Cardiac-Sensitization(CS)
vs.
Physiologically-Based Pharmacokinetic Modeling (PBPK)

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CS vs. PBPK

Objectives:

- Define NOAEL, LOAEL, and cardiac sensitization (CS);
- Describe the use of epinephrine challenge dose to elicit CS in dogs;
- Explain the relationship between the epinephrine challenge dose and chemical agents such as CFC and CF_3I.
- Explain a PBPK Model and its valid use.

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Definitions:

- **NOAEL (No observed adverse effect level): Negative to a test chemical dose administered during epinephrine-challenge; less than the lowest positive dose.**
- **LOAEL (Lowest observed adverse effect level): Positive at the lowest test chemical dose administered during epinephrine-challenge dose that is just below that which will evoke a cardiac response even without a test chemical administered.**

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LOAEL (Lowest observed adverse effect level):

- **The lowest level of test chemical that produced cardiac response when high dose of exogenous epinephrine is given to challenge the experimental animals (called an epinephrine-challenge).**

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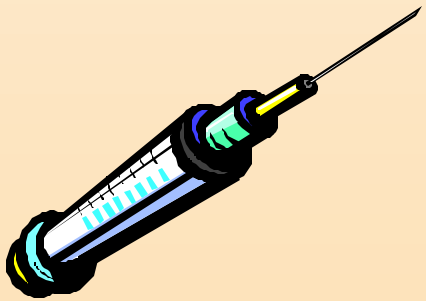
LOAEL (Lowest observed adverse effect level):

- **Without an exogenous epinephrine-challenge, animals will not respond to a test chemical unless much higher dose is given.**
- **Thus, high level of epinephrine-challenge is required in the CS testing to evoke a clinical response.**



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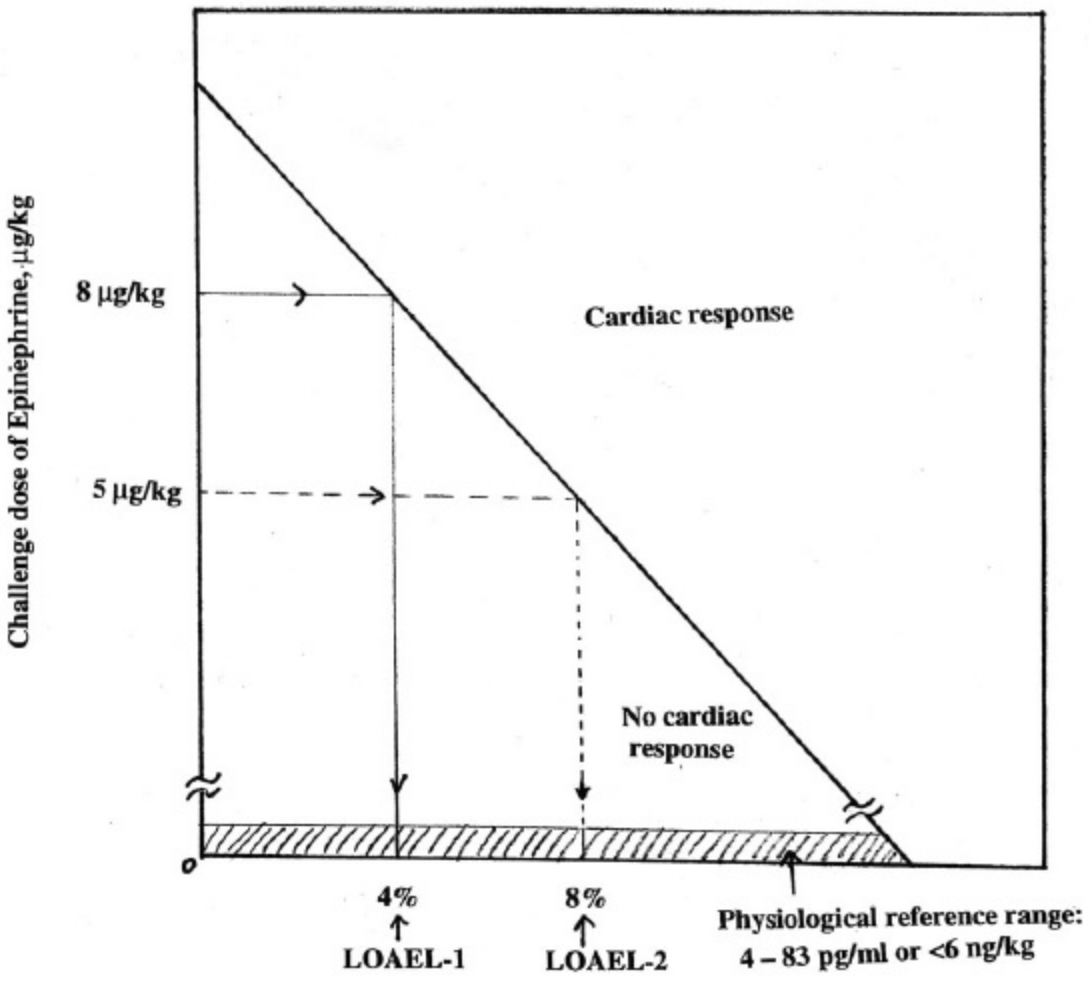
CS Test Protocol:

