

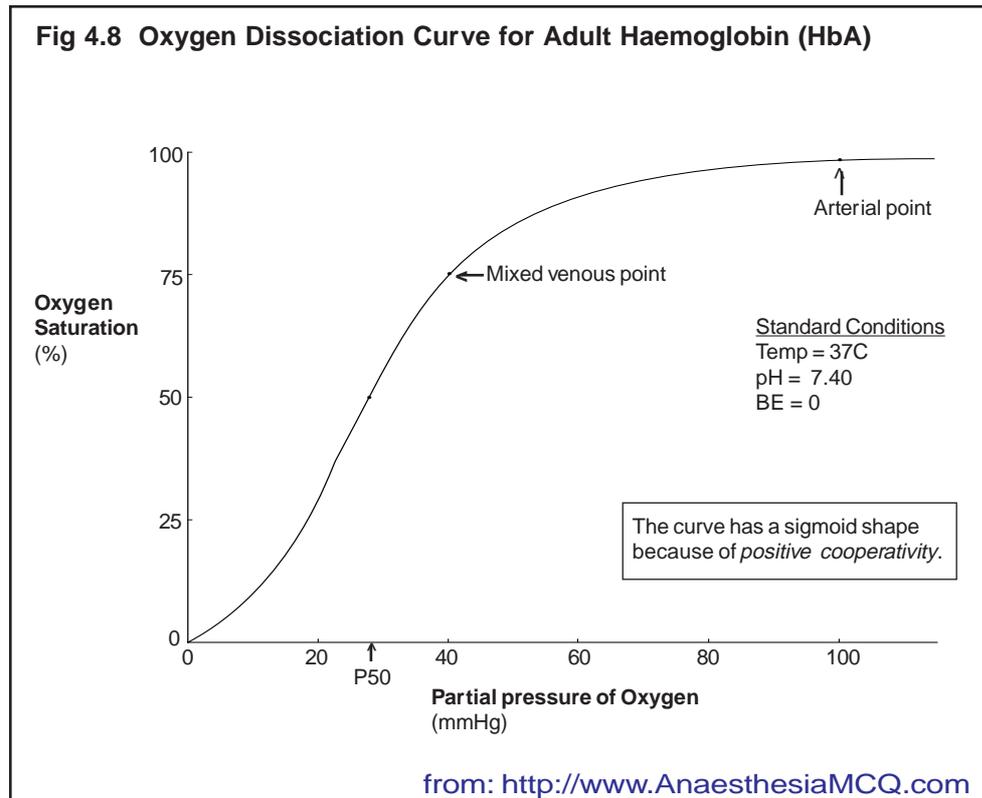
Oxygen Dissociation Curve

Can you draw the oxygen dissociation curve of normal adult haemoglobin?

How many points on the curve can you indicate with values?

The oxygen dissociation curve of adult haemoglobin is a sigmoid curve. The three main points to indicate on the curve are:

- Arterial point: pO_2 100 mmHg with $SO_2 = 97.5\%$
- Mixed venous: pO_2 40 mmHg with $SO_2 = 75\%$
- P50: pO_2 26.6 mmHg with $SO_2 = 50\%$



[Four other simple points to remember to allow rapid and reasonably accurate drawing of the ODC in a viva are:

pO_2 0 mmHg, SO_2 0% - the origin

pO_2 10 mmHg, SO_2 10% - just easy to remember & helps get the sigmoid shape.

pO_2 60 mmHg, SO_2 91% - the 'ICU' point

pO_2 150 mmHg, SO_2 98.8% - shows flat upper part of ODC

The 'ICU point' can be considered as the point on the curve that separates the steep lower part from the flat upper part. This is a bit artificial but a pO_2 of 60 mmHg in this sense is considered as the lowest acceptable pO_2 in an ICU patient because marked desaturation occurs at pO_2 values below this point.]

What is the mixed venous point?

This is the point which represents mixed venous blood. The pO_2 here is 40 mmHg and the haemoglobin saturation is 75%. The oxygen content cannot be specified without further information (eg [Hb])

Note that the mixed venous point does NOT really lie on the normal ODC as above (& in all the texts). The increased pCO_2 and decreased pH in mixed venous blood mean that the mixed venous point must lie on a slightly right shifted ODC rather than the standard ODC. This is the Bohr effect.

