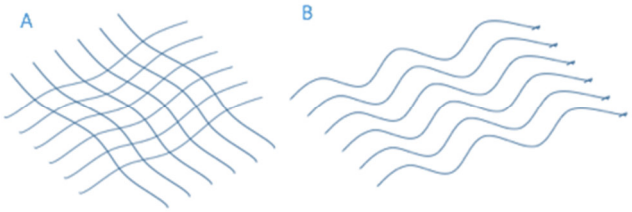
How Contact Angle Measurement Can Help to Understand Scar-free Wound Healing

The most common responses to injury in human tissues and organs are scarring and fibrosis. For the healing process the most favorable outcome would certainly be a complete regeneration, with new tissue retaining the same structural, aesthetic, and functional attributes as the original uninjured tissue. However, it is extremely difficult to achieve complete regeneration which as the ultimate goal it termed “scar-free wound healing” and normally only occurs in invertebrates and lower vertebrates. A scar-free healing requires a precise manipulation of cell migration and protein orientation in the extracellular matrix (ECM) remodeling. In scars collagen is arranged in parallel bundles of collagen fibers while healthy scar-free tissue has a “basket weave” structure (**Picture 1**).

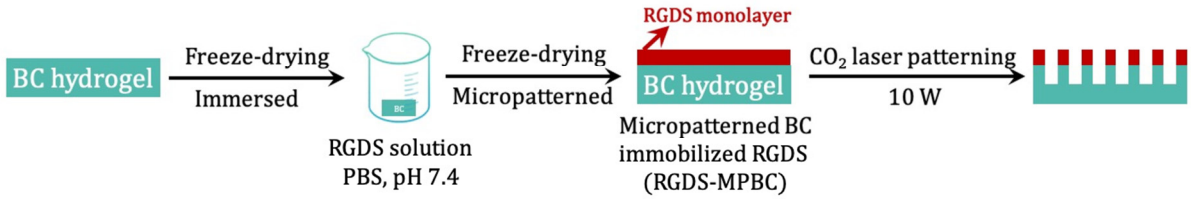


Picture 1: *(A) Collagen fibers in normal skin “basket weave”. (B) Parallel collagen fibers in scar tissue.*

A multitude of therapies have been introduced to treat and/or prevent scars on skin, but the efficiency of commercially available therapies remains limited. Therefore, more and more scientists make efforts in order to provide better solutions. To shed light on how to

directly manipulate the collagen distribution and cell migration, Hu et al. recently reported their research on a material based on spongy bacterial cellulose (BC) for scar-free wound healing.

In this research, the micropatterned BC overlaid with a tetrapeptide (RGDS-MPBC) was fabricated by two steps (**Picture 2**). Field emission scanning electron microscopy data showed that a smooth columns (100×100 µm) surrounded by groove channels (100–150 µm wide) were generated. Low energy CO_2 laser photolithography led to an excellent result, avoiding holes penetrating the BC hydrogel, when compared to the traditional photolithography and soft lithography with higher energy laser sources.



Picture 2: *Preparation process for micropatterned BC immobilized RGDS (RGDS-MPBC)*

A variety of previous studies have testified that surface micropatterns can enhance the surface hydrophobicity due to "lotus effect", which refers to surface roughness-induced ultrahydrophobicity. To verify this, water contact angle measurements were done with an OCA 20 contact angle analyzer from DataPhysics Instruments to study the surface properties of various materials based on BC (**Table 1**).

Table 1: *Characterization of Surface Wettability*

	BC	RGDS-BC	MPBC	RGDS-MPBC
Water contact angles (ϑ)	55.9 ± 2.1°	62.4 ± 8.3°	80.7 ± 1.1°	70.5 ± 0.1°

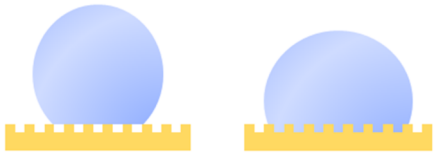
MPBC had the largest contact angle which meant the best hydrophobicity. The contact angle of RGDS-MPBC was decreased due to the hydrophilic nature of amino acid sequences when compared to that of MPBC. However, RGDS-MPBC still showed a better hydrophobicity than pristine BC and RGDS-BC without micropattern. This implies that a

crossed groove/column micropattern on the surface effectively enhanced the surface hydrophobicity.

