Institution:
University of Cambridge

Unit of Assessment:
UoA5

Title of case study:
Development of kisspeptin analogues as therapeutic targets for reproductive disorders

1. Summary of the impact (indicative maximum 100 words)

The work of Colledge and colleagues between 2000 and 2007 has identified and characterised a molecule which is an important regulator of fertility: the neuropeptide kisspeptin.

The identification of its role in fertility has led to kisspeptin and its analogues being tested in clinical trials to make IVF treatment safer (Phase II: one trial), and as therapeutic agents for reproductive system conditions such as delayed puberty, menopause and absence of menstruation (Phase I: four trials). In April 2013, 11 months after the start of the Phase II IVF study, a healthy baby has been born to a participant treated with kisspeptin. Patients enrolled in these fertility trials have testified to the improvement in quality of life which the hope of being able to conceive that this alternative to conventional IVF has brought them.

2. Underpinning research (indicative maximum 500 words)

Disorders of the reproductive system are of increasing prevalence; the number of infertile couples worldwide has increased from 42 million in 1990 to 48.5 million in 2010 (Mascarenhas et al (2012) National, Regional, and Global Trends in Infertility Prevalence Since 1990: A Systematic Analysis of 277 Health Surveys. PLoS Med 9(12): e1001356. doi:10.1371/journal.pmed.1001356). Infertility treatment comes at a cost to the NHS, and carries risks for the patients: *e.g.,* IVF treatment using human chorionic gonadotrophin (hCG) can result in the potentially life threatening condition, ovarian hyperstimulation syndrome (OHSS). To increase positive outcomes to treatment, it is necessary to understand the molecular causes of these disorders allowing the development of novel therapeutic interventions.

Research carried out between 2000 and 2007 in the group of Prof Colledge, Professor of Reproductive Physiology in the Department of Physiology, Development and Neuroscience in Cambridge since 2009 (Reader in Molecular Physiology 2001-2009, Lecturer 1995-2000), identified the critical role that kisspeptins play in regulating the reproductive axis.

Secretion of gonadotrophic releasing hormone (GnRH) from the hypothalamus represents the final step from the brain in the control of reproduction. Prior to the research, it was known that kisspeptins were a ligand for the G protein–coupled receptor GPR54, but the physiological role of this interaction was not known. In research carried out between 2000 and 2003 in the group of Prof Colledge, using *Gpr54*-null mouse models, and in parallel in the group of William Crowley and Stephanie Seminara at Harvard, Boston, USA, on patients lacking pubertal development with mutations in GRP54, the role of GPR54 in activating the reproductive axis through secretion of gonadotrophic releasing hormone (GnRH) from the hypothalamus was identified.

Gpr54-deficient mice were generated in collaboration with Takeda Cambridge Ltd (part of Takeda Pharmaceutical Company, Japan). Characterization of the mutant mice identified that the *Gpr54*-null mice did not display any of the physiologic changes associated with sexual maturation. Mutations in *Gpr54* in mice caused hypogonadotropic hypogonadism, similar to that found in humans with mutations in *GPR54*. Taken together, these observations established that kisspeptin signalling is conserved between mice and humans and is a critical determinant of puberty (Ref 1, Section 3).

Further research carried out in the group of Prof Colledge between 2002 and 2005 on gpr54-/- mice, in collaboration with Alain Caraty from the Institut National de la Recherche Agronomique in France and Takeda Cambridge Ltd, showed that the kisspeptin receptor was expressed by GnRH neurons and kisspeptin required this receptor to directly stimulate GnRH release (Ref 2, Section 3). This was the first paper to show that kisspeptin could stimulate GnRH release which provided important data about using it for this purpose in IVF treatment.

