Tuning the Force, Speed, and Efficiency of an Autonomous Chemically Fueled Information Ratchet

Stefan Borsley, David A. Leigh,* Benjamin M. W. Roberts,§ and Inigo J. Vitorica-Yrezabal

ABSTRACT: Autonomous chemically fueled molecular machines that function through information ratchet mechanisms underpin the nonequilibrium processes that sustain life. These biomolecular motors have evolved to be well-suited to the tasks they perform. Synthetic systems that function through similar mechanisms have recently been developed, and their minimalist structures enable the influence of structural changes on machine performance to be assessed. Here, we probe the effect of changes in the fuel and barrier-forming species on the nonequilibrium operation of a carbodiimide-fueled rotaxane-based information ratchet. We examine the machine’s ability to catalyze the fuel-to-waste reaction and harness energy from it to drive directional displacement of the macrocycle. These characteristics are intrinsically linked to the speed, force, power, and efficiency of the ratchet output. We find that, just as for biomolecular motors and macroscopic machinery, optimization of one feature (such as speed) can compromise other features (such as the force that can be generated by the ratchet). Balancing speed, power, efficiency, and directionality will likely prove important when developing artificial molecular motors for particular applications.

INTRODUCTION

Autonomous chemically fueled information ratchets1,2 have recently been used as engines to drive synthetic molecular motors3−5 and pumps.6−10 Given the ubiquity of such processes in biology,11 understanding how to maximize their performance is important for synthetic molecular nanotechnology12−18 and artificial chemical fueling systems.19−22 With macroscopic machinery, optimizing one aspect of performance often compromises others. For example, diesel engines need to produce substantial torque to pull heavy loads, while gasoline engines are generally designed to maximize speed and/or efficiency.23 Features of biomolecular machines have evolved to similarly suit their functions:24−27 to transport cargo,28,29 a motor needs to be fast to beat diffusion, while the external force it can successfully pull against is less important. Conversely, speed of operation is less consequential for motor proteins in muscle cells,11,30 while the force they can exert is the primary requirement. Often trade-offs come from prioritizing a particular function:24−29,31 for example, for a macroscopic heat engine, conditions that give the maximum power output cannot also result in the most efficient operation.32

Distinct performance characteristics of molecular ratchets include speed, power, and efficiency. For autonomous chemically fueled information ratchets,4−7 these features relate to the directionality achieved and to the rate and efficiency of catalysis of the fuel-to-waste reaction by the ratchet.33−36 Appreciating how to manipulate these features and understanding the connections between them should help in optimizing molecular ratchet designs for specific tasks.

The carboxylate-catalyzed hydration of carbodiimides has emerged as a robust, reliable, and versatile fuel-to-waste process for autonomously fueling molecular machines,5,6 paralleling its use in driving transient bond formation7,37,38 and dissipative assembly.39−41 The fuel cycle is particularly amenable for tuning in rotaxane-based information ratchets as the initially formed O-acyl urea can be displaced with a second nucleophilic catalyst42−44 that can act as a barrier-forming species (Figure 1). This allows the structure of both the fuel and the nucleophilic catalyst to be varied independently without affecting the structure of the carboxylate catalyst (here, a rotaxane ratchet).

Previously, it has been demonstrated that the distribution of the macrocycle between the two fumaramide sites in rotaxane ratchet 1 (Figure 1) can be driven away from the equilibrium by the hydration of disisopropylcarbodiimide (DIC).6 The DIC reacts with the carboxylic acid of the axle, transiently forming an O-acyl urea barrier to macrocycle shuttling. The O-acyl urea

Received: July 24, 2022
Published: September 8, 2022
is quickly displaced by a nucleophilic catalyst [hydroxybenzotriazole (HOBt) or pyridine] to form a barrier that is hydrolyzed slowly on the time scale of macrocycle shuttling kinetics. Directional motion is generated through kinetic asymmetry \(^{1,34\text{-}36}\) in the chemical engine cycle.\(^3\) It arises from differences in rates of the machine co-conformers in both their reaction with DIC and in the hydrolysis of the barrier (the machine is doubly kinetically gated),\(^6\) the former being a classic Curtin–Hammett principle scenario.\(^3,43\)

Here, we explore the relationships between the force, speed, and efficiency in molecular ratchet 1 (Figure 1). The X-ray crystal structure of 1 (Figure 2) shows structural features consistent with how kinetic asymmetry likely arises within the chemical engine cycle (Figure 3). We examine the effect of varying the structure of the fuel (Figure 4) and barrier (Figure 5) on the operation of the ratchet and consider the design implications for the development of future ratchets.

