This guidance note addresses some specific issues concerning the safe use of carbon dioxide in laboratories and outlines some possible scenarios and their consequences. It should be noted that the control measures for minimising risks associated with the use of CO\(_2\) are essentially the same as those for other non-toxic, non-flammable gases. The guidance note complements Guidance Note 25: Safe Handling, Use and Storage of Compressed Gases.

Part 1 – Bottled and Piped Sources of CO\(_2\)

1. General Uses
Carbon dioxide is a common industrial gas used extensively in laboratories throughout the College. In particular, it is used widely within the Faculties of Medicine and Life Sciences as a feed gas for CO\(_2\) incubators employed for tissue culture.

2. Properties and Hazards
   - Colourless gas.
   - Present in the air we breathe at a concentration of approximately 300ppm (parts per million) or 0.03%.
   - Odourless at low concentrations but characteristic odour at higher concentrations (see Table 2).
   - Non-flammable – though like any other compressed gas, exposure to fire may cause the cylinder to rupture or explode.
   - The gas / vapour is heavier than air – relative density = 1.52 (air = 1). May accumulate in confined spaces, particularly at or below ground level.
   - Capable of causing asphyxiation at high concentrations (see Table 2).
   - Though CO\(_2\) is a non-toxic gas, it does have an occupational exposure limit assigned to it under the Control of Substances Hazardous to Health Regulations 1999 as indicated in Table 1:

<table>
<thead>
<tr>
<th>Occupational Exposure Standard (OES)</th>
<th>Concentration (ppm)</th>
<th>Concentration (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Short term exposure limit (STEL)</td>
<td>15000</td>
<td>1.5</td>
</tr>
<tr>
<td>Long term exposure limit (LTEL)</td>
<td>5000</td>
<td>0.5</td>
</tr>
</tbody>
</table>

Occupational Exposure Standards are set to help protect the health of workers and represent the concentrations of hazardous substances in the air averaged over a specific time period (time weighted average – TWA). Two time periods are used: long term (8 hours) and short term (15 minutes). The long term limit of 8 hours represents a typical working day, whilst the short term limits are set to help prevent effects which some substances may have following only a few
minutes of exposure e.g. eye irritation. An OES is set at a level at which (based on current scientific knowledge) there is no indication of risk to the health of workers exposed to inhalation day after day. The effects and symptoms of CO₂ exposure are summarised in Table 2:

<table>
<thead>
<tr>
<th>CO₂ concentration</th>
<th>Effects and Symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td>ppm</td>
<td>% vol</td>
</tr>
<tr>
<td>10000</td>
<td>Slight but un-noticeable increase in breathing rate.</td>
</tr>
<tr>
<td>20000</td>
<td>Breathing becomes deeper – rate increases to 50% above normal. Prolonged exposure (several hours) may cause headache and exhaustion.</td>
</tr>
<tr>
<td>30000</td>
<td>Breathing becomes laboured. Hearing ability reduced, headache experienced with increase in blood pressure and pulse rate.</td>
</tr>
<tr>
<td>40-50000</td>
<td>As above. Signs of intoxication after 30 minutes exposure and slight choking sensation.</td>
</tr>
<tr>
<td>50-100000</td>
<td>Characteristic pungent odour noticeable. Breathing very laboured leading to physical exhaustion. Headache, visual disturbance, ringing in the ears, confusion probably leading to loss of consciousness within minutes.</td>
</tr>
<tr>
<td>100000+</td>
<td>Rapid loss of consciousness with risk of death from respiratory failure.</td>
</tr>
</tbody>
</table>

3. Foreseeable Risks

Uncontrolled release of gas due to:

- regulator failure.
- failure of pipework or tubing connecting cylinder to other equipment.
- damage caused by impact e.g. falling cylinder.
- damage caused by fire.

4. Assessment of Potential for Gas Release

It is difficult to determine exactly what rate CO₂ would be discharged from a cylinder in the event of a tube 'blow-off'. It is estimated that a discharge ranging from anywhere between 1.2 to 84.0 m³ per hour could be anticipated (figures supplied by Gas Safety UK Ltd.). The lowest figure can be considered to be a slow release scenario whereby the contents of a cylinder would be discharged over the course of several hours (i.e. a working day or overnight), while the highest figure equates to a sudden release, whereby the cylinder contents are completely discharged within the space of a few minutes (the ‘worse case’ scenario). Gas suppliers such as BOC will be able to supply information on the quantities of gas contained in different sized cylinders – it can therefore be estimated how long a cylinder would take to empty at a given release rate.

Since the process of risk assessment examines the likelihood of an event occurring, it is perhaps most appropriate to look at slow release scenarios as examples. These are arguably more likely than a sudden or fast release and there is also a greater chance that a slow release would go unnoticed (a sudden failure or rapid release may well give rise to hissing, popping or other audible / visual warnings that would alert anyone in the vicinity to the fact that something was wrong).
Consider the following examples:

Slow release scenario
A laboratory with dimensions 6m x 4m x 3m contains one size K vapour withdrawal CO₂ cylinder connected by a flexible tube to an incubator. The tube accidentally becomes detached and goes unnoticed (there have been reports of this actually happening within the College).

Using the equation:

\[ C = \frac{L}{Vn} \]

where: 
- \( C \) = CO₂ concentration in room
- \( L \) = CO₂ release from cylinder (m³ / hour)
- \( V \) = room volume (m³)
- \( n \) = air changes per hour

\[ C = \frac{1.2}{72} \times 1 = 0.016 \times 100 = 1.66\% \text{ CO}_2 \ (\text{O}_2 \text{ level reduced from 21 to 20.6\%}) \]

A cylinder containing 34Kg of CO₂ (capable of producing 18.2 m³ gas) would take around 16 hours to empty at a release rate of 1.2m³ / hour.