In a third study, carried out from 2003 to 2007 in a collaboration between the group of Prof Colledge and Takeda Cambridge Ltd., mice deficient in the *Kiss1* gene, encoding kisspeptin, were generated to establish whether kisspeptins are the authentic agonists of GPR54 *in vivo* and to determine whether these ligands have additional physiological functions. *Kiss1*-null mice were found to be viable and healthy with no apparent abnormalities, but failed to undergo sexual

maturation (Ref 3, Section 3). The phenotype of *Kiss1*-null mice was virtually identical to that of *Gpr54*-null mice (*c.f.* Ref 1, Section 3). The study provided direct proof that kisspeptins are the true physiological ligand for the GPR54 receptor *in vivo*. *Kiss1* also did not seem to play a vital role in any other physiological processes other than activation of the hypothalamic–pituitary–gonadal axis, and loss of *Kiss1* could not be overcome by compensatory mechanisms (Ref 3, Section 3).

Two further studies were carried out by the group of Prof Colledge between 2005 and 2008, in collaboration with Takeda Cambridge Ltd. The first study tested the mechanism of action of kisspeptin on stimulating GnRH release. The results provided evidence for a potent stimulating effect of kisspeptin at GnRH nerve terminals of the mouse, and suggested a new point at which kisspeptin can act on GnRH neurons (Ref 4, Section 3). The second study addressed, using *Gpr54*- and *Kiss1*-null mice, whether kisspeptin and GPR54 have a key role in the activation of GnRH neurons to generate the luteinizing hormone (LH) surge responsible for ovulation. Whereas wild-type littermates all exhibited LH surges, none of the mutant mice from either line did. These observations provided the first evidence that kisspeptin–GPR54 signaling is essential for GnRH neuron activation that initiates ovulation, and broadened considerably the potential roles and therapeutic possibilities for kisspeptin in fertility regulation (Ref 5, Section 3).

Taken together these findings paved the way for the development of kisspeptins as treatment for disorders of the reproductive system.

3. References to the research (indicative maximum of six references)

1. Seminara, S.B., Messager, S., Chatzidaki, E.E., Thresher, R.R., Acierno, J.S., Shagoury, J.K., Bo-Abbas, Y., Kuohung, W., Schwinof, K.M., Hendrick, A.G., Zahn, D., Dixon, J., Kaiser, U.B., Slaugenhaupt, S.A., Gusella, J.F., O'Rahilly, S., Carlton, M.B., Crowley, W.F., Aparicio, S.A. and **Colledge, W.H.** (2003). The *GPR54* gene as a regulator of puberty. *N. Engl. J. Med.* **349**:1614-1627. DOI: 10.1056/NEJMoa035322

2. Messager S., Chatzidaki E.E., Ma D., Hendrick A.G., Zahn D., Dixon J., Thresher R.R., Malinge I., Lomet D., Carlton M.B., **Colledge W.H.**, Caraty A., Aparicio S.A.J.R. (2005) Kisspeptin directly stimulates gonadotropin-releasing hormone release via G protein-coupled receptor 54. *PNAS* **102**: 1761-1766. DOI:10.1073/pnas.0409330102

3. Xavier d'Anglemont de Tassigny, Lisa A. Fagg, John P. C. Dixon, Kate Day, Harry G. Leitch, Alan G. Hendrick, Dirk Zahn, Isabelle Franceschini, Alain Caraty, Mark B. L. Carlton, Samuel A. J. R. Aparicio, and **William H. Colledge**. (2007). Hypogonadotropic hypogonadism in mice lacking a functional *Kiss1* gene. *Proc Natl Acad Sci* **104**: 10714-10719. DOI: 10.1073/pnas.0704114104

4. de Tassigny, X.A., Fagg, L.A., Carlton, M.B. and Colledge, W.H. (2008). Kisspeptin can stimulate GnRH release by a direct action at GnRH nerve terminals. *Endocrinology* **149**: 3926-3932. DOI: 10.1210/en.2007-1487

