DNA analysis of a single pollen grain to identify the provenance of pollen transported to Antarctica

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This study carried out DNA analysis of a single pollen grain from Antarctic snow. It may allow identification of pollen species and definition of pollen provenance from distribution of the pollen source plants. However, little is known about the pollen concentration and composition in Antarctic snow. This study consequently examined snow samples obtained from three sites in Antarctica to reveal them. The results showed that the pollen concentrations ranged 0.2-1.2 grains/kg. The compositions revealed that *Pinus* pollen was a major component and represented 50-100%. Therefore, we used *Pinus* pollen for the analysis. Three regions: *rbcL* gene (1331 bp), the spacer region between *rpl20* and *rps18* (576 bp) and *clpP* gene (220 bp) in the chloroplast genome were amplified by the polymerase chain reaction (PCR) and sequenced to identify the species. The rates of success during amplifications from the extant pollen of *Pinus resinosa* that were directly collected from the male flowers were 33.3% for *rbcL*, 75.0% for *rpl20-rps18* and 100% for *clpP*, respectively. Higher success rate associated with amplification of shorter fragment was observed. Next, the amplification for *rpl20-rps18* was attempted on 7 *Pinu*s pollen grains from Antarctic snow samples but no success was achieved. In addition, the amplification for *rpl20-rps18* and *clpP* was tested, respectively, on 6 and 27 *Pinus* pollen grains collected from surface snow samples of a Russian Altai mountains' glacier. However it also failed. On the other hand, the existence of DNA in pollen grains from the Russian glacier was confirmed with SYBR Gold-stained. Therefore, the main reason of the unsuccess seems to be a problem with the PCR conditions. Consequently, the multiplex PCR was used for the amplification of Pinus pollen grains from the glacier. So far, sequence data have been obtained at success rate of around 20%. A higher success rate would be achieved by improving the multiplex PCR technique.