Analytical Methods for Total Coliforms and *E. coli*

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TCRDS Federal Advisory Committee Meeting

February 20, 2008

This Presentation

Objectives

Provisions

System compliance

Improvement

Research

Analytical Methods

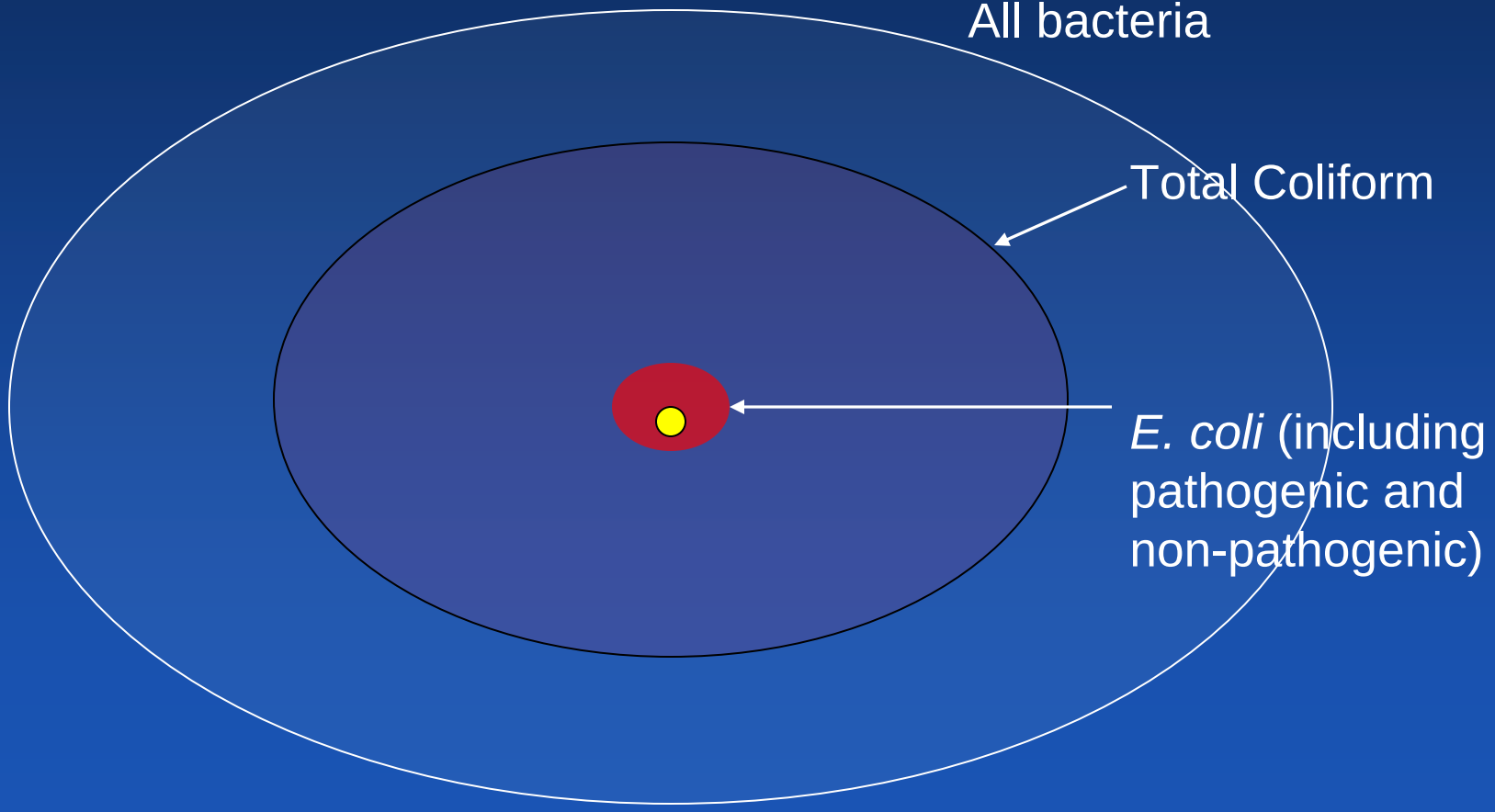
Objectives of Presentation

- Illustrate differences between coliform methods
  - Time to results
  - Accuracy
- Monitoring and analysis timeline

