Editorial on "Genesis on diamonds II: contact with diamond enhances human sperm performance by 300%"

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Chemical sciences generally, and biochemistry among them, are undergoing a remarkable change of paradigm. Around 1985, chemists knew molecules on one side and bulk materials on the other, with a few oddities like colloidal gold or capillary water in between. Today we realise that these 'oddities' were but glimpses of a whole unsuspected world of nanoscience: objects of several nanometer size possessing most peculiar properties (1). As always in such times, everything is in flux, every resolved question poses three new ones, there are unexplained observations [thus, it was repeatedly reported since 1999 that water colloids of nanodiamond particles yielded some enigmatic fibrous structures on storage (2,3). It turned out (4) that nanodiamond facilitates, in yet unexplained way, the infestation by, and growth of, fungi, whose gifs tend to accumulate diamond nanoparticles within them! This episode highlights the importance of multidisciplinary approach, as the early researchers' background in explosive technology and shock-wave physics obviously diverted their attention from biological explanations], and unproven hypotheses everywhere, and it requires no small daring (as well as methodical work) to convert such unfinished science into useful practical applications. The present paper (5) is a brief survey of one such achievement, which resulted from two distinct, but interconnected, lines of research, pursued for a number of years by the team of Dr. Sommer at Ulm University: nanodiamond surfaces and interfacial nanolayers of water.

Two types of nanodiamond are relevant in biomedical field. Detonation nanodiamond (DND) formed during detonation of carbon-rich/oxygen-poor high explosives (6), comprises particles of roughly 5 nm size, which can be used for drug delivery or biolabelling (7). Nanocrystalline diamond layers (8) produced by chemical vapour deposition (CVD) under relatively milder conditions, are widely used as coatings, including those of surgical instruments and bioimplants. It is the latter type of nanodiamond that was used in the discussed work.

Nanoscopic interfacial water layers which form on all solid surfaces, whether hydrophilic or hydrophobic, under air or under (bulk) water (9,10), are known to play immensely important role in a variety of biological processes (11), as was anticipated in the visionary work of Szent-Györgyi (12). Nanodiamond of any kind or, indeed, the surface of bulk diamond (natural or synthetic) is on the nanoscale highly hygroscopic and thus provides an excellent platform for studies of interfacial water, which showed the latter to be quite distinct from bulk water: with denser molecular packing (13), highly ordered and quasi-crystalline (14) [to the extent that hydrated diamond surface could have served as an origin-of-life platform, by imposing order on adsorbed pre-biological molecules (15)]. On the macroscale, however, the nanodiamond surface can present hydrophobic or hydrophilic properties, for instance, when terminated with hydrogen or oxygen, respectively, as reflected by the water contact angle. The corollary of this is the viscosity of interfacial water (generally, not just on nanodiamond) being several orders of magnitude higher than that of bulk water, in particular on the hydrophilic species. This realisation allowed for the first time to make sense of the thermodynamics of ATP synthase rotary nanomotor, previously believed to work at the impossible 100% efficiency, calculated on the assumption that mitochondrial water has the viscosity of bulk water—whereas in fact it is interfacial water (16).

Interfacial water layer on nanodiamond responded to irradiation by red (670 nm laser) or near-IR light by an increase of volume and a corresponding decrease of viscosity (14). Similar irradiation of living cells boosts ATP synthesis—which can be now explained by reduced viscosity of the (interfacial) water around the nanomotor (16).

Presently, these findings are applied to improve the IVF procedure. The curse of the latter are so-called reactive oxygen species (ROS), viz. singlet O_2 molecules, free radicals and peroxides, which are inevitable products of cell metabolism, but damage sperm cells in the way they do not do *in vivo*. This problem of 'oxidative stress' has been recognised for decades (17), but the causes of its aggravation *in vitro* were not well understood. Sommer *et al.* show that a standard polystyrene Petri dish, contrary to common perception, is not inert towards aqueous media but suffers mechanical softening on nanoscale—as measured by an ingenious underwater nanoindentation device (18). This softening makes the surface a trap for ROS which form a cell-damaging nanolayer and aggravate the damage further by increasing water viscosity. The recommended (and tested) answer was to replace polystyrene Petri dish with a CVD-nanodiamond coated quartz one, which, indeed, greatly improved the survival rate of sperm cells (5,19). Admittedly, the title of the paper (5) is something of a misnomer: diamond is not enhancing sperm performance in an absolute sense, but rather doing less damage to it, relative to polystyrene surfaces. A further improvement was achieved (5) by using 670 nm LED irradiation (vide supra) to boost the synthesis of ATP—the fuel for spermatozoa motility. This generates ROS (in an intensity- and dose-dependent manner) as a by-product which would more than counterbalance the positive effect on polystyrene—but not on diamond.

Appreciating the prospects of the new approach, let us survey some of the remaining issues. Firstly, what is the underlying cause of the improvement? Obviously, the supreme hardness and absolute chemical inertness of diamond simply discourages the adsorption and accumulation of ROS on the surface—but can it be more than this? The authors hypothesise that nanodiamond may actively neutralise ROS (5). Can it be related to diamond surface having a unique property of negative electron affinity (20), thus readily releasing electrons into water? If nanodiamond indeed is an active scavenger of ROS—can a suitably prepared DND powder do the same job?

One should not imagine a diamond surface as a mere cross-section through a bulk diamond structure as known from crystallography. Such dissection would leave every second surface atom with a dangling chemical bond pointing outward—a most unstable and chemically reactive state. In reality, the surface is terminated either by C-H bonds in hydrogen-rich environment, or by various oxygen-containing groups (COOH, OH, C=O, etc.) in oxidative one. A 'clean' (non-terminated) diamond surface always structurally adjusts to achieve stability. The adsorptive properties of the surface thus depend strongly on the history of its synthesis, sometimes in anti-intuitive ways. Thus, hydrophobic H-terminated nanodiamond adsorbs a thicker layer of interfacial water than hydrophilic oxidised one, although the water-diamond binding is stronger in the former case (21)! For the same reason, the ideal inertness and biocompatibility of pure diamond should not be ascribed by default to terminated nanodiamond surfaces (7), especially so for DND born in an explosion and purified from graphite by "aqua regis" or perchloric acid (6). DND is never pure diamond—indeed, it is never pure carbon, containing up to 10% or even 15% of impurities: Fe, N, H, but mainly O. In our own experience, DND from various suppliers may differ quite startlingly and proper standardisation is long overdue. However, CVD nanodiamond is produced under much more controllable conditions and does not require wet purification, therefore its standards of uniformity and purity are incomparably better.

Sommer's work invoked concerns (22) that nanodiamond itself may be toxic to sperm cells and fertilised eggs, but such conclusions were all drawn from experiments with DND (23). In view of the above, these DND results need not necessarily apply to CVD nanodiamond. In the present case, biocompatibility was proven rather than presumed, although due caution is of course necessary. In the first place, strict uniformity of nanodiamond Petri dishes used in research and clinical practice must be assured. It is therefore unfortunate that the papers (5,18,19) do not describe how these coatings were made, although detailed information can be found in the thesis of Zhu (24), which is available online.

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Footnote

Conflicts of Interest: The authors have no conflicts of interest to declare.

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