**RESULTS AND DISCUSSION**

**Relating Force, Speed, and Efficiency to the Operation of an Information Ratchet.** Upon fueling with carbodiimide fuels, ratchet 1 performs work by pumping the macrocycle away from the equal distribution between the fumaramide sites on the axle at equilibrium in 1 to a nonequilibrium distribution in the 1/1' mixture (Figure 1).\(^6\) The further the distribution is driven away from the equilibrium value, the more work the ratchet has to do to reach and maintain the out-of-equilibrium state. The ratio of dist-1'/prox-1' (where prime denotes any species where macrocycle shuttling between the fumaramide groups is blocked) in the steady state is a direct measure of the efficiency of the ratchet.

---

**Figure 1.** Chemomechanical cycle of carbodiimide-fueled molecular ratchet 1. The macrocycle in 1 incessantly shuttles between two fumaramide binding sites (green), spaced non-equidistantly from either side of a carboxylate catalyst (red) on the axle. Ratchet 1 reacts with a carbodiimide fuel (blue) and a nucleophile (purple) to form a bulky activated ester, 1', in which the macrocycle is blocked from shuttling between the fumaramide sites. Hydrolysis of the ester under the fueling conditions regenerates 1. Esterification occurs preferentially from co-conformer dist-1 due to the steric clash between the fuel and the macrocycle, while activation of the ester by the hydrogen bonding from the macrocycle results in faster hydrolysis of prox-1' (disfavored, slower reaction pathways shown with faded arrows), resulting in pumping of the macrocycle to the distal fumaramide site. Equilibrium arrows are used for thermodynamic consistency (to obey microscopic reversibility), although under the experimental conditions, the backward steps are negligible. The reaction with p-methoxybenzylamine results in a kinetically stable amide as a permanent barrier, allowing the dist-2/prox-2 ratio to be determined by \(^{1}\)H NMR spectroscopy.
ratcheting constant, $K_r$, (Supporting Information, Section S4.2), which quantifies kinetic asymmetry.\textsuperscript{1,33} Under these circumstances, with a negligible mechanical exchange between the proximal co-conformer of ratchet 1 (the t-butyl ester of 1) and distal co-conformer, $K_r$ quantifies the energy stored by the ratcheting process.\textsuperscript{34,44} In turn, the energy stored is related to the average force a ratchet can exert when pushing against a load, as force can be represented as a change in energy divided by distance. Qualitatively, the higher the energy stored by the ratchet (i.e., the higher the directionality), the higher the force that can be produced by its operation.\textsuperscript{31}

The "speed" of the ratchet operation (i.e., the net forward chemomechanical cycle rate) is the rate of forward cycles that allow work to be done compared to the rate of backward cycles that undo that work and the rate of futile cycles that do no work but still consume fuel.\textsuperscript{2,33,34} This means that both the rate of catalysis and the directionality of the ratchet contribute to the speed of a motor. Likewise, the (potential) power of the ratchet is the force the ratchet produces (or could produce) at the net rate at which it proceeds forward around the chemomechanical cycle.\textsuperscript{31}

"Machine efficiency" is the fraction of the available energy from the fuel-to-waste reaction that is harnessed productively by the machine, that is, the number of forward steps per unit fuel. Machine efficiency can be divided into "catalytic efficiency"—the proportion of the fuel-to-waste reactions that proceed by the machine-catalyzed pathway—and "thermodynamic efficiency"—the proportion of energy released from the fuel-to-waste reaction that is harnessed by the machine. In common with the force, power, and speed, the thermodynamic efficiency is linked to the kinetic asymmetry (and hence the directionality) of the operation.\textsuperscript{31}

Understanding how structural features affect directionality and the rate and efficiency of fuel use by ratchets is therefore key for tuning the force, speed, power, and efficiency of a ratchet in driving nonequilibrium functions and processes.

