



Molecular motors in nanobiotechnology: Protein and DNA based molecular motors: A review

Yathrib Ajaj^a, Zaid H. Mahmoud^b, Ameer Najy Obeed^c, Moatasem Al-Salih^d,
Batool Ali Ahmed^e, Ekhlash Abdallah Hassan^f, Marwa Sabbar Falih^g, Mahdijeh Nosratabadi^h,
Ehsan Kianfar^{i,j,k,*}

^a German University of Technology in Oman, Oman

^b Chemistry Department, College of Science, University of Diyala, Iraq

^c Department of Pharmacy, Al-Hadi University College, Baghdad 10011, Iraq

^d College of Pharmacy, Al-Ayen University, Thi-Qar, Iraq

^e Department of Medical Engineering, Al-Nisour University College, Baghdad, Iraq

^f Department of Chemistry, College of Science, University of Diyala, Baquba, Diyala, Iraq

^g Department of Chemistry, College of Science, Mustansiriyah University, Baghdad, Iraq

^h Department of Materials Engineering, Faculty of Engineering, Shahid Bahonar University of Kerman, Kerman, Iran

ⁱ Department of Chemical Engineering, Arak Branch, Islamic Azad University, Arak, Iran

^j Young Researchers and Elite Club, Gachsaran Branch, Islamic Azad University, Gachsaran, Iran

^k Istanbul Medeniyet University, Department of Mechanical Engineering, Kadikoy, Istanbul, Turkey

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ABSTRACT

Molecular motor can be defined as the collection of a separate number of molecular features (that is, a supra-molecular structure) which has been developed to carry out a function through the mechanical movements of its components, which occurs when it is appropriately stimulated externally. Therefore, molecular motors include a part with a motor, which is a tool with the power of turning energy to mechanical work. Molecular motors and machines operate by means of nuclear rearrangements, which like their macroscopic peers, and are described by the type of energy input provided to make them function, the way in which their operation can be observed, the potential to renew the operation at will, i.e., setting up of a cyclic process, the time scale needed to complete a cycle of operation, and the performed function. Due to progresses made in several branches of chemistry, it has become possible to design and construct basic prototypes of artificial molecular motors and machines so that we can better understand the mechanisms by which molecular motors of the biological world operate. The expansion of the concept of motors to the molecular level has received great importance not only in the whole research areas but also in the advancement of nanoscience and the development of nanotechnology. Today, molecular motors are one of the common areas of research between biotechnology and nanoscience. These devices are nanoscale motors that have the ability to perform mechanical movements in exchange for a suitable external stimulus. This stimulation can be caused by a chemical reaction, environmental changes, or light exposure. To date, a large number of high-efficiency natural molecular motors have been identified that can be imitated to develop a wide range of synthetic molecular motors. Although the net weight of Nano motors is in the range of molecular units, these nanoscale structures are capable of generating force at very small levels of Pico and fete newton, and with their self-assembly ability can produce large forces (such as those produced in human muscle). These motors obtain their energy through chemical or optical excitation and convert it into mechanical work. Also, the amount of productive force and its efficiency were examined. Molecular motors are known for having a vast variety of types. One of the best possible classifications is based on the origin of the motors. Therefore, molecular motors can be divided into natural and artificial. Natural molecular motors are protein-based, and the fuel they need is supplied by ATP. The synthetic type is either based on DNA molecules or based on chemical (inorganic) compounds. DNA-based synthetic molecular motors derive their energy from

Abbreviations: ATP, Adenosine triphosphate; V-ATPase, Vacuolar-type ATPase; RNA, Ribonucleic Acid; MTOC, microtubule organizing center; MTs, move along microtubules; DNA, Deoxyribonucleic Acid.

* Corresponding author.

E-mail address: ehsankianfar775@gmail.com (E. Kianfar).

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DNA. In contrast, the energy of chemical molecular motors can be supplied by various methods such as optical excitation and electrochemical reaction. In this paper, the general introduction of molecular motors and their operating mechanism, as well as the types of molecular motors and electronic and non-electronic applications of molecular motors will be discussed.

Introduction

Molecular motors are natural (biological) or artificial molecular machines that are the essential agents of movement in living organisms. In general terms, a motor is a device that consumes energy in one form and converts it into motion or mechanical work; for example, many protein-based molecular motors harness the chemical free energy released by the hydrolysis of ATP in order to perform mechanical work [1–5]. In terms of energetic efficiency, this type of motor can be superior to currently available man-made motors. One important difference between molecular motors and macroscopic motors is that molecular motors operate in the thermal bath, an environment in which the fluctuations due to thermal noise are significant [6–8].

Molecular motors, which are an important classification of molecular machines, employ considerable variety of energy resources to engender unidirectional mechanical motion [9–12]. The operational principles of these motors, whether biological or synthetic, are disparate from those of man-made macroscopic motors. Molecular motors have nanoscale dimensions and wholly operate in a solution environment where viscosity is dominant [13–16]. Under these low Reynolds number, over-damped conditions, they cannot count on inertia to keep up motion. In addition, they are incessantly agitated by random Brownian motion, which delivers both challenges and opportunities for mechanisms of converting energy [17–20]. Researchers have long been intrigued by the question of how biological molecular motors can operate so well under these peculiar conditions [4,21–23]. In biological systems, molecular motors made of proteins and nucleic acids are frequent, and ordinarily use the chemical energy of ATP or the electrochemical potential of protons across the cell membrane (the so-called proton motive force) as an energy source [24–27]. Well-studied instances of ATP-driven linear molecular motors include myosin's, kinesins, and dynein's responsible for cellular activities such as muscle contraction and intracellular vesicle transport [28–31]. ATP-driven and/or proton-motive force-driven rotary motors are found in ATP synthase, V-ATPase, and the bacterial flagellar motor. ATP synthase and V-ATPase also act as energy converters, in which ATP chemical energy and proton electrochemical potential are converted via mechanical rotation in a reversible [32–36]. The macromolecular machines underlying the dominant dogma of molecular biologist, such as RNA polymerase and the ribosome, are also linear molecular motors that move in a one-way direction along nucleic acid tracks to read and write genetic information [37–40]. Fig. 1 shows

Molecular Engine [15].

Today, biotechnology and nanoscience are intertwined in many fields of application and cannot be separated from each other. In biotechnology, attempts are made to use biomaterial manufacturing units to make bioelectronics devices [41–46]. In contrast, nanoscience seeks to use nanoscale units of natural and laboratory materials to improve the performance of structures and increase their efficiency [47–51]. With this in mind, human advances in the engineering of complex molecular structures with the aim of better understanding and utilizing biotechnology seem essential. Many known biological units can be used as a valuable source of information for nanoscience [21,22,25,52,53]. For example, molecules of DNA, RNA, lipids, and proteins can be used in nanoscience as high-performance compounds with the ability of molecular self-assembly [27,54,55]. This is why researchers today are looking to design nanoscale hybrid structures that are built with a combination of biological and synthesized units and optimized for specific functions [56–59]. Molecular motors are one of the most compelling examples of the relationship between biotechnology and nanoscience [60–62]. “Protein motors” are a kind of biomolecular motors which have been substantially optimized [17,63–66]. These motors are qualified for conversion of chemical energy into highly efficient work done mechanically. In addition to being able to perform mechanical work in an individual manner, they are able to bring about large forces (paramount to the forces produced in human muscle) through their self-assembly capabilities [67–70]. Identifying these motors in nature would be assumed as an advanced stage in the synthesis of synthetic molecular motors [71–75]. So far, many artificial motors have been synthesized by scientists, each of which is comparable with its natural counterparts. In this article, the general definition of molecular motors, their principles, and operating mechanisms will be discussed [76–80]. Attempts are made to discuss the energy source, how energy is turned to work, the amount of power produced, and the amount of efficiency of these motors [47–49]. Molecular motors are devices that can perform mechanical work in molecular dimensions by the interactions of their constituent molecules [81–85]. Molecular dimensions refer to the nanometer scale [86–88]. Hence, molecular motors can be considered as belonging to the field of nanoscience and nanotechnology [89–92]. Molecular motors get their energy from a specific source and use it to change their spatial structure. This restructuring leads to mechanical work [93–95]. The mechanical performance of molecular depends, on the one hand, on the interactions between their constituent molecules and, on the other hand, on the interactions between molecular motors [96–99]. These interactions include ionic and van der Waals forces that affect the spatial shape (geometry) of molecules [100–103]. The energy required for structural changes in molecular motors comes from a variety of sources [104–108]. Depending on the type of molecular motors, this energy can be chemical, light, or environmental changes such as temperature or pH [109–114]. Molecular motors are the active dependable tools of the cells [115,116]. The overall active transport in the cell is pushed by the three classes of molecular motors namely kinesin, dynein, and myosin [117–119]. They have been developed to enable movement on their polymer filaments, either on cellular or supra-cellular levels, and are able to identify the route of movement. Molecular motors employing the cytoskeleton for movement are divided into two categories on the basis their substrates:

(1) actin-based motors such as myosin carry their cargo along short actin filaments [120–122], which are found all over the cell but are highly concentrated on the cell membrane; and (2) microtubule-based motors such as dynein [123] and kinesin [124] move along

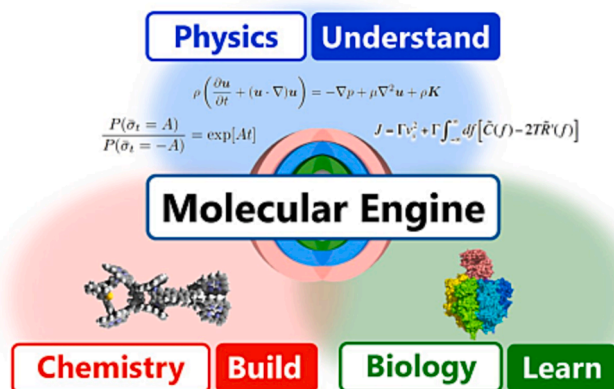


Fig. 1. Molecular Engine [15].

microtubules (MTs), which are long polarized filaments with their plus-ends located in the cell periphery and their minus-ends placed at the microtubule organizing center (MTOC) near the nucleus [125–128]. A great number of these molecular motors are dimers with two ‘heads’ connected together at a ‘stalk’ region and a ‘tail’ domain opposite the heads [129–133]. The core of the molecular motors includes the motor domain that supplies the motion along the filaments (MTs or actins) whereas the tail of the molecular motors contains the subunits held accountable for cargo bonding and organization [134–138].

For millions of years, there have been many different types of molecular motors in nature that have been optimized over the years [139]. As our knowledge of natural molecular motors grows, so does our ability to imitate them to synthesize artificial species [140]. Therefore, molecular motors can be divided into two main groups: natural (protein) motors and artificial motors. Natural motors are often based on ATP [141]. Molecular motors are an important and integral part of living organisms and their job is to create motion. Without molecular motors, no movement occurs in the body of living things, and the result is stagnation and the death of life [142]. Molecular motors receive the energy they need from an external source, convert it into mechanical work, and move the living world [143]. These motors can provide the energy needed by the cell, generate cell translocations, promote cell division, and when biologically assembled together, they can produce biological organs [144]. These capabilities have made molecular motors one of the essential components of living organisms [145]. For example, muscle contraction (movement) that occurs in a complex process is caused by myosin molecular motors. In fact, myosin molecular motors are located between the actin filaments in the muscle and cause them to slide over each other (Fig. 2). Another example of the application of a molecular motor is “filtration”. Molecular motors can confront a solution or suspension stream in the body of living organisms, separate the chemical from the stream, and filter the solution or suspension [146]. Molecular motors, on the other hand, can move and connect tiny components that are self-assembling but unable to move on their own [147].

Organelle transport, ciliary movement and contraction are paradigmatic tasks of cytoskeletal motors, but the motor has more than it would meet the microscopes’ eye. Some motors are involved in the transfer of messenger RNA or macromolecular complexes [148–151]. Others do not

have the ability to move and are still vital for specific cellular activities. Some motor omissions or mutations are likely to be deadly for multicellular organisms, implying that these motors are critical for pivotal phases in development [152]. In other conditions, the losing of specific motors results in rippling diseases. Finally, motors might have a role in cellular homeostasis and cell architecture in manners that expand beyond functions in transfer [153–155]. These are intriguing research areas unearthed in the last few years. New on the agenda of motor functions is participation in mRNA transfer [156–158]. Restricting mRNA translation impedes the subcellular dispensation of the protein product but necessitates the transfer of mRNA to its target point. Depending on the system studied, mRNA transport is achieved by myosin, kinesin, or dynein motors [159–161]. In yeast, for instance, certain mRNAs are transported in a complex with myosin V, while in neurons or insect oocytes, microtubule motors are required [162–164]. In all instances, RNA-binding and adaptor proteins combine the RNA into a ribonucleoprotein transport package, although the exact molecular interactions inside this complex are yet to be determined [165,166]. A framework for the striking centrality of motor-dependent mRNA localization in the *Drosophila* oocyte is that the convergence of Oskar mRNA and associated proteins at the posterior pole are supported by customary kinesin, while Bicoid mRNA-containing complexes are moved by dynein to the anterior pole [167–169]. Their accurate deployment sets up the anterior-posterior axis. There are implications that the setup of dorsoventrally also requires motors. The determination of the left–right axis in mammals is also dependent on the activity of molecular motors, although the nature of the implication is different [170–172]. Left-right patterning was offered to require the transfer of a ‘morphogen’ to the left side of the vertebrate gastrula by the cilia of the embryonic node, an organizing construction in the developing embryo. In supporting this notion, omissions in a gene encode a dynein isoform known as left–right dynein which is a member of the Krp85/95 kinesin family, both of which are required for ciliary development, inactivate nodal cilia, and result in random positioning of internal organs [173–176]. The results from these studies were met with skepticism since the mechanism was said to be too generic for such a role-playing step in development. However, a factitious flow around the nodal cilia, spawned with the use of an innovative micromechanical device, was shown to influence the status of internal organs [177–180]. This is just one intriguing example that a motor flaw can lead to a pathological condition, situs inversus. Work over the past few years has shown motors in a growing number of human diseases (Table 1).

