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Psychobiology Research Group

Trials and pharmaco-kinetics, dynamics and genetics

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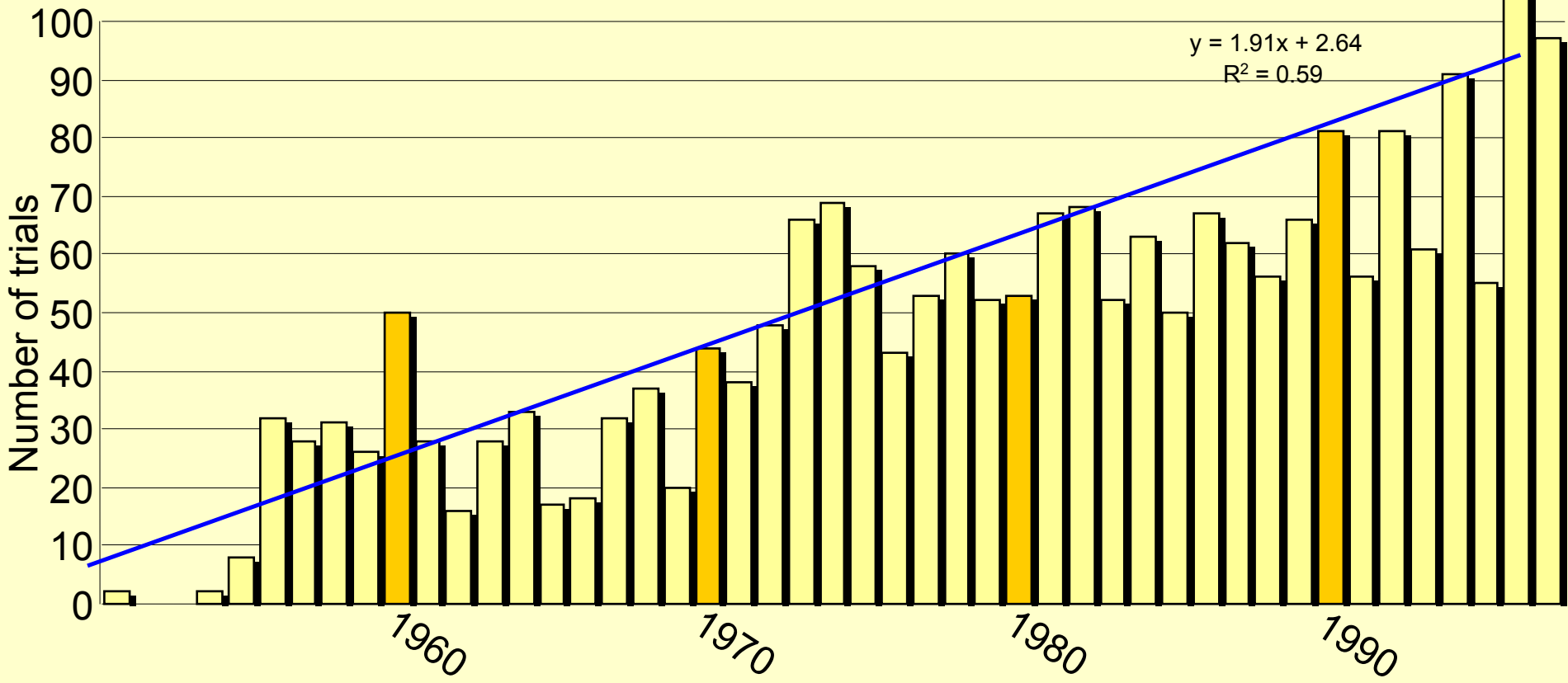
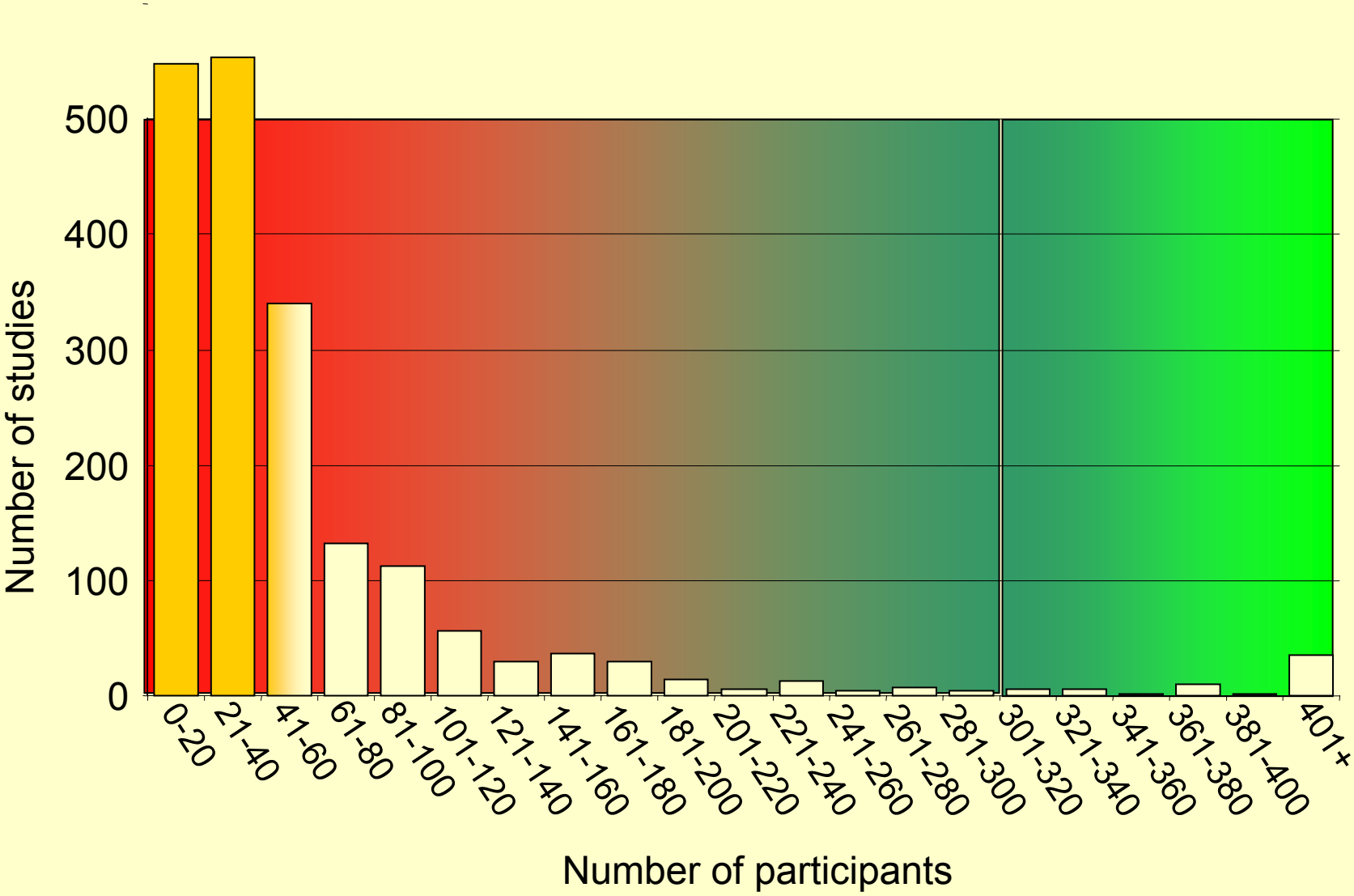
Clinical Trial Methodology (and the placebo effect)

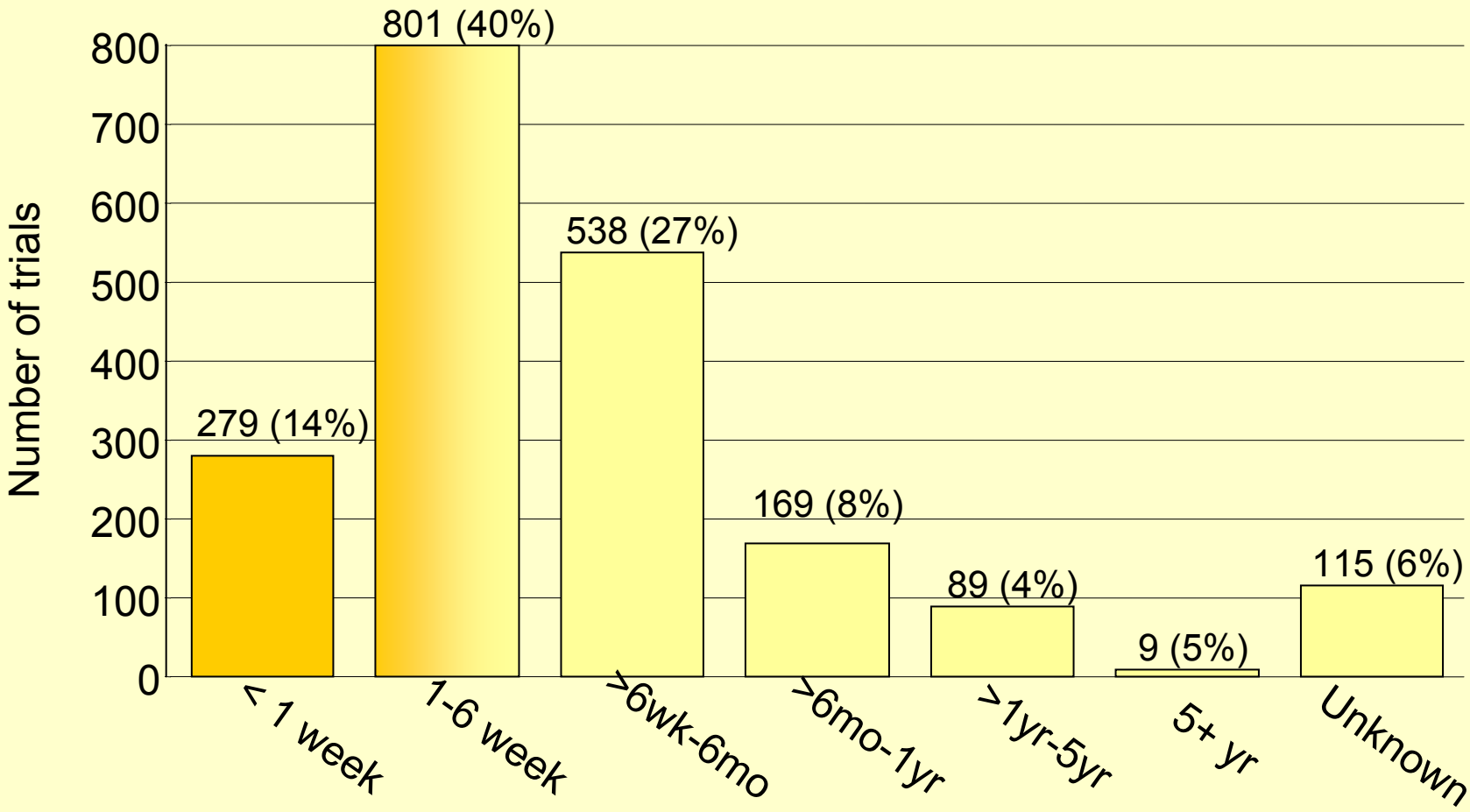
Number of antipsychotic trial reports over time



Frequency of trial size



Numbers of trials by duration



Interventions tested

- 531 interventions in the first 2000 antipsychotic trials

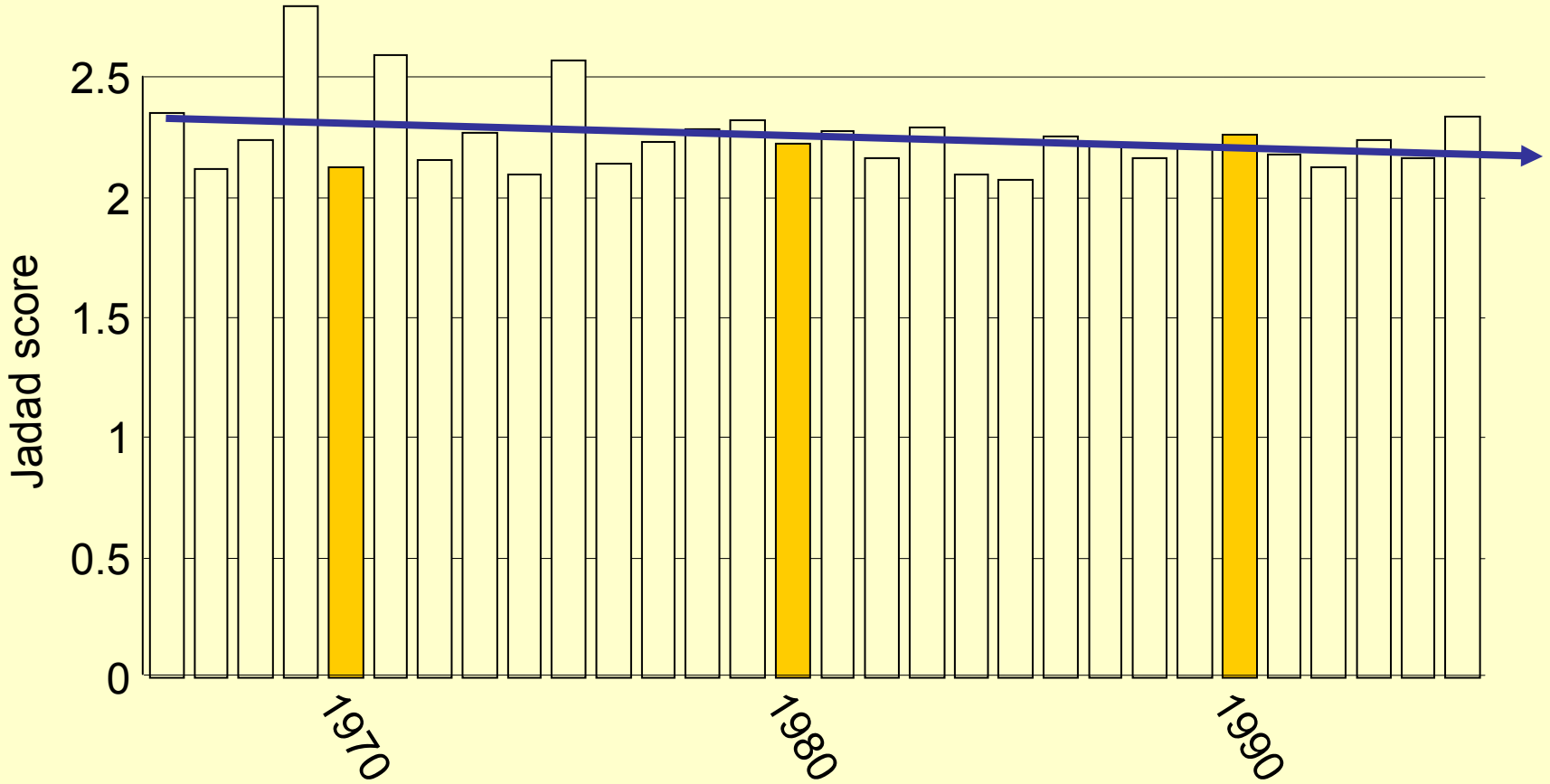
  (Marshall et al., BJPsych, March 2000.)

Outcome Measures used

- 640 different scales in first 2000 trials (Marshall et al., BJPsych, March 2000.)

Mean quality score (1966-95)

(score 0-5; 5 = good)



Types Of Clinical Trial:
By Phase Of Development

Phase I Trials:	Clinical Pharmacology in human volunteers
Phase II Trials:	Early trials in patients to establish indications, dose and efficacy
Phase III Trials:	Large (?) studies in patients to establish comparative efficacy
Phase IV Trials:	Post-marketing surveillance

NB for the development of single drug:

Average Cost: >800 Million Dollars per Drug

Average Time: ~10-15 Years from Discovery to Commercial Availability

Phase I Studies - human pharmacology

1. Absorption, metabolism and excretion
2. Pharmacokinetics
3. Tolerance
4. Adverse effects
5. Physiological (autonomic) effects
6. Central effects
7. Psychomotor function
8. EEG

So:

- single dose built up to adequate dose or detectable effect
- multiple doses
- 24 hour clinical observation
- exclude elderly, young, women, etc

Phase I and early Phase II studies

- Open
- Single blind
- Escalating dose with randomised placebo

Late Phase II and Phase III studies

- Controlled
- Randomised
- Double or triple blind

Phase IV: Methods of post-marketing surveillance

1. **Voluntary Reporting**

   CSM yellow cards; black triangles; red alerts.

2. **Intensive Surveillance**

   Medicines Evaluation and Monitoring Group (MEMO)

3. **Retrospective Methods**

   e.g. case control studies

4. **Prospective Methods**

   Monitored release (Clozapine)

   Cohort study e.g. First 10,000 patients exposed to a certain drug. May detect ADR risk of 0.1%

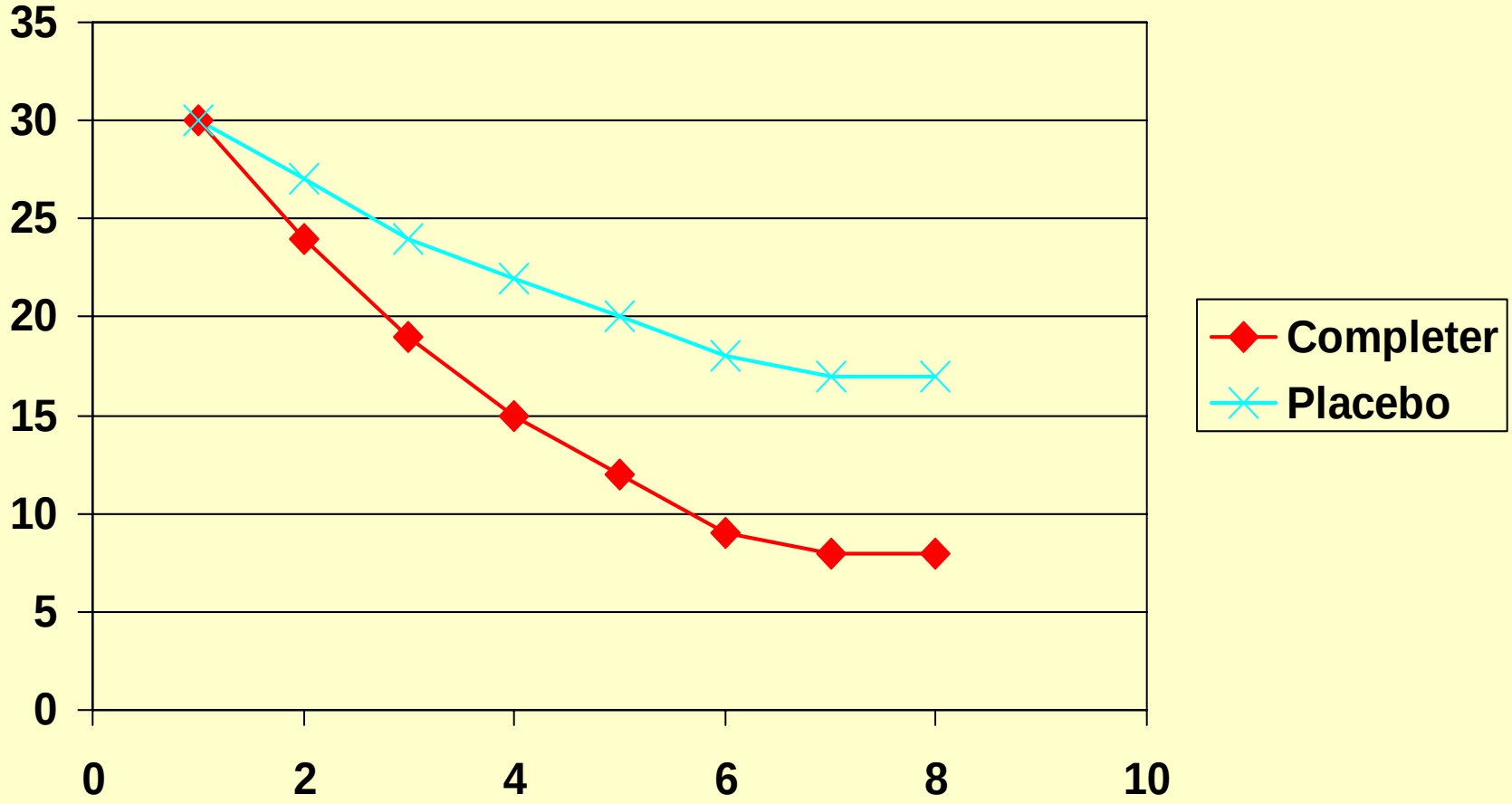
Randomised Controlled Trials

- Advantages
  - Uses hypotheticodeductive reasoning
  - Eradicates bias if properly conducted
  - Allows for meta-analyses
- Problems
  - Expensive and time consuming therefore
    - often not done
    - funded by pharmaceutical industry
    - use surrogate end points rather than clinically valid ones
  - Large studies can find statistically significant but clinically irrelevant differences.
  - Results may not be generalisable
  - Not always ethical

Randomised Controlled Trials: Points to look out for

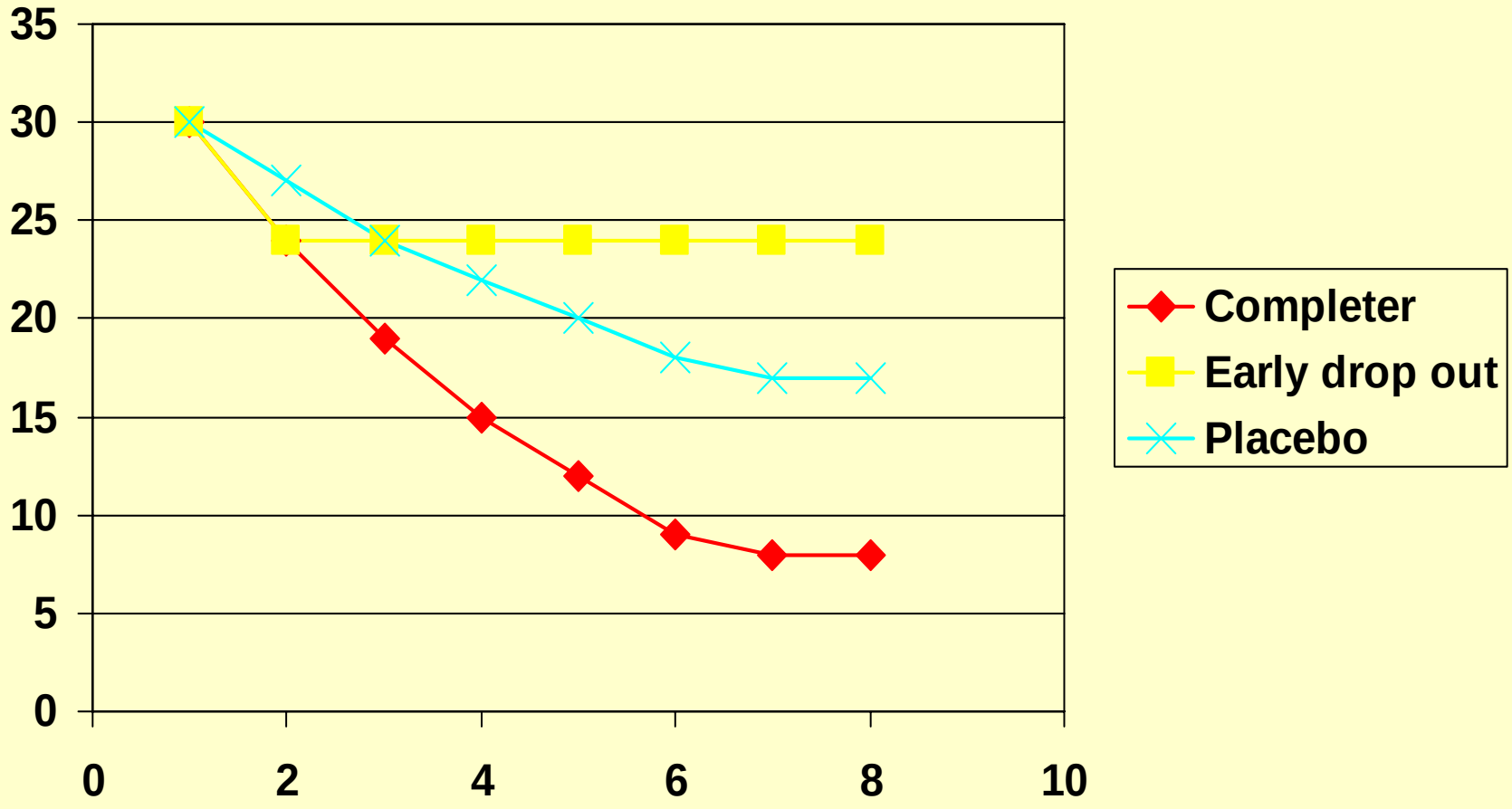
- Was the study population clearly defined?
- Was randomisation correctly carried out or did it introduce hidden bias?
  - e.g. not randomising all eligible patients
- Were patients and assessors truly “blind”?
- Were the clinical assessment scales appropriate?
- How was the data analysed? (e.g. ITT analysis)

Completer analysis, ITT, LOCF and MMRM



MMRM = Mixed model repeated measures

Completer analysis, ITT, LOCF and MMRM



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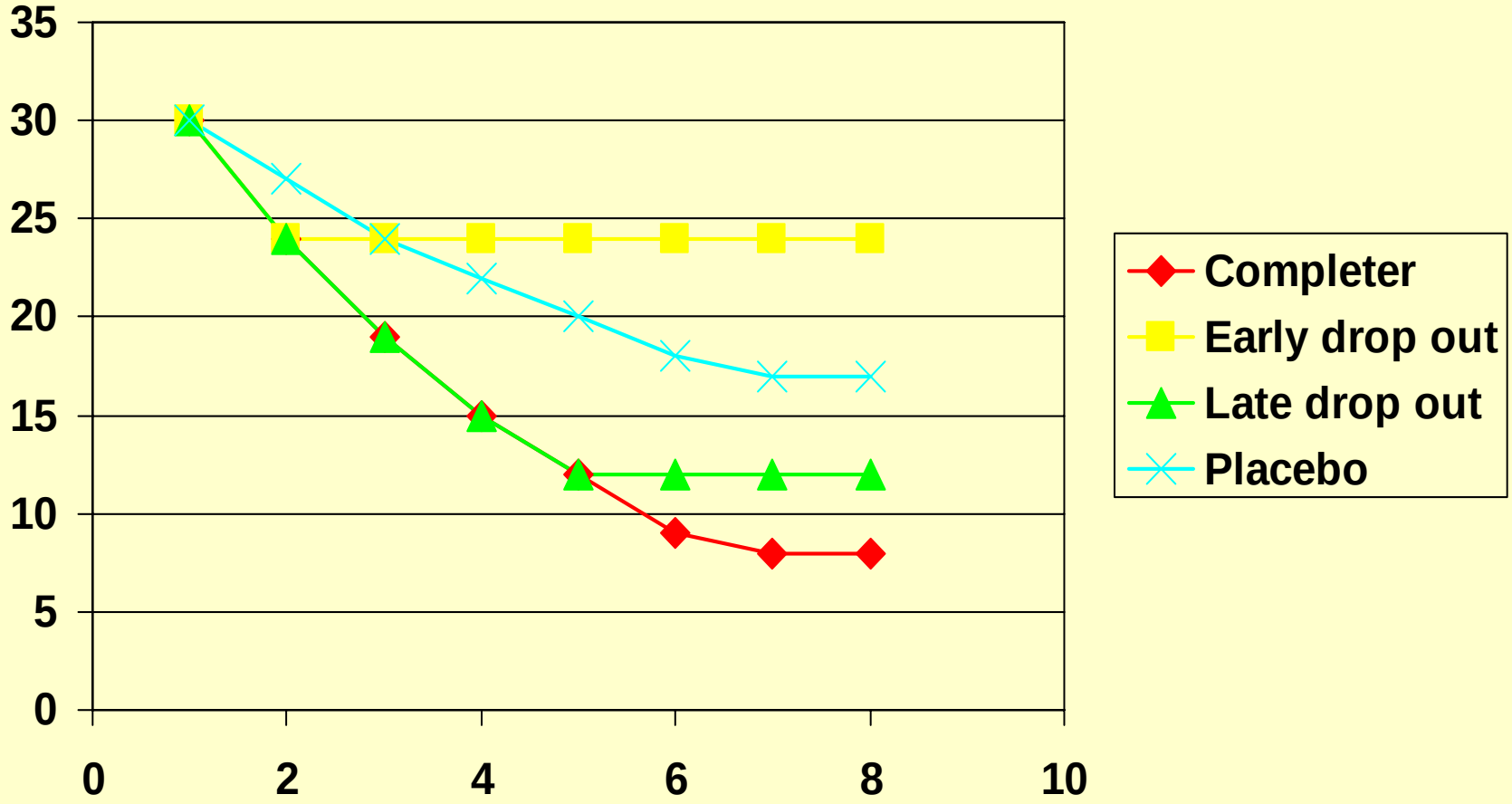


MMRM = Mixed model repeated measures

A note on randomisation

- Rationale – to balance confounders and reduce selection bias
- Conduct – allocation must be concealed (NB this is not the same as blinding)
- Types – simple, blocked, stratified…
- Unit – individuals or groups

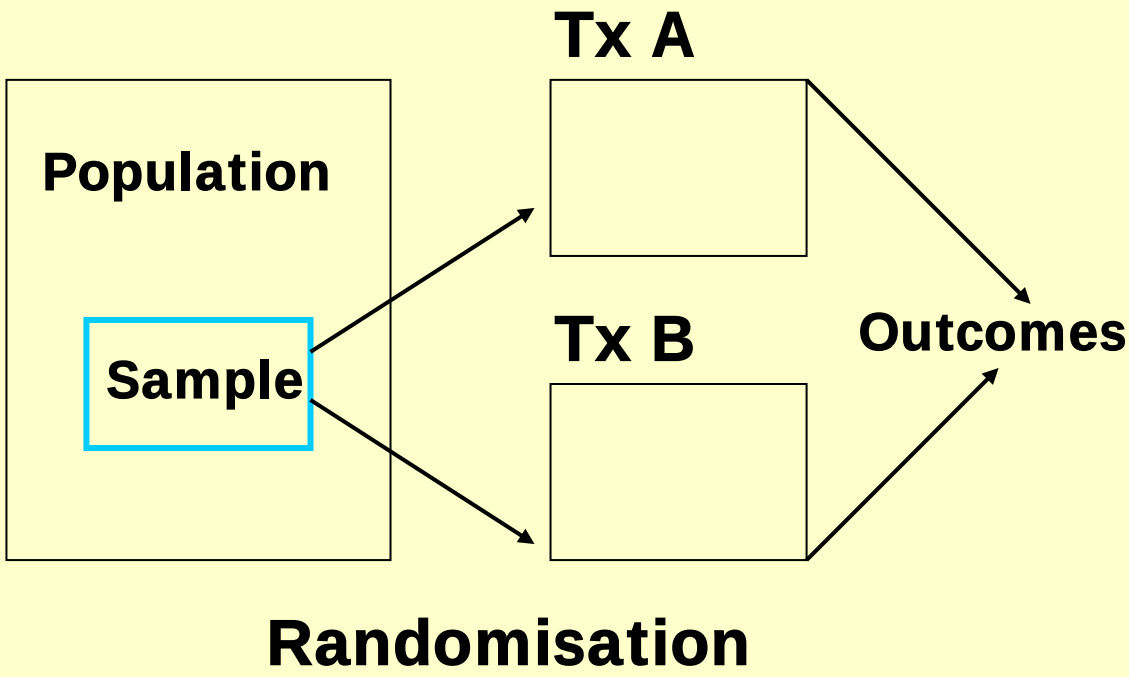
	Randomisation		Non-random assignment
	Cheating difficult (%)	Cheating easy (%)	(%)
Proportion of studies with p=<0.05	8.8	24.4	58.1

From Chalmers et al, NEJM, (1983) 309, 1358-61.