**X-ray Crystal Structure of Ratchet 1.** We previously found\textsuperscript{1} that fueling ratchet 1 with DIC in the presence of HOBT as a barrier-forming species drives the macrocycle distribution from the equilibrium value of 50:50 to 5:95 in favor of the distal co-conformer. The directionality arises from kinetic gating of both the fuel addition and barrier hydrolysis steps. This can be rationalized by the steric clash between the fuel and the macrocycle, resulting in a slower rate of fuel addition to co-conformer prox-1 than to dist-1, while hydrogen bonding between the macrocycle and the carboxylate group results in faster barrier hydrolysis of prox-1 OBt\textsuperscript{●} than of dist-1 OBt\textsuperscript{●}. We attempted X-ray crystallography on a range of derivatives of 1 to see if we could obtain solid-state structural evidence in support of either or both parts of this putative mechanism.

A single crystal of prox-1 OBt\textsuperscript{●} (the t-butyl ester of ratchet 1) suitable for X-ray diffraction was obtained by slow evaporation of a solution of rotaxane in CH\textsubscript{2}Cl\textsubscript{2}/MeOH (Figure 2 and Section S7, Supporting Information, CCDC deposition code: 2191007). The solid-state structure shows four NH···O hydrogen bonds between the amides of the macrocycle and the fumaramide carbonyls on the thread (average NH···O distance = 2.30 Å). The bulky tert-butyl barrier is oriented away from the macrocycle, reflective of the steric clash between the macrocycle and the barrier envisaged as being responsible for the difference in the rate of fuel addition between dist-1 and the more sterically encumbered prox-1 co-conformer (Figure 3a).

In the solid-state structure, hydrogen bonding is also apparent between the macrocycle and the carbonyl of the ester. The macrocycle is puckered, with the upper far side (as viewed in Figure 2B) angled toward the ester. Two hydrogen bonds are present between the macrocycle and the ester carbonyl, from the amide N–H (Figure 2B, green, NH···O distance = 2.53 Å, and angle = 118.6°) and from a benzylic C–H hydrogen atom (Figure 2B, blue, CH···O distance = 2.53 Å, and angle = 126.8°). Such hydrogen bonding would polarize the C=O bond of the ester.\textsuperscript{36} If present during the chemomechanical cycle in solution, this interaction would stabilize the developing negative charge\textsuperscript{36} at that oxygen atom of the proximal co-conformer in the transition state during barrier hydrolysis, leading to faster hydrolysis of dist-1\textsuperscript{●} than of prox-1\textsuperscript{●} (Figure 3B).

**Varying the Fuel Structure.** With X-ray crystallography providing structural evidence in support of the origin of directional selectivity in the operation of information ratchet 1, we next explored the effects of changing the structure of the fuel. The original DIC fuel was compared to two other
common carbodiimide coupling agents,\textsuperscript{51} dicyclohexylcarbodiimide (DCC) and 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide methyliodide (EDC·\textsuperscript{\textbullet}MeI)\textsuperscript{52} (Figure 4). Experiments were conducted using the conditions optimized for directionality\textsuperscript{6} \{[1] = 2.5 mM, [barrier-forming species] = 5.0 mM, and [fuel] = 12.5 mM in 2-(N-morpholino)-ethanesulfonic acid (MES)-buffered (100 mM, pH \textit{obs} 5.36) MeCN-d\textsubscript{3}/H\textsubscript{2}O (7:3 \textit{v}/\textit{v}), Section S2.1, Supporting Information}. These conditions ensure that the vast majority (approximately 95\%) of rotaxane 1/1\textsuperscript{′} is present as 1\textsuperscript{′} while fuel remains during the reaction.\textsuperscript{6} The operating conditions were buffered to acidic conditions at which carbodiimide activation is effective\textsuperscript{53} with phosphate buffers avoided because of the known issue of phosphate-induced decomposition of carbodiimides.\textsuperscript{54} The rate of the fuel-to-waste reaction was monitored by \textsuperscript{1}H nuclear magnetic resonance (NMR) spectroscopy. The macrocycle distribution formed under out-of-equilibrium fueling was kinetically trapped at different stages by the addition of \textit{p}-methoxybenzylamine (Figure 1, bottom), allowing the evolution of directionality to be determined by \textsuperscript{1}H NMR of the kinetically trapped species. The results of the experiments with the different fuels are shown in Table 1.

