THE USE OF CO2 IN CANNABIS CULTIVATION AND EXTRACTION

CONSORTIUM MANAGEMENT GROUP
CARBON DIOXIDE (CO2) MOLECULE

- Carbon dioxide (CO2) is a colorless and odorless gas that is essential to life on Earth.

- Natural sources of CO2 include ocean-atmosphere exchange, plant and animal respiration, soil decomposition, and volcanic eruptions.

- Current atmospheric CO2 levels range between 300-400 parts per million (PPM).

- Plants, algae, and cyanobacteria use light energy to photosynthesize carbohydrate and oxygen from carbon dioxide and water.
**PHOTOSYNTHESIS**

**LIGHT REACTION:**
Light energy is used to convert water and other compounds into oxygen and chemical energy (NADPH, ATP)

**CALVIN CYCLE:**
The chemical energy produced from the Light Reaction is used to convert carbon dioxide and water into carbon compounds which eventually form glucose.
CALVIN CYCLE VS PHOTORESPIRATION

- Chemical energy that is produced from the Light Reaction is used by the enzyme RuBisCO in carbon fixation (Calvin Cycle) or oxygen fixation (Photorespiration).

- Although both reactions eventually produce precursors for glucose, Photorespiration produces these precursors at a reduced rate and higher metabolic cost compared to the Calvin Cycle.

- Photorespiration occurs roughly 25% of the time and results in a net loss of carbon.

- CO₂ enrichment increases the concentration of carbon dioxide and pushes the reaction to favor CO₂-fixation (Calvin Cycle) over O₂-fixation (Photorespiration).
STOMATA: Gas and Water Exchange

- Stomata are pores found in the epidermis of leaves, stems, and other organs used to control gas exchange.
- Stomata can either open to let CO2 in and release H2O and O2, or it can close to conserve H2O.
- Under normal atmospheric concentrations of CO2 (300 PPM), the gain of CO2 also results in the loss of H2O.
- High concentrations of CO2 (1500 PPM), lead to less stomata openings, which optimizes CO2 gain with minimal H2O loss.
- A benefit of CO2 enrichment is allowing for optimal CO2 diffusion with minimal water loss.
CO2 VS GROWTH RATE

- Below 200 PPM, the concentration of CO2 is too low for effective photosynthesis and no substantial growth is observed.
- The 300 PPM range was determined as the ambient atmospheric concentration, and the growth rate at this concentration was chosen to be the 100% growth point.
- At the 1000 to 1500 PPM range, optimal growth rate was observed.
- After 1500 PPM, the percentage growth rate was observed to decrease.
- At around 2000 PPM, the percentage growth rate falls below 100%.
CO2 ENRICHMENT OF GROW ROOMS

- Once other horticultural factors have been optimized (light, nutrients, plant health), CO2 becomes an important factor in increasing crop yield.

- Through its effects on improving the rate of Calvin Cycle reactions and Stoma regulation, CO2 enrichment can increase normal plant growth from 20% to 100%.

- There are various methods of introducing CO2 into indoor cultivation facilities, including: CO2 injection systems, CO2 burners, yeast fermenters, and dry ice.
CO2 ENRICHMENT SYSTEMS:

1. CO2 Injection Systems: CO2 tanks in conjunction with pressure regulators, solenoid valves, and timers can be used to inject CO2 at regular intervals.

2. CO2 Burners: Hydrocarbons such as butane, propane, and natural gas are used in specialized burners that maximize CO2 production and minimize heat generation.

3. CO2 Fermenters: Brewer’s yeast, sugar, and water is combined to produce alcohol and CO2 through fermentation.

4. Dry Ice: Solid dry ice is housed in a container over the canopy, and CO2 flows onto the plant as dry ice sublimates.
CO2 TANK SIZES

- Average pressure @70-75°F: 850-900 psi.
- Safe working pressure of tanks exceeds 1800 psi.
- Burst tested at 3800 psi.
- Common sizes from 2.5lbs to 100lbs.
- Industrial sizes available from 300lbs to 750lbs.
- Commercial uses include:
  - Carbonation of beverages
  - De-caffeinate coffee beans
  - Flavor and fragrance manufacturing
  - Waste water management, re-mineralization of desalted water.
  - Chemical production
  - Welding
  - Semi conductor cleaning

High Pressure Tanks By Size
Average Pressure 850 - 900 psi @ 70°F - 75°F
Safe working pressure exceeds 1800 psi. Burst Tested @ 3800 psi.
CO2 EXPOSURE LEVELS

CO2 Levels
- 300 ppm (.03%) Atmospheric conc.
- ~1,000 ppm (.10 %) in Extraction Room.
- ~1,500 ppm (.15%) in Cultivation Room.
- 5,000 ppm (.50 %) Sensor Triggered to Activate Purging of Cultivation and Extraction Rooms.
- 5,000 ppm (.50 %) OSHA Permissible Exposure Limit (PEL), 8 hours.
- 30,000 ppm (3.00 %) OSHA Short Term Exposure Limit (STEL), 10 minutes.
- 50,000 ppm (5.00 %) NIOSH Intoxication after 30 minute exposure.
- 90,000 ppm (9.00 %) NIOSH Lethal Concentration Low, 5 minutes.
CO2 SAFETY: Monitoring Equipment and Excess Flow Valves

Monitoring Equipment
- Link4 and Atlas are manufacturers of hydroponic control systems that monitor environmental variables such as the ambient concentrations of CO2.
- Purge fans can be connected to monitoring systems, that will trigger a room purge if certain PPM levels are reached.

