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Could Bubble Growth by Rectified Diffusion Produce Macroscopic Bubbles in the Tissues of Beaked Whales?

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COULD BUBBLE GROWTH BY RECITIFIED DIFFUSION PRODUCE MACROSCOPIC BUBBLES IN THE TISSUES OF BEAKED WHALES?

by

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Contents

1. Background

The phenomenon of mass-stranding has been observed in beaked whales since the early 1960s when mid-frequency active sonar was first put into service by naval vessels worldwide (Balcomb & Claridge 2001). This mass-stranding behaviour was immediately of particular interest because the most severely affected beaked whale species – *Ziphius cavirostris* and *Mesoplodon densirostris* – had only ever been observed to strand individually prior to the use of this sonar (Balcomb & Claridge 2001, Friedman 1989). The phenomenon has received increased attention in recent years after Simmonds and Lopez-Juardo noted the spatial and temporal coincidence of mass-stranding events with naval manoeuvres (Simmonds & Lopez-Jurado 1991). Many confirmed incidences have been noted since then, including strandings in Greece in 1996 (Frantzis 1998), Madeira in 2000 (Freitas 2004), the Bahamas in 2000 (Evans *et al.* 1970) and the Canaries in 2002 (Jepson *et al.* 2003, Fernandez *et al.* 2005).

Necropsies conducted on whales stranded during the Canary Islands incident revealed that the whales suffered from symptoms consistent with DCS (Jepson *et al.* 2003, Fernandez *et al.* 2005), including lesions associated with the presence of macroscopic gas bubbles. The discovery of such DCS-like symptoms in beaked whales was both controversial and unexpected, since it had always been assumed that whales would have evolved anatomical, behavioural and physiological adaptations to buffer them against the effects of such diseases (Cox *et al.* 2006). It is speculated that these DCS-like symptoms are directly caused by gas embolism, with the gas bubbles forming in the blood and tissues *in vivo* from stabilised gas bubble nuclei already present in the tissues. Explaining how these nuclei might grow to sizes that can cause such injuries is therefore pivotal to understanding the aetiology of this DCS-like disease in beaked whales, and provides the motivation for this report.

2. Beaked whale injuries

Pathological findings

The injuries associated with mass beaching mentioned in the background to this report were identified during necropsies that were carried out on whale carcases collected from a number of sites from separate mass-stranding events all around the world. The pathologies described here are those identified in beaked whales that stranded during the Canary Islands mass beaching event in 2002, as these findings provide a summary of the typical lesions associated with such events by (Jepson *et al.* 2003)(Fernandez *et al.* 2005). Throughout this section, words indicated by a '*' are defined in the glossary at the end of this report.

On the macroscopic scale, the whales were found to display evidence of acute*, severe and diffuse* congestion* and vascular* haemorrhaging*, most notably around the acoustic jaw fat, ears, lungs, central nervous system (CNS) (including the brain) and kidneys. In the lungs, multiple haemorrhages were accompanied by blood clots in the fine pulmonary capillaries. In the kidneys, the haemorrhages were multifocal*, interstitial*, interlobular* and subcapsular*, and just as in the lungs, were found alongside blood clots. Congestion and haemorrhaging also occurred in the lymphatic system, spleen, and neck (including the pharynx, larynx, thyroid gland and trachea), resulting in the distention of some of the vessels within them. In all cases, haemorrhages were microvascular, i.e. occurred in the small diameter blood vessels within these organs.

In addition to these injuries, evidence of gas-bubble associated lesions* and fat embolism* in the blood, lymphatic vessels and parenchyma (white matter) of several vital organs were also found. The organs affected by gas bubbles in this way included the brain, liver, kidney (subcapsular kidney veins), and lungs (Jepson *et al.* 2003, Fernandez *et al.* 2005). The peribullar* and pterygoid* sinuses* were also full of white froth (Fernandez et al. 2005). These findings were controversial for two reasons. Firstly it is difficult to gain definitive evidence of gas embolism after death (Knight 1996). Secondly, up until the first publication of these results (Jepson *et al.* 2003), it was neither known nor expected that gas bubbles could form *in vivo*, since the assumption had always been made that whales would have evolved adaptations to prevent deleterious gas bubble formation (Cox *et al.* 2006).

Fat emboli were found in the epidural* veins (where epidural adipocytes* associated with haemorrhaging were either enlarged or ruptured); liver sinusoids*; lymph nodes (particularly those of the epidural retia* in which adipocyte necrosis* was associated with acute haemorrhaging); and lungs (in both the capillaries and veins) (Fernandez et al. 2005). With only one exception, fat emboli in the vascular system were found exclusively in veins. No fat emboli were found in the vessels of the CNS, and it is thought that this was because of their filtration by large vascular plexi including the epidural and thoracic retia (Fernandez *et al.* 2005). The exact mechanism of fat emboli formation is not known, but it is thought that in the case of the pulmonary and systemic emboli discussed here, they result from injury to fat deposits (Fernandez *et al.* 2005). None of the whales that beached on the Canary Islands contained pathogenetic* bacteria (Fernandez *et al.* 2005, Jepson *et al.* 2003), or inflammatory* or neoplastic processes* (Fernandez *et al.* 2005), and all were feeding and in good body condition immediately prior to stranding (Fernandez *et al.* 2005) therefore suggesting a very rapid demise from full health.

