Newsletter









COVER STORY Multi-channel data: how to study correlations

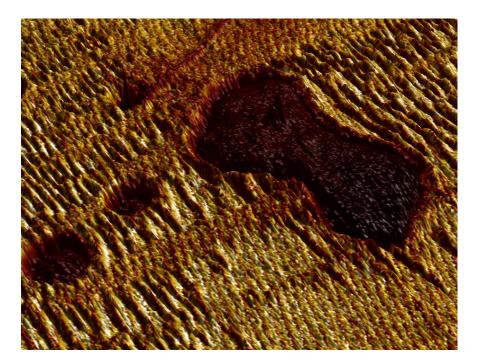
SEM ANALYSIS For improved wound care in burn patients

APPLICATION Exploring hyperspectral maps of 2D materials

SURFACE METROLOGY Q&A The surface metrology quiz is back!

NEWS & SOCIAL Events & product highlights What's hot online?





3D topography, mechanical properties, adhesion, chemical information, conductivity, magnetic force...

SPMs - and particularly AFMs - have the ability to yield many different kinds of data about the properties of a sample. In some applications, it can be useful to analyze some of these different data channels correlatively.

... Turn to page 2 ...

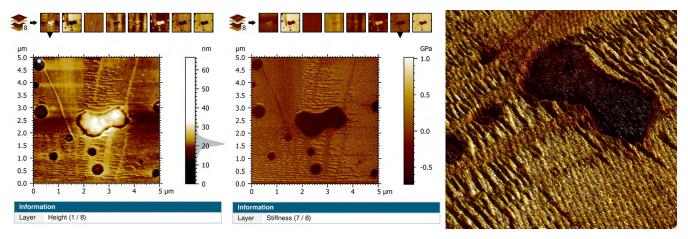


We look forward to seeing you at:

- Materials Research Society Meeting & Exhibit Booth #716 December 3-5, 2019- Boston, MA, USA
- ► DPG Spring Meeting Booth #25 March 15-20, 2020 Dresden, Germany

MULTI-CHANNEL DATA: HOW TO STUDY CORRELATIONS 66

Many scanning probe microscopes can image several interactions simultaneously. Through their different modes, SPMs produce multiple datasets at the same location on the sample. For some applications, it can be meaningful to combine and correlate the information coming from these different channels. Mountains-SPIP® 8 offers many tools for efficiently performing this comparison. Let's take a closer look, with **Isabelle Cauwet**, Digital Surf applications specialist.



Above (left to right). Height data, stiffness data (displayed in 2D) & 3D overlay of stiffness on height.

"MULTI-CHANNEL" IMAGE PROCESSING MADE SIMPLE

Some microscopes generate more than one value for each pixel. One of these "channels" is often topography, i.e. a map of Z heights. However, in scanning probe microscopy (AFM, STM etc.), it is common to obtain other values, such as conductivity or stiffness, which are central for understanding sample properties. MountainsSPIP® 8 allows users to handle multi-channel images as easily and as efficiently as if they were dealing with a simple topography image.

FINDING & VISUALIZING CORRELATIONS

Display your data in Full screen mode and select the channel (or "layer") you wish to visualize using the thumbnails at the top of the frame or the arrows in the contextual ribbon. Alternating the display of different channels is the first step in allowing you to visually identify correlations.

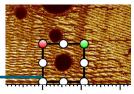
You may display correlations in 3D by superimposing the texture of a layer (in the example above, the stiffness) on a topography layer (the height). In MountainsSPIP®, you can apply color

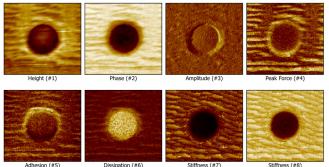
palettes to your data and optimize them to get better contrast. The "SPIP" palette corresponds to colors normally used when studying SPM data but you can choose from a range of many others.

