Otto Loewi and Vagusstoff

One of the more colorful stories in the history of neuroscience was contributed by Otto Loewi, who, working in Austria in the 1920s, showed definitively that synaptic transmission between nerve and heart is chemically mediated. The heart is supplied with two types of innervation; one type speeds the beating of the heart, and the other slows it. The latter type of innervation is supplied by the vagus nerve. Loewi isolated a frog heart with the vagal innervation left intact, stimulated the nerve electrically, and observed the expected effect, the slowing of the heartbeat. The critical demonstration that this effect was chemically mediated came when he took the solution that bathed this heart, applied it to a second isolated frog heart, and found that the beating of this one also slowed. The idea for this experiment had actually come to Loewi in a dream. Below is his own account:

In the night of Easter Sunday, 1921, I awoke, turned on the light, and jotted down a few notes on a tiny slip of paper. Then, I fell asleep again. It occurred to me at six o’clock in the morning that during the night I had written down something most important, but I was unable to decipher the scrawl. That Sunday was the most desperate day in my whole scientific life. During the next night, however, I awoke again, at three o’clock, and I remembered what it was. This time I did not take any risk; I got up immediately, went to the laboratory, made the experiment on the frog’s heart, described above, and at five o’clock the chemical transmission of the nervous impulse was conclusively proved.... Careful consideration in daytime would undoubtedly have rejected the kind of experiment I performed, because it would have seemed most unlikely that if a nervous impulse released a transmitting agent, it would do so not just in sufficient quantity to influence the effector organ, in my case the heart, but indeed in such an excess that it could partly escape into the fluid which filled the heart, and could therefore be detected. Yet the whole nocturnal concept of the experiment was based on this eventuality, and the result proved to be positive, contrary to expectation.

(Loewi, 1953, pp. 33, 34)

The active compound, which Loewi called vagusstoff, turned out to be acetylcholine. As we shall see in this chapter, acetylcholine is also a transmitter at the synapse between nerve and skeletal muscle. Here, unlike at the heart, acetylcholine causes excitation and contraction of the muscle.
Role of Ach-Esterase on EPP time-course

\[
\text{ACh} \xrightarrow{\text{Acetylcholinesterase}} \text{Choline} + \text{acetate}
\]

\[
\text{CH}_3 \cdot \text{N}^+ \cdot \text{CH}_2\text{CH}_2\text{OCOCH}_3 + \text{H}_2\text{O} \rightarrow \text{Choline} + \text{acetate}
\]

Stimulate Motor Axon

Saline + Neostigmine

Saline

1 mV

5 msec
Ach-Esterase and Independence Between Release Sites
Role of Uptake at Glutamatergic and GABAergic CNS Synapses

Also the postsynaptic neuron and surrounding Astrocytes participate in transmitter uptake.

GABAergic Transmission
- GABA uptake blocked
  - Isaacson 1993

Glutamatergic Transmission
- Glutamate uptake blocked
  - Arnth-Jensen 2002
How do Vesicles Interact with the Presynaptic Membrane: Docking

The Snares
- syntaxin
- synaptobrevin
- SNAP-25
Botulinum toxin

clostridium botulinum

Exposure to clostridium botulinum usually through canned food.

Flaccid Paralysis
Botox

Treatment of spasm
Tetanus toxin

Exposure to *Clostridium tetani* (usually through a cut or scratch) causes “lockjaw”
Targets for Clostridial Toxins

Botulinum toxin: BoTX
Tetanus toxin: TeTX
What is the Ca²⁺ Sensor that Triggers Vesicular Fusion?

Diagram showing the interaction between various proteins and calcium ions during vesicular fusion.
Figure 5.6
Various sizes of CNS synapses. Notice that larger synapses have more active zones.

Synapse size

Active zones

Presynaptic terminals

Postsynaptic elements

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Electrical Synapses

(a) Diagram showing a gap junction between Cell 1 and Cell 2.

(b) Diagram of Connexons between the cytoplasm of Cell 1 and Cell 2, with channels formed by pores in each membrane.

