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REVIEW ARTICLE

A methodological combined framework for roadmapping biosensor research: a fault tree analysis approach within a strategic technology evaluation frame

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Abstract

Biosensor technology began in the 1960s to revolutionize instrumentation and measurement. Despite the glucose sensor market success that revolutionized medical diagnostics, and artificial pancreas promise currently the approval stage, the industry is reluctant to capitalize on other relevant university-produced knowledge and innovation. On the other hand, the scientific literature is extensive and persisting, while the number of university-hosted biosensor groups is growing. Considering the limited marketability of biosensors compared to the available research output, the biosensor field has been used by the present authors as a suitable paradigm for developing a methodological combined framework for "roadmapping" university research output in this discipline. This framework adopts the basic principles of the Analytic Hierarchy Process (AHP), replacing the lower level of technology alternatives with internal barriers (drawbacks, limitations, disadvantages), modeled through fault tree analysis (FTA) relying on fuzzy reasoning to count for uncertainty. The proposed methodology is validated retrospectively using ion selective field effect transistor (ISFET) – based biosensors as a case example, and then implemented prospectively membrane biosensors, putting an emphasis on the manufacturability issues. The analysis performed the trajectory of membrane platforms differently than the available market roadmaps that, considering the vast industrial experience in tailoring and handling crystallic forms, suggest the technology path of biomimetic and synthetic materials. The results presented herein indicate that future trajectories lie along with nanotechnology, and especially nanofabrication and nano-bioinformatics, and focused, more on the science-path, that is, on controlling the natural process of self-assembly and the thermodynamics of bioelement-lipid interaction. This retained the nature-derived sensitivity of the biosensor platform, pointing out the differences between the scope of academic research and the market viewpoint.

Keywords

Analytic hierarchy process, biosensors, fault tree analysis, fuzzy reasoning, systems of innovation

History

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Introduction

Innovation systems and science policies frequently focus on linkages between universities and industry, and the commercial translation of academic discoveries. Overlooked in such analyses are the knowledge discovery processes within academic research and the non-commercial aspects of science. The desire to move research into practical applications to capture the benefits derived from scientific discovery has long motivated science funding and program development, especially in the biotechnology sector. Distinctions between ''pure'' and ''applied'' science were common during the 1960s through to the 1980s. However, it is widely recognized that this linear model of innovation, in which pure science is published in the academic literature, then picked-up by industry and developed into useful applications, is unnecessarily simplistic and frequently inaccurate, at least for certain disciplines. More likely, scientific research in these disciplines contains aspects of both basic inquiry (discovery) and practical application (utility) and moves back and forth between the two (Stokes, 1997). These movements take place at a deeper phenomenological level and their traces become somehow evident only to researchers who actually incorporate instinctively the relevant methodological paths into their hypotheses. On the other hand, methodologists or technology managers engaged, respectively, with the theoretical or the practical aspect of research, but not with the research itself, stay usually on a surface level being able to discriminate only quasi-linear segments of a trace, which is usually more complex, sometimes tending to completely cover part of the surface like a Peano curve.

While several tools have been developed to address certain aspects of science-enabled innovation, academic research has not been studied independently but considered either as the problem-solving aspect of technology commercialization (Chang et al., 2009; Kajikawa et al., 2008; Miyazaki & Islam, 2007; Pandza & Holt, 2007) or as the front-end of product development (Fleischer et al., 2008; Oliveira &

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Rozenfeld, 2010). In both approaches, trajectories have been drawn in association with the commercial translation of the scientific research, assigning a role of research regulator to the industrial sector, while various terms have been used over the years to connect discovery and utility, such as ''mission-oriented research'', ''directed research'', ''use-inspired basic research'' and ''strategic research''; the last term has been especially connected with back-casting. This view however neglects the self-regulating role of university research, where technology evolution is defined by the inherent technology barriers. However, the pursuit of strategies to overcome these barriers gives rise to new opportunities, develops extensive networks of collaboration or intensifies dissipative ones.

In no field has the academic research dynamics governing the production of scientific knowledge been more apparent than in biosciences, opened up by the rise of biotechnology, fueling advances in some of the most notable scientific areas to emerge over the last decades, such as clinical diagnostics, environmental monitoring, pharmaceuticals and industrial control. Bio-related academic research has been at the forefront of the scientific and industrial infrastructure for the past two decades now (Supplementary Figure 1), feeding a push-pull mechanism that serves both academic excellence and product innovation through an efficient technology transfer stream (Bishop et al., 2011; Schmoch, 2007). The impact of biotechnology on the pharmaceutical sector is a case in point. The innovative network developed has not only altered the technological profile of the sector, by driving chemical production to bioprocesses. It has also shifted drastically the trajectories from chemistry toward life sciences (through biochemistry) (Gavrilescu & Chisti, 2005). The network's grounding in basic research, together with its multidisciplinary and decentralized dynamics, increased the relevance of academic research outputs, fostered the emergence of specialized biotechnology firms that necessitated (and promoted) intra-industrial cooperations and university–industry links, while obliging the pharmaceutical firms to reposition their strategies (Canongia, 2007; Salicrup & Fedorková, 2006). Markedly, the high transformative capacity of the science-enabled technology, that is, the high capability of the scientific bodies involved to constantly redefine the portfolio of their products based on endogenously produced knowledge (Garud & Nayyar, 1994) brought about significant structural and institutional changes to the sector, giving rise to new scientific and technological opportunities.

There are cases, however, where several new technologies, incubated and fostered within the same evolutionary frame of biosciences, had only an indirect, supportive and subsidiary impact while failing to challenge the sectoral system and its established structures in any substantial way. Biosensor technology, for example, was set off to revolutionize instrumentation and measurement (see e.g. Churchouse et al., 1986; Griffiths & Hall, 1993; Hall, 1986; Petersen, 1996), particularly medical diagnostics and treatment (see e.g. Mascini, 1992; Severinghaus & Astrup, 1986; Turner & Pickup, 1985; Vadgama, 1984; Wilson et al., 1992). Yet, the industry was very reluctant to capitalize on university-produced knowledge and innovation, which gave rise to a, mostly European, paradox where long-term academic excellence has been sustained by a large number of university-hosted biosensor groups with a high absorptive capacity, that is, a marked capability to recognize, value and assimilate exogenous technological change within their scope of research (Cohen & Levinthal, 1990), but low translational capability, that is, low commercialization of the work produced (Luong et al., 2008). In their recent work (Siontorou & Batzias, 2010), the authors have attempted to look into the misalignment of the biosensor field to the related sector dynamics by exploring the reasons behind the limited technology transfer stream and determining the factors, exogenous to biosensor technology (extrinsic barriers), that reduced the likelihood of its successful commercialization. Built to narrow the gaps observed between the technology-driven and industry-related aspects of absorptive capacity, the authors presented a decision-making support tool that the industry can use to strategically model and manage the university output for product innovation.

The present work focuses on the academic research output *per se*, exploring the dynamic structure of the innovation system and attempting to frame it by considering (i) its scientific basis toward (but not through) its commercial applications, and (ii) the factors intrinsic to the technology itself (technology barriers) that could ultimately determine its commercial roadmaps. Roadmapping academic research on biosensors can be a challenging endeavor. Given the complex and dynamic context of the development environment that is oriented more toward knowledge exploration and less to knowledge exploitation, it is hard to anticipate the paths that the emerging technology will follow.

Considering the relatively limited marketability of biosensors (as compared to the available research output), this study delineates the biosensor field as a self-organized university-fostered technology innovation system that is aggregated and dynamic. It firstly critically reviews the early organization of the system within a technological frame that portrays this disruptive technology as providing point solutions to niche problems insufficiently addressed by the conventional (or alternative) technology base and discusses the role of the inherent technology barriers to the compulsory shifting of that frame. It presents a methodological combined framework for strategic technology evaluation and roadmapping of the system, adopting the basic principles of the analytic hierarchy process (AHP) to scientific research output and replacing the lower level of technology alternatives with internal barriers modeled through fault tree analysis (FTA) relying on fuzzy reasoning to account for uncertainty. The framework is validated retrospectively using ion selective field effect transistor (ISFET)-based biosensors as a paradigm and then implemented prospectively for membrane biosensors. A comprehensive discussion on both the methodological tool developed and the future of the biosensor research field is also presented.

Building the biosensor innovation hub – a retrospective analysis

The pioneering work of Leland C. Clark, Jr. in 1960s, which transformed an oxygen probe into a glucose meter (Clark & Lyons, 1962), paved the way for the evolution of biosensor

revolution. The enzyme probe reached the market almost a decade later, when Yellow Springs Instruments (YSI) launched its whole blood glucose analyzer (Model 23 Glucose Analyzer) with a polarographic electrode based on Clark's patent (Clark, 1970). This market success fitted glucose biosensors within a frame that displayed a momentum of its own, built upon what Kuhn (1962) called ''paradigms'', or even more precise, what Lakatos (1970) called ''research programmes.'' The various actors engaged in the emergent techno-economic network drew roadmaps to commercialization that shifted drastically the trajectories of the biomedical industry (Churchouse et al., 1986; Vadgama, 1984). Roadmapping, set in the early 1980s (Boitieux et al., 1985; Nylander, 1985; Severinghaus & Astrup, 1986), followed a strong market-pull mechanism for clinical diagnostics and home-care over-the-counter glucose sensing (Supplementary Figure 2). Three paths emerged, namely on glucose self-monitoring devices, artificial pancreas and continuous (*in vivo*) glucose monitoring. The drivers that created and sustained the three paths were industry, society and the scientific community, respectively, creating *ad hoc* an efficient thick innovation system, that is, a *sui generis* system consisting of actors with endogenous knowledge and interest on glucose sensing, characterized, at least until 1990, by knowledge asymmetry and limited dispersion around the early US biosensor research clusters. Inevitably, the concrete structure of the network pushed and pulled players into finite positions (Siontorou & Batzias, 2010), according to the needs for knowledge absorption and knowledge exploitation, patterning knowledge flows through the mechanisms of demand-driven absorptive capacity and supply-driven transformative capacity, respectively.

Pulling biosensor innovation into the market

During 1970–1989, this mono-disciplinary glucose frame gained its structural properties, outlining scopes, technology strategies, definitions and classifications, focusing on glucose monitoring (Heller & Feldman, 2008), although other possible and plausible fields of biosensing applications started to emerge. Markedly, a strong and effective university–industry alliance had been formed (Siontorou & Batzias, 2010), mostly in the United States, engaging actors with similar awareness of the technology at hand, aiming at rapidly solving scientific and technical problems (vastly on accuracy and sample size, respectively) and at building capabilities in line with customers' requirements. Thereby, this productive coexistence of industry, science and society (Figure 1) outlined, inevitably, a strategic perspective within which the companies strived to align their technology trajectories to sustain the performance of their product concepts and enhance the existing knowledge base.

As more industries became involved, including Ames/Miles, Lifescan, Roche, Boehringer Mannheim, Bayer, Medistron and Johnson & Johnson, more researchers became involved, lifting fast expectations from *ex vivo* measurements to *in vivo* monitoring within less than a decade (Vadgama, 1984). Evidently, industry realized the new scientific field as a discontinuous and disruptive technology during an era of ferment, that is, as an innovation system

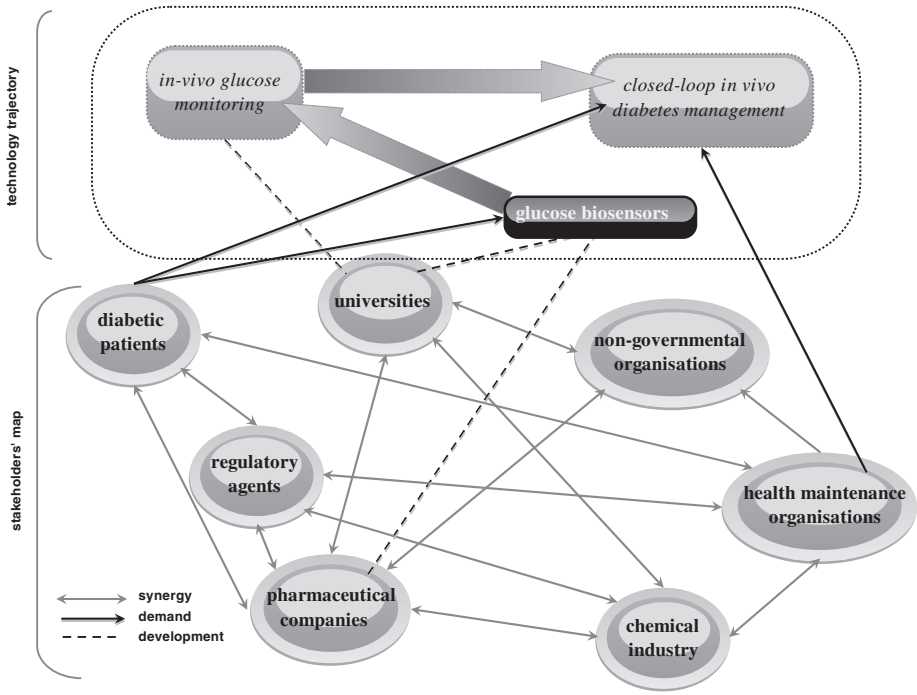

Figure 1. The early system of glucose biosensor innovation, showing the relationships among the drivers and actors of the thick network before the establishment of the business environment, that is, when the dependence of the firms upon the universities was still intense. Interestingly, while both diabetic patients and health organizations demanded radical disease management through the implementation of artificial pancreas (as utmost efficient and cost-effective, respectively), university research was focusing on *in vivo* monitoring as a more attainable short-term goal, based on the long clinical experience of catheters that could be easily adjusted to accommodate the electrodes. Notwithstanding, research on implant sensors produced significant knowledge about online/real-time individualized monitoring, while adding know-how for the design/construction/implementation of the artificial pancreas.

resulting in the generation of new technologies and in changes in the relative weighting of existing technologies (Anderson & Tushman, 1990), presenting superior performance trajectories along critical dimensions that customers (i.e. diabetic patients) value. Larger firms struggled to take advantage of this technology ahead of competitors in the market. Some have developed in-house R&D for capitalizing on own architectural innovation (i.e. of the whole system and its components, according to the definition of Carayannis et al., 2003) and for extending the radical technology; for example, after Roche launched the Haemo-Glucotest in 1968, the company spent 10 years on improving it and needed another 10 years to launch its line of glucose meters (Accu-Chek). Others tried to form direct links with the firms that had already the technology at near to ''fully fledged'' innovations, that is, outsourcing knowledge through acquisitions that enabled rapid market entries secured by proprietary rights. For example, Bayer acquired Ames/Miles in 1978 and launched Glucometer® a few years later, while Johnson & Johnson acquired Medistron and LifeScan in 1986 to have access to the Glucoscan®.