As evidenced by the directionality observed upon the addition of \textit{p}-methoxybenzylamine at 0 h and modeling of the kinetics (Supporting Information, Section S4), only the gating of the fueling step is significantly affected by these variations in the fuel structure (Table 1). This confirms that neither the carbodiimide nor the formation of the urea waste product plays a role in the barrier removal step. Ratchet 1 gives better directionality with either DCC or EDC·MeI (Table 1, entries 2 and 3) than with DIC (Table 1, entry 1) (Figure 4A,B). DCC is bulkier than DIC, which exacerbates the steric clash when the macrocycle is on the proximal fumaramide site (Figure 3B). Steric issues may also contribute to the better fueling gating observed with EDC·MeI (Table 1, entry 3). However, the positive charge on the quaternary ammonium group of this fuel leads to faster reaction rates with carboxylic anions.\textsuperscript{55} This effect may be greater for \textit{dist}-1 than for \textit{prox}-1, if hydrogen bonding similar to that observed with the ester in Figure 2 also polarizes the carboxylate. Both steric and electronic effects likely lead to EDC·MeI providing the best fueling gating of the three carbodiimide fuels investigated.

Directionality is not the only important factor to consider in molecular ratchet design.\textsuperscript{24−29,31} The rate of the machine-catalyzed fuel-to-waste reaction determines how fast a motor can operate.\textsuperscript{1,31,33,36} Here also, EDC·MeI was the preferable carbodiimide fuel as it reacts faster, which combined with the better directionality means that the ratchet undergoes net forward chemomechanical cycles at over double the rate with EDC·MeI compared to that with DIC or DCC (Figure 4C). Although DCC showed better directionality than DIC, they have a similar step rate due to the significantly slower reaction of DCC than that of DIC under these conditions.

Only very modest changes were observed in the overall machine efficiency under the conditions optimized for directionality.\textsuperscript{6} EDC·MeI showed a marginally better catalytic efficiency (Supporting Information, Section S3), with 64\% of the fuel-to-waste reactions proceeding via the machine-catalyzed pathway (compared to 52\% and 54\% for DIC and DCC, respectively). This improved catalytic efficiency and directionality result in the best overall efficiency for EDC·MeI (Figure 4D). However, errors propagated from the directionality measurements (Supporting Information, Section S6)
make it difficult to assess the overall efficiency with a high
degree of accuracy.

**Varying the Barrier Structure.** Altering the structure of
the barrier-forming species provides another opportunity to
influence the operating cycle of the molecular ratchet (Figure
1). The operation of the machine under the optimized
conditions (Section S2.1, Supporting Information) with
different barrier-forming species, HOBt and hydroxyazabenzo-
triazole (HOAt), was examined in combination with both DIC
and EDC·MeI. In line with the proposed mechanism of
operation (Figure 1), varying the barrier structure was found to
principally alter the gating of the ester hydrolysis step (Table
1).

HOBt gave rise to a better overall directionality of the
ratchet than HOAt (Figure S5A,B). However, the time at which
the maximum directional bias (steady state) was reached was
significantly decreased with HOAt. This trend was observed
using either fuel (Figure S5A) but is most obvious using DIC:
the maximum directionality was reached after 18 h with HOBt
but 4× faster (4.5 h) with HOAt [Figure S5A(i)].

The differences in the directionality and rates can be
rationalized from a consideration of the barrier structures. The
activated ester 1OAt is significantly more hydrolytically labile
than the 1OBt counterpart due to both the increased negative
charge stabilization (making OAt a better leaving group) and
hydrogen bonding to water via the pyridyl nitrogen (stabilizing
the transition state for hydrolysis; Figure 3).

Barrier hydrolysis is rate-limiting under these conditions, and
therefore, faster hydrolysis results in a faster machine-catalyzed
fuel-to-waste reaction, giving rise to faster cycling of the
chemical engine and more rapid buildup of directional bias.
The use of HOAt instead of HOBt increases the rate of cycling
3-fold with EDC·MeI fuel or 4-fold with DIC fuel (Figure 5C).

While HOAt results in a faster fuel-to-waste reaction, HOAt
also accelerates the background fuel-to-waste reaction (Supporting Information, Section S3.1), which reduces the
catalytic efficiency of the molecular ratchet (Supporting
Information, Section S3.1). Consequently, no significant
differences are observed in the overall machine efficiency
between HOBt and HOAt (Figure 5D).