The over-riding conclusion from these clinicopathologic* findings was that these lesions were most similar to those reported in acute DCS in human divers (Fernandez *et al.* 2005, Jepson *et al.* 2003). As there are no individual symptoms that are pathognomonic* of DCS (Jepson 2004), a diagnosis of this condition relies on the combination of symptoms found. This, taken together with the fact that DCS is a human disease, means that the whales can only be said to have suffered injuries that were consistent with, but not diagnostic of, a DCS-like condition.

Causes of the injuries

It is thought that the lesions found in cases typified by the ‘Canary Islands whales’ all arise, both directly and indirectly, from acute gas embolism. Most of these gas bubbles are presumed (although not confirmed) to contain nitrogen, although some may contain intestinal gases. They are formed *in vivo* by a yet unknown mechanism, which will be the subject of the next section of this report. What happens to them after this determines the injuries they then go on to cause, and the next few sentences describe current speculations about how this may occur. Since nitrogen is more soluble in lipids than in polar molecules, most of the sources of nitrogen in the body are in fat-rich tissues, so this is where most of the bubbles are thought to form. It can be seen from the documented injuries that gas bubbles cause direct damage to

these fatty tissues – the acoustic jaw fats, the epidural fat surrounding the spinal cord (part of the CNS), the perirenal fat surrounding the kidneys, and the lymphatic system all displayed lesions directly resulting from gas embolism. When the bubbles enter the circulation, they may be transported to capillary dense tissues in organs anywhere in the body where they lodge, resulting in vessel damage and haemorrhaging (Rommel *et al.* 2006), blood clot formation, and / or localised tissue death by ischemia (Rommel *et al.* 2006). If the bubbles then enter the parenchymous tissue of the affected organ, they may cause additional problems by putting pressure on the cells, resulting in the improper function of that organ.

Additionally, gas bubbles may cause damage to the adipocytes in the fatty tissues in which they arise resulting in the production of fat emboli. Having entered the circulation, these fat emboli may themselves lodge in small diameter blood vessels in the vital organs (on the venous side 'downstream' of the organs in which they arise) also causing haemorrhaging and localised cell death by ischemia as an indirect effect of the gas embolism (Rommel *et al.* 2006).

It is because of the gas- and fat embolic nature of this disease, together with the presence of widely disseminated microhaemorrhages in lipid rich tissues on central lines of circulation (e.g. brain and spinal cord) that this condition is thought to be analogous to DCS is humans. Although DCS may be the ultimate cause of death in these whales, it may be another factor that is the proximate cause. Cardiovascular collapse resulting from the stress of beaching, involving hyperthermia* and high endogenous* catecholamine* release, has been suggested (Fernandez *et al.* 2005, Jepson 2004), and was thought to have caused the death of the whales involved in the Bahamas incident (Jepson 2004). However, this has been all but ruled out as an ultimate cause owing to the lack of bruising and the fact that some whales died at sea before subsequently washing up (Fernandez *et al.* 2005).

Similar conditions

It should be noted that evidence of gas embolic disease has also been described in beaked whales that stranded individually in the UK (Jepson *et al.* 2003, Jepson *et al.* 2005). In these cases it was not clear whether sonar was involved, although some of the pathologies described in these cases were similar in nature to those associated with animals that mass stranded. However, there were also important differences that lead to the conclusion that this represented a new form of gas embolism

inconsistent with any so far known in either human or animal pathology, including DCS (Jepson *et al.* 2005). This was because this condition was associated with the presence of large bubbles in the liver and kidneys of 0.2 to 6.0 and 0.2 to 0.9 cm in diameter respectively, that were found to be filled with a colourless and odourless gas and encapsulated by fibrotic tissue. The presence of this fibrotic tissue indicated that this was a chronic condition, and it was suggested that it was initiated via the haematogenous transfer of gas bubbles to the affected organs over a period of several days, with the effect being exacerbated by the cyclical pressure changes associated with repeated diving (Jepson 2007). Importantly for this report, these findings provided support for the theory that whales do have bubble nuclei present within their tissues *in vivo,* and are therefore not innately immune to DCS - a theory additionally supported by evidence of osteonecrosis* and other symptoms in sperm whales whose combined presence is most parsimoniously explained by a condition similar in nature to that just described (Moore & Early 2004). It has been suggested that it is these bubble nuclei that go on to grow either during or following exposure to naval sonar to cause the lesions described in mass stranded whales.