Excess Flow Valves
- Examples of Excess Flow Valves (EFVs) include ball and magnet designs or spring designs.
- In-line devices to limit the upstream or downstream flow when extreme under/over flow rates occur.
SIMPLIFIED CO2 ENRICHMENT SCHEMATIC

1. CO2 ROOM will house CO2 tanks that range up to 750lb capacity. Each tank is fitted with Restrictive Flow Orifice (RFO) that will limit flow to 240CuF/H. Tanks are anchored to the wall.

2. SAFETY MANIFOLD contains a vent line, Excess Flow Valve (EFV), and Safety Shut Off Valve connected to Alarm System.

3. LINK4 SYSTEM is the environmental monitoring system and dosing controller.

4. ENRICHMENT PANEL will contain RFO, EFV, and manual shutoff valves.

5. CULTIVATION ROOM contains CO2 monitors, alarms, and emergency purge fans.
CANNABIS AND CANNABINOIDs

- Cannabis is the genus of flowering plants that include the species Cannabis indica, Cannabis sativa, and Cannabis ruderalis.

- Trichomes are resin glands produced by the cannabis plant. They contain the highest concentration of cannabinoids.

- Cannabinoids are a class of chemical compounds produced by both plants (phytocannabinoids) and by humans (endocannabinoids).

- Cannabinoids such as THC, CBD, and CBN have proven effective in the therapeutic and palliative care of certain diseases and ailments.
BASIC SOLVENT EXTRACTION OF CANNABINOID OVERVIEW

CANNABIS + SOLVENT → FILTERATION → CANNABINOID + SOLVENT → HEAT/EVAPORATION → CANNABINOID (THC, CBD, CBN)

EXHAUSTED PLANT MATTER → SOLVENT
SOLUTE, SOLVENTS, AND SOLVATION

- Solutes are the dissolved substances in a solution. The target solute in this case are cannabinoids.

- Solvents are substances that dissolve solutes in a solution. Commonly used solvents in the extraction of cannabinoids include: butane, ethanol, and CO2.

- Solvation is the dissolution of solute molecules by the surrounding solvent molecules. The homogenous mixture of solute and solvents is called a solution.
‘LIKE DISSOLVES LIKE’: POLAR AND NONPOLAR COMPOUNDS

- Covalent bonds are a type of chemical bond in which atoms share electrons.

- The differences in electronegativity between covalently bonded atoms determine if the bond is polar (unequal sharing of electrons) or nonpolar (equal sharing of electrons).

- Cannabinoids are non-polar compounds. They are primarily hydrocarbons that have no polarity in the sharing of electrons in their chemical bonds.

- Non-polar solvents are generally needed for non-polar solutes. Water (polar) and oil (nonpolar) do not mix.
HEALTH AND SAFETY RATINGS OF COMMONLY USED CANNABIS EXTRACTION SOLVENTS

**CARBON DIOXIDE**

- **BUTANE**
  - PEL: 1,000 PPM
  - IDLH: 19,000 PPM

- **ETHANOL**
  - PEL: 1,000 PPM
  - IDLH: 15,000 PPM

- **LIQUID CO2**
  - PEL: 5,000 PPM
  - IDLH: 40,000 PPM

- **GASEOUS CO2**
  - PEL: 5,000 PPM
  - IDLH: 40,000 PPM

PEL = PERMISSIBLE EXPOSURE LIMIT (OSHA)
IDLH: IMMEDIATELY DANGEROUS TO LIFE OR HEALTH (NIOSH)
BUTANE EXTRACTION

- Butane is extremely flammable and the low barrier of entry for Butane (BHO) extraction poses a public safety risk due to the potential for untrained and improper extraction practices.

- Butane is an attractive solvent for DIY’ers due to its low startup cost and readily available equipment.

- Simple BHO extraction setup:
  - Glass tube with tapered opening
  - Coffee filters
  - Butane canister
  - Pyrex plate for collecting extract
ETHANOL EXTRACTION

- High proof alcohol is used to dissolve cannabinoids from cannabis, and the solution of alcohol and cannabinoids is separated from the raw plant material.

- Alcohol is then distilled from the solution, leaving a highly concentrated cannabinoid product.

