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PERSPECTIVE

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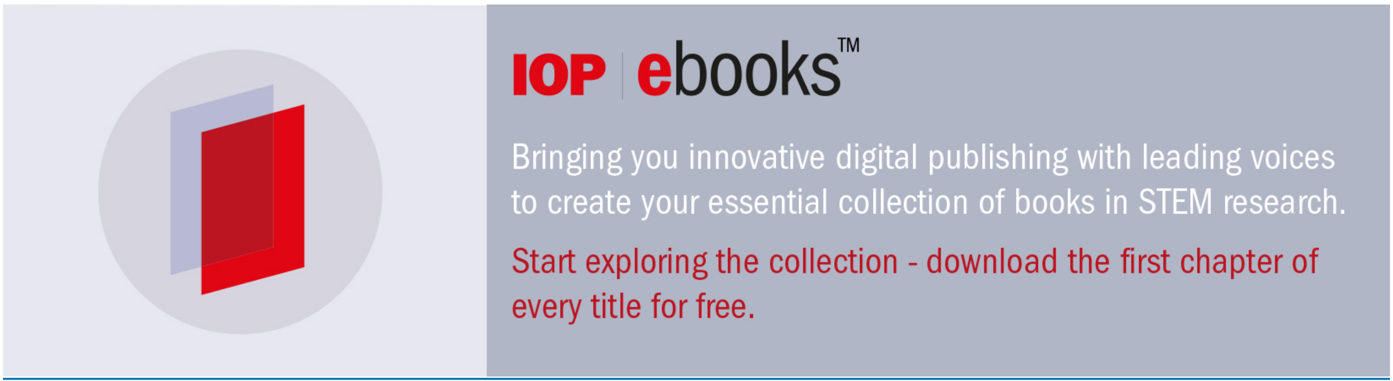

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PERSPECTIVE

Biosynthetic infochemical communication

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Abstract

There is an ever-increasing demand for data to be embedded in our environment at ever-decreasing temporal and spatial scales. Whilst current communication and storage technologies generally exploit the electromagnetic properties of media, chemistry offers us a new alternative for nanoscale signaling using molecules as messengers with high information content. Biological systems effectively overcome the challenges of chemical communication using highly specific biosynthetic pathways for signal generation together with specialized protein receptors and nervous systems. Here we consider a new approach for information transmission based upon nature's quintessential example of infochemical communication, the moth pheromone system. To approach the sensitivity, specificity and versatility of infochemical communication seen in nature, we describe an array of biologically-inspired technologies for the production, transmission, detection, and processing of molecular signals. We show how it is possible to implement each step of the moth pheromone pathway for biosynthesis, transmission, receptor protein binding/transduction, and antennal lobe processing of monomolecular and multimolecular signals. For each implemented step, we discuss the value, current limitations, and challenges for the future development and integration of infochemical communication technologies. Together, these building blocks provide a starting point for future technologies that can utilize programmable emission and detection of multimolecular information for a new and robust means of communicating chemical information.

1. A vision for ratiometric infochemical communication

One of the greatest challenges of the information age is the requirement to transmit, control, and assess ever-increasing streams of data. The computational capacities of various information systems have increased at rates of at least 58% in the last 25 years, whereas communications capabilities have increased by only 28% [1]. There is therefore a need to bridge this gap through the development of new and alternative communication technologies that can work effectively in a variety of media and environments. Moreover, there are circumstances in which electromagnetic data transmission is in itself problematic, such as within liquid media, where an alternative approach is needed.

Whilst our digital world is currently dominated by electromagnetic communication systems, chemicals are the ubiquitous language of nature. What if we could connect our digital world more intrinsically to nature through communication at the molecular level? Molecules are fundamental units of biological information and provide an almost limitless source from which combinations of *infochemicals* precisely delivered in space and time can establish a highly complex and rapid form of communication. Compared to electromagnetic communication, molecules offer a very wide range of symbol values and atomic configurations that can be transported in a variety of media to effect a concomitant variety of biochemical signaling functions. Exploiting molecules as a fundamental unit of information in this way provides the opportunity to create a new form of information transfer that is intrinsically embedded within the physical world. True convergence of information communication technology with biology (bio-ICT) will require new sets of tools and technologies for precise delivery, monitoring, control and detection at the molecular scale that may act as technological translations between conventional electronic and molecular domains.

Interest in molecular communication has expanded rapidly [2], but few empirical studies have been performed [3] and the term currently covers a wide variety of approaches from many areas of science and technology. Molecular motors using cytoskeletal filaments [4] or microplatforms of cells expressing gap junction channels [5] have been proposed for molecular signal propagation, but longer range molecular communication remains a relatively unexplored field [3]. Most progress in molecular communication falls under the realm of synthetic biology, which involves the analysis and engineering of cellular signaling processes (for recent reviews on manipulation of cellular signaling modules, see [6, 7]). Thus, much of the work relating to molecular communication has dealt with signal propagation between these cellular components, including manipulation of metabolic pathways for the construction of gene-metabolic circuits for bacterial communication [8]. Some progress relating to molecular communication is also occurring in the burgeoning field of chemical computing [9], where biomolecular components such as enzymes, DNA, RNA, and cells are used to create computing systems for information processing. As with molecular communication, however, this field is still embryonic [9].

In the past decade, molecular communication has also emerged as a concept for signaling between biological nanomachines [3, 10] whilst infochemistry has been used to describe a method of communication using chemical reactions to transmit alpha-numeric information optically from burning 'infofuses' [11]. In the latter study, the communication media was electromagnetic, but initiated by controlled chemical reactions. A recent bench top chemical communication system [12] has also been reported that focuses on the timing of pulses of high concentration of a single compound, but not the informational content of the molecular structure itself nor possibilities for multimolecular information. These methods both utilized chemical information as a simple binary digital code. Here, we consider an alternative approach—to harness the mechanisms of chemical communication adopted by nature as a technological solution to multimolecular infochemical communication.