What is meant by the term 'P50'?

This term is used in reference to the oxygen dissociation curve. It is defined as the partial pressure of oxygen at which the oxygen carrying protein is 50% saturated. It is usually used in relation to haemoglobin but can also be used for other oxygen binding proteins such as myoglobin. Though often drawn as a point on the dissociation curve, this is incorrect as the P50 is, by definition, a point on the x-axis as it is a particular pO₂ value (& not a pO₂-SO₂ value pair like the mixed venous point for example.)

What is the normal value for the P50 of adult haemoglobin?

The P50 of normal adult haemoglobin is 26.6 mmHg.

What is the P50 used for?**Why was this point on the curve chosen for this purpose?**

The P50 is used to specify the position of the oxygen dissociation curve (or alternatively, the P50 is an *index of oxygen affinity* of the oxygen carrying protein. This is what specifying the position of the curve is really about).

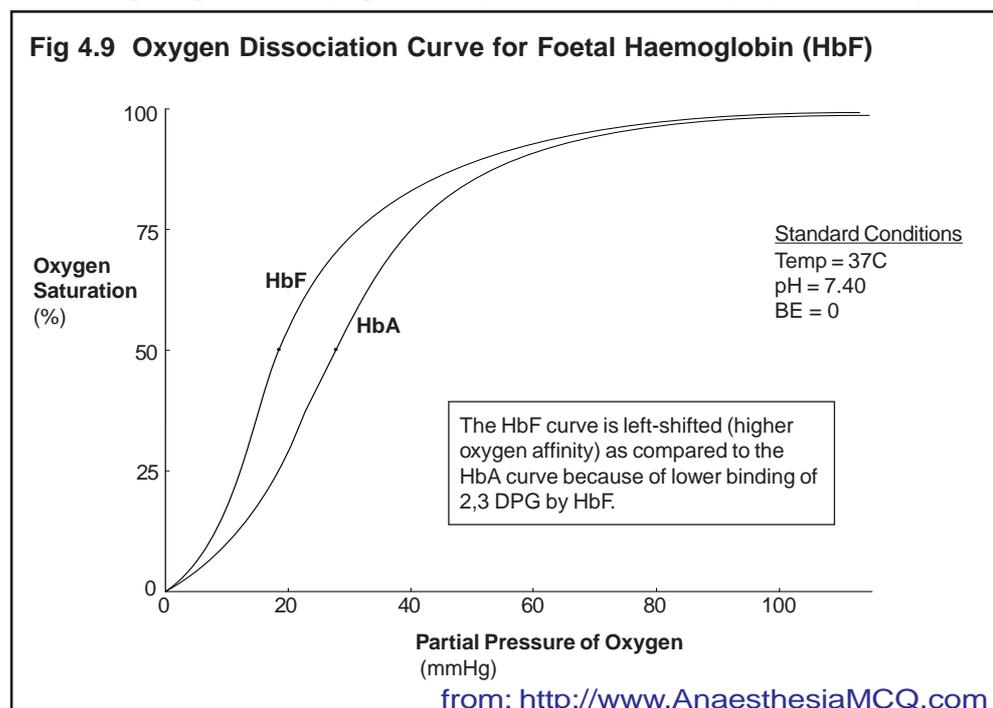
It is the most useful point for specifying the curve's position because it is on the steepest part of the curve. It is therefore the most sensitive point for detecting a shift of the curve. Specifying the P50 of a curve allows comparison with the position of other curves under different conditions.

What does a right shift indicate?**What are the causes of a right shift?**

A right shift indicates decreased oxygen affinity. The P50 is higher for a right shifted curve.

A right shift can be caused by an increase in 4 factors:

- temperature
- [H⁺]
- pCO₂
- red cell 2,3 DPG level.

Can you superimpose on the graph the oxygen dissociation curve for foetal haemoglobin?

How is this curve different?

What is the P50 of foetal haemoglobin?

