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Early origins of respiratory disease

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Abstract: Chronic respiratory morbidity is unfortunately common in childhood, particularly in those born very prematurely or with congenital anomalies affecting pulmonary development and those with sickle cell disease. Our research group, therefore, has focused on the early origins of chronic respiratory disease. This has included assessing antenatal diagnostic techniques and potentially therapeutic interventions in infants with congenital diaphragmatic hernia. Undertaking physiological studies, we have increased the understanding of the premature baby's response to resuscitation and evaluated interventions in the delivery suite. Mechanical ventilation modes have been optimised and randomised controlled trials (RCTs) with short- and long-term outcomes undertaken. Our studies highlighted respiratory syncytial virus lower respiratory tract infections (LRTIs) and other respiratory viral LRTIs had an adverse impact on respiratory outcomes of prematurely born infants, who we demonstrated have a functional and genetic predisposition to respiratory viral LRTIs. We have described the long-term respiratory outcomes for children with sickle cell disease and importantly identified influencing factors. In conclusion, it is essential to undertake long term follow up of infants at high risk of chronic respiratory morbidity if effective preventative strategies are to be developed.

Keywords: congenital diaphragmatic hernia; neonatal ventilation; resuscitation; sickle cell disease.

Introduction

Chronic respiratory morbidity is unfortunately common in childhood, particularly in those born very

prematurely or with congenital anomalies affecting pulmonary development and in those with sickle cell disease. Our research group, therefore, has focused on the early origins of chronic respiratory disease and how to better prevent and treat it. Bronchopulmonary dysplasia (BPD) is the most common adverse outcome of very premature delivery and results in chronic respiratory morbidity, lung function abnormalities and troublesome respiratory symptoms requiring treatment even into adulthood. It is diagnosed in infants who received supplementary oxygen for least 28 days and the severity determined at 36 weeks postmenstrual age. BPD has a multifactorial aetiology which includes volutrauma and oxygen toxicity. As a consequence, a focus of our research has been to use physiological assessments and randomised trials, to develop and optimise the use of respiratory support on the neonatal unit and during resuscitation on the delivery suite. We investigated whether prematurely born infants have a functional and/or genetic predisposition to respiratory syncytial virus (RSV) lower respiratory infections (LRTIs) or other viral LRTIs. In addition, we have determined if respiratory viral infections in infancy in the community or requiring hospital admission have adverse outcomes at school age and if those outcomes could be predicted by biomarkers in infancy. Infants with congenital diaphragmatic hernia (CDH) can suffer a high mortality and morbidity, despite advances in neonatal care. Thus, we have developed and evaluated antenatal diagnostic and therapeutic interventions with the aim of improving outcomes. Furthermore, we have undertaken trials to identify optimal neonatal management. Sickle cell disease can be a devastating condition. To further understand the risk factors for severe lung disease and its course in children, we investigated the impact of sickle cell disease (SCD) on lung function in cohorts of children and adults in the UK and Jamaica. The aim of this review is to describe the results of those research studies and how they have influenced clinical practice.

Neonatal respiratory support

There have been two main strategies to reduce BPD using invasive ventilation. One is to use respiratory support techniques which work synergistically with the infant's

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respiratory efforts – patient triggered ventilation (assist control ventilation (ACV); synchronized intermittent mandatory ventilation (SIMV); pressure support; proportional assist ventilation (PAV) and neurally adjusted ventilatory assist (NAVA)). The other is to minimize excessive tidal volumes: volume targeted ventilation and high frequency oscillation (HFO).

Patient triggered ventilation

ACV and SIMV

During ACV, providing the critical trigger level is exceeded, all the infant's inspiratory efforts will trigger an inflation, whereas during SIMV the clinician presets the number of inflations that can be triggered. Physiological studies demonstrated ACV and SIMV were associated with a lower rate of asynchrony, higher tidal volumes and improved blood gases compared to intermittent positive pressure ventilation (IPPV). Our systematic review, however, demonstrated that the only benefits were a shorter duration of ventilation (ACV/SIMV vs. IPPV), ACV vs. SIMV was associated with a trend towards a shorter duration of weaning and SIMV or SIMV plus PS vs. HFO was associated with a greater risk of moderate/severe BPD and a longer duration of ventilation [1].