(Continuous monitoring)

1. **Start ECG recording at 0 min,**
2. **First epinephrine-challenge injected at 2 min,**
3. **Test chemical inhaled at 7 min,**
4. **Second epinephrine-challenge injected at 12 min,**
5. **Stop test chemical and ECG at 17 min.**

Figure 1. Hypothetical relationship between epinephrine dose and chemical that produces cardiac response.



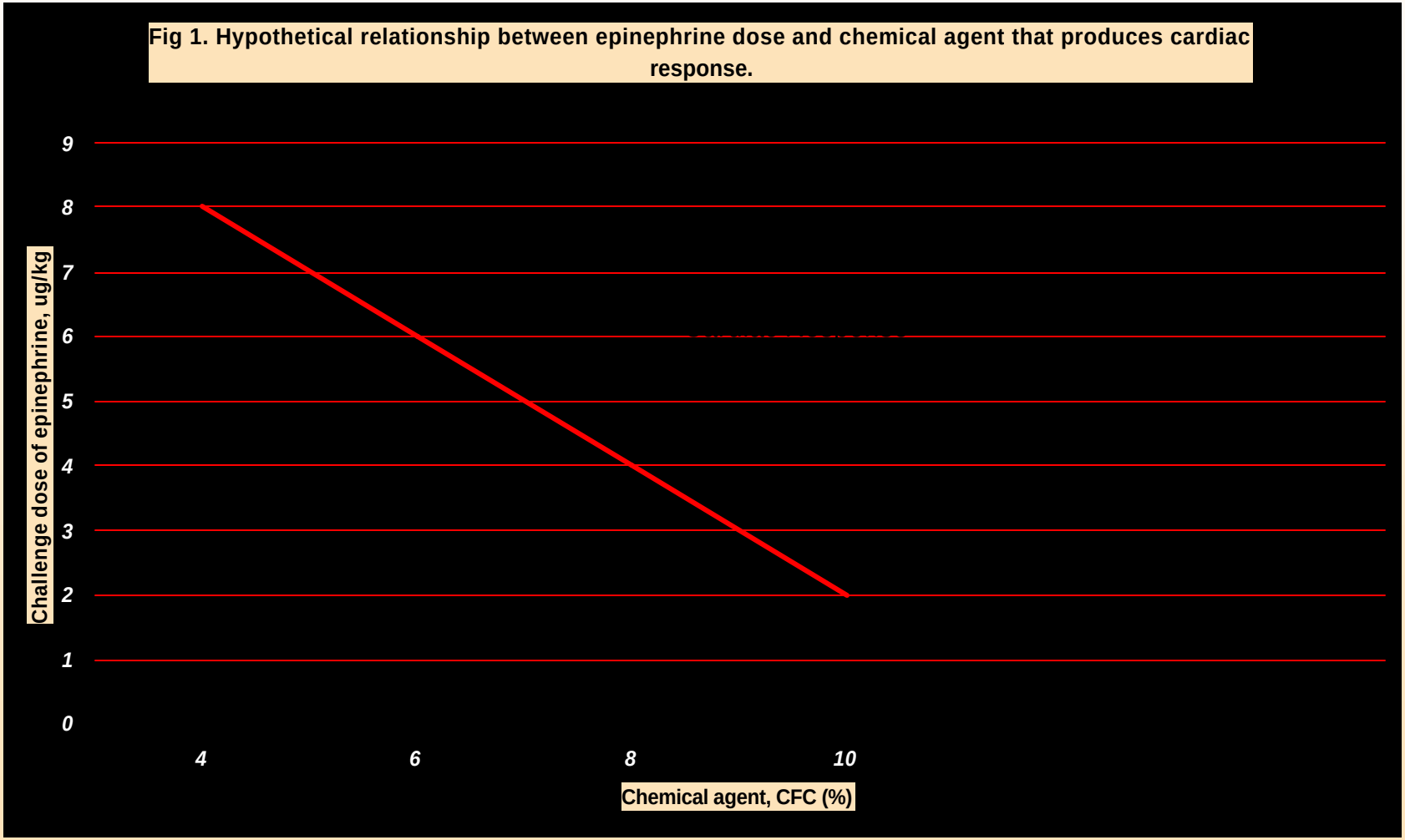


Fig 1. Hypothetical relationship between epinephrine dose and chemical agent that produces cardiac response.
Challenge dose of epinephrine, ug/kg
9
8
7
6
5
4
3
2
1
0
4
6
8
10
Chemical agent, CFC (%)

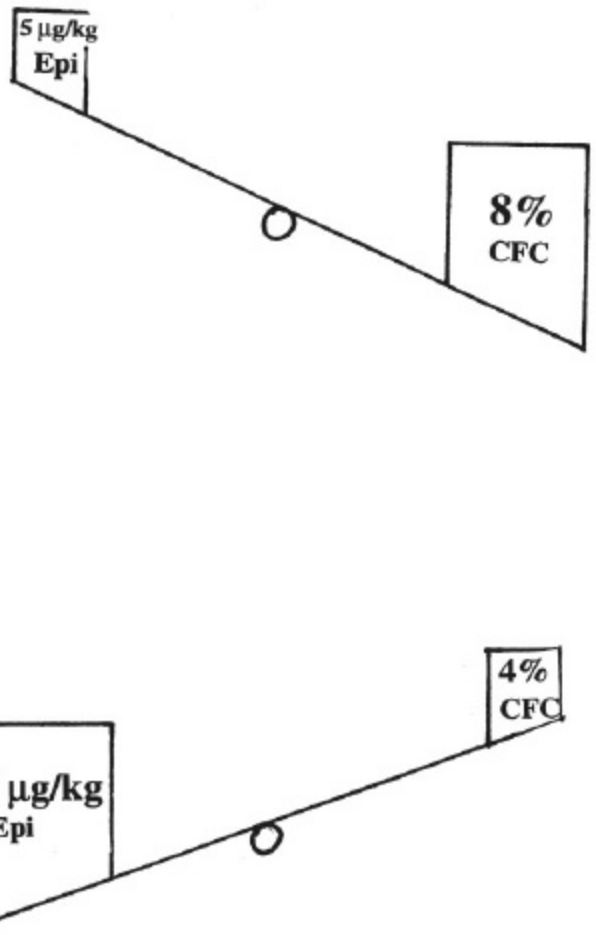


Figure 2. Relationship between epinephrine (Epi) dose vs. chemical agent (CFC).

