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OCULAR TEAR FILM BUBBLE COUNTS AFTER RECREATIONAL COMPRESSED AIR DIVING

Michael H Bennett, David J Doolette and Nicole Heffernan

Key Words

Air, bubbles, decompression, eyes, recreational diving, research.

Abstract

Previous authors have demonstrated an increase in tear film bubble counts following dry, compressed air dives. We examined the lower tear film meniscus for the presence of bubbles in 42 divers after compressed air dives on a single day and in 11 divers undergoing repetitive, multi-day diving exposures over 5 days. Following diving, bubble counts increased significantly ($P < 0.01$) from pre-dive values. From a pre-dive median (inter-quartile range) of 0 (0-0.33) bubbles/eye, single day divers reached a maximum bubble count at 48 hours after diving of 1 (0-2.25) bubbles/eye. Similarly, from a pre-dive count of 0.33 (0-1) bubbles/eye, multi-day divers had increased bubble counts from 24 hours following their first dive until 24 hours following their final dive when counts were 1.67 (0.92-3.08) bubbles/eye. Bubble counts were not significantly correlated with inert gas load, body mass index, age or diving experience. We confirm that tear film bubble counts are raised following wet compressed air diving as previously described following dry diving.

Introduction

Following the serendipitous discovery of bubbles in the ocular tear film of a hyperbaric attendant following a dry chamber exposure to compressed air breathing,[1] several studies have demonstrated a relationship between the appearance of such bubbles and decompression stress in dry chamber dives. Bubble counts are uniformly increased following dry dives to PADI table no-stop limits between 12 and 36 m. Furthermore, counts increase with both increasing bottom times at a given depth and with exercise at depth, and are decreased following periods of oxygen breathing.[2-4] Tear film bubbles may be detectable in the absence of ultrasonic Doppler detectable venous bubbles and may persist for long periods following a compression exposure, typically up to 48 hours.[5]

Three possible practical uses may follow from these interesting observations. Firstly, counts may prove to be useful in estimating the decompression stress following standard depth, time and decompression exposures. This would assist in attempts to compare the risk of decompression sickness (DCS), between alternative diving schedules and in quantifying the risk of particular exposures. Secondly, repeated tear film examinations in individuals subject to frequent periods of compression and decompression might prove to be a sensitive monitor of mounting decompression stress. Finally, on a clinical level, tear film examination in symptomatic individuals may prove to be an important diagnostic aid in DCS.

To date, no data have been published on tear film bubbles in association with wet compressed gas diving. It is the aim of the present two studies to document the number of bubbles appearing in the lower lid tear film meniscus following single, repetitive, and multi-day compressed air diving in the marine environment. The specific hypotheses are that underwater compressed air diving causes an increase in the number of tear film bubbles that is sustained throughout multi-day diving and that bubble number is correlated with decompression stress.

Materials and Methods

Both studies were approved by the South Eastern Sydney Area Health Service Ethics Committee before commencement of enrolment and involved only adult volunteers who were appropriately trained and qualified to undertake scuba diving activity. Volunteers were excluded if they gave any history of ocular disease (apart from refractive errors), tear film dysfunction, medical reasons for not diving on the selected dive day, or if they had undertaken compressed gas breathing in the seven days prior to the study. Divers were equipped with standard recreational open circuit scuba with half-face mask and all dives were in seawater.

Study one: single day diving

45 volunteers were recruited from local dive clubs to undertake two air dives on a single day. A dive leader holding a minimum qualification of dive master was selected for each dive boat and all divers were instructed on the planned depth and time profiles. The first dive was to consist of a 25 minute dive to 25 m, followed by a controlled ascent to a safety stop of 5 minutes at three m. After a surface interval of 1.5 hours, a second dive was made to 20 m for 25 minutes with similar ascent rates and safety stop. Each of these was designed to fall within the maximum no-stop dive time allowed (PADI tables). All dives were recorded on personal depth and time recorders and later downloaded using commercial software (Datatrak, v2.03, Dynatron AG, Zurich). Water temperature varied from 17° to 20° C (63° to 68° F).