**Tuning the Force, Speed, and Efficiency of Ratchet 1.**
For the operation of ratchet 1, EDC·MeI was found to be the
best fuel in terms of directionality (Figure 4B), as well as the
rate (Figure 4C) and efficiency (Figure 4D) with which it
reacts with the ratchet. Consequently, fueling with EDC·MeI
results in the highest force, speed, power, and efficiency of
operation of ratchet 1. Upon varying the barrier structure, the

---

**Figure 4.** Operation of ratchet 1 with different carbodiimide fuels ([I] = 2.5 mM, [HOBt] = 5.0 mM, and [fuel] = 12.5 mM in MES-buffered (100
mM, pHobs 5.36) MeCN-d3/H2O (7.3 v/v)). (A) Graph plotting the directionality as a ratio of the distal and proximal isomers changing as the
ratchet undergoes multiple cycles over time. (B) Maximum directionality reached by ratchet 1 under these conditions, with different carbodiimide
fuels. (C) Net forward cycling rate of machine 1 with different carbodiimide fuels (see Supporting Information Section S5 for details). (D) Net
forward steps per fuel equivalent, which corresponds to the overall efficiency of the machine as a percentage of the overall energy supplied by the
fuel-to-waste reaction that is productively harnessed by machine 1.56
picture is more complex. HOBt gives better directionality, while HOAt enables faster catalyst engine cycles. As the force that can be generated is proportional to directionality, the use of HOBt would be the best for a function that required a high force output. By contrast, if speed was more important (e.g., perhaps for transport using a catenane motor based on ratchet), HOAt would be more suitable as the faster rate of reaction results in faster net forward cycling despite lower directionality.

The changes between HOBt and HOAt illustrate not only the benefits but also the challenges in optimizing for a particular feature; improvements in one aspect of machine operation may lead to sacrifices in other properties. Different interactions with the solvent, varying amounts of the water content, waste production, and the pH may all influence different features of machine performance.

### CONCLUSIONS

In addition to directionality, the characteristics of force, speed, and efficiency of a molecular ratchet (1) can be tuned with a judicious choice of the fuel and barrier structure. While the latter three features are sometimes maximized together (changing the fuel from DIC to EDC-Mel, for example), there may be trade-offs to be made if one trait is particularly desired. The increase in the speed of the engine cycle that arises from using HOAt instead of HOBt is at the expense of poorer directionality. Consequently, the speed of the ratchet is increased, while the potential force output is decreased. For some tasks, the time that a nonequilibrium steady state can be maintained for may also be important.

For chemical engines based on 1, this would be best served by using either DCC or DIC as the fuel, rather than EDC-Mel, as DCC and DIC react more slowly.

---

**Table 1. Experimentally Determined Parameters for Directionality, Speed, and Efficiency of the Operation of Ratchet 1 with Different Fuel/Barrier Combinations**

| entry | fuel | barrier-forming species | overall directionality, dist-1/prox-1 | fuelling gating | waste-forming gating | net forward step rate/x10^2 h^-1 mM^-1 | overall efficiency |
|-------|------|-------------------------|--------------------------------------|----------------|---------------------|----------------------------------------|-------------------|
| 1     | DIC  | HOBt                    | 20.3 ± 2.0:1                         | 2.2 ± 0.03     | 9.3 ± 0.9           | 0.72 ± 0.15                            | 30% ± 6%          |
| 2     | DCC  | HOBt                    | 25.7 ± 2.6:1                         | 2.6 ± 0.04     | 9.4 ± 1.0           | 0.67 ± 0.15                            | 36% ± 8%          |
| 3     | EDC-Mel | HOBt                | 30.9 ± 2.9:1                         | 3.1 ± 0.04     | 9.7 ± 0.9           | 1.83 ± 0.36                            | 43% ± 8%          |
| 4     | DIC  | HOAt                    | 13.7 ± 1.5:1                         | 1.7 ± 0.04     | 8.0 ± 0.9           | 2.45 ± 0.56                            | 32% ± 7%          |
| 5     | EDC-Mel | HOAt                 | 24.3 ± 1.1:1                         | 3.5 ± 0.03     | 7.0 ± 0.3           | 5.03 ± 0.46                            | 40% ± 4%          |
These kinds of trade-offs in aspects of performance are ubiquitous in the design of macro-scale machines and are also evident in the behavior of biological molecular machines.\textsuperscript{12−17} It seems very likely, therefore, that the ability to tune the structures of chemical engines and fuels to suit particular tasks will prove important in the development of artificial molecular nanotechnology.

