WORK INSTRUCTIONS

2.4.3 Qualitative Confirmation of Benzodiazepines in Urine

REFERENCE MATERIAL

Benzodiazepine Positive Control

- *Stock Solutions*
  1 mg/mL single component or 250µg/mL multicomponent benzodiazepine class reference solutions.
- *Direct spiking*
  Spike negative urine with 3µL of each single component stock or 12µL of 250µg/mL stock solution.
- *Working Control Solution (10ng/µL)*
  (Note: A minimum of two benzodiazepine class compounds must be included in the control solution. One of these compounds must form a TMS derivative.)

  Add 100µL each 1mg/mL or 400µL 250µg/mL Stock Solution to ≅ 9mL MeOH in 10mL ball flask. Bring up to volume with MeOH. Solution is stable for one-year when stored under refrigeration.

Conjugated Controls

- Commercially obtained conjugated control (example: Bio-Rad C3)

- *Stock Solution*
  1 mg/mL Oxazepam Glucuronide or 100µg/mL Lorazepam Glucuronide or 100µg/mL Morphine Glucuronide

- *Direct spiking*
  Spike negative urine with 3µL of 1mg/mL stock solution or 30µL of 100µg/mL stock solution.

- *Working Glucuronide Solution (10ng/µL)*
  Add 100µL of 1mg/mL Stock Solution (or 1mL 100µg/mL Stock Solution) to 10mL MeOH. Solution is stable for one year when stored under refrigeration.

Internal Standard

- *Stock Solution*
  1mg/mL Prazepam.

  Direct addition
  Add 3µL of 1mg/mL stock solution to each 6mL urine sample and control.

- *Working Internal Standard Solution*
  Add 100µL Stock Solution to ≅ 9mL MeOH in 10mL ball flask. Bring up to volume with MeOH. Solution is stable for one year when stored under refrigeration.

COMMENTS

This method has instructions for the preparation of both an underivatized and a TMS-derivatized extract. The analyst has the option of preparing either or both of these extracts.

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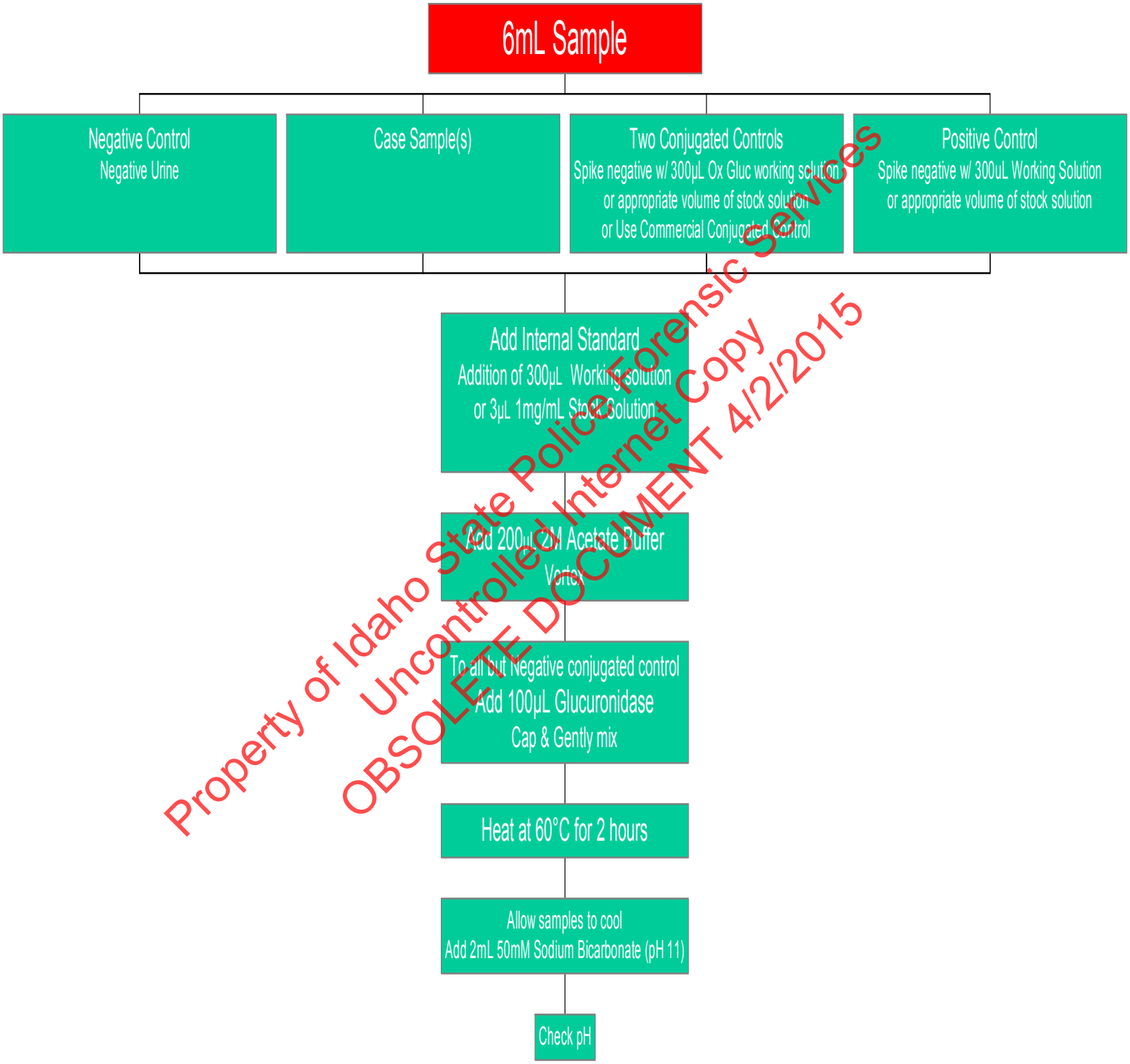
2.4.3 Qualitative Confirmation of Benzodiazepines in Urine

SAMPLE PREPARATION: Glucuronide Cleaving



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WORK INSTRUCTIONS

2.4.3 Qualitative Confirmation of Benzodiazepines in Urine

SAMPLE PREPARATION: Extraction and Reconstitution and/or Derivatization

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