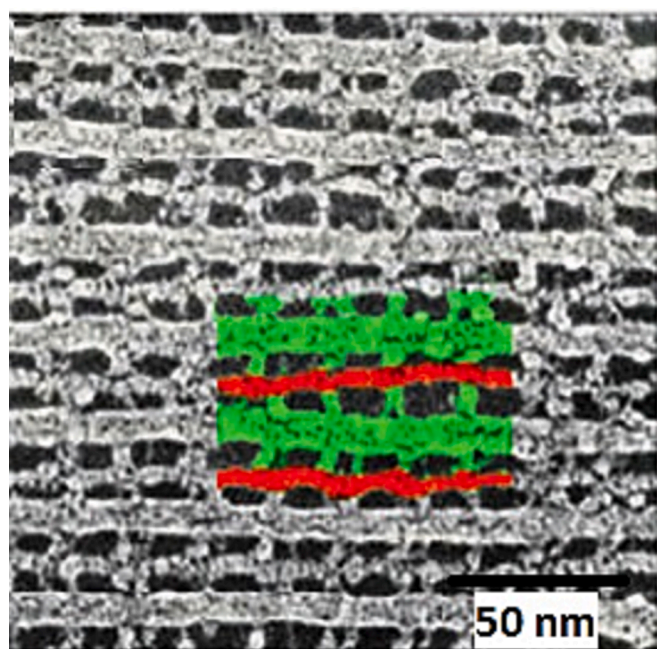


Fig. 2. Myosin motors (green) are located between thick actin filament (green) and thin actin filament (red) [9]. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Table 1
Examples of molecular motors.

Disease or defect	Motor involved	Ref
Myosin myopathies	Myosin I	[66]
Griscelli syndrome (pigmentation disorder)	Myosin V	[67]
Hearing loss	Myosin IIIa (ninaC), myosin VI, myosin VIIa, myosin XVa	[68]
Retinitis pigmentosa (photoreceptor degeneration)	Cytoplasmic dynein, Kinesin Krp85/95	[69]
Primary ciliary dyskinesia	Cytoplasmic dynein, Kinesin Krp85/95	[70]
Kartagener syndrome (situs inversus)	Axonemal dynein	[71]
Polycystic kidney disease	Axonemal dynein, kinesin Krp85/95	[72]
Lissencephaly	Dyneins and kinesins	[73]
Charcot-Marie-Tooth disease type 2A	LI51 (dynein/dynactin interactor)	[74]
Virus transport	KIF1B (Unc104 kinesin family)	[75]
Anthrax susceptibility	Conventional kinesin, cytoplasmic dynein, myosin	[76,77]
Neurodegenerative diseases	KIF1C (Unc104 kinesin family)	[78]
Neurodegenerative diseases	Kinesin	[79]
	Cytoplasmic dynein	[80]

Definition of molecular motors

Life is based on movement. Most of the motions that occur in the peripheral world are caused by protein molecular motors. These motors can move various components in the body of living organisms, provide the energy needed by a cell, promote cell division, and when placed on a large scale, are self-sustaining [78,187–180]. They can move biological organs such as muscles. For example, Fig. 3 shows a picture of a Kinesin molecular motor transporting food in a certain direction. The presence of these motors in the body of living organisms is essential so that any defects in them cause very chronic diseases [18,175–177]. Fig. 4 also shows a diagram of myosin molecular motors that begins to move in a certain direction due to specific chemical reactions. In general, a “molecular machine” consists of one or a set of molecular components at the nanoscale and is homogenous with molecular units designed to perform mechanical movements (output) due to a suitable external stimulus (input) [73]. These types of motors are very similar in performance to conventional (large-scale) motors. The input of molecular motors, like conventional motors, can be chemical energy, light, or other stimuli. As our knowledge of molecular motors in nature increases, so does the ability to use these motors in other environments or to synthesize molecular motors by imitating their natural variants [19].

Examples of biologically important molecular motors

Myosins

Myosins are responsible for muscle contraction, intracellular cargo transport, and producing cellular tension [82].

Kinesin

Kinesin moves cargo inside cells away from the nucleus along microtubules, in anterograde transport [83].

Dynein

Dynein produces the axonemal beating of cilia and flagella and also transports cargo along microtubules towards the cell nucleus, in retrograde transport [84].

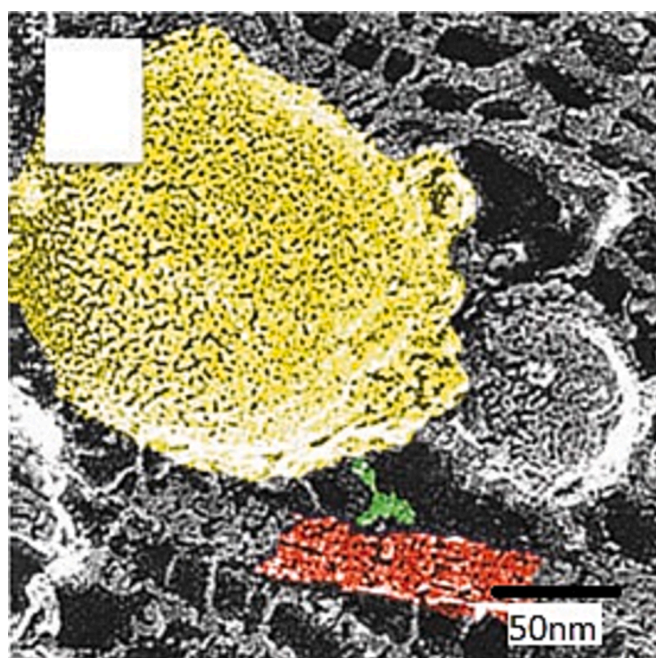


Fig. 3. Microscopic image of a Kinesin motor (green) transporting food (yellow) in a certain direction (red) [1]. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Actin

Actin polymerization generates forces and can be used for propulsion. ATP is used [85].

Microtubule

Microtubule polymerization using Guanosine-5'-triphosphate (GTP) is a purine nucleoside triphosphate. It is one of the building blocks needed for the synthesis of RNA during the transcription process. Its structure is similar to that of the guanosine nucleoside, the only difference being that nucleotides like GTP have phosphates on their ribose sugar. GTP has the guanine nucleobase attached to the 1' carbon of the ribose and it has the triphosphate moiety attached to ribose's 5' carbon [86].

Dynamin

Dynamin is responsible for the separation of clathrinid buds from the plasma membrane. GTP is used [87].

FoF1-ATP synthase

FoF1-ATP synthase family of proteins convert the chemical energy in ATP to the electrochemical potential energy of a proton gradient across a membrane or the other way around. The catalysis of the chemical reaction and the movement of protons are coupled to each other via the mechanical rotation of parts of the complex. This is involved in ATP synthesis in the mitochondria and chloroplasts as well as in pumping of protons across the vacuolar membrane [88].

RNA polymerase

RNA polymerase transcribes RNA from a DNA template [89].

DNA polymerase

DNA polymerase turns single-stranded DNA into double-stranded DNA [90].

Helicases

Helicases separate double strands of nucleic acids prior to transcription or replication. ATP is used [91].

Topoisomerases

Topoisomerases reduce supercoiling of DNA in the cell. ATP is used [92].

RSC and SWI/SNF

RSC and SWI/SNF complexes remodel chromatin in eukaryotic cells. ATP is used [93].

SMC proteins

SMC proteins responsible for chromosome condensation in eukaryotic cells [93].

Power generation and energy conversion in molecular motors

Today, with advances in biotechnology and nanoscience, there is a great deal of information about how molecular motors in nature and synthetic molecular motors work. These motors need initial energy for their linear and rotational motion [74,178–180]. This energy is converted into mechanical energy and used to do the work. In general, molecular motors get their energy from a single source and use it to change their spatial structure [20,181–184]. A change in conformation causes the molecule to move toward one field or one part of the motors to move toward another. The energy required, depending on the type of molecular motors, can be chemical, light, or changes in environmental conditions such as ambient temperature and pH [75]. In fact, protein motors have a specific spatial structure with certain rules that change in temperature or acidity of the environment or external stimuli, changing these rules and spatial arrangement in a part of the molecular motors.

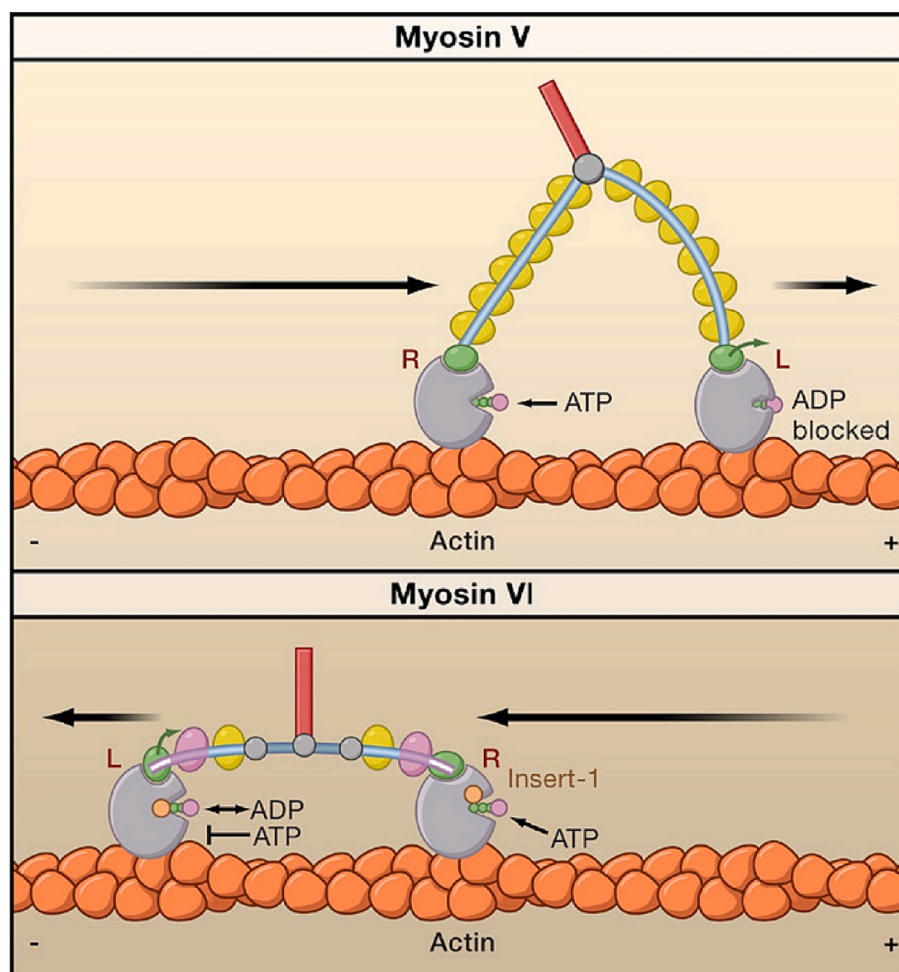
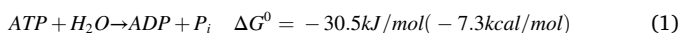


Fig. 4. Schematic of the movement of the myosin molecular motors. Depending on the type of chemical reaction performed, the motors start moving in a certain direction [2].

The result of these interactions is a “mechanical movement”, one of the molecular motors in nature that has linear motion and uses chemical energy as an energy source is the myosin motor. This engine, like other natural protein motors, is based on ATP (Adenosine triphosphate) [21]. ATP is an enzyme that plays a key role in the energy transfer between cells [76]. Myosin molecular motors derive their chemical energy from the hydrolysis of ATP to ADP (Adenosine diphosphate) and phosphate. The following equation shows the corresponding reaction:



of course, the products of this reaction have the ability to combine and produce ATP again. Therefore, these motors do not have any waste products (waste) and this factor increases their efficiency. On the other hand, unlike heat motors, natural molecular motors do not release heat during their operation and the reactions performed are of the heat-absorbing type. Fig. 5 shows the operation cycle of the myosin motors [77,185–187]. As can be seen, this cycle involves complex steps in the reaction of Equation (1), changes in the spatial structure of molecules, and their interaction with the ground (here, actin filaments). That is, the chemical energy generated during the motor’s interactions with the ground is converted into the mechanical energy required to move the motor, which is discussed in detail in another article entitled “Molecular motors: Types.” And will be reviewed [4].

