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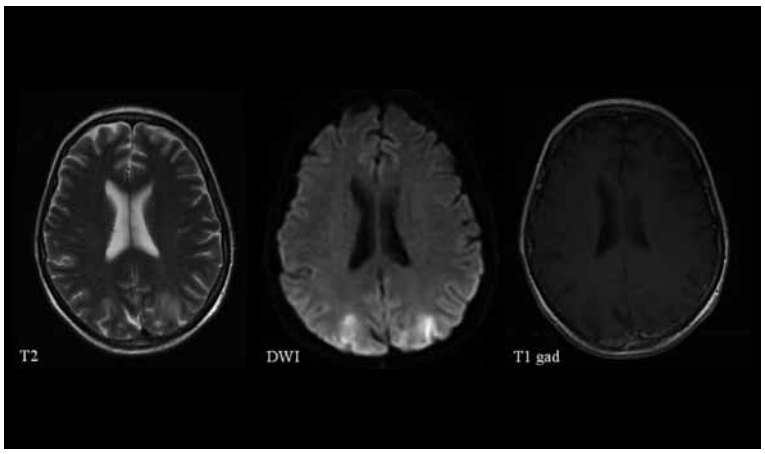

Blindness, confusion and seizures in a cancer patient – what is your diagnosis?

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EDITORIAL

The Netherlands Journal of Medicine – one year in Amsterdam

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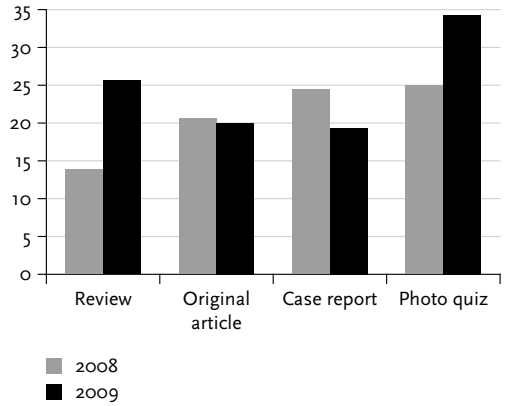
The Amsterdam editorial team of the *Netherlands Journal of Medicine* has now been in office for one year and that provides an opportunity to look back and see how the Journal has evolved.[1] In 2009, the Journal saw a marked increase in submissions. Interestingly, these papers are not only from the Netherlands, but a substantial number come from other parts of the world (*table 1*). The steadily rising impact factor of the Journal and its increasing position on the list of Journals in the field of general medicine may be an important factor here. The journal impact factors for 2009 have not yet been calculated; however, our first predictions show that it is likely that the increase over the last few years will be sustained.

An increasing number of submissions and a fixed space for publication will lead to lower acceptance rates. The acceptance rate of the Journal in 2009 for the various article types, the origins of the submissions and the subdisciplines from which the papers were submitted are shown in *table 1*. The overall acceptance rate has now fallen to 30%, and for specific paper categories it is lower than 20%. In fact, our policy is to only accept case reports (as with many journals the type of paper that is submitted most) if they substantially increase our insight into the pathogenesis or background of a disease or if they report a really original clinical finding. Photo quizzes remain a very popular item of the Journal (also reflected by a high number of 'hits' on our website) and we have decided to publish somewhat more of these often very interesting and illustrative cases. As shown in *figure 1*, we have also published many more review manuscripts than in previous years. We have indeed adopted a policy of actively soliciting review manuscripts from well-known authors on a given subject, both from the Netherlands and abroad. Our group of associate editors has been very helpful in retrieving these manuscripts and we expect to continue this strategy in the coming years.