Gradually distancing itself from the university science base, the industry established a high competition arena in the early 1990s that gave rise to significant advances in technology, with miniaturization of analyzers and a remarkable increase in the number of devices available, nowadays listed by the ''ADA's Buyer's Guide to Diabetes Products'' (American Diabetes Association, 2005). This arena was determined by the timing and impact of the innovation under consideration. The new technology was called to rapidly address the needs of 5% of the US population and 0.4% of the world population, at an increasing rate of 3–5% annually and at a direct health care cost of $2000 per patient (Steck & Rewers, 2004). This innovation gave a straightforward cost reduction trajectory at a performance level higher than that of the earlier processes that it replaced. Four strategies have been consecutively followed in industrial research to deal with the challenges and opportunities of diabetic management (inset to Figure 2), as determined by patent searching (using the methodology proposed by Lee & Su (2011) for mapping scientific knowledge): (i) intensifying the pull-basis toward the performance improvements demanded by the health care providers (R&D on performance), (ii) ascending the trajectory of the push-basis toward higher level of efficacy (R&D on reliability), (iii) aligning with the needs of end customers (R&D on convenience) and

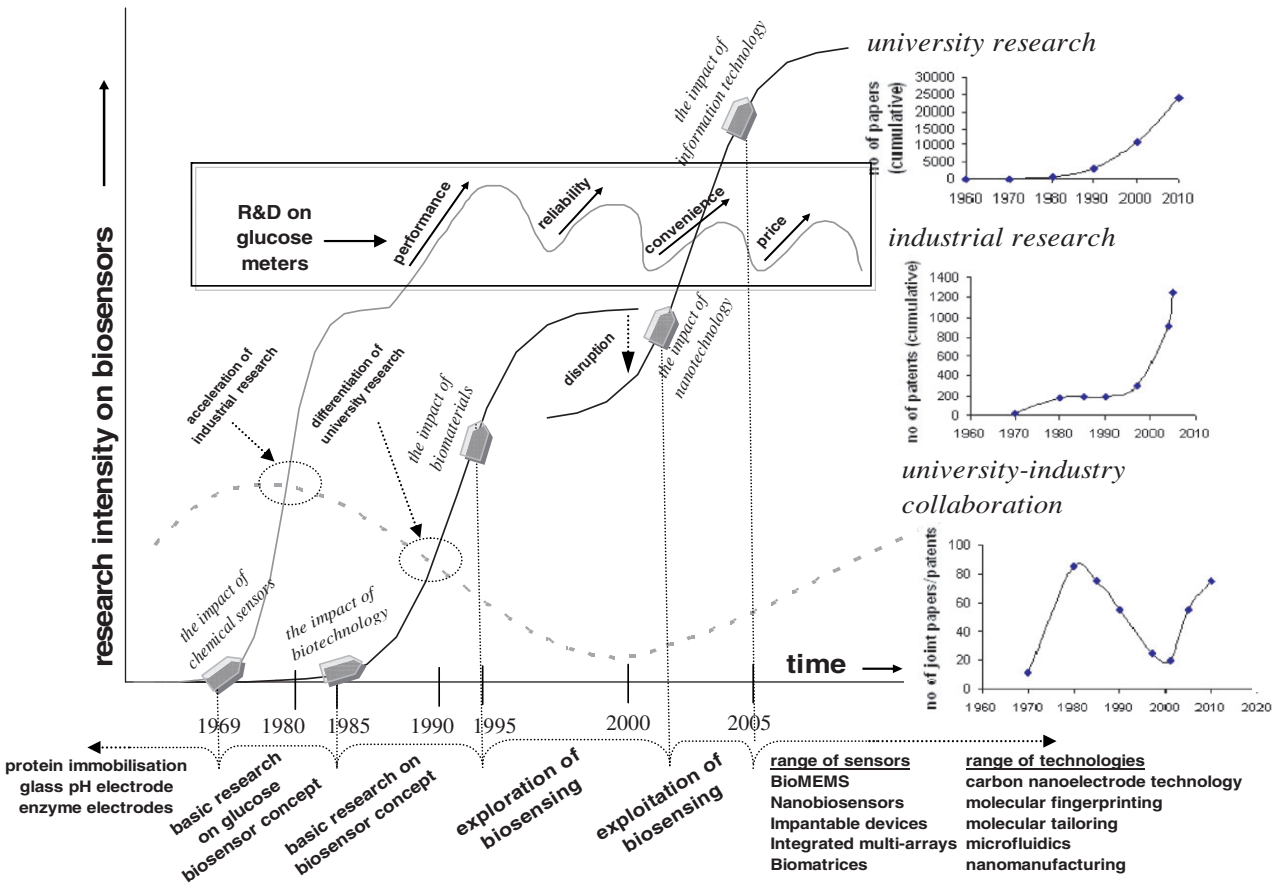

Figure 2. The biosensor technological frame as structured by the paradigm shift of diagnostic glucose monitoring or ''glucometry'' through (a) the technology performance roadmap (gray continuous line) referring mainly to the ''context'' of innovation, that is, the socio-technical perspective drawn by the industry (via a cost reduction trajectory as depicted with the inset) on the basis of the environment in which the innovation emerged, and the effect of that environment on the technology evolution; (b) the technology evolution (black continuous line) referring mainly to the ''content'' of innovation, that is, the scientific perspective drawn by the academic research on the basis of strategic merging with chemical sensors, biotechnology, biomaterials, nanotechnology and bioinformatics. The disruption that occurred around 2000 signifies the accommodation of nanotechnology-based design and production that not only re-boosted biosensing (from both scope and applicability) but also revived the university-industry collaboration (dashed gray line). The publication patterning has been constructed using the methodology of Lee & Su (2011) for mapping scientific knowledge.

(iv) increasing the market share with less costly formats and processes.

When the marketed technologies became comparable in most critical aspects, the real clinical needs for detecting unsuspected hypoglycemia, neonatal screening and adolescence diabetes monitoring urged the industry to look into the science push-pool for alternate testing, giving rise to minimally invasive glucose monitoring (Supplementary Figure 3). Knowing that this shifting was not enough to maintain the competitive advantage, noninvasive ''glucometry'' rapidly became the new target of the industries, enabling the renewed intensification of university-industry relations seen today (note the increase in the number of university-industry joint papers/patents after 2000 in Figure 2).

The changing structure of the scientific frame

Within the techno-economic transition led by the ''glucose sensor'', the academic community realized the innovation as yet another advancement, more or less ''linear'', of the already well-established chemical sensor format: using the same instrumentation and experimental design, the chemical reaction was replaced by a biochemical one to produce the ''enzyme electrode'' or ''enzyme probe''. When Guilbault and Montalvo, for example, reported the first potentiometric enzyme sensor in 1969 – urease immobilized, or, as the authors stated, ''insolubilized'', in a polyacrylamide gel held over the surface of a monovalent cation electrode (Guilbault & Montalvo, 1969) – Guilbault had already completed more than 10 years of research on chemical sensors.

Quite often, university-fostered innovations are incremental, that is, they originate from technological exploitation of available knowledge reserves while they are facilitated through the transfer of tacit knowledge. One of the key reasons why technological progress often proceeds along certain trajectories is that the existing technology and its design has already benefited from all kinds of evolutionary improvements, offering an established research environment in terms of accumulated knowledge, capital outlays, infrastructure and available skills (Kemp, 1994). Therefore, understanding technical change and the creation of a new scientific domain implies creating insight in the relation between incumbent technology and the incumbent (innovation) system in relation to the emerging technology and the emerging innovation system. The following example clarifies why insight in system dynamics and activities is more useful than the study of the system structure alone. Within a time span of 15 years (1990–2005), the criteria used were (a) number of university publications, (b) citations (self-citations exempted) and (c) number of patents (as indicators of a national market strategy). Both Germany and the Netherlands supported biosensor development within their biotechnology portfolio and hosted prominent biosensor research groups; however, Germany is considerably more successful in terms of diffusion rates. A possible explanation lies within the German innovation system that performs best in more traditional technologies, giving incremental advancements rather than radical innovations (Kaiser & Prange, 2004). Given their long history on chemical sensors and actuators, to no surprise did the German groups adopt early non-electrochemical platforms, as surface plasmon resonance and quartz crystal microbalance, providing the suitable electronics-biology interface that linked discovery with utility, that is, they attached actual applicability and marketability to a product that remained (and in certain cases still remains), more or less, for academic consumption only. Within this context the electronic nose and tongue emerged, along with the first university-derived biosensor product, the Automated Water Analyser Computer Supported System (AWACSS) for multi-analyte water monitoring (Tschmelak et al., 2005a, b). The development of the latter followed a problem-solving scheme that remained faithful to its chemical sensor basis (Siontorou & Batzias, 2010).

The drive of the innovation system was indisputably the substitution of a biochemical for the chemical reaction (Palchetti & Mascini, 2010), which actually paved the way for the exploration of natural chemoreceptions or the exploitation of its concept, that is, of the analytical sensitivity and selectivity of biological moieties. Several articles of early to mid-1980s point to the analogy of biosensing with natural olfaction (see e.g. Krull & Thompson, 1985; Krull et al., 1989; Thompson et al., 1986). This analogy gave the necessary incentive and theme differentiation for pulling researchers to the biosensing quest, luring the academia with the advantages of (i) the tremendous prospects related to nature-mimicking, (ii) the already acquired knowledge and expertise on sensor configurations that could be easily adapted to biosensor design and (iii) the vast opportunities revealed on the miniaturization of the bioanalytical laboratory, which in addition to the impact of high-throughput screening, opened up the possibility for multi-analyte assays by micro-dimensions of biosensors.

Needless to say, the scientists that have gained expertise on enzyme electrodes became the leaders of the early biosensor groups that were employed within the chemistry departments (especially within analytical chemistry laboratories) of various universities worldwide (Supplementary Figure 4). Diverse patterns of emerging systems of innovation were seen as an outcome of mutual interaction of academic heritage, mostly organizational path-dependency, with new competencies that fitted perfectly within the knowledge-accumulating institutions. The range of enzymes employed and their physical properties soon enabled the transfer of the original electrochemical sensor design to alternative platforms. This gave rise to ''optrodes'' and rapid immunoassays, or extended the range of available transducers and transduction schemes toward multi-analyte screening. This included the electronic nose and tongue with their sound basis on sensors, and artificial membranes and Langmuir-Blodgett films, rooted deeply in biophysics and physical chemistry. The latter, especially, became rapidly very popular on the premises that the reconstruction of natural membranes provides the suitable environment for enzyme functionality.

Highly regionalized as regards research and early exploitation, and highly globalized in terms of development and knowledge dissemination, these systems have contributed to the aggregation of clusters of scientific competence which enhanced the technological innovative capacity at micro-levels (i.e. at the university level). As a result, codified knowledge, captured through publication and citation

analysis, presents only a partial picture of the formation of the academic innovation system. As a matter of fact, slow knowledge dissemination mechanisms in the pre-Internet period and the limited availability of resources favored knowledge flows through both personal communication channels and long-sustained collaborations and interdependencies (some post-graduate affiliations) linked to ''spin-off'' activity seen at Supplementary Figure 4. The upshot was an elaborate lattice-like structure of relationships built on short cognitive distance that gave rise to a slow nested process of knowledge sharing within a cluster. This was subsequently independently employed to produce new knowledge (at a deeper or an extended level) that fed the cycle again, each time promoting the initial concept (or line of product) one step forward. Four interacting factors facilitated the formation of these clusters: natural propensity, complementary resources, formation of groups in the innovation system and actor-network interactions. The relationships between these determinants are critical in locating concentrations of innovation clusters that were not always regional. While Japanese groups were concentrating on semiconductor technology, European and US groups were engaging in miscellaneous biosensor formats and experimental designs.

The capabilities that have developed over time were dispersed across various clusters that were unaware of the existence of complementary capabilities or were working as competing groups. The clusters formed in the United States were subjected to competing demands from technology areas, as well as, from industry links which acted as powerful forces in determining the eventual success of clusters (Siontorou & Batzias, 2010). In Europe, the creation of a suitable ''innovation climate'' was fostered by collaborative research arrangements between knowledge providers, when the opportunity of EU financing was raised on the premises of European collaboration, especially with developing countries, such as the Copernicus (1994–1997), the Inco-Copernicus (1996–1998), the Biosensors for Environmental Technology (BIOSET) (1997–2000) and others. There appeared ample opportunities for clusters to develop critical technologies simply by forming cross-institutional linkages (Schmoch, 2007). The synergies drawn managed, to some extent, to consolidate the work of those operating within collaborating innovation clusters and contributed effectively to biosensor development. Yet, the major driver was fund raising. The groups which failed to gain funding were either absorbed into successful clusters or remained unsupported. The collaboration between clusters promoted largely the concept of biosensor applicability, shifting the technological frame (Figure 2) from exploration to exploitation, which would become feasible only at the nanotechnology era. Applicability (as a tangible concept and not a possibility) was implicitly associated with marketability (i.e. the direct translation of academic innovation to product), which set off a new period in biosensor innovation problem-solving. The biosensor advantages over conventional technology, more a theoretical discussion than a proven ability, portrayed in every biosensor paper published, had to be somehow implemented, employing skills and competences beyond the capabilities of analytical or solid state chemistry. Biomedical, environmental and industrial aspects necessitated solutions on sensitivity, selectivity, interference and cross-reactivity, stability, integration, miniaturization and manufacturability. In fact, while tremendous improvements have been achieved over the years in the manipulation of biological signals, the common practice of matching first naturally occurring biomolecules to target analytes and then using the intrinsic properties of the biological system in device development has not been as successful as expected, in terms of performance (selectivity and sensitivity) and stability (see e.g. Hellinga et al., 1998; Marvin et al., 1997; Piervincenzi et al., 1998). Thus, it became obvious that while the structure and function of the natural biological macromolecules is impressive, fabrication of biomaterial-based devices is inherently limited by the available diversity, cross-reactivity, surface topography and stability problems of native proteins (see e.g. Aizawa et al., 1998; Gilardi et al., 1994; Hock et al., 2002). This realization has led some clusters to embark on the development of a new generation of biorecognition elements that are not naturally occurring but have been molecularly engineered and synthesized in the laboratory. Thus, research trends in biosensor design and fabrication have been shifted from modifying sensing surfaces toward the engineering (design and synthesis) of suitable interfacial recognition nanobiomaterials. The impact of nanotechnology (Figure 2) is also evident in transduction schemes and integration strategies, making miniaturization feasible, which, in turn, caught anew the interest of industry.

Regardless, the major posit of low-cost substitution of conventional analytical equipment, that is, the cost-reduction strategy bequeathed to biosensors by the glucose monitoring frame, has largely failed to persuade industry (Luong at al., 2008), although it is not totally fallacious. Compared to other bio-research areas, biosensor technology requires fewer resources to provide rapidly a significant academic output (Siontorou & Batzias, 2010). In theory, and, to a certain extent in practice, as well, any analyte can be detected by a biochemical system, which can be coupled to a suitable transducer. It follows that the same transducer can host various biochemical systems, extending the range of analytes that can be detected (i.e. the ''applicability'', in the academic sense), while numerous optimization strategies can be devised at the bench scale (by highly trained personnel), even if they cannot be actually implemented at the industrial scale.