5. Clarkson, J., d'Anglemont de Tassigny, X., Moreno, A.S., Colledge, W.H. and Allan E. Herbison. (2008). Kisspeptin–GPR54 signaling is essential for preovulatory GnRH neuron activation and the luteinizing hormone surge. J. Neurosci. **28(35)**: 8691– 8697. DOI:10.1523/JNEUROSCI.1775-08.2008

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- Ford Physiology Fund University Endowment; allocation of £30,000 to Prof William Colledge, 2000-2004
- Financial support from Takeda Cambridge Ltd (formerly Paradigm Therapeutics), Cambridge towards animal maintenance costs and consumables, 2000-2008, £55,000
- BBSRC grant BB/C003861/1 "Investigating the role of the G-protein coupled receptor GPR54 in regulating the mammalian reproductive axis" to Prof William Colledge, 2005-2008, £279,709
- BBSRC grant BB/F01936X/1 "Determining the role of kisspeptins in the peripheral control of ovarian physiology and pregnancy" to Prof William Colledge, 2008-2011 £482,149

4. Details of the impact (indicative maximum 750 words)

IMPACTS ON HEALTH AND WELFARE:

New clinical intervention is being trialled

The basic research carried out by Prof Colledge identified a completely new molecule involved in activating the reproductive axis at puberty. The knowledge gained from this research allowed the development of a **new clinical intervention** which has been **trialled with patients**:

IVF treatment

A Phase II clinical trial was started by Dr Waljit S Dhillo, Imperial College London, in 2012 at Hammersmith Hospital London, to investigate whether administration of kisspeptin to women can result in oocyte maturation (NCT01667406; Ref 1, Section 5). The significant expected advantage (to be demonstrated through this trial) of kisspeptin over current treatments to stimulate ovulation during IVF treatment (ie administering human chorionic gonadotrophin (hCG)) is a more physiological increase in reproductive hormones and oocyte maturation during IVF treatment. So far, early results in 30 women who have participated showed kisspeptin could be used to stimulate egg release in a gentler, more physiological way, without leading to ovarian hyperstimulation syndrome. Kisspeptin stimulated egg release in 29 of the 30 women, and 28 of the women were then able to use their eggs to attempt IVF (Ref 7 (BBC news 18/6/2013), Section 5).

Reproductive disorders

The research identifying kisspeptin as an activator of the reproductive axis has opened the possibility to use kisspeptin analogues as tools for characterizing certain reproductive disorders. Four Phase I clinical trials of a truncated form of kisspeptin (kisspeptin112-121), in combination with GnRH (NCT00914823; Ref 2, Section 5), or on its own (NCT01438073, NCT01438034, NCT01862094; Refs 3-5, Section 5) are being conducted at Massachusetts General Hospital or at the Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD) in the US, for conditions including Hypogonadotropic Hypogonadism and Delayed Puberty. Completion dates range from Dec 2013 to March 2015.

In addition, pilot studies carried out at Imperial College London, led by Dr Waljit S Dhillo, have identified that a single kisspeptin-54 injection acutely stimulates the release of reproductive hormones in women with hypothalamic amenorrhea (HA), a commonly occurring condition characterized by absence of menstruation. Women with HA who were treated with twice-weekly KP-54 injections had significantly elevated levels of reproductive hormones after 8 weeks as compared with treatment with saline. No adverse effects were observed (Ref 6, Section 5).

Outcomes for patients or related groups have improved

The trial started in June 2012, and by July 2013 one baby - a **healthy boy** called Heath - has been born. “Heath's mother, Suzie Kidd who is 34 and from Hitchen, says she is ecstatic that she was chosen to take part in the trial. [..] "We are so, so grateful."” (Ref 7 (BBC news 18/6/2013), Section 5). Women enrolled in this clinical trial have expressed what an **improvement in quality of life** the hope of successful treatment is giving them through a blog that was started on 6/5/13; by 31/7/13 the blog had 900 entries (Ref 8, Section 5; quotes: “This is an amazing opportunity for us” Entry #7, 9/3/13; “I had my EC today and we have 11 eggs!!!!!!! We are so pleased.” Entry #32, 21/3/13; “Just had a call from the hospital and we have 9 embryo's!!! Very happy” Entry #34, 22/3/13).

