

## **MICROFLUIDIC ACTUATION IN LIVING ORGANISMS: A BIOMIMETIC CATALOGUE**

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### **KEY WORDS**

Microfluidics, biomimetics, cilia, flagella, classification.

### **ABSTRACT**

*Biomimetic engineering requires reliable and relevant sources of biological information as a spring-board for technology. However, biological classifications are made by biologists for biologists. There are two main groups of biological taxonomies: generic (based on phylogeny) and morphological (based on morphology). But this arrangement of information is useless for engineering, which needs an arrangement based on a functional-morphological analysis requiring a totally different classification. We show that it is possible to create an extremely focused framework, which removes the enormous diversity of living forms, illustrated by the development of such a framework for mechanisms operating with liquids at the micro scale.*

### **1. INTRODUCTION**

The ARTIC project aims to create microfluidic actuators for moving, pumping and mixing liquids at the micro-litre scale, for which living systems possess a wide range of methods. Some of them are very ancient and universal, which proves their reliability. For example, organisms of all sizes move liquids with cilia and flagella and their derivatives. Microfluidic actuation in engineering is a relatively young technology. It can therefore learn from biology. The transfer of biological principles into the sphere of technology is the realm of biomimetics. But the procedures of searching, comparing and selecting an appropriate living prototype, its analysis and exposure of required mechanisms, are still obscure, so this is not a trivial task. This procedure is not yet well defined in contemporary biomimetics. In most cases, direct and verbatim copying is not very successful because of the enormous complexity of living systems and the impossibility of making a perfect functional copy. A simplified version may well decrease the effectiveness of the mechanism and discredit the concept of taking ideas from nature. Moreover, if we create the most perfect copy of some living creature, we must realise that it will inherit not only the advantages of the prototype, but the shortcomings as well (e.g., do we need the toothache which would accompany a perfect denture?). That means that “interpretation” from biological “language” into a technological one is much more promising [1]. Even when the prototype has been chosen, there is still the question of how to implement biological ideas in technology. We suggest a system that helps find novel combinations of the existing properties of the most widespread and universal biological microfluidic actuators: cilia, flagella, cirri and membranelli.

## 2. CILIAR ACTUATORS IN BIOLOGY

All biological processes involve the manipulation of aqueous solutions, starting from micro- and nano- scales as the most ancient. The mechanisms involved have survived millions of years of natural selection and so are extremely reliable.

Existing lists, catalogues and classifications of biological systems, and their functions and effects, are not particularly useful for engineers, because they are made by biologists mostly for the needs of biology. The main differences of the requirements of biologists and engineers are shown in table 1.

Classification for a biologist	Classification for an engineer
Descriptive information	Prescriptive information
Classification is genetic (taking into account the kin-relations) and therefore objective. This is not always related to a single function or context.	Context-oriented and therefore subjective. Can be adjusted for convenience of use.
The purpose is purely theoretical, detecting and discovering the most fundamental laws of matter and natural mechanisms that generate the diversity of life.	Highly practical. Quick, easy, correct search/access (based on various parameters and for different contexts) of a relevant prototype.
Classification is based on morphology.	Classification is based on function and context.
Attention is focused on the past and present, detailing the palaeontology and phylogenesis of the system.	Attention is focused onto the future which is where the reward and benefit are.
The enormous number and diversity of species, and the inferred existence of unknown extinct but relevant organisms, make classification difficult.	The enormous number and diversity of species, makes selection of the best biological prototype very difficult.
Biological diversity is "good". A biologist searches for and describes new entities to make the list as full as possible and tries to classify the new objects as precisely, as possible. Valuation of the biological entities is absent.	Biological diversity is "bad". An engineer is overloaded by the biological diversity, thus his choice of a prototype is often nearly random, because of lack of objective criteria for selection and valuation. Thus biological evaluation for biomimetic classification is essential.

**Table 1.** Comparison of biological classification and desirable technological classification

There is no proper technological classification of biological entities. We propose desirable features of a classification that would satisfy engineers. Ideally such a classification should be open and allow the inclusions of new entities and also allow options that are not present in contemporary organisms. These options are the main resource for biomimetic devices to be developed as they use the advantages of both life and technology.

