Introduction to Achaea

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ARCHAEA…………………

Archaea is divided into 3 major phyla: Euryarchaeota, Crenarchaeota & Thaumarchaeota . Newly recognised group Nanoarchaeota & Korarchaeota

Archaea

Phyla

Euryarchaeota	Nanoarchaeota	Korarchaeota	Crenarchaeota	Thumarchaeota
Halobaterium	Nanoarchaeum	Korarchaeum	Thermoproteus	Nitrososphaera
Halococcus			Pyrodictium	Nitrosopumilus
Natronococcus			Sulfolobus	Cenarchaeum
Methanobacterium			Desulfurococcus	
Methanocladococcus				
Methanosarcina				
Pyrococcus				
Thermoplsma				
Picrophilus				

Unique features

- Chemoorganotrophic or Chemolithtrophic metabolism
- H_2 as common e^- donor; NH_3 oxidation
- S^o as terminal e^- acceptor
- Methanogenesis

Crenarchaeota:

Members are found in Solfataras (hot; sulphur rich environments)

- Hyperthermophilic orders- Thermoproteales, Sulfolobales & Desulfurococcales
- At present ~ 25 genera are included here. *Sulfolobus & Thermoproteus* are most studied.
- Members of this group can survive well at 100°C and most extreme is a member of Pyrodictiaceae family. Optimum growth temp. is 105°C & autoclaving at 121°C for several hours fails to kill it! Found in a Hydrothermal vent of Pacific ocean.
- Some members are called Thermoacidophiles. Optimum growth conditions are 80°C and pH 2-3.
- Produce H2SO4 from biological oxidation of H_2S & S^o.
- Obligate anaerobes except a few members.
- *Sulfolobus*: Temperature up to 90˚C & pH 1-5; mostly spherical cells with lobes and adhere to sulphur crystals. *Sulfolobus* can grow aerobically with S^o as e^- donor and O_2 as e- acceptor & in anaerobic condition it uses other componds like NO_3^- as e^- acceptor!
- *Thermproteus*: Rod shaped cells; inhabits mostly neutral or slightly acidic hot springs. They are strickt anaerobes with S^o based respiration. Chemolitho or organotrophic growth on either H_2 or formate, small peptdes, ethanol, malate *etc.*

Euryarchaeota:

- Composed of 10 orders- Thermococcales, Methanopyrales, Methanobacteriales, Thermoplasmatales, Archaeoglobales, Methanosarcinales, Methanomicrobiales & Halobacteriales.
- They are broadly classified into

✓ Methanogens- Use H_2 & CO_2 or small organics to produce CH_4. It is the largest group of studied archaea. Eg. *Methanococcus* sp. , *Methanospirillum* sp.

✓ Halobacteria- Extreme halophiles; require at least 1.5 M NaCl for survival & optimum being 4 M NaCl. Presence of light harvesting pigment Bacteriorhodopsin at cell membrane: Purple Membrane. Eg. *Halobacterium salinarium*.

✓ Methanogens are most strict anaerobes while extreme halophiles are obligate aerobes!

EXTREME HALOPHILICS: *Haloarchaea*

❖ Key members- *Halobacterium, Halococcus* & *Natronobacterium*

❖ Inhabits the highly salty environments like salt lakes; heavily salted foods like several fish & meat products.

❖ 1.5M [NaCl] (~9%) or more for optimum growth. Mostly they require 2-4M NaCl: also can grow at 32% NaCl (saturation level).

❖ *Natronobacterium, Haloferax* generally grow at slightly low salinity. Natonobacterium, Natonomonas are highly alkalophilic nature: growth in “Soda” lakes (pH 9-11).

