



Editorial: Elevated Carbon Dioxide Sensing and Physiologic Effects

Eoin P. Cummins^{1*}, Ankit Bharat^{2,3}, Jacob I. Sznajder³ and István Vadász^{4,5,6}

¹School of Medicine and Conway Institute of Biomolecular and Biomedical Research, University College Dublin, Dublin, Ireland, ²Division of Thoracic Surgery, Feinberg School of Medicine, Northwestern University, Chicago, IL, United States, ³Division of Pulmonary and Critical Care Medicine, Feinberg School of Medicine, Northwestern University, Chicago, IL, United States, ⁴Department of Internal Medicine, Member of the German Center for Lung Research (DZL), Justus Liebig University, Universities of Giessen and Marburg Lung Center (UGMLC), Giessen, Germany, ⁵The Cardio-Pulmonary Institute (CPI), Giessen, Germany, ⁶Institute for Lung Health (ILH), Giessen, Germany

Keywords: hypercapnia, alveolar epithelium, skeletal muscle, ubiquitination, carbamate

Editorial on the Research Topic

Elevated Carbon Dioxide Sensing and Physiologic Effects

Hypercapnia, an elevated level of carbon dioxide (CO₂) in blood and tissue, is similarly to hypoxemia a fundamental gas exchange disturbance and is a hallmark of various pulmonary diseases (Vadász et al., 2012). Although hypoxia and hypoxemia have been very much in the focus of biological and clinical research in the last decades, much less attention has been afforded to hypercapnia. It is well known that CO₂ is a product of cellular oxidative respiration, and is primarily eliminated in mammals by the lungs. CO₂ homeostasis is indispensable for life, whereas alterations in CO₂ concentrations are sensed by cells, initiating specific signaling events (Cummins et al., 2020). In this Research Topic contributed by an international consortium of researchers, in forms of original research manuscripts, reviews and perspectives, several novel or most recently discovered aspects of CO₂ sensing and signaling mechanisms are covered. Here, we briefly summarize some of the main findings and themes discussed in the articles and outline some potential future directions for the field.

OPEN ACCESS

Edited and reviewed by:

Andrew John Halayko,
University of Manitoba, Canada

*Correspondence:

Eoin P. Cummins
eoin.cummins@ucd.ie

Specialty section:

This article was submitted to
Respiratory Physiology and
Pathophysiology,
a section of the journal
Frontiers in Physiology

Received: 11 March 2022

Accepted: 22 March 2022

Published: 27 April 2022

Citation:

Cummins EP, Bharat A, Sznajder JI
and Vadász I (2022) Editorial: Elevated
Carbon Dioxide Sensing and
Physiologic Effects.
Front. Physiol. 13:894222.
doi: 10.3389/fphys.2022.894222

ORIGINAL RESEARCH PAPERS

Gabrielli et al. presented a study on the mechanisms underpinning the endocytosis of the Na,K-ATPase via the ubiquitin pathway. The authors using buffered CO₂ conditions equivalent 110 mmHg demonstrated TRAF2-dependent ubiquitination of lysine 5 and 7 on the β-subunit of the Na,K-ATPase under conditions of elevated CO₂, downstream of PKC phosphorylation. TRAF2 is well known as an E3-ubiquitin ligase capable of facilitating K63 polyubiquitination, and also as a ubiquitin ligase important for NF-kappaB-dependent signaling cascades (Zhou et al., 2013). These findings linking hypercapnia and an E3-ubiquitin ligase are of particular interest given the recent findings by the Cann group in Durham University implicating ubiquitin itself as a direct target for post-translational carboxylation (Linthwaite et al., 2021).

Ceco et al. focused their research on the molecular mechanisms underpinning skeletal muscle function. Work from the Sznajder group has linked hypercapnia with airway smooth muscle and skeletal muscle dysfunction e.g., impaired function, differentiation, regeneration and recovery (Jaitovich et al., 2015; Shigemura et al., 2018; Shigemura and Sznajder, 2021). In this study, the authors investigated the metabolic phenotype of muscle exposed to hypercapnia and identified metabolic changes resulting in elevated fatty acid oxidation. Interestingly, use of an inhibitor of fatty acid shuttling, etomoxir, reversed the CO₂-dependent phenotype. The authors proposed that hypercapnia “overloads” mitochondrial metabolism in skeletal muscle.