Design of RCTs

- Parallel
- Withdrawal
- Run-in
- Cross over
- Factorial

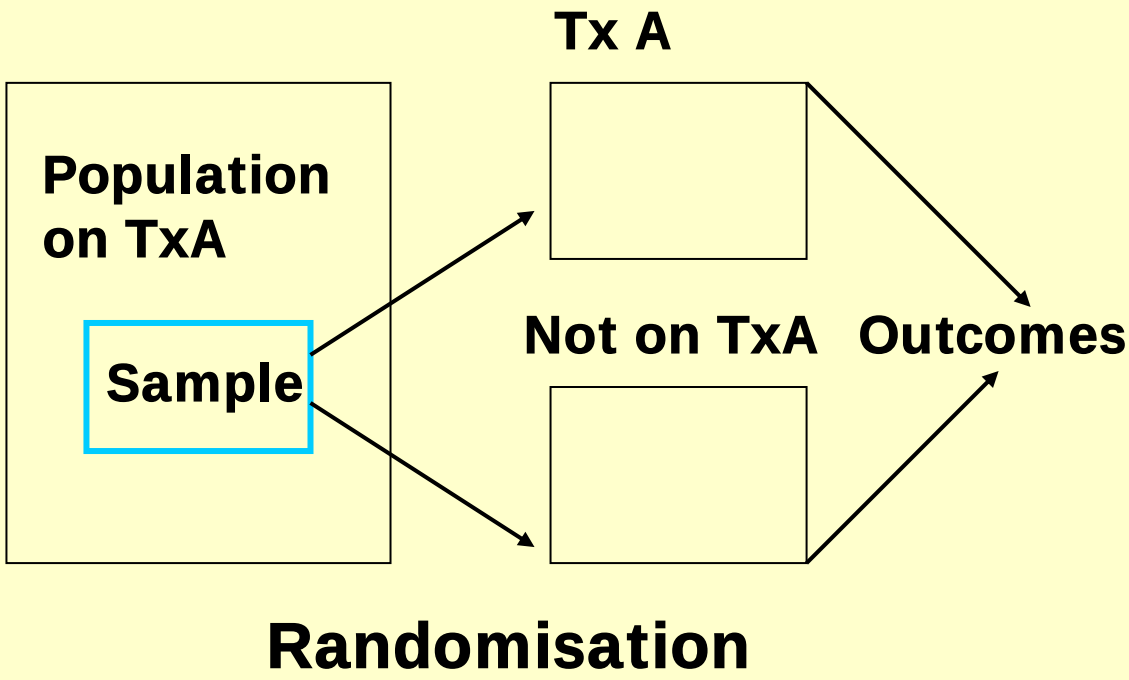
Design of RCTs: Parallel



Parallel Design

- Advantages
  - Few Assumptions
  - Simple Statistical Approach
  - Can be used in all situations
- Disadvantages
  - Time
  - Cost

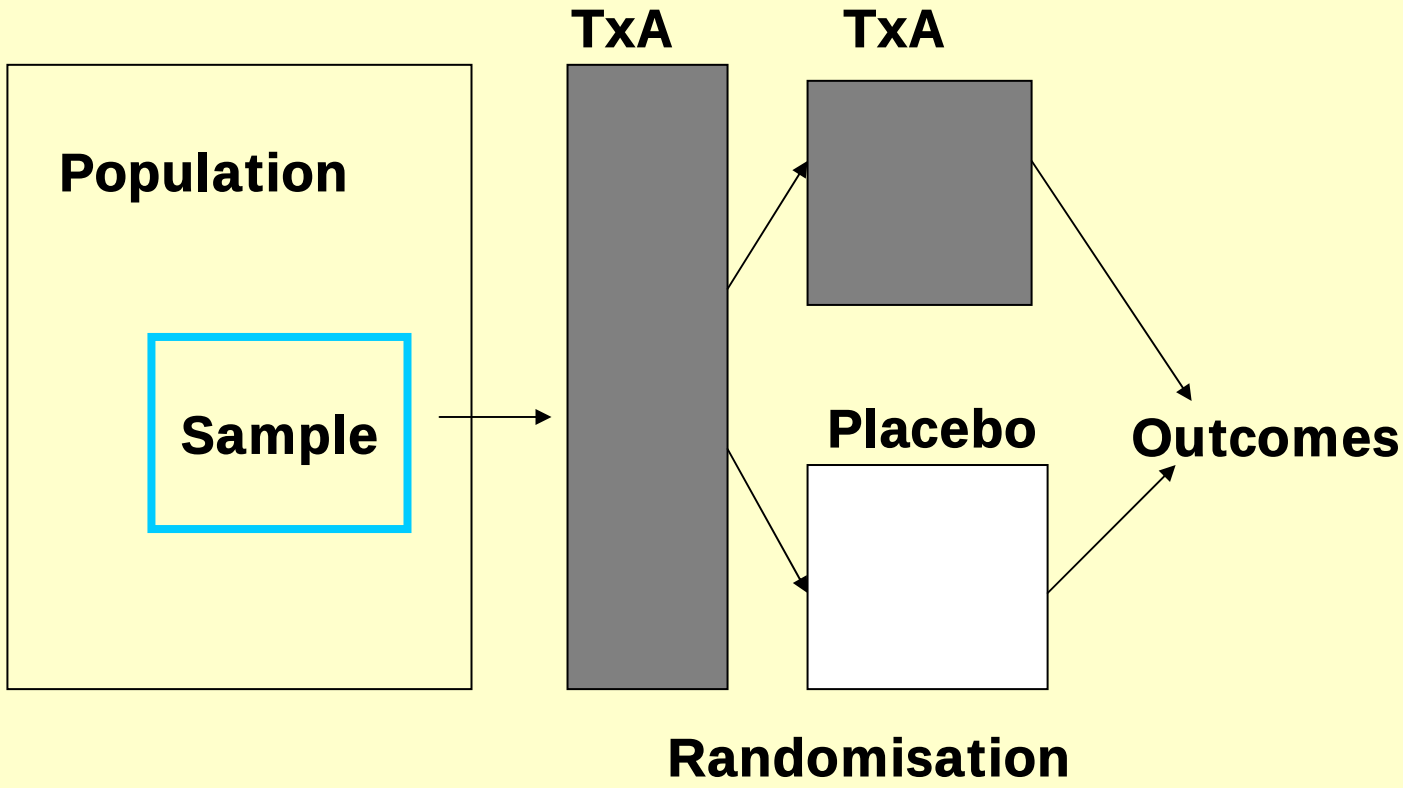
Design of RCTs: Withdrawal



Withdrawal Design

- Advantages
  - Easy access to subjects
  - Show whether proven treatment remains beneficial
- Disadvantages
  - Selected population
  - Different stages of disease
  - Maintenance vs continuation

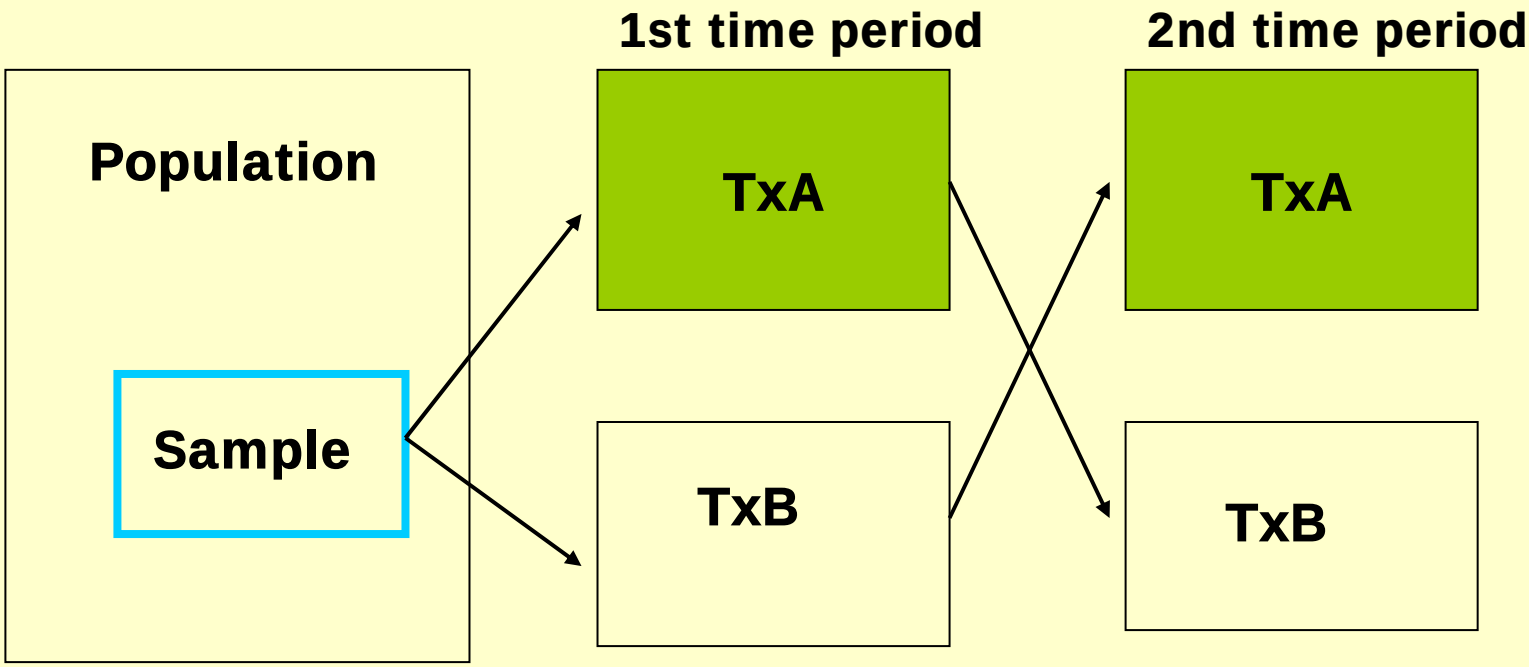
Design of RCTs: Run in



Run In Design

- Advantages
  - Excludes “non-compliant”, treatment “intolerant” subjects or placebo responders
  - Reduces drop outs
- Disadvantages
  - Delay in randomization
    - Time
    - Loss of events
  - Increased chance of unmasking
  - More selective sample

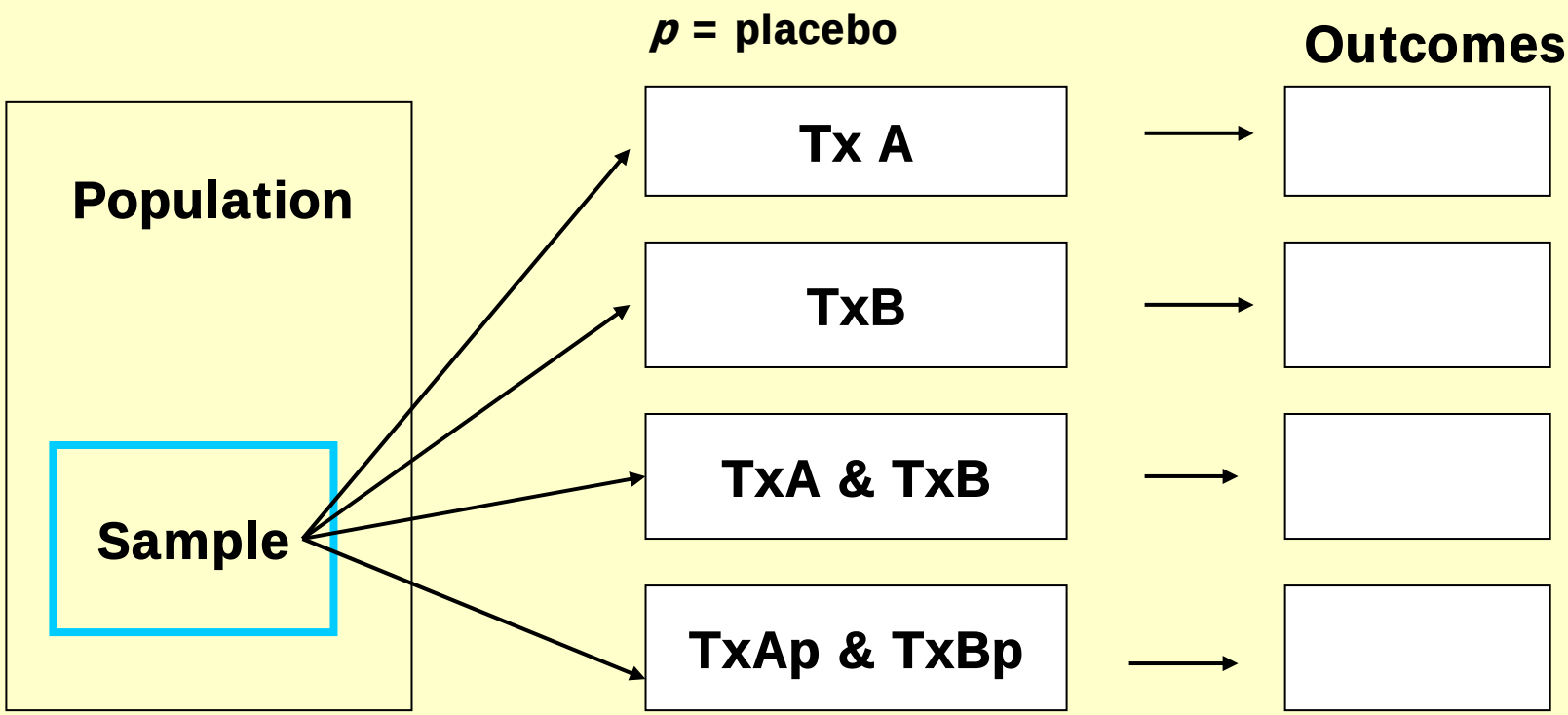
Design of RCTs: Cross Over



Cross Over Design

- Advantages
  - Use of the same sample twice
  - Reduces variability as patient acts as own control
- Disadvantages
  - Assumes no carryover effect
  - Assumes stability of disease process

Design of RCTs: Factorial



Factorial Design

- Advantages
  - Addresses two (or more) questions at the same time
  - Offers information on interactions
  - Get information whether

    two treatments are better than one
- Disadvantages
  - Complexity
  - Harder recruitment
  - Impact on compliance
  - Polypharmacy

Types of research design

- Randomised controlled trials
- Cohort studies
- Case controlled studies
- Cross sectional surveys
- Single case studies

Cohort Studies

- Also referred to as prospective, follow up, or outcome studies
- Group identified and watched to see what happens
- Best practical way of identifying risks and prognosis

- Advantages
  - Large studies provide powerful evidence
- Problems
  - Can take a long time
  - Difficult not to loose large proportion of cases over time (leads to bias)
  - Differences prior to recruitment may effect results
  - Observed association between two variables may be due to a third (confounding) non-observed variable

Case Controlled Studies

- Also known as case-comparison or retrospective study
- A group of individuals (defined by characteristic of interest) compared to a group of controls
- Useful for examining associations of rare conditions

- Advantages
  - Quick, cheap and easy (relatively)
  - Usually the only option for rare conditions
- Problems
  - May rely on accurate notes and retrospective diagnoses
  - May rely on long term memory of doctors and patients
  - Questionnaire bias, recall bias, surveillance bias
  - Often don’t have much statistical power
  - Observed association between two variables may be due to a third (confounding) non-observed variable

Efficacy V Effectiveness

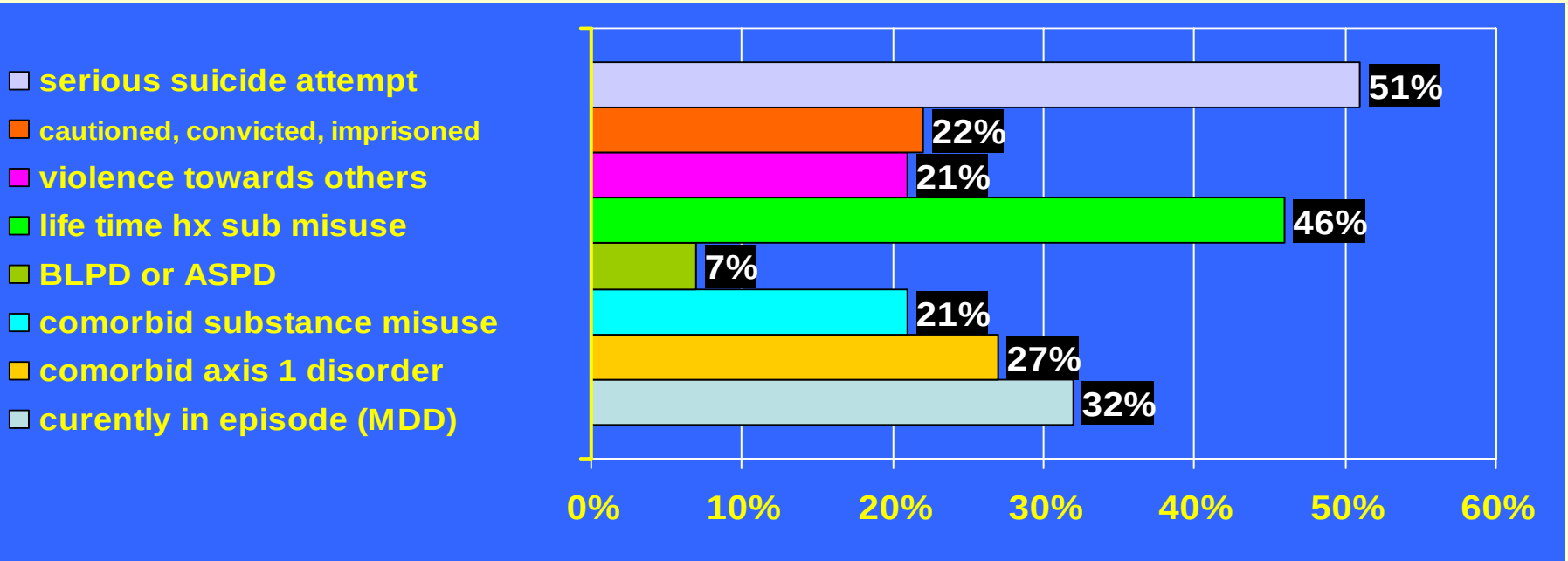
- Efficacy studies – homogenous population, euthymic, no comorbidity etc important to establish treatment is beneficial

- Effectiveness- heterogenous (messy) reflect clinical practice, pragmatic- important in telling us what happens in reality

MRC STUDY- Scott et al, 2006

- 5 centres: 4 provincial cities, 1 university town
- 253 subjects, recruited via case registers, hospital data, CPA
- At least 1 manic episode in 12 ms, but not currently manic
- Mean Age 42ys, Median Onset 25ys, Median Episodes>20

80% >=2 & 70% >=3 of the following Clinical Features-



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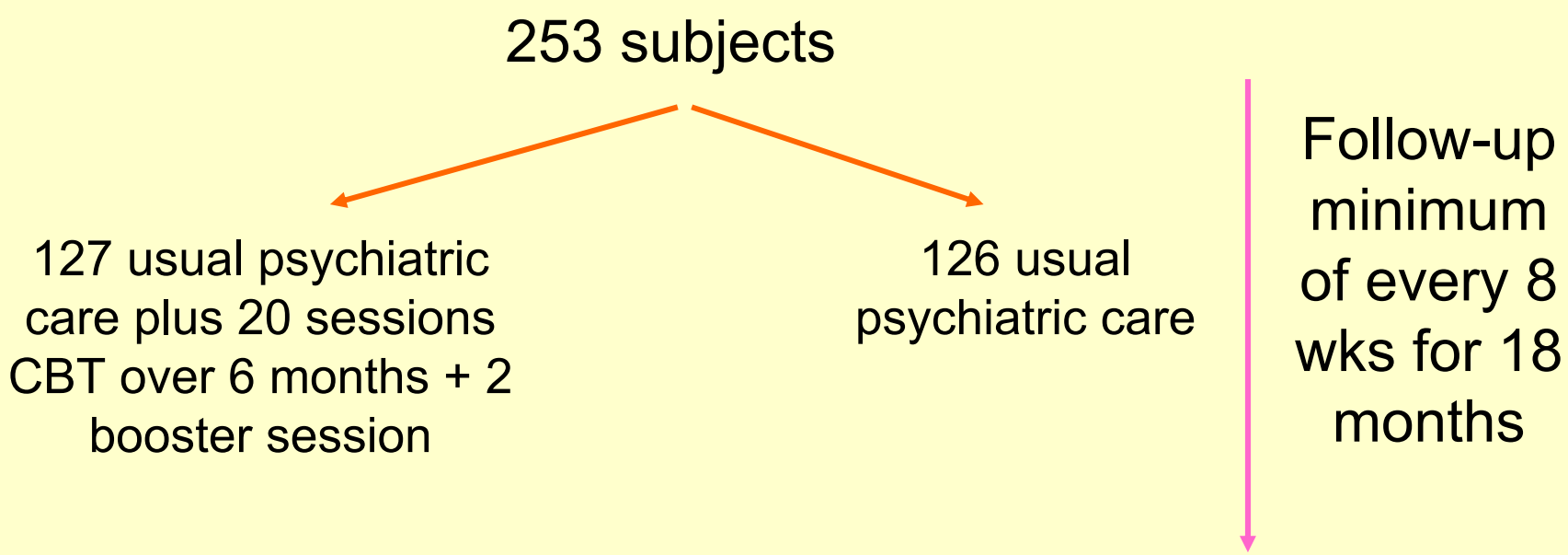
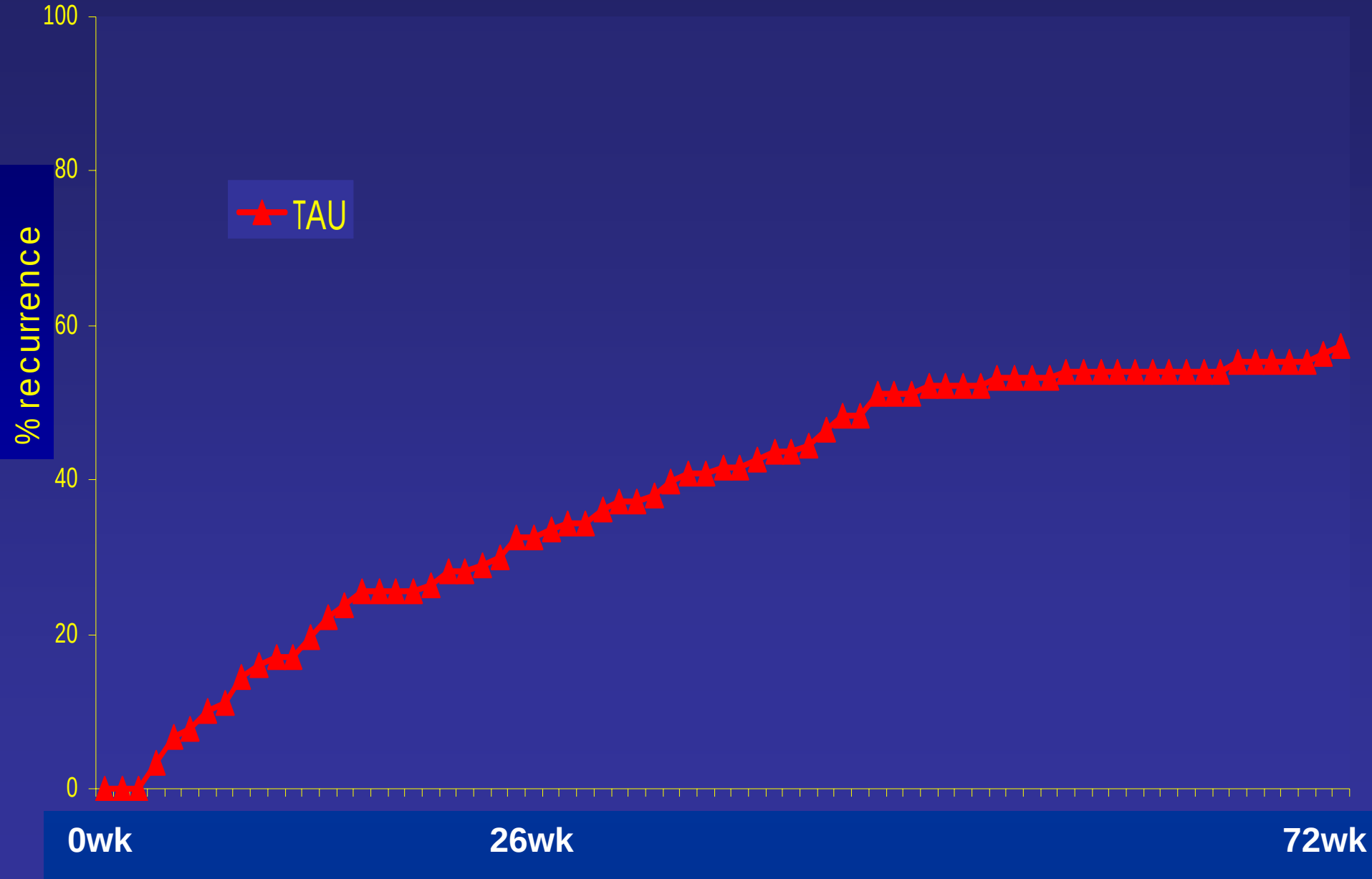


Actuarial cumulative recurrence curves (Kaplan Meier): ITT analysis of any recurrence (Scott et al, 2006)