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CF_3I (Dodd and Vinegar, Drug and Chemical Toxicology, 21(2): 137 - 149, 1998)

- **Used a total of 9 pure-bread male Beagle dogs,**
- **Each dog received varying doses of epinephrine (1, 4, 8, and 12㎍g/kg),**
- **Each dog received a dose of CF_3I where the test is performed in singlicate, n = 1.**

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CF_3I (Dodd and Vinegar, Drug and Chemical Toxicology, 1998):

- Three dogs (33%) were rejected due to adverse response to an epinephrine reaction before CF_3I given.
- Two dogs were struggling even at 1 μg/kg epinephrine),
- Thus, 5/9 (56%) dogs showed clinical effects without CF_3I given.

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Comments of the CF_3I Experimentation

- **Manipulated Experiment: An excess level of exogenous epinephrine-challenge doses that correspond to a normal human plasma (4-83 pg/ml):**

µg/kg	Human Eql.
1	167 X
4	670 X
8	1,300 X
12	2,000 X

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Comments of the CF_3I Experimentation

- **Biological Variability:**

  The dogs were showing clinical effect caused by epinephrine-challenge without CF_3I given. The other biological variability includes gender, age, nutrition, health state, psychological factors, and physiological difference.

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Comments of the CF_3I Experimentation

- **Statistical Significance:**

  Only one dog is used per CF_3I dose in which one died at 0.4% (at 8 mg/kg epinephrine-challenge). The test was performed in singlicate, not using multiple dogs for the CF_3I (0.4%) in question to verify the reproducibility of test results.

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Comments of the CF_3I Experimentation:

- Based on this one experiment, the NOAEL was then assigned as 0.2% (2,000 ppm) and the LOAEL, 0.4% (4,000 ppm).

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Comments of the CF_3I Experimentation

- However, when 2.5% CF_3I was exposed to dogs without epinephrine-challenge, no cardiac response observed (12.5 times higher than the NOAEL). Tachycardia began to show at 5% level . . . ICF Kaiser/Huntingdon, 1998 in Clewell, H. and Lawrence, G., May 21, 1999.

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Comments of the CF_3I Experimentation

- **Scientific confirmation of data: As of Aug 20, 2002, there is no other publication repeating and reproducing the same experimental results.**

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Comments of the CF_3I Experimentation

- No arterial epinephrine level was determined during the experiment. As a result, I cannot determine what really killed the dog (additional endogenous epinephrine produced by the dog during the stress or the actual test chemical administered).

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Comments of the CF_3I Experimentation

- Mode of administration of exogenous epinephrine: More epinephrine will reach the heart if injected in the artery than in the venous vein.

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Comments of the CF_3I Experimentation

- Also one dog died when 2% of CFC-11 (12 mg/kg epinephrine) was administered. Does this mean that the NOAEL for CFC-11 is 2% or 20,000 ppm (the accepted NOAEL is 0.34% or 3,400 ppm). Note: The NOAEL for CF_3I was determined at 8 mg/kg epinephrine-challenge. Then, why the CFC-11 study was performed at high epinephrine-challenge of 12 mg/kg? The dose-response between the epinephrine dose and test chemical administered is inversely related.

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Comments of the CF_3I Experimentation

- **CFC-11 and CFC-12 are used in human oral inhalation propellant for Albuterol (asthma medication) as well as refrigerants even though the NOAEL is similar to CF_3I.**

NOAEL: CFC-11 = 3,400 ppm

CF_3I = 2,000 ppm

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Comments of the CF_3I Experimentation

- If CFC-11 and CFC-12 are adequately safe for human use, then CF_3I should be safe. The toxic data profile of CF_3I obtained from an animal model falls within the range of toxicity data profiles of currently used inhalation propellant medications, fire extinguishants and refrigerants.

