

SNS COLLEGE OF PHARMACY AND HEALTH SCIENCES

Affiliated To The Tamil Nadu Dr. MGR Medical University, Chennai

Approved by Pharmacy Council of India, New Delhi.

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COURSE NAME : PHARMACEUTICAL MICROBIOLOGY - BP303 T

B.PHARM II YEAR / III SEM

UNIT 3

SUB TOPIC :FACTORS INFLUENCING DISINFECTION, ANTISEPTICS AND THEIR EVALUATION

A. Concentration of Agent

Higher Concentration → Faster & Stronger Action

Some agents effective only at optimum concentration (e.g, alcohol 70%)



B. Duration of Exposure

Longer Contact Time → Greater Microbial Kill



Insufficient Time → Sub-Lethal Effect

C. Type of Microorganism

Most Resistant

Least Resistant



Spores



Mycobacteria



Gram-Negative



Gram-Positive



Fungi



Viruses

D. Microbial Load

Higher Number of Organisms ↓ Reduced Effectiveness



High Microbial Load



Low Microbial Load

E. Presence of Organic Matter

Blood, Pus, Feces, Proteins reduce activity

Especially affects Halogens & Phenolics



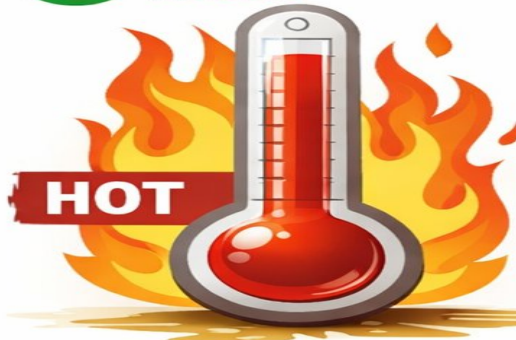
F. Temperature

Higher Temperature → Increased Killing Rate

Excess Heat may Inactivate Some Disinfectants



**Faster
Action**



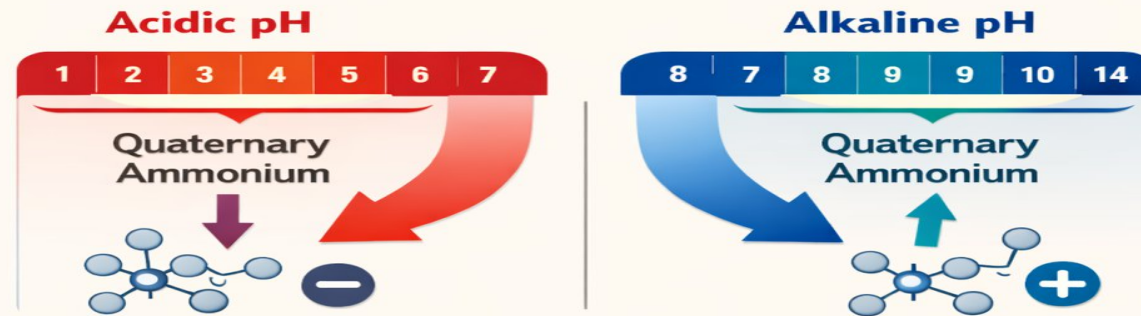
**Disinfectant
Breakdown**



G. pH of Medium

Acidic or alkaline pH alters ionization & activity

Example: Quaternary ammonium compounds ↓ in acidic pH

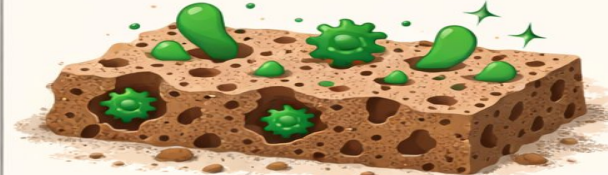


H. Nature of Surface

Smooth surfaces
→ Better Action

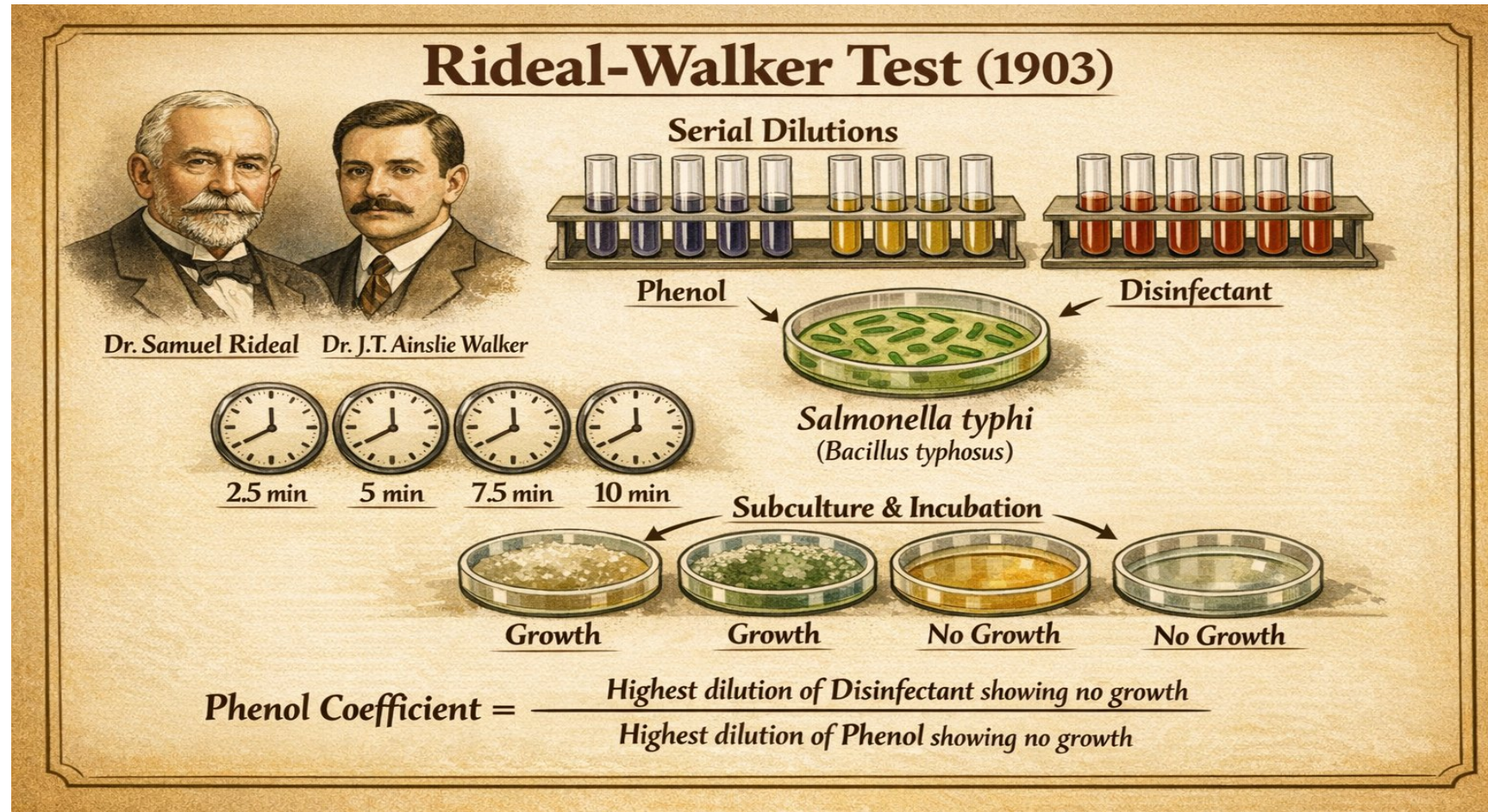


Porous surfaces
→ Reduced Penetration



Evaluation of Disinfectants & Antiseptics

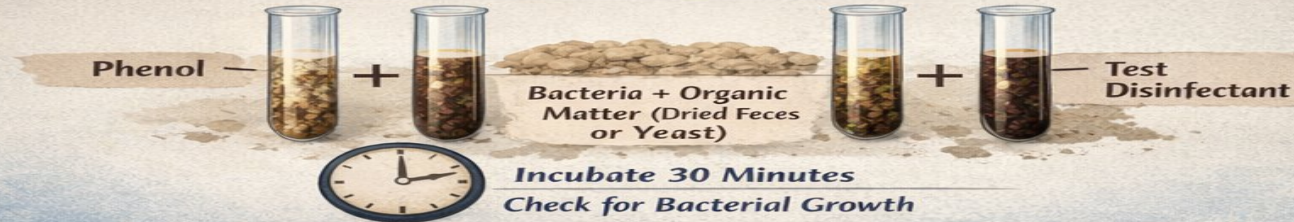
A. Phenol Coefficient Tests



The Chick-Martin Test (1908)

