DNA-BASED MOLECULAR ECOSYSTEM ON A CHIP

Adrien Padirac*, André Estévez-Torres, Teruo Fujii*, Yannick Rondelez***
** LIMMS/CNRS-IIS, Institute of Industrial Science, The University of Tokyo, Japan*
*** Laboratoire de Photonique et de Nanostructures, CNRS, France*

ABSTRACT
In nature, Predator-Prey (PP) systems are extremely hard to observe because of their large scale in both time and space [1]; we propose here a micro-ecosystem, based on biochemical reactions, that is able to emulate a two-dimensional *in vitro* PP ecosystem under a microscope. This molecular *in vitro* implementation accurately reproduces the dynamics of PP ecosystems, and is greatly scaled-down in time and space. Moreover, it experimentally shows that by controlling both reaction and diffusion in a microfluidic set-up, it is possible to trigger the emergence of complex spatial patterns.

KEYWORDS
Reaction-Diffusion, Predator-Prey, Ecosystem, Reaction Networks.

A DNA-BASED IMPLEMENTATION OF PREDATOR-PREY RELATIONSHIP
Predator-Prey (PP) networks have been mostly studied for their ecological relevance. Following the century-old PP equations [2], PP networks are known to be able to display complex behaviors –including oscillations– under a large range of conditions. They, however, have never found any chemical *in vitro* implementation. PP systems can be defined in terms of a set of reactions: preys autocatalytic reproduction, predation of preys by predators, and decay of both species. By using a set of simple DNA-based enzymatic reaction [3], we have developed an *in vitro* system that encodes PP relationship [4]: predator (fox) and prey (rabbit) are both DNA species, and their DNA sequences define their trophic relationship. Experimentally, DNA-foxes and DNA-rabbits interact in a closed environment: in the presence of grass DNA sequence, rabbits autocatalytically reproduce (Figure 1-A). Grass sequence is labeled in 3'-end with a fluorescent dye, which allows –through nucleobase quenching [5]– to monitor the rabbits as they hybridize to the grass. A fox finding a rabbit produces two foxes (Figure 1-B); both foxes and rabbits die (Figure 1-C), whereas the amount of grass stays constant across generations. In 0-dimensional (well mixed) conditions, this system is an accurate chemical *in vitro* model of Lotka-Volterra PP equations [2], and displays sustained oscillations of fox and rabbit populations (Figure 1-D).

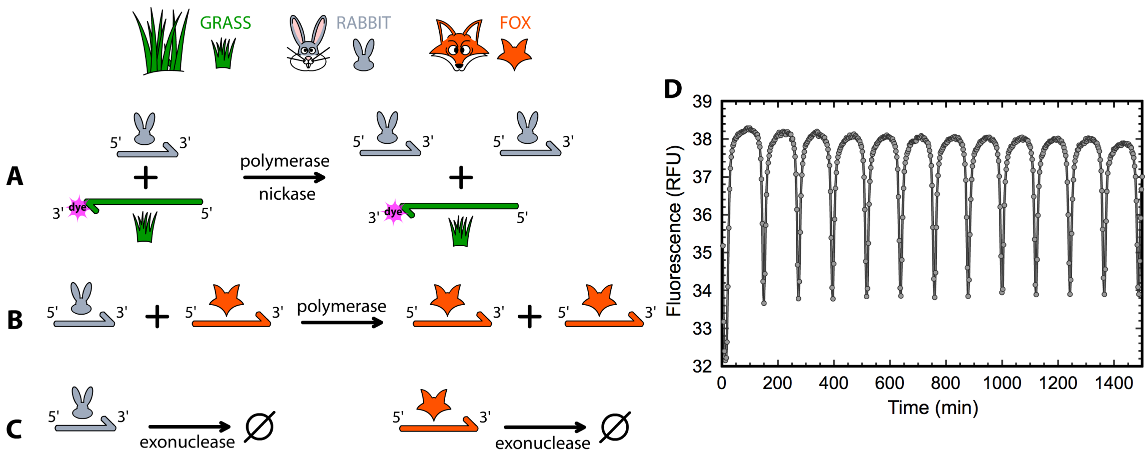

Figure 1. DNA-based Predator-Prey system. (A) In presence of the grass DNA strand, rabbits autocatalytic reproduce. (B) Predation of rabbits by foxes. (C) Death of both rabbits and foxes. (D) Experimental curve in 0-dimensional milieu (test tube) using N-quenching [5], showing oscillations of rabbit population: fluorescence decreases as concentration of rabbit increases.

ECOSYSTEM ON A CHIP
In the natural world, spatial effects are believed to be responsible for a large part of the dynamic complexity observed among animal populations [6]. It was shown that, for instance, different spreading rates for prey and predator potentially lead to complex spatial patterning [6]. Therefore, we implement our DNA-based PP system in a two-dimensional environment, where both foxes and rabbits can freely diffuse.

The molecular PP ecosystem is set in a thin (~250µm) chamber filled with a few microliters of reacting mix and enclosed between two glass slides. The chamber is first shaped in Parafilm® M, then sandwiched between two glass slides and baked a few minutes at 55°C. We follow the reaction with a fluorescent microscope, using a purposely-developed quencher-free reporter of rabbit concentration [5]. From initially localized populations of rabbits and foxes, rabbit and fox populations evolve in the pseudo two-dimensional landscape. As an example, traveling waves of rabbits colliding with the walls of the ecosystem or with other waves can be seen on Figure 2: rabbits reproduce (darker area), foxes increase as they hunt and eat rabbits, rabbits migrate (traveling wave) but eventually get surrounded by foxes (wave extinction). As foxes cannot find rabbits anymore, they starve, and rabbits recover, starting the cycle again. These complex spatial behaviors are supported by simulations that predict emergence of pattern –such as travelling and colliding waves– for a locally perturbed two-dimensional PP ecosystem (Figure 3).