The development of a biomimetic infochemical technology requires consideration of the entire cascade of communication events from signal encoding and controlled emission (i.e. transmission) to sensitive and specific signal detection and decoding (i.e. reception). Such technology must precisely sequence and coordinate all necessary events from programmable encoding through to millisecond molecular detection and subsequent decoding to complete a chain of high-bandwidth infochemical communication from a set of diverse *infochemicals*. This introduces a significant set of challenges. First, the encoding process depends upon either ready availability or real-time production of a diverse set of molecules supplied to a microactuator capable of generating a unique infochemical message localized in both space and time. To achieve sufficient bandwidth, such a system should precisely control blends of molecules for release into the environment in a programmable manner to encode specific messages over time. At the receiver side, the diverse set of chemical ratios must be accurately detected with a sensitive and specific microsensor array that minimizes crosstalk interference between molecular types acting as separate channels of information. This multimolecular stream must finally be decoded in real-time to extract the original message. The performance of the overall system depends upon the diversity of the chemical library deployed, the discrete temporal resolution of signal delivery and detection, noise in the receiver, interchannel crosstalk, and the encoding strategy used. Some mathematical treatment of stochastic signal generation, encoding schemes, trafficking and detection of molecular signals has been reported [2, 13].

Realizing this vision for multimolecular infochemical messaging requires a fresh approach to developing technologies for encoding and decoding diverse sets of molecules. Instead of embarking on an endless rational design of perfectly specific sensors for a potentially infinite set of infochemicals, we propose a solution for communication provided by biology. Biological systems effectively overcome the challenges of chemical communication using highly specific biosynthetic pathways coupled with specialized protein receptors and nervous systems for efficient decoding [14]. The olfactory system of animals deploys a set of olfactory receptors (ORs) such that the pattern of receptor population activity uniquely encodes an astonishing variety of chemical blends. Animals possess both ORs tuned to diverse chemical information in their environment, as well as a central nervous system evolved to fuse and extract information about specific molecular combinations. These tiny, specialized, and low-power biological systems thus offer an ideal model on which to base technologies for energy-efficient multimolecular communication.

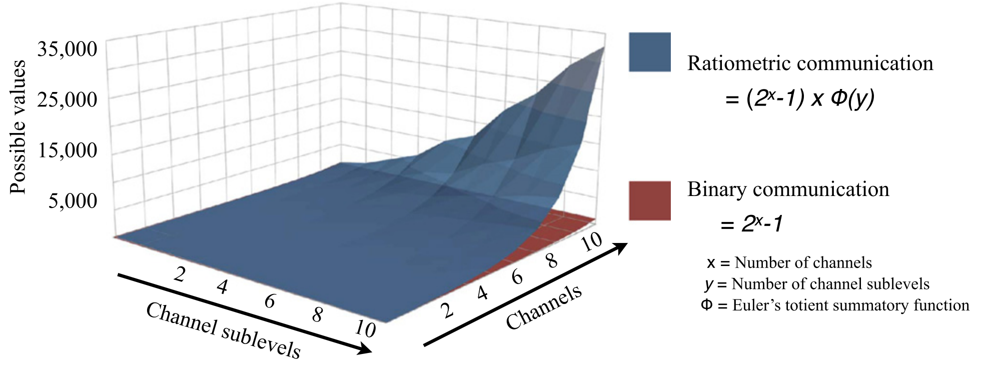

Figure 1. Ratiometric communication. For x channels, traditional binary (digital) communication (red) allows $2^x - 1$ possible values of information. In contrast, ratiometric signals (blue) provide $(2^x - 1) \times \Phi(y)$ where Φ is Euler's totient summatory function $\left[\sum_{n=1}^{y} \phi(n)\right]$ and y is the number of sublevels within each channel (between on—100% and off—0%) that can be detected (e.g. 1, 50%; 2, 33%, 66%; 3, 25%, 50%, 75%; etc). Thus, for five channels, binary communication offers 31 possible values. However, ratio-based communication provides up to 124 unique values with three additional sublevels of detection (i.e. 25, 50, 75%). Note that the values obtained for ratiometric signals are smaller than $(y + 2)^x - 1$ as the channel, x, and sublevels, y, are not independent of each other but are instead a ratio to the other channels. Thus, ratiometric communication does not distinguish, for instance, 2:2:2:2:2 from 1:1:1:1:1, and therefore sacrifices amplitude information for concentration invariance, a key feature required for turbulent, real-world environments.

2. The moth pheromone system: a model for multimolecular infochemical communication

Infochemical communication is ubiquitous amongst living organisms [15, 16]. Insects, in particular, are pre-eminent infochemical communicators of the natural world. Because the sense of smell is a primary modality for insects [17], infochemical blends must be constantly decoded in order to locate food, predators, or sexual mates with millisecond precision in a tiny olfactory system. The moth pheromone system is proof *par excellence* of multimolecular infochemical communication. Moth sex pheromone blends generally comprise a small number of closely related but distinct compounds of different carbon chain lengths, geometric configurations, functional groups and levels of desaturation that are released by the female and detected by the male in specific ratios. Indeed, this ratio-based communication is a hallmark of many pheromone systems [18]. Female moths can produce an 'almost unlimited' number of these chemical blends by altering the variety and order of enzymatic activity through *de novo* biosynthesis from fatty acid precursors [18]. In fact, 1572 species of moths [19] have evolved over 2931 combinations from only 377 chemical compounds [20] by modulating the concentration ratios of these constituents within the blend [21]. Male moths correspondingly possess component-specific receptor proteins in antennal sensory neurons [22] whose signals are processed into a unified, ratio-based signal [23] within the antennal lobe (AL) of the brain [24].