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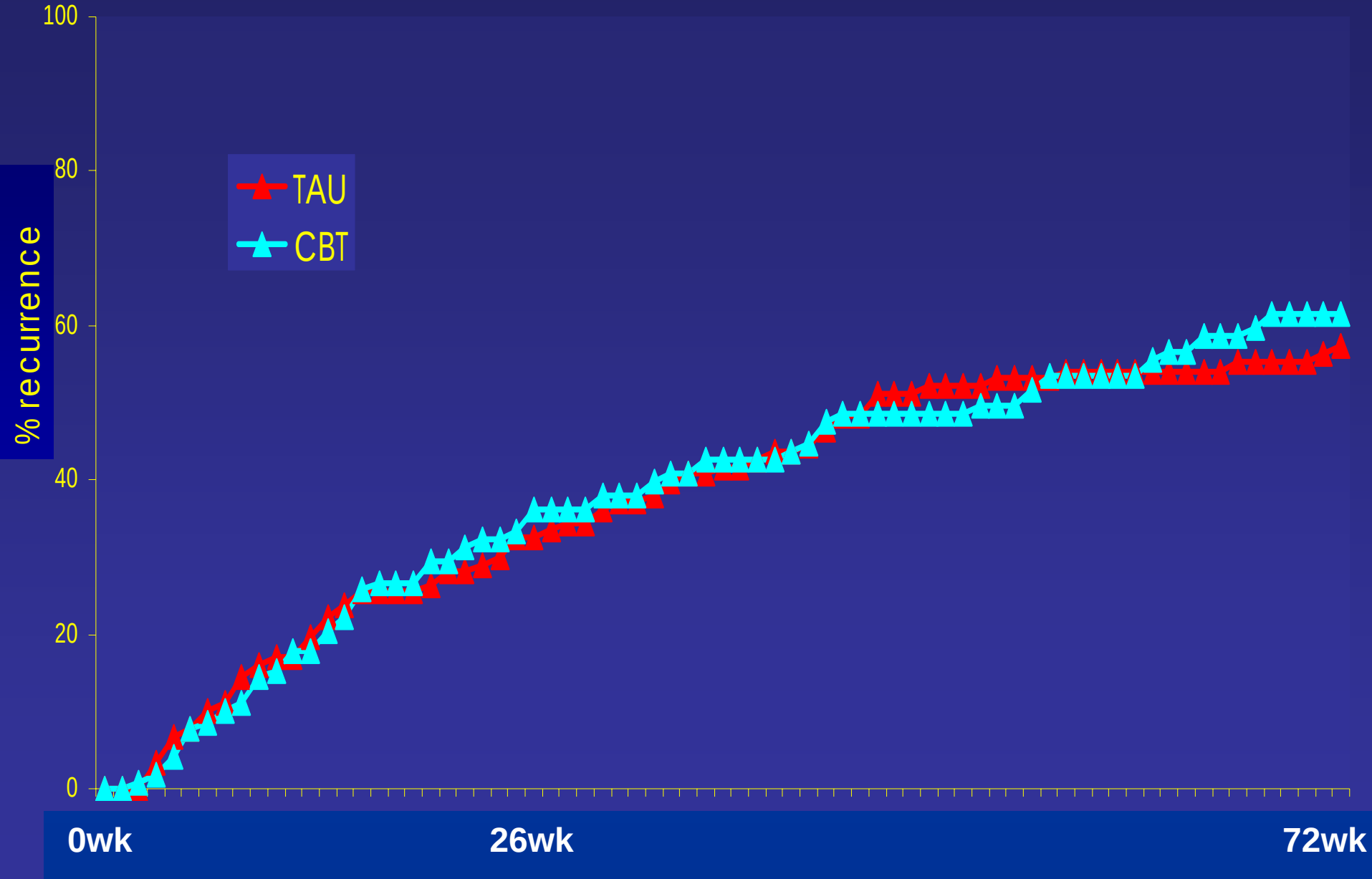
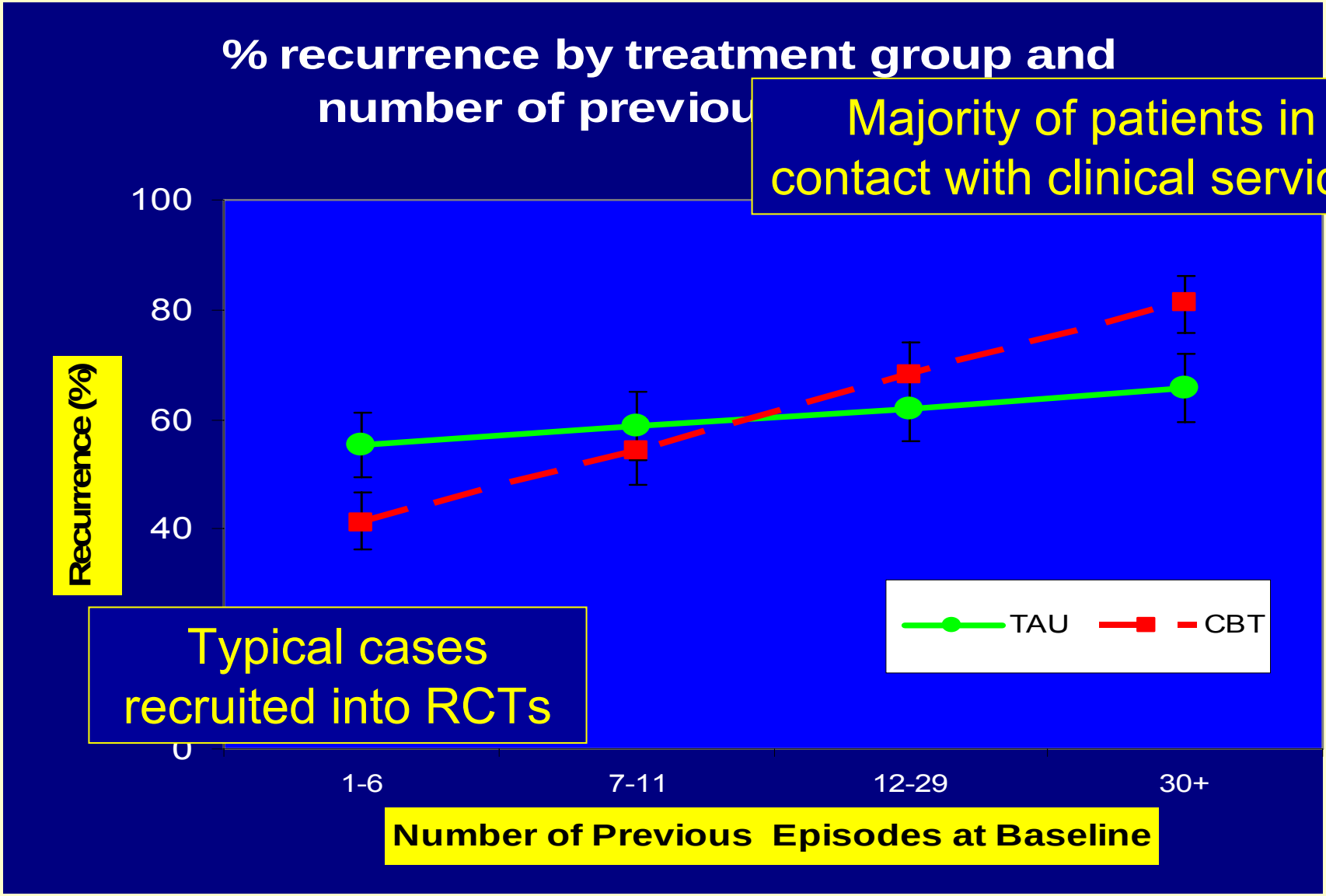


Secondary Analysis



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The placebo effect

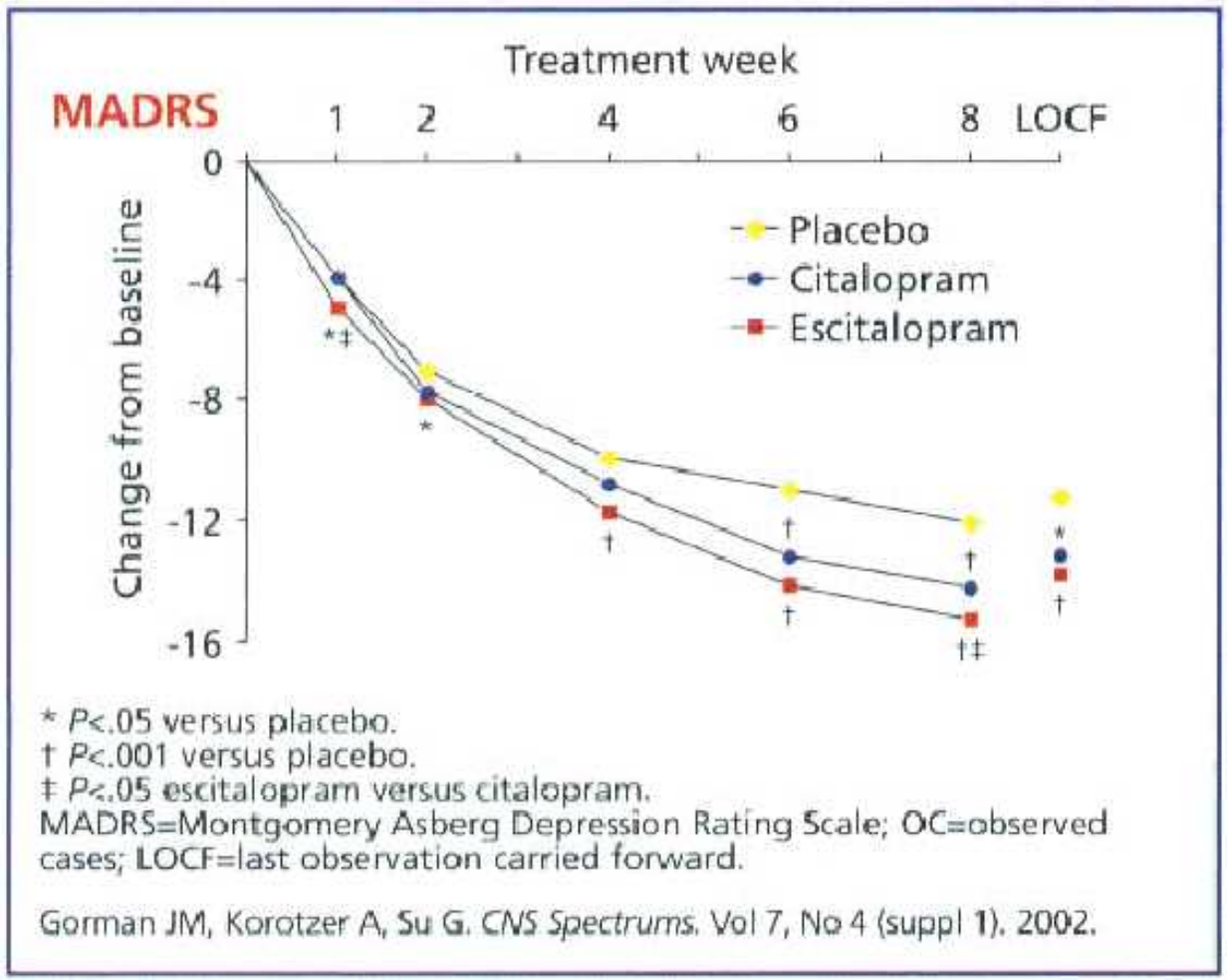
“Astonishing Medical Fact: Placebos Work! So Why not use them as medicine?”

New York Times Magazine January 2000

Placebo – I will please

- 1785 Motherby's New Medical Dictionary- 'a commonplace method or medicine calculated to amuse for a time, rather than for any other purpose'
- C19 - 'an inactive substance', a 'makebelieve medication'
- By early 20C - equating of placebo with sham treatment acting by suggestion
- WHR Rivers 1908 – first recorded use of inert substance as a control for effect of alcohol and stimulants on fatigue
- Linked to development of clinical trials and the RCT
- Henry Beecher 1955 – 'The Powerful Placebo'

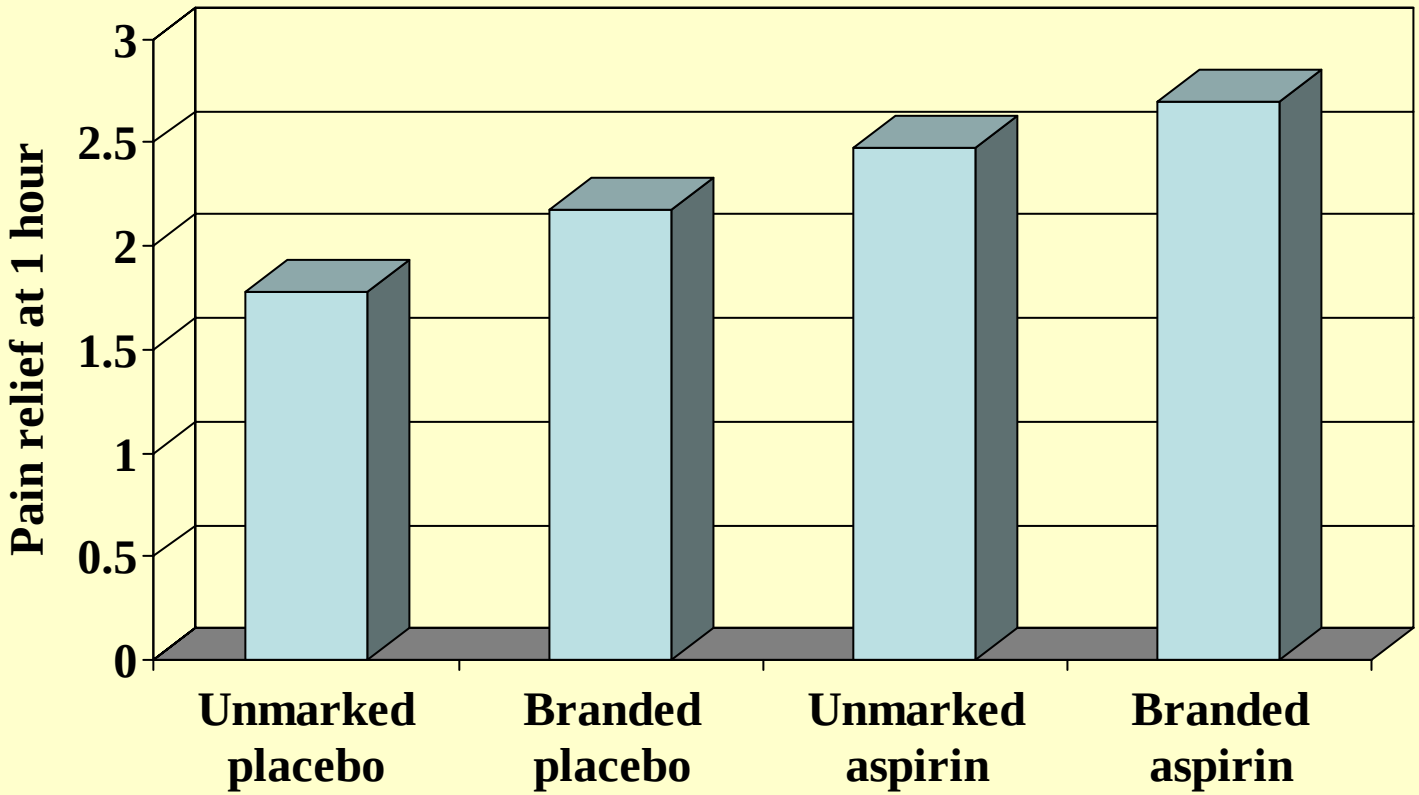
Efficacy of escitalopram vs citalopram vs placebo



One should use drugs quickly before they lose their effectiveness (William Osler)

Placebo and branding effects on analgesia for headache

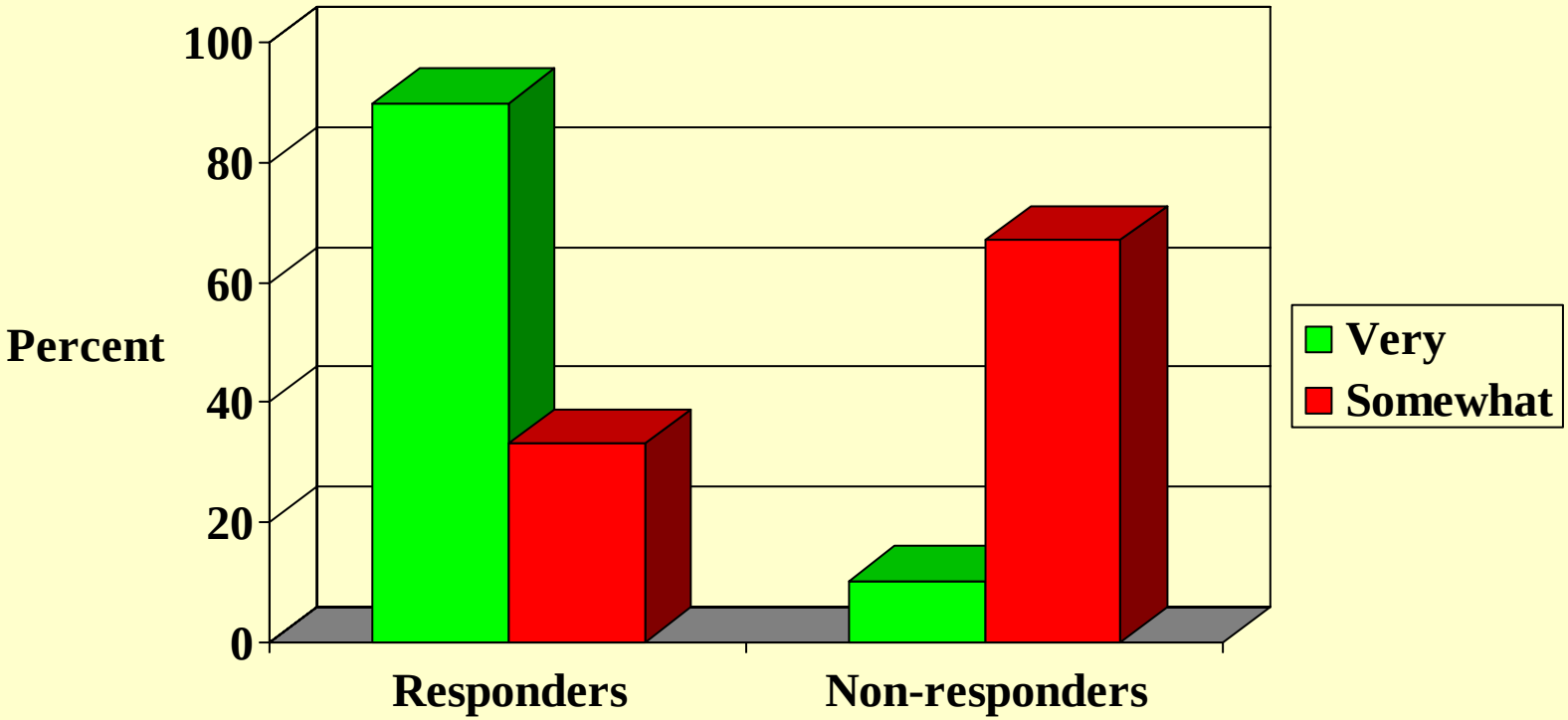
Significant effects of brand vs unmarked and aspirin vs placebo



Branthwaite et al 1981

Patient expectations and response to reboxetine

N=25, Single bind 8 week study, response = HAMD<11
Expectations: 10 very effective, 15 somewhat effective



Krell et al APA 2003

Interaction between drug and placebo effects

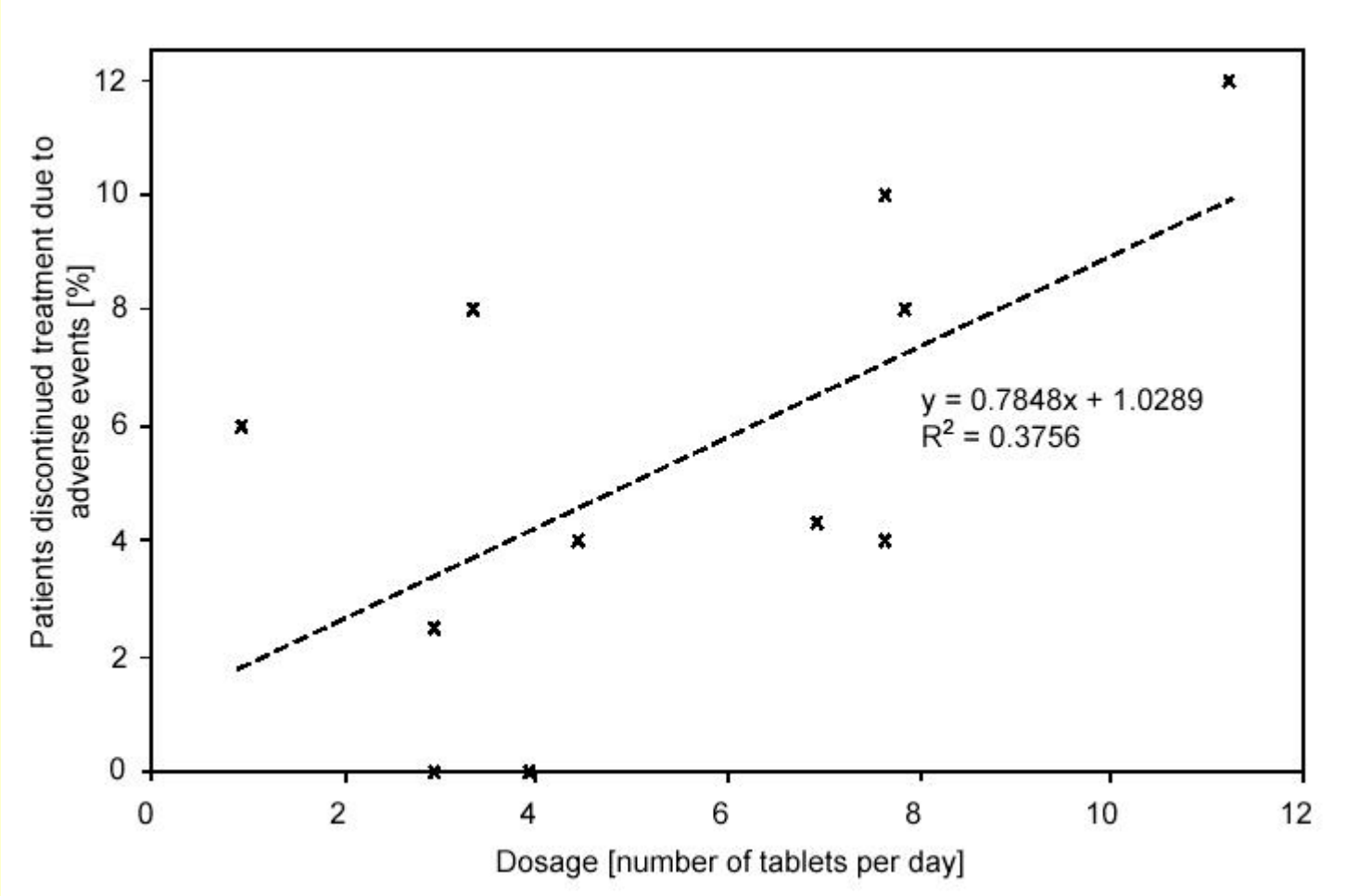
Table 4.1 "Adherence" and response to drug and placebo.

Drug tested	N	Outcome	Drug group		Placebo group	
			Adherent*	Not adherent	Adherent*	Not adherent
clofibrate	3760	5 year mortality	15·0%	24·6%	15·1%	28·2%
antibiotics	150	infection after chemotherapy	18·1%	53·0%	32·2%	64·0%
chlorpromazine	374	1 year relapse	13·0%	57·0%	40·0%	80·0%
propranolol (men)	2175	1 year mortality	1·4%	4·2%	3·0%	7·0%
propranolol (women)	602	1 year mortality	4·5%	8·7%	6·8%	19·0%
amiodarone	1141	2 year all cause mortality	7·4%	14·8%	8·8%	18·7%

*Adherent patients took more than 75% or 80% of medication (65% in amiodarone study); non adherent patients took less.

