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Characterization of Electrolyte Content in Urine Samples through a Differential Microfluidic Sensor Based on Dumbbell-Shaped Defected Ground Structures

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In this paper, a differential microfluidic sensor and comparator based on a pair of microstrip lines loaded with dumbbell-shaped defected ground structure (DB-DGS) resonators is applied to the characterization of electrolyte concentration in samples of horse urine. Since variations in the total electrolyte content in urine may be indicative of certain pathologies, the interest is to use the device as comparator, in order to determine changes in the electrolyte concentration as compared to a reference level. To validate the approach, we have made differential measurements of a set of urine samples with different electrolyte concentrations (which have been previously obtained by means of electrochemical methods). The obtained results correlate with the nominal electrolyte concentrations of the samples, thereby pointing out the potential of the approach as a low-cost pre-screening method (or complementary diagnosis system) to detect potential pathologies or diseases in horses and other animals.

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I. INTRODUCTION

Electrolytes such as sodium (Na^+), calcium (Ca^{2+}), potassium (K^+), chloride (Cl^-) and bicarbonate (HCO_3^-) are present in blood and urine, and play an important role in several vital functions, such as body hydration, blood pH and pressure control, nerve and muscle functions, etc. [1]. Indeed, excessive imbalances in the concentration of certain electrolytes (known as anion gap [2]), as well as an excess or defect of the total concentration of electrolytes in blood and urine, may be indicative of certain disorders. Thus, monitoring the concentration of ions in blood and urine is important for medical diagnosis and tailored fluid therapies. Currently available methods for that purpose use ion selective electrodes (ISE) [3]. Such methods are able to individually determine the concentration of specific electrolytes. However, such electrochemical system is expensive, and it is not compatible with the increasing demand of real-time monitoring of blood or urine bio-samples. Within this context, the development of alternative low-cost and real-time measurement methods for the characterization of electrolyte concentration in urine and blood is of high interest.

The presence of ions and other compounds in urine or blood determines their physical properties, in particular the conductivity (or loss factor) and the dielectric constant. Therefore, the total concentration of electrolytes in bio-samples can potentially be inferred by means of methods sensitive to such variables (conductivity and dielectric constant), and particularly through microwaves. Although microwave-based sensors are not able to selectively provide the specific concentration of the different ions in blood or urine, but the total concentration, microwave methods satisfy the above-cited demands of low cost and fast measurement. Thus, microwave sensors can be considered useful complementary diagnosis tools for diseases related to the alteration of blood or urine composition. Moreover, the electromagnetic (sensing) elements and the associated circuitry of microwave sensors are compatible with handheld solutions. Thus, in this paper we will apply a differential microwave sensor/comparator to the characterization of electrolyte content in urine, and particularly horse urine.

In recent years, significant efforts have been dedicated to the research and development of microwave sensors for material characterization, including bio-samples. Of particular interest are those sensors based on planar structures and electrically small resonators, due to their low cost, low profile, compatibility with printing fabrication processes (including flexible and conformal substrates), integration with sensor hardware, and high sensitivity, among other advantageous aspects. The sensing strategies can be categorized in various groups, to be discussed next.

One approach exploits the variation in the resonance frequency and magnitude experienced by the sensing resonator when it is loaded with the material (or sample) under test (SUT) [4]-[15]. Such technique is simple as far as a single resonator, typically loading a transmission line, suffices for sensing purposes. However, frequency variation sensors are subjected to cross-sensitivities, e.g., caused by changes in environmental factors (temperature and moisture, for instance). Therefore, such sensors need calibration before their use, in order to avoid false readouts of the variable of interest (measurand).

