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What can be learned from the comparison between culture grown bacteria on selective media and environmental uncultured samples for the assessment of the bacterial diversity?

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Advances in culture independent methods have revolutionized microbial world. However, they cannot make links to bacterial physiology that culturing provides and there is an increasing need to understand the limits and benefits of culture media. Here, we compared the bacterial community structure of 3 freshwater samples from different altitudes in the catchment of Durance. Diversity was assessed through analysis of the V3-V4 region of 16S rRNA gene of total DNA extracted from samples and compare to those observed following cultivation on 3 media (1/10TSA, a broad range medium, KBC and CVP two semi selective media adapted respectively to *Pseudomonas syringae* group and to pectinolytic bacteria of the genera *Pectobacterium* and *Dickeya*). The 3 uncultured environmental water samples showed a dramatic decrease in diversity and evenness from the highest to the lowest sampling point correlated with a qualitative change in organic matter. Cultivation of these 3 samples alters the recovered bacterial community whatever the culture medium used, diversity and evenness being dramatically lower and erases the diversity differences observed between the three uncultured freshwater samples.

As expected, calculated diversity and evenness were higher on 1/10 TSA generalist culture medium compared with those observed for the semi selective KBC and CVP culture media, both showing similar diversity and evenness index. Qualitative analysis of phyla and genera lead to unexpected results

P. syringae and Pectinolytic bacteria were nevertheless easily identified and isolated on respectively KBC and CVP media. On CVP medium, pectinolytic bacteria create holes in the medium through polypectate degradation and it correlates with the occurrence of numerous OTU belonging to the *Pectobacterium* genera following CVP cultivation. As such OTU were not detected in uncultured freshwater samples, it indicates that *Pectobacterium* bacteria belong to the rare biosphere in freshwater samples.