IMPACTS ON SOCIETY, CULTURE AND CREATIVITY:

Prof Colledge has discussed his work on the role of kisspeptin in reproduction from 2008 to 2011 at the Science Festival in Cambridge, reaching 1600 members of the public overall. The breakthroughs of the Phase I and II clinical trials of kisspeptin in IVF treatment have been showcased on the BBC News, on BBC Radio 4 Women’s Hour, in articles in the Guardian, the Telegraph, the Daily Mail, and a plethora of international online health information/news sites, women’s / family magazines, specialist fertility publications, science / research news sites and blogs (examples are given in Refs 7-10, Section 5). Through the coverage in the Guardian, Telegraph and Daily Mail alone (based on their traffic figures) an estimated 3 million people have been informed of the importance of kisspeptin, millions more through the BBC programmes and online media, and the blogs provide evidence of debate that has been stimulated.

5. Sources to corroborate the impact (indicative maximum of 10 references)

Trials:

1. *Drug*: Kisspeptin; *Study title:* The Use of the Hormone Kisspeptin in IVF Treatment (Phase II); *Sponsor / collaborators:* Imperial College London; *Dates*: 01/06/2012-01/06/2015
http://clinicaltrials.gov/show/NCT01667406
2. *Drug*: Kisspeptin 112-121 / GnRH; *Study title:* Administration of Kisspeptin 112-121 to Healthy Subjects and Subjects With Hypogonadotropic Hypogonadism (Phase I); *Sponsor / collaborators:* Massachusetts General Hospital; *Dates*:01/03/2009-01/12/2013
http://clinicaltrials.gov/ct2/show/NCT00914823
3. *Drug*: Kisspeptin 112-121; *Study title:* Elucidating Kisspeptin Physiology by Blocking Kisspeptin

Signaling (Phase I); *Sponsor / collaborators:* Massachusetts General Hospital; *Dates*: 01/09/2011-01/09/2013; http://clinicaltrials.gov/ct2/show/NCT01438073
4. *Drug*: Kisspeptin 112-121; *Study title:* Kisspeptin in the Evaluation of Delayed Puberty (Phase I); *Sponsor / collaborators:* Massachusetts General Hospital; *Dates*: 01/11/2011-01/11/2014*;* http://clinicaltrials.gov/ct2/show/NCT01438034
5. *Drug*: Kisspeptin 112-121; *Study title:* Blockade of Kisspeptin Signaling in Postmenopausal Women (Phase I); *Sponsor / collaborators:* Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD) ; *Dates*: 01/05/2013-01/03/2015 http://clinicaltrials.gov/ct2/show/NCT01862094
6. Jayasena et al (2010) Twice-Weekly Administration of Kisspeptin-54 for 8 Weeks Stimulates Release of Reproductive Hormones in Women With Hypothalamic Amenorrhea. Clin Pharmacol Ther 88: 840-847 DOI: 10.1038/clpt.2010.204

Examples of Press Coverage

7. *BBC News:*
17/3/2009: Hormone 'to restart reproduction' http://news.bbc.co.uk/1/hi/health/7945600.stm
18/6/2013: IVF: First baby born using 'safer' method http://www.bbc.co.uk/news/health-22935211
8. *Blog of women enrolled in Phase II clinical trial:*
http://babyandbump.momtastic.com/ttc-groups/1766667-hammersmith-kisspeptin-ivfers.html
9. *BBC Radio 4*: Women's hour, 17 March 2009: Kisspeptin: New Infertility Treatment
http://www.bbc.co.uk/radio4/womanshour/04/2009_11_tue.shtml
10. *The Guardian:* http://www.guardian.co.uk/society/2013/jun/18/hormone-breakthrough-fertility-kisspeptin