To alleviate the cross sensitivity to ambient conditions, sensors exploiting symmetry properties have been reported (see [16]-[18]). These sensors are based on symmetry disruption. Since symmetry is invariant to changes in environmental conditions, it follows that symmetry-based sensors are robust against the above-cited cross sensitivities. These symmetry-based sensors can be divided in three main categories: coupling modulation sensors [16],[19]-[26], frequency splitting sensors [27]-[33], and differential-mode sensors [34]-[46]. In coupling modulation sensors, a symmetric resonator symmetrically loads a transmission line. The line and the resonant element should not be arbitrarily selected. That is, for sensor functionality, the symmetry planes of both elements (the resonator and the line) must behave as electromagnetic walls of different sort (one an electric wall and the other one a magnetic wall) [16]-[18]. By this means, electromagnetic coupling between the resonator and the line is prevented, and the line is transparent. By contrast, when symmetry is truncated, e.g., by means of an asymmetric dielectric load, or by means of a relative (angular or linear) displacement between the line and the resonator, line-to-resonator coupling arises, and a notch in the transmission coefficient of the line is generated. The magnitude of this notch is related to the level of asymmetry and thereby it can be used as output variable for sensing purposes. This type of sensors can be applied to material characterization, but most of these sensors have been focused on the measurement of spatial variables and velocities [19]-[26]. The main limitation of these sensors is that measurement of notch magnitude is more sensitive to noise, as compared to frequency measurement.

Frequency splitting sensors consist of a transmission line structure symmetrically loaded with a pair of resonant elements (not necessarily symmetric) [29]-[30]. These sensors are similar to differential sensors as far as one resonant element is for the reference (REF) material, or sample, whereas the other one should be loaded with the (SUT). If the samples are identical, a single notch in the transmission coefficient arises. However, this notch splits in two notches, provided symmetry is truncated. The level of asymmetry dictates the difference in the notch frequencies, and therefore such frequency difference can be considered the main output variable for sensing [31]-[33].

Finally, in differential sensors, two independent sensors are used, one for the REF material and the other one for the SUT. Several implementations of these sensors have been reported, including sensors based on meandered lines [41],[44] on resonator loaded lines [35],[37]-[40],[42],[45]-[47], and sensors based on artificial transmission lines [34],[43]. In these sensors, the output variable can be the phase difference between the sensing lines [36],[41], but, recently, sensors based on the measurement of the cross-mode transmission coefficient have been reported [35],[37]-[39],[40],[43],[45]-[47]. Moreover, in a very recent implementation, differential sensors with enhanced sensitivity based on simple two-port measurements have been presented [44]. Sensors based on the measurement of the cross-mode transmission coefficient have demonstrated to exhibit good levels of sensitivity and resolution. It is remarkable, for instance, the sensor presented in [45], where a resolution as small as 0.125g/L (5.44 mEq/L) of electrolyte concentration in DI water was demonstrated. It is also worth-mentioning the sensor reported in [43], with very high sensitivity achieved thanks to the high dispersion characteristics of electro-inductive wave transmission lines [48] (the complementary counterpart of magneto-inductive wave transmission lines [49]-[54]).

As an extended paper of the conference paper [46], in this work we apply the differential-mode sensor first presented in [46], and based on a pair of microstrip lines loaded with a dumbbell-shaped defected ground structure (DB-DGS), to the characterization of urine samples. The main aim is to demonstrate that the sensor is sensitive to variations in the electrolyte content of the considered samples. Since such samples have been achieved from horses (in some cases suffering medical disorders), it follows that the sensor can be used as a method for real-time monitoring changes in the total electrolyte concentration in urine. Many other sensors focused on the characterization of liquids and bio-samples have been reported (see, e.g., [55]-[61]).

II. THE PROPOSED SENSOR

The differential-mode sensor used for the characterization of urine samples was the one first reported in [46], and then studied in detail in [40]. The topology of such sensor, including relevant dimensions, is depicted in Fig. 1. The sensor consists of a pair of microstrip lines, each one loaded with a dumbbell-shaped defected ground structure (DB-DGS) transversally oriented to the axis of the lines. For liquid characterization, fluidic channels on top of both DB-DGSs, plus the necessary accessories for liquid injection and for providing mechanical stability, are needed (see Fig. 2, and [40], where further details of the fluidic part of the sensor are reported). In [40], an exhaustive analysis relative to sensitivity improvement was carried out. It was concluded from that analysis that, for sensitivity optimization, the sensor substrate must exhibit a small dielectric constant. Moreover, the ratio between the inductance and the capacitance of the DB-DGS must be as small as possible. In practice, this is achieved by means of elongated topologies, as the one visible in Fig. 1 [40],[46].

