PATHOLOGY IN AQUACULTURE

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Health Management

- Prevent introduction of disease to healthy animals.
- Prevent propagation of existing disease agents.
- Production of healthy, high quality fish.

INTRODUCTION

Aquaculture ...

- ***Asia - cradle of aquaculture***
- ***Demand of aqua. products,***
- ***Intensification …***

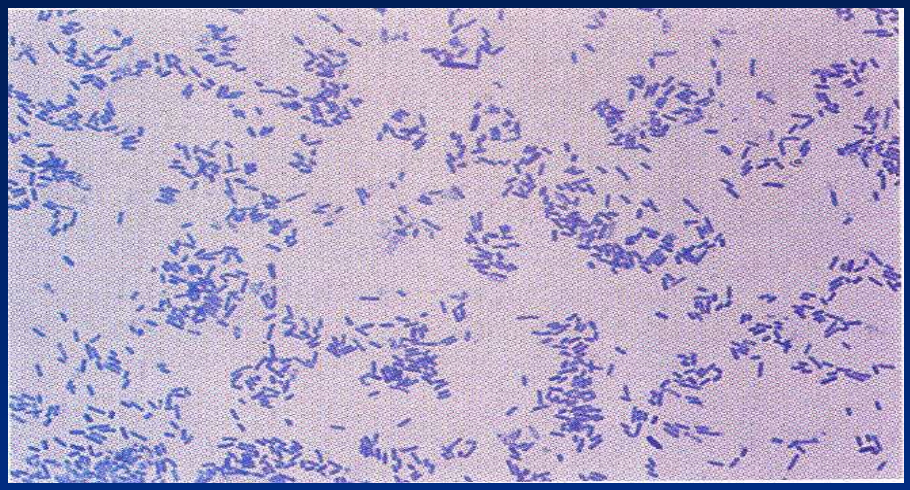
What is disease?

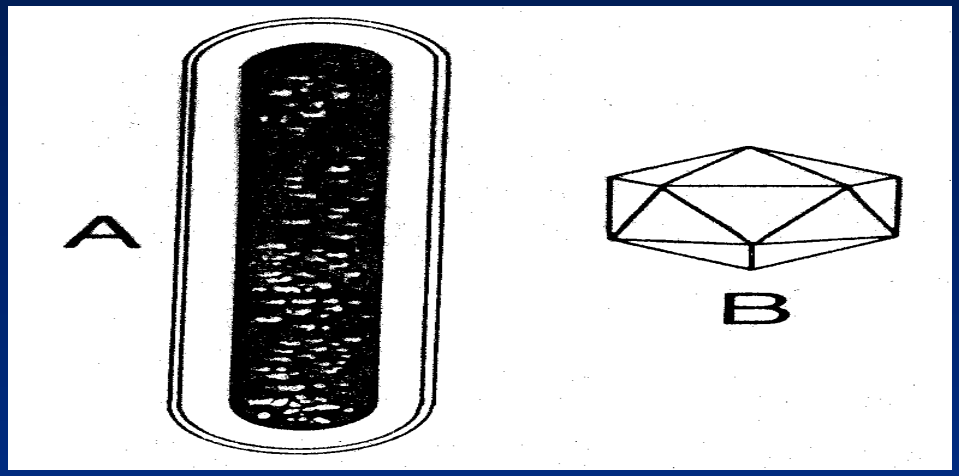
- **Deranging of normal health**
  - **Structural**
  - **Behaviourable**

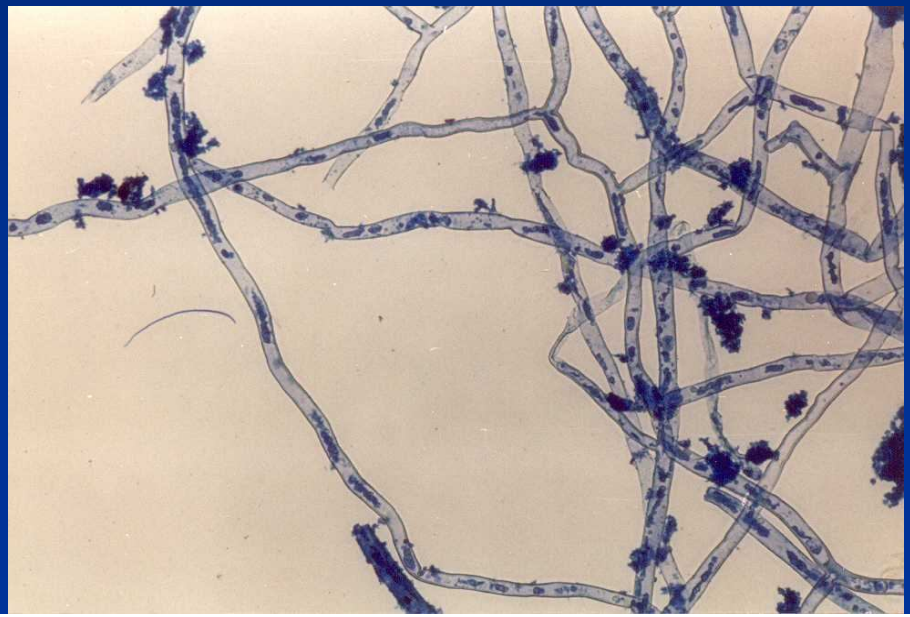
When Disease Occur ?

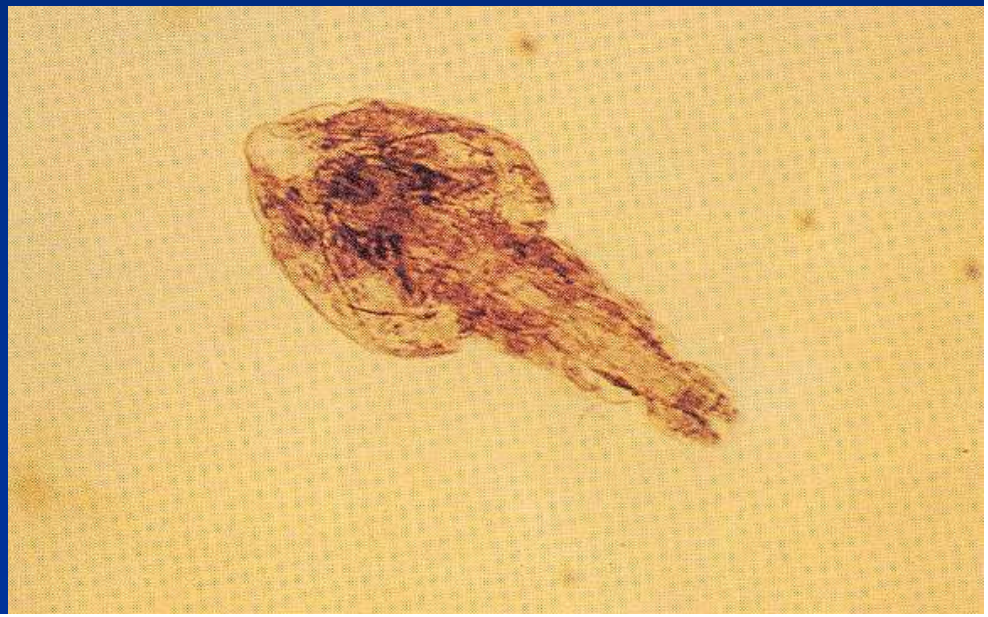
- **Inability of the host**
- **Dominance or Virulence of the pathogens**

What are pathogens ?









DISEASE IS THE MAJOR DINOMINATOR IN AQUACULTURE SUCCESS

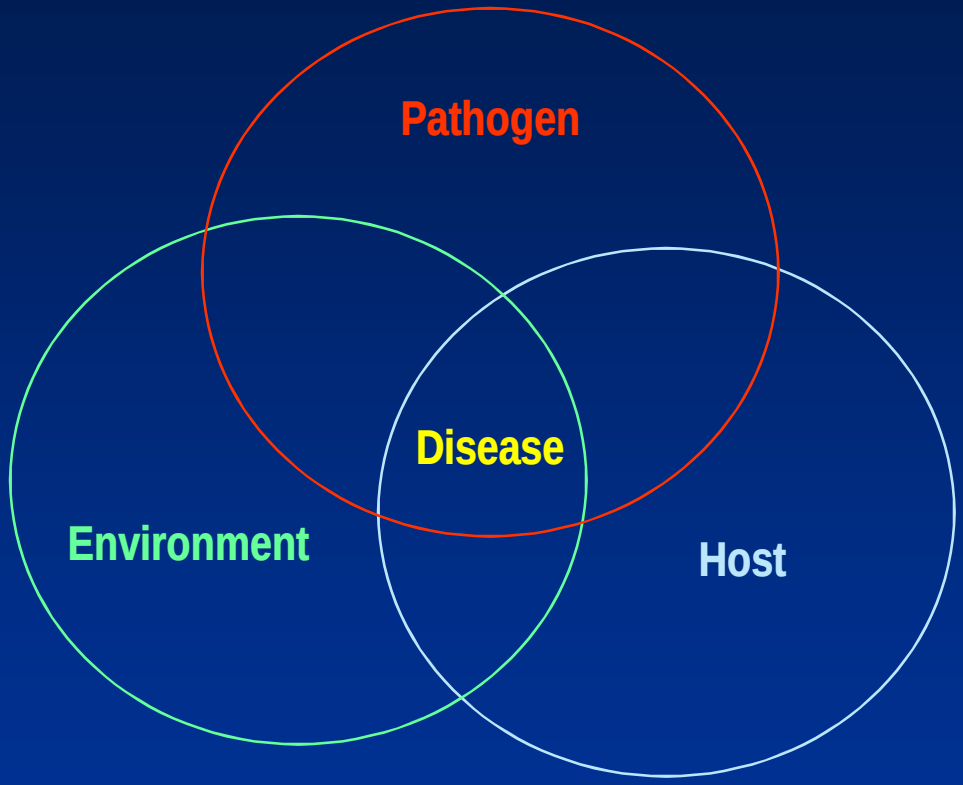
Disease outbreak in cultivable organism has leads to severe losses across different geographical locations since early 1990s