The ocular tear film bubbles on each eye were counted prior to diving, then again at 1 hour, 12, 24, 48 and 72 hours post-diving using a slit lamp (SL900, Haag-Streit, Switzerland) employing a standard technique described elsewhere.[6] Briefly, the subject is examined by sweeping the slit across slowly from the medial to the lateral border of the inferior tear film gutter, counting bubbles, if any. It is important to limit the inspection to the gutter itself in order to standardise the examination and because small bubbles on the lid itself are not uncommon as a result of physical foaming after blinking. The subject is asked to close their eyes for 5 seconds, open them again and the examination is repeated. Three sweeps are made and the bubble count averaged before the procedure is repeated in the other eye.

Study two: repetitive, multi-day diving

Eleven adult divers who were planning to undertake a series of ten compressed air dives over a five day period were recruited. No attempt was made to standardise the individual dives, however, all were undertaken in the same series of dive sites and the depth and time profiles were recorded as for study one. Water temperature for this series of dives was 24° to 28° C (75° to 82° F).

Ocular tear film examinations were made, by the standard method described above, prior to the first dive, within two hours of completion of each day's diving and then at 24, 48 and 72 hours following the last dive. All volunteers completed at least 7 of the planned dives and 9 of 11 divers completed all examinations.

Statistical analysis

Both volunteer groups were recruited as a convenience sample and no power calculations were made concerning any magnitude of difference in tear film bubble counts. Median tear film bubble counts per eye were compared using the Friedman test for multiple comparisons (a non-parametric equivalent of ANOVA). The Shapiro-Wilk W test for non-normality was employed on bubble count distributions and the Mann-Whitney U test for individual comparisons of bubble counts when distributions were unlikely to be normal.

In study one, the effect of potentially important determinants of bubbles in individual divers (age, body mass index [BMI] and previous diving experience) was examined using non-parametric correlation and regression (Kendall's rank correlation method, which provides a distribution free measure of the strength of dependence between two variables). In study two, the possible relationship between bubble count in individuals and decompression stress was examined using the same statistical methods. An index of tissue inert gas loading was used as an estimate of decompression stress for each dive. The gas-loading index was calculated as the product of maximum gauge pressure (in metres of seawater) during a dive and the square root of total bottom time (P√T).[7] For each diver, P√T was summed for all dives each day (daily gas load) for comparison with daily bubble scores and P√T was summed for all dives in the 5 day period (cumulative gas load) for comparison with peak and cumulative bubble scores.

All calculations were made using StatsDirect statistical software, version 1.611, (Iain Buchan, 2000).

Results

STUDY ONE

45 divers entered the water for the first dive and the spread of maximum depths and in-water times recorded are shown in Figure 1. Three individuals had markedly different profiles (In-water time <12 mins and/or maximum depth <20m) and these individuals were excluded from further analysis.

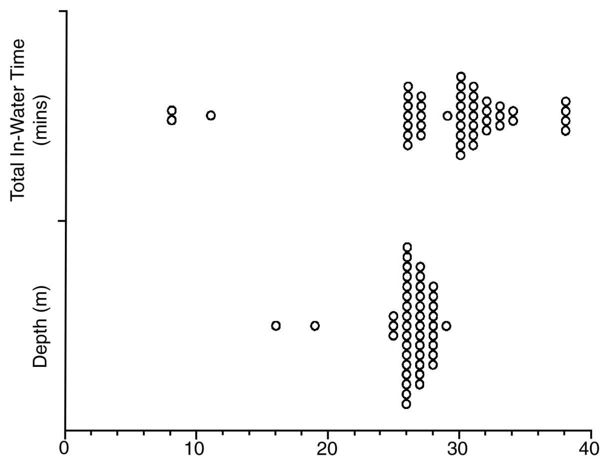

Figure 1. Dive one: depth and time exposures for all divers in study 1. X-axis is metres of seawater or minutes. The three individuals with dive time less than 12 minutes were excluded from analysis.