The continually updated content of the scientific frame

Biosensor science and technology were widely acknowledged as having a larger impact to many areas of scientific research (such as physics, chemistry, material sciences, biology and engineering) and potential for many technological applications (such as healthcare and life sciences, energy and environment, electronics, communications and computing, manufacturing and materials). Although there is no sharp distinction between them, biosensor science is concerned with understanding some phenomena (such as surface tension/properties, quantum effects, molecular assembly) and their influence on the properties of materials. Whereas biosensor technology aims to exploit these effects to create structures, devices and systems with novel and significantly improved

properties and functions due to their size. At times, the science part, rationally, preceded the technology part, even with very short lag times, but usually technology preceded scientific explanation. Much of the pioneering and fundamental research had a strong edisonian character. Through trial and error, the many possibilities of the biosensors have been disclosed. Following the enzymes, antibodies have been implemented, establishing a new, prompt (less than 5-min assay), direct and label-free (especially of the electrochemical type) immunochemical platform. Biosensor exploration rapidly moved to tissues, whole cells, microorganisms, receptors, protein channels, DNA and many other proteinaceous moieties that have been extensively studied as per their suitability for sensing and their potential for integration, establishing a curiosity-driven era.

Inevitably, different research streams emerged and developed that gave rise to numerous paths. Their content and dynamics are difficult to track at a time when they are struggling to define what they are, what they include and exclude, and how they organize and classify themselves internally. Frequent re-labeling and flexible taxonomic criteria make it difficult to use worldwide databases, such as those of the ISI/WoS, to track the dynamics of science and technology. The term ''biosensor'' is systematically used only after 1985, while a partial definition (related to electrochemical devices only[1]) is given in 1999 (Thévenot et al., 1999). Even today there seems to be no consensus on either terminology or its scientific connotation; what constitutes or not a biosensor (or biological sensor, or biological sensory system, or bioprobe, or biosensoric device, or bioanalytical device, etc.) has yet to be resolved, as the IUPAC definition erroneously rejects (i) biosensor systems that use some kind of sample pretreatment (and most devices do) as dialysis or pre-concentration, (ii) biosensor systems that utilize some kind of immobilization matrix that conveys (and even amplifies) the biochemical information to the transducer, (iii) biosensor systems that employ biomimicking moieties, such as liquid crystals that resemble the physical chemistry of lipid molecules but at higher robustness, (iv) biochemical assays, as well as, the glucose meters which are based on disposable enzyme electrodes, and most of the immunological schemes that require more than one step (Scheller et al., 2001).

Using the data search strategy proposed by Mogoutov & Kahane (2007), the intelligent agent developed by Batzias & Markoulaki (2002) and the aid of seven biosensor experts (to determine technology boundaries and growth and to decide on the incorporation of new terms and concepts), the dynamics of the research paths have been examined. In general, these different paths suggest that the technological dominances are derived from a sociocultural evolutionary process consisting of variation, selection and retention (Campbell, 1969). Variation emerged through stochastic technological breakthroughs which led to technological rivalry between alternatives. The groups with the higher absorptive capacity progressed the sector rapidly, adopting developments on chemical sensors, innovations on biomaterials and the new tools provided by biotechnology, nanotechnology and, later, by information technology (Figure 3). The great variety in technological trajectories at the beginning of the scientific sector has been, thus, gradually reduced to those (few) trajectories that concentrated research clusters toward overcoming technological bottlenecks. This convergence toward common technological trajectories and practices within the sector contributed to higher productivity of knowledge, thereby resulting in extending the available resources required to generate new technological trajectories that, in their turn, raised the overall variety in the system.

Knowledge production followed a strong problem solving strategy, mostly divided in three optimization aspects: transduction, interfacing (the problem of bioelement immobilization), cost (referring to cost of research), regardless of the paths' orientation (transducer-oriented, bioelement-oriented, analyte-oriented, biosystem-oriented, etc.). It is worthwhile noting that the in-depth analysis of the content of biosensor innovation presented herein relied more on the expert's opinion and less on data mining techniques.

The scientific map of surface plasmon resonance biosensors (Supplementary Figure 5) reveals five trails: (i) performance optimization (decrease of refractive index resolution and increase of sensitivity), (ii) functionalization improvement (resolving the trade-off of strong ligation versus optimal kinetics), (iii) non-specific binding (interference) reduction, (iv) miniaturization/compactness and (v) high-throughput fabrication with lower cost. Amperometric systems followed a similar research strategy toward fault remediation, focusing on the significant drawbacks of the well-established and widely used transduction system: sensitivity improvement and interference minimization. The so-called first generation biosensors (ca. till 1985), in which detection was based on the shuttle mechanism, that is, on freely diffusing redox species, had many shortcoming (Dzyadevych et al., 2008), as: (i) the electrode geometry effect on sensor response that necessitated electrode pre-treatment for obtaining reproducible surfaces, (ii) the effect of particle mass transfer through biocatalytic and semipermeable membranes on biosensor response, (iii) the electrode drift and (iv) the matrix effect. To avoid the first drawback, the surface was polished and processed to heat or chemical treatment. These techniques often resulted in improvement of response reproducibility; however, this effect vanished at long-term exposition of an electrode to the tested solution. A promising approach to overcome this obstacle was the chemical modification of the surface (Albery & Hillman, 1984; Lorenzo et al., 1998), using high-conductive organic materials (Albery & Craston, 1987; Li et al., 1999) or redox-polymers (Cosnier, 1999), that gave rise to the ''second generation'' of biosensors, which utilized electron mediators (electron wiring); the resulting platforms exhibited, however, low stability, prompting the use of conductive polymers (Gerard et al., 2002).

[1]Thévenot et al. (1999, p. 2334): ''An electrochemical biosensor is a self-contained integrated device, which is capable of providing specific quantitative or semi-quantitative analytical information using a biological recognition element (biochemical receptor) which is retained in direct spatial contact with an electrochemical transduction element. Because of its ability to be repeatedly calibrated, we recommend that a biosensor should be clearly distinguished from a bioanalytical system, which requires additional processing steps, such as reagent addition. A device which is both disposable after one measurement, i.e., single use, and unable to monitor the analyte concentration continuously or after rapid and reproducible regeneration should be designated a single use biosensor.''

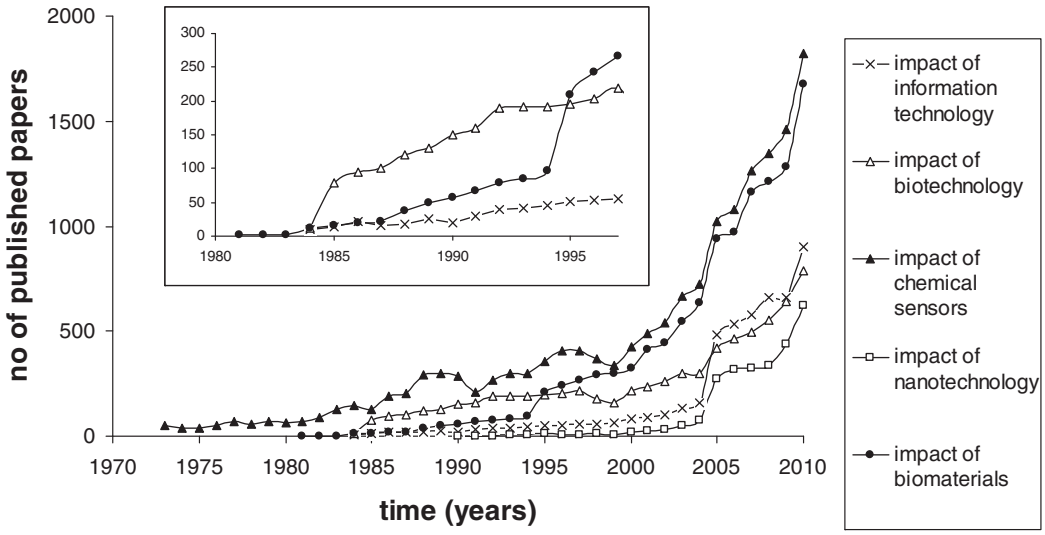

Figure 3. Biosensor research trends (1973–2010) putting emphasis on the clusters with the higher adsorptive capacity, that is, those that adopted/adapted rapidly scientific breakthroughs to biosensor formats.

In the majority of biosensor breakthroughs, even by early 2000, significant knowledge gaps limited the exploitation of the concept. The mechanism of signal generation was largely speculated (or erroneously presumed), prohibiting, thereby, effective optimization (e.g. signal amplification or elimination of non-specific adsorption). When thin film characterization techniques evolved, the science-part has been enriched and the respective technology part has been revisited. The knowledge acquired encouraged the shifting of the research strategy toward more oriented and systematic frameworks, enabling biosensor exploitation. The unsolved problems have been studied again by interdisciplinary teams, and thesis-driven (rational) research has not only boosted the number of academic publications but also revived industrial interest. Nanotechnology and information technology have mostly impacted the sector, as they came to solve transduction and manufacturability problems. Many issues are still pending, for example, protein interactions with the transducer surface (the interface problem) are yet to be resolved (e.g., Lin et al., 2011), while diffusion remains a controversial issue (Wheatley, 1998).

In effect, biosensor technology is built upon many sciences and technologies and is inherently complex. In biosensors there can be both, a mechanistic and a biomimetic version; while the former is more materials science and microelectronic inspired. It has new or significantly improved mechanical, electrical and chemical properties or functions. The latter leans toward biotechnology, having control of biological systems to achieve desired and designed outcomes. In all views, however, academic research pursues the technology explanation (in-depth and high granularity) to acquire the knowledge required for innovative problem solving in fault detection and remediation.

The methodological combined framework

The Analytic Hierarchy Process (AHP) is a comprehensive framework designed to cope with the intuitive, the rational and the irrational when decision makers make multi-objective, multi-criteria and multi-factor decisions, with or without certainty, about any number of alternatives (Harker & Vargas, 1987). In the technology management area, there have been several studies applying the AHP approach to the evaluation of technologies or for roadmapping sectors. In those studies, the hierarchical model for the evaluation and assessment of technologies is constructed on three main levels, namely: objectives, criteria (and in some cases sub-criteria) and technology alternatives. The series of comparative judgments is analyzed to determine the relative impact of alternative technologies on the objective. However, to enhance the dynamic and flexibility features of the AHP framework so that it could be adopted for roadmapping academic research on biosensors. The conventional approach of structuring a hierarchical decision model with the technology alternatives at the lowest level is not rigorous enough, because the whole series of comparative judgments needs to be repeated every time a change in the development of any particular technology is captured or a new technology emerges, both are frequently seen in academia as the output of the continuous knowledge creation process.

A new structure of the hierarchical model approach is proposed by replacing technology alternatives at the lowest level with technology internal barriers (Figure 4), modeled through fault tree analysis (FTA) relying on fuzzy reasoning to count for uncertainty, while (i) criteria refer to performance and/or quality specifications set by the intended use of the technology, and (ii) factors refer to aspects of technology directly influencing the criteria.

The modified AHP framework

The AHP framework, as implemented herein, is structured by obtaining strategic information on the development of the various technologies (or research paths) fostered within academia and then using this information to evaluate the value of each technology based on the impact of its characteristics on the sector's objective (defined by the technology creators and the market) in each time period (Supplementary Figure 6). The Delphi method and AHP are

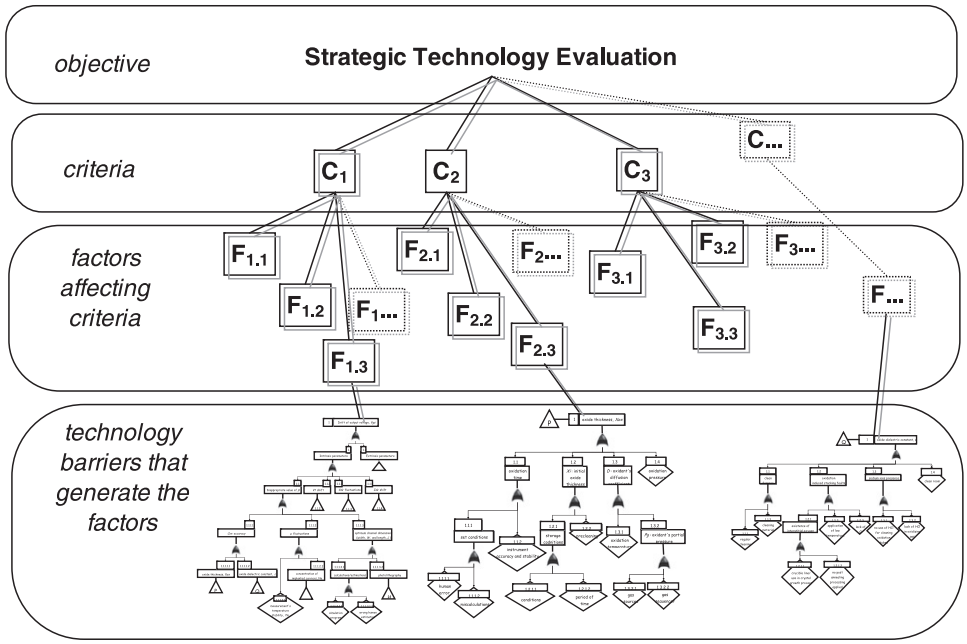

Figure 4. Overview of the hierarchical model approach; technology alternatives at the lowest level with technology internal barriers identified and quantified by means of fuzzy fault tree analysis.

combined as two parts of the framework. Delphi method is used for obtaining the strategic information about the future development of technologies from a group of experts, selected from academia, involved in developing these technologies, while AHP is used for evaluating the impact of these technologies. A series of judgment quantifications is also obtained from the experts in implementing the research output into products.

By tracking the development progress of each research path over time (stage 2), any changes observed on the value of technology, especially in the case of inferior technology paths (classified as such in the frame of reference) achieving dominancy (stage 6), would represent the degree to which the biosensor society perceives the values on the improvement of technologies (stage 9). The development of the AHP model is achieved in three steps: (a) technology characterization (stages 1–11), (b) hierarchical modeling (stages 12–15) and (c) technology evaluation (stages 16–21), relying on fuzzy fault tree analysis (stages 15 and 16). The processes of identifying components and quantifying the relative importance of components in each hierarchy are based on a set of criteria and the corresponding factors (stage 12). The experts provided comparative judgments for each set of criteria and factors, finally selected those factors that weigh most for each criterion (stage 14) (Table 1).

