Themenkreis B: Biodiversität

BSL 7 Taxon identification of plant tissue-containing herbal mixtures using single molecule real time sequencing



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Species identification often is a challenging and time-consuming enterprise that requires on the one hand taxonomic expertise, especially in case of young species rich radiations, and on the other hand most often well-developed fertile specimens. Considering this, an exact identification of plant fragments, pollen, seeds, or roots on the species level using traditional (e.g. microscopic, phytochemistry etc.) methods is nearly impossible. Genetic barcoding, however, can aid this. In recent years, DNA-barcoding revolutionized taxon identification using specific regions of the genome that are able to discriminate species. The main advantage is not only the underlying option of a cost effective and fast high-throughput process, but also the fact that any kind of tissue can be analyzed. Various national DNA-barcoding projects around the world, such as the German Barcode of Life (GBoL) initiative (www.bolgermany.de), compile a worldwide searchable database containing the DNA-barcodes of all species on Earth. However, the problem arises with herbal drug mixtures, e. g. herbal teas, dietary supplements, or herb and spice mixtures, as the standardized DNA barcoding routine cannot be applied anymore. Instead, additional methodological steps (i.e., cloning) need to be introduced, which are laborious and costly, due to the high number of clones required to statistically represent the taxonomic breadth of the sample. In contrast, well-established next generation sequencing (NGS) platforms, such as HiSeq or MiSeq, can provide coverage but they fail to deliver sufficient read length. Because of those read length problem, only a fraction of the targeted DNA barcode can be obtained, limiting the resolution power. Here, we present a strategy that will allow a precise species level identification of herbal mixtures using the established full-length DNA barcoding regions via single molecule real time sequencing (SMRT), which provides full length sequences of the DNA-barcode regions – irrespective of its length - and coverage for statistical analyses.