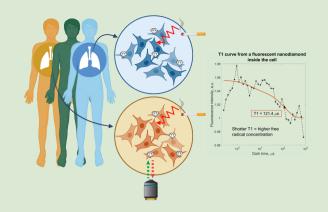
Nanodiamonds can be used for direct, all-optical, highly sensitive monitoring of free radical production inside live primary cells in health and disease





about our research?

Harnessing the power of nanodiamond magnetometry for free radical detection in primary cells

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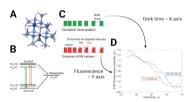
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BACKGROUND

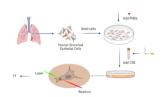
Nanodiamond magnetometry is a new technique for free radical detection. It senses the spins of unpaired electrons in the sample and can, in principle, achieve single-spin resolution1, while requiring as little as one cell for the analysis. Our group has successfully applied nanodiamond magnetometry to various live cells2-5.

In this study, we use it to detect free radicals in primary human airway epithelial cells exposed to cigarette smoke extract (CSE).



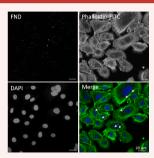
Fluorescence intensity of nitrogen-vacancy centers in nanodiamonds (A) depends on their quantum state (B), which, in turn, is affected by external spins. With a specific laser pulsing sequence (C) we can read out these quantum states and infer the free radical concentration around the ticle (D). orter T1 = more radicals!

METHODS

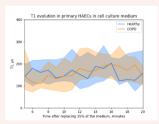


Cells from **3 healthy and 3 COPD donors** were incubated with nanodiamonds and then exposed to CSE (0%, 5%, 15%, 35%). T1 changes were monitored from the start of the exposure for 20 minutes. We validated the method in BEAS-2B cell line. using DCFDA assav as a control.

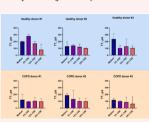
RESULTS



Fluorescent nanodiamonds are successfully internalized by the primary human airway epithelial cells after 2 hours of incubation (1 µg/mL in the cell culture

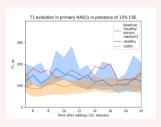


Without the trigger, there are no significant differences in T1 values between the cells from COPD patients and cells from healthy donors (p=0.6990).

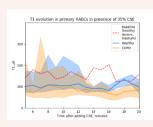


Cells from different donors with the same health status show slightly different response to CSE.

Adding CSE to the cells from COPD patients significantly shortens T1 (p=0.0007 from two-way ANOVA). Cells from COPD patients are sensitive to lower CSE concentrations than cells from healthy donors (at 5%, p=0.0968).

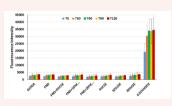


The differences between healthy donors and COPD patients are the most pronounced at **15% CSE** (p=0.0247).

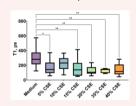


At high CSE concentration (35%), cells from healthy and COPD donors show the same response (p=0.3189).

VALIDATION IN BEAS-2B



Conventional assays, such as DCFDA, do not show the effect of CSE within 2 hours. Diamond magnetometry shows a drop in T1 after 10 minutes of CSE exposure.



CONCLUSIONS

- Nanodiamond magnetometry shows increased free radical production in the cells exposed to CSE within the first 20 minutes - earlier than othe conventional assays, such as DCFDA
- Cells from COPD donors do not show higher free radical load without the stressor.
- At the same time, cells from COPD donors are more sensitive to CSE than cells from healthy donors. Further research is needed into the underlying molecular mechanisms.
- Nanodiamond magnetometry captures the cell variability between different donors with the same health status. It might be a useful tool for personalized medicine.

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