Developed by Harriette Chick & Charles James Martin

Phenol vs. Disinfectant in Dirty Conditions



Key Differences from Rideal-Walker Test

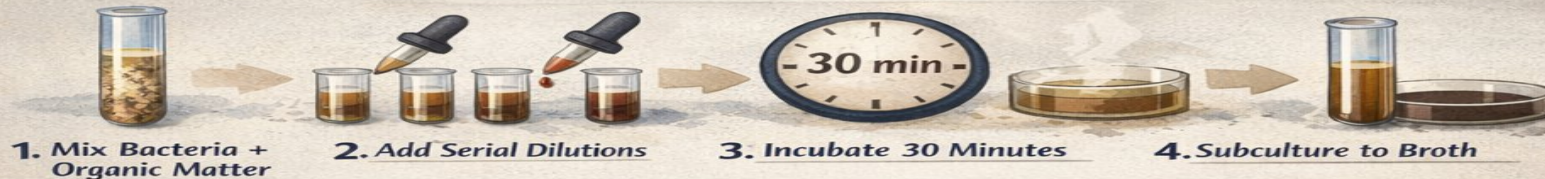
Rideal-Walker Test (1903)

- Clean Conditions
- Short Contact Time
- *Salmonella typhi* Only
- High Phenol Coefficient

Chick-Martin Test (1908)

- Organic Matter Added
- Fixed 30-Minute Time
- *Salmonella typhi* & *Staphylococcus aureus*
- Lower Phenol Coefficient

Procedure Overview



Calculating Phenol Coefficient

$$\text{Phenol Coefficient} = \frac{\text{Mean Phenol Concentration Allowing Survival}}{\text{Mean Disinfectant Concentration Allowing Survival}}$$

OR

$$\frac{\text{Effective Dilution of Disinfectant}}{\text{Effective Dilution of Phenol}}$$

Results: >1 = More Effective than Phenol <1 = Less Effective than Phenol

Pioneering Test for Disinfectants in Realistic, Dirty Conditions

In-Use Test

Monitoring Disinfectant Contamination

1 mL Sample in Broth
Dilute & Neutralize

Incubate & Count Colonies
• 37°C | 3 Days

**Room Temp
7 Days**

Plate Drops on Agar

Pass
Less than 5 Colonies

Fail
5 or More Colonies

Routine Hospital Testing

Ensure Disinfectant Safety

Kelsey-Sykes Capacity Test

Testing Disinfectant Efficacy

1. Prepare Disinfectant Dilution



2. Add Bacterial Suspension



3. First Challenge

8 min, Subculture to Broth



4. Second Challenge

10 min: Add Bacteria



5. Third Challenge

20 min: Add Bacteria



6. Incubate & Evaluate

Check for Growth



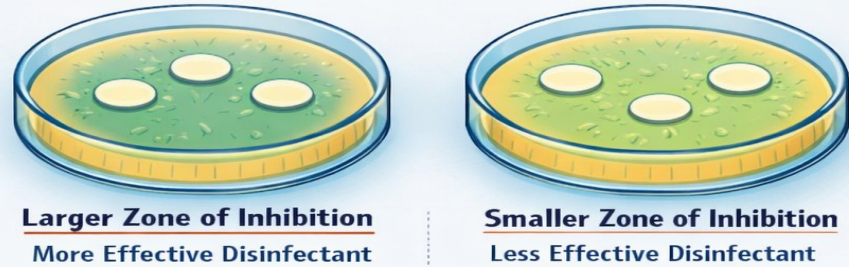
Test Organisms: *S. aureus*, *E. coli*, *P. aeruginosa*, *P. vulgaris*

Agar Diffusion Method for Disinfectant Comparison

Procedure Steps



Zone of Inhibition Comparison



Key Considerations & Limitations



Diffusion Variability

Different chemicals diffuse at different rates



Solubility Issues

Poorly soluble agents may not diffuse



Bacteriostatic vs. Bactericidal

Inhibits growth vs. Kills bacteria

A BRIEF SUMMARY OF DISINFECTANTS & ANTISEPTICS

Key: USED FOR ANTISEPSIS USED TO DISINFECT SURFACES USED FOR STERILISATION (E.G. MEDICAL INSTRUMENTS) USED FOR PRESERVATION