- Haloarchaea are generally Gram –ve & non spore formers.
- Cells may be rod (*Halobacterium*)or coccus (*Halococcus*) shaped.
- Genomes contain large plasmid having high G+C content (>60%) with ~30% of the total DNA content.
- Use amino acids & organic acids as e^- donors and require vitamins as growth factors.
- The requiremet of Na^+ cannot be satisfied by any other ion, even K^+.
- To maintain and withstand this high [salt], 'Compatible Solute' concept is key for survival. Cytoplasm of the cells synthesize similar amount of salt so that osmotic balance can be maintained.
- Uptake of large amounts of K+ inside the cell is essential to maintain the osmo-balance.
- Presence of extremely high quantities of glu & asp in glycoproteins of cell surface of Halobacterium signifies the need of Na^+ since the loss of bound Na^+ leads to decrease in integrity of cell surface structure as –vely charged proteins repel each other.
- All the cytosolic proteins are acidic and kept functionally active by the presence of K^+.
- Proteins are generally almost devoid of hydrophobic amino acids and also lysine.
- Ribosomes need high [KCl] for integrity and stability.
- Bacterioruberins (C_{50} carotenoid) can be used as light harvesting pigment in some members.
- Bacteriorhodopsin-
  - Light harvesting integral membrane protein in *Halobacterium salinarium* : structural & functional similarity to rhodopsin.
  - Presence of retinal enables bacteriorhodopsin to pump H+ across the membrane and also give a purple hue: Purple membrane.
  - Synthesis of bactriorhodopsin is induced in low Oxygen environment.

Methanogens

- Habitat: Anoxic sediments- marsh lands, swamp, paddy fields; rumen of ruminant animals, caecum of caecal animals, large intestine of monogastric animals such as human, dog; geothermal vents; as endosymbionts of various protozoa

- Morphology: Cell wall is made up of pseudomurein, methanochondroitin & glycoprotein. Presence of S-layer in some of the members.

- Physiology: They are obligate anaerobes, most of them are mesophilic and non-halophilic with several exceptions.

- Biochemistry: Some unique substrates (unlike glucose which is common) are generally converted into CH_4. 3 types of substrates are mainly acted upon by methanogens

a. CO_2 type- CO_2 using H_2 as e^- donor; formate; CO. *Methanobacterium sp.*
b. Methylated type- Methanol, methylamines, methylmercaptan (CH_3SH), dimethylsulphide. *Methanosarcina sp.*
c. Acetotrophic type- Acetate, pyruvate. *Methanococcus sp.*

Methanogens

Methanobacteriales

Methanobacterium
Methanobrevibacter
Methanosphaera

Methanomicrobiales

Methanomicrobium
Methanogenium
Methanoplanus

Methanopyrales

Methanopyrus

Methanococcales

Methanococcus
Methanocladococcus

Methanosarcinales

Methanosarcina
Methanolobus
Methanohalophilus
Methanomicrococcus

Methanocladococcus jannaschii-

- Hyperthermophilic methanogen
- 1.66 Mbp circular genome with ~ 1700 genes
- Biochemical pathways similar to those of bacteria
- Molecular processes like transcription, translation resemble those of eukaryotes