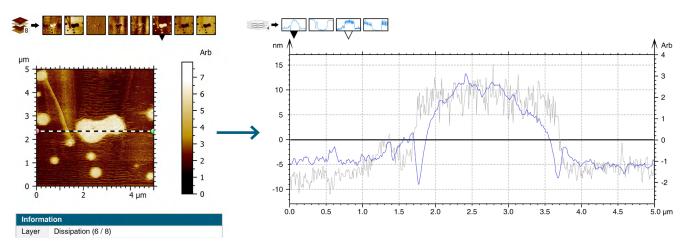
DYNAMICALLY ANALYZING **REGIONS OF INTEREST**

Using the Extract area operator, you can select a region of interest that will be effective on all channels simultaneously. You can freely move

the selection rectangle about, all further dependent (multi-channel) analysis steps will be updated accordingly in real time.







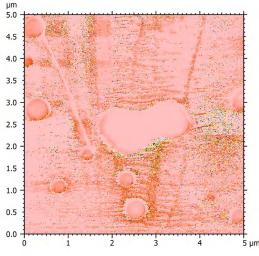
Above. The Extract profile tool lets users display correlations between different channels (here, height compared to dissipation).

EXPLORE CORRELATION IN GREATER DETAIL

The Extract profile operator lets you display the signal of the different channels on the same graph, and explore correlation in more detail. Above, we display the correlation graph between height (in blue) and dissipation (in gray). Once again, you can freely move the line representing the profile extraction (dotted line) and the graph will be updated automatically.

QUANTIFY CORRELATIONS

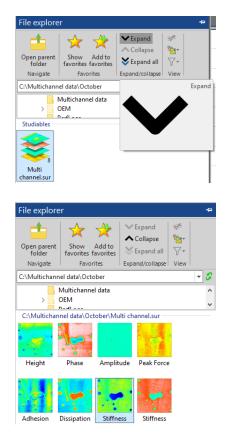
Finally, correlations between two different channels can be calculated, for example using a point by point ratio and analyzing the mean and distribution of this ratio. This is possible with the Mathematical function operator (see below).



Above. f(m)=Height/dissipation

BUT WHAT IF I ONLY WANT TO STUDY ONE CHANNEL?

All the signals contained in SPM data are not always useful. If you prefer to open only one channel, go to the File explorer, select your file and click on Expand (see image below).



Now, you just have to double click, or drag and drop, the channel(s) you want to analyze.



See dynamic tools for studying correlations in video

Many of the tools shown in this article are interactive so they are easier to see and understand in video format.

www.digitalsurf.com/news/multi-channel-data-how-to-study-correlations/

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SEM ANALYSIS FOR IMPROVED WOUND CARE IN BURN PATIENTS



Bacterial infection of wounds is a major risk for patients undergoing skin grafts following severe burn injuries. **Drs Monica Iliescu Nelea (left) & Michel Alain Danino,** of the Plastic and Reconstructive Surgery Department at the University of Montreal Hospital Center (CHUM), Montreal, Canada are part of a group of researchers working on furthering medical understanding of this phenomenon.

A biofilm is a layer of bacterial microorganisms that have aggregated to form a colony. Biofilms can develop on many surfaces, including wound dressings. This can be particularly critical for patients undergoing skin grafting for burn wounds since they often suffer from a weak or compromised immune system.

The areas where healthy skin is taken from, known as donor sites, are particularly vulnerable to biofilm formation, the presence of which strongly compromises normal wound healing. This in turn can become a source of pain for the patient and ultimately affects scar aesthetics.

THE SEM: A STATE-OF-THE-ART TOOL FOR BIOMATERIAL SURFACE CHARACTERIZATION

Using the Thermo Scientific™ (former FEI) Quanta™ 450 FEG Environmental Scanning Electron

Microscope (ESEM™), researchers compared the efficacy of two types of wound dressings used in preventing the formation of bacterial biofilm on burn patient skin graft donor sites.

One dressing contained bismuth tribromophenate at a concentration of 3% which confers it bacteriostatic (antibacterial) properties (XeroformTM). The other was an absorptive alginate calcium sodium dressing (KaltostatTM).

Right. Bacterial biofilms can develop on many surfaces, including wound dressings.

Dressing samples derived directly from actual burn patient donor sites were analyzed for bacterial biofilm density and then compared.