3. Bubble nuclei and bubble growth

Bubble nuclei

It is clear from the proposed disease aetiologies described above that gas emboli are favoured as being the underlying cause of the injuries observed in mass stranded animals. It is widely assumed that these gas emboli grow from a population of stable bubble precursors, often referred to as bubble 'seeds' or 'nuclei', which exist permanently within the relevant biological tissues *in vivo*. This is a concept that is not universally accepted, but nevertheless generally considered to be true (Jepson *et al.* 2005, Daniels & ter Haar 1986, Houser *et al.* 2001). The nature of these bubbles is not fully understood, since ordinarily a bubble will collapse under the Laplace pressure, which opposes the inwardly acting force of surface tension, forcing gas out of the bubble. These populations of bubbles seem to be stable against dissolution, (but not buoyancy), indefinitely. Proposed mechanisms of stabilisation of individual bubbles include the role of surfactant in making the bubbles impermeable to gas and thereby preventing its movement either into or out of the bubble (Potter 2004, Leighton 1994); and the presence of microscopic crevices within the tissue in which stable bubbles can exist (Leighton 1994).

The alternative explanation for the permanent existence of these bubble nuclei in the tissues that does not require any stabilisation mechanism, is that of spontaneous, or homogenous nucleation, wherein the population is stabilised by the formation of new nuclei to replace old ones (Church 2002). Bubble nuclei are hypothesised to form as a result of the transient concentration of high energy molecules within the tissue due to the uneven distribution of energy between the molecules of that tissue as described by the Boltzman distribution. These concentrated 'pockets' or inclusions of high energy molecules arise randomly and continually, possibly as a result of exposure to cosmic rays (Leighton 1994), before dissolving, so even though an individual inclusion exists only transiently, bubble nucleus inclusions are continually present within the tissue.

Whether any of these explanations describes the true mechanism by which bubble nuclei may be continually present in the tissues of the whales is not yet known, but it can be seen that there are a number of plausible explanations that could explain their existence. The assumption that these bubble nuclei do exist permanently within the tissues in some way therefore seems a reasonable one.

Rectified diffusion

Rectified diffusion is one of the proposed mechanisms by which bubble nuclei might grow to form the gas emboli found in the mass stranded whales. This mechanism involves acoustically driven bubble growth resulting directly from exposure to sound, which in the case of the whales could be the naval sonar acting on the tissues. It occurs when the alternate high and low pressure phases of an acoustic wave drive the pulsed net expansion of a pre-existing bubble nuclei. During the high pressure compression phase of the wave, the pressure inside the bubble increases, leading to the extra dissolution of the gases therein according to Henry's Law. Correspondingly, during the rarefaction phase, the pressure inside the bubble drops and gases dissolved in the surrounding medium re-enter the gas phase, resulting in the expansion of the bubble. The pulsatile nature of the bubble growth results from the entry of more gas into the bubble during the rarefaction phase than leaves it during the compression phase – a result both of the larger surface area for diffusion during expansion (referred to as the 'area effect'), and the high concentration of gas that is maintained in the layer immediately surrounding the bubble as it expands (referred to as the 'shell effect'). As stated above, for this to be a viable mechanism of sonar-induced bubble growth in the whales, a population of stabilised gas nuclei

must already exist in the tissues and the sound would have to destabilise these gas nuclei in some way, by disrupting whichever mechanism of stabilisation was in place. Exactly how this might occur is not known, but it is well established that sound can enhance bubble growth by rectified diffusion (Leighton 1994, Crum & Mao 1996), including in biological material (Crum *et al.* 2005), making rectified diffusion a candidate mechanism for bubble growth in the whales.

Static diffusion

Static diffusion is an alternative means by which gas emboli could grow from bubble nuclei. This mechanism involves the passive movement of a gas from an area of high concentration to an area of lower concentration down its concentration gradient. Since the concentration of a given gas in the tissues depends upon the ambient pressure according to Henry's Law, the potential for static diffusion-induced bubble growth is directly related to the changes in ambient pressure resulting from dive behaviour. As the animal rises towards the surface, bubbles expand under the reduced static pressure. This expansion is reinforced by the exsolution of previously dissolved gas into the bubble, since expansion of the latter causes the gas pressure to drop below the gas pressure that was in equilibrium with the species dissolved in the surrounding tissues. It has therefore been suggested that behavioural responses to sonar exposure during which the normal diving behaviour of the whales changes could result in the growth of gas nuclei in the tissues of the whales and therefore the injuries observed in mass stranded animals.

4. Bubble growth in the tissues of the whales

The question of whether rectified diffusion or static diffusion is the mechanism of bubble growth in the tissues of whales exposed to naval sonar has been the subject of much debate. A number of mathematical models have been proposed describing bubble growth, but there are inherent problems with these. The background population of stabilised gas nuclei in biological tissues is not currently characterised, and this fundamentally limits the power of these models. In addition to this, assumptions have to be made in order to simplify the equations upon which these models are built – assumptions such as the values for parameters constituting the nature of the sound source and the initial diameter of the bubble nuclei – and these also severely reduce the reliability and applicability of these models. Nevertheless,

such models are still useful for laying the foundations upon which future work can be built.