Unlike natural molecular motors, “synthetic molecular motors” require fuel to use chemical energy to move. Fuel supply and consumption by the motors lead to the production and accumulation of

waste materials and damage to the body or any other environment. In other words, these motors have not yet reached a level of efficiency where they can regenerate and consume fuel from their reaction products [78,188–191]. Therefore, scientists tend to develop motors that use other energy sources (other than chemical energy) to perform mechanical work. Optical energy is one of these sources. Most artificial motors today use this type of energy [22–24]. For example, rotenone molecular motors perform mechanical work when stimulated by light waves.

Force scale

Marden & Allen [4] showed that the relationship between the maximum force output of a linear motor and its mass is a scaling one (Fig. 6), which holds over more than 25 orders of magnitude in mass and is written as:

$$f = (m^* 26600 N / 2/3 \text{ Kg} - 1)^{2/3} = (M^* 1.4 f N / 3 / 2 Da - 1)^{2/3} \quad (2)$$

Although a significant overlap seems to exist at the macroscale between biological motors (e.g., muscles) and synthetic motors (e.g., winches), research data hints at the molecular scale stem entirely from linear motor proteins. Recent progresses by the Stoddart group have made it possible to remotely compare them with synthetic molecular motors. Rotaxanes, photo-activated synthetic molecular motors that have been successfully assembled into artificial muscles [22], in theory can bring forces of up to approximately count 30 pN [23]. Practically speaking, an initial average contractile force of 10 pN per motor lowered to less than

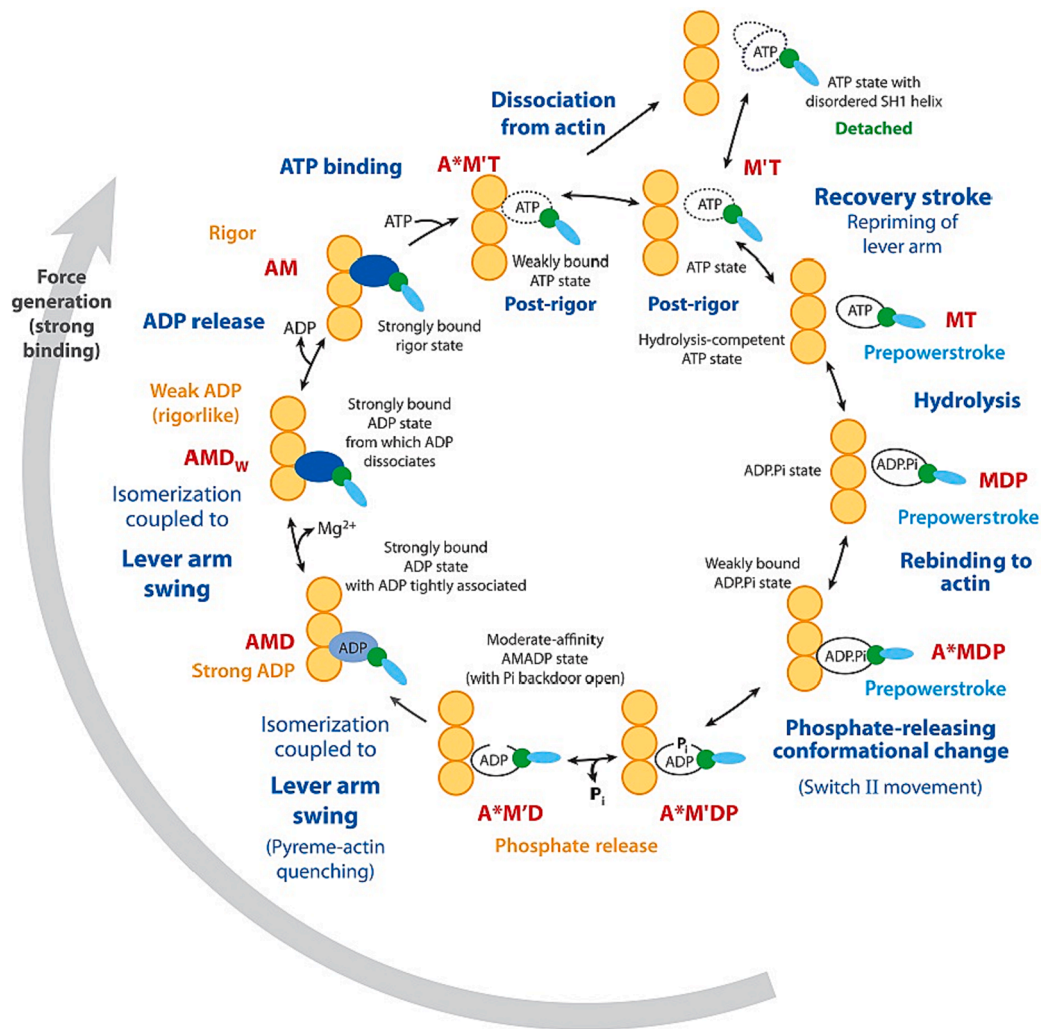


Fig. 5. Scheme of the movement cycle of the myosin motor on the actin filament [3].

half after only 25 activation cycles, which indicates that an enormous number of motors lose their activity in each cycle [22]. Fig. 5 suggests that a functional motor with a mass of 4 kDa should be designed for a maximum force of 0.3 pN. Neither the existence nor the exponent of this scaling relationship is entirely surprising. The exponent of 2/3 arises from the fact that the cross-section of a structure that is scaled linearly in mass will scale with an exponent of 2/3, which enables the supplied forces to raise with an exponent of 2/3 without any change in stress. Considering the relatively narrow distribution of materials around a line in an Ashby diagram plotting strength versus density [24], the disparate materials employed in constructing molecular motors and ship engines produce a low effect. However, the coefficient relating to force and mass is beyond expectation. Even a mechanically insignificant polymer such as poly (ethylene terephthalate) has a tensile strength of more than 50 MPa, which means that a cube of this material with a density of 1.4 g/cm³ is able support a tensile force of

$$f = \sigma \left(\frac{m}{\rho} \right)^{\frac{2}{3}} = (m \cdot 2.5 \cdot 10^8 N^{\frac{2}{3}} K g^{-1})^{\frac{2}{3}} = (M \cdot 13000 fN^{\frac{2}{3}} D a^{-1})^{\frac{2}{3}}$$

In other words, the stress averaged over the whole cross-section of the motors is 500-fold smaller than the strength of typical engineering material. This output gap probably originated from a safety factor to account for fatigue as well as from the fact that only a small fraction of the motor cross-sections are available to support the generated force at certain internal locations, such as the connecting rod in a piston engine

[79,192–194]. Designs for synthetic molecular motors will likely have to consider this engineering rule of thumb that connects force and mass in order to create reliable nanoscale devices. The identification of force-limiting weak elements in molecular motor designs will be critical for substantial improvements [80,195–197]. A revolutionary design would be likely to significantly reduce the output gap without a compromise in a life span [4].

Efficiency of molecular motors

One of the most important parameters for evaluating the performance of molecular motors is their efficiency in converting chemical or electrical energy into mechanical energy. Most existing heat is less than 60 % efficient. In most cases, as the size of the motors becomes smaller, their efficiency decreases due to increased energy waste [49,198–204]. However, the efficiency of molecular motors, despite their very small geometric dimensions, is relatively high (albeit in the natural type and not artificial) [50,205–211]. The efficiency of such motors can reach 100 %. For example, the efficiency of the F1-ATPase motors is 100 %. From this point of view, molecular motors look very efficient and attractive. This efficiency increases when the energy efficiency of every single motor is at least equal to the average efficiency of all the motors housed in a self-contained assembly [51,212–217]. The highest efficiency of a molecular motor is achieved when its chemical and mechanical cycles are well designed. What matters about these motors is their efficiency at maximum power, not maximum efficiency at lower

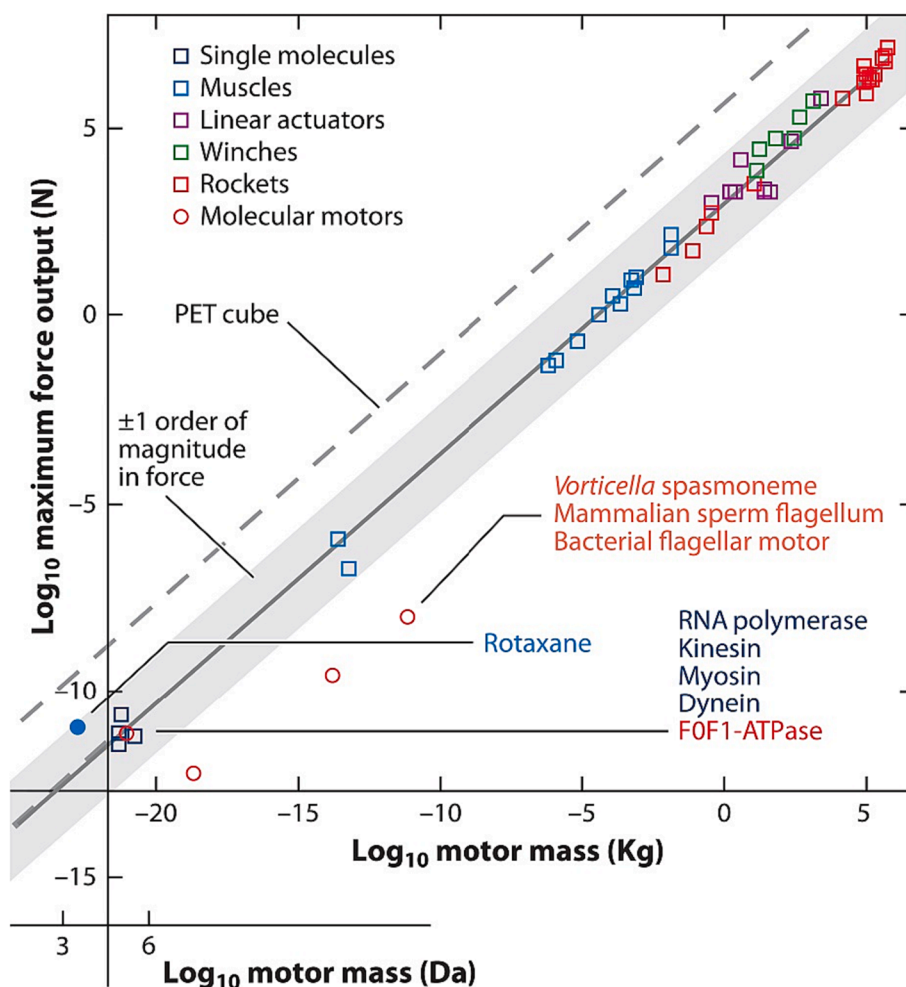


Fig. 6. Maximum production force by various types of artificial and natural motors on a molecular scale and larger in terms of their weight. The data for most motors are on a line with or near a slope of 23 (Marden-Allen relationship). The dotted line is related to the tolerable force of PET polymer in different masses [4].

power [21,218–220]. Studies show that the “power stroke” mechanism, in which the input energy is suddenly converted into mechanical work, increases the efficiency of the maximum productive force. Fuel consumption is shown for different transmission modes of the molecular motors. Transitional states are states in which the motor converts the received energy into mechanical work [22,221–224]. The lower the transfer of the motor modes to converting energy into mechanical work, the higher the efficiency at maximum output (Fig. 7). In the mechanism of “sudden release of energy” there is no transition mode and the input energy is immediately converted into mechanical work. Therefore, the molecular motor will have the highest efficiency in the maximum productive force [52]. Although natural molecular motors largely follow the “sudden energy release mechanism”, most synthesized molecular motors are not subject to this mechanism [53]. Deviation from this mechanism reduces the efficiency of molecular motors [33,34]. DNA-based molecular motors, for example, operate on the basis of “diffusive motion.” This mechanism is exactly the opposite of the mechanism of the sudden release of energy. In the intrusion mechanism, there are different transfer modes that the motors convert the input energy into mechanical work by going through these transmissions [25]. These existing transition modes reduce the efficiency of the molecular motor. It should be noted that if the efficiency of a molecular motor cannot be kept equal to the efficiency of the whole system (including an array of motors), the efficiency of individual molecular motors will no longer be important. In this case, the main purpose of the designs will be to optimize the final efficiency of the system. For example, consider the regular arrays of molecular motors that make up muscle tissue

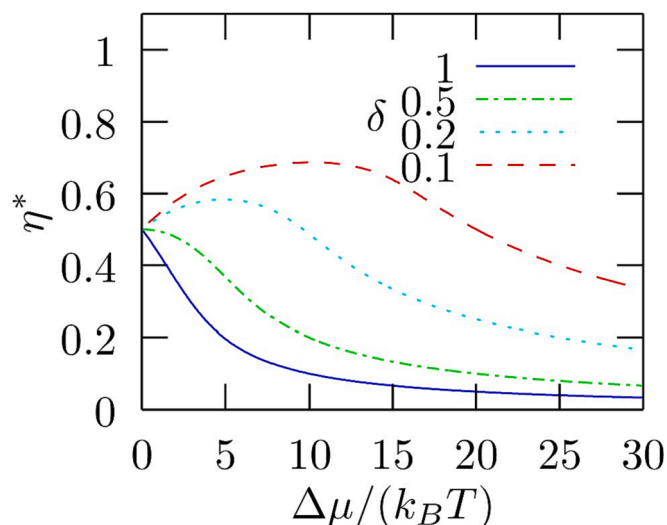


Fig. 7. Diagram of the maximum efficiency of a molecular motor at maximum output force (η^*) as a function of the difference in chemical potential of the fuel at different transition states (δ) during the reaction steps. $\eta = 0$ is the state in which the mechanism of sudden energy release (power stroke) occurs. In fact, the reaction energy is suddenly released in one step. In this case, the return is equal to 1 [5].