The CO₂ concentration within the laboratory reaches around 1.7% - exceeding both the short term and long term exposure limits (remember, this is the lowest estimated discharge in a room with 1 air change per hour). If the room is below ground level, has no windows or has other factors affecting air circulation, the air changes may be even lower – typically 0.4 per hour. The same scenario would then give rise to a CO₂ concentration exceeding 4% (O₂ level reduced from 21 to 20.2%) - at this level, signs of intoxication would be evident after 30 minutes exposure.

The above examples illustrate that even at these low release rates, the levels of CO₂ in the room can become unacceptably high. It is also evident that it is the rise in CO₂ levels and not the level of oxygen depletion that is the issue.

5. Control Measures

- Ventilation – ensure that it is adequate. Estimations can be made using the above guidelines. If natural ventilation is poor, consider forced ventilation / extraction.
- Consider fitting safety devices such as excess flow valves - these detect drops in pressure and halt or drastically reduce the flow of gas from the cylinder. They are commercially available, easy to fit and relatively inexpensive.
- If flexible tubing is used, ensure that it is securely connected to equipment (e.g. with a cable tie or ‘jubilee’ clip).
- Consider fixed-point gas detection monitors / alarms.
- Ensure that cylinders are secure and away from sources of heat.
- Ensure that there is a regime in place for proper systematic checking and maintenance of regulators.
- Training – ensure that users are familiar with the equipment and the properties of all substances that they are handling and know what to do in an emergency.
Part 2 – Dry Ice

1. General Uses

Commonly used as a cooling / freezing agent in laboratory procedures or for transportation of samples.

2. Properties and Hazards

- Solid form of CO$_2$ – very cold (-78°C), can cause cryogenic burns.
- Normally supplied as pellets, flakes or blocks.
- Sublimes (changes state from solid to gas – the product will not melt to a liquid) – gaseous CO$_2$ presents the same hazards as outlined in Part 1.

3. Foreseeable Risks

- Burns due to skin contact.
- Sublimation resulting in accumulation of gas in poorly ventilated areas.
- Explosion due to pressure build up if kept in sealed containers.
- Manual handling of large bags of dry ice (10kg is the standard package weight).

4. Assessment of Potential for Gas Release

It is difficult to evaluate the rate at which the solid form will convert to the gaseous form since this will be dependent on a number of variables:

- The form of the dry ice – pellets or flakes, for instance, will sublime at a faster rate than blocks.
- The ambient temperature – sublimation will proceed faster at higher temperatures.
- The degree of insulation provided by the container.

However, the data below can be used to make some approximate estimates as to what concentration of gas will be generated in particular circumstances.

- 1kg of dry ice will produce 0.45 m$^3$ of gas (figure supplied by Gas Safety UK Ltd.).
- Dry ice to CO$_2$ sublimation rate = approx. 1% of total mass per hour in an insulated container (figure supplied by Gas Safety UK Ltd. - source: Federal Aviation Administration in USA).
- Dry ice to CO$_2$ sublimation rate = approx. 14% of total mass per hour at room temperature in the open (figure supplied by Gas Safety UK Ltd. - source: a study published in Aviation, Space and Environmental Medicine 1977).

Consider the following example:

A number of specimens packed in dry ice are being transported by car from one location to another. The container is well insulated and is positioned on the back seat of the car - the cars windows are closed. The journey takes 1 hour. Assuming the interior volume of the car is 2.8 m$^3$:

<table>
<thead>
<tr>
<th>Quantity of dry ice (Kg)</th>
<th>Volume of gas produced during journey (m$^3$)</th>
<th>Concentration of CO$_2$ in car atmosphere (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>0.023</td>
<td>0.8</td>
</tr>
</tbody>
</table>
The concentration of CO2 in the car reaches 0.8% - between the short term and long term exposure limits and within a range where it will not cause any adverse effects. The concentration would be increased by any combination of the following:

- increasing the quantity of dry ice used.
- using a poorly insulated or open container.
- using a smaller car.
- lengthening the journey time.

It can therefore be seen that providing all the factors are taken into account, a reasonable quantitative assessment of risk can be carried out.

5. Control Measures

- Do not handle with bare hands – use cryogenic gloves.
- Avoid carrying dry ice in the cab of a truck or the passenger compartment of a car. If this is not possible, use as little dry ice as possible, ensure that the container is well insulated (though not tightly sealed) and ensure that the compartment is well ventilated (open windows, ensure ventilation system is set to draw fresh air from outside).
- Unload the material as soon as possible at the end of a journey.
- Store dry ice in well ventilated areas away from direct sunlight and sources of heat.
- Use suitable storage containers (there are commercially available insulated containers with vented seals specifically designed for storing dry ice).
- Secure to prevent any unauthorised access.
- Use appropriate warning signage where necessary.
- When opening lids to storage containers, allow a few seconds for gas to dissipate and do not lean in for longer than necessary.
- Do not store or use dry ice in any gas tight container.
- Do not store dry ice in a working refrigerator or freezer – it will sublimate at a faster rate than in an insulated storage container and the extremely cold temperature may cause the thermostat to cut out.
- Do not play games with dry ice.
- Dispose of unwanted dry ice by allowing it to evaporate in a well ventilated area – it will sublime leaving no residue.
- Carry out manual handling assessment of bags if necessary.
- Ensure that all users of dry ice are familiar with the hazards and necessary precautions.