They are major constrains to aquaculture production and trade, affecting socio economic development in the sector worldwide

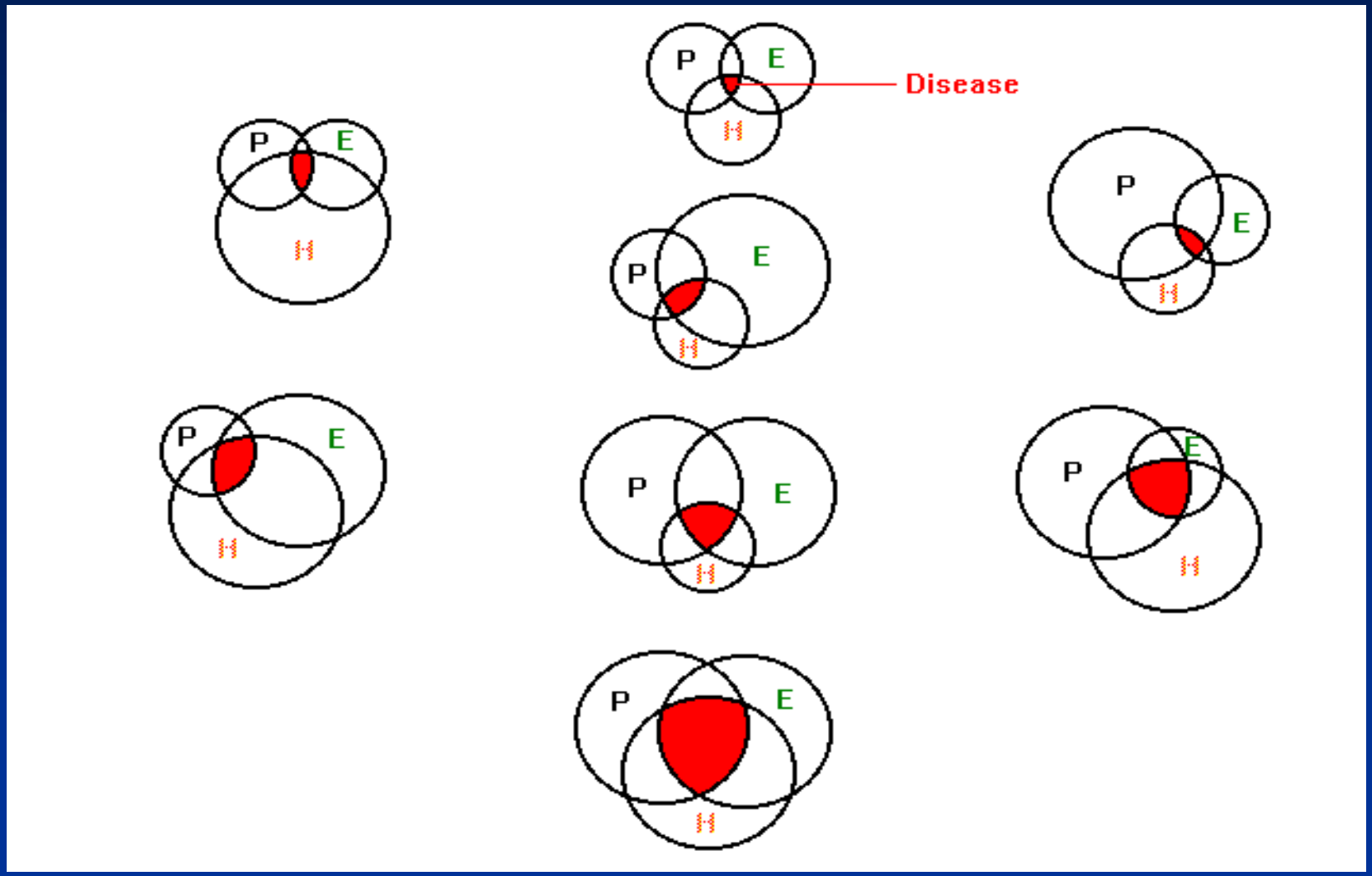
The risk of disease losses is likely to increase due to new bacterial and fungal pathogens being identified every year

Estimated annual loss in aquaculture production 3 trillion USD due to disease and environmental related problems

Disease manifestation process



DMP – Four “K”



Factors Influencing Disease Transmission

Agent

- Infectivity
- Pathogenicity
- Virulence
- Immunogenicity
- Antigenic stability
- Survival

Environment

- Weather
- Housing
- Geography
- Occupational setting
- Air quality
- Food

Host

- G Age
- Sexe
- Behaviour
- Nutritional status
- Health status

Sign and symptoms

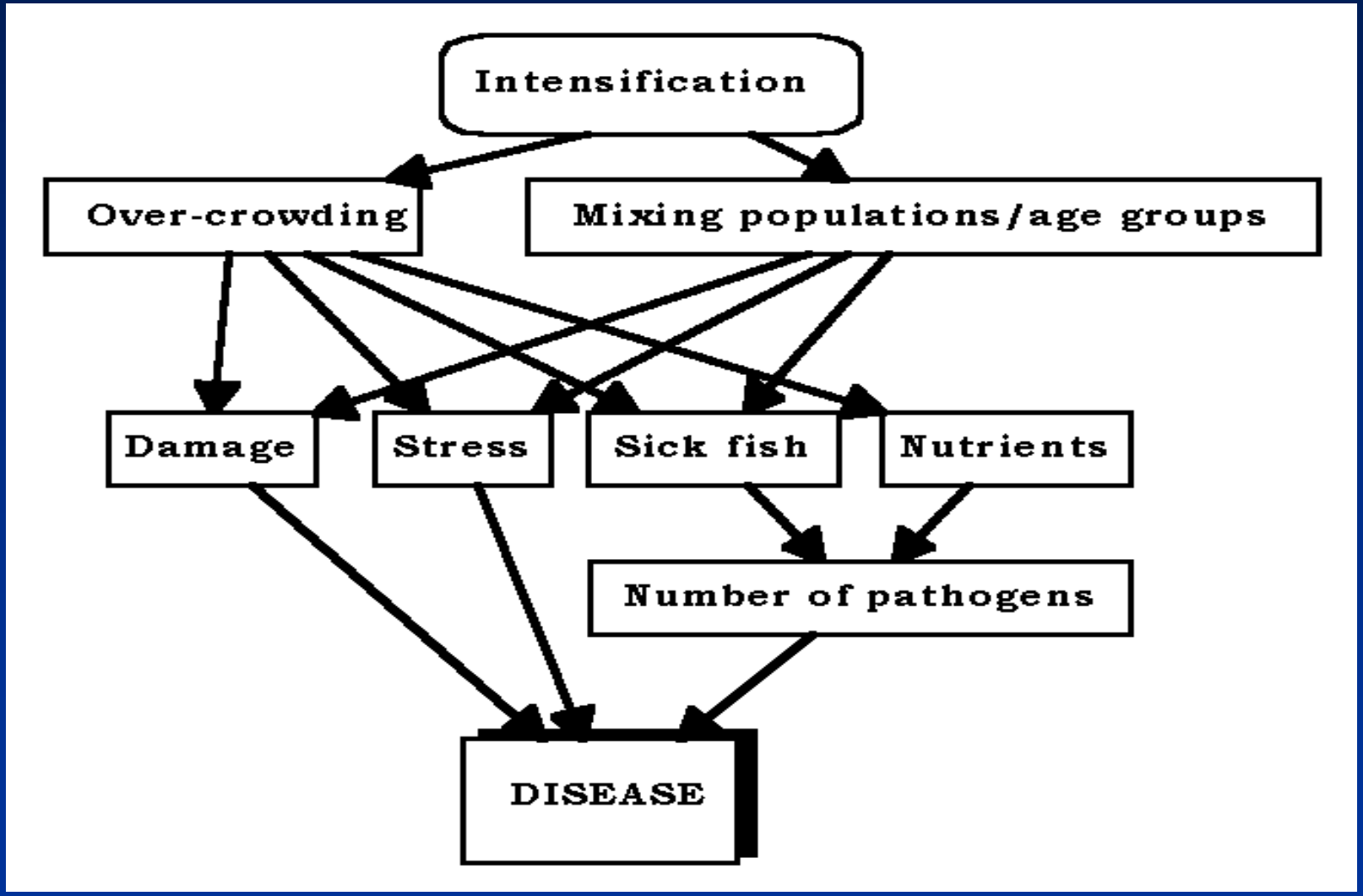
CLINICAL EFFECTS OF INFECTION ON THE BODY -
Acute

- Fever; anorexia, protein catabolism, acute-phase protein response, hypoalbuminaemia, low serum iron, sequestration of iron, anaemia, neutrophilia
- Inflammation; pain, dysfunction, tissue damage
- Convulsions; especially in children
- Confusion; especially in the elderly
- Shock.
- Haemorrhage; haemolytic anaemia, intravascular coagulation
- Organ failure.