Nearly every living creature moves liquids with cilia and flagella and their derivatives. We have considered all biological mechanisms for moving water at the microscale, concentrating on features of design that may be helpful for the engineering of artificial cilia. To enable this we have developed a logical framework that allows quick searching for the parameters of ciliary movement together with the requirements of engineering. We have already made an "engineer-friendly" catalogue of methods of manipulating liquids in living systems [2]. But it is comprised only of existing organisms. Engineering is aimed at creating something that does not yet exist and improving on the predecessors.

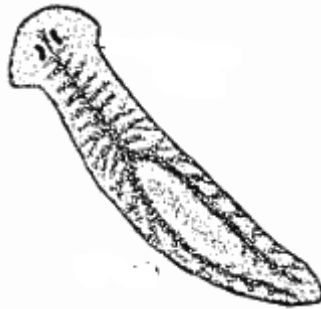
This was the starting point for creating a biomimetics tool for extracting required effects and transferring them into an engineering context. Basing on the system approach we defined first the structural level at which to consider the biological entities. As we were tasked to develop an artificial ciliar actuator, we decided to concentrate on cilia and the basement membrane into which they are inserted. This step is arbitrary and directed only by the function which is to be transferred. In order to produce a generalized model for technology transfer, we will call the combination [cilia + basement membrane] our *system*. We could choose any level for our system, e.g. the level of internal structure of a single cilium (which is a *sub-system* of the system we have chosen) or the levels of organs and organism (which contain the system we have chosen and so can be considered *super-system* and *super-super-system*). The next step is to describe the basic physical components and features of the system. This will give us a list of parameters: a cilium is dynamic, internally driven (i.e. active), flexible, morphologically symmetrical (radially symmetrical cross-

section), etc. The analogous list of properties is made for the other component of our system – the basement membrane. This can be dynamic or static, passive or active, flexible or stiff, elastic and continuous. All these parameters are about mechanical properties and the list can be easily and naturally extended by adding the opposite ones when and if absent (e.g., plastic – elastic, symmetrical – asymmetrical, etc.). The next step is to arrange this list in the form of binary tree of paired opposites (which is a thesis – antithesis structure, typical of many types of classification). Since we have two independent components in the system we can make a table or matrix very easily (table 2). This represents our design space. Note that if we had more components in our system we would need more dimensions in the matrix. It may be convenient to limit the number of components at the definition of the system, but this might cause problems by forcing us to ignore important components. Now we can take all the known biological examples and plot them into the cells of this table according to the parameters listed. Let us consider the most typical ones.

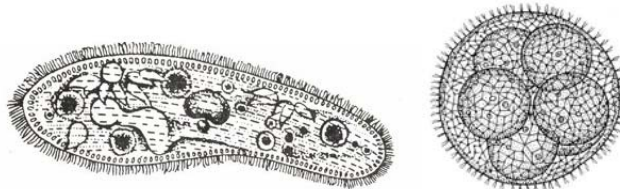
Flat worms (e.g., *Planaria maculata*) live in or near water and are totally covered with cilia. The concerted metachronal (wave-like) movement of the cilia provides smooth locomotion along surfaces. Most ciliated infusorians (e.g., *Paramecium caudatum*) and flagellated organisms (e.g., *Volvox globator*) provide locomotion with cilia and/or flagella. The body of the animal does not usually move as well, for instance by wriggling. Infusorians with cirri (e.g., *Stylonichia mytilus*) are not very common. Cirri are longer and thicker than cilia and are rigid. This shows that small effectors such as cilia can be amalgamated into tiny legs that can walk on a solid surface.