An artificial system based on moth pheromone communication would offer a flexible, scalable, and modularized process of controlled synthesis, release, detection and processing to effectively solve the challenges of multimolecular communication. Chemical mixtures released in different ratios allow us to consider a new approach to chemical signaling in which an individual molecular compound has context in time and space with others, hereafter referred to as a 'ratiometric' signal. It is important to note that natural airborne odour mixtures are transported by advection in turbulent plumes [25, 26]. This means that mixtures are transported in turbulent eddies within the airstream, and individual mixture filaments can remain relatively compact and undiluted for several metres from the odour source [26]. Signal transport is therefore not limited by chemical and physical properties of the mixture components, such as would occur through diffusion over far longer timescales. As such, infochemical choice can be made relatively independent of molecular identity. In addition, infochemicals derived from common precursors, such as those discussed here, will often share similar diffusion and solvation properties. Approached in this way, unique ratio combinations of infochemicals can provide specific meanings in much the same way that, for example, letters in specific combinations and sequences create words and sentences. By making an appropriate selection of the total set of ligands deployed in the environment, such a strategy offers the potential for concentration-invariant information transfer and exponential information growth (figure 1). Note that concentration-invariance does not suggest concentration-independence. In fact, ratiometric coding requires that precise relative concentrations of infochemicals be controlled. However, as long as the relative ratios of the individual components are maintained, the information transfer will remain the same regardless of overall signal strength. As figure 1 shows, this phenomenon will also be preserved regardless of the number of signal components.

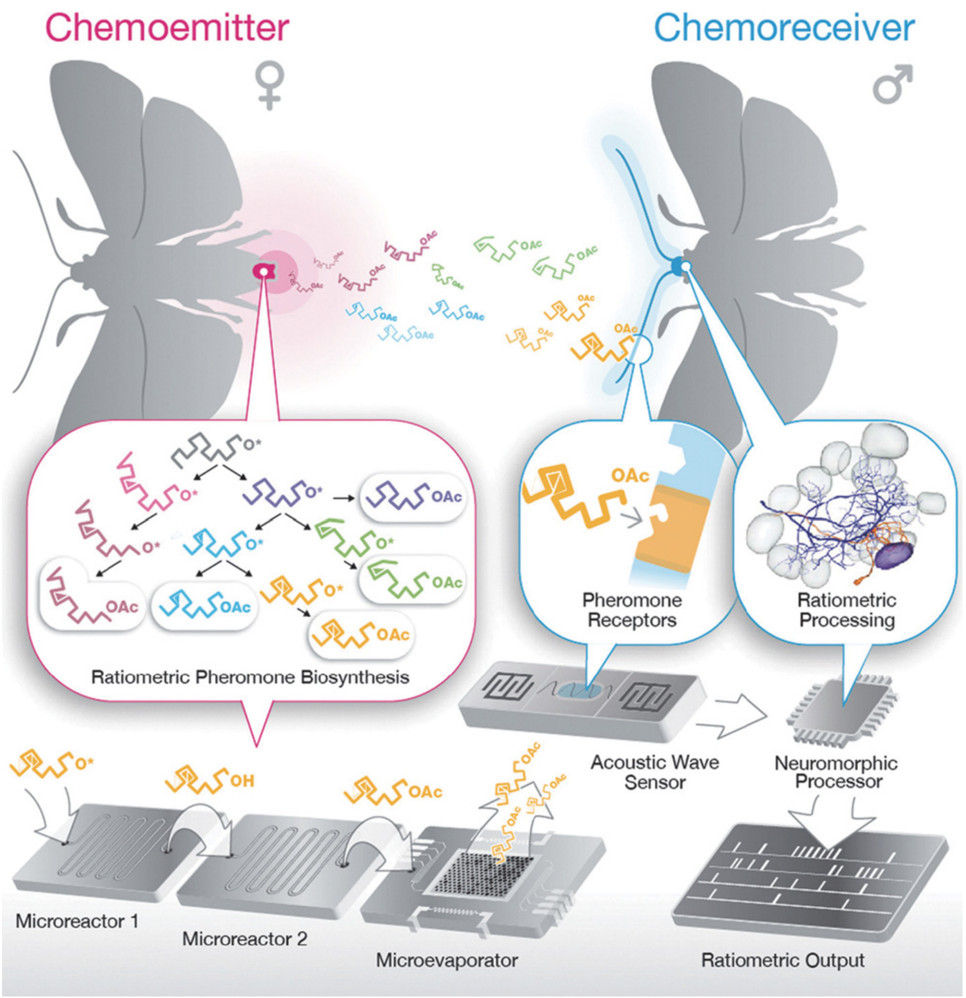

Figure 2. Ratiometric infochemical communication, conceptual overview. (Top) Ratiometric blends are synthesized within the female moth pheromone gland from fatty acid precursors and released into the air. The male moth detects the blend using highly specific protein receptors in antennal sensory neurons and processes the ratiometric signal within the antennal lobe of the brain (neural reconstruction originally published in [66] and reproduced with permission). (Bottom) Our technological concept includes the fabrication of a 'chemoemitter', a microsystem capable of producing and releasing a precise mix of biosynthetic compounds using combined microreactor and evaporator components, and a 'chemoreceiver', a surface acoustic wave sensor system capable of detecting the biosynthetic ligands and decoding ratiometric information using a neuromorphic processor.

In this perspective, we offer a stepwise, biologically-based approach [27] towards a moth-based infochemical communication system, translating the moth pheromone pathway into separate technologies for biosynthesis, transmission, receptor protein binding/transduction, and AL signaling (figure 2). We summarize our current progress in implementing each step and explore how these steps may be combined to create a prototype system as proof of principle of biomimetic multimolecular infochemical signaling. For each step, we also discuss its value, current limitations, and future challenges for the development of infochemical communications technologies. This new approach requires a multidisciplinary convergence of the latest advances in chemistry, biochemistry, ecology, materials science, neuroscience, ICT, and microsystems engineering to connect biology and technology through a common 'chemical language' for innumerable medical, environmental, and manufacturing applications.

3. Signal generation, encoding and emission: biosynthesis and airborne transmission

An efficient infochemical messaging system requires an encoder capable of both molecular production on demand, and programmable delivery of precise molecular information. This system must also be able to dynamically alter the timing and combinations of chemical information to deliver different molecular messages rapidly in space and time in a programmable fashion. The generated signal must ultimately be trafficked and terminated in the airborne environment.