Pressure support (PS)

During PS, the beginning and end of the infant's spontaneous breath determines the duration of ventilator inflation. In a randomised crossover study, we demonstrated that the work of breathing was lower during 1 h on SIMV with PS than SIMV alone [2]. Those results emphasize the beneficial effect of supporting all of a prematurely born infant's respiratory efforts during mechanical ventilation.

PAV and NAVA

During PAV, the applied pressure is servo controlled throughout each spontaneous breath. The applied pressures increase in proportion to the tidal volume (elastic unloading) or flow (resistive unloading) generated by the patient. The frequency, timing and magnitude of lung inflation are controlled by the patient [3]. Two of our recent studies in infants with evolving or established BPD demonstrated that PAV was associated with a reduction in the oxygenation index compared to assist control

ventilation (ACV) when each were studied in 1 h epochs [4] and similarly when each were studied for 4 h epochs [5]. In addition, the WOB was lower after 1 h of PAV compared to 1 h of ACV [4]. During NAVA, the diaphragmatic electrical (Edi) activity triggers ventilator inflations [6] and the infant's respiratory abnormalities can be "unloaded" by delivering pressure proportional to the Edi signal. The clinician can set the "NAVA level" to increase or decrease the level of respiratory support delivered by the ventilator. There has, however, been a paucity of evidence on NAVA from RCTs. Indeed, our Cochrane review identified only one RCT which failed to highlight any important outcomes [7]. In a randomised crossover study of preterm infants with evolving or established BPD, nine infants were randomised to receive assist control ventilation (ACV) or NAVA for 1 h and then switched to the other mode of ventilation for a subsequent hour. The oxygenation index was significantly lower on NAVA [8]. In a physiological study, in which PAV and NAVA were compared in infants with evolving/established BPD, both ventilation modes improved oxygenation. The A-a gradient, however, was better on NAVA likely reflecting the shorter trigger delay [9].

Volume guarantee ventilation

During VTV there is a feedback loop for automatic adjustment of the peak inflation pressure (PIP) so that a set tidal volume is delivered regardless of variation in lung compliance and airway resistance. Systematic reviews have shown that use of VTV is associated with reduction in the incidences of BPD, periventricular leukomalacia (PVL), intraventricular haemorrhage (IVH) and pneumothorax and duration of mechanical ventilation, but the studies included in the review used a range of volume targeted volumes from 3 to 10 mL/kg. We, therefore, have undertaken a series of physiological studies to determine the most effective volume targeted volume in specific diseases. We assessed the work of breathing (WOB) determined by measurement of the pressure time product of the diaphragm. During weaning and acute RDS 6 mL/kg compared to 4 or 5 mL/kg was associated with the lowest WOB [10]. Amongst term born infants with a variety of diagnoses including meconium aspiration syndrome and pneumonia, but not CDH, the WOB was lowest at 6 mL/kg [11]. In a study of 18 infants with evolving or established BPD the WOB, was assessed at different levels of targeted volume ventilation. Only at a target tidal volume of 7 mL/kg was the WOB reduced significantly below baseline [12], that

result likely reflects the increased physiological dead space in infants with BPD [13]. In CDH infants, both 5 and 6 mL/kg were associated with a significant lower WOB than 4 mL/kg. As CDH infants are likely to have pulmonary hypoplasia, we would recommend use of 5 mL/kg [14].

High frequency oscillation (HFO)

During HFO, small tidal volumes are delivered at frequencies between 10 and 20 Hz. In a Cochrane review, prophylactic HFO (started in the first 12 h after birth) was associated with modest, but significant reduction in BPD, but a meta-analysis of patient-level data did not show any advantage of HFOV over conventional ventilation with respect to short term outcomes including BPD. We undertook a randomised, international multicentre trial (United Kingdom Oscillation Study, UKOS), 798 infants completed the trial. UKOS differed from the other randomised trials included in the systematic review in that the infants were on their randomised mode of ventilation within the first hour after birth. We had hypothesised that the nature of respiratory support in the first few hours might be crucial to lung damage. There were, however, no significant differences in the primary outcomes of UKOS [15]. Subsequently, an observational study reported that at follow-up between six and 12 months of age, infants supported by HFO did not suffer the deterioration in small airway function experienced by those supported by conventional ventilation [16]. This was not a randomised trial, but those who received HFO tended to be the smallest and sickest. As a consequence, we undertook a follow-up study of the UKOS participants to test the hypothesis that HFO might protect small airway function. Comprehensive respiratory and neurodevelopmental assessments of children aged 11–14 years were undertaken. The results demonstrated the children who had been supported by HFO in the neonatal period had superior lung function with no increase in adverse neurodevelopmental outcomes [17]. *In vitro* studies using alveolar analogue cells demonstrated that IL-8 and IL-6 release was lower in a model of cell stretching which mimicked HFO rather than conventional ventilation [18]. It has been previously shown that increased cytokine release can disrupt angiogenesis and alveolarization [19], which could explain the *in vivo* effects we demonstrated. Assessing the UKOS cohort at 16–19 years, however, found no significant differences in lung function or neurodevelopmental outcomes between the two groups [20]. During puberty, there is a lung growth spurt and this may have resulted in “catch up” of the conventional ventilation