An important model was produced by Crum and Mao in 1996 (Crum & Mao 1996), which applied old equations describing bubble growth via rectified diffusion to estimate the conditions necessary to obtain such growth as a result of exposure to low-frequency sonar propagation in the ocean. The conclusion from this model was that under the range of conditions they assumed to be realistic - sonar frequencies in the range of 300-500 Hz; received levels at the location of the whales of 150-220 dB; modest nitrogen saturation levels in the tissues of 100-223% of those at 1 ATA; and initial bubble nucleus diameters of 1-10 μm - significant bubble growth would only be expected to occur at received levels in excess of 210 dB, with no bubble growth predicted below received levels of 190 dB. The implication of this result was that it is highly unlikely that sonar could have directly caused the death of all of the whales involved in each of the mass stranding events in which it was used, since whales were affected over much larger distances, (in the order of kilometres), than the areas within which the received levels would have exceeded the 210 dB threshold.

However, work carried out since the publication of this paper has revealed the assumptions of the model to be wrong. The frequencies used in the model were lower than those now known to have been used in mass stranding events - all of which were within the mid frequency range 0.5-10.0 kHz (Cox *et al.* 2006) – although this may not be a problem since bubble growth is thought to be frequency independent within this range (Crum & Mao 1996). In addition to this, the range of nitrogen saturation levels in the tissues, (treated as a generic whole in the model of Crum and Mao (1996)), were lower than those more recently predicted by Houser *et al.* (2001), to arise in the muscles of diving beaked whales. Houser *et al.* estimated the intramuscular nitrogen tensions of several cetacean species, including the beaked whale species *Hyperoodon ampullatus*, using a model developed by Ridgeway and Howard (1979) - which was itself based on the nitrogen tensions they measured in dolphins trained to dive repeatedly to 100 m. Their model predicted that muscle nitrogen tensions in beaked whale tissues could reach 300% of their initial value at the surface by the end of a sequence of dives (Houser *et al.* 2001). This was an important finding because the gas concentrations outside the bubbles determine the diffusion gradient for the gas and therefore the growth of the bubbles. Since nitrogen both comprises roughly 79% of air and accumulates in the tissues because it is biologically inert, the concentration of nitrogen in the tissues is

particularly important in predicting bubble growth there. Again, the model of Houser *et al.* (2001) is limited by the assumptions it relies upon, but as yet, it is our most accurate estimate. Crum and Mao were unaware of the study by Ridgeway and Howard (Houser *et al.* 2001) and therefore did not estimate and include the nitrogen tensions later predicted by Houser *et al.* in their model, although they recognised that this initial condition would be critical in determining the thresholds for bubble growth that it predicted (Crum & Mao 1996).

In 2004, Potter developed a model to assess initial micro-bubble growth in the tissues according to the model of Crum and Mao, substituting in values of tissue saturations of 200-300% to determine whether insonification was required to cause bubble growth (via rectified diffusion), or just to bring about the acoustic activation of the pre-existing bubble nuclei in order that bubble growth might occur by static diffusion driven by the nitrogen tensions in the tissues (Potter 2004) - 'acoustic activation' referring here to the destabilisation of the bubble nuclei by sound. He found that so long as the acoustic source in some way activated the gas nuclei in the tissues, additional insonification to enable rectified diffusion to occur was unnecessary to cause the bubbles to grow to a large enough size to cause DCS, resulting in doubt as to whether rectified diffusion does in fact play a role in the growth of bubbles in the tissues. These results also imply that static diffusion plays a critical role in bubble growth following on from the destabilisation of the gas bubble nuclei. Overall they therefore support of the suggestion by Houser et al. that static diffusion may play a pivotal role in bubble growth in the tissues instead of, or alongside, that played by rectified diffusion. In more detail, Potter found that following initial bubble activation by a transient pulse of sound, bubbles could reach a size sufficient to cause DCS-like symptoms within 10 minutes (Potter 2004). This lead to the suggestion by Houser et al. that once stable gas nuclei were 'activated' (destabilised) by an acoustic signal, (presumably involving some initial expansion, possibly by rectified diffusion), the concentrations of nitrogen they had predicted would be sufficient to cause rapid bubble growth by static diffusion, which could produce bubbles big enough to exert large pressures on the tissues over localised areas, ultimately resulting in the DCS-like lesions seen in the whales exposed to sonar (Houser *et al.* 2001). Crum and Mao also stated that bubble growth by rectified diffusion could only produce bubbles of a finite size, (which was essentially limited by the bubble's natural resonance size), whereas growth of bubbles by static diffusion could result in their growth to an indefinite size (Crum & Mao 1996). This effect of static diffusion on bubble growth is additional to the bubble growth caused

by changes in hydrostatic pressure associated with the changes in depth during a dive, which occur independent of mass flux. It is important to note that continued acoustic exposure was not needed to maintain bubble growth by static diffusion, and so once activated, growth could continue in the absence of the sonar source. This would make sonar particularly dangerous for beaked whales, since if static diffusion does play such a key role, once they are exposed to the sonar, they are likely to suffer from the DCS-like lesions as a result of the nitrogen tensions in their tissues.