The curve has a sigmoid shape very similar to the normal ODC for adult haemoglobin but slightly left shifted. The P50 is lower at about 18 mmHg. (Values between 18 and 20mmHg are quoted in various sources.)

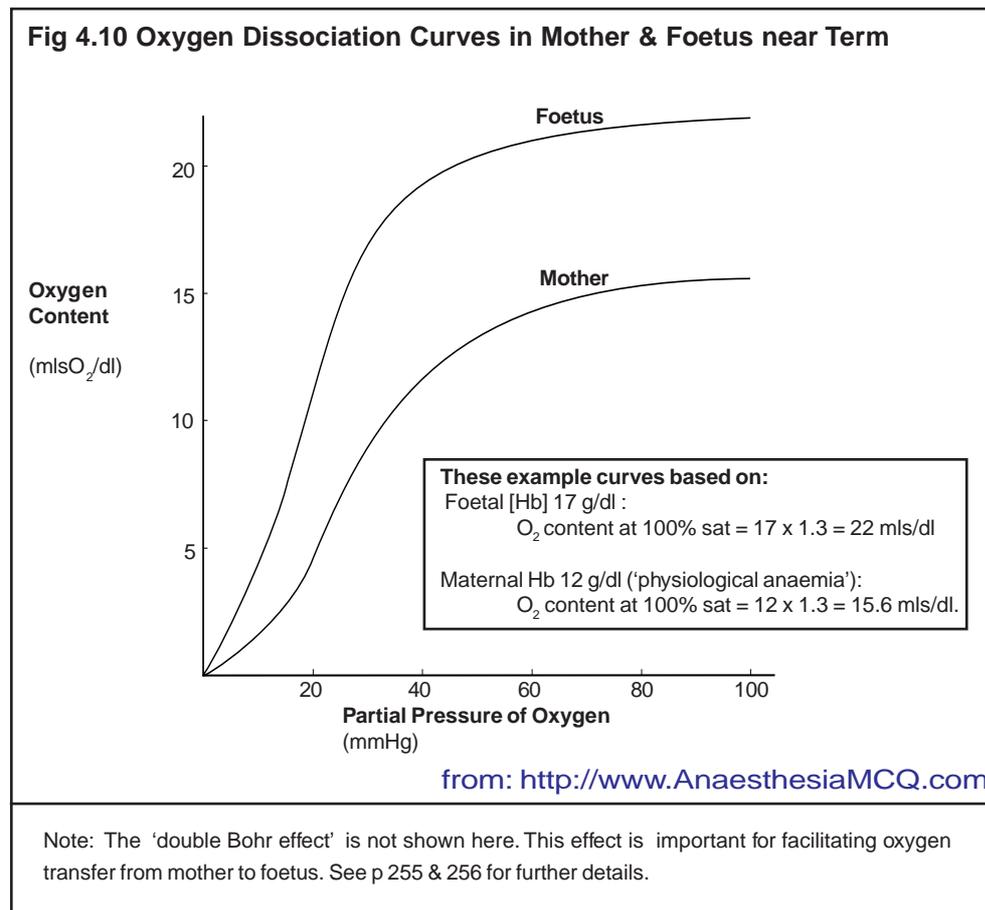
Why is the foetal haemoglobin curve shifted to the left?

The lower P50 value indicates that the curve is left shifted as compared to the adult curve (ie foetal Hb has a higher oxygen affinity). The reason for this is the reduced binding of 2,3 diphosphoglycerate (2,3 DPG) to foetal haemoglobin. 2,3 DPG binds best to the beta chains of adult haemoglobin and this shifts the curve to the right indicating a decrease in oxygen affinity. In fact, 2,3 DPG binds most avidly to the beta chains of *deoxyhaemoglobin*.

Foetal haemoglobin consists of 2 alpha chains and 2 gamma chains (ie there are no beta chains). Consequently the P50 is lower than that of adult haemoglobin because HbF is less sensitive to the effects of 2,3 DPG.

Could you draw the oxygen dissociation curves for a foetus at term and for the mother at term using oxygen content on the y-axis (rather than saturation)?