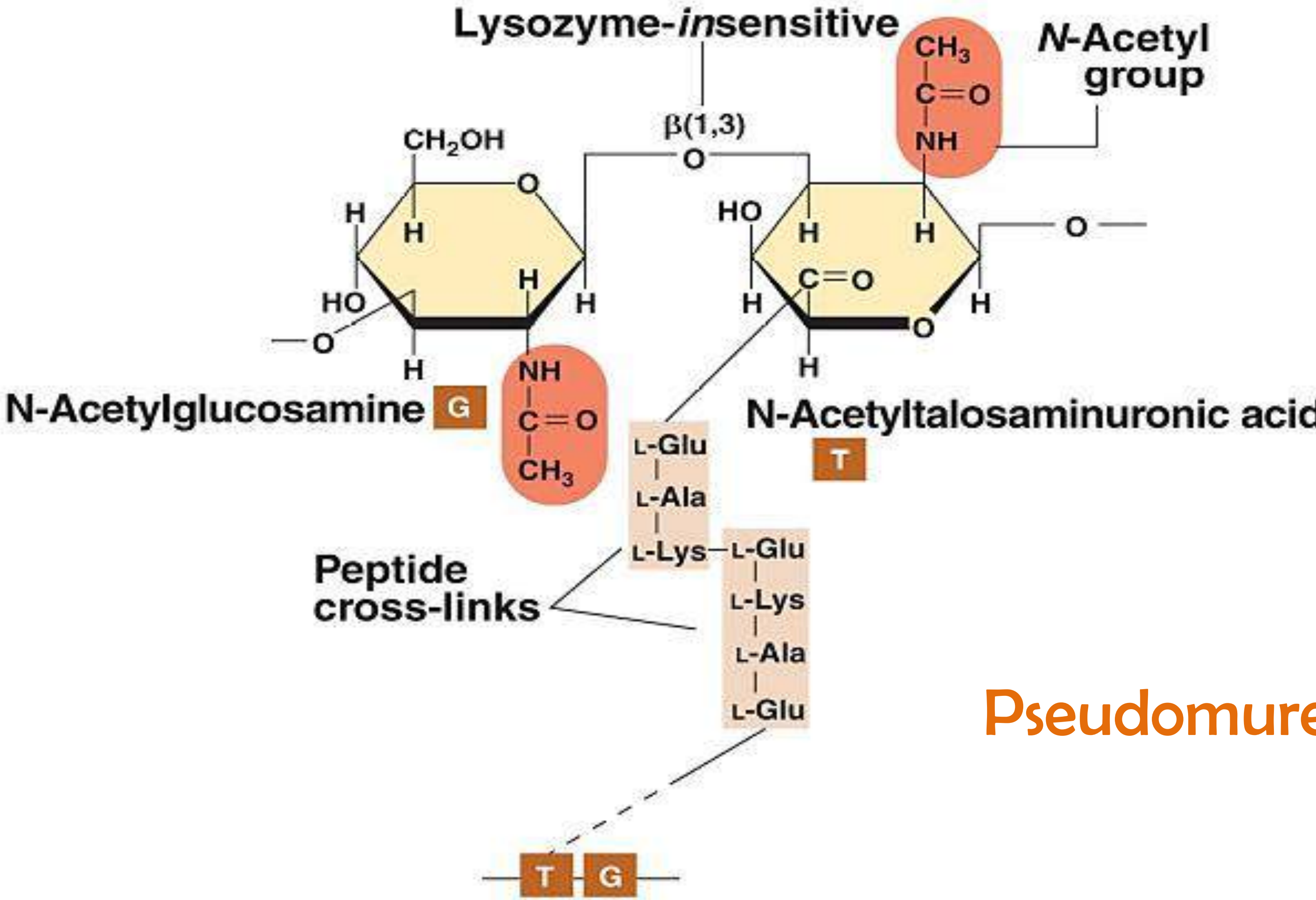
Pseudomurein

- The cell walls of certain methanogenic Archaea contain a molecule that is remarkably similar to peptidoglycan, a polysaccharide called pseudomurein (the term “murein” is from the Latin word for “wall” and was an old term for peptidoglycan

- The backbone of pseudomurein is composed of alternating repeats of N-acetylglucosamine and N-acetyltalosaminuronic acid; the latter replaces the Nacetylmuramic acid of peptidoglycan.

- Pseudomurein also differs from peptidoglycan in that the glycosidic bonds between the sugar derivatives are β-1,3 instead of β-1,4, and the amino acids are all of the L stereoisomer.

- It is thought that peptidoglycan and pseudomurein either arose by convergent evolution after Bacteria and Archaea had diverged or, more likely, by evolution from a common polysaccharide present in the cell walls of the common ancestor of the domains Bacteria and Archaea.



Pseudomurein

Other Polysaccharide Walls

Instead of Pseudomurein : other polysaccharides.

❖ *Methanosarcina sp*.

thick polysaccharide walls composed of polymers of glucose, glucuronic acid, galactosamine uronic acid, and acetate.

❖ *Halococcus sp.*

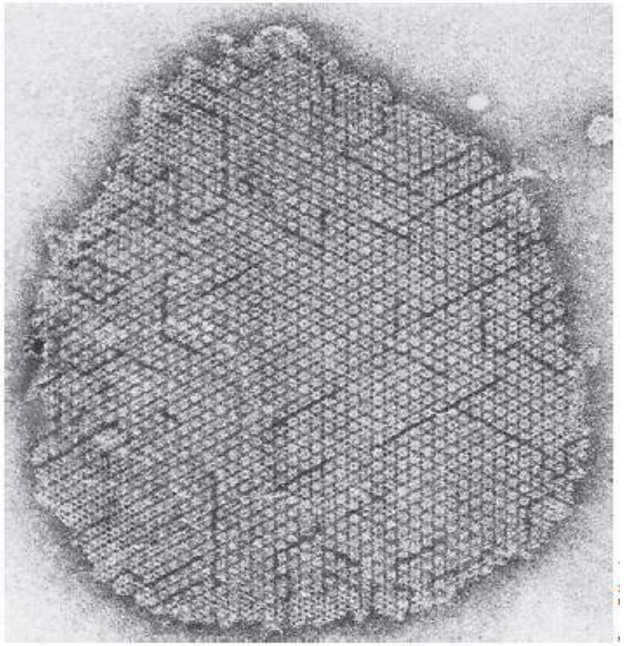
similar cell walls that also contain sulfate ($SO4^{2-}$). The negative charge on the sulfates bind the high concentration of Na^+ present in the habitats of *Halococcus sp.*

Methanobacterium sp.

