

**1. Procedure summary**

This procedure describes how to maintain culture nutrition at the IABR in Columbus, NM.

**1.1. Related Procedures**

Safe traffic flow

CB-03-006-001

Nurse Trailer Operation

CB-01-002-009

**1.2. Procedure impacts and concerns**

Safety

Personnel must be fully equipped with appropriate PPE in regards to each chemical's MSDS.

PPE: Standard PPE + N95 Respirator, Nitrile gloves

Quality

Inaccurate concentrations of nutrient addition could lead to toxicities or deficiencies that will result in changes in culture health.

Delivery

NA

Environmental

Loss of containment

Cost

NA

Compliance

Compliance with OSHA's Hazardous Waste Operations and Response, and Hazardous Communication Standard in addition to the Sapphire Energy, Inc. Chemical Hygiene Plan is required. See 29 CFR 1910.120 and 1200. An AUL list, MSDSs and label information will be available for easy reference in a binder in the administration building

**1.3. Responsibilities and owners**

Document Owner

Manage contentment and distribution

Jose Perez

Process Owner

Responsible for content and process validation

Rebecca White

Site Manager

Responsible for implementation and conformance

Rebecca White

**2. Process****2.1. Process description**

Pond health and productivity can be affected by fluctuations in concentrations of essential nutrients. Nutrient Replenishment is necessary to maintain target nutrient concentrations that are depleted as the culture grows and consumes these nutrients. This procedure describes the process of replenishing nutrients.

## 2.2. Process diagram: Work Instruction

## 2.3. Process steps

### 2.3.1 Nutrient Replenishment

#### 2.3.1.1 Alkalinity Addition

**2.3.1.2.** Transport necessary alkalinity component(s) to pond using F-450 flat bed or flatbed trailer make sure that flat bed or flatbed trailer are not overloaded. Refer to safety traffic flow R2 for traveling in the vicinity.

**Note:** If the f-450 is not available use flatbed trailer. Or you may use both.

**2.3.1.3.** Stage super sacks of bicarbonate and sodium sulfate at designated area.



**2.3.1.4.** Dispense alkalinity powder into pond extending boom using tele handler behind the paddle wheel.



**Note:** If you only need to use half a supersack, cut bag in the center and tilt tele-handler front and back.

**NOTE:** If you do not extend boom out on tele-handler immediately after opening supersack chemicals will spill on ground causing a loss of containment.

**NOTE:** Amount of alkalinity added to each pond is dependent on QAQC.

**2.3.1.5.** Rinse liner using nurse trailer with fresh water after dispensing sodium bicarb or sodium sulfate into pond. Refer to nurse trailer operation procedure SOP.



**NOTE:** If Wagner Pump available you may use to help dissolve and rinse liner.

**2.3.1.2. Nutrient Addition** (and other nutrients in 50 to 55 lb. bags)



**2.3.1.2.1.** Stage 50 lb. bags to where the ends of the bags are on pond liner slope.



Figure –Shows where bags are laid out when staging.

**2.3.1.2.2** Cut bags to dispense into the pond and rinse nitrate beads into pond using nurse trailer with freshwater. Refer to Nurse trailer operation procedure SOP.

**NOTE:** Clean up bags as you go.



### **2.3.1.3. Phosphate Addition**

**2.3.1.3.1.** Refer to Loading nurse trailer from enclosed system SOP for loading Phosphates into nurse trailer and Safe traffic flow procedure for traveling in between ponds.

**2.3.1.3.2.** Dispense phosphate solution into adjacent tanks at each pond individually. According to feeding sheet.

**2.3.1.3.3.** Open valve so that phosphate solution will dispense into culture slowly.

**2.3.1.3.4.** After all ponds have been fed and phosphate has dispensed completely into pond triple rinse adjacent tanks with fresh water using acid nurse trailer to ensure that adjacent tanks and nurse trailer are both triple rinsed.

