Electronics for electrophysiologists II

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Plan

- Filtering
- Aliasing
- Extracellular stimulation
- Loose-cell attached stimulation and recording
- Grounding and shielding
- Data acquisition
- Analysis environments

Linear systems

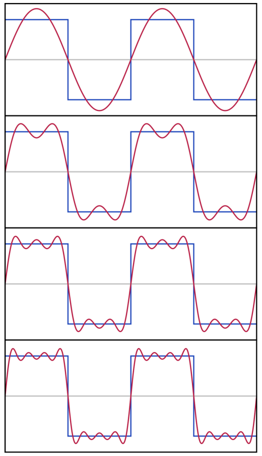
Filters are linear systems. Define a function f with inputs a, b and a multiplicative constant k. f is linear when the following are true:

- $f(a) + f(b) = f(a + b)$ (Superposition)
- $f(ka) = kf(a)$ (Homogeneity)

Linearity may require a restricted domain (e.g. within power supply limits of an electronic circuit).
Nonlinear functions are approximately linear for small changes (derives from series expansion).
a, b, f can be functions of time.
Filters are also *shift invariant*—properties are not time dependent.
Thus *LSI*: Linear Shift Invariant.

Fourier transform

Any periodic signal can be represented as a series of sinusoids.



Using sinusoid inputs to characterize an LSI system

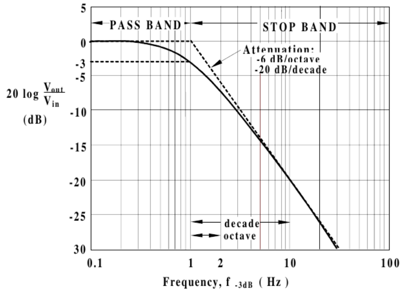
- Sinusoid input $\rightarrow$ LSI $\rightarrow$ sinusoid output *at same frequency*
- Amplitude and phase may change

Combining Fourier theory and properties of LSI systems, we can predict the response of a filter to any input if we know its response to sine waves.

- Decompose arbitrary input into sine waves
- Apply filter action to each sine wave (altering amplitude and phase)
- Recompose output by superposition

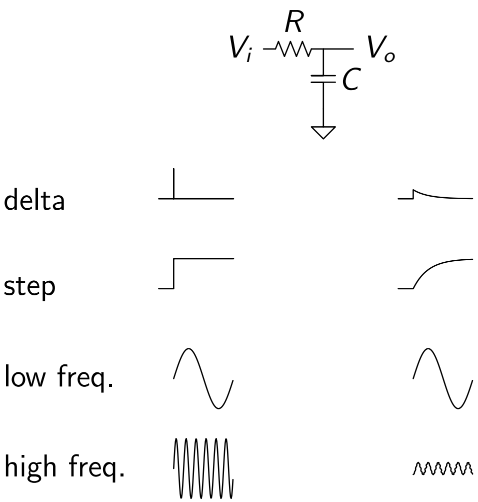
Filters are characterised by Bode plots

- Amplitude (dB) and phase (angle) vs frequency (logarithmic)

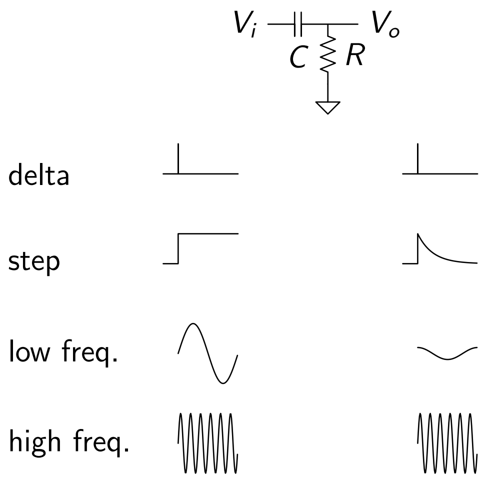


- decibels (dB) $= 20\log_{10}(A_o/A_i)$
- 6 dB = 2-fold
- 20 dB = 10-fold
- Passband, corner frequency (-3 dB), rolloff (dB/decade), stopband