<p>ALCOHOLS</p> <p>USED IN ALCOHOL-BASED SANITARY HAND GELS PRESENT IN HOSPITALS</p> <div> <chem>CCO</chem> ETHANOL </div> <div> <chem>CC(C)O</chem> ISOPROPANOL </div> <div> </div> <p>Kill many bacteria and fungi, and some viruses, when used at suitably high concentrations (usually 60-90% solution). Slow-acting, and evaporate easily, so lack residual action. Can't be used to sterilise.</p>	<p>ALDEHYDES</p> <p>MAINLY USED FOR DISINFECTION & STERILISATION OF MEDICAL INSTRUMENTS</p> <div> <chem>HCHO</chem> FORMALDEHYDE </div> <div> <chem>O=CC(=O)CCC=O</chem> GLUTARALDEHYDE </div> <div> </div> <p>Show broad activity against bacteria, fungi, and viruses. Not used for general antiseptics, due to their high toxicity. Due to the relatively long contact times required to disinfect, other agents are often preferred.</p>	<p>BIGUANIDES</p> <p>USED IN CREAMS & FOR SKIN ANTISEPSIS IN SURGICAL PROCEDURES</p> <div> <chem>Clc1ccc(cc1)Nc2ccc(cc2)Nc3ccc(cc3)Nc4ccc(cc4)Nc5ccc(cc5)Nc6ccc(cc6)Nc7ccc(cc7)Nc8ccc(cc8)Nc9ccc(cc9)Nc10ccc(cc10)Nc11ccc(cc11)Nc12ccc(cc12)Nc13ccc(cc13)Nc14ccc(cc14)Nc15ccc(cc15)Nc16ccc(cc16)Nc17ccc(cc17)Nc18ccc(cc18)Nc19ccc(cc19)Nc20ccc(cc20)Nc21ccc(cc21)Nc22ccc(cc22)Nc23ccc(cc23)Nc24ccc(cc24)Nc25ccc(cc25)Nc26ccc(cc26)Nc27ccc(cc27)Nc28ccc(cc28)Nc29ccc(cc29)Nc30ccc(cc30)Nc31ccc(cc31)Nc32ccc(cc32)Nc33ccc(cc33)Nc34ccc(cc34)Nc35ccc(cc35)Nc36ccc(cc36)Nc37ccc(cc37)Nc38ccc(cc38)Nc39ccc(cc39)Nc40ccc(cc40)Nc41ccc(cc41)Nc42ccc(cc42)Nc43ccc(cc43)Nc44ccc(cc44)Nc45ccc(cc45)Nc46ccc(cc46)Nc47ccc(cc47)Nc48ccc(cc48)Nc49ccc(cc49)Nc50ccc(cc50)Nc51ccc(cc51)Nc52ccc(cc52)Nc53ccc(cc53)Nc54ccc(cc54)Nc55ccc(cc55)Nc56ccc(cc56)Nc57ccc(cc57)Nc58ccc(cc58)Nc59ccc(cc59)Nc60ccc(cc60)Nc61ccc(cc61)Nc62ccc(cc62)Nc63ccc(cc63)Nc64ccc(cc64)Nc65ccc(cc65)Nc66ccc(cc66)Nc67ccc(cc67)Nc68ccc(cc68)Nc69ccc(cc69)Nc70ccc(cc70)Nc71ccc(cc71)Nc72ccc(cc72)Nc73ccc(cc73)Nc74ccc(cc74)Nc75ccc(cc75)Nc76ccc(cc76)Nc77ccc(cc77)Nc78ccc(cc78)Nc79ccc(cc79)Nc80ccc(cc80)Nc81ccc(cc81)Nc82ccc(cc82)Nc83ccc(cc83)Nc84ccc(cc84)Nc85ccc(cc85