\section*{ASSOCIATED CONTENT}

\subsection*{Supporting Information}

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/jacs.2c07633.

Experimental methods and operations data and X-ray crystal structure determination method and data (PDF)

Accession Codes

CCDC 2191007 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via www.ccdc.cam.ac.uk/data_request/cif, or by emailing data_request@ccdc.cam.ac.uk, or by contacting The Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; fax: +44 1223 336033.

\section*{AUTHOR INFORMATION}

\subsection*{Corresponding Author}

David A. Leigh — Department of Chemistry, University of Manchester, Manchester M13 9PL, U.K.; School of Chemistry and Molecular Engineering, East China Normal University, Shanghai 200062, China; orcid.org/0000-0002-1202-4507; Email: david.leigh@manchester.ac.uk

\subsection*{Authors}

Stefan Borsley — Department of Chemistry, University of Manchester, Manchester M13 9PL, U.K.

Benjamin M. W. Roberts — Department of Chemistry, University of Manchester, Manchester M13 9PL, U.K.; orcid.org/0000-0003-2820-8359

Igigo J. Vitorica-Yrezabal — Department of Chemistry, University of Manchester, Manchester M13 9PL, U.K.

Complete contact information is available at: https://pubs.acs.org/10.1021/jacs.2c07633

\subsection*{Author Contributions}

*B.B. and B.M.W.R. contributed equally.

\subsection*{Notes}

The authors declare no competing financial interest.

\section*{ACKNOWLEDGMENTS}

We thank the Engineering and Physical Sciences Research Council (EPSRC; EP/P027067/1) and the European Research Council (ERC Advanced grant 786630) for funding. D.A.L. is a Royal Society Research Professor.

\section*{ABBREVIATIONS}

\begin{itemize}
  \item DCC: dicyclohexylcarbodiimide
  \item DIC: diisopropylcarbodiimide
  \item DMSO: dimethyl sulfoxide
  \item EDC: 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide
  \item HOAt: hydroxyazabenzotriazole
  \item HBOt: hydroxybenzotriazole
  \item MES: 2-(N-morpholino)ethanesulfonic acid
  \item NMR: nuclear magnetic resonance
\end{itemize}

