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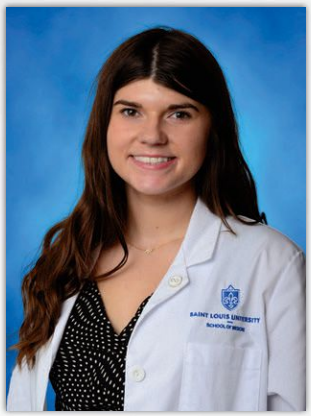
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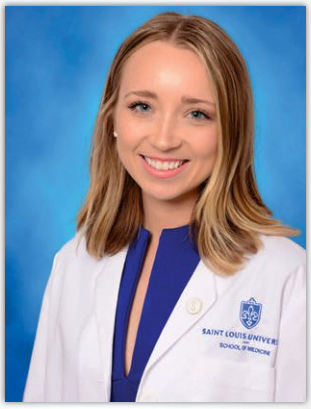
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A Tsunami of Concern: The Lasting Impacts of COVID Isolation and School Closures on Youth Mental Health



by Erick Messias, MD, PhD, Leticia Ugarte, MD, Enoch Azasu, PhD & Andrew J. White, MD

The COVID-19 pandemic disrupted lives in many ways across countries, cultures and age groups. Those years brought not only infectious risks but also a series of factors undermining the mental health of our youth.

In an attempt to mitigate the spread of the virus, lockdowns, quarantine measures, and school closures were implemented, at various levels, worldwide. At the time, these measures were deemed necessary to safeguard public health. However, the impact of these actions on the younger generation has been profound, and in some cases, damaging to their psychological health and development. A large amount of concerning information has been emerging regarding the delayed effects of COVID isolation and school closures on today's young people, encompassing mental health, socialization access and skills, academic performance, and suicide. Compounding these issues are the recent deregulation of cannabis, its ease of access, and increased use among teenagers. This perspective aims to provide an overview of these impacts, paying special attention to the role of COVID isolation and cannabis use in youth suicide. Post hoc analyses questioning the necessity of school closures are tempting, but fraught with difficulty. What is clear, however, is that resources supporting youth mental health were insufficient to prevent the consequences of the pandemic isolation, and that insufficiency persists today.



Erick Messias, MD, MPH, PhD, is the Samuel W. Fordyce Professor and Chair, Department of Psychiatry and Behavioral Neuroscience; Leticia Ugarte, MD is Assistant Professor, and Enoch Azasu, PhD, are in the Department of Psychiatry and Behavioral Neuroscience; **Andrew J. White, M**D, (pictured), is the Robert W. Wilmott Endowed Professor, Chair, Department of Pediatrics; all are at the Saint Louis University School of Medicine, St. Louis, Missouri.

It is crucial to prioritize the mental and emotional well-being of young people, not only during the pandemic but also in its aftermath.

Mental Health: The Silent Pandemic

The mental health of young people is an issue of ongoing and accelerating concern. The isolation brought about by the pandemic has contributed to a mental health crisis among the youth which is now echoing years after schools have reopened. Many studies and reports suggest that isolation and loneliness are associated with increased depression and anxiety prevalence among youth.[1] A large survey of the UK population showed that clinically significant levels of mental distress rose from 18.9% in 2018–19 to 27.3% in April 2020, one month into UK lockdown. The same survey found increases were greatest in 18 to 34 year-olds, women, and people living with young children.[2] It is essential to consider the long-term consequences of this mental health crisis, which could persist long after the pandemic ends.

Declining Socialization Skills: The Lost Art of Connection

Another concerning aspect of COVID isolation and school closures is the impact on the acquisition and development of basic socialization skills. Human interaction is fundamental for the development of these skills and the abrupt interruption of in-person socialization opportunities may have long lasting detrimental consequences. Young children and adolescents need to learn these essential social skills through peer interaction, which includes understanding social cues, working on 'in-the-moment' conflict resolution, and witnessing and fostering empathy.

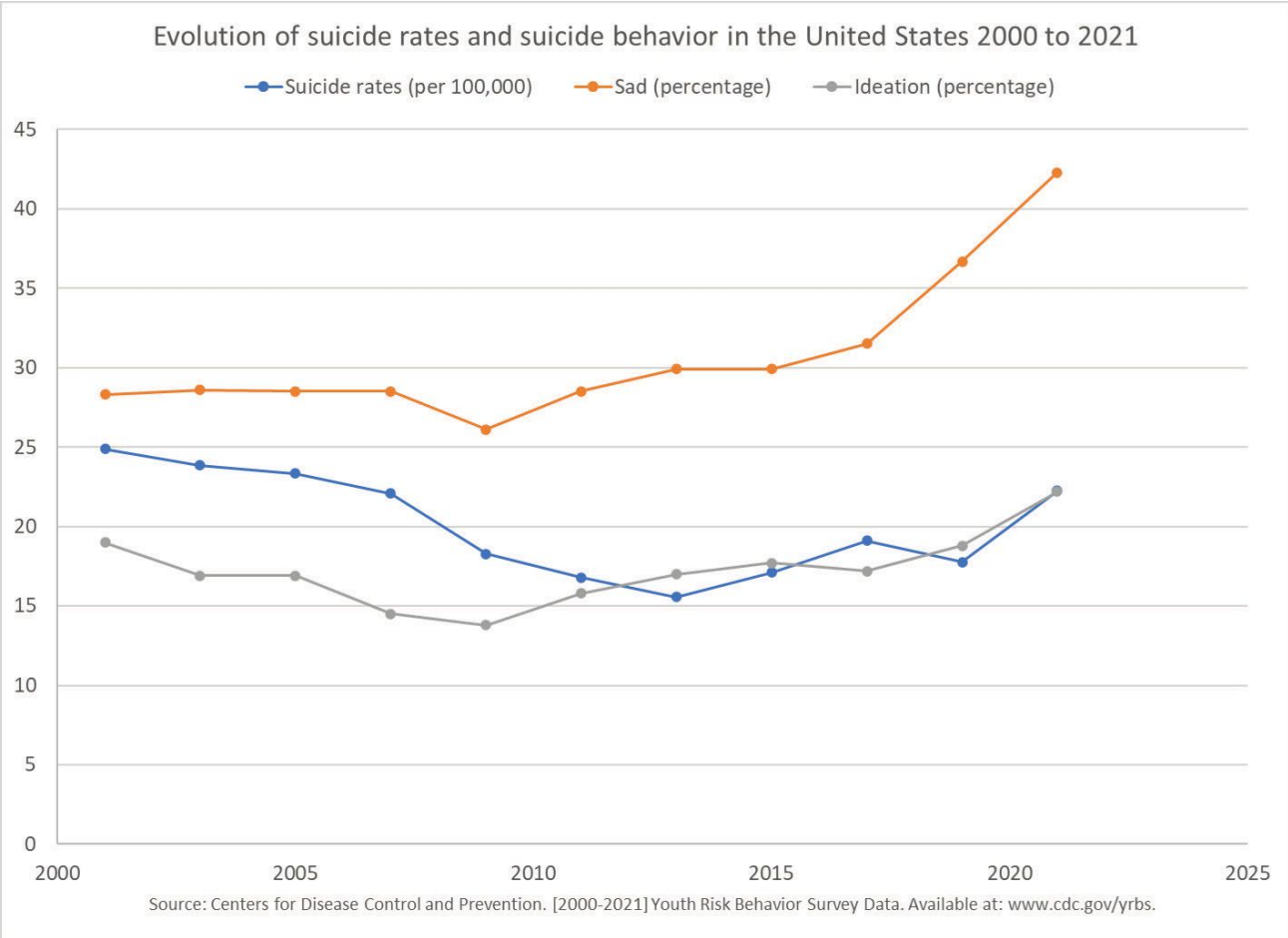
Isolation during the pandemic deprived children of these crucial learning experiences. Social isolation has also been association with numerous negative outcomes across the lifespan including depression, poor sleep quality, impaired executive function, accelerated cognitive decline, unfavorable cardiovascular function, impaired immunity, altered hypothalamic pituitary-adrenocortical activity, a pro-inflammatory gene expression profile, and earlier mortality.[3]

Furthermore, the timing of the pandemic came about just as studies started to link evidence between increased screen time with negative developmental and psychological outcomes. While technology can be a valuable tool for staying connected, excessive screen time can have adverse effects on social and psychological development and has been shown to be associated with lower psychological well-being, including less curiosity, lower self-control, more distractibility, more difficulty making friends, less emotional stability, being more difficult to care for, and inability to finish tasks.[4] It is essential to strike a balance between the use of technology for socialization on one hand, and in-person interaction on the other.

Scholastic Challenges: The Impact on Learning

One of the most visible impacts of COVID isolation and school closures is the disruption of education. The abrupt shift to remote learning challenged students, parents, and educators alike. It exposed the digital divide, where not all students had access to the necessary technology and internet connectivity for effective online learning. This digital divide disproportionately affected vulnerable and marginalized communities, exacerbating educational inequalities.[5]

Studies have shown that prolonged school closures can lead to learning loss, particularly in core subjects such as math and reading.[6] Examining US household search activity for online learning resources during the pandemic closures revealed that areas of the country with higher income, better internet access and fewer rural schools saw substantially larger increases in search intensity.[7]



One aspect to consider is the link between remote learning and academic performance. Many students have struggled to adapt to remote learning, and the impact on academic achievement has been disproportionately higher among minority students, students in low-income communities, and children who speak English as a second language. Furthermore, math learning scores are more likely to be affected than language and arts scores by pandemic changes.[8]

The disruption of traditional learning environments has also had an impact on students' motivation and engagement. Surveys among college students have documented a significant decrease in motivation in the context of online educational options during the pandemic.[9,10] This decreased motivation could have long-term consequences on educational attainment and career prospects.

COVID Isolation and Youth Suicide: A Disturbing Connection

Perhaps one of the most alarming consequences of COVID isolation among young people is the potential link to increased suicide rates. The isolation, stress and uncertainty brought about by the pandemic have created a perfect storm for mental health crises, and suicide is an extreme manifestation of these challenges.

Data from the first year of the pandemic showed that male youth, aged 5 to 12 years, and 18 to 24 years, non-Hispanic AI/AN youth, Black youth, and youth who died by firearms, experienced significantly more suicides than expected.[11] Further analyses of the suicide cases show that youth who were impacted by COVID experienced significantly more isolation, school problems, depression, and anxiety disorder diagnoses.[12]

These trends are deeply concerning and require urgent attention from policymakers, healthcare providers, and educators.

Cannabis Use and Youth Mental Health: A Complex Relationship

The role of cannabis use in youth mental health during COVID isolation is a complex and controversial topic. Cannabis is one of the most commonly used psychoactive substances among young people, and its potential role in exacerbating mental health issues should not be ignored.[13]

While some studies have suggested a link between cannabis use and an increased risk of mental health problems, including depression and anxiety,[13] the relationship between cannabis use and suicide is less clear. It is wise to approach this topic with caution and rely on evidence-based research. A large population-based Finnish study found an association between youth cannabis use and self-harm later in life but not with suicide.[14] A US epidemiological survey of adolescents found cannabis use associated with higher odds of depression and depression severity, and depression and cannabis use was independently associated with higher odds of suicide attempt.[15] Another large study looking at youth ages 10 to 24 with mood disorders found that cannabis use is a risk marker for self-harm, all-cause mortality, death by unintentional overdose and homicide.[16]

Finally, a meta-analysis of studies looking at adolescent (18 and younger) cannabis use and young adult (18 to 34) mental health outcomes concluded with a warning that high prevalence of adolescents consuming cannabis generates a large number of young people who could develop depression and suicidality attributable to cannabis.[17] These results should alert us to the possibility of long term consequences of adolescent cannabis use.

The Importance of a Holistic Approach to Youth Mental Health

Addressing the multifaceted impact of COVID isolation and school closures on youth mental health requires a multiprong approach. Policymakers, educators, parents, and healthcare providers must collaborate to provide comprehensive support to mitigate the negative consequences of these many factors.

1. Mental Health Services

Access to mental health services must be prioritized. Schools can play a crucial role in identifying and addressing mental health issues among students, but parents, providers and friends may all need to play a part. Telehealth services can also improve access to mental health support, particularly in underserved communities and studies have shown good acceptability both by parents[18] and youth, but reliable and well distributed technology infrastructure is not present.[19] A complicating factor is the shortage of mental health care providers. Recognizing this issue, the American Board of Pediatrics and the American Psychiatric Association, among others, have called for policies to increase the number of physicians 'in training' pursuing fields focusing on youth mental health. The shortage of providers, however, is an enormous problem not likely to be solved solely with an increase in psychiatrists, and other strategies are being put forth. These include shifting the skill sets of today's pediatricians, adding more focus and attention to mental health topics, rotations, lectures, and skills that they learn during their residency training years, so that they are better equipped to handle these issues in their primary care offices. Downstream changes in compensation, workflow and career satisfaction, however, may influence these plans.

2. Socialization Opportunities

Schools should prioritize opportunities for socialization and emotional development. Social and emotional learning (SEL) programs can help students develop essential skills for building healthy relationships and address social withdrawal and anxiety.[20]

3. Academic Support

Academic support should be augmented, and interventions should be implemented to address learning gaps caused by school closures. Individualized learning plans, small group learning sessions, and tutoring programs can be effective in helping students catch up. This is particularly important for low-income students but may be more challenging in school districts with budgetary restraints.

4. Digital Equity

Efforts to bridge the digital divide are essential to ensure all students have access to online learning

resources. This includes providing devices and internet connectivity to underserved communities.[21]

5. Substance Use Prevention

Substance use prevention programs should be integrated into school curricula. It is essential to educate students about the risks associated with cannabis and other drugs and to provide resources for those struggling with substance use. One example of such a program is the School-based Prevention Intervention and Resources Initiative (SPIRIT) supported by the Missouri Department of Mental Health's (DMH) Division of Behavioral Health (DBH).

6. Community Engagement

Community-based organizations and local governments can play a vital role in providing extracurricular activities and support services for young people. These programs can help combat social isolation along with providing a sense of belonging.[22] Expansion of such services with an increased emphasis on participation may yield tremendous benefits to our youth.

Conclusion

The tsunami of bad news following the years of COVID isolation and school closures on the mental health of young people is a call to action. The mental health challenges, declining socialization skills, academic setbacks, and increased suicide rates are alarming trends that demand our collective attention and response. While this paper does not provide new scientific research, it highlights the existing body of knowledge and emphasizes the need for evidence-based interventions and comprehensive support for young people.

Addressing these challenges requires a collaborative effort from policymakers, educators, healthcare providers, parents, and communities. It is crucial to prioritize the mental and emotional well-being of young people, not only during the pandemic but also in its aftermath. By taking proactive measures to support our youth, we can mitigate the long-term consequences of this crisis and build a more resilient and mentally healthy generation to inherit a better and happier world.

References

1. Loades, M. E. et al. Rapid Systematic Review: The Impact of Social Isolation and Loneliness on the Mental Health of Children and Adolescents in the Context of COVID-19. J. Am. Acad. Child Adolesc. Psychiatry 59, 1218–1239.e3 (2020).
2. Pierce, M. et al. Mental health before and during the COVID-19 pandemic: a longitudinal probability sample survey of the UK population. Lancet Psychiatry 7, 883–892 (2020).
3. Hawkley, L. C. & Capitanio, J. P. Perceived social isolation, evolutionary fitness and health outcomes: a lifespan approach. Philos. Trans. R. Soc. Lond. B Biol. Sci. 370, (2015).
4. Twenge, J. M. & Campbell, W. K. Associations between screen time and lower psychological well-being among children and adolescents: Evidence from a population-based study. Prev Med Rep 12, 271–283 (2018).
5. Haderlein, S. K. et al. Disparities in Educational Access in the Time of COVID: Evidence From a Nationally Representative Panel of American Families. AERA Open 7, 23328584211041350 (2021).
6. Kuhfeld, M. et al. Projecting the Potential Impact of COVID-19 School Closures on Academic Achievement. Educ. Res. 49, 549–565 (2020).
7. Bacher-Hicks, A., Goodman, J. & Mulhern, C. Inequality in household adaptation to schooling shocks: Covid-induced online learning engagement in real time. http://www.nber.org/papers/w27555.pdf (2020) doi:10.3386/w27555.
8. Hoofman, J. & Secord, E. The Effect of COVID-19 on Education. Pediatr. Clin. North Am. 68, 1071–1079 (2021).
9. Means, B., Neisler, J. & Others. Suddenly online: A national survey of undergraduates during the COVID-19 pandemic. https://digitalpromise.dspacedirect.org/handle/20.500.12265/98 (2020).
10. Corpus, J. H., Robinson, K. A. & Liu, Z. Comparing college students' motivation trajectories before and during COVID-19: A Self-Determination Theory approach. Front. Educ. 7, (2022).
11. Bridge, J. A. et al. Youth Suicide During the First Year of the COVID-19 Pandemic. Pediatrics 151, (2023).
12. Schnitzer, P. G., Dykstra, H. & Collier, A. The COVID-19 Pandemic and Youth Suicide: 2020-2021. Pediatrics 151, (2023).
13. Volkow, N. D., Baler, R. D., Compton, W. M. & Weiss, S. R. B. Adverse health effects of marijuana use. N. Engl. J. Med. 370, 2219–2227 (2014).
14. Denissoff, A. et al. Does cannabis use in adolescence predict self-harm or suicide? Results from a Finnish Birth Cohort Study. Acta Psychiatr. Scand. 145, 234–243 (2022).
15. Hinckley, J. D. et al. Cannabis Use Is Associated With Depression Severity and Suicidality in the National Comorbidity Survey-Adolescent Supplement. JAACAP Open 1, 24–35 (2023).
16. Fontanella, C. A. et al. Association of Cannabis Use With Self-harm and Mortality Risk Among Youths With Mood Disorders. JAMA Pediatr. 175, 377–384 (2021).
17. Gobbi, G. et al. Association of Cannabis Use in Adolescence and Risk of Depression, Anxiety, and Suicidality in Young Adulthood: A Systematic Review and Meta-analysis. JAMA Psychiatry 76, 426–434 (2019).
18. Olateju, A., Cervantes, M., Dowshen, N., Kuhns, L. M. & Dhar, C. P. Acceptability of Telemedicine Among Parents of Adolescent Patients in an Adolescent Clinic: Cross-sectional Survey Study. JMIR Pediatr Parent 5, e39704 (2022).
19. Hawke, L. D., Sheikhan, N. Y., MacCon, K. & Henderson, J. Going virtual: youth attitudes toward and experiences of virtual mental health and substance use services during the COVID-19 pandemic. BMC Health Serv. Res. 21, 340 (2021).
20. Sousa, V., Silva, P. R., Romão, A. M. & Coelho, V. A. Can an Universal School-Based Social Emotional Learning Program Reduce Adolescents' Social Withdrawal and Social Anxiety? J. Youth Adolesc. (2023) doi:10.1007/s10964-023-01840-4.
21. Gottschalk, F. & Weise, C. Digital equity and inclusion in education. https://www.oecd-ilibrary.org/content/paper/7cb15030-en (2023) doi:10.1787/7cb15030-en.
22. Oberle, E., Ji, X. R., Guhn, M., Schonert-Reichl, K. A. & Gadermann, A. M. Benefits of Extracurricular Participation in Early Adolescence: Associations with Peer Belonging and Mental Health. J. Youth Adolesc. 48, 2255–2270 (2019).

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Locum Tenens: An Evolving Paradigm of Care

by Randy Jotte, MD, Gary Gaddis MD, Larry Lewis, MD & Evan Schwarz, MD

Introduction

Locum tenens physician employment arrangements have their supporters and their detractors. Some deride them as a potential "error-producing condition,"[1] yet others laud them as an option to reduce or ameliorate physician "burnout," low patient satisfaction, and disparities in health care access.[2]

Many views are anecdotal or provided by *locum tenens* staffing companies. Empirical literature is limited and often methodologically weak.[3]



Randy Jotte, MD, (pictured) and **Larry Lewis, MD**, are Professors of Emergency Medicine and are at Washington University School of Medicine in St. Louis, Missouri. **Gary Gaddis, MD**, is Professor of Biomedical and Health Informatics, University of Missouri-Kansas City School of Medicine and Volunteer Clinical Professor of Emergency Medicine, University of California-Irvine School of Medicine. Evan Schwarz, MD, is an Associate Professor of Emergency Medicine at the David Geffen School of Medicine, UCLA, Los Angeles California.

Nonetheless, consideration of the seismic shifts in the delivery of healthcare in the last quarter-century in the US requires recognition of the substantial and increasing role that *locum tenens* physician coverage plays in both community and hospital-based healthcare today. Those considering a *locum tenens* experience during their medical career might benefit from the advice of Malcolm Gladwell: "Truly successful decision-making relies on a balance between deliberate and instinctive thinking." Whether you are considering hiring *locum tenens* physicians within your practice or seeking such employment yourself, what follows aspires to assist you as you make your deliberations.

Historical Context

Merriam-Webster defines *locum tenens* as "one filling an office for a time or temporarily taking the place of another."[4] *Locum tenens* became a healthcare delivery model in the 1970s initially to address rural physician burnout in Utah. With funding from the Robert Wood Johnson Foundation, the Health System Research Institute at the University of Utah College of Medicine created a network of physicians who covered rural practices while the rural physicians accessed

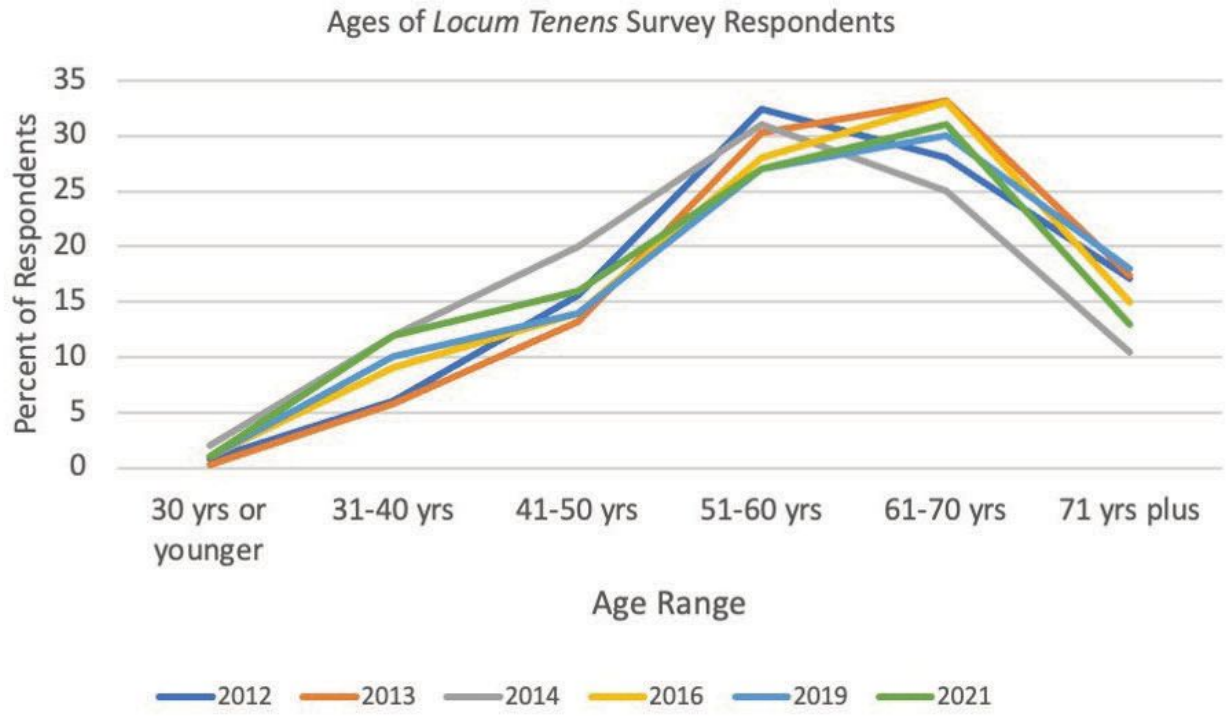


Figure 1. Age Ranges of *Locum Tenens* Physicians: 2012-2021[11, 12]

continuing education. One of the two physician founders of *locum tenens*, Alan Kronhaus, recounted that the scheduling of his *locum tenens* shifts allowed him to nurture mutual passions of patient care and skiing, with six-month stints of each.[5]

While features of the original model persist, *locum tenens* as a healthcare delivery model has grown and advanced dramatically in ensuing decades. The *Wall Street Journal* reported in 2023 that 50,000 doctors, or 7% of the US physician workforce not including foreign medical-school graduates, currently practice medicine via temporary assignments.[6] Nearly 10,000 physician *locum tenens* jobs are listed with one agency alone, with specialties ranging from family practice, emergency medicine and hospital medicine to transplant surgery, interventional radiology and hospice and palliative care.[7] As reported by Staffing Industry Analysts in "Largest Healthcare Staffing Firms in the US: 2019 Update," 49 staffing firms generated at least $50 million each in US healthcare staffing revenue in 2018. Together, these firms generated an estimated $12.7 billion in revenue and comprised 75% of the market.[8]

Characteristics of the *Locum Tenens* Workforce

A 2017 survey of 897 *locum tenens* physicians was performed by Staff Care for the purpose of characterizing their physician workforce. Approximately 60% were medical specialists.[9] Close to half, 48%, were 61 years or older and three-quarters were 51 years or older. Sixty-five percent had been in practice for at least 21 years. Nine of ten physicians surveyed had worked in permanent practice at some point during their careers. As illustrated in Figure 1, the age ranges of *locum tenens* physicians responding to a staffing company survey has varied little over the last decade. Approximately half of those surveyed reported that they began working as a *locum tenens* physician during the middle of their careers, while 36% started taking *locum tenens* positions after retiring from permanent practice. The most common factors influencing respondents' decisions about which *locum tenens* positions to take were location (89%), pay rate (67%), length of assignment (60%), and patient load (38%).[10] See Figure 1.[11,12]

Formed as a response to standardize *locum tenens* services, the National Association of Locum Tenens Organizations, NALTO, was established in 2001 to

both create and enforce strong industry standards for the profession, stressing honesty, objectivity, integrity, and competency. Best practice guidelines primarily address contractual expectations, dispute resolution, and documentation of communications rather than delving into potentially controversial matters regarding clinical practice. As NALTO's Board of Directors are affiliated primarily with staffing organizations and lack physician representation, development of organizations promoting the interests and perspectives of *locum tenens* physicians is warranted.[13]

Quality of *Locum Tenens* Care

Few studies exist to support or refute the quality of care delivered by physicians working in *locum tenens* positions. Some have attributed prescribing errors to *locum tenens* staff,[14] ascribed in part to a lack of familiarity among *locum tenens* physicians with local teams, processes, practices, and guidelines. In England, *locum tenens* physicians are more likely to be the subject of complaints, more likely to have those complaints subsequently investigated and more likely to receive sanctions, although the reasons behind such complaints are neither well-documented nor understood.[15] The General Medical Council reviewing such complaints concluded: "Doctors who act solely as *locums* or work for *locum* agencies have a greater risk of being complained about, and complaints about them are more likely to reach the threshold for a full investigation. A wider discussion is essential to understand the root causes of this and to explore whether further professional and workplace support for *locums* can reduce the risk to patients."[16]

An assessment of the quality and cost of neurosurgical care between *locum tenens* and non-*locum tenens* neurosurgeons in the US reviewed 2005-2011 Medicare claims data and found no difference in short-term complication rates, lengths of stay, or costs between *locum tenens* and non-*locum tenens* neurosurgeons.[17]

A 2020 prospective cohort study compared treatment by 22 employed hospitalists and 24 *locum tenens* hospitalists at Promedica Toledo Hospital in Ohio. Primary outcome was adjusted length-of-stay and secondary outcomes were hospital cost, inpatient mortality, 30-day all-cause readmission, and 30-day mortality. Patients treated by *locum tenens* hospitalists had one-day shorter length-of-stay and $1,339 decreased hospital costs but no statistically significant difference in mortality or readmissions.[18]

In another large retrospective study, researchers at Harvard University, Columbia University, UCLA and the National Bureau of Economic Research analyzed national Medicare data to characterize *locum tenens* physicians' patterns, quality and costs of care relative to non-*locum tenens* physicians. Randomizing a sample of 1,818,873 Medicare fee-for-service beneficiaries hospitalized and treated by a general internist during 2009-2014, 38,475 of them (2.1%) received care from a *locum tenens* physician. In addition, 9.3% (4,123/44,520) of general internists were temporarily covered by a *locum tenens* physician at some point. The primary outcome was 30-day mortality. Secondary outcomes included inpatient Medicare Part B spending, length-of-stay, and 30-day readmissions. In this retrospective cohort analysis, there was no significant difference in 30-day mortality among patients treated by *locum tenens* physicians compared with those treated by non–*locum tenens* physicians (8.83% vs 8.70%). Regarding secondary outcomes, patients treated by *locum tenens* physicians had significantly higher adjusted total Part B charges ($1,836 vs $1,712), longer length-of-stay, (5.64 days vs 5.21 days), and significantly lower readmission rates (22.80% vs 23.83%) than patients treated by non–*locum tenens* physicians.[19]

Based on our review of the literature, data with which to compare quality indictors between *locum tenens* and non-*locum tenens* physicians is limited and conflicting. Further, results may differ based on the specialty of care. Do results in general internal medicine translate to emergency medicine, anesthesiology, or critical care? The use of *locum tenens*, at a minimum, may impact patient outcomes, nursing and other healthcare team members' satisfaction and wellbeing, hospital financial margins, and the community mission of hospitals. Studies to better inform decisions regarding the use of *locum tenens* are needed. Aside from mortality data and cost analyses, comparison of the satisfaction of patients, nurses, other physicians, and all healthcare team members who works side-by-side with *locum tenens* physicians is warranted. Finally, additional data is needed to inform physicians as to the pros and cons of *locum tenens* work at various stages of one's career.

Pros and Cons of *Locum Tenens*

From a physician perspective, potential benefits of *locum tenens* work could include greater scheduling flexibility, reduced administrative burdens, enhanced pay, and opportunity for social and cultural experiences in multiple locales. Disadvantages include the

need to secure hospital credentials for each facility of employment and a state medical license for assignments outside one's primary state of practice. In addition, credentialling for clinical privileges at many hospitals requires a detailed chronology of prior work experiences, possibly supported by identification of professional references for care rendered. The "paper trail" of *locum tenens* positions could become progressively cumbersome depending on the number and duration of assignments.

As independent contractors, *locum tenens* physicians are responsible for filing their own taxes and securing those "fringe" benefits typically provided by an employer, including life insurance, disability, health insurance, and retirement plans.[20] Some *locum tenens* physicians choose to incorporate to reduce taxation and limit liability. While the hourly wage might be higher for *locum tenens* work, the Bureau of Labor Statistics reports that total benefits for employed workers in the ninetieth wage percentile constitute 37% of their total compensation, a factor to deliberate when considering high *locum tenens* pay sans benefits.[21] Professional liability insurance, whether claims with a tail or occurrence, traditionally is the responsibility of the *locum tenens* company. Should "tail" coverage become problematic, costs could be prohibitive. Simply acquiring retrospective "tail" coverage as an independent contractor might be unfeasible. Paid vacation, sick, personal, and holiday leave are not provided. While anecdotal, some physicians pursuing *locum tenens* work during the COVID pandemic found themselves unemployed once staffing needs were reduced, facing long stretches of unplanned and unpaid sabbatical.

Recommendation for *Locum Tenens*

If considering contracting physicians for temporary clinical coverage, *locum tenens* remains a viable option. Both the physician workforce and the corporations arranging and securing such coverage have improved substantially over the last half-century. While empiric evidence evaluating *locum tenens* is limited, two cohort studies noted above identified no significant difference in 30-day mortality between care provided by *locum tenens* and non-*locum tenens* physicians.

If contemplating ongoing work as a *locum tenens* physician, we believe the role remains best suited for those later in their career when approaching retirement, as a post-retirement clinical activity or perhaps as a sabbatical earlier in one's career. While the professional benefits secured through traditional employment ultimately can be established by an independent contractor pursuing full-time *locum tenens* work, the hassles of doing so can be substantial. In addition, the financial impact of unexpected unemployment, whether from an injury, illness, or pandemic, can be devastating. Many recognize such drawbacks. While media reports suggest "gig work" has become increasingly popular among younger physicians,[22] survey results of practicing locum physicians suggest otherwise.[23,24]

Lifestyle benefits of *locum tenens* employment certainly exist for younger physicians desiring clinical practice without an ongoing commitment to a locale, schedule, colleagues, hospital, or patient population. This must be weighed against the advantages of a less itinerant practice, which include not only the employment benefits packages noted above, but also the satisfaction of enduring relationships with patients and colleagues, a sense of building and maintaining a core clinical practice, and the opportunity to branch out in other medical pursuits such as health policy, education, and research. While most of this manuscript's authors have grayed to some degree, all readily recall early career frustrations: a sense of under-appreciation, lack of fulfillment, few options for advancement, and ultimately, self-doubt. Not limited to the practice of medicine, others have noted that "No one is immune from frustration, but the savviest people understand it's an essential part of success."[25] Most of us who have chosen the more traditional path for our medical career have successfully pursued a diversity of interests beyond our core clinical practice. Given time, opportunity eventually knocks, sometimes repeatedly.

References

1. Dean B, Schachter M, Vincent C, Barber N. Causes of prescribing errors in hospital inpatients: a prospective study. Lancet. 2002 Apr 20;359(9315):1373-8. doi: 10.1016/S0140-6736(02)08350-2. PMID: 11978334.
2. A prologue to locum tenens, locumstory.com, CHG Healthcare, accessed June 21, 2023, https://locumstory.com/story
3. Ferguson J, Walshe K. The quality and safety of locum doctors: a narrative review. J R Soc Med. 2019 Nov;112(11):462-471. doi: 10.1177/0141076819877539. PMID: 31710823; PMCID: PMC6851536.
4. "Locum tenens." *Merriam-Webster.com,* 2023. https://www.merriam-webster.com. (June 23, 2023).
5. A history of locum tenens: the origins of a new approach to healthcare. CompHealth. A CHG Company. August 13, 2017. https://comphealth.com/resources/the-origin-of-locum-tenens
6. Tarrant, Gretchen. Burned Out, Doctors Turn to Temp Work: A growing group of physicians are ditching medicine's traditional career path and hitting the road as temporary doctors-for-hire." The Wall Street Journal. [New York, NY], June 6, 2023.
7. Jobs. CompHealth. A CHG Company. Accessed June 21, 2023. https://comphealth.com/jobs/physician
8. Healthcare Staffing Report: October 10, 2019. Largest US Healthcare Staffing Firms List Includes 49 Firms: SIA. Staffing Industry Analysts. October 9, 2019. https://www2.staffingindustry.com/Editorial/Healthcare-

Staffing-Report/Archive-Healthcare-Staffing-Report/2019/Oct.-10-2019/Largest-US-healthcare-staffing-firms-list-includes-49-firms-SIA#:~:text=October%209%2C%202019&text=AMN%20Healthcare%20Services%20Inc.,the%20fourth%20quarter%20of%202017.
9. White Paper Series: Locum Tenens and the Emerging Shortage of Medical Specialists. Staff Care. An AMN Healthcare Company. Accessed June 22, 2013. https://www.staffcare.com/siteassets/resources/thought-leadership/white-paper-locum-tenens-and-emerging-shortage-of-medical-specialists.pdf
10. Blumenthal, DM, Tsugawa Y, Olenski AR, Jena AB. Substitute Doctors Are Becoming More Common. What Do We Know About their Quality of Care? Harvard Business Review. 2017
11. 2017 Survey of Temporary Physician Staffing Rents. Staff Care. An AMN Healthcare Company. Accessed June 23, 2023. https://www.staffcare.com/siteassets/resources/thought-leadership/2017-survey-temporary-physician-staffing-trends.pdf
12. 2021 Survey of Locum Tenens Physicians and Advanced Practitioners. Staff Care. An AMN Healthcare Company. Accessed June 23, 2023. https://www.amnhealthcare.com/amn-insights/physician/surveys/
13. Best Practice Guidelines. NALTO. The National Association of Locum Tenens Organizations. Accessed June 22, 2023. https://www.nalto.org/best-practice-guidelines/
14. Dean B, Schachter M, Vincent C, Barber N. Causes of prescribing errors in hospital inpatients: a prospective study. Lancet. 2002 Apr 20;359(9315):1373-8. doi: 10.1016/S0140-6736(02)08350-2. PMID: 11978334.
15. Ferguson J, Walshe K. The quality and safety of locum doctors: a narrative review. J R Soc Med. 2019 Nov;112(11):462-471. doi: 10.1177/0141076819877539. PMID: 31710823; PMCID: PMC6851536.
16. General Medical Council. What Our Data Tells Us about Locum Doctors. Manchester: General Medical Council, 2018.
17. Chiu RG, Nunna RS, Siddiqui N, Khalid SI, Behbahani M, Mehta AI. Locum Tenens Neurosurgery in the United States: A Medicare Claims Analysis of Outcomes, Complications, and Cost of Care. World Neurosurg. 2020 Oct;142:e210-e214. doi: 10.1016/j.wneu.2020.06.169. Epub 2020 Jun 27. PMID: 32599195.
18. Mustafa Ali MK, Sabha MM, Mustafa SK, Banifadel M, Ghazaleh S, Aburayyan KM, Ghanim MT, Awad MT, Gekonde DN, Ambati AR, Ramahi A, Elzanaty AM, Nesheiwat Z, Shastri PM, Al-Sarie M, McGready J. Hospitalization and Post-hospitalization Outcomes Among Teaching Internal Medicine, Employed Hospitalist, and Locum Tenens Hospitalist Services in a Tertiary Center: a Prospective Cohort Study. J Gen Intern Med. 2021 Oct;36(10):3040-3051. doi: 10.1007/s11606-020-06578-4. Epub 2021 Jan 25. PMID: 33495887; PMCID: PMC7832420.
19. Blumenthal DM, Olenski AR, Tsugawa Y, Jena AB. Association Between Treatment by Locum Tenens Internal Medicine Physicians and 30-Day Mortality Among Hospitalized Medicare Beneficiaries. *JAMA.* 2017;318(21):2119–2129. doi:10.1001/jama.2017.17925
20. The pros and cons of locum tenens. Weatherby Healthcare. A CHG Company. Accessed June 23, 2023. https://weatherbyhealthcare.com/blog/pros-and-cons-of-locum-tenens
21. Employer Costs for Employee Compensation – March 2023. Bureau of Labor Statistics. U.S. Department of Labor. June 16, 2023. https://www.bls.gov/news.release/pdf/ecec.pdf
22. Tarrant, Gretchen. Burned Out, Doctors Turn to Temp Work: A growing group of physicians are ditching medicine's traditional career path and hitting the road as temporary doctors-for-hire." The Wall Street Journal. [New York, NY], June 6, 2023.
23. 2017 Survey of Temporary Physician Staffing Rents. Staff Care. An AMN Healthcare Company. Accessed June 23, 2023. https://www.staffcare.com/siteassets/resources/thought-leadership/2017-survey-temporary-physician-staffing-trends.pdf
24. 2021 Survey of Locum Tenens Physicians and Advanced Practitioners. Staff Care. An AMN Healthcare Company. Accessed June 23, 2023. https://www.amnhealthcare.com/amn-insights/physician/surveys/
25. Blaschka, A. (September 9, 2022). 4 Sneaky Ways Frustration Can Help You make Career Progress. *Forbes.* https://www.forbes.com/sites/amyblaschka/2022/09/09/4-sneaky-ways-frustration-can-help-you-make-career-progress/?sh=6a21f972be6e

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Community Practice to Academe: A Road Less Traveled

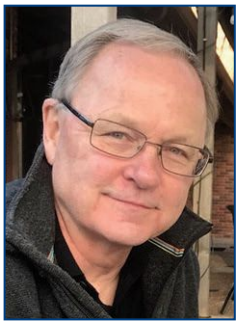
by David A. Fleming, MD

The decision was made after 20 years in private practice, but the transition began long before. The lure of academia was very strong in my year as Chief Resident, where I enjoyed opportunities to teach, publish, and stretch my wings in leadership. That year was a heady time, spawning a deep sense of professional accomplishment and contribution. For many residency programs, including internal medicine, Chief Resident status is a significant predictor of moving seamlessly into an academic career post residency.[1,2] But my professional future was preordained for a very different trajectory.