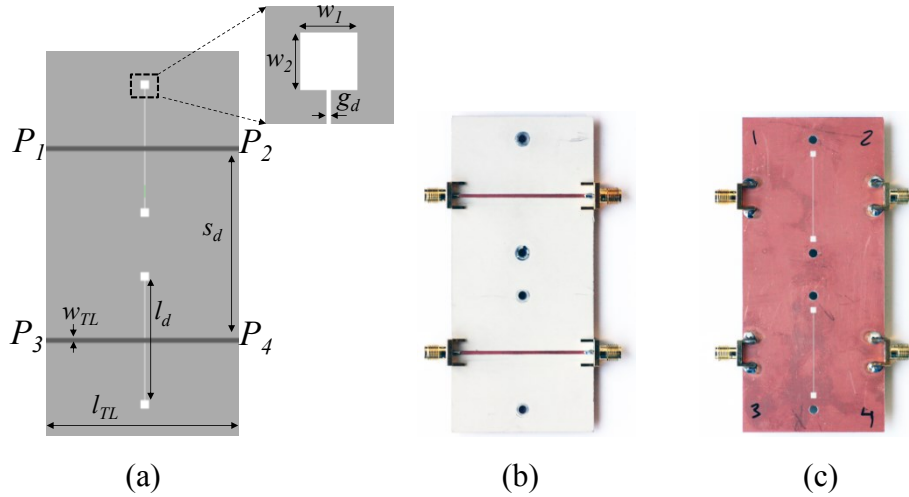


Fig. 1. The proposed DB-DGS based differential sensor. (a) Layout of the microwave part; (b) fabricated device (top); (c) fabricated device (bottom). Dimensions (in mm) are: $w_1 = w_2 = 2$, $w_{TL} = 1.14$, $l_{LT} = 50$, $l_d = 28$, $g_d = 0.2$, and $S_d = 44$. The considered substrate is the *Rogers RO3010* with thickness $h = 1.27$ mm, dielectric constant $\epsilon_r = 10.2$ and loss tangent $\tan\delta = 0.0035$.

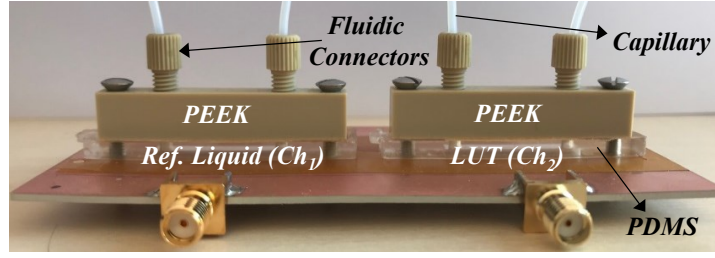


Fig. 2. Perspective view of the fabricated microfluidic sensor including the fluidic part.

The high sensitivity of the sensor of Fig. 1 was demonstrated in [40],[46], where it was used as a comparator, able to discriminate the presence of tiny defects in solid samples [46], as well as a measuring device, able to provide the concentration of NaCl in aqueous solutions. In this paper, the aim is to demonstrate the potential of the device to characterize urine samples, and particularly to infer total concentration of electrolytes, to be discussed in the next section.

III. CHARACTERIZATION OF URINE SAMPLES

The Faculty of Veterinary Sciences at the *Universitat Autònoma de Barcelona* has provided us the urine samples. Such samples have been obtained from horses suffering different disorders. The list of samples and the corresponding total concentration of electrolytes (in mEq/L) is shown in Table I, where the samples have been sorted in ascending order of electrolyte concentration. The nominal electrolyte concentrations in urine have been inferred from electrochemical methods by determination of sodium, potassium, chloride, calcium and magnesium concentrations, particularly ISE, as reported in the introduction (this aspect is out of the scope of this paper).

