

# 10

## EXPERIMENT TEN

# Mammalian Uterus

The isolated uterus of a rat or guinea pig provides a good example of a tissue containing inhibitory  $\beta_2$ -adrenoreceptors. Stimulatory agonists such as oxytocin can also be studied in this preparation.

Sympathomimetic amines act at uterine  $\beta$ -adrenoceptors to relax muscle that has been precontracted with potassium chloride. Oxytocin stimulates both the frequency and force of slow waves of contraction in uterine smooth muscle. These effects are highly dependent on the presence of oestrogen, the immature uterus being quite resistant. Oestrogen may increase the oxytocin receptor numbers, and enhance intercellular coupling by inducing the formation of gap junctions.

### Dissection

Untreated, disoestrous guinea pigs at days 6–10 of the 16-day cycle may be used. It is very difficult to obtain rats at the same cycle stage out of the 4–5 day cycle, so they should be pretreated with oestradiol cypionate (20  $\mu\text{g}/\text{kg}$ , subcutaneous) 24 hr before use. Each uterine horn is removed and mounted in an organ bath, with a thread at one end arranged so that longitudinal contractions are recorded by a force transducer. Allow 45 min equilibration, washing at 15-minute intervals.

Plate 16 shows the dissection of a female rat, with the uterine horns visible (the forked structure); Plate 17 shows the uterus dissected out.

### Experiment

#### Chart Settings

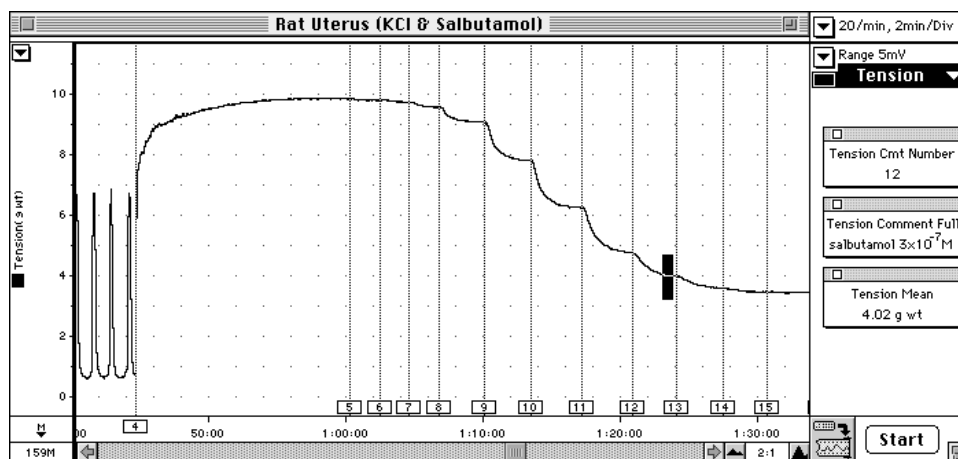
This is a single-channel arrangement with a PowerLab, Bridge Amp, and force transducer. In Chart, the range should be chosen to suit the maximum force to be exerted on the transducer. The final scale after units conversion and so on should be 1 to 10 g wt. The sampling rate can be as low as 20 samples/min (contractions are very slow). The view compression should be 1:1 to 5:1. A high-pass filter of 2 Hz should be chosen in the Bridge Amplifier dialog box. Data Pad miniwindows can be set up to show the comment number and text, and the mean of the selection or value at the active point (using the Mean function).

## Protocol

After equilibration, add KCl 40 mM to precontract the muscle. After a further 10–15 min, when the contraction has stabilised, inhibitory agonists can be tried. The inhibition shows little fade, so it is easy to construct a cumulative dose–response curve. For both adrenaline and the  $\beta_2$  agonist salbutamol, satisfactory test concentrations are  $10^{-10}$  M,  $3 \times 10^{-10}$  M,  $10^{-9}$  M,  $3 \times 10^{-9}$  M, and so on, through to  $10^{-5}$  M. Figure 10–1 shows contractions of rat uterine muscle, with spontaneous activity at the left. KCl was added to precontract the muscle, followed by increasing concentrations of salbutamol to show the cumulative dose–response relation. Antagonism by a beta-blocker can be demonstrated with propranolol  $10^{-7}$  M, allowed to equilibrate for at least 20 min.

**Figure 10–1**

Relaxation of precontracted rat uterine muscle, with spontaneous contractile activity at left. KCl (40 mM) added at comment #4 precontracts the muscle. Increasing concentrations of salbutamol ( $10^{-10}$  M to  $10^{-5}$  M) added at the other comments show a cumulative effect.

