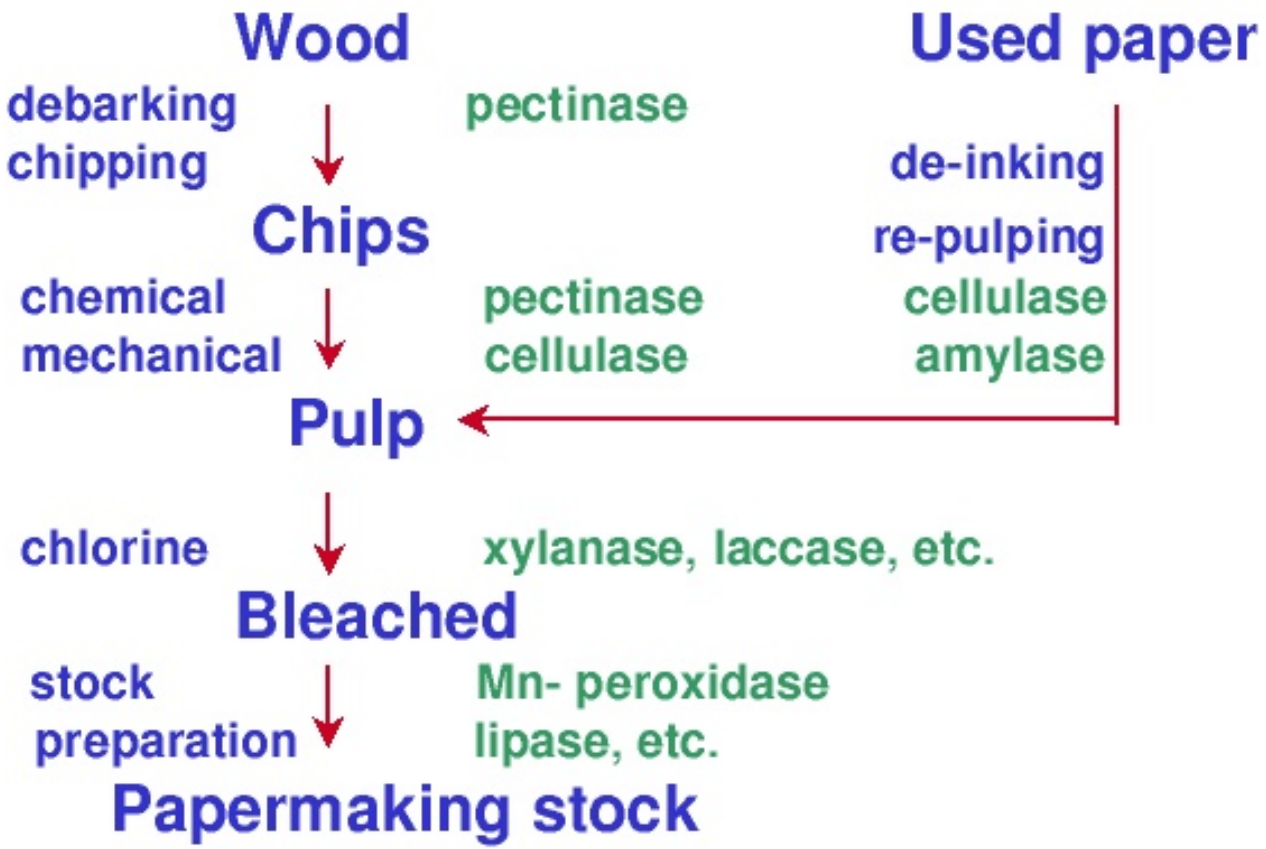
pharmaceutical and fine chemicals	cytochrome P450 oxygenases, glutathione transferases, lactases, alpha-galactosidases	biotransformation of antibiotics, antitumourals, steroids, use of enzyme in therapy etc.
textile	cellulases, proteases, amylases, catalases	dye removal, de-sizing of starch, improve colour brightness and smoothness, degradation of hydrogen peroxide, degumming
leather processing	proteases and lipases	removing fat and hair from skin and hide
personal care	proteases, glucoamylases, glucose oxidases, catalases	tooth paste, cleaning solution for contact lens