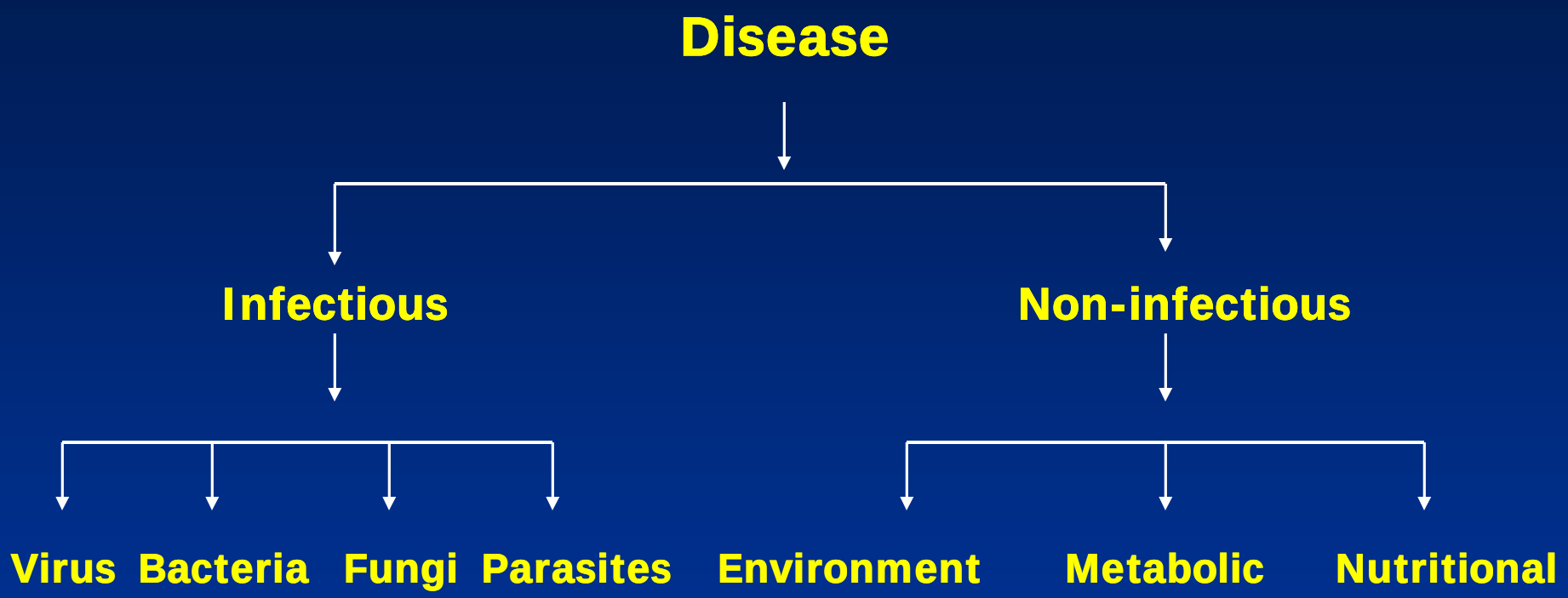
CLINICAL EFFECTS OF INFECTION ON THE BODY- **Chronic :**

- Weight loss and muscle-wasting
- Malnutrition; especially associated with diarrhoea
- Retardation of growth and intellect in children
- Anaemia; iron sequestration, maturation arrest in marrow, folate deficiency
- Tissue destruction; e.g. lung in pneumonia or tuberculosis, nerves in leprosy, liver in hepatitis B
- Post-infective syndromes; e.g. lactose intolerance, malabsorption, irritable colon, depression, post-viral fatigue syndrome

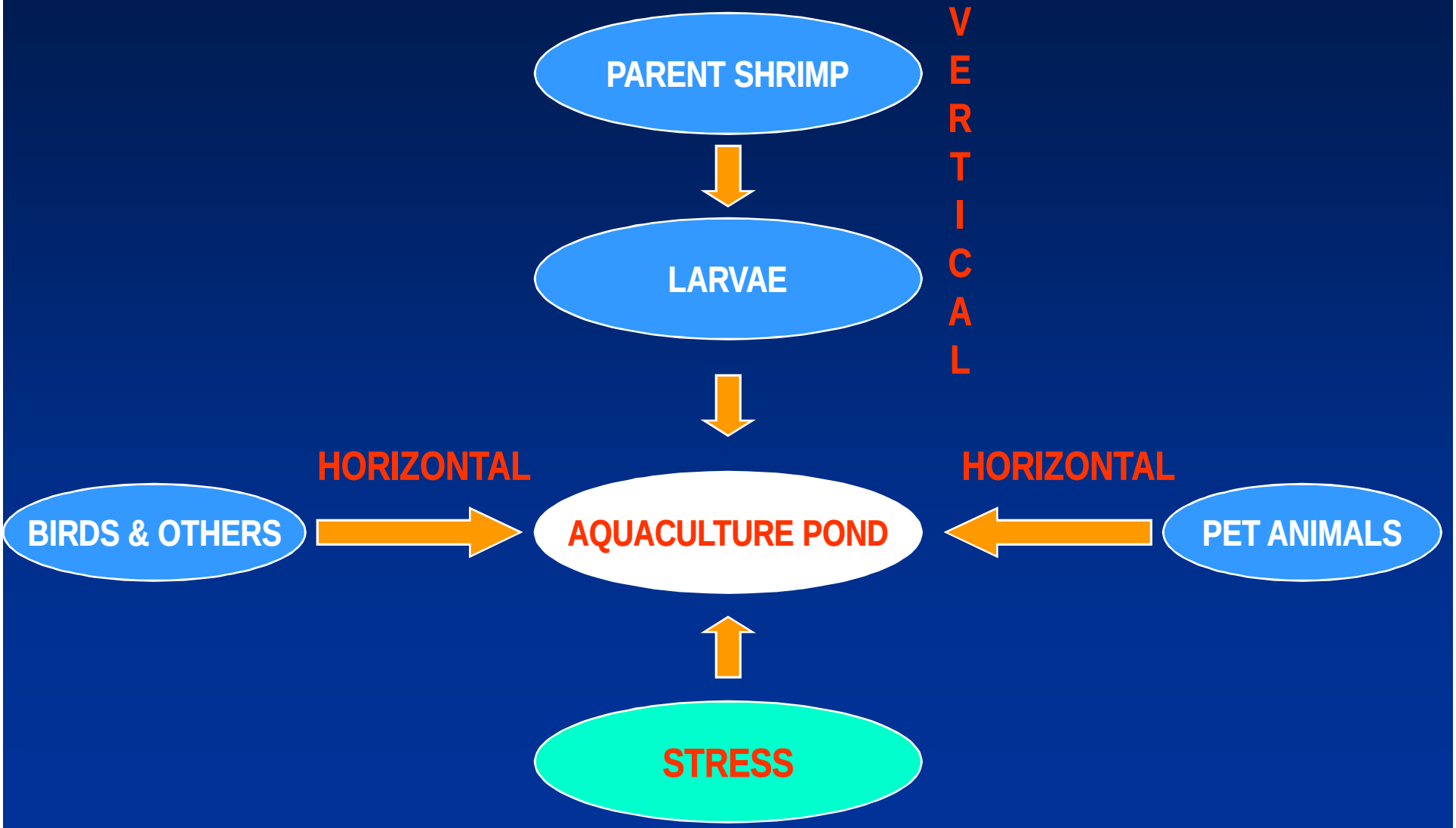
Pathogen


Intensification
Over-crowding
Mixing populations/age groups
Damage
Stress
Sick fish
Nutrients
Number of pathogens
DISEASE

Types of Diseases



Types of disease transmission



Means of Transmission of Infectious Diseases

Contact	Requires direct or indirect contact (fomite, blood, or body fluid)
Food or Water	Ingestion of contaminated food or water
Airborne	Inhalation of contaminated air
Vector-borne	Dependent on biology of vector as well as infectivity of organism
Perinatal	Similar to contact infection, however, the contact may occur in utero or during delivery.
Sexual	transmission by sexual intercourse.