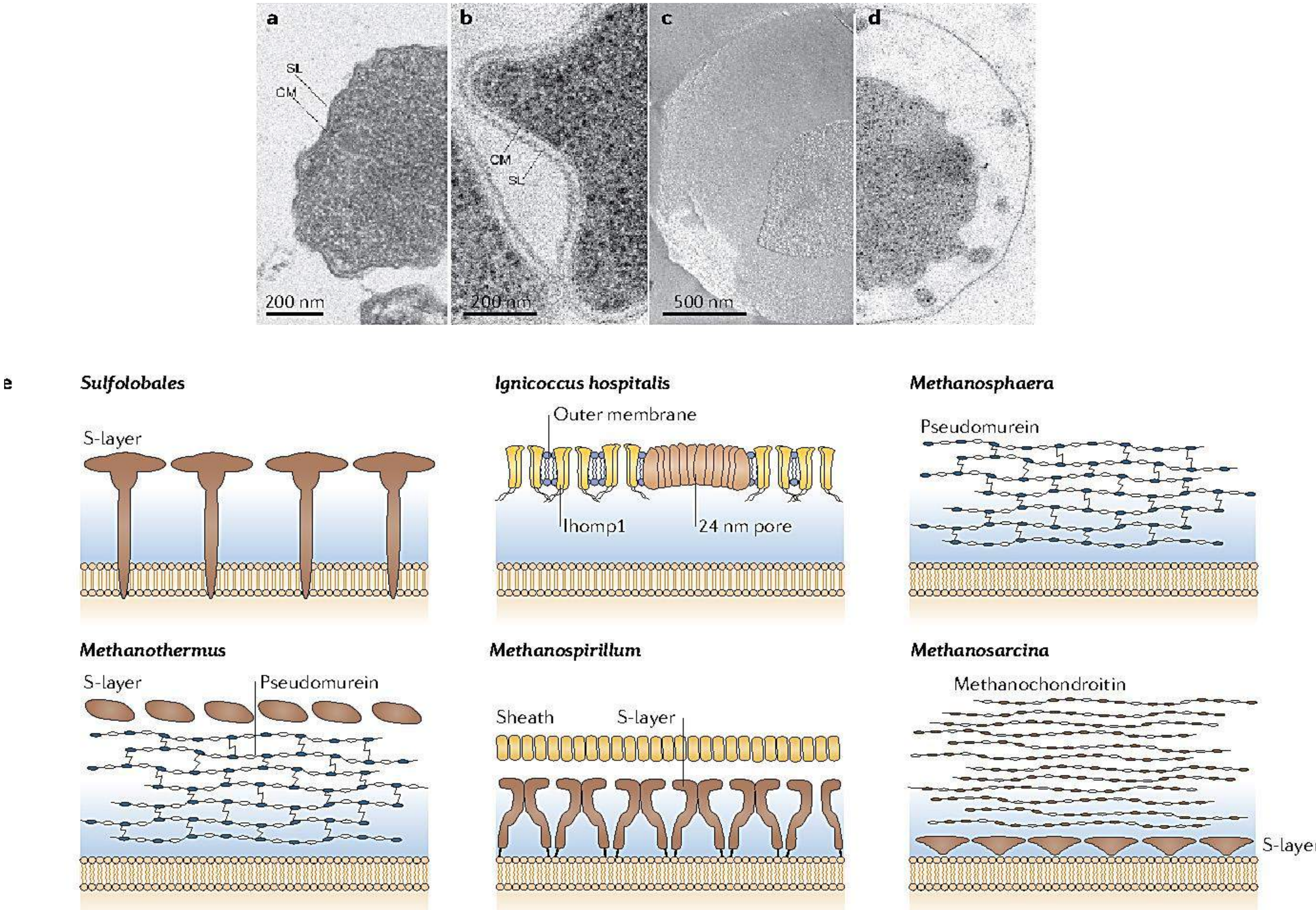
Pseudomurein walls composed of methanochondroitin (structural resemblance to chondroitin, the connective tissue polymer of vertebrate animals)

