

Erythropoietin concentrations in obstructive sleep apnoea

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Abstract

Eight patients with obstructive sleep apnoea and a normal haemoglobin concentration underwent nocturnal studies during which oxyhaemoglobin saturation was recorded continuously with an ear oximeter and serum erythropoietin concentration was measured hourly by means of a radioimmunoassay. Serum erythropoietin concentrations remained within the normal range throughout the study despite falls in oxyhaemoglobin saturation in individuals to 33-78%. There was no relation between the degree of nocturnal hypoxaemia and serum erythropoietin concentrations. The brief cyclical episodes of hypoxaemia typical of obstructive sleep apnoea may not be a sufficient stimulus for erythropoietin secretion.

Hypoxaemia in normal subjects (for example, at high altitude) stimulates renal erythropoietin production and release with subsequent increases in bone marrow red cell production and circulating red cell mass.^{1,2} Secondary polycythaemia in chronic obstructive lung disease is generally believed to be mediated through this mechanism. Only a proportion of patients with hypoxaemia and lung disease, however, develop polycythaemia and only about half of these have raised serum concentrations of erythropoietin.³ Severe nocturnal hypoxaemia has been postulated as the cause of polycythaemia in chronic obstructive lung disease³ and it has been suggested that intermittent hypoxia may stimulate erythropoietin production.⁴ Patients with obstructive sleep apnoea rarely develop

polycythaemia despite repeated falls in arterial oxyhaemoglobin saturation (Sao₂) during six to eight hours of sleep. These patients provide a model for studying the characteristics of erythropoietin release in response to intermittent severe hypoxaemia in man.

Methods

Eight men with newly diagnosed and untreated obstructive sleep apnoea underwent all night sleep studies. Four of the patients had daytime hypoxaemia (arterial oxygen tension (PaO₂) less than 10 kPa) and two had airflow obstruction with a forced expiratory volume in one second (FEV₁) below 80% predicted; none was polycythaemic. The mean age of the patients was 52.5 (range 42-70) years and three were current smokers. The results of their haematological, biochemical, and physiological tests are summarised in the table. Daytime oxyhaemoglobin saturation was calculated from the arterial oxygen tension measured during the day. Arterial oxyhaemoglobin saturation was estimated during sleep with an ear oximeter (Ohmeda Biox IIa) accurate to within 2.5% down to an Sao₂ of 60%. A compressed summary of the all night recording of Sao₂ was constructed by joining the peak and trough values from each episode of apnoea.

Serial venous blood samples were taken from an indwelling intravenous cannula before the onset of sleep, at roughly hourly intervals during sleep (more frequently in patient 5) and the following morning after waking. The total volume of blood taken was 60 ml in all subjects except patient 5, who had 120 ml removed. Samples were allowed to clot at room temperature. Serum was separated

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Baseline results and serum erythropoietin (sEPO) concentrations during wakefulness in eight patients with obstructive sleep apnoea

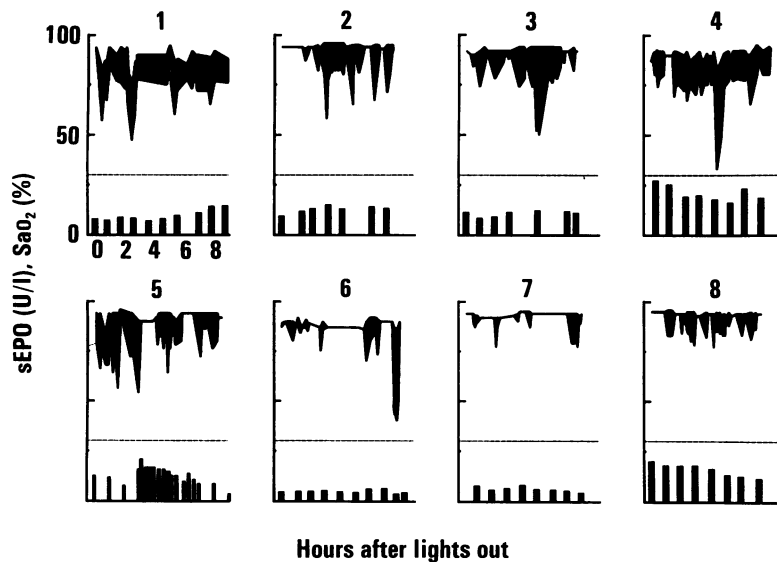
Patient No	Age (y)	PaO ₂ (kPa)	PaCO ₂ (kPa)	Sao ₂ (%)	Hb (g/dl)	Packed cell volume	FEV ₁ (% pred)	Creatinine (mmol/l)	Daytime sEPO (U/l)	
									10-12 am	8-10 pm
1*	42	8.1	6.5	90	16.6	0.49	73	71	8.2	—
2	62	13.6	5.6	97	14.4	0.44	94	111	11.9	10.6
3	70	9.5	6.0	93	13.8	0.42	83	71	12.5	11.8
4*	47	8.5	7.9	92	13.1	0.42	83	90	22.3	26.2
5	46	11.6	6.1	96	12.9	0.37	87	74	4.0	—
6*	56	6.9	6.4	89	15.0	0.48	58	140	—	4.9
7	49	14.0	5.5	98	15.0	0.48	90	97	9.6	—
8	48	10.1	5.5	95	15.8	0.49	92	68	11.9	12.2