Aquatic Animal Diseases (global trends & spread & emerging threats)

OIE Listed Aquatic Animal Diseases

FINFISH DISEASES
Epizootic haematopoietic necrosis
Epizootic ulcerative syndrome (red-spot disease)
Gyrodactylosis
Infectious haematopoietic necrosis
Infectious salmon anaemia
Koi herpesvirus disease
Red sea bream iridoviral disease
Spring viraemia of carp
Viral haemorrhagic septicaemia

MOLLUSCS DISEASES
Infection with Abalone herpes-like virus (newly adopted)
Infection with *Bonamia exitiosa*
Infection with *Bonamia ostreae*
Infection with *Marteilia refringens*
Infection with *Perkinsus marinus*
Infection with *Perkinsus olseni*
Infection with *Xenohaliotis californiensis*

CRUSTACEAN DISEASES
Crayfish plague
Infectious hypodermal and haematopoietic necrosis
Infectious myonecrosis (IMN)
Necrotising hepatopancreatitis (newly adopted)
Taura syndrome
White spot disease
White tail disease (MrNV)
Yellowhead disease

AMPHIBIAN DISEASES
Infection with *Batrachochytrium dendrobatidis*(newly adopted)
Infection with ranavirus(newly adopted)

Non-OIE Listed Aquatic Animal Diseases*

FINFISH DISEASES
Bacterial kidney disease (BKD)*
Channel catfish virus disease*
Grouper iridoviral disease
Oncorhynchus masou virus disease*
Viral encephalopathy and retinopathy*
Infectious pancreatic necrosis
Pancreas disease (PD)/Sleeping disease
Cardiomyopathy syndrome (CMS)
Heart and skeletal muscle inflammation (HSMI)
Enteric septicaemia of catfish*
Streptococcus ineae (Tilapia)
Amoebic gill disease
Kudoa thyrsites

MOLLUSCS DISEASES
Infection with *Bonamia roughleyi**
Infection with *Marteilla sydneyi**
Infection with *Haplosporidium costale**
Infection with *Haplosporidium nelsoni**
Infection with *Mikrocytos mackini**
Infection with *Marteilioides chungmuensis*

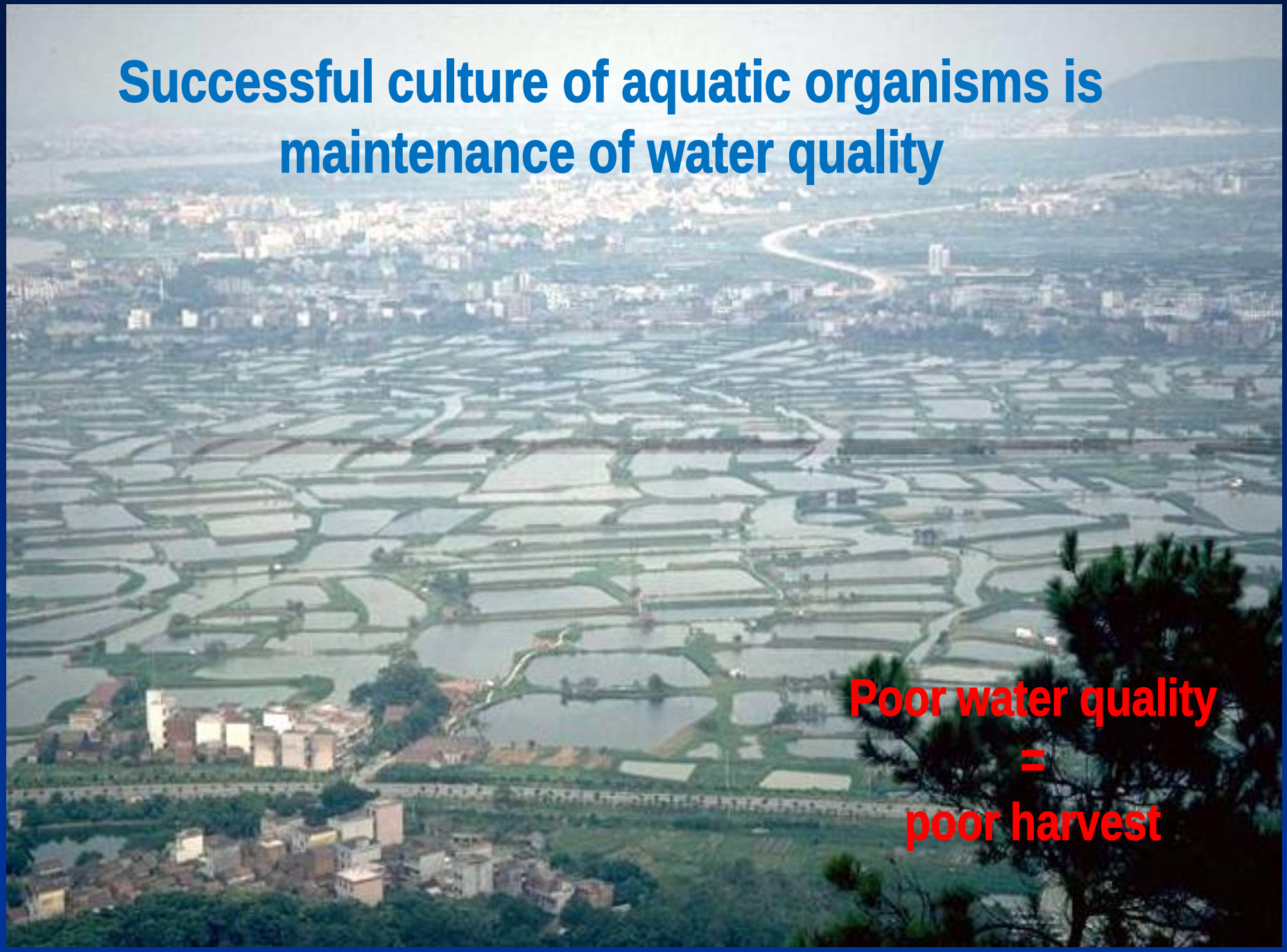
CRUSTACEAN DISEASES
Acute viral necrosis (in scallops)
Akoya oyster disease
Monodon slow growth syndrome
Milky haemolymph disease of spiny lobster (*Panulirus* spp.)
Spherical baculovirosis*
Tetrahedral baculovirosis*
Microsporidiosis (cotton shrimp disease)

*OIE delisted disease for which there is a designated OIE Reference Laboratory

STRESSORS…

Some of these can be easily and cost effectively controlled and others cannot at any cost

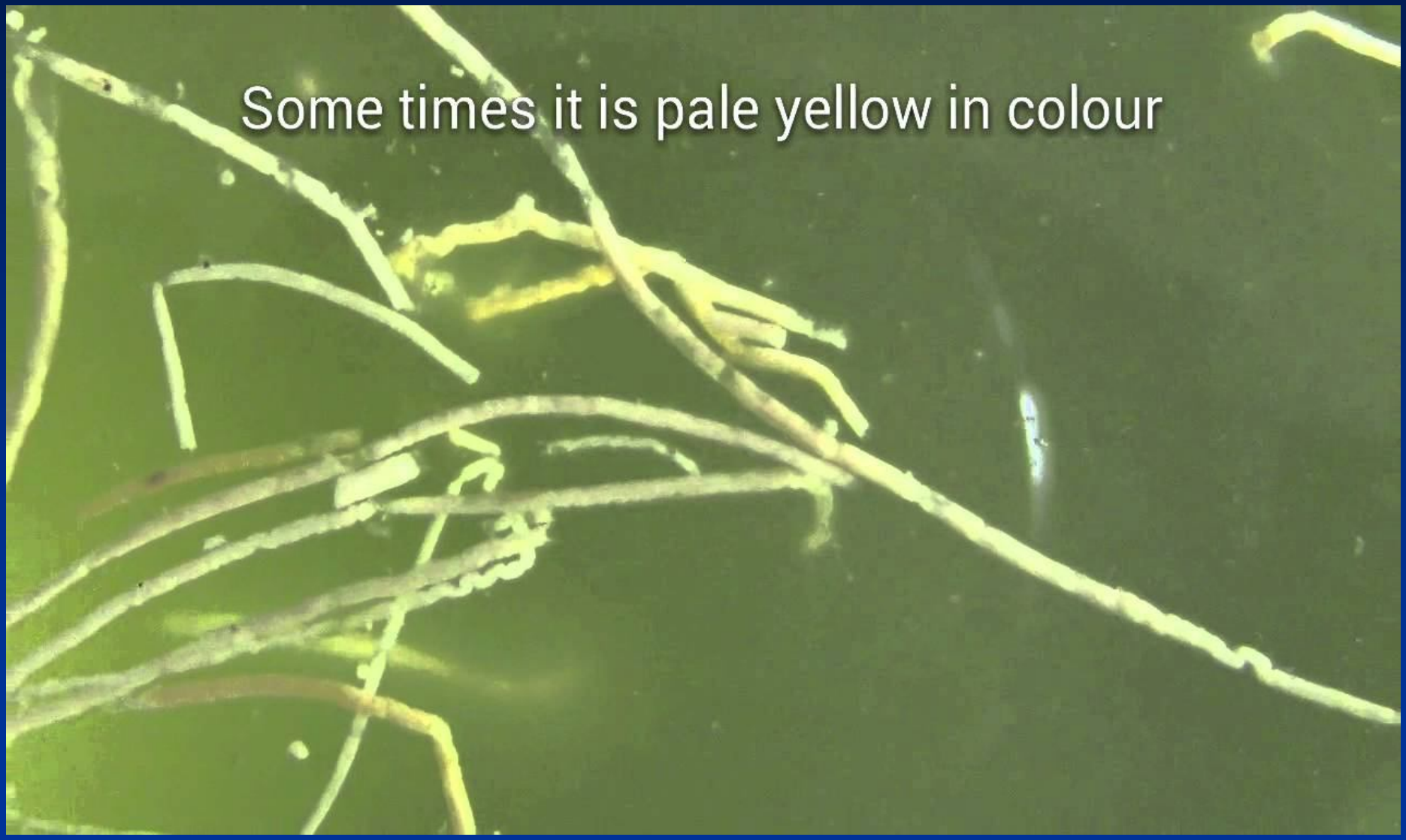
1. Rapid changes in temperature
2. Rapid changes in pH
3. High salinity fluctuations
4. Insufficient oxygen
5. Elevated CO2
6. High suspended solid loads
7. Unionized ammonia
8. Nitrites
9. Hydrogen sulfide
10. Density (crowding)
11. Pesticides
12. Heavy metals
13. Nutrition
14. Moulting
15. Handling
16. Conditions like continuous rains


