

Robert Geoffrey Edwards and Patrick Christopher Steptoe's Clinical Research in Human in vitro Fertilization and Embryo Transfer, 1969-1980

The biomedical accomplishment of human in vitro fertilization and embryo transfer (IVF-ET) took years to become the successful technique that presently enables infertile couples to have their own children. In 1969, more than ten years after the first attempts to treat infertilities with IVF technologies, the British developmental biologist Robert Geoffrey Edwards fertilized human oocytes in a Petri dish for the first time. In 1970, Edwards and his research partner, gynecologist and surgeon Patrick Christopher Steptoe started working with human patients with complicated and individualized gynecological conditions. It took Edwards and Steptoe another eight years of modifying medical procedures, as well as of dealing with the ups and downs of funding situations and public opinions before they could celebrate birth of the first baby conceived through IVF-ET in 1978.

In 1969, Edwards and his research team fertilized human ova with the aid of a designed culture medium, Bavister's medium. The human oocytes they fertilized were different from the eggs that are fertilized within the human body, as they had matured in vitro instead of in the ovarian follicles. Edwards soon found these in vitro eggs had difficulty in developing into blastocysts and often displayed aberrant morphologies. Informed by relevant literature on in vitro egg abnormalities seen with rabbits and pigs, Edwards believed that normal development and pregnancy could only be established when human ova have matured in vivo. He concluded that for IVF-ET to work as a viable fertility treatment, the matured eggs had to be retrieved directly from female patients.

Edwards's research partner, Patrick Steptoe, working at the Oldham General Hospital, in Oldham, England, had been using a laparoscope to examine pelvic cavities and conduct intrapelvic operations. Through the use of laparoscopy, he developed a procedure to retrieve maturing eggs from human ovaries. Also, he carefully selected woman volunteers, either infertile themselves or having infertile husbands, and gave them doses of the fertility drug Pergonal. This drug contains hormones that stimulate the growth of ovary follicles early in the menstrual cycle. Between day nine and day eleven of the women's menstrual cycles, Steptoe administered another fertility drug, Pregnyl, composed of human chorionic gonadotropin (hCG), to stimulate ovulation. Steptoe's new hormonal regimen often resulted in the ovulation of four to six eggs, instead of only one egg in a usual menstrual cycle, a case of superovulation.

The day after the injection of hCG into a woman, Steptoe and his surgical team examined the woman's ovaries and aspirated the swelling blue-reddish follicles. The surgeons then isolated the maturing oocytes from the follicle fluids. At the same time, Edwards had a culture of spermatozoa from the woman volunteer's husband ready. He transferred the retrieved eggs into the spermatozoon culture and finally incubated them for fertilization and for the resulting zygotes' development.

By April 1970, Steptoe and Edwards had obtained eggs from forty-six women. With these volunteers' help, Edwards and Steptoe monitored the effects of different doses of stimulating hormones and measured the varied fertilization rates in different culture media. As they kept modifying and optimizing the IVF procedures, they saw many fertilized eggs divide into 2-, 4-, 8-, and 16-cell stages. Four of these embryos formed blastocysts in culture five days after fertilization. Although Edwards was eager to transfer the embryos into their mothers' uteri for further development, he squashed, stained, and examined the embryos microscopically to make sure that the embryos developed in vitro were normal.

Planning for transferring the embryos from the dish into the patient's uterus, Edwards and Steptoe felt the need for better facilities to assist with their work. The Oldham General Hospital did not have a laboratory that could sustain the level of sterility required by clinical work in IVF and embryo transfer. Taking advantage of his professional network, Edwards acquired permission for developing an IVF research laboratory, attached to an operating room, in Newmarket General Hospital in Cambridge.

Confident about the prospect of their work, Edwards and Steptoe submitted a grant proposal to the Medical Research Council applying for funds to subsidize the cost in building new facilities. This application was turned down based on the ethical concerns about using human subjects for IVF-ET research. In its rejection letter, the MRC expressed concerns about potential risks involved in human IVF research and suggested that work on other primates be done first. Edwards and Steptoe found the MRC's reasoning hard to accept. They noted that monkeys were rather bad model organism for studying human IVF: monkeys do not react to fertility drugs and their eggs were difficult to fertilize in vitro.

In the early 1970s, public concerns about Edwards and Steptoe's work in human IVF arose in many forms. Newspapers such as Daily Express, The Times, and The Sun published dozens of articles making claims about the moral issues involved in the human IVF research and social perils that the research's future applications might bring about. Edwards and Steptoe strived to address these concerns by clarifying their goals and their important scientific achievements. They did not expect that their proposal to the MRC would be rejected based on what they regarded as ungrounded moral issues.

Eventually, with the financial supports from the Oldham Area Health Authority and from a few American philanthropists who became interested in the human IVF research, Edwards and Steptoe managed to establish an adequate facility at Kershaw's Cottage Hospital in Royton in 1971. There, they started their trials to transfer 8-cell embryos into volunteer mothers. Steptoe used a specially designed cannula to guide the developing tiny embryo carried by a drop of culture medium through the cervical canal into the recesses in the uterus.

The move to the new research facility did not, however, bring about an immediate propitious start. Quite the contrary, the different working location, and equipment, ranging from culture dishes, pipettes, medical appliance, batches of culture media, and brands of other solutions, seemed to have jointly altered the experimental conditions. The fertilization rates in vitro significantly declined from what had previously been attained. Even the few fertilized eggs did not lodge to the wombs, and the follow-up hormonal pregnancy tests after embryo transfers constantly gave negative results.