Table 1. *Number of submissions to the Netherlands Journal of Medicine in 2009 and acceptance rate (= published papers divided by submitted papers)*

	Submitted	Acceptance rate
Total	328	30%
Article type		
• Review	35	74%
• Original article	107	19%
• Case report	136	14%
• Photo quiz	50	68%
Origin		
• Netherlands	61%	39%
• Other European countries	16%	23%
• North America	7%	30%
• Rest of the world	16%	4%
Subdiscipline		
• Cardiovascular	75	36%
• Respiratory	14	14%
• Gastroenterology	38	34%
• Intensive care	44	52%
• Haematology/Oncology	56	20%
• Rheumatology/ Immunology	21	29%
• Nephrology	23	22%
• Endocrinology	33	24%
• Infectious disease	21	24%
• Other	3	0%

Figure 1. *Article types published in the Netherlands Journal of Medicine in 2008 and 2009*



Acceptance or rejection of a manuscript is a result of intensive peer review, and we thank the many reviewers of the *Netherlands Journal of Medicine*, who have helped us tremendously in the last year. Also, our highly active group of junior associate editors, composed of residents in training for Internal Medicine who have themselves been very active in research over the last few years, is invaluable for guiding the review process, in particular for case reports and photo quizzes.

Apart from citations in other journals (which underlie the impact factor of a journal), downloading of our articles from the Journal's website may be another measure of the 'impact' of a journal.[2] In *table 2* we report the ten most downloaded papers in the *Netherlands Journal of Medicine* in 2009. It is not known whether the number of downloads correlates with the number of citations, but we intend to analyse this for our Journal and we will report about this in one of the coming issues.

On behalf of the entire editorial team in Amsterdam, I can say without any reservation that editing the *Netherlands Journal of Medicine* is a great pleasure and we hope and expect that 2010 will be another good year for the Journal with many interesting publications.

Table 2. *Most downloaded articles in the Netherlands Journal of Medicine in 2009*

1.	Kuipers MT, *et al.* Hypomagnesaemia due to proton pump inhibitors – a review.[3]
2.	Rahnama'i MS, *et al.* Amoxycillin/clavulanate-resistant *E. coli* in bacterial peritonitis after abdominal surgery.[4]
3.	Bhat SA, *et al.* Novel antibodies in the treatment of non-Hodgkin's lymphoma.[5]
4.	Hoeks MP, *et al.* Adult issues in phenylketonuria.[6]
5.	Jaspers H, *et al.* Bilateral swollen eyelids occurring during adjuvant treatment with tamoxifen.[7]
6.	Koopmans PP, *et al.* Should antiretroviral therapy for HIV infection be tailored for intracerebral penetration?[8]
7.	Schrauwen RW, *et al.* Seven days PPI-triple therapy with levofloxacin is very effective for *H. pylori* eradication.[9]
8.	van Meerten T, *et al.* CD20-targeted therapy: a breakthrough in the treatment of non-Hodgkin's lymphoma.[10]
9.	Khan FY, *et al.* Rhabdomyolysis: a review of the literature.[11]
10.	Velema MS, *et al.* DRESS syndrome caused by nitrofurantoin.[12]

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REVIEW

Clinical implications of heart-lung interactions during mechanical ventilation: an update

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ABSTRACT

As opposed to spontaneous respiration wherein small cyclic changes in transpulmonary, negative pressure coincide with lung volume changes, positive pressure (mechanical) ventilation results in a simultaneous rise in transpulmonary pressure and lung volumes. The changes may affect biventricular cardiac loading and function in dissimilar ways, depending on baseline cardiopulmonary function. This review is intended to update current knowledge on the pathophysiology of these heart-lung interactions in helping to explain the common circulatory alterations occurring during airway pressure changes and to better understand mechanisms of disease and modes of action of treatments, during spontaneous and mechanical ventilation.

KEYWORDS

Cardiopulmonary interactions, cardiac function, mechanical ventilation, pulsus paradoxus.

