Alcohol Pharmacology and Analysis Methods

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Ethanol

- Ethanol (Ethyl Alcohol, C_2H_5OH) is derived from fermentation of sugars in fruits, cereals, and vegetables.
- Commercial beer, wine and liquors contain various amounts of ethanol.
- Ethanol is also found in variety of perfumes, after-shaves, mouthwashes, some rubbing alcohols, pharmaceutical preparations (elixirs) and may other products.
- Ethanol used as a solvent and antiseptic in industry and medicine.


SPRENGEL
16 feine Trüffel umhüllt von feiner Schokolade


PASSPORT SCOTCH
HAIG & HAIG
CATTOS
PRIME


PASHA
VODKA
CHIVAS REGAL
12

What is called Alcoholic Beverage?

- An **alcoholic beverage** is a drink which contains a substantial amount of the ethanol (0.5-95% V/V).
- Also, they classified as fermented (beers and wines) and distilled (liqours) alcoholic beverages.
- Alcoholic beverages typically contain between 3% and 40% alcohol by volume.

Alcoholic beverages classification

- *1- Fermented beverages*

  Wine, champagne, sherry

  Beer

  Cider

  Mead

- 2- *Distilled beverages*

  Whisky (40-55%)

  Vodka (60p-90%)

  Rum (90-95%)

  Brandy (40-50%)

Ethanolic Products

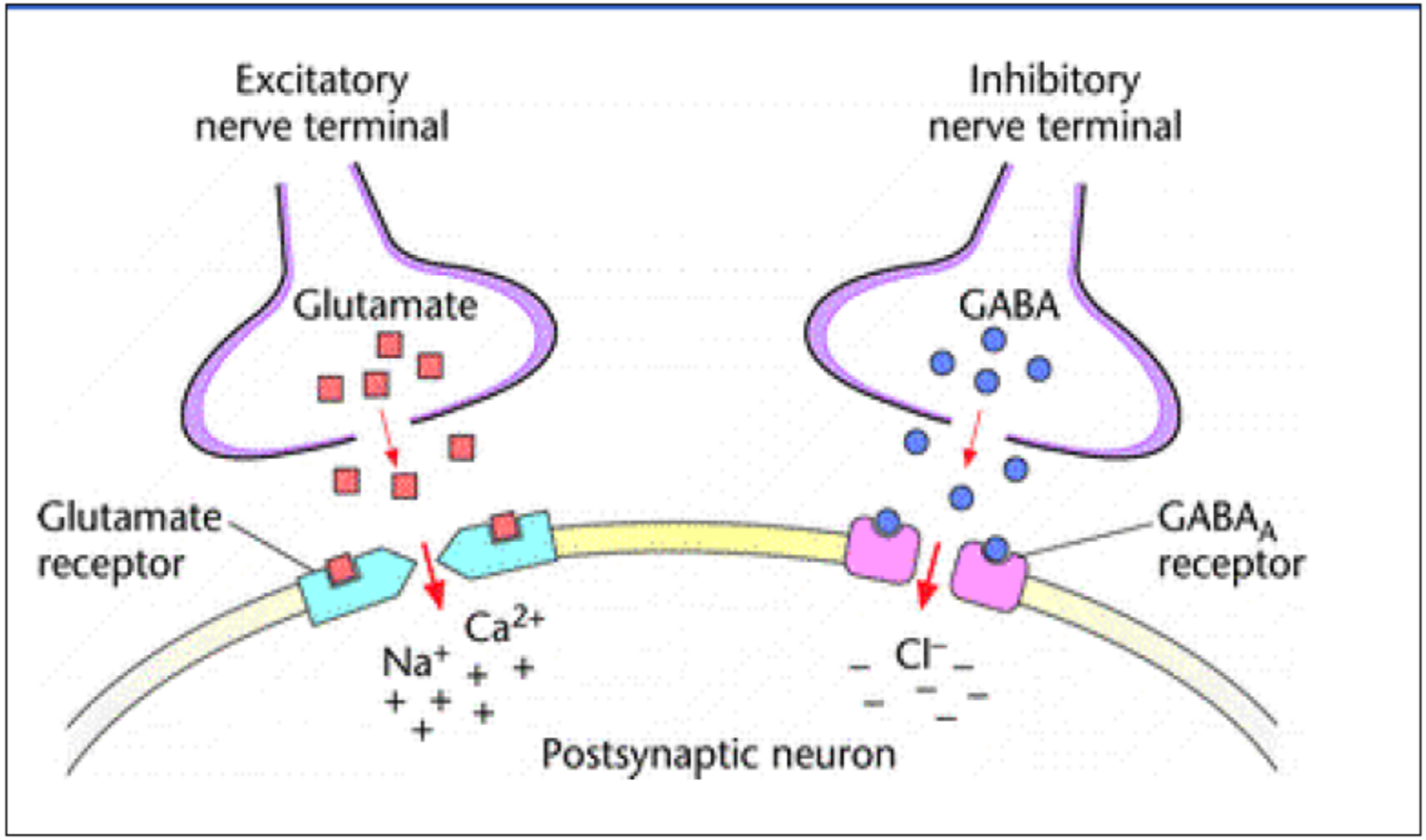
Beer 3-6%	Antiseptic 10-70%
Wine 16%	Perfume 40-80%
Whisky 40%	Aftershave 40-80%
Vodka 60-90%	Mouthwash 15-25%

Ethanol Pharmacodynamics

Pharmacodynamics

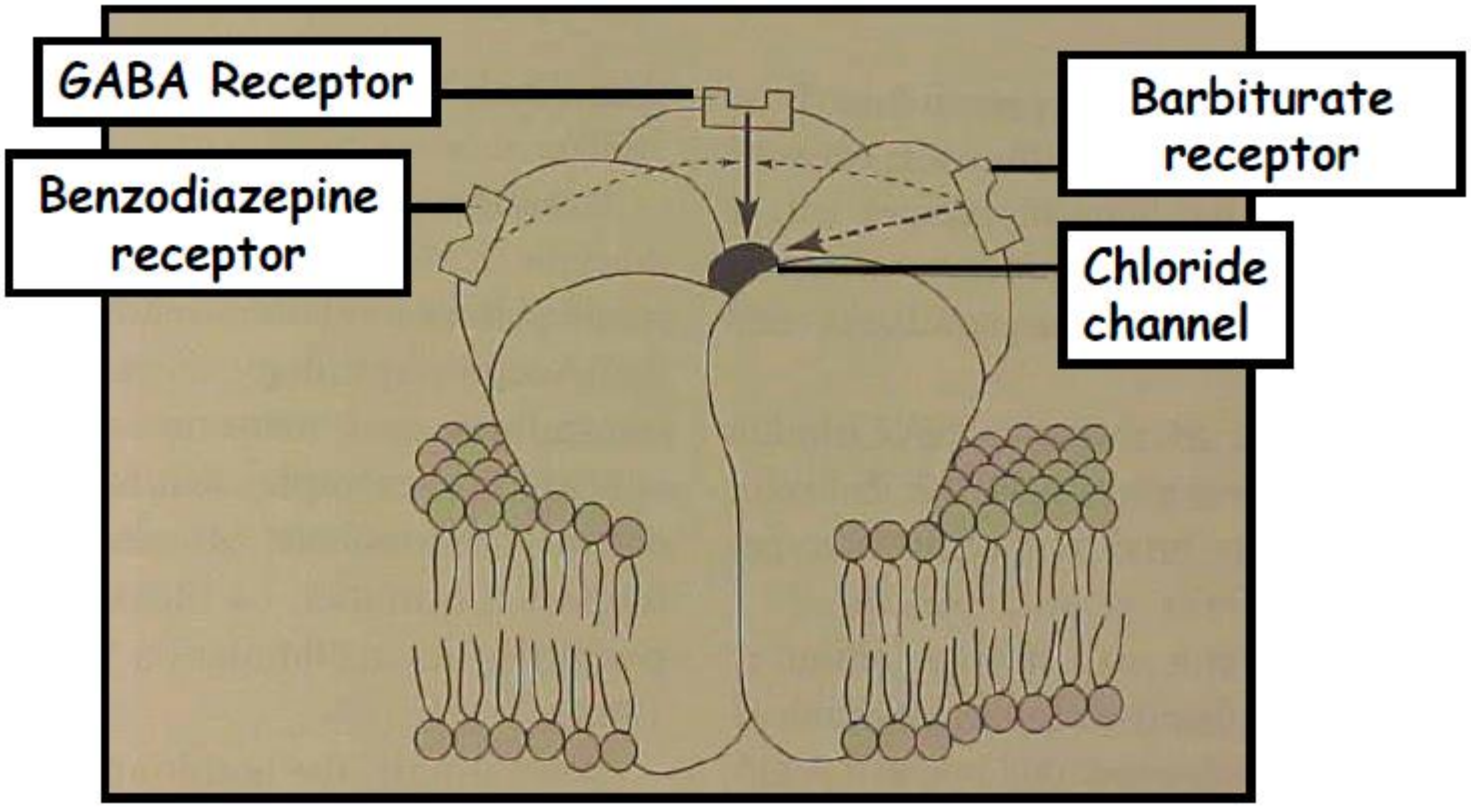
Studies of biochemical and physiological effects of drugs on living organisms including mechanisms of action, dose response relationships, and drug-effects on behavior in relation to chemical structure and dosage form.