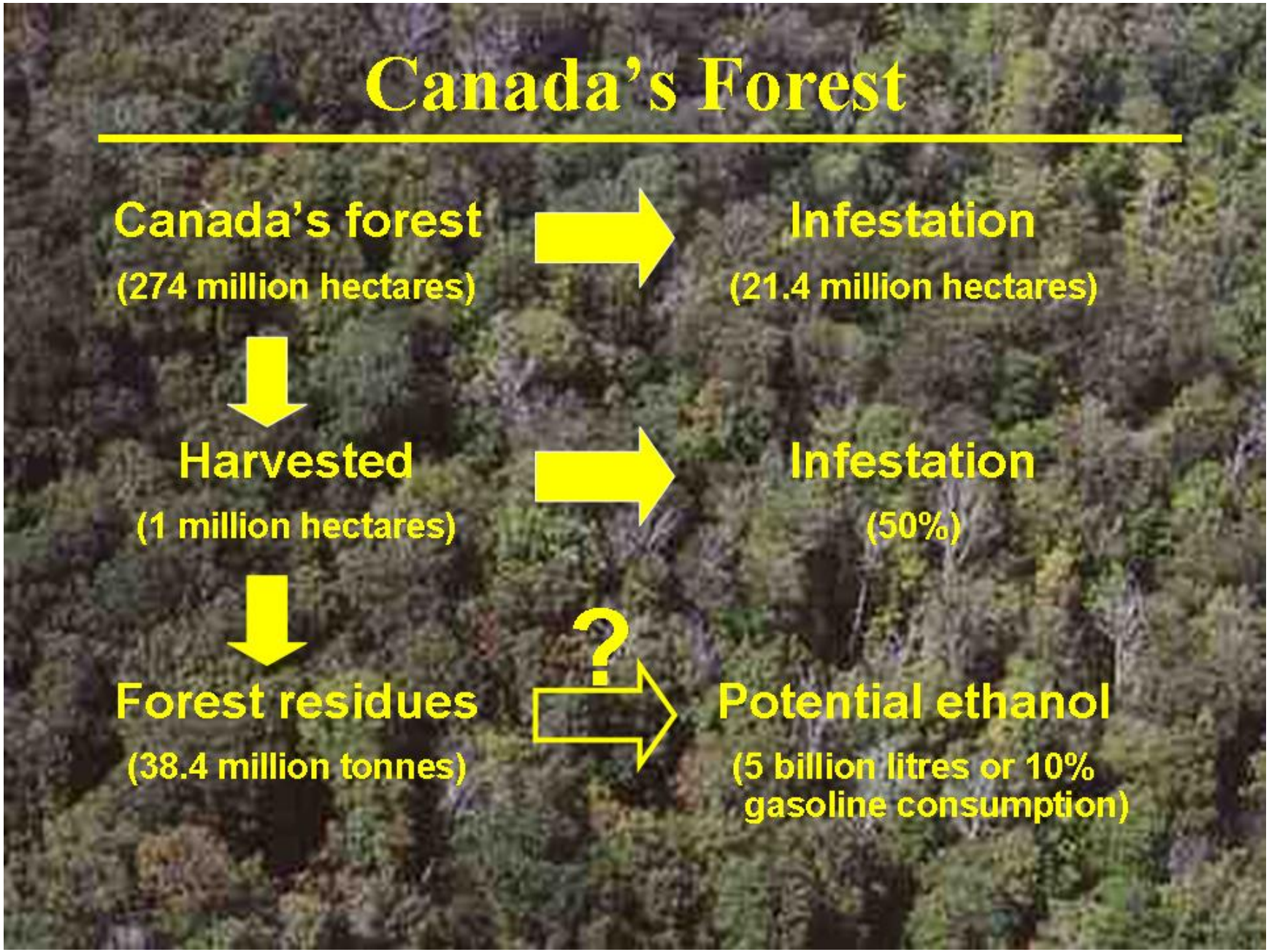
Industrial Processing of Trees



Industrial Uses of Enzymes: Paper



Industrial Uses of Enzymes: Bio-Ethanol from Forestry Waste



Does Biotechnology for Sustainability Work?

OECD report on 21 case studies

wide range of industrial sectors: pharmaceuticals, fine chemicals, bulk chemicals, food and feed, textiles, pulp and paper, minerals and energy.

in Austria, Canada, Germany, Japan, the Netherlands, the United Kingdom, the United States and South Africa.

"their use invariably leads to reduction in operating costs or capital costs or both"

"New "bio-processes" can substantially reduce emissions and the use of hazardous raw materials."

"They result in fewer by-products, generate fewer waste materials and consume less energy."