Using the moth pheromone system as a template, the chosen biological model for our prototype technologies was the Egyptian cotton leafworm moth *Spodoptera littoralis*, a species with well-known pheromone biosynthesis and detection pathways (see [28–31] for recent studies). Starting from a single

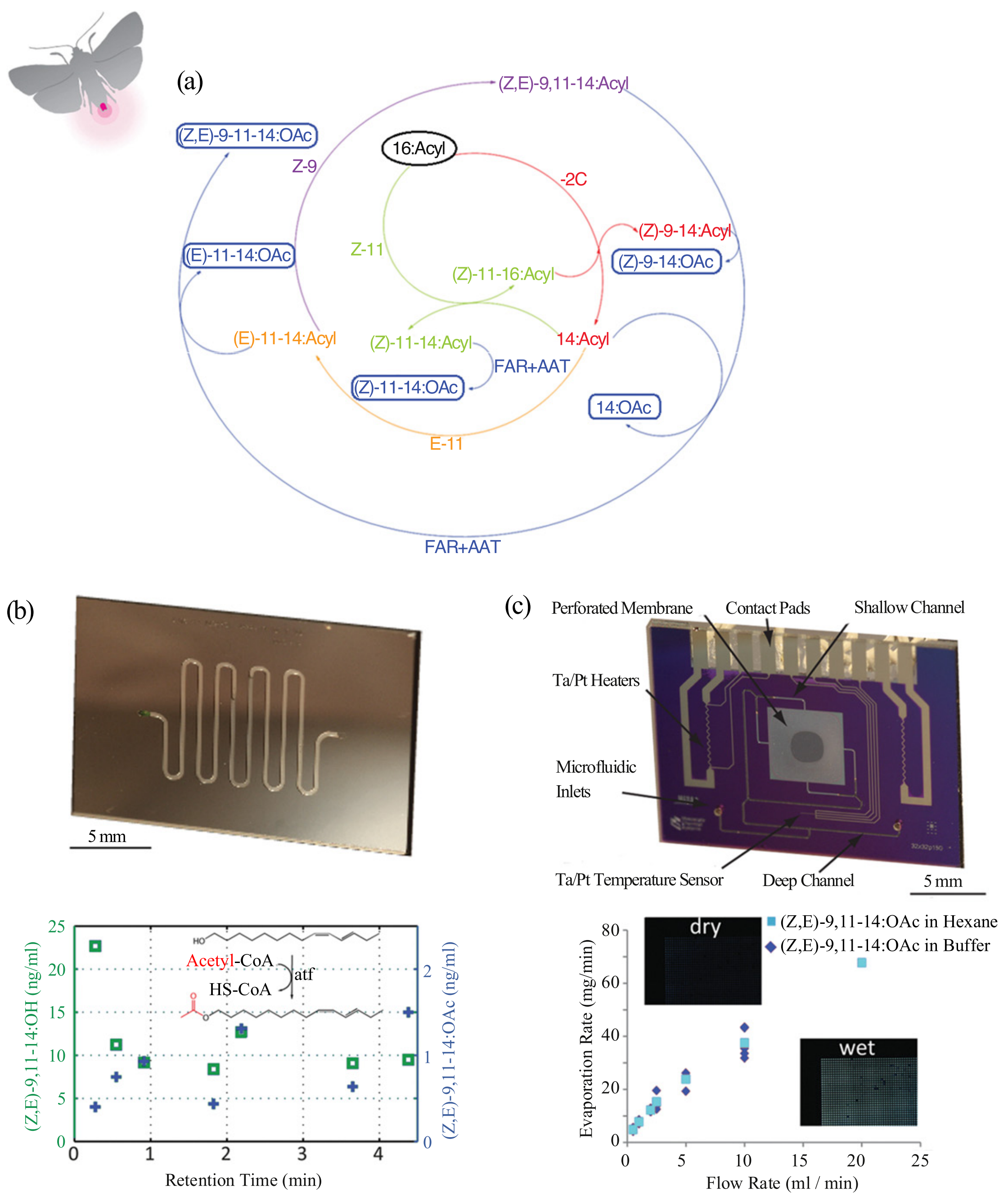

Figure 3. Infochemical synthesis and release technologies. (a) Biosynthetic pathway of *Spodoptera littoralis* main sex pheromone components. Arrow colours represent different biosynthetic steps with enzymes as follows: chain shortening enzyme (−2C), desaturase enzymes (E-11, Z-11 and Z-9 depending on the enzyme stereospecificity), fatty acyl reductase (FAR) and acetyl transferase (*atf*). (b) Photograph of simple silicon–glass based microreactor. Graph adapted with permission from [39]. (c) Photograph of silicon–glass based microevaporator using an integrated electrical heater.

precursor, *S. littoralis* females are able to produce a large variety of compounds by varying the sequence of enzymatic activity [31]. Through a combination of desaturases, reductases and acetyl transferases (figure 3(a)), *S. littoralis* females produce several unsaturated 14-carbon acetates in specific ratios from the same fatty acid pathway [31]. In moths, desaturase enzymes such as $\Delta 5$, $\Delta 9$, $\Delta 10$, $\Delta 11$, and $\Delta 14$ introduce double bonds at different carbon positions in the fully saturated fatty acid chain. Chain shortening enzymes, which have yet to be characterised at the enzymatic level in insects, reduce the carbon chain length. Finally, the carbonyl carbon of the chain is modified to form a functional group, firstly through reduction of the fatty acid to the corresponding alcohol via reductases. Then, an acetyl transferase, an alcohol oxidase/dehydrogenase and/or an acetate esterase may induce the formation of specific oxygenated functional groups. In addition, specific aldehydes are decarbonylated to form hydrocarbons, which can be used to produce epoxide pheromones. As a consequence, the interplay of this small set of biochemical reactions in different sequences can potentially create a library with an enormous diversity of signaling molecules (figure 3(a)).

By mimicking this modular enzymatic process, it should be possible to synthesize a very wide variety of infochemical compounds (figure 2; bottom). To explore the viability of a microtechnology for controlled, on-demand biosynthetic signal generation, we implemented a single step of this biosynthesis pathway, a functional group modification, by utilizing immobilized biosynthetic enzymes within a microreactor. Enzyme immobilization has been studied extensively over the last decade [32] and several immobilization strategies have also been applied in microfluidic systems [33]. In particular, a His_6 tag that reversibly binds to Ni^{2+} chelated in nitrilotriacetic acid is a popular genetically encoded affinity tag [34]; well-known in the field of immobilized metal affinity chromatography for reversible capture and purification of proteins [35]. This immobilization approach was also previously applied to microfluidic and sensing systems [36, 37]. Thus, this method offers a promising approach for on-demand molecular signaling via artificial infochemical biosynthesis.