The criteria of performance (C1) and reliability (C3) are commonly applied in analytical chemistry, although their straightforward implementation in biosensors is not easy. In electrochemical sensors, for example, the term ''stability'' refers to both the structural stability (shelf life) of the bioelement (F1.15.04) and the reproducibility of response (or precision F3.04.11), partly attributed to the electrode drift (F3.04.12) (Thévenot et al., 1999), whereas ''operational stability'' (F.3.14.02) and bioelement stability (F1.15.04) are frequently used interchangeably. Nonetheless, the term is yet to be defined and evaluated, as most devices have not been tested extensively at real operation conditions[2]. Geometry (C2) plays an important role in miniaturization (F2.28) and also in facilitating the free association of the analyte with the bioelement, while restricted access of intereferents; in most set-ups, the transducer is the main determinant, as the size of the bioelements is extremely small, but in integrated microchips these differences fade (F2.39). Criterion C4 refers to the cost of university research. Although the resources required to upgrade instrumentation in a university laboratory are scarce, inter-university collaborations mitigate this problem. Serviceability (C5) includes the installation and maintenance complexity of the transducer and supporting equipment (F5.01.07). Academic research favors the interchangeability of components (F5.04.14) and handmade configurations for extending the range of experimental formats and allowing manipulability. Flexibility (C7) favors mainly the incorporation of various bioelements on a transducer type (F7.15.12), giving rise to numerous biosensors and/or optimization options.

The inputs were analyzed to determine the relative priority of criteria as well as the relative importance of factors associated with each criterion (stage 15). The results indicate that among the seven criteria, the experts placed their emphasis on performance, reliability and manufacturability. This result also reflects the researchers' mission in

[2]These misconceptions on terminology may be justified by the scale that these devices are usually developed and tested within the university environment. Because bench-scale devices (experimental set-ups or prototypes) exhibit limited operation time at carefully controlled environments, lifetime is indeed determined primarily by the structural stability of the biological element, and secondarily by the effect of the electrode drift on the calibration curve (commonly, whatever strikes first). At real operation, however, extreme conditions may prevail (e.g. temperature, unanticipated interference, overflows, pH), even as abrupt changes, that may be proven detrimental to the device itself, let alone its operation (see e.g. Batzias and Siontorou, 2005; Khanna, 2007; Siontorou et al., 2010).

Table 1. Sets of criteria and respective factors used in hierarchical modeling.

Criteria	Factors[a]
C1: performance	F1.03.02: specificity F1.03.10: sensitivity F1.03.18: detection range F1.03.22: selectivity F1.06.12: ruggedness F1.06.17: robustness F1.07: assay simplicity F1.13: signal amplification F1.15.04: bioelement stability F1.15.14: functionalization
C2: geometry	F2.28: miniaturization F2.32: intended use F2.39: intergradability
C3: reliability	F3.04.03: accuracy F3.04.11: precision F3.04.12: drift F3.14.02: operational stability F3.15.06: signal scattering F3.24.07: nonspecific adsorption F3.24.08: cross-reactivity
C4: cost	F4.01.26: cost of instrumentation F4.02.02: cost of fabrication F4.11.09: cost of reagents F4.13.05: cost of knowledge management F4.17: cost of quality control
C5: serviceability	F5.01.07: installation and maintenance complexity F5.04.14: interchangeability of components
C6: manufacturability	F6.02.05: design ruggedness/robustness F6.02.21: simplicity F6.03.05: versatility F6.03.12: scalability F6.14: mass production
C7: flexibility	F7.15.12: physical moldability F7.19: tailoring F7.23.04: adaptability F7.23.09: upgradeability

[a]Code numbers are provided by the dedicated Knowledge Base developed by the authors, based on the hierarchical taxonomy performed by the experts; terminology is based, whenever applicable, on the International Union of Pure and Applied Chemistry Compendium of Chemical Terminology – the Gold Book.

maintaining the biosensor state-of-the-art as a performance leader in diagnostics.

Technology internal barriers

Technology evaluation (stages 16–21) is commonly thought of its societal impact, that is, its ability to integrate with relevant impacts and markets. University research, on the other hand, may appear as chaotic and unorganized, guided more by expectations and visions and less by actual admissible product-shaping strategies. It encompasses a high degree of uncertainty in innovativeness, number of constituent technologies, manufacturing difficulties and institutional changes. From the market view, there is certainly a lack of insight on the process of idea development and potential device application that concomitantly leads to difficulties in ascribing market value to a given idea. There is often a misperception of the valuation of university-derived technology resulting either to undervaluation (and early rejection) or to overvaluation (and market failure). Academia, on the other hand, profiles technologies instinctively, by means of ongoing dynamics, and although it is sometimes biased toward easily quantified outputs (decision node D), it perceives the innovation chain through concerted research on ''real'' obstacles. These obstacles can be considered at various levels, starting from the micro-level and ending up at the macro-level. Given the multitude of barriers, the externalities of the system of innovation (stage 10) and its affordance structures (stage 8), the identification of the technology barriers for each alternative path (stage 15) and their decomposition into micro-level problems/gaps/obstacles (stage 16) necessitates meta-analysis on the self-regulating paths in order to capture the effective deployment of technology.

Looking into the published university output can only serve effectively certain aspects of this task, such as problem classifications, categorization of research efforts or trends, whereas the valuable piece of information is linked more to the implied concepts that the overt message. In fact, research efforts are mostly concentrated around the challenges (obstacles/faults/defects), either in terms of causality (a question-driven process toward requiring in-depth investigation, parameter identification and high granularity level information retrieval) or resolution (an open-ended empirical process, usually only partially supported by in-depth information). Clearly, the identification of the internal technology barriers for technology evaluation requires a suitable tool that supports a hierarchical structure to fit the AHP framework and to handle rigorously system functions and cause-consequence relations.

The analytical techniques initially considered for implementing stage 16 were Fault Tree Analysis (FTA), Failure Modes and Effects Analysis (FMEA), Hazard and Operability (HAZOP) study and Management Oversight and Risk Tree (MORT) Analysis. The former, being the only one built on a deductive basis (Adler et al., 2003), is more compatible with AHP as it allows some *a priori* categorization at the first levels of its dendrtitic structure. The fuzzy version of FTA (FFTA) was further selected to account for uncertainty (Batzias & Siontorou, 2005; Siontorou & Batzias, 2008; Siontorou et al., 2010).

The tree (i.e. the content part of analysis) is structured in a top-down direction by deduction, identifying the events that directly contribute to the appearance of the top events, the immediate causes of these events and so on, until the final sources of error (usually at deep phenomenological levels) are reached. The tree may be revised by induction (bottom-up), provided that the necessary information is available. This scheme, involving top-down analysis and bottom-up verification, permits the introduction of deeper knowledge into the surface knowledge level, usually characterizing conventional FTA (Batzias & Siontorou, 2005; Siontorou & Batzias, 2008; Siontorou et al., 2010). Each branch has been analyzed to the deeper possible level of knowledge (ultimate event) to facilitate the identification of those technology barriers that are due to a lack of either appropriate technology or knowledge. It is worthwhile noting that for avoiding the problem of circularity, that is, when the ascendant event becomes the cause of its descendant, frequently observed

at extended trees, decomposition procedures have to be employed, similar to the ones suggested by Camarinopoulos & Yllera (1985; 1986).

At its final form (in content and architecture), the tree is quantified through a modified 4-stage Delphi method by using the experts' opinion to partition the space of variables for each node and determine the fuzzy rules serving as an inference engine for diagnosis (i.e. the formal part of the analysis). Running the fuzzy FTA bottom-up, using the final events (alone or in combination) that are considered most responsible for the appearance of the top event (as per strength and frequency), a crisp number is calculated for the latter and compared to a pre-specified threshold. Likewise, based on a nested loop mechanism, all combinations of value ranges are examined, until the most critical events, that is, those that produce the higher value of the top event, are determined.

Each tree node has been quantified using ranges of values expressed by the linguistic terms (fuzzy sets) *low*, *medium*, and *high*. The degree (or weighting) that any value belongs to a fuzzy set is represented by the membership function, which takes values between 0 and 1. Each term is schematically presented as a triangle, formed by the lower and higher value that the node may exhibit, peaking at its most probable or usual value (membership function 1). The overlapping of the space of variables depends on the uncertainty of the measure or effect of the event. Working in a tree-like structure, the quantification of each node depends on its direct descendant nodes (i.e. its immediate causes), given by a series of fuzzy rules (considering combinations of values at varying significance indices) that determines the output of the node as fuzzy set. Using the inference engine, the basal events are entered as crisp numbers (arithmetic values) to produce as output the fuzzy sets and membership functions of their immediate ascendants, which become the input (without defuzzification) for the next level, and so-forth until a fuzzy set (expressed as crisp value after defuzzification) for the top event is yielded.

Framework evaluation – ISFET-based biosensors

The proposed methodology has been evaluated retrospectively on the ion selective field effect transistor (ISFET) technology output at early to mid-1990s. The experts employed had either participated or lead university research groups with high activity on ISFETs at that time. ISFETs have been selected because (i) the extended use of field effect transistors in chemical sensing provided a well-established scientific and technical background at a high degree of technology exploitation that most biosensor groups familiarized with easily (stage 3), and (ii) ISFET technology represented at the time (i.e. at a time that nanotechnology had not been in any practical applicability phase), along with probes (i.e. of the catheter type) the only formats feasible enough for clinical diagnostic and portable environmental device applications (stage 9). The technology characterization (stages 1–11) revealed that the relatively short-term instability or drift (factor F3.04.12), which can also be interpreted as lack of repeatability (Fraden, 1996), imposed severe limitations on sensor reliability (criterion C3). Drift is essentially equivalent to instability in the DC operating point of the ISFET. Therefore, depending on the particular mode of operation[3], the characterization of the drift behavior involved measuring the temporal change in the ISFET's DC operating point. Looking extensively into the literature of 1990–1994, the two modes of operation were essentially equivalent inasmuch as sensor reliability is concerned, because the ratio, which denotes the ratio of device response arising from pH variations to that arising from drift, has been shown to be the same in both modes of operation (Janata & Huber, 1985).

The drift data published for pH-sensitive biosensors revealed that over time durations, in the order of seconds, the gate voltage in the feedback mode of operation exhibited only random fluctuations, attributed mainly to fundamental sources of noise (Atkinson et al., 1994) and/or DC power supply variations (Mittermeier et al., 1995). Furthermore, the time constant characterizing drift was reported of the order of several hours (Dun et al., 1991), which implies that over a time interval of several seconds, drift was negligible. The relatively slow, temporal variation characterizing long-term drift (stage 15) formed the basis of several analytical approaches proposed at the time to correct instability, using the Shockley model (Allen & Holberg, 1987) for managing and/or manipulating sensor's behavior:

$$I_{ds} = \beta\left[\left(V_{gs} - V_t\right)V_{ds} - \frac{1}{2}V_{ds}^2\right] \Rightarrow V_{gs} = \frac{I_{ds}}{\beta V_{ds}} + V_t + \frac{1}{2}V_{ds} \quad (1)$$

where, β is the geometric sensitivity parameter, V_{ds} is the drain to source voltage, I_{ds} is the drain to source current and V_t is the threshold voltage. Tree synthesis (stage 16) was based more on dependence, drawn from the theoretical (or science) aspects of ISFET used in its modeling, and less on causal relations. Taking the V_{gs} parameter value as system output, this work addressed the modeled (intrinsic) parameters (node 1.1) related to ''drift of output voltage, V_{gs}'', taken as ''top event'', to highlight: (i) the critical technological problems and (ii) the role of the fabrication/assembly processes (used at mid-1990s) in delivering reliable devices. The small dimensions of ISFETs demanded strictly controlled fabrication and extreme accuracy in handling for yielding reliable bench-scale devices. It was quite later that ''operability'' (node 1.2) was included in the research agenda (Batzias & Siontorou, 2005).

The top event has been analyzed (i.e. decomposed) into first-level faults related to or arising from the constant β (node 1.1.1), the threshold voltage, V_t (node 1.1.2), the drain to source voltage, V_{ds} (node 1.1.3) and the drain to source current, I_{ds} (node 1.1.4) (Figure 5). Apparently, the precise estimation of constant β (node 1.1.1), related to oxidation, implantation and photolithography processes, depends on manufacturing parameters (see e.g. nodes 1.1.1.2.2, 1.1.1.1.1.4, 1.1.1.1.1.3.1 and

[3]ISFETs commonly operate in two modes. In the feedback mode, the drain current, I_{ds}, is kept constant by applying a compensating feedback voltage, V_{gs}, to the solution side of the gate (e.g. using a double junction type calomel reference electrode). Ideally, the changes in this feedback voltage correspond to ISFET's response to variations in the ion concentration. Alternatively, a constant gate voltage can be applied to the reference electrode using a DC power supply, in which case ISFET's response is characterized by changes in the drain current.