The Application of Biotechnology to Industrial Sustainability, 148 pages, OECD, Paris 2001

The Fungal Genomics Project

Identify useful enzymes for industrial applications

secreted by 14 species of fungi

by constructing cDNA libraries with 5000 genes per species

and detecting genes similar to known enzymes

and

by constructing cDNA microarray for each species

and detecting genes expressed under related conditions

Characterize selected enzymes

Fungal Species

White-rot fungi:
- *Phanerochaete chrysosporium*
- *Trametes versicolor*
- *Lentinula edodes*

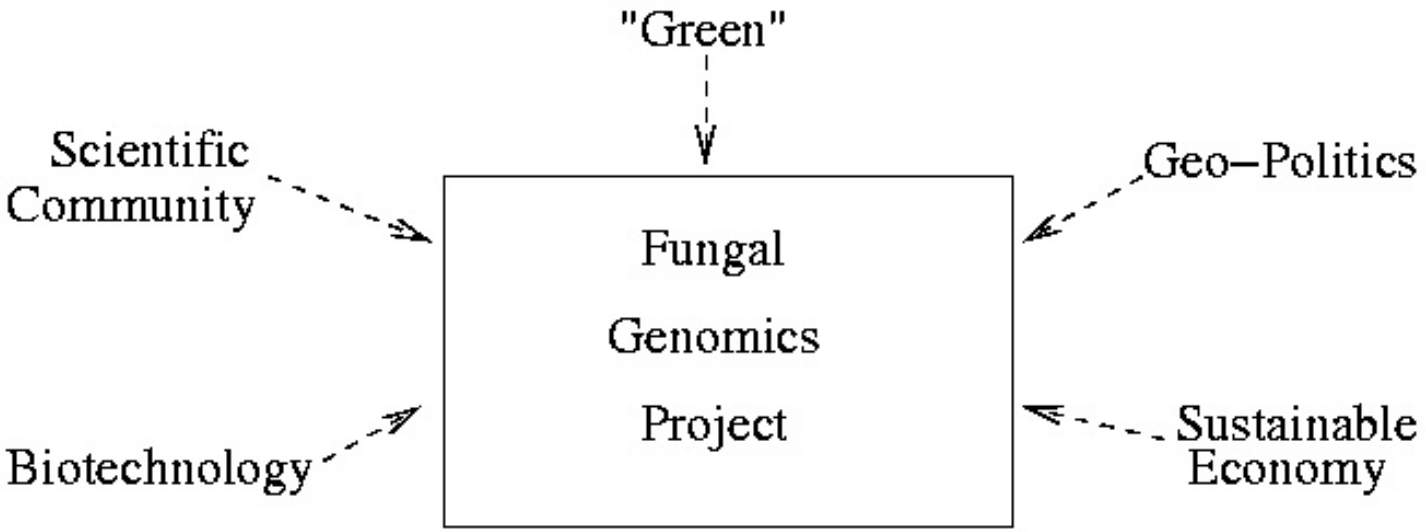
Other lignin and pitch degrading fungi:
- *Gloeophyllum trabeum*
- *Ophiostoma piliferum*
- *Corpinus cinereus*

Pollutant degrading fungi:
- *Aureobasidium pullulans*
- *Amorphotheca resinae*
- *Leucosporidium scottii*
- *Cunninghamella elegans*