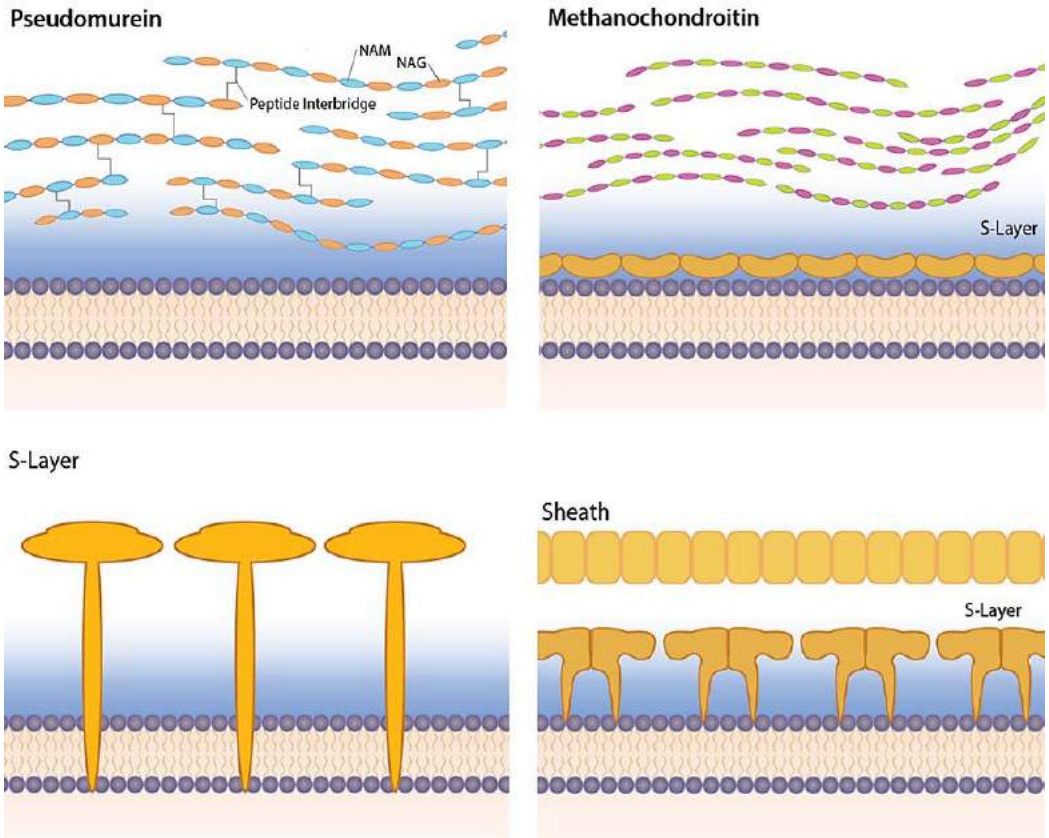
S-Layers

- Commonest cell wall in species of Archaea is the Paracrystalline Surface Layer, or **S-layer**.
- Consisting of interlocking protein or glycoprotein molecules that show an ordered appearance when viewed with the electron microscope
- The paracrystalline structure of S-layers is arranged to yield various symmetries, such as hexagonal, tetragonal, or trimeric, depending upon the number and structure of the protein or glycoprotein subunits of which they are composed.
- Methanogen *Methanocaldococcus jannaschii,* consist only of an S-layer.
- S-layers are sufficiently strong to withstand osmotic bursting. However, when an S-layer is present along with other wall components, the S-layer is always the outermost wall layer, the layer that is in direct contact with the environment.
- Since the interface between the cell and its environment, it is likely that the S-layer functions as a selective sieve, allowing the passage of low-molecular-weight solutes while excluding large molecules and structures (such as viruses).