Our prototype technology (see [38, 39] for detailed methodology) focused on the conversion of (*Z*,*E*)-9,11-tetradecadien-1-ol (*Z*9,*E*11-14:OH) into the main pheromone component (*Z*,*E*)-9,11-tetradecadienyl acetate (*Z*9,*E*11-14:OAc) through a diacylglycerol acyl transferase (*atf*, wax ester synthase) from *Acinetobacter sp.* [38]. MEMS-based microreactors (figure 3(b)) were fabricated with established lithographic methods [40] followed by deep reactive ion etching and anodic wafer bonding. To establish controlled component ratios, partial transformation of the dienic alcohol into the corresponding acetate was achieved by regulating the flow inside the microreactor with immobilized *atf* [38], thus demonstrating the feasibility of the approach for generating temporally precise multicomponent signals. New ratiometric signals could potentially be created by altering the components and flow rates of the system, and several reactors could theoretically be placed in series or parallel to replicate the multistep synthesis of the biological system. (figure 2, bottom).

Following signal generation, creating a programmable and reproducible technology that can propagate spatially and temporally precise signals into a moving airflow is itself a significant challenge. Female moths are thought to release controlled quantities of the pheromone blend as small droplets through fine pores in the pheromone gland cuticle [41]. Inspired by biology, we created a micromachined evaporator capable of a controlled release of the pheromone into the air. A silicon–glass prototype evaporator (‘artificial gland’) was fabricated consisting of a silicon membrane surface perforated with ~40 000 micromachined vias or ‘pores’ (figure 3(c)) [39]. Rectangular microfluidic channels delivered the eluent of the microreactor from two inlets to the reservoir located under the membrane. Fast and accurate temperature regulation through integrated thin-film heaters allowed for timely and precise evaporation of different compounds over a broad range of flow rates (see [42] for detailed information). The microevaporator was also evaluated in behavioural assays, and the released pheromone exhibited an attractant effect similar to that elicited by three virgin females, showing that the artificial chemoemitter was an effective mimic for the biological system [39]. As a result, our ‘artificial gland’ achieved a reproducible, airborne infochemical signal regulated in space and time. We also demonstrated the effectiveness of the microevaporator for controlled pheromone delivery with another moth, the silkmoth, *Bombyx mori* [43].

A major challenge in establishing these technologies was the development of a functional enzyme microreactor that produced controlled infochemical products while reducing the adsorption and subsequent loss of both enzyme substrate and product on the surface of the microreactor. This problem has not always been properly addressed in recent literature on enzyme microreactors. In fact, the development of suitable microreactor coatings for these relatively unstable and complex biosynthetic enzymes is a significant challenge. The development of multiple microbioreactors (figure 2) with suitable coatings for all desaturases, reductases, and acetyl transferases used in the biological system remains a significant obstacle for successful infochemical communication. However, the controlled chemical production and release established by such biomimetic technologies provides an efficient template for the development of such multicomponent systems in the future.

4. Signal detection and termination: receptor protein binding/transduction

A key to the strength and versatility of an infochemical system is its ability to deliver and detect a wide variety of infochemical messages. This is accomplished in the infochemical encoder through programmable delivery and emission of a virtually limitless variety of chemical inputs (see above). On the infochemical decoder side, however, we require a sensor system both specific enough to detect molecular levels of chemicals as well as broad enough to detect a wide variety of different signals. Although several sensitive and specific chemical sensor systems, such as electronic nose technology are currently available [44–46], such systems fail to accurately discriminate multimolecular chemical signals in some real-world tasks [47, 48], and e-nose systems that discriminate ratiometric chemical signals have not yet been widely explored.

One approach to overcoming these limitations is through the creation of biosensor technologies that directly incorporate insect ORs themselves. Insects have efficiently solved the apparent tradeoff between sensor sensitivity and selectivity through a system utilizing sensitive, yet broadly-tuned ORs with non-overlapping response profiles (see [49, 50]). Male moths also possess distinct protein receptors specific to each pheromone component to detect the ratiometric signal (figure 2 top right). There are at least three principles used in biological olfactory systems to ensure efficient termination of the chemosensory signals. Firstly, rather than using small numbers of receptors with high binding affinity to different ligands, olfactory systems instead deploy large populations of diverse ORs that generally have medium to low binding affinities to a wide array of ligands. In general, low-affinity binding involves a shorter residence time for the ligand, allowing for a faster termination of the olfactory signal. Therefore OR based detection strategies inherit this advantage. Secondly, transduction pathways and intracellular processes in olfactory sensory neurons (OSNs) include adaptation processes that terminate signalling for specific ligands to constant chemical input. For this reason, adaptation processes in OSNs improve signal termination performance. Finally, both insects and mammals may use odour degrading enzymes to inactivate the molecular signal [51]. These enzymes could also be employed in future infochemical communication systems should the first two strategies provide insufficient signal termination.

The use of biological ORs in chemical sensing technology is gaining popularity in recent years because they are, in general, orders of magnitude more sensitive than the most advanced physical approaches such as chemical ‘noses’ or gas-chromatography/mass spectrometry techniques [52]. Biosensors incorporating heterologously expressed ORs are also generally more sensitive and specific than ‘e-noses’, and engender much more practicality than using the organism itself as the detector [52]. A variety of transduction technologies are currently being pursued for ‘olfactory biosensing’, including optical detection with surface plasmon resonance, fluorescence and luminescence, resonance technologies such as bulk and surface acoustic wave (BAW/SAW) resonators or mechanical cantilevers, and electrochemical methods that assess conductance, resistance, current, or pH such as microelectrodes and potentiometric sensors [52]. Several of these methods have already been achieved, including the use of bioluminescence resonance (BRET) [53] or SAW [54] with *C. elegans* worm receptors, carbon nanotube transistors for mouse receptors [55], and microelectrode voltage clamp [56] and SAW [57, 58] for insect receptors (see [52] for recent review of all current techniques). However, olfactory biosensor development has been hampered by the lack of a small, portable transduction system with high sensitivity and selectivity and minimal sample preparation (e.g. labeling) [52]. Thus, most transduction systems still utilize relatively complex technologies that are laboratory-based [52]. Resonant sensors based on acoustic waves (such as SAW) can offer sensitivity, selectivity, portability and biocompatibility with heterologously-expressed insect receptors [57, 58].