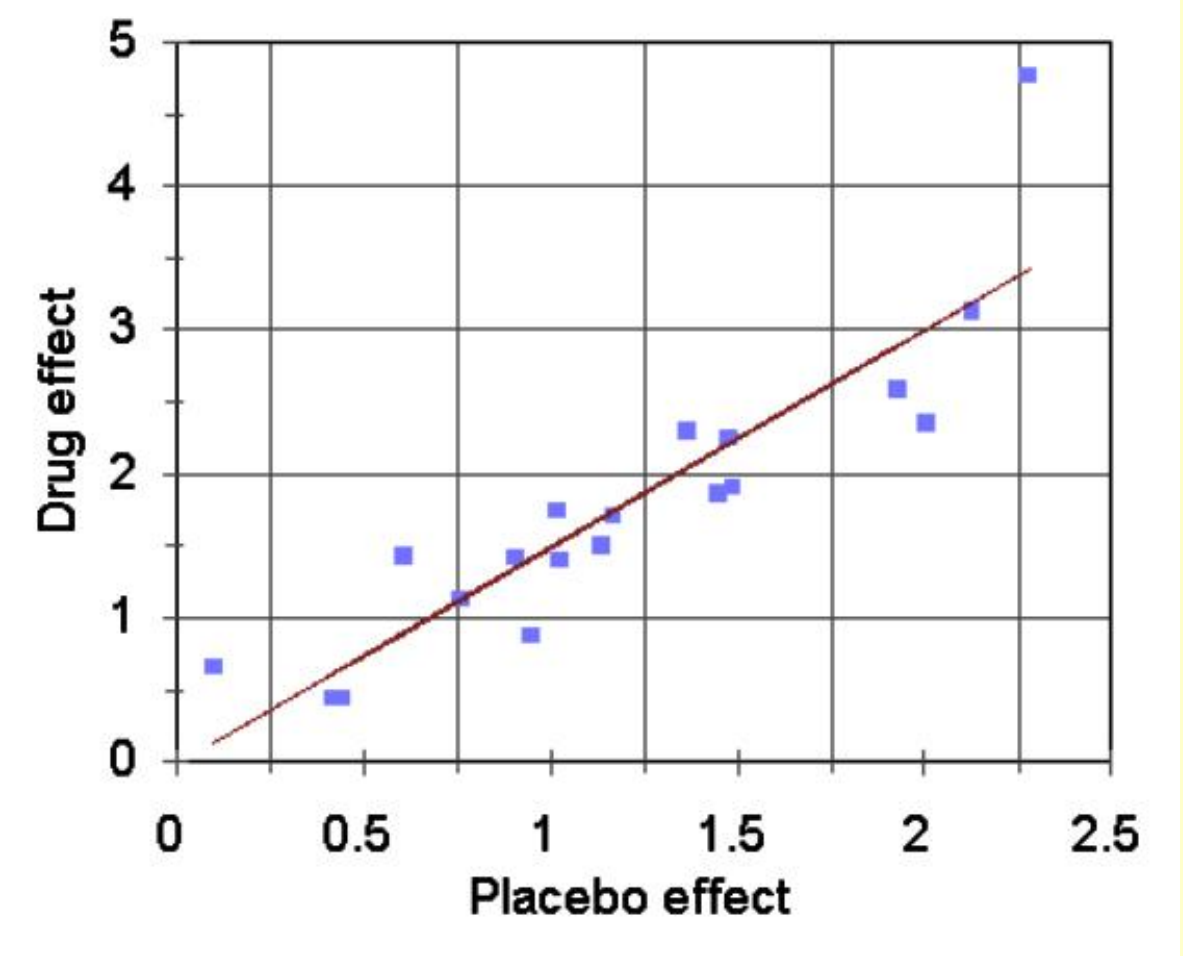
Moerman 2002

Effect of dose of placebo on side-effect dropouts



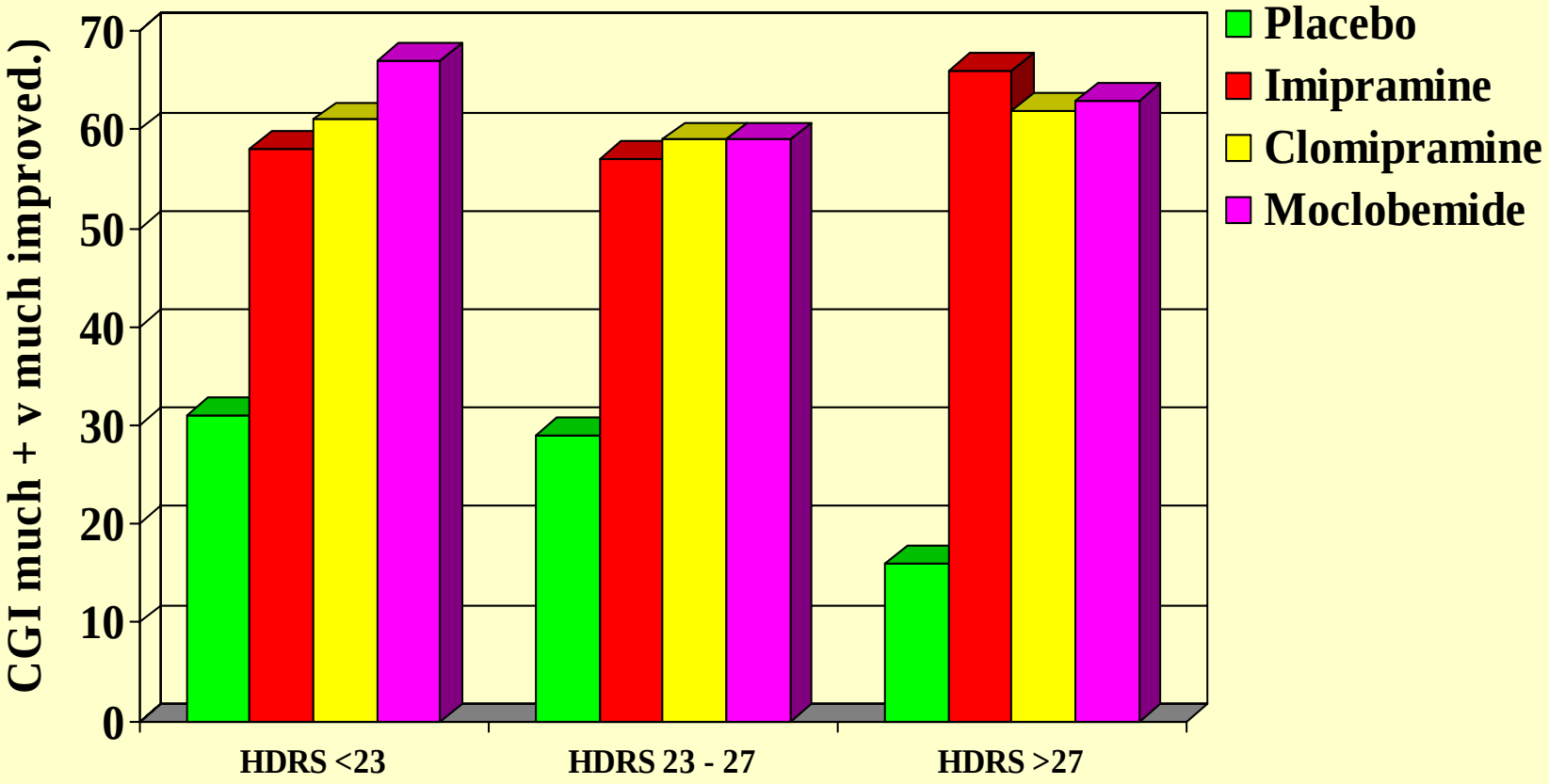
Lebeda et al APA 2003

Relationship between anti-depressant and placebo response



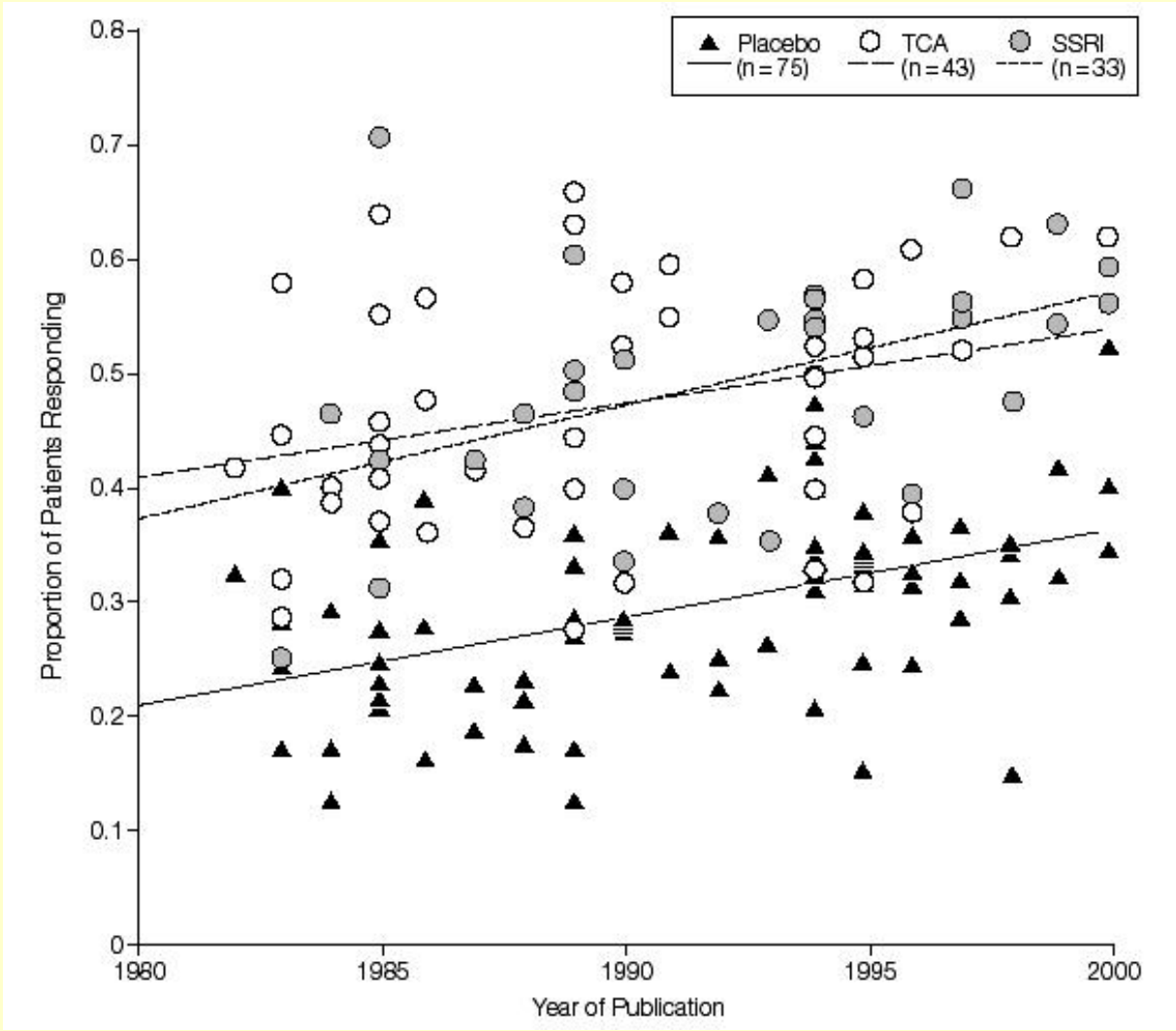
Kirsch et al 1998

Response by severity of depression



Angst et al 1993

Responses over time

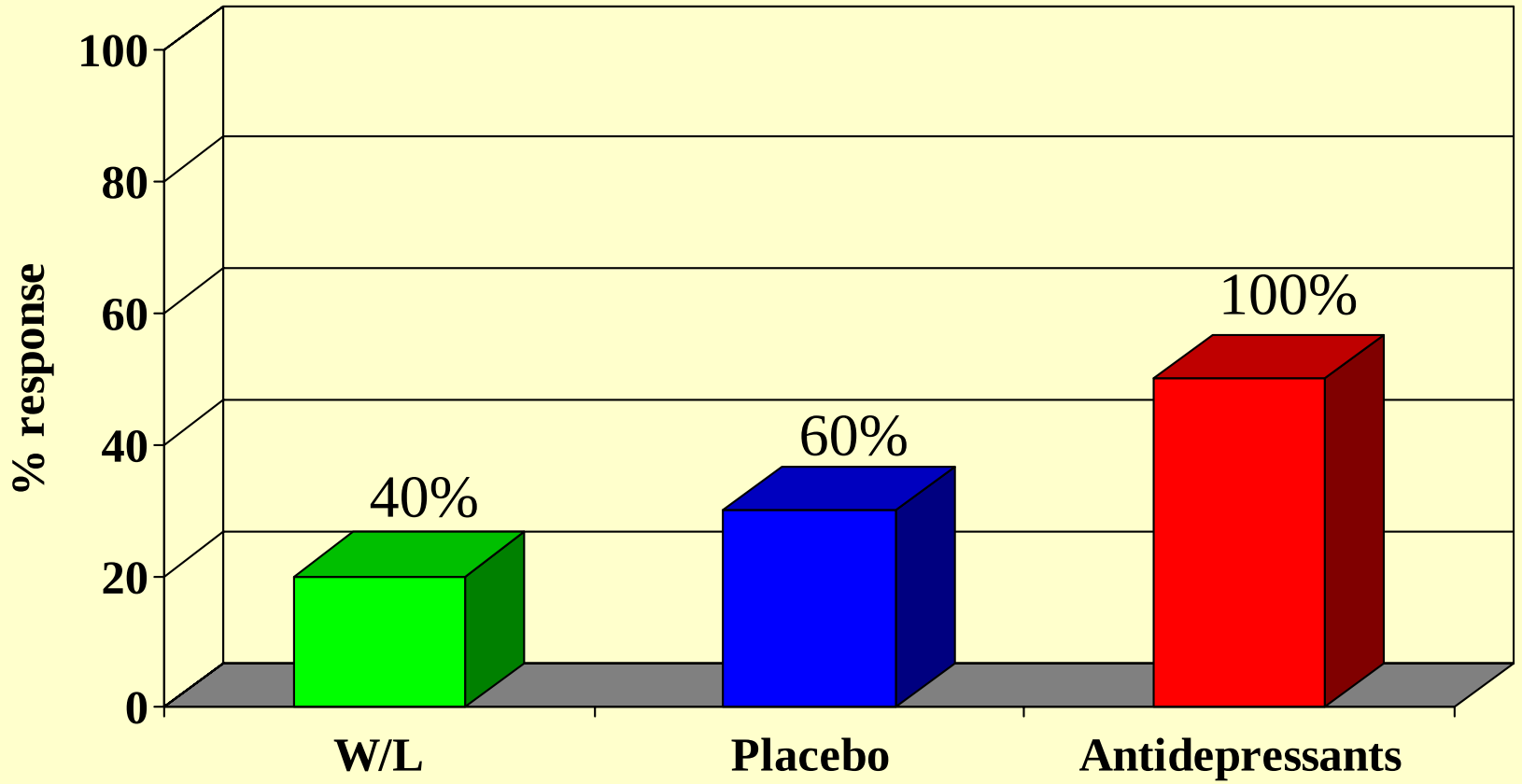


Antidepressants
r=0.26, p=0.02

Placebo
r=0.45, p<0.001

Walsh et al 2002

Response rates in depression



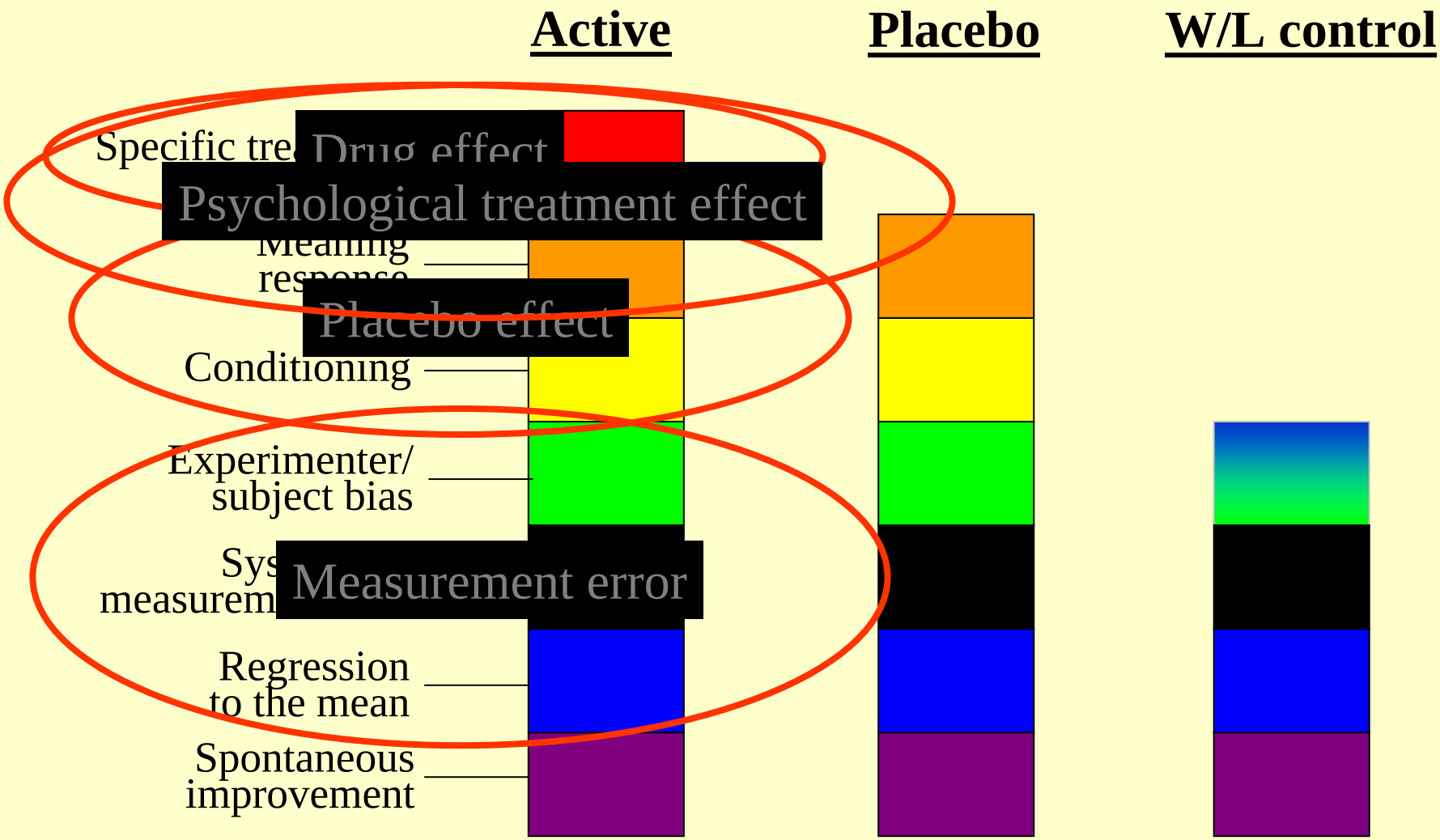
Waiting list – Posternak & Miller 2001
Placebo/antidepressants – Walsh et al 2002

Approaches to explaining the placebo effect

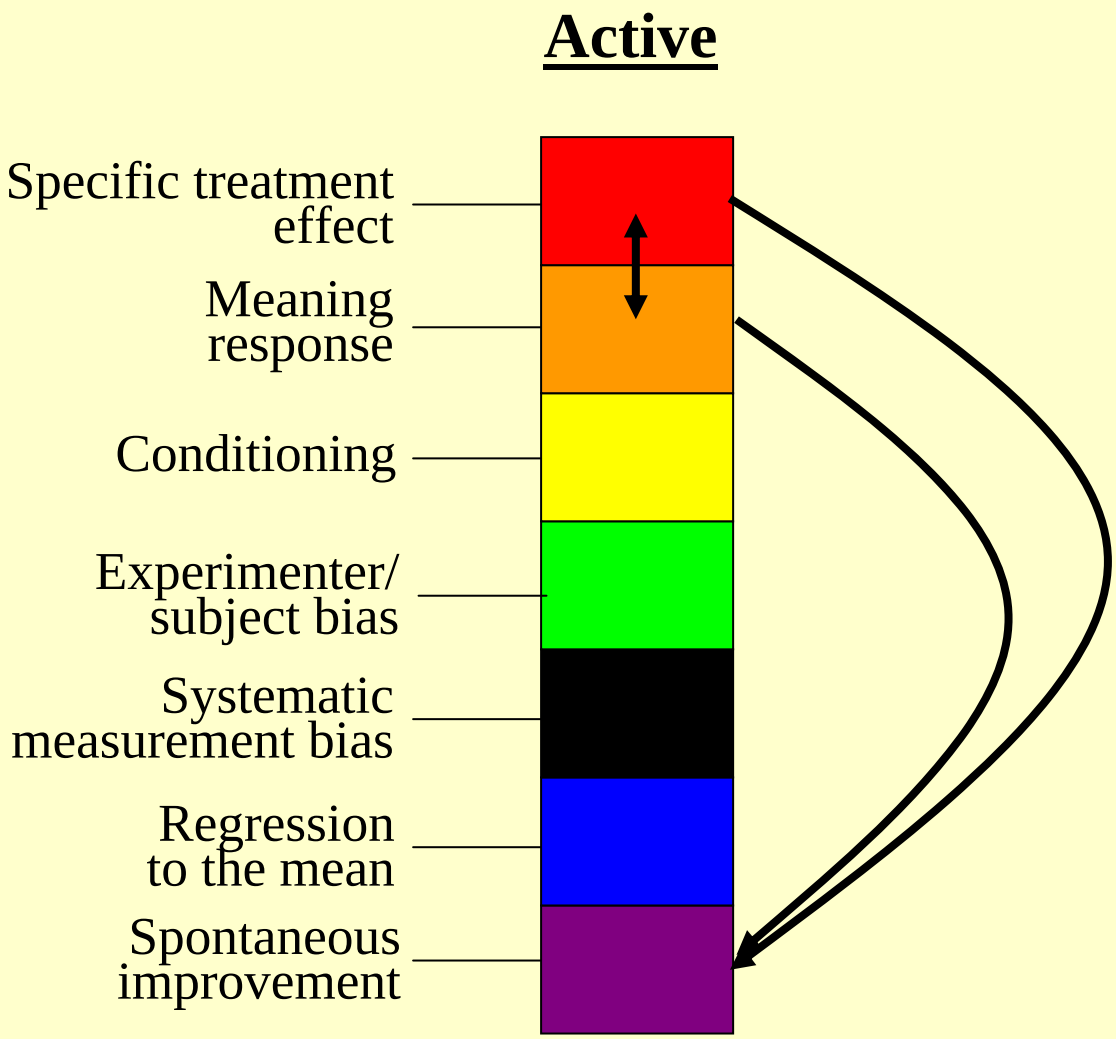
- Individual differences: the ‘placebo responder’
- Individual/collective beliefs
- Interpersonal dynamics: the doctor-patient relationship
- Expectancy
- Attribution theory
- Emotional processes: anxiety reduction/hope
- Brain biochemistry: eg endorphins
- Conditioning

after Moerman 2002

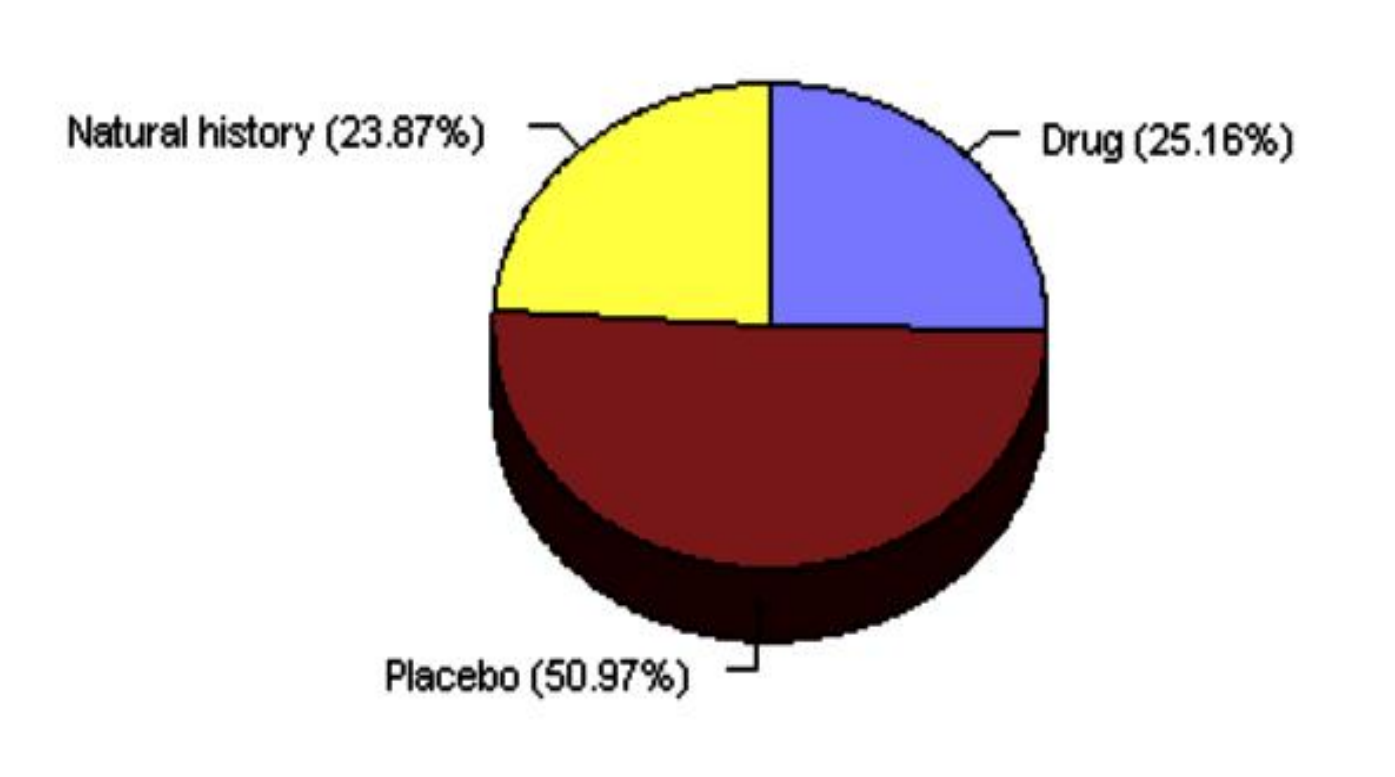
Components of a response



Additive or interactive?



Contributions to the response to antidepressants in depression



But assumes additive rather than interactive model and ignores measurement errors

Kirsch et al 1998

Approaches to explaining the placebo effect

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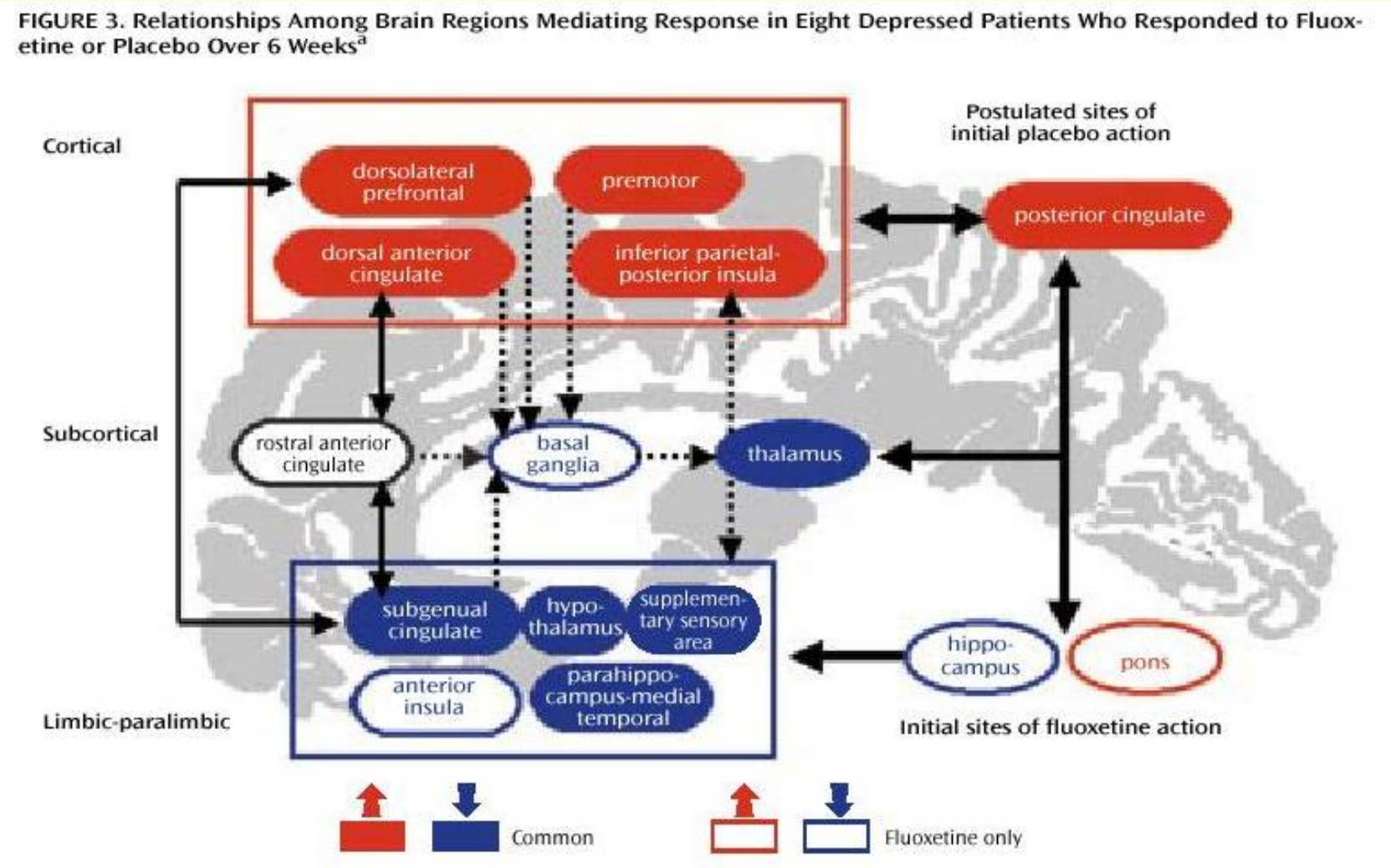
after Moerman 2002

Functional neuroanatomy of drug vs placebo treatment



Mayberg et al 2002

Ethics/implications

Placebo

Clinical trials

- Control for placebo effect to determine specific effects
- Involves standardising treatment
- Regulatory need
- More proof of concept than measure of effectiveness
- Conflicts with the Declaration of Helsinki
- Alternative approaches

Therapeutics

- Maximise placebo effect to improve outcome
- Involves individualising treatment
- Historically –'lie like a doctor'
- Deceptive use of placebo now seen to violate informed consent
- Placebo use of active treatments still an issue

Conclusions

- There is a need to distinguish placebo effects from measurement error and natural history
- Placebo and ‘active’ effects are unlikely to be simply additive
- Placebo effects are more than simply an irritant to be controlled for; they challenge our concepts of mechanism of action
- In therapy the placebo effect presents a paradox
- An important aspect of EBM may be maximising the placebo effect

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Pharmaco-kinetics, dynamics and genetics

‘Clinical Pharmacology’

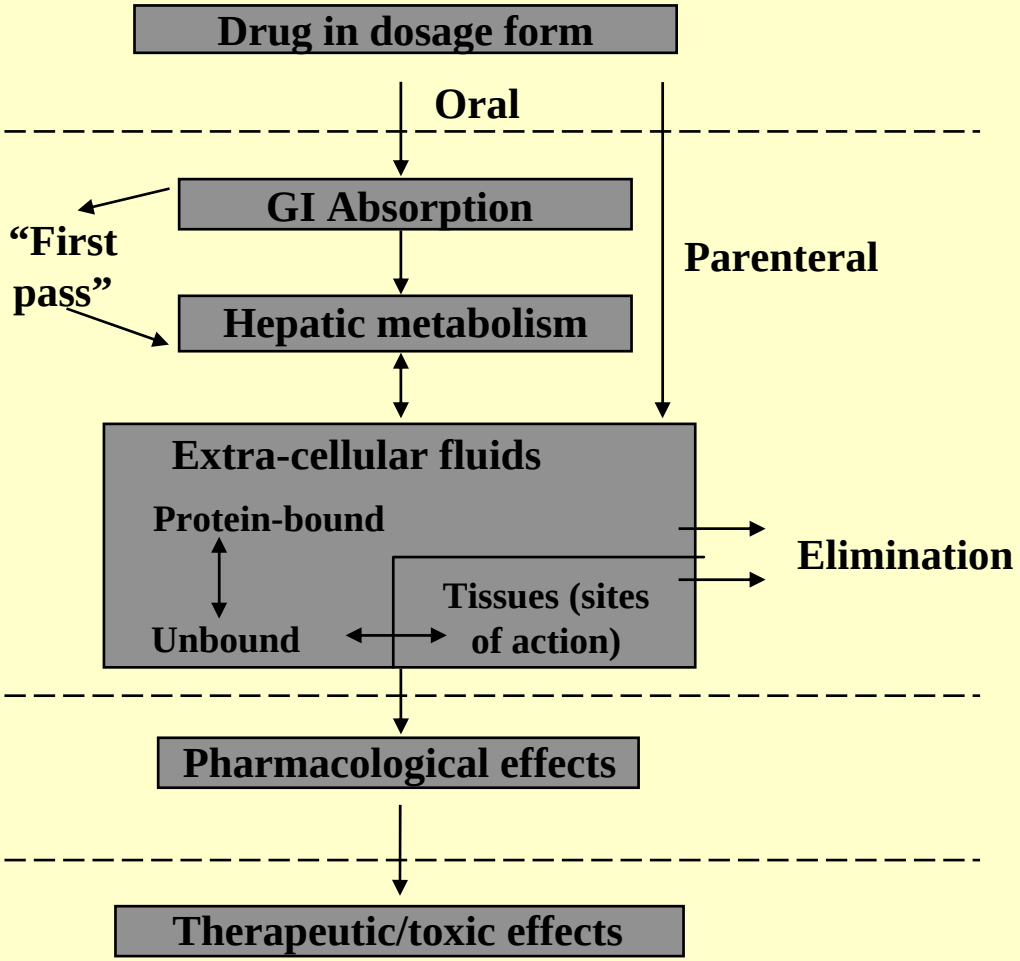
(taken from the Oxford Textbook of Clinical Pharmacology and Drug Therapy)

PHARMACEUTICAL PROCESS
‘Is the drug getting into the patient?’

PHARMACOKINETIC PROCESS
‘Is the drug getting to its site of action?’

PHARMACODYNAMIC PROCESS
‘Is the drug producing the required pharmacological effect?’