Receptors involved in some of the actions of ethanol



Effects on ligand-gated ion channels

$GABA_A$ inhibitory receptor EtOH potentiates the effect of GABA

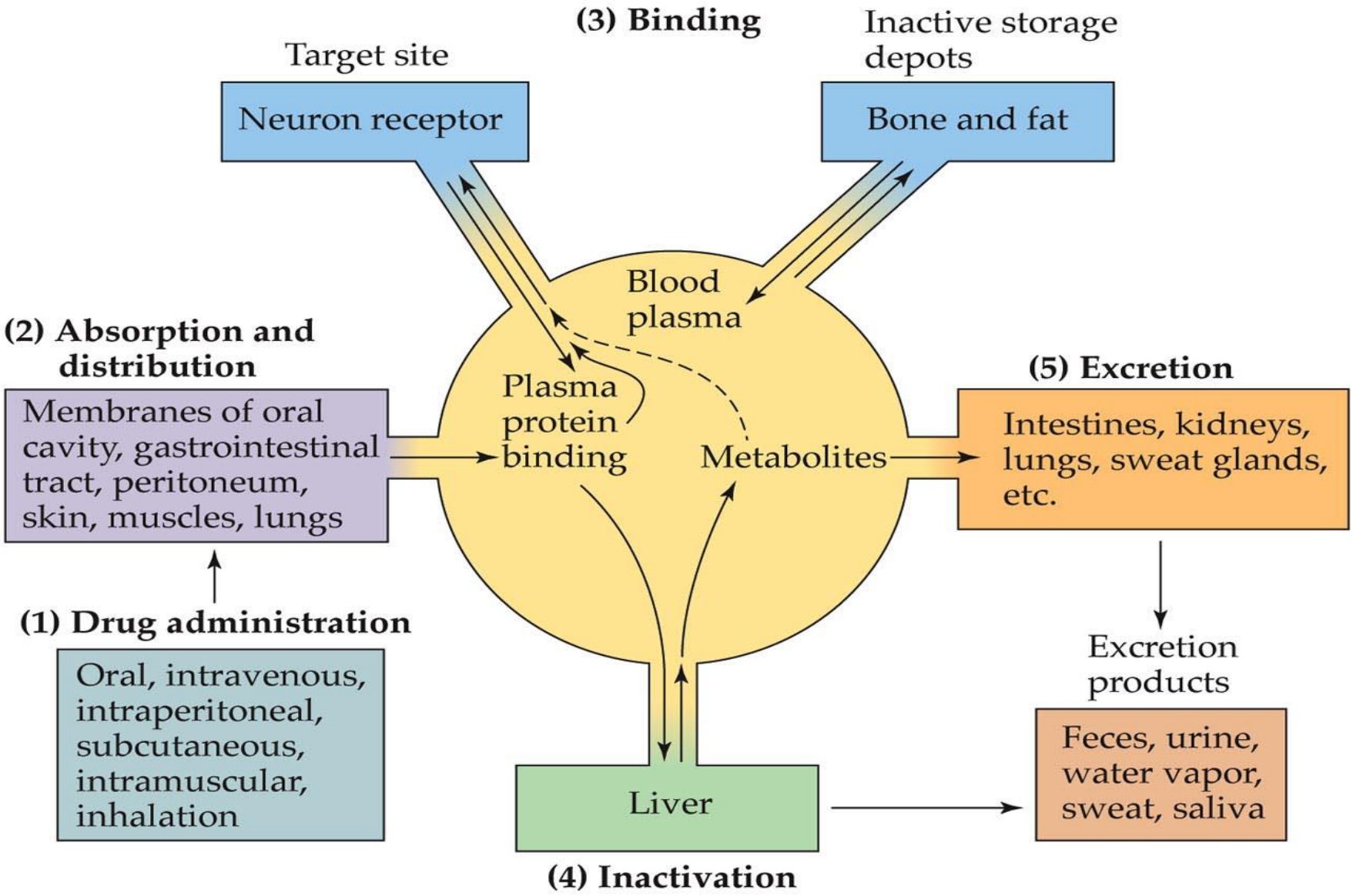


Signs & symptoms of intoxication and BAC

<25 mg%	*sense of warmth, wellbeing, talkativeness, self confidence*
25-50 mg%	*Euphoria, decreased judgment and control*
50-100 mg%	*Ataxia, decreased reflexes/increased reaction time*
100-250 mg%	*Ataxia, diplopia, slurred speech, nystagmus*
250-400 mg%	*Stupor, coma, nausea, vomiting*
>400 mg%	*Respiratory paralysis, hypothermia, death*

Ethanol Pharmacokinetics

Pharmacokinetics theory



Ethanol absorption

- The alcohol molecule is a small polar molecule with both lipophilic and hydrophilic characteristics.
- The amphipathic qualities of alcohol help to explain its pharmacokinetics within the body.

