Finding, understanding, and applying primary research literature for the herbalist
Refining strategic approaches to exploring that space-in-between primary research and traditional knowledge

Michael Tims, Ph.D.
Academic Director, Herbal Program
Maryland University of Integrative Health

Primary research can provide additional layers of information to inform the decision making of medicine maker or clinician. Overlapping data from primary research can often support traditional knowledge. However, what do you do when such confluence is lacking or contradicts your own understanding? Let’s redefine some strategies together for creatively exploring that space-in-between.

Strategic Overview: Ecosystem Thinking
- Improvise that space-in-between
- Life asks questions
- Collaboration of ideas is more powerful than competition
- Does it lead to a compelling question?

This lecture will review four herbs, moving from simpler to more complex examples of evidence based research being applied to clinical settings. We will explore how evidenced based data provides a basis for herbalist to explore explicit questions in clinical settings.

1) Ephedra sinica
Ephedra clinical data – two phase clinical design (control vs. treatment)
Extraction and HPLC measurement of blood sera (blind) and Ephedra capsules
Ephedrine was the only alkaloid detected
\( K_a \) = rate of absorption
\( T_{max} \) = how long it stayed around

<table>
<thead>
<tr>
<th>Ephedrine treatment</th>
<th>( K_a ) (hr(^{-1}))</th>
<th>( T_{max} ) (hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Powdered herb</td>
<td>0.49</td>
<td>3.9</td>
</tr>
<tr>
<td>Extracted ephedra in formula</td>
<td>1.36</td>
<td>2.8</td>
</tr>
<tr>
<td>Ephedrine in tablet</td>
<td>1.73</td>
<td>1.69</td>
</tr>
<tr>
<td>Ephedrine in solution</td>
<td>2.35</td>
<td>1.81</td>
</tr>
</tbody>
</table>

2) Sambucus nigra
Combining studies on pharmacokinetics and in vitro studies
Used Madin-Darby Canine Kidney Epithelial Cells (MDCK) infected with KAN-1 (a human H5N1 virus) and propagated for 48 h with Rubini extract. Control untreated virus and cells were used for infection. The level of KAN-1 infected cells was significantly reduced with elderberry treatment. ($p < 0.07$)

Gram-positive and negative bacteria exposed to varied concentrations elderberry extract in liquid culture showed a steep decline at exposure to 5% elderberry extract.


(A) plasma before consumption of the elderberry extract

(B) plasma 30 min after

(C) plasma 60 min after

(D) elderberry extract containing 1.2 μg of anthocyanins
Plasma samples from elderly women of 720 mg anthocyanins.

(A) before

(B) 10 min after

(C) 20 min after

(D) 45 min after consumption

The 720 mg total anthocyanin is equivalent of 80 ml of extract. How do we relate the 5% bacterial exposure rate to a dosing regimen?

3) *Hypericum perforatum*


The assay design utilized radioligand binding and quantitative methods for determining substrate-enzyme binding to determine whether *Hypericum* based substrates bound active sites of receptors. Inhibitors were expected to bind more tightly than activating or functional substrates. The assay included the following individual receptors:

- G-protein-coupled receptors (GPCRs) - 5-HT, adrenergic, opioid, histamine, metabotropic glutamate, muscarinic acetylcholine
- Ligand-gated ion channels including GABA receptors
- Various neurotransmitter transporters

Bioactive compounds found in crude extract and tested:

- phenylpropanes
- flavonol derivatives
- biflavones
- proanthocyanidines
- xanthones
- phloroglucinols - hypercin
- amino acids
- naphthodianthrones - hyperforin
- essential oil constituents

The results identified several compounds in SJW that interacted with different *in vitro* receptors and transporters, particularly hyperforin that bound various serotonergic, noradrenergic, dopaminergic, cholinergic, and opioid receptors. The effects occurred via competitive inhibition, some were unanticipated binding events, the most potent included biflavonoid, amentoflavone, hypericin and
pseudohypericin. Hyperforin and the flavonoids showed only weak binding inhibition. This would seem to indicate that production via marker method is not always a sound choice.

Researchers described distinct spectra of activities across the assay system, with activity at dopaminergic, adrenergic, or opioid receptor sites. This last result speaks to a pattern of effect of which herbalist are quite familiar, and couches it in terminology accessible to the allopathic community.

Of note, concentration of compounds required for half-maximal receptor occupancy was quite high. Do we need to compare *in vivo* plasma concentrations following application of clinically relevant doses How does this inform requirements for growing conditions?

4) *Panax quinquefolius*

Investigated ginseng saponins, or ginsenosides, particularly the protopanaxadiols (Rb₁, Rb₂, Rd) and protopanaxatriols - Re, Rg₁, which together makeup 90% of ginsenoside content.


Tachikawa et al. hypothesized that gut flora play a role previously ignored, and thus the active compounds are metabolites of prodrug. They decided to focus on M for this study.

The adrenal medulla secretes catecholamine in response to stress. Over secretion can lead to exhaustion of target organ. The most abundant catecholamines are epinephrine (adrenaline), norepinephrine and dopamine. Ginseng saponins effectively inhibited the catecholamine secretion induced by acetylcholamine (ACh) by blockading ACh-induced Na⁺ influx into the cells through nicotinic ACh receptor-operated cation channels or receptors.

The found that catecholamine secretion was inhibited in a concentration dependent manner (cdm); that M had greatest inhibition vs. other metabolites and that the effect not completely reversible; the duration of exposure determined the potency of the effect; M reduced ACh induced ion movement in (cdm), supporting the Na⁺ efflux hypothesis. They concluded that ginsenoside metabolites inhibit secretion of catecholamines and may require interaction with microbiome for activation. Several interesting question remain, however. Can human gut flora be altered to more effectively bio-transform the ginsenosides? What initial concentration of ginseng will provide active levels of the metabolite? What role do multi-constituent ratios play in the final biotransformation?