Table I. List of urine samples and the corresponding electrolyte concentration

<i>Urine sample</i>	<i>Electrolyte concentration (mEq/L)</i>
5341	130.05
5349	133.66
5344	155.35
5346	213.31
5345	311.14
5343	417.05
5347	552.14
5342	580.51
5348	757.2

Since the main purpose of this study is to demonstrate the potential of the approach as a method to monitor changes in the total concentration of electrolytes in urine, we have opted to consider sample #5341, the one with lower electrolyte content, as reference (REF) sample. In a real scenario, the interest is monitoring the potential changes of the electrolyte content in a diseased animal during a certain interval of time (e.g., during hospitalization). Consequently,

the REF sample is the urine at the beginning of the monitoring time interval. Each urine sample has been injected in the SUT channel, with the REF sample in the corresponding channel, and, after injection, we have obtained the cross-mode transmission coefficient. The results are depicted in Fig. 3, where it can be appreciated that the noise level is situated at roughly 25.9 dB. This is the maximum value of the cross-mode transmission coefficient corresponding to the symmetric case (i.e. with the REF sample in both channels). Figure 4 depicts the maximum value of the cross-mode transmission coefficient for the different samples (the x -axis corresponds to the total concentration of electrolytes of the samples). As it can be seen, there is in general a correlation between the electrolyte content and the maximum value of the cross-mode transmission coefficient, although one sample (#5342) does not follow this trend. This is thought to be due to the presence of other substances, in particular sediments, which are visible and may affect somehow the complex permittivity of the SUT. Nevertheless, these results indicate that the system is able to detect small changes in total concentration of electrolytes. Actually, we have repeated the measurement four times, in order to ensure that the results are repetitive. The error bars, included in Fig.4, indicate that the results are repetitive to a good extent.

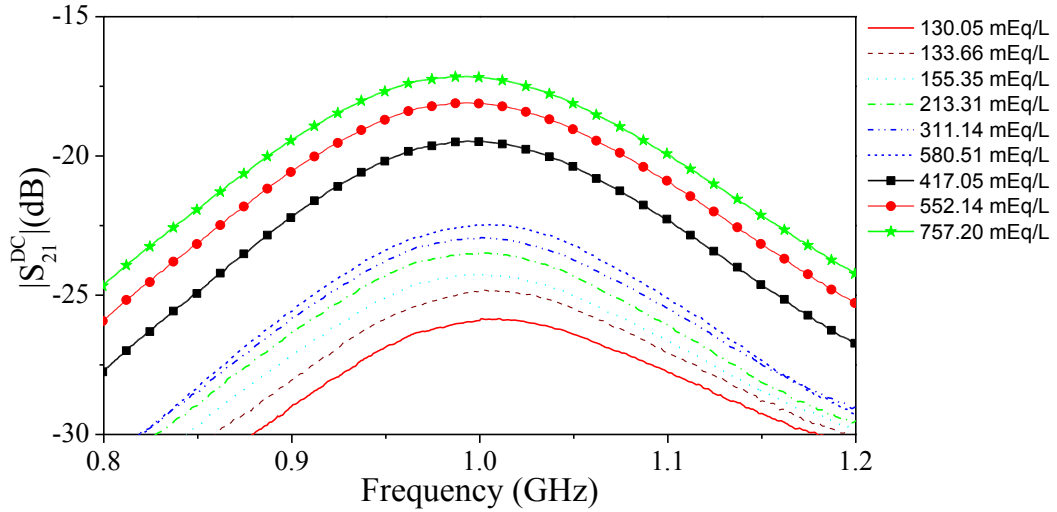


Fig. 3. Cross-mode transmission coefficient for the different SUT samples.

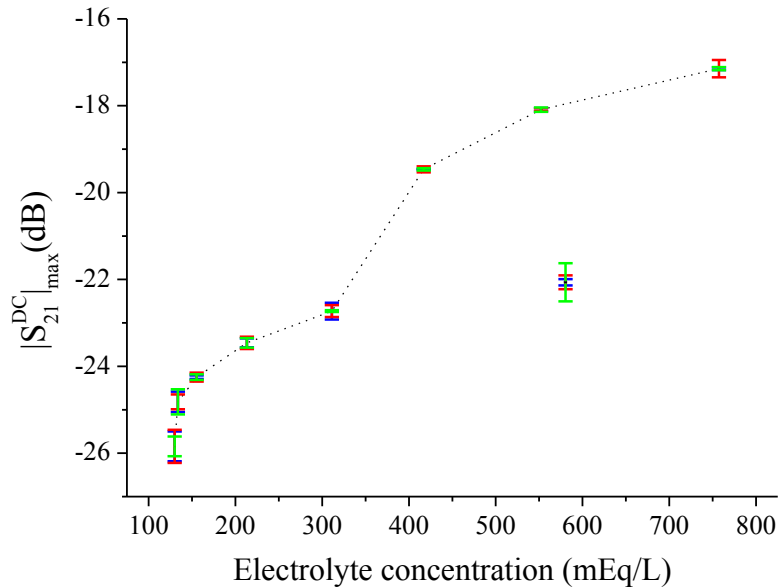


Fig. 4. Maximum value of the cross-mode transmission coefficient for the different samples.