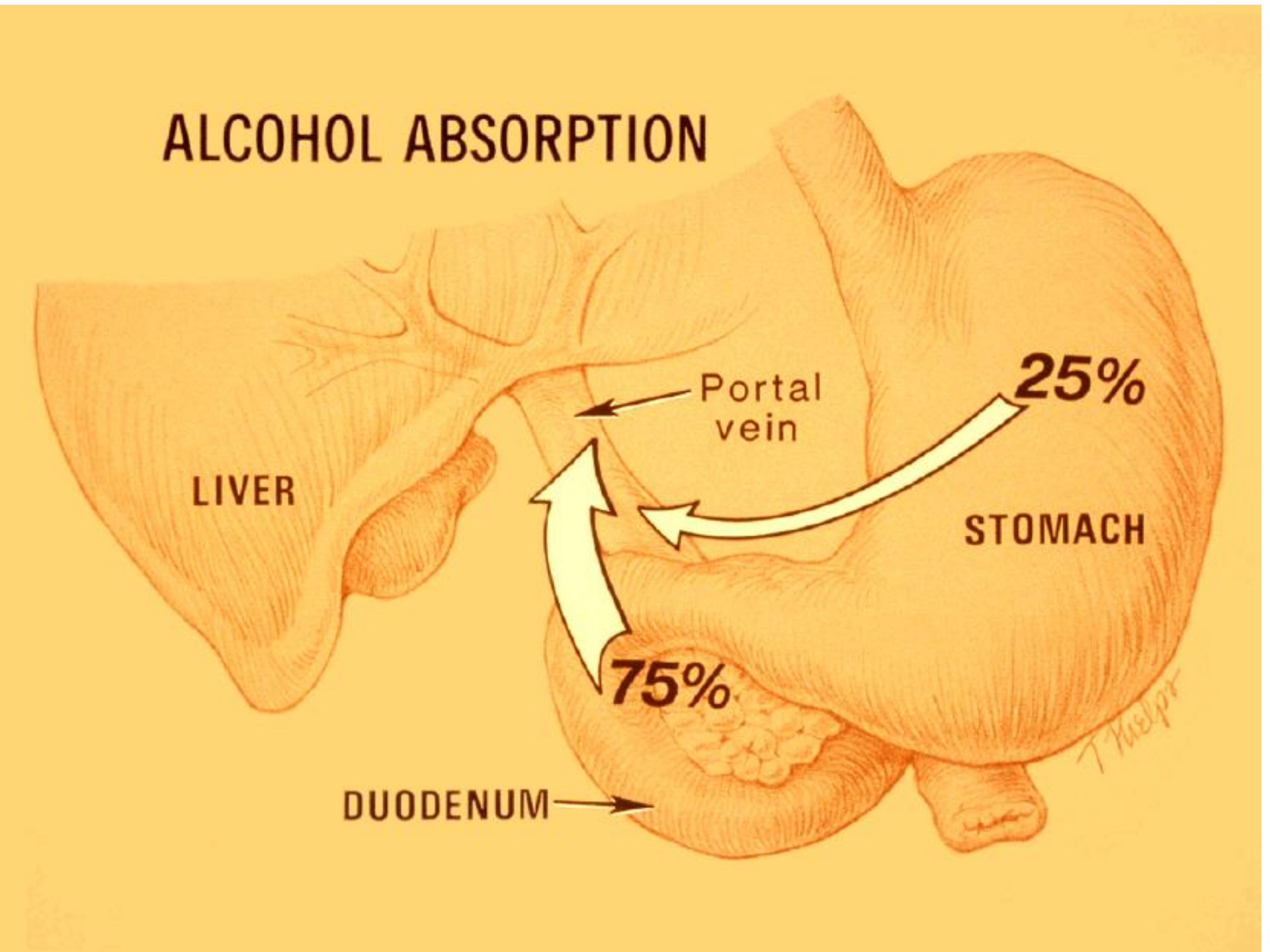
- The hydrophilic combined with the polar properties of the alcohol molecule explain how alcohol is completely soluble in water and thus has a similar volume of distribution (Vd) to total body water (TBW).

As alcohol is a small water soluble molecule that can cross cell membranes, it is absorbed from both the stomach (20 %) and the upper small intestine (80 %).

The time from the last drink to maximal concentration in blood usually ranges from 30 to 90 minutes.

Factors affecting:

Concentration of ethanol, Fasting, Delaying the stomach emptying, Co ingestion of some drugs.


ALCOHOL ABSORPTION
Portal vein
25%
LIVER
STOMACH
75%
DUODENUM

BAC is dependent to many factors

- Blood alcohol concentration (BAC) is determined by the various factors that affect the rate at which alcohol is absorbed, distributed, metabolised and excreted from the body.
- Following oral administration absorption and distribution determines the proportion and rate at which orally ingested alcohol reaches the blood and body tissues (bioavailability).

Factors affecting Blood Alcohol Concentration

Variable	Primary reason
Gender	Differences in TBW and gastric ADH
Ethnicity Different	Sensitivities to alcohol
Type of alcohol	Amount & strength can affect absorption
Mixer	Can affect absorption
Stomach content	Timing & meal type (e.g. fat content) affect absorption

Intra-individual and Inter-individual variations

- The rate of absorption varies significantly in both intra-individual and inter-individual comparisons even after standardized conditions.
- Intra-individual variability is due to variation in gastrointestinal function (gastric emptying, intestinal transit time, and portal blood flow).

Effect of gastric emptying

- The rate of **gastric emptying** has a significant impact on the speed at which alcohol is absorbed, because alcohol is absorbed much faster from the small intestine, than it is from the stomach.

Effect of gastric emptying

- Factors which affect alcohol availability and gastric emptying will greatly influence the rate of absorption.
- For example, the consumption of alcohol with food inhibits absorption because approximately 20% of the ingested alcohol is oxidised before it can be absorbed.

Type of drink

- The type of drink consumed also plays a role.
- Drinks with alcohol content between 20-30% are absorbed quickest.
- Whereas drinks with a higher alcohol content are absorbed more slowly, because an alcohol content over 30% **irritates the gastric mucosa** increasing mucus secretion and decreasing gastric emptying.

Effect of mixer

- Thus drinks with an alcohol content above 30% can cause a faster rise in BAC if served diluted with a mixer, than if they are served without dilution.
- This is especially true if the mixer is a carbonated drink as this can also increase the rate of absorption.

Ethanol fatal dose

The fatal dose of ethanol is 300-400 mL of pure ethanol (600-800 mL of 50% spirits), for the average adult if consumed ***in less than one hour.***

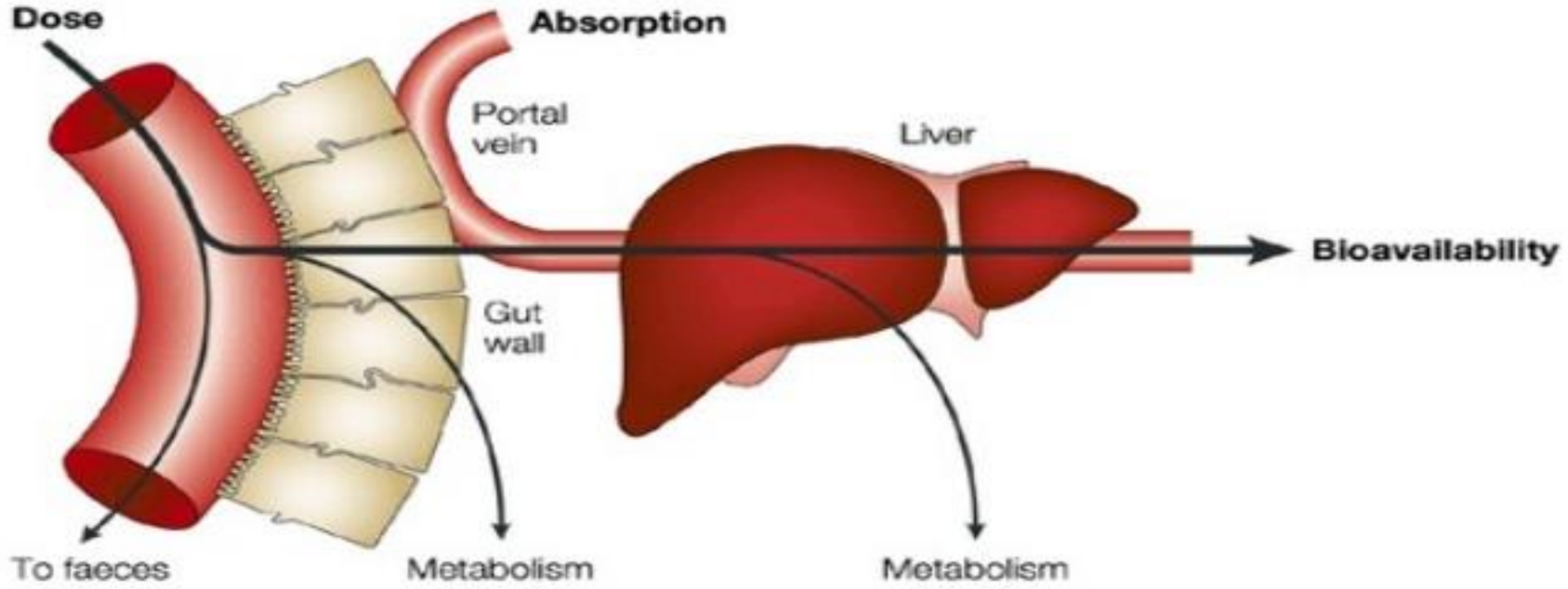
Estimating blood alcohol concentration

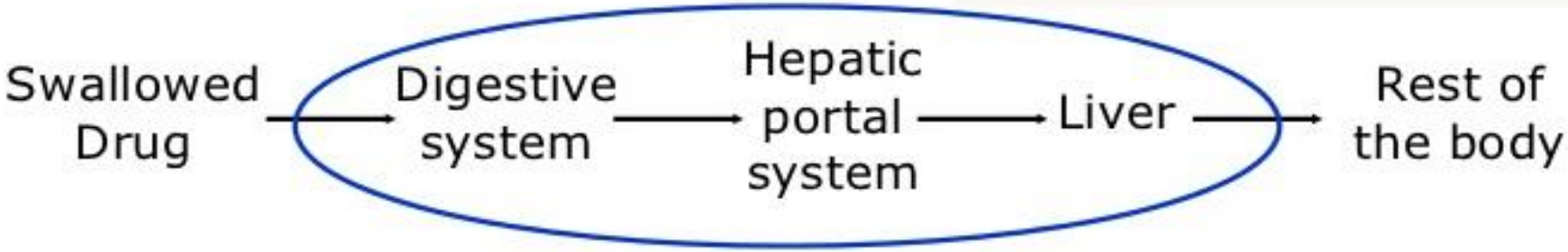
$$\text{EBAC} = \frac{\text{Et (mL)} \times \text{concentration (\%)} \times 800}{\text{Vd (L/Kg)} \times \text{Weight (Kg)} \times 10}$$