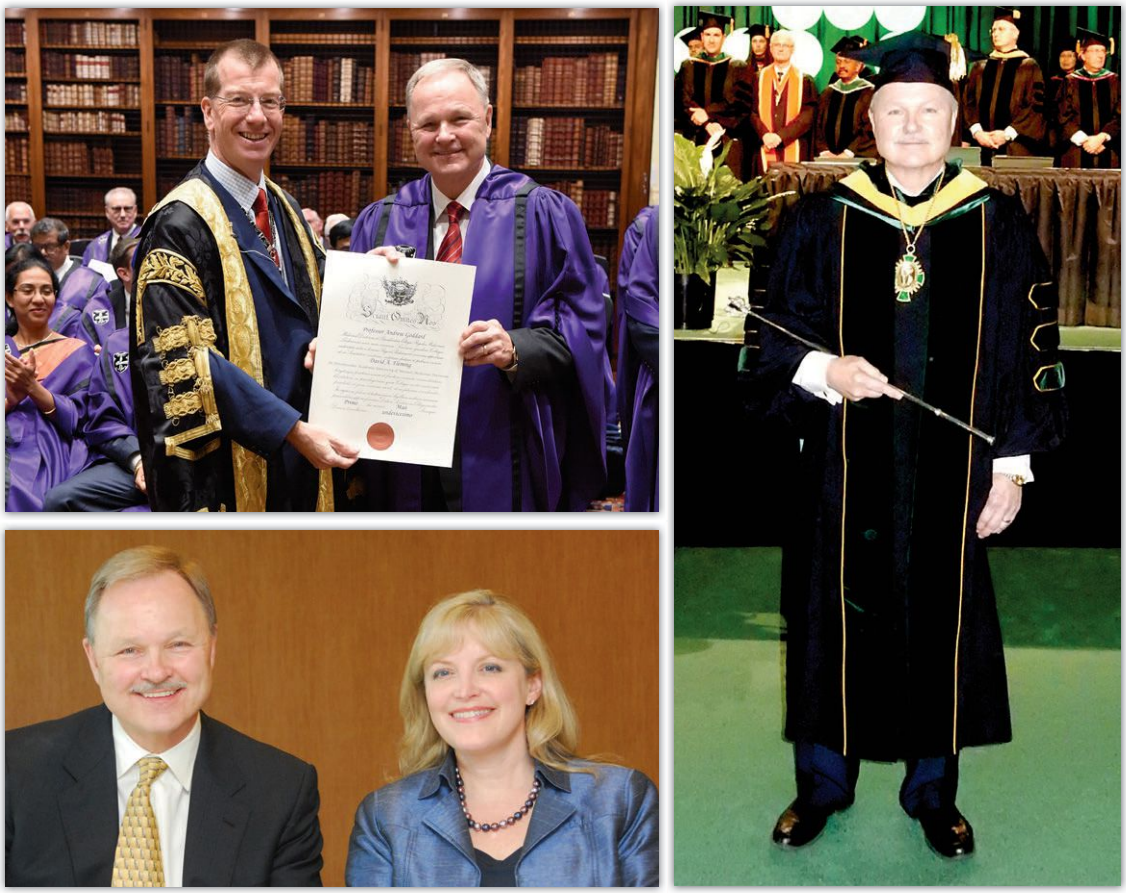
Like many, my commitment to medicine began as a young lad and that focus of intensity never wavered. Knowing the challenges of getting there, I made a personal commitment that if accepted to medical school, I would become a primary care physician and return to the hometown that had given me so much in my formative years. I grew up in a small rural community with existential challenges like many small towns, desperately striving to have and hold a good hospital staffed by good and committed health care professionals. When I returned home post residency a new hospital and clinic were being built. The future for primary and specialty care abounded with opportunities to grow and better serve that region of our state. In a few years the small internal medicine group we had formed was well intrenched, growing, and thriving. Like many small group practices, however, unintended change was inevitable.

As in many professions, physicians seek change from time to time. And for various reasons, often financial, some academic physicians chose to leave for private practice. Clinician educators are particularly vulnerable to the lure. This is often a consequence of dissatisfaction due to academic institutions demanding clinical productivity while failing to recognize and reward clinical and teaching excellence.[3] Lack of professional and advancement opportunities, salary concerns, and personal/family reasons are often cited.[4] There has also been a significant shift in recent years of physicians, both academic and private, seeking the security of contracted employment with large groups or institutions. Analysis by the American Medical Association shows that more than half of patient care physicians are now contracted with corporate entities outside of physician-owned medical practices like ours.[5]

Historically, it has seemed less likely that physicians well entrenched in private practice would transition to the rarified world of academia. Indeed, when starting out, the thought of leaving my private practice to go anywhere else was unimaginable. In recent years, however, most academic medical centers have been increasing their faculties by adding community physicians (85%).[6] This blending process has not been an easy task. When placed in the same practice plan, efforts to synergize the divergent cultures of academic and community physicians often have significant challenges, including allocation of clinical effort; education expectations (training medical students, residents and/or other clinical providers); remuneration (contracting); and to whom and how to fairly allocate academic rank and status (which promotion track and who is expected to go



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Clockwise from top left: Dr. Fleming receiving his Fellowship in the Royal College of Physicians (London); as President of the American College of Physicians; and alongside his wife, Karen Edison, MD, who is Professor and Chair Emerita of the Department of Dermatology, Founding Director of the Center for Health Policy, and Senior Medical Director for the Missouri Telehealth Network at MU.

to whose office for meetings). Of note, fewer than 50% of community physicians brought into academic medical practices are given academic titles, further widening the cultural divide.

My opportunity came 15 years into a thriving practice and, yes, there were challenges. I had maintained an adjunct faculty position with my medical school, attended weekly medicine grand rounds, and enjoyed having medical students and other learners with me on a regular basis. My patients came to expect and enjoyed having learners at the bedside. At the time we had a strong, visionary, and charismatic school of medicine dean whose goal was to develop a robust statewide network of community clinics to improve access to quality health care, enhance learning opportunities, and advance the academic and service missions of the school. This dean strove to enhance his core faculty by attracting experienced clinician educators who brought both wisdom and knowledge, and who knew the rigors and joys of true community practice, a rare commodity in the academic community. As part of the university community practice our small group maintained its community presence but with enhanced opportunities to teach and pursue other academic goals if desired. A perfect fit, or so it seemed.

The University Community Practice was a large group of like-minded colleagues and friends. We were a convivial and seasoned group of committed physicians from around the state and became the largest "department" in the medical school. But few were seen as peers in the "real" academic departments. The community physicians, though clearly impactful for patients and learners, were typically not seen as incorporate to the tightly woven academic fiber of university physicians who had paid their dues and bore the scars from climbing the ladder to attain rank and status. In later years, as the ROI waned, the university divested itself of community clinics and physicians.

Though most of my community-based colleagues ultimately parted ways I stayed on, finding opportunities to grow academically while maintaining my practice and persona as a community physician. The traditional faculty (mostly tenure track) often found my nontraditional presence disconcerting, even those who knew me well. The clinical faculty (nontenure track) primarily involved in clinical care and teaching, seemed much more welcoming and eager to share both clinical and the teaching loads, though still cautious of my intentions as an "outsider" who had yet to earn his academic spurs. It took time and effort to garner trust and open doors but open they did with most faculty. Eventually, I found greater opportunities to teach, pursue research, and assume leadership roles, both at home and nationally.

Five years into my transition I was accepted for a two-year Health and Human Services research fellowship on the East Coast. On completion, I was recruited back, this time by a new vice chancellor and dean, both equally visionary and eager to garner leadership potential and innovation. With their support I was enabled to develop an academic center that remains a mainstay of clinical and scholarly excellence. After several years and many turns in the road, professionally, I returned to my academic home feeling more comfortable in my academic skin, but still guided by core values of community practice that define and sustain me as a physician.

My life took one final turn in the last decade of my career when as Center Director and Chair of Medicine, I was elected President of the American College of Physicians. Indeed, these years were very busy and demanding, but it was in that epoch time that I sensed faculty, especially of my own department, finally closing the loop of awareness in seeing added value when community physicians such as myself become full members of the academic community.

My way has been atypical, circuitous, fulfilling, and at times heartbreaking—and was spawned not insignificantly by a healthy mixture of tenacity and luck in the choices made. But opening those doors also required the presence of rare leaders who, springing from the inevitable revolving door of academic leadership, "get it" as to the value of inclusion and acceptance. In the end, success in my case was fostered by a few open-minded individuals among the many who have little understanding of community practice, what we do, or what we have to offer.

Many years ago, as a senior resident presenting during morning report, I was called out by my chair (who a few years later joined me in private practice) for using pejorative terminology when referring to a patient's "Local Doc". To this man, one I revered and feared, I gave a firm response that I was raised by local docs and one day would be a "Local Doc" caring for patients and at times referring them to this medical center.

As I reflect on a career sequentially immersed in two professional cultures, I see similarities between the "Ivory Tower" bias that many in private practice have toward academic physicians and the "Local Doc" bias that many academic physicians have toward community physicians. Having lived and worked in both, I must admit to having similar feelings in both roles from time to time. However, I have always known in my heart that my colleagues in both tribes are knowledgeable, skilled, and caring. Most importantly we all share a universal grounding of commitment to our profession and the welfare of patients.

When planned and executed well, academic-community partnerships can lead to true integration with physicians in both cultures working together to improve the health of their community at large. For me, it's been an amazing ride and if I had it all to do over again…I wouldn't change a thing!

References

1. Jordan J, Hopson L, Clarke S, et al. Academic springboard: The chief resident position correlates with career path in emergency medicine. AEM Educ Train. 2021;5:e10639
2. Garg M, Kompala T, Hurley M, López L. Characterization of Internal Medicine Chief Resident Administrative, Educational, and Clinical Experiences. JAMA Netw Open. 2022;5(3):e223882
3. Aronoff D. And then there were none: the consequences of academia losing clinically excellent physicians. Clin Med Res. 2009;7(4):125-6
4. Girod S, Fassiotto M, Menorca R, Etzkowitz H, Wren S. Reasons for faculty departures from an academic medical center: a survey and comparison across faculty lines. BMC Med Educ. 2017;17(1):8
5. Kane, C. Recent Changes in Physician Practice Arrangements: Private Practice Dropped to Less Than 50 Percent of Physicians in 2020 [Internet]. Chicago (IL): American Medical Association: www.amaassn.org/system/files/2021-05/2020-prp-physician-practice-arrangements.pdf
6. Stamy CD, Schwartz CC, Han LP, Schwinn DA. Community and Academic Physicians Working Together in Integrated Health Care Systems. Mayo Clin Proc Innov Qual Outcomes. 2021;5(5):951-960 **MM**



MSMA: Your Oxygen Mask

by Lancer G. Gates, DO

High Flyers

Whiteman Air Force base, located in Knob Noster, Missouri, is home to the Northrup Grumman B-2 Spirit, also known as the Stealth bomber. Each day these bombers fly sorties around the globe. The B-2 Stealth bomber pilots don't take-off to high altitude flights without a co-pilot, and although they may refuel in-flight, they need to intermittently land for plane maintenance and for restorative sleep.



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The medical profession has also flown to new heights. We have immediate access to the latest medical research, the latest medical technology, and instantaneous communication with our peers. Despite these advances, we cannot stay up in this rarified air for perpetuity. We, the high-flying physicians, need to take a lesson from the high-flying pilots and intermittently get grounded for maintenance and rest.

Put on Your Own Oxygen Mask First

Before every passenger airline flight departs, the flight attendants provide passengers with a list of safety information to be utilized in the event of an in-flight emergency. One of the instructions is "put on your own oxygen mask first, before you try to help others."

The airline industry's candid approach of promoting passengers to help themselves first is honest, practical, and … a breath of fresh air.

Contrast this candid approach with the altruistic approach taken by many public health officials during the recent pandemic that mandated individuals wear masks to first and foremost protect other members of the public, rather than to first and foremost protect themselves.

Most medical students are blessed with an abundance of altruism. Following training, many physicians take their altruism to another level, aptly called "the Nurture Effect." The nurture effect is the observation that "people who take care of others on a regular basis are generally less likely to take care of themselves."[1]

It is time to acknowledge that we are not superhuman, and that we cannot take care of others if we don't take care of ourselves. We need to put on our own oxygen mask first. Our oxygen mask is our MSMA.

The air in airplanes' oxygen masks contains a mixture of gases including oxygen, nitrogen, water vapor, argon, and carbon dioxide. The air in MSMA's oxygen mask contains a mixture of advocacy and networking which when inhaled together provide humors of wellness.

Securely Fasten Your Seatbelt

Flight attendants also request that you securely fasten your seatbelt. If we are going to free ourselves from "the Nurture Effect" then we need to strap ourselves into a daily selfcare routine which includes elements of physical activity, stretching, spirituality, meditation, and nutrition. We can incorporate a few of these during our workday, e.g. taking the stairs, and eating nutritious meals; and we can do others at home with our families. If there is any time left, then we can cultivate our other pursuits.

Shut Off All Personal Electronic Devices

Many primary care physicians experience a layover of their workday as they spend time each night documenting in their patient's electronic medical records (aptly called "pajama time"). During take-offs and landings passengers accept that the airline industry doesn't tolerate airwave interference. During our workday If we have fewer interruptions then we will be in a better position to complete our medical records before we leave work, prior to the weekends, and prior to our vacations.

During our vacations and our time away from work we need to unplug; and patients, colleagues, and administrators just need to accept that we are not to be interrupted.

Take Care of Your Co-Pilots

Pilots look out for the welfare of their co-pilots. We need to apply the "Golden Rule"[2] and treat our fellow physicians the way that we would like to be treated. Following each patient encounter while the details are fresh in our minds, we should complete our documentation. This practice will improve the quality of the documentation, and provide our colleagues with timely access to our assessments and recommendations. We should avoid consulting physicians on weekends and holidays for non-emergent medical issues. We should respect our fellow physicians' vacations and days off-work by calling their office or referencing their schedule before we attempt to contact them on their personal devices. We should address as many patient issues as possible before we sign out for the weekend or leave for vacation. We should plan ahead and try our best to limit the number of patients that we handoff to our colleagues.

We should make scheduling equity our goal because shared experiences lead to camaraderie. We, the experienced physicians, should avoid the temptation of legacy scheduling, whereby we opt out of night and weekend call because we have "paid our dues." Likewise, the younger generation of physicians should understand that work-life balance isn't a concept reserved for their generation, thereby leaving the older generation of physicians to work more and longer shifts because "they are used to working all the time." Physician compensation formulas that account for any scheduling inequities due to work-life choice diversity within a physician group can significantly decrease compensation animosity.

Hospital and medical practice administrators should limit the administrative burden for their physicians. Recently the Joint Commission and Centers for Medicare and Medicaid approved that physician recredentialing in hospitals may be done every three years rather than every two years.

The MSMA advocates for a sustainable physician practice environment in Missouri. If the DEA only

requires recredentialing every three years, then why does the Missouri BNDD require recredentialing every year? Similarly, why can't the Missouri Board of Healing Arts recredential physicians every three years rather than every year? If these two Missouri government agencies were to make these changes and thereby cut their annual workload by two-thirds, they might be able to license new physicians more quickly. This would provide relief for Missouri physicians who are overwhelmed in their practice, and just waiting for new physicians to get credentialed.

MSMA also advocates for Missouri solutions to federal problems. Earlier this year Missouri passed SB 106, a $2.3M State funded GME program which increases the number of primary care residency positions in Missouri.

Rely on Your Co-Pilots & Ground Crew

Pilots entrust the lives of their passengers with their co-pilots. We entrust the care of our patients with our physician colleagues. Primary Care physicians rely on Emergency Medicine physicians to triage, evaluate, and stabilize their unstable patients. Emergency Medicine physicians rely on Hospitalists to assume the care of patients that require hospital admission. Hospitalists rely on Primary Care physicians to provide outpatient care for their patients following hospitalization. All Primary Care physicians, Emergency Medicine physicians, and Hospitalists rely on the expertise of highly trained Consultants.

If we can entrust the care of our patients with our physician colleagues, then we can surely share our professional and non-patient related practice responsibilities with them. We need to get beyond the days of giving more and more responsibilities to the same physicians because they have the requisite knowledge and experience. Professional and non-clinical responsibilities are like a glass brimming with water. If we add a drop of water to this glass, then a drop of water will surely spill out. Whenever we take on a new professional role or responsibility, we should give up one of our other roles or responsibilities. For example, if we take on a new committee assignment, then we should hand over one of our other committee assignments to a physician that is new to practice. As younger physicians take on more roles and responsibilities, they too will gain knowledge and experience, and they will grow professionally.

We also need to rely on our ground crew, our families. One of my former physician partners likened weekend hospital rounding to mowing the yard. Well, we can't be mowing our yard at home if we are mowing the yard at the hospital, so teach a family member how to mow the yard or contract it out. We should also outsource some of our other domestic responsibilities to family members. There will be a learning curve and we will need to provide grace while our family members acquire the skills that we have honed over the years.

Know Where the Exits Are Located

Flight attendants also point out the locations of all of the exits. All MSMA members are provided with a "parachute" if they are facing a personal or professional crisis. The Missouri Physicians Health Program has been operating for decades and provides professional and confidential services for our colleagues who are facing an emergency landing.

Ground Control is Clear for Take-Off

Pilots are required to log their flight hours, and these hours are limited by the federal aviation association. We should encourage our practice administrators to limit the duration of our shifts and to not assign post-call responsibilities. We should encourage our practice administrators to solicit all the physicians in our groups for their time-off requests so that they can be incorporated into the practice schedule (holiday time-off requests need to be shared equally by the physicians over the years). We should encourage our practice administrators to develop a protocol for backup physician coverage in the event of a high patient census or if a physician is unable to perform his/her shift. Finally, we should encourage our practice administrators to limit the number of meetings that require in-person physician attendance, and instead hold these meetings virtually.

Occasionally our physician colleagues will retire or transition to another professional opportunity. We should encourage our practice

administrators to replace these physicians with physicians, not with physician extenders. The passenger airline industry doesn't replace pilots with flight attendants.

Flying First Class

Fly "First Class" with MSMA and your complimentary MSMA oxygen mask. Your membership in this exclusive club costs only $450 per year for active members, $300 for physicians in their second year in practice, and $75 for retired physicians. You may elect for auto-renewal to avoid annual dues statements. Physicians in their first year of practice, fellows, residents, and students fly for free.

Upgrades

I encourage you to upgrade your MSMA experience by contributing to the Missouri Medicine Political Action Committee (MMPAC) which enhances your advocacy efforts. You may also contribute to the annual MSMA Alliance Holiday Sharing Card, a fundraiser for the Missouri State Medical Foundation which provides scholarships to students attending medical school in Missouri. Finally, you can upgrade your spouse's experience with a membership in the MSMA Alliance, a venue where they can share their challenges and joys with other physician spouses.

Enjoy Flights to Your Preferred MSMA Destinations!

Each year the MSMA holds events for its members. Please consider booking your reservations for these upcoming events: the Advocacy Training Workshop on October 14, 2023, at the Courtyard by Marriott in Jefferson City; the Physician Wellness Seminar co-hosted by MAOPS and MSMA on October 20-22, 2023, at Margaritaville, Lake of the Ozarks; our Physician Advocacy Day on March 4, 2024, at the Capitol in Jefferson City; and the MSMA Annual Convention on April 5-7, 2024, at the Renaissance Airport Hotel in St. Louis. If you don't like to travel then tune into the monthly "Fireside Chats" held every third Wednesday of every month from 7:00 p.m. - 8:00 p.m. Book your flights at www.msma.org/events. Many are free or discounted with your MSMA membership.

References

1. 78% of Houston hospital workers overweight, study says, citing "nurture effect"'. Healthcare Finance News. Accessed July 18, 2023. https://www.healthcarefinancenews.com/news/78-houston-hospital-workers-overweight-study-says-citing-nurture-effect
2. The Gospel of Matthew (7:12)

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Lessons Learned: Avoiding Risks When Using Social Media

by Michaeli Hennessy, JD, Julianne Story, JD & Pete Enko, JD

Social media is an increasingly common resource for people to learn about medical care and their health conditions. Patients can find healthcare professionals actively posting on Twitter, Facebook, Instagram, TikTok, and other social media platforms, offering their expertise, debating policies and practices, promoting patient care and education, and raising awareness of new health-related developments. With this wealth of resources readily accessible to patients, it is not surprising that a study has revealed that one in five Americans turn to TikTok before calling their own doctor.[1] To ensure that information being shared via social media is doing the general public and the practitioner good, however, attention should be paid to a few regulatory and industry recommendations, which if not followed, can lead to costly legal problems.





Julianne Story, JD (left), Labor and Employment Partner; Peter Enko, JD (center), Healthcare Regulatory & Compliance Counseling Partner; and Michaeli Hennessy, JD (right), Labor and Employment Associate, from the Kansas City office of Husch Blackwell, LLP, wrote this article. Husch Blackwell represents a full spectrum of healthcare providers and other businesses in developing compliance strategies, preliminary enforcement measures, employment concerns, and litigation matters. The information contained in this article should not be construed as legal advice or a legal opinion on any specific facts or circumstances. The contents are intended for general information purposes only, and readers are encouraged to consult their attorney concerning specific situation and specific legal questions.

Table 1. 2022 penalty chart for HIPAA violations under 45 C.F.R. 102

Penalty Tier	Level of Culpability	Minimum Penalty Per Violation	Maximum Penalty Per Violation	Annual Penalty Limit
Tier 1	Lack of Knowledge	$127	$31,987	$31,987
Tier 2	Reasonable Cause	$1,280	$63,973	$127,974
Tier 3	Willful Neglect	$12,794	$63,973	$319,865
Tier 4	Willful Neglect (not corrected within 30 days)	$63,973	$63,973	$1,919,173

Pay Attention to HIPAA Pitfalls

It should go without saying that healthcare professionals must comply with privacy and security requirements of the Health Insurance Portability and Accountability Act of 1996 ("HIPAA"),[2] as modified by the Health Information Technology for Economic and Clinical Health Act,[3] if they are using and disclosing protected health information on their social media platforms.

Unfortunately, many providers are unaware of how broadly the term "protected health information" can be construed. Under HIPAA, protected health information encompasses any individually identifiable health information, which relates to a particular patient.[4] Consequently, in addition to details of a patient's past, present or future treatment, all names, photos, and other tidbits about a patient (including things like tattoos) should be kept off a practitioner's social media unless they have first obtained a valid authorization from the patient. Among other things, such authorization must meaningfully describe the information the provider intends to use and disclose and their reasoning for doing so.[5] That said, healthcare professionals on social media should also know that HIPAA does not restrict the use or disclosure of "de-identified" health information, which "neither identifies nor provides a reasonable basis to identify an individual."[6] To de-identify information, the healthcare provider can either receive a formal determination by a statistician or, more commonly, remove all specifics related to the individual, including their family history, household members, employer information, and any other recognizable depictions.[7]

As a dental practice in Texas found out not too long ago, express patient authorization is a standard that cannot be ignored, no matter the situation.[8] In responding to a patient's negative Yelp review, the dental practice disclosed the patient's last name and details of the patient's health condition.[9] The Office of Civil Rights ("OCR") of the Department of Health and Human Services (the federal agency which oversees HIPAA) issued the dental practice a fine of $10,000 for the unconsented disclosure of the patient's protected health information.[10] Similarly, in June 2023, the OCR fined a New Jersey healthcare provider $30,000 for impermissibly disclosing patient information, including the patient's mental health diagnosis and subsequent treatment, while responding to the patient's negative online review.[11] And, in neighboring Rhode Island, the Department of Health Board of Medical Licensure and Discipline (the "Board") fined a physician $500 and required attendance at a confidentiality course for revealing personal identifiable information about her patients on Facebook.[12] According to the Board, the physician described the patients' injuries with such depth, that the patients could be identified by unauthorized third parties.[13]

These civil fines, though serious in nature, do not come close to the maximum permissible amounts. There are four tiers to the sanctions available under HIPAA, each of which ratchets up with increasing levels of culpability (Table 1).

Additionally, criminal penalties can come into play if a person knowingly obtains and discloses protected health information in violation of HIPAA's requirements.[14]

These examples underscore the importance of understanding where HIPAA protected health information begins and ends and the perils of failing to obtain appropriate authorization. To combat HIPAA risks associated with their use of social media, it is critical that healthcare providers implement policies, which discourage all types of specific patient references and spell out procedures for obtaining valid authorizations. In addition, training on what is and is not permitted under HIPAA should be rolled out across the entire practice—even to those personnel who are not typically interacting with patients and their information.

Counter the Spread of Medical Misinformation

One of the benefits and banes of social media is that it offers the rapid dissemination and consumption of information. But the on-demand nature of social media also gives rise to concerns of accuracy, credibility, and reliability—concerns which are often neglected before information is published but can lead to significant liability exposure when they are called out afterwards.

The COVID-19 pandemic magnified the problem of medical misinformation on social media. One study found that nearly eight in ten adults heard of or consumed medical misinformation related to the pandemic.[15] One chiropractor notably found himself faced with a federal lawsuit resulting from his failure to verify the accuracy of the content he promoted on social media. During the pandemic, the chiropractor marketed vitamin D and zinc products, claiming they were more effective at treating COVID than vaccines.[16] Shortly thereafter, the Federal Trade Commission sued the chiropractor for violating the COVID Consumer Protection Act of 2021.[17] Finding that the chiropractor engaged in deceptive advertising, a federal judge ordered him to pay an $80,000 fine and permanently restrained him from promoting his products as ones that prevented, treated, or reduced the severity of COVID.[18]

An important takeaway from this enforcement action is that medical misinformation has the power to negatively impact the general public, the reputations of individual practitioners and the larger healthcare community. As such, providers must be diligent in ensuring that all their healthcare-related social media posts contain scientifically accurate content.

Avoid Potential Conflicts of Interest

Many social media users have built lucrative online careers. For healthcare professionals, social media stardom and paid marketing campaigns carry risks and must be thoroughly considered and vetted.

As a current case in point, a licensed nurse practitioner specializing in dermatology gained notoriety as a social media influencer by posting skincare and beauty content.[19] Achieving popularity, the nurse practitioner began offering virtual consultations and announced a "Skincare Expert Course," which provided lessons to healthcare providers and skincare enthusiasts about acne, healthcare routines, and dermatology principles used to treat patients.[20] The nurse practitioner's employer terminated her employment, finding that she breached her employment contract by competing with the employer's business and overall "hurting the field [of dermatology] more broadly by sidestepping having to refer to a dermatologist altogether."[21]

As this situation shows, when using social media, healthcare professionals must consider not only their ethical and regulatory compliance obligations, but also the duties owed to their employers and the profession at large.

Develop and Maintain Social Media Policies

As a nursing facility in Winston-Salem recently learned the hard way, social media policies provide an effective tool to address unprofessional online conduct. The nursing facility employed a nurse who regularly posted about her job on TikTok,[22] amassing a following of more than 38,000 active viewers.[23] In one video, the nurse is seen holding a pill bottle, with an overlay caption stating, "me making sure all my patients sleep all night cause they kept me up last night."[24] Other videos included purported jokes of lying about vital signs and unplugging a patient's ventilator to charge her phone.[25] The nurse claimed that her TikTok videos were merely "just dark humor."[26] While none of the videos identified patients or disclosed any patient health information, the nursing facility nonetheless terminated the nurse's employment for violating the company's code of conduct and harming society's perception of the nursing profession.

To sidestep these lapses in behavior, healthcare employers should develop and maintain up-to-date polices that acknowledge the importance of social media, but also contain clear statements regarding the entity's culture and expectations for professionalism. Such policies should also specify the types of information that can and cannot be shared on social media and identify those who are authorized to post on behalf of an employer—making clear that employee posts reflect the personal opinions of the employee and not the employer. Finally, any social media policy should comply with the National Labor Relations Act,[27] which gives employees the right to join together to improve their working conditions. This "right to join" includes activities occurring in cyberspace, such as on Facebook, Twitter, Instagram, or even TikTok.[28] In this way, regardless of whether they are represented by a union, employees have the legal right to engage in discussion with each other about their pay, benefits, and working conditions with coworkers on social media and any policy must not have a chilling effect on such protected activities.

Once drafted, the employer should provide the social media policy to all employees, obtain employees' express acknowledgement and agreement to comply, and ensure employee understanding through robust and regular trainings.

Conclusion

Deployed wisely, social media can greatly benefit access to and delivery of healthcare by creating connections, offering on-demand information, and promoting innovation. To achieve these aims, however, physicians and their employers must manage the risks social media poses by establishing policies and training programs, which assure patient confidentiality and safety and foster professional online presences that enhance the reputations of individuals and the industry alike.

References

1. *The Shifting Role of Influence and Authority in the Rx Drug & Health Supplement Market*, charityrx (https://www.charityrx.com/blog/the-shifting-role-of-influence-and-authority-in-the-rx-drug-health-supplement-market/) (Sept. 21, 2022).
2. 42 U.S.C.A. § 101-512.
3. 42 U.S.C.A Ch. 6a, Subch. XV.
4. 45 CFR § 160.103.
5. 45 CFR § 164.508.
6. 45 CFR §§ 164.502(d), 164.514(a) and (b). *See also, Summary of the HIPAA Privacy Rule*, US. Dept. of Health and Human Services (https://www.hhs.gov/hipaa/for-professionals/privacy/laws-regulations/index.html) (Oct. 19, 2022).
7. *Id.*
8. Steve Alder, *Dental Practice Fined $10,000 for PHI Disclosures on Yelp*, The HIPAA Journal (Oct. 3, 2019).
9. *Id.*
10. *Id.*
11. Steve Alder, $30,000 Penalty for Disclosing PHI Online in Response to Negative Reviews, the HIPAA journal (June 6, 2023).
12. Molly Merrill, *"Coffee Shop Test" May Have Prevented RI Doc's Facebook Debacle*, Healthcare IT News (April 21, 2011).
13. *Id.*
14. *Summary of the HIPAA Privacy Rule*, *supra* note 6.
15. Madeline Barron, *How to Spot and Combat Health Misinformation*, American Society for Microbiology (Sept. 9, 2022).
16. Monica Obradovic¸ *St. Louis Chiropractor Fined $80K for Spreading Covid Misinformation*, RiverFront Times (Aug. 3, 2023).
17. *United States of America v. Quickwork, LLC and Eric Anthony Neptune*, No. 4:21-cv-00437 (E.D. Mo. May 5, 2021).
18. *Id.*
19. Alexandra Levine, *Doctors and Nurses Are Becoming Internet Stars. Some Are Losing Their Jobs Over It*, Forbes (Dec. 21, 2022).
20. *Id.*
21. *Id.*
22. Ford Hatchett, *Triad Nurse Says She Was Suspended Over TikTok Videos*, WXII (June 28, 2021); *see also* Leila Kozma, *A Triad Nurse Came Under Fire for Posting TikToks about Workplace Misconduct*, Distractify (June 30, 2021).
23. *Id.*
24. *Id.*
25. *Id.*
26. Amanda D'Ambrosio, *Nurse Suspended Over TikTok Videos about Patients*, MedPage Today (July 7, 2021).
27. 29 U.S.C. §§ 151-166 Suppl. 2.
28. *What's the Law: Social Media*, National Labor Relations Board (https://www.nlrb.gov/about-nlrb/rights-we-protect/the-law/employees/social-media-0).

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Future of AI in Medicine: New Opportunities & Challenges

Artificial intelligence (AI) is an ever-advancing domain with the immense capacity to revolutionize numerous facets of human life. The advancement of AI and Natural Language Processing (NLP) has given rise to innovative tools that can aid in this endeavor. Among them is ChatGPT, developed by OpenAI, Inc. in California, which is a large language model that generates human-like text based on given prompts. In the conversation between Dr. Hagan and ChatGPT regarding the potential role of AI in medicine, ChatGPT exhibits remarkable intellect and provides a comprehensive discussion on the advantages and drawbacks that AI technology brings to patient care and medical education.[1] This dialogue offers an insightful exploration into the promise and potential perils that come hand in hand with integrating AI into the field of medicine. ChatGPT explains how AI algorithms can improve patient care and outcome by quickly and accurately analyzing medical images, helping doctors detect diseases, and developing appropriate treatment plans with cost reduction.[1] However, alongside these benefits, during the conversation, ChatGPT doesn't overlook the concerns and dangers associated with the use of AI in medicine. It empathizes with Dr. Hagan's concern about plagiarism facilitated by AI systems, acknowledging the potential for individuals to exploit AI-generated content without detection.[1]

AI's potential to provide unique perspectives through instantaneous data retrieval from vast datasets greatly benefits researchers. Writing a research paper is a technical undertaking for students and researchers, demanding meticulous organization of substantial data and proper formatting. Large language models can generate extensive amounts of coherent and grammatically accurate text, closely emulating human writing style and language patterns. As a result, AI and NLP can rapidly produce research paper paragraphs or even entire sections that are challenging to distinguish from human-written content. It significantly saves researchers and students valuable time during the writing process and streamlines the peer review procedure. One of the main advantages brought up by ChatGPT is the potential for AI to enhance medical education and the effectiveness of AI in analyzing vast amounts of data in the research fields.[2,3] Despite significant advancements, Natural Language Processing (NLP) lacks the innate understanding of humans.[4] AI-based models are not immune to errors and can misinterpret context; these models might even fabricate entirely fictional research, complete with imaginary study subjects and fabricated statistics in extreme cases.[2,4]

AI-generated plagiarism is a significant issue. To protect intellectual property rights, legal frameworks must adapt to address AI-generated content, establishing ownership regulations and liability attribution.[2,5] AI lacks the criteria for traditional authorship, such as making substantial contributions and being accountable for accuracy and integrity.[2,3] Properly acknowledging AI's role, comprehensive discussions about authorship policies without compromising human authors' credibility are urgent in the current era.[3,5] AI algorithms learn from training data, and if that data contains biases, AI can amplify and perpetuate these biases, skewing research results. Addressing bias in AI is vital and requires meticulous data selection, ongoing monitoring for biases, and using fairness-aware algorithms to mitigate bias. Natural Language Processing (NLP) systems lack human comprehension capabilities and might easily overlook crucial contextual details. To address this challenge, some counterbalancing AI detector tools have been developed to distinguish between human-generated and AI-generated text. [4] While these tools provide a certain level of mitigation, a cause for concern arises with the availability of online paraphrasing tools, which can be employed to evade detection.

