Experiment 24	Swarming and gliding microorganisms
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Reading	Chapters in BBOM 9th: 3.11 p. 79, 13.10 p. 481-482, 13.16 p. 495-499 and 13.24 p. 526 Chapters in BBOM 10 th : 4.10-4.11, p. 82-87, 12.11, p 379, 12.17, p. 393-396, 12.25, p. 422-423 BBOM : Madigan M.T., J.M. Martinko and J. Parker: "Brock - Biology of Microorganisms", 9 th Edition (1999) or 10 th Edition (2003), Prentice Hall.
Objectives	 We will enrich for and grow swarming and gliding microorganisms on solidified agar and study the growth patterns and the organisms by microscopy. We will analyze possible reasons for such phenomena with the aid of published reports. We will propose further experiments to investigate bacterial swarming and pattern formation
Background	 Bacteria have evolved a number of motility strategies in order to move to a new and better environment. The most common one is swimming, but other moving strategies such as twitching, swarming and gliding can also be observed. Here, we focus on swarming and gliding, two motility strategies on hard surfaces which can lead to pattern formation with certain organisms. Like swimming, swarming is performed by flagella. Many different species including gramnegative as well as gram-positive species are able to swarm. Swarming cells are often rather large and have numerous flagella, some express a filamentous growth form. They move together as a colony on a hard surface. Organized moving leads to pattern formation. Shape and morphology of the pattern is very variable, but under the same conditions different species may also exhibit similar patterns. Moreover, one species may change the type of pattern when transferred to a different environment.
	 In contrast to swarming, gliding is not dependent on flagella. For gliding, the hard surface must be covered with a liquid film. Gliders can be divided into fast and slow gliders. Some cyanobacteria may glide with the help of filamentous sheaths, true rotating flagella have never been found in gliding bacteria. Pattern formation is dependent on a variety of parameters such as diffusion, surface tension, signal transduction and cell-to-cell communication. Sometimes for pattern formation cooperation between autonomous cells is required. These microorganisms show a multicellular behaviour. Generic models for predicting pattern formation have been developed by Gutnick and Ben-Jacob.
	 Physiologically, swarming organisms are not distinct from other organisms. They are found almost everywhere. Even swimming species like <i>Escherichia coli</i> are able to differentiate into swarming cells. Little is known so far about the reasons for and the mechanisms of swarming and gliding. The abilities to swarm and glide are of great importance for the organisms involved, in infection mechanisms, in colonization of new habitats and in establishing new growth on surfaces (biofilm formation).
Literature	 The chapters of BBOM 9th mentioned above. Zopfi K., 1992. Schwärmende Mikroorganismen im Auseesediment, Semesterarbeit Mikrobielle Ökologie Gutnick D. L., Ben-Jacob E., 1999. Complex pattern formation and cooperative organization of bacterial colonies. Microbial Ecology and Infectious Disease, American Society of microorganisms, Washington D.C., Rosenberg E., ed. p. 284 - 299 Eisenbach M., 2001. Bacterial Chemotaxis. Encyclopedia of life sciences, Nature Publishing Group
Links	http://www-micro.msb.le.ac.uk/video/motility.html http://www.aip.org/mgr/png/2001/138.htm http://www.amsta.leeds.ac.uk/Euromech422
Practical work	The students prepare agar plates and grow microorganisms from sediment and compost samples under different growth conditions. They look for pattern forming species by microscopy and discuss possible reasons for swarming and gliding.

Material and Experimental Protocols	• For each sample pour an 1.7% agar plate with the following concentrations: 1x LB medium, 1xLB medium + 2x PPY, 1xLB medium + 12.5 mM sucrose, 1xLB medium + 25 mM sucrose.
	• Liquify the LB agar medium in the microwave (notice that bubbling can be omitted), add the ingredients (PPY, sucrose) from concentrated stock solutions and pour the liquid agar into the petri dishes (around 15 ml per dish). Remove bubbles with the flame of the Bunsen burner without melting the plastic petri dish. Let the agar solidify.
	• Plate the inocula on the agar as demonstrated by the instructors. Incubate at RT for seven days. Look out for swarming colonies. Observe the formed colonies and the organisms which form them under the microscope and take pictures.
	• Analyse your observations.
	• Discuss swarming hypotheses.
	Material:
	samples from sediment, compost, spring water, etc.
	1x LB agar medium
	dH ₂ O
	sterile tap water in capped test tubes
	PPY 20x (Potassium phosphate 100mM pH 7, peptone 20 g/l, yeast extract 20 g/l) autoclaved
	sucrose 250 mM in distilled water, autoclaved
	technical alcohol
	microscope + camera
	binocular
	microwave
	incubator
	plastic petri dishes
	bunsen burner
	test tubes, test tube rack
	glass container with lid (for alcohol)
	water quentch (45°C)
	tweezers
	spatula
	hockey stick spreader
	inoculation loop
	pipettor 10 ml, sterile, with tips
	household paper, kleenex, aluminium foil, parafilm, marker pen, gloves
Laboratory Rules & Precautions	Don't eat in the lab. Wash your hands before leaving the lab and work aseptically. Wear your lab coat and gloves while working in the lab.
Experiences gained	• How to prepare agar plates containing different concentrations of ingredients for enrichment of swarming bacteria.
	• How to grow organisms from a natural sample.
Timing	120 min
Reporting	Take photographs, report the growth under different nutrient concentrations and discuss the possible hypotheses with the aid of the literature.
Questions to be answered	 Which further experiments could be done to understand swarming phenomena ? What could be reasons for different growth and different organisms which emerge under different conditions ?