In addition to modelling work, laboratory-based experiments have also been conducted to identify the relative roles of rectified and static diffusion in the growth of bubbles in the tissues of beaked whales exposed to sonar. In 2005, Crum conducted an experiment in which he exposed excised bovine and porcine tissues to transient bursts of ultrasound, and found that bubbles did grow, but only if the tissues were saturated with gas (Crum *et al.* 2005). It is noteworthy that the bubbles did not grow unless the gas concentrations were supportive of static diffusion, providing further evidence for the involvement of this form of bubble growth. However, the test conditions that were used were not consistent with those that a beaked whale performing a typical dive cycle exposed to sonar would encounter, and so these results are not entirely applicable to the case of the beaked whales.

The aim of the work conducted as part of this report was to produce a model incorporating all of these findings by building on those of both Potter and Crum and Mao, whilst excluding some of the less accurate assumptions that have commonly featured in models of this kind. The model is then used to identify the roles of rectified and static diffusion in the growth of bubble nuclei in the tissues of whales exposed to naval sonar.

5. The model

For the reasons described in the previous section, a model of bubble growth via rectified diffusion was produced based upon the model of Crum and Mao (Crum & Mao 1996), but with the inclusion of tissue nitrogen saturations of 300% relative to the saturations at one atmosphere of pressure in line with the results of Houser et al. (Houser *et al.* 2001). This was used to make quantitative predictions as to the conditions under which macroscopic bubbles would be expected to grow, and the size that these bubbles would eventually reach for bubbles of initial radius 10 μm. Instead of using conventional equations to describe the growth of a bubble in a

sound field, modified versions of these equations that took into account the dependence of the initial equilibrium bubble radius, R_0, on time were used in the generation of this model. The derivation of both the conventional equations used and those used here are presented in the appendix of this report.

The results of this model revealed that bubble growth by rectified diffusion would only be expected to occur at received levels of more than 200 dB, with growth by static diffusion alone explaining the growth below these intensities. This result is shown by the identical bubble growth curves that resulted from exposure to sonar at received levels of 180 dB and 200 dB as opposed to the increased growth that resulted from received levels of 220 dB, as can be seen in figure 1 below.

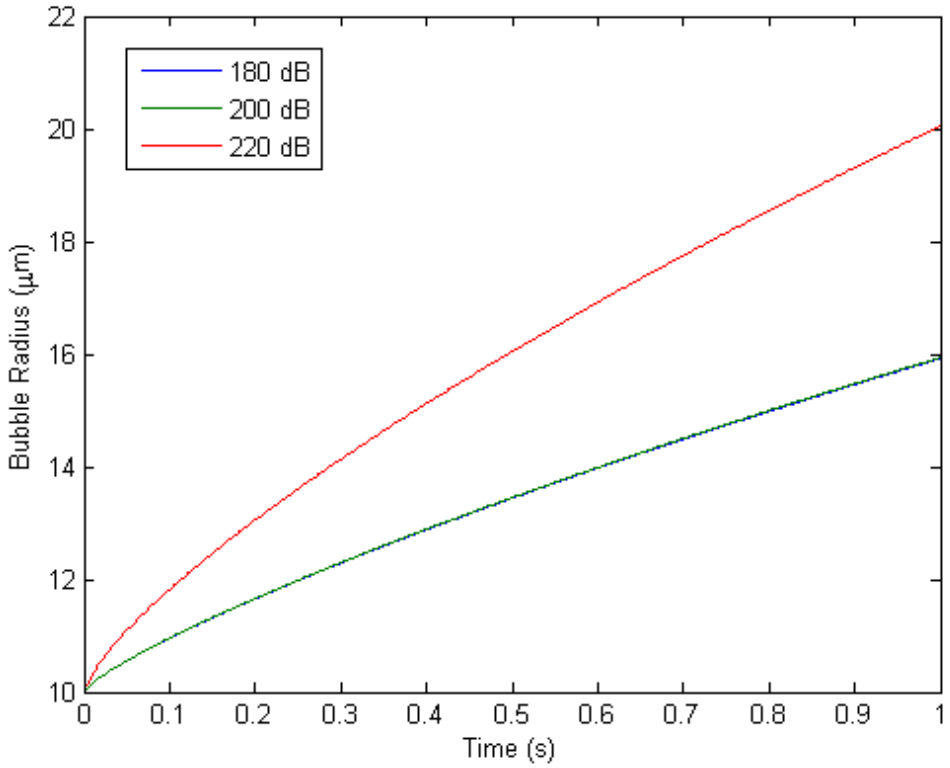

Figure 1. Predicted growth of a bubble of initial radius 10 µm in an acoustic field of different amplitudes. (Note that the 180 dB (blue line) and 200 dB (green line) curves almost overlay each other, they are the lower curves that terminate at 16 mm at 1 s.)