)Nc86ccc(cc86)Nc87ccc(cc87)Nc88ccc(cc88)Nc89ccc(cc89)Nc90ccc(cc90)Nc91ccc(cc91)Nc92ccc(cc92)Nc93ccc(cc93)Nc94ccc(cc94)Nc95ccc(cc95)Nc96ccc(cc96)Nc97ccc(cc97)Nc98ccc(cc98)Nc99ccc(cc99)Nc100ccc(cc100)Nc101ccc(cc101)Nc102ccc(cc102)Nc103ccc(cc103)Nc104ccc(cc104)Nc105ccc(cc105)Nc106ccc(cc106)Nc107ccc(cc107)Nc108ccc(cc108)Nc109ccc(cc109)Nc110ccc(cc110)Nc111ccc(cc111)Nc112ccc(cc112)Nc113ccc(cc113)Nc114ccc(cc114)Nc115ccc(cc115)Nc116ccc(cc116)Nc117ccc(cc117)Nc118ccc(cc118)Nc119ccc(cc119)Nc120ccc(cc120)Nc121ccc(cc121)Nc122ccc(cc122)Nc123ccc(cc123)Nc124ccc(cc124)Nc125ccc(cc125)Nc126ccc(cc126)Nc127ccc(cc127)Nc128ccc(cc128)Nc129ccc(cc129)Nc130ccc(cc130)Nc131ccc(cc131)Nc132ccc(cc132)Nc133ccc(cc133)Nc134ccc(cc134)Nc135ccc(cc135)Nc136ccc(cc136)Nc137ccc(cc137)Nc138ccc(cc138)Nc139ccc(cc139)Nc140ccc(cc140)Nc141ccc(cc141)Nc142ccc(cc142)Nc143ccc(cc143)Nc144ccc(cc144)Nc145ccc(cc145)Nc146ccc(cc146)Nc147ccc(cc147)Nc148ccc(cc148)Nc149ccc(cc149)Nc150ccc(cc150)Nc151ccc(cc151)Nc152ccc(cc152)Nc153ccc(cc153)Nc154ccc(cc154)Nc155ccc(cc155)Nc156ccc(cc156)Nc157ccc(cc157)Nc158ccc(cc158)Nc159ccc(cc159)Nc160ccc(cc160)Nc161ccc(cc161)Nc162ccc(cc162)Nc163ccc(cc163)Nc164ccc(cc164)Nc165ccc(cc165)Nc166ccc(cc166)Nc167ccc(cc167)Nc168ccc(cc168)Nc169ccc(cc169)Nc170ccc(cc170)Nc171ccc(cc171)Nc172ccc(cc172)Nc173ccc(cc173)Nc174ccc(cc174)Nc175ccc(cc175)Nc176ccc(cc176)Nc177ccc(cc177)Nc178ccc(cc178)Nc179ccc(cc179)Nc180ccc(cc180)Nc181ccc(cc181)Nc182ccc(cc182)Nc183ccc(cc183)Nc184ccc(cc184)Nc185ccc(cc185)Nc186ccc(cc186)Nc187ccc(cc187)Nc188ccc(cc188)Nc189ccc(cc189)Nc190ccc(cc190)Nc191ccc(cc191)Nc192ccc(cc192)Nc193ccc(cc193)Nc194ccc(cc194)Nc195ccc(cc195)Nc196ccc(cc196)Nc197ccc(cc197)Nc198ccc(cc198)Nc199ccc(cc199)Nc200ccc(cc200)Nc201ccc(cc201)Nc202ccc(cc202)Nc203ccc(cc203)Nc204ccc(cc204)Nc205ccc(cc205)Nc206ccc(cc206)Nc207ccc(cc207)Nc208ccc(cc208)Nc209ccc(cc209)Nc210ccc(cc210)Nc211ccc(cc211)Nc212ccc(cc212)Nc213ccc(cc213)Nc214ccc(cc214)Nc215ccc(cc215)Nc216ccc(cc216)Nc217ccc(cc217)Nc218ccc(cc218)Nc219ccc(cc219)Nc220ccc(cc220)Nc221ccc(cc221)Nc222ccc(cc222)Nc223ccc(cc223)Nc224ccc(cc224)Nc225ccc(cc225)Nc226ccc(cc226)Nc227ccc(cc227)Nc228ccc(cc228)Nc229ccc(cc229)Nc230ccc(cc230)Nc231ccc(cc231)Nc232ccc(cc232)Nc233ccc(cc233)Nc234ccc(cc234)Nc235ccc(cc235)Nc236ccc(cc236)Nc237ccc(cc237)Nc238ccc(cc238)Nc239ccc(cc239)Nc240ccc(cc240)Nc241ccc(cc241)Nc242ccc(cc242)Nc243ccc(cc243)Nc244ccc(cc244)Nc245ccc(cc245)Nc246ccc(cc246)Nc247ccc(cc247)Nc248ccc(cc248)Nc249ccc(cc249)Nc250ccc(cc250)Nc251ccc(cc251)Nc252ccc(cc252)Nc253ccc(cc253)Nc254ccc(cc254)Nc255ccc(cc255)Nc256ccc(cc256)Nc257ccc(cc257)Nc258ccc(cc258)Nc259ccc(cc259)Nc260ccc(cc260)Nc261ccc(cc261)Nc262ccc(cc262)Nc263ccc(cc263)Nc264ccc(cc264)Nc265ccc(cc265)Nc266ccc(cc266)Nc267ccc(cc267)Nc268ccc(cc268)Nc269ccc(cc269)Nc270ccc(cc270)Nc271ccc(cc271)Nc272ccc(cc272)Nc273ccc(cc273)Nc274ccc(cc274)Nc275ccc(cc275)Nc276ccc(cc276)Nc277ccc(cc277)Nc278ccc(cc278)Nc279ccc(cc279)Nc280ccc(cc280)Nc281ccc(cc281)Nc282ccc(cc282)Nc283ccc(cc283)Nc284ccc(cc284)Nc285ccc(cc285)Nc286ccc(cc286)Nc287ccc(cc287)Nc288ccc(cc288)Nc289ccc(cc289)Nc290ccc(cc290)Nc291ccc(cc291)Nc292ccc(cc292)Nc293ccc(cc293)Nc294ccc(cc294)Nc295ccc(cc295)Nc296ccc(cc296)Nc297ccc(cc297)Nc298ccc(cc298)Nc299ccc(cc299)Nc300ccc(cc300)Nc301ccc(cc301)Nc302ccc(cc302)Nc303ccc(cc303)Nc304ccc(cc304)Nc305ccc(cc305)Nc306ccc(cc306)Nc307ccc(cc307)Nc308ccc(cc308)Nc309ccc(cc309)Nc310ccc(cc310)Nc311ccc(cc311)Nc312ccc(cc312)Nc313ccc(cc313)Nc314ccc(cc314)Nc315ccc(cc315)Nc316ccc(cc316)Nc317ccc(cc317)Nc318ccc(cc318)Nc319ccc(cc319)Nc320ccc(cc320)Nc321ccc(cc321)Nc322ccc(cc322)Nc323ccc(cc323)Nc324ccc(cc324)Nc325ccc(cc325)Nc326ccc(cc326)Nc327ccc(cc327)Nc328ccc(cc328)Nc329ccc(cc329)Nc330ccc(cc330)Nc331ccc(cc331)Nc332ccc(cc332)Nc333ccc(cc333)Nc334ccc(cc334)Nc335ccc(cc335)Nc336ccc(cc336)Nc337ccc(cc337)Nc338ccc(cc338)Nc339ccc(cc339)Nc340ccc(cc340)Nc341ccc(cc341)Nc342ccc(cc342)Nc343ccc(cc343)Nc344ccc(cc344)Nc345ccc(cc345)Nc346ccc(cc346)Nc347ccc(cc347)Nc34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Ideal Properties of Disinfectants / Antiseptics