*Current smoker.

PaO₂—arterial oxygen tension; PaCO₂—arterial carbon dioxide tension; Sao₂—arterial oxyhaemoglobin saturation; Hb—haemoglobin; FEV₁—forced expiratory volume in one second.

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Changes in oxyhaemoglobin saturation (SaO_2) and serum erythropoietin (sEPO) concentrations with time during an all night study of eight patients with obstructive sleep apnoea. SaO_2 is represented by the traces at the top and serum erythropoietin by the bars at the bottom of each section. The light horizontal line indicates the upper limit of the reference range of serum erythropoietin.

within four hours of collection and stored at -20°C until the time of assay. All samples from an individual were estimated at the same time and all the assays were completed in three runs.

Erythropoietin levels were measured by a modification of the radioimmunoassay method described by Egrie *et al.*,⁵ which uses reagents derived from recombinant human erythropoietin with delayed addition of the labelled antigen. The curve derived from the recombinant human erythropoietin was standardised against the World Health Organisation's second international reference preparation of human urinary erythropoietin. The reference range for serum erythropoietin in a population of 107 healthy individuals was 6–30 mU/ml (geometric mean 13.34). The intra-assay coefficient of variation was 8% and the interassay value 13% for serum erythropoietin concentrations of 10–100 mU/ml.

For statistical analysis simple regression was used to assess correlation between measurements and Student's paired *t* test to compare morning and evening erythropoietin concentrations. Analysis of variance was used to compare areas under the curve of erythropoietin concentration against time and SaO_2 against time.

Results

All eight patients had repeated episodes of hypoxaemia during sleep (figure). The lowest SaO_2 recorded during sleep in individual subjects ranged from 33% to 78%. The mean duration of an episode of desaturation (defined as a fall of 4% or more in SaO_2 from the baseline level while they were lying supine) was 50 seconds and the longest lasted three minutes.

The serum erythropoietin measurements were within the defined normal range in all the patients at all times. There was no relation between the serum erythropoietin concentration, haemoglobin, or "awake" SaO_2 and Pao_2

(table). Serum erythropoietin concentrations in blood samples taken between 10.00 and 12.00 hours the morning after the study did not correlate with the degree or duration of nocturnal hypoxaemia. Four patients (2, 3, 4, 8) had samples taken for measurement of serum erythropoietin concentrations while they were awake before (21.00–22.00 hours) and after the sleep study (10.00–12.00 hours). There was no significant difference in the values (mean (SD) 15.2 (7.3) and 14.6 (5.1) mU/ml).

There was no significant correlation between mean overnight erythropoietin concentration or any of the following measures of SaO_2 : mean SaO_2 ($r = 0.17$), mean SaO_2 at arousal ($r = 0.30$), mean SaO_2 between episodes of apnoea ($r = 0.21$), total hypoxic dose ($r = 0.12$), lowest SaO_2 ($r = 0.18$). There was no significant increase in erythropoietin overnight with time. There was also no significant correlation between the areas under the curves for SaO_2 against time or for serum erythropoietin concentrations against time (analysis of variance: $R^2 = 0.138$; $F = 0.962$, $p < 0.25$).