Approved Methods

- Two types of methods:
  - Methods based on growth and fermentation
  - Methods based on growth and enzyme production/activity
- Methods vary in terms of
  - Time to result
  - Whether get TC/EC results in one test or two
  - Accuracy
- See handout “Methods approved for the detection of total coliforms and *E. coli* under the current TCR” for full details

Coverage of Different Methods



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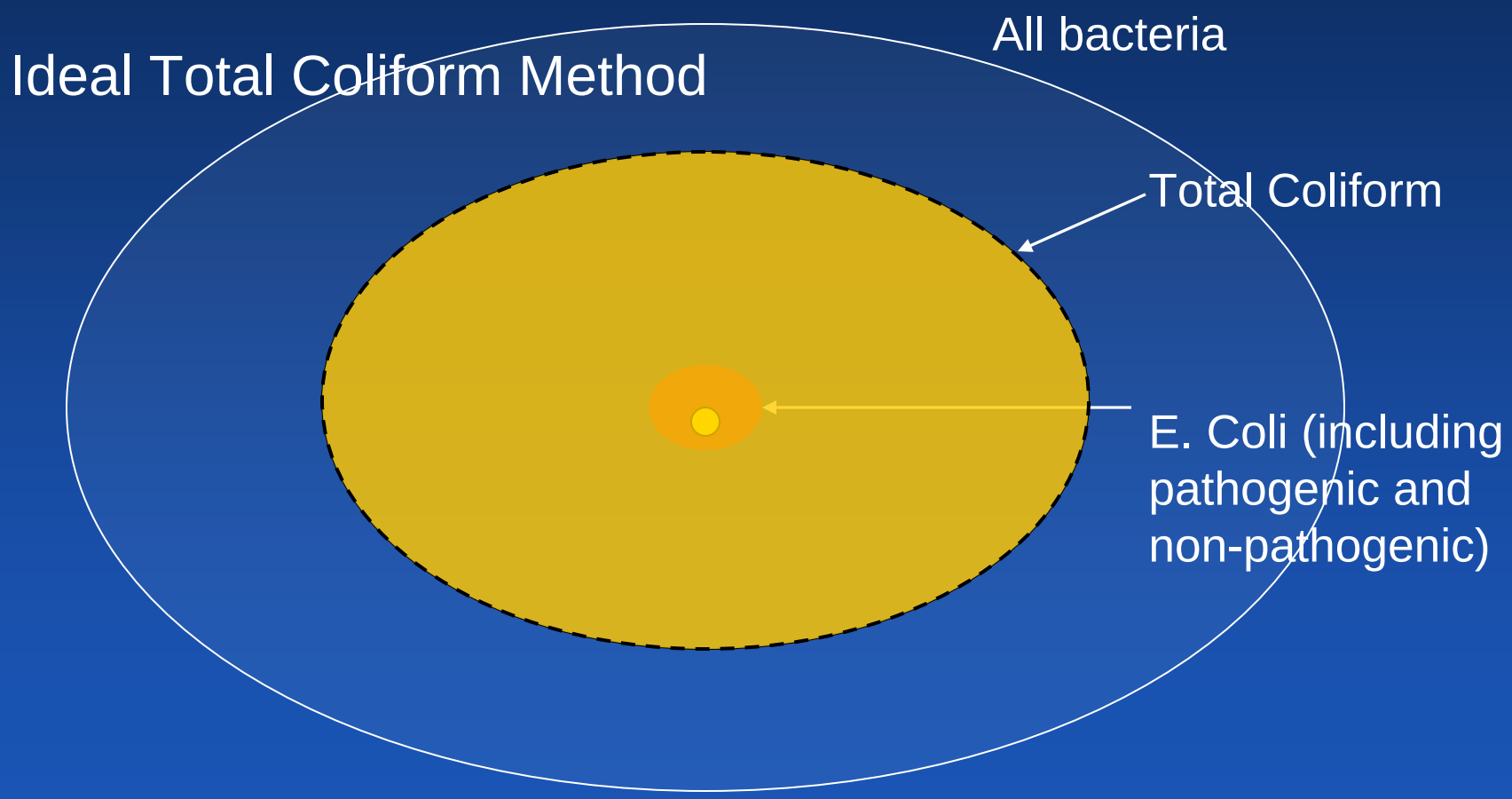