**2.3.1.3.5.** Turn in completed feeding sheet to team lead or supervisor when task completed.

**NOTE:** Open valve **AFTER** desired amount of phosphate is dispensed into adjacent tank.

#### **2.3.1.4. Trace and Iron Addition**

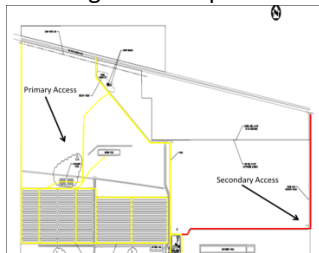
**2.3.1.4.1.** Refer to Trace and Iron Stock Solution Mixing for details on mixing stock solutions.



**NOTE:** Iron color- Red  
 Trace color- Dependent on recipe sheet.

**2.3.1.4.1.2.** Pump Stock solutions into respective portable tanks on Trace and iron trailer. Refer to trace metal and iron stock mixing station.

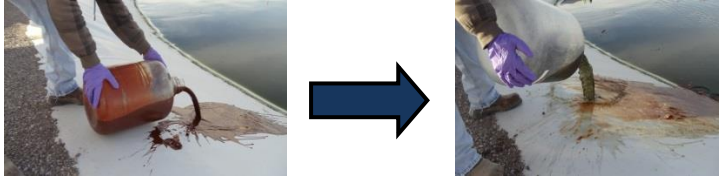
**2.3.1.4.1.3.** Refer to safe traffic flow Procedure for details on traveling between ponds.



**2.3.1.4.1.4.** When at desired pond location open valve on respective portable tanks on trace and iron trailer and begin dispensing stock solution into graduated cylinder. See nutrient calculation sheet for proper amount according to pond number.



**2.3.1.5.** Use graduated cylinder to dispense solution near adjacent tank on liner into pond. Always dose iron first then trace at same location.



**2.3.1.5.1.** Rinse liner using fresh water dispensing tank on trailer.



#### **2.3.1.6 UAN-28 and Urea Addition**

**2.3.1.6.1.** Refer to Loading nurse trailer from enclosed system procedure and Safe traffic flow Procedure SOP for traveling between ponds.

**2.3.1.6.2.** Dispense nitrogen(s) into adjacent tanks at each pond individually. According to feeding sheet.

**2.3.1.6.3.** Open valve so that nitrogen solution will dispense into culture slowly.

**2.3.1.6.4.** After all ponds have been fed and nitrogen has dispensed completely into pond triple rinse adjacent tanks with fresh water using same nurse trailer to ensure that adjacent tanks and nurse trailer are both triple rinsed.

**2.3.1.6.5.** Turn in completed feeding sheet to team lead or supervisor when task completed.

**NOTE:** Proper PPE required dependent on feeding urea or UAN-28.

#### **Volume Calculations**

**2.3.1.6.1** Determine volume of pond that you will be dosing based on the current operating depth and the corresponding strapping table.

##### **2.3.1.6.2 Nutrient Analysis and Calculations**

**2.3.1.6.3** Obtain Alkalinity, NO<sub>2</sub>/NO<sub>3</sub>, PO<sub>4</sub>, Urea/Ammonia concentration output from lab QA/QC. (Communicated by Lab, Team Lead or Director of Cultivation).

**NOTE:** Open valve AFTER desired amount of nitrogen is dispensed into adjacent tank.

**Required documents****2.4. Input documents**

Alkalinity Sheet  
Phosphate Sheet  
Trace and iron Recipe  
Urea Sheet  
UAN-28 Sheet

**2.5. Output documents**

Production Log  
Replenishment Sheet

**3. Document control****3.1. Revision history**

R0 – Initial Release – Andy Randall (Rob McBride)	02-16-2012
R1 – Adriana Rascon	07-03-2012
R2-Adriana Rascon	09-20-2012
R3-Magdalena Pacheco	01-22-2015

**3.2. Document approval****3.3. Document reviewers****4. Risk analysis**

&lt;Risk name&gt;

&lt;Mitigation plan&gt;

&lt;Owner&gt;

&lt;RPN&gt;