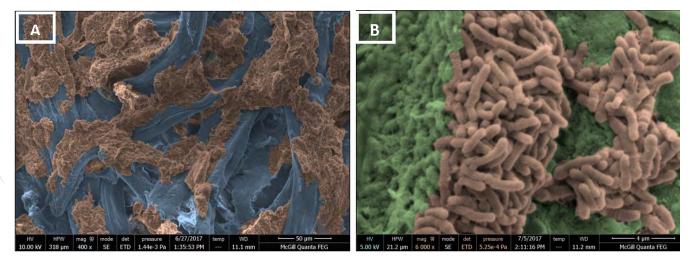
The samples were prepared for analysis under the scanning electron microscope (SEM) using an original method developed by the research team that aims to maintain the integrity of the biofilm microstructure.

To this day, SEM imaging has rarely been employed for dressing analysis and this is the first time that it has been used for in-situ biofilm visualization for this particular application.

ANALYZING RESULTS

In the SEM images obtained of the wound dressing samples, the research team assigned a score based on the predominance of bacteria and the presence of bacterial biofilm.





Above. Colorized SEM images of the Kaltostat™ wound dressing at different magnifications. Colorization was performed with TopoMaps software based on Mountains®. Bacterial biofilms are made visible by colorization in light brown.

Different types of bacteria were easily identifiable in the SEM images, both in isolated forms and in clusters within biofilm.

For example, it was possible to observe cocci-type bacteria establishing biofilm. Bacilli-type biofilm and streptococci were also observed. All these types of bacteria were detected for both dressings, the main difference being that they were highly visible in the Kaltostat[™] dressing and were less notable in both quantity and frequency in the Xeroform[™] dressing.

The images above, colorized using Mountains®based TopoMaps software¹, attest to the presence and extent of bacterial biofilm in a lower magnification view (A) where bacterial biofilm is colored in brown and wound dressing fibers in blue.

Furthermore, a higher magnification view image (B) shows bacterial clusters (colored in brown) as well as the extracellular polysaccharide network (EPS matrix, colored in green) which partially surrounds the bacteria, forming the biofilm.

"Colorizing the images helped us to better highlight the presence of bacterial biofilms on SEM images at low magnification" stated Dr Iliescu, "It also helped to enhance the presence of bacterial aggregates (clusters) and the formation of the EPS matrix on SEM images at high magnification."

FURTHER POSSIBILITIES IN BIOMEDICAL RESEARCH

This study allowed the team to show that the scanning electron microscope is undoubtedly an immensely valuable tool for the analysis of wound dressing samples for which it had not been previously used.

¹ www.fei.com/software/topomaps-for-materials-science/

This imaging technique permits researchers to visualize and characterize bacterial biofilms, provided that the preparation technique is appropriate and that results are correctly analyzed.

The team was able to observe that while donor sites covered with the commonly used wound dressing Kaltostat[™] often became breeding grounds for microorganisms, the dressing with bacteriostatic properties (Xeroform[™]) hindered bacterial proliferation and accelerated wound healing. In other words, bacterial biofilm formation was more pronounced in the wound dressing without bacteriostatic properties.

READ MORE

► In-situ characterization of the bacterial biofilm associated with Xeroform™ and Kaltostat™ dressings and evaluation of their effectiveness on thin skin engraftment donor sites in burn patients. Monica Iliescu Nelea, Laurence Paek, Lan Dao, Nathalie Rouchet, Johnny I. Efanov, Coeugniet Édouard, Michel A. Danino. Burns, Volume 45, Issue 5, August 2019, Pages 1122-1130. doi.org/10.1016/j.burns.2019.02.024

COLORIZE YOUR SEM IMAGES IN JUST A FEW CLICKS

Quick & easy SEM image colorization explained in video: <u>https://youtu.be/KCvIZwg6ZRs</u>



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EXPLORING HYPERSPECTRAL MAPS OF 2D MATERIALS



Tailoring 2D semiconductor heterostructures with specific bandgaps is a key aspect of leveraging new quantum materials for electronics and optoelectronics, one of the hot topics for researchers currently working in nanotech. **Craig Wall**, applications scientist at Montana Instruments, recently investigated the subject using Mountains[®] software to analyze results from Raman spectroscopy and photoluminescence.

2D HETEROSTRUCTURE ASSEMBLY IN A VARIABLE TEMPERATURE ENVIRONMENT

The basic heterostructure assembly principle is simple: exfoliate, for example, a monolayer of MoS₂ (molybdenum disulfide), put it on top of another mono- or few- layer crystal, such as WS₂ (tungsten disufide), add another 2D crystal, and so on. The resulting heterostructure represents an artificial material assembled in a specified sequence with single layer precision, held together by van der Waals forces.