We have also characterized the same urine samples by means of the differential-mode sensor reported in [39]. The measured cross-mode transmission coefficients are depicted in Fig. 5, whereas the maximum value of the cross-mode transmission coefficient is shown in Fig. 6. For this sensing device, also the sample #5342 (with a nominal concentration of electrolytes of 580.51 mEq/L) does not correlate with the other values. Therefore, these results support that the presence of visible sediments in the urine sample is the cause of the uncorrelated value of the cross-mode transmission coefficient. The fact that the results obtained from both independent differential sensors exhibit good correlation (with the above-cited exception of the altered sample #5342), indicate that the proposed differential sensing method, based on the measurement of the cross-mode transmission coefficient, is useful to real-time monitoring potential changes in the total electrolyte content of urine in animal patients.

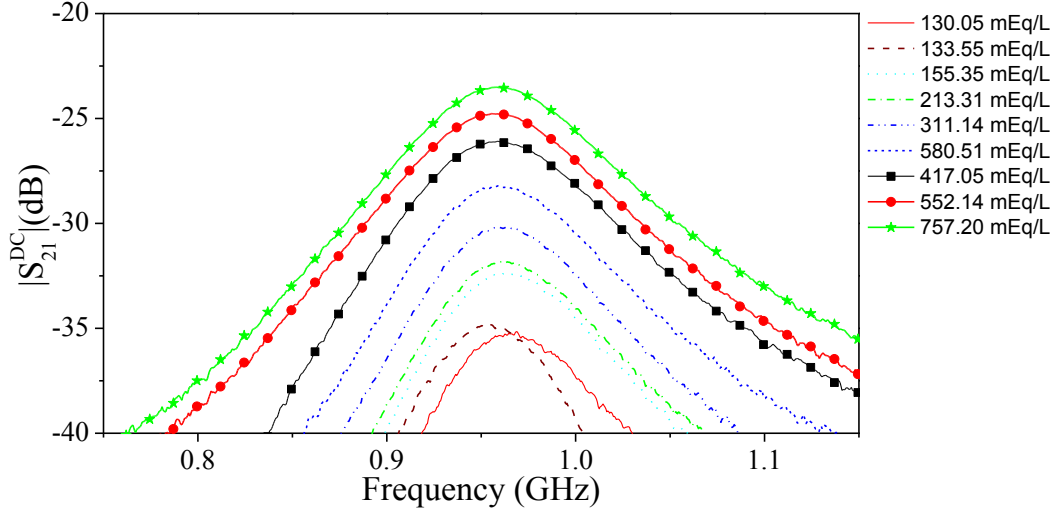


Fig. 5. Cross-mode transmission coefficient for the different SUT samples inferred by means of the sensor system reported in [39].