Successful culture of aquatic organisms is maintenance of water quality
Poor water quality
=
poor harvest

White feces with shrimp

Figure 3 White feces with shrimp

White feces



WHITE SPOT SYNDROME

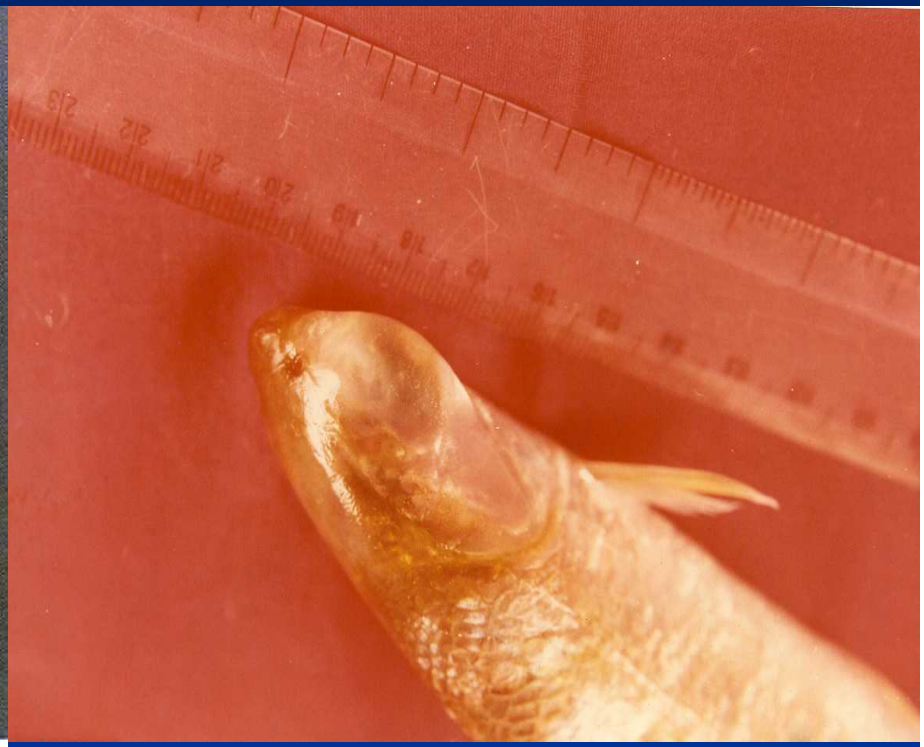
Infectious fin fish diseases…,

EUS

"A seasonal epizootic condition of freshwater and estuarine warm water fish of **complex infectious etiology** characterized by the presence of invasive ***Aphanomyces*** infection and necrotizing ulcerative lesions typically leading to **granulomatous response**"

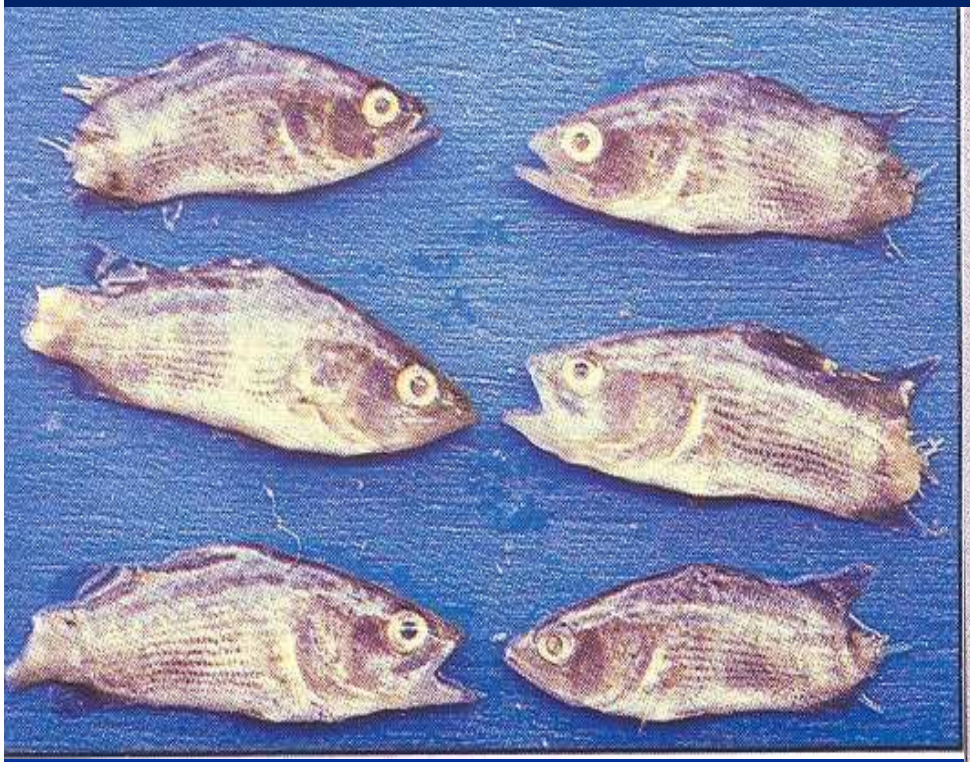
EXOPHTHALMIA OR “POPEYE”

Sign and symptoms: **In the initial stage, affected fish stopped feeding and became inactive and the descement’s membrane of the eye was ruptured, the gas bulged the cornea out, creating either an unilateral or “Popeye”**



FIN AND TAIL ROT DISEASE

Sign and symptoms: **The affected areas will turn black and look as if they were burnt and then the blackened part of the tail will be sloughed off**



Disease diagnosis – *Why?*

- **To Control and prevent the diseases**
- **Cardinal step - in health management programme**
  - **to recognize the occurrence of an abnormality**
  - **to identify the etiologic agent**
- **Failure of the accurate diagnosis …**
  - **to the large-scale mortality**
  - **to indiscriminate use of drugs and chemicals**
  - **causing environmental contamination**
  - **drug residual effects**
  - **drug resistant pathogens**

Stages in Epizootic (disease)