Figure 1. White light image of MoS₂-WS₂ multilayer heterostructure



Often, additional or new information about a sample can be revealed with temperaturedependent measurements. In this study, we were able to demonstrate efficient hyperspectral mapping of 2D materials with the **microReveal Raman spectroscopy and photoluminescence platform** which maintains high spatial resolution and collection efficiency across the entire temperature range (4K to 500K). The system overcomes challenges associated with variable temperature sample drift by incorporating a low thermal mass sample stage, in-vacuum high NA objective, and coupling optics to a low astigmatism, broadband spectrograph. The exciton recombination dynamics of stacked MoS₂-WS₂ heterostructures have previously been studied at room temperature.1-2 The maximum of the valence band and the minimum of the conduction band are separated in the layers of WS2 and MoS2, respectively. The reported energy gap for MoS_2 is 2.39eV and for WS_2 is 2.31eV. The energy difference between the maximum valence bands of MoS₂ and WS₂ is about 350 meV. Photoexcited free electron-hole pairs prefer to stay separated at layer interfaces. The excited electrons in WS₂ tend to accumulate in the conduction band of MoS₂ while holes generated in the valence band of MoS₂ tend to transfer to WS₂ at the interface. The interlayer radiative recombination of spatially separated carriers can lead to the extra peaks in the photoluminescence spectra. In our investigations, we were able to observe the interlayer exciton in the hyperspectral mapping study which is at lower energy compared with pure monolayer MoS₂ and WS₂.

CONCLUSIONS

Thermomechanical drift and stability become more complex to control when systematically varying temperature throughout an experiment. The hyperspectral maps (see opposite page) are 200µm x 200µm with 1µm pixels and the collection of the 40,000 spectra that compose each map necessitates ultra-low thermomechanical drift, achievable with the microReveal.

Studies of the epitaxial and non-epitaxial heterostructures of MoS₂-WS₂ on silicon suggest that interlayer exciton relaxation is independent of stacking epitaxy and orientation, see Fig. 1.³ For the non-epitaxial heterostructures, raising the temperature from 5K to 150K revealed edge effects on the intensity of the PL from the base WS₂ layer.

color

Green

Yellow

Orange

Blue

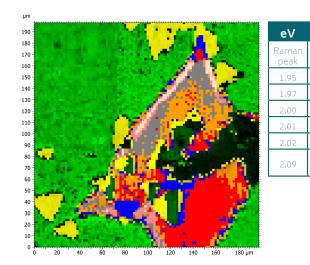
Red

HYPERSPECTRAL PHOTOLUMINESCENCE MAPS

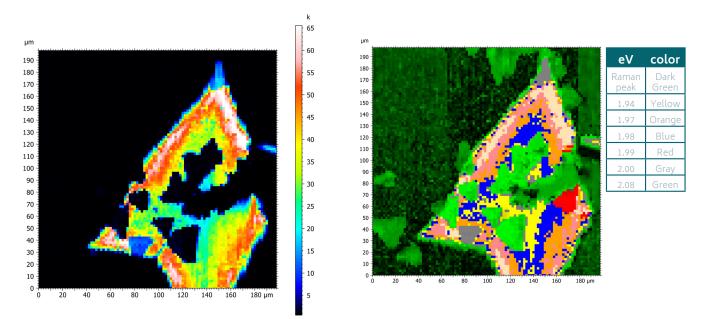
μm ò 180 µm

PRINCIPAL COMPONENT ANALYSIS

with k means clustering illustrating unique spectral contributions



Above. Non-epitaxial MoS₂-WS₂ at 5K.





Hyperspectral maps and advanced principal component analysis (PCA) were performed with Mountains® software.



REFERENCES

- 1. J. Zhang, et. al., **Observation of Strong Interlayer Coupling in MoS**₂**/WS**₂ **Heterostructures.** Adv. Mater. 2016, 28, 1950–1956.
- M. Thripuranthaka, et. al., Temperature dependent Raman spectroscopy of chemically derived few layer MoS₂ and WS₂ nanosheets. Appl. Phys. Lett. 2014, 104, 081911.
- 3. Y. Yifei, et. al., Equally Efficient Interlayer Exciton Relaxation and Improved Absorption in Epitaxial and Nonepitaxial MoS²/WS² Heterostructures. Nano Lett. 2015, 15, 1, 486-491.