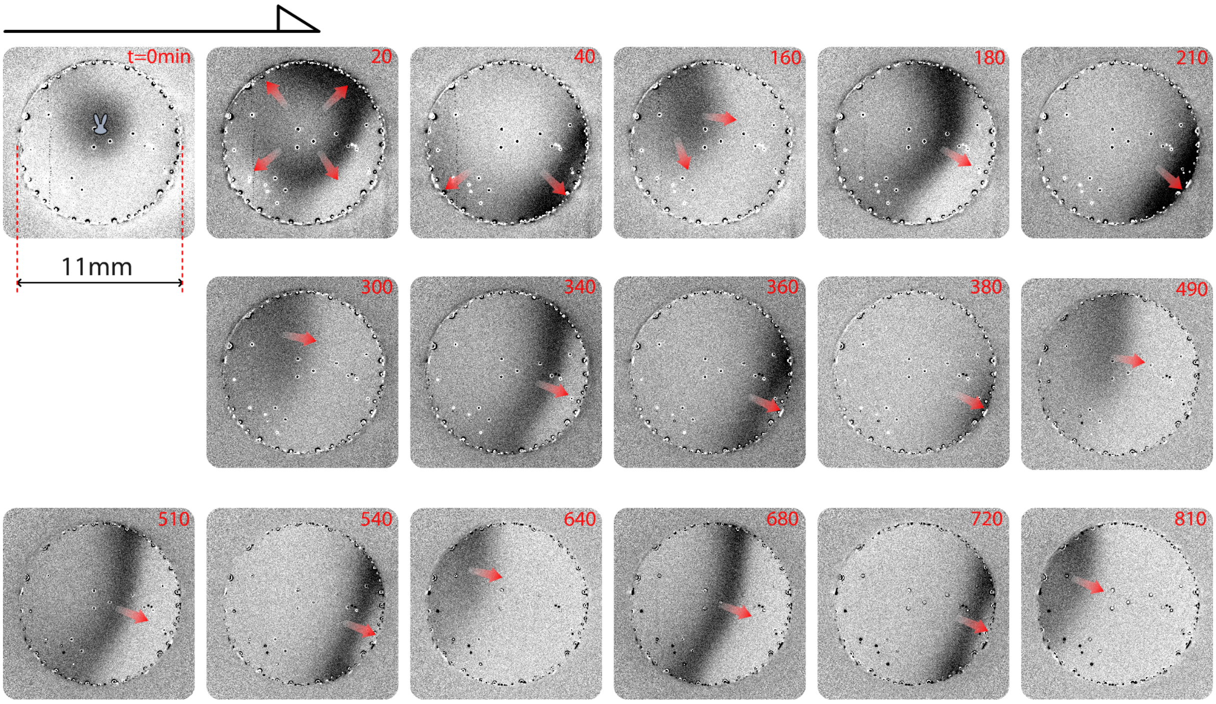

Figure 2. Time-evolution of a two-dimensional PP ecosystem starting with a localized population of rabbits: dark areas are areas of high rabbit concentration monitored with N-quenching [5]. Red arrows show the waves movement.

This micro-ecosystem enables easy two-dimensional *in vitro* study of PP relationship and its extensive range of dynamics arising, for instance, from environmental perturbations. It can be used to explore the landscape-dynamics relationships for complex PP ecosystems, by implementing different environment topology (micro-ecosystem shape), or localizing some of the resources at particular positions of the microchip (patchy systems).
Our report also opens the rational programming of emergent patterns in a microfluidic environment, with possible applications for on-chip computation and self-organizing devices.

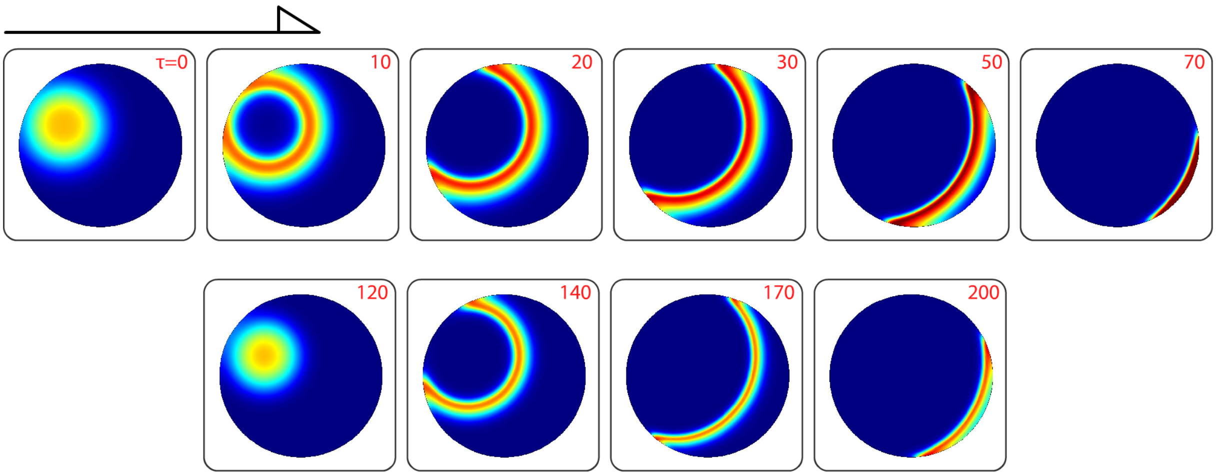

Figure 3. Finite-element simulation (non-dimensional model) showing the rabbit population evolving in a two-dimensional round arena.

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CONTACT

Yannick Rondelez 81-3-5452-6213 or rondelez@iis.u-tokyo.ac.jp