[27,225–228]. The muscle usually consists of 1020 molecular motors with an average efficiency of 20 to 40 %. This average efficiency is approximately equal to the efficiency of the individual motors that make it up [35].

ATP-based protein molecular motors

These molecular motors, as their name implies, are made of protein and derive their energy from ATP molecules. ATP is an enzyme with three phosphate molecules. These molecules play an essential role in energy production. The most important natural protein motors are Ethiopia synthase (Fo-F1-ATPase), kinesin, and myosin. All of these motors need ATP, directly or indirectly, to receive energy and perform mechanical work [54].

Fo-F1-ATPase motors

Nature employs molecular machines to carry out complicated functions in almost all substantial biological processes and to strengthen macroscopic motion in organisms. Biomolecular motor proteins undertake enormous tasks including cell division, intracellular transfer, and mechanical actuation in biological cells [55,229–231]. The rotary motor FoF1-ATPase is one of the most conclusively explored biomolecular machines resulting from its essential physiological function. Research on the function of FoF1-ATPase can help to more extensively find more about the deep biological processes and may also lead to the flourishing of biological molecular motor-based devices or even encourage the inventive design and construction of artificial molecular machines. Biomolecular motors could be separated into two general categories: linear molecular motors (for example, kinesin, dynein, and myosin) and rotary molecular motors (ATPase and bacterial flagella). The rotary molecular motor ATPase is the most extensively studied natural molecular machine because of its presence across the whole world and wide availability from natural resources [56,232–235].

ATP is a common unit of energy in biological molecular systems. This molecule consists of 3 phosphate ions that are attached to the enzymatic base of the molecule (Fig. 8). By hydrolyzing ATP and breaking the bond between the “phosphate ions” and the “enzymatic base of the ATP molecule” (i.e. ADP), the energy of this molecule is released and can advance biological processes. This reaction is summarized in Equation (3):



As can be seen, breaking the ATP bond (i.e. consuming it) produces the molecule ADP (adenosine diphosphate) and the phosphate ion (Pi). Combining these two products can regenerate the ATP molecule. Thus, Equation (3) is a reversible reaction. Unlike the process of hydrolysis of ATP, which produces energy (i.e., Equation (1) is spontaneous), the recombination of reaction products and the reproduction of ATP require energy. This reaction is the basis of the performance of the Fo-F1-ATPase molecular motor. In fact, the Fo-F1-ATPase molecular motor is a

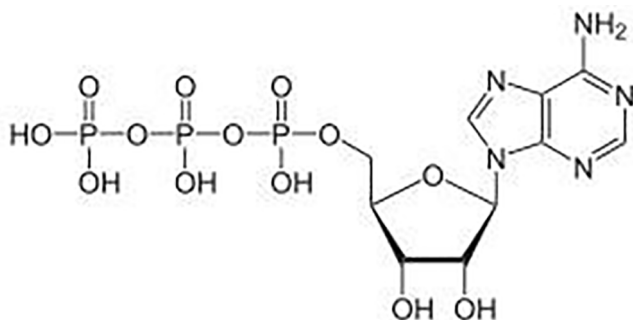


Fig. 8. Molecular structure of ATP molecule [10].

combination of two F0-ATPase (hydrophobic transmembrane) and the F1-ATPase globular motor, which have different structures and functions. The Fo engine has different spatial structures in different organs, and how it works depends on the energy source within that organ [57]. For example, in the mitochondria of cells that make up animal tissue (which is an organ inside the cells that converts food to ATP), the Fo motor is located in the inner membrane of the mitochondria and causes the ion-motive to move. In fact, the difference in ion concentration on both sides of the motor creates the driving force of these was only “ion propulsion from hydrogen ions”, today studies have shown that sodium ions also cause them to move. This ionic driving force is defined as working on an ion unit that an ion can produce by moving through its membrane [10,37].

F1 motor's structure

This motor, which derives its energy from ATP hydrolysis, consists of a central protein column called the γ -subunit. This column is surrounded by 6 other subunits called α and β . The α and β subunits are placed symmetrically and symmetrically next to each other. Also, the subunit δ is connected to the circumference of the α - β cylinders and the subunit ϵ is located below or next to the γ column. The α and β subunits contain specific sites for enzyme binding. These sites are a good place for ATP / ADP molecules to bind. Enzyme binding sites in α subunits allow only enzymes to physically bind, while binding sites in β subunits act as catalysts for reaction (conversion of ATP to ADP or vice versa). Fig. 9 shows a diagram of the structure of the F1 motor, which is part of a bacterium [11].

F1 motor performance

According to the proposed mechanism for this motor, each of the β

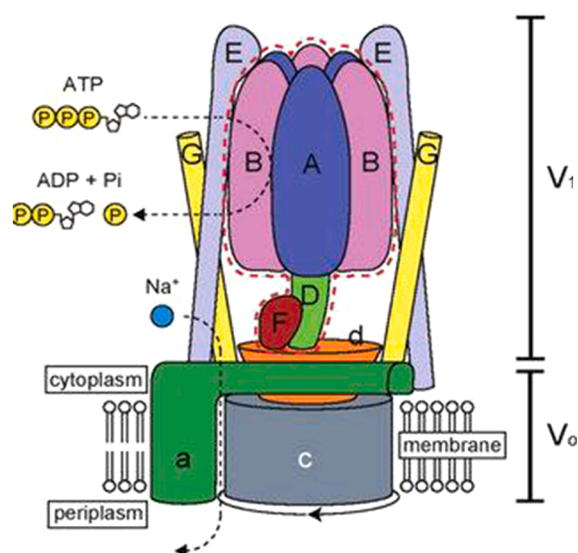


Fig. 9. Schematic of the F1 and F0 motor structures that form part of the structure of a bacterium. Here, the F0 motor moves in the direction shown due to the difference in the concentration of sodium ions inside and outside the cell, leading to the production of ATP in the F1 motor [11]. The V1 domain of V-ATPase is composed of a hexameric arrangement of alternating A and B subunits responsible for ATP binding and hydrolysis; it also contains the DF subunits (shown by a dotted red line), the focus of this study. The V o domain of V-ATPase comprises a subunit and an attached membrane c ring. The V 1 and V o domains are connected by a central stalk, which is composed of D, F, and d subunits, and 2 peripheral stalks assembled from the E and G subunits of V 1. ATP hydrolysis induces the rotation of the central axis (DFd complex) together with the c ring, which causes Na + to be pumped through the channel at the interface between the c ring and a subunit [81]. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

subunits can be in one of three modes: O (open), L (loose), and T (closed). When the β subunit is in the O state, the motor is not catalytically active and will have very little tendency to bind to molecules (ADP and P_i). In the L state, the β subunit forms a weak bond with the molecules, although it is still catalytically inactive. In the T state, the ADP molecule and the P_i ion bind together to form the ATP molecule. Finally, a change in the spatial shape of the motor converts the T mode to O, allowing the ATP molecule produced to separate from the motor [60]. Fig. 9 shows the different stages of this process. The F₀ motor causes the rotation of the γ -column. The rotation of the nanometer column γ , which connects the F₁ motor to the F₀ motor, causes a spatial deformation in the F₁ motor, resulting in a change in the state of the subunits. As shown in Fig. 10, the ion flux (and in some cases, the ion flux from the hydrogen ion) causes the F₀ motor to rotate along the membrane [12,38,39].

Myosin and kinesin motors

In addition to the F₀-F₁-ATPase protein motors that rotate, there are other protein motors with linear motion in nature. These motors, which are responsible for transporting various components such as food, muscle movement, and cell division, require the ATP molecule to move. These small molecular machines are divided into three main families, the “kinesins”, the myosin” and the “dynein’s”.

Myosin motors

These motors are a broad family of protein motors capable of transporting various components such as food along the actin filament. Actin filaments are tiny protein tubes through which myosin molecular motors move. These motors get the energy they need to move by hydrolyzing or consuming ATP molecules [61,62]. A diagram of the myosin motor is shown in Fig. 11. These motors have two similar spherical heads. Each of these two, which operate independently of each other, is connected to a polymer string by a lever arm (level arm) made of polymer. The two polymer strands are wound in a spiral, holding the two ends together. The motor’s tail is usually attached to the transmission. It should be noted that while moving, only one end of the motor operates and the other end is free and without connection to the actin filament [13,40].

Fig. 12(a) shows the myosin motor with an active head (green). The motor moves on actin filament (blue). The lever arm in this figure is red, which connects the motor head to the green helix. The reason for naming it “arm” is that this part of the motor has the ability to deform spatially; Although not seen as a real arm. According to the proposed mechanism, the motor headfirst reacts with ATP to produce ADP and P_i . At this time, because the motorhead is connected to ADP, it does not want to connect to the thin actin filament. When the active motor accidentally collides with the actin filament, the P_i ion left over from the reaction is released and a bond is established (Fig. 12(b)). The release of phosphate ions changes the spatial deformation of the lever arm and rotates it counterclockwise. This rotation moves the molecular motor by 10 nm along the actin filament (Fig. 12(c)). The ADP catalytic center then releases itself and is replaced by a new ATP, separating the motor from the thin actin filament (Fig. 12(d)). The ATP is then converted to ADP and P_i , and the process is repeated. If the function of the myosin motor is to move the muscles, the end of the polymer chain is attached to a thin actin filament called the myosin filament, instead of being attached to the

transported component. This filament is parallel to the actinic fixed filament. In this case, the movement of the motor causes the myosin filament to slip relative to the fixed actin filament. The result of this slip is muscle movement [14,41,237–240].

Kinesin motors

These motors move different components by moving linearly along the microtubule. Resin tubes are tubes 25 nm in diameter made of a special protein called tubulin. Resolutions are polar; That is, they have a negative head and a positive head. Kinesins move from negative to positive. Like myosin’s, these motors have two heads, except that both heads are involved in the movement. An overview of the Kinesin motor is shown in Fig. 13. Each of the two ends of the Kinesin motor is connected to the engine shaft by a neck connection [17,63]. The transported particles can also be attached to the tail of the motor. In the free state, both ends of the motor are attached to separate ADP molecules and move randomly [15,42].

When one end of the motor hits the resonator, it attaches to it and releases its ADP molecule. A new ATP molecule then replaces the ADP molecule (Fig. 14a). This reaction causes the neck joint to rotate. The neck connection causes the other end of the motor to rotate and come forward by bypassing the motor attached to the resonator (Fig. 14b) and attaching it to the front junction along the resonator [242–244]. The first head of the engine, which is connected to the ATP, then hydrolyzes it, separates the resulting P_i , and holds the generated ADP together. The ADP front motor is then replaced with a new ATP (Fig. 14c), which rotates the neck joint to bring the rear head forward (Fig. 14d). By repeating this cycle, the Kinesin motor rotates on both heads by rotating its head. A Kinesin motor is capable of moving 100 steps without detaching from the microtubule. Also, the speed of such motors can reach 1000 nm per second. Production capacity can also be around 5 to 6 picoton [14].

DNA-based molecular motors

The main function of DNA in nature is to store and transmit information. But proteins are responsible for mechanical work. Maybe that’s why DNA has a simpler structure than proteins. DNA molecules are made up of only four types of nucleotides, marked A, T, G, and C. Nucleotides are organic molecules that are known in nature as the building blocks of DNA. DNA is called ss-DNA and two strands of DNA are wrapped around each other and their nucleotides are complementary to each other, called “ds-DNA”. In the structure of this type of DNA, nucleotides A are attached to T and C to G. Since ds-DNA is more robust than other types, it can be a good choice for molecular motors. In contrast, ss-DNA, which has more flexibility, can be used in the flexible parts of molecular motors. The DNA molecule has two important and practical capabilities: (1) molecular recognition, and (2) self-assembly. The same capabilities can be used to build molecular motors. Nucleotides A and T, as well as C and G, tend to bind to two different ss-DNAs. A stable and effective ds-DNA is formed only when the sequence of nucleotides on each strand of ss-DNA is complementary; That is, the nucleotides in the two branches facing each other are complementary (A versus T, and C versus G). Thus, when the complement of the ss-DNA molecule is in a solution, they eventually find each other, bind to each other, and form the stable ds-DNA molecule. From a mechanical point of

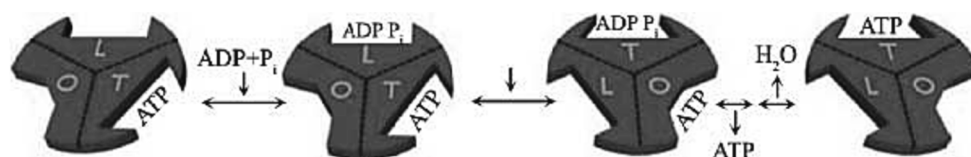


Fig. 10. Mechanism of operation of F₁ motor [12].