AI is a powerful tool that holds enormous potential to enhance patient care, medical education, research, diagnosis, treatment, and cost reduction. To combat AI-related misconduct, it is fundamental to establish robust and transparent mechanisms to detect and address pressing issues like plagiarism, fabrication, or falsification. Human oversight and ethical guidelines are essential to ensure that AI complements rather than replaces medical professionals' expertise and intellectual rigor to ensure its responsible utilization for the betterment of healthcare.

References

1. Hagan J, ChatGPT. The Promise & Perils of Artificial Intelligence: A Conversation with ChatGPT. Mo Med. 2023.
2. Dupps WJ Jr. Artificial intelligence and academic publishing. J Cataract Refract Surg. 2023;49(7):655-656.
3. Liebrenz M, Schleifer R, Buadze A, Bhugra D, Smith A. Generating scholarly content with ChatGPT: ethical challenges for medical publishing. Lancet Digit Health. 2023;5(3):e105-e106.
4. Narayanaswamy CS. Can We Write a Research Paper Using Artificial Intelligence? 2023;81(5):524-526.
5. Brian L. Frye and Chat GPT, Should Using an AI Text Generator to Produce Academic Writing Be Plagiarism?, 33 Fordham Intell. Prop. Media & Ent. L.J. 946.

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Errata

In the July/August 2023 *Missouri Medicine*, 120:4, there are two errata.

1. In the Medical-Legal article titled: "Stop, Collaborate, and Listen: Missouri Requirements Regarding Collaboration With and Supervision of Advanced Practice Nurses" by Christina B. Moore, JD and Sydney Miller, JD, the correct wording on page 255 should be: *In other words, the physician had the duty to supervise and could not rely on inaction as a defense.*

2. In the Science of Medicine article titled: "In the Household Income and Parental Educational Level Affect Post-Operative Opioid Usage in Pediatric Orthopaedic Patients" by Michael A. Ewing MD, Lauren N. Livesay, BS, James L. Cook, DVM, PhD, Emily V. Leary, PhD, Daniel G. Hoernschemeyer, MD, and Sumit K. Gupta, MD, an author is missing from the author list and about the author information and should include *Tyler Swearingin, BS, University of Missouri - Columbia School of Medicine.* **MM**

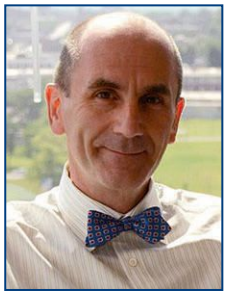
"For the Furtherance of Research" The Second Century of Biochemistry and Molecular Biology at Saint Louis University School of Medicine

by Joel C. Eissenberg, PhD, Christine K. Jacobs, MD & Enrico Di Cera, MD

On the occasion of the department's 100th anniversary, we summarize highlights of the department's first century, and then focus on a vision of the future of Biochemistry and Molecular Biology and the ways in which the department is positioning itself to help lead the future of biomedical research.

The First Century of Biochemistry and Molecular Biology at Saint Louis University

The Saint Louis University Department of Biochemistry was the brainchild of Hanau Loeb, MD, an outstanding physician and visionary Dean who understood the value of basic sciences for the future of medical research and education. He recruited Edward Doisy, MD, then a young Associate Professor in the department of Biological Chemistry at Washington University in St. Louis, to become the first chairman of the newly founded department of biochemistry at Saint Louis University. Loeb's decision was the Zeitenwende of biomedical research in the School of Medicine. Dr. Doisy rapidly established a national and international reputation for the department first with the discovery of estrogens and then with the synthesis of vitamin K that earned him the Nobel Prize in Physiology or Medicine in 1943. Notably, he patented the process for purification of conjugated estrogen from pregnant mare urine and donated the proceeds to Saint Louis University "for the furtherance of research, particularly in the Department of Biochemistry." The department was named after Dr. Doisy in 1955 in recognition of his generosity. Dr. Doisy's successor, Robert Olson, MD, continued a tradition of excellence in metabolic biochemistry while also hiring the first molecular biologists in the department. International prestige of the department grew further with William S. Sly, MD, a member of the National Academy of Sciences and the department's third chairman, who greatly expanded the department's cadre of molecular biologists. The current chair, Enrico Di Cera, MD, is a Fellow of the American Association for the Advancement of Sciences and a structural enzymologist.



Enrico Di Cera, MD, (pictured), is the Alice A. Doisy professor and Chair of the Department of Biochemistry and Molecular Biology. Joel Eissenberg, PhD, is Professor, and **Christine K. Jacobs, MD**, is Vice President of Medical Affairs and Dean of the School of Medicine. All are at Saint Louis University, St. Louis, Missouri.

Technology for the Future

Cutting edge research requires cutting edge tools and technology. The last couple of decades have seen a major investment in infrastructure to keep

"The frontiers in structural biology include mapping the entire conformational landscape, and getting away from the static view of the molecule."

- Henry Chapman, Professor of Physics, University of Hamburg

the department at the forefront of transformational research. The department houses its own full-service genomics core facility that provides investigators with whole genome analysis, supported by an in-house bioinformatics facility that offers complete analysis of high-throughput expression data, including gene ontology enrichment analysis. It also offers next-generation genomic sequence assembly with access to the department's high performance-computing cluster.

The department has established a significant footprint for structural biology in the region.

A state-of-the-art X-ray crystallographic facility houses two X-ray generators and imagers, as well as robotic instrumentation for the preparation, screening and imaging of crystals. A confocal microscope supports FLIM-FRET, Intensity FRET, and Anisotropy imaging. The department is the only location in the state that houses an Electronic Paramagnetic Resonance (EPR) Facility to measure protein dynamics and interactions at the atomic scale. A collaboration with the Washington University Center for Cellular Imaging gives faculty at Saint Louis University access to cryo-EM.

A Focus on Structural Biology

There is something powerful and elegant about structural biology. Visualizing the catalytic site of an enzyme not only unlocks the atomic secrets of the catalytic mechanism, but offers a detailed molecular target for drugs that can control that mechanism. For decades, the key to solving macromolecular structures at the atomic level was X-ray crystallography. which provides the highest resolution picture of molecules ranging from short peptides and nucleic acids up to complex macromolecular machines like the ribosome, the platform for protein biosynthesis. To date, well over 100,000 protein structures alone have been solved using X-ray crystallography. But this technology rests on the ability to form crystals of highly purified molecules, which remains something of an art.

Recently, advances in electron microscopy (EM), coupled with computational image analysis, has begun to supplant X-ray crystallography. Single particle cryo-EM avoids the need for crystallization by spreading purified molecules on a grid and imaging thousands of individual molecules in random orientations. Computer software aligns and classifies the images into consensus structures that approach the resolution of X-ray crystallographic structures without the necessity of crystal formation.

The promise of structural biology in medicine has also been greatly aided by the revolution in computer speed and memory that now permits so-called "in silico" screening of drug libraries using virtual docking algorithms that lower the cost and time to identify candidate drugs for novel targets.

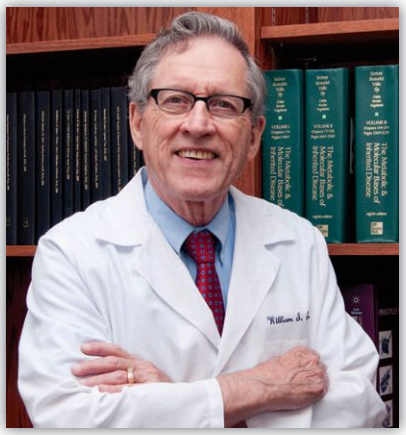
The Second Century of the Department

While space doesn't permit a summary of all the diverse and impactful research in the department, this issue of Missouri Medicine highlights several examples.

Research in the laboratory of Reza Dastvan, PhD, combines Electron Paramagnetic Resonance restraint-assisted modeling with structural studies to define the conformational rearrangements triggering extracellular release of the creatine kinase and sphingosine kinase isoforms as key therapeutic targets in various neoplastic, cardiovascular and neurodegenerative diseases. The laboratory of Kyle McCommis, PhD, focuses on mitochondrial metabolism and its role in human disease. In particular, his research explores the role of mitochondrial metabolism in heart failure and in the

Sly Syndrome Named After Doisy Chair

William S. Sly, M.D., was chair of the Edward A. Doisy Deparment of Biochemistry and Molecular Biology from 1984 until 2010. He achieved international distinction for his work on lysosomal storage diseases.



In 1973, Dr. Sly described a novel human mucopolysaccharidosis, MPS VII (also called Sly Syndrome), caused by a defect in cellular β-glucuronidase activity. He subsequently undertook a series of investigations to understand the molecular basis for this disorder. In the course of this work, Dr. Sly's group cloned the human β-glucuronidase gene, characterized the causative mutations in Sly Syndrome patients, defined a glycoprotein recognition system for trafficking of lysosomal hydrolases and cloned and characterized the human IGFII/ Mannose 6-P receptor implicated in lysosomal enzyme trafficking. The studies led to seminal discoveries on the trafficking of cellular proteins to the lysosome, including the role of mannose 6-phosphate residues in protein targeting. In recognition of his discoveries, Dr. Sly was elected to the National Academy of Sciences in 1989.

Working with collaborators at the Jackson Labs, Dr. Sly characterized the mouse model of Sly Syndrome and used this model to evaluate the effects of enzyme therapy, bone marrow transplantation and gene therapy. He devised a novel approach to overcome one of the difficulties of using animal model systems to study enzyme replacement by developing an immunotolerant transgenic mouse.

Dr. Sly's research on delivering lysosomal enzymes across the blood-brain barrier in the mouse model led to the discovery of a previously unknown mechanism for protein entry into the central nervous system. These studies opened new approaches to the challenge of delivering a therapeutic enzyme to the brain to treat various lysosomal storage diseases.

This decades-long journey culminated in 2015 in a publication reporting the first treatment for Sly syndrome, Mepsevii, developed by Ultragenyx and now approved by the FDA and in Europe. Mepsevii is a recombinant form of the β-glucuronidase enzyme designed to treat non-neurological symptoms of the disease.

role of hepatic stellate cell activation in liver fibrosis. The lab of Nicola Pozzi, PhD, studies the dynamics of proteins involved in Antiphospholipid Syndrome, such as thiol-oxidoreductases, complement and clotting factors. The laboratory of Gabriel Dai, PhD, combines cutting-edge biophysical and biochemical techniques to understand how ion channels are controlled by membrane voltages, the binding of intracellular ligands and the dynamics of cellular lipids. These mechanisms are important for neuropathic pain, epilepsy, Alzheimer's disease, and cardiac arrhythmia. The lab of Yuna Ayala, PhD, investigates the cellular function and molecular structure of RNA binding proteins, such as TDP-43, to understand basic mechanisms of RNA processing and pathogenesis of neurodegenerative disorders like amyotrophic lateral sclerosis (Lou Gehrig's disease) and frontotemporal lobar degeneration. The goal of the lab of Edwin Antony, PhD, is to build quantitative models of enzyme activity to understand how enzymes that orchestrate specific DNA repair and recombination processes recognize DNA damage and coordinate its repair, and how ATP is used by two oxidoreductases, Nitrogenase and DPOR, to orchestrate substrate reduction. **MM**

Metabolic Drivers and Rescuers of Heart Failure

by Rachel C. Weiss, Thiago N. Menezes, PhD & Kyle S. McCommis, PhD

Both preclinical animal models and human data strongly suggest a connection between altered myocardial metabolism and heart failure.



Rachel C. Weiss, Thiago N. Menezes, PhD & Kyle S. McCommis, PhD (pictured), are in the Edward A. Doisy Department of Biochemistry & Molecular Biology, Saint Louis University School of Medicine, St. Louis, Missouri.

Abstract

Cardiac hypertrophy and heart failure involve a number of metabolic alterations. Human genetic mutations and murine genetic deficiency models of metabolic enzymes or transporters largely suggest that these alterations in metabolism are maladaptive and contribute to the cardiac remodeling and dysfunction. Here, we discuss insights into metabolic alterations identified in cardiac hypertrophy and failure, as well as dietary and pharmacologic therapies that counteract these metabolic alterations and have been shown to significantly improve heart failure.

Introduction

As the end stage of virtually all forms of chronic cardiac pathologies, including hypertension, myocardial infarction, and valvular disease among others, heart failure affects an estimated 6.7 million adults in the US alone.[1] Despite existing treatments predominantly aimed at the neurohumoral systems regulating fluid volume and blood pressure, outcomes and five-year survival rates have not significantly improved in decades.[1] Better understanding of the molecular processes driving heart failure are sorely needed to develop improved therapies. Due to the metabolic alterations described below, targeting these changes in cardiac metabolism is increasingly considered an attractive therapeutic modality.

The heart requires a continuous supply of chemical energy due to its constant contractile work. While the preferred fuel source is fatty acids, the normal adult myocardium is metabolically flexible and can metabolize alternative fuels, such as glucose, lactate, amino acids, and ketone bodies in proportion to their delivery.[2] The vast majority of high-energy adenosine triphosphate (ATP) produced in the heart arises from mitochondrial oxidative metabolism, with only ~5% of ATP produced by glycolysis. The majority of cardiac ATP (60-70%) is used in myosin ATP hydrolysis to fuel contraction, while the remaining ATP is used for enzymatic reactions and various ion pumps, particularly the sarcoplasmic reticulum Ca^{2+}-ATPase pump (SERCA) to regulate cytoplasmic Ca^{2+} concentrations. Phosphocreatine (PCr) also plays an important role in buffering the high-energy phosphates in

the heart, with (PCr) concentrations ~1.5-fold higher than ATP concentrations.[3] Due to extremely high ATP hydrolysis rates and relatively low PCr and ATP pool sizes, constant ATP generation is required so that high-energy phosphate pools are not depleted within a matter of seconds.[4] The principal cardiac metabolic pathways and how they are altered in hypertrophy and heart failure are summarized in Figure 1.

Altered Metabolism in Hypertrophy and Heart Failure

Decreased Fat Oxidation and Overall Oxidative Capacity

In both human heart failure patients and animal models of hypertrophy/failure, one of the most consistent metabolic findings is a reduction in cardiac fatty acid uptake and oxidation.[5-7] This decrease in fat oxidation is due, at least in part, to a transcriptional downregulation of fat oxidation enzymes and transporters, and other mitochondrial enzymes.[6,8-10] As the heart normally relies on fat oxidation for the majority of ATP synthesis, unsurprisingly, failing human hearts contain significantly decreased ATP and PCr levels.[3] Patients with fatty acid oxidation disorders as well as mouse models of cardiac fat oxidation enzyme/transporter deficiency can develop cardiomyopathy,[11-14] suggesting that loss of fat oxidation directly contributes to heart failure pathology.

Alterations in Glycolysis and Glucose Oxidation

Changes in glucose metabolism are less consistent and may depend not only on the etiology but also the stage/severity of heart failure. In general, many studies observe increased glucose uptake and glycolysis in hypertrophy and heart failure.[4,7] Glucose oxidation can be decreased, or unchanged in the failing heart, although unchanged glucose oxidation despite increased glycolytic flux represents a prominent mismatch between glucose utilization and oxidation.[15] In relation to this mismatch, we and others recently described spontaneous heart failure development in mice with cardiac deletion of either mitochondrial pyruvate carrier (MPC) subunit.[16-18]

Glucose uptake in the heart is mediated by glucose transporters 1 and 4, and phosphorylation by hexokinase into glucose 6-phosphate (G6P) prevents the molecule from being transported back outside of

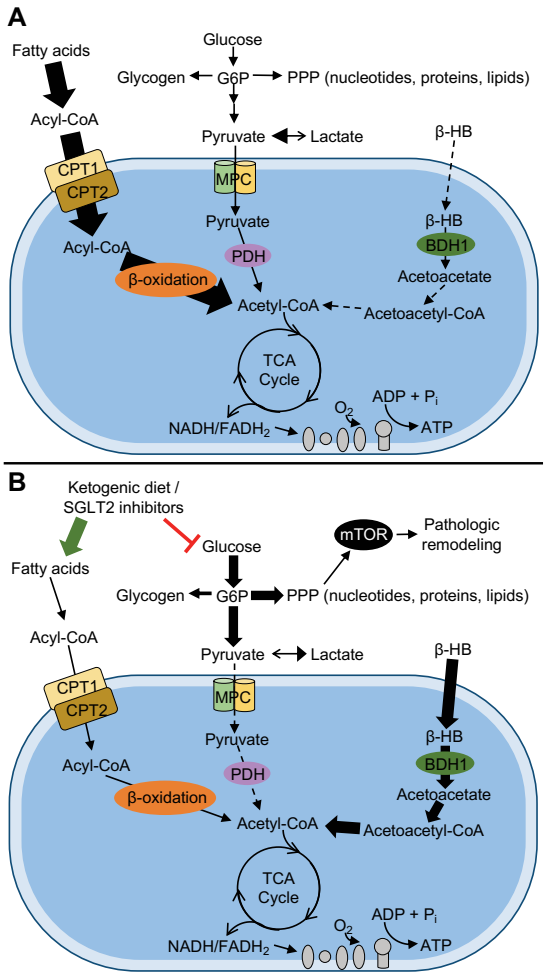


Figure 1. The principal cardiac metabolic pathways and how they are altered in hypertrophy and heart failure.

the cell. G6P can be used in glycolysis, or enter several "accessory" metabolic pathways, including conversion to glycogen or the pentose phosphate pathway.[2] More recently, studies seem to suggest that this increase in glucose utilization and flux through accessory glucose pathways is critical to providing the anabolic building blocks that drive hypertrophic growth.[19,20] For example, flux through the pentose phosphate pathway may drive the synthesis of nucleotides, proteins, and lipids required for pathologic growth of the heart. Additionally, these studies show that accumulation of G6P can lead to increased protein synthesis via activation of the mammalian target of rapamycin (mTOR), the master regulator of anabolic cellular processes.[19] Interestingly, increasing cardiac fat oxidation was sufficient to decrease glucose utilization

and improve hypertrophic protein synthesis.[20] It has also been described that these glucose utilization changes precede pathologic remodeling,[21] suggesting that preventing these metabolic changes may prevent the development of pathological hypertrophy.

Glycogen has not been extensively studied in hypertrophied/failing hearts. With increased glucose uptake and decreased glucose oxidation, one would expect glycogen stores to increase, as we have found in failing mitochondrial pyruvate carrier-deficient hearts.[16,17] Indeed, G6P is an allosteric activator of glycogen synthase, the rate-limiting step of glycogen formation.[22] Interestingly, in hypertrophied hearts, it has been suggested that glycolysis is increased from exogenous glucose, but glycogenolysis remains normal.[23] Typically, glycogen stores are very low in cardiomyocytes, and it is well-established that glycogen storage diseases are associated with cardiac hypertrophy and cardiomyopathy.[24] One potential mechanism for cardiac glycogen accumulation affecting hypertrophic growth would be that glycogen can sequester and inhibit the activation of the energy sensor adenosine monophosphate-activated protein kinase (AMPK),[25] leading to mTOR activation and anabolic growth. More work is required in this area and is one of the current research focuses of our laboratory.

Ketone Body Metabolism in Heart Failure

A number of recent studies have suggested that failing hearts increase ketone body utilization.[26,27] In the normal, mostly postprandial state, circulating ketone body concentrations are low and therefore cardiac ketone metabolism is limited. However, both animal models and human heart failure patients are known to have increased plasma ketone concentrations,[28,29] and cardiac ketone extraction and oxidation is increased.[30,31] It should be noted that the degree of ketosis associates with the extent of cardiac dysfunction and neurohumoral activation,[28,32] and therefore the degree of ketosis also insinuates increased risk of cardiovascular events and death.[33] That said, mice with cardiac deficiency of ketone oxidation enzymes display worsened heart failure,[34,35] while mice overexpressing the ketolytic enzyme β-hydroxybutyrate dehydrogenase 1 are protected against heart failure from pressure overload.[36] As failing hearts display decreased oxidation of fatty acids and glucose, and increased metabolism of ketone bodies, there is great excitement for enhancing ketone body metabolism to treat heart failure as described in more as follows.

Nutritional and Pharmacologic Therapies Targeting Altered Metabolism in Heart Failure

Ketogenic Diet and Ketone Supplements

There are two main ways to chronically elevate ketosis or blood ketone body concentrations. The first route is via consumption of a high fat—low carbohydrate "ketogenic" diet, which mimics the fasting condition and provides the liver with excess fatty acids that get converted to the ketone bodies acetoacetate and predominantly β-hydroxybutyrate. Recently, we and others successfully prevented or reversed murine heart failure from mitochondrial pyruvate carrier deficiency,[16,17] and others described improved cardiac remodeling in models of hypertension or aortic constriction pressure-overload by feeding a ketogenic diet.[34,37-39] However, it has become apparent that in the setting of a ketogenic diet (or fasting), the heart downregulates the enzymatic machinery for metabolizing ketone bodies, and significantly enhances fat oxidation.[16,17,40] Thus, there is little evidence to suggest that improvements in heart failure with a ketogenic diet are driven by ketone metabolism. In support of this, it should be noted that mice with cardiac deletion of carnitine palmitoyltransferase 2 and defective fat oxidation develop heart failure that is not rescued by a ketogenic diet.[13] Due to the severe carbohydrate restriction involved with a ketogenic diet, glycemia and insulinemia levels are decreased, likely leading to significantly reduced cardiac glucose metabolism. Therefore, in our opinion a ketogenic diet improves heart failure by both limiting flux through the glucose utilization pathways that appear to drive hypertrophic growth, as well as reinvigorating cardiac fat uptake and oxidation.

The second mechanism to increase ketosis, apart from modulating dietary fat content, involves consumption of ketone salt or ketone ester supplements commonly referred to as "nutritional ketosis". These supplements have been tested in a handful of studies, which largely suggest improvements in heart failure or cardiac function.[41-44] Acute three-hour ketone body infusions have also been shown to improve cardiac hemodynamics in human heart failure patients.[44] However, it is difficult to extrapolate findings from acute infusions to the settings of chronic ketosis. Indeed, acute ketone infusion is known to decrease cardiac glucose uptake and improve myocardial blood

flow.[45] Therefore, even though ketone supplements do not involve modulation of other dietary factors, further study is required to investigate whether improvements in heart failure are due to ketone metabolism vs other factors. Of note, a search of clinicaltrials.gov currently identifies over 50 studies investigating the effects of increased ketosis by either ketone supplements or ketogenic diet on cardiac outcomes.

SGLT2 Inhibitors

A recent revolution in heart failure pharmacotherapy involves a drug that was originally designed to decrease glycemia in diabetic subjects. Sodium glucose transporter 2 (SGLT2) inhibitors prevent the renal reabsorption of glucose and can therefore force hundreds of grams of glucose to be excreted in urine daily. One of the most profound findings from SGLT2 inhibitor trials is the striking decrease in heart failure outcomes and mortality in both diabetic and non-diabetic subjects.[46] The chronic excretion of glucose lowers glycemia and insulinemia and thus increases fatty acid utilization and enhances ketosis to a degree. However, several animal studies of heart failure suggest that the improvements with SGLT2 inhibitors are not dependent on enhanced cardiac ketone metabolism.[47-49] Conversely, SGLT2 inhibitors significantly increase myocardial fat uptake and severely decrease glucose uptake.[50] Therefore, like a ketogenic diet, SGLT2 inhibitors may improve heart failure via a combination of enhancing cardiac fat oxidation and limiting excessive glucose flux. However, SGLT2 inhibitors also produce other effects that may significantly contribute to the cardiovascular benefits, such as blood volume and pressure control due to osmotic diuresis and natriuresis, as well as inducing moderate weight loss.

Conclusion

Both preclinical animal models and human data strongly suggest a connection between altered myocardial metabolism and heart failure. A decrease in oxidative capacity, particularly of fatty acids is most likely responsible for the decrease in cardiac energetics of failing hearts. Conversely, the enhanced flux through glucose pathways that generate anabolic building blocks appear to help drive pathologic cardiac growth. Existing data from ketogenic diet as well as SGLT2 inhibitors suggests that heart failure is improved likely through a combination of enhancing cardiac fat oxidation and decreasing excessive glucose flux. More work is required to uncover the specific mechanisms of these interventions. Additionally, these dietary or pharmacologic therapies will require increased testing in different forms of heart failure, such as ischemic versus non-ischemic, systolic versus diastolic dysfunction, etc.

References

1. Tsao CW, Aday AW, Almarzooq ZI, et al. Heart Disease and Stroke Statistics-2023 Update: A Report From the American Heart Association. Circulation. Feb 21 2023;147(8):e93-e621. doi:10.1161/CIR.0000000000001123
2. De Jong KA, Lopaschuk GD. Complex Energy Metabolic Changes in Heart Failure With Preserved Ejection Fraction and Heart Failure With Reduced Ejection Fraction. Can J Cardiol. Jul 2017;33(7):860-871. doi:10.1016/j.cjca.2017.03.009
3. Beer M, Seyfarth T, Sandstede J, et al. Absolute concentrations of high-energy phosphate metabolites in normal, hypertrophied, and failing human myocardium measured noninvasively with (31)P-SLOOP magnetic resonance spectroscopy. J Am Coll Cardiol. Oct 2 2002;40(7):1267-74. doi:10.1016/s0735-1097(02)02160-5
4. Doenst T, Nguyen TD, Abel ED. Cardiac metabolism in heart failure: implications beyond ATP production. Circ Res. Aug 30 2013;113(6):709-24. doi:10.1161/CIRCRESAHA.113.300376
5. Kato T, Niizuma S, Inuzuka Y, et al. Analysis of metabolic remodeling in compensated left ventricular hypertrophy and heart failure. Circ Heart Fail. May 2010;3(3):420-30. doi:10.1161/CIRCHEARTFAILURE.109.888479
6. Osorio JC, Stanley WC, Linke A, et al. Impaired myocardial fatty acid oxidation and reduced protein expression of retinoid X receptor-alpha in pacing-induced heart failure. Circulation. Jul 30 2002;106(5):606-12. doi:10.1161/01.cir.0000023531.22727.c1
7. Davila-Roman VG, Vedala G, Herrero P, et al. Altered myocardial fatty acid and glucose metabolism in idiopathic dilated cardiomyopathy. J Am Coll Cardiol. Jul 17 2002;40(2):271-7.
8. Garnier A, Fortin D, Delomenie C, Momken I, Veksler V, Ventura-Clapier R. Depressed mitochondrial transcription factors and oxidative capacity in rat failing cardiac and skeletal muscles. J Physiol. Sep 1 2003;551(Pt 2):491-501. doi:10.1113/jphysiol.2003.045104
9. Sack MN, Rader TA, Park S, Bastin J, McCune SA, Kelly DP. Fatty acid oxidation enzyme gene expression is downregulated in the failing heart. Circulation. Dec 1 1996;94(11):2837-42. doi:10.1161/01.cir.94.11.2837
10. Barger PM, Brandt JM, Leone TC, Weinheimer CJ, Kelly DP. Deactivation of peroxisome proliferator-activated receptor-alpha during cardiac hypertrophic growth. Research Support, Non-U.S. Gov't Research Support, U.S. Gov't, P.H.S. J Clin Invest. Jun 2000;105(12):1723-30. doi:10.1172/JCI9056
11. Knottnerus SJG, Bleeker JC, Wust RCI, et al. Disorders of mitochondrial long-chain fatty acid oxidation and the carnitine shuttle. Rev Endocr Metab Disord. Mar 2018;19(1):93-106. doi:10.1007/s11154-018-9448-1
12. He L, Kim T, Long Q, et al. Carnitine palmitoyltransferase-1b deficiency aggravates pressure overload-induced cardiac hypertrophy caused by lipotoxicity. Circulation. Oct 2 2012;126(14):1705-16. doi:10.1161/CIRCULATIONAHA.111.075978
13. Pereyra AS, Hasek LY, Harris KL, et al. Loss of cardiac carnitine palmitoyltransferase 2 results in rapamycin-resistant, acetylation-independent hypertrophy. J Biol Chem. Nov 10 2017;292(45):18443-18456. doi:10.1074/jbc.M117.800839
14. Xiong D, He H, James J, et al. Cardiac-specific VLCAD deficiency induces dilated cardiomyopathy and cold intolerance. Am J Physiol

Heart Circ Physiol. Feb 2014;306(3):H326-38. doi:10.1152/ajpheart.00931.2012
15. Fillmore N, Levasseur JL, Fukushima A, et al. Uncoupling of glycolysis from glucose oxidation accompanies the development of heart failure with preserved ejection fraction. Mol Med. Mar 15 2018;24(1):3. doi:10.1186/s10020-018-0005-x
16. McCommis KS, Kovacs A, Weinheimer CJ, et al. Nutritional modulation of heart failure in mitochondrial pyruvate carrier-deficient mice. Nat Metab. Nov 2020;2(11):1232-1247. doi:10.1038/s42255-020-00296-1
17. Zhang Y, Taufalele PV, Cochran JD, et al. Mitochondrial pyruvate carriers are required for myocardial stress adaptation. Nat Metab. Nov 2020;2(11):1248-1264. doi:10.1038/s42255-020-00288-1
18. Fernandez-Caggiano M, Kamynina A, Francois AA, et al. Mitochondrial pyruvate carrier abundance mediates pathological cardiac hypertrophy. Nat Metab. Nov 2020;2(11):1223-1231. doi:10.1038/s42255-020-00276-5
19. Karlstaedt A, Khanna R, Thangam M, Taegtmeyer H. Glucose 6-Phosphate Accumulates via Phosphoglucose Isomerase Inhibition in Heart Muscle. Circ Res. Jan 3 2020;126(1):60-74. doi:10.1161/CIRCRESAHA.119.315180
20. Ritterhoff J, Young S, Villet O, et al. Metabolic Remodeling Promotes Cardiac Hypertrophy by Directing Glucose to Aspartate Biosynthesis. Circ Res. Jan 17 2020;126(2):182-196. doi:10.1161/CIRCRESAHA.119.315483
21. Davogustto GE, Salazar RL, Vasquez HG, et al. Metabolic remodeling precedes mTORC1-mediated cardiac hypertrophy. J Mol Cell Cardiol. Sep 2021;158:115-127. doi:10.1016/j.yjmcc.2021.05.016
22. Roach PJ, Depaoli-Roach AA, Hurley TD, Tagliabracci VS. Glycogen and its metabolism: some new developments and old themes. Biochem J. Feb 1 2012;441(3):763-87. doi:10.1042/BJ20111416
23. Allard MF, Henning SL, Wambolt RB, Granleese SR, English DR, Lopaschuk GD. Glycogen metabolism in the aerobic hypertrophied rat heart. Circulation. Jul 15 1997;96(2):676-82. doi:10.1161/01.cir.96.2.676
24. Arad M, Maron BJ, Gorham JM, et al. Glycogen storage diseases presenting as hypertrophic cardiomyopathy. N Engl J Med. Jan 27 2005;352(4):362-72. doi:10.1056/NEJMoa033349
25. McBride A, Ghilagaber S, Nikolaev A, Hardie DG. The glycogen-binding domain on the AMPK beta subunit allows the kinase to act as a glycogen sensor. Cell Metab. Jan 7 2009;9(1):23-34. doi:10.1016/j.cmet.2008.11.008
26. Aubert G, Martin OJ, Horton JL, et al. The Failing Heart Relies on Ketone Bodies as a Fuel. Circulation. Jan 27 2016;133(8):698-705. doi:10.1161/CIRCULATIONAHA.115.017355
27. Bedi KC, Jr., Snyder NW, Brandimarto J, et al. Evidence for Intramyocardial Disruption of Lipid Metabolism and Increased Myocardial Ketone Utilization in Advanced Human Heart Failure. Circulation. Feb 23 2016;133(8):706-16. doi:10.1161/CIRCULATIONAHA.115.017545
28. Lommi J, Kupari M, Koskinen P, et al. Blood ketone bodies in congestive heart failure. J Am Coll Cardiol. Sep 1996;28(3):665-72. doi:10.1016/0735-1097(96)00214-8
29. Lommi J, Koskinen P, Naveri H, Harkonen M, Kupari M. Heart failure ketosis. J Intern Med. Sep 1997;242(3):231-8.
30. Monzo L, Sedlacek K, Hromanikova K, et al. Myocardial ketone body utilization in patients with heart failure: The impact of oral ketone ester. Metabolism. Nov 26 2020;115:154452. doi:10.1016/j.metabol.2020.154452
31. Murashige D, Jang C, Neinast M, et al. Comprehensive quantification of fuel use by the failing and nonfailing human heart. Science. Oct 16 2020;370(6514):364-368. doi:10.1126/science.abc8861
32. Marcondes-Braga FG, Gutz IGR, Batista GL, et al. Exhaled acetone as a new biomaker of heart failure severity. Chest. Aug 2012;142(2):457-466. doi:10.1378/chest.11-2892
33. Shemesh E, Chevli PA, Islam T, et al. Circulating ketone bodies and cardiovascular outcomes: the MESA study. Eur Heart J. Mar 6 2023;doi:10.1093/eurheartj/ehad087
34. Horton JL, Davidson MT, Kurishima C, et al. The failing heart utilizes 3-hydroxybutyrate as a metabolic stress defense. JCI Insight. Feb 21 2019;4(4):e124079. doi:10.1172/jci.insight.124079
35. Schugar RC, Moll AR, Andre d'Avignon D, Weinheimer CJ, Kovacs A, Crawford PA. Cardiomyocyte-specific deficiency of ketone body metabolism promotes accelerated pathological remodeling. Mol Metab. Oct 2014;3(7):754-69. doi:10.1016/j.molmet.2014.07.010
36. Uchihashi M, Hoshino A, Okawa Y, et al. Cardiac-Specific Bdh1 Overexpression Ameliorates Oxidative Stress and Cardiac Remodeling in Pressure Overload-Induced Heart Failure. Circ Heart Fail. Dec 2017;10(12)doi:10.1161/CIRCHEARTFAILURE.117.004417
37. Sharma N, Okere IC, Barrows BR, et al. High-sugar diets increase cardiac dysfunction and mortality in hypertension compared to low-carbohydrate or high-starch diets. J Hypertens. Jul 2008;26(7):1402-10. doi:10.1097/HJH.0b013e3283007dda
38. Duda MK, O'Shea KM, Lei B, et al. Low-carbohydrate/high-fat diet attenuates pressure overload-induced ventricular remodeling and dysfunction. J Card Fail. May 2008;14(4):327-35. doi:10.1016/j.cardfail.2007.11.003
39. Okere IC, Young ME, McElfresh TA, et al. Low carbohydrate/high-fat diet attenuates cardiac hypertrophy, remodeling, and altered gene expression in hypertension. Hypertension. Dec 2006;48(6):1116-23. doi:10.1161/01.HYP.0000248430.26229.0f
40. Wentz AE, d'Avignon DA, Weber ML, et al. Adaptation of myocardial substrate metabolism to a ketogenic nutrient environment. J Biol Chem. Aug 6 2010;285(32):24447-56. doi:10.1074/jbc.M110.100651
41. Yurista SR, Matsuura TR, Sillje HHW, et al. Ketone Ester Treatment Improves Cardiac Function and Reduces Pathologic Remodeling in Preclinical Models of Heart Failure. Circ Heart Fail. Dec 28 2020;14(1):e007684. doi:10.1161/CIRCHEARTFAILURE.120.007684
42. Takahara S, Soni S, Phaterpekar K, et al. Chronic exogenous ketone supplementation blunts the decline of cardiac function in the failing heart. ESC Heart Fail. Dec 2021;8(6):5606-5612. doi:10.1002/ehf2.13634
43. Thai PN, Miller CV, King MT, et al. Ketone Ester D-beta-Hydroxybutyrate-(R)-1,3 Butanediol Prevents Decline in Cardiac Function in Type 2 Diabetic Mice. J Am Heart Assoc. Oct 5 2021;10(19):e020729. doi:10.1161/JAHA.120.020729
44. Nielsen R, Moller N, Gormsen LC, et al. Cardiovascular Effects of Treatment With the Ketone Body 3-Hydroxybutyrate in Chronic Heart Failure Patients. Circulation. Apr 30 2019;139(18):2129-2141. doi:10.1161/CIRCULATIONAHA.118.036459
45. Gormsen LC, Svart M, Thomsen HH, et al. Ketone Body Infusion With 3-Hydroxybutyrate Reduces Myocardial Glucose Uptake and Increases Blood Flow in Humans: A Positron Emission Tomography Study. J Am Heart Assoc. Feb 27 2017;6(3)doi:10.1161/JAHA.116.005066
46. McMurray JJV, Solomon SD, Inzucchi SE, et al. Dapagliflozin in Patients with Heart Failure and Reduced Ejection Fraction. N Engl J Med. Nov 21 2019;381(21):1995-2008. doi:10.1056/NEJMoa1911303
47. Verma S, Rawat S, Ho KL, et al. Empagliflozin Increases Cardiac Energy Production in Diabetes: Novel Translational Insights Into the Heart Failure Benefits of SGLT2 Inhibitors. JACC Basic Transl Sci. Oct 2018;3(5):575-587. doi:10.1016/j.jacbts.2018.07.006
48. Byrne NJ, Parajuli N, Levasseur JL, et al. Empagliflozin Prevents Worsening of Cardiac Function in an Experimental Model of Pressure Overload-Induced Heart Failure. JACC Basic Transl Sci. Aug 2017;2(4):347-354. doi:10.1016/j.jacbts.2017.07.003
49. Abdurrachim D, Teo XQ, Woo CC, et al. Empagliflozin reduces myocardial ketone utilization while preserving glucose utilization in diabetic hypertensive heart disease: A hyperpolarized (13) C magnetic resonance spectroscopy study. Diabetes Obes Metab. Feb 2019;21(2):357-365. doi:10.1111/dom.13536
50. Santos-Gallego CG, Requena-Ibanez JA, San Antonio R, et al. Empagliflozin Ameliorates Adverse Left Ventricular Remodeling in Nondiabetic Heart Failure by Enhancing Myocardial Energetics. J Am Coll Cardiol. Apr 23 2019;73(15):1931-1944. doi:10.1016/j.jacc.2019.01.056

MM

Forecasting the Future of Antiphospholipid Syndrome: Prospects and Challenges

by Aberdeen Taylor, Suresh Kumar, PhD & Nicola Pozzi, PhD

Antiphospholipid syndrome is a life-threatening autoimmune disease resulting in thrombosis of arteries and veins in young patients and obstetric complications such as miscarriage, stillbirth, and preeclampsia.