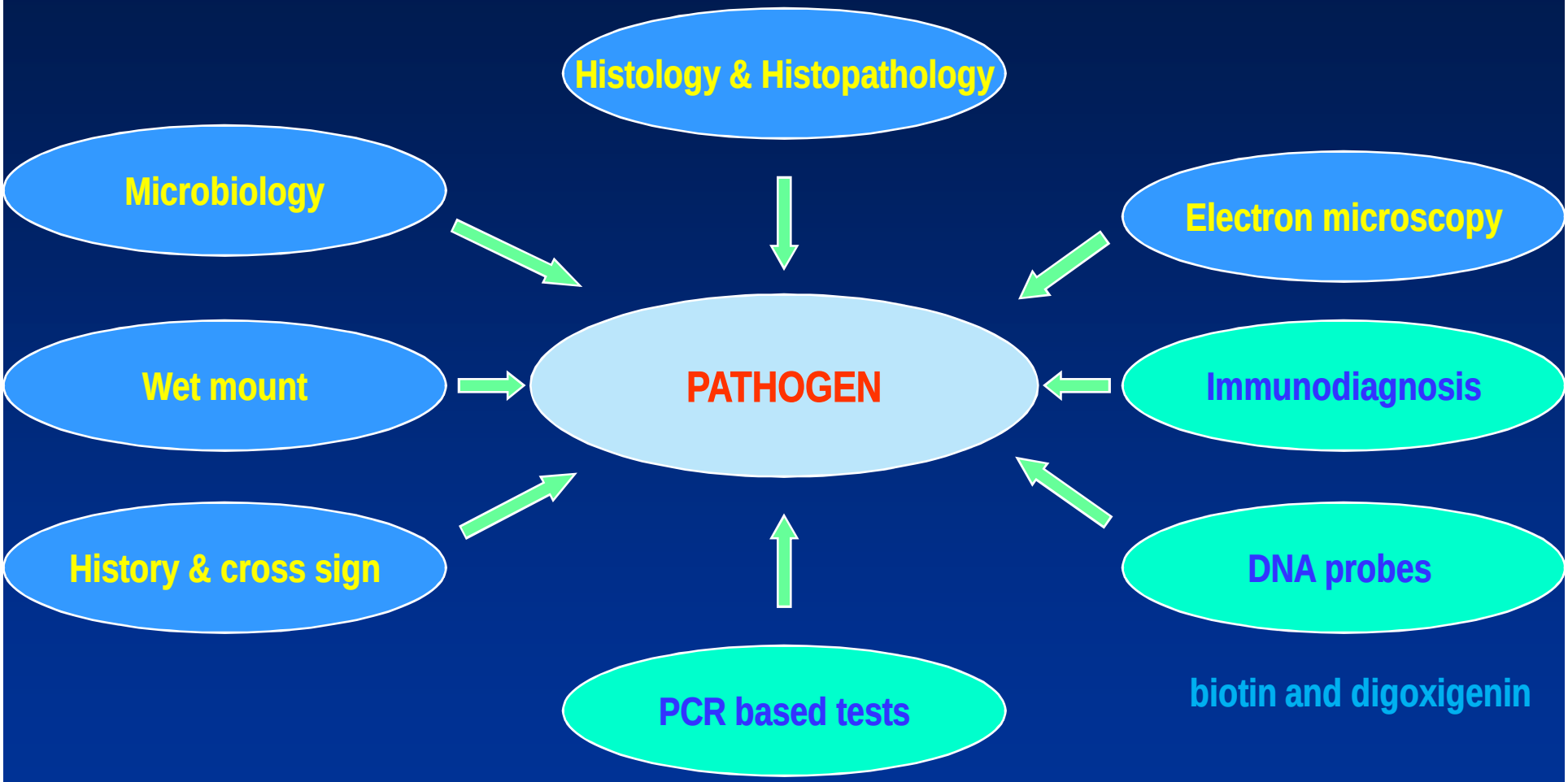
1. **incubatory**: agent has penetrated host barrier, found home and multiplying
2. **clinical or subclinical**: host adversely affected (manifestations)
   - depression (reduced activity)
   - color change
   - interrupted feeding behavior
   - body contortions
   - respiratory change
   - mortality

Potential for Disease via Infection: contributors

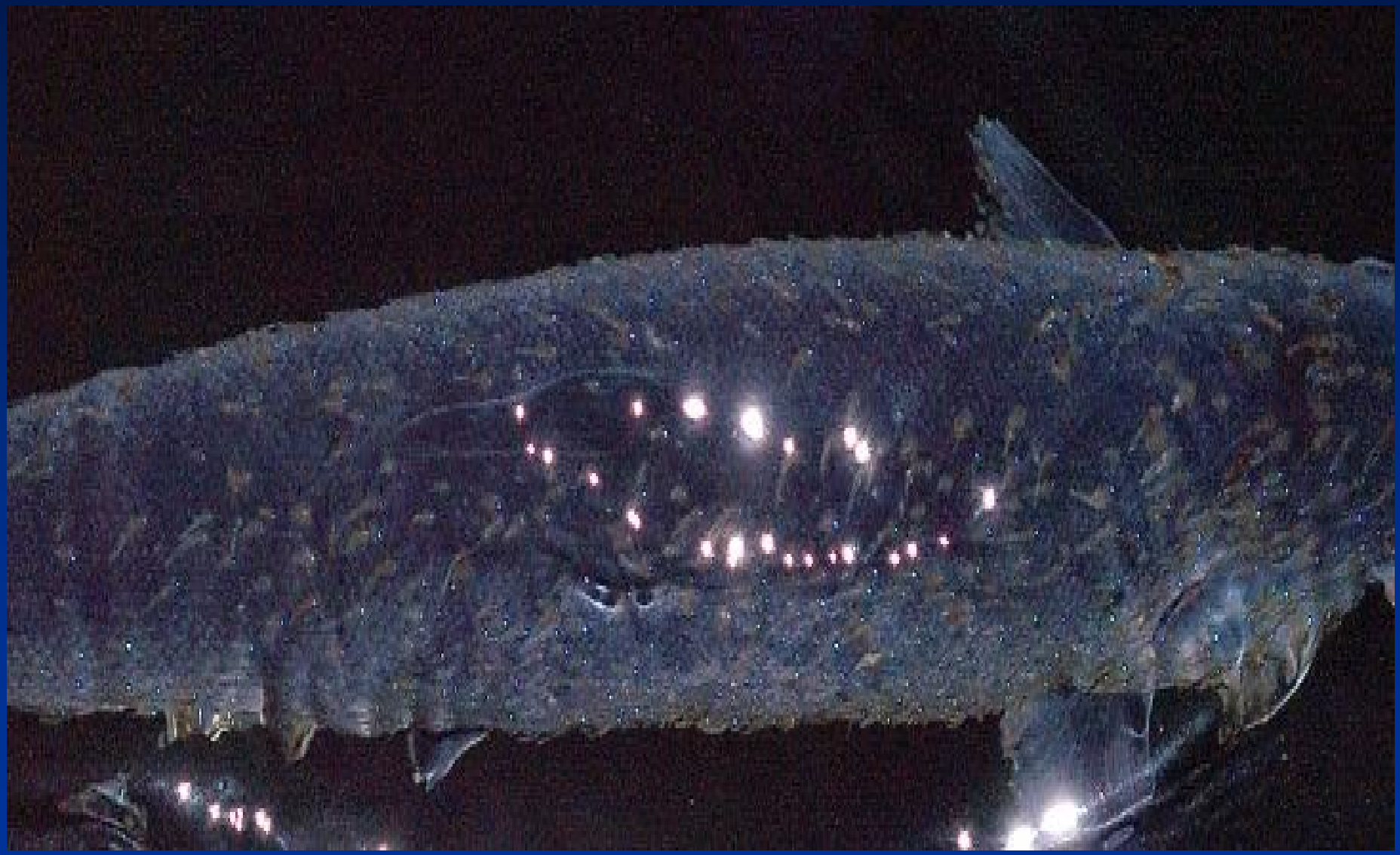
1. number of organisms (overwhelming)
2. infectivity (ability to get in)
3. virulence (ability to produce disease)
4. susceptibility of the host
5. agent's ability to overcome host's defenses
6. level of stress

- probablility of disease (Theobald Smith Model) = (# agents x virulence of agents)÷(resistance of host)

Methods…


Histology & Histopathology
Microbiology
Electron microscopy
Wet mount
PATHOGEN
Immunodiagnosis
History & cross sign
DNA probes
PCR based tests
biotin and digoxigenin

Specific diagnostic challenges



Problems with diagnosis!

- Identifying aquatic health is difficult
  - Cannot see the animals
    - Abnormal behaviour
    - Mortalities
  - Feeding often only time to observe
- Clinical signs / examination not very useful
  - Farmers may use CS to spot a problem
  - Cannot often use CS to diagnose a problem
    - Aquatic animals limited capacity to express CS
    - Same CS different disease or same disease different CS

Limitations in diagnosis

Test limitations

- False negatives (sensitivity)
- False positives (specificity)

- Best tests including PCR
  - 95% sensitive and specific
  - Get 5% false negatives and 5% false positives

Koch's Method (Postulates)

- 1) find the organism common to all infected animals, demonstrate its absence in healthy ones
- 2) isolate the organism in pure culture
- 3) reproduce the disease in suitable experimental animals
- 4) reisolate the same organism from experimentally infected animals

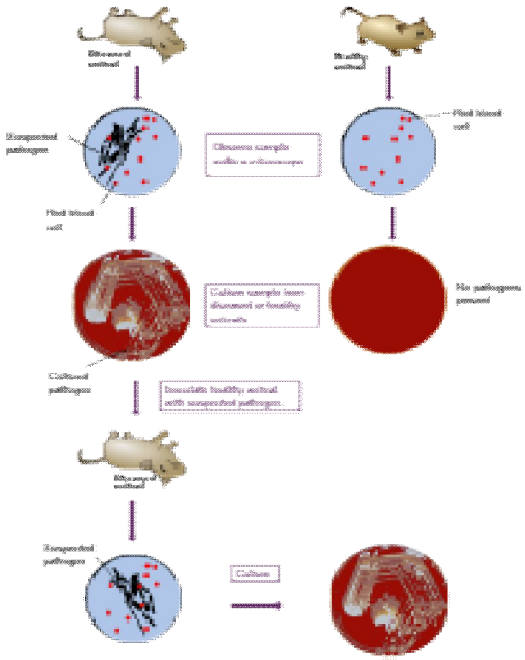
Koch's Postulates:

1. The microorganism must be found in abundance in all organisms suffering from the disease, but should not be found in healthy organisms.