group. Whether this effect remains we intend to investigate now the cohort are 20–24 years of age.

Follow-up of the UKOS cohort allowed us to determine the effect of being born small for gestational age (SGA) and male sex on subsequent lung function and to determine the trajectory of lung function in those born very prematurely. At 16–19 years of age, amongst those born at less than 29 weeks of gestational age, those born SGA had poorer lung function [21]. We have demonstrated that males, born very prematurely had worse outcomes in the first 2 years after birth and poorer lung function at 11–14 years of age [22]. Comparison of lung function results at 11–14 years and 16–19 years in the UKOS cohort demonstrated that deterioration in lung function was associated with male sex, white ethnicity, lower gestational age at birth, postnatal corticosteroid administration, BPD and lower birthweight [23]. In addition, exercise capacity assessed using a modified shuttle test was shown to be significantly poorer in those with worse lung function at 16–19 years [24]. We demonstrated that those who had received corticosteroids on the neonatal unit had significantly worse lung function at 11–14 years [25]. Those results were not from a randomised trial, as the infants had received corticosteroids because of the severity of their respiratory disease. The data, however, were analysed using propensity score matching and the mean lung function was lower as the number of courses of dexamethasone increased. Furthermore, between 11 to 14 years and 16–19 years whereas lung function improved in the group not exposed to postnatal corticosteroids, forced expiratory flow at 75% of the expired vital capacity and forced expiratory volume in 1 s deteriorated in the exposed group [26]. Those results suggest that prematurely born young people who received postnatal corticosteroids may be at risk of premature onset of chronic obstructive pulmonary disease.

Respiratory monitoring in the delivery suite

Ten percent of newborns require respiratory support after birth. The International Liaison Committee on Resuscitation has made clear recommendations on the resuscitation of newborns and all neonatal trainees receive standard training in resuscitation (Resuscitation Council UK Newborn Life Support). Our survey of all UK neonatal units, however, highlighted that there was variation in practice between levels of units in with regard to whether an FiO₂ of 0.21 was used initially in term born and

prematurely born infants, if they used a device to detect exhaled CO_2 and whether positive end expiratory pressure (PEEP) was used [27]. We, therefore, used a respiratory function monitor to determine the responses to resuscitation of prematurely born infants in the delivery suite and what practices were undertaken. We first demonstrated that the magnitude of pressure and time of the first five inflations was very variable. Importantly, we demonstrated that expired tidal volumes were significantly greater if the infant inspired during the inflation [28]. There was no significant relationship between the inflation time and the inflation flow time, but there was a significant relationship between the inflation pressure and the inflation flow time. Those results suggest that prolonging inflation times during face mask resuscitation of prematurely born infants would not improve ventilation as prolonged inflation did not lead to longer inflation flow times [29].

Immediately after birth, carbon dioxide elimination only occurs if there is effective ventilation of the lungs and associated vasodilation of the pulmonary vascular bed. We demonstrated that during face mask resuscitation, improved carbon dioxide elimination occurred with the onset of the infant's respiratory efforts [30]. The time for ETCO_2 to be detected following intubation in the delivery suite was found to vary emphasising the importance of using clinical indicators to assess correct endotracheal tube position in addition to ETCO_2 monitoring. We highlighted that capnography detected ETCO_2 levels faster than colorimetric devices [31]. Airway obstruction can occur during face mask resuscitation of preterm infants at birth. We demonstrated similar expired tidal volume levels and similar pressures in those resuscitated via an endotracheal tube or via a facemask. Thus, respiratory function monitoring during initial resuscitation can objectively identify infants who may require escalation of inflation pressures [32]. Initial resuscitation via an endotracheal tube using currently recommended pressures rarely produced adequate tidal volumes, but resuscitation via an endotracheal tube or face mask was most effective when the infant's respiratory efforts coincided with an inflation [33].