Due to inclement weather and water conditions, only 31 divers undertook the second dive. Most completed the planned depth and time exposures, although three individuals spent 47 to 48 minutes at 15 to 16 m and one further individual spent 34 minutes at a maximum depth of 28 m. These individuals were retained in the analysis. The spread of exposures to both depth and time is shown in Figure 2.

Of the 42 divers included in the analysis, 10 were female. The included group had a mean age of 32.7 yrs

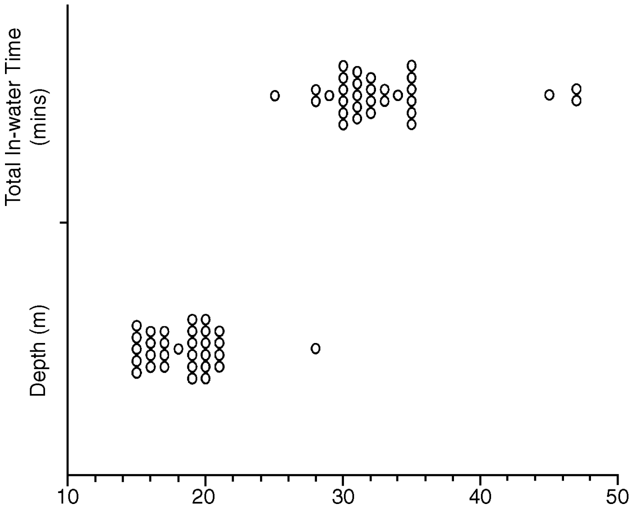

Figure 2. Dive two: depth and time exposures for all divers in study 1. X-axis is metres of seawater or minutes.

(SD 9.0 yrs, n=42), average body mass index (BMI) of 25.6 (SD 4.6, n=42) and a wide range of previous diving history. Median years of diving activity were 4 years (range 0.4 to 41) and median number of previous dives was 100 (range 3 to 5,000). Four subjects were regular smokers.

Fifty six of the 84 eyes (66.7%) showed no bubbles on examination prior to diving and 47.6% of subjects (20/42) showed no bubbles in either eye at this time. Maximum bubble numbers were seen at 48 hours following diving, although the numbers of eyes and individuals who remained bubble free was still substantial at 34.5% of eyes (29/84) and 23.8% of individuals (10/42). The distribution of bubbles prior to diving and at 48 and 72 hours is shown in Figure 3. The distributions are unlikely to be normal at any examination (*P* ≤0.0001, Shapiro-Wilks W test).

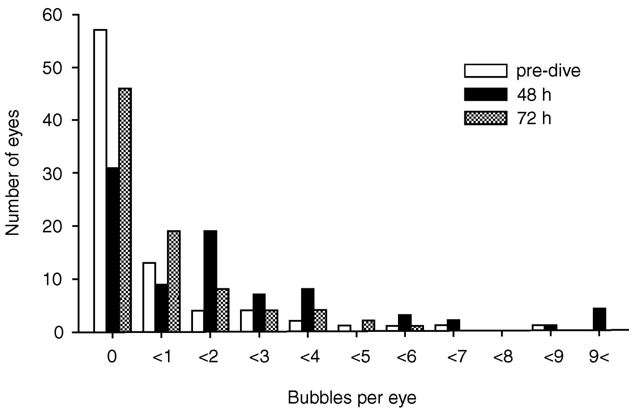

Figure 3. Bubble counts in individual eyes at pre-dive, 48 hours and 72 hours following diving in study 1. Bubble counts are the average of 3 sweeps and are grouped from left to right as 0, 0< to <1, 1 to <2, 2 to <3, etc.