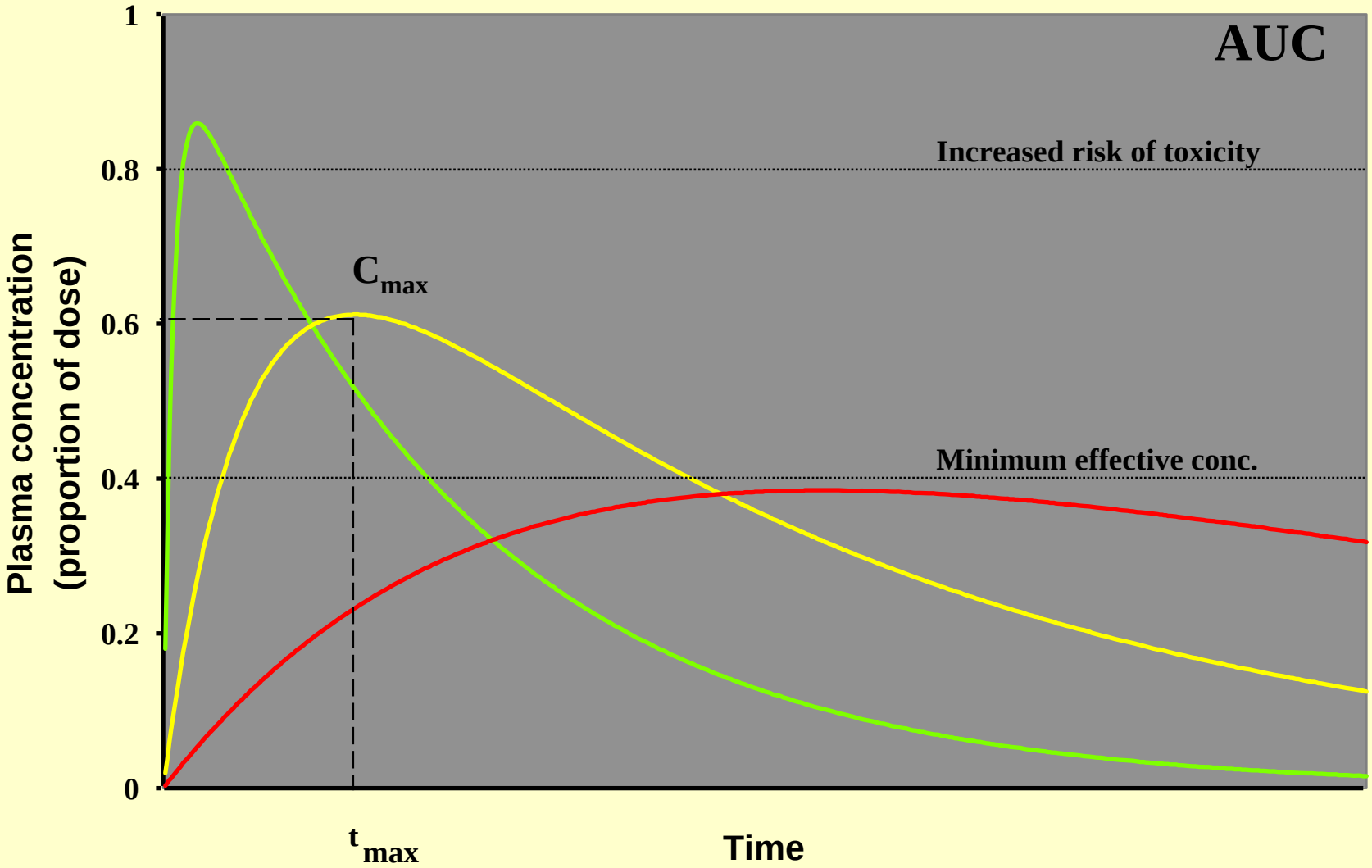
THERAPEUTIC PROCESS
‘Is the pharmacological effect being translated into a therapeutic effect?’



Pharmacokinetics

- Bioavaliability
  - Absorption
    - General principles
    - Specific clinically relevant examples
  - Distribution
  - Metabolism
  - Elimination

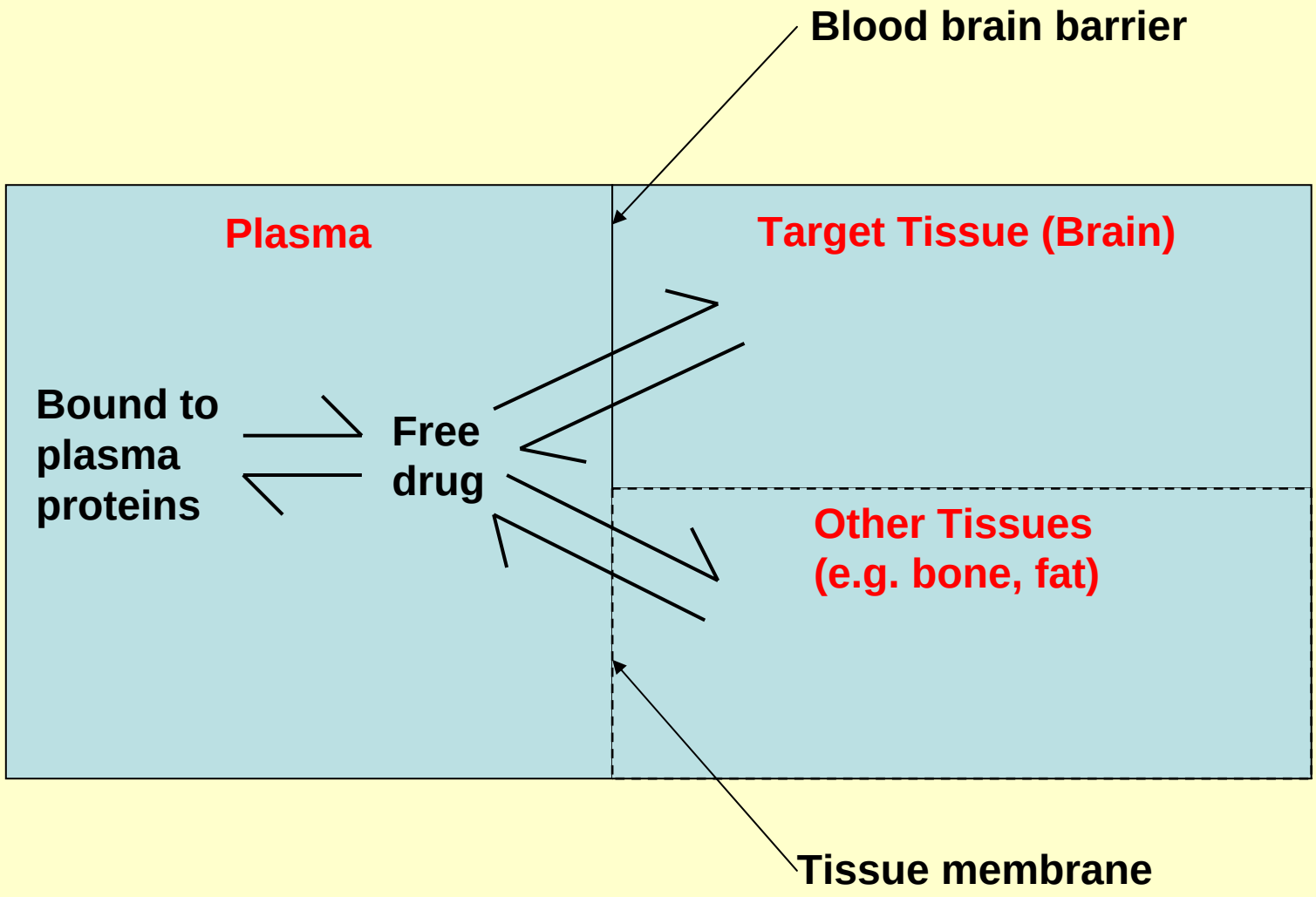
Theoretical plasma concentrations of three drugs with different rates of absorption



Absorption of TCAs

- t_{max}
  - tertiary amines: 1 - 3 hours
  - secondary amines: 4 - 8 hours
- Clinical relevance:
  - shorter t_{max} leads to higher C_{max}
  - most side effects (e.g. sedation, postural hypotension, membrane stabilisation) are dependent on the plasma concentration
  - therefore give sedative TCA all in one dose at night (and postural hypotension occurs while lying down!)
  - secondary amines often associated with fewer side effects

Distribution



Distribution

- Factors influencing distribution
  - Plasma protein biding
  - Tissue perfusion
  - Permeability of tissue membranes
  - Active transport out of tissues (p-glycoproteins)
- Volume of distribution calculated from quantity of drug and plasma concentration
  - $V_d = Q/C_p$
  - V_d is high if drug has high affinity for tissues (e.g. is fat soluble and hence much is in brain and fat)

Blood Brain Barrier

- Consequence of the special nature of capillaries in the brain and only allows lipid soluble molecules into the brain
  - Non-lipid soluble drugs require transport systems which can be active (e.g. L-tryptophan, L-dopa) or passive (e.g. Li)
  - P-glycoproteins in endothelial membranes pump drugs out of capillary cells and prevent some drugs getting into the brain
  - Areas of the brain not protected:
    - Median eminence of the hypothalamus
    - Vomiting centre

Metabolism of drugs

- Occurs mainly in the liver by P450 isoenzymes
- ‘First pass’ metabolism reduces the amount of drug reaching the systemic circulation (bioavailability)
- Two ‘types’ of metabolism:
  - Type 1 - metabolic modification
    - e.g. oxidation, reduction, hydrolysis
    - metabolites often have pharmacological activity
  - Type 2 - conjugation
    - e.g. with glucoronic acid, glycine, sulphate
    - metabolite water soluble and inactive

Metabolism of TCAs

- Extensive ‘first pass’ metabolism (40-50%)
  - decreased by:
    - primary liver disease
    - impaired right ventricular function
    - increased age (over 60s twice average plasma concentrations)
    - acute ingestion of alcohol
    - neuroleptics, SSRIs (see below)
  - increased by (hepatic induction):
    - subchronic alcohol
    - carbamazepine

Metabolism of TCAs

- Type 1 metabolism converts tertiary to secondary amines, eg.
  - Amitriptyline → Nortiptyline
  - Imipramine → Desipramine
  - Clomipramine → Desmethylclomipramine
- Tertiary amines generally more potent 5-HT uptake blockers, secondary amines more potent NA uptake blockers
  - Up to 70% of clomipramine may be converted to desmethylclomipramine
    - may lead to lack of efficacy in OCD

Metabolism of SSRIs - 2
P450 isoenzyme inhibition

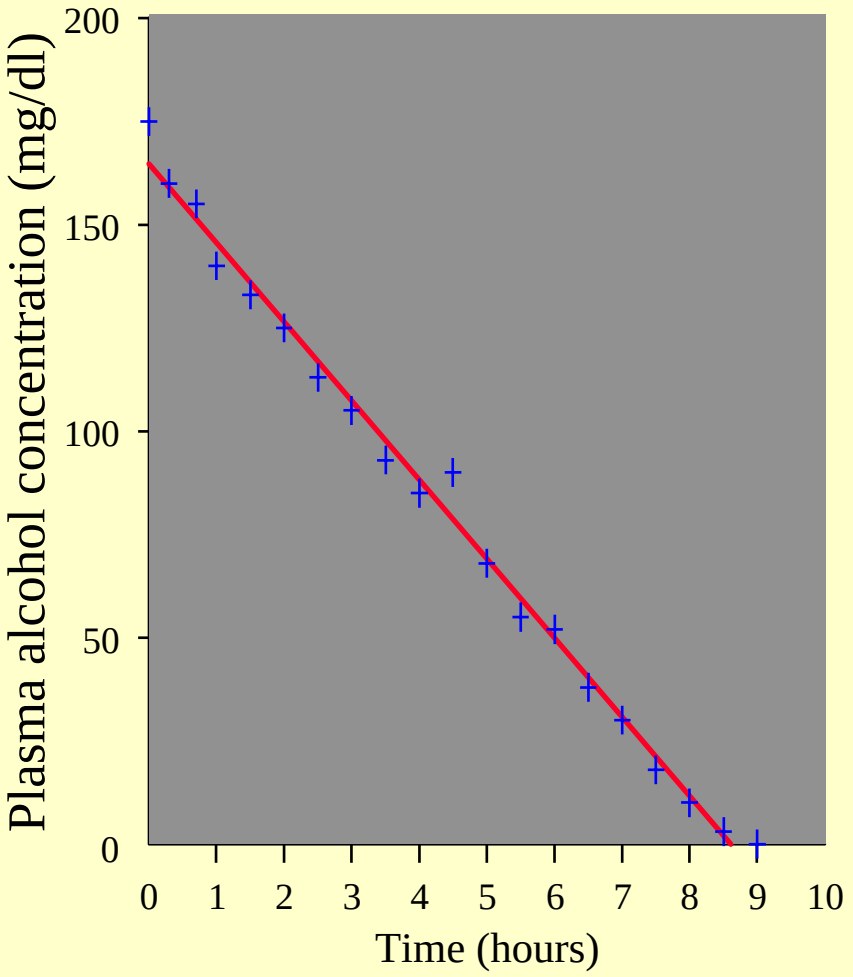
	1A2 inhib.	2D6 inhib.
Fluoxetine	+	+++
Fluvoxamine	+++	+
Paroxetine	+	+++
Sertraline	+	+
Citalopram	+	+

- 1A2 inhibition leads to increased levels of
  - caffeine, clozapine, theophyline
- 2D6 inhibition leads to increased levels of
  - β-blockers, neuroleptics, TCAs, opiates

Elimination of drugs

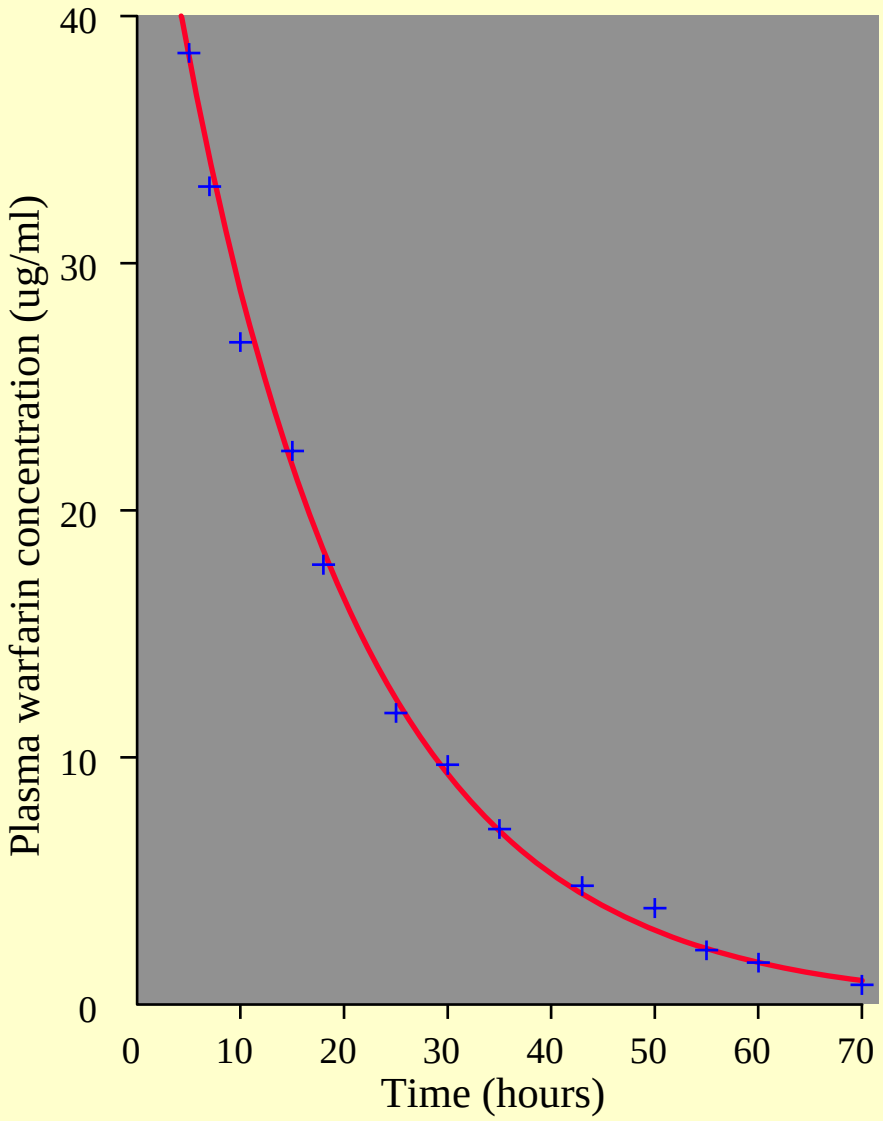
- Primarily via the kidney
  - Metabolism of drug usually has to occur first to produce a water soluble compound
  - This is usually the rate limiting step
  - Factors slowing metabolism will increase the elimination time
- Kinetics
  - Usually ‘first order’
  - Influences the dosing schedule
  - Influences the possibility of withdrawal problems

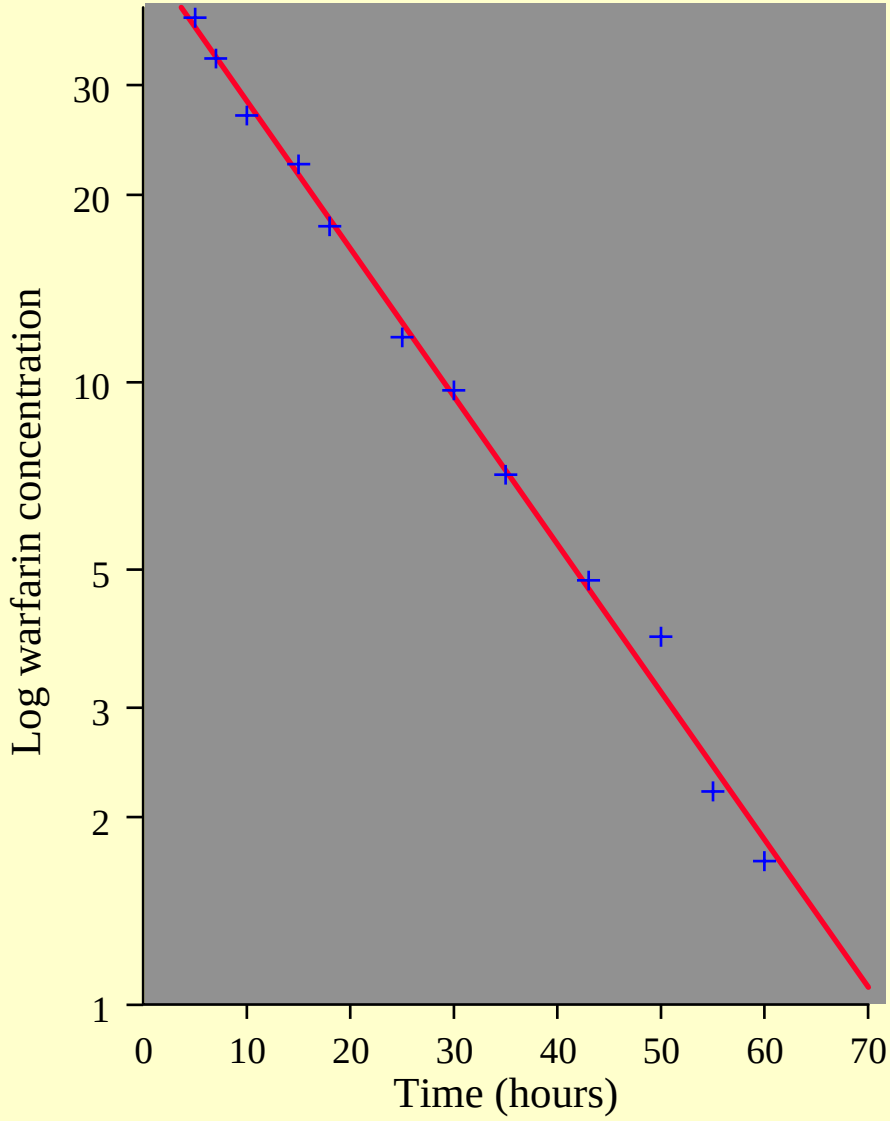
Zero order kinetics



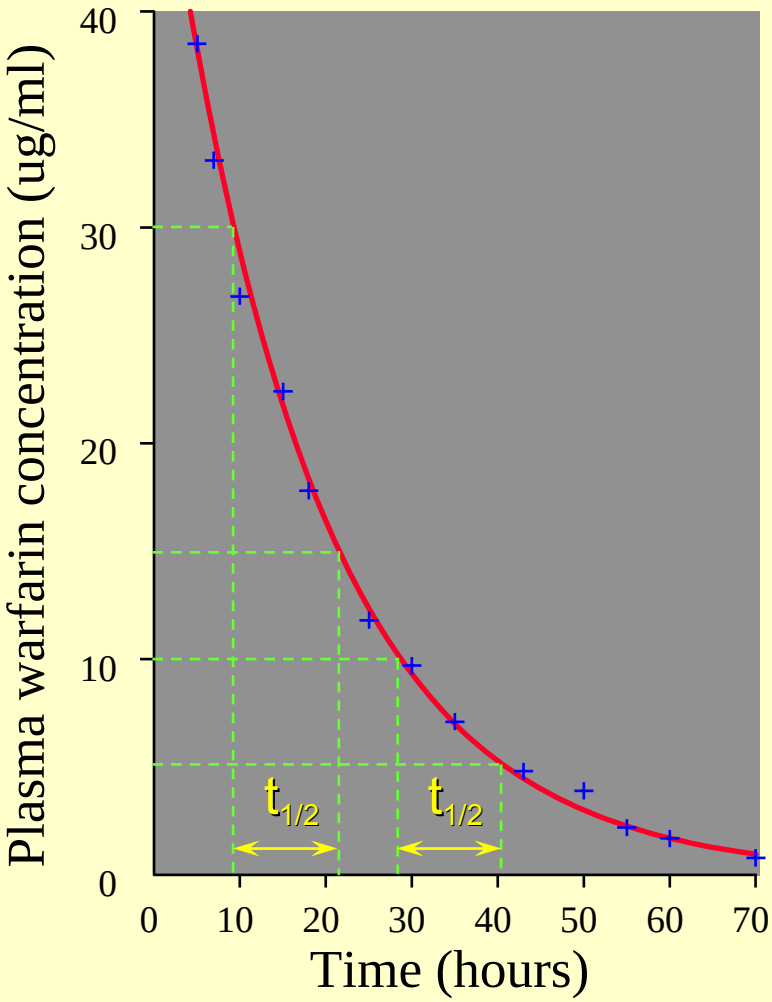
- The rate of elimination is <u>independent</u> of plasma concentration
- A small change in dose can produce a big change in plasma concentration
- Rare except if elimination process is saturated (can occur with TCAs)

Plasma concentration of warfarin following bolus i.v. infusion



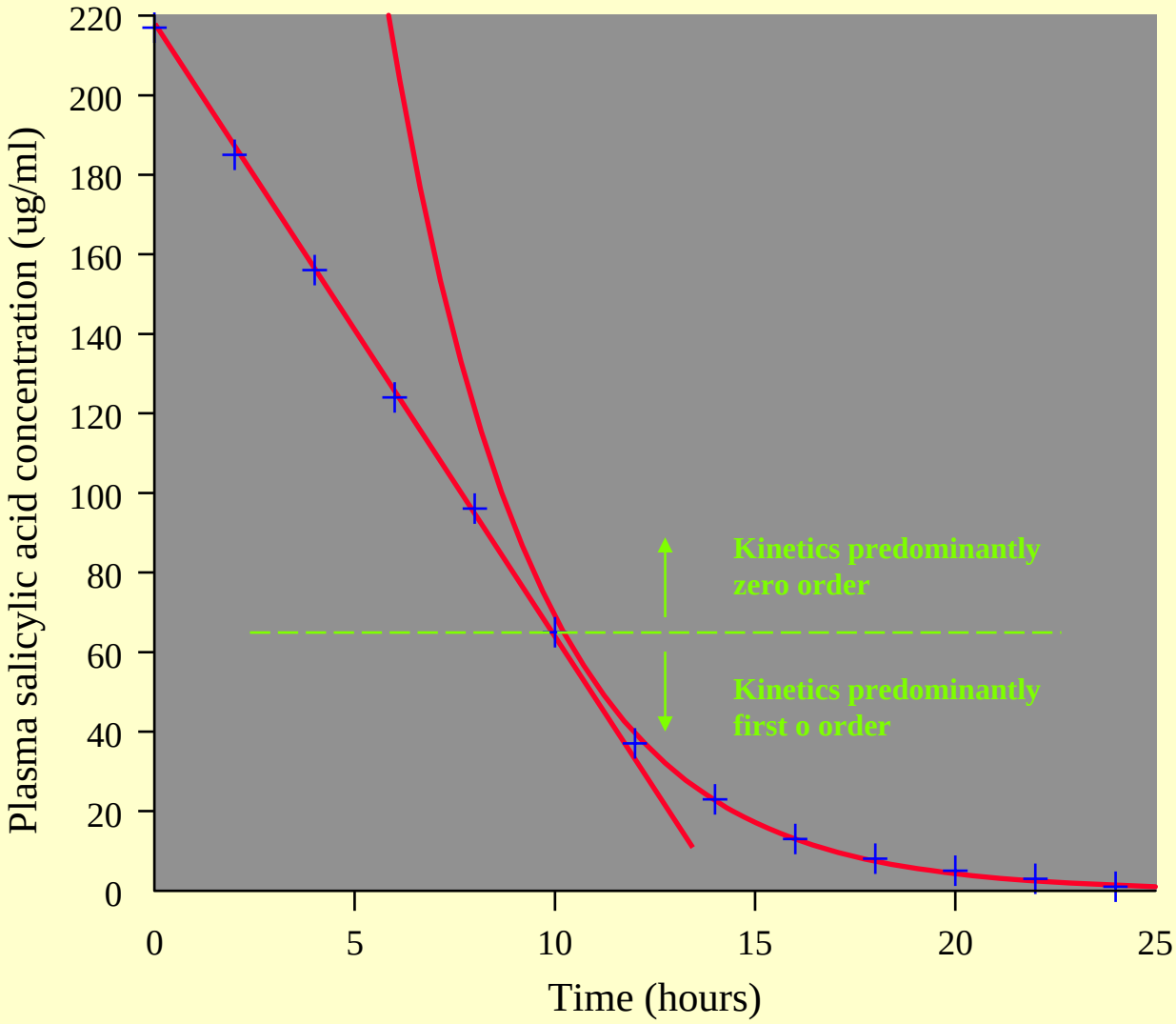


First order kinetics



- The rate of elimination is <u>proportional</u> to the plasma concentration
- Elimination rate quantified by ‘half life’
- The majority of drugs have first order kinetics

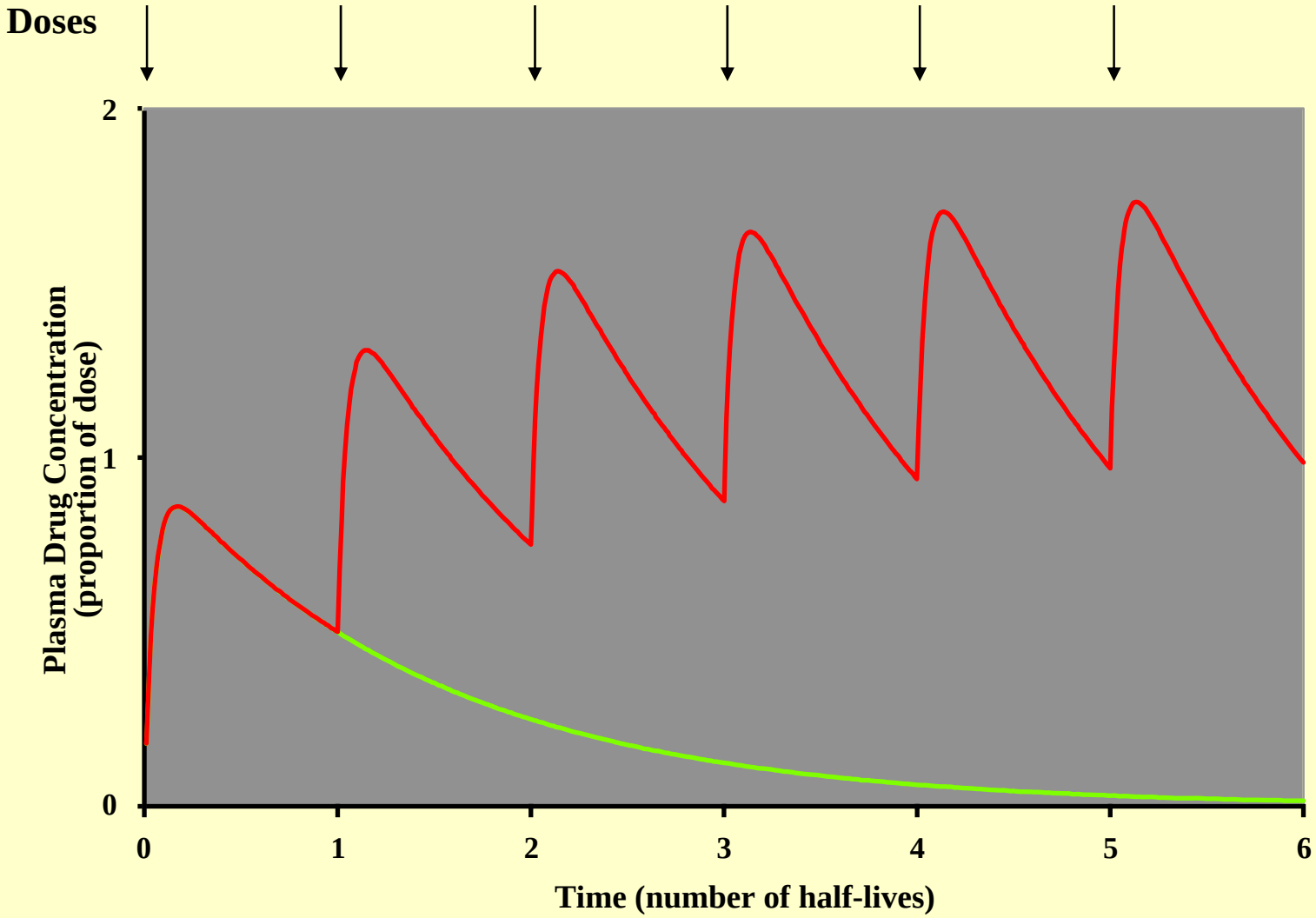
Plasma concentration of salicylic acid following bolus i.v. infusion