INTRODUCTION

Spontaneous respiration as well as mechanical ventilatory support can alter cardiac loading and function and thereby contribute to circulatory alterations. This may particularly occur in some disease states. If severe, as in critically ill patients in the intensive care unit, the changes may be associated with diminished tissue oxygen delivery and distant organ dysfunctions, and thereby contribute to morbidity and mortality. This update is meant to explain these frequently encountered alterations to provide a rationale for treatment. We will summarise current knowledge on the effects of airway pressures on the right and left ventricle separately (*table 1*) and will focus on animal and human studies with major mechanistic or therapeutic implications. We will not discuss the effect of airway pressure on the peripheral circulation and for more in-depth physiological reviews the reader is referred to recent publications.[1-4]

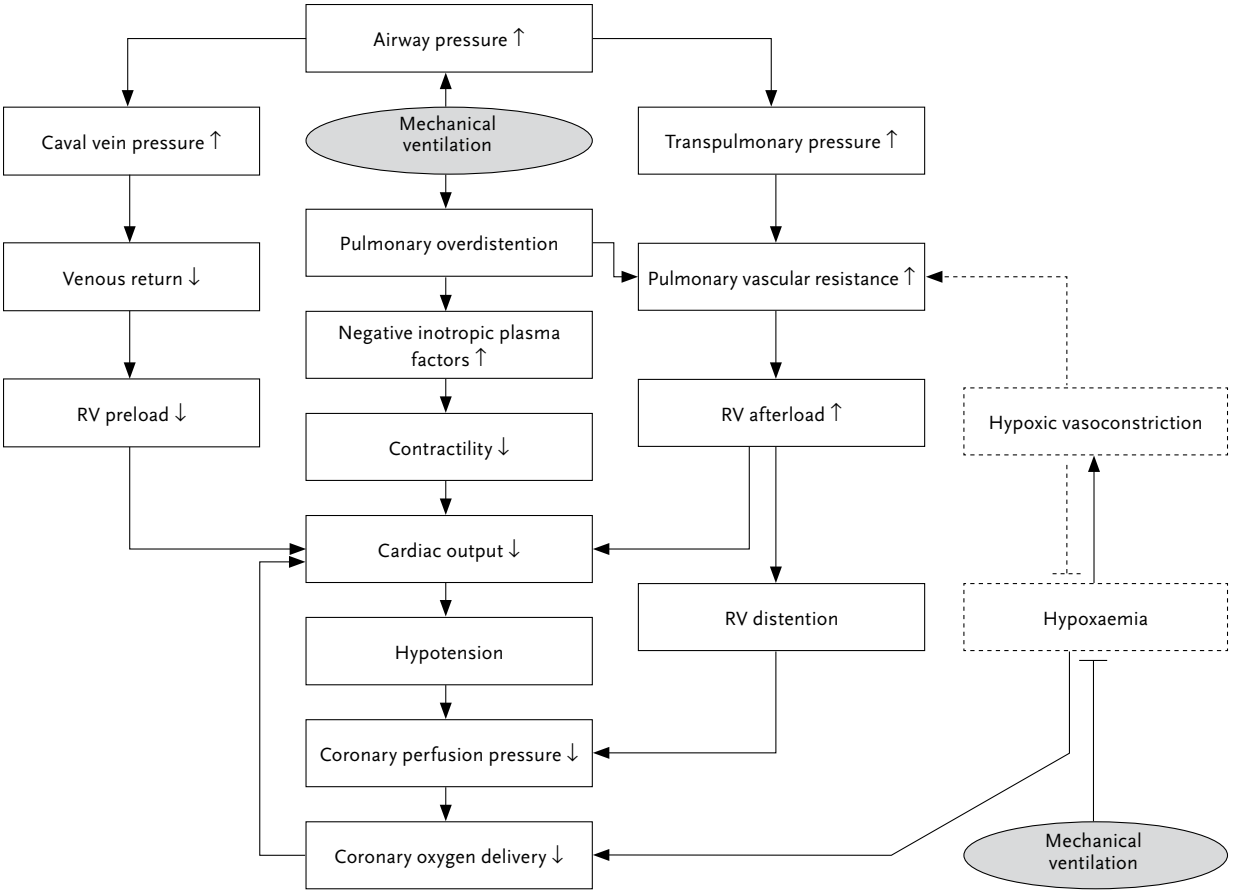
Table 1. *Effects of increase in airway pressure and volume*

