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## Real-time electrochemical detection of paracetamol interaction with intestinal tissue

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#### **Title:**

**Real-time electrochemical detection of paracetamol interaction with intestinal tissue**

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1. The title should be as brief as possible but long enough to indicate clearly the nature of the study. Capitalise the first letter of the first word ONLY (place names excluded). No full stop at the end.

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Oral drug delivery is the preferred route for drug administration mainly due to good patient compliance. A myriad of approaches is already in use to study the effect of drugs in biological systems, however, there is a constant need for new methods and tools to study their interaction in various parts of the gastrointestinal tract<sup>1,2</sup>. Several *ex-vivo* approaches are used for the evaluation of drug permeation through the small intestine. However, very little is known about the effect and interaction of drugs between the enzymes (e.g., native catalase) present in the intestine. In the current study, we show the application of an electrochemical O<sub>2</sub> sensor for studying the effect of paracetamol on the native catalase in the intestinal tissues.

The sensors, prepared by mounting the cleaned tissue isolated from porcine on a custom-made Clark type electrode, (Fig. 1a) were exposed to different concentrations of H<sub>2</sub>O<sub>2</sub>, the substrate for catalase. Addition of H<sub>2</sub>O<sub>2</sub> can also mimic local changes in redox environment in the tissue, like in the case of inflammation, which results in increased H<sub>2</sub>O<sub>2</sub> levels<sup>3</sup>.

The observed current change, production of O<sub>2</sub>, is due to the reaction of H<sub>2</sub>O<sub>2</sub> with catalase (Fig. 1b). Our experiments indicate that the intestinal tissue contains a significant amount of catalase, considering the current generated after the addition of 500 μM substrate. We observe that there is a linear relationship between H<sub>2</sub>O<sub>2</sub> concentration and current response (Fig. 1c). Additionally, we demonstrated the antioxidant capacity of paracetamol, which can be seen from decreasing O<sub>2</sub> production (Fig. 1d).

To the best of our knowledge, we present for the first time a method for direct electrochemical measurement of drug-catalase interaction in intestine. As a next step, we intend to further study the effects of other antioxidants and pharmaceuticals on catalase activity in intact tissues.

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