2. The microorganism must be isolated from a diseased organism and grown in pure culture.

3. The cultured microorganism should cause disease when introduced into a healthy organism.

4. The microorganism must be reisolated from the inoculated, diseased experimental host and identified as being identical to the original specific causative agent.



Disease Prevention…

"prevention is better than cure"

1. Location of the farms
   * ***Good quality water, Free from IAD pollution***
2. Pond preparation
   * ***Thoroughly drained, sun dried, Black soil, liming***
3. Stocking
   * ***Healthy PL, density***
4. Feeding
   * ***Balanced diet, quantity,***
   * ***Avoid accumulation, old, rancid, mouldy feeds***
5. Water quality management
   * ***Physico-Chemical parameters***

Disease Control…

1. Quarantine measures
   * *Seed and brood stock*
   * *Pathogenic microbes*
2. Use of genetically resistant stock
   * *Species specific (Yellow head disease)*
3. Use of prophylactic vaccines
   * *Commercial vaccine*
4. Use of drugs & chemicals
   * *Malachite green, $KMno_4$, Liming, Bleaching powder*
   * *Probiotics (Bioremediation), Immunostimulants*

Bioremediation

“Microbial transformation of harmful materials into harmless molecules”

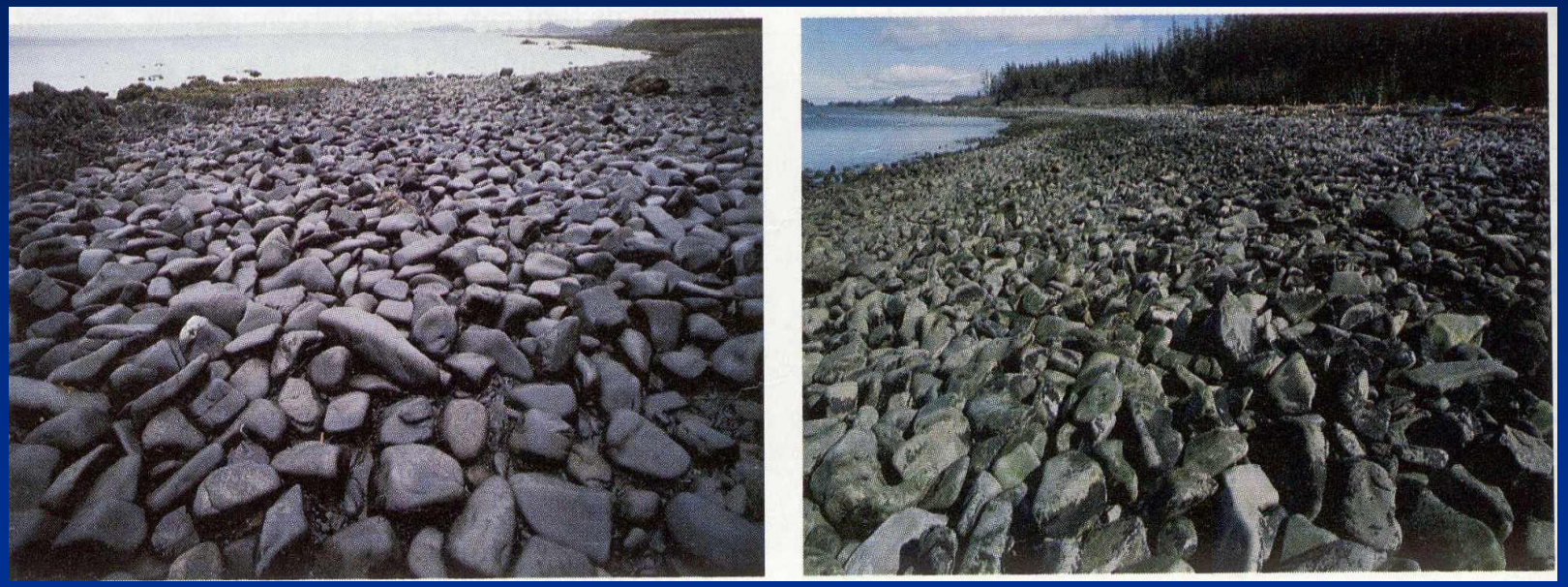
- **To help our pollution problems by digest organic and inorganic pollutants**

Toxic organic compounds provide energy and carbon
Inorganic substances either transform into insoluble or volatile compounds or accumulate them intracellularly.

Bioaugmentation…?

"The introduction of microbes into a contaminated environment for the purpose of detoxification is termed bioaugmentation"

(a)



(b)

Petroleum spill (a) before and (b) after bioremediation cleanup
Using a petroleum-digesting strain of *Pseudomonas*

Bioremediation in Aquaculture

- **To improve the water quality by application of probiotics and enzymes to the pond.**

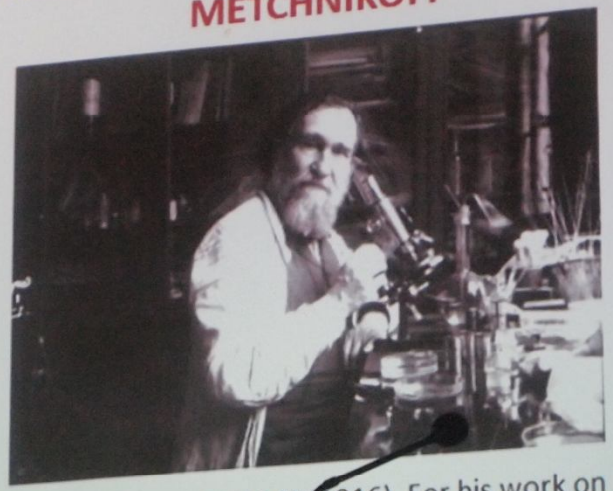
- **This type of biotechnology is known as bioremediation, which involves manipulation of microorganizms in ponds to enhance mineralization of organic matter to get rid of undesirable waste compounds and to eliminate the pathogens.**

PROBIOTICS…?

The intestine harbors trillion of commensal bacteria that participate in digestive functions and help to protect the host from the aggression of several entero-pathogens

The beneficial effects of the micro-biota on the host immune system have allowed the use of PROBIOTICS in improving health and protection against infectious agents

The first microbe used specifically for this purpose was *Lactobacillus bulgaricus,* discovered in 1908 by **METCHNIKOFF**



Elie Metchnikoff (1845-1916). For his work on immunity, Metchnikoff shared with Paul Ehrlich the 1908

Definition of Probiotics?

- **A bacterial supplement of a single or mixed culture of selected non-pathogenic bacterial strains was termed probiotics**
- **Probiotics' - coined by Parker (1974) – 'for life'**
- **Includes bacteria, cyanobacteria, fungi *etc.*,**
- **Synonymous terms used for ...**
  - **Normal micro biota**
  - **Effective micro biota**
  - **Probiont**
  - **Beneficial bacteria**
  - **Friendly bacteria**

Types of probiotics…

- **Non-viable probiotics** : ***These are dead***
- **Freeze-dried probiotics** : ***These will die rapidly upon leaving refrigeration***
- **Fermentation probiotics** : ***These are produced through fermentation***
- **Viable probiotics** : ***These are live with guaranteed shelf life, guaranteed number of organisms, have a protocol for counting and to be very stable and efficacious***

Types of probiotics…

Soil probiotics:

- **Mixture of live bacterial cultures used to break the heavy organic load**
- **Soil discolours due to anaerobic bacteria**
- **Hydrogen sulphide production (obnoxious odour) due to Sulphur bacteria**
- **Microaerophyllic microbes break down organic load and remove hydrogen sulphide**

Types of probiotics…

Water probiotics: **These are live microbial preparations**

- **Which utilize nitrogen-bearing wastes in their metabolic process**
- **Suppress the growth of harmful bacteria such as *Vibrio* and inhibits their growth by producing certain vibriocidal substances**
- **High enzymatic activities removes the organic load**

Types of probiotics…

Feed probiotic: **Which consists mainly Lactic acid Bacillus or *Lactobacillus sporogens***

- **Which are predominant members of the normal intestinal flora**
- **Adverse conditions like stress, diseases and antibiotic therapy disturbs the balance of the intestinal flora leading to detrimental effects on the system**

- **Produce biological lactic acid digestive enzymes such as amylases, proteases and lipases for better digestion**
- **Inhibits growth of pathogenic organisms**
- **Maintains normalcy in the gastrointestinal tract**