Surface Roughness Induced Hydrophobicity

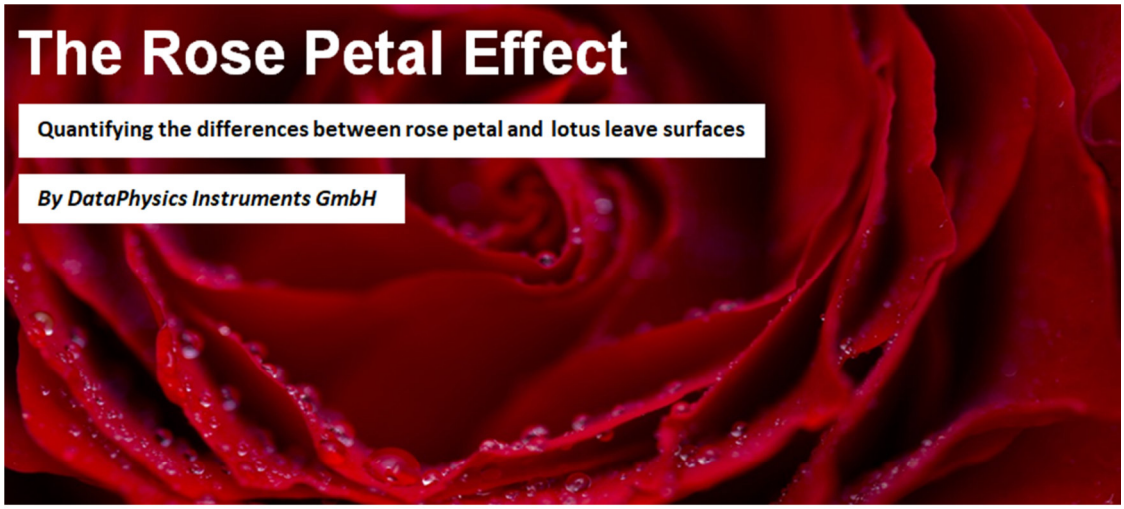
A micropattern on the surface can lead to an increased contact angle resulting in a gain of hydrophobicity. In order to simply elucidate the surface roughness and structured character of a surface the change in absolute contact angle value or the contact angle hysteresis are valuable indictors.

For microstructured/rough surfaces mainly two states exist: The Cassie-Baxter state where the drop floats on the peaks of the surface and does not wet the groves in between and the Wenzel state where the also the groves are wetted. This has a strong impact on the adhesive properties of the surface as introduced in our article “The Rose Petal Effect”.



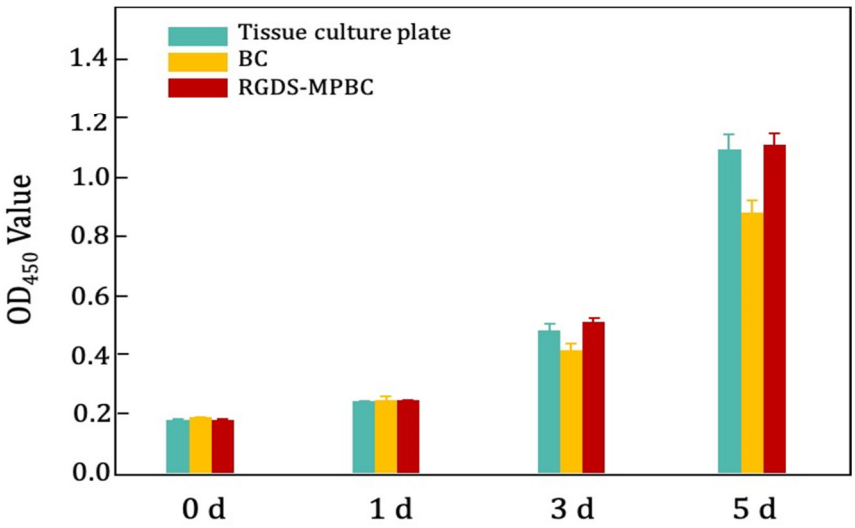
Cassie-Baxter state

Wenzel state



The hydrophilicity of a biomaterial is crucial to it´s usefulness because cells prefer to attach to hydrophilic surfaces rather than hydrophobic surface. In addition, the use of RGDS can improve cell attachment. On the other hand, the adsorption of collagen prefers hydrophobic surfaces. Therefore, RGDS-MPBC materials with RGDS immobilized column surfaces and hydrophobic groove channels were an ideal candidate to evaluate the effect on guiding cell migration and collagen distribution. *In vitro* cell culture assays revealed that fibroblasts L929 cells showed better viability in the extract liquid of RGDS-MPBC ($OD_{450,5d}$ ~ 1.1) than that of the pristine BC ($OD_{450,5d}$ ~ 0.88) after 5 days (**Picture 3**). In addition, the ordered aggregation of human skin fibroblast (HSF) cells was found on the column platform surface of RGDS-

MPBC, not in the groove channels, speaking for an excellent control of cell migration thus underlining the effectiveness of these materials to enhance scar-free wound healing processes.



Picture 3: *Mouse fibroblasts L929 cell viability of extracted liquids from tissue culture plate (as a control), BC and RGDS-MPBC.*

This research presents a promising approach toward scar-free wound healing, through induction of "basket-woven" collagen organization compared to a parallel aggregation of collagen fibers like in normal wound healing. The authors provide an effective solution for guiding cell migration and collagen distribution by fabricating micropatterned surfaces. The micropatterned surface offers hydrophilic columns where cells aggregate and more hydrophobic grooves which are beneficial for collagen organization.

An OCA 20 Contact Angle Analyser (DataPhysics Instruments GmbH, Germany) was used in this research.

For more information, please refer to the following article:

Surface engineering of spongy bacterial cellulose *via* constructing crossed groove/column micropattern by low-energy CO_2 laser photolithography toward scar-free wound healing; Yang Hu, Haiyan Liu, Xin Zhou, Haobo Pan, Xiuping Wu, Noureddine Abidi, Yongjun Zhu, Jinhui Wang; *Materials Science & Engineering C* **2019**, 99, 333-343; DOI: 10.1016/j.msec.2019.01.116