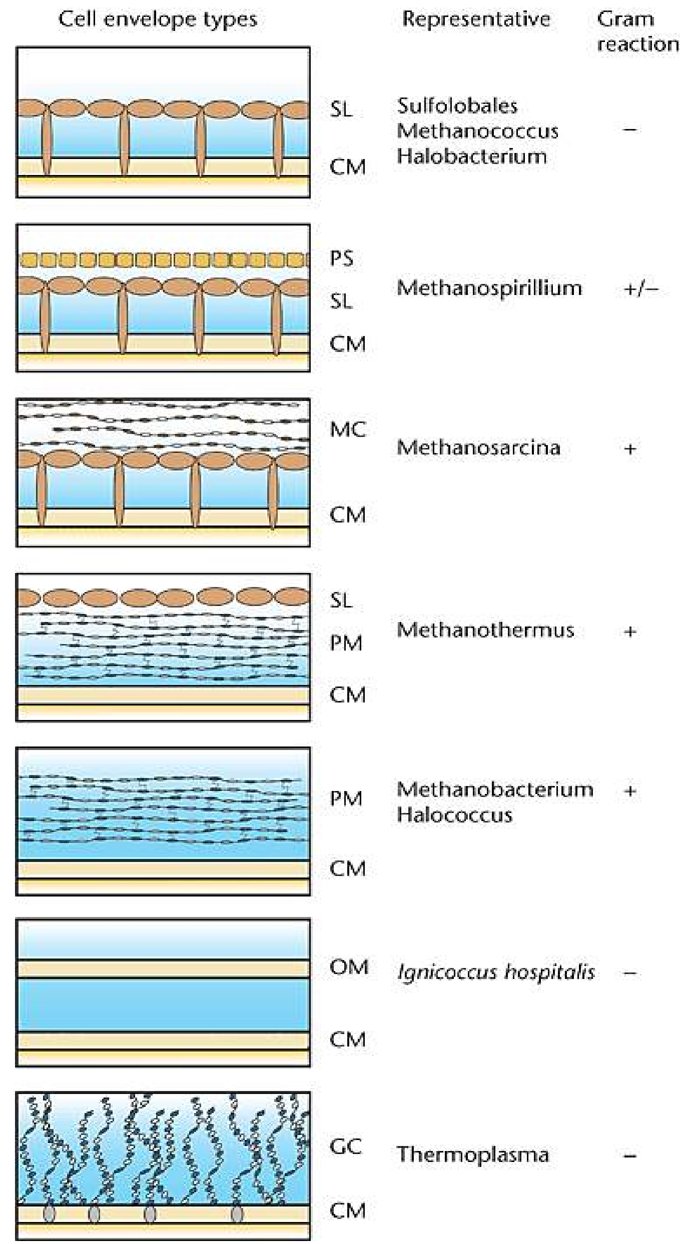


SEM of S- Layer


a
SL
CM
200 nm
b
CM
SL
200 nm
c
500 nm
d
e
Sulfolobales
S-layer
Ignicoccus hospitalis
Outer membrane
Ihomp1
24 nm pore
Methanosphaera
Pseudomurein
Methanothermus
S-layer
Pseudomurein
Methanospirillum
Sheath
S-layer
Methanosarcina
Methanochondroitin
S-layer



Cell Wall Structures



Thermophiles

- ✓ Thermoplasms- Fond of FeS; optimum growth pH is 1.5 & temp. being 60°C. Eg. *Picrophilus* sp.can grow at pH 0 even!!
- ✓ Extremely thermophilic S° reducers- Can reduce elemental Sulphur to H_2S and growth occurs at 88-100°C. Eg. *Pyrococcus furiosa*
- ✓ Sulphate reducers- Eg. *Archaeoglobus* sp. ; optimum growth temp. ~ 85°C & pH is 3.5.