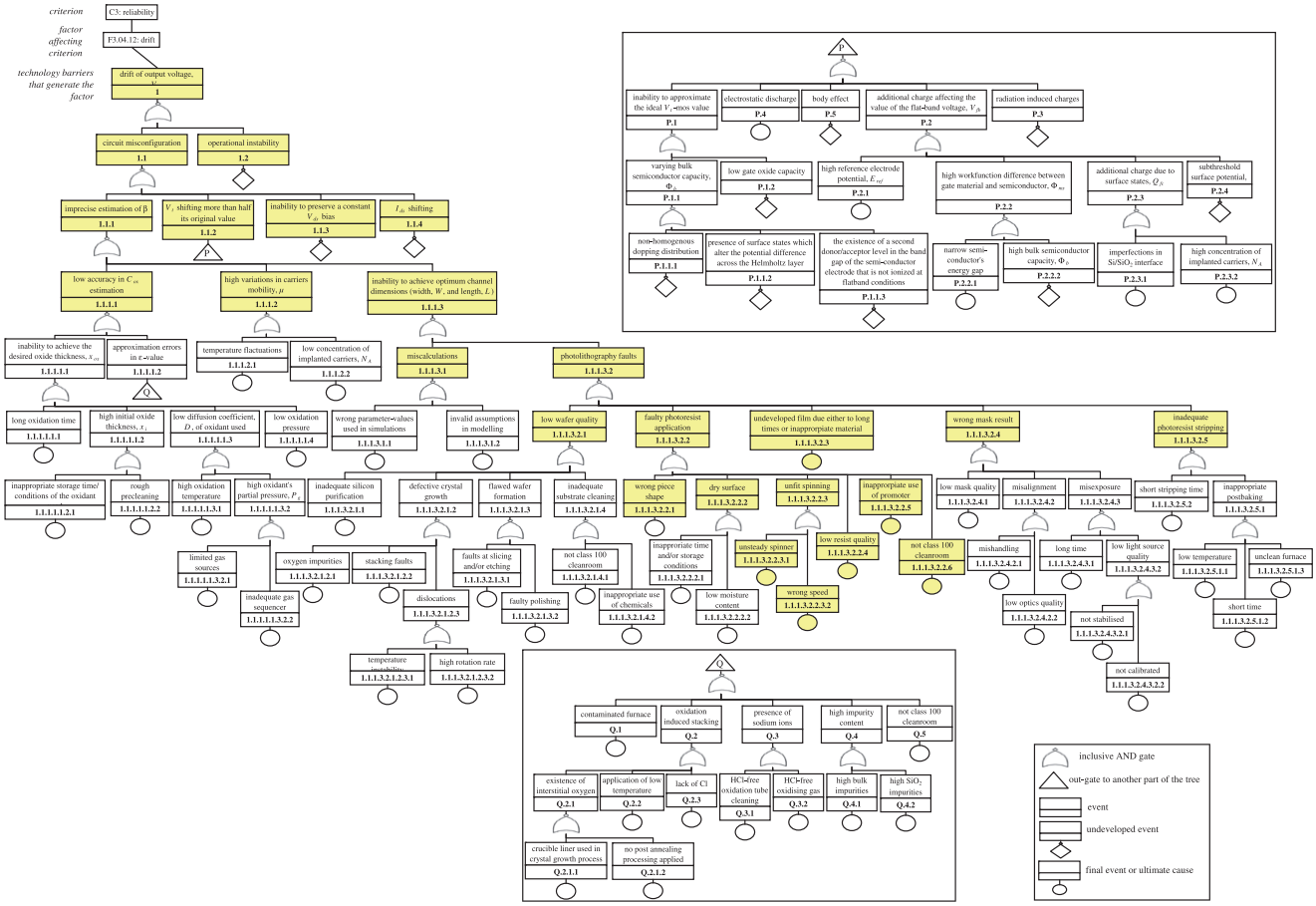

Figure 5. The qualitative upper part of the dependence fault tree (constituting the content part of the methodological framework) synthesized for investigating the causes of drift in ISFETs. Constant β (node 1.1.1) is given by $\beta = C_{ox}\mu W/L$, where C_{ox} (node 1.1.1.1) is the gate oxide capacity per unit area, else $C_{ox} = \varepsilon/x_{ox}$, where ε is the gate oxide dielectric constant (node 1.1.1.1.2) and x_{ox} is the gate oxide thickness (node 1.1.1.1.1). Note that, because the fault behavior characterizes the FET's current in the non-saturation region, the channel-length modulation due to changes in the drain to source voltage, V_{ds}, is ignored. The threshold voltage, V_t (node 1.1.2), is given by $V_t = V_{t-mos} + V_{fb}$, where $V_{t\text{-}mos}$ stands for the voltage threshold of an ideal MOS. The first parameter is calculated by the following: $V_{t-mos} = 2\phi_b + (Q_b/C_{ox})$, $\phi_b = (kT/q)\ln(N_A/N_i)$ and $Q_b = N_A 2\phi_b\sqrt{(2\varepsilon_{Si}q)}$, where q is the charge of an electron, ϕ_b is the semiconductor's bulk potential (node P.1.1) depending on carrier doping, N_A (node P.1.1.1) and Q_b, which is the bulk charge (Schepel et al., 1984; Weste & Eshraghian, 1993). The flat-band voltage, V_{fb} (node P.2), is given by $V_{fb} = E_{ref} - \Psi + x_{sol}\left[(\Phi_{Si}/q) - (Q_{fc}/C_{ox})\right]$, where Ψ is the surface potential, x_{sol} is the surface dipole potential determining the interface potential at the gate oxide/electrolyte interface and is considered to be constant, Φ_{Si} is the silicon's work function, considered to be constant, E_{ref} (node P.2.1) is the reference electrode's potential relative to vacuum and is considered to be depended on circuitry and the work function difference between the gate material and the silicon, Φ_{ms} (node P.2.2) is given by $\Phi_{ms} = -\left[(E_g/2) + \phi_b\right]$, where E_g stands for the semiconductor's energy gap (node P.2.2.1). The threshold voltage is also affected by the so-called ''body effect'' (Weste & Eshraghian, 1993) occurring in the case of series connected devices on a common substrate; the body effect factor, γ (node P.5), is expressed by $\gamma = (x_{ox}/\varepsilon_{ox})\sqrt{2q\varepsilon_{Si}N_A}$.

the Q sub-tree). Similarly, the gate oxide thickness, x_{ox} (node 1.1.1.1.1), depends on the oxidation time (node 1.1.1.1.1.1), the initial oxide on the gate area (1.1.1.1.1.2), the oxidation pressure (node 1.1.1.1.1.4) and the diffusion coefficient of the oxidant used (1.1.1.1.1.3); the initial oxide on the gate area could affect the calculations for determining the desired oxide thickness. The value of the dielectric constant, ε, (node 1.1.1.1.2 or Q), deriving from the insulating quality of the oxide, is susceptible to approximation errors at oxide contamination, commonly occurring due to furnace (node Q.1) or room (node Q.5) cleanliness. Furthermore, the variability of the processes used seems to affect negatively the value of ε. For example, hydrochloride ions were commonly used to sequester sodium ions either for cleaning the oxidation tubes or as part of the oxidizing gas. This treatment, induced, though, segregation and diffusivity phenomena (especially critical when high sensitivity was sought for), and sometimes it was omitted (nodes Q.3.1 and Q.3.2), leaving sodium ions (node Q.3) to degrade the performance of the sensor (Chovelon et al., 1992). Moreover, these processes may induce stacking faults (node Q.2) arisen from interstitial oxygen (node Q.2.1) already built in the wafer during the initial crystal growth process (Kriegler, 1973). Later, these processes have been replaced by dry furnace oxidation in pure O_2 atmosphere at 820 °C, wet furnace oxidation in an H_2O atmosphere at 600 °C and rapid thermal process oxidation in a mixed atmosphere of O_2 and N_2O (1:1) at 950 °C (Jaeger, 2001).

The shifting of the threshold voltage, V_t (node 1.1.2 or P), is also linked to fabrication processes. Non-homogeneous doping distribution (node P.1.1.1) could not permit the stabilization of the bulk potential (node P.1.1), thus misjudging the ideal voltage threshold (node P.1). Radiation (node P.3) has been also reported to induce charges in the device (Khazan, 1994; Sze, 1981), leading in the presence of excessive carriers. This effect was considered critical as it could alter the expected electrical characteristics

(conductivity), resulting in fallacious value of threshold voltage, *Vt*; thus, the device needed radiation isolation apart from thermal isolation, although the encapsulation method was difficult (Ho & Kratochvil, 1983).

Photolithography faults have been frequently reported to hamper the setting of optimal values for the dimensions of the gate channel (node 1.1.1.3.2). The initial surface should be as dry as possible (node 1.1.1.3.2.2.2) and free of organic contaminants. Because SiO_2 (commonly used at the time) is hydrophilic, a special pre-treatment should be carried out, such as 300 °C–1000 °C baking to dehydrate the surface, mechanical scrubbing or even the use of a promoter just before photoresist application (Bergveld, 2003), which added considerably to defect formation. Moreover, the shape of the substrate piece (node 1.1.1.3.2.2.1) determined the profile of the applied photoresist film (Damon, 1969) raising various problems; for example, non-round substrates made the film inhomogeneous due to turbulence in the photoresist caused by the corners; round substrates, on the other hand, necessitated optimum spinning speed and high acceleration (node 1.1.1.3.2.2.3.2). Otherwise, photoresist was building up either at the edges (at low speeds) or at the centre (at high speeds).

The mask alignment and exposure (node 1.1.1.3.2.4) process that follows the photoresist application were critical steps for the formation of the FET's pattern. Initially, a high-quality mask was needed with well-defined patterns and a clean surface; alignment seems to be the most probable step responsible for inaccurate patterning (node 1.1.1.3.2.4.2). Exposure (node 1.1.1.3.2.4.3) parameters determined the efficient change of the photoresist chemical characteristics to become soluble during developing; frequent calibration was required (node 1.1.1.3.2.4.3.2.2) to achieve reliable radiation. During the developing step (node 1.1.1.3.2.3), special care should be taken in setting the right time and in selecting the right developer material; in case of residuals, a second development dip was performed, adding, unavoidably, to surface inhomogeneity. During photoresist stripping (node 1.1.1.3.2.5), the film should be hardened and fastened to the substrate, usually by post baking (node 1.1.1.3.2.5.1) that frequently induced extreme hardening (Koryta, 1986) making impossible to remove the photoresist during stripping. It is worthwhile noting that, stripping should be performed at a temperature at least 20 °C less than the initial dehydration baking temperature (Colclaser, 1990) to assure that the gases absorbed at the substrate surface would not lift-off the photoresist. Although plasma stripping was more accurate and controllable, evidence suggested mobile ion contamination of the gate oxide (Maddox & Parker, 1978), leaving wet cleaning technology to dominate until late 1990s (Kim et al., 1997) that later turned to ECR (electron cyclotron resonance) hydrogen or oxygen plasma cleaning.

A sample of the fuzzy rules used (Figure 6), representing a chain leading from the eighth-level final event 1.1.1.3.2.2.3.2 (wrong spinning speed) to the top event (the highlighted part of Figure 5), is presented below:

IF 1.1.1.3.2.2.3.2 is *medium* AND 1.1.1.3.2.2.3.1 is *low* THEN 1.1.1.3.2.2.3 is *medium*.

IF 1.1.1.3.2.2.3 is *low* OR *medium* AND 1.1.1.3.2.2.1 is *low* OR *medium* OR *high* AND 1.1.1.3.2.2.2 AND 1.1.1.3.2.2.4 AND 1.1.1.3.2.2.5 are *low* THEN 1.1.1.3.2.2 is *low.*

IF 1.1.1.3.2.2 is *low* AND 1.1.1.3.2.1 AND 1.1.1.3.2.3 And 1.1.1.3.2.4 AND 1.1.1.3.2.5 are *low* or *medium* THEN 1.1.1.3.2 is *low.*

IF 1.1.1.3.1 is *high* AND 1.1.1.3.2 is *low* THEN 1.1.1.3 is *low.*

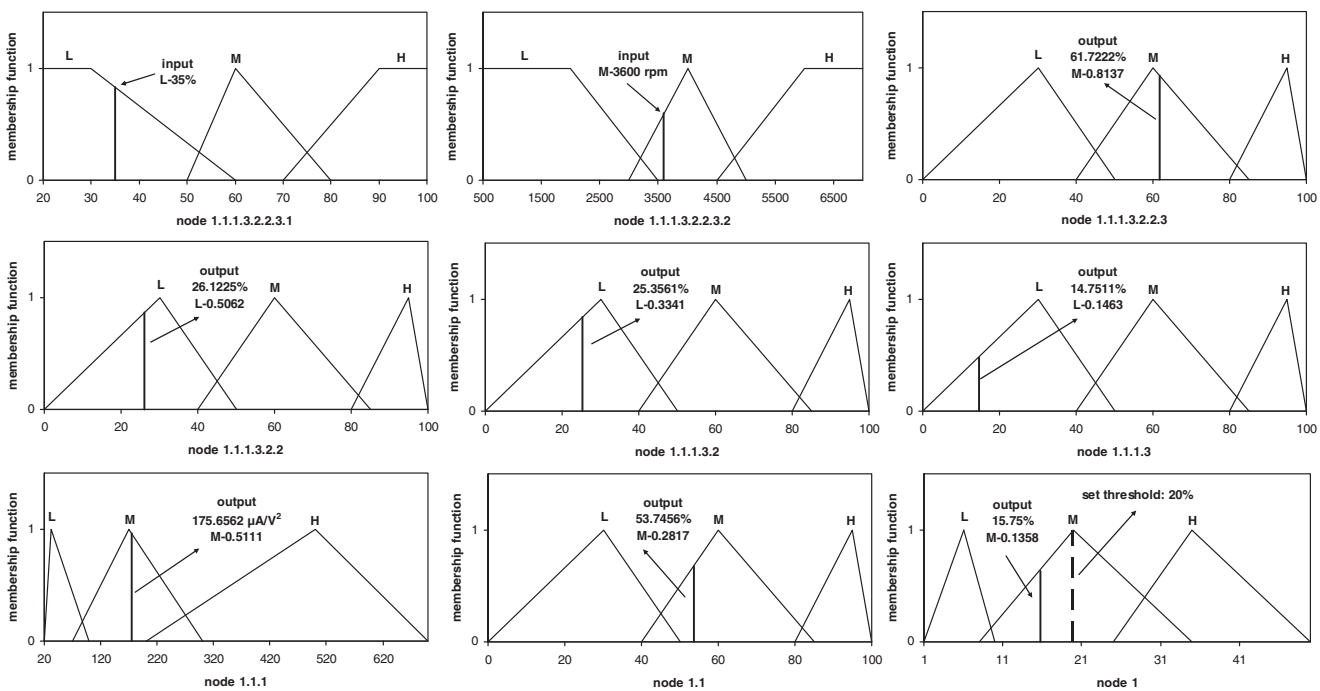

Figure 6. Membership function of some of the variables participating within the fuzzy chain leading from the final event 1.1.1.3.2.2.3.2 (spinning) to the top event ''drift of output voltage, V_{gs}'' (highlighted path in Figure 5). The solid line represents output values (intermediate and final) except from the basal events (input value). The symbols L, M, H stand for the linguistic terms *low*, *medium*, *high*, respectively, obtained by the partitioning of the space of variables. For each output, the value after defuzzification is given along with its membership function.

IF 1.1.1.1 AND 1.1.1.2 are *medium* AND 1.1.1.3 is *low* THEN 1.1.1 is *medium*.
IF 1.1.1 AND 1.1.2 AND 1.1.3 are *medium* AND 1.1.4 is *high* THEN 1.1 is *medium*.
IF 1.1 is *medium* AND 1.2 is *low* THEN 1 is *medium*.

The partitioning of the space of variables by means of the linguistic terms *low*, *medium*, and *high* was performed by the participating experts. For example, node 1.1.1.3.2.2.3.2 (spinning speed) has been partitioned to 1000–3500 rpm for the *low* region (peaking at 2000 rpm, which represents the most frequently used speed), 3000–5000 rpm for the *medium* region (peaking at 4000 rpm), and 4500–7000 rpm for the *high* region (peaking at 6000 rpm).

The threshold value set by the experts for the ''top event'' was 20% (1 ± 0.2 V), as experimentally suitable enough for preserving the sensor's reliability (*medium* range). The problem investigated refers mainly to the significance of the spinning on the homogeneity of photoresist application, also taking into account the variability of the other parameters. Twenty cases have been tested aiming at examining all possible combinations and influences. The results have shown that if all the involved parameters were kept *low*, the spinner speed (node 1.1.1.3.2.2.3.2) and stability (1.1.1.3.2.2.3.1) did not influence *Vgs* (node 1) significantly. If, however, photoresist stripping faults were *high* (node 1.1.1.3.2.5), a significant drift of *Vgs* occurred at *high* values of 1.1.1.3.2.2.3.2. Furthermore, if node 1.1.1.3.2.4 (mask result) was *medium* and all other parameters kept in the upper *low* levels, *Vgs* reached its upper limit of the threshold value. These results indicate that although the wafer quality (node 1.1.1.3.2.1) was considered important and received much attention, it was actually the mask quality (node 1.1.1.3.2.4) that weighted mostly on the accuracy of the sensor's output. Moving up the tree, the influence of C_{ox} accuracy (node 1.1.1.1) outranks the other branches: at *high* values, with all other branches at *low*, the impact on the top event was calculated to 35%.