Cilia in a macro-organism create a constant flow of liquid with suspended particles. Normally the direction of flow is constant. Membranelli (flat triangular structures formed by the amalgamation of several cilia) create a paddle or blade, which has a bigger area and thus can be more effective than a single cilium. Cilia and membranelli of sessile aquatic animals (e.g., *Stentor globator*) operate as manipulators, but are obviously not locomotory since the organism is anchored to the substrate. The enormous diversity of living creatures that use cilia employs only a surprisingly small number of the available parameters. The same parsimony has been found in other biological structures, e.g. the morphology of coiled shells [3]. This type of analysis thus allows us to escape from the biological paradigm with its inherent limitations due to morphology and phylogeny since we are presented with more possibilities than exist in nature.

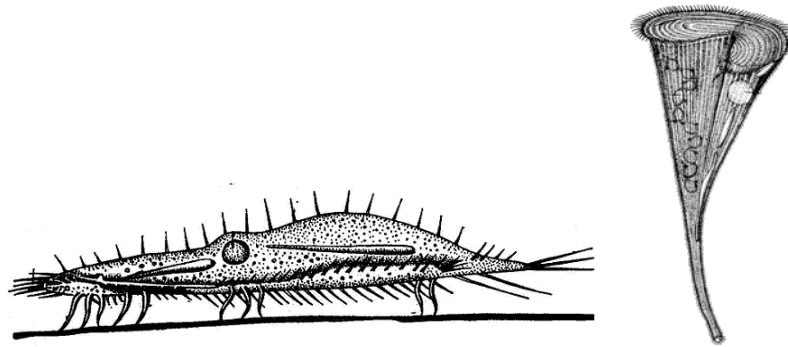
Now we see all the existing possibilities of the combinations of the parameters to be applied for the design, we can start the constructive part of the process, assembling novel actuators using the properties provided by the table 2.



**Figure 1:** Flat worm, *Planaria maculata*



**Figure 2:** Ciliated infusoria *Paramecium caudatum*, and flagellated organism *Volvox globator*



**Figure 3:** Stylonichia mytilus (left and, Stentor polymorphus (right).

### 3. BIOMIMETIC CILIA

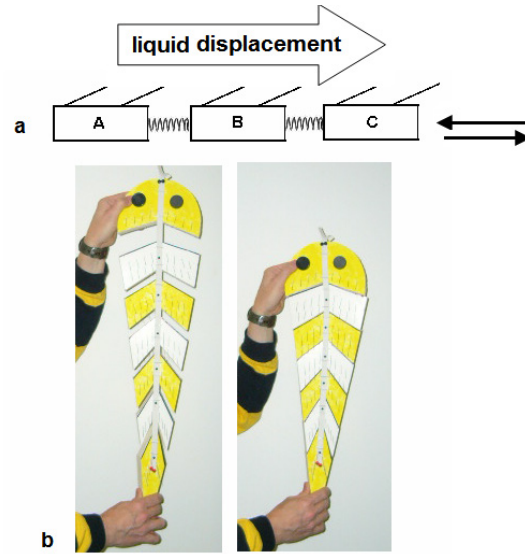
To borrow ideas from nature for our artificial cilia we first need to know what we want to change in the standard engineering approach. What is the ideal device to transport water on a micro-scale (table 3)? It seems that not all the features of natural cilia are suitable as they are either difficult (and therefore expensive) to implement or require complicated control mechanisms. Ideally we need to achieve the advantages of natural cilia by the means that currently available in engineering.

Using the field of possibilities (table 2), we decided to make a model of such an ideal ciliary actuator. As it is very difficult to actuate individual cilia and to control them, we decided to make the main driving part not the cilia, but the basement. As a result of combining various desirable (and also logically possible, practical and affordable) parameters we developed a device with an externally driven (passive) stiff asymmetrical cilium on an active flexible elastic actuating basement. We called it “Creepy” (Figure 4, a, b). The main body of the device is segmented perpendicularly to its longitudinal axis and the segments are joined by elastic elements. Every segment is provided with rigid needles (“cilia”) bent in one direction at an angle of 30 degrees. Being attached at the point A the whole array can be stretched and then returned to its initial position. All segments begin moving synchronously. This reciprocating movement can move objects placed on the surface with the rigid asymmetrical needles due to their polarised direction.