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Abstract

Antiphospholipid syndrome (APS) is an autoimmune condition affecting young patients, primarily women, negatively impacting their quality of life. APS is under-recognized and underdiagnosed and can have devastating results if untreated, mainly due to uncontrolled thrombosis. Research in the past decades has led to several breakthroughs with important implications for clinical practice. Here, we summarize the state of APS diagnosis, treatment, pathophysiology, and research directions that hold promise for advancing diagnosis and treatment.

The Current Atate of APS Diagnosis and Treatment

Definition and Epidemiology

Antiphospholipid syndrome (APS) is a life-threatening autoimmune disease defined by the presence of antiphospholipid antibodies, correlating with a hypercoagulable state that often manifests as thrombosis of arteries and veins in young patients, including obstetric complications such as miscarriage. It is associated with a 50-80% increased mortality compared with the general population, with the mean age of death in one study being 59 years.[1]

The prevalence of APS in the United States is estimated to be 40-50 in 100,000, while the incidence of new diagnoses is approximately two cases per 100,000.[2] While APS can occur as a primary disease, most APS patients concurrently carry other autoimmune diseases like Systemic Lupus Erythematosus (SLE), which is implicated in approximately 35% of cases.[1,2] Thus, APS patients are generally referred to and cared for by rheumatologists, although hematologists and obstetricians/gynecologists are also involved in the management of APS patients.

Clinical Manifestations

APS manifestations can be grouped into three primary types:

1) Thrombotic APS, which involves arterial or venous thrombosis with persistence of antiphospholipid antibodies for >12 weeks;

2) Obstetric APS, which includes patients with pregnancy morbidity (spontaneous abortion after 10 weeks, prematurity due to severe preeclampsia or placental insufficiency, or multiple early

pregnancy losses in addition to persistence of antiphospholipid antibodies for >12 weeks); and

3) Catastrophic APS, which is a life-threatening form of APS involving small and large vessel occlusions that affect multiple organs and occurs simultaneously and rapidly. Catastrophic APS is rare, accounting for <1% of APS cases.

The most common presenting site of thrombosis in APS is deep veins of the lower extremities. Arterial thromboses, while less common, are potentially life or limb-threatening; these include ischemic stroke (the most common of the arterial complications), myocardial infarction, renal artery thrombosis, and limb ischemia.[3,4] In addition to the hallmark thrombotic manifestations, non-thrombotic clinical manifestations such as thrombocytopenia, livedo reticularis, cardiac valve abnormalities, and cognitive and kidney dysfunction are common clinical features.[5]

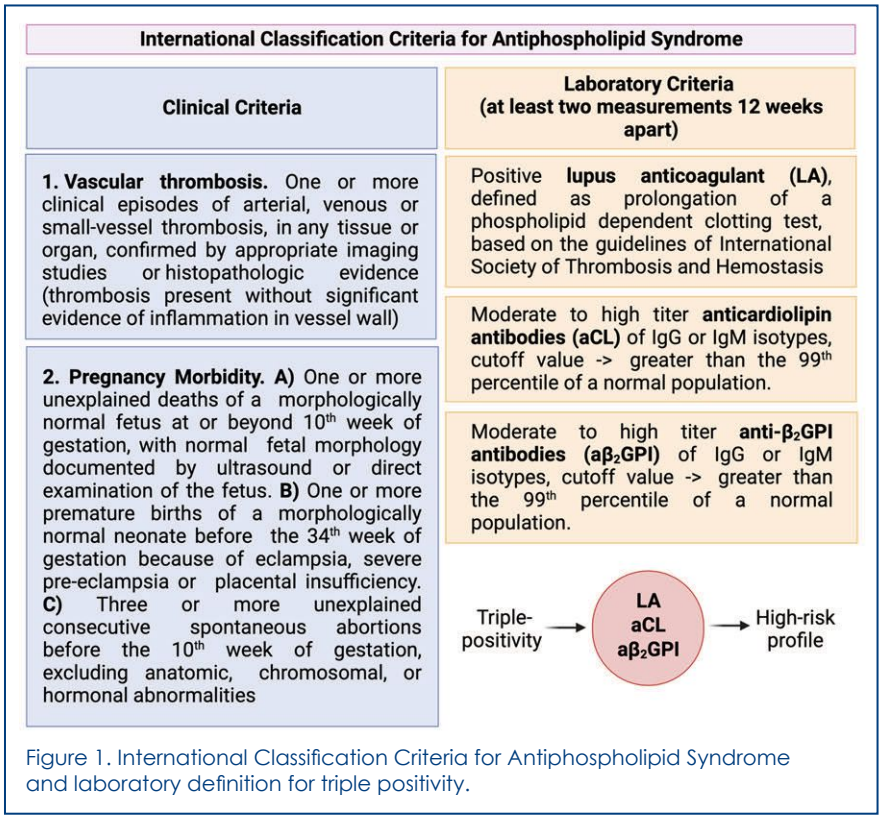


Figure 1. International Classification Criteria for Antiphospholipid Syndrome and laboratory definition for triple positivity.

Diagnosis

Diagnosis of APS should be suspected when one or more unprovoked thrombotic events occur in young patients (<50 years of age) with no apparent cardiovascular risk factors, in patients with cardiovascular risk factors in which thrombosis occurs at unusual sites, or in any case of unexplained pregnancy morbidity[6] (Figure 1). Non-thrombotic clinical manifestations described above, or the presence of SLE should also increase suspicion.

In addition to a history of at least one clinical event, diagnosis requires the persistent presence of one or more types of antiphospholipid antibodies at least 12 weeks apart. In fact, antiphospholipid antibodies can appear during infections,[7] but those are usually transient and/or different from those traditionally detected in thrombotic APS patients, as our studies have recently shown in the case of COVID-19.[8]

Three types of laboratory tests have clinical significance and are part of the International Classification Criteria[9] (Figure 1): lupus anticoagulant, anticardiolipin antibodies (aCL), and anti-β_2-glycoprotein I antibodies (aβ_2GPI).

Lupus anticoagulant is a functional test that detects all antiphospholipid antibodies. As such, lupus anticoagulant positivity is regarded as the most important risk factor for APS-related thrombotic events. Detection of lupus anticoagulant typically involves two functional coagulation assays that measure the ability of antiphospholipid antibodies to prolong the phospholipiddependent clotting time: diluted Russell viper venom time (dRVVT) and the activated partial thromboplastin time (aPTT). An individual is considered positive for lupus anticoagulant if at least one of these tests is positive. Positivity for lupus anticoagulant should be interpreted with caution, since elevated levels of acute-phase reactants and anticoagulant therapy interfere with the test results.

Unlike lupus anticoagulant, aCL and aβ_2GPI measure a subset of antiphospholipid antibodies. These antibodies target the negatively charged phospholipid cardiolipin (aCL) and the protein cofactor $\beta_2$glycoprotein I (aβ_2GPI), respectively.

However, it is important to keep in mind that since β_2GPI binds to cardiolipin, and detection of aCL occurs in the presence of β_2GPI, there is a significant overlap between these two groups, and concerns remain about differences and similarities. Anticardiolipin antibodies and aβ_2GPI are measured by solid phase immunoassays, in which cardiolipin or $\beta_2$glycoprotein I are coated onto plastic plates and patients' plasma serves as a source of autoantibodies.[10] In this case, the presence of either IgG or IgM isotypes is considered diagnostic, although IgG antibodies have more clinical relevance than IgM.

Since antiphospholipid antibodies are a heterogeneous group of autoantibodies with overlapping but not identical characteristics, there are other factors to consider when making a diagnosis. The risk associated with vascular complications is likely to depend on the patient's presentation and specific antiphospholipid antibody profile[11]. Patients with more than one positive test, particularly those positive for lupus anticoagulant aCL, and aβ_2GPI, show the strongest association with thromboses (Figure 1). These patients, commonly referred to as "triple-positive," are categorized as having a high-risk antiphospholipid antibody profile[12], and are managed more aggressively.

Treatment

Since the biggest concern in APS patients is vascular thromboses, treatment consists of strategies to lower thrombotic risk. However, since clinical manifestations are diverse and laboratory testing takes up to 12 weeks to confirm the presence of antiphospholipid antibodies, management of suspected APS patients remains challenging. Three major scenarios can be envisioned: 1) patients with persistently positive antiphospholipid antibodies who have not had a clinical thrombotic event; 2) patients with acute thromboses in which APS is suspected; and 3) patients with a definite APS diagnosis.

Primary thrombosis prophylaxis should be considered for patients with persistently positive antiphospholipid antibodies who have not had a clinical thrombotic event. However, whether or not it is appropriate to treat these individuals prophylactically is a clinical decision dependent on patient factors such as presence of systemic rheumatic disease like SLE, history of obstetric APS, triple positivity, as well as the additional risk factors for thrombosis (major surgery, estrogen use, hyperlipidemia, tobacco use).[6] In general, the use of antiplatelets or anticoagulation for primary prevention in patients without prior thrombosis is not recommended in the US, although some international agencies have recently endorsed the use of low-dose aspirin for subjects with a high-risk profile, defined as persistently positive for lupus anticoagulant or with triple positive status, or both.[13] In the context of primary thrombosis prophylaxis, statins may also be considered in antiphospholipid-positive patients with additional cardiovascular risks factors,[13,14] whereas hydroxychloroquine may be considered among antiphospholipid-positive patients with underlying systemic autoimmune diseases such as SLE.[13]

For acute thromboses in suspected APS, therapeutic interventions and anticoagulation are initiated prior to confirmatory testing for a diagnosis of APS since most patients will not have been tested for antiphospholipid antibodies previously, and the diagnosis will only be confirmed with a positive follow-up test for antiphospholipid antibodies 12 weeks after the initial event.[9] Acute thromboses (MI, PE, ischemic stroke, limb ischemia) should be promptly managed according to current guidelines for the respective diagnoses. Long-term anticoagulation may be considered for other prothrombotic conditions, as well as for a probable diagnosis of APS, while confirmatory testing is pending. Currently, warfarin is preferred over direct oral anticoagulants (DOACs) such as dabigatran, rivaroxaban, and apixaban for non-pregnant patients where APS is likely.[6,13] Patient factors that increase the likelihood of APS and support the use of warfarin include a high-risk antiphospholipid antibody profile, arterial rather than venous thrombosis, presence of systemic rheumatic diseases such as SLE, or prior history of obstetric morbidity or APS during pregnancy. Patient factors that suggest a lower likelihood of APS and promote the use of a DOAC include thrombotic events during an acute viral infection (which can transiently elevate antiphospholipid antibodies), venous thromboembolism after surgery or prolonged immobility, or arterial thrombosis in the presence of atherosclerosis.[15] Heparin and its derivatives, such as fondaparinux, are recommended for use during pregnancy due to warfarin's teratogenic effects.

Dabigatran, rivaroxaban, and apixaban remain contraindicated in pregnancy and for use in the post-partum nursing mother. After birth, warfarin, heparin, and fondaparinux are safe for breastfeeding mothers.[16]

After the initial three- to six-month treatment phase following an acute thrombotic event, testing positive for antiphospholipid antibodies at 12 weeks is diagnostic of APS and determines the need for secondary thrombosis prophylaxis. Current therapeutic recommendations for patients with only venous thrombosis include warfarin with a goal INR of 2-3.[17] For patients with arterial thromboembolism, warfarin therapy with a target INR of 2-3 plus aspirin is indicated.[18] Current guidelines do not recommend DOACs in these patients with APS, as studies have demonstrated inferiority when compared to warfarin therapy.[19-22] However, this remains a matter of debate. For now, DOACs may be considered only in select patients who are at low risk for recurrent thrombotic events or who cannot tolerate warfarin or the required monitoring. For most patients with an unprovoked thrombotic event and laboratory evidence of persistent antiphospholipid antibodies, lifelong anticoagulation is recommended due to the lifelong morbidity and risk of mortality associated with recurrent thrombosis.[23] A high-risk antiphospholipid antibody profile remains one of the greatest risk factors for recurrent thrombosis. Conversely, cessation of anticoagulation therapy may be considered in patients with low or normal repeat antiphospholipid antibody titers.

Antiphospholipid Antibodies

Etiology of Antiphospholipid Antibodies

Antiphospholipid antibodies are a heterogeneous group of antibodies.[7] Most bind to phospholipid-binding proteins, like β_2-glycoprotein I,[24] but some bind directly to neutral and anionic phospholipids that are exposed on cell surfaces, like cardiolipin[25] and lysobisphosphatidic acid.[26] Several hypotheses have been advanced to explain their existence and heterogeneity. A theory is that antiphospholipid antibodies are natural antibodies generated by B1 lymphocytes. At low concentrations, they might contribute to the normal hemostatic balance present in healthy individuals,[27] bridging the time needed by adaptive immunity to develop specific antibodies. However, upregulation of their plasma levels and subsequent antigen-driven maturation would transform them from potentially protective to pathogenic. Mechanisms leading to their upregulation and maturation are unclear, but clinical observations show a relationship between the presence of antiphospholipid antibodies and infections, suggesting there are structural similarities between proteins expressed by pathogens and autoantigens, a concept known as molecular mimicry. Recent work showing that persistent immune reactivation from gut microbiota[28] results in antiphospholipid antibodies reinforces this relationship and extends this concept from pathogens to commensal bacteria. However, other correlations beyond molecular mimicry exist, as antiphospholipid antibodies have also been found in many types of malignancy and after treatment with certain drugs.

Pathophysiology of APS

Since antiphospholipid antibodies are persistently present but thrombotic events occur only occasionally, a "two-hit" model has been proposed to explain thrombotic complications in APS patients.[7] This model argues that the presence of antiphospholipid antibodies ("first hit") is necessary but not sufficient to trigger thrombosis; a secondary insult ("the second hit") is required. Mechanisms for the "second hit" include activation of endovascular and immune cells via surface adhesion molecules and cytokines and the release of cellular debris to induce inflammation and the response of neutrophils and platelets. Such a pro-inflammatory state significantly increases the risk of developing thrombosis in individuals exposed to infection, surgery, or acute endovascular injury but also in individuals with subclinical chronic inflammation and stasis.

Although the precise mechanism by which antiphospholipid antibodies tip the hemostatic balance toward a procoagulant state remains incompletely understood, studies have captured the activation of endothelial cells[29,30] monocytes[31] and neutrophils[32] by aβ_2GPI antibodies, causing expression of tissue factor and adhesion molecules, which are known activators of platelet aggregation and the coagulation cascade. Elevated complement activity was also measured in thrombotic APS

patients, with recent studies suggesting a possible link between β_2GPI and mannose-binding lectin.[33] Finally, neutrophils may play an additional role by releasing neutrophil extracellular traps (NETs), which are webs of chromatin and bactericidal proteins,[34] in response to aβ_2GPI antibodies.[35] These NETs could, in turn, contribute to an inflammatory response that could induce thrombosis. Furthermore, as recently shown in catastrophic APS, mutations in complement regulatory genes, hence a predisposed genetic background, might play a key role.[36]

Molecular Targets

Many antigens of antiphospholipid antibodies have been proposed over the years, but biochemical validation for several of them remains elusive. Also, little evidence exists regarding how antiphospholipid antibodies interact with their targets, even those that have been known for decades. Coagulation and complement factors like complement factor H, complement factor I, prothrombin, annexin A5 and endothelial protein C receptor are likely to play a role; however, the most studied of them all is beta-2 glycoprotein I (β_2GPI), a circulating phospholipid-binding glycoprotein with an unknown physiological role.[37]

β_2GPI features 5 domains arranged like beads on a string, the last of which is a positively charged domain that interacts with negatively charged phospholipids on cell membranes. Because of its string-like structure, it is a flexible protein. Various studies have captured evidence of J-shaped (open), S-twisted, and O-shaped (closed) conformations, each of which could have different roles in APS pathology.[38-40] A schematic of the J conformation obtained by X-ray crystallography, which is the only conformation so far captured at atomic resolution, is shown in Figure 2. There remains controversy regarding the conformational changes that induce the disease; one hypothesis postulates a switch from the O to the J conformation upon membrane binding, thus exposing a cryptic binding site for antiphospholipid antibodies in domain one (DI), ultimately inducing the coagulation cascade and causing thrombosis.[39] Another hypothesis supported by our recent studies[40,41] describes the prevalence of J-shaped form in solution, which then binds to anionic membranes allowing for the locally increased binding affinity of

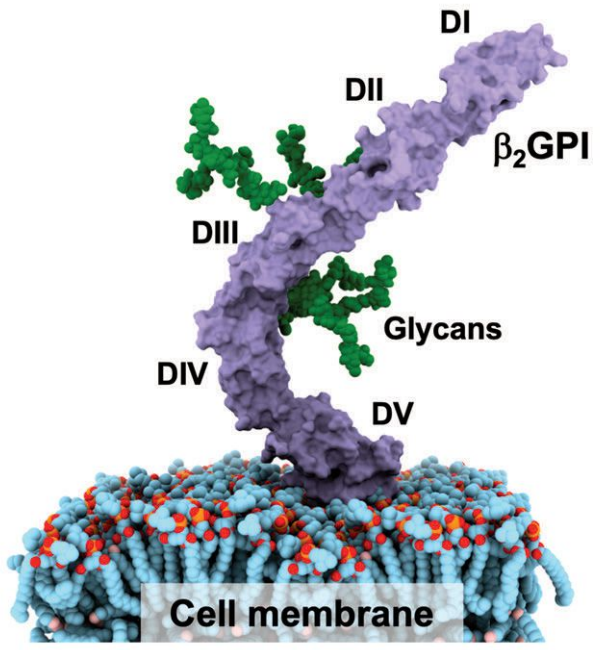


Figure 2. Structural model of the J-elongated form of beta-2 glycoprotein I (β2GPI) bound to the cell membrane. β2GPI is comprised of 5 domains (DI-DV) organized to form a J-shape conformation. Sugars (glycans) occupy the waist of the molecule, forming a molecular belt providing physical separation between DI and DV. Domain V (DV) interacts with the negatively charged cell membranes, composed of phospholipids (blue).

antiphospholipid antibodies. Since, in our studies, the interaction between antiphospholipid antibodies and β_2GPI can occur in solution, as it does not require large-scale conformational changes of the antigen, these results provide new ideas on how to develop technologies for blocking antigen-autoantibody interaction and accumulation on cell membranes, thus reducing downstream effects of antiphospholipid antibodies.

Future Directions and Challenges

What's New in APS Diagnosis?

The heterogeneity of clinical presentation and antiphospholipid antibodies make a timely and accurate diagnosis of APS challenging for clinicians. Identifying patients at higher risk for severe APS and vascular events also often requires multiple tests, including aβ_2GPI, anti-cardiolipin antibodies, and the lupus anticoagulant, all of which are included in the international criteria. However, these are not

diagnostic criteria, and it is left to the physician to interpret the clinical picture and laboratory data as a whole. This leads to a lack of standardization in testing obtained and diagnosis.

Standardization of how the test themselves are conducted in the lab is an important area of improvement.[10,42] In fact, guidelines on how antiphospholipid antibody testing should be conducted across different laboratories are continuously being updated. Of the three diagnostics tests, lupus anticoagulant is potentially the most predictive but also the most puzzling to interpret for physicians because of the many confounding factors that can falsify the test. The name itself is paradoxical since "lupus anticoagulant" stands for the functional ability of certain types of antiphospholipid antibodies to prolong, in vitro, phospholipiddependent clotting time. However, in vivo, these antibodies result in a hypercoagulable state. While new conditions and new reagents are constantly being tested to improve reproducibility and minimize false positive and negative results, immunologic assays might one day replace this test.

Emerging diagnostic tools accessible to physicians that have shown utility in identifying APS patients but have yet to be commented on in the classification criteria include anti-domain I antibodies (a subgroup of aβ_2GPI antibodies with proven pathogenic potential[43,44]) and anti-phosphatidylserine/prothrombin antibodies (shown to be associated with thrombotic APS[45] and triple-positivity[46,47]). IgA aβ_2GPI antibodies also correlate with APS symptoms, especially in lupus patients, but no strong evidence for the diagnostic value in thrombotic or obstetric clinical events exists.[10] Currently, these tests serve as supplemental evidence to consolidate diagnosis in patients with conflicting antiphospholipid antibody profiles.

Diagnostic tools that are not yet commercially available but are under development by different groups focus on newly engineered proteins[48] and carefully designed peptide libraries.[49] The goal of these tests is twofold. First, to define in greater detail the characteristics of circulating antiphospholipid antibodies in each individual. This will likely result in more accurate risk stratification and better clinical decisions. Second, to control the chemical and conformational properties of the antigens. This will likely improve standardization between different laboratories as well as the interpretation of results. A novel ELISA assay in which the antigen conformation and orientation are well-defined and density is tunable provides a proof of concept for this new diagnostic approach.[48] This assay was developed recently in our laboratory and is accessible to interested investigators.

What's New in treatment?

The mainstay of APS treatment remains anticoagulation, most often with warfarin. However, several existing drugs are under investigation.

Hydroxychloroquine is routinely used in the treatment of lupus and has shown reduction in the risk of thrombosis in thrombotic APS and asymptomatic carriers of antiphospholipid antibodies.[50] However, there remains a need for multicenter clinical trials assessing the efficacy of this treatment outside of APS patients with SLE.

Rituximab is often used to treat other systemic rheumatic diseases, with evidence that rituximab can reduce antibody titers in several antibody-mediated autoimmune conditions. It can be used in antiphospholipid antibody-positive APS patients.[51] However, insufficient data recommends routine use of rituximab in thrombotic APS. One study utilizing Rituxumab for treatment of 19 APS patients with noncriteria manifestations found that rituximab was safe and particularly effective for skin manifestations. However, rituximab had no effect on antiphospholipid antibody levels in this study.

Eculizumab is a monoclonal antibody that is active against the complement system and has been used in refractory APS and catastrophic APS, with most case reports describing favorable clinical responses. The evidence is, however, limited to case series with a handful of patients, and it remains unclear how early eculizumab must be given in the clinical course to be effective.[52]

There are also investigations for non-preapproved therapies that have shown promising results in preclinical studies. One such therapy is redirecting T cells to selectively kill anti-β_2GPI antibody-producing B cells using chimeric autoantigen-TCRs (CATCRs), allowing T cells to bind autoantigen-specific B-cell receptors (BCRs) and induce selective cell death. Other B-cell therapies include monoclonal antibodies to B-cell activating factor (BAFF), which has been

shown to exist at higher levels in many types of autoimmune diseases, including APS.[53]

Pathways to disrupt neutrophilic involvement in APS thrombosis are also being plied.[54] Neutrophil extracellular traps (NETs) have recently received increasing attention as drivers of thromboinflammation in APS. One proposed regulatory mechanism of NETosis involves surface adenosine A2A receptors, which are abundant on the surface of neutrophils and act as a brake on inflammation.[55] Two FDA-approved medications with antineutrophil properties, defibrotide and dipyridamole, have antineutrophilic properties and a protective effect against endothelial activation by agonism via different mechanisms with adenosine receptors. Finally, knockout of KLF2, a transcription factor critical for organization of the neutrophil's actin cytoskeleton, resulted in a neutralizing effect of the cells' adhesive properties and, thereby, thrombogenic activity in both arterial and venous vascular beds[56]. This selective modulation of immune cell activity could become a powerful therapy for APS as well as other procoagulant states.

Conclusion

APS is a life-threatening autoimmune disease resulting in thrombosis of arteries and veins in young patients and obstetric complications such as miscarriage, stillbirth, and preeclampsia. Epidemiologic and treatment center data specific to Missouri's population is currently unavailable, underscoring the need for continued investigation of this disease in our state. APS is mediated by the binding of antiphospholipid antibodies to lipids and proteins like β_2GPI that interact with cell membranes, resulting in the activation of the complement and coagulation cascades. Diagnosis of this disease can be difficult due to great heterogeneity in molecular hallmarks of the disease, and the need for follow-up testing after an acute thrombosis. The International Classification for laboratory evidence of antiphospholipid antibodies provides helpful insight to physicians looking to diagnose APS, but fails to bridge the gap between the molecular basis of the disease and patient factors such as presentation and comorbidities when determining the overall risk of thrombosis recurrence. The current treatment of APS is non-specific, limited to anticoagulation to prevent thrombosis and improve pregnancy outcomes. Over the last three decades, advancement in basic science technologies has made the study of the underlying molecular mechanisms of the disease more tractable. Elucidating the molecular drivers of the disease using integrated multi-omics approaches and defining interactions between antigens and antibodies are exciting new avenues in APS research. Basic knowledge to emerge from these studies will open the door to more specific and targeted diagnostic and therapeutic approaches.

References

1. Cervera R, Serrano R, Pons-Estel GJ, et al. Morbidity and mortality in the antiphospholipid syndrome during a 10-year period: a multicentre prospective study of 1000 patients. Ann Rheum Dis. 2015;74(6):1011-1018.
2. Dabit JY, Valenzuela-Almada MO, Vallejo-Ramos S, Duarte-Garcia A. Epidemiology of Antiphospholipid Syndrome in the General Population. Curr Rheumatol Rep. 2022;23(12):85.
3. Miranda S, Park J, Le Gal G, et al. Prevalence of confirmed antiphospholipid syndrome in 18-50 years unselected patients with first unprovoked venous thromboembolism. J Thromb Haemost. 2020;18(4):926-930.
4. Cervera R, Piette JC, Font J, et al. Antiphospholipid syndrome: clinical and immunologic manifestations and patterns of disease expression in a cohort of 1,000 patients. Arthritis Rheum. 2002;46(4):1019-1027.
5. Ruiz-Irastorza G, Crowther M, Branch W, Khamashta MA. Antiphospholipid syndrome. Lancet. 2010;376(9751):1498-1509.
6. Garcia D, Erkan D. Diagnosis and Management of the Antiphospholipid Syndrome. N Engl J Med. 2018;378(21):2010-2021.
7. Meroni PL, Borghi MO, Raschi E, Tedesco F. Pathogenesis of antiphospholipid syndrome: understanding the antibodies. Nat Rev Rheumatol. 2011;7(6):330-339.
8. Borghi MO, Beltagy A, Garrafa E, et al. Anti-Phospholipid Antibodies in COVID-19 Are Different From Those Detectable in the Anti-Phospholipid Syndrome. Front Immunol. 2020;11:584241.
9. Miyakis S, Lockshin MD, Atsumi T, et al. International consensus statement on an update of the classification criteria for definite antiphospholipid syndrome (APS). J Thromb Haemost. 2006;4(2):295-306.
10. Devreese KMJ. Solid Phase Assays for Antiphospholipid Antibodies. Semin Thromb Hemost. 2022;48(6):661-671.
11. Galli M, Luciani D, Bertolini G, Barbui T. Lupus anticoagulants are stronger risk factors for thrombosis than anticardiolipin antibodies in the antiphospholipid syndrome: a systematic review of the literature. Blood. 2003;101(5):1827-1832.
12. Pengo V, Ruffatti A, Legnani C, et al. Clinical course of high-risk patients diagnosed with antiphospholipid syndrome. J Thromb Haemost. 2010;8(2):237-242.
13. Cohen H, Cuadrado MJ, Erkan D, et al. 16th International Congress on Antiphospholipid Antibodies Task Force Report on Antiphospholipid Syndrome Treatment Trends. Lupus. 2020;29(12):1571-1593.
14. Drosos GC, Vedder D, Houben E, et al. EULAR recommendations for cardiovascular risk management in rheumatic and musculoskeletal diseases, including systemic lupus erythematosus and antiphospholipid syndrome. Ann Rheum Dis. 2022;81(6):768-779.
15. Schulz A, Herrmann E, Ott O, Lindhoff-Last E. Thromboembolic Antiphospholipid Syndrome (APS): Efficacy and Safety of Different Anticoagulants-Results of the APSantiCO Registry. J Clin Med. 2022;11(16).
16. Bates SM, Rajasekhar A, Middeldorp S, et al. American Society of Hematology 2018 guidelines for management of venous thromboembolism:

venous thromboembolism in the context of pregnancy. Blood Adv. 2018;2(22):3317-3359.
17. Crowther MA, Ginsberg JS, Julian J, et al. A comparison of two intensities of warfarin for the prevention of recurrent thrombosis in patients with the antiphospholipid antibody syndrome. N Engl J Med. 2003;349(12):1133-1138.
18. Tektonidou MG, Tincani A, Ward MM. Response to: 'Correspondence on 'EULAR recommendations for the management of antiphospholipid syndrome in adults" by Gao and Qin. Ann Rheum Dis. 2021.
19. Cohen H, Hunt BJ, Efthymiou M, et al. Rivaroxaban versus warfarin to treat patients with thrombotic antiphospholipid syndrome, with or without systemic lupus erythematosus (RAPS): a randomised, controlled, open-label, phase 2/3, non-inferiority trial. Lancet Haematol. 2016;3(9):e426-436.
20. Dufrost V, Risse J, Zuily S, Wahl D. Direct Oral Anticoagulants Use in Antiphospholipid Syndrome: Are These Drugs an Effective and Safe Alternative to Warfarin? A Systematic Review of the Literature. Curr Rheumatol Rep. 2016;18(12):74.
21. Dufrost V, Risse J, Reshetnyak T, et al. Increased risk of thrombosis in antiphospholipid syndrome patients treated with direct oral anticoagulants. Results from an international patient-level data meta-analysis. Autoimmun Rev. 2018;17(10):1011-1021.
22. Pengo V, Denas G, Zoppellaro G, et al. Rivaroxaban vs warfarin in high-risk patients with antiphospholipid syndrome. Blood. 2018;132(13):1365-1371.
23. Khamashta MA, Cuadrado MJ, Mujic F, Taub NA, Hunt BJ, Hughes GR. The management of thrombosis in the antiphospholipid-antibody syndrome. N Engl J Med. 1995;332(15):993-997.
24. de Groot PG, Urbanus RT. The significance of autoantibodies against beta2-glycoprotein I. Blood. 2012;120(2):266-274.
25. Manukyan D, Muller-Calleja N, Jackel S, et al. Cofactor-independent human antiphospholipid antibodies induce venous thrombosis in mice. J Thromb Haemost. 2016;14(5):1011-1020.
26. Muller-Calleja N, Hollerbach A, Royce J, et al. Lipid presentation by the protein C receptor links coagulation with autoimmunity. Science. 2021;371(6534).
27. Lackner KJ, Muller-Calleja N. Antiphospholipid Antibodies: Their Origin and Development. Antibodies (Basel). 2016;5(2).
28. Ruff WE, Vieira SM, Kriegel MA. The role of the gut microbiota in the pathogenesis of antiphospholipid syndrome. Curr Rheumatol Rep. 2015;17(1):472.
29. Simantov R, LaSala JM, Lo SK, et al. Activation of cultured vascular endothelial cells by antiphospholipid antibodies. J Clin Invest. 1995;96(5):2211-2219.
30. Del Papa N, Guidali L, Sala A, et al. Endothelial cells as target for antiphospholipid antibodies. Human polyclonal and monoclonal anti-beta 2-glycoprotein I antibodies react in vitro with endothelial cells through adherent beta 2-glycoprotein I and induce endothelial activation. Arthritis Rheum. 1997;40(3):551-561.
31. Amengual O, Atsumi T, Khamashta MA, Hughes GR. The role of the tissue factor pathway in the hypercoagulable state in patients with the antiphospholipid syndrome. Thromb Haemost. 1998;79(2):276-281.
32. Ritis K, Doumas M, Mastellos D, et al. A novel C5a receptor-tissue factor cross-talk in neutrophils links innate immunity to coagulation pathways. J Immunol. 2006;177(7):4794-4802.
33. Durigutto P, Macor P, Pozzi N, et al. Complement Activation and Thrombin Generation by MBL Bound to beta2-Glycoprotein I. J Immunol. 2020;205(5):1385-1392.
34. Brinkmann V, Reichard U, Goosmann C, et al. Neutrophil extracellular traps kill bacteria. Science. 2004;303(5663):1532-1535.
35. Yalavarthi S, Gould TJ, Rao AN, et al. Release of neutrophil extracellular traps by neutrophils stimulated with antiphospholipid antibodies: a newly identified mechanism of thrombosis in the antiphospholipid syndrome. Arthritis Rheumatol. 2015;67(11):2990-3003.
36. Chaturvedi S, Braunstein EM, Yuan X, et al. Complement activity and complement regulatory gene mutations are associated with thrombosis in APS and CAPS. Blood. 2020;135(4):239-251.
37. de Groot PG, Meijers JC. beta(2) -Glycoprotein I: evolution, structure and function. J Thromb Haemost. 2011;9(7):1275-1284.
38. Hammel M, Kriechbaum M, Gries A, Kostner GM, Laggner P, Prassl R. Solution structure of human and bovine beta(2)-glycoprotein I revealed by small-angle X-ray scattering. J Mol Biol. 2002;321(1):85-97.
39. Agar C, van Os GM, Morgelin M, et al. Beta2-glycoprotein I can exist in 2 conformations: implications for our understanding of the antiphospholipid syndrome. Blood. 2010;116(8):1336-1343.
40. Ruben E, Planer W, Chinnaraj M, et al. The J-elongated conformation of beta(2)-glycoprotein I predominates in solution: implications for our understanding of antiphospholipid syndrome. J Biol Chem. 2020;295(31):10794-10806.
41. Kumar S, Chinnaraj M, Planer W, et al. An allosteric redox switch in domain V of beta(2)-glycoprotein I controls membrane binding and anti-domain I autoantibody recognition. J Biol Chem. 2021;297(2):100890.
42. Devreese KMJ. Testing for antiphospholipid antibodies: Advances and best practices. Int J Lab Hematol. 2020;42 Suppl 1:49-58.
43. Yin D, Chayoua W, Kelchtermans H, et al. Detection of anti-domain I antibodies by chemiluminescence enables the identification of high-risk antiphospholipid syndrome patients: A multicenter multiplatform study. J Thromb Haemost. 2020;18(2):463-478.
44. Banzato A, Pozzi N, Frasson R, et al. Antibodies to Domain I of beta(2)Glycoprotein I are in close relation to patients risk categories in Antiphospholipid Syndrome (APS). Thromb Res. 2011;128(6):583-586.
45. Atsumi T, Ieko M, Bertolaccini ML, et al. Association of autoantibodies against the phosphatidylserine-prothrombin complex with manifestations of the antiphospholipid syndrome and with the presence of lupus anticoagulant. Arthritis Rheum. 2000;43(9):1982-1993.
46. Vandevelde A, Chayoua W, de Laat B, et al. Added value of antiphosphatidylserine/prothrombin antibodies in the workup of thrombotic antiphospholipid syndrome: Communication from the ISTH SSC Subcommittee on Lupus Anticoagulant/Antiphospholipid Antibodies. J Thromb Haemost. 2022;20(9):2136-2150.
47. Chinnaraj M, Planer W, Pengo V, Pozzi N. Discovery and characterization of 2 novel subpopulations of aPS/PT antibodies in patients at high risk of thrombosis. Blood Adv. 2019;3(11):1738-1749.
48. Chinnaraj M, Pengo V, Pozzi N. A Novel ELISA Assay for the Detection of Anti-Prothrombin Antibodies in Antiphospholipid Syndrome Patients at High Risk of Thrombosis. Front Immunol. 2021;12:741589.
49. Moghbel M, Roth A, Baptista D, et al. Epitope of antiphospholipid antibodies retrieved from peptide microarray based on R39-R43 of beta2-glycoprotein I. Res Pract Thromb Haemost. 2022;6(7):e12828.
50. Arachchillage DJ, Laffan M, Pericleous C. Hydroxychloroquine as an Immunomodulatory and Antithrombotic Treatment in Antiphospholipid Syndrome. Int J Mol Sci. 2023;24(2).
51. Hubben A, McCrae KR. Emerging Therapies in Antiphospholipid Syndrome. Transfus Med Rev. 2022;36(4):195-203.
52. Andrade D, Tektonidou M. Emerging Therapies in Antiphospholipid Syndrome. Curr Rheumatol Rep. 2016;18(4):22.
53. van den Hoogen LL, Bisoendial RJ. B-Cells and BAFF in Primary Antiphospholipid Syndrome, Targets for Therapy? J Clin Med. 2022;12(1).
54. Knight JS, Kanthi Y. Mechanisms of immunothrombosis and vasculopathy in antiphospholipid syndrome. Semin Immunopathol. 2022;44(3):347-362.
55. Ali RA, Gandhi AA, Meng H, et al. Adenosine receptor agonism protects against NETosis and thrombosis in antiphospholipid syndrome. Nat Commun. 2019;10(1):1916.
56. Nayak L, Sweet DR, Thomas A, et al. A targetable pathway in neutrophils mitigates both arterial and venous thrombosis. Sci Transl Med. 2022;14(660):eabj7465.