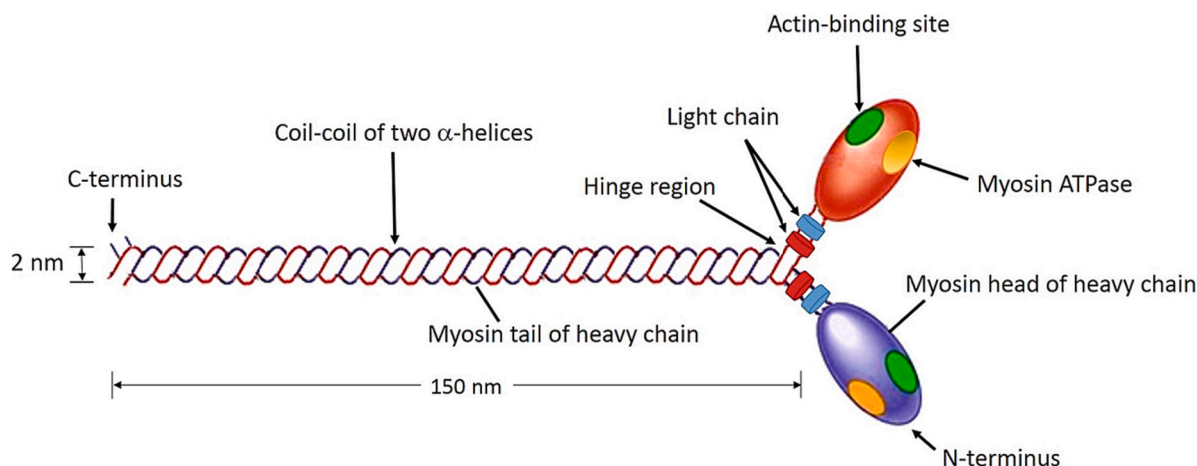


Fig. 11. Scheme of myosin molecular motor and its structure [13].

view, if the energy released by the binding (or zipping) of two complementary DNA molecules is used to lift a hypothetical load, it can produce a force of up to 15 piconewtons [42,43]. This amount of force is equal to the forces produced by protein molecular motors. DNA Tweezers is a DNA-based synthesized motor whose fuel is DNA molecules. This motor consists of three strands of ss-DNA with the letters A, B, and C. Strings B and C are zipped locally on the central string A (Fig. 15-a). The fuel for this motor is a single strand of ss-DNA called F. The design of this single thread is such that it can be zipped (paired) with unzipped parts B and C. Therefore, when the fuel enters the environment in which the motor is located (Fig. 15-b), it is paired with unzipped parts B and C, and the motor takes on a closed shape (Fig. 15-c). At this time, if fuel supplement F enters the environment, it separates the fuel from the motor and the motor assumes an open state (initial state). These switchable motors can be used to identify a variety of fuels; Because a fuel (ss-DNA) is only useful for one type of motor. In other words, a given fuel can only be paired with a specific motor and can be operated [17].

Natural molecular motors and machines

In the last few years, there has been lots of progress in elucidation of the moving mechanisms of motor biomolecules, because of the fact that besides the founded physiological and biochemical methods, novel in vitro techniques have been honed which mix optical and mechanical methods to bring under observation the behavior of a single protein. The most significant and well-known natural molecular motors are ATP synthase, myosin, and kinesin [21,22,185,186]. ATP synthase is the widespread enzyme that manufactures adenosine triphosphate (ATP) and is a rotary motor supplied with a proton gradient [25,27,52–54]. The enzymes of the myosin and the kinesin families are linear motors that proceed along polymer substrates (actin filaments for myosin and microtubules for kinesin), metamorphose the energy of ATP hydrolysis into mechanical work [55,56]. Motion originates from a mechanic chemical cycle, during which the motor protein tethers to successive sites along the substrate in such a way as to move forward on average. Several other biological processes are based on motions, including protein folding and unfolding. Another instance is RNA polymerase, which moves along DNA while carrying out transcriptions, thus acting as a molecular motor. Suitable engineering of natural molecular motors and/or integration of motor proteins within artificial Nanodevices has also been obtained [57,58,236,245,246], thereby creating the possibility of building functional hybrid devices. Fig. 16 shows Natural molecular machines are products of evolution [59].

Rotaxanes and catenanes as artificial molecular machines

Principally, molecular motors and machines can be designed starting from several kinds of molecular [59] and supramolecular [59] systems, including DNA [59]. However, most of the systems constructed so far are on the basis of rotaxanes, catenanes, and related structures. Some relevant components of these multicomponent systems will therefore be explained in summary. The names of these compounds derive from the Latin word's Rota and axis for wheel and axle, and catena for the chain. Rotaxanes [60] are minimally composed (Fig. 17a) of a dumbbell-shaped molecule surrounded by a macrocyclic compound and terminated by bulky groups (stoppers) that prevent disassembly; catenanes [60] are made of (at least) two interlocked macrocycles or "rings" (Fig. 17b). Rotaxanes and catenanes are appealing systems for the construction of molecular machines because of the peculiar mechanical bonds that keep the molecular components together; hence, relative motions of such molecular components can be easily imagined (Fig. 18).

Inorganic (chemical) molecular motor

So far, a wide range of chemical (synthetic) molecular motors has been synthesized by imitating or inspiring natural species. These motors are often similar to today's large-scale machines such as gears, propellers, and shuttles. These devices are easy to synthesize and may even be more productive than natural species [64–66]. Electrostatic interactions and hydrogen and covalent bonds of these elements play an important role in engine performance. Synthetic molecular motors can be activated in a variety of ways, including chemical, electrochemical, and photochemical (light-stimulated) methods. The Rotaxanes base motor, for example, is one of the synthesized molecular motors that can be switched. An illustration of this motor is shown in Fig. 19. These molecular motors are usually composed of two parts: (1) a dumbbell molecule with two heavy chemical groups at both ends; And (2) a light ring-shaped molecule that is locked around the dumbbell-shaped molecule between the two ends of the dumbbell molecule. This set (combination of bar and dumbbell) contains "two specific places with selective absorption" and can be attached to loops [44]. This connection can be polar and van der Waals. Normally, the circular composition is connected to only one of these two locations. With a suitable external stimulus, this connection can be changed so that the ring moves to another active location to maintain the stability of the molecule [67–69]. Therefore, with proper external stimulation, the ring can be moved between two active locations and lead to the motor switch between these two modes. Appropriate external stimulation can be chemical, electrochemical, or optical in nature. Also, how the stimulus is applied can affect motorcycles and speed [16].

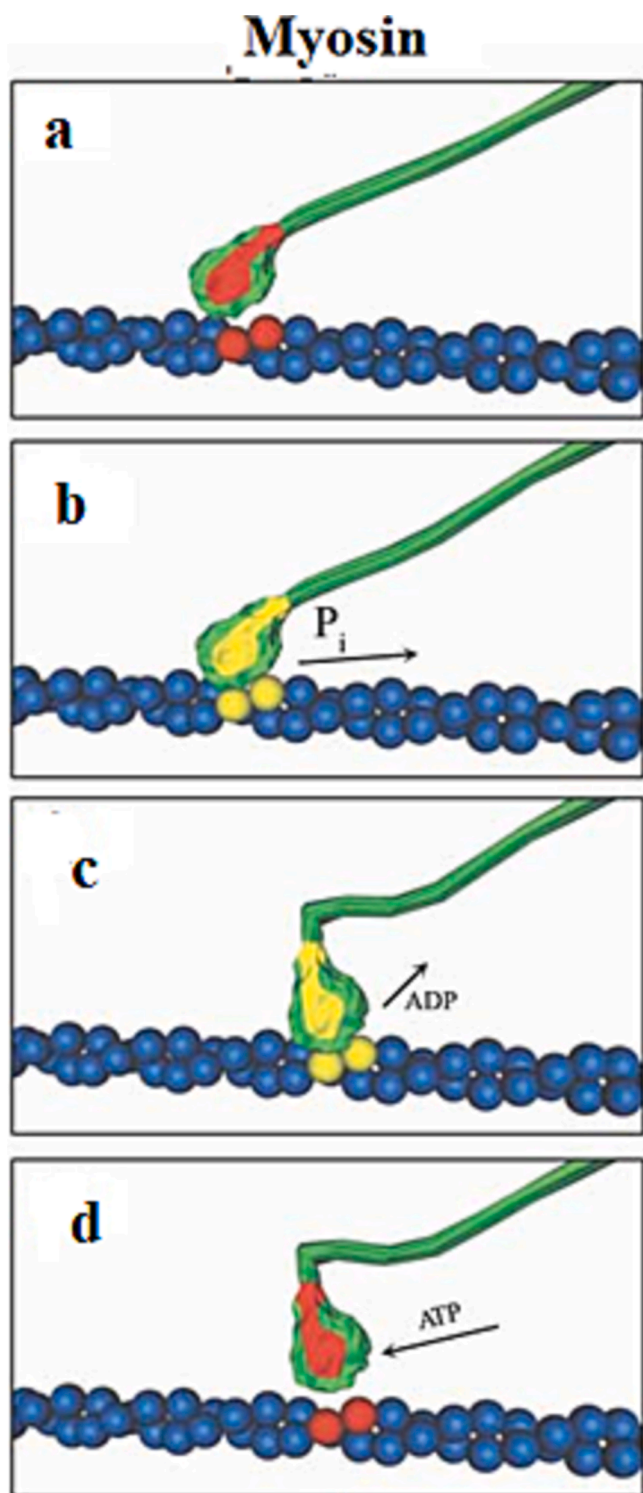


Fig. 12. Scheme of the stages of motion of the myosin molecular motor [12]. (a) myosin motor with an active head (green) (b) When the active motor accidentally collides with the actin filament, the P_i ion left over from the reaction is released and a bond is established (c) This rotation moves the molecular motor by 10 nm along the actin filament (d) The ADP catalytic center then releases itself and is replaced by a new ATP, separating the motor from the thin actin filament. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

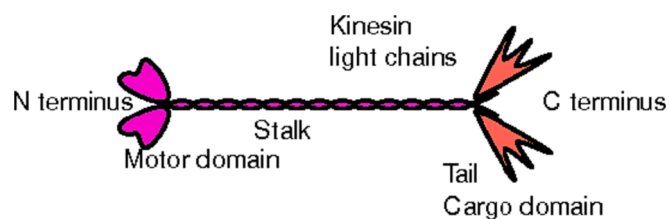


Fig. 13. An overview of the Kinesin molecular motor and its various parts [15].

Because of their special structure, at least two intriguing molecular motions can be imagined in rotaxanes, namely (i) translation, i.e. shuttling [51], of the ring along the axle (Fig. 19a); and (ii) rotation of the macrocyclic ring around the axle (Fig. 19b). Hence, rotaxanes are good prototypes for the construction of both rotary and linear molecular motors. Systems of type (i), termed molecular shuttles, make up indeed the most common implementation of the molecular motor concept with rotaxanes. If two identical recognition sites for the ring, i.e., “stations”, are located within the dumbbell component, the result is a degenerate, conformational equilibrium state in which the ring spontaneously shuttles back and forth along the axle [51]. When the two recognition sites on the dumbbell component are different, a rotaxane can exist as two different equilibrating conformations, the population of which reflects their relative free energy as determined primarily by the strengths of the two different sets of non-covalent bonding interactions. In the schematic representation shown in Fig. 19, the molecular ring stands preferentially around station A (state 0), until a stimulus is applied that switches off this recognition site. The rotaxane then equilibrates according to the new potential energy landscape, and the molecular ring moves by Brownian motion to the second recognition site (station B, state 1). If station A is switched on again by an opposite stimulus, the original potential energy landscape is restored, and another conformational equilibration occurs through the shuttling of the ring back to station A by Brownian motion. Inappropriately designed rotaxanes, the switching process can be controlled by reversible chemical reactions (protonation-deprotonation, reduction-oxidation, isomerization) caused by chemical, electrochemical, or photochemical stimulation.