Broad-Spectrum Activity
Kills Bacteria, Viruses & Fungi



Rapid Action
Works Quickly



Stable & Non-Toxic
Safe for Humans



**Effective in Presence of
Organic Matter**
Works in Dirty Conditions



Non-Corrosive & Non-Staining
Gentle on Surfaces



Economical & Easy to Use
Affordable & Convenient

The Alcohol Confusion Test

You disinfect your hands with 100% alcohol because “more is always better.” After a while, microbes start laughing instead of dying. Why?



- A. Microbes are immune to pure alcohol**
- B. 100% alcohol evaporates too fast and fails to penetrate**
- C. Alcohol becomes water-resistant**
- D. Microbes enjoy strong drinks**



Hurry-Up Disinfection

You spray disinfectant on a table and wipe it off immediately because you're late for class. What most likely happened to the microbes?



- A.** They were completely destroyed
- B.** They became superheroes
- C.** They experienced a sub-lethal effect and survived
- D.** They instantly mutated

MICROBIAL GYM SESSION

Low concentration disinfectant is used daily for a very short time.
Microbes exposed to this treatment will most likely:

- A. Die instantly
- B. Become weaker and disappear
- C. Get "training sessions" and survive better next time
- D. Start producing antibiotics

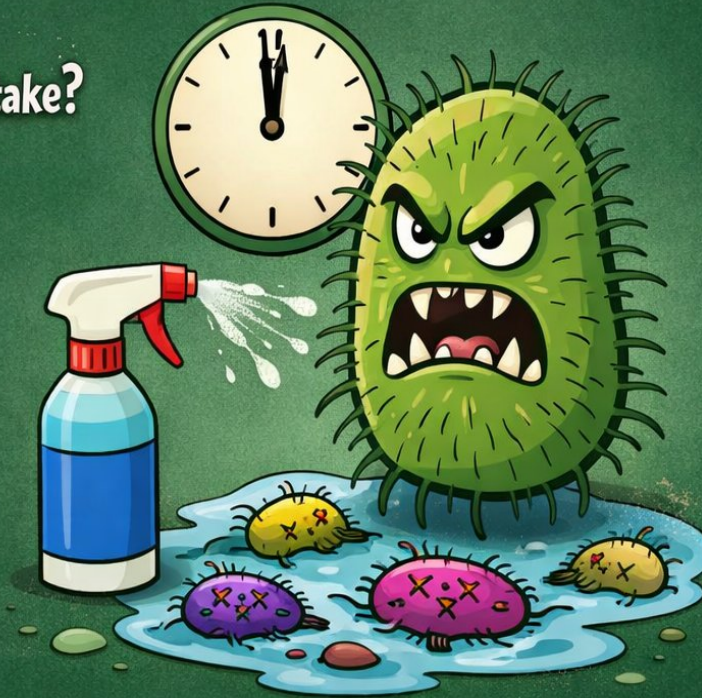


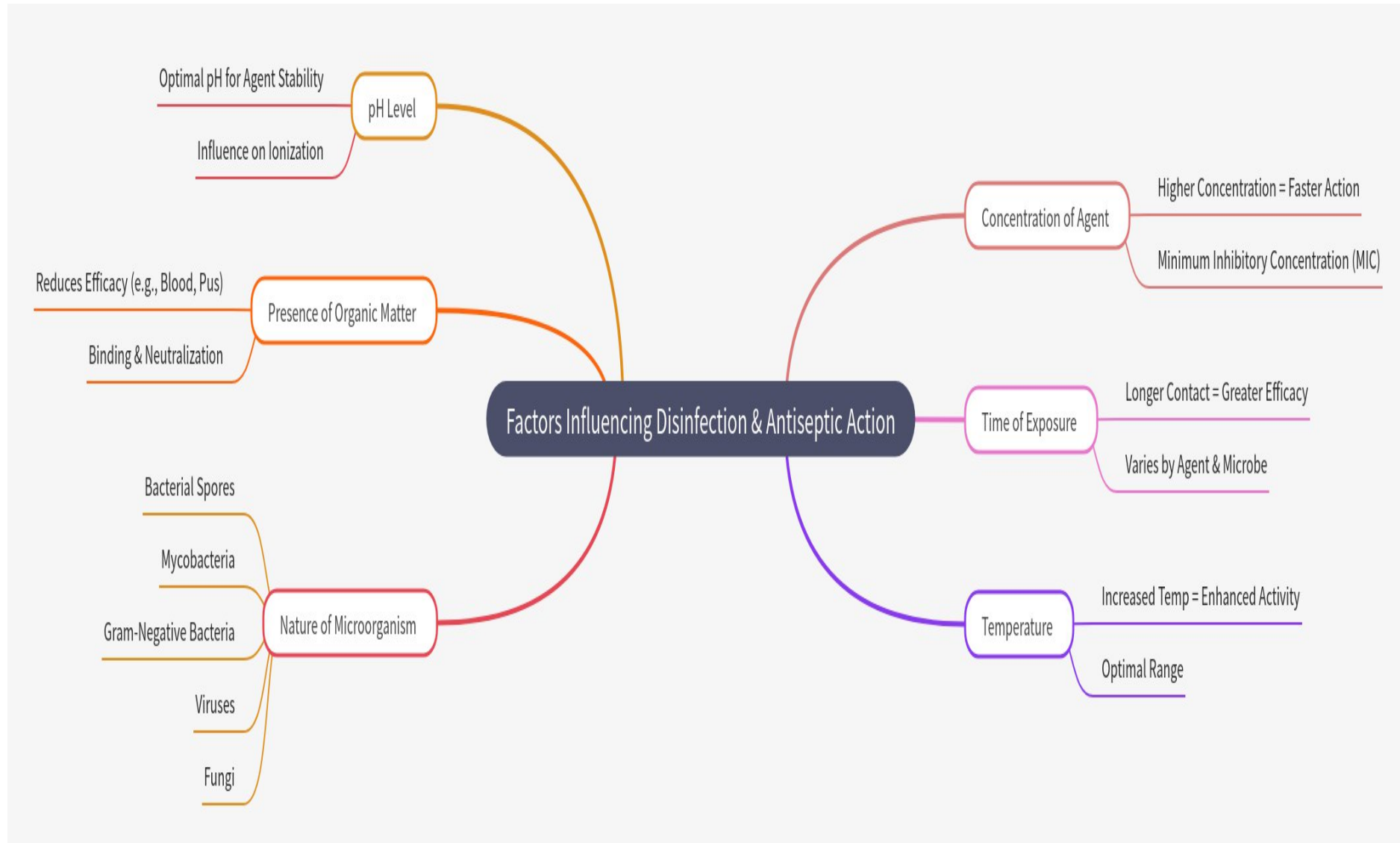
The 5-Second Rule (Microbiology Edition)