First pass metabolism

- The bioavailability of alcohol is reduced by first pass metabolism (FPM).
- Oxidation of alcohol by gastric alcohol dehydrogenase (ADH) in the gastric mucosa accounts for a small proportion of FPM, but the majority occurs via oxidation by ADH in the liver hepatocytes.

First Pass Metabolism





Distribution

- The proportion of alcohol that is absorbed, and escapes FPM enters the systemic circulation and is rapidly distributed throughout the body tissues via the blood plasma until an equilibrium between the BAC and tissue concentration is reached.
- The time until equilibrium is dependant upon the permeability (**water content**) and **rate of blood flow** but is generally achieved within 1-2 hours.

The concentration of alcohol in blood and tissue depends on the amount of total body water, since alcohol is soluble in water.

$$V_d = 0.6\ L/Kg$$

Gender differences

- Gender differences in responses to ethanol are well recognized.
- Women are more sensitive to alcohol, and exhibit higher mortality at lower levels of consumption than men.

Gender differences

- Women exhibit somewhat higher blood levels than men following ingestion of equivalent doses of ethanol.

- This phenomenon appears to be due in part to more extensive ADH-catalyzed metabolism of ethanol by the male gastric mucosa. This would increase the bioavailability of alcohol resulting in increased BAC.

- A second factor contributing to the higher blood ethanol levels and greater CNS effects in women is their smaller volume of distribution (higher body fat) for relatively polar solvents such as alcohols.

Ethanol metabolism

Different metabolic pathways for ethanol

liver is the major organ to metabolize and eliminate alcohol.

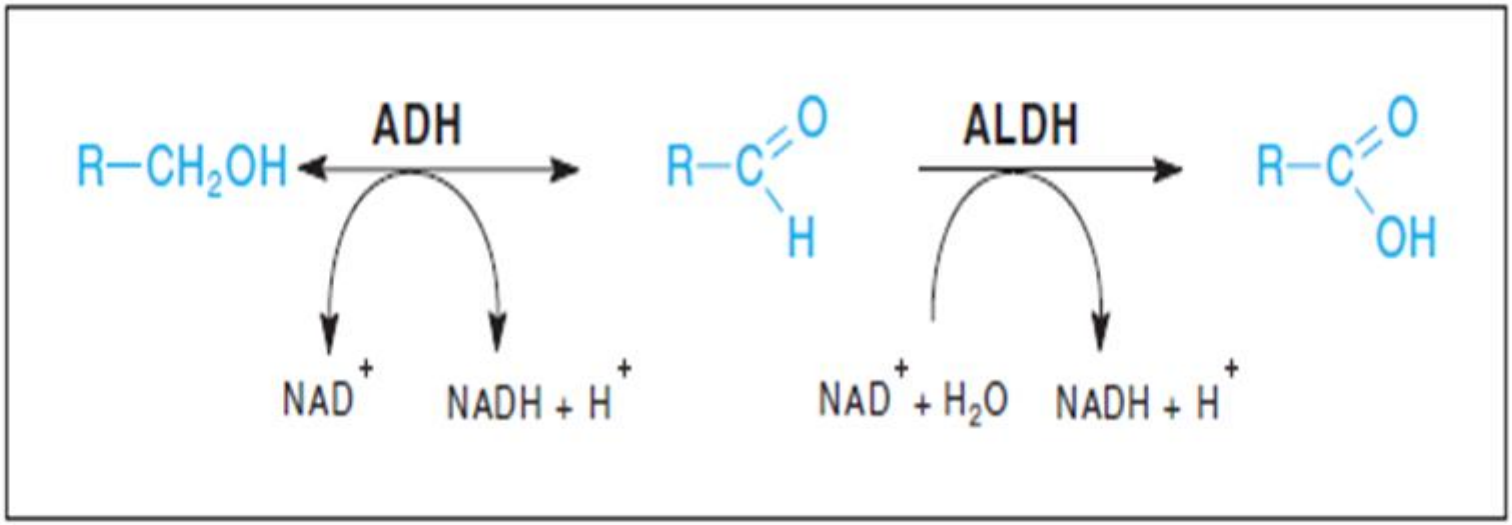
1- Hydrogen peroxidase

2- MEOS (inducible)

3- Alcohol dehydrogenase

- A small proportion (2-5%) of the alcohol absorbed is excreted unchanged in the urine, sweat or breath but the majority (~ 90 %) is removed via oxidation by ADH.
- This can occur in various organs such as the stomach and small intestine but is primarily carried out by **hepatic ADH.**

ALCOHOL METABOLISM



Oxidation of alcohols to aldehydes and carboxylic acids by alcohol dehydrogenase (ADH) and aldehyde dehydrogenase (ALDH).

Zero-order kinetics

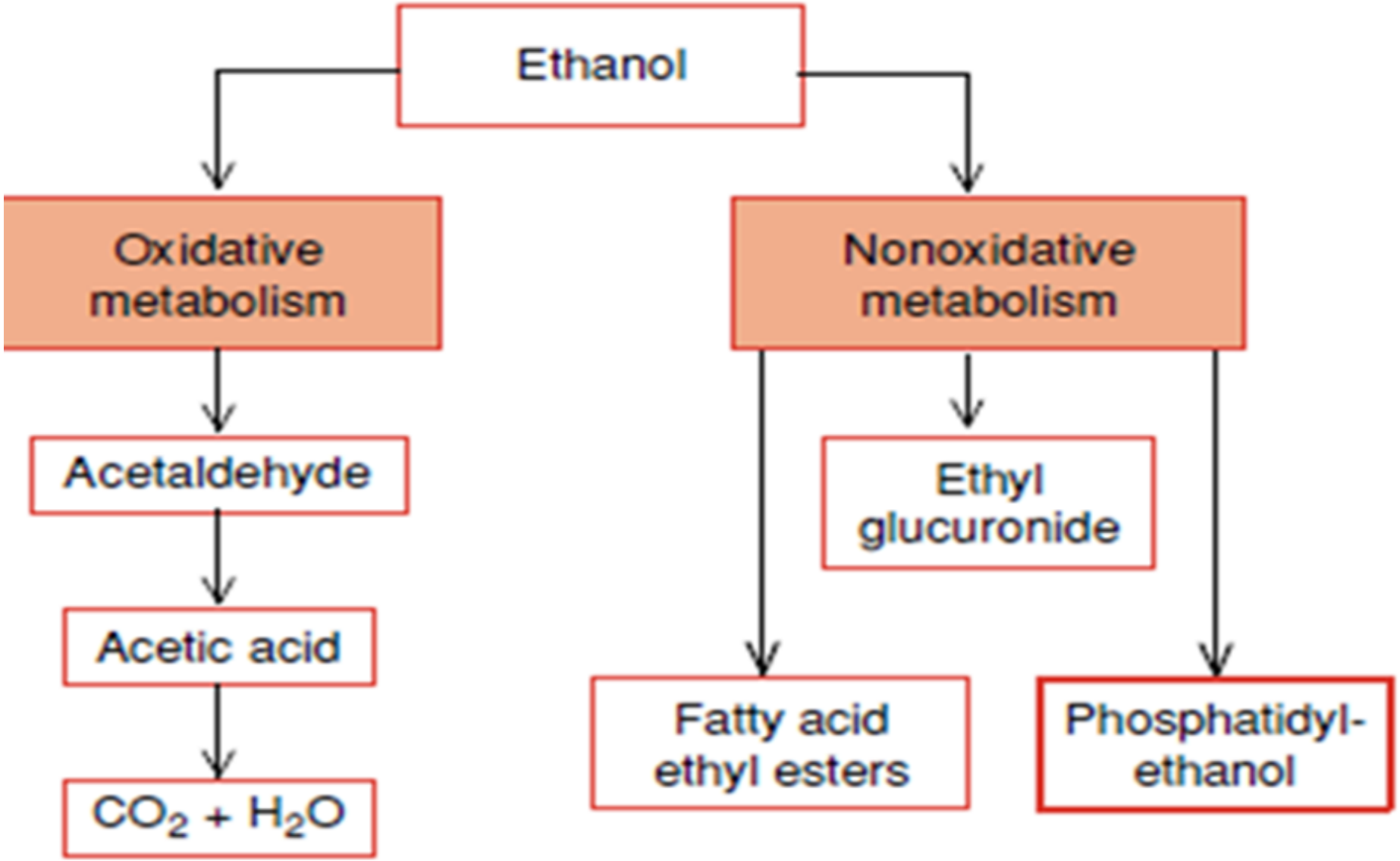
- The process that takes place at a constant rate independent of drug concentration involved in the process.