The results obtained correlate well with the evolution of photolithographic patterning, especially after the utilization of nanotechnology design and production tools (stage 20). Nanopatterning presents its own drawbacks, especially in low-voltage applications (e.g. when monitoring a biochemical reaction), achieving only the lessening of the significance (weight) of the 1.1.1.3.2-branch to the top event. Lithography-based patterning in a nano-scale reveals mostly a strong dependence of apparatus cost with downsizing, given an already expensive mould (Zaborowski, et al., 2011). To avoid the high cost of patterning, a number of techniques have been developed for determining the pattern width by alternative methods. A widely used spacer technique (Choi et al., 2002) is a good example, although with limited applicability (Zaremba-Tymieniecki et al., 2010).

In anticipation of broad-applicability technological developments of the 1.1.1.3.2-branch, research efforts have also considered new materials in sensing in an attempt to remediate node 1.1.1.1.2 or Q (dielectric constant approximation, Figure 5). Various solid-state metal oxides have been proposed as pH-sensing films (Gerlach et al., 2005; Webster, 1999), including PtO_2, IrOx, RuO_2, OsO_2, Ta_2O_5, RhO_2, TiO_2 and SnO_2; IrOx, RuO_2 and SnO_2 have been proven relatively more efficient (Yao et al., 2001). Ruthenium (Liao & Chou, 2007) and stannic (Tsai et al., 2005) oxides exhibit near-Nernstian responses in wide pH ranges, however, at high hysteresis and drift that may lead to calibration issues and electrode instability. Iridium oxide films showed very good stability over wide pH ranges, rapid responses, less potential drift and high durability, even at high temperatures (Nishio and Tsuchiya, 2001). Various fabrication methods have been proposed for IrOx films (see e.g. Huang et al., 2011), most quite costly, while cost reductions during mass production remain to be seen. Two techniques appear to be quite promising: thermal oxidation and sol-gel. The latter seems to gain a competitive advantage since thermal oxidation requires high temperatures (500–800 °C) (Yao et al., 2001) to make the films thicker for providing more stable potentials, but with a tendency to crack that becomes an issue for sensing. Furthermore, high temperatures limit (a) the use of polymer and photoresist as sacrificial materials for defining film electrode patterns, because these materials usually cannot survive at a temperature above 200 °C, and (b) integration, especially with the sensing film (Huang et al., 2011).

Framework implementation – membrane-based biosensors

Membrane biosensors closely mimic biological sensory functions (Krull & Thompson 1985; Krull et al., 1989; Thompson et al., 1986). The development of ion-channel switch biosensor (Cornell, 1997; Downard, 1998) took 10 years, a team of 60 scientists and engineers, and at considerable cost that, however, could not outbid the success of converting a concept into a practical device. The mechanism of response (signal generation) relies on the changes of transmembrane ion flux induced by the interaction between a target analyte and an artificial cell membrane, detected as changes in the membrane's electrical conductance that provides efficient and rapid screening of samples.

Lipid membranes are two-dimensional fluid nanostructures where two, preferably, lipid layers are held together by non-covalent hydrophobic interactions of amphipathic molecules. The films have a thickness of about 5 nm, varying with the lipid tail length. Pure lipid bilayers, spanning an aperture that separates two electrolyte solutions (freely suspended BLMs), exhibit a resistance of $100\,M\Omega\,cm^2$ allowing for a small (on the order of few picoamperes) transmembrane ion current (Tien & Ottova-Leitmannova, 2000). For analytical applications, various biological moieties (e.g. enzymes, ion carriers, receptors, DNA) are incorporated into the membranes to add functionality (Ottova & Tien, 1997). Although sensor's sensitivity and selectivity toward a given analyte are endowed by the biological moiety (dependent on its affinity for the target), speed of response and reversibility rests on the lipid film. In any solid-state sensor, analyte molecules have to diffuse into and react with the biosensing component and any products of the reaction must diffuse out (Batzias & Siontorou, 2005). It, therefore, follows that the thinner the sensing layer the less time this will take and speed and reversibility of sensor response may well be improved. Needless to say that ultrathin films make the use of even highly expensive molecules (as DNA) economically possible. For example, a typical self-assembled monolayer

has a mass of approximately 2×10^{-7} g/cm^2 (Davis & Higson, 2008).

A wide range of materials have been used (Bartlett & Cooper, 1993; Scouten et al., 1995; Wink et al., 1997), employing various techniques for guiding thin film formation (Roberts, 1990; Tredgold, 1994; Ulman, 1998). Most of the proposed methods produce relatively thick layers compared with natural lipid films (at Å dimensions), thus lacking in sensitivity but gaining in stability and ruggedness. Other less thick systems can be used for the immobilization of biomolecules, such as lipid cast films, cross-linked with glutaraldehyde or dicyclohexyl carbodiimide (DCC), or multi-layered polylysine casts, extensively used for the production of DNA microarrays owing to their high affinity for biomolecules (Davis & Higson, 2008). A number of analytical tools are available for thin film interrogation, for example, electrochemistry, optical spectroscopy, surface plasmon resonance, quartz crystal microbalance.

Molecular self-assembly, a key link between physics, chemistry and biology, can be used to create novel structures, materials and devices for use in biosensors (Boozer et al., 2003; Jianrong et al., 2004). Various self-assembled structures have been studied for biosensing, especially bilayer membranes (Nikolelis & Siontorou, 1995; Siontorou et al., 1997, 2000). The hydrophilic/hydrophobic characteristics of the lipid molecules allow them to spontaneously form organized structures on supports, given that adequate hydration is provided. Simulating cell membrane functions, the bilayer structures provide a natural environment for membrane-bound moieties (e.g. proteins, receptors, membrane/tissue fragments and entire cells) under non-denaturing conditions and in a well-defined orientation (Göpel & Heiduschka, 1995; Ramsden, 1998). From the soap bubble of Hooke (1672), research performed on this concept aided the understanding of membrane functions and provided thermodynamic data, assisted the optimization of bioelement incorporation onto transducer surfaces and clarified (to a certain extent) the mechanisms of signal generation. The paths followed (stage 5) soon thinned to two trajectories, namely, stability and manufacturability (Supplementary Figure 7), because other performance requirements, such as exceptional selectivity and sensitivity, had been already demonstrated in laboratory set-ups. The current membrane university network (Figure 7) was structured by applying science overlay mapping (Leydesdorff & Rafols, 2009) to locate researchers amongst the academic disciplines (stage 3). Apparently, global research on membrane biosensors involves a very extensive range of research fields. It is concentrated on the

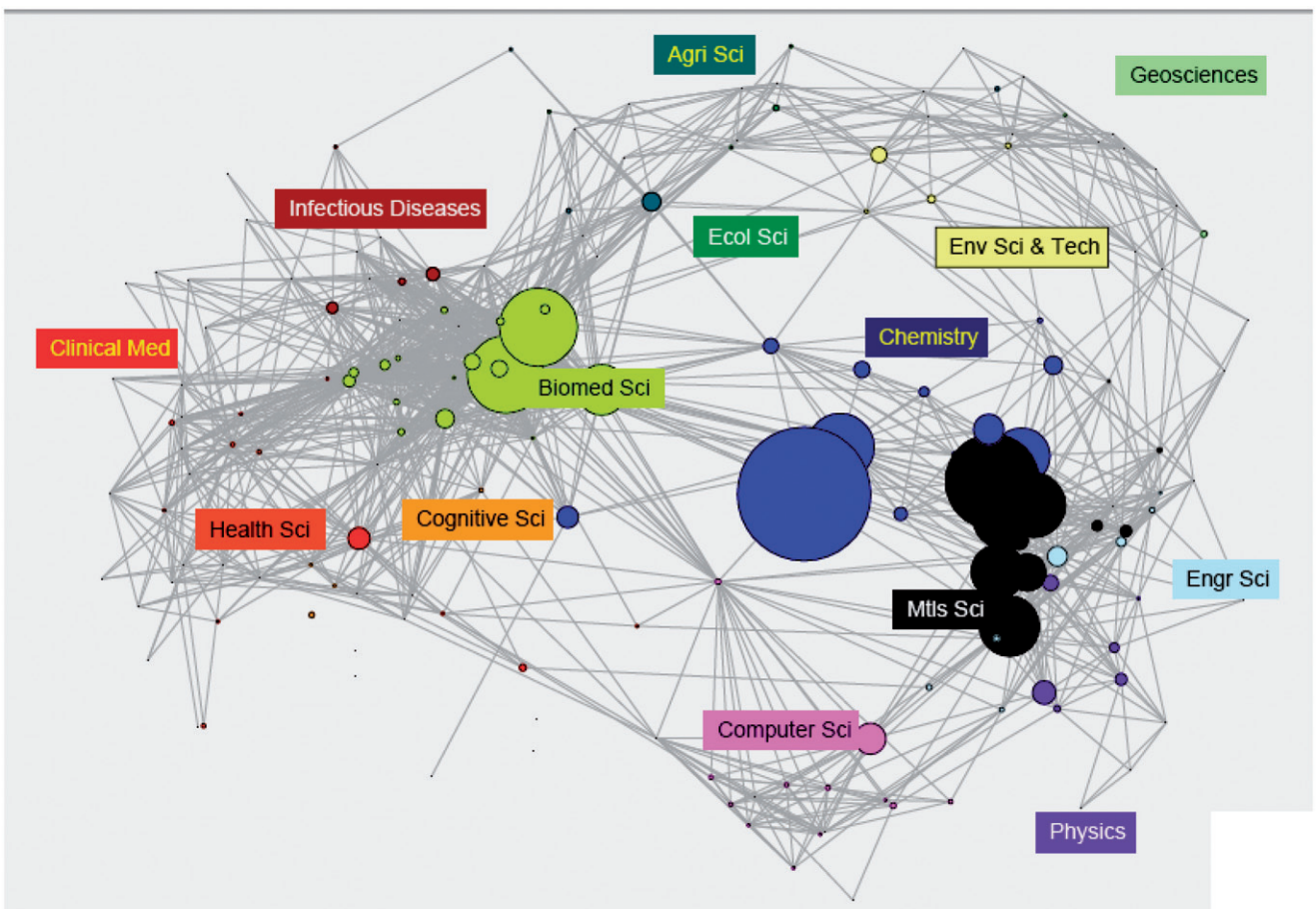

Figure 7. Science overlay mapping of the present membrane biosensor network. This approach (based on the model of Leydesdorff and Rafols, 2009) uses the subject categories that Web of Science assigns to journals: for a set of publications indexed by Web of Science (in this case, by the Science Citation Index, SCI, which is part of Web of Science), membrane biosensors have been located via the journals in which the appear. Membrane biosensor research overlies a base map reflecting the 175 subject categories shown by the background intersecting arcs. The subject categories were grouped into ''macro-disciplines'' and became the labels in the figure. The research clusters appear as nodes in the map, with larger nodes reflecting greater numbers of publications.

materials science and chemistry macro-disciplines, also involving a number of biomedical sciences and various other fields. This analysis helped to understand the science and technology fields involved (stage 10), which subsequently fed the fault tree analysis.

Looking into the content of this innovative biosensor frame (stage 14), the various research streams struggled over stability (criterion C3: reliability, factor: F3.14.02), causing the rivalry of altering lipid composition (with cholesterol, for instance) versus supporting membranes on solid supports. There has been an increasing emphasis on the latter rival because surface-based membrane platforms offer inherent stability because of the underlying supporting surface, as the lipid membrane remained intact. These platforms had the competitive advantage of preserving the outstanding nature-derived sensitivity, although the supporting chemistry and engineering hampered transduction, whereas the asymmetric hydration could not host transmembrane alterations (Disalvo et al., 2008; Goto et al., 2008; Kiessling et al., 2009). Nevertheless, stability remained an issue and 10 years later research revisited rigidity, aided by the availability of more suitable materials, as chitosan or polymerizable lipids, drawn deep by the science basis. The general conditions for stability of a system, particularly a membranous one, are given by the thermodynamics. The membrane is stable when its free energy has a minimum value in the space of the independent thermodynamic variables (Almeida, 2009). This means that any infinitesimal change of the independent parameters (pressure, temperature, electrical potential, surface tension, etc.) should lead to an increase of the free energy of the system. However, in many cases, as during protein function or flip-flop, the membrane can be unstable with respect to some of the thermodynamic parameters. However, the rate of change of the membrane state is so small that during the characteristic time-scale of the experiment, this membrane behaves as stable that is the case of ''meta-stable'' membranes.

Membrane instability can be brought about by various reasons (Dimitrov & Jain, 1984), including osmotic pressure difference, hydrodynamic instability or dielectric breakdown that will alter the surface tension of the lipid film, leading initially to molecular re-arrangement and finally to membrane discontinuity. While the former does not alter substantially the functionality of the membrane, the latter is irreversible and results in terminal rupture. The organization of the lipids into the bilayer structure during membrane formation plays an important role in the film tolerance toward rupture, as the viscoelasticity properties of the films are built-in and defined during the process of reconstitution (White 1972; Ziebert & Lacoste, 2011). Thus, it soon became evident that stability is not only directly related to factor F3.14.02, but also depends upon fabrication parameters relevant to criterion C6 (manufacturability). The lack of manufacturability, allegedly still prohibiting industrial production, has been analyzed with fault tree analysis (stage 16), as per the three factors more critically related (directly or indirectly) to stability, that is, F6.02.21 (simplicity), F6.02.05 (ruggedness/robustness) and F6.14 (mass production) (Supplementary Figure 8).

The results, provided by the inference engine and evaluated by the experts, indicate that F6.02.05 (directly related to stability) weighs mostly on the manufacturability problem, yet this fault is yielded by the strict conditions required for the formation of the bilayer (node A1.1.3): phospholipids self-organize to lipid bilayers, stably accommodated at room temperature and balanced electric field, hydrostatic pressure and ion gradients (Stern et al., 1992). Contrariwise, high temperatures (node A1.1.3.1.1.2.2) (Quinn, 1989), hydrostatic pressure (node A1.1.3.1.1.2.3) (Brooks et al., 2011) and electrolyte imbalance (node A1.1.3.1.1.2.1) (Gurtovenko, 2005), along with other membrane-inherent thermodynamic states (Almeida, 2009) and environmental conditions (Suresh & Edwardson, 2010; Zhu et al., 2012) (nodes A1.1.3.1.1.2.4 and A1.1.3.1.1.2.5, respectively), lead to irreversible phase separation of lipid components within the membrane (node A1.1.3.1.1.2). Moreover, the membrane may be impacted by the externally applied electric field (node D.2.3) (Sens & Isambert, 2002), either creating high transmembrane potentials or assuming high values that overcome the energetic barrier for hydrophile pore formation, destabilizing surface energy enhancing undulations (node D.2.2.1) and facilitating multiple pore formation (node D.2.2.2) as a result of electroporation (Tarek, 2005).