Disclosure

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Signaling by Ion Channels: Pathways, Dynamics and Channelopathies

by Gucan Dai, PhD

Ion channels are precisely organized and play a crucial role in dynamic signaling, which is often unique to the animal kingdom.



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Abstract

Charged ions and ion channels play a critical role in regulating the electrical activities of excitable cells. This review discusses the principles of ion channel regulation in the time domain, as well as the diseases that can arise from channel dysfunction and disturbances in ionic balance. Ion channel signaling is a dynamic process that is essential for various physiological functions, including pain sensation, motor control, and the body's response to stress, such as fight-or-flight response.

Introduction

Ion channels are membrane proteins that create subnanometer-diameter pores that allow specific ions to permeate the cell membrane.[1] More than 300 types of ion channel genes have been identified in the human genome, and dysfunctions in these channels, known as channelopathies, are implicated in a wide range of diseases. They are also an important drug target, with numerous therapeutics designed to modulate their activity. Channel regulation is mediated by a wide range of physiological stimuli, including transmembrane voltage, temperature, neurotransmitters, and mechanical force. Recent research has revealed that membrane lipids and lipid-bilayer structures also play a key role in modulating channel function and electrical excitability, allowing for fine-tuning of channel activity during complex physiological responses.

The Roles of Ion Channels: from Nerve Impulses to Cystic Fibrosis

Ionic fluxes from voltage-activated sodium (Na^+) and potassium (K^+) channels establish the rise and fall of action potentials, commonly referred to as "nerve impulses" or "spikes" (Figure 1). These impulses are crucial to nearly all physiological processes. They underlie the transmission of information between neurons, allowing us to perceive the world around us, and to think. Furthermore, ion channels are essential to the function of other excitable cells, such as muscle cells. In muscle cells, action potentials are responsible for the contraction of muscles, allowing us to move and exert force.

Apart from the channels that shape nerve impulses, there are other channels that are well-known to medical doctors. These channels include the renal outer medullary K^+ channel (ROMK) that secretes potassium in the collecting duct system of the kidney, the cystic fibrosis transmembrane conductance regulator (CFTR) chloride channels that controls epithelial mucus transport in the lung, the calcium-activated K^+ channels that are important for lacrimal or salivary

secretion, and the voltage-activated calcium (Ca^{2+}) channels that regulate the release of hormones from endocrine cells. Mutations in these channels can result in serious diseases such as cystic fibrosis.

Understanding the molecular and structural mechanisms of ion channels has led to the development of treatments for many channel-related diseases. Cystic fibrosis, a life-threatening disease caused by congenital CFTR mutations, is one such condition affecting more than 80,000 people globally. It is characterized by the buildup of sticky mucus, lung infection, and lung damage. With gene therapies targeting CFTR, this disease is now mostly treatable.

Ion Channels Process Different Sensory Signals

Ion channels are crucial for processing sensory information, including pH, osmolarity, pressure, and temperature. Most ion channels have evolved the ability to respond to multiple external stimuli. One example is the transient receptor potential (TRP) channels, whose name is derived from a mutant strain of the fruit fly *Drosophila* that loses sustained light response in their photoreceptors. TRP channels can be activated by temperature, voltage, and lipids, as well as capsaicin, menthol, and cannabidiol (CBD). Particularly, capsaicin is a compound found in chili peppers and is known to activate the TRPV1 channel. It is used as a topical analgesic for the treatment of neuropathic pain (Figure 2). In addition, nabiximols (Sativex) is an oromucosal spray that contains CBD and activates the TRPV1 channel. It is approved for the treatment of multiple sclerosis-related spasticity and neuropathic pain.

Ion channels achieve their multimodal property through different protein domain evolution or modification. It has been proposed that TRP channels lost some voltage-sensing amino acids and gained hydrophobic/hydrophilic ones elsewhere, which conferred temperature sensitivity on TRP channels.[2] The exact molecular mechanism of the TRP thermo-sensing remains debatable. In general, channels are most often found to be dually activated by voltage and ligand. This multimodal feature of ion channels greatly diversifies the mechanism whereby they could be intricately and dynamically modulated to control cellular excitability. Furthermore, because of this feature and the measurable channel activity at ultra-high temporal resolution (through on-line patch-clamp electrophysiology), ion channels have become one of the favored macromolecules by protein biochemists to study protein allostery, ligand-binding mechanisms and protein thermodynamics.

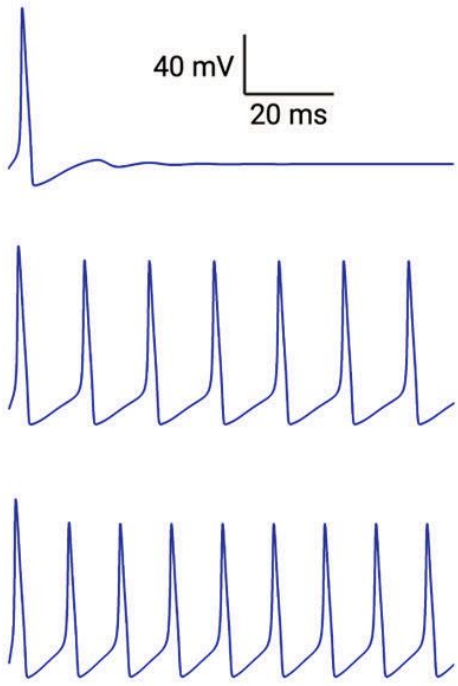


Figure 1. Simulated action potentials generated using the Hodgkin-Huxley model. Different amounts of current were applied to stimulate trains of action potentials with varying frequencies. The current stimulus ranged from low to high (5 pA, 10 pA, and 20 pA from top to bottom), and the parameters used were based on action potential of the squid giant axon and obtained from the Handbook of Physiology - the Nervous System, Chapter 4, authored by Bertil Hille (1977).

Multimodal properties partially explain why the same channel could be therapeutic targets for seemingly different diseases or channelopathies. For example, not only has the TRPV1 channel been a therapeutic target to treat pain, but also to atopic dermatitis, overactive bladder, and cluster headaches. Moreover, the hyperpolarization-activated cyclic nucleotide (HCN)-gated channel, sometimes referred to as "pacemaker channel," is dually regulated by voltage and ligand. Congenital mutations of HCN channels lead to epilepsy, chronic pain and heart diseases such as cardiac arrhythmia.[3] Recent research suggests that HCN2 channels expressed in small nociceptor neurons of the dorsal root ganglion (DRG) can be targeted for treating neuropathic pain.[4]

Nearly All Second Messengers Regulate Ion Channels

Our body requires signaling molecules, also known as messengers, to function properly. While ion channels respond to more than a thousand forms of

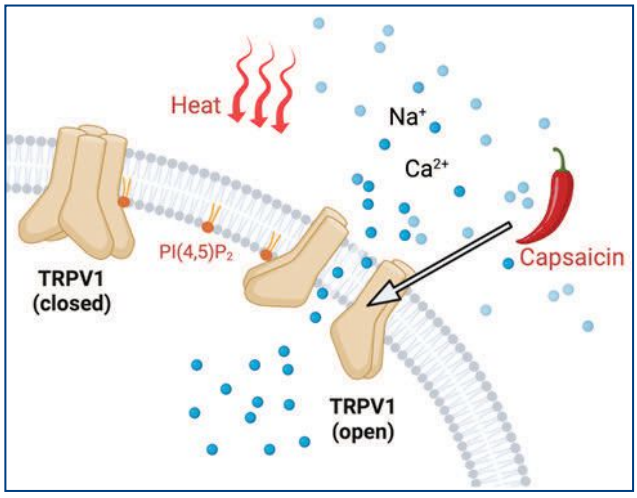


Figure 2. Schematic cartoon showing the multimodal activation of TRPV1 channels by both heat and capsaicin binding. Membrane lipid $PI(4,5)P_2$ binds to TRPV1 channels directly.

first messengers such as extracellular neurotransmitters, odorants, hormones, and pheromones, only a few intracellular second messengers are commonly used in mammalian cells, resulting in a significant convergence in signal transduction. The second messengers utilized in mammalian cells include cyclic adenosine monophosphate (cAMP), cyclic guanosine monophosphate (cGMP), inositol 1,4,5-triphosphate (IP_3), phosphoinositides, diacylglycerol (DAG), G-protein βγ subunits, and cationic Ca^{2+}, Mg^{2+}, and Na^+ ions.

Medical drugs have been developed to manipulate second-messenger systems. For instance, phosphodiesterase (PDE) inhibitors such as sildenafil (Viagra) can increase the levels of cAMP or cGMP by inhibiting the breakdown of these second messengers by phosphodiesterase enzymes.[5] Furthermore, almost all of the second messengers can regulate specific channels through direct binding. For example, cationic Ca^{2+} and Na^+ ions bind to and potentiate Slowpoke family voltage-activated K^+ channels, which are widely expressed in the nervous system.[1,6] The increase in intracellular concentration of second messengers often requires rapid action. Ca^{2+} and Na^+ can enter cells through cation channels such as voltage-gated Ca^{2+} channels, Na^+ channels, or TRP channels, diffuse quickly, and reach their effectors within microseconds, perfectly meeting the requirements of fast signal transduction.

In contrast, other second messengers generally relay to their effectors in signaling cascades within seconds to minutes, which is slower than the cationic second messengers. In some cases, the level of second messengers decreases in the cell to deactivate downstream channels. For instance, decreasing the level of cGMP turns off cyclic nucleotide-gated (CNG) channels to achieve phototransduction in the retina, while decreasing signaling phosphoinositides (particularly the key lipid phosphatidylinositol 4,5-bisphosphate $PI(4,5)P_2$) upon the activation of $G_{q/11}$-type G protein-coupled receptors (GPCRs) closes K^+ and Ca^{2+} channels. From an evolutionary perspective, the "reduction of second messengers" strategy, using PDE or phospholipases (PLC), does not require direct consumption of ATP, making it more energy-efficient.

Mood disorders are often associated with disturbances in the second messenger system, particularly the phosphoinositide-based system. The "inositol depletion hypothesis" has been proposed to provide a mechanistic understanding of mood disorders, especially bipolar disorder and depression. This hypothesis is partly supported by the mood-stabilizing agent lithium, which specifically inhibits inositol recycling. Recent findings have further underscored the medical importance of inositol. For example, supplementing myo-inositols can increase cellular phosphoinositide levels, potentiate various types of K^+ channels, and reduce the hyperexcitability of neurons.[7] Inositol was not previously considered an essential nutrient as it can be synthesized from glucose. However, recent studies have shown that certain individuals, particularly those with insulin resistance and diabetes, may develop inositol deficiencies. This deficiency may be related to the connection between diabetes and mood disorders, commonly referred to as "diabetes mood swings." For pancreatic β-cells under insulin resistance, low cellular glucose levels may lead to low myo-inositol and phosphoinositide levels, which can suppress ATP-sensitive potassium (K_{ATP}) channels,[8] leading to a vicious cycle of high insulin secretion. Inositol may have broad health benefits beyond its currently accepted role in treating polycystic ovary syndrome, a common comorbidity of insulin resistance. Further research is needed to establish the connection between phosphoinositide-based second messengers, ion channels, and mood disorders.

Fast and Slow G Protein-Dependent Signaling Pathways Regulate Ion Channels

When compared to the immediate effects on ion channels caused by voltage, temperature, and mechanical force, or by ligand binding to ionotropic receptors, GPCR- and G protein-mediated signaling pathways to channels are

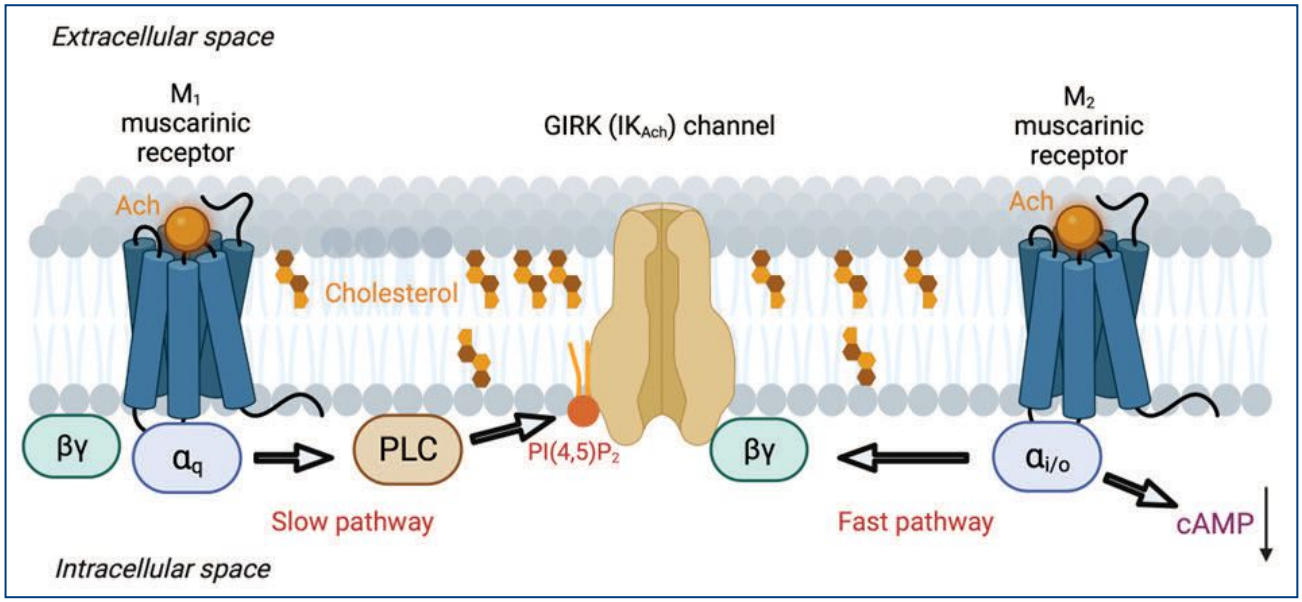


Figure 3. Fast and slow G protein-dependent signaling pathways for modulating ion channels. Activation of the $G_{i/o}$-coupled M2 muscarinic acetylcholine receptors releases the G protein βγ subunits, which activates GIRK channels. On the other hand, activation of the G_q-coupled M1 muscarinic acetylcholine receptors activates PLC, which hydrolyzes membrane lipid $PI(4,5)P_2$, an essential activator of GIRK channels.

generally slower. The channel conformational change is typically the only rate-limiting factor for these immediate effects. The G protein-dependent signaling has significant interest to medical researchers since GPCR is the largest family of proteins that are targeted by FDA-approved therapeutic drugs. Among the G protein-dependent pathways, the relatively fast pathway is mediated by the G-protein βγ subunits released by $G_{i/o}$-coupled GPCR (*i* being *inhibiting* adenylyl cyclases) in a membrane-delimited and pertussis toxin-sensitive manner[9,10] (Figure 3). The type of channel activated by βγ subunits is called the G-protein activated inwardly rectifying K^+ channel, GIRK[10]. It only takes one to five seconds from the application of GPCR agonist to observe a peak of GIRK current in live-cell in-vitro systems;[7] this process can be as fast as a few hundred milliseconds. GIRK channel is also called IK_{Ach} channel because vagal nerve stimulation releases acetylcholine (Ach) to reduce heart rate by activating IK_{Ach}. It was thought that this fast diffusion-limited signaling is partially due to the overabundance of $G\alpha_{i/o}$ subunits relative to other Gα subunits. The GIRK channel is not alone; Gβγ subunits regulate voltage-gated Ca^{2+} channels and TRPM3 channels via a similar mechanism.[11,12]

Scientists have also discovered a slower G protein-dependent signaling pathway in the sympathetic neurons of the superior cervical ganglion (SCG) that helps our bodies deal with stress. This pathway is mediated by the activation of $G_{q/11}$-coupled GPCR and the subsequent recruitment of phospholipase C to hydrolyze $PI(4,5)P_2$ (Figure 3). It is pertussis toxin insensitive and is about five-fold slower than the Gβγ subunit-mediated pathway *in vitro*.[7,13] During this process, IP_3 and DAG are generated, both of which have downstream channels as their effectors. One physiological significance of the PLC/$PI(4,5)P_2$ pathway, which has been a popular area of focus for the past two decades, is its role in regulating over 100 types of ion channels. These channels are directly controlled by $PI(4,5)P_2$ as their ligand, and include HCN, TRP, inward rectifiers, and many other voltage-gated channels.[13,14] In most cases, when $PI(4,5)P_2$ is hydrolyzed, the channel activities subside.[7] Furthermore, regarding the G_s-coupled GPCR pathway (e.g., β-adrenergic receptors, and *s* being *stimulating* adenylyl cyclases), cAMP levels are elevated with similar kinetics (~20 to 50 s), and the effect can be more sustainable. Importantly, the modulation of HCN pacemaker channels through cAMP is crucial for the sympathetic regulation of the heartbeat. Collectively, the varied kinetics of G protein-dependent regulation of ion channels ensures that the cellular excitability can be modulated with high spatiotemporal precision.

Table 1: Calcium-permeable ion channels and the related channelopathies

Channel Type	Main Functions	Related Diseases
Voltage-activated Ca^{2+} channels	Presynaptic neurotransmitter release, excitation-contraction (EC) coupling in cardiac and skeletal muscles	Hypokalemic periodic paralysis, ataxia, migraine
TRP channels	Thermo-sensation, Ca^{2+} release from intracellular organelles	Chronic pain, episodic pain, mucolipidosis, melanoma, dermatitis, carcinoma, asthma, chronic obstructive pulmonary disease, ischemia
Cyclic-nucleotide gated channel	Retinal phototransduction	Retinitis pigmentosa, achromatopsia, macular degeneration
Orai (CRAC) channel	T-cell activation, SOCE	Immunodeficiency, myopathy, and muscular hypotonia
Mitochondrial calcium uniporter	Ca^{2+} uptake to mitochondria	Neurodegeneration, ataxia
IP_3 receptor	ER/SR Ca^{2+} release	Neurodegeneration, taste perception deficit, heart failure and hypertension
Ryanodine receptor	ER/SR Ca^{2+} release, EC coupling	Malignant hyperthermia, central core disease
NMDA receptor	Ca^{2+} entry in synapse, postsynaptic potential, synaptic plasticity, learning and memory	Alzheimer's disease, Parkinson's disease, schizophrenia, epilepsy
Piezo channel	Mechano-sensation	Hereditary xerocytosis, lymphedema, osteoarthritis
CatSper channel	Ca^{2+} entry to sperm flagella	Male infertility

Mobilizing Local Ca^{2+} Is the Goal for Channel Signaling

Adequate calcium intake is essential for the proper functioning of muscles, nerves, and the heart. Because of the 20,000-fold concentration gradient of extracellular (2 mM) versus intracellular (resting 100 nM) Ca^{2+} levels, Ca^{2+} is ideally suited to be a sensitive and dynamic signaling messenger. Ca^{2+} is essential for achieving many crucial physiological functions, for example, the release of neurotransmitter vesicles at presynaptic terminals in the brain and the excitation-contraction coupling for cardiac and skeletal muscle contraction.[1] Cells often use local Ca^{2+} release to achieve fast signaling. In other words, Ca^{2+} does not need to travel far to produce a signaling effect. Besides voltage-activated Ca^{2+} channels, nonselective cation channels like TRPV channels, and neuronal NMDA glutamate receptors serve the role of permitting Ca^{2+} entry into cells.[1] In addition, IP_3 receptors and ryanodine receptors (also known as the ionotropic receptor of caffeine) mobilize Ca^{2+} from the intracellular endoplasmic reticulum ER (and sarcoplasmic reticulum, SR) Ca^{2+} store. The release of Ca^{2+} from IP_3 receptors upon the activation of $G_{q/11}$-type GPCRs coincide temporally with the hydrolysis of plasma membrane $PI(4,5)P_2$ and the rise of IP_3. Nevertheless, Ca^{2+} release from the ER could also exhibit prolonged oscillation due to feedback regulation of IP_3 receptors by local Ca^{2+}. Furthermore, immune cells, like cytotoxic T cells, do not use voltage-activated Ca^{2+} channels, but rather use Ca^{2+} release-activated Ca^{2+} (CRAC) channels[15] identified as Orai channels to allow the influx of extracellular Ca^{2+}. Orai activation is initiated by its direct interaction with the ER-residing protein STIM, which senses the low Ca^{2+} level in the ER store of calcium.[15] Therefore, this process is also called the store-operated Ca^{2+} entry (SOCE) and happens with a timescale of ~ 50 seconds. Elevated cytosolic free Ca^{2+} ions are regulated by Ca^{2+}-binding proteins like calmodulin and buffered by calbindin/parvalbumin in neurons. Ca^{2+} ions can also undergo the mitochondrial Ca^{2+} uptake quickly (less than 100 s) by mitochondrial calcium uniporter (MCU).[1] Other intracellular Ca^{2+} stores such as endosomes and lysosomes can release local Ca^{2+} through TRPML and TRPM2 channels. Besides IP_3, notable Ca^{2+}-mobilizing second messengers include ADP-ribose (ADPR) and nicotinic acid adenine dinucleotide phosphate (NAADP). ADPR is an agonist of Ca^{2+}-permeable TRPM2 channels whereas the ionotropic receptor of NAADP remains uncertain.

There are many diseases linked to Ca^{2+}-permeable channels and imbalances in Ca^{2+} homeostasis (Table 1). Alzheimer's and Parkinson's disease, along with other neurodegenerative diseases, are believed to be connected to alterations in calcium signaling, even though the pathophysiological mechanisms are not yet well understood. The major element in the

Alzheimer's γ-secretase protein complex presenilin-1 was proposed as an ER Ca^{2+}-leak channel, but it could instead function as a Ca^{2+}-leak enhancer by alternative mechanisms. In general, Ca^{2+} signaling through glutamate receptors are important for synaptic plasticity, learning and memory. Furthermore, mutations in TRPML (ML being mucolipin) are associated with a type of neurodevelopmental disorder called mucolipidosis, whereas mutations in TRPM (M being melastatin) are associated with hypomagnesemia and melanoma. Moreover, mutations in genes encoding the Orai/STIM CRAC system are linked to autoimmune diseases, myopathy, and muscular hypotonia[15].

Mechanical Force Activates Ion Channels

A new frontier in drug discovery involves targeting newly characterized channels that detect lipid membrane curvature and deformation caused by changes of cell shape. These channels, known as mechanosensitive channels, produce rapid and often transient currents when subjected to mechanical force applied to the cell membrane. The discovery of human mechanosensitive channels, referred to as "Piezo," represents an exciting new avenue for investigating the mechanism underlying the dynamic impact of the lipid bilayer on ion channels.[16] The opening of Piezo channels is directly coupled to a change in the membrane curvature caused by mechanical force, happening within a few milliseconds after a mechanical stimulus.[17] These channels are not only necessary for touch sensation but also act as sensors for red blood cell volume, morphological changes of endothelial cells in blood vessels, pressure in pancreatic ducts, and stretching of the bladder wall. The physiological significance of these findings is profound, and many diseases caused by dysfunctional mechano-transduction are associated with mutations in Piezo genes. Furthermore, hair cells in the inner ear also use mechanical force through the tip-links between hair bundles to gate a type of channel called TMC (transmembrane channel-like protein), whose identity had been a persistent mystery in auditory research.[18] The latest research suggests that TMC's multiple auxiliary subunits, particularly TMIE, lipids, and lipid membrane thickness could all contribute to achieve the opening of TMC channels[19]. Besides Piezo and TMC, channels like the volume-activated chloride channel, TRPN and the K_{2P} (TREK subtype) channel are other reported mechano-sensors.[20]

In addition, the dynamic heterogeneity of the lipid-bilayer membrane regulates ion channels. Particularly, membrane nanodomains (often less than 250 nm in size) enriched with sphingomyelin (SM) and cholesterol create membrane compartmentalization to gather molecules for effective signaling.[21] These organized nanodomains, previously called "lipid rafts," could rearrange and coalesce under certain pathophysiological conditions, e.g., inflammation, neuropathy and age-related neurodegeneration. Furthermore, drugs that target cholesterols such as statin could change these domains. In general, many more types of channels could be sensitive to membrane deformation and stretching, considering that transmembrane helices tend to rearrange differently depending on the membrane thickness and curvature. It remains elusive how the lipid disposition of membrane nanodomains as well as the line tension at the boundary of different lipid domains could affect the mechano-sensitivity of ion channels.

Blood Flow and Blood Coagulation Factors Regulate Ion Channels in the Brain

The blood flow in the brain is tightly coupled to neuronal activity, as it supplies oxygen and nutrients to neurons. This coupling, so-called "neurovascular coupling," involves a variety of ion channels. One fast pathway is that extracellular K^+ ions, released from astrocytes in response to neuronal activity, act like first messengers to dilate blood vessels (less than a minute) in the brain and increase cerebral blood flow.[22] This effect is achieved by a swift rise in K^+ levels, causing the removal of the blockage of inwardly rectifying K_{ir} channels by intracellular magnesium (Mg^{2+}) and polyamines.[22] It is worth noting that Mg^{2+} also blocks NMDA glutamate receptors. Therefore, K^+ and Mg^{2+} act as signaling ions that dynamically modulate cellular excitability in the brain. The potentiation of inwardly rectifying channels by K^+, particularly in endothelium cells of blood vessels, provides a positive feedback mechanism to supply more blood to more active brain regions. Interestingly, because of the dependence of K_{ir} channels on $PI(4,5)P_2$, this effect is also sensitive to the $G_{q/11}$-coupled GPCR-mediated $PI(4,5)P_2$ dynamics. Furthermore, molecules in the blood-coagulation system, particularly thrombin, regulate voltage-gated ion channels in the brain. Blood coagulation factors released from broken blood vessels (hemorrhage), often caused by stroke and brain trauma, could induce epileptic seizures.[23] Similar to the potentiation of cardiac voltage-gated Na^+

channels by thrombin[24], neuronal Na^+ channels are also potentiated by thrombin.[23] These results suggest that thrombin, a protein that itself is allosterically activated by Na^+ binding,[25] can rapidly potentiate Na^+ channels and cause epileptic seizures that are associated with intracerebral hemorrhage.[23] The exact molecular mechanism underlying the thrombin potentiation of ion channels is uncertain. Essentially, the effect of blood supply to the brain can impact the excitability of neurons through ion-channel mechanisms, leading to significant physiological consequences.

Conclusion

Since Hodgkin and Huxley's discovery of the separate Na^+ and K^+ conductance in nerve action potentials in the 1950s, the study of ion channels has advanced significantly. As of 2023, multiple high-resolution protein structures exist for nearly all channel subfamilies in various states. Ion channels are precisely organized and play a crucial role in dynamic signaling, which is often unique to the animal kingdom. Despite these advancements, significant questions remain regarding the signaling complexes formed by ion channels in native membranes, their conformational dynamics during physiological adaptation, allostery of the multimodal protein regulation, and how they interact with other molecules such as membrane lipids.

References

1. Hille, B. Ion Channels of Excitable Membranes 3rd edn, (Sinauer Associates, 2001).
2. Chowdhury, S., Jarecki, B. W. & Chanda, B. A molecular framework for temperature-dependent gating of ion channels. Cell 158, 1148-1158 (2014). https://doi.org:10.1016/j.cell.2014.07.026
3. Robinson, R. B. & Siegelbaum, S. A. Hyperpolarization-activated cation currents: from molecules to physiological function. Annu Rev Physiol 65, 453-480 (2003). https://doi.org:10.1146/annurev.physiol.65.092101.142734
4. Emery, E. C., Young, G. T., Berrocoso, E. M., Chen, L. & McNaughton, P. A. HCN2 ion channels play a central role in inflammatory and neuropathic pain. Science 333, 1462-1466 (2011). https://doi.org:10.1126/science.1206243
5. Bobin, P. et al. Cyclic nucleotide phosphodiesterases in heart and vessels: A therapeutic perspective. Arch Cardiovasc Dis 109, 431-443 (2016). https://doi.org:10.1016/j.acvd.2016.02.004
6. Butler, A., Tsunoda, S., McCobb, D. P., Wei, A. & Salkoff, L. mSlo, a complex mouse gene encoding "maxi" calcium-activated potassium channels. Science 261, 221-224 (1993). https://doi.org:10.1126/science.7687074
7. Dai, G., Yu, H., Kruse, M., Traynor-Kaplan, A. & Hille, B. Osmoregulatory inositol transporter SMIT1 modulates electrical activity by adjusting PI(4,5)P2 levels. Proc Natl Acad Sci U S A 113, E3290-3299 (2016). https://doi.org:10.1073/pnas.1606348113
8. Ashcroft, F. M. ATP-sensitive potassium channelopathies: focus on insulin secretion. J Clin Invest 115, 2047-2058 (2005). https://doi.org:10.1172/JCI25495
9. Pfaffinger, P. J., Martin, J. M., Hunter, D. D., Nathanson, N. M. & Hille, B. GTP-binding proteins couple cardiac muscarinic receptors to a K channel. Nature 317, 536-538 (1985). https://doi.org:10.1038/317536a0
10. Logothetis, D. E., Kurachi, Y., Galper, J., Neer, E. J. & Clapham, D. E. The βγ subunits of GTP-binding proteins activate the muscarinic K^+ channel in heart. Nature 325, 321-326 (1987). https://doi.org:10.1038/325321a0
11. Herlitze, S. et al. Modulation of Ca^{2+} channels by G-protein βγ subunits. Nature 380, 258-262 (1996). https://doi.org:10.1038/380258a0
12. Badheka, D. et al. Inhibition of Transient Receptor Potential Melastatin 3 ion channels by G-protein βγ subunits. Elife 6 (2017). https://doi.org:10.7554/eLife.26147
13. Hille, B., Dickson, E. J., Kruse, M., Vivas, O. & Suh, B. C. Phosphoinositides regulate ion channels. Biochim Biophys Acta 1851, 844-856 (2015). https://doi.org:10.1016/j.bbalip.2014.09.010
14. Hilgemann, D. W. & Ball, R. Regulation of cardiac Na^+,Ca^{2+} exchange and K_{ATP} potassium channels by PIP_2. Science 273, 956-959 (1996). https://doi.org:10.1126/science.273.5277.956
15. Lewis, R. S. Store-Operated Calcium Channels: From Function to Structure and Back Again. Cold Spring Harb Perspect Biol 12 (2020). https://doi.org:10.1101/cshperspect.a035055
16. Coste, B. et al. Piezo proteins are pore-forming subunits of mechanically activated channels. Nature 483, 176-181 (2012). https://doi.org:10.1038/nature10812
17. Lin, Y. C. et al. Force-induced conformational changes in PIEZO1. Nature 573, 230-234 (2019). https://doi.org:10.1038/s41586-019-1499-2
18. Pan, B. et al. TMC1 Forms the Pore of Mechanosensory Transduction Channels in Vertebrate Inner Ear Hair Cells. Neuron 99, 736-753 e736 (2018). https://doi.org:10.1016/j.neuron.2018.07.033
19. Jeong, H. et al. Structures of the TMC-1 complex illuminate mechanosensory transduction. Nature 610, 796-803 (2022). https://doi.org:10.1038/s41586-022-05314-8
20. Jin, P., Jan, L. Y. & Jan, Y. N. Mechanosensitive Ion Channels: Structural Features Relevant to Mechanotransduction Mechanisms. Annu Rev Neurosci 43, 207-229 (2020). https://doi.org:10.1146/annurev-neuro-070918-050509
21. Hilgemann, D. W. et al. Lipid signaling to membrane proteins: From second messengers to membrane domains and adapter-free endocytosis. J Gen Physiol 150, 211-224 (2018). https://doi.org:10.1085/jgp.201711875
22. Longden, T. A., Hill-Eubanks, D. C. & Nelson, M. T. Ion channel networks in the control of cerebral blood flow. J Cereb Blood Flow Metab 36, 492-512 (2016). https://doi.org:10.1177/0271678X15616138
23. Isaeva, E., Hernan, A., Isaev, D. & Holmes, G. L. Thrombin facilitates seizures through activation of persistent sodium current. Ann Neurol 72, 192-198 (2012). https://doi.org:10.1002/ana.23587
24. Pinet, C., Le Grand, B., John, G. W. & Coulombe, A. Thrombin facilitation of voltage-gated sodium channel activation in human cardiomyocytes: implications for ischemic sodium loading. Circulation 106, 2098-2103 (2002). https://doi.org:10.1161/01.cir.0000034510.64828.96
25. Di Cera, E. Thrombin: a paradigm for enzymes allosterically activated by monovalent cations. C R Biol 327, 1065-1076 (2004). https://doi.org:10.1016/j.crvi.2004.07.011

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Disclosure

Figures were created using BioRender. **MM**

Uncovering Critical Roles for RNA in Neurodegeneration

by Yuna M. Ayala, PhD

The identification of previously uncovered regulatory factors will highlight new mechanisms in fundamental biological processes and innovative strategies for therapy against neurodegenerative disease.



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Abstract

RNA-binding proteins, in particular TDP-43, are key players in neurodegenerative disorders, mainly amyotrophic lateral sclerosis and frontotemporal dementia. We aim to elucidate how TDP-43 dysfunction alters cell metabolism and to identify mechanisms linked to aberrant behavior. We find that RNA binding plays a key role in maintaining TDP-43 homeostasis and in controlling cellular organization, two processes of essential importance to TDP-43 pathology. This research will provide insight into pathogenesis and help develop therapeutic interventions.

Significance of TDP-43 in Neurodegeneration

Investigation of RNA-binding protein biology has surged because of its connection to neurodegeneration. In particular, the RNA-binding protein TDP-43, or TAR DNA binding protein, is a key player in neurotoxicity associated with two devastating neurodegenerative disorders, amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD). ALS and FTD are viewed as a disease spectrum for which little therapeutic intervention and diagnostic tools are available. The accumulation of TDP-43 aggregates in the cytoplasm of neurons and glial cells, accompanied by decreased TDP-43 activity, are hallmark features of these disorders.[1] In addition to its key role in ALS and FTD, TDP-43 is also implicated in other neurodegenerative diseases. TDP-43 pathology is observed in approximately half of Alzheimer's disease (AD) cases,[2-6] chronic traumatic encephalopathy (CTE)[7] and multisystem proteinopathy (MSP).[8] Importantly, the prevalence of TDP-43 pathology is widespread in Limbic-predominant Age-related TDP-43 Encephalopathy (LATE), an AD-like disorder affecting the oldest population.[9] TDP-43 mutations are causative of 2-5% of familial ALS. However, TDP-43 aggregates with or without TDP-43 mutations accumulate in almost all ALS patients.

Following the discovery of TDP-43 pathology in ALS and FTD, the list of RNA-associated proteins shown to aggregate and found to have disease-linked mutations has steadily increased, underscoring the importance of RNA metabolism and of RNA-binding protein function in disease. This class of proteins is now recognized to undergo assembly processes that may increase their susceptibility

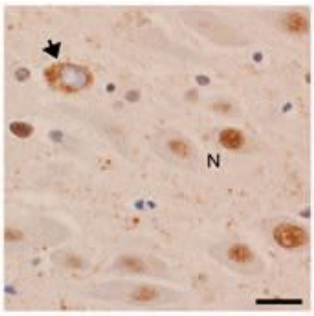


Figure 1. Cytoplasmic TDP-43 aggregates and loss of nuclear localization are characteristic features of pathology in TDP-43 proteinopathies. Immunohistochemistry of ALS spinal cord motor neurons showing aberrant cytoplasmic TDP-43 mislocalization, corresponding to loss of nuclear protein detection (arrow). Compare this to normal TDP-43 staining predominantly in the nucleus (N). Scale, 20 µm. Courtesy of Dr. Dennis Dickson (Mayo Clinic, Jacksonville).

to aggregation. RNA-binding proteins linked to ALS/FTD, including TDP-43, undergo a process termed liquid-liquid phase separation (LLPS) or biomolecular condensation. LLPS mediates the formation of membraneless organelles in eukaryotic and prokaryotic cells through an assembly process/condensation that generates a dense phase in equilibrium with a more dilute phase. This mechanism underlies the biogenesis, function and cellular organization of RNA and protein components in ribonucleoprotein (RNP) granules.[10] Importantly, aberrant LLPS results in condensates that are less dynamic and acquire solid-like properties leading to the accumulation of irreversible aggregates. Therefore, uncovering the mechanisms that control LLPS will contribute to developing potential therapeutic tools for the associated neurodegenerative disorders.