We investigated whether neonatal trainees found respiratory function monitoring (RFM) helpful during resuscitation of prematurely born infants, what decisions were made on the basis of the RFM and whether those decisions were helpful. We demonstrated that the decisions they made were not evidence based and further training is required [34].

We demonstrated that the response to resuscitation in infants with CDH differed between those who did and did not survive. Infants who died had lower expired tidal volumes, poorer compliance and ETCO_2 values in the first

minute and the last minute of recorded resuscitation [35]. Using assessments in the delivery suite, we have demonstrated that CDH infants who survived had a larger anatomical deadspace suggesting they had less lung hypoplasia [36].

Abnormal levels of ETCO_2 during resuscitation are associated with IVH development [37]. We determined whether CO_2 levels in the first three days after birth reflected abnormal ETCO_2 levels in the delivery suite and hence, a prolonged rather than an early insult resulted in IVH. We demonstrated that there were no significant differences in NICU CO_2 levels between those who did and did not develop an IVH. Large fluctuations in ETCO_2 during resuscitation in the delivery suite, however, were highly predictive of IVH development in preterm infants [38].

Sustained inflations in the delivery suite at initial stabilisation may reduce the need for intubation and result in a shorter duration of initial ventilation, but had not been compared to routine UK practice. We, therefore, undertook a RCT in infants born at less than 34 weeks of gestational age comparing 15 s inflations to five inflations lasting up to 3 s. The sustained inflation group had a shorter duration of mechanical ventilation in the first 48 h after birth [39]. The SAIL study reported an excess of mortality in their sustained inflation group and hence the study was terminated prematurely [40]. The SAIL study recruited infants born before 23 and 27 weeks of gestation, whereas in our study we recruited infants of less than 34 weeks of gestational age. We did not report an excess of mortality in our trial, but the study was not powered to detect a significant difference.

We have used respiratory function monitoring to assess interventions in the delivery suite. Surfactant delivery via an endotracheal tube resulted in increased ventilation inhomogeneity, likely due to airway obstruction caused by liquid surfactant present in the airways [41]. A prospective cohort study was undertaken which demonstrated less invasive surfactant administration (LISA) was feasible on the delivery suite following simulation training [42]. A case control study of "LISA" infants to historical controls matched for gestational age, birth-weight and gender demonstrated LISA was associated with a reduction in respiratory support and FiO_2 requirement in the first 2 min post LISA. Compared to historical controls LISA administration was associated with a reduction in the need for mechanical ventilation within the first 72 h after birth, the incidence of moderate to severe BPD and the costs of neonatal intensive care [43]. Whether LISA in the delivery suite improves long term outcomes is now being assessed. Furthermore, we are determining if it has benefits in near term or term born infants.

Respiratory viral infections

We demonstrated in a cohort of BPD infants from four centres, that those who had had a hospitalization for respiratory syncytial virus (RSV) infection had greater healthcare utilization and associated costs in the first two years after birth compared to those who had a probable bronchiolitis admission, a respiratory admission or no admission [44]. When the cohort were re-examined at five years of age, those who had an RSV hospitalization in the first two years had greater chronic respiratory morbidity and increased cost of care throughout the preschool years [45]. At 8–10 years of age, those with an RSV hospitalization in the first 2 years had a greater cost of care for outpatient attendances and reduced airway calibre [46].