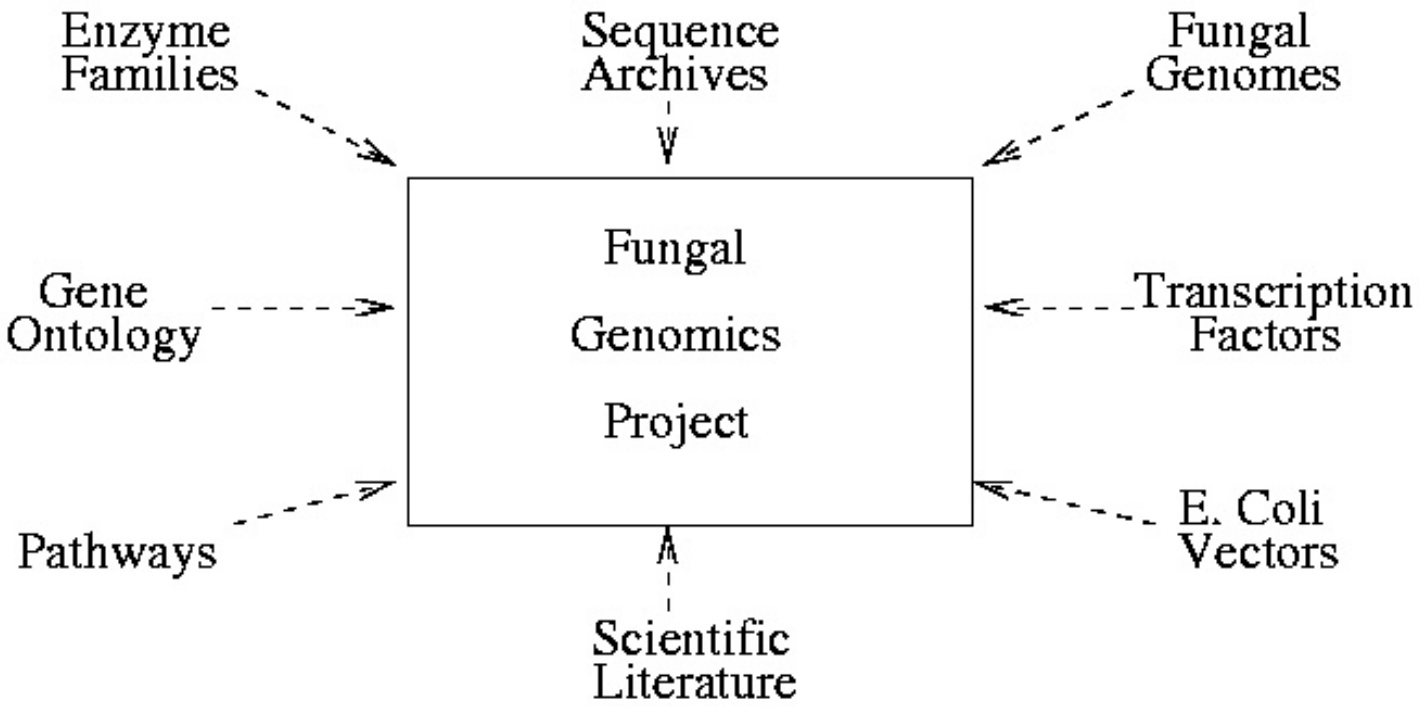
Freeze-tolerant fungi:
- *Chrysosporium pannorum*
- *Cryptococcus laurentii*

Thermophilic composters:
- *Thermomyces lanuginosa* (60^0C)
- *Chaetomium thermophile* (50^0C)

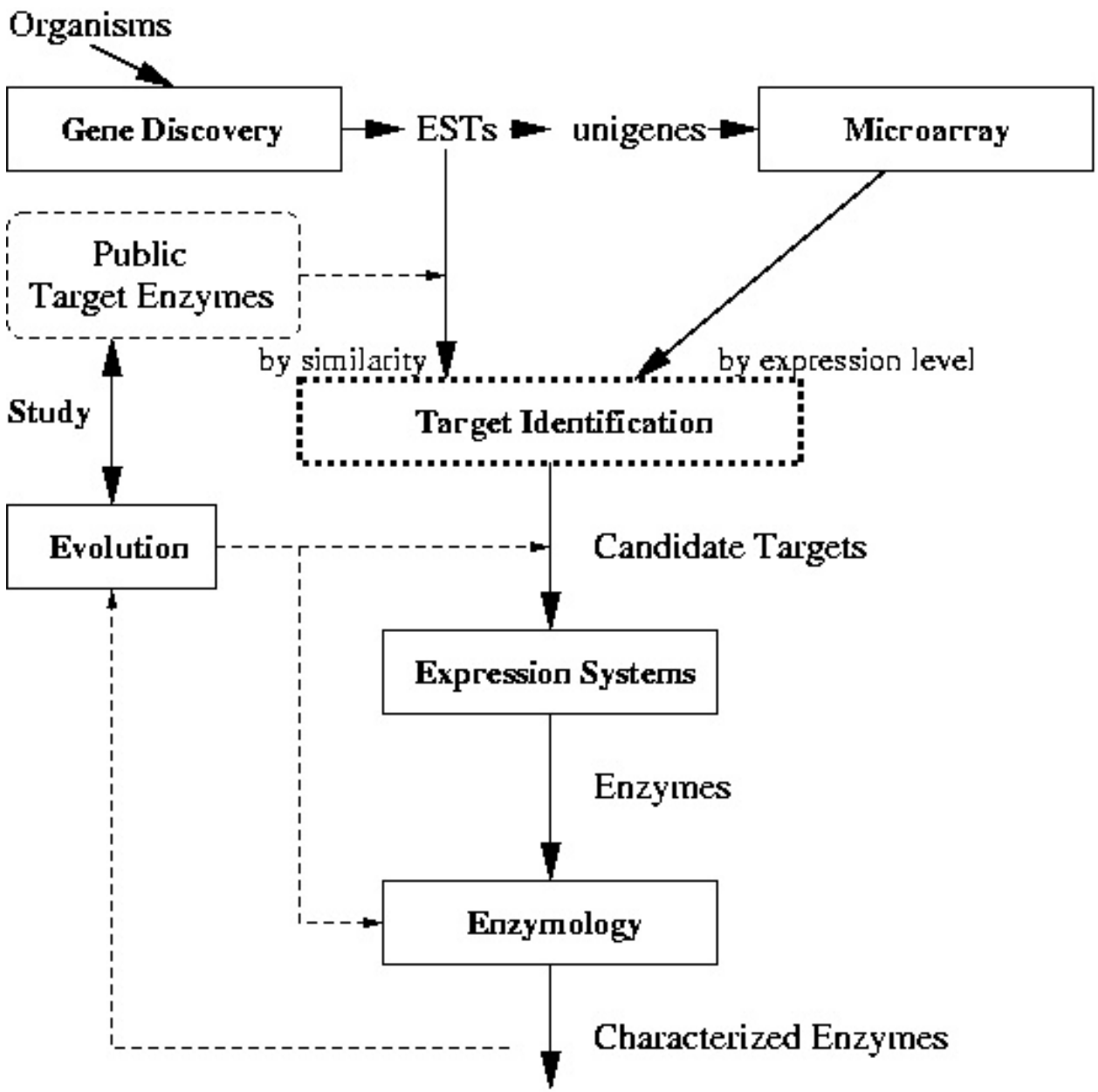
External Context of Fungal Genomics Project