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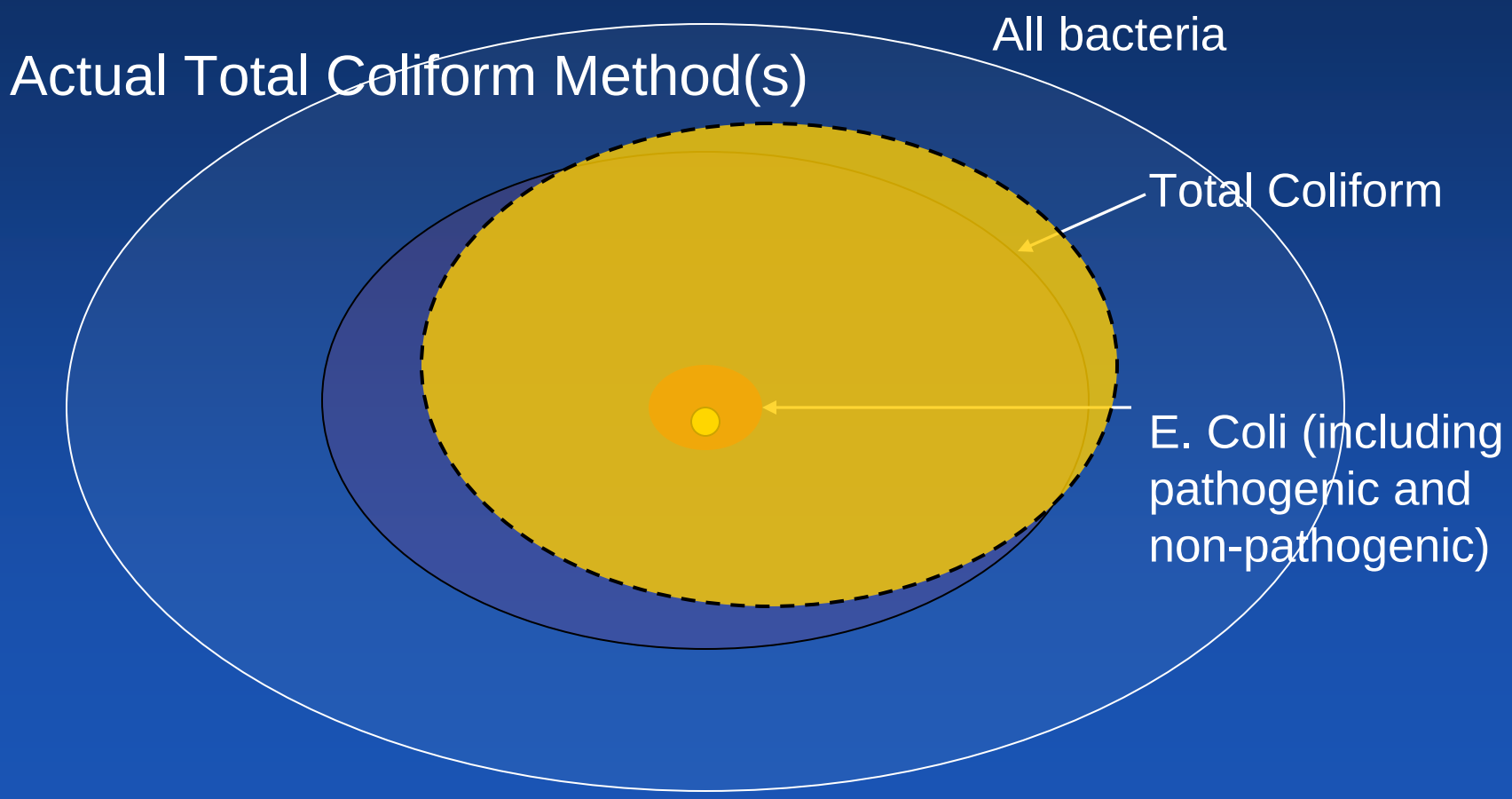