Cellular displacement

One of the most important functions of molecular motors in nature is the transfer of molecules, supermolecules, various compounds, micron particles, and nanometers. There are a wide variety of particles in the body of living organisms that must be moved between different places with high speed and accuracy [70,71]. These nanoscale particles, like a variety of molecules in living cells, are transported in two ways: (1) by molecular motors; And (2) by transmission by shuttle systems. In the first method, the particle is attached directly to the molecular motor, and as the motor moves in a certain direction, the particle moves along with it. The molecular motors that make this transfer are generally from the Kinesin family of molecular motors [72–74]. These motors, with their two active heads, can move on microtubules in the desired direction. They have the ability to attach a variety of particles to their flexible tail. Fig. 20 shows a picture of the transfer of dunk water (bag, vesicle) by a Kinesin engine onto a static micro tube. The shuttle system is another mechanism for particle transport. Molecular shuttles are nanoscale transmitters capable of transporting micro, millimeter, and even centimeter particles. Shuttles (actin filaments or residues) receive the power needed for transmission from molecular motors. In these systems, molecular protein motors (usually of the myosin family) are attached to a patterned surface from the top of the head, and the shuttles move freely on their tail portions (Fig. 21). The method of particle transfer by shuttles is very similar to the previous method (direct particle transfer by molecular motors); the difference is that in this method, the molecular motors are attached to a fixed surface from the tail area and the

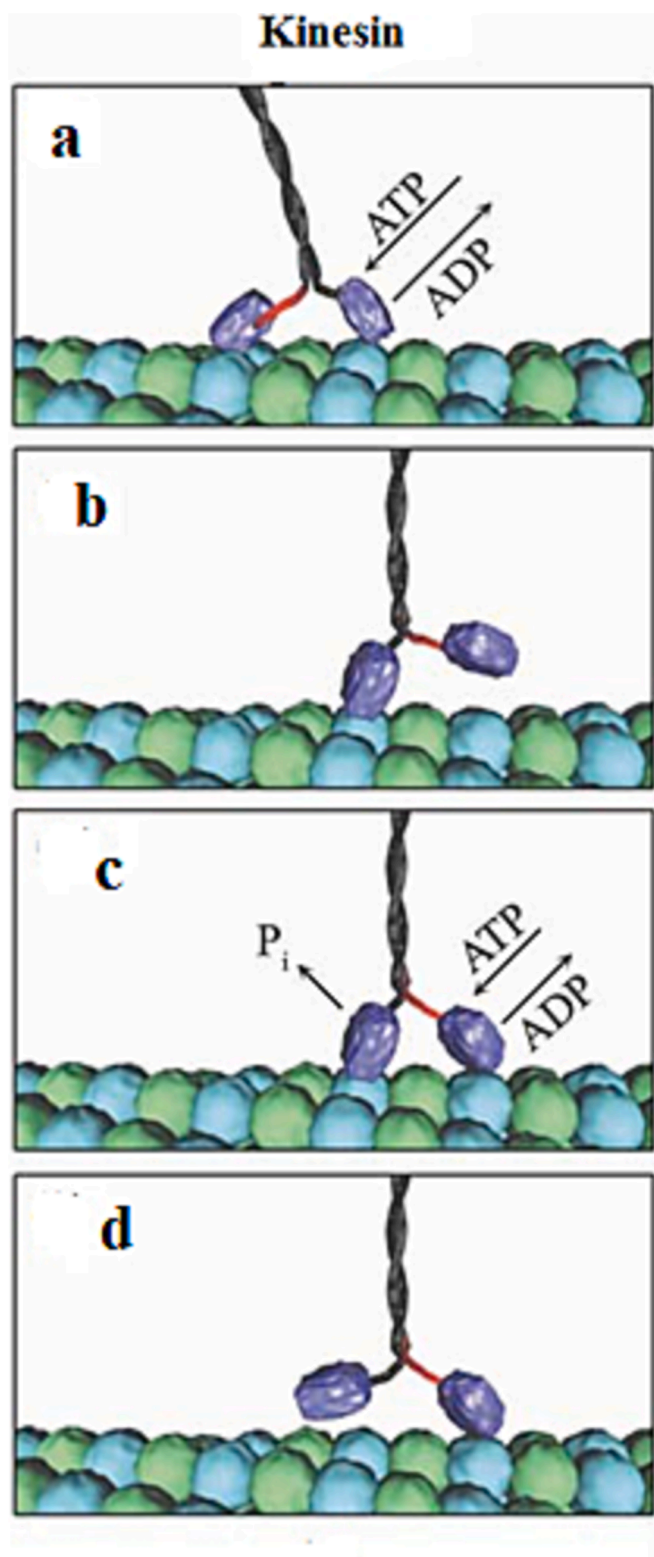


Fig. 14. Scheme of the stages of motion of the Kinesin molecular motor [12]. (a) When one end of the motor hits the resonator, it attaches to it and releases its ADP molecule. A new ATP molecule then replaces the ADP molecule (b) The neck connection causes the other end of the motor to rotate and come forward by bypassing the motor attached to the resonator (c) The ADP front motor is then replaced with a new ATP (d) which rotates the neck joint to bring the rear head forward.

residuals are attached to the other end are moved [75,76]. In fact, the motors are stationary and the shuttles are moving. In this method, the transported components (particles, molecules, etc.) can be attached to the filaments and residues and move with them. In the direct transmission system by the motors, the particles are attached directly to the tail of the motor, and the motor moves with its head on a fixed filament or resin [46]. In the shuttle transmission system, part of the filament or resin is modified so that particles and molecules adhere to the shuttle with ionic, van der Waals, or covalent forces. It should be noted that parts of the shuttles are often left free to be connected to molecular motors. For more emphasis, the shuttle can be likened to a boat floating on water. The inhabitants of the boat act as particles, and the boat motor acts as a molecular motor [77]. Owing to the power created by the motors, the hull of the boat moves and with the movement of the boat, the inhabitants also move. The type and characteristics of the bond between particles and shuttles (filaments and residues) greatly affect the performance of the transmission system [78]. Actin filaments, for example, have a helical structure, which causes the filament to spin once per micrometer on myosin molecular motors. Meanwhile, if the particles attached to the filament also experience this rotation, there may be interference with the movement of the filament.

A molecular motor is usually mounted on a surface with a specific pattern that directs the shuttle movement. These patterns are usually caused by chemical changes or topographic changes on the surface. The connection of particles to the surface of the shuttles is made of ionic and hydrogen bonds and can be done by surface modifiers on the filaments. These connections are removed by adding a special buffer, and particles can be separated from the shuttle at the destination point. These systems are commonly used to transport large particles over long distances. If the transmission distance (transmission length) and particle size is less than a certain value, normal transmission (direct transmission by molecular motor or by free flow) will be faster than shuttle-based transmission [47]. Studies have shown that, approximately, the transfer time multiplied by the particle diameter should be greater than 1000 nm (s.nm) so that the average displacement through the shuttles is greater than the average displacement through free penetration (moving in the direction of the concentration gradient in The environment is generally blue [18,19].

Biosensors

Shuttles can transmit a variety of proteins and viruses. For this reason, the shuttle transmission system can be used as biosensors. In chemistry, chemicals that are examined qualitatively and quantitatively are called “analyses”. Analyses in a biosensor are usually captured by receptor molecules and then tagged. Optical labels emit light of a different wavelength (fluorescent radiation) when they receive the light of a certain wavelength. By studying the light radiation, the analyses can be identified. One of the most common ways to build optical biosensors to identify molecules, proteins, and viruses is to use “double-antibody-sandwich assays.” In this method, the analysis is exposed to the sensor and attached to its receptor molecules (antibodies). After the analytic is attached to the antibody, the additional (unbound) analyses are washed and attached to special optical tags that have entered the system. After that, the extra labels are washed and removed. Finally, the light is emitted and the labels attached to the analyses are identified. Figs. 22-a shows the mentioned process. These steps include receiving analyses, washing additional analyses, labeling analyses attached to optical labels, washing additional optical labels, and identifying analyses. Changing solutions and washing is done manually. The shuttle transmission system can be used in this identification method; In this way, the antibodies are attached to the surface of the shuttles and by absorbing the analytic, they are transported by molecular motors to be labeled in the medium containing the optical label. They are then transferred to another section to be detected by light radiation. In this case, there will be no need to wash. In fact, the use of molecular shuttles arranges the steps of “receive-

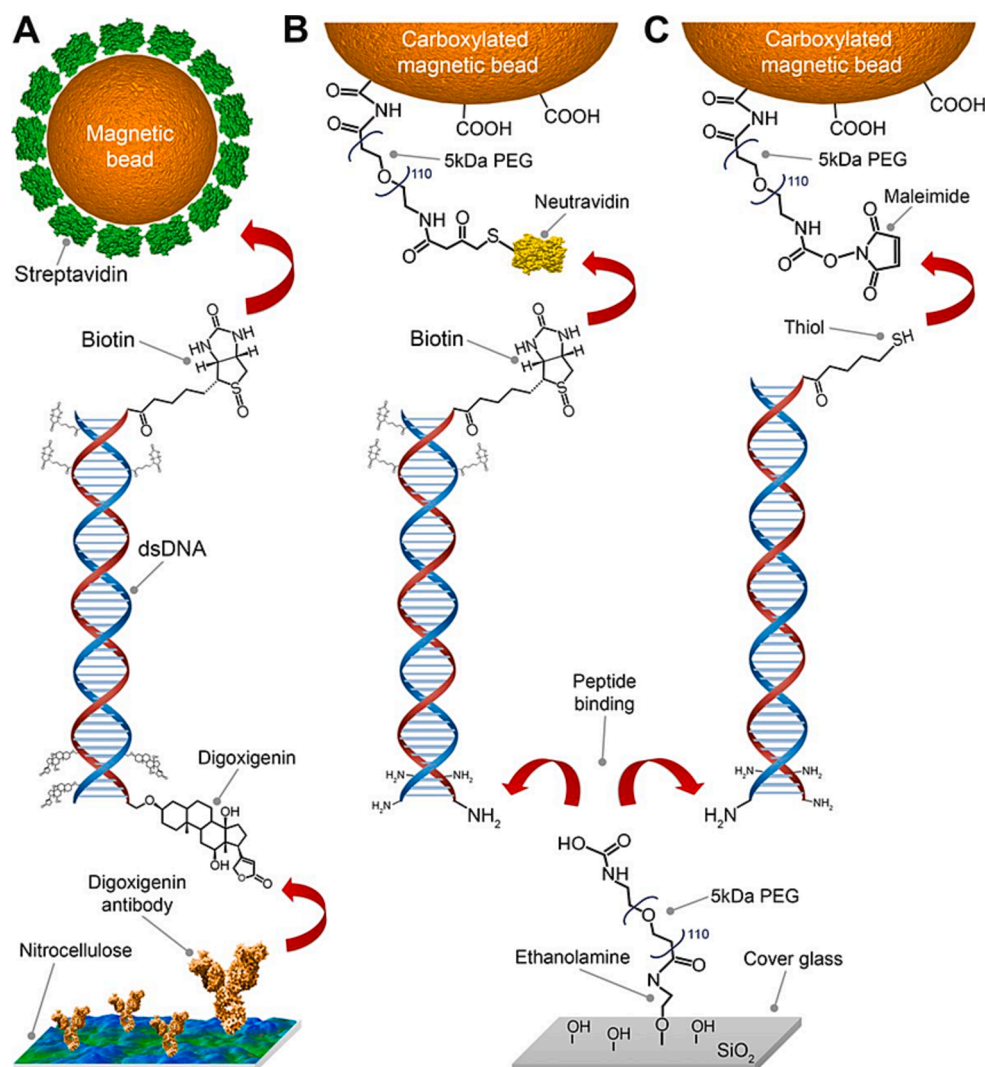


Fig. 15. Schematic of DNA-based operation of Tweezers motor [17].

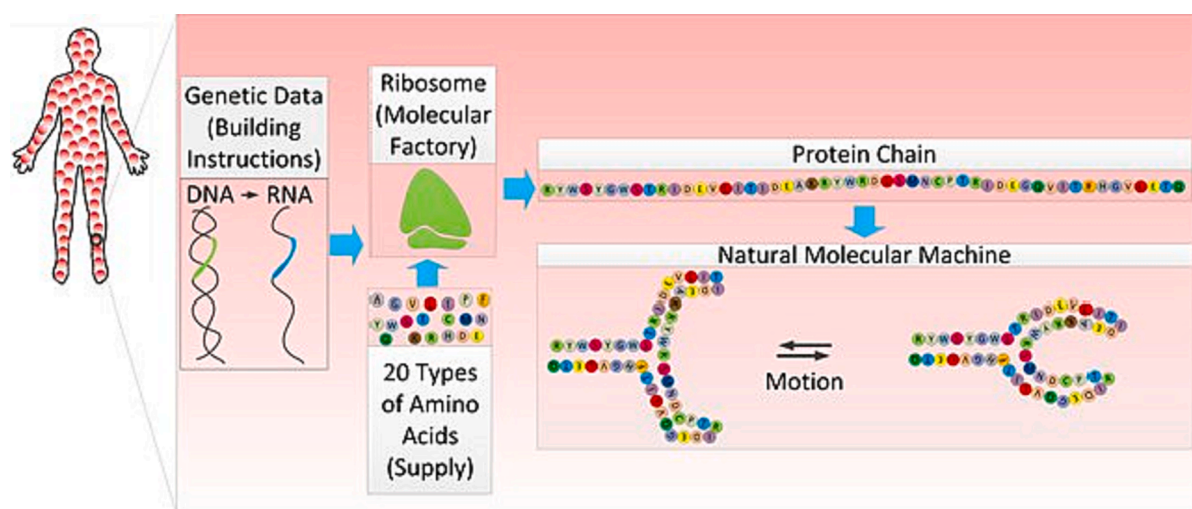


Fig. 16. Natural molecular machines are products of evolution [59].

wash-label-wash-identify” in order for “receive-transfer-label-transfer-identification” Changes (Fig. 22-b). The use of molecular shuttles in biosensors increases their efficiency and ease of detection. Shuttles can

also be concentrated in a very small space. This increases the accuracy of the identification [20,47,48].