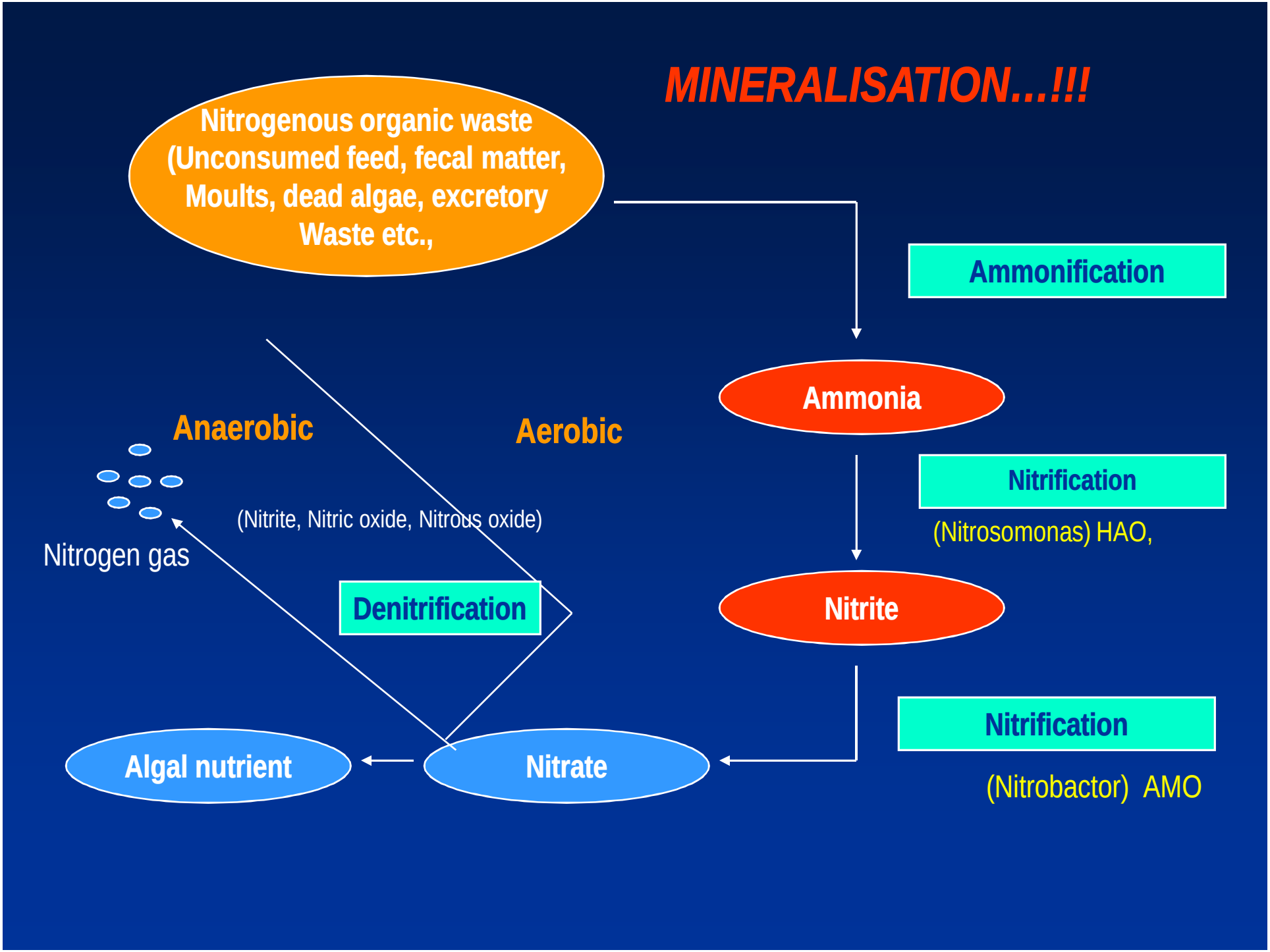
HOW DO PROBIOTICS WORK???

- About one third of immune system is localized in the gastrointestinal tract
- One of the major mechanisms of probiotic action is through the regulation of host immune response
- Influence the innate and adaptive immunity by modifying the composition and activity of the gut

•PROBITICS increase IgA counts and other immunoglobulins-secreting cells In the intestinal mucosa and stimulates the local release of interferon

Probiotics and their role

- ***Bacillus*** : **Mineralization and breakage of proteins**
- ***Nitrosomonas*** : **Oxidation of ammonia**
- ***Nitrobacter*** : **Oxidation of nitrites**
- ***Aerobacter*** : **Reduction of organic matter**
- ***Cellulomonas*** : **Breakage of plant material**


MINERALISATION...!!!
Nitrogenous organic waste (Unconsumed feed, fecal matter, Moults, dead algae, excretory Waste etc.,
Ammonification
Ammonia
Anaerobic
Aerobic
Nitrification
(Nitrosomonas) HAO,
(Nitrite, Nitric oxide, Nitrous oxide)
Nitrogen gas
Denitrification
Nitrite
Nitrification
(Nitrobactor) AMO
Algal nutrient
Nitrate

Chemical reaction…

An obligate aerobic & oxidizing process

NITRIFICATION

$NH_3 \rightarrow No_2 \rightarrow No_3$

An anaerobic & Reducing process

DE NITRIFICATION

N_2
N_2O ← NH_3 ← No_2 ← No_3
NO

Probiotics activities:

- Probiotic bacteria competitively exclude the pathogenic bacteria or produce substances that inhibit the growth of the pathogenic bacteria
- Provide essential nutrients to enhance the nutrition of the cultured animals through nitrogen cycle
- Provide digestive enzymes to enhance the digestion of the cultured animals
- Probiotic bacteria directly uptake or decompose the organic matter or toxic material in the water and hence improving the quality of the water

Beneficial effects of probiotics may be mediated by...

- Neutralization of toxin
- Suppression of viable count
- Production of antibacterial compounds
- Competition for adhesion sites
- Alternation of microbial metabolism
- Stimulation of immunity of the host
- Accelerate the sediment decomposition

Other application of probiotics...

- Minimizes frequent water exchange
- Reduces BOD and COD
- Regulates pH and stabilizes bloom
- Improve survival and growth rate
- No residue, No pollution, No odour, No side effect
- Reduces operational & maintenance costs
- Secures better yields and larger profits

Infectious Diseases

Barriers for Defense Against Infection:

1. Skin:
   - Prevents entry of infectious organisms, unless injured.
   - Severe burn patients who die are usually killed by infections. So much skin is damaged they are very vulnerable to infections.
2. Mucus membrane:
   - Mucous is usually rich in enzymes that will kill many pathogens
3. Cilia:
   - These are hair-like structures lining the respiratory tract. They work to sweep foreign particles out of the respiratory tract.
   - Damaged by smoking, leaving smokers more vulnerable to infections.
4. Coughing:
   - Helps remove foreign material from respiratory tract.
5. Personal Hygiene
   - Helps reduce the number of pathogenic organisms on the skin and other surfaces of the body.

SUCCESSFUL FARMING

- Plan ahead.
- Good health records for each pond.
- Good observations.
- Good feed.
- Water quality/quantity.
- Stay on top of things!!


UNITY IS OUR STRENGTH

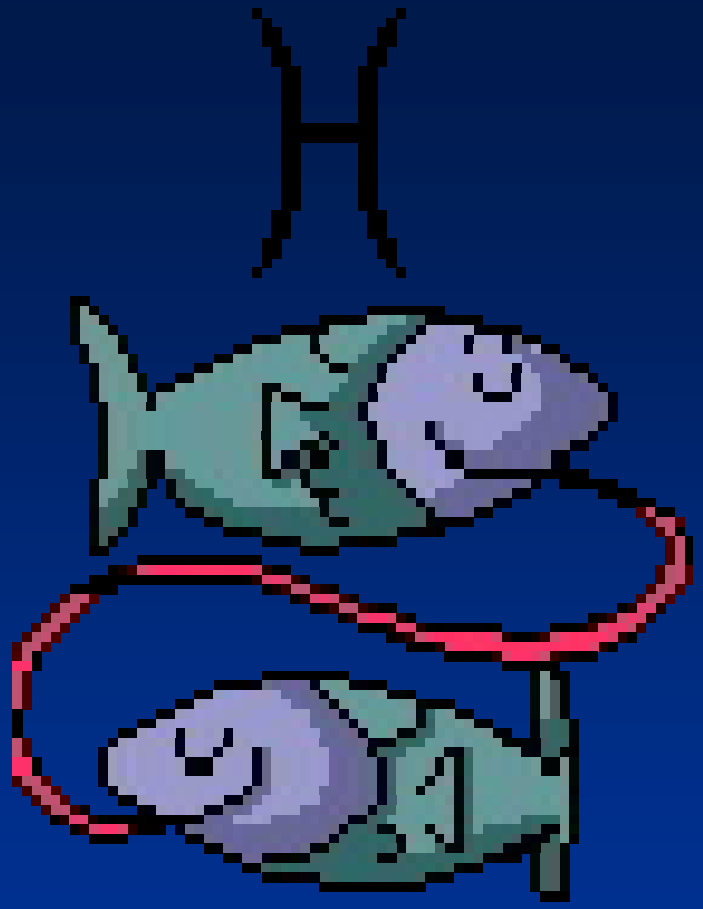
always right, your friend and a sensitive partner.....”

“If you take care of your microbial friends they will take care of your future”

Dr. D. Perlman

ACKNOWLEDGEMENTS

ANNAMALAI UNIVERSITY



Thank you...