This prediction was consistent with mounting evidence suggesting that rectified diffusion plays no role in bubble growth at received levels of less than 200 dB at the sonar frequencies that the whales were exposed to shortly before mass beaching. The source levels of the sonar used in these cases were less than 230 dB SPL (re: 1 µPa at 1 m) (Cox *et al.* 2006) and received levels decay as a function of the

reciprocal of intensity squared. It therefore seems highly improbable that rectified diffusion could cause the macroscopic bubbles discovered in all of the whales, as they are unlikely to have occupied only the area within a 100 m radius of the source where received levels would have been sufficiently high to cause bubble growth by rectified diffusion in their tissues. This study therefore provides evidence, confirmed by other research groups, that rectified diffusion is unlikely to play an important role in the formation of macroscopic bubbles in cetaceans in the presence of mid-frequency sonar. If it is not rectified diffusion that causes bubble growth, then the only remaining plausible candidate mechanism for bubble growth is static diffusion.

6. Conclusions

Although there remains the scope for rectified diffusion to play a role in the initial destabilisation of bubble nuclei present in the tissues of beaked whales exposed to naval sonar, it has been shown here, in line with mounting evidence from other authors (Crum *et al.* 2005, Potter 2004), that static diffusion is the primary mechanism of such growth. As a result of this work, models designed to estimate how the partial pressure of nitrogen, as the inert and largest component of air, changes in the tissues of whales during diving will be developed so that the resulting risk of static diffusion-induced bubble growth can be estimated. This will provide a means of assessing the role of any behavioural responses to sonar elicited by the whales and any resulting risk of bubble growth by static diffusion to cause the DCS-like pathologies observed in mass stranded whales.

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Glossary

Acute (symptoms): those that arise over a short period of time

Adipocytes: fat cells

Catecholamine: any of a group of amines composed of a pyrocatechol molecule and the aliphatic portion of an amine that have important physiological effects as neurotransmitters and hormones, such as adrenaline and noradrenaline

Clinicopathologic: relating to the signs and symptoms that are observed in a patient, in conjunction with the results of laboratory examination

Congestion: abnormal accumulation of blood or fluid in part of the body

Diffuse (damage): occurring throughout the body

Embolism: blockage of a blood vessel by a blood clot, a piece of tissue, an air bubble, or a foreign object

Endogenous: originating or produced within an organism, tissue or cell

Epidural: the space where spinal fluid circulates around the cord

Haemorrhaging: heavy bleeding

Inflammatory: characterised by inflammation

Interlobular: between the lobes (of the liver)

Interstitial: pertaining to or located between the small spaces and gaps

Lesion: an injury or wound

Multifocal: occurring in more than one location

Necrosis: cell death

Neoplastic processes: a group of cells undergoing abnormal and uncontrolled cell growth

Osteonecrosis: the death of osteocytes (bone cells)

Pathogenetic: disease-causing

Pathognomonic: characteristic or symptomatic of a particular disease or condition

Peribullar and pterygoid sinuses: air cavities within the skull which are lined by mucous membranes

Retia: a network of blood vessels or nerves

Sinusoids: tiny endothelium-lined passages for blood in the tissue of an organ

Subcapsular: beneath the capsule of tissue around the outside of an organ

Vascular: pertaining to blood vessels

Appendix

The derivations of both the conventional equation that describes bubble growth within a sound field and the alternative form of that equation that was newly derived here are described in this appendix. A list of the nomenclature used is provided below:

C = concentration of gas dissolved within a liquid

C_{R_0} = dissolved gas concentration in liquid at bubble wall when bubble has equilibrium radius

C_∞ = initial uniform gas dissolved within a liquid

D = diffusion coefficient for dissolved gas within a liquid

R = radius of curvature of liquid/gas interface (e.g. radius of a spherical bubble)

R_g = the gas constant (8.31441 ± 0.00026 $JK^{-1}mol^{-1}$)

R_0 = the initial equilibrium radius of the bubble

N_m = total number of moles contained within a bubble

$N_{m,i}$ = initial number of moles contained within a bubble

T = absolute temperature (measured in Kelvin)

V = bubble volume

U_C = the difference in the mass of dissolved gas (rather than the concentration) from the initial conditions

h_R = a substitution parameter

r = the radial coordinate in the spherical frame, with origin at centre of a bubble (if present)

t = time

β = a resistive constant leading to damping

γ = ratio of specific heat of a gas at constant pressure to that at constant volume

ξ = substitution parameter

ρ = density (generally of a fluid, specifically of the fluid surrounding a bubble)

ρ_0 = equilibrium fluid density

σ = surface tension of a liquid

Derivation of the conventional equation

The following sketches the deviation of the conventional rectified diffusion model described by Leighton (1994, p383-401); the final expression obtained being equation (4.332) in (Leighton, 1994), which was original due to Church (1988).