Zero-order ethanol metabolism

- The rate of ethanol metabolism is linear.
- Chemists refer to this as a Zero Order Reaction.
- Because the primary decay product of alcohol metabolism-**acetaldehyde**-is poisonous.
- The body must eliminate the acetaldehyde produced by the breakdown of alcohol before any more alcohol can be processed in order to avoid acetaldehyde poisoning.
- This slows down the rate of alcohol metabolism to a Zero Order Reaction rather than a First Order Reaction.

Microsomal ethanol oxidizing system

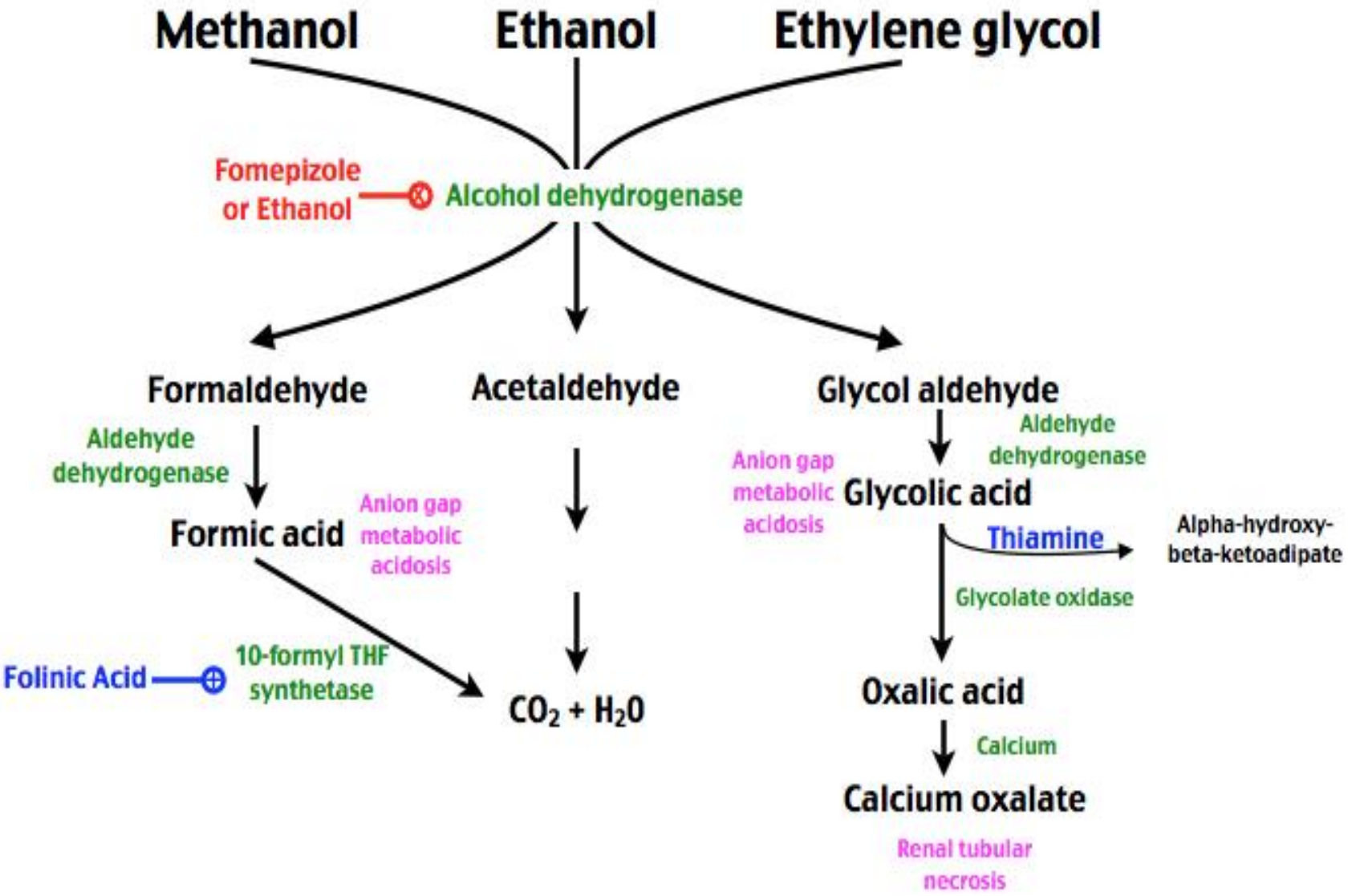
- A secondary oxidation pathway for alcohol metabolism is the microsomal alcohol oxidizing system (involving microsomal cytochrome P450 (CYP) 2E1), which due to its low affinity for alcohol (about 10 fold lower than ADH) only accounts for ~10 % of total alcohol clearance by the liver at low concentrations.


Ethanol
Oxidative metabolism
Nonoxidative metabolism
Acetaldehyde
Ethyl glucuronide
Acetic acid
Fatty acid ethyl esters
Phosphatidyl-ethanol
$CO_2 + H_2O$

Use of metabolic pathway in treatment

- Ethanol interacts with other solvents that are also metabolized by ADH and CYP2E1.
- Ethanol can be an effective **antidote** for poisoning by methanol, ethylene glycol, and diethylene glycol.
- As ethyl alcohol has a relatively high affinity for ADH, it competitively inhibits the metabolic activation of other alcohols and glycols.