It seems, therefore, that future trends on manufacturability should better follow the science path than the technology path (stage 20). These findings fully support the notion that scientific research is multi-dimensional and nonlinear, often following unforeseen paths refuting trajectories that were based on frequently obscure relations between the various determinants. Certainly, trajectories lie along with nanotechnology, and especially nanofabrication and nano-bioinformatics (stage 19). The various roadmaps that have recently been presented in market reports, include membrane biosensors (see e.g. McWilliams, 2008; Rajaram, 2011) seen possible for future implementation (because no relevant commercial device currently exists), owing to the persisting extensive scientific literature. They suggest that biomimetic and synthetic materials should be sought after, considering that the significant industrial experience in tailoring and handling crystallic forms could facilitate mass production. The analysis presented, however, suggests that nanofabrication should be focused on controlling the natural process of self-assembly and the thermodynamics of bioelement-lipid interaction (Figure 8), thus retaining (and fully exploit) the nature-derived sensitivity of the biosensor platform. Undoubtedly, the interest of university researchers concentrates on the elucidation of natural processes, whereas the industry is market/applicability-oriented. Yet, the implementation of biomimetic materials necessitates deep knowledge on biological mechanisms, down to molecular level, which is, currently, not available.

Discussion

This work points toward a generalized process or model for roadmapping academic research based on the identification of technology clusters and the dynamics of the innovation trajectories. The proposed combined framework is designed to promote the university output for commercial use, not only when industry sets the targets and employs scientific

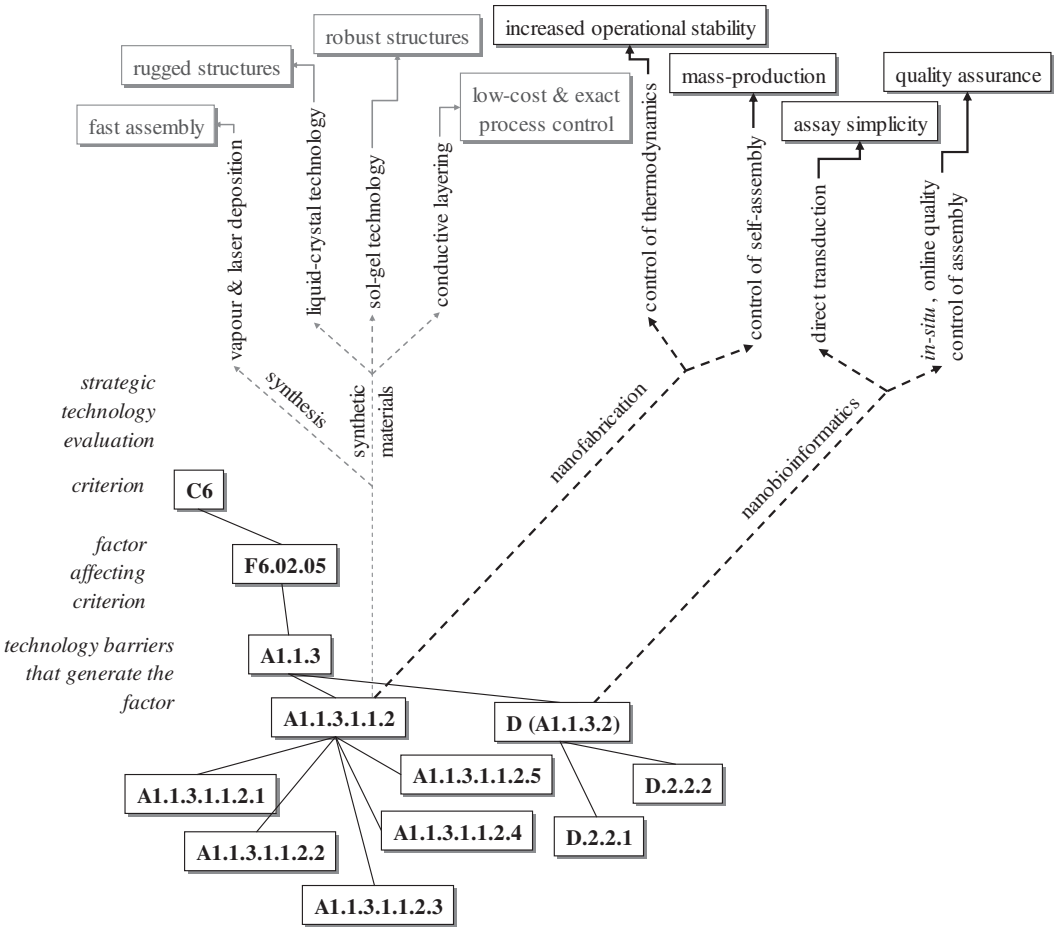

Figure 8. Excerpt from the strategic technology evaluation of membrane biosensors related to factor F6.02.05 (design ruggedness/robustness; A1-branch at Supplementary Figure 8). The technology barriers shown, A1.1.3.1.1.2 and A1.1.3.2 (or D), have been identified by inference engine (and agreed upon by the experts) as most probable and/or critical causes of the manufacturability issues (criterion C3). The right part (gray lines) progress is less probable to succeed its goals or proceed the left part (bold lines) development scenarios, although it is aligned with the roadmaps that have been recently presented in market reports.

breakthroughs (as seen in the case of the glucometers), but also when the science-push base is the right choice, that is, when shifting from mission-oriented to diffusion-oriented paradigms.

A university-fostered technology roadmap and its processes differ from sustaining roadmaps and emergent technology roadmaps that are based on market competencies and interests. Academy is focused on knowledge production, not technology exploitation, following many research paths and opening many possibilities. Academic research is defined herein as research that frames, analyzes and processes an emergent technology with a view to: (i) grasping the technology's complexity, (ii) considering and deploying its diverse perspectives, (iii) linking abstract and case-specific knowledge and (iv) applying descriptive, normative and practical knowledge to address the relevant bottlenecks. The biosensor technology field has been selected as an exemplar owing to its rapid emergence from a revolutionary breakthrough and the high absorptive capacity of the self-organized, aggregated, complex and dynamic innovation system.

Framing the university output is not an easy task; bibliometrics, scientometrics and patent analysis can only seize the impact of produced knowledge, not only its dynamics or its trajectories. Looking into the content of the scientific frame, this work revealed that although the drive of the innovation system was the exploration of natural chemoreception, trajectories moved along problem solving in an interchangeable shifting between science and technology. The AHP model approach adapted herein, where technology alternatives at the lowest level have been replaced with technology internal barriers, is most suitable for drawing trajectories, inasmuch as this nested loop mechanism of knowledge production will be maintained as part of academic research policies.

Modeling involved a collective mapping of projected research-strategy paths (stage 5) and a reflection on the future technical path or entanglements which are projected (stage 11), that is, foreseen or sought, on the premises that the sector will be oriented toward performance engineering. The multi-path analysis allows the mapping of these projections, under which there exist convoluted socio-technical links and dynamics that could either enable or constrain some of these research paths (stages 6–10). When the path created is the aggregate outcome of activities at the so-called basic research levels, then any research stream identified at stage 4 can potentially become the core for developing the new technology.

Technology barriers have been identified with fault tree analysis (stage 16), which has been frequently applied in determining causal or dependence relations in shortcoming

and deficiencies, handling efficiently multifaceted problems, inter-disciplinary frames and loose system boundaries. Adopted in its fuzzy version to count for uncertainty, it has been retrospectively validated using the ISFET technology output at early to mid-1990s. It has been proved suitable to appreciate the complexity of biosensors and correctly draw the trajectories that the field relies on in the coming years by identifying the technology barriers that the researchers would, inevitably, focus upon. The structure of the tree remains valid till today, further supporting the value of the proposed tool, as the problems of the past have been only partly solved today and researchers are still studying the same issues, although from different angles, with an extended knowledge background and more sophisticated instrumentation. Attempting an inductive generalization, the self-regulating role of university research incorporates, inevitably, at some point, self-evaluation which gives rise to a continuous dialectic trade-off between science and technology, in analogy to the Nonaka's continuous interplay between tacit and explicit knowledge. Self-evaluation alone cannot push developments forward, but it provides a holistic and judgmental view of the system of innovations that, although it retains its status, it does so at a more advanced knowledge level (Nonaka's spiraling) that pushes developments forward.

The proposed methodology has been also implemented prospectively for membrane biosensors. Factors driving the current development of lipid membrane sensors involve stability and manufacturability. Using FTA in studying manufacturability issues, the control of self-assembly complexity was highlighted, suggesting that manufacturability and stability are actually linked. To date, no single approach has successfully combined the sensitivity of the black lipid films with a robust design. Through combination of nanofabrication and nano-bioinformatics with self-assembly of free-spanning membranes, it is suggested that in recent years membrane biosensors might find their way toward commercialization. This analysis, however, positions the trajectory of membrane platforms differently than the market reports do, pointing out the differences between the scope of academic research and the market point of view. Many groups are currently working on both paths hopefully to soon assist the deployment of natural cellular processes to commercial detectors.

Because of the exploratory nature of the ISFET-biosensors case, the experts selected for determining the dynamics of the technology and for identifying the research paths had either participated or lead university research groups with high activity on ISFETs at the given time frame. Similarly, the implementation case on membrane biosensors relied on university researchers, selected from highly ranked lipid membrane biosensor groups. For reflexive alignment within research networks (stages 10, 11 and 19), however, the participation of experts on loosely related subjects (e.g. experts in other biosensor formats or in chemical sensors or other interested groups, as defined at stage 3) would seem advantageous in setting strategies for further research or for offering solutions. This network-level strategy support system represents an oblique and indirect approach to specific technological issues (well-defined and formally categorized topics within tight academic networks), such as the cell-on-a-chip. In a way, it is a bottom-up approach for methodology/tool building, growing with each new fault tree added, raising the FTA modeling tool to the network level that will facilitate knowledge flow and symmetry of knowledge dissemination.

Thereby, a mechanism will be structured to permit multiple model management and conceptual knowledge-level integration. Knowledge can be represented seamlessly from the abstract/common level to domain/path-specific level only within a properly designed ontological platform that will allow knowledge to be used not only for representation but, also, for reasoning at conceptual level. Such a platform can support a more computer-aided roadmapping tool, provided that, the inference engine could successfully handle: (i) vague, imprecise, interchangeable or controversial classifications and terminology, (ii) multiple definitions of scope, purpose or operation of the same entity, even if the physical laws governing the entity at its various functions are differentiated, complementary or follow different levels of knowledge, (iii) interdisciplinary scientific inputs and multifaceted applicability of the same product structure and (iv) interfacial phenomena. Furthermore, it should easily provide and explore knowledge from different levels, domains of discourse, and complex dynamic environments, contributing substantially to framing the knowledge creation and innovation process. However, the cost of collecting knowledge could still be too expensive in comparison with benefits obtained from such customization. Clearly, a trade-off exists, on the one hand, it is important to generate the maximum added value from accumulated knowledge, but on the other, knowledge accumulation efforts should be made easier with intelligent computational supports.

Conclusions and perspectives

This study suggests a comprehensive combined framework for roadmapping the university research output, in which the Analytic Hierarchy Process is developed *ad hoc* on the basis of the emerging internal technology barriers. This re-orientates the various productive research streams (trajectories) that are structured within the academic fields establishing their rigid thematic structure. In collaboration with experts, the authors have used the biosensor field as a suitable paradigm for developing this framework, proven suitable to accommodate the multifaceted and interrelated issues of the university-produced innovation, considered within its inherent context, that is, fostered and managed within a technology cluster as part of a continuously reforming and redefining research agenda. Fault tree analysis has been used to extrapolate the underlying causes impeding the full realization of the field's potential. These causes, actually, delineate the university research paths, drawing trajectories toward commercialization that may dynamically change in the time course as knowledge accumulates.

The evidence provided so far indicates that in any attempt to analyze an academic innovation system one should consider and evaluate that university research is mostly self-regulated, dynamically oriented toward strategic targets set *a priori* using continually changing tactics and multiple hypotheses testing (push-basis), until knowledge accumulation adequately supports certain research paths to become dominant (pull-basis). This is particularly evident for the

biosensor field, where its founding target, that is, natural chemoreception, is not only valid today but it has also affected significantly other fields, such as nanotechnology and bioinformatics. Notwithstanding, certain areas have presently reached a marketable state-of-the-art, as the case of ISFETs, while others, such as the thin film platforms, need further research on their scientific bases, to the degree and extent feasible by modern technology limitations. The strategic priorities of academic innovation may not be understood or clarified when the frame is structured or when the clusters are defining their scope, inasmuch as academia purports to enhance knowledge creation/exploration. This may or may not align well with the pull-basis, yet it preserves enough degrees of freedom to generate disruptive concepts (e.g. the successful case of glucose sensing and the soon-to-be artificial pancreas). At the time course, however, the frame becomes well- and strictly-defined with limited ability to generate disruptive concepts on its own, necessitating the shifting of the frame toward transdisciplinarity to allow for new scientific and technological opportunities. The shifting of the biosensor scientific frame from biomedical to chemometrics (at mid-1990s), and the alignment of the sector with the nanotechnology trajectories (at early 2000s) extended considerably the scope of the field (application area) at more plausible and feasible specifications for ''intended use.''

While the suggested framework comes closer to the nature of knowledge production within academia, roadmapping its research output may not be sufficiently credible for long-term projections. A roadmap presupposes collectively agreed destinations and futures with academic research; being diffusive and elusive, it is difficult to draw trajectories toward a common operating context in the future without carrying along high degrees of uncertainty. The scheme presented, focusing on the current problem-solving strategies pursued by the researchers, manages to capture the analytical (slow, conscious and consistent) aspects of innovation to lessen near-term future uncertainty, provided that the roadmapping process is kept on an ongoing basis. Undoubtedly, framing the intuitive (rapid, unconscious, combinatorial) thinking makes the art of science difficult. Yet, the combined framework suggested herein may facilitate the handling of this parameter by including knowledge at deeper phenomenological levels, provided by fault tree analysis, in roadmapping. With the clear understanding of the dynamic linkage and relationship among the key players that form the scientific network, long-term roadmap credibility and utilization can be improved, while knowledge can be more effectively shared and transferred.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this article.

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Appendix A

Notes on the history of glucose biosensing (Supplementary Figure 2) – extract from the dedicated Knowledge Base developed by the authors.