Drug kinetics

- The variation of plasma concentration over time
  - zero-order:
    - concentration falls at a constant rate
  - first-order:
    - concentration falls at a rate proportional to the concentration

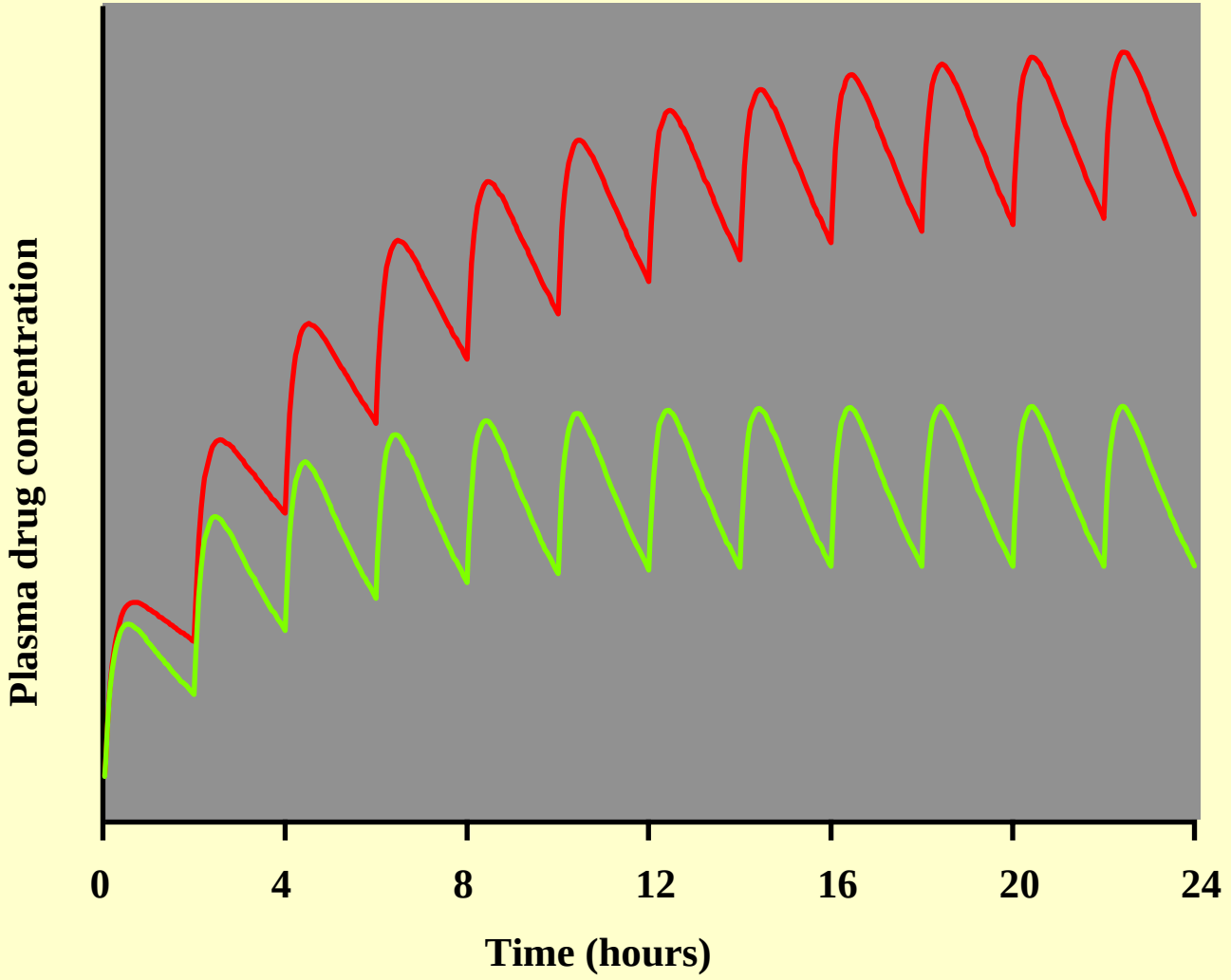
Theoretical plasma concentration of a first order drug after single or repeated doses



Elimination / accumulation of first-order drugs

No. of half lifes	% Eliminated/accumulated	
1	50	=50%
2	50+25	=75%
3	50+25+12.5	=87.5%
4	50+25+12.5+6.25	=93.75%
5	50+25+12.5+6.25+3.125	=96.9%

Effect of reduced metabolism of a drug on its steady state concentration



Half lives of TCAs

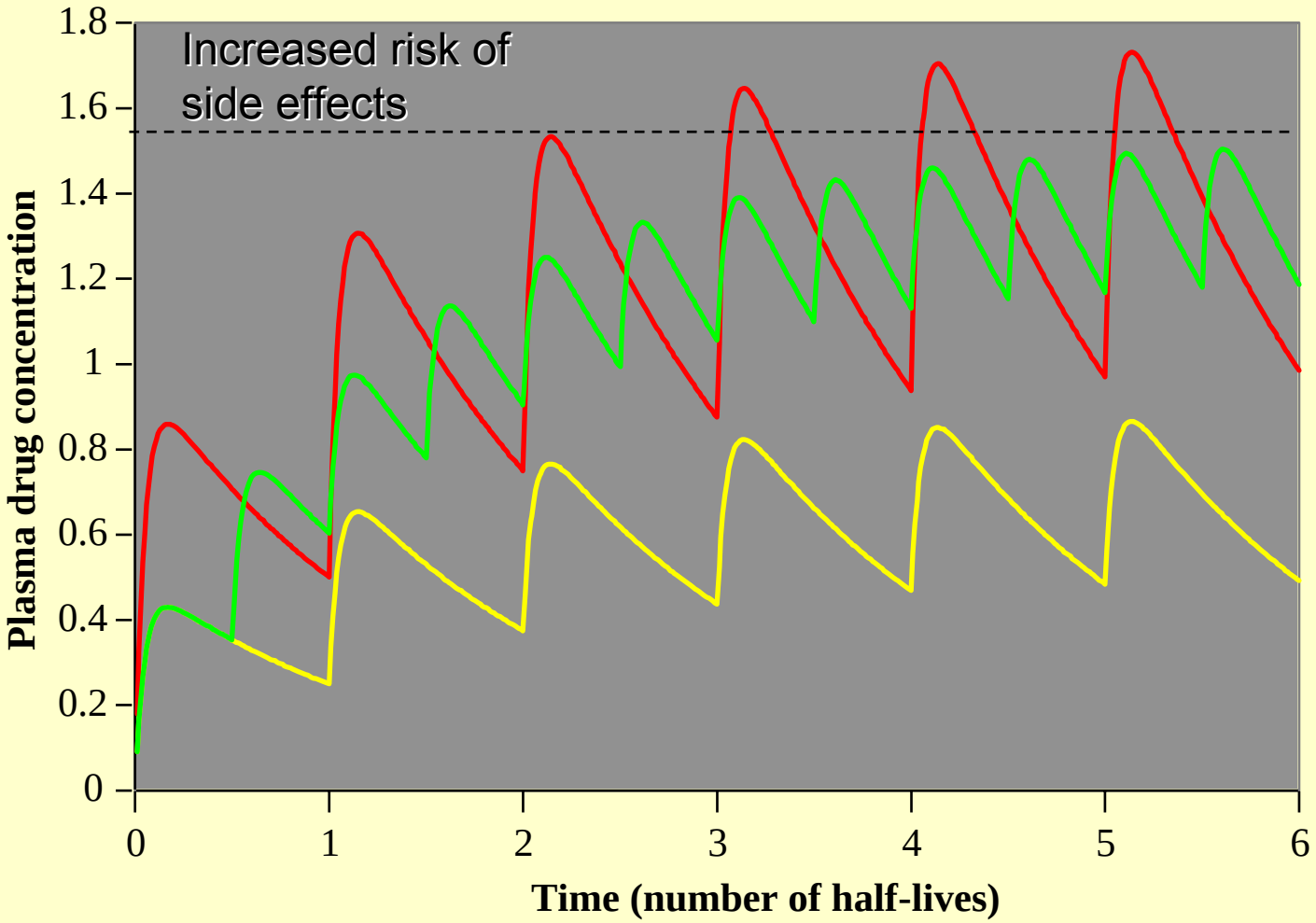
	Half Life (hours - approx)	Metabolite
Amitriptyline	16	Nortriptyline
Imipramine	12	Desipramine
Clomipramine	18	DMC
Nortriptyline	60	
Desipramine	50	
DMC	45	
Lofepramine	5	Desipramine

"…prescribing phenothiazines and tricyclic antidepressants three times a day is simply a public display of pharmacological ignorance…"

R.E. Kendell (1993)

Companion to Psychiatric Studies, 5th Ed. p 419

Effect of varying dose and frequency of administration of a first order drug



Half dose, twice as often

Control

Half dose, same freq.

Half lives of SSRIs - 1

	Half life (hrs) (Active metab.)
Fluoxetine	45-72 (150-200)
Sertraline	25 (66)
Citalopram	36 (?)
Paroxetine	10-20
Fluvoxamine	15

- Note inter-drug and -individual variation
- Fluoxetine and paroxetine
  - $t_{1/2}$ increases with dose and time
- Paroxetine and citalopram
  - $t_{1/2}$ increases with age
- Fluvoxamine and sertraline
  - $t_{1/2}$ lower in men

Half lives of SSRIs - 2

Clinical Relevance

- Fluoxetine/norfluoxetine long half life consequences:
  - 5+ weeks to steady state
  - late emergence of plasma level dependent side effects
  - prolonged washout period
    - N.B. delayed CYP2D6 inhibition
  - benefit for poor compliers
  - little risk of discontinuation syndrome

Half lives of SSRIs - 3
Discontinuation syndrome

- Seen with TCAs, MAOIs and others
- SSRI discontinuation syndrome
  - does not imply dependence
  - starts within few days of stopping treatment
  - usually resolves spontaneously within 3 weeks
  - characteristic symptoms
    - dizziness, nausea, lethargy, headaches, paraesthesia
  - SSRI reinstatement leads to rapid resolution
  - Most common with paroxetine, least with fluoxetine
    - thought to be related to differences in half life
      - N.B. paroxetine also anti-cholinergic

Pharmacokinetics Summary

- A knowledge of the pharmacokinetics of antidepressants can improve their clinical usage e.g. by:
  - minimising side effects associated with Cmax
    - split dosages
    - choice of drug (secondary versus tertiary TCA)
  - adjusting dosages appropriately for age and sex
  - avoiding pharmacokinetic interactions
  - being aware of discontinuation phenomena
  - considering therapeutic monitoring if indicated

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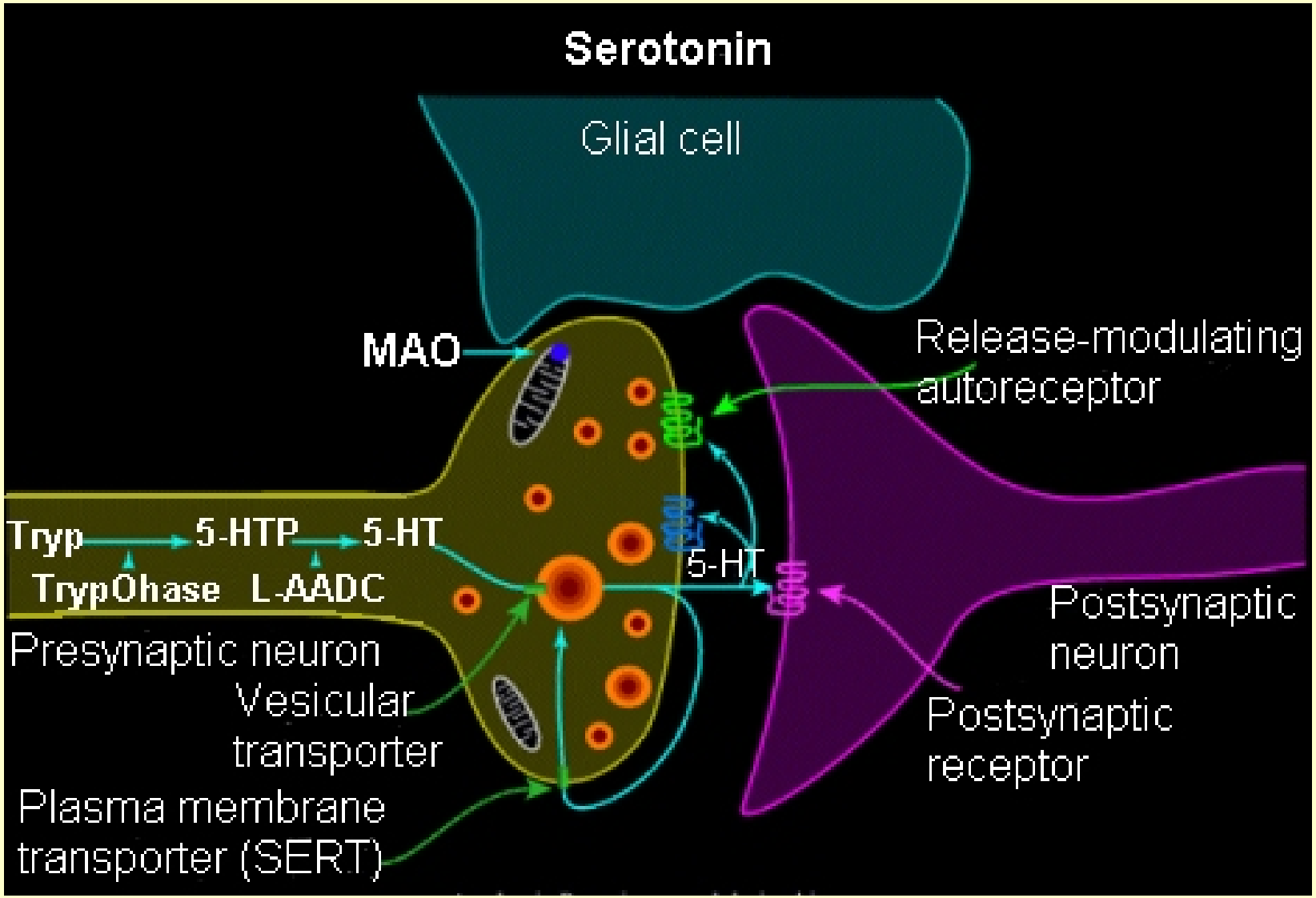
Psychobiology Research Group

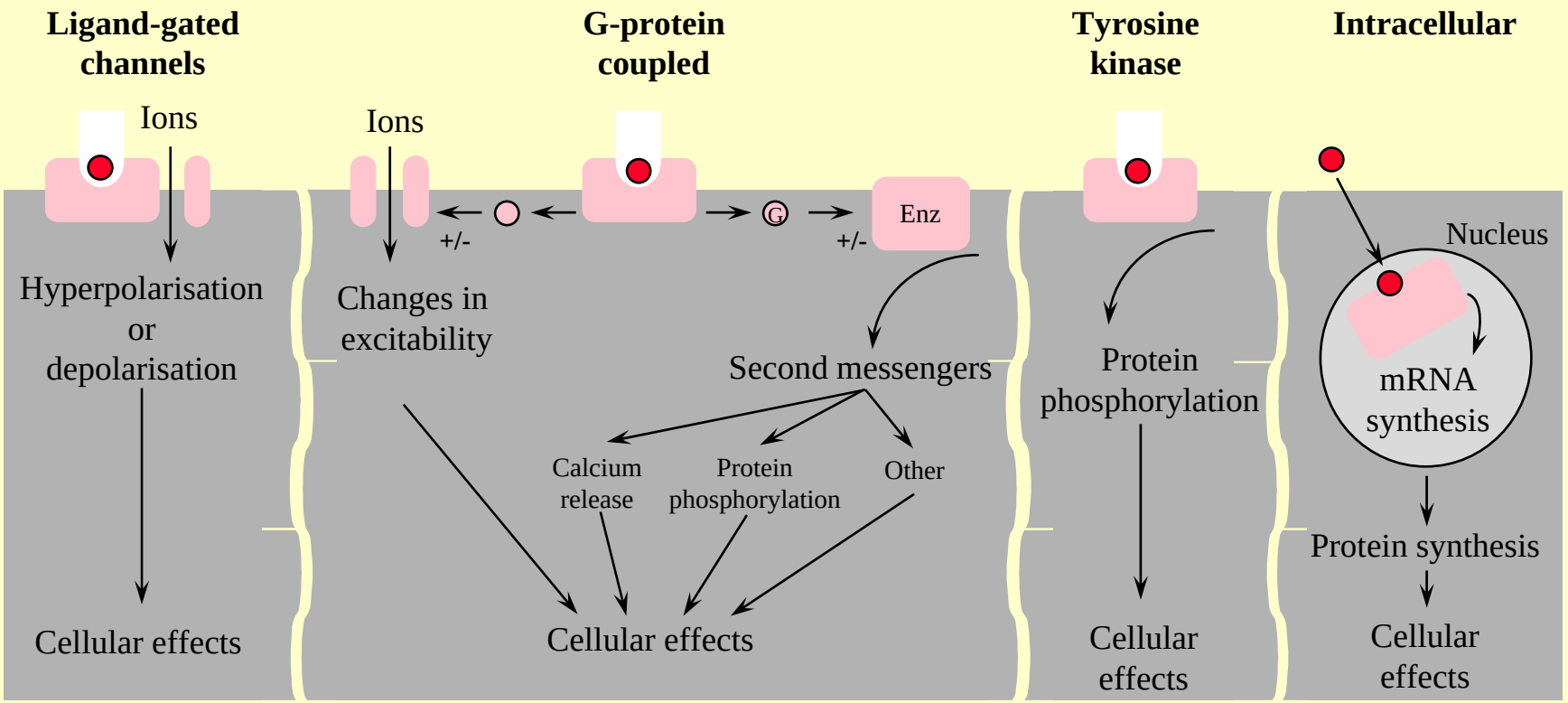
Pharmacodynamics

Neurotransmission
How drugs act
Potency
Dose-response relationships

5-HT neurotransmission



What happens when a receptor is activated ?



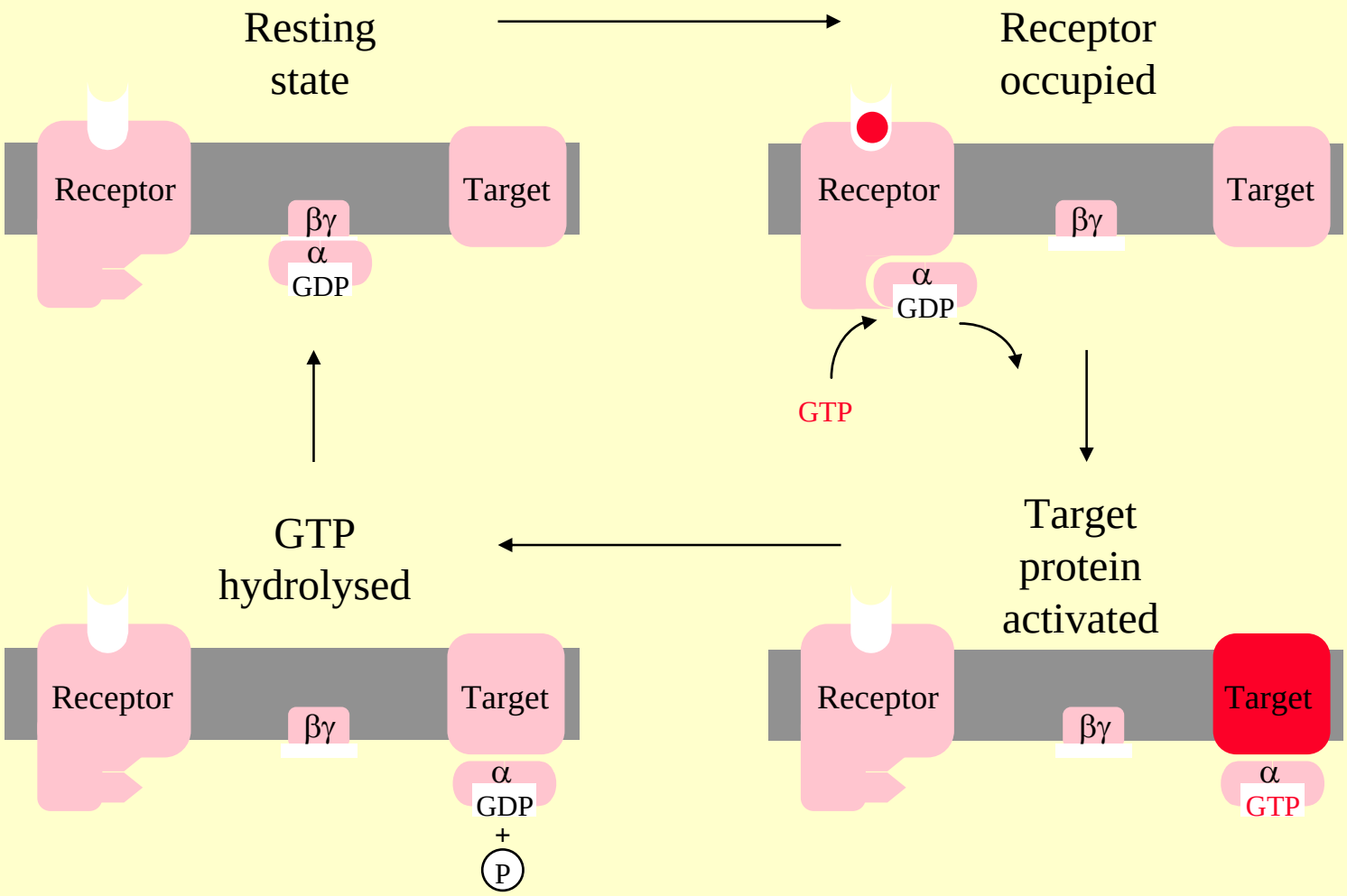
Time Scale

milliseconds	seconds	minutes	hours

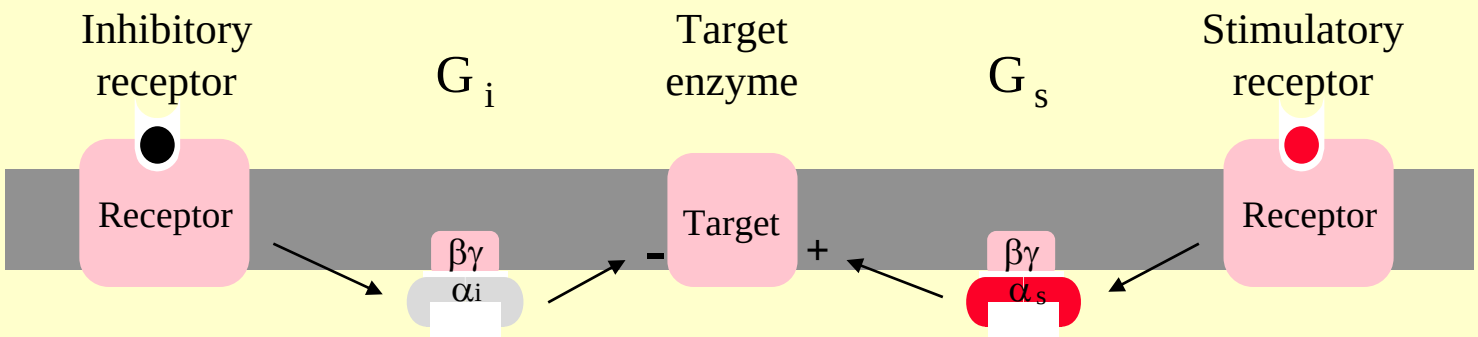
Examples

nACh $GABA_A$ NMDA	mACh $GABA_B$ $5\text{-}HT_{1A}$	Insulin	Glucocorticoid Mineralocorticoid

Function of G-proteins



Bidirectional Control of Adenylate Cyclase by G_s and G_i



How do drugs act?

