

Locating a stress sensor!

SINCE bacteria have decided not to be insensitive about such serious situations, they are used to developing combating responses to aid survival during stress. Recent emergence of pathogenic microorganisms that are able to resist some food preservation regimes, e.g. acid resistance of *Escherichia coli* O157, make the need for understanding the mechanisms of bacterial stress responses not only necessary for fundamental knowledge but for practical purposes as well.

Over its mostly short life span a bacterial cell is inevitably prone to physical and chemical environmental hardships. This is commonly the case when food processors apply heat, cold, salt, etc, to ensure the safety of their produce or when the human body expresses defence mechanisms to combat bacterial infection.

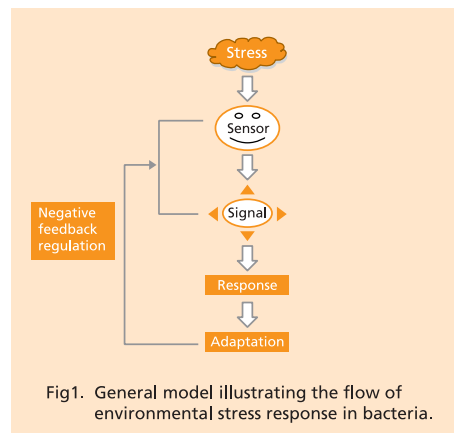


Fig1. General model illustrating the flow of environmental stress response in bacteria.

Figure 1 illustrates a rather simplified model of the most plausible and popular stress response cascade. In this, a bacterial structure senses the unfavourable change in the environment (stress) and emits a signal that induces the expression of cellular mechanisms in order to cope with the emergent hardship. Attaining satisfactory adaptation state would stimulate a feedback regulation to switch off or slow down the signal emanation. Obviously, sensing stress is the start point. While this is reasonably certain, various conflicting rationales have been introduced to define where stress is sensed by cell.

Stress sensing via cytoplasmic membrane.

Because of its location and components, the cytoplasmic membrane has been traditionally suggested to sense environmental changes through certain proteins that expand into the periplasm to interact with stress. The introduction of the two-component signal transduction

model with its fascinating and elegant concepts has been useful to explain the mechanism of the membrane in some cases. There are two main components in this system: sensor and regulator. The sensor is located in the cytoplasmic membrane and contains an input domain that senses the stimulus and passes it to a transmitter (**figure 2**). Through a series of phosphorylation and dephosphorylation reactions, the latter forwards a signal to the regular, located in cytoplasm, which induces the expression of certain genes involved in the stress response.

This system was found to serve well in mediating responses to phosphate or nitrogen starvation, responses to oxygen limitation, responses to changes in carbon, nitrogen sources, and very recently it was shown to be involved in heat response. However, some people have not been happy with such an appealing story and have provided evidence that it is not necessarily the membrane.

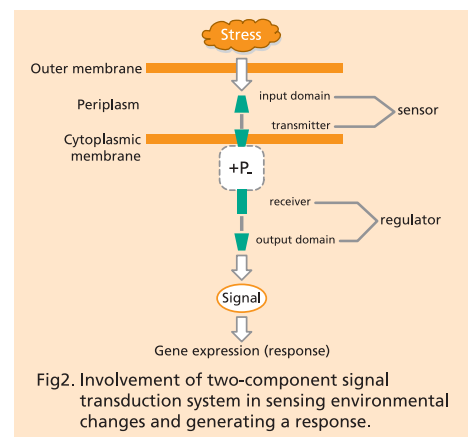


Fig2. Involvement of two-component signal transduction system in sensing environmental changes and generating a response.

Could ribosomes sense stress?

Given that it is at the ribosome that gene messages are translated into proteins, their role could be envisaged to be rather effective at later stages in the stress response cascade. However, the intriguing observations of VanBogelen and Neidhardt demonstrated that ribosomes can fulfil the sensor's motifs during heat or cold shock. They found that the addition of certain antibiotics that target ribosomes stimulated the expression of the same arrays of proteins induced on temperature up- or downshift (the so-called heat or cold shock

proteins). Startlingly, the patterns of the antibiotic-induced proteins simulated those of mild or severe temperature shift proteins depending on the drug concentration.

The heat or cold shock proteins (HSPs or CSHs) have been presumed to aid a cell's adaptation by allosterically preventing and / or repairing damage to cellular molecules following abrupt temperature change. This could lead to the situation represented in **figure 3**.

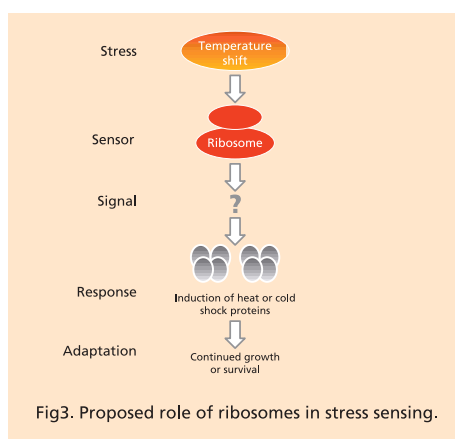


Fig3. Proposed role of ribosomes in stress sensing.