Consider Fick's Law controlling mass flux, (1), expressed in a spherical polar co-ordinate system.

$$\frac{\partial C}{\partial t}+V(r)\frac{\partial C}{\partial r}=D\left(\frac{\partial^2 C}{\partial r^2}+\frac{2}{r}\frac{\partial C}{\partial r}\right) \tag{1}$$

Applying the substitution used by Eller and Flynn (1965) leads to

$$D\left(1+\frac{3h_R}{R^3}\right)^{4/3}\frac{\partial^2 U_C}{\partial {h_R}^2}=\frac{\partial U_C}{\partial \xi} \tag{2}$$

Expanding using the Taylor series, solving each order independently and combining the zeroth and first term gives:

$$N_m-N_{m,i}=4\left(C_\infty-C_{R_0}\frac{\langle R/R_0\rangle}{\left\langle\left(R/R_0\right)^4\right\rangle}\right)\left(2{R_0}^2\sqrt{\pi Dt\left\langle\left(R/R_0\right)^4\right\rangle}+\pi DtR_0\langle R/R_0\rangle\right) \tag{3}$$

Let

$$\bar{R}_1=\langle R/R_0\rangle;\quad \bar{R}_4=\left\langle\left(R/R_0\right)^4\right\rangle$$

This produces:

$$N_m-N_{m,i}=4\left(C_\infty-C_{R_0}\frac{\bar{R}_1}{\bar{R}_4}\right)\left(2{R_0}^2\sqrt{\pi Dt\bar{R}_4}+\pi DtR_0\bar{R}_1\right) \tag{4}$$

Assuming R_0 is constant (discussed later), differentiation with respect to t gives:

$$\frac{dN_m}{dt}=4\pi DR_0\left(C_\infty-C_{R_0}\frac{\bar{R}_1}{\bar{R}_4}\right)\left(R_0\sqrt{\frac{\bar{R}_4}{\pi Dt}}+\bar{R}_1\right) \tag{5}$$

It is also known that C_{R_0} can be expressed in terms of C_0 using the following equation:

$$C_{R_0} = \frac{C_0}{p_0}\left(p_0 + \frac{2\sigma}{R_0}\right) = C_0\left(1 + \frac{2\sigma}{p_0 R_0}\right) \tag{6}$$

Substituting this expression for C_0 into (5) produces the following equation:

$$\frac{dN_m}{dt} = 4\pi D R_0\left(C_\infty - C_0\left(1 + \frac{2\sigma}{p_0 R_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\right)\left(R_0\sqrt{\frac{\bar{R}_4}{\pi D t}} + \bar{R}_1\right) \tag{7}$$

This equation is expressed in terms of dN_m/dt, but for an equation describing bubble growth, an equation expressed in terms of dR_0/dt is required. Expressing (7) in terms of the rate of change of the radius using the (isothermal) Ideal Gas Law is therefore employed as this provides a relationship between N and R:

$$pV = \frac{4\pi R_0^{\,3}}{3}\left(p_0 + \frac{2\sigma}{R_0}\right) = \frac{4\pi}{3}R_0^{\,3}p_0 + \frac{8\pi}{3}\sigma R_0^{\,2} = N_m R_g T \tag{8}$$

This can then be re-arranged to give equation (10) below, which is effectively (4) expressed with dR_0 as the subject in place of dN_m:

$$dN_m = \frac{1}{R_g T}\left(4\pi R_0^{\,2} p_0 dR_0 + \frac{16\pi}{3}\sigma R_0 dR_0\right) = \frac{4\pi R_0}{R_g T}\left(R_0 p_0 + \frac{4}{3}\sigma\right)dR_0 \tag{9}$$

$$\frac{4\pi R_0}{R_g T}\left(R_0 p_0 + \frac{4}{3}\sigma\right)\frac{dR_0}{dt}$$
$$= 4\pi D R_0 C_0\left(1 + \frac{2\sigma}{p_0 R_0}\right)\left(\frac{C_\infty}{C_0}\left(1 + \frac{2\sigma}{p_0 R_0}\right)^{-1} - \frac{\bar{R}_1}{\bar{R}_4}\right)\left(R_0\sqrt{\frac{\bar{R}_4}{\pi D t}} + \bar{R}_1\right)$$

$$\frac{dR_0}{dt} = \frac{D R_g T}{R_0 p_0\left(1 + \frac{4\sigma}{3R_0 p_0}\right)} C_0\left(1 + \frac{2\sigma}{p_0 R_0}\right)\left(\frac{C_\infty}{C_0}\left(1 + \frac{2\sigma}{p_0 R_0}\right)^{-1} - \frac{\bar{R}_1}{\bar{R}_4}\right)\left(R_0\sqrt{\frac{\bar{R}_4}{\pi D t}} + \bar{R}_1\right) \tag{10}$$

This is what we refer to as the conventional equation used to model the growth of a bubble in a sound field.

Alternative model of Rectified Diffusion

This section details the derivations of an alternative equation describing bubble growth through rectified diffusion. The derivation of (10) contains an inherent contradiction. To obtain (4) from (5), the differentiation assumes that R_0 is constant, which is at odds with the final result (10), which defines the rate of change of R_0. This contradiction is unnecessary and this subsection derives an alternative expression for bubble growth in which the assumption of constant R_0 is avoided.