Median and inter-quartile range tear film bubble counts per eye for all examinations are shown in Figure 4. The Friedman test yields a significant value ($P \leq 0.0001$), suggesting a true difference in bubble counts between at least two groups. Friedman multiple comparisons model yields statistically significant differences between pre-dive bubble counts and counts at all examinations except 72 hours, (versus post-dive $P < 0.0001$, 12 hours $P = 0.001$, 48 hours $P < 0.0001$, 72 hours, $P = 0.09$).

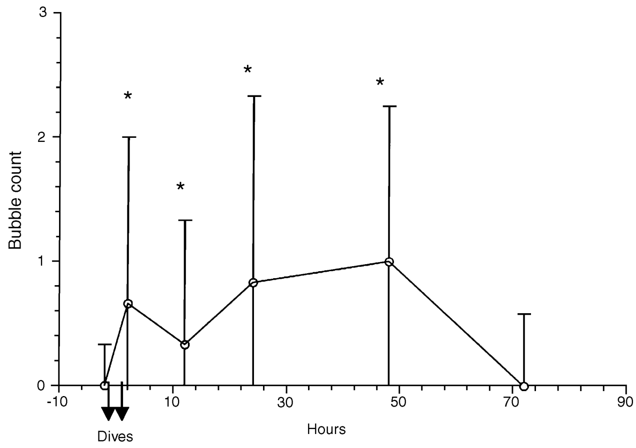

Figure 4. Median bubble counts in individual eyes before and after single day diving in study 1. Arrows on x-axis indicate the time of the two dives. Error bars are inter-quartile range. Asterisks indicate significant different from pre-dive (P<0.05).

Bubble counts at 48 hours after diving were compared between those completing one dive versus those completing both dives, however there was no significant difference ($P = 0.82$, Mann-Whitney U test). A similar analysis was made to compare bubble numbers in males versus females and those who wore contact lenses. While there was a trend to more bubbles in females, this was not statistically significant (median difference females 0.5 bubbles more per individual, 95% CI -0.33 to 2.0, $P = 0.14$). Eight divers wore soft contact lenses while diving and all kept them in during the examinations. The area under the contact lens was not examined for bubbles and there were no significant differences in inferior tear film meniscus bubble counts between those with and without lenses (at 48 hours, 1.3 bubbles/person with lenses, versus 1.6 without, $P = 0.69$, Mann-Whitney U test).

The relationship between bubble counts at 48 hours and age, diving experience and BMI were examined using Kendall's rank correlation coefficient (RCC) and non-parametric linear regression. No significant relationships were evident (with age: $P = 0.91$, RCC -0.01, total dives logged: *P* = 0.53, RCC 0.54 and BMI: *P* = 0.36, RCC -0.11).

STUDY TWO

Eight divers completed all 10 dives, one diver completed only 7 dives, one completed 8 dives, and another 9 dives. Overall, the mean maximum depth of each dive was 23.0 metres (SD 4.6m, n=104) and the mean time underwater for each dive was 44.0 minutes (SD 5.5 min, n=104). Mean P√T for each individual dive was 151.4 (SD 27.8, n=104) and mean daily gas load was 288.2 (SD 69.3, n=55). The cumulative gas load for the 5 day period for each diver is shown in Figure 5.

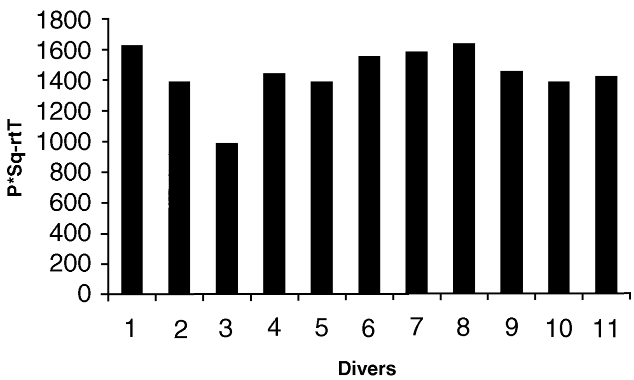

Figure 5. Cumulative inert gas load index for each diver in study 2. Bars show the sum for all dives during the 5 day period.

