pH: Definitions & Concepts

$$HA \leftrightarrow H^+ + A^-$$

Acid — Conjugate Base

$$B + H^+ \leftrightarrow BH^+$$

Base — Conjugate Acid

acid: donates protons (H^+)

base: accepts protons (H^+)

<u>strong acid/base</u>

- **completely dissociates**

<u>weak acid/base</u>

- **partially dissociates (i.e., equilibrium)**

$$pH = -\log[H^+]$$

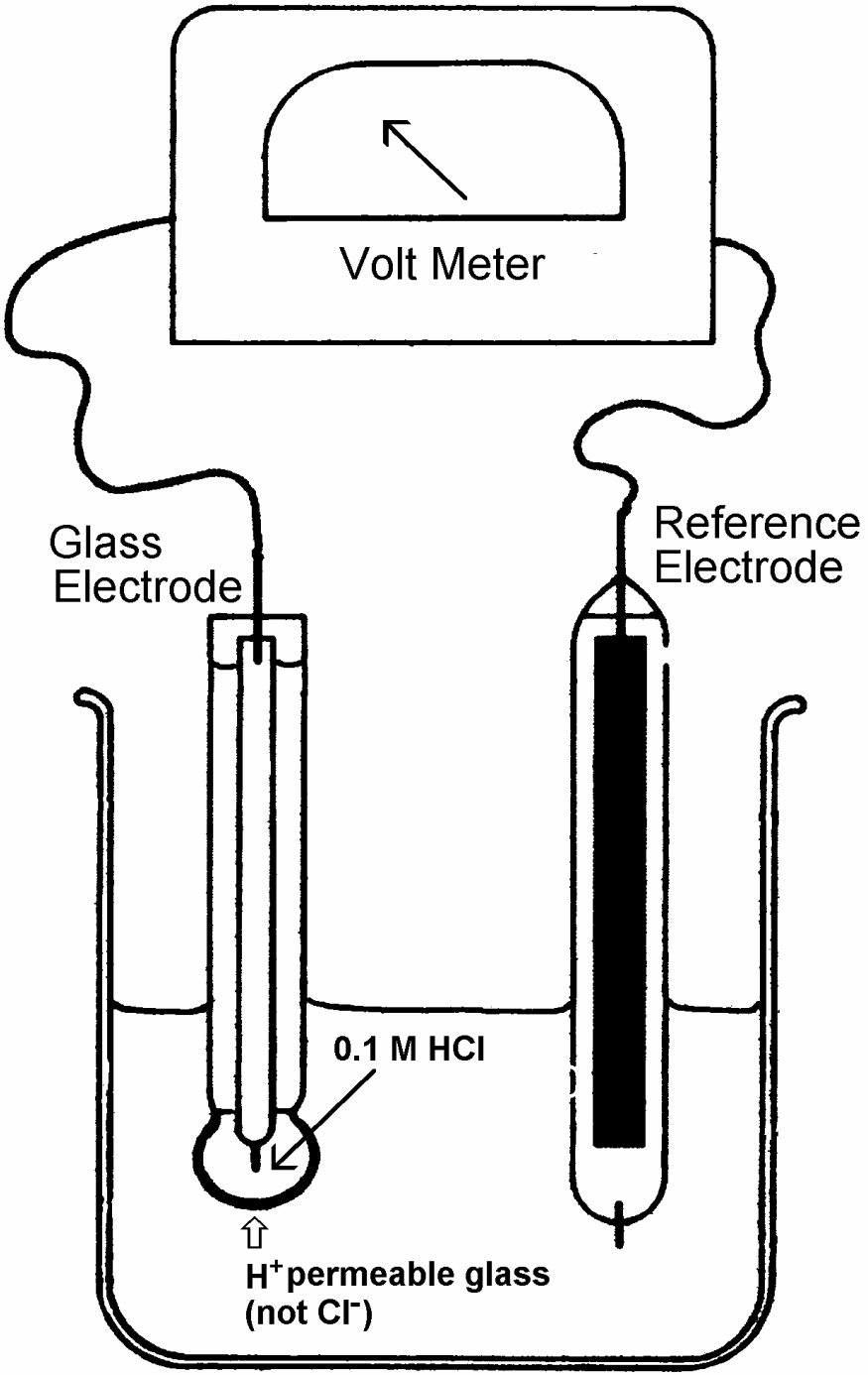
Henderson-Hasselbach

$$\mathbf{pH = pK_a + \log[A^-]/[HA]}$$

A^- = conjugate base (or salt), base

HA = acid, conjugate acid

pK_a = -log of dissociation constant



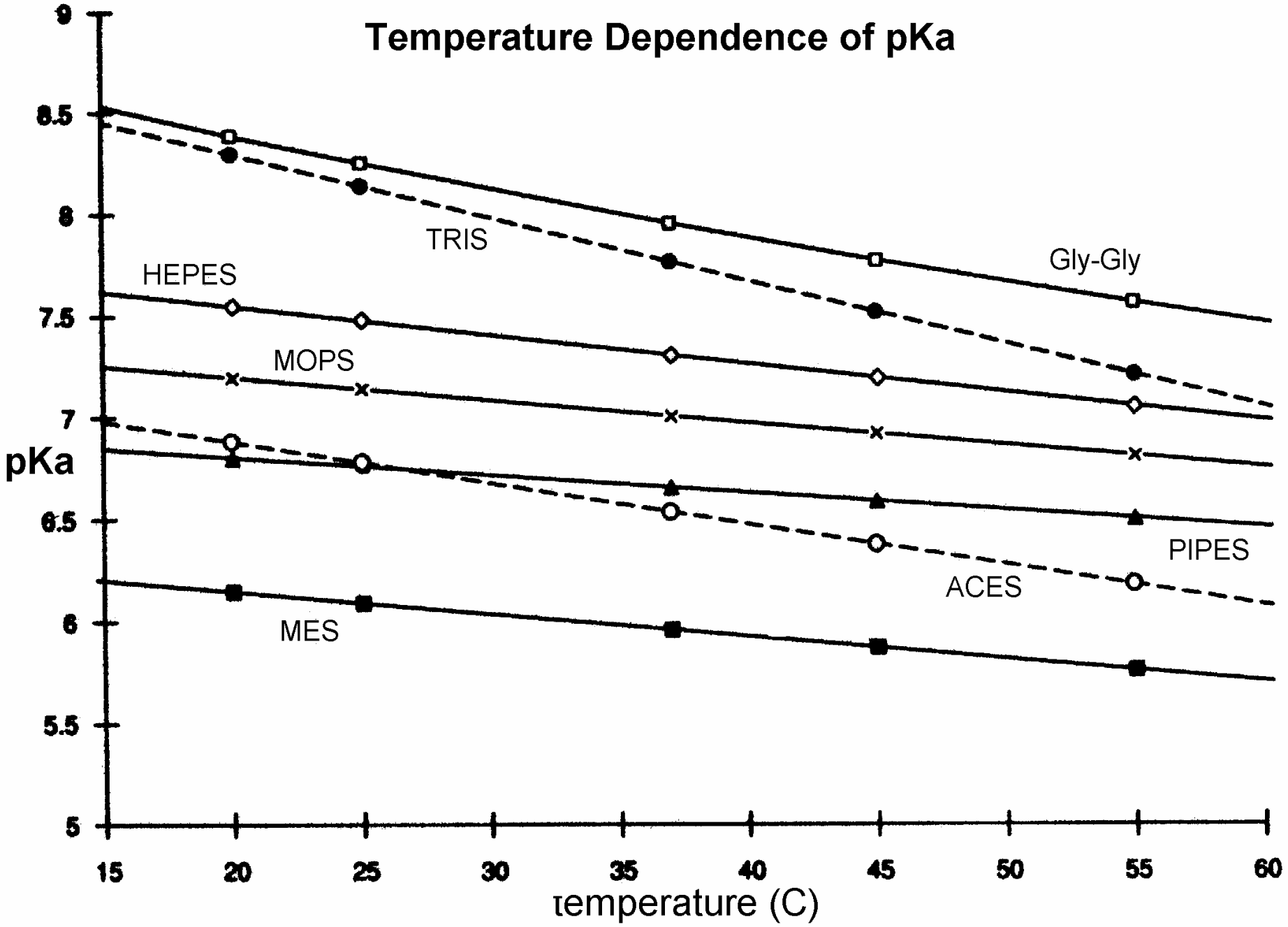
pH Measurement

$$E = E^o + (2.3RT/F) \cdot pH$$

E = electrical potential (or voltage)

Factors Affecting:

- ionic concentration
- Na^+ error
- temperature


Temperature Dependence of pKa
9
8.5
8
7.5
7
6.5
6
5.5
5
pKa
15
20
25
30
35
40
45
50
55
60
Temperature (C)
TRIS
Gly-Gly
HEPES
MOPS
PIPES
ACES
MES

Choosing a Buffer

- **literature or empirical**
- **$\pm$ 1 pH unit from pK_a**
- **buffering capacity ($\beta = \Delta B/\Delta pH$)**
- **compensate for known changes in H^+**
- **compatibility with experimental system**

Good's Buffer Criteria

- pKa between 6 and 8
- high solubility in aqueous solutions
- excluded by biological membranes
- minimal effects due to conc., temp. and ionic composition
- well-defined or non-existent interactions with cations
- chemical stability
- insignificant absorption between 240 and 700 nm
- readily available in pure form

Zwitterionic Buffers (aka Good's Buffers)

Name	pK_a	range
MES	6.15	5.8- 6.5
ADA	6.62	6.2- 7.2
PIPES	6.80	6.4- 7.2
MOPS	7.20	6.5- 7.9
HEPES	7.55	7.0- 8.0
EPPS	8.00	7.6- 8.6
Tricine	8.15	7.6- 8.8
Bicine	8.35	7.8- 8.0
CHES	9.55	9.0-10.1
CAPS	10.4	9.7-11.1