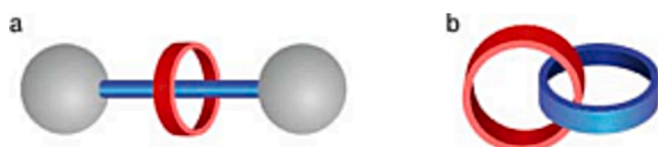


Fig. 17. Schematic representation of a rotaxane (a) and a catenane (b) [59].

Drug

In addition to the tasks and applications mentioned, molecular motors have recently been used to perform specific functions in cells, such as non-viral gene therapy and interneuron delivery. To date, most gene therapy methods have used viral vectors. These carriers are highly

efficient at transmitting gene material to the infected host. Unfortunately, out of 300 conventional gene therapy methods, ten patients are treated for thousands of patients. The reason for the failure of most of these methods is the toxicity of viruses and the natural resistance of the immune system against their entry. Difficult synthesis of transmitters, low transmission capacity, and unstable genes are other limitations of this method of drug delivery [76]. Hence, much research is currently being done on non-viral drug delivery using synthetic vectors. However, there are still many obstacles in the way. The reason for the failure of most of these methods is the toxicity of viruses and the natural resistance of the immune system against their entry. Difficult synthesis of transmitters, low transmission capacity, and unstable genes are other limitations of this method of drug delivery [77]. Hence, much research is currently being done on non-viral drug delivery using synthetic vectors.

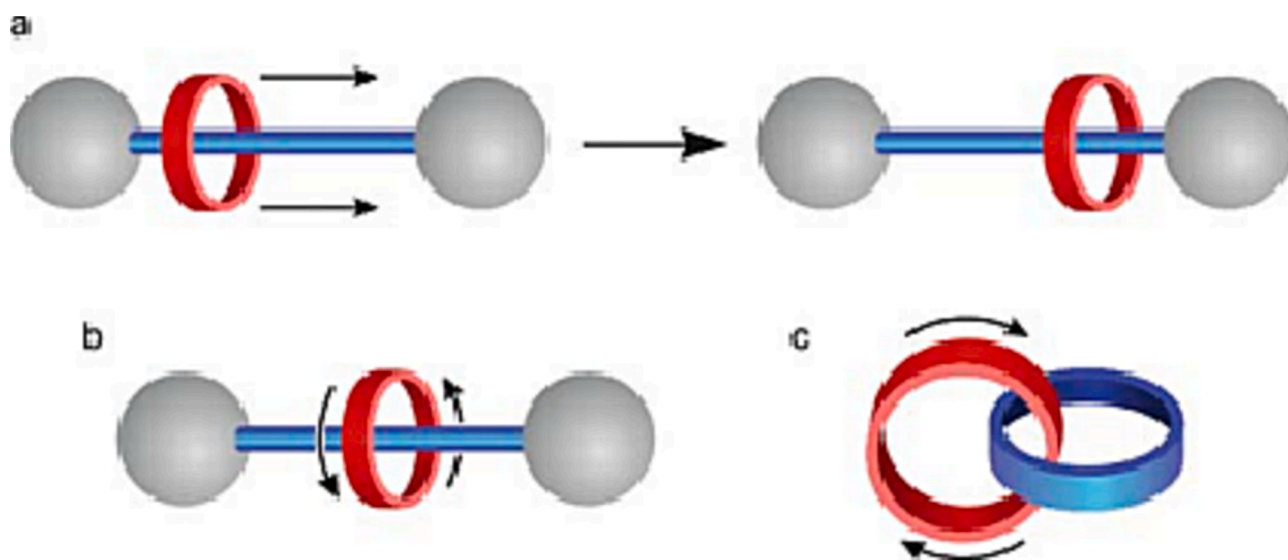


Fig. 18. Some of the intercomponent motions that can be obtained with rotaxanes and catenanes: are shuttling (a) and ring rotation (b, c) [59].

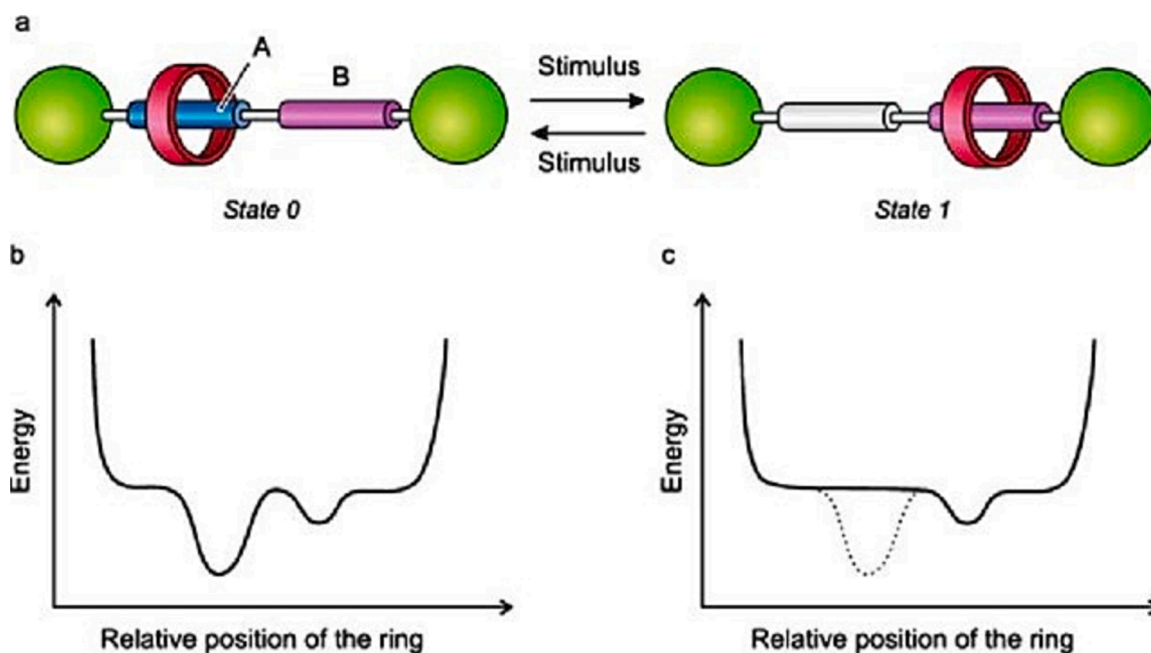


Fig. 19. Schematic of the two-station rotaxane and its operation as a controllable molecular shuttle (a). The figure is a simplified representation of the potential energy of the system as a function of the position of the ring relative to the axle before (b) after (c) switching off station A. An alternative approach would be to modify station B through an external stimulus in order to make it a stronger recognition site compared to station A. [16,50].

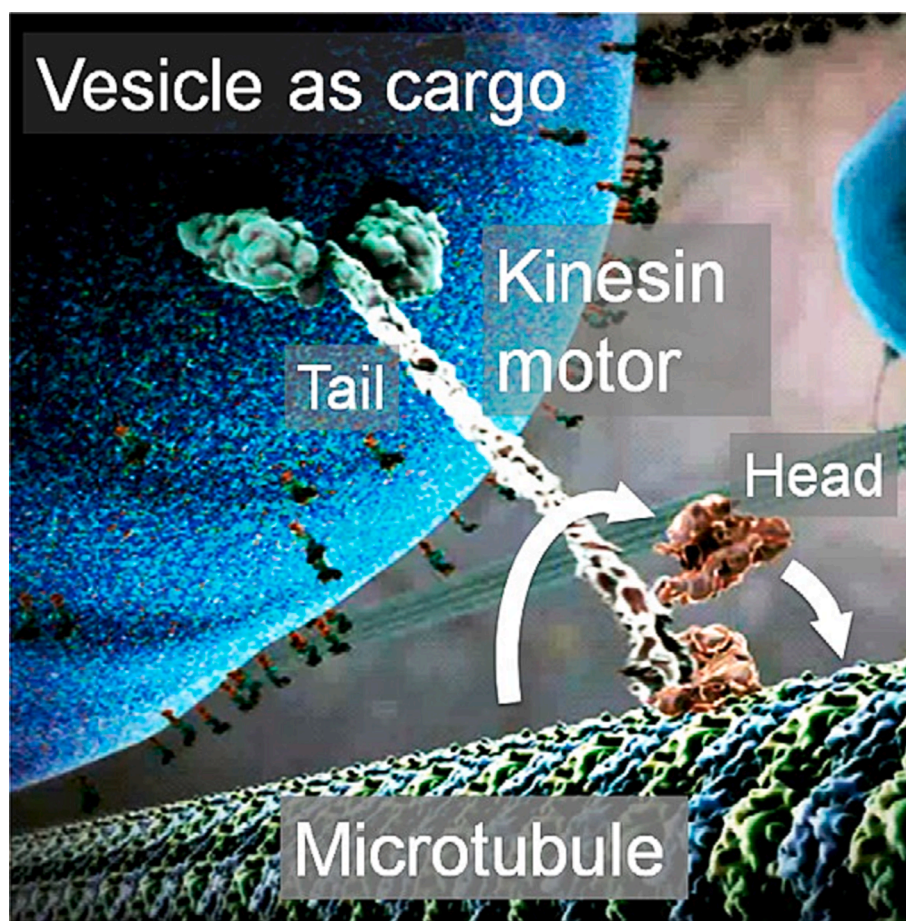


Fig. 20. Image of the transfer of dunk water (bag, vesicle) by the Kinesin motor on a stationary resin [18].

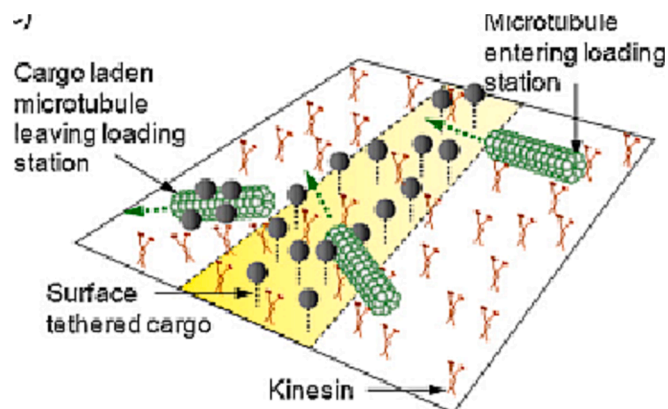


Fig. 21. Scheme of the shuttle transmission system: Particle-free shuttles (resins and filaments) enter the yellow part containing the particles by moving the myosin molecular motors. These particles attach to the shuttles and are transported by the shuttle transmission system to the desired sections that contain the least amount of particles. In fact, the yellow part is where the shuttles are loaded [19]. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

However, there are still many obstacles in the way. One of these barriers is the difficulty of transferring DNA to the cell nucleus. Studies have shown that the very low mobility of DNA in the cytosol portion of the cell (cytosol) is one of the main barriers to this transfer. The cytosol is the intracellular fluid in the cytoplasm of the cell. One good way to get around this barrier is to use molecular motors. Currently, there are at

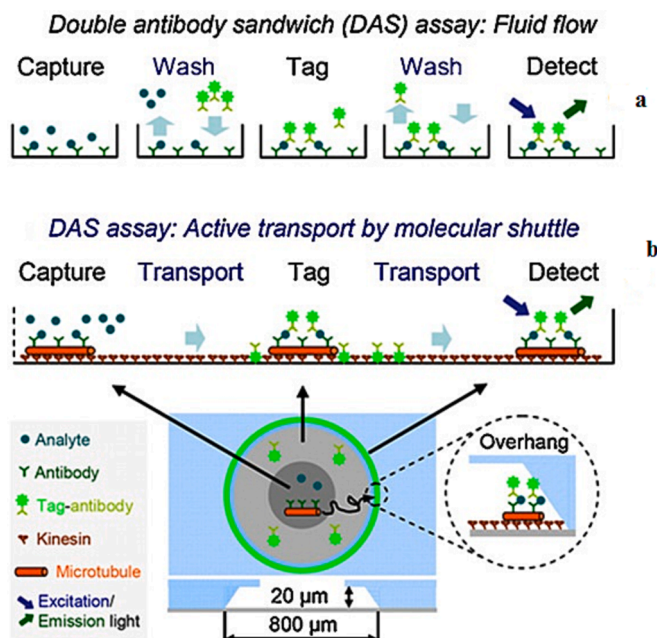


Fig. 22. Sensor array based on antibody sandwich: (a) ordinary type and (b) type based on molecular shuttles [20].

least three approaches to using molecular motors to deliver DNA and other drugs to the cell nucleus [49]:

- (1) Creating a new one-way path near the nucleus and placing a synthesized molecular motor on it;
- (2) Using existing natural routes and placing a synthesized engine on them.
- (3) Using a natural molecular motor with a natural path already in place in the cell.

Synthesized adapters are used to connect DNA to natural motors. These adapters are small pieces of complete DNA or RNA that are used to bind DNA. Fig. 23 shows a diagram of the last two methods that use natural pathways such as residues [21].