Molecular interaction of the drug

↓

Cell and tissue biochemistry manipulated

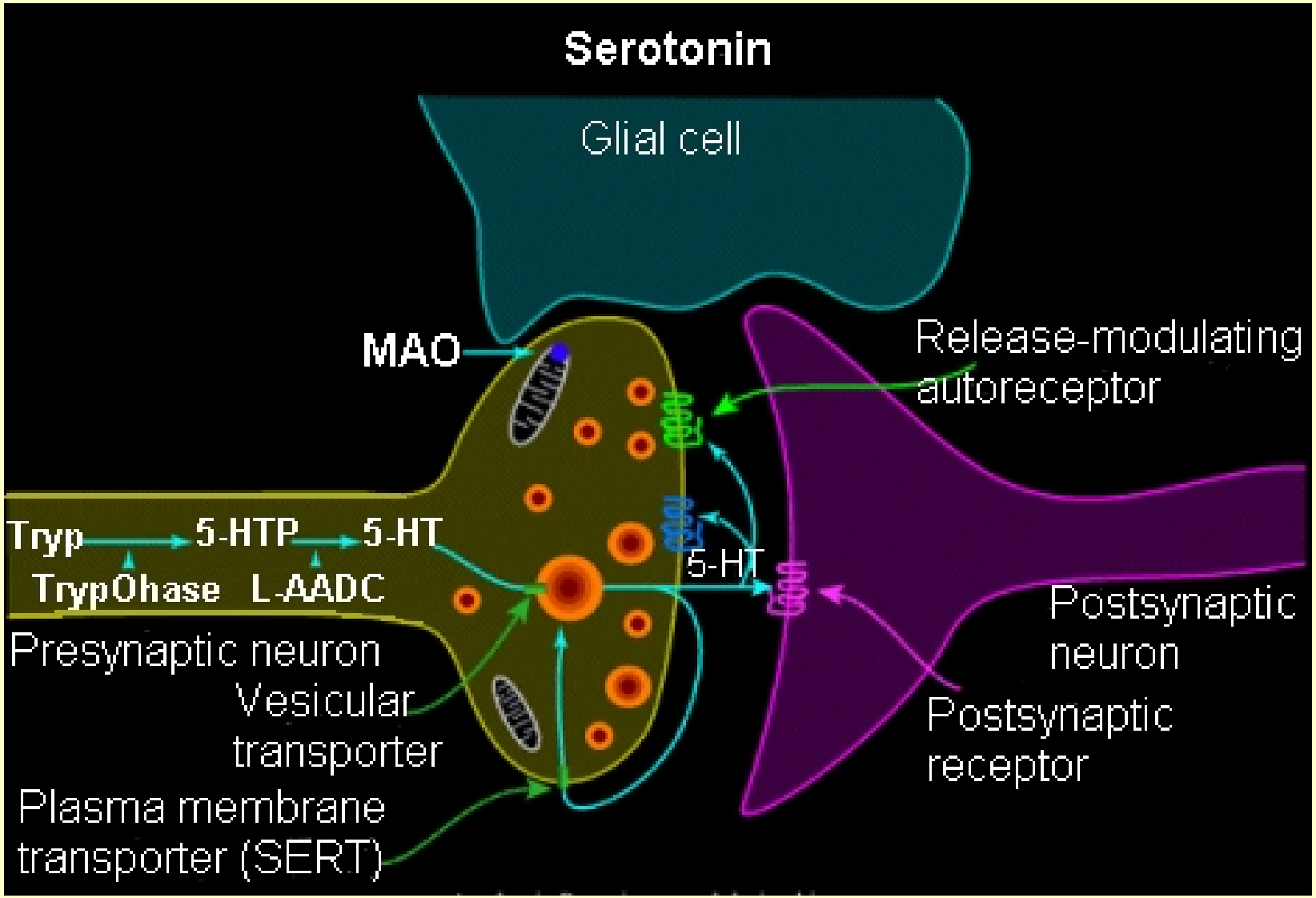
↓

Organ physiological function manipulated

↓

Therapeutic benefit to patient

5-HT neurotransmission

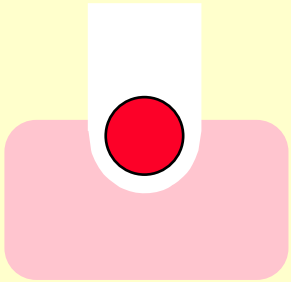


Targets for drug action

1. Synthesis (e.g. l-tryptophan)
2. Storage (e.g. reserpine)
3. Release (e.g. amphetamine)
4. Receptors (e.g. mirtazepine)

Targets for drug action
1. Receptors

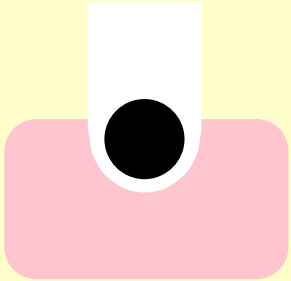
Agonist



Direct → Ion channel opening

Transduction mechanisms
- Enzyme activation/inhibition
- Ion channel modulation
- DNA transcription

Antagonist



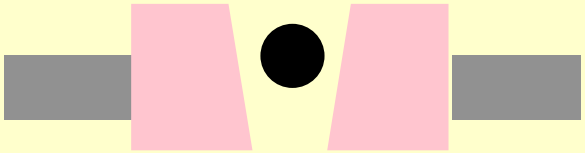
→ No effect - Endogenous mediator blocked

Targets for drug action

1. Synthesis (e.g. l-tryptophan)
2. Storage (e.g. reserpine)
3. Release (e.g. amphetamine)
4. Receptors (e.g. mirtazepine)
5. Ion channels (e.g. verapamil)

Targets for drug action
2. Ion Channels

Blockers



Ion flux blocked

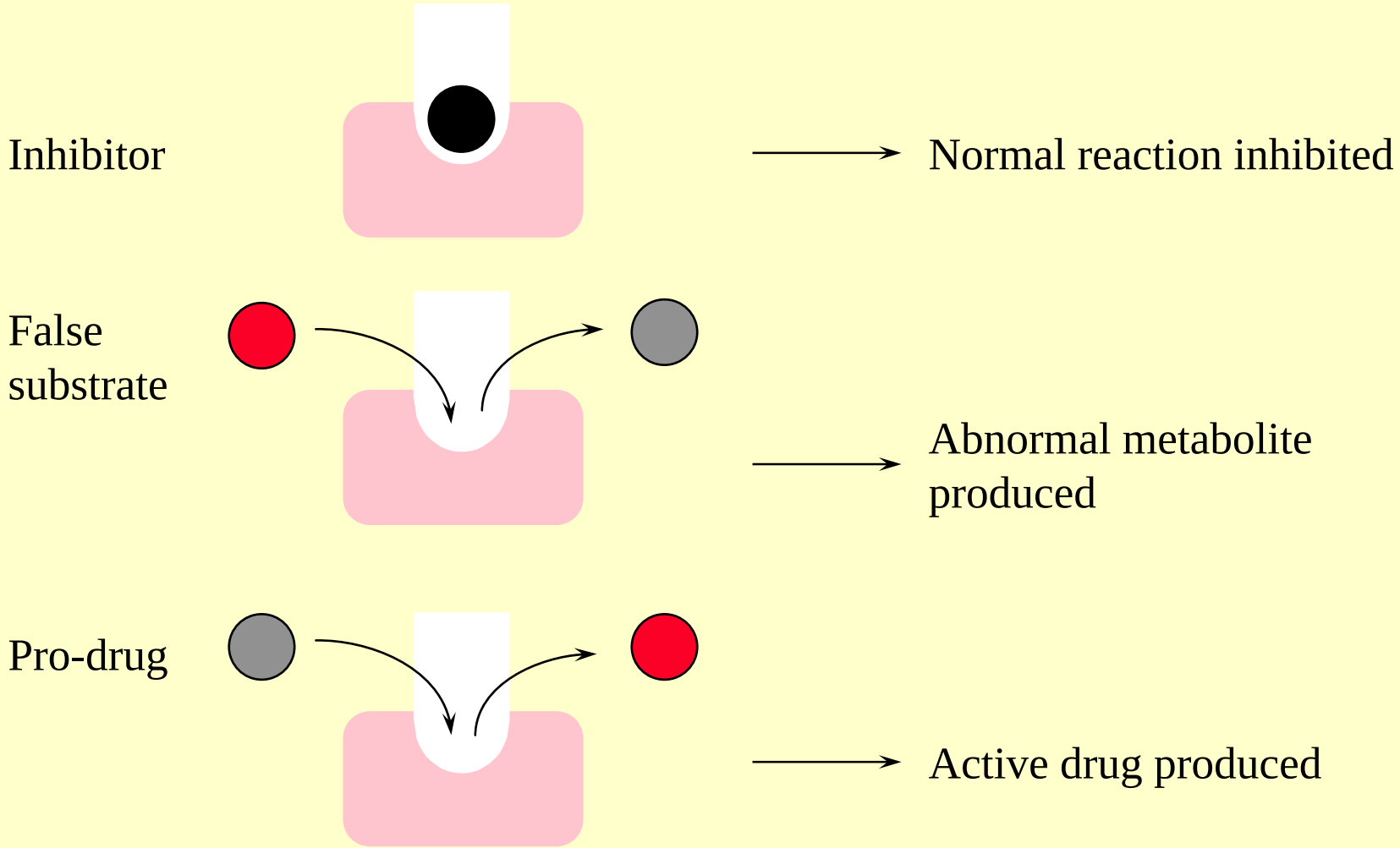
Modulators

Increased or decreased opening probability

Targets for drug action

1. Synthesis (e.g. l-tryptophan)
2. Storage (e.g. reserpine)
3. Release (e.g. amphetamine)
4. Receptors (e.g. mirtazepine)
5. Ion channels (e.g. verapamil)
6. Second messenger systems (e.g. lithium)
7. Re-uptake (e.g. SSRIs)

Targets for drug action
3. Enzymes

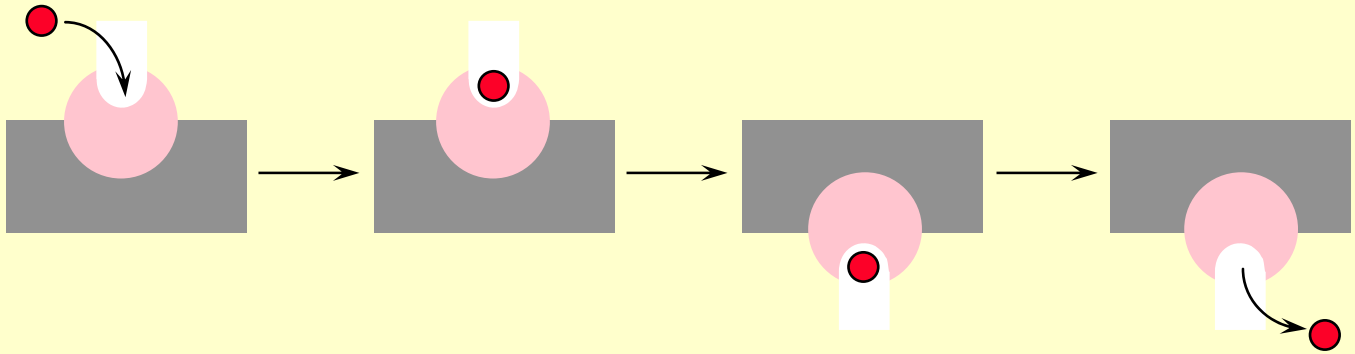


Targets for drug action

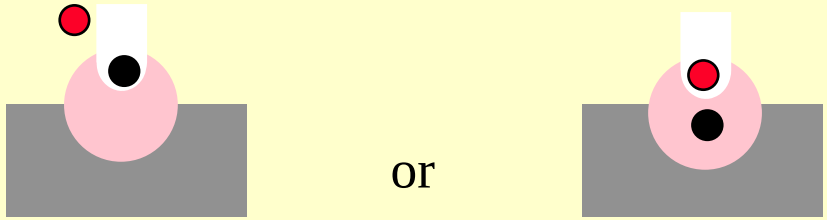
1. Synthesis (e.g. l-tryptophan)
2. Storage (e.g. reserpine)
3. Release (e.g. amphetamine)
4. Receptors (e.g. mirtazepine)
5. Ion channels (e.g. verapamil)
6. Second messenger systems (e.g. lithium)
7. Re-uptake (e.g. SSRIs)
8. Degradation (e.g. MAOIs)

Targets for drug action
4. Carriers

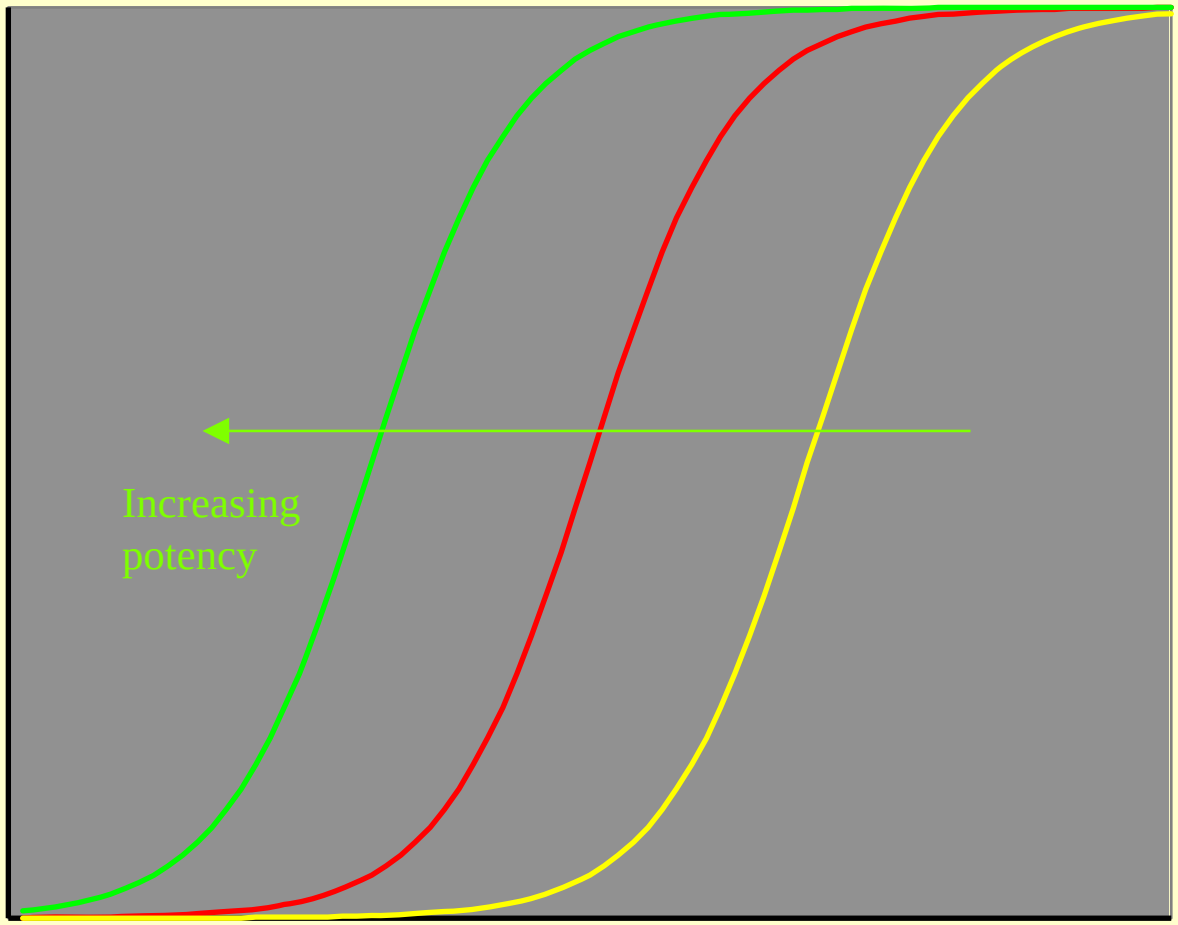
Normal transport



Inhibitor



Potency of receptor agonists



'Potency' of drugs acting at receptors

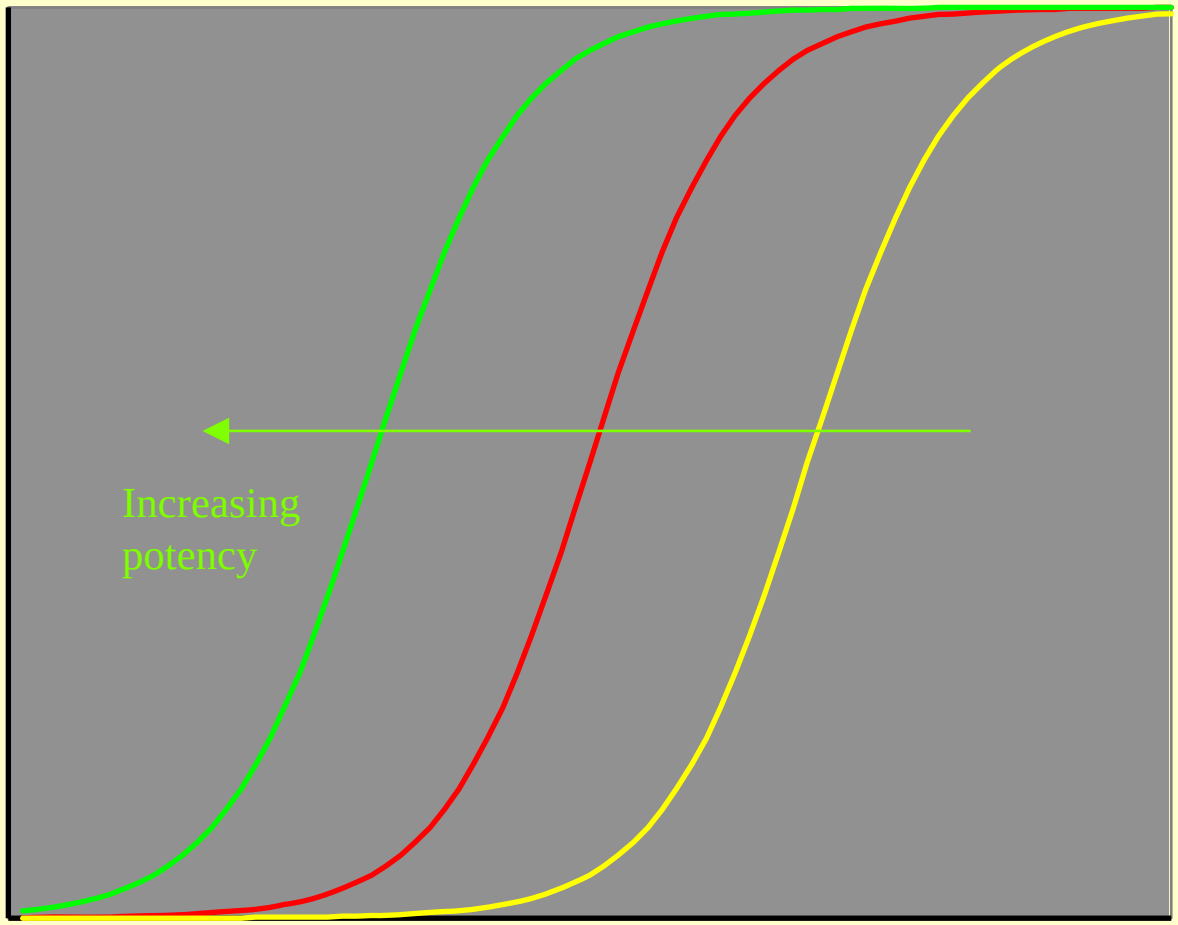
- Antagonist
  - depends solely on affinity for receptor
- Agonist
  - depends on combination of affinity and 'intrisic activity' or 'efficacy'