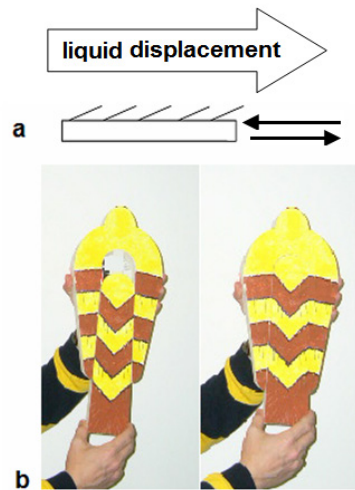
Animate biological cilia	Preferred artificial cilia
Morphology: radial symmetry giving 3D operation	Structure: axial symmetry and lateral asymmetry giving 2-D operation.
Functionally universal	Functionally specialized
Shape of a cilium is typically round.	Cilium should be flat.
Metachronal reciprocal movement is combined with other types of actuation and/or with hierarchically organised geometrical effects.	Typically synchrony. Type of actuation: normally all types of rotation.
Reversible switch to any direction	Actuation in a single direction
Active (internally actuated)	Passively driven (externally actuated)
Function of liquid transport is supported on multiple hierarchical levels of “a device”.	What do we want? Many functions?

**Table 3.** Artificial cilia: facts and artefacts (Types and prototypes.)

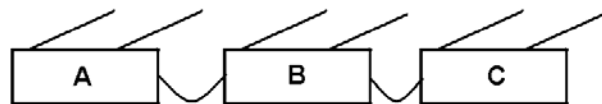




**Figure 4:** “Creepy” – a ciliar actuator. (a - side view shows segments A, B, C, rigid “cilia” and elastic elements connecting segments; b - model in stretched and contracted positions – segments are painted in contrasting colours).



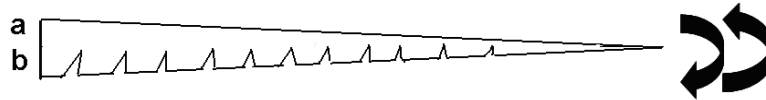
**Figure 5:** “Crawly” – another ciliar actuator (a - side view; b - model showing longitudinal segmentation – the chevrons show the displacement of segmented parts). The central part of the device moves to-and-fro relatively lateral parts. As liquid at the microscale level possesses high viscosity, it will be moved by the rigid asymmetrical cilia, which cover all the surface (moving and non-moving parts) of “Crawley”. The central moving part is made as a wedge for easier moving.



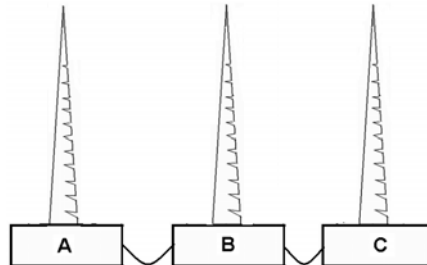
**Figure 6:** Metachronal wave is allowed by the looseness of the joints.



**Figure 7:** The difference in stiffness between the two sides (a, b) of a cilium provides asymmetry for effective and recovery strokes.



**Figure 8:** Different behaviour of cilium in effective and recovery stroke due to one sided segmentation of a cilium.



**Figure 9:** “Metachrone” actuator with morphologically asymmetrical cilia.

We also tried different combination of the parameters and designed another biomimetic device with passive rigid asymmetrical cilia and an active rigid segmented actuating basement (“Crawly”, Figure 5, a, b). The foundation is segmented longitudinally. It is provided with needles bent at 30 degrees and the moving parts are joined with elastic elements. All segments begin moving synchronously. The reciprocating movement transports an item placed on the needled surface functioning like a conveyor belt.

Finally we built a device with an active fragmented actuating basement with loose joints providing the metachronal wave. We called this one “Metachrone” (Figure 6, a, b). The moving parts of these devices work synchronously, but this array provides a wave, which itself moves. It is actuated as in the previous devices by reciprocation.