Simple low-pass filter



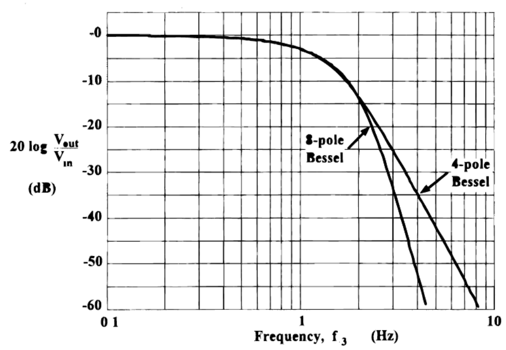
Simple high-pass filter



Cascading filters

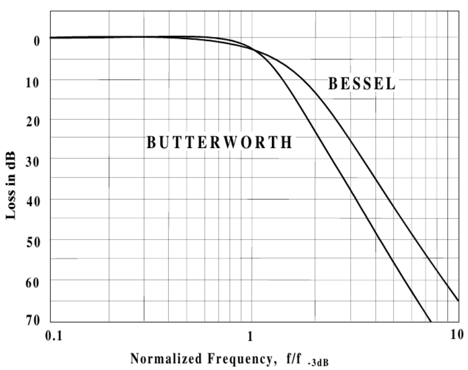
- Simple RC filters have a very slow rolloff (also poor output impedance)
- Cascading filters can produce faster rolloff
- Each "RC equivalent" is called a "pole"
- Making multipole filters from passive components (R, C, L) is very difficult
- Active filters are universally used (usually 2 poles per op-amp)
- 2 to 10 pole filters are common
- Many different types of multipole filters

Multipole filters—poles



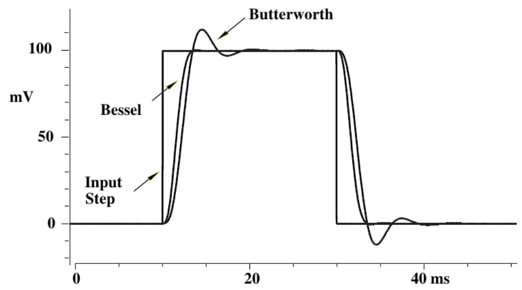
- Limiting slope $= -$poles * 20 dB/decade

Multipole filters—rolloff



- Bessel filters have a soft corner...

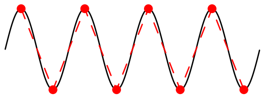
Multipole filters—pulse response



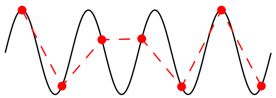
- ...but pulse response of Bessel filter does not ring
- Electronic low pass filters delay signals
- (Some offline digital filters—e.g. Gaussian—do not delay)

Aliasing

- Continuous signals are digitised (analog to digital conversion)



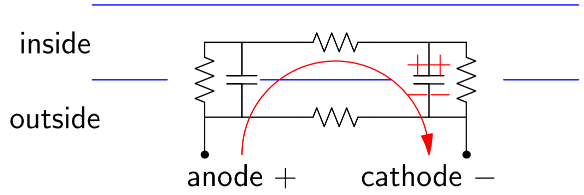
$f_{\text{sample}} = 2f_{\text{signal}}$ (Nyquist limit)



$f_{\text{sample}} < 2f_{\text{signal}}$ (Aliasing)

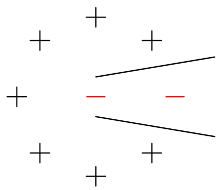
- Ideal: $f_{\text{sample}} > 2f_{\text{signal}}$, but there is often HF noise
- Use anti-aliasing filter to remove high-freq. signals
- **Signal is *not* negligible at filter *corner* frequency**
- Use f_{sample} 5–10*corner frequency of a multipole filter

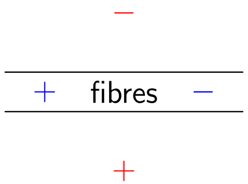
Extracellular stimulation



- Cathode stimulates (but cf. anode-break excitation)
- Neural extension in direction of voltage field required
- Current stimulation independent of electrode resistance (reproducible)
- Voltage stimulation has shorter artefact (charge dissipated by stimulator not electrode)
- Shield cables, even electrodes (but capacitance is problem in current mode)