Inset:

A. Macromolecules and Micromolecules
B. OUTSIDE INSIDE
C. NH₂ – COOH
Gap junctions between dendrites of inhibitory neocortical interneurons.  a | Electron-microscopic photograph showing a gap junction (gj) between two dendrites of basket cells (d1 and d2) in layer 4 of the adult primate motor cortex (magnification x 25,000).  b | Higher magnification of the gap junction (gj) illustrated in panel a. Note the characteristic multilayered structure and the typical accumulation of dense material in the dendritic cytoplasm adjacent to the gap junction (magnification 180,000) (From: Nature Reviews Neuroscience 2; 425-433 (2001);
The coupling coefficient

Neuron 1

Neuron 2

b

I_1

V_1

5 mV

V_2

2 mV

c

I_2

V_2

V_1
Because the input resistances $R_1$ and $R_2$ are not necessarily the same, $K_{21}$ and $K_{12}$ do not have to be equal.

The coupling coefficient

Diagram and electrical model of an electrical synapse

For a voltage at $V_1$: \[ \frac{V_2}{V_1} = \frac{R_2}{R_2 + R_c} = K_{12}. \]

For a voltage at $V_2$: \[ \frac{V_1}{V_2} = \frac{R_1}{R_1 + R_c} = K_{21}. \]
Both Neurons are voltage clamped. The potential of Neuron 2 (V₂) is kept at -60 mV. The potential of Neuron 1 (V₁) is depolarized and hyperpolarized from -60 mV in increments of 10 mV (ΔV₁). Because Neuron 1 and Neuron 2 are coupled via Gap junctions, currents injections (ΔI₂) are necessary to keep Neuron 2 at -60 mV, while Neuron 1 is being clamped at different potentials. The amount of ΔI₂ is proportional to the conductance of the gap junctions. The conductance (ΔI/ΔV; the reciprocal of the resistance) is given by the slope of the graph and is measured in siemens (1S = 1A/V = 1/Ω). In this particular example the conductance of the Gap junction Gc is about 1.17 nS (or the Junctional Resistance R Rc is 855 MΩ).

Measuring Rc (the junctional resistance) or Gc (the junctional conductance (1/Rc)).
Gap Junction Synchronize the Activity of Neuronal Populations
## Properties of chemical and electrical transmission

<table>
<thead>
<tr>
<th>Chemical</th>
<th>Electrical</th>
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<tbody>
<tr>
<td>Unidirectional</td>
<td>Can be bidirectional or unidirectional</td>
</tr>
<tr>
<td>Excitatory or Inhibitory</td>
<td>Sign conserving. Depolarization (or hyperpolarization) in one neuron leads to depolarization (or hyperpolarization) in the other</td>
</tr>
<tr>
<td>Delay of (\sim 0.5-1.0 ) msec</td>
<td>No delay other than from low pass filtering. Ideal for rapid communication</td>
</tr>
<tr>
<td>Amplification. Uses energy from ion gradients and can prolong response</td>
<td>Dissipative. Signals usually smaller in the coupled neuron. Time course limited by membrane time constant</td>
</tr>
<tr>
<td>Efficient for impedance mismatches</td>
<td>Inefficient when coupling between neurons of different input resistances</td>
</tr>
<tr>
<td>Plasticity (use dependent)</td>
<td>Coupling can be modulated by chemicals</td>
</tr>
<tr>
<td>Chemical communication primarily by exocytosis</td>
<td>Small molecules can pass between neurons</td>
</tr>
<tr>
<td>Chemical</td>
<td>Electrical</td>
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<tr>
<td>--------------------------------------</td>
<td>----------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Measurement of a reversal potential</td>
<td>Direct electrical coupling using simultaneous recordings between two or more cells</td>
</tr>
<tr>
<td>Small synaptic delay</td>
<td>No delay</td>
</tr>
<tr>
<td>Pharmacological experiments. Use antagonists of neurotransmitter receptors</td>
<td>Fluorescent dye injected into one neuron diffuses and fills other neurons</td>
</tr>
<tr>
<td>High Mg^{2+}/low Ca^{2+} will block transmission</td>
<td>High Mg^{2+}/low Ca^{2+} will not block transmission</td>
</tr>
<tr>
<td>Morphology. Separation of pre- and post-synaptic membranes</td>
<td>Presence of gap junctions</td>
</tr>
</tbody>
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