Investigation of surface properties (self-propelled probe)

One of the applications of molecular motor and shuttle transmission systems is to study the surface properties and topography of different surfaces. To enter the discussion, consider a surface that has topography and elevation (Fig. 24-a). In this case, kinesin molecular motors can be attached to the surface, and then fluorescent impregnated residues can be added to the surface. Naturally, the pellets are moved on this surface by the Kinesin motor. In this case, the residues go to different parts of the surface that are covered by molecular motors, and the higher the altitude, the fewer residues move to those areas. Now, if we take several hundred images from the surface at different times and place them on top of each other, the places that are less accessible to the resurfaces will be dark (Fig. 24-a). In fact, the higher the altitude, the less residue passes through and the darker it looks. As mentioned, these residues are labeled with fluorescent labels. Therefore, by emitting a certain light beam, the particles emit light with a different wavelength (but a certain) that is recorded by a special camera or photographic screen [46,47]. More accurate information on surface topography can be obtained using

fluorescence-interference contrast (FLIC) microscopy. This technique provides useful information on the surface topography with nanometer resolution using the interference effect of excitation light waves (input) and emitted light waves (output). For 3D imaging, the pattern of the surface to be studied is created on a thin layer of SiO₂. The reason for using silicon oxide is that the light reflected from it is a good criterion for examining the interference of input and output waves. The residues move due to motion on the kinetic and change their distance from the silicon oxide surface according to the surface topography. If they have a high surface concentration, the residues will completely absorb the surface topography (Fig. 20-b). Now, with the FLIC technique and the study of the interference of the incoming waves reflected from the silicon surface and the reflected waves from the residuals, the topography of the surface is obtained with nanometer accuracy. Strong interactions between kinetics and microscopes can be used as a technique to detect the surface concentration of molecular motors, especially when they have very low surface densities (Fig. 24-c). Measuring the rate at which the solutes in the solution bind to the surface containing kinesin motors (or other molecular motors) leads to the identification of the surface density of molecular motors. The accuracy of this method can reach several motors per 1 μm [49]. From this method, the area of the defective parts of a coated surface can be obtained (Fig. 24-c). Molecular motors have the ability to connect defective and uncoated parts. Of course, this method is used for those biological coatings that molecular motors cannot adhere to. Therefore, by measuring the surface concentration of molecular motors, the area of uncoated parts can be obtained.

Applications of molecular motors

Rotaxane dendrimers

Rotaxane dendrimers are a newly emerging large family of mechanically interlocked molecules (MIMs), which combine the concept of molecular switching properties into hyperbranched dendrimers to

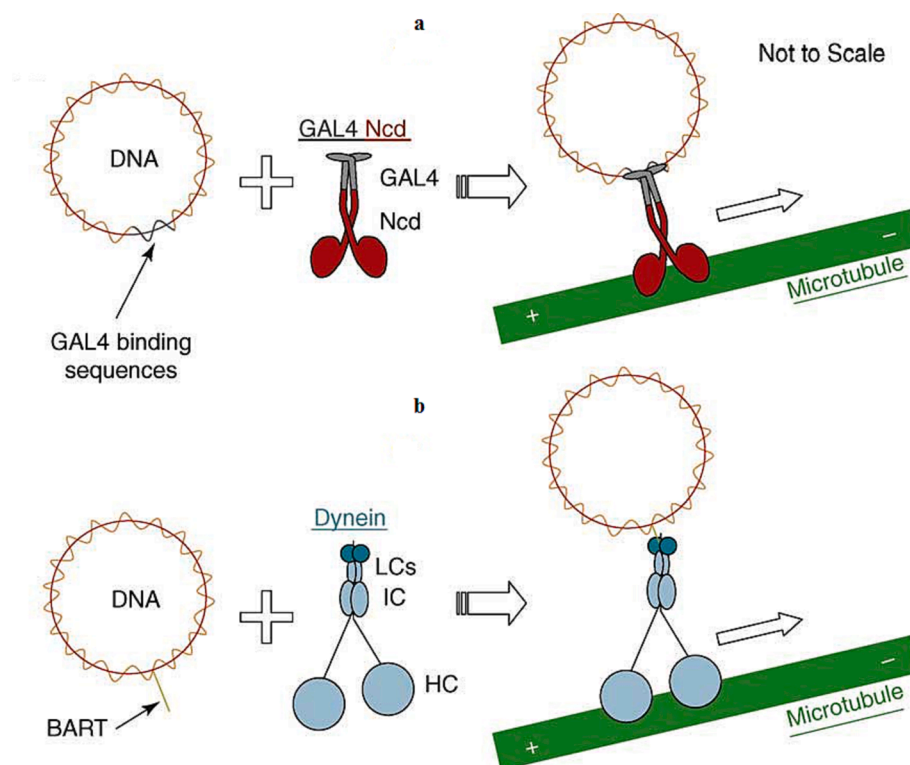


Fig. 23. Use of molecular motors for gene therapy: DNA transfer and delivery to the cell nucleus through the natural path of the residue using a molecular motor connected to DNA (a) and a natural Dining motor (b) by special adapters (BART) bind to DNA [21].

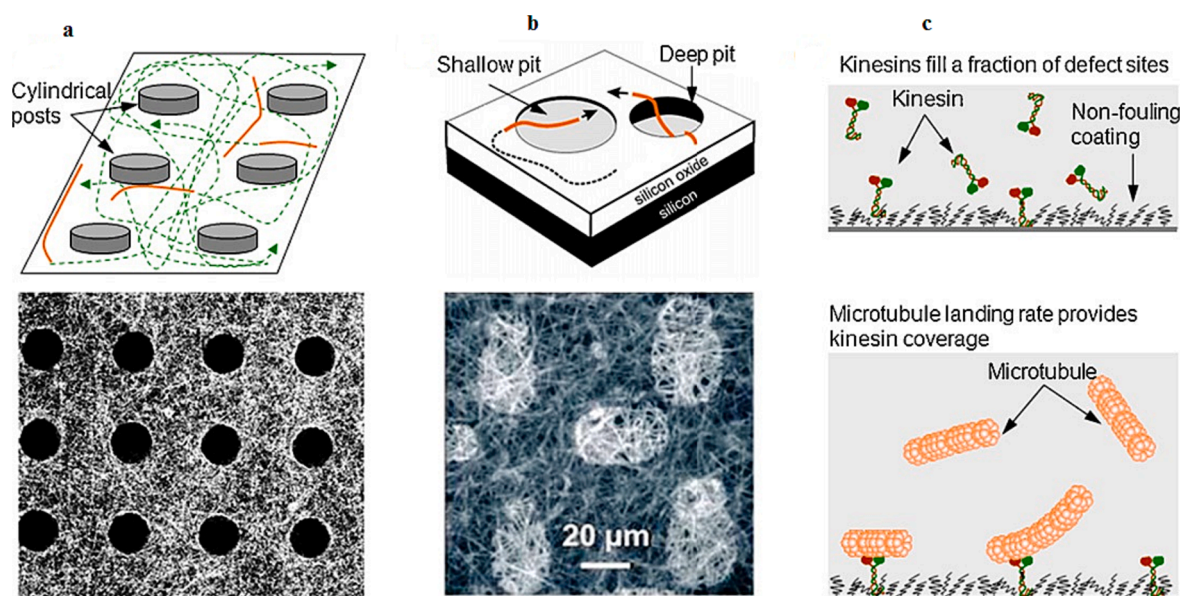


Fig. 24. Survey of surface properties using molecular and shuttles: (a) Topographic information of a surface can be obtained with different images of the random motion of fluorescent reservoirs at different time intervals; (B) The topographic information of a surface can be obtained with very high accuracy using the FLIC imaging technique; (C) Calculate the area of the defective parts of the coating by measuring the concentration of the molecular motors connected to them [4,18–22].

render new macromolecular machines. Basically, rotaxane dendrimers are divided into three types and each type represents a special connection between the mechanical bonds, macrocycles and rods. In the past few years, with the methodology development of rotaxane synthesis, more complex and high molecular weight rotaxane dendrimers have been designed and synthesized, especially the most complicated Type III rotaxane dendrimers. In particular, higher generation sophisticated rotaxane dendrimers bring advancements from 1-dimensional to 3-dimensional molecular switching, and have been applied in advanced functional materials for various applications towards drug delivery. This review surveys the development and advancement of each type of rotaxane dendrimer, from the simple dendron-grafted Type I to intricate Type III dendritic polyrotaxane dendrimers with future perspectives [243].

Knotaxane

Two classes of mechanically interlocked molecules, [3] rotaxanes and knotted [1]rotaxanes, were the subject of this investigation. The necessary building blocks, alkyne-terminated axles containing two ammonium ions and azide-terminated stoppers, and azide-containing substituted macrocycles, have been synthesized and characterized. Different [3] rotaxanes were synthesized by copper-catalyzed “click” reactions between the azide stoppers and [3] pseudorotaxanes formed from the dialkyne axles and crown ethers (DB24C8). Methylation of the triazoles formed by the “click” reaction introduced a second binding site, and switching via deprotonation/protonation was investigated. In preliminary tests for the synthesis of a knotted [1]rotaxane, pseudorotaxanes were formed from azide-containing substituted macrocycles and dialkyne substituted diammonium axles, and copper-catalyzed “click” reactions were carried out. Mass spectral analyses showed successful double “click” reactions between two modified macrocycles and one axle. Whether a knotted [1] rotaxane was formed could not be determined.

Borromean rings

In addition to a parent zinc(II) Borromean ring (BR) complex, the preparation and characterization of two hex substituted BR complexes

with either 4-acetoxymethylphenyl or 4-methylthiophenyl substituents associated in turn with all six pyridyl rings has been achieved convergently in good yields by appealing to the dynamic features of the reactions between primary amino groups in a preformed acyclic ligand and 2,6-diformylpyridine. Two molecules of the acyclic ligands react with two molecules of 2,6-diformylpyridine to form a cyclic [2 + 2] tetraimine in the presence of Zn(II) ions as templates in 2-propanol at 70 °C. The successful preparation of the two derivatives by convergent template-directed syntheses opens up opportunities to self-assemble, under equilibrium control, numerous nanoscale metallo-organic particles with potentially useful properties [244].

Outlook of molecular motors

Molecular motors are strong multifunctional components involved in numerous cellular processes. As such, they require regular activity to improve the timing and impact of their contributions. Exploring future has always been challenging and often created wrong results. Using present work as a guide, four main areas of future research on molecular motors can be recognized. First, even though we seem to have a common idea of motor chemo mechanics, considerable details still need to be worked out. Atomic resolution structures will be the guide. Combining with single-molecule techniques of improved spatiotemporal resolution and sensitivity and the ratio of motor mutants, common principles of motor physiology will emerge. Second, many motors are known only by sequence, especially in plants, so this is a fruitful playground for the cell biological hunter-gatherer. Molecular motors distributed throughout a material enable the controlled modulation of optical and mechanical properties. Biological proof-of-principle examples are the modulation of color in melanophores via the aggregation and dispersion of pigment granules by motor proteins [239] and the modulation of the mechanical properties of muscle as a result of contraction. The demonstration of color tuning of a liquid crystal by activation of synthetic molecular motors has been a major milestone in demonstrating potential applications of molecular motors [240]. However, given the advanced state of solid-state photonics, the demands on competing technologies with respect to switching speed and on/off ratios are particularly high. At the same time, photovoltaics may offer applications for relatively slow tuning of optical properties, similar to the optimization of chloroplast

position by motor proteins in green leaves [241]. Materials with controllable mechanical properties, such as magnetorheological fluids, find widespread use in engineering. Molecular motors provide the added functionality of converting chemical energy into mechanical work and thereby drive energy-dependent property changes such as shape changes against a load. In vitro experiments using myosin motor proteins and actin gels have demonstrated that motors can modulate stiffness by more than two orders of magnitude [242].

Conclusion

1. In this paper, the general principles of molecular motors are introduced. It was said that these motors have the ability to work and move (rotational and linear) at the nanoscale. They need input energy to move, which can be provided by a chemical reaction, stimulation with light, or stimulation by environmental changes such as temperature changes or environmental acidity. Molecular motors perform mechanical work (translational, rotational, etc.) by receiving energy and changing their spatial structure. Although the force generated by a single molecular motor is the power of Pico and Femto newton, much larger forces are produced by putting them together and forming self-assembly structures (such as human muscle tissue). In a regular array of molecular motors, the efficiency of each individual motor is approximately equal to the average efficiency of the entire array. In general, the efficiency of natural molecular motors is much higher than that of synthesized species.
2. Molecular motors come in a wide variety of types. So far, a wide range of motors have been discovered in nature and have been well studied. Natural molecular motors are protein-based, and most fuel is ATP. ATP is the energy source of living systems. One way to produce this substance is through the function of the ATPase molecular machine. Today, many artificial motors have been made by recognizing and imitating natural molecular motors; that can produce forces similar to or even greater than those produced by natural species. These synthetic motors are either DNA-based or chemical. The main fuel for type I motors is DNA molecules, but type II motors obtain their energy through chemical, electrochemical, light, or environmental changes.
3. Molecular motors have many functions and applications in the nature and body of living organisms. For example, myosin motors contract between the actin filaments of muscles, causing them to contract and produce force in the muscle. These are capable of transporting a variety of molecules, particles and chemicals directly or through molecular shuttles. This transmission capability makes it possible to build sensors and deliver drugs using molecular motors. Other applications of these motors include surveying topography of different levels.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The data that has been used is confidential.

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