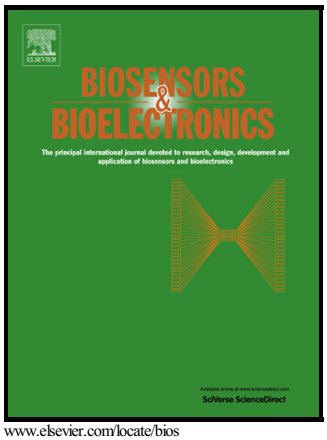

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Fenton Fragmentation for Faster Electrophoretic On Chip Purification of Amplifiable Genomic DNA

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With a rapid and simple actuation protocol electrophoretic nucleic acid extraction is easy automatable, requires no moving parts, is easy to miniaturize and furthermore possesses a size dependent cut-off filter adjustable by the pore size of the hydrogel. However electrophoretic nucleic acid extraction from bacteria has so far been applied mainly for short RNA targets. One of the reasons is that electrophoretic processing of unfragmented genomic DNA strands is time-consuming, because of the length. Here DNA fragmentation would accelerate extraction and isolation. We introduce on-chip lysis and non-enzymatic DNA cleavage directly followed by a purifying step for receiving amplifiable DNA fragments from bacteria in less than 25 minutes. In contrast to restriction enzymes the Fenton reaction is known to cleave DNA without nucleotide specificity. The reaction mix contains iron (II) EDTA, sodium ascorbate, hydrogen peroxide and lysozyme. The degree of fragmentation can be adjusted by the concentration of reagents. The results enable electrophoretic extraction methods to unspecifically process long genomic DNA in a short time frame, e.g. for pathogen detection in a lab-on-a-chip format.

Highlights:

- We obtain amplifiable DNA fragments from bacteria in less than 25 minutes.
- Lysis, DNA fragmentation and on-chip electrophoretic purification are combined.
- Inexpensive, non-enzymatic and sequence unspecific fragmentation is applied.
- Electrophoretic purification of long DNA strands can be accelerated that way.
- The degree of fragmentation can be adjusted by the reagent concentration.