Toxic Alcohol Metabolism

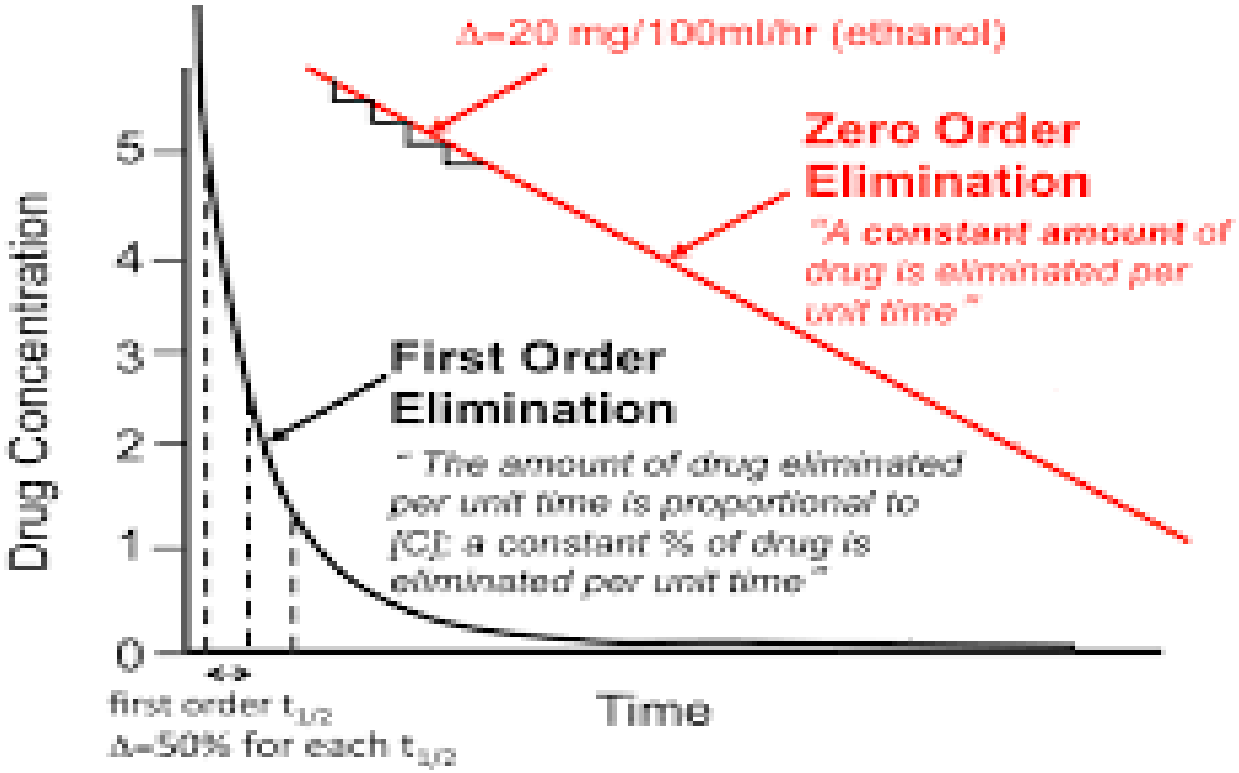


Excretion

Zero order elimination process

- Alcohol elimination was originally believed to be a zero-order process, meaning that alcohol was removed from the body **at a constant rate**, **independent of the concentration of alcohol**.
- ADH is saturated at low concentrations of alcohol, hence, the overall elimination process proceeds at maximal velocity and is independent of the alcohol concentration.

Zero and first order kinetics



Pharmacokinetic

- Excretion

  some unchanged in breath and urine

  In nonalcoholic adults: 15-24 mg%/h

  In social drinkers: 15 mg%/h

  In alcoholic adults: 15-49 mg%/h

  In children: 20-30 mg%/h

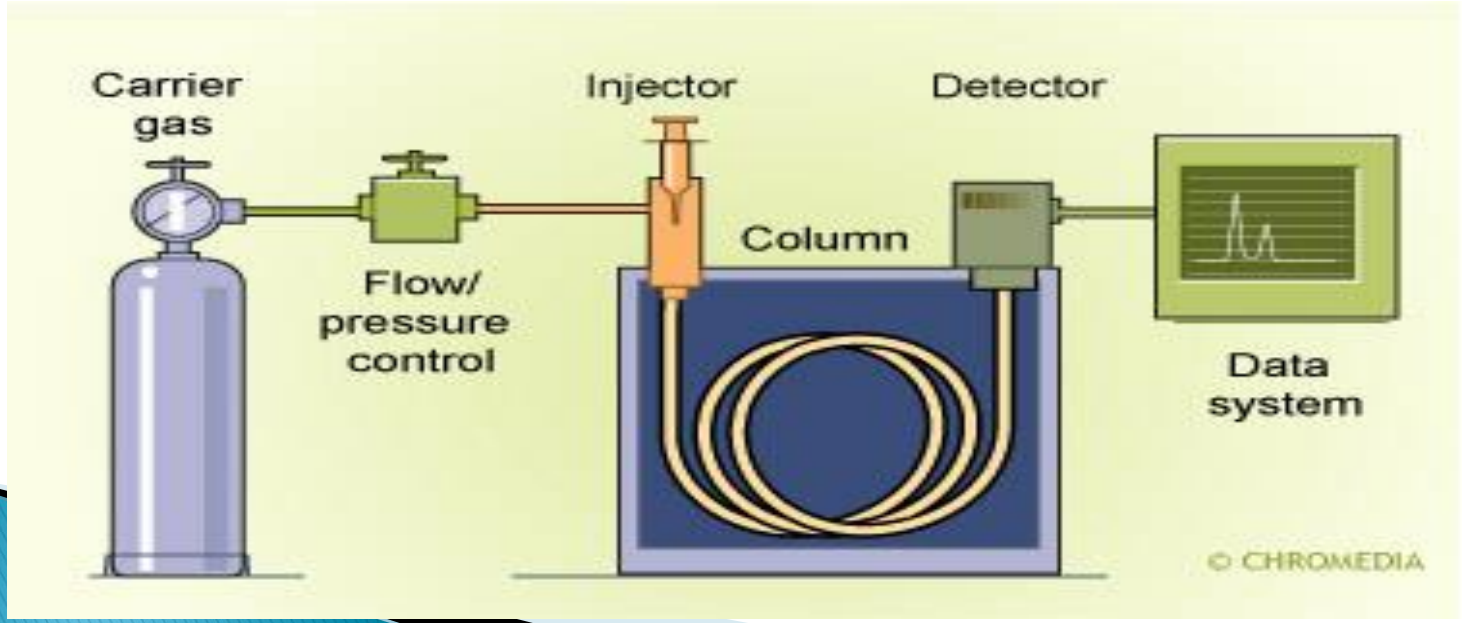
Analytical methods for forensic alcohol analysis

Assay Methodologies

- Gas chromatography
- Enzymatic oxidation
- Chemical reaction
- Breath alcohol analysis

Gas chromatography (GC)

- **Gas chromatography (GC)**, is a common type of chromatography used in analytical chemistry for separating and analyzing compounds that can be vaporized without decomposition.



Gas Chromatography

- **Advantages:**
- Specificity for ethanol, methanol and other types of alcohol **identification** and **quantitation**.
- Enhanced with the use of multiple columns or varying chromatographic conditions.

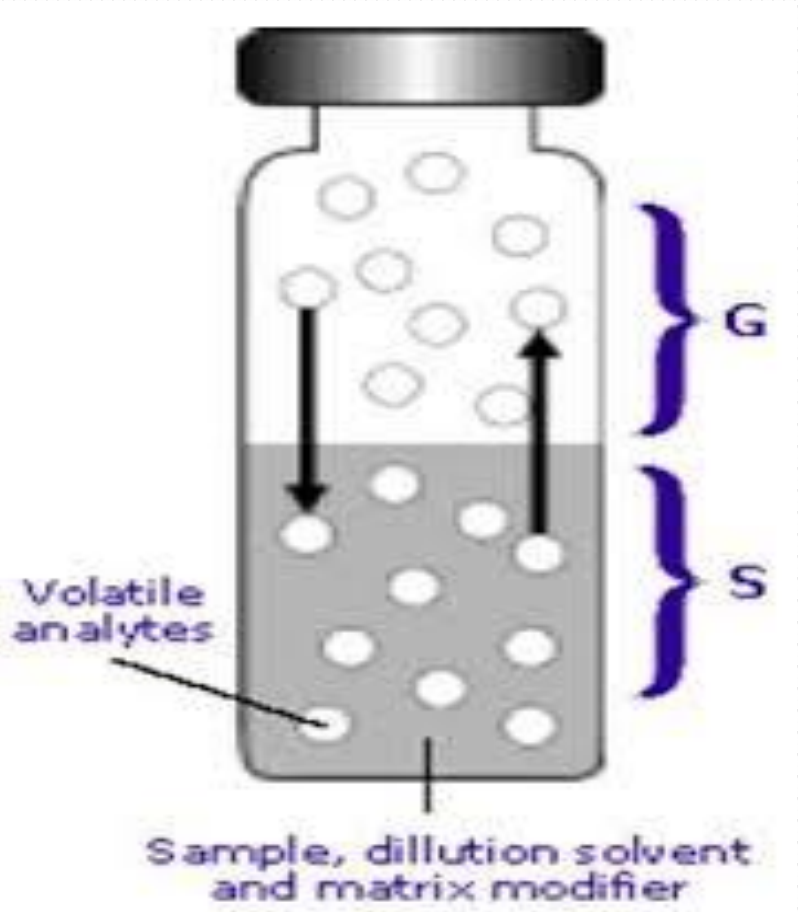
Gas Chromatography

- **Disadvantages:**
- Requires specialized instrumentation (gas chromatograph)
- Requires highly trained technical staff
- Analysis slower than enzymatic assay

A gas chromatograph with a headspace sampler

Headspace Gas Chromatography definition

- "Headspace" is the gas space above the sample in a chromatography vial.
- Volatile sample components diffuse into the gas phase, forming the headspace gas.
- Headspace analysis is therefore the analysis of the components present in that gas.