\section*{REFERENCES}

\begin{enumerate}
  \item Astumian, R. D. Kinetic asymmetry allows macromolecular catalysts to drive an information ratchet. \textit{Nat. Commun.} \textbf{2019}, 10, 3837.
  \item Kay, E. R.; Leigh, D. A.; Zerbetto, F. Synthetic molecular motors and mechanical machines. \textit{Angew. Chem., Int. Ed.} \textbf{2007}, 46, 72−191.
  \item Amano, S.; Borsley, S.; Leigh, D. A.; Sun, Z. Chemical engines: driving systems away from equilibrium through catalyst reaction cycles. \textit{Nat. Nanotechnol.} \textbf{2021}, 16, 1057−1067.
  \item Wilson, M. R.; Solà, J.; Carlone, A.; Goldup, S. M.; Lebrasseur, N.; Leigh, D. A. An autonomous chemically fuelled small-molecule motor. \textit{Nature} \textbf{2016}, \textit{534}, 235−240.
  \item Borsley, S.; Kreidt, E.; Leigh, D. A.; Roberts, B. M. W. Autonomous chemically fuelled directional rotation about a single bond. \textit{Nature} \textbf{2022}, 604, 80−85.
  \item Borsley, S.; Leigh, D. A.; Roberts, B. M. W. A doubly kinetically-gated information ratchet autonomously driven by carbodiimide hydration. \textit{J. Am. Chem. Soc.} \textbf{2021}, 143, 4414−4420.
  \item Amano, S.; Fiedlen, S. D. P.; Leigh, D. A. A catalysis-driven artificial molecular pump. \textit{Nature} \textbf{2021}, 594, 529−534.
  \item Erbans-Cakmak, S.; Leigh, D. A.; McTernan, C. T.; Nussbaumer, A. L. Artificial molecular machines. \textit{Chem. Rev.} \textbf{2015}, \textit{115}, 10081−10206.
  \item Aprahamian, I. The future of molecular machines. \textit{ACS Cent. Sci.} \textbf{2020}, 6, 347−358.
  \item Feng, Y.; Ovalle, M.; Seale, J. S. W.; Lee, C. K.; Kim, D. J.; Astumian, J. F.; Stoddart, J. F. Molecular pumps and motors. \textit{J. Am. Chem. Soc.} \textbf{2021}, 143, 5569−5591.
  \item Schliwa, M.; Woeihke, G. Molecular motors. \textit{Nature} \textbf{2003}, 422, 759−765.
  \item Sauvage, J.-P. From chemical topology to molecular machines (Nobel lecture). \textit{Angew. Chem., Int. Ed.} \textbf{2017}, 56, 11080−11093.
  \item Stoddart, J. F. Mechanically interlocked molecules (MICs)—Molecular shuttles, switches, and machines (Nobel lecture). \textit{Angew. Chem., Int. Ed.} \textbf{2017}, 56, 11094−11125.
  \item Feringa, B. L. The art of building small: From molecular switches to motors (Nobel lecture). \textit{Angew. Chem., Int. Ed.} \textbf{2017}, 56, 11060−11078.
  \item Astumian, R. D. How molecular motors work—insights from the molecular machinist’s toolbox: the Nobel prize in Chemistry 2016. \textit{Chem. Sci.} \textbf{2017}, 8, 840−845.
  \item Zhang, L.; Marcos, V.; Leigh, D. A. Molecular machines with bio-inspired mechanisms. \textit{Proc. Natl. Acad. Sci. U.S.A.} \textbf{2018}, 115, 9397−9404.
  \item Lancia, F.; Ryabchun, A.; Katsonis, N. Life-like motion driven by artificial molecular machines. \textit{Nat. Rev. Chem.} \textbf{2019}, 3, 536−551.
  \item Heerd, A. W.; Goldup, S. M. Simplicity in the design, operation, and applications of mechanically interlocked molecular machines. \textit{ACS Cent. Sci.} \textbf{2020}, 6, 117−128.
  \item Kelly, T. R.; Cai, X.; Damkaci, F.; Panicker, S. B.; Tu, B.; Bushell, S. M.; Cornella, I.; Piggott, M. J.; Salves, R.; Cavero, M.; Zhao, Y.; Jasmin, S. Progress toward a rationally designed, chemically powered rotary molecular motor. \textit{J. Am. Chem. Soc.} \textbf{2007}, 129, 376−386.
  \item Berna, J.; Alajarin, M.; Orenes, R. A. Azodicarboxamides as template binding motifs for the building of hydrogen-bonded molecular shuttles. \textit{J. Am. Chem. Soc.} \textbf{2010}, 132, 10741−10747.
  \item Berrocal, J. A.; Biagini, C.; Mandolini, L.; Di Stefano, S. Coupling of the decarboxylation of 2-cyanophenylpropanoic acid to large-amplitude motions: a convenient fuel for an acid-base-operated molecular switch. \textit{Angew. Chem., Int. Ed.} \textbf{2016}, \textit{55}, 6997−7001.
  \item Biagini, C.; Di Stefano, S. Abiotic chemical fuels for the operation of molecular machines. \textit{Angew. Chem., Int. Ed.} \textbf{2020}, \textit{59}, 8344−8354.
  \item Department of Energy, Vehicle Technologies Office and Office of Energy Efficiency and Renewable Energy U.S. Davis, S. C.; Boundy, R. G. \textit{Transportation Energy Data Book}, 39th ed.; Oak Ridge National
\end{enumerate}
(24) Efremov, A.; Wang, Z. Universal optimal working cycles of molecular motors. Phys. Chem. Chem. Phys. 2011, 13, 6223–6233.

(25) Wagoner, J. A.; Dill, K. A. Opposing pressures of speed and efficiency guide the evolution of molecular machines. Mol. Biol. Evol. 2019, 36, 2813–2822.

(26) Wagoner, J. A.; Dill, K. A. Mechanisms for achieving high speed and efficiency in biomolecular machines. Proc. Natl. Acad. Sci. U.S.A. 2019, 116, 5902–5907.

(27) Brown, A. I.; Sivak, D. A. Theory of nonequilibrium free energy transport by molecular machines. Chem. Rev. 2020, 120, 434–459.

