



Abant Tıp Dergisi

Abant Medical Journal

Abant Med J 2024;13(2):45-46, doi:10.47493/abantmedj.1529498



A Call for Innovation in Drug Screening: Integrating Genetically Encoded Biosensors for Deeper Insights

İlaç Taramasında Yenilik Çağrısı: Daha Derin Bilgiler İçin Genetik Olarak Kodlanmış Biyosensörlerin Entegrasyonu

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Geliş Tarihi (Received): 07.08.2024

Kabul Tarihi (Accepted): 18.08.2024

Yayın Tarihi (Published): 29.08.2024

Dear Editor,

We are writing to express our views on the urgent need for more informative and efficient methods in drug screening and testing within preclinical and translational medicine. The current state of the art primarily involves 2,5-diphenyl-2H-tetrazolium bromide (MTT) or similar cell viability assays, which provide limited information, predominantly focusing on cell toxicity. These methods are poor in providing insights into specific cellular mechanisms, long-term effects, or the drug's mode of action, thereby failing to deliver the comprehensive analysis required in modern drug development (1).

In vivo experiments, specifically in the context of drug development, are increasingly being abandoned in both the EU and the USA due to ethical concerns and regulatory changes. As a result, the reliance on in vitro systems and the development of innovative tools for drug screening have become more crucial than ever. Among these, genetically encoded biosensors for high-content drug screening represent a highly promising yet underutilized approach in the field.

Genetically encoded biosensors are advanced tools designed to provide real-time insights into cellular processes. Engineered from natural or synthetic genetic elements, these biosensors are optimized for high-throughput applications, allowing detailed monitoring of cellular responses. They track various biological signals or molecules, offering precise data on drug mechanisms and interactions at the molecular level (2). Integrating these biosensors into drug screening protocols can significantly enhance the efficiency and effectiveness of drug development processes by providing critical, functional insights into how drugs impact cellular functions.

Some of our previous studies have highlighted the significant value and effectiveness of genetically encoded biosensors in this field. These biosensors have proven particularly valuable in studying nitric oxide (NO), a crucial signaling molecule in the cardiovascular system (3). NO is essential for regulating vascular tone, blood pressure, and overall cardiovascular health by activating soluble guanylate cyclase (sGC), leading to vasodilation and maintaining endothelial function and vascular homeostasis. In the context of cardiovascular therapeutics, nitroglycerin (GTN) is a well-established antianginal drug that exerts its effects through NO release. The bioactivation of GTN by aldehyde dehydrogenase-2 (ALDH2) converts it to NO, which then activates sGC and induces vasodilation. Despite GTN's clinical success, understanding the precise mechanisms of its NO release and its impact on NO signaling pathways has been challenging due to limitations in traditional measurement techniques. Our research has demonstrated how advancements in genetically encoded biosensors can overcome these challenges. These innovative tools provide important insights into the real-time dynamics and spatial distribution of NO within cells, enabling a more precise visualization of NO production and its effects on cellular signaling. Our study, employing NO biosensors, has notably advanced the understanding of NO's role in cardiovascular pharmacology.

Atıf/Cite as: Eroğlu E. A Call for Innovation in Drug Screening: Integrating Genetically Encoded Biosensors for Deeper Insights. Abant Med J. 13(2):96-97. doi:10.47493/abantmedj.1529498

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It has addressed longstanding questions about NO signaling in response to GTN, highlighting the intricate interplay between drug metabolism and NO production in the vascular system (4).

In another study, we assessed the efficacy of two approved iron supplement drugs – Venofer and Ferinject – using our genetically encoded biosensor approach. This method effectively detects available iron (II) in cultured cells, demonstrating that genetically encoded biosensors are valuable tools for drug screening and testing at the (sub)cellular level (5).

Another crucial point we address is the critical importance of physiological oxygen levels in drug testing. We discovered that cells cultured under physiological normoxia (5 kPa O₂) require notably lower iron supplementation to activate metalloproteins compared to cells grown in standard room air (18 kPa O₂) (6). Additionally, we used genetically encoded biosensors to investigate the complex interaction between hydrogen peroxide (H₂O₂) and nitric oxide (NO) in endothelial cells under varying oxygen conditions. Traditionally, simultaneous measurement of these molecules has been technically challenging, leading to inconsistent results. By employing advanced biosensors -HyPer7 for H₂O₂ and geNOps for NO- for concurrent imaging in single cells, we found that, under ambient oxygen conditions, H₂O₂ did not significantly impact NO production. However, under physiological normoxia, we observed distinct oxidative stress and nuanced NO responses to H₂O₂ (7). These results underscore the necessity of incorporating physiological conditions into cellular studies to obtain accurate, meaningful insights, thereby enhancing the reliability of drug testing protocols.

We believe that integrating genetically encoded biosensors into standard drug screening protocols could significantly enhance the development of safer and more effective therapeutics. Their ability to provide detailed functional insights at the cellular level addresses a critical gap left by traditional drug testing methods.

Thank you for considering our perspectives on this matter. We look forward to further discussions and collaborations to advance the field of drug screening and testing.

Sincerely,

Informed Consent: Written consent was obtained from the participants.

Conflict of Interest: Authors declared no conflict of interest.

Financial Disclosure: Authors declared no financial support.

Author Contributions: Idea/Concept: E.E.; Design: E.E.; Supervision: E.E.; Funding: E.E.; Materials: E.E.; Data Collection/Processing: E.E.; Analysis/Interpretation: E.E.; Literature Review: E.E.; Drafting/Writing: E.E.; Critical Review: E.E. The authors have accepted responsibility for the entire content of this manuscript and approved its submission.

References

1. Ghasemi M, Turnbull T, Sebastian S, Kempson I. The MTT assay: Utility, limitations, pitfalls, and interpretation in bulk and single-cell analysis. *Int J Mol Sci.* 2021; 22(23):12827.
2. Potekhina ES, Bass DY, Kelmanson IV, Bessmertnykh-Vass YE, Skobeleva OI, Krylov AV. Drug screening with genetically encoded fluorescent sensors: today and tomorrow. *Int J Mol Sci.* 2021; 22(1):148.
3. Eroglu E, Gottschalk B, Charoensin S, Lim J, Smith JD, Lee M. Development of novel FP-based probes for live-cell imaging of nitric oxide dynamics. *Nat Commun.* 2016; 7(1):1–11.
4. Opelt M, Eroglu E, Waldeck-Weiermair M, De Jongh S, Toth L, Mukherjee S. Formation of nitric oxide by aldehyde dehydrogenase-2 is necessary and sufficient for vascular bioactivation of nitroglycerin. *J Biol Chem.* 2016; 291(46):24076–84.
5. Sevimli G, Alston AE, Funk F, Zhang M, Liao Y, Wang H. Probing subcellular iron availability with genetically encoded nitric oxide biosensors. *Biosensors.* 2022; 12(10):903.
6. Sevimli G, Smith MJ, Caglar TA, Chen Y, Liu Y, Li Z. Nitric oxide biosensor uncovers diminished ferrous iron-dependency of cultured cells adapted to physiological oxygen levels. *Redox Biol.* 2022; 53:102319.
7. Altun HY, Secilmis M, Yang F, Lu Y, Zhang X, Zhao H. Visualizing hydrogen peroxide and nitric oxide dynamics in endothelial cells using multispectral imaging under controlled oxygen conditions. *Free Radic Biol Med.* 2024; 221:89–97.