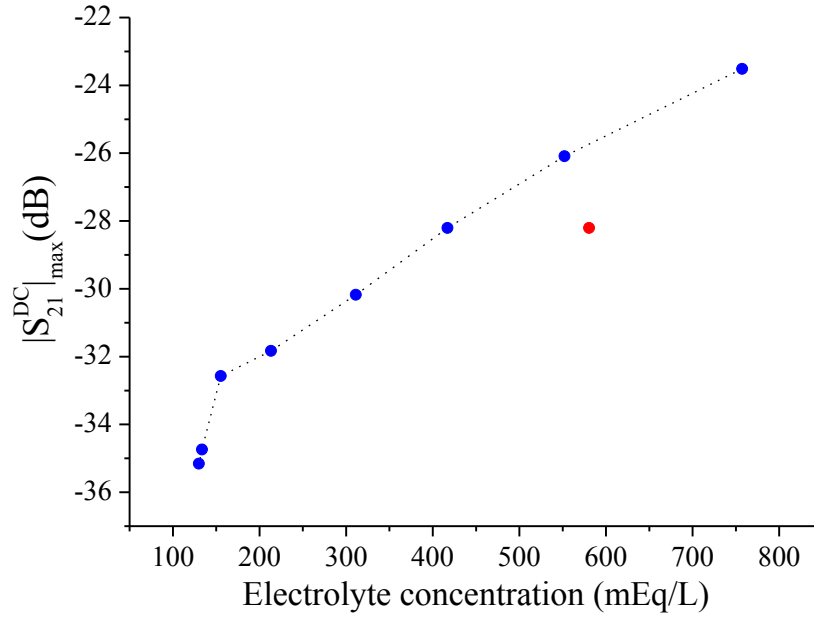


Fig. 6. Maximum value of the cross-mode transmission coefficient for the different samples, as inferred from the sensor reported in [39].

IV. DISCUSSION

In general, microfluidic sensors based on DGS structures as sensing elements are interesting as far as the upper side of the substrate is kept unaltered, i.e., without the presence of the fluidic channel, and this eases, in general, sensor design. There are other DGS structures of interest for sensing, for example, complementary split ring resonators (CSRRs) [62]. CSRR-based sensors with good sensitivity have been reported. However, as discussed in [63], the sensitivity of the resonance frequency of DB-DGSs with the dielectric constant of the SUT is, in general, superior to the one of CSRRs. This is because in a DGS structure, the varying capacitance directly affects the resonance frequency, whereas in a CSRR there is a coupling capacitance that adds to the varying capacitance, and this obscures somehow the effects of the dielectric constant of the SUT on the resonance frequency, thereby limiting the sensitivity.

In [40], an exhaustive comparative analysis of various types of sensors for the measurement of solute content (mainly NaCl and glucose) in DI water was carried out, and it was concluded from that analysis that the sensor of Fig. 2 and the one in [39] (used to obtain the results depicted in Figs. 5 and 6), offer a very competitive combination of resolution, sensitivity and dynamic range. For that main reason, the measurements of the cross-mode transmission coefficient for the different urine samples have been obtained by considering not only the DGS-based sensor (Fig. 2), but also the SRR based sensor of [39]. Indeed, the results in terms of performance are very similar, as it can be appreciated by comparing Figs. 4 and 6 (note, however, that a true comparison is not easy since the substrate materials are different, as discussed in [40]). It can be concluded from the results of this paper, and from the above-cited comparative analysis, that the sensor of Fig. 2 is very useful to monitor variations of electrolyte concentrations in urine, the main intended application. It is also a good candidate for that purpose the sensor reported in [39], though in this case the fluidic part should be placed at the same substrate face than the line strip.

Let us further emphasize that this work represents a first stage for the development of low-cost sensors for monitoring changes in the electrolyte content of urine in diseased animals, particularly horses, in real time. Monitoring these potential changes in the electrolyte content is of interest as a pre-screening method to detect possible pathologies related to variations in electrolyte content. In a real scenario, the REF sample should be the urine of the animal at the beginning of the monitoring time. Nevertheless, in this paper, we have opted to consider as REF sample one specific sample, i.e., the one with smaller electrolyte content, as a way to emulate real operating conditions of the sensor.

V. CONCLUSION

In conclusion, the microwave comparator presented in [46], based on a pair of DB-DGS-loaded microstrip lines, has been applied to the characterization of urine samples in this paper. The samples contain different concentrations of electrolytes and have been obtained from horses suffering different diseases. We have considered as REF sample the one with smaller concentration of urine, and it has been found that the maximum value of the cross-mode transmission coefficient for the different SUT samples exhibits a good correlation with the electrolyte content (with one exception attributed to the presence of sediments in the sample).