Median tear film bubble numbers and inter-quartile range for each examination are shown in Figure 6. The bubble counts rose over the period of diving (days 1-5) and then decreased until at 72 hours (day 8 examination) they were not significantly different from the pre-dive count. As with study one, the distributions of bubbles at each examination were unlikely to be normal (*P* < 0.05, Shapiro-Wilks W test), making comparisons using ANOVA unhelpful.

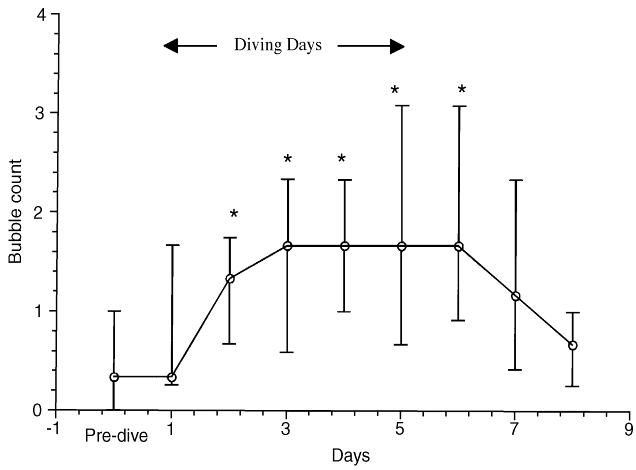

Figure 6. Median bubble counts in individual eyes during study 2. Dives were conducted on days 1-5 as indicated by the arrows. Error bars are inter-quartile range. Asterisks indicate significant different from pre-dive (P<0.05).

The Friedman test yields a significant value ($P \leq 0.0001$) and suggests a true difference in bubble counts between at least two groups. Friedman multiple comparison model yields statistically significant differences between pre-dive bubble counts and counts at days 2 to 6 (versus day 1 *P* = 0.21, day 2 *P* = 0.0009, day 3 *P* <0.0001, day 4 *P* < 0.0001, day 5 *P* < 0.001, day 6 *P* < 0.0001). Similar comparisons with counts on days 7 and 8 could not be made because of data loss.

Three possible relationships between gas load and bubble counts were examined using non-parametric regression and correlation. Daily gas load was correlated with the bubble count taken 24 hours following the end of that day's diving. However, this correlation is difficult to interpret because tear film bubbles following diving persist for longer than the interval between the daily diving exposures. Therefore, for each diver, cumulative gas load was correlated against both peak (day 6) bubble counts and against cumulative bubble counts (sum of bubble counts for days 1-6). Table 1 lists the assumptions implicit in these models and the results of these analyses, none yielded any significant relationship.

TABLE 1

REGRESSIONS OF BUBBLE COUNTS AGAINST P√T IN STUDY 2

Correlation and Regression	Assumption Regarding Diving	Kendall Regression Correlation Co-efficient (RCC)	*P*-value
Bubbles Day 6 and cumulative PT	Decompression stress cumulative	-0.17	0.45
Bubbles 24 hours after each diving day and P√T for that day	Decompression stress each day independent	-0.01	0.88
Cumulative total bubbles and cumulative P√T	No assumption	0.05	0.88

INTER-OPERATOR VARIABILITY

In study one, four slit-lamp operators were involved in the tear film examinations. Two examiners performed the majority of eye examinations (386/540, 71.5% and 120/540, 22.2% of examinations respectively). There was some inter-examiner variability between these two operators. The principle operator counted a median of 0.33 bubbles less per eye than the second operator, 95% CI -0.66 to 0 bubbles, $P < 0.01$ (Mann-Whitney U test). In study two, a single operator (MB) performed 91% of examinations. Two others performed the remaining 9%. There were no significant differences between operators and bubble counts recorded in this study between the principal operator and all others ($P = 0.92$, Mann-Whitney U test).