Right ventricle	Left ventricle
Decreased preload	Decreased preload
Increased afterload	Decreased compliance
Reduced contractility	Variable effects on (autonomous nervous system control of) contractility
Compression of heart in cardiac fossa	Decreased afterload
	Compression of heart in cardiac fossa

THE RIGHT VENTRICLE

After instituting mechanical ventilation for acute respiratory failure, arterial blood pressure is often observed to fall. This can, most commonly and importantly, be attributed, at least in part, to a fall in right ventricular filling following impeded venous return, depending on the transmission of airway to juxtacardiac pressure and on systemic filling (abdominal) pressure (*figure 1*).[5,6] The difference between transpulmonary and systemic filling pressures is indeed the gradient for venous return. The transmission can be assessed by measuring pleural or pericardial (rather than oesophageal) pressure and is normally about 50% of prevailing airway pressure.[7] Therefore, right ventricular end-diastolic volume generally falls upon increases in airway pressure as a reflection of a fall in venous return and a resultant fall in right ventricular preload, which largely explains the fall in stroke volume and cardiac output that is dependent on (transmission of) mean airway pressure during the respiratory cycle.[5-9] Also contributing to a fall in cardiac

Figure 1. *Cardiovascular effects of mechanical ventilation and application of PEEP. Mechanical ventilation alters intrathoracic pressures and thereby affects the cardiovascular system, mainly the right ventricle*



preload is compression of the heart by the lungs in the cardiac fossa,[7] so that haemodynamic alterations in response to airway pressure may be greater in closed than in open chest conditions. The latter may be relevant for conditions after sternotomy during cardiac surgery.[10] Airway pressure transmission is expected to decrease with decreasing lung (or increasing chest wall) compliance. If, however, lung (tidal) volume is kept constant in diseased lungs, the atmospheric-pressure referenced pleural, pericardial and thus right atrial pressures will rise, since airway pressure is increased in face of diminished compliance. Then, the effect of altered airway pressure transmission on net right ventricular preload may be negligible. Conversely, open abdomens may be associated with increased chest wall compliance.

In addition to a pressure-related fall in right ventricular preload, a volume-induced rise in pulmonary vascular resistance may, particularly with inspiration, result in a rise in right ventricular afterload. The latter phenomenon may in turn attenuate a fall in right ventricular end-diastolic volume, for instance during inspiration as compared with expiration, but may aggravate a fall in cardiac output. The preload effect may prevail with rises in mean airway pressure upon altering ventilatory settings, while the rise in inspiratory afterload may further modulate this effect. However, if an increase in right ventricular afterload predominates over the fall in venous return, right ventricular end-diastolic volume may increase with rising airway pressures, and indeed features of acute cor pulmonale may develop in up to 25% of patients with acute lung injury on mechanical ventilation, as supported by echocardiography.[11] This may render the right ventricle susceptible to an imbalance in coronary blood supply to demand and may further depress function, particularly in case of arterial hypotension and coronary hypoperfusion (*figure 1*). Occurrence of tricuspid regurgitation may further confound haemodynamics. The acute cor pulmonale is potentially ameliorated or prevented by lung-protective and low (*vs* high) tidal volumes during mechanical ventilation.[11] Also, too much positive end-expiratory pressure (PEEP) and presumably resultant overdistention of the lung may liberate plasma factors that may have negative inotropic actions on the heart, possibly by spillover of the inflammatory response to overdistention, at least in experimental animals.[12]