In a prospective hospital and community follow up study, we demonstrated that RSV infection was associated with increased respiratory morbidity in prematurely born infants, even if a hospital admission was not required [47]. Lung function measurements at one year highlighted that in infants born before 32 weeks of gestation, that those who had an RSV LRTI or a human metapneumovirus (hMPV) LRTI had poorer lung function at follow up, more days of wheeze and bronchodilator requirement [48]. We highlighted that prematurely born infants were functionally and genetically predisposed to RSV LRTIs. Lung function (functional residual capacity, resistance (RRS) and compliance (CRS) of the respiratory system) measured at 36 weeks postmenstrual age showed that those who were admitted to hospital with an RSV or other viral LRTI had significantly higher RRS [49]. Amongst 146 infants born at less than 36 weeks of gestation, a SNP in ADAM 33 was associated with an increased risk of developing RSV LRTIs. SNPs in several genes were associated with increased chronic respiratory morbidity (interleukin 10 (IL-10), nitric oxide synthase 2A (NOS2A), surfactant protein C (SFTPC), matrix metalloproteinase 16 (MMP16) and vitamin D receptor (VDR)) and reduced lung function at one year (MMP16, NOS2A, SFTPC and VDR) in infants who had RSV LRTIs [50]. In a prospective follow up study, prematurely born infants who developed a human rhinovirus (HRV) LRTI had lower CRS before maternity unit discharge. A SNP in the gene coding for the vitamin D receptor was associated with the development of HRV LRTIs and other respiratory viral LRTIs in prematurely born infants [51]. From our prospectively followed cohort, we demonstrated that those who had RV LRTI compared to RSV LRTI had more respiratory related outpatient attendances suggesting they had greater chronic respiratory morbidity [52]. We also highlighted that HRVC rather than HRVA was associated with chronic

respiratory morbidity [53]. Follow up of 51 of the cohort to five to seven years of age demonstrated that those who had had an RV infection in the first 2 years had poorer lung function (lower FEV₁) at follow up and the RV and RSV groups has higher non-respiratory medication costs. Those results further the suggestion that symptomatic viral LRTIs are more likely to occur in a vulnerable group, that is who were requiring non respiratory medications for co-morbidities [54].

Antenatal and neonatal management of infants with congenital diaphragmatic hernia

Congenital diaphragmatic hernia (CDH) affects approximately 2.4–4 per 10,000 live births. It has a high mortality and morbidity related to pulmonary hypoplasia and pulmonary hypertension. Our group has evaluated antenatal diagnostic and intervention techniques and aimed to optimize postnatal management. Estimation of the fetal lung volumes (FLV) can be undertaken by 3D ultrasound with virtual organ computer aided analysis. We demonstrated that FLVs were strongly correlated with the duration of oxygen dependency and lung function in the perinatal period [55]. Our tertiary perinatal unit is a referral centre for the antenatal intervention of fetal endoscopic tracheal occlusion (FETO). During FETO, percutaneous endoscopy is used to insert a latex balloon into the fetal airway. The physiological effects of tracheal occlusion have been thought to lead to pulmonary hyperplasia due to the accumulation of fluid produced and trapped inside the lungs, elevating intrapulmonary pressure and stimulating growth. Although in observational studies FETO was associated with improved survival, we highlighted that “FETO” infants tended to deliver prematurely and if that occurred before 35 weeks of gestation, survival was poor [56]. In an observational study, comparison of CDH infants with and without FETO demonstrated that the FETO infants did not have a higher mortality, but did have a higher morbidity. They required significantly longer durations of ventilation and supplementary oxygen and hospital stay. The lowest OI in the first 24 h after birth was the best predictor of survival [57]. Tracheomegaly has been reported following FETO, but in 70 CDH infants we found that it did not correlate with the duration of respiratory support or hospital stay [58]. Comparing outcomes by CDH side in 193 CDH infants demonstrated no significant differences in mortality between R and L CDH infants who had undergone

FETO. The right sided CDH infants, however, had greater hernia occurrence, pectus deformity, scoliosis and chronic respiratory morbidity [59].

As a member of the CDH Euro Consortium, we contributed to establishing a consensus regarding the postnatal management of CDH patients. This identified a lack of evidence base regarding the optimum ventilation strategy for infants with CDH [60]. As a consequence, the consortium undertook a RCT (the VICI trial) comparing HFOV and conventional mechanical ventilation (CMV). There were no significant differences in the primary outcome BPD or death or the duration of survival, but the CMV group had a lower duration of ventilation and were less likely to require ECMO or pulmonary vasodilation [61]. There was no consensus or evidence as to whether a neuromuscular blocking agent should be used during the initial resuscitation of infants with CDH in the delivery suite. We demonstrated that infants with CDH, particularly those who had undergone FETO had a low lung compliance at birth and this was further reduced by administration of a neuromuscular blocking agent [62]. As a consequence, the consensus statement was revised to include the avoidance of neuromuscular blocking agents at delivery [63]. We have demonstrated the response to resuscitation differed significantly between those who did and did not survive. Comparison of the results of four high volume centres demonstrated that there was significant variability in survival over time and between centres which should be taken into consideration in the planning of future trials [64].