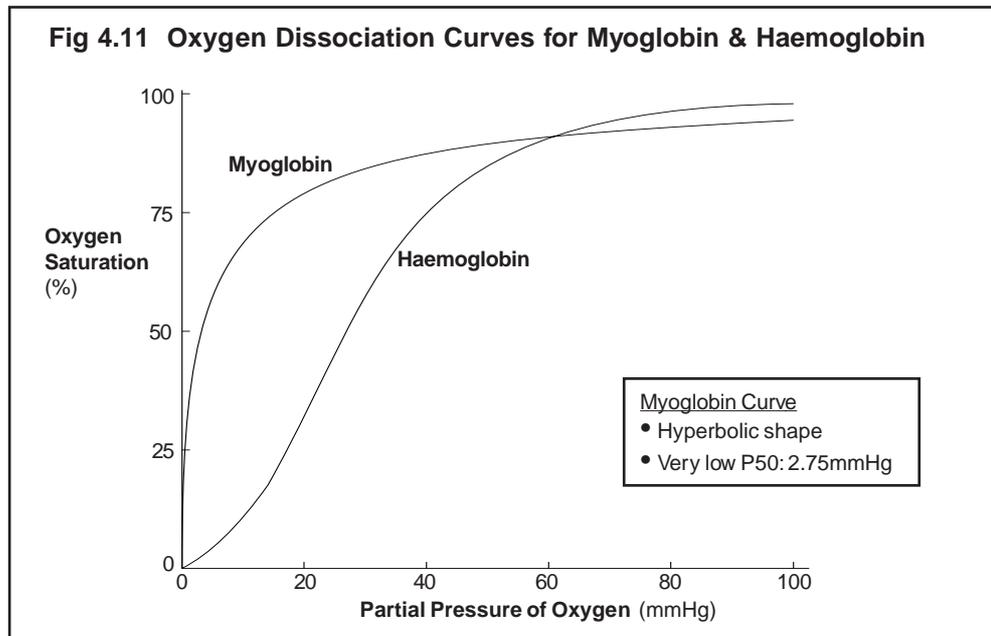
The curves are different because of the left shift of foetal haemoglobin, the increased [Hb] of foetal blood and the decreased [Hb] of maternal blood (*physiological anaemia*).



What is myoglobin?

Myoglobin is a haem-containing oxygen binding protein that is present in skeletal muscle. It has a role as an oxygen store.

Can you draw the oxygen dissociation curve for myoglobin?



What is the P50 of the myoglobin curve?

What is the shape of this curve?

Why is it different from the shape of the haemoglobin curve?

What is 'positive cooperativity'?

The myoglobin curve is a rectangular hyperbola with a very low P50 (2.75 mmHg). It lies well to the left of the sigmoid-shaped haemoglobin curve. It has a much higher oxygen affinity.

The physiological reason (the advantage): Myoglobin needs to have a P50 less than haemoglobin so it can take up oxygen from it. Also, myoglobin needs to be able to load and unload oxygen in the range of pO₂ values that occur within the cell. If its P50 was say 20 mmHg and intracellular pO₂ was 1 to 5 mmHg, then the myoglobin could never load oxygen. Intracellular pO₂ does vary between different cells and within the same cell, but is typically low. Oxidative phosphorylation ceases below a pO₂ of about 1 mmHg. It can be seen that myoglobin with a P50 of 2.75 mmHg is well matched to intracellular needs in muscle cells. It can load oxygen from haemoglobin and can unload its oxygen as cytoplasmic pO₂ falls to low levels.

The chemical reason (the cause): The reason why the curves are different is because of the different structures of myoglobin and haemoglobin. Myoglobin contains only a single globin chain: its dissociation curve is a rectangular hyperbola. Haemoglobin contains four globin chains and the oxygenation of each chain causes structural changes which increase the affinity of the haem of the remaining chains for oxygen. This consequence of sub-unit interaction is known as **positive cooperativity** and this increasing oxygen affinity as oxygen loads is the cause of the sigmoid shape of the dissociation curve.

What is the physiological significance of the shape of the oxygen dissociation curve for adult haemoglobin?