Accumulation of intracellular, cytoplasmic TDP-43 aggregates correlates with the loss of normal nuclear localization of the protein,[1,11] suggesting that pathology also results from a loss of TDP-43 function (Figure 1). The mechanisms by which TDP-43 aggregates are linked to decreased nuclear localization are unknown. One prevalent idea is that accumulation of the cytoplasmic TDP-43 inclusions sequesters nuclear protein. This is supported by the ability of TDP-43 aggregates to spread through prion-like mechanisms, as shown in vitro and in cell-based models.[12] Moreover, treatment of a neuronal-like model with exogenous pre-formed aggregates seeds cytoplasmic inclusions, resulting in loss of nuclear TDP-43.[13] The consequences of decreased nuclear TDP-43 in disease were demonstrated by Liu et al., whereby FTD cells with lower nuclear TDP-43 accumulate widespread changes in gene expression, including alterations in many TDP-43-regulated transcripts.[14] TDP-43 is highly conserved among higher eukaryotes and its loss is deleterious and lethal in cells and in multiple in vivo models.[15-18 19,20 21-24] TDP-43 expression is also essential in neurons, as demonstrated by the toxicity and cell death caused by TDP-43 knockdown.[20,25] Altogether, these findings underscore the urgency to prevent or reverse TDP-43 aggregate accumulation because of its potential intrinsic toxicity and also because of the importance of restoring TDP-43 nuclear function.

The Role of RNA Binding on TDP-43 Solubility and Condensation

One of the major interests in our lab is to elucidate the mechanism(s) of TDP-43 aggregation, including identifying structures of intermediates that participate in this process, which remain largely unknown. Using purified full-length TDP-43, we showed that TDP-43 assembles into misfolded oligomeric species during the early phase of aggregation.[12] These oligomeric aggregates are different from soluble, functional TDP-43 higher order assemblies. The oligomeric aggregates represent misfolded species whose accumulation is irreversible and resistant to denaturing conditions. Ordered oligomerization upon misfolding of proteins linked to neurodegeneration has been previously observed. These oligomers, such as in the case of alpha-synuclein and tau, precede the larger species detected as inclusions in pathology.[26-29] These aggregate species are believed to drive neurodegeneration because they are capable of seeding prion-like conversion and show greater cytotoxicity compared to larger aggregates. In the case of TDP-43, we similarly observed that oligomeric species seed de novo aggregation in human cells and are necessary for the formation of larger TDP-43 aggregates.[12] In our efforts to determine the mechanism of TDP-43 aggregate formation, we discovered that the RNA recognition motifs (RRMs) (Figure 2) are crucial

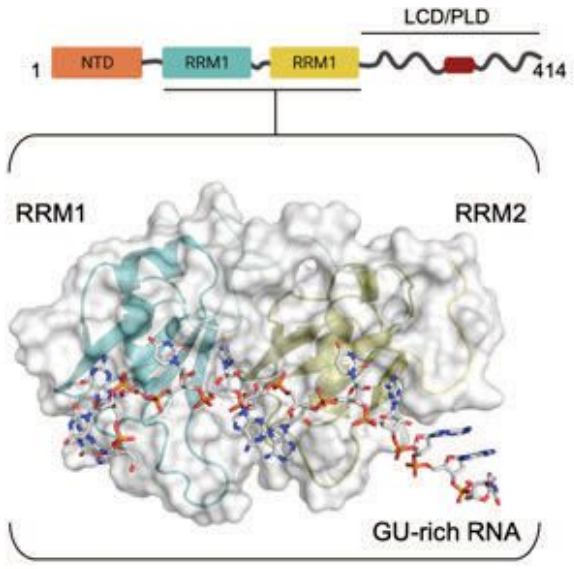


Figure 2. TDP-43 binds with strong preference to GU-rich RNA. TDP-43 is organized into three independently folded regions: N-terminal domain (NTD) and two RNA recognition motifs (RRMs). The mostly disordered C-terminal region is referred to as low complexity domain (LCD) or prion-like domain (PLD). The solution structure of RRM1 and 2 bound to GU-rich RNA was solved by NMR, showing multiple sequence-specific contacts.[8]

in triggering protein misfolding. Furthermore, we found that GU-rich RNA molecules specifically, which bind TDP-43 with high affinity, potently inhibit TDP-43 aggregation. This is observed using purified protein as well as in human cells. These findings were unexpected, as most of the aggregation-prone function had been previously focused on the disordered C-terminal tail of TDP-43. We also demonstrated that GU-rich RNA increases the liquid/dynamic properties of TDP-43 condensates that form through LLPS.[30] Our results help explain how RNA-binding proteins that are prone to aggregation maintain solubility under physiological conditions. Additionally, these findings help explain why TDP-43 accumulation in the cytoplasm, where RNA is less abundant relative to the nucleus, increases TDP-43 aggregation propensity. These findings point to a crucial and previously unrecognized role of RNA binding in preventing the misfolding of partner proteins. One of our primary goals is to understand how specific RNA binding increases the liquid properties of TDP-43 condensates and inhibits aggregation. We will harness this information to develop strategies in preventing and decreasing TDP-43 pathology.

Regulation of TDP-43 Function and Nuclear-Cytoplasmic Distribution

TDP-43 binds to hundreds of coding and non-coding RNA transcripts identified in human cells and mouse brain and these interactions control RNA processing and gene expression.[31,32] TDP-43 modulates multiple RNA processing mechanisms, although its most well-established function is regulation of RNA splicing and polyadenylation.[31,33,34] The majority of TDP-43 targets are composed of long GU-rich sequences and GU dinucleotide repeats.[31,35,36] Based on previous biochemical and structural data,[37,38] this sequence composition suggests that these transcripts accommodate multiple TDP-43 molecules. Such complexes are likely to form condensates according to our recent work. We demonstrated that GU-repeat RNA promotes TDP-43 LLPS in a sequence and length-dependent manner, whereby increasing the number of binding sites strongly induces TDP-43 condensation.[30] These RNA-bound condensates form through multivalent interactions that also require TDP-43 N-terminal domain (NTD) oligomerization and assembly of the low complexity C-terminal domain (CTD). Therefore, our results predict that TDP-43 association with many of its RNA targets involves, and may depend on, condensate formation. Consistent with this idea data from our lab and others indicate that RNA-mediated TDP-43 condensation is key in controlling RNA processing function.[39,40]

TDP-43 is predominantly localized in the nucleus of normal cells, although it shuttles continuously between nuclear and cytoplasmic compartments.[41] As earlier discussed, the distribution of TDP-43 between these compartments is important in maintaining TDP-43 proteostasis and preventing aggregation. Specifically, increased cytoplasmic accumulation is linked to TDP-43 aggregation and neuronal

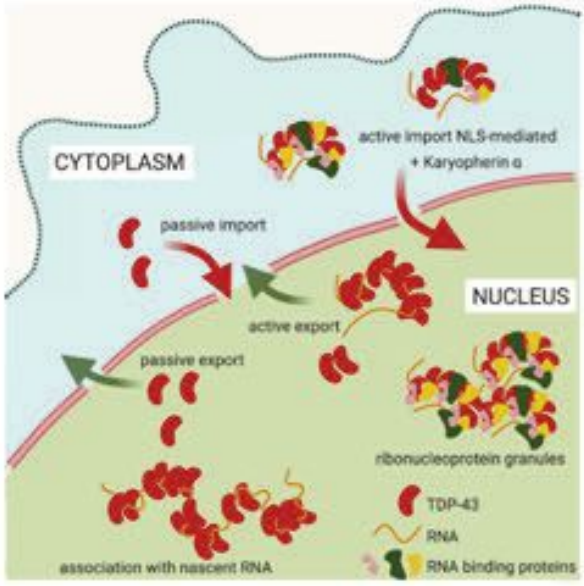


Figure 3. Regulation of TDP-43 nuclear-cytoplasmic trafficking in cells. Based on our findings and others, we postulate that TDP-43 nuclear import and exit are regulated by active and passive mechanisms that are size dependent. In particular, we find that RNA binding largely contributes to nuclear retention through the formation of large ribonucleoprotein assemblies.

cell death, as seen in cell and in vivo models.[42,43] Cytoplasmic TDP-43 is more susceptible to proteotoxic conditions, such as inhibition of degradation pathways and seeding with pre-formed TDP-43 aggregates.[12,44,45] The effect of increased cytoplasmic TDP-43 pathology, accompanied by the loss of nuclear localization in disease,[1,11] is recapitulated in a mouse model overexpressing cytoplasmic human TDP-43 in neurons.[46] These mice show strong neurotoxicity and shortened lifespan. The significance of TDP-43 nuclear-cytoplasmic trafficking in pathogenesis is further underscored by evidence that nuclear pore/receptor factors are disrupted in ALS-FTD patients with TDP-43 pathology.[47,48] In addition, impaired nuclear import is observed in the most common inherited mutation in ALS and FTD patients, caused by C9Orf72 hexanucleotide repeat expansion.[49-52] These alterations disrupt the cellular dynamics of RNA-binding proteins and may at least partially explain the accumulation of cytoplasmic TDP-43 pathology linked to the C9Orf72 mutation. The collection of these findings strongly suggests that abnormally elevated TDP-43 concentration in the cytoplasm promotes TDP-43 pathology through enhanced aggregation and loss of function.

Despite its importance, the regulation of TDP-43 nuclear import, nuclear retention and export remain ill-defined. Improved understanding of the control of TDP-43 nucleo/cytoplasmic dynamics will provide new avenues to restore TDP-43 cellular proteostasis as a potential therapeutic approach (Figure 3). A classic basic bipartite nuclear localization signal (NLS) in the linker region between the NTD and RRM1 mediates TDP-43 nuclear import[53]. The NLS binds the nuclear-import receptor importin α (karyopherin α)[54]. Importin α and karyopherin β1 not only mediate TDP-43 nuclear import, but also act as chaperones and are able to prevent aggregation of purified TDP-43.[55] This suggests that nuclear transport receptor binding to TDP-43 is a protective mechanism preventing unfavorable interactions and perhaps masking sensitive TDP-43 regions until its import to the nucleus. Curiously, the NLS may be only partially responsible for TDP-43 nuclear import, as significant levels of nuclear TDP-43 are observed despite disruption of the signal in various cell models and transgenic mice.[41,44,56-58] The partial nuclear localization of the TDP-43 NLS-defective mutant is also observed upon knockdown of endogenous TDP-43, indicating that NLS-deficient TDP-43 is not imported to the nucleus through association with endogenous TDP-43.[58] This suggests the existence of an additional nuclear import signal. Alternatively, considering that the molecular mass of a single TDP-43 molecule is at the limit size for passive trafficking through the nuclear pore, it is possible that a portion of TDP-43 molecules passively diffuses through the pore, as suggested.[58-60] In this case, active nuclear import may be necessary to transport higher order TDP-43 assemblies, including RNA and protein-bound complexes. A nuclear export signal (NES) proposed to bind the export factor exportin 1 (XPO1/CRM1) was previously mapped to RRM2, using sequence comparison.[53] However, this idea has been revised based on evidence from multiple

groups showing that the putative NES is not required for nuclear exit and does not significantly bind the nuclear export protein exportin 1 (XPO1).[58,61] Instead, TDP-43 seems to undergo passive nuclear egress that is size-dependent.[58,60] We previously found that transcription inhibition, or disruption of RNA-binding function significantly promotes TDP-43 nuclear exit.[41] This and more recent evidence,[62] including our work,[63] support the intriguing model that RNA interactions are capable of modulating TDP-43 nuclear-cytoplasmic trafficking. According to this idea, recruitment of TDP-43 to nuclear RNA transcripts composed of multiple binding sites generates large macromolecular assemblies that are unable to passively go through the nuclear pore. In agreement with this hypothesis, addition of high levels of GU-repeat RNA ($(GU)_6$), a short oligonucleotide predicted to bind one molecule of the protein, increases cytoplasmic TDP-43.[62] This result suggests that $(GU)_6$ competes with nuclear transcripts for TDP-43 binding and increases the pool of monomeric protein passively diffusing through the nuclear pore. Interestingly, the majority of long non-coding RNAs (lncRNAs) express nuclear retention signals usually encoded by internal transposable elements, which promote nuclear localization[64] (reviewed in[65]). Deletion of a transposable element in lncRNA Malat1, which directly binds TDP-43, promotes TDP-43 export to the cytoplasm and increases the formation of TDP-43 inclusions in this compartment.[6]. Therefore, it is possible that TDP-43 association with nascent RNA and recruitment to RNA molecules with nuclear retention properties (e.g., lncRNAs) maintain high nuclear TDP-43 levels.[67]. Altogether, these findings provide strong evidence that RNA binding, through the formation of higher order macromolecular complexes, e.g., RNP assemblies, plays a critical role in controlling nuclear-cytoplasmic TDP-43 distribution. We are currently investigating how factors controlling RNP complex biogenesis modulate TDP-43 nuclear-cytoplasmic trafficking. The identification of previously uncovered regulatory factors will highlight new mechanisms in fundamental biological processes and innovative strategies for therapy against neurodegenerative disease.

References

1. Neumann, M., Sampathu, D. M., Kwong, L. K., Truax, A. C., Micsenyi, M. C., Chou, T. T., Bruce, J., Schuck, T., Grossman, M., Clark, C. M., McCluskey, L. F., Miller, B. L., Masliah, E., Mackenzie, I. R., Feldman, H., Feiden, W., Kretzschmar, H. A., Trojanowski, J. Q. & Lee, V. M. Ubiquitinated TDP-43 in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Science 314, 130-133 (2006).
2. Amador-Ortiz, C., Lin, W. L., Ahmed, Z., Personett, D., Davies, P., Duara, R., Graff-Radford, N. R., Hutton, M. L. & Dickson, D. W. TDP-43 immunoreactivity in hippocampal sclerosis and Alzheimer's disease. Ann Neurol 61, 435-445, doi:10.1002/ana.21154 (2007).
3. Josephs, K. A., Murray, M. E., Whitwell, J. L., Parisi, J. E., Petrucelli, L., Jack, C. R., Petersen, R. C. & Dickson, D. W. Staging TDP-43 pathology in Alzheimer's disease. Acta Neuropathol 127, 441-450, doi:10.1007/s00401-013-1211-9 (2014).
4. Robinson, J. L., Lee, E. B., Xie, S. X., Rennert, L., Suh, E., Bredenberg, C., Caswell, C., Van Deerlin, V. M., Yan, N., Yousef, A., Hurtig, H. I., Siderowf, A., Grossman, M., McMillan, C. T., Miller, B., Duda, J. E., Irwin, D. J., Wolk, D., Elman, L., McCluskey, L., Chen-Plotkin, A., Weintraub, D., Arnold, S. E., Brettschneider, J., Lee, V. M. & Trojanowski, J. Q. Neurodegenerative disease concomitant proteinopathies are prevalent, age-related and APOE4-associated. Brain 141, 2181-2193, doi:10.1093/brain/awy146 (2018).
5. Wennberg, A. M., Tosakulwong, N., Lesnick, T. G., Murray, M. E., Whitwell, J. L., Liesinger, A. M., Petrucelli, L., Boeve, B. F., Parisi, J. E., Knopman, D. S., Petersen, R. C., Dickson, D. W. & Josephs, K. A. Association of Apolipoprotein E epsilon4 With Transactive Response DNA-Binding Protein 43. JAMA Neurol 75, 1347-1354, doi:10.1001/jamaneurol.2018.3139 (2018).
6. Yang, H. S., Yu, L., White, C. C., Chibnik, L. B., Chhatwal, J. P., Sperling, R. A., Bennett, D. A., Schneider, J. A. & De Jager, P. L. Evaluation of TDP-43 proteinopathy and hippocampal sclerosis in relation to APOE epsilon4 haplotype status: a community-based cohort study. Lancet Neurol 17, 773-781, doi:10.1016/S1474-4422(18)30251-5 (2018).
7. McKee, A. C., Gavett, B. E., Stern, R. A., Nowinski, C. J., Cantu, R. C., Kowall, N. W., Perl, D. P., Hedley-Whyte, E. T., Price, B., Sullivan, C., Morin, P., Lee, H. S., Kubilus, C. A., Daneshvar, D. H., Wulff, M. & Budson, A. E. TDP-43 proteinopathy and motor neuron disease in chronic traumatic encephalopathy. J Neuropathol Exp Neurol 69, 918-929, doi:10.1097/NEN.0b013e3181ee7d85 (2010).
8. Weihl, C. C., Pestronk, A. & Kimonis, V. E. Valosin-containing protein disease: inclusion body myopathy with Paget's disease of the bone and fronto-temporal dementia. Neuromuscul Disord 19, 308-315, doi:10.1016/j.nmd.2009.01.009 (2009).
9. Nelson, P. T., Dickson, D. W., Trojanowski, J. Q., Jack, C. R., Boyle, P. A., Arfanakis, K., Rademakers, R., Alafuzoff, I., Attems, J., Brayne, C., Coyle-Gilchrist, I. T. S., Chui, H. C., Fardo, D. W., Flanagan, M. E., Halliday, G., Hokkanen, S. R. K., Hunter, S., Jicha, G. A., Katsumata, Y., Kawas, C. H., Keene, C. D., Kovacs, G. G., Kukull, W. A., Levey, A. I., Makkinejad, N., Montine, T. J., Murayama, S., Murray, M. E., Nag, S., Rissman, R. A., Seeley, W. W., Sperling, R. A., White, C. L., 3rd, Yu, L. & Schneider, J. A. Limbic-predominant age-related TDP-43 encephalopathy (LATE): consensus working group report. Brain 142, 1503-1527, doi:10.1093/brain/awz099 (2019).
10. Strom, A. R. & Brangwynne, C. P. The liquid nucleome - phase transitions in the nucleus at a glance. J Cell Sci 132, doi:10.1242/jcs.235093 (2019).
11. Arai, T., Hasegawa, M., Akiyama, H., Ikeda, K., Nonaka, T., Mori, H., Mann, D., Tsuchiya, K., Yoshida, M., Hashizume, Y. & Oda, T. TDP-43 is a component of ubiquitin-positive tau-negative inclusions in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Biochem Biophys Res Commun 351, 602-611 (2006).
12. French, R. L., Grese, Z. R., Aligireddy, H., Dhavale, D. D., Reeb, A. N., Kedia, N., Kotzbauer, P. T., Bieschke, J. & Ayala, Y. M. Detection of TAR DNA-binding protein 43 (TDP-43) oligomers as

initial intermediate species during aggregate formation. J Biol Chem 294, 6696-6709, doi:10.1074/jbc.RA118.005889 (2019).
13. Gasset-Rosa, F., Lu, S., Yu, H., Chen, C., Melamed, Z., Guo, L., Shorter, J., Da Cruz, S. & Cleveland, D. W. Cytoplasmic TDP-43 De-mixing Independent of Stress Granules Drives Inhibition of Nuclear Import, Loss of Nuclear TDP-43, and Cell Death. Neuron 102, 339-357 e337, doi:10.1016/j.neuron.2019.02.038 (2019).
14. Liu, E. Y., Russ, J., Cali, C. P., Phan, J. M., Amlie-Wolf, A. & Lee, E. B. Loss of Nuclear TDP-43 Is Associated with Decondensation of LINE Retrotransposons. Cell Rep 27, 1409-1421 e1406, doi:10.1016/j.celrep.2019.04.003 (2019).
15. Wood, M., Quinet, A., Lin, Y. L., Davis, A. A., Pasero, P., Ayala, Y. M. & Vindigni, A. TDP-43 dysfunction results in R-loop accumulation and DNA replication defects. J Cell Sci 133, doi:10.1242/jcs.244129 (2020).
16. Wu, L. S., Cheng, W. C., Hou, S. C., Yan, Y. T., Jiang, S. T. & Shen, C. K. TDP-43, a neuro-pathosignature factor, is essential for early mouse embryogenesis. Genesis 48, 56-62, doi:10.1002/dvg.20584 (2010).
17. Kraemer, B. C., Schuck, T., Wheeler, J. M., Robinson, L. C., Trojanowski, J. Q., Lee, V. M. & Schellenberg, G. D. Loss of murine TDP-43 disrupts motor function and plays an essential role in embryogenesis. Acta neuropathologica 119, 409-419, doi:10.1007/s00401-010-0659-0 (2010).
18. Sephton, C. F., Good, S. K., Atkin, S., Dewey, C. M., Mayer, P., 3rd, Herz, J. & Yu, G. TDP-43 is a developmentally regulated protein essential for early embryonic development. The Journal of biological chemistry 285, 6826-6834, doi:10.1074/jbc.M109.061846 (2010).
19. Chiang, P. M., Ling, J., Jeong, Y. H., Price, D. L., Aja, S. M. & Wong, P. C. Deletion of TDP-43 down-regulates Tbc1d1, a gene linked to obesity, and alters body fat metabolism. Proceedings of the National Academy of Sciences of the United States of America 107, 16320-16324, doi:10.1073/pnas.1002176107 (2010).
20. Iguchi, Y., Katsuno, M., Niwa, J., Takagi, S., Ishigaki, S., Ikenaka, K., Kawai, K., Watanabe, H., Yamanaka, K., Takahashi, R., Misawa, H., Sasaki, S., Tanaka, F. & Sobue, G. Loss of TDP-43 causes age-dependent progressive motor neuron degeneration. Brain 136, 1371-1382, doi:10.1093/brain/awt029 (2013).
21. Feiguin, F., Godena, V. K., Romano, G., D'Ambrogio, A., Klima, R. & Baralle, F. E. Depletion of TDP-43 affects Drosophila motoneurons terminal synapsis and locomotive behavior. FEBS Lett 583, 1586-1592, doi:S0014-5793(09)00298-1 [pii] 10.1016/j.febslet.2009.04.019 (2009).
22. Diaper, D. C., Adachi, Y., Sutcliffe, B., Humphrey, D. M., Elliott, C. J., Stepto, A., Ludlow, Z. N., Vanden Broeck, L., Callaerts, P., Dermaut, B., Al-Chalabi, A., Shaw, C. E., Robinson, I. M. & Hirth, F. Loss and gain of Drosophila TDP-43 impair synaptic efficacy and motor control leading to age-related neurodegeneration by loss-of-function phenotypes. Hum Mol Genet 22, 1539-1557, doi:10.1093/hmg/ddt005 (2013).
23. Bose, P., Armstrong, G. A. B. & Drapeau, P. Neuromuscular junction abnormalities in a zebrafish loss-of-function model of TDP-43. J Neurophysiol 121, 285-297, doi:10.1152/jn.00265.2018 (2019).
24. Zhang, T., Hwang, H. Y., Hao, H., Talbot, C., Jr. & Wang, J. Caenorhabditis elegans RNA-processing protein TDP-1 regulates protein homeostasis and life span. J Biol Chem 287, 8371-8382, doi:10.1074/jbc.M111.311977 (2012).
25. Wu, L. S., Cheng, W. C. & Shen, C. K. Targeted depletion of TDP-43 expression in the spinal cord motor neurons leads to the development of amyotrophic lateral sclerosis-like phenotypes in mice. J Biol Chem 287, 27335-27344, doi:10.1074/jbc.M112.359000 (2012).
26. Conway, K. A., Harper, J. D. & Lansbury, P. T. Accelerated in vitro fibril formation by a mutant alpha-synuclein linked to early-onset Parkinson disease. Nat Med 4, 1318-1320, doi:10.1038/3311 (1998).
27. Sanchez, I., Mahlke, C. & Yuan, J. Pivotal role of oligomerization in expanded polyglutamine neurodegenerative disorders. Nature 421, 373-379, doi:10.1038/nature01301 (2003).
28. Lesne, S., Koh, M. T., Kotilinek, L., Kayed, R., Glabe, C. G., Yang, A., Gallagher, M. & Ashe, K. H. A specific amyloid-beta protein assembly in the brain impairs memory. Nature 440, 352-357, doi:10.1038/nature04533 (2006).
29. Klyubin, I., Walsh, D. M., Lemere, C. A., Cullen, W. K., Shankar, G. M., Betts, V., Spooner, E. T., Jiang, L., Anwyl, R., Selkoe, D. J. & Rowan, M. J. Amyloid beta protein immunotherapy neutralizes Abeta oligomers that disrupt synaptic plasticity in vivo. Nat Med 11, 556-561, doi:10.1038/nm1234 (2005).
30. Grese, Z. R., Bastos, A. C., Mamede, L. D., French, R. L., Miller, T. M. & Ayala, Y. M. Specific RNA interactions promote TDP-43 multivalent phase separation and maintain liquid properties. EMBO Rep 22, e53632, doi:10.15252/embr.202153632 (2021).
31. Tollervey, J. R., Curk, T., Rogelj, B., Briese, M., Cereda, M., Kayikci, M., Konig, J., Hortobagyi, T., Nishimura, A. L., Zupunski, V., Patani, R., Chandran, S., Rot, G., Zupan, B., Shaw, C. E. & Ule, J. Characterizing the RNA targets and position-dependent splicing regulation by TDP-43. Nat Neurosci 14, 452-458, doi:10.1038/nn.2778 (2011).
32. Polymenidou, M., Lagier-Tourenne, C., Hutt, K. R., Huelga, S. C., Moran, J., Liang, T. Y., Ling, S. C., Sun, E., Wancewicz, E., Mazur, C., Kordasiewicz, H., Sedaghat, Y., Donohue, J. P., Shiue, L., Bennett, C. F., Yeo, G. W. & Cleveland, D. W. Long pre-mRNA depletion and RNA missplicing contribute to neuronal vulnerability from loss of TDP-43. Nat Neurosci 14, 459-468, doi:10.1038/nn.2779 (2011).
33. Rot, G., Wang, Z., Huppertz, I., Modic, M., Lence, T., Hallegger, M., Haberman, N., Curk, T., von Mering, C. & Ule, J. High-Resolution RNA Maps Suggest Common Principles of Splicing and Polyadenylation Regulation by TDP-43. Cell Rep 19, 1056-1067, doi:10.1016/j.celrep.2017.04.028 (2017).
34. Ling, J. P., Pletnikova, O., Troncoso, J. C. & Wong, P. C. TDP-43 repression of nonconserved cryptic exons is compromised in ALS-FTD. Science 349, 650-655, doi:10.1126/science.aab0983 (2015).
35. Sephton, C. F., Cenik, C., Kucukural, A., Dammer, E. B., Cenik, B., Han, Y., Dewey, C. M., Roth, F. P., Herz, J., Peng, J., Moore, M. J. & Yu, G. Identification of neuronal RNA targets of TDP-43-containing ribonucleoprotein complexes. The Journal of biological chemistry 286, 1204-1215, doi:10.1074/jbc.M110.190884 (2011).
36. Polymenidou, M. & Cleveland, D. W. The seeds of neurodegeneration: prion-like spreading in ALS. Cell 147, 498-508, doi:10.1016/j.cell.2011.10.011 (2011).
37. Ayala, Y. M., Pantano, S., D'Ambrogio, A., Buratti, E., Brindisi, A., Marchetti, C., Romano, M. & Baralle, F. E. Human, Drosophila, and C.elegans TDP43: nucleic acid binding properties and splicing regulatory function. J Mol Biol 348, 575-588, doi:10.1016/j.jmb.2005.02.038 (2005).
38. Lukavsky, P. J., Daujotyte, D., Tollervey, J. R., Ule, J., Stuani, C., Buratti, E., Baralle, F. E., Damberger, F. F. & Allain, F. H. Molecular basis of UG-rich RNA recognition by the human splicing factor TDP-43. Nat Struct Mol Biol, doi:10.1038/nsmb.2698 (2013).
39. Koehler, L. C., Grese, Z. R., Bastos, A. C. S., Mamede, L. D., Heyduk, T. & Ayala, Y. M. TDP-43 Oligomerization and Phase Separation Properties Are Necessary for Autoregulation. Frontiers in Neuroscience 16, doi:10.3389/fnins.2022.818655 (2022).
40. Hallegger, M., Chakrabarti, A. M., Lee, F. C. Y., Lee, B. L., Amalietti, A. G., Odeh, H. M., Copley, K. E., Rubien, J. D., Portz, B., Kuret, K., Huppertz, I., Rau, F., Patani, R., Fawzi, N. L., Shorter, J., Luscombe, N. M. & Ule, J. TDP-43 condensation properties specify its RNA-binding and regulatory repertoire. Cell 184, 4680-4696 e4622, doi:10.1016/j.cell.2021.07.018 (2021).
41. Ayala, Y. M., Zago, P., D'Ambrogio, A., Xu, Y. F., Petrucelli, L., Buratti, E. & Baralle, F. E. Structural determinants of the cellular localization and shuttling of TDP-43. J Cell Sci 121, 3778-3785, doi:10.1242/jcs.038950 (2008).
42. Igaz, L. M., Kwong, L. K., Lee, E. B., Chen-Plotkin, A., Swanson, E., Unger, T., Malunda, J., Xu, Y., Winton, M. J., Trojanowski, J. Q.

& Lee, V. M. Dysregulation of the ALS-associated gene TDP-43 leads to neuronal death and degeneration in mice. The Journal of clinical investigation 121, 726-738, doi:10.1172/JCI44867 (2011).
43. Barmada, S. J., Skibinski, G., Korb, E., Rao, E. J., Wu, J. Y. & Finkbeiner, S. Cytoplasmic mislocalization of TDP-43 is toxic to neurons and enhanced by a mutation associated with familial amyotrophic lateral sclerosis. J Neurosci 30, 639-649, doi:30/2/639 [pii]
10.1523/JNEUROSCI.4988-09.2010.
44. Scotter, E. L., Vance, C., Nishimura, A. L., Lee, Y. B., Chen, H. J., Urwin, H., Sardone, V., Mitchell, J. C., Rogelj, B., Rubinsztein, D. C. & Shaw, C. E. Differential roles of the ubiquitin proteasome system and autophagy in the clearance of soluble and aggregated TDP-43 species. Journal of cell science 127, 1263-1278, doi:10.1242/jcs.140087 (2014).
45. Porta, S., Xu, Y., Restrepo, C. R., Kwong, L. K., Zhang, B., Brown, H. J., Lee, E. B., Trojanowski, J. Q. & Lee, V. M. Patient-derived frontotemporal lobar degeneration brain extracts induce formation and spreading of TDP-43 pathology in vivo. Nat Commun 9, 4220, doi:10.1038/s41467-018-06548-9 (2018).
46. Walker, A. K., Spiller, K. J., Ge, G., Zheng, A., Xu, Y., Zhou, M., Tripathy, K., Kwong, L. K., Trojanowski, J. Q. & Lee, V. M. Functional recovery in new mouse models of ALS/FTLD after clearance of pathological cytoplasmic TDP-43. Acta Neuropathol 130, 643-660, doi:10.1007/s00401-015-1460-x (2015).
47. Chou, C. C., Zhang, Y., Umoh, M. E., Vaughan, S. W., Lorenzini, I., Liu, F., Sayegh, M., Donlin-Asp, P. G., Chen, Y. H., Duong, D. M., Seyfried, N. T., Powers, M. A., Kukar, T., Hales, C. M., Gearing, M., Cairns, N. J., Boylan, K. B., Dickson, D. W., Rademakers, R., Zhang, Y. J., Petrucelli, L., Sattler, R., Zarnescu, D. C., Glass, J. D. & Rossoll, W. TDP-43 pathology disrupts nuclear pore complexes and nucleocytoplasmic transport in ALS/FTD. Nat Neurosci 21, 228-239, doi:10.1038/s41593-017-0047-3 (2018).
48. Kim, H. J. & Taylor, J. P. Lost in Transportation: Nucleocytoplasmic Transport Defects in ALS and Other Neurodegenerative Diseases. Neuron 96, 285-297, doi:10.1016/j.neuron.2017.07.029 (2017).
49. Freibaum, B. D., Lu, Y., Lopez-Gonzalez, R., Kim, N. C., Almeida, S., Lee, K. H., Badders, N., Valentine, M., Miller, B. L., Wong, P. C., Petrucelli, L., Kim, H. J., Gao, F. B. & Taylor, J. P. GGGGCC repeat expansion in C9orf72 compromises nucleocytoplasmic transport. Nature 525, 129-133, doi:10.1038/nature14974 (2015).
50. Jovicic, A., Mertens, J., Boeynaems, S., Bogaert, E., Chai, N., Yamada, S. B., Paul, J. W., 3rd, Sun, S., Herdy, J. R., Bieri, G., Kramer, N. J., Gage, F. H., Van Den Bosch, L., Robberecht, W. & Gitler, A. D. Modifiers of C9orf72 dipeptide repeat toxicity connect nucleocytoplasmic transport defects to FTD/ALS. Nature neuroscience 18, 1226-1229, doi:10.1038/nn.4085 (2015).
51. Zhang, K., Donnelly, C. J., Haeusler, A. R., Grima, J. C., Machamer, J. B., Steinwald, P., Daley, E. L., Miller, S. J., Cunningham, K. M., Vidensky, S., Gupta, S., Thomas, M. A., Hong, I., Chiu, S. L., Huganir, R. L., Ostrow, L. W., Matunis, M. J., Wang, J., Sattler, R., Lloyd, T. E. & Rothstein, J. D. The C9orf72 repeat expansion disrupts nucleocytoplasmic transport. Nature 525, 56-61, doi:10.1038/nature14973 (2015).
52. Jovicic, A., Paul, J. W., 3rd & Gitler, A. D. Nuclear transport dysfunction: a common theme in amyotrophic lateral sclerosis and frontotemporal dementia. J Neurochem 138 Suppl 1, 134-144, doi:10.1111/jnc.13642 (2016).
53. Winton, M. J., Igaz, L. M., Wong, M. M., Kwong, L. K., Trojanowski, J. Q. & Lee, V. M. Disturbance of Nuclear and Cytoplasmic TAR DNA-binding Protein (TDP-43) Induces Disease-like Redistribution, Sequestration, and Aggregate Formation. J Biol Chem 283, 13302-13309 (2008).
54. Nishimura, A. L., Zupunski, V., Troakes, C., Kathe, C., Fratta, P., Howell, M., Gallo, J. M., Hortobagyi, T., Shaw, C. E. & Rogelj, B. Nuclear import impairment causes cytoplasmic trans-activation response DNA-binding protein accumulation and is associated with frontotemporal lobar degeneration. Brain 133, 1763-1771, doi:10.1093/brain/awq111 (2010).
55. Guo, L., Kim, H. J., Wang, H., Monaghan, J., Freyermuth, F., Sung, J. C., O'Donovan, K., Fare, C. M., Diaz, Z., Singh, N., Zhang, Z. C., Coughlin, M., Sweeny, E. A., DeSantis, M. E., Jackrel, M. E., Rodell, C. B., Burdick, J. A., King, O. D., Gitler, A. D., Lagier-Tourenne, C., Pandey, U. B., Chook, Y. M., Taylor, J. P. & Shorter, J. Nuclear-Import Receptors Reverse Aberrant Phase Transitions of RNA-Binding Proteins with Prion-like Domains. Cell 173, 677-692 e620, doi:10.1016/j.cell.2018.03.002 (2018).
56. Bentmann, E., Neumann, M., Tahirovic, S., Rodde, R., Dormann, D. & Haass, C. Requirements for stress granule recruitment of fused in sarcoma (FUS) and TAR DNA-binding protein of 43 kDa (TDP-43). J Biol Chem 287, 23079-23094, doi:10.1074/jbc.M111.328757 (2012).
57. Amlie-Wolf, A., Ryvkin, P., Tong, R., Dragomir, I., Suh, E., Xu, Y., Van Deerlin, V. M., Gregory, B. D., Kwong, L. K., Trojanowski, J. Q., Lee, V. M., Wang, L. S. & Lee, E. B. Transcriptomic Changes Due to Cytoplasmic TDP-43 Expression Reveal Dysregulation of Histone Transcripts and Nuclear Chromatin. PLoS One 10, e0141836, doi:10.1371/journal.pone.0141836 (2015).
58. Pinarbasi, E. S., Cagatay, T., Fung, H. Y. J., Li, Y. C., Chook, Y. M. & Thomas, P. J. Active nuclear import and passive nuclear export are the primary determinants of TDP-43 localization. Sci Rep 8, 7083, doi:10.1038/s41598-018-25008-4 (2018).
59. Timney, B. L., Raveh, B., Mironska, R., Trivedi, J. M., Kim, S. J., Russel, D., Wente, S. R., Sali, A. & Rout, M. P. Simple rules for passive diffusion through the nuclear pore complex. J Cell Biol 215, 57-76, doi:10.1083/jcb.201601004 (2016).
60. Ederle, H., Funk, C., Abou-Ajram, C., Hutten, S., Funk, E. B. E., Kehlenbach, R. H., Bailer, S. M. & Dormann, D. Nuclear egress of TDP-43 and FUS occurs independently of Exportin-1/CRM1. Sci Rep 8, 7084, doi:10.1038/s41598-018-25007-5 (2018).
61. Archbold, H. C., Jackson, K. L., Arora, A., Weskamp, K., Tank, E. M., Li, X., Miguez, R., Dayton, R. D., Tamir, S., Klein, R. L. & Barmada, S. J. TDP43 nuclear export and neurodegeneration in models of amyotrophic lateral sclerosis and frontotemporal dementia. Sci Rep 8, 4606, doi:10.1038/s41598-018-22858-w (2018).
62. Duan, L., Zaepfel, B. L., Aksenova, V., Dasso, M., Rothstein, J. D., Kalab, P. & Hayes, L. R. Nuclear RNA binding regulates TDP-43 nuclear localization and passive nuclear export. Cell Rep 40, 111106, doi:10.1016/j.celrep.2022.111106 (2022).
63. Dos Passos, P. M., Hemamali, E. H., Mamede, L. D., Hayes, L. R. & Ayala, Y. M. RNA-mediated ribonucleoprotein assembly controls TDP-43 nuclear retention. bioRxiv, doi:10.1101/2023.08.06.552215 (2023).
64. Chen, L. L. & Carmichael, G. G. Altered nuclear retention of mRNAs containing inverted repeats in human embryonic stem cells: functional role of a nuclear noncoding RNA. Mol Cell 35, 467-478, doi:10.1016/j.molcel.2009.06.027 (2009).
65. Palazzo, A. F. & Lee, E. S. Sequence Determinants for Nuclear Retention and Cytoplasmic Export of mRNAs and lncRNAs. Front Genet 9, 440, doi:10.3389/fgene.2018.00440 (2018).
66. Nguyen, T. M., Kabotyanski, E. B., Reineke, L. C., Shao, J., Xiong, F., Lee, J. H., Dubrulle, J., Johnson, H., Stossi, F., Tsoi, P. S., Choi, K. J., Ellis, A. G., Zhao, N., Cao, J., Adewunmi, O., Ferreon, J. C., Ferreon, A. C. M., Neilson, J. R., Mancini, M. A., Chen, X., Kim, J., Ma, L., Li, W. & Rosen, J. M. The SINEB1 element in the long non-coding RNA Malat1 is necessary for TDP-43 proteostasis. Nucleic Acids Res 48, 2621-2642, doi:10.1093/nar/gkz1176 (2020).
67. Bahar Halpern, K., Caspi, I., Lemze, D., Levy, M., Landen, S., Elinav, E., Ulitsky, I. & Itzkovitz, S. Nuclear Retention of mRNA in Mammalian Tissues. Cell Rep 13, 2653-2662, doi:10.1016/j.celrep.2015.11.036 (2015).