- Alcohol is less selective than butane, requiring more nuanced methodologies to produce a high quality extract.
CO2 EXTRACTION

- CO2 is readily available, inexpensive, nontoxic, nonflammable, chemically inert under many conditions, environmentally acceptable, and liquefiable at reasonable pressures and temperatures.

- In order for CO2 to be a viable solvent for cannabinoids, it needs to reach a supercritical fluid state in which it displays the properties of both a liquid and a gas. This is achieved through temperature and pressure control.

- CO2 extraction system tend to be more expensive (~$160K model pictured), complex, and acquisition is heavily restricted. The higher requirements for acquisition limits unqualified usage.
CARBON DIOXIDE (CO₂) MOLECULE

- The Carbon – Oxygen bond is polar, but the linear geometry of the molecule results in “no net dipole moment”, and makes CO₂ a nonpolar molecule.

- “Like dissolves like”, nonpolar solvents can be used to extract cannabinoids and other oils from the cannabis plant.

- CO₂’s low molar mass and boiling point aids in the solvent/solute separation and purification process.

(a) No net dipole moment

1. **Molar mass**: 44.01 g/mol
2. **Formula**: CO₂
3. **Boiling point**: -109.3°F (-78.5°C)
4. **Melting point**: -68.08°F (-55.6°C)
SUPERCritical FLUID
CO2

- CO2 is in a Supercritical Fluid (SCF) state at 88 degrees Fahrenheit and 1073 PSI.
- Compared to H2O, which reaches a SCF state at 705 degrees Fahrenheit and 3210 PSI.
- SCF CO2 exhibit both gas-like properties (high diffusivity and low viscosity) and liquid-like properties (high relative density and rate of absorption).
- SCF CO2 allows for waste minimization and solvent recycling.
- SCF CO2 is currently being used in the industrial extraction of caffeine from coffee beans. Providing a safer solvent alternative to dichloromethane (carcinogenic).
OVERVIEW: CLOSED LOOP CO2 CYCLE

1. CO2 is pumped from the CO2 Tank into the Accumulator Vessel
2. CO2 is pumped from the CO2 Tank into the Extraction Vessel
3. CO2 and Extract mixture is pumped into the Separator Vessel 1
4. CO2 and Extract mixture is separated, Extract stays in Separator Vessel 1, and CO2 is pumped to Separator Vessel 2
5. CO2 is pumped back to Accumulator Vessel for condensation to Liquid State
6. CO2 is pumped back to Extraction Vessel, completing a closed loop cycle.
EXTRACTION VESSEL

- **PRESSURE**
- **TEMPERATURE**
- CO2 is brought to a Supercritical State in the Extraction Vessel, solvating non-polar compounds including: cannabinoids, terpenes, and plant waxes.
- Raw plant material is packed into the extraction vessel. 20L (~9lbs) raw material capacity.
- Safety mechanism prevents opening the vessel without proper depressurization.
- Connected to Water Bath for temperature manipulation.
- **1500-2000 PSI** target use
- **100 F**
SEPARATOR VESSEL 1 & SEPARATOR VESSEL 2

- **PRESSURE**
- **TEMPERATURE**
- CO2 is separated from the targeted extraction compounds in the Separator Vessel.
- CO2 separates from the solution and is pumped to Accumulator Vessel.
- Cannabinoids, terpenes, and plant waxes collect in Separator Vessel 1 for retrieval.
- Separator 2 has a pressure relief valve to vent excess CO2, this will be connected to a hose that will vent CO2 out.
- Connected to Water Bath for temperature manipulation.
- **1000-1500 PSI** target use
- **135 F**
ACCUMULATOR VESSEL

- ↓ PRESSURE
- ↓ TEMPERATURE
- CO2 is condensed back into a Liquid State in the Accumulator Vessel.
- CO2 from the Accumulator Vessel is pumped back into the Extraction Vessel, completing a full closed loop cycle.
- Connected to Water Chiller for temperature manipulation.
- 500-1000 PSI target use
- 45 F
OVERVIEW: CLOSED LOOP EXTRACTION

- CO2 is turned into a supercritical fluid in the Extraction Vessel, and is used to solvate cannabinoids.
- The SCF CO2 and cannabinoid mixture flows to the Separator Vessel 1 and 2 for separation of CO2 and cannabinoids.
- Cannabinoids is collected in Separation Vessel 1.
- CO2 flows to the Accumulator Vessel and is collected back into a liquid state.
- Liquid CO2 flows is pumped back into Extraction Vessel for further solvation of cannabinoids, completing one closed loop cycle.
CO2 EXTRACTED CANNABIS OILS

- CO2 extracted Cannabis Oil can be used as the active component for various ancillary products, including: Tinctures, Vape Pens, Capsules, Ointments, and Salves.

- Capsules and Tinctures provide an easy to dose delivery method for MMJ patients that cannot smoke due to respiratory concerns.

- Ointment and Salves work well for topical ailments.

- Vape Pens provide an alternative to those who prefer to smoke but are concerned with inhaling byproducts of combusting the cannabis plant.