(1) Clark LC Jr, Gollan F, Gupta VB. (1950). The oxygenation of blood by gas dispersion. Science, 111, 85–7. *Clark developed his first bubble oxygenator for use in cardiac surgery. However, when he came to publish his results, his article was refused by the editor since the oxygen tension in the blood coming out from the device could not be measured. Clark found a Van Slyke apparatus and learned how to run it; within a couple of weeks he had oxygen values and sent the manuscript back to Science. Clark, however, wanted continuous oxygen monitoring for using his device in humans. He tried at first optically using an oximeter. This was no help after the blood was fully saturated. He decided to adopt polarography instead that led to the oxygen electrode.*

(2) Davies PW, Brink F Jr. (1942). Microelectrodes for measuring local O_2 tension in tissues. Rev Sci Instrum, 13, 524-33. *Davies and his collaborators studied oxygen tension and metabolism at the surface of the mammalian cerebral cortex adopting an electrolytic method for measuring dissolved oxygen proposed at 1939–1940. The used platinum electrodes made in the form of a catheter for making very localized measurements. Davies and Brink were the first to use membrane-covered oxygen electrodes, although unlike Clark's later insulating membranes, theirs were collodion, and were permeable to ions as well as to oxygen. Such electrodes tended to be free from ''electrode ageing'', a phenomenon which had made long-duration measurements difficult, particularly in blood.*

(3) Clark LC Jr, Wolf R, Granger D, Taylor Z. (1953). Continuous recording of blood oxygen tensions by polarography. J Appl Physiol, 6, 189–93. *Clark used a two electrode system containing a Pt anode as working electrode surrounded by a silver/silver chloride (Ag/AgCl) reference electrode to determine the oxygenation of blood. His first sensor failed because blood components were adsorbed on the electrode's surface, and this adsorption distorted the signal. Clark then had the ingenious idea of using the cellophane wrapper of a cigarette packet on his sensor. Only low molecular weight substances, mainly oxygen, could reach the electrode and be measured. The reduction current indicated the oxygen concentration, and so the Clark electrode was created.*

(4) Clark LC, Lyons C. (1962). Electrode systems for continuous monitoring in cardiovascular surgery. Ann NY Acad Sci, 102, 29–45. *To calibrate his sensor, Clark added an enzyme, glucose oxidase (GOD), to the solution. Clark then developed the sensor further by entrapping concentrated GOD with another semi-permeable membrane in front of the electrode. The electrode could then be re-used for multiple glucose measurements. The enzyme layer became an integrated part of the sensor. Clark went on to coin the term ''enzyme electrode'' at a meeting of the New York Academy of Sciences in 1962. Clark's original patent (US patent 3,539,455, 1970) covers the use of one or more enzymes for converting electroinactive substrates to electroactive products. The effect of interferences was corrected by using two electrodes (one covered with GOx) and measuring the differential current. Clark's technology was subsequently transferred to Yellow Spring Instrument Company that launched in 1975 the first dedicated glucose analyzer for the direct measurement of glucose in 25 mL samples of whole blood.*

(5) Warburg OH, Christian W. (1932). Uber ein neues oxydationsferment und sein absorptionsspektrum. Biochem Z, 254, 438–58. *In their study, Warburg and Christian showed that the ''yellow enzyme'' from yeast was rendered colorless upon reduction by its then still undefined substrate(s), and that its color was restored upon its reoxidation by shaking with gaseous O_2.*

(6) Theorell AHT. (1936). Die physiologische reoxydation des reduzierten gelden ferments. Biochem Z, 288, 317–28. *Theorell showed that the reaction of the reduced ''yellow enzyme'' with O_2 produced H_2O_2.*

(7) Franke W, Deffner M. Zur Kenntnis der sog. (1939). Glucose-oxydase. II. Ann., 541, 117–50. *Franke and Deffner isolated in 1939 yellow glucose oxidase (GOx) from Aspergillus niger and established that the GOx reaction center contained flavin. They also showed that the glucose-reduced flavin of GOx was oxidized by O_2, by cytochrome C, and by quinonoid dyes like toluylene blue, thionine, methylene blue, pyocyanine, and safranine T.*

(8) Malmstadt HV, Pardue HL. (1961). Quantitative analysis by an automatic potentiometric reaction rate method. Specific enzymatic determination of glucose. Anal Chem, 33, 1040–7. *The basis for building GOx-based electrodes for monitoring glucose potentiometrically through measuring the drop in O_2 partial pressure upon its consumption, or amperometrically, upon the electrooxidation of the H_2O_2 produced, existed already in 1939. Nevertheless, two decades past before Malmstadt and Pardue reported on the first glucose electrochemical assay. They used molybdate-catalyzed H_2O_2-oxidation of I^- to I_2, to enable determination of the H_2O_2 concentration by I^-/I_2 potentiometry.*

(9) Kajihara T, Hagihara B. Serum glucose measurement by oxygen electrode and glucose oxidase. Rinsho Byori, 14, 322–27; Makino Y, Konno K. (1967). Measurement of blood glucose by oxygen electrodes. Rinsho Byori, 15, 391–4. *Kajihara and Hagihara, then Makino and Konno, initiating what would later become the Japanese school in biosensors, monitored glucose concentrations through O_2 consumption in combination with, catalase, decomposing the H_2O_2 produced to water and O_2.*

(10) Updike SJ, Hicks GP. (1967). The enzyme electrode. Nature, 214, 986–8; (1967). Reagentless substrate analysis with immobilized enzymes. Science, 158, 270–2. *Updike and Hicks significantly simplified the electrochemical glucose assay by immobilizing and thereby also stabilizing GOx on Clark's principle. They developed further this principle by using two oxygen working electrodes (one covered with the enzyme) and measuring the differential current for correcting for the oxygen background variation in samples.*

(11) Silverman HP, Brake JM. (1970). Method of determining microbial populations, enzyme activities, and substrate concentrations by electrochemical analysis. U.S. Patent 3,506,544. *They described redox-couple mediated electrooxidation of glucose, based on the findings of Warburg and Christian (1932)[5] and Franke and Deffner (1939)[7].*

(12) Guilbault GG, Lubrano GJ. (1973). An enzyme electrode for the amperometric determination of glucose. Anal Chim Acta 64, 439–55. *They described an enzyme electrode for the determination of blood glucose based on amperometric (anodic) monitoring of the liberated hydrogen peroxide. Good precision and accuracy were obtained in connection to 100 μL blood samples.*

(13) Skeggs, LT. (1960). The determination of carbon dioxide in blood serum. Ann NY Acad Sci, 87, 650–7. *The double-lumen catheter for continuous blood sampling restricts the presence of anticoagulant to the area of blood withdrawal without its spreading into the general circulation. This device paved the way for in vivo and continuous glucose monitoring and artificial pancreas.*

(14) Bessman SP, Schultz RD. (1972). Sugar electrode sensor for the artificial pancreas. Horm Metab Res, 4, 413–7. *The possibility of measuring glucose levels in diabetes patients has been regarded as an opportunity for the artificial pancreas as early as 1965. But it soon became obvious that for in vivo monitoring of tissue glucose concentration the glucose transducer should be independent of oxygen tension. By recording the difference between the output of the two electrodes, Updike and Hicks[10] obtained an enzyme electrode sensitive to changes in glucose concentration, but relatively insensitive to changes in oxygen tension. However fouling of the electrode and rapid enzyme denaturation made the sensor inappropriate for long-term in vivo monitoring in body fluids. To overcome this problem Bessman and Schultz suggested a membrane covered oxygen electrode coated externally with a noble metal catalyst for the rapid continuous measurement of dissolved reducing sugar. However, body fluids quickly poisoned the catalyst in spite of electrochemical attempts to regenerate the electrodes with frequent negative electrical pulses.*

(15) Soeldner JS, Chang KW, Aisenberg S, Hiebert JM. (1973). Progress towards an implantable glucose sensor and an artificial beta cell. In: Urquhart J, Yates FE, ed. Temporal aspects of therapeutics. New York: Plenum Publishing, 181–204. *They devised a specially designed fuel cell that generated electrical energy by the process of auto-electro-chemical oxidation of glucose. The maximum current that could be sustained by the fuel cell was expected to be proportional to the glucose concentration. However, implantation of the fuel-cell glucose sensors into animals resulted in a very marked foreign body reaction after 27 days which was reduced by using a sulphonated*

polyelectrolyte complex as a semipermeable membrane instead of cellulose.

(16) Layne EC, Schultz RD, Thomas LJ, Slama G, Sayler DF, Bessman SP. (1976). Continuous extracorporeal monitoring of animal blood using the glucose electrode. Diabetes, 25, 81–9. *The dual cathode oxygen sensor of Updike and Hicks[10] was further developed and intended as a prototype for implantation for the artificial pancreas, as in the continuous extracorporeal monitoring of animal blood. Its response was apparently nonlinearly proportional to the glucose concentration.*

(17) Botz CK. (1976). An improved control algorithm for an artificial β-cell. IEEE Trans BioMed Eng, 23, 252–5. *It was early appreciated that computer algorithms were necessary for calculating the amount of insulin required for infusion with respect to the concentration of glucose level measured with the electrode, taken also into account the levels of two hormones: insulin if the level is too high and glucagon (or dextrose) if the level is too low.*

(18) Keen H, Knight RK. (1962). Self-sampling for blood-sugar. Lancet, 1, 1037–40. *Self-sampling for blood sugar determination was first introduced in 1962 by Keen and Knight, who developed a method whereby the patient while at home applied a drop of blood onto a small filter paper pad, which was then allowed to dry before being dispatched by mail to the hospital laboratory for analysis along with the patient's details, date, and time of sampling. The method of glucose estimation was colorimetric and considered adequate for clinical purposes when assayed within a week of sampling.*

(19) Rennie IDB, Keen H, Southon A. (1964). Rapid enzyme-strip method for estimating blood sugar. Lancet, 2, 884–6. *Rennie et al. described a rapid (1-min) enzyme test strip assay – based semiquantitative method for estimating capillary whole blood sugar.*

(20) Sönksen PH, Judd SL, Lowy C. (1978). Home monitoring of blood-glucose. Method for improving diabetic control. Lancet, 1, 729–32; Walford S, Gale EA, Allison SP, Tattersall RB. (1978). Self-monitoring of blood-glucose. Improvement of diabetic control. Lancet, 1, 732–5. *In the late 1970s, after several attempts at publishing and presenting their results, two simultaneous articles appeared in the Lancet demonstrating that rapid methods of SMBG, predominantly in persons with type 1 diabetes, were a means of improving diabetes control.*

(21) Cohen M, Zimmet P. (1983). Self-monitoring of blood glucose levels in non-insulin-dependent diabetes mellitus. Med J Aust, 2, 377–80. *They introduced the idea of glucose self-monitoring in patients with Type II diabetes.*

(22) Lubbers DW, Opitz D. (1976). Quantitative fluorescence photometry with biological fluids and gases. Arch Exp Med Biol, 75, 6511–5. *In 1793, Lubbers and Opitz presented an optical glucose chemical sensor; they also coined the term optode to spectrophotometric-based probes.*

(23) Kulp TJ, Camins I, Angel SM, Munkholm C, Walt DR. (1987). Polymer immobilized enzyme optodes for the detection of penicillin. Anal Chem, 59, 2849–53. *Walt and colleagues further elaborated the concept of optodes by placing enzymes at the tip of optical fibres.*

(24) Pickup J. (1993). Developing glucose sensors for *in vivo* use. Trends Biotechnol, 11, 285–91.

(25) Fischer U, Abel P. (1982). A membrane combination for implantable glucose sensors. Measurements in undiluted biological fluids. Trans Am Soc Artif Intern Organs, 28, 245–8; Shichiri M, Kawamori R, Yamasaki Y, Hakui N, Abe H. (1982). Wearable artificial endocrine pancreas with needle-type glucose sensor. Lancet, 2, 1129–31. *In 1982, Fischer's group in East Germany and Shichiri's group in Japan draw the path for implantable glucose sensors, suggesting anti-fouling strategies.*

Comments

In 1941 Clinitest® effervescent urine sugar testing tablets launched by Ames (a subdivision of Miles Laboratories) used alkaline copper sulphate and Na citrate that form color in re-hydrated state dependent on glucose content – 'non-specific' test subject to interference

In 1954 Glucotest/Testape roll licensed by Eli Lilly to Boehringer Mannheim

In 1964 Earnest C Adams developed Dextrostix (Ames – Miles Laboratories) Patent No 3,092,465 issued 4th April 1963)

In 1968 the Haemo-Glukotest (Roche Diagnostics, Mannheim, Germany), developed and improved in 1979, which still remains the gold standard of accuracy for purely visual blood glucose determination.

Appendix B

Notes on the technology map of surface plasmon resonance biosensors and amperometric biosensors (Supplementary Figure 5) – extract from the dedicated Knowledge Base developed by the authors.

(1) Kretshmann E, Raether H. (1968). Radiative decay of nonradiative surface plasmons excited by light. Z Naturforsch, 23A, 2135–6.

(2) Jeldly, TA, Nagy, K Johannessen, JS (1979). Solid-state ion-selective electrodes with integrated electronics. J Electrochem Soc, 126, 793–5.

(3) Giuliani JF, Wohltjen H, Jarvis NL. (1983). Reversible optical waveguide sensor for ammonia vapors. Optics Lett, 8, 54–6.

(4) Bradley RA, Drake RAL, Shanks IA, Smith AM, Stephenson PR. (1986). Optical biosensors for immunoassays. Royal Society Discussion Meeting, London, May 1986.

(5) Raether H. (1977). Surface plasma oscillations and their applications. In: Hass G, ed. Physics of thin films Vol. 9. New York:Academic Press, 145–261.

(6) Liedberg B, Nylander C, Lundstrom I. (1983). Surface plasmon resonance for gas detection and biosensing. Sens Actuat, 4, 299–304.

(7) Flanagan MT, Pantell RH. (1984). Surface plasmon resonance and immunosensors. Electron Lett, 20, 968–70.

(8) Cullen DC, Brown RGW, Lowe CR. (1988). Detection of immunocomplex formation via surface plasmon resonance on gold-coated diffraction gratings. Biosensors, 3, 211–25.

(9) Sarid D, Deck RT, Craig AE, Hickernell RK, Jameson RS, Fasano J. (1982). Optical field enhancement by long-range surface-plasma waves. J Appl Opt, 21, 3993–5.

(10) Matsubara K, Kawata S, Minami S. (1990). Multilayer system for a high-precision surface plasmon resonance sensor. Opt Lett, 15, 75–7.

(11) Nenninger G, Tobiska P, Homola J, Yee S. (2001). Long-range surface plasmons for high-resolution surface plasmon resonance sensors. Sens Actuat B-Chem, 74, 145–51.

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Appendix C

Notes on the scientific map of lipid membrane biosensors till early 1990s (Supplementary Figure 7) – extract from the dedicated Knowledge Base developed by the authors.

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