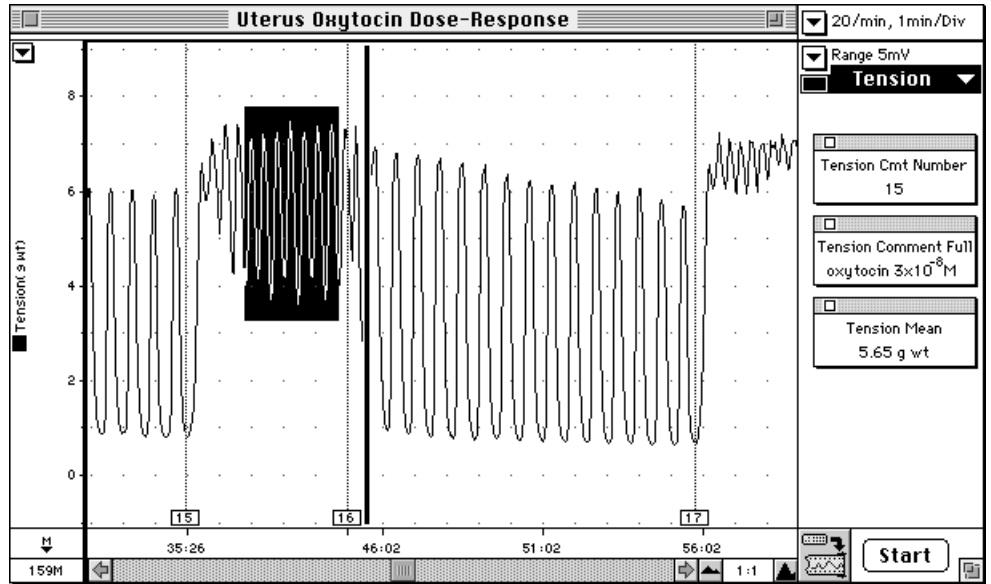


## Further Work

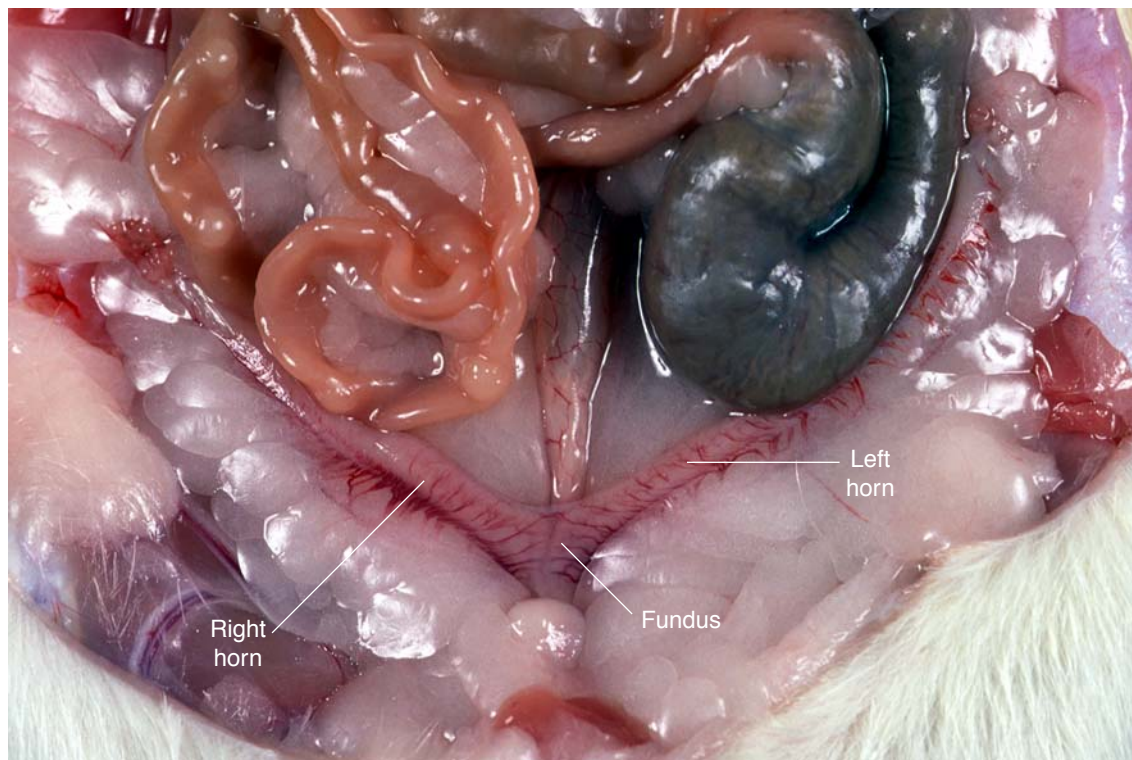
Stimulatory agonist effects can also be recorded. For oxytocin, a suitable range of concentrations is  $10^{-10}$  M to  $10^{-6}$  M. For carbachol, a suitable range is  $10^{-9}$  M to  $3 \times 10^{-5}$  M. Since the effect of these agonists is to increase both spontaneous activity and resting tone, there are difficulties in quantifying the responses. One approach is to measure the mean tension over several minutes. The Data Pad can be set up so that a miniwindow shows this information. The effect of indomethacin  $10^{-5}$  M on oxytocin responses can be used to investigate the possible role of prostaglandin synthesis in oxytocin agonism.

Figure 10–2 shows contractions of rat uterine muscle, with spontaneous activity at left. A miniwindow shows the mean of the selected data during the response to oxytocin. A similar selection would be used to quantify the control value (in the absence of an agonist).

**Figure 10-2**  
Contractions of rat uterine muscle, with spontaneous activity at left. Oxytocin ( $3 \times 10^{-8}$  M) was added at comment #15.



**Plate 16.** Female rat genitalia; dissection of uterine horns in situ.



**Plate 17.** Rat uterus dissected out.