  Note: I am not advocating this fire suppressant, CF_3I, for human medical use.

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Comments of the CF_3I Experimentation

- According to Skaggs and Rubenstein (Setting the Occupational Exposure Limit for CF_3I, Halon Options Technical Conference, 27-29 APR 1999), the dogs without epinephrine-challenge when exposed to 5% CF_3I showed no adverse cardiac effects. This is about 12.5 times above the LOAEL (0.4%).

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Recommendations:

- **A standardized protocol for CS study is needed:**

  - Inverse dose relationship between the epinephrine-challenge and the test chemical administered,

  - Determine an appropriate epinephrine-challenge dose,

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Recommendations:

A standardized protocol for CS study is needed:

- **Inclusion of positive and negative controls for precision study,**

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Recommendations:

- **A standardized protocol for CS study:**

- Mode of epinephrine injection: venous vs. arterial,

- Blood level of epinephrine before exposing to a test chemical,

- Additional sensors such as BP and heart rate to monitor heart functions beside ECG,

- Use multiple animals to confirm the positive results,

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Recommendations:

A standardized protocol for CS study is needed:

- **Assess reproducibility of the positive test results by another laboratory,**

-The CS data must not be used as an absolute basis to determine the safe use of a chemical in humans.

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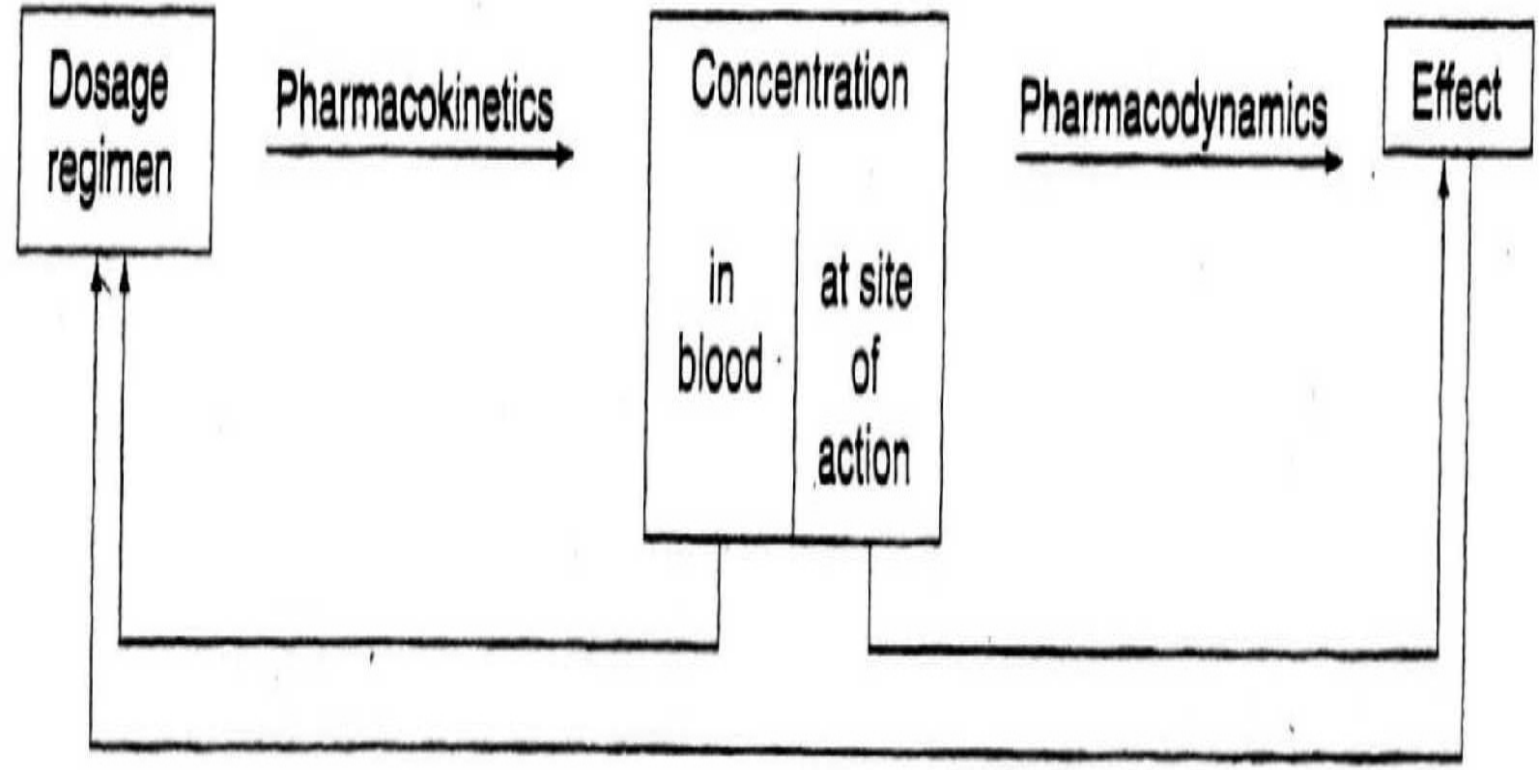
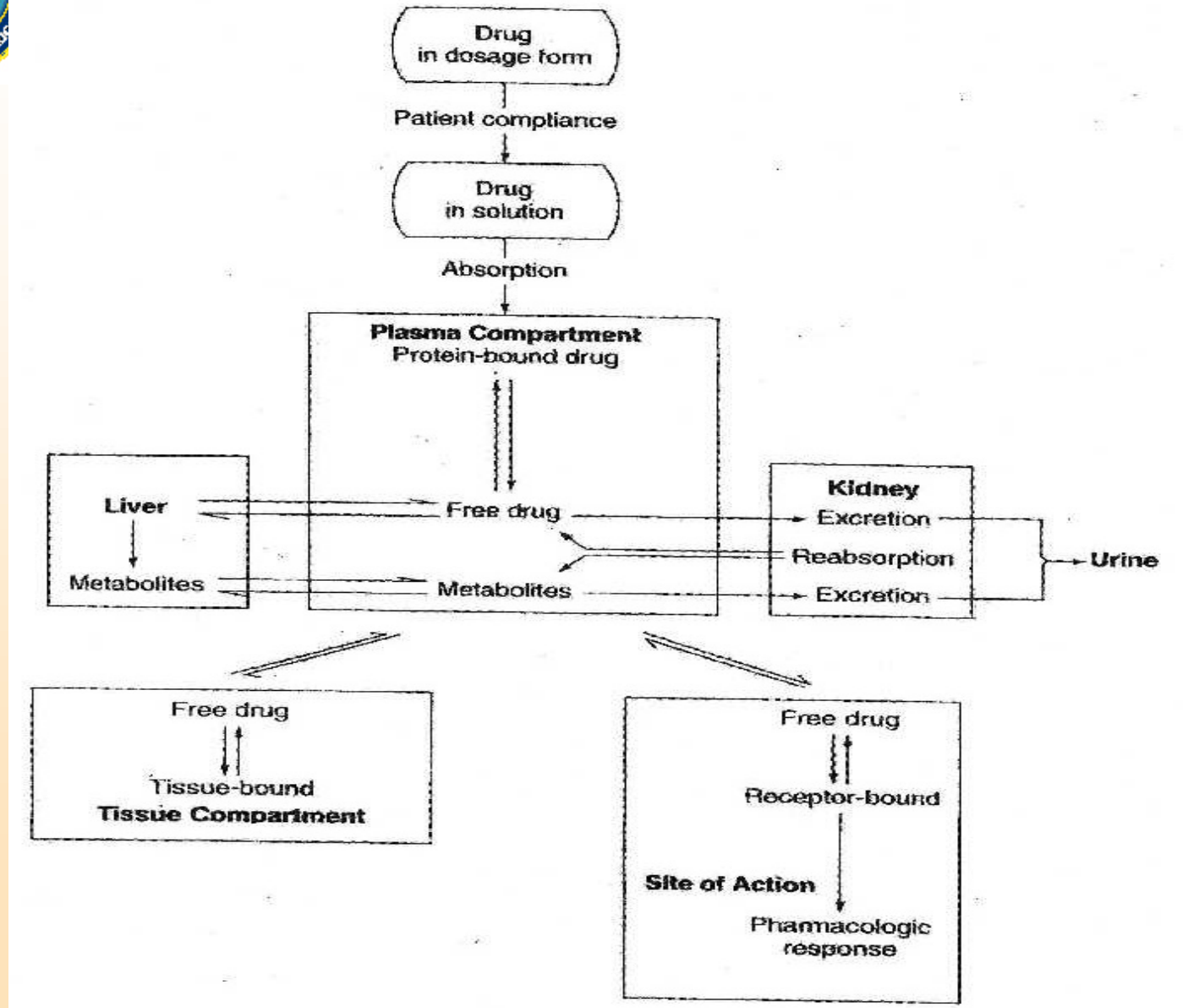
- **Physiologically-Based Pharmacokinetic (PBPK) Model:**
  - **Mathematical description of:**
    - **» Uptake,**
    - **» Absorption,**
    - **» Distribution,**
    - **» Pharmacodynamics,**
    - **» Metabolism,**
    - **» Elimination.**