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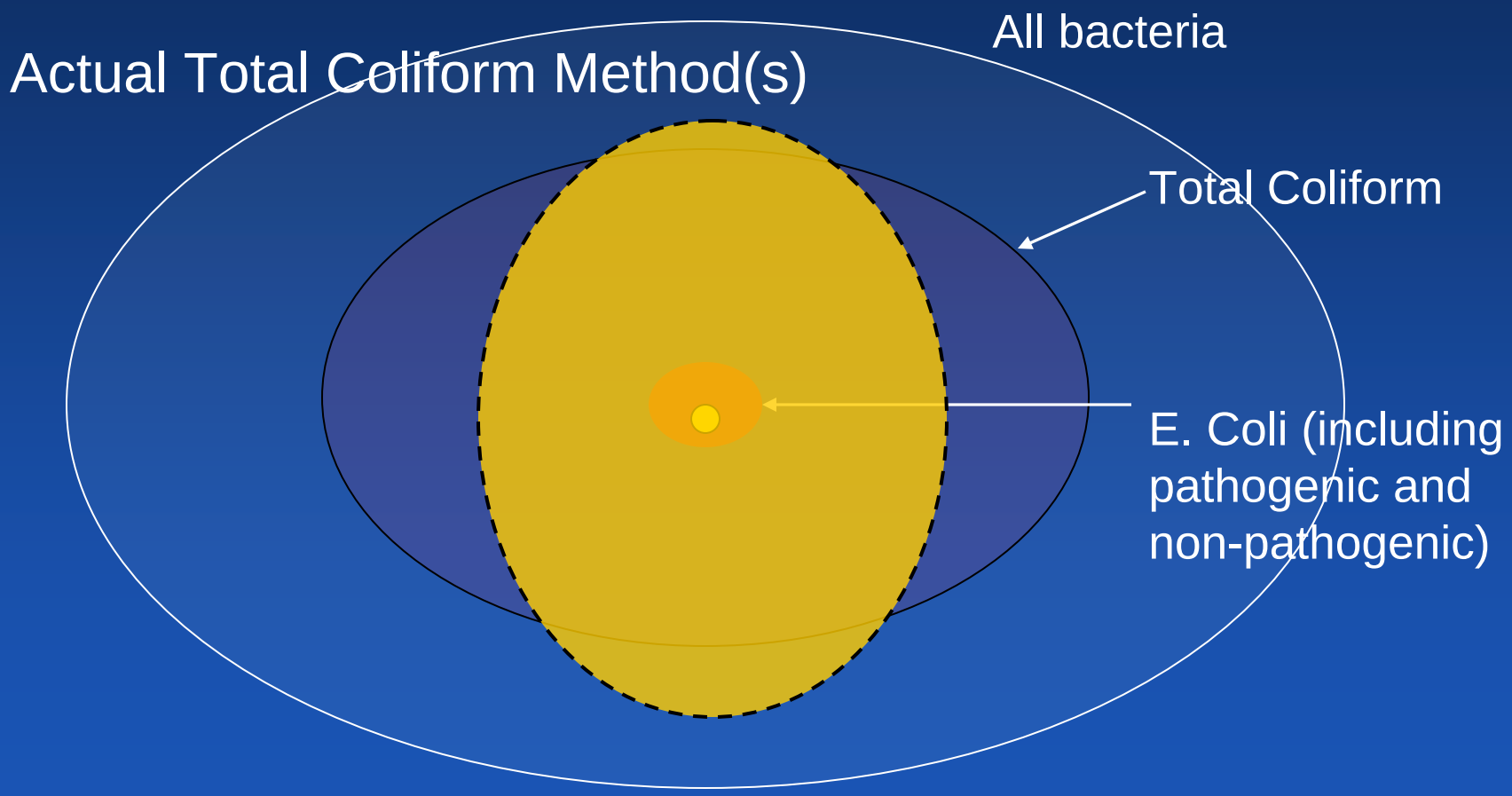