To get maximum of beating cilia must show asymmetry in effective and recovery strokes. In the course of effective stroke they must be stiff, but in recovery stroke – relaxed and flexible. Living cilium provide these properties functionally. But in our case it is much easier to make it with the morphological means. So to get maximum similarity with a biological ciliar surface the segments must be provided with morphologically asymmetrical cilia. This gives a metachronal wave not only by segmental movement, but also by the entire surface covered by cilia. Note that this functional effect is achieved by totally different morphological structures and actuation from living prototypes.

The main function of a cilium is creating fluid flow as a result of asymmetrical beating – effective and recovery strokes. The effective stroke requires the cilium to be rigid and the recovery stroke demands a relaxed flexible structure. In the living cilia this is provided by the functional asymmetry of the morphologically symmetrical internal structure, which is the actuator itself – an analogue of a linear electric motor, where linear elements are actively sliding or fixed relative to one another at different stages of the motion cycle. An artificial cilium, which is inevitably simpler, is more likely to be a passive device driven by external sources of energy – electromagnetic or electro-static fields, active basement, etc., but must also provide asymmetrical effective and recovery strokes. This can be achieved if the two sides of the cilium are of different stiffness (Figure 7). This can be achieved by small cuts perpendicular to the main axis along the cilium (Figure 8).

These morphologically asymmetrical cilia can be attached to the basement of “Metachrone” (Figure 9), which will provide a real metachronal wave of asymmetrically moving cilia – the main requirement of the current microfluidic project and the necessary condition for creating a flow of liquid.

#### 4. CONCLUSIONS

Direct copying of natural prototypes is not the easiest and most efficient biomimetic strategy. We suggest the following steps.

1. Define the main function of a device, its environment, time, and size scale.
2. Look for prototypes in biology. “Dissect”, analyze and classify them according to the main functional requirements. This is the *system*.
3. Make a list of parameters that are essential for the performance of the main function.

4. Extend the list of the properties by adding the opposites to every parameter.
5. Arrange the parameters in a table with the two parts of our system variables in columns and rows.
6. Find a combination of the most desirable, possible and affordable parameters for a biomimetic device that does not exist in nature, but can be implemented with the help of engineering.

Our models of artificial ciliar devices prove once again that biomimetics should not copy natural systems, but extract the most essential and significant features from the realm of biology and translate them into the language of engineering.

Biomimetic approach to engineering does not end with choosing right living prototype. It opens new engineering opportunities in design as we can see the whole range of possibilities that are not realised yet in living nature. Such non-existing combinations of various parameters inspire engineers to develop a prototype of potential artificial actuators, where advantages of living and engineering systems are merged together. “Creepy” and “Crawly” are biomimetic creatures that employ in their principle of action those mechanisms which are not in use in living nature, but which are possible to implement with the means of contemporary engineering. The next stage of the work will be to design a set of experiments to vary parameters of the devices to identify its most optimum action, e.g., density of the ciliar “carpet”, angles of cilia, dimensions of cilia (length, width/diameter, shape of the cross-section of a cilium, etc.). It is obvious that different liquids will require displacement of some above-mentioned parameters (moving, pumping and mixing). Scaling effects should be also envisaged, but carefully adjusted to the parameters of media where the device is operating (liquids, gels, emulsions, suspensions and various inclusions of bigger size). In spite of the fact that biological systems operate with or within liquids with wide variety of parameters and in different scales, the set of actuators and principles of action is amazingly similar. Cilia, bristles, tentacles, paddles, membranes/fins create actuating poly-systems at any scale level as well, as wave-like movements are creating undulation, metachronism, peristalsis and their derivatives. Similar morphological structures (tubes, funnels, branching pipe works, spirals/helices, suckers, etc.) also can be found nearly at any size levels in living systems. This gives us an idea that the functional properties of all those mechanisms have much in common under various conditions and contexts and can be employed for biomimetic developments at all ranges of scales.

All these issues are supposed to be considered as further steps in the design of biomimetic microfluidic actuators.

## ACKNOWLEDGEMENTS

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