The period between 1971 and 1974 was difficult for Edwards and Steptoe's research. Ethical criticisms propagated and intensified in the media. The variations in the new experimental setting stymied research progress. Steptoe's increasingly painful arthritis condition made the research ever more taxing. Edwards and Steptoe nevertheless persisted. As they checked one solution after another for the causes of the drifting of experimental conditions, they also began to realize that the fertility drugs for stimulating superovulation had the side effect of shortening the menstrual period. So, by the time that researchers had made an 8-celled embryo ready for transfer, the mother's womb was just in the phase of preparing to shed its linings, making implantation of the embryo impossible.

Knowing that hCG was often prescribed to protect early pregnancy, Edwards and Steptoe started to use a hormonal regimen that continued hCG injections until seven weeks after embryo transfer, hoping that the sustained level of hCG would keep the lining of the uterus intact. This regimen worked in some cases. In the summer of 1975, Edwards and Steptoe obtained their first positive result from a urine pregnancy test of a patient. Five weeks later, Steptoe could examine the developing embryo through ultrasound. Although it was eventually shown to be an ectopic tubal pregnancy and was aborted, the proof-of-concept pregnancy greatly encouraged Edwards and Steptoe.

Using the prolonged hCG regimen, Edwards and Steptoe achieved two more pregnancies by 1976. But these implanted embryos were soon rejected by the mother's womb and spontaneously aborted. The researchers started to recognize that the hormonal dynamic that caused the uterus to retain the implanted embryo was forbiddingly complex. The hCG could not always do the trick—as it lengthens

the menstrual cycles, it often interrupts the balance of other ovarian hormones such as estrogen and progesterone.

Edwards and Steptoe began to try out solutions that did not use hormonal stimulations. They froze some developing embryos to recover them when the mother's uterus was ready, but failed in reinitiating development after thawing the embryos. With no alternative, Edwards and Steptoe finally decided to give up superovulation and follow the timing of the patient's own menstrual cycle. They set up a plan to retrieve eggs from the single developing follicle in ovaries during the late follicular phase of menstrual cycle so that, when the embryo was introduced, the uterus linings would still be thick without the disturbance from excessive hormones.

Edwards and Steptoe's new plan posed heavy demands on both the researchers and patients. As ovulation happens about 36 hours after the rapid increase of luteinizing hormone level, the LH surge, researchers had to monitor the LH concentrations to appropriately calculate the timing for ovum retrieval. Accordingly, the patients had to reliably collect their urine samples once every three hours for inspection. Submitting to the rhythm of human hormonal changes, the surgical team had to be on call around the clock.

In the fall of 1976, Lesley Brown, a female patient with blocked and distorted fallopian tubes, was admitted to the Kershaw's Cottage Hospital. Seeking a child of her own, in November 1977, she went through the laparoscope-aided ovum-retrieval operation that was scheduled according to her own hormonal cycle. One week later, Steptoe introduced into her uterus an 8-celled embryo that was developed from the zygote formed through the in vitro fusion of her retrieved egg and her husband, John Brown's, spermatozoon. Lesley Brown was the second patient to complete the new IVF plan without hormonal intervention and the first one to bring an IVF embryo to term. She delivered Louise Joy Brown, the first human baby generated from IVF, on 25 July 1978.

As the world marveled at the birth of what the media called the first test-tube baby, the Browns were overjoyed with the child they thought that they could never have. Although delighted in their triumph, Edwards and Steptoe reported their success in *Lancet* letter. With many patients becoming hopeful after the birth of Louise, the biologist and the doctor aimed to establish an IVF-ET routine that could attain a stable and acceptable pregnancy rate. By 1980, they would carry out their hectic IVF-ET strategy on 68 patients and uncover even more insights about human reproduction, as summarized in their 1980 paper "Establishing Full-Term Human Pregnancies Using Cleaving Embryos Grown in vitro."

Sources

1. Edwards, Robert G. "The Early Days of in vitro Fertilization." In *Pioneers in In Vitro Fertilization: The Proceedings of a Symposium held in Oss, The Netherlands, November 5, 1993*, eds. A. Th. Alberda, R. A. Gan, and H. M. Vemer, 7-24. The Parthenon Publishing Group, 1995.
2. Edwards, Robert G. and Patrick C. Steptoe. *A Matter of Life: The Story of a Medical Breakthrough*. William Morrow, 1980, 87-156.
3. Edwards, Robert G., Patrick C. Steptoe, and Jean M. Purdy, "Establishing Full-Term Human Pregnancies Using Cleaving Embryos Grown in vitro." *British Journal of Obstetrics and Gynaecology* 87 (1980): 737-56.
4. Edwards, Robert G., Patrick C. Steptoe, and Jean M. Purdy, "Fertilization and Cleavage in vitro of Preovulator Human Oocytes." *Nature* 227 (1970): 1307-09.
5. Maienschein, Jane. *Whose View of Life? Embryos, Cloning, and Stem Cells*. Harvard University Press, 2003.
6. Steptoe, Patrick C., and Robert G. Edwards, "Birth after the Reimplantation of a Human Embryo." *The Lancet* 2 (1978): 366.
7. Steptoe, Patrick C., and Robert G. Edwards, "Laparoscopic Recovery of Preovulatory Human Oocytes after Priming of Ovaries with Gonadotrophins," *The Lancet* 295 (1970): 683-89.
8. Steptoe, Patrick C. and Robert G. Edwards, "Reimplantation of a Human Embryo with Subsequent Tubal Pregnancy," *The Lancet* 307 (1976): 880-82.

9. Steptoe, Patrick C., Robert G. Edwards, and Jean M. Purdy, "Human Blastocysts Grown in Culture," *Nature* 229 (1971): 132-33.