▪Thermophilic & extremely acidophilic archaea: most acidphilic archaea known

Thermoplasmatales

- Thermoplasma
- Picrophilus
- Ferroplasma

▪Cell wall-less microorganisms- *Thermoplasma & Ferroplasma*

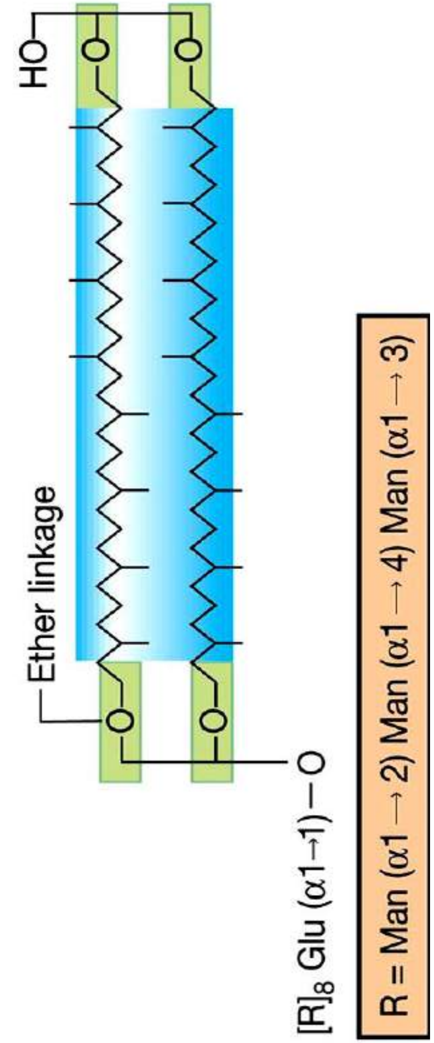
▪ *Thermoplasma*

1) *Chemoorganotroph*, optimum growth condition. 55˚C &pH=2 in complex medium

2) Facultative aerobes: grow either aerobically or anaerobically by sulphur respiration

3) Mainly FeS_2 is used up by these microbes in coal refuse piles; heating up of refuse coal mines due to microbial metabolism t combustion temperature

4) *T.acidophilum & T.volcanicum*
5) Unique technique to withstand these very harse conditions: specialised cytoplasmic membrane structure: **LIPOGLYCAN-** tetraether lipid monolayer membrane with mannose & glucose.
6) Presence of glycoproteins not sterols in membrane

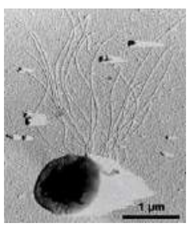


1) Small genome & DNA is complexed and packed with highly basic DNA-binding proteins → Globular particulate structure : Nucleosome like form; some what similar to HU of bacteria but other members of Euryarchaeota contains DNA complexing proteins similar to eukaryotic histones!

2) Picrophilus: optimal growth pH is 0.7 and even at pH 0

3) Consisting of S-layer and much lower GC content compared to *Thermoplasma* or *Ferroplasma*

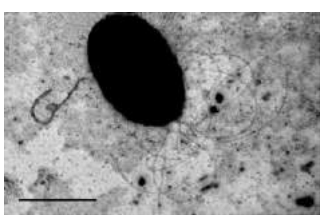
4) Moderate acidity such as~ pH 4.0- disintegration of picrophilus cell membrane



Thermococcales → *Thermococcus* {55°-95° C}

- Spherical hyperthermophile; anoxic habitat
- Tufts of polar flagella & highly motile
- Obligate anaerobe, chemoorganotroph metabolising proteins & other organics using S^0 as e^- acceptor

Thermococcales → *Pyrococcus* {70°-105° C}



- Metabolically, digest proteins, starch or maltose are oxidised using S^0 as e^- acceptor
- Absence of S^0 as e^- acceptor, leads to H_2 liberation

❖ *Pyrococcus furiosus*: Pfu DNA polymerase equipped with proofreading activity

Nanoarchaeota: **parasite /symbiont of Archaea!!**

✓Only a single species: *Nanoarchaeum equitans*, one of the smallest organisms known

✓Smallest genome amongst archaea- 0.49Mb; Coccoid cells- 0.4µM diameter

✓Growth only possible when attached to host organism *Ignicoccus hospitalis* another hyperthermophilic member of Crenarchaeota