To demonstrate the feasibility of OR-based signalling for infochemical communication, we combined SAW-cell-based resonant sensors with pheromone receptors as a specific detection solution (see [27, 57–59] for details). Here the *Drosophila melanogaster* pheromone receptor Or67d, which specifically responds to the pheromone cis-11-vaccenyl acetate (*Z*11-18:OAc) [60], was heterologously expressed in Sf9 (*Spodoptera frugiperda*) cells and coupled to shear horizontal mode dual surface acoustic wave (SH-SAW) resonator-based microsensors (figures 4(a) and (b)). Sf9 cells expressing the Or67d protein were coated onto the surface of the sensing side of a dual 60 MHz SH-SAW resonant sensor. The other side of the dual sensor contained non-transfected cells and provided a reference signal. A reliable and reproducible ligand–biosensor interaction was enabled using an automated microfluidic system. The differential frequency response of ca. 600 Hz–100 μM 11-cis-vaccenyl acetate (figure 4(b)) indicated that receptor-ligand binding events could be accurately detected, therefore establishing an effective olfactory biosensor using highly-specialized pheromone receptor proteins.

The genetic basis of pheromone reception in our model species *S. littoralis* is a subject of current study [28], and deorphanization of these receptors is still underway. For this reason, we were required to choose a *Drosophila melanogaster* receptor, which remains one of the only insect species with an elucidated chemosensory repertoire. In addition, the topology and signal transduction of insect receptors is a current subject of study and controversy [61–63], which can limit their expression in heterologous systems. As such, although our technology exhibited effective chemical sensing capabilities, we must increase our knowledge of insect chemoreceptors to efficiently replicate the broad infochemical detection capabilities of the insect antenna.

5. Signal decoding: AL processing

At the final stage of information transmission, our complex multimolecular message must be decoded into a unified signal. In the olfactory processing centers of animals, a series of separate sensory input channels, corresponding to individual receptor/sensor signals, are translated into a single’ odour object’ [64] in the same way object percepts are generated from a visual scene. Meeting this challenge requires appropriate sensor fusion of multichannel input capable of robust processing of corrupted data, scale/position invariance across multiple sensors, and noise