Discussion

With these studies we have established for the first time the presence of an increased number of bubbles in the lower tear film meniscus following compressed air breathing in the marine environment. The numbers of bubbles detected are a little lower than those found after dry dives in a hyperbaric chamber (1,2,3), and it is possible our technique for the detection of bubbles is somewhat less sensitive than those described by these authors. The persistence of bubbles in significant numbers for 48 hours after decompression is consistent with previous findings.[5] Regardless of continued diving (within PADI table limits), bubble counts rise 24 hours after the first dive and remain elevated and stable while diving continues and for 24-48 hours after the last dive.

Contrary to previous reports, no correlation was found between number of tear film bubbles following diving and different levels of decompression stress.[2] This is probably a limitation of the present studies. In study two, it is clear from the coefficient of variation of the maximum diving depth (20%), time underwater (13%), and daily decompression stress (24%) that the pattern of diving was relatively homogeneous. Tear film bubble count may be insensitive to small variations in decompression stress. Similarly in study one, no difference in tear film bubble number was found between divers who completed one or two dives, but both the single and repetitive dives schedules were at the no-stop limit and may result in similar decompression stress.

The persistence of tear film bubbles for at least 48 hours following diving also complicates the correlation of daily bubble counts with daily decompression stress during multi-day diving. Firstly, bubbles detected on any particular day may represent the decompression stress 24 or 48 hours previously. Correlation of cumulative bubble counts with cumulative decompression stress (both sums across all days) is free from any assumed temporal relationship, but was not significant. Secondly, although common decompression algorithms do not account for cumulative decompression stress between dives separated by more than 18 hours, decompression stress may accumulate, for instance in the presence of stable tissue-bubble complexes. Correlation of peak (day 6) bubble counts with cumulative decompression stress assumes bubble counts represent a cumulative effect of all dives, but was not significant. However, in both cases tear film bubble count may be insensitive to the relatively small variation in cumulative decompression stress (coefficient of variation = 12%).

$P\sqrt{T}$ is an index of diffusion limited inert gas uptake at the end of dives with bottom time of less than 100 minutes duration.[7] As the actual depth/time profile and subsequent decompression is not considered, it is not a true measure of decompression stress per se. However, such classification of dives according to maximum depth and duration has proved useful as an index of decompression severity even amongst quite different decompression practices.[8] In study two, diving practices were sufficiently similar to justify use of $P\sqrt{T}$.

While the differences described in these studies are statistically significant, they are small in magnitude. There is no known pathological implication from the finding of increased bubble numbers of any magnitude in an individual. Furthermore, the observation of increased tear film bubbles following diving does not prove a causal relationship between compressed air breathing and ocular bubbles. A series of further studies are under way at the Prince of Wales Hospital to investigate any correlation between such bubbles and other activities such as running, swimming or snorkelling. If bubble counts are not raised after these activities, the case for a causal relationship between bubbles and diving will be strengthened.

Doubt remains as to the origin of these bubbles. Four possible sources have been suggested, none of which is supported by good evidence at this point. The bubbles may arise from vascular structures in the conjunctivae or from the aqueous humour and move by diffusion to the interstitium of the conjunctivae and thence into the tear film. This is most useful in explaining early bubbles following a decompression such as those described in previous small studies of dry diving. The time course of such bubbling is likely to be short and parallel the detection of central venous ultrasonic Doppler bubbles after decompression. The persistence of bubbles for 48 hours is more difficult to account for by this mechanism, although secondary diffusion from a more remote source is one possibility.