Method Performance Studies

- Recall from December meeting:
  - Data from the ATP shows that all coliform methods have varying levels of false positive and false negative results.
  - Olstadt (2007) and Fricker (2003 and 2007) demonstrate that the performance of coliform methods vary in the field.
    - Olstadt suggests that method performance can be influenced by water matrix effects.
  - Some studies have suggested that false positive results could be due to variations in the manufacture of the media.
    - Amount/type of antibiotic may be an issue under some circumstances.
- Preliminary data from ongoing AwwaRF study reinforces these same points

Factors Impacting Timeframe

- Analytical method
  - Time required to get results ranges from 18-48+ hrs
  - TCR specifies that the same method be used for routine and repeat samples

Factors Impacting Timeframe

- In-house vs. contract lab
  - TCR does not define a timeframe by which the laboratory must report results to the PWS
  - Some contract labs do not accept shipments Friday-Sunday
  - For affected systems, repeat samples are typically collected the week following a positive routine sample
    - Note: TCR specifies that repeat samples be collected within 24 hrs, but States have the flexibility to extend this timeline

Factors Impacting Timeframe

- Reporting method
  - Electronic
  - Phone
  - Email
  - Hard copy in regular mail

Factors Impacting Timeframe

- Scheduling/Shipping
  - Small, rural systems may have difficulty scheduling someone to collect repeat samples or making shipping arrangements in the 24 hrs specified in the TCR
  - Systems using contract labs do not always stock extra sample bottles so repeat samples require shipment of bottles from lab before sampling

Timeframe Scenarios

Time* until:	Fastest Possible Scenario (In-house lab, 24-hr method)	Longer Scenario (Contract lab closed on weekends, 48-hr method)
Routine Sample Collected	Monday	Monday
Notified of routine TC+/EC+	Tuesday afternoon (Day 2)	Thursday afternoon (Day 4)
Collect repeat samples	Wednesday morning (Day 3)	Monday morning (Day 8)
Notified of repeat TC+/EC+	Thursday afternoon (Day 4)	Thursday afternoon (Day 11)
Public Notification	Friday afternoon (Day 5)	Friday afternoon (Day 12)

* Note: Times can vary depending on other factors listed in previous slide but not included in these examples.

Summary of Fermentation Methods

Method	Time to Result	Simultaneous TC & EC in Initial Test
Presence-Absence Broth	TC: 48-96 hrs EC: 48-72 hrs	No
Multiple-Tube Fermentation	TC: 48-96 hrs EC: 48-72 hrs	No
M-endo / Membrane Filtration	TC: 48-72 hrs EC: 48 hrs	No
M-ColiBlue	TC: 24 hrs EC: 24 hrs	Yes

Summary of Enzyme Methods

Method	Time to Result	Simultaneous TC & EC in Initial Test
MI medium	TC: 24 hrs EC: 24 hrs	Yes
Coliscan	TC: 24 hrs EC: 24 hrs	Yes
Colilert 18	TC: 18-22 hrs EC: 18-22 hrs	Yes
Colilert	TC: 24-28 hrs EC: 24-28 hrs	Yes
Colisure	TC: 24-48 hrs EC: 24-48 hrs	Yes
Readycult	TC: 24 hrs EC: 24 hrs	Yes
Colitag	TC: 24 hrs EC: 24 hrs	Yes
E*Colite	TC: 28 hrs EC: 28 hrs	Yes

Conclusions

- TWG recommends reviewing the method approval (ATP) criteria
  - Criteria needs to be relevant to frequency of detection of organisms in drinking waters (including matrix effects)
  - Differences in methods may be important and need to be investigated
- Impact of time on recovery of injured organisms and die-off (method time, holding time)
- Choice of analytical method influences false+/false- rates in the field but we do not have data to understand this
- Methods alone do not address the other factors that add to the overall timeframe to get to notification under the current rule

Conclusions Continued

- For rule options, need to consider the methods in the context of the consequences of the results, for example:
  - If TC is not linked to MCL and public notification, does TC method accuracy become more/less important?
  - If TC is linked to a follow-up action, does time to result become more/less important than accuracy?
  - Should different performance criteria be developed EC methods than for TC methods?
  - Is there an impact on current users of specific methods and the manufacturers?