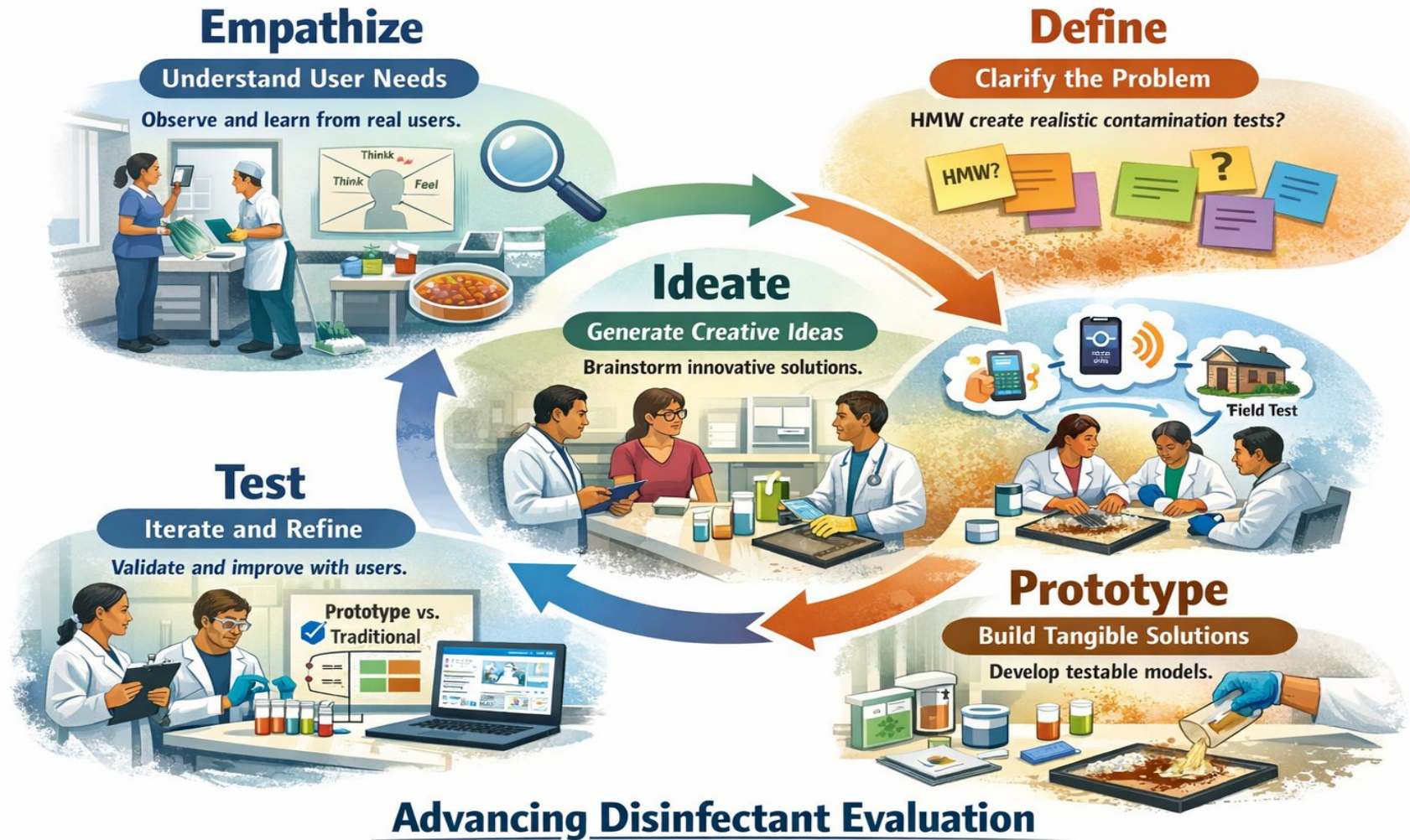
A disinfectant needs 10 minutes contact time,
but you give it only 1 minute.

What is the scientific name for this mistake?

- A.** Time optimization
- B.** Incomplete sterilization
- C.** Sub-lethal exposure
leading to survival
- D.** Speed disinfection technology







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