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Environmental Metabolomics for the layperson

What is Environmental Metabolomics?

Metabolomics is a new experimental technique that is becoming widely used in biology, medicine and the environmental sciences for studying living organisms. This method measures the concentrations of the large numbers of naturally occurring small molecules (called metabolites) that are present in blood, urine, and tissues.

The pattern, or fingerprint, of metabolites in the biological sample can be used to learn about the "health" of the organism. This approach is being used to study human diseases, with the goal to find unique patterns of metabolites that could be used to diagnose specific diseases. In a similar manner, *environmental metabolomics* is being used to study the effects of environmental stress – such as pollution and climate change – on the health of fish and invertebrate organisms that live in our natural environment. Areas of application of environmental metabolomics include aquatic toxicology, terrestrial toxicology, fish diseases, aquaculture, environmental monitoring and ecological risk assessment.

The tools used to measure metabolite levels are more commonly associated with chemistry laboratories, and include nuclear magnetic resonance (NMR) spectroscopy and mass spectrometry. Also, because of the immense amount of data that is collected in a typical experiment, experts in mathematics and computer science are needed to analyse the data. Finally, biologists are needed to design the experiments and to help interpret the results.

For a basic introduction to the application of metabolomics to environmental studies, read "**Environmental Metabolomics: The Study of Disease and Toxicity in Wildlife (http://www.actionbioscience.org/genomic/viant.html)**".

For a general introduction to metabolomics, that requires a basic level of scientific knowledge, read "**Metabolomics: What's Happening Downstream of DNA (pdfs/Metabolomics%20Review.pdf)**".

For a more thorough introduction to environmental metabolomics, read:
"Metabolomics of aquatic organisms: the new omics on the block", *Mar. Ecol. Prog. Series* **332**, 301-306 (2007).

"Metabolomics: Methodologies and Applications in the Environmental Sciences", *J. Pestic. Sci.* **31**, 245-251 (2006).

Components of a Metabolomics Study

There are four components to most metabolomics experiments:

1. **Good experimental design** - this is critical if you are to successfully answer the biological questions posed. The design includes the choice of species, the collection and treatment of those organisms, the choice of method to extract metabolites from the biological samples (see 2), the choice of tool to measure the metabolites (see 3), and the choice of mathematical method to use to analyse the resulting data. Note that a metabolomics experiment does not have to test a specific hypothesis, but rather if the experiments are designed correctly they can generate new hypotheses to test in subsequent targeted experiments. This is in contrast to "traditional" studies in biology which always have an hypothesis that the researcher attempts to prove or disprove.
2. **Collection of samples and extraction of the metabolites** - for this part of the study it is critical that the methods used are highly reproducible. Once samples have been collected a range of methods can be used to chemically extract the metabolites from the rest of the cellular material (e.g. discard the DNA, RNA and proteins). The extraction protocol often employs organic solvents such as methanol.
3. **Measurement of metabolites** - many different techniques can be used to detect and characterise the levels of metabolites in biological samples. The two most common approaches are NMR spectroscopy and mass spectrometry. Different methods have their own advantages and disadvantages and so choosing the right technique to answer the biological questions of interest is of crucial importance.
4. **Data analysis (bioinformatics)** - a wide range of tools exist for analysing metabolomics data. It is again important to select the appropriate method to answer the particular question(s) that you are investigating. The overall goal of many analyses is to sieve through all the metabolic measurements and to discover the often small number of metabolites that are different between the groups of organisms that are being studied. Those metabolites can then potentially be used as *biomarkers* for that particular stress or disease.

Challenges in Environmental Metabolomics

There are several challenges and difficulties in environmental metabolomics that are hindering the widespread introduction of this approach into the environmental sciences.

1. **Communication between scientists** - as highlighted above, this field is highly multi-disciplinary. Unfortunately, just as different countries speak different languages, so different fields of science also use different languages. Metabolomics requires that scientists from diverse backgrounds such as chemistry, biology and computer science are able to discuss projects together, which can be quite challenging. There is a need for multi-disciplinary training for young researchers.
2. **Further development of tools to measure metabolites** - since this field is still in its infancy, there is an on-going development of several different tools used to detect and characterise metabolite levels. Although this is a necessary process, it creates considerable confusion for researchers entering the field of environmental metabolomics. Furthermore, compared to many of the methods used in biology, both NMR spectroscopy and mass spectrometry are technically demanding approaches that require considerable training. There is a need to establish centres of excellence in environmental metabolomics to facilitate the more widespread application of this approach.
3. **Standardisation and validation of environmental metabolomics** - currently different research groups are using different experimental designs, different analytical tools and different data analysis tools to conduct their metabolomics research. This makes it difficult to compare results between different studies. Furthermore, there has been no validation to confirm that even if environmental scientists use the same tools that the same results will be achieved (which is of course critical for a "validated method"). These issues are now beginning to be addressed.
4. **Identification of metabolites** - The unambiguous identification of metabolites (in an NMR or mass spectrum) is a crucial component of any metabolomics study, and is needed to provide the greatest possible amount of biological information and new knowledge. However, few publicly available libraries exist that contain the information needed to achieve this, in particular for "exotic" metabolites that may occur in non-mammalian organisms. There is a need to construct databases containing spectra of pure metabolites to enable unambiguous metabolite identification.

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