$$\text{Response} = f\left(\frac{\varepsilon\ N_{tot}\ x_A}{x_A + K_A}\right)$$

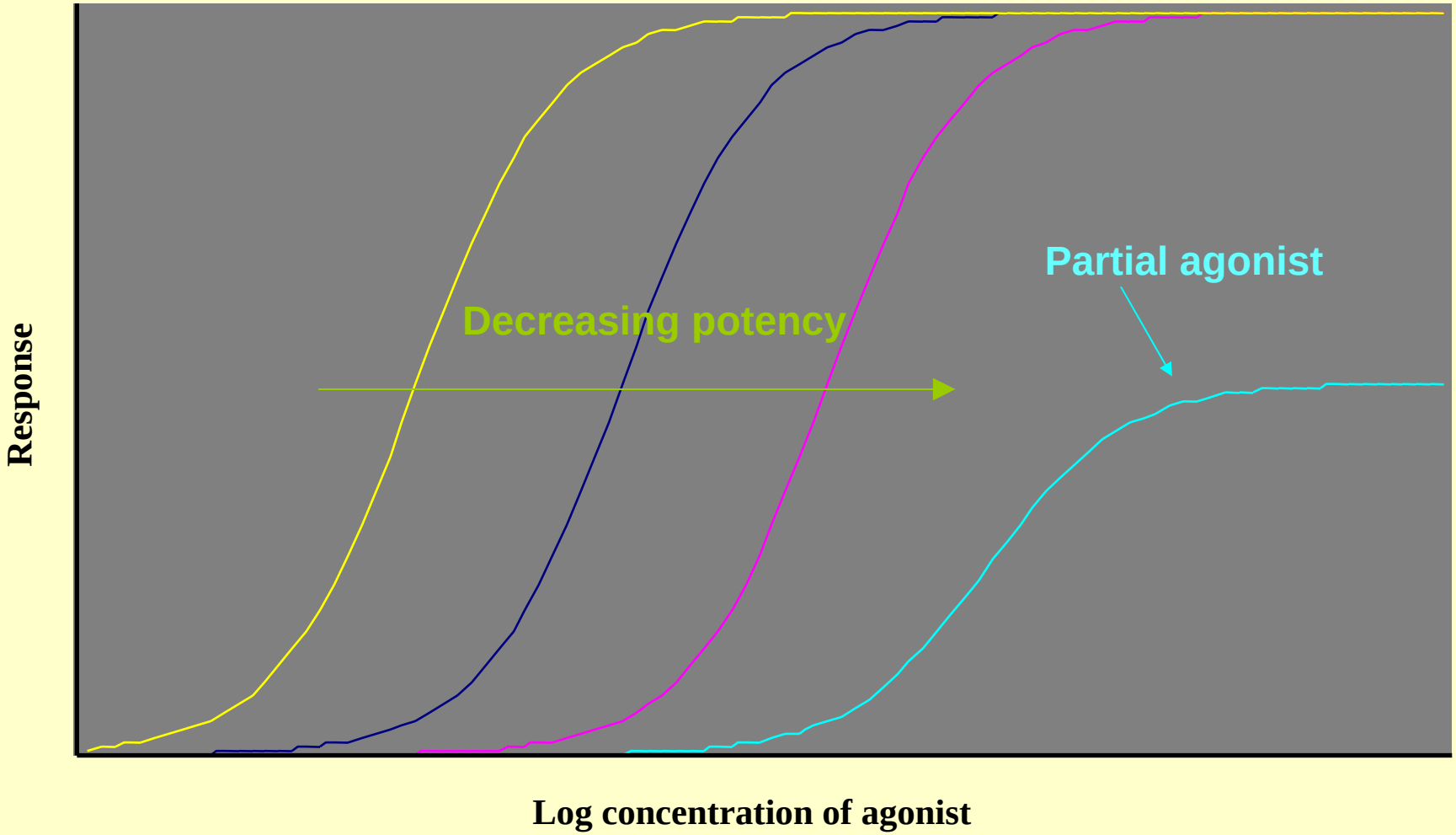
Red = property of drug

Green = property of tissue

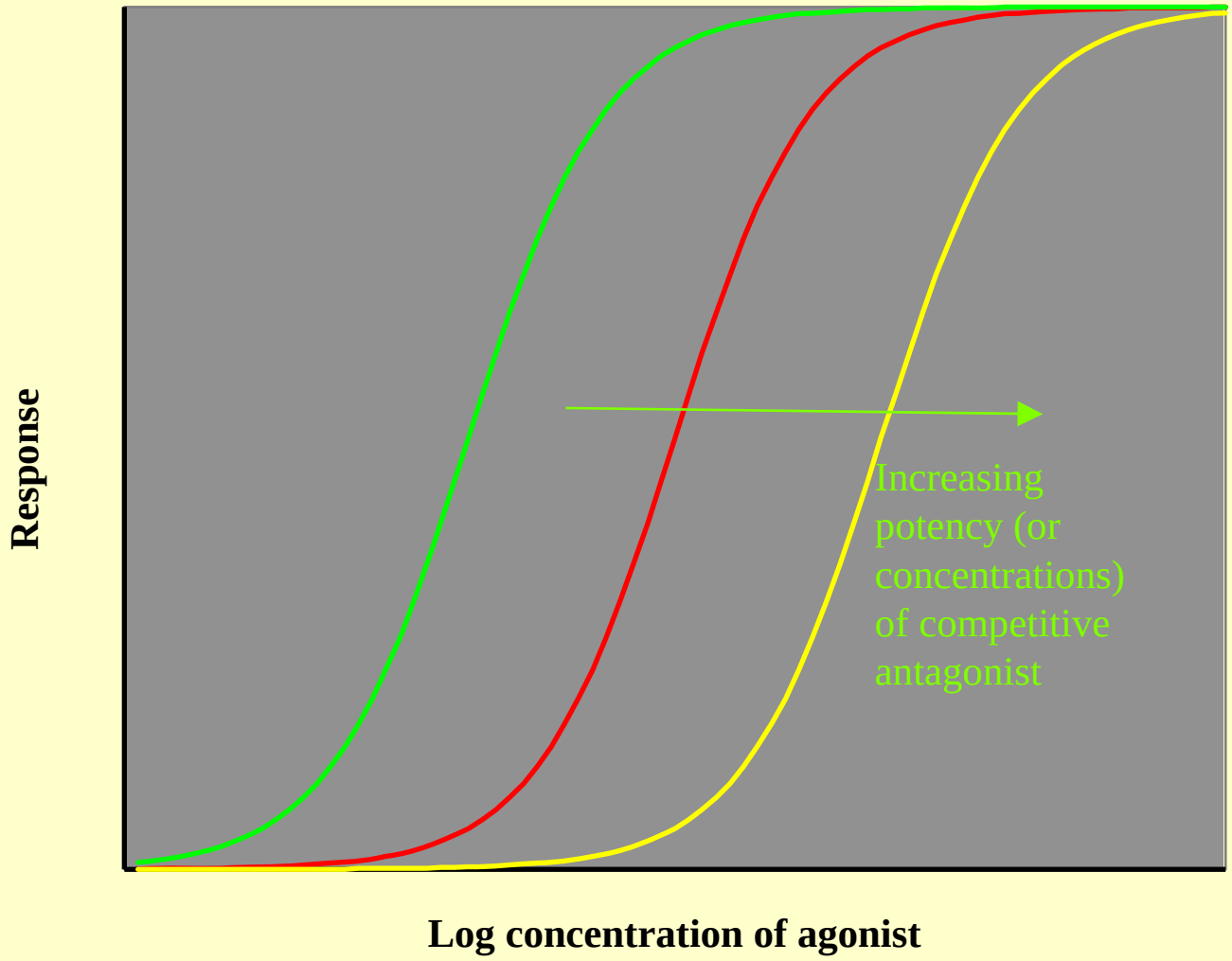
Potency of receptor agonists



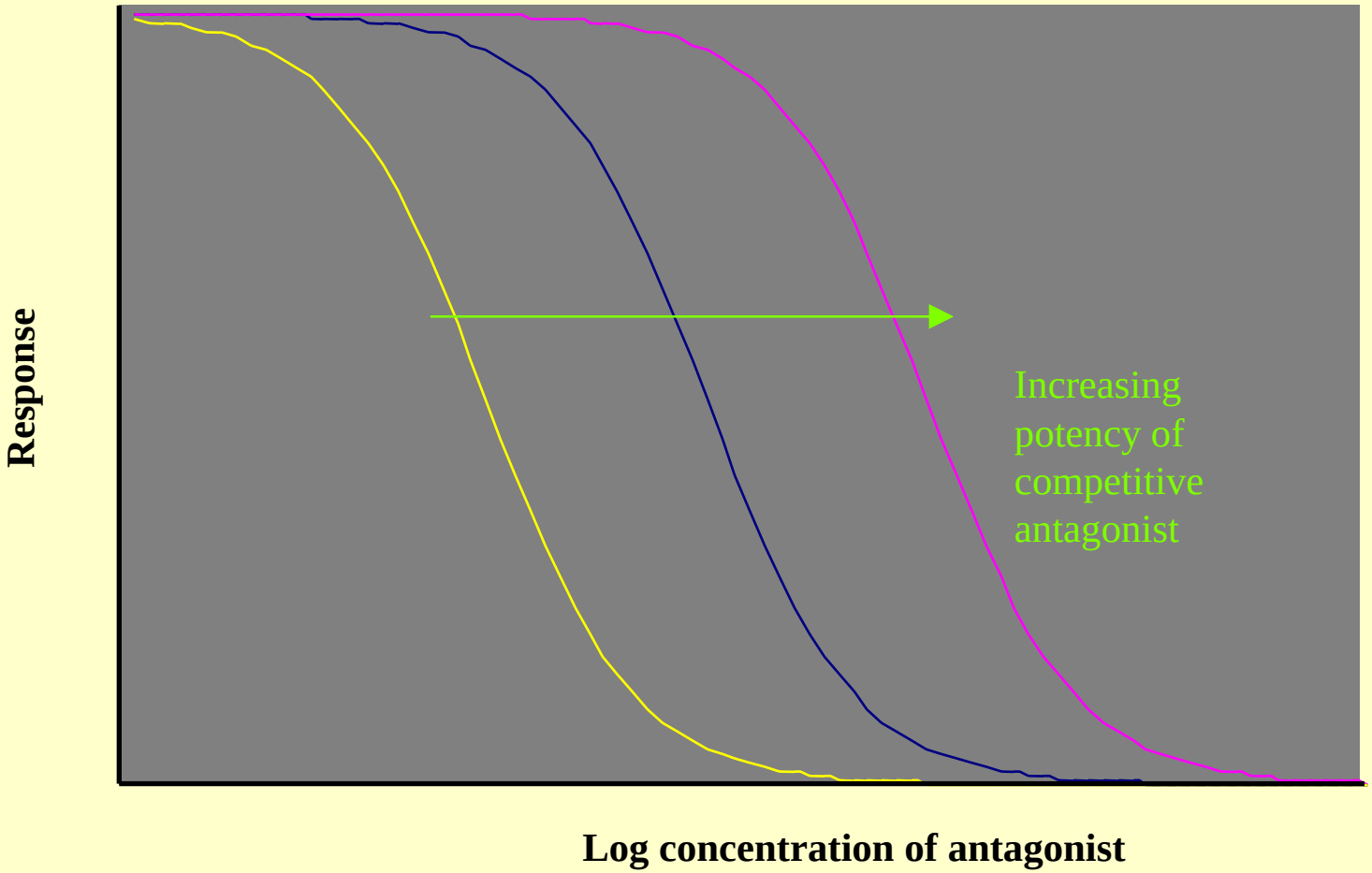
Partial agonists



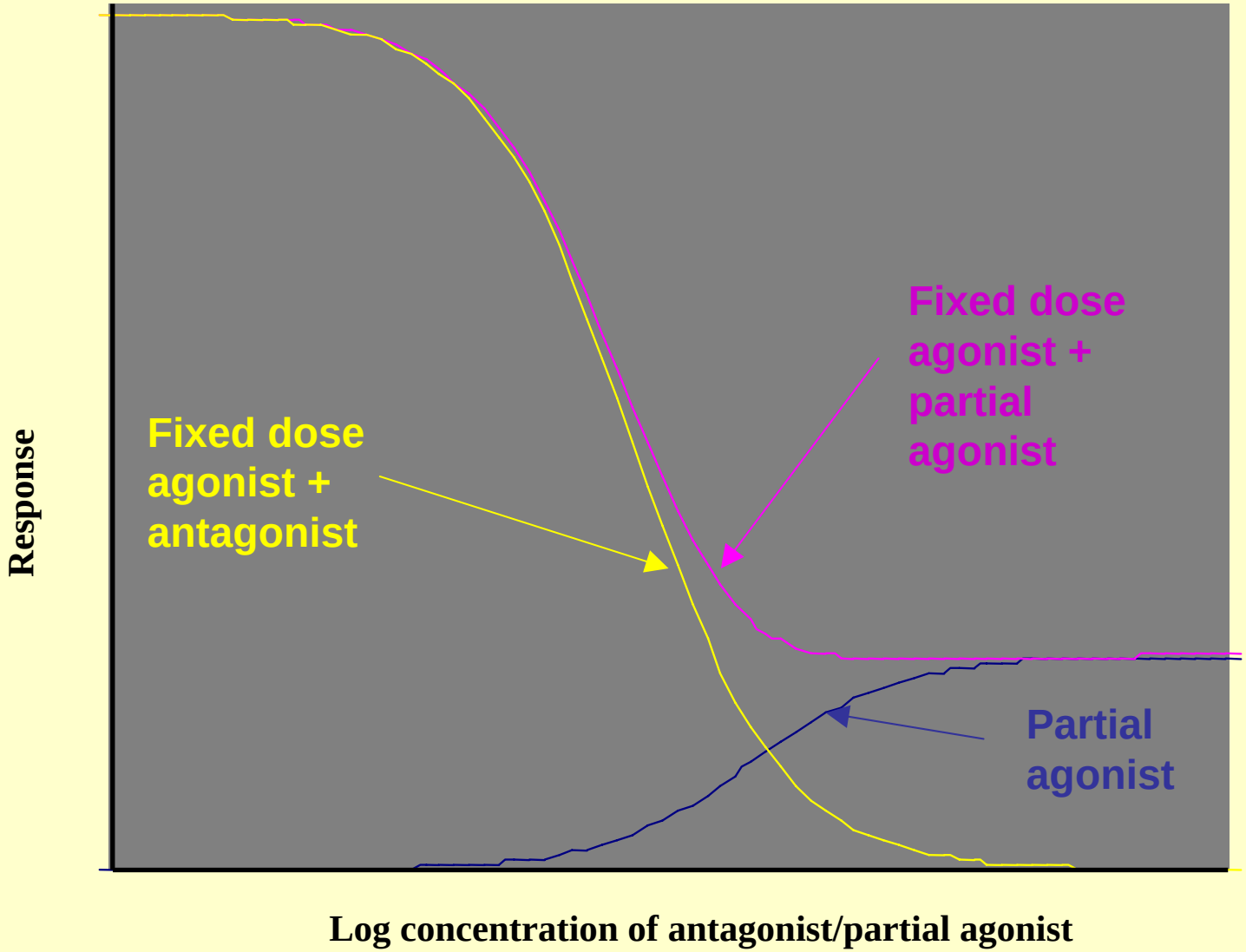
Potency of antagonists



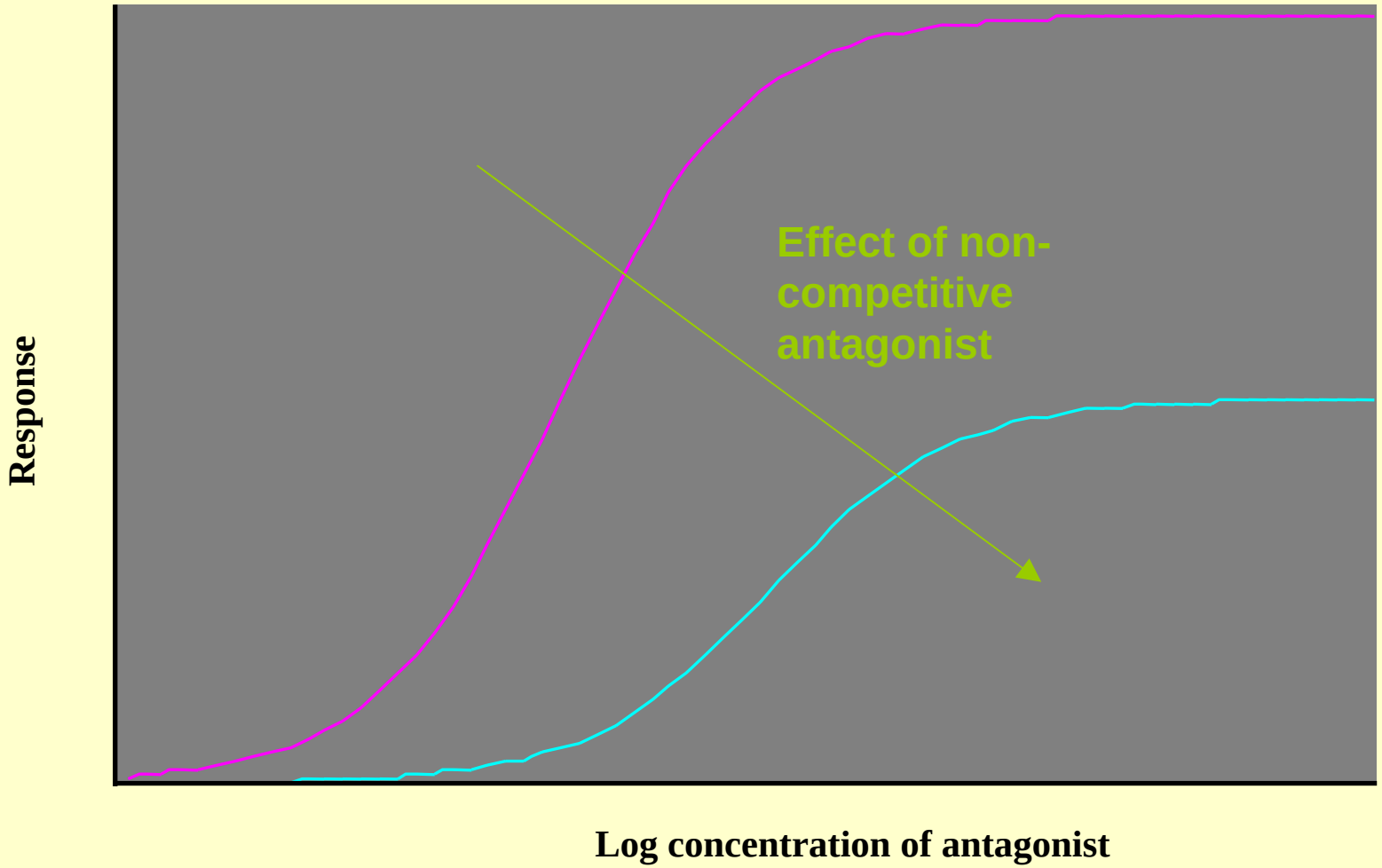
Potency of antagonists (Fixed dose of agonist)



Effect of partial agonist on agonist



Non-competitive antagonists



UNIVERSITY OF NEWCASTLE

Psychobiology Research Group

Pharmacogenetics: Any relevance to clinical practice?

Plan

- What is pharmacogenetics and why might it be of interest in psychiatry?
- What are some of the findings in pharmacogenetics esp. relating to antidepressants and antipsychotics?
- Do these have any current or future clinical relevance?

Pharmacogenetics

- Hypothesis
  - Variability in response, toxicity and adverse effects following drug treatment is influenced by genetic variation
- Advantages
  - Genotyping can be done any time
  - Not influenced by current treatment
  - Can be measured very reliably
  - Genome fully sequenced
  - Easy to do – peripheral blood sample

Heritability – a starting point

- FHx of response or side effects
  - Poor man's pharmacogenetics?
- Antidepressants
  - 38 family pairs concordant for response to Imipramine (Angst, 1964)
  - 12/12 and 10/12 concordance of first degree relatives (Pare et al. 1962; Pare & Mack, 1971)
  - Retrospective study in 4 families who responded to tranylcypromine but not other ADs (O'Reilly et al. 1994)
  - 67% of 1° rels of fluvoxamine responders responded (Franchini et al. 1998)
- Antipsychotics
  - Afro-Caribbean greater acute response than Caucasians (Emsley et al. 2002)
  - Little other supportive data

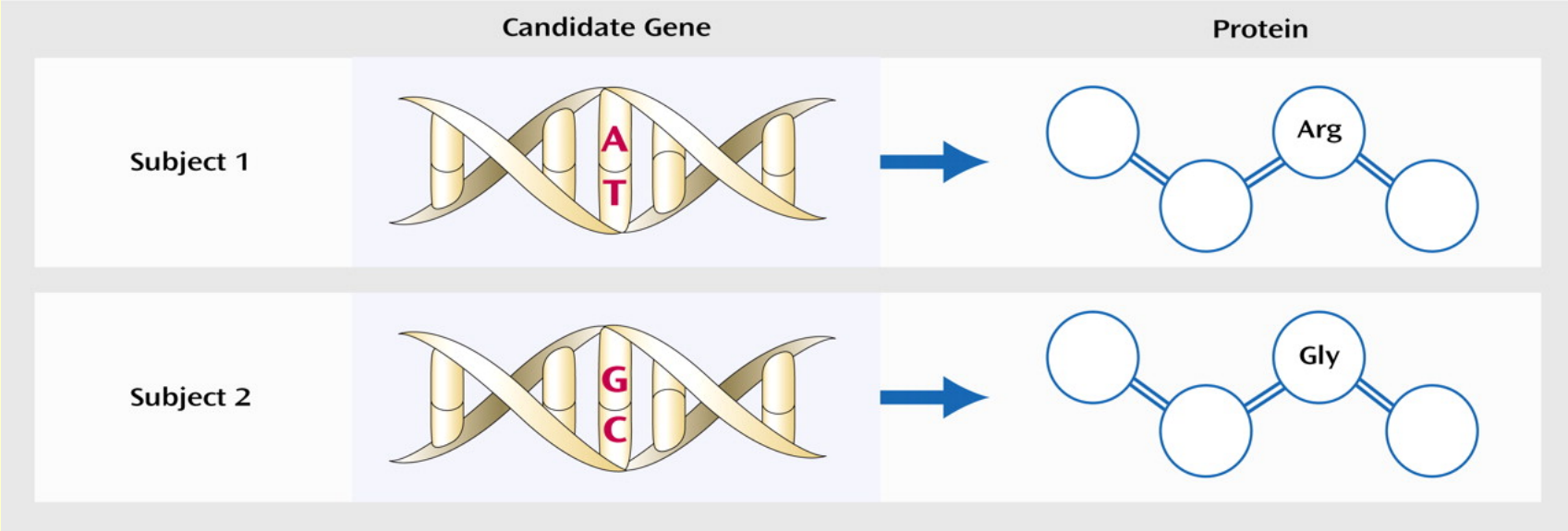
Definition of some terms

- Pharmacogenetics
  - The study of candidate genes that may influence drug effects and metabolism
- Pharmacogenomics
  - The study of all genes (and their expression) in the genome that may influence drug effects and metabolism
  - Needs large-scale high-through put techniques to screen the genome

Genetic Variation

- Polymorphism
  - Genetic variation that occurs with a frequency ≥ 1% in the population
  - Various types
    - SNPs (Single nucleotide polymorphisms)
    - Repetitive DNA sequences
  - Must be functional (?)
    - Alter the expression levels or conformation of a drug-related protein

Single Nucleotide Polymorphism (SNP) in the Coding Region of a Gene



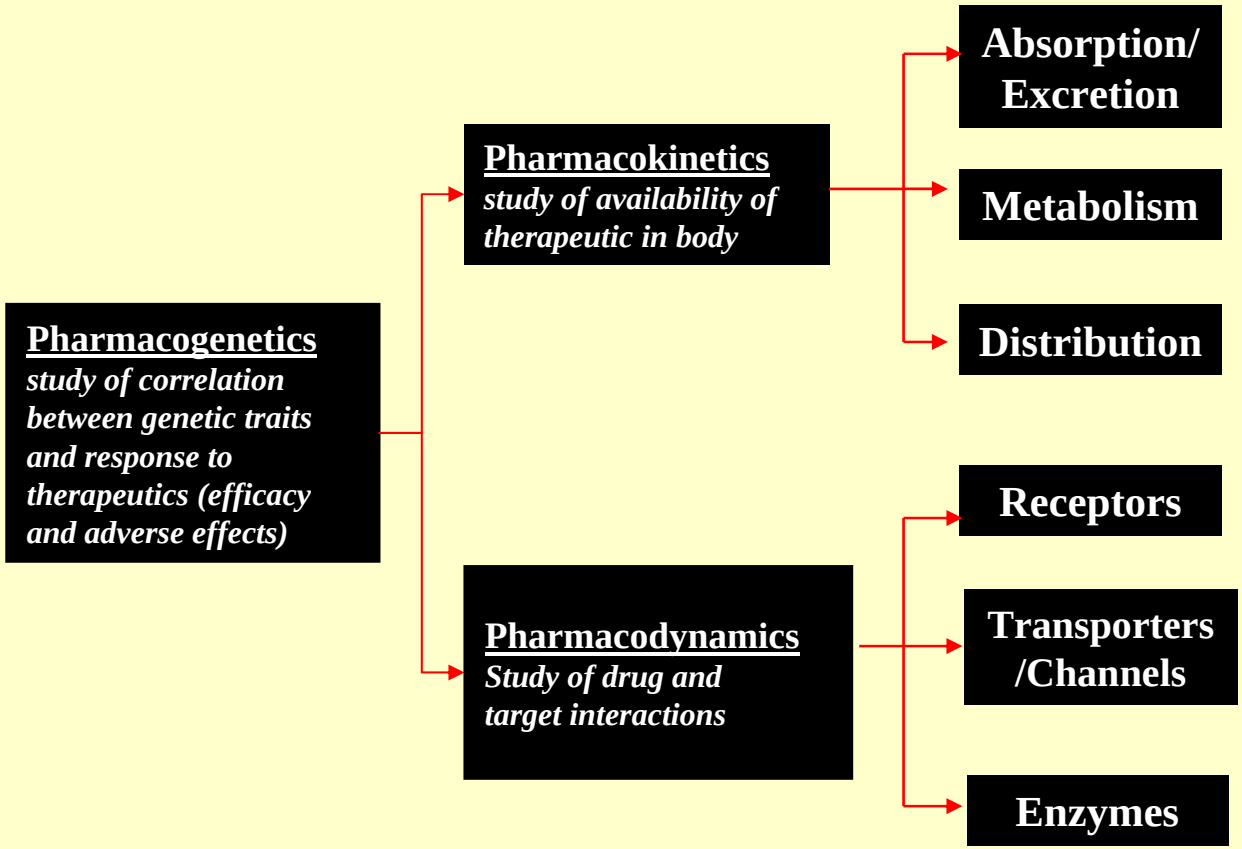
- SNP results in alteration of the amino acid sequence of the corresponding protein
  - arginine (Arg) substituted for glycine (Gly)
  - Distinct protein structures could result in phenotypic differences between the subjects, such as variation in response to medication.

Taken from Malhotra et al. 2004 Am.J.Psych.

Pharmacogenetics: Association studies

- Association of polymorphisms of candidate genes in individuals with different responses to treatment
- Useful for identifying genes of major effect
- Problems
  - Definition of phenotype
    - Diagnostic heterogeneity
    - Response assessment
    - Placebo response
    - Effects of previous treatments
  - Sample size
  - Duration of treatment
  - Ethnicity
  - Comorbid illness and concomitant medication
  - Response probably determined by multiple genes of small effect

Pharmacogenetic tree



CYP450:

- **76%** of ADRs are dose dependent
- **CYP450** is one of the best characterized metabolic protein complexes

Pharmacokinetics: CYP450

- CYP450 enzymes polymorphic with e.g. 70+ variants of CYP2D6 gene
  - Some of these affect functionality
  - Reduced activity
    - CYP1A2*1C, CYP2D6*10B, CYP2C9*2
  - Increased activity
    - CYP1A2*1F, CYP2D6*2xn

Drug Concentrations by Genotype

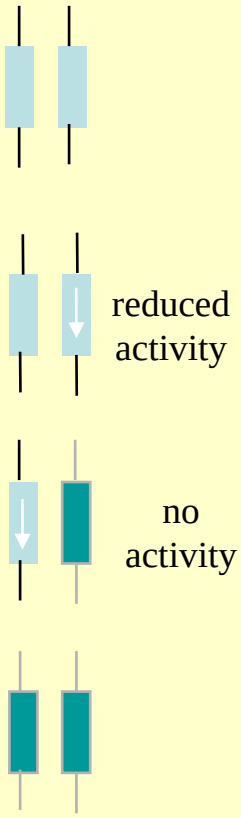
Metabolizer Status	Genotype	Response to average daily dose

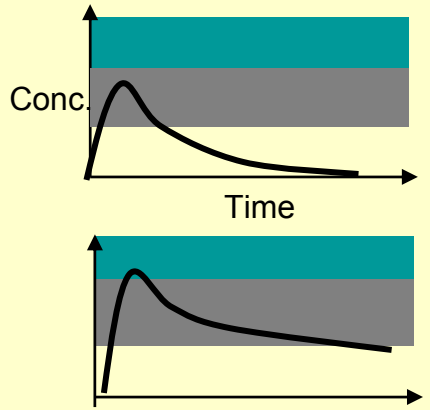
Ultrarapid

Extensive

Intermediate

Poor



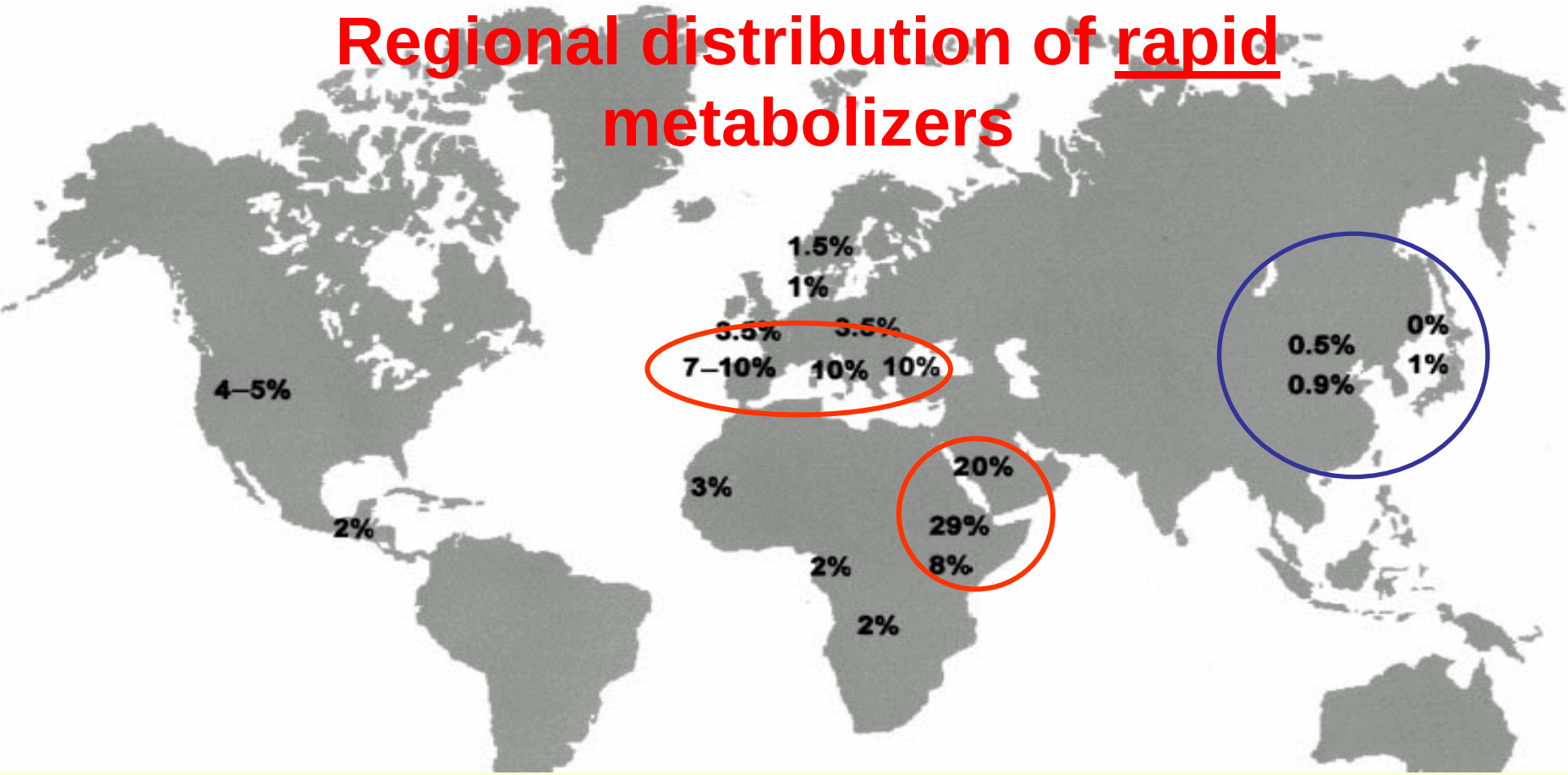


= Adverse Events

= Therapeutic Window

= Ineffective

Regional distribution of rapid metabolizers



Ingelman-Sundberg (2001) Journal of Internal Medicine 250: 186

CYP2D6 and dosing of antidepressants

Genetic analysis may allow for appropriate dosing:

	Percent of normal dose			
Drug	UM	EM	IM	PM
Venlafaxine	-	130%	80%	20%
Desipramine	260%	130%	80%	20%
Fluoxatine	-	120%	-	60%
Mianserin	300%	110%	-	70%

Source: Kirchheiner et al., Acta Psychiatr. Scand 2001: 104: 173-192

CYP450 Polymorphism Findings

- No association between CYP450 polymorphisms and response to antipsychotics or antidepressants identified to date
- CYP2D6 and CYP1A2 associated with increased side effects of antipsychotics (TD and PSx)(Basile et al. 2000; Lam et al. 2001)
- CYP2D6 and CYP2C19 associated with increased side effects with sertraline (Wang et al. 2001)
  - N.B. wide therapeutic index with SSRIs

Roche

Diagnostics

AMPLICHIP

Powered by Affymetrix

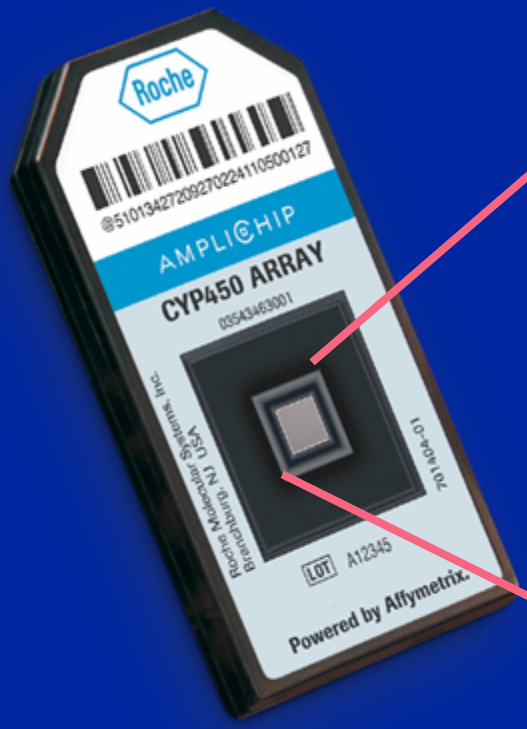
The Way Ahead™

The AmpliChip tests are based on Affymetrix microarray technology

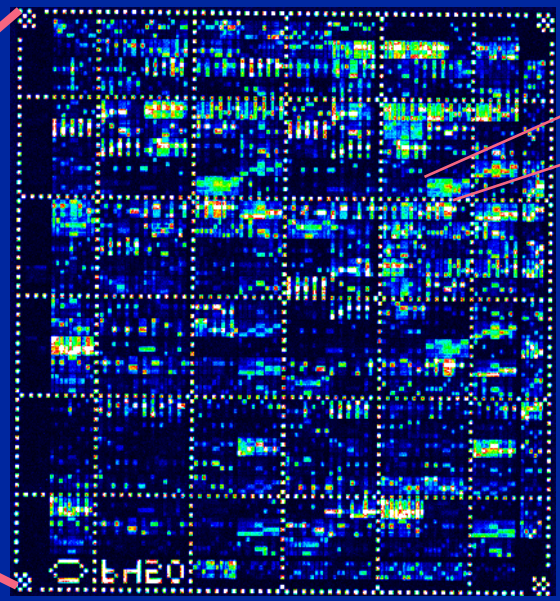
AmpliChip CYP450 CE-IVD

Roche

Diagnostics

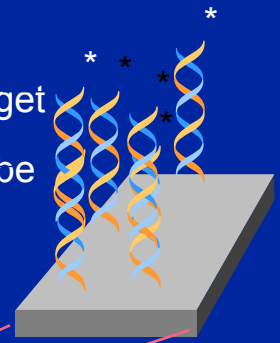


CYP450 *2D6 & 2C19*



labeled DNA target

Oligonucleotide probe



To address the relevant genetic variations, each array contains over 15,000 different probes complementary to sense and anti-sense P450 genomic DNA. Probes range in length from 18mer to 22mer

AMPLICHIP

Pharmacodynamics

- The interaction of a drug with a target molecule
  - Receptors, enzymes, transporters, ion channels
- Leads to therapeutic effects
- Can lead to side effects

Dopamine receptors and antipsychotics

- DRD2 polymorphisms
  - Associated with early response to HDL and Risperidone (Malhotra et al. 1999; Schafer et al. 2001; Mata et al. 2002)
  - **Not** long-term clozapine response (Arranz et al. 1998)
  - **Not** TD (Kaiser et al. 2002)
  - BUT N.B. largest DRD gene with many polymorphisms
- DRD3 polymorphisms
  - Associated with clozapine response (Scharfetter et al. 1998)
  - Effect of olanzapine on +ve symptoms (Staddon et al. 2002)
  - Meta-analysis shows small risk of TD (Lerer et al. 2002)
- DRD4 polymorphisms
  - No consistent association with clozapine response (Malhotra et al. 2004)

5-HT Receptors and antipsychotics

- 5-HT_{2A} polymorphisms
  - 2 different ones associated with clozapine non-response in European and American populations (Arranz et al. 1995; Masellis et al. 1998)
  - 1 associated with risperidone and clozapine response in Chinese populations (Lane et al. 2002)
  - Increased risk of TD (Tan et al. 2001)
- 5-HT_{2C} polymorphisms
  - Meta-analysis suggests role in clozapine response (Sodhi et al. 1999)
  - Risk of TD (Segman et al. 2000)
  - Risk of weight gain (Reynolds et al. 2003)
- 5-HT_{6}
  - ?Clozapine response (Yu et al. 1999; Masellis et al. 2001)

5-HT Receptors and antidepressants

- 5-HT_{2A} polymorphisms
  - Marginal association with SSRI response (Cusin et al. 2002)
- 5-HT_{1A} polymorphism
  - Functional
  - Associated with alterations in expression of 5-HT_{1A} receptors (Lemonde et al. 2003)
  - Associated with response to TCAs and SSRIs (Serretti et al. 2004; Lemonde et al. 2004)

5-HT Transporter

- Polymorphism in the 5-HTT promoter region (5-HTTLPR) – s and l forms
  - s/s associated with an stress X genetic interaction in vulnerability for depression (Wilhelm et al. 2006)
  - l/l associated with SSRI greater response in Caucasians (Smeraldi et al. 1998)
    - Response also faster (Pollock et al. 2000; Zanardi et al. 2000)
    - ? Effect in Asians - non-response (Kim et al. 2000) but response in Chinese (Yu et al. 2002)
  - s/s associated with antidepressant induced mania (Mundo et al. 2001)

Other proteins

- Tryptophan Hydroxylase polymorphism
  - Poor response to fluvoxamine and paroxetine (Serretti et al. 2001)
  - N.B. non-functional
- MAO-A polymorphism
  - No association with antidepressant response (Serretti et al. 2004)
- G-protein polymorphisms
  - Association with depression and antidepressant response (Zill et al. 2000; Exton et al. 2003)
- BDNF polymorphism
  - Trend for association with SSRI response (Tsai et al. 2003)
- Inositol phosphate polymorphism
  - Inconsistent data with lithium (Steen et al. 1998)

Combinations of genes

- Combining information from key response-related genes
  - Can constantly refine predictions by adding additional genes
  - Will need adjustments for ethnic mix
- Examples:
  - DRD3 and 5-HT_{2C} polymorphisms have additive effects on risk of TD (Segman & Lerer 2002)
  - DRD3 and CYP1A2 polymorphisms additive effects on risk of TD (Basile et al. 2000)
  - Response in Alzheimers predicted by combination of polymorphisms of APOE, PS1 and PS2 (Cacabelos et al. 2000)
- Problems
  - What statistical methods should be used?
  - Disequilibrium
  - Effects additive or synergistic?

Prediction of Clozapine response (Arranz et al. 2000)

- 200 schizophrenia patients (all white Caucasians of British origin) treated with clozapine (133 responded)
- 19 polymorphisms analysed
- 6 with strongest association with response (5-HT_{2A} X 2, 5-HT_{2C} X 2, 5-HTT, H_2) combined
  - PPV: 0.76 ± 0.08
  - NPV: 0.82 ± 0.16
  - Sensitivity 95.9% ± 0.04% (for identifying “satisfactory” responders)
  - Specificity 38.3 % ± 0.14% (for identifying poor responders)
- Utility?
  - “benefit of persevering with treatment in poor responders”
  - “more patients will benefit from clozapine if a positive response is predicted”
  - Other drugs
    - Olanz 70% correct predictions; add in DRD3 increases to 76%

Where to next?

- Independent replication of results needed
- Clarification of ethnic differences
- DNA micro-arrays for high through put analysis for a wider search of the genome
- Newer candidates
  - Genes who's expression is altered by disease
  - Signal transduction proteins

Conclusions

Pharmacogenetics:
Any relevance to clinical practice?

Possibly….

- CYP450 chip technology may be helpful for a minority of patients
  - A pragmatic trial is about to start
- Use of pharmacogenetics for efficacy predictions (e.g. for clozapine) less clear

- The future (5-10 years) does potentially look very interesting