Using the definitions below and substituting these and equation (6) into equation (3) so that all of the dependencies on R_0 are explicit gives:

$$\bar{R}_1 = \frac{\tilde{R}_1}{R_0}; \quad \bar{R}_4 = \frac{\tilde{R}_4}{R_0^4}$$

$$\tilde{R}_1 = \langle R \rangle; \quad \tilde{R}_4 = \langle R^4 \rangle$$

$$N_m = 4\left(C_\infty - C_0 \left(1 + \frac{2\sigma}{R_0 p_0} \right) \frac{\bar{R}_1}{\bar{R}_4} \right) \left(2R_0^2 \sqrt{\pi D t \bar{R}_4} + \pi D t R_0 \bar{R}_1 \right) + N_{m,i} \tag{11}$$

For the purposes of simplification we define

$$\gamma(t) = \left(2R_0^2 \sqrt{\pi D t \bar{R}_4} + \pi D t R_0 \bar{R}_1 \right)$$

Allowing (11) to be written as

$$N_m = 4\left(C_\infty - C_0 \left(1 + \frac{2\sigma}{R_0 p_0} \right) \frac{\bar{R}_1}{\bar{R}_4} \right) \gamma(t) + N_{m,i}$$

Differentiating this with respect to t gives:

$$\frac{dN_m}{dt} = 4\left(C_\infty - C_0 \left(1 + \frac{2\sigma}{R_0 p_0} \right) \frac{\bar{R}_1}{\bar{R}_4} \right) \dot{\gamma}(t) + 4\gamma(t) C_0 \left(\frac{2\sigma}{R_0^2 p_0} \right) \frac{\bar{R}_1}{\bar{R}_4} \dot{R}_0 \tag{12}$$

where

$$\dot{\gamma}(t) = 4R_0 \dot{R}_0 \sqrt{\pi D t \bar{R}_4} + R_0^2 \sqrt{\frac{\pi D \bar{R}_4}{t}} + \pi D R_0 \bar{R}_1 + \pi D t \dot{R}_0 \bar{R}_1$$

$$= R_0^{\,2}\sqrt{\frac{\pi D\bar{R}_4}{t}} + \pi D R_0 \bar{R}_1 + \left(4R_0\sqrt{\pi Dt\bar{R}_4} + \pi D\bar{R}_1\right)\dot{R}_0$$

Which can be written as

$$\dot{\gamma}(t) = \dot{\gamma}_1(t) + \dot{\gamma}_2(t)\dot{R}_0$$

In which

$$\dot{\gamma}_1(t) = R_0\left(R_0\sqrt{\frac{\pi D\bar{R}_4}{t}} + \pi D\bar{R}_1\right)$$

$$\dot{\gamma}_2(t) = 4R_0\sqrt{\pi Dt\bar{R}_4} + \pi Dt\bar{R}_1$$

Equations (9) and (12) can be combined to eliminate dN_m/dt leading to:

$$\left(C_\infty - C_0\left(1+\frac{2\sigma}{R_0 p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\right)\dot{\gamma}(t) + \gamma(t)C_0\left(\frac{2\sigma}{R_0^{\,2}p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\dot{R}_0 = \frac{\pi R_0}{R_g T}\left(R_0 p_0 + \frac{4}{3}\sigma\right)\dot{R}_0$$

Re-arrangement to dR_0/dt the subject:

$$\beta_1\dot{\gamma}(t) + \beta_2\dot{R}_0 = \beta_1\dot{\gamma}_1(t) + \beta_1\dot{\gamma}_2(t)\dot{R}_0 + \beta_2\dot{R}_0 = \beta_3\dot{R}_0$$

$$\beta_1 = \left(C_\infty - C_0\left(1+\frac{2\sigma}{R_0 p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\right);\quad \beta_2 = \gamma(t)C_0\left(\frac{2\sigma}{R_0^{\,2}p_0}\right)\frac{\bar{R}_1}{\bar{R}_4};\quad \beta_3 = \frac{\pi R_0}{R_g T}\left(R_0 p_0 + \frac{4}{3}\sigma\right)$$

$$\left(\beta_3 - \beta_1\dot{\gamma}_2(t) - \beta_2\right)\dot{R}_0 = \beta_1\dot{\gamma}_1(t) \quad\Rightarrow\quad \frac{dR_0}{dt} = \frac{\beta_1\dot{\gamma}_1(t)}{\beta_3 - \beta_1\dot{\gamma}_2(t) - \beta_2}$$

$$\frac{dR_0}{dt} = \frac{\left(C_\infty - C_0\left(1+\frac{2\sigma}{R_0 p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\right)\left(R_0^{\,2}\sqrt{\frac{\bar{R}_4}{D\pi t}} + R_0\bar{R}_1\right)}{\frac{R_0}{DR_g T}\left(R_0 p_0 + \frac{4}{3}\sigma\right) - \left(C_\infty - C_0\left(1+\frac{2\sigma}{R_0 p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}\right)\left(4R_0\sqrt{\frac{t\bar{R}_4}{D\pi}} + t\bar{R}_1\right) - \left(2R_0^{\,2}\sqrt{\pi Dt\bar{R}_4} + \pi Dt R_0\bar{R}_1\right)C_0\left(\frac{2\sigma}{R_0^{\,2}p_0}\right)\frac{\bar{R}_1}{\bar{R}_4}}$$

This is the expression that was used to describe the growth of a bubble in a sound field in the model presented in this report.

Comparing this with the original model, (10) one can see that this modification has resulted in two extra terms appearing in the denominator.