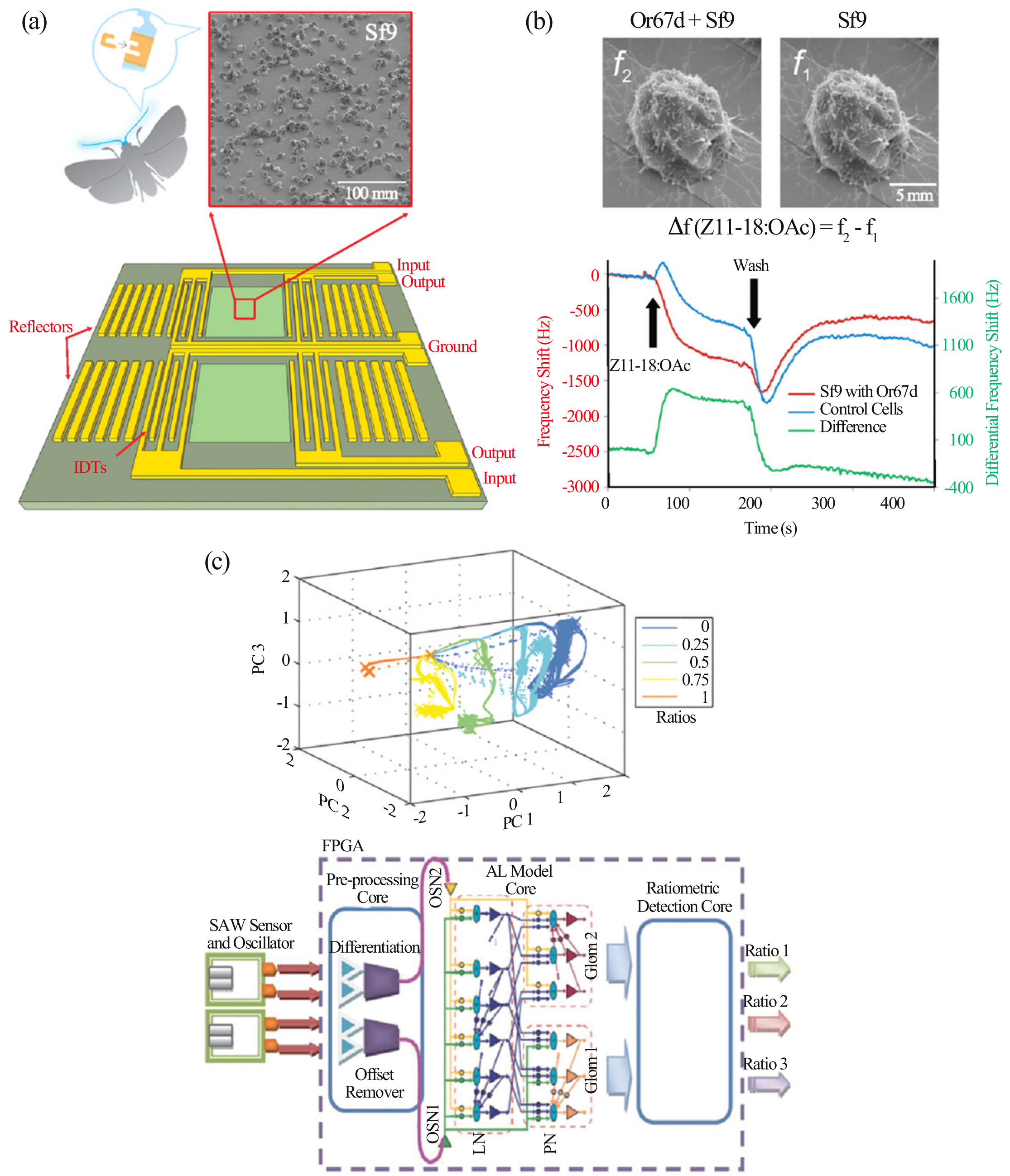

Figure 4. Infochemical detection and processing technologies. (a) Sf9 cells (top) are deposited over the sensing areas of SAW resonator sensors made from thin gold interdigitated (IDT) electrodes and reflectors on a lithium tantalate piezoelectric substrate (bottom). (b) Cells attach themselves to both gold IDT and bare piezoelectric substrate (top) with piezoelectric evanescent wave penetration depth approximating cell diameter. The simultaneous response of the SAW sensors (bottom) to 100 μM of pheromone (Z11-18:OAc) is shown for transfected (red) and control Sf9 cells (blue). Differential measurement (green), $\Delta f(Z11\text{-}18{:}OAc)$, removes non-specific sensor responses to, e.g. pH and temperature. (c) (Top) Projection neuron (PN) population trajectories from the neuromorphic antennal lobe model behave according to a limit cycle attractor that varies smoothly as a function of input ratio. (Bottom) Implementation of the neuromorphic model in FPGA and coupling to SAW sensors. (c) Originally published in [74] and reproduced with permission.

suppression, among others. Biology exploits local nonlinear processing during the early stages of the olfactory pathway (e.g. [65–67]) and highly evolved parallel architectures to sequentially reduce the dimensionality of sensory information for blend and component information extraction. The massively dimensional sensor fusion problem is rendered tractable in insects through parallel subsystems that give rise to increasingly sparse representations at progressive levels within the nervous system, allowing processing of both different sensory stimuli and different features of those stimuli [24, 68].

In insects, separate receptor neuron signals are unified in the moth brain at the first synapse of the insect olfactory system, the AL [23]. Using a combination of neurophysiological experiments [66, 69] and computational modelling [67, 70], we found that the ratiometric signal transmitted by the AL is a unique entity predominantly driven by interneuron connectivity through local interneurons [66, 67, 69]. Projection neurons (PNs) then relay this composite signal to higher brain centres. PNs relaying pheromonal information have also been shown to be maximally responsive to pheromone blends when their components are presented in behaviourally active ratios [71].

As a strategy for rapidly decoding ratiometric infochemical signals in real-time, we explored the use of a dynamical neuronal model constructed with a connectivity pattern from the male moth pheromone processing region, the macroglomerular complex (MGC) [70]. After describing the MGC network as a set of coupled first-order ordinary differential equations, we found that the resulting network dynamics encode precise ratios of activity across OR input channels through a set of unique spatiotemporal trajectories. This computational model was then implemented in programmable logic (field programmable gate arrays) using the standard leaky integrate-and-fire spiking neuron model formalism (see [72, 73] for details) as a real-time neuromorphic processor for ratiometric infochemical information (figure 4(c)). Programmable logic hardware is an ideal technology to fulfil these requirements. These devices are fast due to fine grained parallelism. They can also be clocked at a wide variety of clock speeds to match the dynamics of chemical communication in real-world environments, can be reprogrammed in milliseconds, and offer vast numbers of individual circuit elements that are inherently parallel in nature and may be configured arbitrarily. By driving this processor with chemosensory input in real-time, we have shown how this neuromorphic processor can extract ratiometric information from a blend faster and with more accuracy than linear machine learning methods applied to the same data [73]. Our neuromorphic solution can thus provide an effective strategy for recovering infochemical blend information.

6. System integration and future directions: ratiometric infochemical communication

As a final proof-of-concept of our overall infochemical communication approach, a series of wind tunnel experiments were conducted using two mobile robots (chemoemitter and chemoreceiver robots) to assess airborne infochemical communication with binary blends in different ratios transported through naturally turbulent plumes (movie S1). The necessary infochemical emission, detection and processing hardware based on our developed technologies was integrated on the robot platforms (figure 5(a)). A controlled series of pulses of binary blends were injected into the turbulent flow by the chemoemitter robot and the corresponding ratios independently recovered in the chemoreceiver by classifying PN population trajectories produced by the neuromorphic model running in real-time as a programmable logic core (figure 5(b)). Specific motor commands were then executed on the chemoreceiver robot depending on these classified ratios (see [74] for details), demonstrating how ratiometric chemical messages can be communicated between transmitter and receiver and subsequently translated into robust behavioural responses under turbulent, real-world conditions. Movie (S1) shows that distinct component ratios presented by a chemoemitter robot could be reliably decoded in real time to execute corresponding preprogrammed behavioural responses in the chemoreceiver robot using its neuromorphic processor, demonstrating successful infochemical communication through the decoding of specific ratiometric messages.

Here, we have explored the development of a set of technologies based on the entire pheromone signalling pathway of moths to address each of the major challenges to infochemical communication (figure 6): infochemical production (biosynthetic pheromone microreactor), release (‘artificial gland’ microevaporator), detection (OR-based biosensor) and processing (neuromorphic AL processor). We have also demonstrated our insect-based approach for infochemical communication under turbulent, real-world conditions using a coupled robotic emitter and receiver. These developments are notwithstanding several obstacles that must be overcome to achieve a fully-integrated technology for deployment in a variety of contexts and spatial scales. In addition to the specific scientific and technological hurdles we have considered above for each component, the most significant challenge remains the precise sequencing of encoding, delivery, propagation, rapid detection, synchronization and real-time decoding of diverse sets of molecular types to achieve multi-channel broadband infochemical messaging.