Dosage regimen
Pharmacokinetics
Concentration
in blood
at site of action
Pharmacodynamics
Effect


Drug
in dosage form
Patient comptiance
Drug
in solution
Absorption
Plasma Compartment
Protein-bound drug
Liver
Free drug
Kidney
Excretion
Reabsorption
Urine
Metabolites
Metabolites
Excretion
Free drug
Tissue-bound
Tissue Compartment
Free drug
Receptor-bound
Site of Action
Pharmacologic
response

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- Physiological and anatomical properties of animals,
- Specific chemical properties,
- Chemical concentrations in blood and duration of exposure (inhalation during the first second to minute),
- The link among the following data:
  - CS end-point in animals,
  - LOAEL at 5-minute exposure,
  - Human arterial concentration data obtained from:
    - Halothane, Isoflurane, and Desflurane,
    - CFC-11 (IV and Inhalation)
    - Monte Carlo Simulations (95 and 99%).

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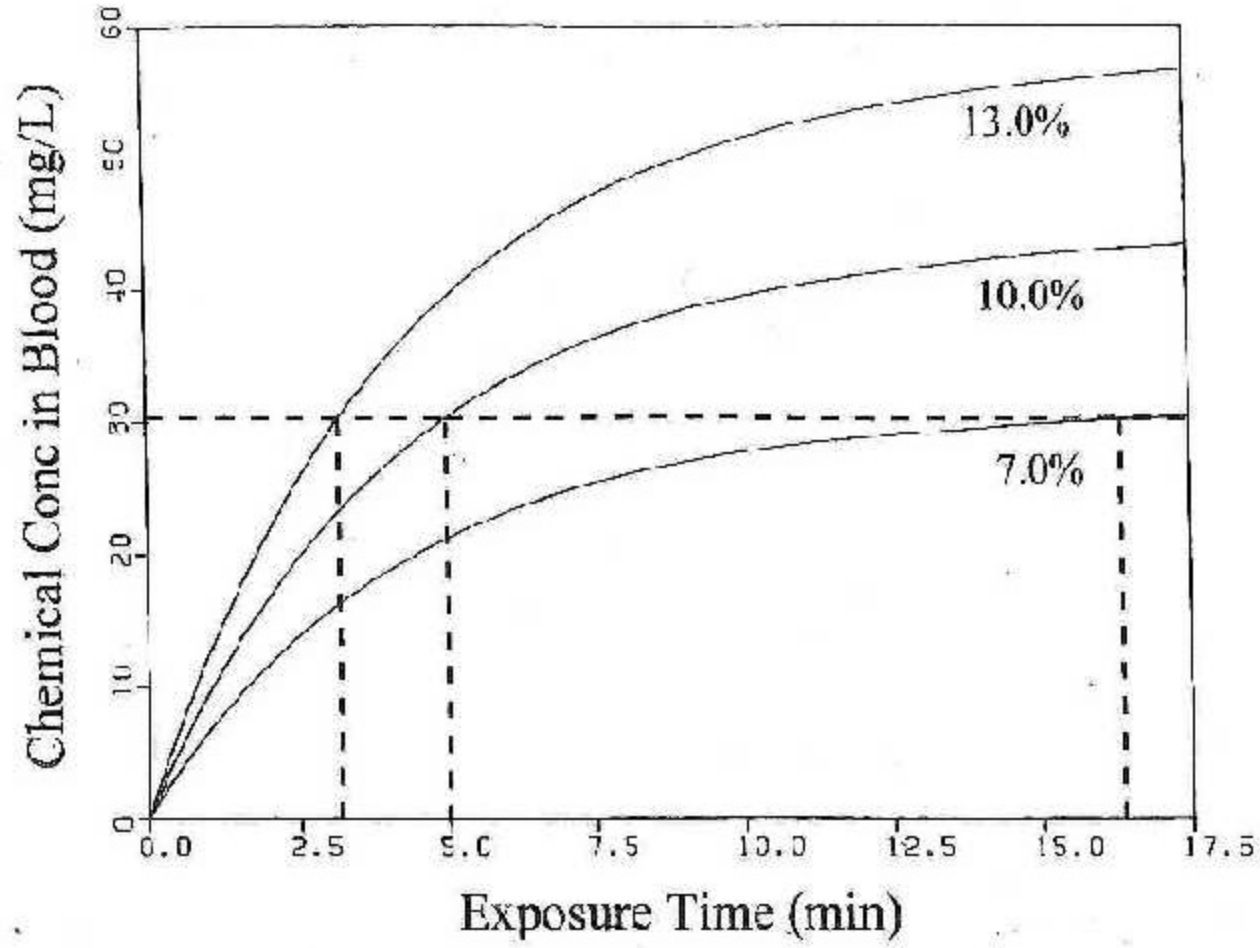
- **Resting and moderate activity level conditions,**
- **Based on 70 kg man.**


Chemical Conc in Blood (mg/L)
0
10
20
30
40
50
60
13.0%
10.0%
7.0%
0.0
2.5
5.0
7.5
10.0
12.5
15.0
17.5
Exposure Time (min)

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- **Time for Safe Human Exposure for CF_3I”**

% v/v	PPM	Human Exposure Time (Minute)
0.20	2,000	5.00
0.25	2,500	5.00
0.30	3,000	5.00
0.35	3,500	4.30
0.40	4,000	0.85
0.45	4,500	0.49
0.50	5,000	0.35

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- **General guidelines for Occupied vs. Unoccupied areas:**
  - **If chemical concentration < LOAEL, use in Occupied area.**
  - **If chemical concentrations > LOAEL, use in Unoccupied areas.**

- **Concentrations > LOAEL: < 30 sec to egress the area;**
- **Concentrations < LOAEL: < 60 sec to egress the area;**
- **No information of LOAEL: < 30 sec to egress the area.**

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- **PBPK currently in use:**

  (NFPA 2001 Standard)

  - **US Environmental Protection Agency, EPA,**
  - **National Fire Protection Agency, NFPA (NFPA 2001 Standard),**
  - **Occupational Safety and Health Administration, OSHA,**
  - **National Institute for Occupational Safety and Health, NOISH,**
  - **US Dept of Transportation, FAA.**