Disclosure

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Current Understanding of Dietary Fiber and Its Role in Chronic Diseases

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Current studies have shown the greatest associations between dietary fiber intake and risk reduction in obesity and cardio-vasular disease.



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Abstract

Dietary fiber (DF) is an essential, albeit under-consumed, component in the North American diet. DF is thought to have anti-inflammatory disease-modifying effects via DF-related gut microbiota degradation products called short chain fatty acids. Thus far studies have shown the greatest associations between DF intake and risk reduction in obesity, improved weight loss outcomes, and risk reduction of cardio-vascular disease (CVD). There is weak evidence associating DF intake and inflammatory bowel disease (IBD) risk, IBD remission, reduced risk of Crohn's disease (CD flares, and no evidence showing any benefit towards ulcerative colitis (UC) specifically. Evidence on DF intake and the risk reduction of colorectal cancer (CRC) has been equivocal. Studies were limited by a lack of randomization or in controlling fiber types and sources. Based on the current beneficial associations of DF on obesity management and CVD, counseling patients to increase DF intake may be a cost-effective measure to decrease the burden of chronic disease.

Introduction

Dietary fiber (DF) is an essential part of a healthy human diet. It is associated with diversification of gut microbiota and is thought to play a role in various pathological conditions.[1] The average American DF intake ranges between 11.8g-19.8g, while the daily intake recommendation is 25-35grams. The gut microbiome plays an important role in degrading dietary fiber into short-chain fatty acids (SCFAs) which have several anti-inflammatory effects associated with modifying various chronic diseases. Many studies have shown an association between increased DF intake and reduced risk of cardiovascular disease and obesity. There is also some weak evidence indicating that DF intake is associated with reduced risk of development of inflammatory bowel disease and the risk of Crohn's disease flares. This review aims to discuss the current understanding of DF and the role it may play in obesity, cardio-vascular disease (CVD), colorectal cancer (CRC), and inflammatory bowel disease (IBD). -

Classification

There are numerous definitions of DF based on

factors such as the size of the carbohydrate polymer, solubility, and the location of fermentation in the gut. According to the Codex Alimentarius Commission, which is a collection of internationally adopted food standards, DF includes carbohydrate polymers with ten or more monomeric units that are neither digested nor absorbed by the endogenous enzymes of the small intestine of the human gastrointestinal system.[2] The definition of the United States Food and Drug Administration (FDA) differs slightly and also includes carbohydrates of three to nine units.[3] DF can also be classified based on factors such as their primary food source, chemical structure, water solubility, viscosity, and fermentability. Commonly, based on its water solubility, DF is classified as insoluble fiber (IF) or soluble fiber (SF; Figure 1).[4] The solubility of fiber influences its physiological role as described further below.

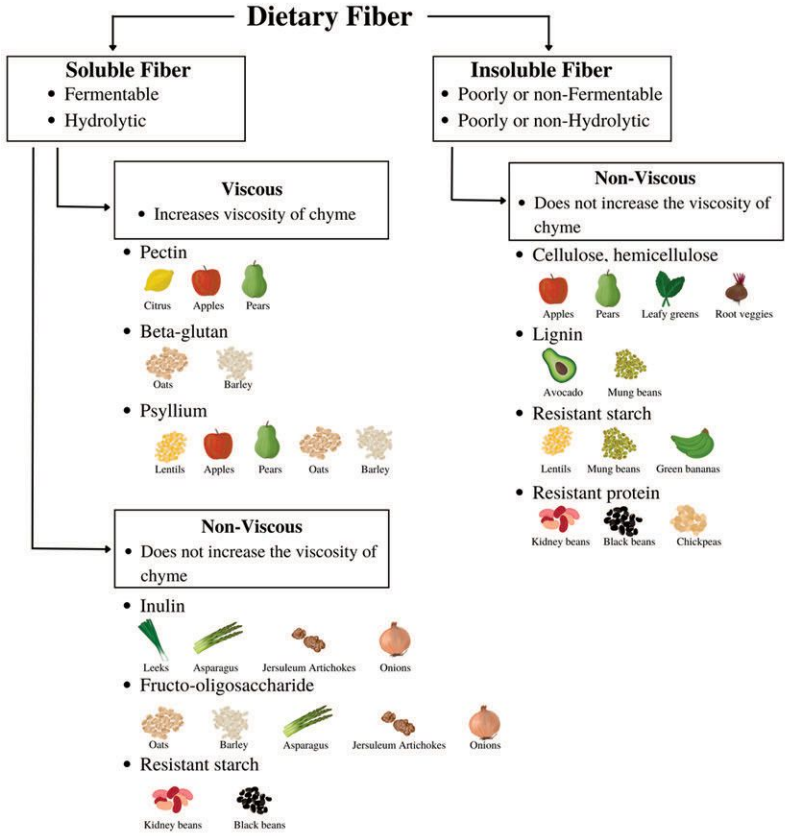


Figure 1. Classifications of DF types and their sources. Fibers can be classified further based on the ability to change the viscosity of chyme: the partially digested food materials and digestive secretions during digestion. Note that "resistant" refers to products that can resist breakdown during digestion.

Main Food Sources of Dietary Fiber

Edible fiber includes fiber naturally found in food, as well as those synthetically or enzymatically produced. Most edible DF is found in natural sources. Both SF and IF can be found in legumes, vegetables, nuts/seeds, fruits, and synthetically in cereals (Figure 1).[5] Insoluble fiber can bypass digestion in the small intestine and are fermented by specialized microorganisms. They are usually found in cereals, legumes, ripe fruits, and green bananas (Figure 1).[5]

Current Fiber Recommendations and Intake

The FDA recommends 28g of dietary fiber per day for a 2000 kcal/day diet.[6] The American Dietetic Association recommends 14g/1,000 kcal or 25g for adult women and 38g for adult men.[7] These recommendations are based on studies showing associations with the protective effects of DF against cardiovascular disease.[7] Recommendations vary in children and the elderly based on age. Infants below the age of six months who are exclusively breastfed do not require additional DF intake. Adequate data is not available for infants from the age of 7-12 months. After the age of two years, adequate DF intake is equal to or greater than their age, plus 5g/day. In the elderly, lower values of 14g/1000 kcal are recommended.[7]

The average daily DF intake in the US is lower than recommended. According to the National Health and Nutrition Examination Survey's most recent 2017-2018 report on DF intake by Americans, the fiber intake in males ranges from 12.4-19.8g

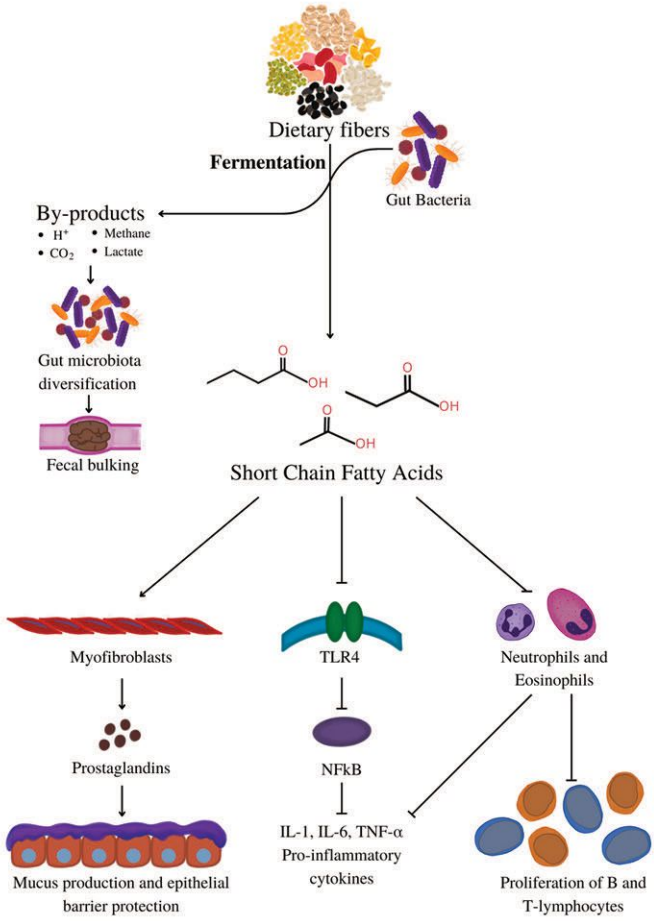


Figure 2. DF fermentation byproducts, protons (H^+), carbon dioxide (CO_2), methane, and lactate, aid in the diversification of gut microbiota resulting in increased fecal bulking. SCFAs act on myofibroblasts to produce prostaglandins that produce mucus membranes and provide epithelial barrier protection. Inhibition of toll-like receptor 4 (TLR4) downregulates nuclear factor kappa-light-chain-enhancer of activated B cells (NFkB) to attenuate the production of proinflammatory cytokines such as interleukin-1(IL-1), IL-6, and tumor necrosis factor-alpha (TNF-α). Inhibition of the activation of neutrophils and eosinophils prevents further cytokine activation and proliferation of B and T cells.

on average and lower in females from 11.8-16g.[8] This reported intake is several grams lower than the recommended daily intake. In comparison, the European Food Safety Authorization reports an average daily DF intake of 16-29g for an adult.[9] This is a notable increase in the average consumption of fiber relative to the US.

Normal Digestive Physiology and Pathology

The digestive physiology of DF is dependent on the fiber type, location within the small and large intestines, and specific gut microbiota. DF, while thought to be resistant to absorption, does indeed undergo some chemical and structural changes in the gastrointestinal system, such as hydrolysis. Solubility has been shown to have an association with hydrolysis, with increasing resistance to hydrolysis seen with decreasing water solubility.[1] IF is resistant to hydrolysis in the small intestine due to its hydrophobic and crystalline structure formed by hydrogen bonds. This results in a bulking effect, as it can bypass the small intestine with minimal breakdown.[1] DF that is resistant to breakdown in the small intestine undergoes fermentation by the gut microbiome of the large intestine. The by-products produced through fermentation provide energy that is required by gut microbes to survive and grow.[5] This plays a vital role in the diversification of the gut microbiota and further aids in fecal bulking.[1] Some studies indicate that the effect of DF on the diversification of gut microbiota may play a role in disease development and progression.[10,11]

DF undergoes anaerobic fermentation by the gut microbiome to be degraded into products known as short-chain fatty acids (SCFAs) including butyrate, propionic acid, and acetic acid. SCFAs have several effects including anti-inflammatory actions through the inhibition of cytokines, activation of dendritic cells that are involved in anti-viral mechanisms, epithelial barrier protection, boosting IgA secretions, and mucus production.[10, 11] The microbes of the gut play a crucial role in the metabolism of DF, which in turn likely contribute to their associated protective role in chronic inflammatory conditions (Figure 2).[10, 11] We further describe the role of dietary fiber in chronic disease below.

Obesity

Obesity has become a pandemic worldwide due to the increasing availability of calorie-dense food and simple carbohydrates. There is a growing body of research showing the association between DF and

obesity. Existing literature suggests a protective effect of high DF intake on ideal body weight through the generation of SCFAs.[12] Some hypothesized mechanisms to explain the obesity modulating effects of SCFA include that they provide extra sources of energy, suppress insulin-mediated fat accumulation via G-protein coupled receptor-43 expressed in the intestinal epithelium, immune cells, and adipocytes, regulate fasting-induced adipose factor, and regulate leptin secretion via free fatty acid receptor 3.[13, 14, 15]

In a 12-week, single-center, double-blind, placebo-controlled trial, 53 adults who had overweight or obesity were randomly assigned to a pea fiber (PF) or a control diet.[16] Pea fiber intake significantly altered the types of fecal SCFAs and bile acid (BA) levels. The PF group was found to have higher acetate levels and reduced isovalerate, cholate, deoxycholate, and total BA content. Although the exact mechanism of action and role of these metabolite changes are unclear, these SCFAs and BAs may be involved in obesity by acting on signaling pathways, gut epithelium barrier function, and inhibition of proinflammatory cytokine production.[16] The correlation of PF consumption, and DF in general, with alterations in the gut microbe metabolites may be a possible mechanism involved in the obesity modulating effects of DF.

Several studies have examined the role of DF on weight management outcomes. A prospective study with a sample size of 120,877 non-obese individuals observed an inverse correlation between long-term weight gain and intake of DF.[17] In another study, children between the ages of 7–12 years old with overweight or obesity were randomly assigned to receive either oligofructose-enriched inulin or a maltodextrin placebo once daily for 16 weeks. Oligofructose enriched inulin is a mixture of non-digestible carbohydrates, containing oligosaccharide and inulin, both of which are fermentable plant fructan DF. Reduced fat mass was observed in the inulin group, suggesting that increased intake of DF may have beneficial effects on obesity.[18] More recently, the RESOLVE study initiated a two-phase experimental protocol to examine the role of lifestyle modifications on weight loss outcomes. Phase one consisted of a three-week intervention in diet, exercise, and education under a controlled environment with close supervision. This was followed by a second phase wherein participants maintained these lifestyle changes under distant supervision for 12 months. It was found that fiber intake was the only nutritional variable that systematically predicted a significant reduction in health indicators like weight, central fat, and body mass index (BMI).[19]

A limitation of the RESOLVE study was the second phase of distant supervision, which was not in a controlled setting. The above studies were also limited by varying DF sources and types, limiting generalizability. For example, some studies have examined individual fiber types, such as inulin, [18] others with a pea fiber source,[16] and the RESOLVE study examined generalized DF content. [19] With the plethora of the types and sources of DF, it is unclear whether certain DF types or sources have the same obesity modulating effects.

From the above limited studies, DF intake has been associated with reduced obesity and improved weight loss outcomes. Gut microbe-related DF degradation products, such as SCFAs and BAs are thought to be the physiological drivers in these obesity modulating effects. Further studies that include randomization are required to further elucidate whether individual fiber types or sources play an obesity modulating role.

Cardio-Vascular Disease

Coronary artery disease (CAD) is the leading cause of mortality worldwide. Its etiology is a culmination of factors involving lifestyle, environment, and genetics. Diet has been widely regarded as a modifiable risk factor for the prevention of CVD. A high fiber intake may modify risk factors that can contribute to atherosclerotic disease including dyslipidemia, obesity, hypertension, and diabetes.[20,21] Current evidence suggests an inverse relationship between CVD and intake of DF. The Pooling Project of Cohort Studies on Diet and Coronary Disease pooled data across 10 prospective cohort studies. This study found a significant inverse relationship between DF intake and the risk of CAD.[22] There was a dose-response relationship for every 10g/day increase in DF intake, with a 14% reduction in risk of all coronary events, and a 27%

reduction in the risk of coronary mortality.[22] This association was independent of age, sex, CVD risk factors such as BMI and hypertension, lifestyle factors such as smoking and physical activity, and other dietary factors such as energy and fat intake.[22] Another meta-analysis analyzed the effects of 10g/day increment increases of DF and found a pooled relative risk of 0.91 for CVD.[23] A meta-analysis of 19 studies indicated that an increased intake of total fiber, IF, and fiber from cereals, vegetables, and fruits lowered the risk of CVD and coronary heart disease (CHD).[24] There was a reduction of risk with each increase of 7g/day in total fiber intake, with relative risks of 0.91 for both CVD and CHD.[24]

There are several limitations to the studies associating DF to CVD risk. Given that intake of DF was not randomized, causality is difficult to establish. The definition of DF and the definition and measurement of specific fiber sources such as cereal fiber, vegetable fiber, and whole grains were not standardized. For example, Pereira et al. found significant associations between cereal and fruit DF intake in reducing CVD risk, however non-significant findings were associated with vegetable fiber intake.[22] Further studies controlling for fiber type and source may be warranted. Many of the studies also failed to control other dietary variables including vitamins, minerals, and beta-carotene. Considering there is some evidence for the contribution of these nutrients in decreasing CVD risk, failing to control for them can potentially impact findings.[25] Finally, the influence of socioeconomic factors remains unclear as most studies were in a predominantly Caucasian population in the US and Europe. Given that the food trends and sources of fiber intake are different in different ethnicities and races, the relationship needs to be adjusted for these factors as well.[25]

From the above studies, DF intake has been associated with a reduction in the risk of CVD. Currently, the consensus on the amount and type of fiber has yet to be reached, but as described above, an increase in 7-10g/day was associated with a reduction in CVD risk. These studies are limited by a lack of randomization, control for DF types and sources, control of various socioeconomic factors, and control for other dietary nutrients that impact CVD. Further studies are required to better elucidate the long-term effects of DF on CVD risk.

Colorectal Cancer

Colorectal cancer (CRC) is one of the leading causes of cancer deaths in the world. The recent intense screening measures have reduced the incidence, but CRC remains the third leading cause of cancer in men and women.[26] Understanding the preventative factors of CRC may help to reduce its disease burden. Some components of DF such as wheat bran and phytic acid (inositol hexaphosphate) were found to have chemo-preventive properties against colon carcinogenesis.[27]

The evidence on the protective effects of DF intake on CRC has been inconsistent due to evidence being mostly observational. Case-control studies were generally affirmative but most observational cohort studies challenged an association of DF to CRC. [28, 29] In a large prospective study, the NIH-AARP Diet and Health Study, analysis of half a million adults for five years found no association between total DF intake and CRC. However, an inverse association was observed between the intake of fiber from grains and CRC.[26] Another study conducted with a 15-year follow-up period showed a significant decrease in the incidence of CRC with the intake of whole wheat.[30] A similar association was not seen when looking at the incidence of CRC with overall increased DF intake.[30] Current studies investigating the role of DF in the development of CRC have been equivocal, and further studies are required to elucidate whether DF has long-term effects in decreasing the risk for CRC.

Inflammatory Bowel Disease

The incidence of IBD has been increasing over recent decades.[31] IBD is comprised of CD and ulcerative colitis (UC). Currently, the mainstay treatments for patients with IBD have been biologics to arrest progression and maintain remission. Modifiable factors such as DF intake have been investigated to prevent the development and progression of IBD. The potential role of DF lies in the production of SCFAs. SCFAs such as butyrate has been shown to regulate cytokines involved in intestinal inflammation such as IL-10, IFNγ, and IL-1β.[32, 33] In a study comparing women in the highest quantile of fiber intake and women in the lowest quantile of fiber intake, a 40% decrease in risk of CD development was found.[33] The benefit was highest with fiber derived from fruits. However,

Table 1. Practical Approach to Counselling on Dietary Fiber Intake

- Investigate the types of fibers in patient's current diet
- Discuss short- and long-term effects of low fiber diets including increased risk of chronic diseases and decreased gut health
- Provide resources such as print copies of fiber examples and link to Dietary Guidelines for Americans website
- Provide examples of recommended portion sizes
- Work with the patient to make progressive steps to increasing fiber intake such as initially replacing snacks with high fiber options such as fruits, vegetables, nuts, and seeds.

a significant association was not seen in the risk of UC development. Similarly, another study found DF to be protective against CD.[34] Data from a systemic review showed that adults with IBD consume significantly less DF compared to healthy adults, and the presence of an inverse association between DF intake and the risk of CD.[31]

Beyond the risk of developing IBD, studies have also shown a decreased risk of CD flares with fiber intake.[35] A small study found a lower rate of treatment failure for the psyllium plus mesalamine group versus only mesalamine (28% versus 35%).[36] This association was not seen with UC.

While these studies show some beneficial association between high fiber intake and IBD, the impact on causality is yet to be understood. Significant associations between DF were only shown with CD, whereas no associations with UC were found. The effects on IBD remission have been varied and require further investigation.

Clinical Strategies to Improve Dietary Fiber Intake

The intake of DF is limited in the average Western diet. In a clinical setting, a comprehensive approach toward disease management includes counseling patients on dietary modification, an important part of preventative medicine (Table 1). Advising patients to increase overall DF intake is an easy, cost-effective, and low-harm measure, and may aid in the holistic management of obesity, CVD, and other chronic diseases. Counselling should include investigating the dietary components of a patient's average meal. Resources regarding fiber-rich food via print materials, links to websites, along with resources on the beneficial effects of DF can be provided to patients. Dietary Guidelines for Americans have information about food sources that are rich in DF along with recommended portion sizes, and the information is detailed yet simple to understand.[37] Simple behavioral measures such as replacing processed food snacks with small servings of fruits, eating vegetables or salads, and reading food labels may be beneficial.

Conclusion

Current studies have shown the greatest associations between DF intake and risk reduction in obesity and CVD. There is some weak evidence through observational studies indicating that DF intake is associated with a reduced risk of development of IBD and the risk of CD flares. Associations with CRC were equivocal. See Table 2 for a summary of the studies noted in this review. Based on the current findings associating DF intake with reduced risk for obesity and CAD, counselling patients and providing patients with educational resources to increase DF intake may provide long-term benefits. Studies were limited by a lack of randomization or in controlling fiber types and sources. Further investigation is needed into understanding the role of DF in the development of chronic diseases and their implication for treatment.

References

1. Capuano, E. (2017). The behavior of dietary fiber in the gastrointestinal tract determines its physiological effect. Critical Reviews in Food Science and Nutrition, 16, 3543–3564. https://doi.org/10.1080/10408398.2016.1180501

2. Jones, J. M. (2014). CODEX-aligned dietary fiber definitions help to bridge the 'fiber gap.' Nutrition Journal, 1. https://doi.org/10.1186/1475-2891-13-34

3. Questions and Answers on Dietary Fiber | FDA. (n.d.). U.S. Food and Drug Administration; FDA. Retrieved April 17, 2022, from https://www.fda.gov/food/food-labeling-nutrition/questions-and-answers-dietary-fiber#define_dietary_fiber

4. Deehan, E. C., Duar, R. M., Armet, A. M., Perez-Muñoz, M. E., Jin, M., & Walter, J. (2017). Modulation of the Gastrointestinal Microbiome with Nondigestible Fermentable Carbohydrates To Improve Human Health. Microbiology Spectrum, 5.

5. Stephen, A. M., Champ, M. M.-J., Cloran, S. J., Fleith, M., van Lieshout, L., Mejborn, H., & Burley, V. J. (2017). Dietary fibre in Europe:

Table 2. Summary of the Studies Noted in This Review

Study	Study design	Results
Obesity		
Kim et al. [12]	Meta-analysis of 21 randomized control trials, with a median intervention duration of 6 weeks.	- Significant weight reduction associated with diets containing pulse fiber (nonoil seeds of legumes such as beans, lentils, chickpeas)
Mozaffarian et al.[17]	Prospective trial with 120,877 healthy individuals followed over four years	- Inverse correlation between long-term weight gain and DF
Nicolucci et al. [18]	Single-center, double-blind, placebo-controlled trial including children 7-12 years old with overweight or obesity	- Reduced fat mass in the DF group
RESOLVE [19]	Randomized control trial with 87 participants 3-week intensive diet–exercise intervention	- DF was the only variable that predicted weight reduction.
Cardio-vascular disease		
Pereira et al. [22]	Pooled cohort analysis 10 prospective cohort studies with a 6-10 year follow-up	- Inverse relationship between DF intake and risk of CAD
Kim et al. [25]	Meta-analysis of 15 studies	- High DF intake associated with a reduced risk of mortality from CVD
Threapleton et al. [24]	Meta-analysis of 22 cohort studies	- DF inversely associated with risk of CVD
Colorectal cancer		
NIH-AARP Diet and Health Study [26]	Prospective trial including 291,988 men and 197,623 women aged 50–71 years old.	- Total DF intake was not associated with CRC - DF from grains was associated with a lower risk of CRC, and whole-grain intake was inversely associated with CRC risk
NIH-AARP Diet and Health Study [30]	Same as above	- Total DF was not associated with CRC - Whole grains specifically were inversely associated with the incidence of CRC
Inflammatory Bowel Disease		
Ananthakrishnan et al. [33]	Prospective study 170,776 women, followed up over 26 years	- Intake of the highest quintile (median of 24.3 g/day) was associated with a 40% reduction in risk of CD
Amre et al. [34]	Case control study of 130 CD patients and 202 controls	- DF was protective against CD
Hou et al. [31]	Systematic review 19 studies, 2609 IBD patients	- Inverse association between DF intake and the risk of CD
Brotherton et al. [35]	Longitudinal internet-based cohort of more than 14,000 participants with IBD	- Patients with CD in the highest quartile of DF intake were less likely to have a flare - CD patients who reported that they did not avoid high-fiber foods were ~40% less likely to have a flare.

CAD= Coronary Artery Disease; CD= Crohn's Disease; CRC= Colorectal carcinoma; CVD= Coronary Vascular Disease; DF= Dietary fiber; IBD= Inflammatory Bowel Disease; UC= Ulcerative Colitis

current state of knowledge on definitions, sources, recommendations, intakes and relationships to health. Nutrition Research Reviews, 2, 149–190. https://doi.org/10.1017/s095442241700004x
6. Korczak, Renee, and Joanne L. Slavin. "Definitions, Regulations, and New Frontiers for Dietary Fiber and Whole Grains." Nutrition Reviews, no. Supplement_1, Oxford University Press (OUP), July 2020, pp. 6–12. Crossref, doi:10.1093/nutrit/nuz061.
7. Slavin, Joanne. "Position of the American Dietetic Association: Health Implications of Dietary Fiber." Journal of the American Dietetic Association, no. 10, Elsevier BV, Oct. 2008, pp. 1716–31. Crossref, doi:10.1016/j.jada.2008.08.007.
8. U.S. Department of Agriculture, Agricultural Research Service. 2020. Nutrient Intakes from Food and Beverages: Mean Amounts Consumed per Individual, by Gender and Age, What We Eat in America, NHANES 2017-2018.
9. Scientific Opinion on Dietary Reference Values for carbohydrates and dietary fibre. (2010). EFSA Journal, 3. https://doi.org/10.2903/j.efsa.2010.1462
10. Chénard, T., Prévost, K., Dubé, J., & Massé, E. (2020). Immune System Modulations by Products of the Gut Microbiota. Vaccines, 3, 461. https://doi.org/10.3390/vaccines8030461
11. Shenoy, S. (2021). Gut microbiome, Vitamin D, ACE2 interactions are critical factors in immune-senescence and inflammaging: key for vaccine response and severity of COVID-19 infection. Inflammation Research, 1, 13–26. https://doi.org/10.1007/s00011-021-01510-w
12. Kim, S. J., de Souza, R. J., Choo, V. L., Ha, V., Cozma, A. I.,

Chiavaroli, L., Mirrahimi, A., Blanco Mejia, S., Di Buono, M., Bernstein, A. M., Leiter, L. A., Kris-Etherton, P. M., Vuksan, V., Beyene, J., Kendall, C. W., Jenkins, D. J., & Sievenpiper, J. L. (2016a). Effects of dietary pulse consumption on body weight: a systematic review and meta-analysis of randomized controlled trials. The American Journal of Clinical Nutrition, 5, 1213–1223. https://doi.org/10.3945/ajcn.115.124677
13. Kimura, I., Ozawa, K., Inoue, D., Imamura, T., Kimura, K., Maeda, T., Terasawa, K., Kashihara, D., Hirano, K., Tani, T., Takahashi, T., Miyauchi, S., Shioi, G., Inoue, H., & Tsujimoto, G. (2013). The gut microbiota suppresses insulin-mediated fat accumulation via the short-chain fatty acid receptor GPR43. Nature Communications, 1. https://doi.org/10.1038/ncomms2852
14. Kasubuchi, M., Hasegawa, S., Hiramatsu, T., Ichimura, A., & Kimura, I. (2015). Dietary Gut Microbial Metabolites, Short-chain Fatty Acids, and Host Metabolic Regulation. Nutrients, 4, 2839–2849. https://doi.org/10.3390/nu7042839
15. Zaibi, M. S., Stocker, C. J., O'Dowd, J., Davies, A., Bellahcene, M., Cawthorne, M. A., Brown, A. J. H., Smith, D. M., & Arch, J. R. S. (2010). Roles of GPR41 and GPR43 in leptin secretory responses of murine adipocytes to short chain fatty acids. FEBS Letters, 11, 2381–2386. https://doi.org/10.1016/j.febslet.2010.04.027
16. Mayengbam, S., Lambert, J. E., Parnell, J. A., Tunnicliffe, J. M., Nicolucci, A. C., Han, J., Sturzenegger, T., Shearer, J., Mickiewicz, B., Vogel, H. J., Madsen, K. L., & Reimer, R. A. (2019). Impact of dietary fiber supplementation on modulating microbiota–host–metabolic axes in obesity. The Journal of Nutritional Biochemistry, 228–236. https://doi.org/10.1016/j.jnutbio.2018.11.003
17. Mozaffarian, D., Hao, T., Rimm, E. B., Willett, W. C., & Hu, F. B. (2011). Changes in Diet and Lifestyle and Long-Term Weight Gain in Women and Men. New England Journal of Medicine, 25, 2392–2404. https://doi.org/10.1056/nejmoa1014296
18. Nicolucci, A. C., Hume, M. P., Martínez, I., Mayengbam, S., Walter, J., & Reimer, R. A. (2017). Prebiotics Reduce Body Fat and Alter Intestinal Microbiota in Children Who Are Overweight or with Obesity. Gastroenterology, 3, 711–722. https://doi.org/10.1053/j.gastro.2017.05.055
19. Tremblay, A., Clinchamps, M., Pereira, B., Courteix, D., Lesourd, B., Chapier, R., Obert, P., Vinet, A., Walther, G., Chaplais, E., Bagheri, R., Baker, J. S., Thivel, D., Drapeau, V., & Dutheil, F. (2020). Dietary Fibres and the Management of Obesity and Metabolic Syndrome: The RESOLVE Study. Nutrients, 10, 2911. https://doi.org/10.3390/nu12102911
20. Lairon, D., Arnault, N., Bertrais, S., Planells, R., Clero, E., Hercberg, S., & Boutron-Ruault, M.-C. (2005). Dietary fiber intake and risk factors for cardiovascular disease in French adults. The American Journal of Clinical Nutrition, 6, 1185–1194. https://doi.org/10.1093/ajcn/82.6.1185
21. Soliman, G. A. (2019). Dietary Fiber, Atherosclerosis, and Cardiovascular Disease. Nutrients, 5, 1155. https://doi.org/10.3390/nu11051155
22. Pereira, M. A., O'Reilly, E., Augustsson, K., Fraser, G. E., Goldbourt, U., Heitmann, B. L., Hallmans, G., Knekt, P., Liu, S., Pietinen, P., Spiegelman, D., Stevens, J., Virtamo, J., Willett, W. C., & Ascherio, A. (2004). Dietary Fiber and Risk of Coronary Heart Disease. Archives of Internal Medicine, 4, 370. https://doi.org/10.1001/archinte.164.4.370
23. Kim, Y., & Je, Y. (2016). Dietary fibre intake and mortality from cardiovascular disease and all cancers: A meta-analysis of prospective cohort studies. Archives of Cardiovascular Diseases, 1, 39–54. https://doi.org/10.1016/j.acvd.2015.09.005
24. Threapleton, D. E., Greenwood, D. C., Evans, C. E. L., Cleghorn, C. L., Nykjaer, C., Woodhead, C., Cade, J. E., Gale, C. P., & Burley, V. J. (2013). Dietary fibre intake and risk of cardiovascular disease: systematic review and meta-analysis. BMJ, dec19 2, f6879–f6879. https://doi.org/10.1136/bmj.f6879
25. Satija, A., & Hu, F. B. (2012). Cardiovascular Benefits of Dietary Fiber. Current Atherosclerosis Reports, 6, 505–514. https://doi.org/10.1007/s11883-012-0275-7
26. Schatzkin, A., Mouw, T., Park, Y., Subar, A. F., Kipnis, V., Hollenbeck, A., Leitzmann, M. F., & Thompson, F. E. (2007). Dietary fiber and whole-grain consumption in relation to colorectal cancer in the NIH-AARP Diet and Health Study. The American Journal of Clinical Nutrition, 5, 1353–1360. https://doi.org/10.1093/ajcn/85.5.1353
27. Reddy B. S. (1999). Prevention of colon carcinogenesis by components of dietary fiber. Anticancer research, 19(5A), 3681–3683.
28. Howe, G. R., Benito, E., Castelleto, R., Cornée, J., Estève, J., Gallagher, R. P., Iscovich, J. M., Deng-ao, J., Kaaks, R., Kune, G. A., Kune, S., L'Abbé, K. A., Lee, H. P., Lee, M., Miller, A. B., Peters, R. K., Potter, J. D., Riboli, E., Slattery, M. L., … Shu, Z. (1992). Dietary Intake of Fiber and Decreased Risk of Cancers of the Colon and Rectum: Evidence From the Combined Analysis of 13 Case-Control Studies. JNCI: Journal of the National Cancer Institute, 24, 1887–1896. https://doi.org/10.1093/jnci/84.24.1887
29. Bazyk, A. E., Olson, J. E., Sellers, T. A., Kushi, L. H., Anderson, K. E., Lazovich, D., Folsom, A. R., & Bostick, R. M. (1998). Diet and risk of colon cancer in a large prospective study of older women: an analysis stratified on family history (Iowa, United States). Cancer Causes and Control, 4, 357–367. https://doi.org/10.1023/a:1008886715597
30. Hullings, A. G., Sinha, R., Liao, L. M., Freedman, N. D., Graubard, B. I., & Loftfield, E. (2020). Whole grain and dietary fiber intake and risk of colorectal cancer in the NIH-AARP Diet and Health Study cohort. The American Journal of Clinical Nutrition, 3, 603–612. https://doi.org/10.1093/ajcn/nqaa161
31. Hou, J. K., Abraham, B., & El-Serag, H. (2011). Dietary Intake and Risk of Developing Inflammatory Bowel Disease: A Systematic Review of the Literature. American Journal of Gastroenterology, 4, 563–573. https://doi.org/10.1038/ajg.2011.44
32. Asarat, M., Apostolopoulos, V., Vasiljevic, T., & Donkor, O. (2016). Short-Chain Fatty Acids Regulate Cytokines and Th17/Treg Cells in Human Peripheral Blood Mononuclear Cellsin vitro. Immunological Investigations, 3, 205–222. https://doi.org/10.3109/08820139.2015.1122613
33. Ananthakrishnan, A. N., Khalili, H., Konijeti, G. G., Higuchi, L. M., de Silva, P., Korzenik, J. R., Fuchs, C. S., Willett, W. C., Richter, J. M., & Chan, A. T. (2013). A prospective study of long-term intake of dietary fiber and risk of Crohn's disease and ulcerative colitis. Gastroenterology, 145(5), 970–977. https://doi.org/10.1053/j.gastro.2013.07.050
34. Amre, D. K., D'Souza, S., Morgan, K., Seidman, G., Lambrette, P., Grimard, G., Israel, D., Mack, D., Ghadirian, P., Deslandres, C., Chotard, V., Budai, B., Law, L., Levy, E., & Seidman, E. G. (2007). Imbalances in dietary consumption of fatty acids, vegetables, and fruits are associated with risk for Crohn's disease in children. The American journal of gastroenterology, 102(9), 2016–2025. https://doi.org/10.1111/j.1572-0241.2007.01411.x
35. Brotherton, C. S., Martin, C. A., Long, M. D., Kappelman, M. D., & Sandler, R. S. (2016). Avoidance of Fiber Is Associated With Greater Risk of Crohn's Disease Flare in a 6-Month Period. Clinical Gastroenterology and Hepatology, 8, 1130–1136. https://doi.org/10.1016/j.cgh.2015.12.029
36. Copaci, I., et al. "Maintenance of Remission of Ulcerative Colitis (UC): Mesalamine, Dietary Fiber, S. Boulardi." Digestive and Liver Disease, Elsevier BV, May 2000, p. A24. Crossref, doi:10.1016/s1590-8658(00)80138-3.
37. "Food Sources of Dietary Fiber | Dietary Guidelines for Americans." Home | Dietary Guidelines for Americans, https://www.dietaryguidelines.gov/resources/2020-2025-dietary-guidelines-online-materials/food-sources-select-nutrients/food-0. Accessed 4 Aug. 2022.