The hospitable fireball

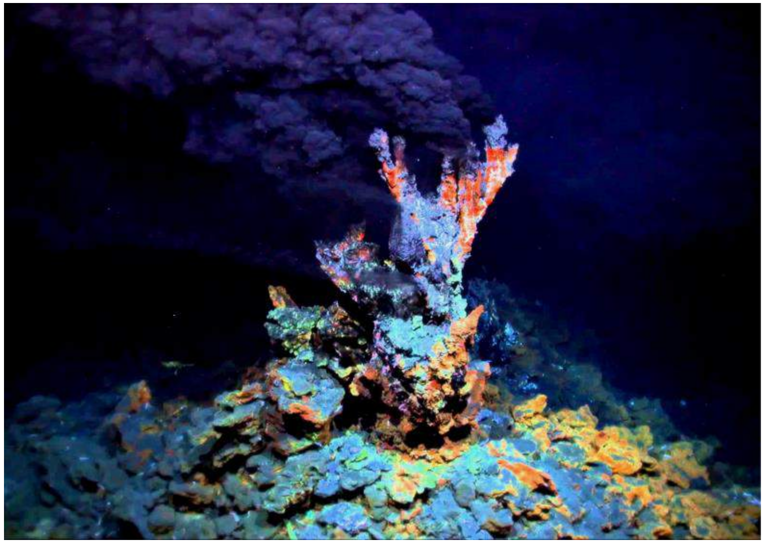
✓Isolated from geothermal vent along with the host

✓Optimum temp. 90˚C {70˚C -98˚C }

✓Generally depends on Ignicoccus (Autotroph, H_2 : e^- donor; S^o: e^- acceptor), supplies Nanoarchaeum with organic C

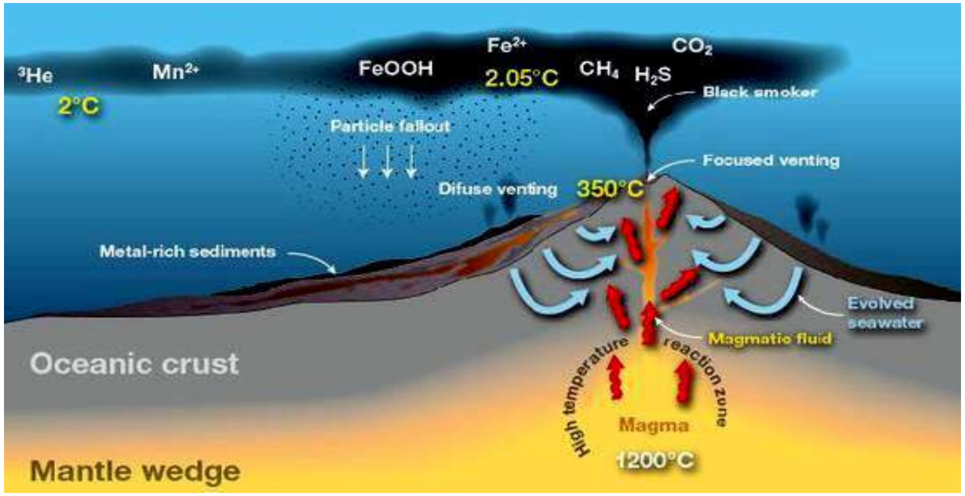
✓Most other metabolic features are yet to be confirmed properly

✓Cell wall composed of S-layer upon a periplasm type space



Life above ~130°C: Any limit of Height!

- *Pyrobolus fumarii*- 113°C
- *Methanopyrus kandleri*- 122°C
- Guess: life is possible above ~150°C too
- Presence of biochemical markers in measurable amount in vent discharges?? DNA/RNA/Protein!



- ○Degradation of ATP at temp. ~150°C in laboratory conditions
- ○How it may be functioning above temp. ??

↓

○Some kind of adaptation:

↓

Purely different biomolecules altogether or protection by any means??

↓

Stability

Protein structure:

a. Unique Folding; hydrophobic cores; salt bridges at surfaces

b. Chaperonins- Thermosome in *Pyrodictium abysii*

High level of thermosome content at 110°C & save the cell after a shock of autoclaving → Renaturing most of the proteins

DNA Stability: DNA binding proteins

a. Increase of cellular solute level: K-cyclic 2,3-bisphosphoglycerate & K-di-*myo*-inositol phosphate

b. High [Spermidine]: stability of ribosomes & nucleic acids

c. *REVERSE GYRASE*: Introduction of +ve supercoils in DNA; stabilising it at very high temp…. Absence of Rev. gyrase in life forms having temp. optima <80°C

d. Presence of highly basic DNA-binding proteins in Euryarchaeota- forming nucleosome like structure & ds stability of DNA

Lipid & Ribosome stability: Dibiphytanyl tetra ether type- lipid monolayer

~15% greater amount of GC content in small ribosomal RNA; low GC content in DNA