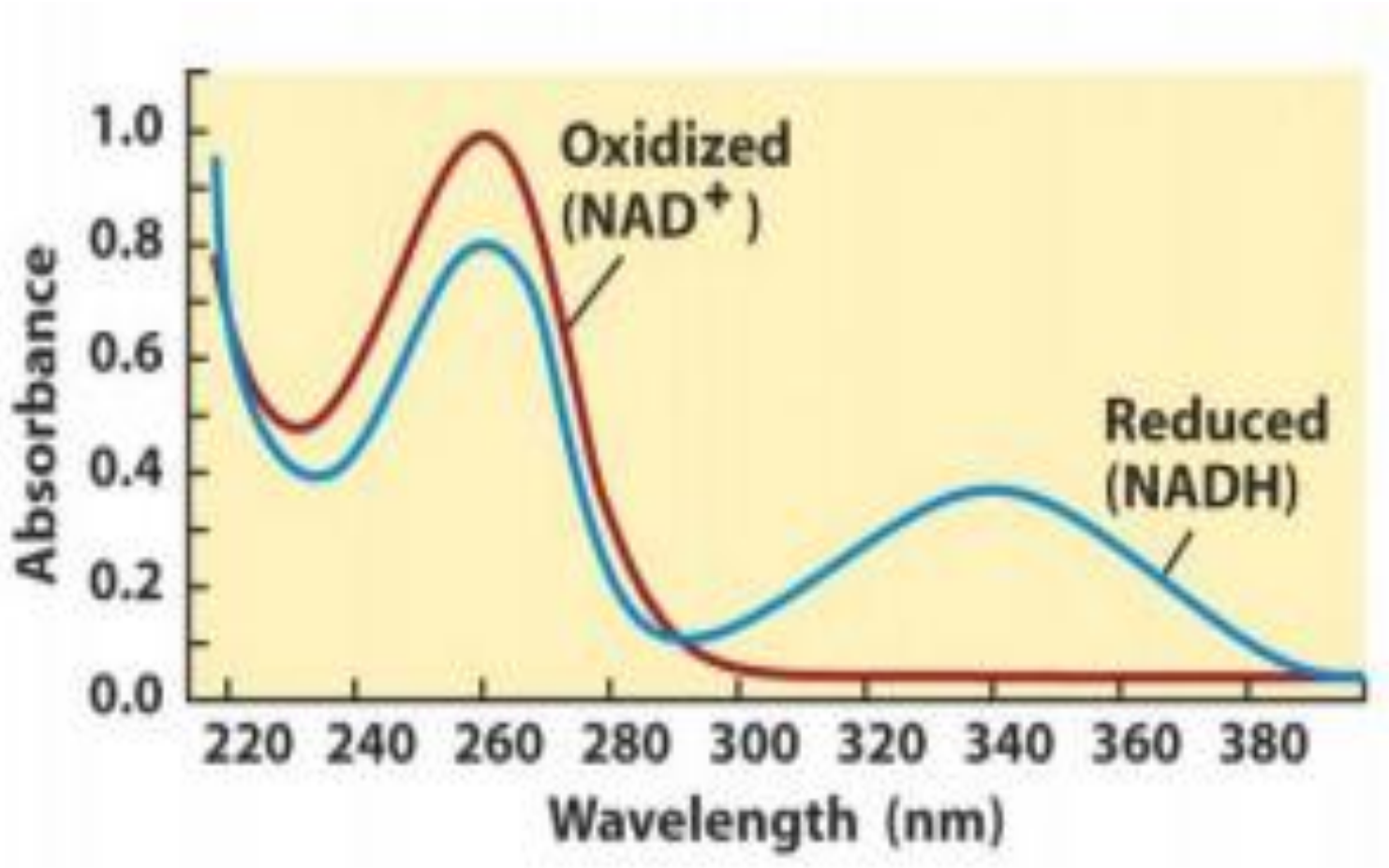
Enzymatic Oxidation Assay

- Most of the commercial kits use alcohol dehydrogenase (ADH):

- $C_2H_5OH + NAD^+ \xrightarrow{ADH} CH_3CHO + NADH + H^+$

- The reaction is monitored following the absorbance of NADH at 340 nm or that of a color product at a higher (visible) wavelength formed by reacting NADH with a dye.

Absorbance of NADH at 340 nm



Enzymatic Oxidation Assay

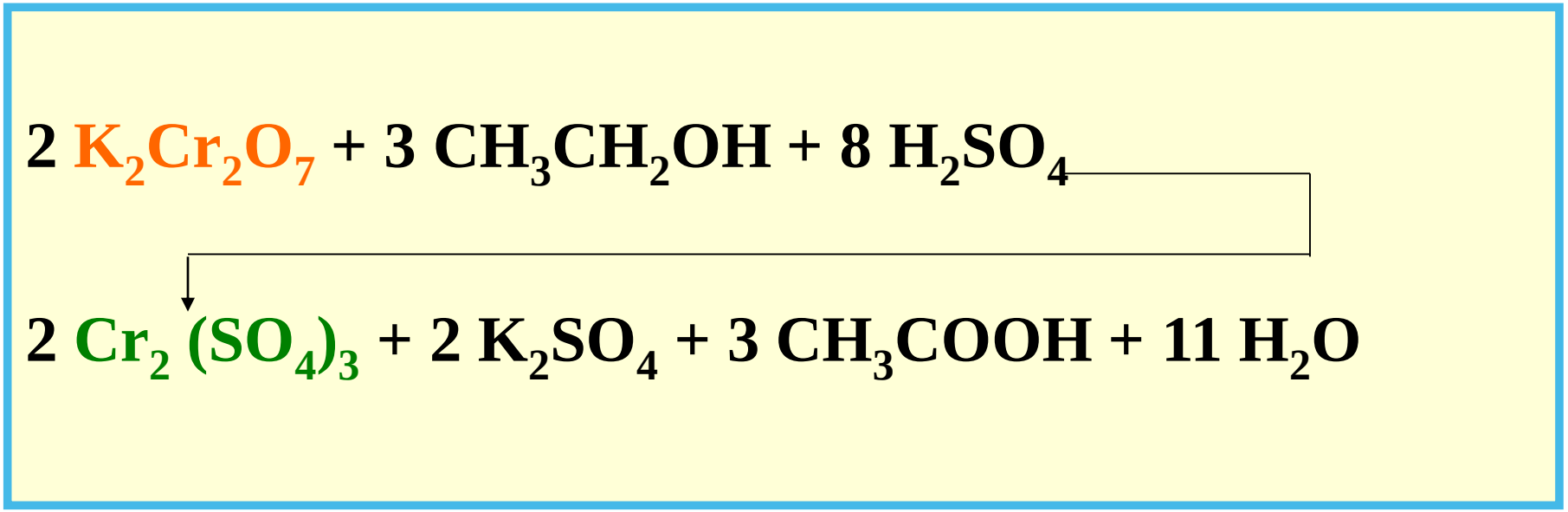
- **Advantages**:
- Rapid, easy to use kits are widely available
- This allows the smallest of clinical laboratories to perform stat quantitative alcohol test
- **Disadvantages**:
- Not specific for ethanol. Other alcohols can interfere at high concentrations
- Will miss methanol and isopropanol overdose