External Context — Public Data



The Fungal Genomics Project



What is Bioinformatics?

To store, organize & analyze biological data

Models — mathematical, statistical, physical

Algorithms — string, tree, graph, patterns

Database technology

Workflow and computational grids

Intelligent reasoning — expert systems, agents

Web access, visualization, usability

Rapid system development and evolution

Bioinformatics for the Fungal Genomics Project

Aims

ensure high-quality experimental data is collected

know quality, confidence, provenance of data

automate data analysis & report generation

Concerns

Hide the technology, Do the science

Confidence in results/interpretation

Flexibility, staying ahead of the curve

Bang for the buck
computational resources can be huge

Bioinformatics for the Fungal Genomics Project

Bioinformatics — Current Needs

- Materials Tracking
- Data Collection
- Quality Control of Data and Process
- Analysis of Sequences — Automated Annotation
- Analysis of Target Sequences — Manual Curation
- Analysis of Microarray Data — Basic Expression

Bioinformatics — Future Needs

- Analysis of Microarray Data — Profiling
- Analysis of Microarray Data — Integrating other Evidence
- Enzyme Families — Phylogeny, Multiple Alignment, Classifiers
- Enzyme Families — Predicting Kinetics

Bioinformatics Platform

Bioinformatics — Housekeeping Tasks

Materials Tracking

identify (bar code labels) and record materials

know location of all relevant physical materials

know mappings caused by physical transfer of materials

Data Collection

keep all data secure and accessible

know quality and provenance of all data

support analysis and interpretation of data

Quality Control

know the quality of data

provide reports to monitor quality of lab processes

assist in diagnosis of problems with lab processes

Bioinformatics — Analysis Tasks

Sequence Analysis

determine high-quality sequence segment

... base call quality, remove contaminants, trim

assemble ESTs into unigenes

Sequence Annotation

similarity against known nucleic and protein sequences

... especially against targetted enzyme families

search for protein motifs and domains

is it secreted enzyme? other localization info?

classify to Gene Ontology category

Microarray Data Analysis

normalize: QC determines bad spots and dynamic range

set threshold for significant expression levels

determine highly expressed genes

Conclusion

Bio-Processing using fungal enzymes is a way to sustainability.

Bioinformatics for finding and understanding fungal enzymes is a rich, challenging area for interdisciplinary research

Long-term Challenges in Bioinformatics:

- Enzyme Family Data — be able to predict kinetics
- Scientific Literature — be able to mine knowledge
- Gene Expression Data — be able to predict function
  — be able to predict regulation
- ... Systems Biology

Exciting times ahead !!!

Acknowledgements

Thank you!

Questions?