Existing approaches to messaging, interfacing or communicating with molecules have been largely theoretical, monomolecular, and do not consider this precise sequencing of communication events, that points to future technologies operating at the molecular scale. To address this issue, we must provide a system where molecules are actively transported over very brief time periods. Biological factors inherently function at the molecular scale, and can complement nano-engineered components to overcome temporal limitations of biologically-based transport systems in certain contexts, such as e.g. biomolecular-motor-based molecular propagation systems [4], offering precise, low emission, high bandwidth release of infochemical messages into both air and liquid media. On the receiver side, most current chemical sensors rely upon a passive diffusion process through a sensitive layer. Sensor response time consequently depends upon the physical dimensions of the device, in particular layer thickness. At reduced spatial scales, future nanoengineered chemosensors offer the possibility for chemical detection that is less diffusion limited and thus can dramatically improve response time and

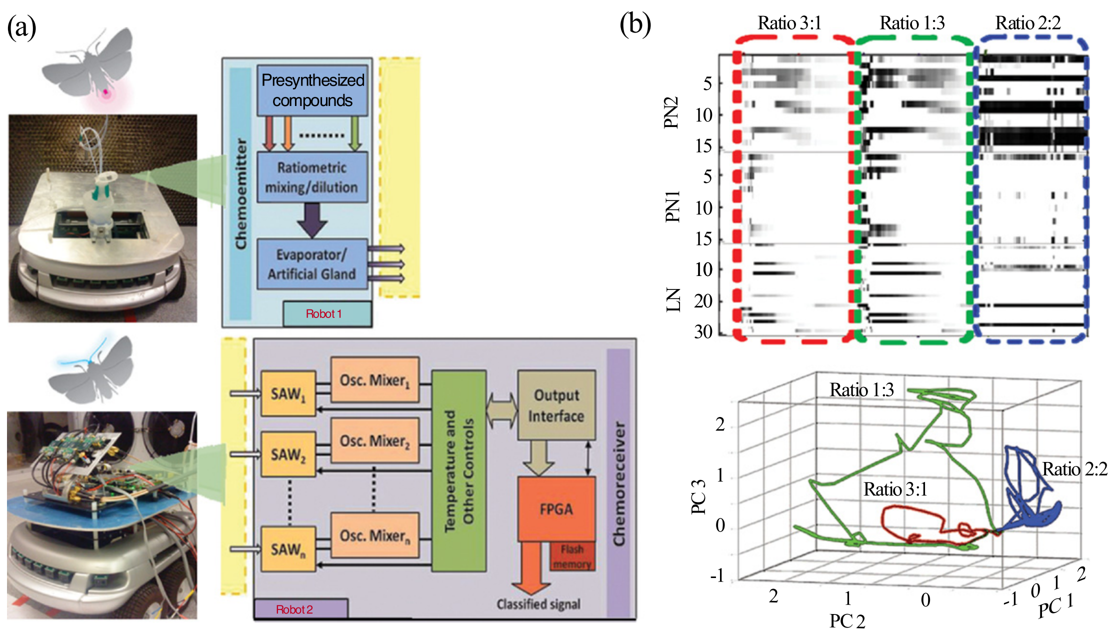

Figure 5. Ratiometric infochemical communication. (a) Implementation of chemoemitter (female) and chemoreceiver (male) robots for communication experiments. (b) Model PN population trajectory to a series of controlled chemical ratios in naturalistic plumes showing recovered ratios in the chemoreceiver (male) robot platform during infochemical communication. Figures originally published in [74] and reproduced with permission.

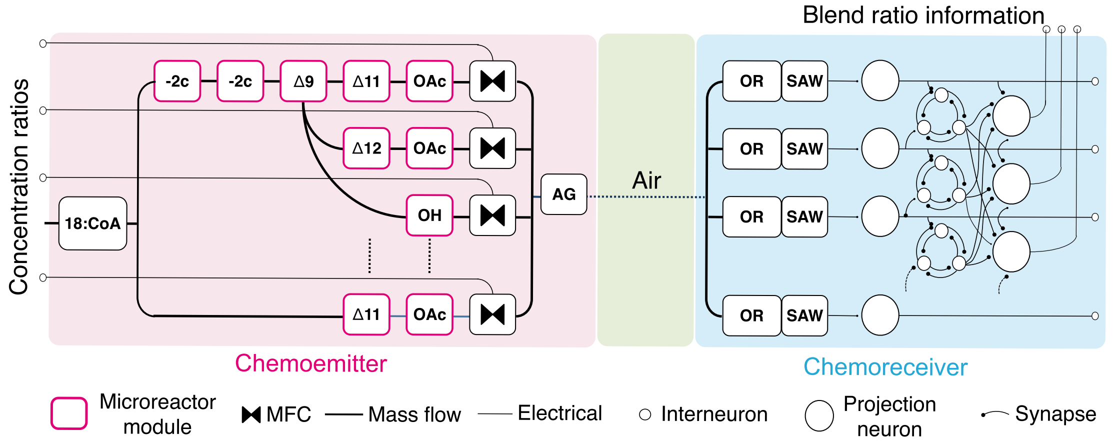

Figure 6. Theoretical infochemical system schematic. The chemoemitter interchanges various biosynthetic modules to produce blend components from a fatty acid precursor (18:CoA) based upon known enzymatic activity within the moth (figure 2; e.g. chain shortening (−2c), desaturase (Δn), and functional group modification: alcohol (OH), aldehyde (Ald), hydrocarbon (Hc), or acetate ester (OAc)). A microevaporator (AG) releases the blend into the air. The chemoreceiver exploits heterologously expressed pheromone receptors (OR) that are transduced using sound acoustic wave sensors (SAW). The ratiometric signal is then processed using a neuronal model based on the moth antennal lobe.

sensitivity. More generally, reducing spatial scales within nano-engineered systems can improve temporal precision at each step in the messaging process. By addressing temporal precision, we can effectively overcome this significant obstacle for accurate sequencing of messaging events to achieve relatively low latency and high bandwidth infochemical communication.

Biomimetic approaches can revolutionize messaging by harnessing the biological factors themselves [3]. Biological components consume extremely low power compared to electronic-based networks—a single molecular reaction consumes 10 000 times less energy than a micro-electronic transistor (see references in [3]). Tapping into and harnessing these biological factors will increase our ability to interact, exploit and control biological processes and integrate these into a range of existing technologies. Such biomimetic information and communications technologies can unlock new opportunities for broadband chemical communication at multiple scales ranging from environmental monitoring to intracellular therapy to nanoscale communication using multi-molecular messengers.

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