ABOUT MONTANA INSTRUMENTS

Montana Instruments[®] Corporation designs and manufactures high-precision electrical, optical, and cryogenic systems for quantum materials research and the quantum computing, sensing and networking industries. Find out more: <u>www.montanainstruments.com</u>

SURFACE METROLOGY QUIZ #2 HOW MUCH DO YOU KNOW?



Working or studying in the field of surface metrology? How well do you know your stuff?

This edition of *Surface Newsletter* sees the return of the Surface Metrology Quiz. Test your knowledge by answering the questions on profile parameters and areal parameters devised by **François Blateyron**, Digital Surf's surface metrology expert.

Please note that for some questions, there are several correct answers.

QUESTION 1

WHICH PARAMETER IS BEST SUITED FOR CHECKING THE SEALING EFFICIENCY OF A GASKET?

A. Rq B. Pz C. Wt D. RSm

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► QUESTION 2

WHICH PARAMETER IS THE MOST SENSITIVE TO OUTLIERS?

A. Rq

B. Wz

C. Rku

D. Pa

QUESTION 3

WHICH OF THE FOLLOWING PARAMETERS CAN BE USED TO CONTROL LUBRICATION EFFICIENCY?

A. Rv B. Rvk C. Rdc D. Rpk

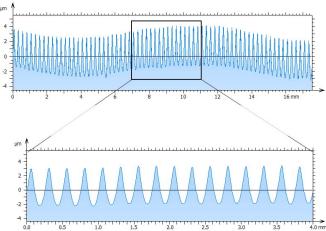


QUESTION 4

WHICH OF THE FOLLOWING FILTRATION CONDITIONS ARE BEST SUITED FOR THE CALIBRATION OF THE VERTICAL AMPLIFICATION COEFFICIENT ON A PROFILOMETER?

- A. Roughness, λc = 0.8 mm
- B. Roughness, λc = 0.25 mm
- C. Roughness, λc= 2.5 mm

D. Waviness, λc= 0.8 mm



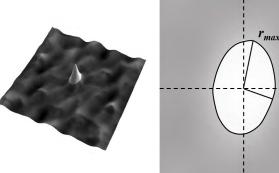
► QUESTION 5 WHICH OF THE FOLLOWING STATEMENTS ARE TRUE?

- A. It is always mandatory to apply a λs filter
- B. The λ s filter is sometimes forbidden
- C.The λs filter is not necessary for waviness parameters
- D.The λs filter can sometimes destroy useful information

► QUESTION 6

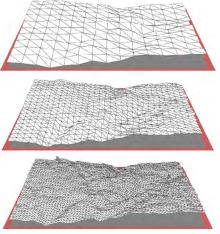
WHICH OF THE FOLLOWING PARAMETERS ARE CALCULATED FROM AN AUTOCORRELATION?

- A. Std
- B. Sal
- C. Str
- D. Sfd



► QUESTION 7 WHICH OF THE FOLLOWING METHODS ARE RELATED TO FRACTAL ANALYSIS?

- A. Morphological envelopes
- B. Length-scale analysis
- C. Area-scale analysis
- D. Box method



► QUESTION 8

WHICH OF THE FOLLOWING PARAMETERS ARE NOT DEFINED IN ISO 25178?

- A. Sbi
- B. Sdv
- C. S3z
- D. S5v

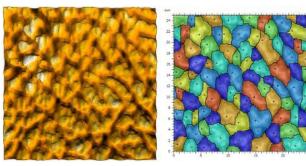
▶ QUESTION 9

WHICH OF THE FOLLOWING PARAMETERS ARE FEATURE PARAMETERS?

A. Spc

r_{min}

- B. Sda
- C. Std
- D. Spd



QUESTION 10

WHICH FAMOUS METROLOGIST ONCE SPOKE ABOUT "PARAMETER RASH"?

