Getting Focused on Reproduction

This issue of Reproduction sees the first in a new initiative from its Editors – the Focus issues. Focus issues are designed to bring together reviews and research papers in areas of Reproductive Biology that have seen recent rapid developments. The first of these is Focus on Fertilization. This will be followed in September by Focus on ART and in December by Focus on Implantation. For each issue a ‘guest’ editor will commission 3–4 reviews and we will be inviting research paper submissions on the topic. If you are reading this in the Journal more information on submission deadlines for these Focus issues is available via www.reproduction-online.org. To satisfy the wider interests of the readers of Reproduction each Focus issue will also contain research articles on the broad range of subjects we strive to cover each month. The Focus issues will provide a resource for experts, students and teachers alike and will form the basis of Focus collections, where past papers and any future papers on the theme will be bundled together online for ease of access.

Focus on Fertilization

Anyone who has been fortunate enough to attend the Fertilization Gordon Conference will know that there are three questions that never fail to invoke lively discussions that last well into the night, often threatening the strict New Hampshire curfew times. In their simplest terms the questions are:

i) How the sperm and egg recognise each other?

ii) How the sperm and egg fuse?

iii) How the sperm triggers egg activation?

Each question represents a research field in its own right and for the most part no research groups have had the inclination, quite sensibly, to diversify into more than one of them.

These are long-standing problems that have thrown up a host of candidate molecules and mechanisms, often with some fanfare, that are subsequently resigned to the sidelines when something new comes along. However, in recent years significant insights have been provided by access to extensive sequence information, the development of well-designed molecular tools and the relative clarity offered by molecular genetics. These insights have led to the identification of new molecules and new mechanisms that have brought us much closer to answering each of these questions. Molecular genetics in particular has proven incisive in unravelling some of the mechanisms of fertilization. For this reason, the first review in this Focus issue provides an overview of the different genetically tractable systems that have contributed to identifying molecules involved in fertilization (Geldziler et al. 2004). Each of the remaining reviews in this Focus issue is dedicated to one of the three questions highlighted above.

Sperm–egg recognition underwent a major leap forward, at least in mammals, with the now classic work identifying the glycoproteins of the zona pellucida. These studies went on to demonstrate the role of zona proteins in sperm binding (Wassarman et al. 2001). In this issue Hoodbhoy & Dean (2004) argue that, rather than any single protein or glycoconjugate being responsible for sperm–zona binding, it is the three-dimensional structure of the zona pellucida that is critical. This hypothesis is drawn from their recent data showing that taxon-specific sperm binding and fertility are not inhibited by replacing mouse ZP2 and ZP3 with the human proteins (Rankin et al. 2003). Thus, while it is clear that ZP3 and ZP2 are the egg receptors for sperm, precisely how they act to bind sperm is not yet fully understood.

The molecules on the sperm surface responsible for interacting with the zona pellucida have also proven to be somewhat elusive. But on this question too there have been exciting new developments. One of the favourite candidates for the sperm receptor for ZP3 is β1, 4-galactosyltransferase I (GalT I). However, GalT I(-/-) males are fertile (Lu & Shur 1997), suggesting that other factors may be involved. Very recent studies have provided a new player. SED1 is a protein containing Notch-like EGF repeats that are expressed on the surface of sperm. Inhibition of SED1 function in vitro reduces sperm–egg binding and inhibits fertilization. In vivo, SED1(-/-) mice have reduced fertility and the sperm fail to bind to the zona pellucida in vitro (Ensslin & Shur 2003). It seems no single sperm protein will be solely responsible for tethering the sperm to the zona pellucida, rather the strategy seems to involve utilising a combination of interactions. The inability of GalT I(-/-) sperm to compete with wild-type sperm in achieving fertilization graphically demonstrates the evolutionary and physiological advantages of increasing the number of mechanisms available for sperm–egg interactions (Lu & Shur 1997).

Having bound and traversed the zona pellucida the fertilizing sperm next comes across the egg plasma membrane. What happens next is a rare event in cell biology – the fusion of two different cell types. The molecules that mediate this fusion event have been the subject of much debate. Inhibitor studies using antibodies suggested that the egg integrin α6β1 was essential for sperm–egg fusion (Almeida et al. 1995). Normal fertility of β1(+/-) mice sheds some doubts on this and further suggests that none of the integrins expressed by eggs are essential for sperm–egg fusion (He et al. 2003). During the rise and fall of integrins, four papers appeared in 2000 to show that the tetraspanin, CD9, was essential for sperm–egg fusion (Kaji et al. 2000, Le Naour et al. 2000, Miller et al. 2000, Hackney et al. 2000).
lar Ca2+ activation is an increase in the concentration of intracellular Ca2+. This is essentially a problem of signal transduction and different species appear to have adopted different approaches to triggering the Ca2+ increase (Stricker 1999, Runft et al. 2002). In mammals, the sperm does not appear to provide the signal by a conventional interaction with a receptor linked to the production of the Ca2+-releasing messenger, inositol 1,4,5 trisphosphate, rather, it uses a more direct route. It first fuses then introduces a soluble signalling molecule that triggers Ca2+-release, a mode of signalling that has no precedent in cell biology. Like the problems outlined above candidate molecules have come and gone. The discovery of high PLC activity in sperm (Jones et al. 1998) and the subsequent cloning of a novel sperm-specific PLC, PLCζ, has provided the best candidate to date (Cox et al. 2002, Saunders et al. 2002). The final proof for PLCζ awaits the knock-out mouse. The case for PLCζ being the Ca2+-releasing factor that triggers Ca2+ oscillations at fertilization is made by Swann and colleagues (2004) in the last review of this Focus issue.

The initiation of Ca2+ oscillations by the fertilizing sperm sets the egg on its path through embryogenesis. The downstream effectors of Ca2+ release and how they lead to the initiation of embryogenesis are starting to be uncovered. This topic is considered by two research papers in this issue that address the roles of Src-related kinases in mammalian fertilization. The outcome of these papers is that, unlike the situation in some invertebrates (Stricker 1999, Runft et al. 2002), Src-related kinases do not take part in the generation of Ca2+ release in mammals (Kurokawa et al. 2004). Instead, it plays a role downstream of Ca2+ in the initiation of cell cycle resumption (Talmor-Cohen et al. 2004).

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