Further work should reveal whether this metabolic phenotype unique to skeletal muscle or is more universal.

Üçpunar et al. focused their work on CO₂ sensitivity and signaling in *Drosophila*. While *Drosophila*-based studies on CO₂ sensitivity and sensing have traditionally focused on attraction to rotting fruit or elevated CO₂ (van Breugel et al., 2018), instead this study focused on the response of *Drosophila* to current ambient atmospheric CO₂ (400 ppm) compared to CO₂-free air. Evidence from this study suggests that ambient levels of CO₂ are sensed, and that the flies have a preference for levels of CO₂ that are lower than our current environment. Thus, this paper raised the intriguing question as to how an odor (CO₂) can be both attractive and aversive in the same animal. As part of the explanation, the authors proposed a circuit dependent on GR63a/GR21a for environmental preference to sub-atmospheric CO₂ levels.

LITERATURE REVIEWS AND PERSPECTIVES

Kryvenko et al. reviewed the role of the endoplasmic reticulum (ER) as a central signaling organelle in the context of CO₂-dependent signal transduction (Jaitovich et al., 2015). Recent evidence suggest that elevated Ca²⁺ downstream of hypercapnia may derive from the ER. Normal ER function is reliant on sufficient ATP and consequently hypercapnia-dependent metabolic changes can impinge on normal ER function and potentially lead to ER stress (Depaoli et al., 2019; Kryvenko et al., 2020). Dysregulation of Ca²⁺ and ATP-dependent chaperones can thus potentially contribute to aberrant protein folding in the ER under conditions of hypercapnia. Furthermore, the authors delineated the interplay between potentially maladaptive cellular responses to CO₂ in the context of the ER.

Shigemura et al. reviewed the impact of CO₂-dependent gene expression in different tissues, with a focus on the lung, skeletal muscle and innate immune signaling. The authors highlighted that many of the observed responses to hypercapnia are maladaptive, discussing recent clinical trial data in patients with severe lung diseases (Nin et al., 2017). It is clear that certain tissue types elicit signaling in a tissue-specific manner e.g., MuRF-1 is a key player in skeletal muscle dysfunction but is specific to that tissue type (Files et al., 2012). Interestingly, MuRF-1 is an E3 ligase like TRAF-2 and suggests that the ubiquitin proteasome system may be a common signaling thread in the response to CO₂. Another common and conserved signaling component involved in the cellular response to CO₂ is the Wnt signaling pathway, which is highly conserved in animals and has been shown to be sensitive to CO₂ across species (Shigemura et al., 2019).

Blake and Cann reviewed the role of carbamylation, a post-translational modification at N-terminal α -amino groups or lysine ϵ -amino groups of target proteins by CO₂. This direct interaction of proteins and CO₂ may alter protein function and might play a role in CO₂ sensing and initiation of subsequent signaling events. In particular, the review discusses the regulation of several proteins by carbamate post-translational modification as well as novel methodological advances in uncovering protein carbamylation at the level of the proteome. Among others, the

manuscript details the role of carbamylation in the mechanism by which CO₂ alters affinity of hemoglobin for oxygen [Bohr effect; reviewed in (West, 2019)], thereby linking carbamate post-translational modification to impaired gas exchange. Other examples of proteins functions of which are altered by carbamylation discussed in the review include, ribulose 1,5-bisphosphate carboxylase-oxygenase, which is responsible for atmospheric CO₂ elimination during photosynthesis (Andersson and Backlund, 2008), bacterial β -lactamases that enable resistance to β -lactam antibiotics as well as ureases, which catalyze hydrolysis of urea to carbonic acid and ammonia with relevance to human disease. Moreover, the regulation of connexin 26, a transmembrane protein that form hexameric connexons of gap junctions or hemichannels and which undergo conformational changes upon carbamylation is described, thus identifying a potential CO₂ sensing molecule.