- A. Professor Tom Thomas
- B. Professor David Whitehouse
- C. Professor Ken Stout
- D. Professor F.A. Firestone



RESOURCES & ANSWERS

- You will find help on the questions in this quiz, as well as a wealth of other information on surface metrology, within the pages of the Digital Surf Surface Metrology Guide: <u>www.digitalsurf.com/guide</u>
- Solutions to the quiz : guide.digitalsurf.com/en/guide-qa-quiz-solutions2.html

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EVENTS & PRODUCTS HIGHLIGHTS

LOOKING BACK ON A BUSY SUMMER TRADE SHOW SEASON

Digital Surf was thrilled to participate in the 1D2D-Nanomat Summer School held in Corsica, France, July 2-12, 2019. This event, chaired by Bruno Grandidier, renowned scientist and group leader at the Institute of Electronics, Microelectronics and Nanotechnology (IEMN) in Lille brought together an international group of 81 researchers to discuss developments in the field of 1D and 2D semiconductor materials.



The program featured a presentation by Digital Surf's SPM specialist, Nicolas, on the main features of new MountainsSPIP® software. Attendees were invited to create a 3D view of their data using Mountains® software. A Facebook contest was organized to elect the public's favorites. Digital Surf was delighted to offer a prize to the winners at the evening gala reception and to 3D-print the winning models.

Over the summer, Digital Surf was also present at the annual Microscopy & Microanalysis (M&M) conference & exhibit held this year in Portland (Oregon), USA, August 4-8 2019. Anne, François P. and Cyrille (see photo above) were on hand to demonstrate the new features of Mountains[®] 8 software and discuss latest analysis methods.

Finally, in September, alongside 478 companies and 23,409 visitors, Digital Surf took part in JASIS (Japan Analytical & Scientific Instruments Show) in Tokyo. Arnaud, Damien and our valued interpreter Sato-san welcomed visitors to the Digital Surf booth which showcased Mountains[®] 8 capabilities, for scanning probe and scanning electron microscopy.

PRODUCT NEWS

MAKING THE SWITCH FROM SPIP[™] 6 TO MOUNTAINSPIP[®] 8

Recently-released MountainsSPIP[®] 8 software is the next-generation update of SPIP[™] 6. It contains most of the best SPIP[™] features (more will be added soon) and also adds the power and automation of the widely-reputed Mountains[®] platform for surface and image analysis.

SPIP[™] users with an active maintenance contract are entitled to a MountainsSPIP®8 license valid until

the end of 2020 free of charge in addition to their SPIP[™] license. Customers wishing to benefit from this offer are invited to contact <u>sales@digitalsurf.com</u> for further details and conditions.

To help make the transition as smooth as possible, the Digital Surf team has concocted some specific resources for SPIP[™] users including a "Survival Guide". The technical support team (<u>support@digitalsurf.com</u>) are also available to assist with any specific questions.



WHAT'S HOT ONLINE



SEEN ON LINKEDIN

Nov 1, 2019: Exactly 30 years since Digital Surf was founded in Besançon by Christophe Mignot and Bertrand Bellaton.

Thank you to all our friends, partners and customers for 30 extraordinary years. This is only the beginning of the story as we continue to grow and imagine the innovative solutions of tomorrow in surface and image analysis.

bit.ly/2pw4fKG





POPULAR ON FACEBOOK

October 18, 2019: team #DigitalSurf took part in the Amical Business Cup football tournament in Besançon. OK, so they may not have won but what a great atmosphere amongst the team and their supporters on the day. Vive le football!

bit.ly/33py2U5

Have you visited our YouTube channel recently?



Watch new Mountains® 8 features video tutorials to help you get started using the new version.

Check them out:

<u>https://youtu.be/</u> <u>UXZ0EnK8hxc</u>



Surface Newsletter

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Take Mountains[®] 8 for a test drive Visit <u>www.digitalsurf.com/free-trial</u>



CONTACT US FOR AN UPDATE

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MEET DIGITAL SURF

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HQ, R&D Center 16 rue Lavoisier 25000 Besançon - France Tel: +33 38150 4800 contact@digitalsurf.com



Surface Newsletter, November 2019

Editor : Christophe Mignot Content editor : Clare Jamet Contributors : François Blateyron, Lucas Bolle-Reddat, Isabelle Cauwet, Adeline Fuzenot, Monica Illiescu Nelea, Craig Wall