The curve can be considered to consist of two parts:

- the flat upper part
- the steep lower part

The flat upper part acts as a buffer in the sense that the pO₂ can drop to about 80 mmHg and yet the haemoglobin will still remain highly saturated (96%) with oxygen. This keeps the arterial oxygen concentration high despite impairment in saturation in the lung.

from: <http://www.AnaesthesiaMCQ.com>

The steep lower part means that if the tissues require more oxygen, substantial amounts of oxygen can be removed from haemoglobin without much further drop in pO₂. The pressure gradient for diffusion of oxygen from capillary to cell tends to be relatively well maintained despite the much increased oxygen extraction. (There are other mechanisms which increase blood flow and thus oxygen supply to the tissues and these are compensatory mechanisms additional to the increased oxygen extraction.)

In summary, the shape of the ODC provides this double buffering effect because:

- The flat upper part tends to 'buffer' haemoglobin saturation against a substantial drop in pO₂. This is useful in the lungs to maintain the arterial haemoglobin saturation.
- The steep lower part has 2 advantages: Large O₂ unloading & a maintained O₂ diffusion gradient (ie the pO₂ gradient from capillary to cell).

How is the 'saturation' of haemoglobin defined?

$$\text{Oxygen saturation (\%)} = \frac{\text{Actual oxygen content of haemoglobin} \times 100}{\text{Maximum oxygen content of haemoglobin}}$$

What is the effect of acute anaemia on the ODC of haemoglobin? (eg if the [Hb] dropped from 15 g/dl to 7.5 g/dl)

The curve would not be altered if it was drawn as Saturation (y-axis) versus pO₂ (x-axis).

If drawn as Oxygen content (y-axis) versus pO₂ (x-axis), the content value at each pO₂ would be halved. The shape would not be altered.

[In chronic anaemia, red cell 2,3 DPG levels rise and the curve will be right shifted.]

What is the effect of carbon monoxide on the ODC? (eg if 33% of the [Hb] of 15 g/dl was present as carboxyhaemoglobin.)

Draw the haemoglobin dissociation curve with oxygen saturation on the y-axis.

Now draw this curve with oxygen content on the y-axis.

There are 2 effects:

- The curve is shifted to the left
- The O₂ content is reduced

(Note the difference in the curves on the next page: this is a frequent source of confusion.)

The left shift occurs because the binding of carbon monoxide causes a conformational change in the haemoglobin causing increased affinity for oxygen by the other subunits.

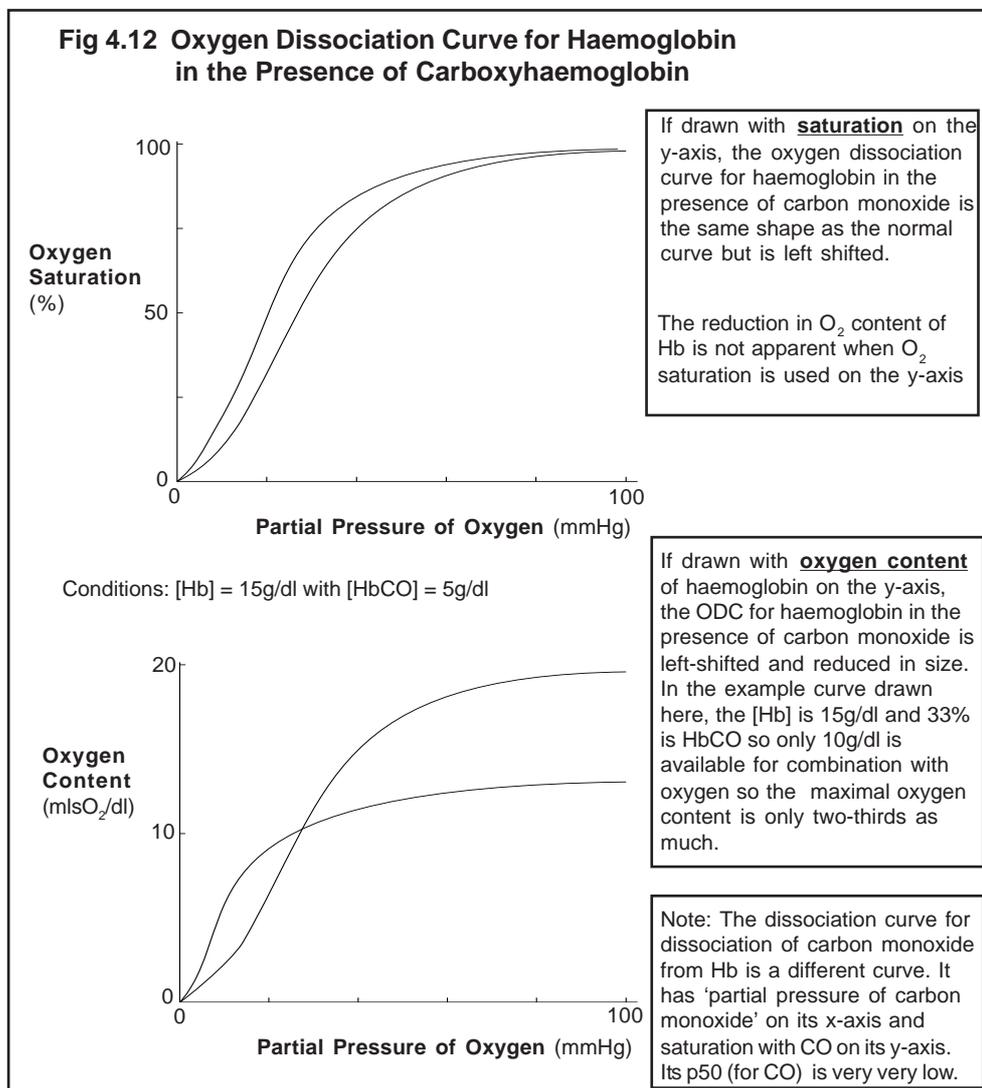
How is the carboxyhaemoglobin dissociation curve different from these two curves?