Balnis et al. built on existing limitations of certain animal models to develop a new paradigm where animals demonstrate chronic obstructive pulmonary disease (COPD)-driven muscle dysfunction, but exhibit normal pCO₂ levels. Thus, hypercapnia can be superimposed on this model to examine the cross talk between the distinct stimuli. Interestingly, the authors reported a relatively “stepwise” decrease in absolute force generation with COPD, hypercapnia and COPD + hypercapnia, in contrast to specific force generation where a synergistic reduction in force was observed when COPD and hypercapnia were combined. The authors highlighted that these data underlie a complex phenotype that may relate to hypercapnia-dependent differences in ECM gene expression in the murine COPD model.

CONCLUSION AND FUTURE DIRECTIONS

Taken together, this Research Topic on “Elevated Carbon Dioxide Sensing and Physiologic Effects” addresses some of the state of the art topics in the field. How is CO₂ sensed acutely and more chronically? What are the mechanisms underpinning these responses e.g., neuronal receptors, signaling cascades, post-translational mechanisms and transcription factors? What are the consequences of these signaling mechanisms for the organism? Is the response to elevated CO₂ adaptive or maladaptive or are these consequences context-specific? Is the organismal response to CO₂ dependent on other factors such as the severity and nature of disease? This is relevant to the current coronavirus disease (COVID-19) pandemic as hypercapnia is often an inevitable clinical consequence of lung-protective mechanical ventilation strategies to reduce the incidence of ventilator-induced lung injury in patients with acute respiratory distress syndrome (ARDS). The relatively long duration of mechanical ventilation in patients with COVID-19 compared with patients with other causes of ARDS [median ~14 compared with ~4 days (Grant et al., 2021)] increases the duration of exposure to hypercapnia and thus has deleterious effects on lung repair after injury. Based on this Research Topic, a strong argument is made for 1) further examination of the ER as a central signaling organelle in hypercapnia and 2) altered ubiquitin-dependent signaling. The cross-talk between these two elements is worthy of future research,

particularly in conjunction CO₂-dependent cellular metabolic changes, which can influence both pathways and are of clinical relevance to patients with ARDS and COVID-19. The CO₂-field is ripe for attracting further excellent research in this area.

AUTHOR CONTRIBUTIONS

EC drafted and AB, JS and IV edited the manuscript. All authors approved the final version of the manuscript.

REFERENCES

- Andersson, I., and Backlund, A. (2008). Structure and Function of Rubisco. *Plant Physiol. Biochem.* 46, 275–291. doi:10.1016/j.plaphy.2008.01.001
- Cummins, E. P., Strowitzki, M. J., and Taylor, C. T. (2020). Mechanisms and Consequences of Oxygen and Carbon Dioxide Sensing in Mammals. *Physiol. Rev.* 100, 463–488. doi:10.1152/physrev.00003.2019
- Depaoli, M. R., Hay, J. C., Graier, W. F., and Malli, R. (2019). The Enigmatic ATP Supply of the Endoplasmic Reticulum. *Biol. Rev.* 94, 610–628. doi:10.1111/brv.12469
- Files, D. C., D'Alessio, F. R., Johnston, L. F., Kesari, P., Aggarwal, N. R., Garibaldi, B. T., et al. (2012). A Critical Role for Muscle Ring finger-1 in Acute Lung Injury-Associated Skeletal Muscle Wasting. *Am. J. Respir. Crit. Care Med.* 185, 825–834. doi:10.1164/rccm.201106-1150oc
- Grant, R. A., Morales-Nebreda, L., Morales-Nebreda, L., Markov, N. S., Swaminathan, S., Querrey, M., et al. (2021). Circuits between Infected Macrophages and T Cells in SARS-CoV-2 Pneumonia. *Nature* 590, 635–641. doi:10.1038/s41586-020-03148-w
- Jaitovich, A., Angulo, M., Lecuona, E., Dada, L. A., Welch, L. C., Cheng, Y., et al. (2015). High CO₂ Levels Cause Skeletal Muscle Atrophy via AMP-Activated Kinase (AMPK), FoxO3a Protein, and Muscle-specific Ring finger Protein 1 (MuRF1). *J. Biol. Chem.* 290, 9183–9194. doi:10.1074/jbc.m114.625715
- Kryvenko, V., Wessendorf, M., Morty, R. E., Herold, S., Seeger, W., Vagin, O., et al. (2020). Hypercapnia Impairs Na,K-ATPase Function by Inducing Endoplasmic Reticulum Retention of the β -Subunit of the Enzyme in Alveolar Epithelial Cells. *Int. J. Mol. Sci.* 21. doi:10.3390/ijms21041467
- Linthwaite, V. L., Pawloski, W., Pegg, H. B., Townsend, P. D., Thomas, M. J., So, V. K. H., et al. (2021). Ubiquitin Is a Carbon Dioxide-Binding Protein. *Sci. Adv.* 7, eabi5507. doi:10.1126/sciadv.abi5507
- Nin, N., Muriel, A., Muriel, A., Peñuelas, O., Brochard, L., Lorente, J. A., et al. (2017). Severe Hypercapnia and Outcome of Mechanically Ventilated Patients with Moderate or Severe Acute Respiratory Distress Syndrome. *Intensive Care Med.* 43, 200–208. doi:10.1007/s00134-016-4611-1
- Shigemura, M., Lecuona, E., Angulo, M., Homma, T., Rodríguez, D. A., Gonzalez-Gonzalez, F. J., et al. (2018). Hypercapnia Increases Airway Smooth Muscle Contractility via Caspase-7-Mediated miR-133a-RhoA Signaling. *Sci. Transl. Med.* 10. doi:10.1126/scitranslmed.aat1662