(28) Marciniak, A.; Chodnicki, P.; Hossain, K. A.; Slabonska, J.; Czub, J. Determinants of directionality and efficiency of the ATP synthase F0 motor at atomic resolution. J. Phys. Chem. Lett. 2022, 13, 387–392.

(29) Schimert, K. I.; Budaitis, B. G.; Reineimann, D. N.; Lang, M. J.; Verhey, K. J. Intracellular cargo transport by single-headed kinesin motors. Proc. Natl. Acad. Sci. U.S.A. 2019, 116, 6152–6161.

(30) Kull, F. J.; Endow, S. A. Force generation by kinesin and myosin cytoskeletal motor proteins. J. Cell Sci. 2013, 126, 9–19.

(31) Amano, S.; Esposito, M.; Kredet, E.; Leigh, D. A.; Penocchio, E.; Roberts, B. M. W. Insights from an information thermodynamics analysis of a synthetic molecular motor. Nat. Chem. 2022, 14, 530–537.

(32) Seifert, S. Stochastic thermodynamics, fluctuation theorems and molecular machines. Rep. Prog. Phys. 2012, 75, 126001.

(33) Astumian, R. D.; Mukherjee, S.; Wark, A. R. The physics and physical chemistry of molecular machines. ChemPhysChem 2016, 17, 1719–1741.

(34) Ragazzon, G.; Prins, L. J. Energy consumption in chemical fuel-driven self-assembly. Nat. Nanotechnol. 2018, 13, 882–889.

(35) Das, K.; Gabrèli, L.; Prins, L. J. Chemically Fueled Self-Assembly in Biology and Chemistry. Angew. Chem., Int. Ed. 2021, 60, 20120–20143.

(36) Borsley, S.; Leigh, D. A.; Roberts, B. M. W. Chemical fuels for molecular machinery. Nat. Chem. 2022, 14, 728–738.

(37) Kariyawasam, L. S.; Hartley, C. S. Dissipative assembly of aqueous carboxylic acid anhydrides fueled by carbodiimides. J. Am. Chem. Soc. 2017, 139, 11949–11955.

(38) Kariyawasam, L. S.; Hossain, M. M.; Hartley, C. S. The transient covalent bond in abiotic nonequilibrium systems. Angew. Chem., Int. Ed. 2021, 60, 12648–12658.

(39) Tena-Solsona, M.; Rieß, B.; Grötsch, R. K.; Lührer, F. C.; Wanzke, C.; Käsdorf, B.; Bausch, A. R.; Müller-Buschbaum, P.; Lieleg, O.; Boekhoven, J. Non-equilibrium dissipative supramolecular materials with a tunable lifetime. Nat. Commun. 2017, 8, 15895.

(40) Bal, S.; Das, K.; Ahmed, S.; Das, D. Chemically fueled dissipative self-assembly that exploits cooperative catalysis. Angew. Chem., Int. Ed. 2019, 58, 244–247.

(41) Rieß, B.; Grötsch, R. K.; Boekhoven, J. The design of dissipative molecular assemblies driven by chemical reaction cycles. Chem. 2020, 6, 552–578.

(42) Schwarz, P. S.; Tena-Solsona, M.; Dai, K.; Boekhoven, J. Carbodiimide-fueled catalytic reaction cycles to regulate supramolecular processes. Chem. Commun. 2022, 58, 1284–1297.

(43) Seeman, J. I. Effect of conformational change on reactivity in organic chemistry. Evaluations, application, and extensions of Curtin–Hammett/Winstein–Holness kinetics. Chem. Rev. 1983, 83, 83–134.

(44) Penocchio, E.; Rao, R.; Esposito, M. Thermodynamic efficiency in dissipative chemistry. Nat. Commun. 2019, 10, 3865.

(45) Astumian, R. D. Irrelevance of the power stroke for the directionality, stopping force, and optimal efficiency of chemically driven molecular machines. Biophys. J. 2015, 108, 291–303.

(46) Li, Q. G.; Fuks, E.; Moulin, M.; Maaloum, M.; Rawiso, I.; Kulic, J. T.; Foy, N.; Giuseppone, N. Macroscopic contraction of a gel induced by the integrated motion of light-driven molecular motors. Nat. Nanotechnol. 2015, 10, 161–165.