Monopolar vs bipolar stimulation

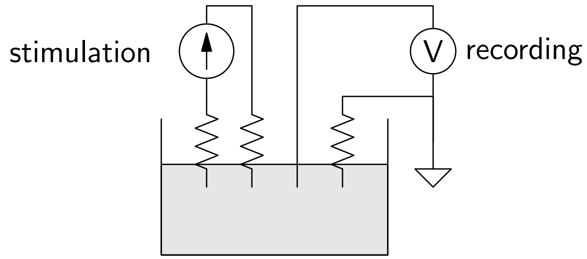




- Usually patch electrode and bath electrode ("∞")
- Very local
- Large artefact (high resistance, unbalanced voltage)

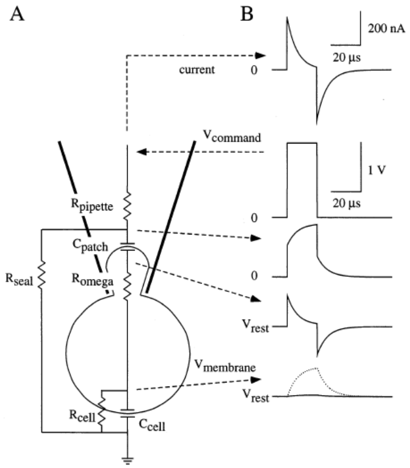
- Usually large metal structures
- Directed field (arrange field along fibres!)
- Smaller artefact

Isolated stimulation



- Stimulating current must not flow in bath electrode (large voltage drop)
- Stimulator must be isolated from ground
- Can use non-isolated stimulation with virtual ground

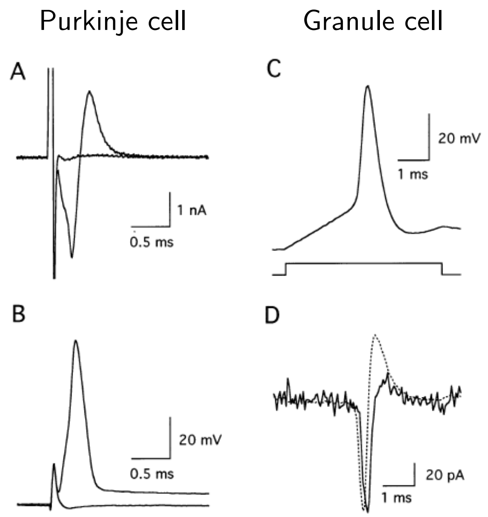
Loose cell-attached stimulation and recording



- Reuse dirty electrode
- No dialysis
- Requires 1–3 V (NPI ELC03)
- Cells usually die from electroporation

(Barbour & Isope, 2000)

Loose cell-attached stimulation and recording



Grounding and shielding

Electrostatic interference - grounded shielding is effective (95 % of problems)

- Grounded Faraday cage
- Ground everything metal near recording to prevent antenna effect (much modern equipment constructed of many isolated parts)
- Beware solution lines, bottles (drip chambers)

Magnetic interference (ground loops e.g. from multiple BNC shields) - grounded shielding is ineffective

- In principle cutting loops, reducing loop areas (twisted pairs) helps
- In practice find source (power supply) and remove it

Low cost data acquisition

- National Instruments card $<$ \$1000 (USD not pesos)
- WinWCP (electrophysiology) or WinEDR (combined with imaging) freeware (written by John Dempster, University of Strathclyde)
- Neuromatic (Jason Rothman, University College London) within IGOR Pro (Wavemetrics)
- LabView (NI)

Analysis environments

- PClamp etc
- IGOR Pro
- Matlab
- GNU R (excellent for statistics)
- Python (numpy, scipy, matplotlib)

Anlysis advice

Important to script (program) your analyses:

- Reproducible
- Verifiable
- Modifiable
- Objective

Comment and document your code! Even you won't understand it 6 months later.

Use simple, standard and compatible formats for data and results:

- Text (ascii, utf8)
- tiff, jpeg etc
- Simple and documented binary formats
- Easy to check and reuse in the future