Discussion

Erythropoietin production in response to induced hypoxia has been investigated in animals but cannot readily be studied in man. Patients with obstructive sleep apnoea have recurrent spontaneous episodes of severe hypoxaemia and offer a human model for the study of erythropoietin secretion. Our results showed no relation between serum erythropoietin concentration and oxyhaemoglobin saturation.

We measured serum erythropoietin using a modification of a well established radioimmunoassay method⁵ that was both sensitive and reliable. Samples were taken at appropriate times to detect a rise in erythropoietin, as this has been reported to begin after as little as one hour of hypoxaemia in animal models.⁴ We do not believe that we missed bursts of secretion initiated by each individual apnoeic episode: the plasma half life of endogenous erythropoietin is at least three hours,¹ and we took samples as frequently as every five minutes in patient 5 (for two hours) and hourly in the rest. We postulated that there might be a cumulative effect secondary to night time hypoxaemia, but neither morning nor evening erythropoietin concentrations were significantly raised, which supports previous findings in patients with chronic obstructive lung disease.³ There was no significant difference between pre-study and post-study concentrations of erythropoietin (evening and morning), suggesting an absence of diurnal variation in our subjects. Early evidence was against the presence of diurnal variation in erythropoietin concentrations,⁶ apart from one report on a single subject.⁷ Recent work using the more sensitive radioimmunoassay showed clear evidence of diurnal variation in serum erythropoietin in 27 patients in hospital⁸ and these results have subsequently been repeated (P J Gomes, personal communication). The lack of diurnal variation in our patients with obstructive sleep apnoea

contrasts with these findings and may be explained by the disruption of sleep by cyclical hypoxaemia and consequent arousal. An alternative explanation would be that our subjects remained awake during their studies. As we did not perform electroencephalography to stage sleep we do not have documentary evidence to refute this suggestion; but the observations of our experienced sleep laboratory staff and the pattern of the oximetry recordings are against it.

A direct relation between the degree of hypoxaemia and erythropoietin concentrations has been described,¹ but despite falls in SaO_2 to below 50% in four patients we could detect no such change. Possibly our patients had adapted to their nocturnal hypoxaemia, as normal individuals do at altitude. In these circumstances there is an initial rise in erythropoietin but secretion then falls to within the normal range,² though polycythaemia is maintained. The patients in our study were not polycythaemic, which would seem to argue against this "adaptive" explanation. Longer episodes of constant rather than episodic hypoxaemia may be required to initiate erythropoietin secretion and increase red cell mass, such as occur in the "overlap" syndrome of obstructive sleep apnoea and chronic obstructive lung disease.⁹ It is, however, puzzling that only half of patients with chronic obstructive lung disease and polycythaemia have raised erythropoietin concentrations.³ Perhaps, as in normal individuals exposed to hypoxia, this is due to down regulation of erythropoietin secretion after an initial rise.^{1,2} None of our patients was polycythaemic, though four had some degree of daytime hypoxaemia. It therefore seems probable that sustained nocturnal hypoxaemia is needed to stimulate erythropoietin release and initiate an increase in red cell mass.

Other factors may also be important in the generation of polycythaemia. In patients with chronic obstructive lung disease carboxyhaemoglobin is the only variable that has been shown to correlate with erythropoietin concentration,⁶ and there is a clear association between

cigarette smoking and polycythaemia in this condition.¹⁰ Patients with sleep hypoxaemia often have low testosterone concentrations,¹¹ and as androgens stimulate erythropoiesis by an unknown mechanism androgen deficiency associated with obstructive sleep apnoea is possibly responsible for a reduction in erythropoietin production.

In conclusion, we did not find raised erythropoietin concentrations in patients with obstructive sleep apnoea and recurrent short episodes of severe nocturnal hypoxaemia. We suggest that prolonged episodes of nocturnal hypoxaemia may be necessary to stimulate erythropoietin production. These findings may explain why polycythaemia is uncommon in patients with obstructive sleep apnoea.

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