Chemical Reaction (Widmark method)

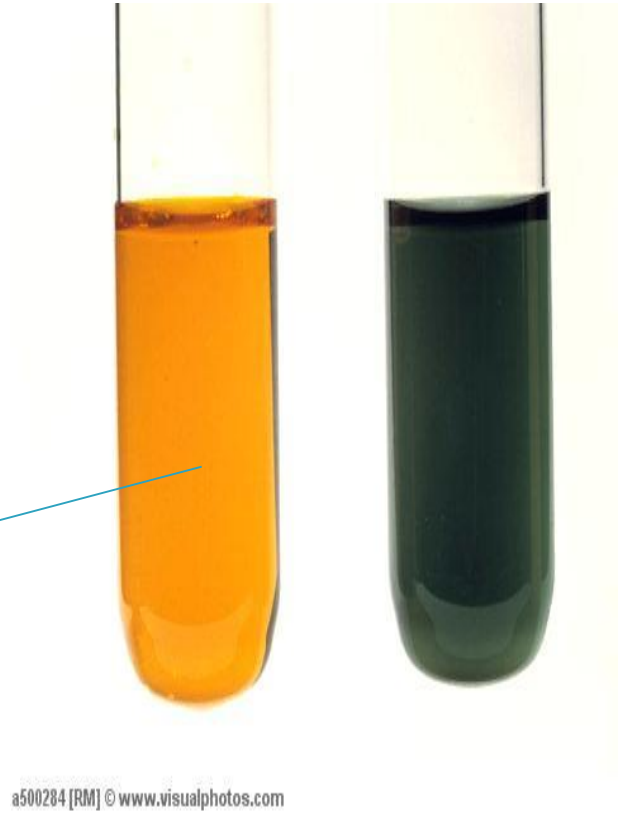
- this is a method for quantifying alcohol based on the oxidation of potassium dichromate in the presence of sulphuric acid, followed by a titrimetric analysis.

- It is non-specific, as alcohols other than ethanol (eg. methanol) and related compounds such as acetone and ether can all be involved in the oxidation reaction.

Chemical Reaction (Widmark method)

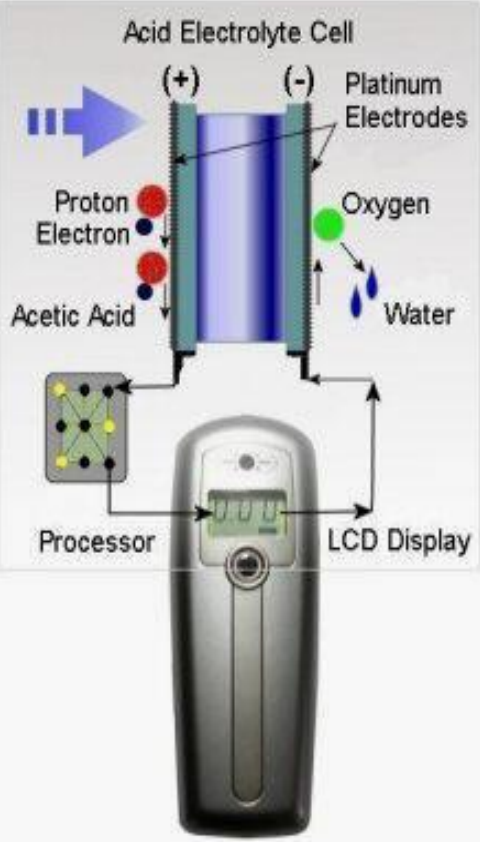


Potassium dichromate
conversion to Chromium sulfate



Breath alcohol analyser

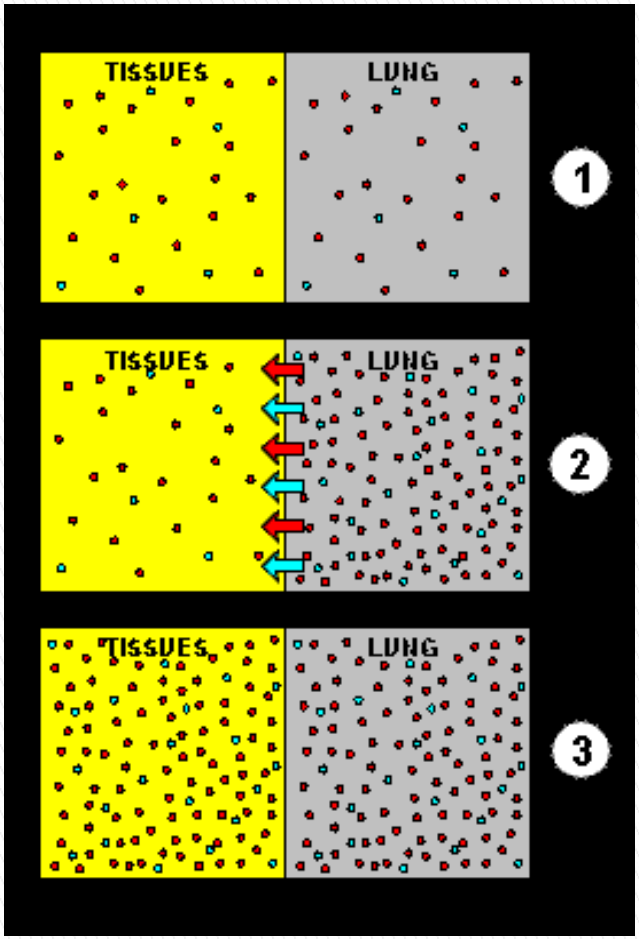
Fuel Cell Detectors



- Apparatus consists of two platinum electrodes with acidic electrolyte material between them
- Ethanol in breath **oxidized** at surface of **anode** to give acetic acid, protons, and electrons
- Atmospheric oxygen **reduced** at **cathode** to give two oxygen atoms
- Protons and electrons from anode travel to the cathode and combine with oxygen to form **water**
- Movement of electrons produces a **current** that is proportional to the amount of alcohol in the breath sample
- Microprocessor measures the current and calculates **BAC**

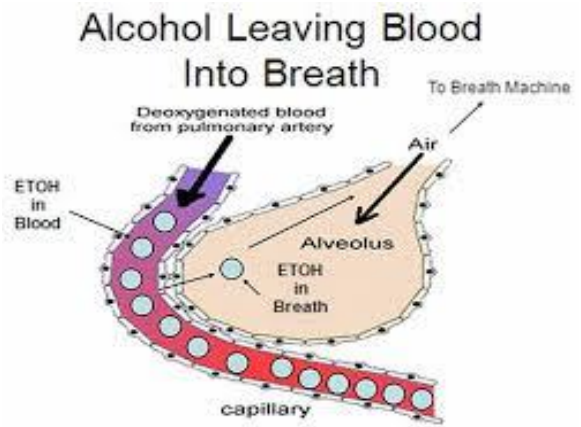
Henry's law and breath alcohol test

- Solubility of gas in a liquid is proportional to the partial pressure of gas over liquid in a *closed system* under *constant temperature*.



Basic Principle of Breath Alcohol Testing

- Following oral consumption, alcohol is absorbed from the gastro-intestinal tract and distributed throughout the body by the circulatory system.
- Alcohol diffuses freely and is found in relative concentrations according to the water content of the various tissues.
- Alcohol conc. in end-expiratory breath (BrAC) is proportional to alcohol conc. in the blood (BAC) suffusing the alveolar bed.



Breath Alcohol Concentration (BrAC) Measurement

- Advantages:

•Breath collection is *noninvasive*

•Collection does not require phlebotomist; can be performed by many more people

•Instrument designed for portability and *easy* breath collection; onsite testing

•Collection and test can be done *simultaneously* with immediate result