Despite advances in neonatal management, there still remains a high mortality amongst CDH patients. As a consequence, we have undertaken a number of studies to understand associations of the mortality and what is the best predictor of outcome. We highlighted that neither the volume of intravenous fluids administered nor the duration of anaesthesia was associated with post operative death [65]. The OI at 24 h post surgery was the best predictor of an increased risk of mortality [65]. Retrospectively assessing the results of CDH infants over a 10 year period, we demonstrated the highest OI in the first 24 h after birth was a predictor of survival, as was the response to iNO [66]. In view of the high ongoing morbidity of CDH patients it is essential they receive long term multidisciplinary follow up [67].

Lung function abnormalities in sickle cell disease

Life expectancy is only 48 years for women and 42 years for men with HbSS (sickle cell disease, SCD). Sickle chronic

lung disease (SCLD) and acute chest syndrome (ACS) are the leading causes of premature death and ACS episodes are the major cause of SCLD. In young adults, we demonstrated that lung function reduction was correlated with the number of ACS episodes [68]. Lung function declines with increasing age starting in childhood [69]. Children more frequently have obstructive lung function abnormalities, whereas adolescents and adults tend to have restrictive abnormalities. We prospectively followed two cohorts and demonstrated that the rate of decline was greater in the younger cohort who had more ACS episodes [70]. Those results emphasize the need for more effective methods of preventing and treating ACS episodes.

A physician's diagnosis of asthma has been significantly related to an increased rate of ACS episodes, but whether anti-asthma therapy reduces ACS episodes has not been definitely evaluated. We compared two cohorts of children and found those living in Jamaica were more likely to have had ACS episodes and asthma compared to those in the UK. It appeared that the onset of asthma preceded the development of ACS episodes [71]. Subsequently, many studies demonstrated that a physician's diagnosis of asthma was related to increased SCD complications. Wheezing in SCD patients, however, may be due to a number of causes other than asthma. Indeed, wheezing per se is an independent predictor of SCD morbidity. Furthermore, we demonstrated that airway nitric oxide (NO) which is elevated in asthmatics was not elevated in SCD children, but alveolar NO was and is an indicator of a hyperdynamic pulmonary circulation [72]. Patients with SCD can suffer chronic anaemia and as a result have a hyperdynamic circulation. In SCD adults, we demonstrated an association between small vessel pulmonary vascular dimensions on HRCT reflecting pulmonary vascular volume, lung function abnormalities and echocardiographic estimates [73]. We, therefore, postulated a pulmonary circulation airway interaction. To test that hypothesis, we measured pulmonary capillary blood volume and found in SCD children, but not controls, it significantly correlated with oscillometric resistance at 5 Hz [74]. Furthermore, we demonstrated airway resistance increased as did the pulmonary capillary blood volume following blood transfusion [75]. Those results emphasize in a child with SCD and wheezing, a diagnosis of asthma should not be assumed and children must be appropriately investigated before anti-asthma therapy is commenced.

Although obstructive lung abnormalities are commoner in children and in adults restrictive abnormalities are more common, there are a mixture of lung function abnormalities seen in SCD patients. We, therefore, hypothesized that cluster analysis of 11 key respiratory

function and haematological biomarkers would reveal different phenotypes in children and young adults with SCD. Three clusters were detected. Individuals in cluster one had moderate to severe anaemia, an elevated pulmonary capillary blood volume (attributed to an anaemia-related increase in cardiac output), mixed obstructive/restrictive physiology with increased respiratory system resistance and hypoxia. Those in cluster two were older with restrictive lung disease and reductions in diffusion capacity. Cluster three consisted of mainly younger patients with baseline obstruction, bronchodilator reversibility (reflective of airway hyper-responsiveness) and elevated serum lactate dehydrogenase levels, suggestive of increased haemolysis [76]. Pathophysiologically, this clustering technique supports the emerging concept that the mechanism of airway obstruction in SCD may be specifically related to the haemoglobinopathy. The pattern observed in cluster one suggests that increased airway obstruction and resistance reflects extrinsic small airway compression by increased pulmonary vascular engorgement and this may mediate abnormal gas exchange. The airway hyper-reactivity observed in cluster three is suggestive of a haemolysis-related dysregulated inflammation aetiology. Such phenotyping also may inform more personalised management strategies to improve outcomes.

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