Disclosure

None reported.

MM

A Review of the Role of Robotics in Surgery: To DaVinci and Beyond!

by Patrick Probst, MD

With new systems and applications routinely coming to market, it continues to be an exciting avenue for new research and development as well as improving the patient care experience.



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Abstract

Since its inception in 1985, robotic surgery has evolved into a mainstream surgical approach that has become virtually synonymous with minimally invasive surgery (MIS) and adopted across several specialties offering decreased patient morbidity and improved post-operative outcomes. This article discusses the current role of robotics in MIS and its varied applications, prevalence in the community and the future of the field.

Background

Advancements in surgery tend to occur within one of several broad categories – cost reduction, improved patient satisfaction, and improvement in clinical outcomes. Historically, progress was achieved by individuals altering surgical technique or implementing lessons accrued from their own post-operative experiences into intra-operative decision making. While this granular improvement is vital and continues in modern surgery, paradigm shifts in the foundation of surgical principles and instrumentation have accelerated the evolution of modern surgical medicine.

Numerous significant shifts in surgery have occurred in the past few decades including the introduction of endoscopic equipment for not only diagnostic but therapeutic management of disease. Since the dissemination of laparoscopy, surgeons have continued to explore less invasive approaches now in the form of robotic assisted surgery.

It is the natural progression for the evolution of surgery to push the degree of invasiveness towards the extreme of the spectrum over time in order to improve patient satisfaction and clinical outcomes. The typical hindrance of such innovation is related to cost.

However, with modern advancements in optics, mechanics, energy, computing power, and more, this progression and adaptation into everyday practice is occurring at an ever-increasing speed. Robotic surgery continues to be at the forefront of MIS innovation given its ability to minimize human error and increase surgical precision and operative standardization. But, it is important to define what constitutes robotic surgery. Currently, "robotic surgery" is often used synonymously with the most widely recognizable robot, the DaVinci surgical system. This platform by Intuitive consists of three components that allow the surgeon to indirectly control robotic arms that grasp, transect, coagulate, staple, clip and suture. While this master-slave or passive apparatus

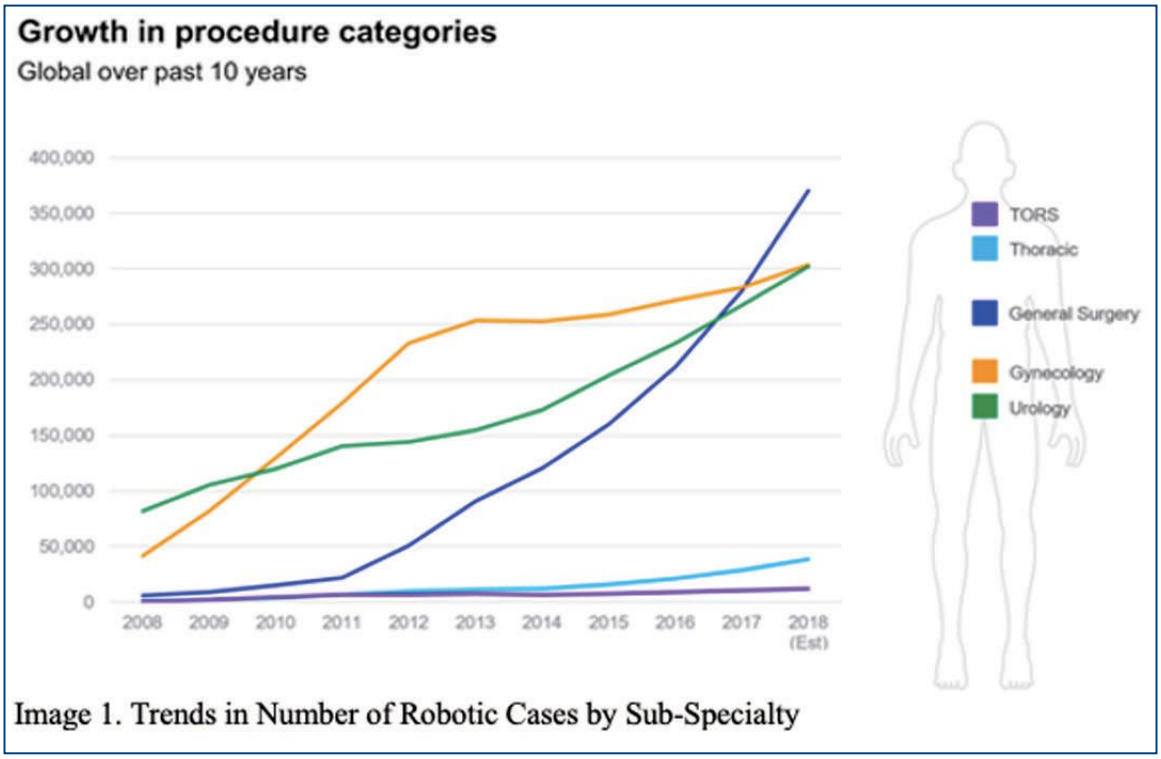


Image 1. Trends in Number of Robotic Cases by Sub-Specialty

that allows for telesurgery remains the most popular globally, other surgical robots can be classified into two other categories: supervisory-controlled, and shared control.[1]

Supervisory-controlled systems allow pre-operative planning with robotic execution under close supervision, such as with aquablation of the prostate in urology. Shared-control systems allow the surgeon and robot to function simultaneously, such as with spine and arthroplasty robots in neurosurgery and orthopedics.[1] This article will highlight the current role of robotic systems in minimally invasive surgery.

Robotic-Assisted Laparoscopic Surgery

DaVinci Laparoscopy

The DaVinci robotic surgical platform (Intuitive surgical Inc., Sunnyvale, CA, USA) was first made commercially available in 2001. It has subsequently undergone three iterations and its most modern format is the DaVinci Xi. Since its inception, this platform has been widely adopted and now used in over 10 million cases over two decades, in five specialties and grown to dominate the robotic assisted laparoscopic (RAL) market (Image 1).[2] At least 85% of prostatectomies are performed robotically and the number of hysterectomies performed robotically has increased by 300% over the last decade.[3,4] In comparison to open surgery, use of the DaVinci robot has shown significant improvement in clinical outcomes often demonstrating less blood loss, a shorter length of stay and less opioid consumption as supported by numerous multi-specialty reports in the literature.[5,6,7,8]

These well-described clinical benefits and rapid adoption by a growing number of surgeons ensures that, for the foreseeable future, the DaVinci robot will continue to be a mainstay in the minimally invasive surgical management of oncologic and reconstructive pathologies in cardiothoracic surgery, general surgery, urology and gynecology. In fact, multiple medical specialties now incorporate training modules and robotic skills courses into their curriculum and require residents to have robotic platform specific training in order to graduate.

However, the original DaVinci patents have now expired and significant robotic system development and competition is expected, particularly in

Robot	Uses	Port	Arms	Haptic Feedback	Eye Tracking	US FDA Approval
DaVinci Xi	MIS	MP	4	No	No	Yes
DaVinci SP	MIS NOTES LESS	SP	1	No	No	Yes
Senhance	MIS	MP	4	Yes	Yes	Yes
Revo-i	MIS	MP	4	Yes	No	Pending (Korea)
SPORT	MIS NOTES LESS	SP	1	No	No	Pre-Clinical
Versius	MIS	MP	4	No	No	Pending (Europe)
MiroSurge	MIS	MP	3	Yes	No	Pre-Clinical
SurgiBot	LESS	SP	1	N/a	N/a	Pending (China)
NeoGuide	NOTES	N/a	1	No	N/a	Yes
Invendoscopy	NOTES	N/a	1	No	N/a	Yes
Flex Robotic	NOTES	N/a	1	No	N/a	Yes
Monarch	NOTES	N/a	1	No	N/a	Yes
Ion	NOTES	N/a	1	No	N/a	Yes
PROCEPT	NOTES	N/a	1	N/a	N/a	Yes
Roboflex	NOTES	N/a	1	No	N/a	Yes

Table 1. Current and Popular Robotic Surgical Systems Worldwide (MP – multiport, N/a – not applicable)[60]

regards to novel technology, reduced cost and size reduction.[9] Current DaVinci platforms do not utilize haptic feedback, the experience of touch by applying resistance to the user, or biometric integration such as eye tracking cameras or head tracking robotic arms that can better replicate a traditional open surgical environment.[10] Additionally, the sizable physical footprint of the DaVinci robot tower, arms and console has proven restrictive in its implementation as well as the significant cost of the system, as the Xi still costs approximately $1.5 million per system.[11] With the expected increase in number of robotic laparoscopic cases over the next decade, numerous companies have entered and expanded the market (Table 1).

Laparo-endoscopic Single-site Surgery (LESS)

The SinglePort (SP) system, created in 2018, is Intuitive's venture into the LESS space. Through a 2.5cm incision, a solitary port releases 1 camera and 3 robotic arms which are controlled from the console by the surgeon similar to the Xi. This approach allows for better cosmetic outcomes – a single incision, quicker patient recovery, and similar optics and dexterity of the instruments. Clinical adoption has continued to increase. Between 2020 and 2021, there has been a 56% increase in the number of SP robotic systems nationwide.

Few companies thus far have made a meaningful impact in the LESS space. However, the STRAS system, version 2 (iCUBE, Strasbourg, France) is a flexible endoscopic system capable of single port intraluminal surgery. Although still in preclinical development, its main advantage over current LESS platforms is its significantly smaller size and the option for table mounting arms. The Single Port Orifice Robotic Technology (SPORT) Surgical System (Titan Medical Inc., Toronto, Canada) also uses a 2.5cm incision to deliver two articulating instruments and a camera for LESS. It is pending FDA approval but has demonstrated success in a single port partial nephrectomy in animal models.[11]

Non-Laparoscopic Robotic Surgical Platforms

Natural Orifice Translumenal Endoscopic Surgery (NOTES)

Robotic NOTES is an exciting area of research and advancement in MIS. The NeoGuide Endoscopy System (Intuitive Surgical Inc., Sunnyvale, CA) and the Invendoscopy E210 System (Ambu, ballerup, Denmark) are flexible self-propelling colonoscopes that have had FDA approval since 2006 and 2016, respectively.[12,13] The lightweight systems are easier to manipulate and apply less force to the colon wall to reduce the colonic looping phenomena that is responsible for the majority of post-operative pain. Similarly, the Flex Robotic system (Medrobotics Corp., Raynham, MA, USA) is a robotic platform intended to increase accessibility to deep organs. Its current indications include transoral procedures and recent feasibility studies demonstrate a role for rectal cancer resection.[14] This platform has had FDA approval since 2015.

Although these systems are segregated into their own unique approaches, their enhanced visualization has spurred ongoing research to assess the potential to combine NOTES, LESS and other laparoscopic

robotic approaches for more diverse and complex surgical applications.

Bronchoscopy Platforms

Bronchoscopy and transthoracic needle aspiration are the two main approaches for diagnostic biopsy of peripheral lung lesions. However, the diagnostic yield of bronchoscopy ranges from 67-84% compared to 92% in needle aspiration due to "getting lost" in the peripheral airways.[15,16] The addition of robotic guidance systems, Monarch platform by Aurius health in 2018 and Ion Endoluminal System by Intuitive Surgical in 2019, has the potential to increase yields close to 95% and increase the ability to localize and precisely puncture peripheral nodules.[17,18] These systems increase structural support with a locking outer sheath but have an inner flexible controllable bronchoscope with electromagnetic navigation guidance and continuous visualization. This allows for 4-way adjustable angulation to reach farther than conventional bronchoscopy, prevent accidental displacement during sampling, and visualize and tamponade bleeding.[19] As this technology expands, there is the potential for adding ablative therapies for the treatment of oligometastatic or inoperable peripheral lung tumors.[20]

Ureteroscopy Platforms

Ergonomic deficiencies during stone manipulation, laser disintegration, surgeon fatigue, exposure to radiation during fluoroscopy and the need for assistance while performing ureteroscopy has led to the development of robotic ureteroscopes. The first clinical application of robotic ureteroscopy (URS) was introduced in 2008 by Desai and colleagues with the Sensei-Magellan system.[21] Since that time the Avicenna Roboflex system, introduced in 2013, has been the only system that remains in clinical use.[22] The surgeon sits at the console controlling a flexible arm that can rotate, advance, retract and deflect as well as manipulate irrigation, lasers and stone baskets. Treatment times, safety profiles, and three-month stone free rates were similar to conventional URS.[23] As these systems develop, the ability to have tactile feedback from the tip of the ureteroscope, 3D positioning and memory to find stones based off of pre-operative imaging, and adaptive intelligent control of the laser settings as the stone is being fragmented could allow for further utility and offset the higher cost of using a robotic system.[22]



Image 2. The Procept aquabeam handpiece

Aquablation

Aquablation (AA) is the newest robotic platform being used in the field of urology. It is a surgeon-planned, ultrasound-guided, robotically executed technique to resect prostate tissue athermally using a high velocity water jet.[24] It is approved for use in men with benign prostatic hyperplasia with 30-150g prostates with or without a median lobe.

The AA system, developed by Procept, uses a transrectal ultrasound to visualize the prostate. The proprietary software is then used to map the prostate dimensions and the anatomic areas of the median lobe, transitional zone and peri-verumontanum tissue (Image 2, 3). A robotically controlled transurethral waterjet is then programmed by the surgeon to resect a specific amount of prostate tissue in a precise location, up to 0.25mm at a time. The robotic system is then activated by the surgeon and the pre-determined tissue is removed within minutes.

This has quickly gained popularity in the urologic community given its short learning curve,

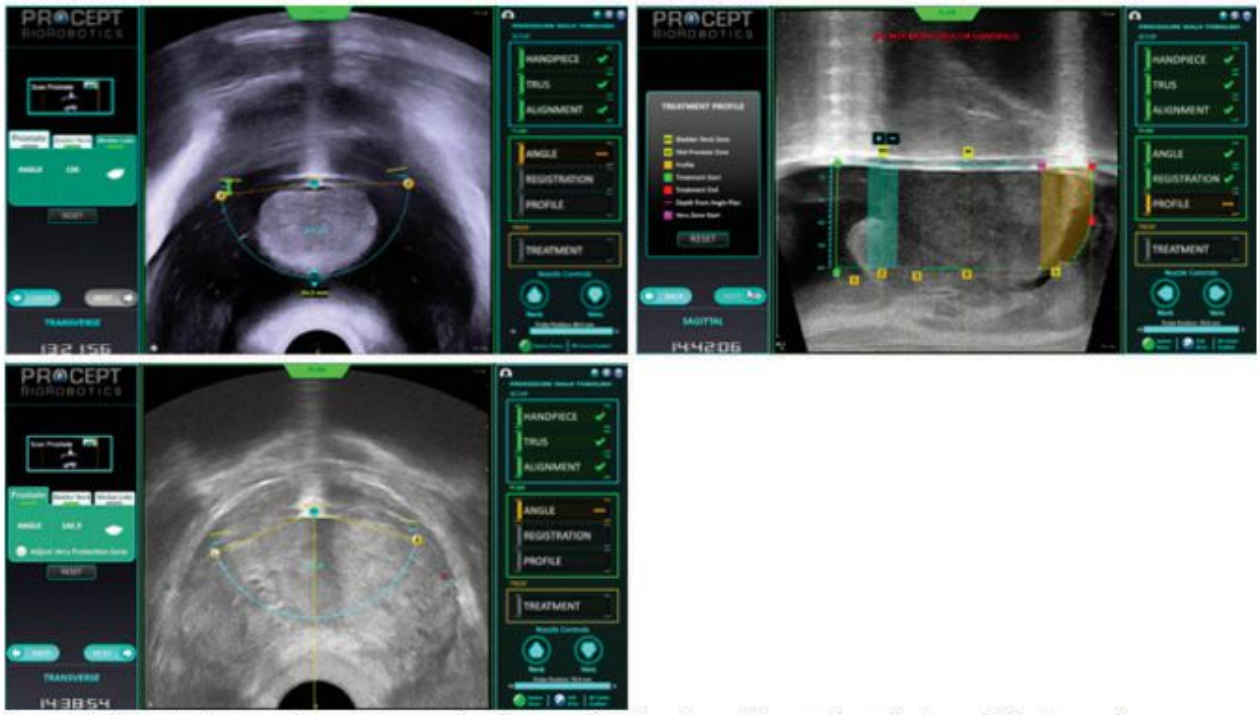


Image 3. Prostatic mapping of the median lobe, mid-gland, and dynamic points – sphincter and bladder neck

use of common urologic skills (transrectal ultrasound and rigid cystoscopy), short operative times, wider range of patient inclusion and encouraging clinical outcomes. There is a 6% incidence of restarting medications or requiring surgical retreatments compared to 12.3% with the gold standard TURP, preservation of sexual function and a 50% reduction in retrograde ejaculation compared to alternative surgical approaches for BPH.[25,26] Due to these advantages, the number of AA systems has increased by 52% over the last 9 months.

Robotic Arm-assisted Arthroplasty

The success of knee and hip arthroplasty relies on surgeons' technical ability to achieve optimal position and alignment of the prosthesis.[27] Computer-assistance with navigation allows for pre-planning and real-time intra-operative feedback while the robotic arm stabilizes and optimizes positioning of the prosthesis and surgical instruments. ROBODOC (Integrated surgical systems, Davis, CA, USA), introduced in 1992, was an active system independently performing the osteotomy and only allowed for the surgeon to start and stop the procedure.

A semi-active system, such as RIO by MAKO Surgical Corp., is now the most widely used system in modern orthopedics. Using a pre-operative extremity CT scan, a 3D model with pre-programmed boundaries is created that limits the range of movement of the surgical instruments controlled by the robotic arm.[28,29] All actions, such as reaming and osteotomy, are performed according to pre-operative planning, but the final manipulation depends on surgeon execution.

Compared to traditional total hip arthroplasty, the MAKO system can reduce varus and valgus deformities of the femoral prosthesis, restore the offset, and more accurately position the acetabular cup prosthesis in the safety zone.[30,31,32,33] Although several studies report the benefit for less intra-operative blood loss, low complication rate and shorter hospital stay with robotic systems, the literature doesn't report a significant difference in short-term clinical efficacy through the Harris hip score or other scoring systems.[33,34]

The FDA has approved MAKO and NAVIO surgical robots for uni-compartmental/total knee

arthroplasty (UKA/TKA) as well. These systems employ visual, tactile and auditory feedback to improve surgical efficiency and achieve a more precise osteotomy alignment.[35]

Over the last decade, use of robotic systems for UKA has more than doubled in certain regions of the US due to the definite and repeatable evidence of improved femoral and tibial prosthetic positioning accuracy by 3 and 3.4 times, respectively.[36,37] Additionally, soft tissue balance improves and the joint dynamics of the knee have better function and greater longevity.[35]

Patients report superior pain scores at 2 months and functional scores at 3 months when compared with traditional approaches.[38] Continued long-term, prospective and randomized-controlled trials are needed to further study the use of MAKO in TKA patients.

Spine Robot Platforms

The Mazor robotic platform (Medtronic Navigation, Louisville, CO, USA; Medtronic Spine, Memphis, TN, USA) was first FDA approved in 2004 and is currently one of the most popular systems in the world for spinal pedicle screw placement. The initial system, SpineAssist, had a patient-mounted track that used pre-operative or intra-operative CT imaging to plan hardware trajectory. The current model, Mazor X Stealth Edition, released in 2019, no longer requires a patient-mounted track, has faster computing speed, the vertebral bodies can be registered individually, and an optic camera allows for self-detection of the robot to avoid intra-operative collisions.

ExcelsiusGPS (Globus Medical, Inc., Audubon, PA, USA) was the first spine robot with a fully integrated navigation platform, real-time instrument tracking and pedicle screw placement without guidewires. Similar to the Mazor X Stealth Edition, intra-operative fluoroscopic imaging can be merged with pre-operative CT scans (Image 3).

These robotic systems aid in maintaining a fixed working angle to reduce inaccuracies and tremors by the surgeon and thus allow for more consistent, safe and improved patient outcomes. While, studies demonstrate a high accuracy rate of robotic assisted pedicle screw placement (91-98%) comparisons with traditional approaches have not shown an overwhelming improvement in accuracy.[39,40,41,42] A recent meta-analysis has demonstrated decreased screw revision risk when using robotic-assisted and navigated screw placement over free-hand techniques.[43] Additionally, robotic assistance can reduce the risk of proximal facet joint violation, compared to freehand techniques, which can minimize the risk of adjacent segment disease.[44,45,46]

Limitations to robotic spinal surgery include cost, lack of diverse indications for its use, increased operative times and lack of direct evidence of benefit. In fact, several database studies report increased risk of re-operation, 30 day readmission rate and complication rate with robotic assistance in lumbar spinal fusion.[47,48] While the technology continues to improve, more research is needed to increase the implementation of robotics in spine surgery.

The Future

Two on-going areas of intense research include robotic telesurgery and micro-robotics. The new 5G network deployed by telecom companies worldwide offers rapid communication and the potential for telesurgery in order to reduce health care costs and improve patient access to quality care. Studies have determined that a lag time <400ms is imperceptible to the surgeon and this can be achieved using 5G networks. Telesurgery has been performed in China, Germany, Italy and Spain with promising results. Remote nephrectomy was safely performed without complication and conversion with a median distance of 187km between surgeon and patient. The median round trip delay was 26ms and total delay was 200ms.[49,50] Over time, the extent and feasibility of telesurgery will continue to be tested. This is promising for use on aircraft carriers, for future space travel and in underserved areas worldwide. As of this year, however, a radical cystectomy was performed remotely from a distance of nearly 3000km with an average total delay lag time of 254ms.[51]

Investigative micro-robotic prototypes are freely mobile capsule endoscopes that offer a variety of diagnostic, targeted drug delivery and surgical applications.[52] One such robot is a millimeter in size and has been, in porcine models, directed with extracorporeal magnets to apply a single functional nitinol clip and stop colonic bleeding following biopsy.[53]

Research is ongoing around four categories specific to micro-robotics: contained propulsion, miniaturized functionality, accurate telemanipulation and consistent visualization.[2]

Propulsion can be achieved externally via electromagnetic fields or ultrasonographic energy while internally driven systems are more restricted as they require chemical reactions for motion and a separate navigational source.[9,54] Several studies have demonstrated proof-of-concept for cutting, grasping and ablation on the micro-scale.[55] There is also early promise with phototaxis of polysterene beads and magnetically directed chrome spheres using MRI for telemanipulation. Finally, studies have demonstrated the ability for live-tracking of ferromagnetically labeled microalgae to model microbots in rats, X-ray angiography tracking of a radio-dense robot in the aorta of a rabbit, and US tracking of a magnetically-labeled robot through muscle tissue in a chicken model.[56,57,58,59]

The age of microbots is clearly in its infancy. However, as further research is performed to address safety to the patient and operator, cost and accessibility, creation of 3D tracking and the potential role of fluorescence, significant advancements could create another major paradigm shift in minimally invasive surgery.

Conclusion

Robotic technology has created a unique paradigm shift in the surgical care of the patient beginning several decades ago. With new systems and applications routinely coming to market, it continues to be an exciting avenue for new research and development as well as improving the patient care experience. As advancements in computer processing, optics, mechanics and haptic feedback continue to rapidly progress, robotic surgery has cemented its role in modern surgical medicine and its near universal adoption across surgical and medical specialties offers promise to continue to improve medical care around the world.

References

1. Alluri RK, Avrumova F, Sivaganesan A, Vaishnav AS, Lebl DR, Qureshi SA. Overview of Robotic Technology in Spine Surgery. HSS J. 2021 Oct;17(3):308-316. doi: 10.1177/15563316211026647. Epub 2021 Jul 1.
2. Khandalavala K, Shimon T, Flores L, Armijo PR, Oleynikov D. Emerging surgical robotic technology: a progression toward microbots. Ann Laparosc Endosc Surg 202;5:3.
3. Golla V, Williams SB. Cost-effectiveness of Robotic-Assisted Prostatectomy in the UK—Are We Doing Enough? JAMA Netw Open. 2022;5(4):e225747.
4. Carbonnel M, Moawad GN, Tarazi MM, Revaux A, Kennel T, Favre-Inhofer A, Ayoubi JM. Robotic Hysterectomy for Benign Indications: What Have We Learned from a Decade? JSLS. 2021 Jan-Mar;25(1):e2020.00091.
5. Lantz A, Bock D, Akre O, Angenete E, Bjartell A, Carlsson S, Modig KK, Nyberg M, Kollberg KS, Steineck G, Stranne J, Wiklund P, Haglind E. Functional and Oncological Outcomes After Open Versus Robot-assisted Laparoscopic Radical Prostatectomy for Localised Prostate Cancer: 8-Year Follow-up. Eur Urol. 2021.
6. Ramirez PT, Frumovitz M, Pareja R, Lopez A, Vieira M, Ribeiro R, Buda A, Yan X, Shuzhong Y, Chetty N, Isla D, Tamura M, Zhu T, Robledo KP, Gebski V, Asher R, Behan V, Nicklin JL, Coleman RL, Obermair A. Minimally Invasive versus Abdominal Radical Hysterectomy for Cervical Cancer. N Engl J Med. 2018 Nov 15;379(20):1895-1904.
7. Shearman AD, Sephton BM, Wilson J, Nathwani DK. Robotic-assisted unicompartmental knee arthroplasty is associated with earlier discharge from physiotherapy and reduced length-of-stay compared to conventional navigated techniques. Arch Orthop Trauma Surg. 2021 Dec;141(12):2147-2153.
8. Witt RG, Hirata Y, Prakash LR, Newhook TE, Maxwell JE, Kim MP, Tran Cao HS, Lee JE, Vauthey JN, Katz MHG, Tzeng CD, Ikoma N. Comparative analysis of opioid use between robotic and open pancreatoduodenectomy. J Hepatobiliary Pancreat Sci. 2022 Jul 7.
9. Ceylan H, Yasa IC, Kilic U, et al. Translational prospects of untethered medical microrobots. Progress in Biomedical Engineering 2019;1:012002.
10. Cadeddu JA. Re: Early Experience with the Senhance®-Laparoscopic/Robotic Platform in the US. J Urol 2019:10109701JU 0000576800809701c.
11. Rassweiler JJ, Autorino R, Klein J, et al. Future of robotic surgery in urology. BJU Int 2017;120:822-41.
12. Eickhoff A, van Dam J, Jakobs R, et al. Computer-assisted colonoscopy (the NeoGuide Endoscopy System): results of the first human clinical trial ("PACE study"). Am J Gastroenterol 2007;102:261-6.
13. Groth S, Rex DK, Rösch T, et al. High cecal intubation rates with a new computer-assisted colonoscope: a feasibility study. Am J Gastroenterol 2011;106:1075-80.
14. Carmichael H, D'Andrea AP, Skancke M, et al. Feasibility of transanal total mesorectal excision (taTME) using the Medrobotics Flex® System. Surg Endosc 2020;34:485-91.
15. Folch EE, Pritchett MA, Nead MA, et al. Electromagnetic Navigation Bronchoscopy for Peripheral Pulmonary Lesions: One-Year Results of the Prospective, Multicenter NAVIGATE Study. J Thorac Oncol 2019;14:445-58.
16. DiBardino DM, Yarmus LB, Semaan RW. Transthoracic needle biopsy of the lung. J Thorac Dis 2015;7:S304-16.
17. Chen A, Silvestri GA, Gildea TR, et al. Robotic Bronchoscopy for Peripheral Pulmonary Lesions: A Multicenter Pilot and Feasibility Study (BENEFIT). Chest New Orleans.
18. Yarmus LB, Mallow C, Pastis N, et al. First-in-Human Use of a

Hybrid Real-Time Ultrasound-Guided Fine-Needle Acquisition System for Peripheral Pulmonary Lesions: A Multicenter Pilot Study. Respiration 2019;98:527-33.
19. Chen AC, Gillespie CT. Robotic Endoscopic Airway Challenge: REACH Assessment. Ann Thorac Surg2018;106:293-7.
20. Agrawal A, Hogarth DK, Murgu S. Robotic bronchoscopy for pulmonary lesions: a review of existing technologies and clinical data. J Thorac Dis. 2020 Jun;12(6):3279-3286.
21. Aron M, Desai MM. Flexible robotics. Urol Clin North Am 2009;36:157-62, viii.
22. Rassweiler-Seyfried MC, Herrmann J, Klein J, Michel MS, Rassweiler J, Grüne B. Robot-assisted flexible ureterorenoscopy: state of the art in 2022. Mini-invasive Surg 2022;6:41.
23. Geavlete P et al. Robotic flexible ureteroscopy versus classic flexible ureteroscopy in renal stones: The initial Romanian experience. Chir 2016;111:326-9.
24. Probst P, Desai M. Expectations Facing Reality: Complication Management after Aquablation Treatment for Lower Urinary Tract Symptoms. Eur Urol Focus. 2022 May 16:S2405-4569(22)00109-2.
25. Gilling, Peter. WATER: A Double-Blind, Randomized, Controlled Trial of Aquablation vs Transurethral Resection of the Prostate in Benign Prostatic Hyperplasia. J Urol 2018; 199: 1252-1261.
26. Desai, Mihir. Aquablation for benign prostatic hyperplasia in large prostate (80-150mL): 6-month results from the WATER II trial. BJU Int 2019; 8(1):e1498.
27. Mason J.B., Fehring T.K., Estok R., et al. Meta-analysis of alignment outcomes in computer-assisted total knee arthroplasty surgery. J Arthroplasty. 2007;22:1097–1106. doi: 10.1016/j.arth.2007.08.001.
28. Bargar W.L. Robots in orthopaedic surgery: past, present, and future. Clin Orthop Relat Res. 2007;463:31–36.
29. van der List J.P., Chawla H., Joskowicz L., et al. Current state of computer navigation and robotics in unicompartmental and total knee arthroplasty: a systematic review with meta-analysis. Knee Surg Sports Traumatol Arthrosc. 2016;24:3482–3495.
30. Amirouche F., Solitro G.F., Chandrasekaran S., et al. Validating a modified circle theorem method for the measurement of acetabular cup anteversion on plain radiography with intra-operative data from robotic assisted total hip arthroplasty. J Arthroplasty. 2016;31:323–329.
31. Domb B.G., Redmond J.M., Louis S.S., et al. Accuracy of component positioning in 1980 total hip arthroplasties: a comparative analysis by surgical technique and mode of guidance. J Arthroplasty. 2015;30:2208–2218.
32. Lim S.J., Ko K.R., Park C.W., et al. Robot-assisted primary cementless total hip arthroplasty with a short femoral stem: a prospective randomized short-term outcome study. Comput Aided Surg. 2015;20:41–46.
33. Chen X., Xiong J., Wang P., et al. Robotic-assisted compared with conventional total hip arthroplasty: systematic review and meta-analysis. Postgrad Med. 2018;94:335–341.
34. Siebel T., Käfer W. Clinical outcome following robotic assisted versus conventional total hip arthroplasty: a controlled and prospective study of seventy-one patients. Z Orthop Ihre Grenzgeb. 2005;143:391–398.
35. Chen X, Deng S, Sun ML, He R. Robotic arm-assisted arthroplasty: The latest developments. Chin J Traumatol. 2022 May;25(3):125-131.
36. Boylan M., Suchman K., Vigdorchik J., et al. Technology-assisted hip and knee arthroplasties: an analysis of utilization trends. J Arthroplasty. 2017;33:1019–1023.
37. Citak M., Suero E.M., Citak M., et al. Unicompartmental knee arthroplasty: is robotic technology more accurate than conventional technique? Knee. 2013;20:268–271.
38. Blyth M.J.G., Anthony I., Rowe P., et al. Robotic arm assisted versus conventional unicompartmental knee arthroplasty: exploratory secondary analysis of a randomised controlled trial. Bone Joint Res. 2017;6:631–639.
39. Devito DP, Kaplan L, Dietl R, et al. Clinical acceptance and accuracy assessment of spinal implants guided with SpineAssist surgical robot: retrospective study. Spine (Phila Pa 1976). 2010;35:2109–2115.
40. Kantelhardt SR, Martinez R, Baerwinkel S, Burger R, Giese A, Rohde V. Perioperative course and accuracy of screw positioning in conventional, open robotic-guided and percutaneous robotic-guided, pedicle screw placement. Eur Spine J. 2011;20:860–868.
41. Keric N, Doenitz C, Haj A, et al. Evaluation of robot-guided minimally invasive implantation of 2067 pedicle screws. Neurosurg Focus. 2017;42:E11.
42. Ringel F, Stuer C, Reinke A, et al. Accuracy of robot-assisted placement of lumbar and sacral pedicle screws: a prospective randomized comparison to conventional freehand screw implantation. Spine (Phila Pa 1976). 2012;37(8):E496–E501.
43. Staartjes VE, Klukowska AM, Schroder ML. Pedicle screw revision in robot-guided, navigated, and freehand thoracolumbar instrumentation: a systematic review and meta-analysis. World Neurosurg. 2018;116:433–443.
44. Hyun SJ, Kim KJ, Jahng TA, Kim HJ. Minimally invasive robotic versus open fluoroscopic-guided spinal instrumented fusions: a randomized controlled trial. Spine (Phila Pa 1976). 2017;42(6):353–358.
45. Li HM, Zhang RJ, Shen CL. Accuracy of pedicle screw placement and clinical outcomes of robot-assisted technique versus conventional freehand technique in spine surgery from nine randomized controlled trials: a meta-analysis. Spine (Phila Pa 1976). 2020;45:E111–E119.
46. Zhou LP, Zhang RJ, Li HM, Shen CL. Comparison of cranial facet joint violation rate and four other clinical indexes between robot-assisted and freehand pedicle screw placement in spine surgery: a meta-analysis. Spine (Phila Pa 1976). 2020;45(22):E1532–E1540.
47. Lieber AM, Kirchner GJ, Kerbel YE, Khalsa AS. Robotic-assisted pedicle screw placement fails to reduce overall postoperative complications in fusion surgery. Spine J. 2019;19(2):212–217.
48. Yang DS, Li NY, Kleinhenz DT, Patel S, Daniels AH. Risk of postoperative complications and revision surgery following robot-assisted posterior lumbar spinal fusion. Spine (Phila Pa 1976). 2020;45(24):E1692–E1698.
49. Legeza P, Britz GW, Shah A, Sconzert K, Sungur JM, Chinnadurai P, Sinha K, Lumsden AB. Impact of network performance on remote robotic-assisted endovascular interventions in porcine model. J Robot Surg. 2022 Feb;16(1):29-35.
50. Li J, Yang X, Chu G, et al. Application of Improved Robot-assisted Laparoscopic Telesurgery with 5G Technology in Urology. Eur Urol. 2022 Jul8:S0302-2838(22)02469-1.
51. Yang X, Wang Y, Jiao W, et al. Application of 5G technology to conduct tele-surgical robot-assisted laparoscopic radical cystectomy. Int J Med Robot. 2022 Aug;18(4):e2412.
52. Ciuti G, Caliò R, Camboni D, et al. Frontiers of robotic endoscopic capsules: a review. J Microbio Robot 2016;11:1-18.
53. Valdastri P, Quaglia C, Susilo E, et al. Wireless therapeutic endoscopic capsule: in vivo experiment. Endoscopy 2008;40:979-82.
54. Chautems C, Zeydan B, Charreyron S, et al. Magnetically powered microrobots: a medical revolution underway? Eur J Cardiothorac Surg 2017;51:405-7.
55. Kirson ED, Yaari Y. A novel technique for micro-dissection of neuronal processes. J Neurosci Methods 2000;98:119-22.
56. Martel S. Swimming microorganisms acting as nanorobots versus artificial nanorobotic agents: A perspective view from an historical retrospective on the future of medical nanorobotics in the largest known three-dimensional biomicrofluidic networks. Biomicrofluidics 2016;10:021301.
57. Belharet K, Folio D, Ferreira A. MRI-based microrobotic system for the propulsion and navigation of ferromagnetic microcapsules. Minim Invasive Ther Allied Technol 2010;19:157-69.
58. Park S, Cha K, Park J. Development of Biomedical Microrobot for Intravascular Therapy. Int J Adv Robot Syst 2010;7:1.
59. Hu W, Lum GZ, Mastrangeli M, et al. Small-scale soft-bodied robot with multimodal locomotion. Nature 2018;554:81-5.
60. Almujalhem A, Rha KH. Surgical robotic systems: What we have now? A urological perspective. BJUI Compass 2020; 1: 152–159.

Disclosure

None reported. **MM**



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