The increase in the synthesis of these proteins following temperature shock was found to be transient, which may refer to the feedback-repressing signal. Recently, HSPs and CSPs were shown to be involved in facilitating ribosomal jobs during stress; initiation of translation, coupling transcription with translation or recycling ribosomes after the completion of polypeptide synthesis. This may reveal another dimension in the mechanism - that ribosomes sense stress and generate signal since they need the response.

How far could ribosomes be implicated in sensing other stresses? Another research group (Zhang *et al.*) have recently revisited this model and hypothesized that ribosomes could be a general sensor for various environmental assaults experienced by *Bacillus subtilis*. Indeed, the evidence for this was indirect.

Sensing stress extracellularly!

Based on a series of related studies undertaken by his group, Professor Robin Rowbury developed a rather revolutionary concept of stress sensing by *Escherichia coli*. He suggested that bacteria would not wait until the stressing stimulus got through to trigger intracellular components; i.e. membrane proteins or ribosomes as this would delay the response to stress. Instead, cells may constitutively synthesise extracellular proteins that could detect environmental

stress in the growth medium. These proteins have been called "extracellular sensing components (ESCs)". They are presumed to be activated by stress which changes their structure as to be able to interact with cell surface receptors inducing appropriate response. At this stage, ESCs are termed extracellular induction components (EICs)- in other words, ESCs are precursors of EICs. Because of their diffusibility, EICs can also travel to other unstressed cells and serve as "alarmones" of anticipated stress.

This model proved useful to explain some interesting observations found in Rowbury's lab; among them was the increase in acid tolerance of *E. coli* following previous exposure to mildly low pH (acid habituation). Another observation was the ability of EICs, produced by acid habituated cells, to confer acid endurance on unadapted organisms. Relying upon the observation that agents which suppress the synthesis of ESCs, and accordingly EICs, also retard acid habituation, it was suggested that these extracellular components were necessarily involved in acid habituation phenomenon. A major reservation of this conclusion is the completion of acid adaptation within 20-30 minutes of an organisms exposure to mildly low pH (mostly pH 5), while in most experiments, the extracellular components were prepared from cultures grown at the same pH for 90 or 120 min.

Beside the need for a more precise structural description of ESCs and EICs, and resolving whether different growth media formulas could affect their synthesis and function, more details on the induction of cell response by the EICs are required. There is no obvious

evidence that EICs are sensed by cell surface components and if the alternative were cytoplasmic membrane sensors, the model would lose a prime advantage which is the early alert of environmental insults.

Single or multi stress sensors?

In each of the above three models, proposing a stress sensing mechanism was the outcome of studying bacterial responses under certain environmental changes; e.g. temperature shock for ribosome sensing. This raises the question of whether there are different sensors for different stresses. "Yes" might be the answer in some contexts of bacterial behaviour. When bacteria are grown in differently supplemented media, their rate of growth changes correspondingly with changes in the macromolecular composition of the cells. Changing the growth temperature can also effect this variation in growth rate, yet this would not involve similar modifications in cellular molecules. Slowing down the growth rate in poor media or under unfavourable temperatures could be seen as an adapting mechanism given that rapidly growing cells are more vulnerable to stress than dormant ones. So, it seems that different mechanisms might serve during various stresses to end up with an adapting response. It is also possible that these three systems might coordinate together for sensing environmental changes. □

Walid M El-Sharoud

School of Food Biosciences,
The University of Reading, UK

Further reading:

- Neidhardt, F.C. 2002. **Microbial reaction to environment: bacterial stress responses revisited in the genomic-proteomic era**, p. 1-18. In D.A. Hodgson and C.M. Thomas (ed.), Signals, switches, regulons and cascades: control of bacterial gene expression. Cambridge University Press, Cambridge.
- Parkinson, J. S. 1995. **Genetic approaches for signalling pathways and proteins**, p. 9-23. In J. A. Hoch and T. J. Silhavy (ed.), Two-component signal transduction. ASM Press, Washington, D.C.
- Rowbury, R. J. 2001. **Cross-talk involving extracellular sensors and extracellular alarmones gives early warning to unstressed *Escherichia coli* of impending lethal chemical stress and leads to induction of tolerance responses**. *Journal of Applied Microbiology* **90**: 677-695.
- VanBogelen, R. A. and F. C. Neidhardt. 1990. **Ribosomes as sensors of heat and cold shock in *Escherichia coli***. PNAS **87**:5589-5593.
- Zhang, S., J. M. Scott and W. G. Haldenwang. 2001. **Loss of ribosomal protein L11 blocks stress activation of the *Bacillus subtilis* transcription factor sigma (B)**. *J. Bacteriol.* **183**: 2316-2321.