This is a dissociation curve for the dissociation of *carbon monoxide* from haemoglobin whereas the curves for the above answer are curves for the dissociation of *oxygen* from haemoglobin (in the presence of carbon monoxide as a modifying factor).

The differences in appearance of the curves are:

- The axes are different: saturation of Hb with CO on the y-axis and partial pressure of carbon monoxide (pCO) on the x-axis.
- The extremely high affinity of carbon monoxide for haemoglobin means that the curve is extremely left-shifted and is a rectangular hyperbola.

[**Note:** A frequent source of confusion in viva candidates is in distinguishing 'carboxyhaemoglobin' from 'carbaminohaemoglobin'. The first is Hb combined with CO and the second is Hb carrying CO₂.]



What is the difference between 'functional saturation' and 'fractional saturation'?

Is this clinically important?

The definition of oxygen saturation given earlier is the definition for 'functional saturation' and is the traditional way of considering saturation (ie saturation is content x 100/capacity). Saturation of haemoglobin was originally determined by measuring the actual oxygen content of a blood sample and by measuring the oxygen content of another blood sample after equilibrating the sample with room air (pO₂ 149mmHg). But blood may typically contain up to 4 haemoglobin species: oxyHb (HbO₂), deoxyHb, MetHb & COHb - and only one of these carries oxygen. The definitions used for saturation are:

- Functional saturation = ([HbO₂] x 100 / ([HbO₂] + [DeoxyHb])
- Fractional saturation = ([HbO₂] x 100 / Total [Hb])
 where Total [Hb] = [HbO₂] + [DeoxyHb] + [MetHb] + [COHb].

Clinically it may be more useful to consider fractional saturation. This is because we tend to use the oxygen saturation value as an index of blood oxygen content (ie Sat x [Hb]). If large amounts of MetHb and/or COHb were present, then fractional saturation is useful in this way but functional saturation would be very misleading (eg 99% functional saturation could be associated with quite a low oxygen content if a high percent of Hb was HbCO or MetHb). Another clinically relevant point: the value displayed by a pulse oximeter is neither functional nor fractional saturation but depending on the calibration used for the particular brand may be close to one or the other of these two saturations. [As a two wavelength device, the pulse oximeter is limited to measuring only two Hb species - HbO₂ and deoxyHb.]

Also consider what saturation value is reported on blood gas reports in your hospital.