It has been suggested the bubbles may arise from the Meibomian (Tarsal) glands located in the upper and lower lids.[3] These are enormously enlarged holocrine sebaceous glands and open directly onto the palpebral margin. The glandular cells have an increasingly high lipid content as they mature and are the source of the lipid layer of the tear film, probably introduced to the other layers with blinking.[9,10] After a variable maturation period from days to weeks, the gland cells rupture into a short central duct and thence into the tear film. It is possible that bubbles may arise in these lipid-containing cells on decompression and remain stabilised in the lipid environment until discharged into the tear film with the lipid cell contents. While this more easily explains the persistence of bubbles than the vascular theory, such bubbles have not been reported emerging from the Meibomian glands during our study. On the other hand, bubbles and glandular excreta following pressure to the area of the glands has been reported.[3]

A third mechanism of bubble generation into the tear film may be the evolution of gas from the peri-scleral lipid tissue or sclera itself. The relatively long time course for the detection of these bubbles may reflect the ability of lipid structures to retain stabilised bubbles over this period. The fourth potential source of bubbles is the lacrimal gland, source of the aqueous and most voluminous of the tear film layers. There does not appear to be any substantial support for this proposition either experimentally or physiologically.

One third of individuals in study one had some bubbles in their tear films prior to diving. Whatever the actual source of bubbles following diving, this figure would indicate a mechanism for the generation of bubbles independent of compressed air breathing. Most ophthalmologists the authors have questioned have not previously noted bubbles during routine slit-lamp examinations. This may not be surprising, however, as these bubbles have no known significance and may pass unnoticed even when present. It seems most likely that the occasional bubbles seen prior to diving are caused by mechanical action during blinking or other ocular and peri-ocular movement. It may be that increased counts following diving merely reflect an increase in mechanical activity in and around the tear film meniscus. A study underway at present in our unit is an attempt to answer the question as to whether exposure to the marine environment and exercise in the water also raise tear film bubble counts.

Slit-lamp operator error is a possible confounder in the present studies, particularly as the operators were not masked to diving exposure. The two principle operators in study one detected bubbles to a significantly different degree in the eyes they examined. The technique described is likely to be highly subject to variations due to different sweep speeds, vigilance and familiarity with the instrument. We have overcome these sources of error as much as possible by developing a standard written protocol and frequent comparisons of technique through the study period. The practical limitations of the study, however, did not allow us to produce multiple operator data for each subject at each examination, or to develop a masked comparison with a control group. Further studies should develop such methodologies or adopt a more objective and standard technique for bubble estimation. A technique using photography and volumetric measurements of bubbles has been reported.[3] We feel the advantage of our approach lies in the minimal equipment required, allowing the possibility of remote clinical diagnosis of significant decompression stress.

It is certainly our clinical impression from a small number of divers attending for the treatment of DCS, that bubble counts may be raised on presentation and subside with oxygen breathing at depth. If tear film bubble counts are to prove useful as a diagnostic tool, future studies will have to demonstrate a significant relationship between decompression stress and bubble counts by examining patients presenting for treatment with clinical evidence of DCS.

We have demonstrated a modest, but statistically significant, rise in bubble counts following compressed air diving within PADI table limits. These modest rises may be of use in comparing decompression stress for putative or experimental dive profiles. Future studies may attempt to develop tear film bubble counting as a method of differentiating those with DCS from those in which DCS is unlikely.

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Declaration

The authors declare that we have no financial interest in any commercial product involved in this research and received no financial assistance for the conduct of these studies.

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NEUROLOGICAL SYMPTOMS DEVELOPING WHILE DIVING

R M Bateman and R N Sawyer Jr

A 25 year old woman presented with left sided weakness and a patchy right sided sensory loss, which developed while diving in Egypt.

Recompression therapy was started, leading to an unsustained improvement in symptoms and signs.

Magnetic resonance imaging for here cervical spine subsequently showed a lesion at the level of C4 (Figure 1) and transverse myelitis was diagnosed. The patient may a nearly full recovery after steroids were given.

This case illustrates the importance of a thorough neurological examination for all suspected cases of decompression illness. It is also a poignant reminder that neurological symptoms and signs in divers may be due to neurological disease other than decompression sickness.

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