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Bibliographies



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Eduard Jose-Cunilleras was born in Barcelona, Spain, in 1974. He received the degree in Veterinary Sciences from the Universitat Autònoma de Barcelona, Barcelona, in 1997. He was awarded with a postgraduate fellowship by the Fulbright Fellowship from 1997 to 1999 to complete an internship in Equine Medicine and Surgery and MSc from The Ohio State University, USA. From 1999 to 2004 he completed a three year residency in Equine Internal Medicine and obtained a PhD degree in Veterinary Clinical Sciences from The Ohio State University. In 2004 he joined the “Animal Health Trust”, UK, as senior equine clinician, and from 2005 to 2008 he continued his professional career as equine veterinary surgeon in equine hospitals in the private sector. In 2008 he returned to the Universitat Autònoma de Barcelona as Lecturer in Equine Medicine and senior clinician in the Veterinary Hospital, and as Senior Lecturer from 2016 to present date. His current research interests include inflammatory markers, coagulation disorders, acid-base and electrolytes disorders in equids and in laboratory animals. He has authored over 60 peer-reviewed journal papers and book chapters. In addition he is head of Department and head of the Equine medicine research group.



Anna Bassols was born in Barcelona, Spain, in 1958. She received a Degree in Pharmacy (University of Barcelona) in 1980 and a PhD in Pharmacy in 1986 with a PhD Thesis on the regulation of skeletal muscle glucose metabolism. In 1986-1987 she earned a Fulbright Fellowship for a postdoctoral stay at the Department of Biochemistry (Massachusetts University Medical Center, USA) to investigate the mechanism of action of TGF- β in the regulation of the extracellular matrix. In 1988 he was named Associate Professor at the Department of Biochemistry and Molecular Biology, at the School of Veterinary Medicine (Universitat Autònoma de Barcelona.) Since 2009, she is Full Professor in 2009 at the same place. Her research career was first focused in extracellular matrix in cancer and other diseases, in humans and other animal species, as dogs, pigs and cattle. In 2006, she started a new research line on the applications of biochemical approaches and techniques to veterinary problems, especially for the search of biomarkers and the development of methods for their determination. On the other hand, her research has focused on the application of proteomics and metabolomics to animal science, related to the identification of biomarkers in farm animals. She is also interested on the central nervous system evaluating changes in neurotransmitter profiles and the brain proteome as markers for stress and welfare. Since 1992, she is also the Scientific Director of the Veterinary Clinical Biochemistry Service, at UAB, a leading diagnostic laboratory for biochemical analysis in domestic and farm animals. She has participated in 25 research national and international projects and she is author or coauthor of

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Ferran Martín (M'04-SM'08-F'12) was born in Barakaldo (Vizcaya), Spain in 1965. He received the B.S. Degree in Physics from the Universitat Autònoma de Barcelona (UAB) in 1988 and the PhD degree in 1992. From 1994 up to 2006 he was Associate Professor in Electronics at the Departament d'Enginyeria Electrònica (Universitat Autònoma de Barcelona), and since 2007 he is Full Professor of Electronics. In recent years, he has been involved in different research activities including modelling and simulation of electron devices for high frequency applications, millimeter wave and THz generation systems, and the application of electromagnetic bandgaps to microwave and millimeter wave circuits. He is now very active in the field of metamaterials and their application to the miniaturization and optimization of microwave circuits and antennas. Other topics of interest include microwave sensors and RFID systems, with special emphasis on the development of high data capacity chipless-RFID tags. He is the head of the Microwave Engineering, Metamaterials and Antennas Group (GEMMA Group) at UAB, and director of CIMITEC, a research Center on Metamaterials supported by TECNIO (Generalitat de Catalunya). He has organized several international events related to metamaterials and related topics, including Workshops at the IEEE International Microwave Symposium (years 2005 and 2007) and European Microwave Conference (2009, 2015 and 2017), and the Fifth International Congress on Advanced Electromagnetic Materials in Microwaves and Optics (Metamaterials 2011), where he acted as Chair of the Local Organizing Committee. He has acted as Guest Editor for six Special Issues on metamaterials and sensors in five International Journals. He has authored and co-authored over 600 technical conference, letter, journal papers and book chapters, he is co-author of the book on Metamaterials entitled *Metamaterials with Negative Parameters: Theory, Design and Microwave Applications* (John Wiley & Sons Inc.), author of the book *Artificial Transmission Lines for RF and Microwave Applications* (John Wiley & Sons Inc.), and co-editor of the book *Balanced Microwave Filters* (Wiley/IEEE Press). Ferran Martín has generated 19 PhDs, has filed several patents on metamaterials and has headed several Development Contracts.

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