FUNDING

This work was supported by a Science Foundation Ireland Career Development Award (15/CDA/3490) (to EC); by grants from the National Institutes of Health (HL-147070) (to JS); the Federal Ministry of Education and Research [German Center for Lung Research (DZL/ALI 1.5, 3.3 and 3.4)], the von Behring Röntgen Foundation (Project 66-LV07) and the German Research Foundation (DFG/KFO309, P5 and The Cardio-Pulmonary Institute (EXC 2026; Project ID: 390649896) (to IV).

- Shigemura, M., Lecuona, E., Angulo, M., Dada, L. A., Edwards, M. B., Welch, L. C., et al. (2019). Elevated CO₂ Regulates the Wnt Signaling Pathway in Mammals, *Drosophila melanogaster* and *Caenorhabditis elegans*. *Sci. Rep.* 9, 18251. doi:10.1038/s41598-019-54683-0
- Shigemura, M., and Sznajder, J. I. (2021). Elevated CO₂ Modulates Airway Contractility. *Interf. Focus.* 11, 20200021. doi:10.1098/rsfs.2020.0021
- Vadász, I., Hubmayr, R. D., Nin, N., Sporn, P. H. S., and Sznajder, J. I. (2012). Hypercapnia: a Nonpermissive Environment for the Lung. *Am. J. Respir. Cell Mol. Biol.* 46, 417–421. doi:10.1165/rcmb.2011-0395ps
- van Breugel, F., Huda, A., and Dickinson, M. H. (2018). Distinct Activity-Gated Pathways Mediate Attraction and Aversion to CO₂ in *Drosophila*. *Nature* 564, 420–424. doi:10.1038/s41586-018-0732-8
- West, J. B. (2019). Three Classical Papers in Respiratory Physiology by Christian Bohr (1855–1911) Whose Work Is Frequently Cited but Seldom Read. *Am. J. Physiology-Lung Cell Mol. Physiol.* 316, L585–L588. doi:10.1152/ajplung.00527.2018
- Zhou, A. Y., Shen, R. R., Kim, E., Lock, Y. J., Xu, M., Chen, Z. J., et al. (2013). Ikk ϵ -Mediated Tumorigenesis Requires K63-Linked Polyubiquitination by a cIAP1/cIAP2/TRAF2 E3 Ubiquitin Ligase Complex. *Cel. Rep.* 3, 724–733. doi:10.1016/j.celrep.2013.01.031

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2022 Cummins, Bharat, Sznajder and Vadász. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.