The response of the right ventricle to increasing airway pressure and PEEP may depend on underlying conditions and the effect of lung volume on pulmonary vascular resistance. Pulmonary hypertension is a common complication of chronic lung disease. Exacerbations and resultant hypoxaemia and hypoxic vasoconstriction may further elevate pulmonary artery pressure and thus afterload upon the right ventricle. A component of acute cor pulmonale can then be superimposed upon chronic cor pulmonale during mechanical ventilation.[11] Underlying intrinsic (right ventricular) disease, for instance in coronary artery (surgery) patients, may also affect cardiac loading and function alterations caused by elevated airway pressures, increasing the risk for right ventricular ischaemia.[8] One may also speculate that overdistention by mechanical ventilation will increase vascular resistance and that recruitment of previously collapsed alveoli ('opening the lung') may decrease resistance following amelioration of hypoxic vasoconstriction and improved CO_2 removal.[11,13,14] Hence, baseline pulmonary pressure/volume relations and effects of altering airway pressures hereon may determine if a right ventricular preload or afterload effect predominates during increases in airway pressure and PEEP. Conversely, the effect of tidal inflation and plateau pressure may depend on baseline PEEP, since PEEP increases plateau airway pressure during pressure-controlled ventilation and may thereby accentuate inspiratory effects. Respiratory compliance-dependent airway pressure transmission may further confound heart-lung interactions. On the other hand, hypovolaemia may aggravate the negative effect of PEEP on cardiac output.[15] Together, the factors may at least explain some of the varying results in the literature on steady state and cyclic changes in right ventricular dimensions during alterations in airway pressures.

THE LEFT VENTRICLE

In patients with severe left ventricular dysfunction that is sensitive to afterload changes, application of positive (end-expiratory) airway pressure, as in a Valsalva manoeuvre, may decrease transmural aortic pressure and thereby afterload. Hence, under these circumstances, PEEP may increase rather than decrease cardiac output, in spite of a reduction in right ventricular preload by the rise in airway pressure.

Increased vagal nerve afferent and efferent activity via lung stretch and stimulation of stretch receptors (by lung volume) may depress biventricular function and lower peripheral vascular resistance, during increments of airway pressure (PEEP), independently of altered loading.[16] Finally, right and left heart function are tied together in ventricular interdependence via a series effect, pericardial constraint, systolic augmentation, diastolic septal interaction, or combinations.[17] Indeed, right ventricular distention may flatten the septum and decrease left ventricular compliance, thereby contributing to a reduction in left ventricular filling and output.[9]

CLINICAL IMPLICATIONS

Imaging techniques to assess biventricular loading consist of echocardiography, magnetic resonance imaging, radionuclide imaging, and determinations of ventricular volumes by thermodilution, but some of these techniques may not be routinely applicable at the bedside. The imaging techniques can thus be helpful to document cardiac loading alterations, for instance during changes in ventilation modes or settings. Indeed, atmospheric pressure-referenced, end-expiratory right atrial and pulmonary capillary wedge pressures, when measured, do not indicate transmural filling pressure of the right and left ventricle, respectively, in the presence of PEEP. The effect of transmitted airway pressure on these measurements is unpredictable and hard to account for, although bedside methods for adjustment have been described.[18] Indeed, when subtracting transmitted airway pressure from atmospheric pressure-referenced pressures in the thorax, transmural cardiac filling pressures can be calculated that may fall upon increases in airway pressure, i.e. PEEP, when pressures are measured at the end of expiration.[18] Finally, because of cyclic variations within the respiratory cycle, bolus thermodilution measurements of cardiac output are determined by the phase in which the measurements are done.[19,20] The practical implication is that right-sided bolus thermodilution cardiac output measurements should be done, particularly in mechanically ventilated patients, at three to four equally spaced time intervals or from four to five random injections throughout the respiratory cycle to obtain a true mean value over that cycle.[19,20]

We will now discuss clinical implications of heart-lung interactions on specific disease manifestations and treatment, during spontaneous and mechanical ventilation.

BRONCHO-OBSTRUCTIVE SYNDROMES AND PERICARDIAL DISEASE

During spontaneous respiration, cyclic changes in pleural pressure occur, which may alter cardiac loading and function and result in the classical pulsus paradoxus, with increases in right ventricular filling upon inspiration (as compared with expiration) while left ventricular filling decreases concomitantly. Pulsus paradoxus refers to an inspiratory fall of 10 mmHg or more of systolic arterial

blood pressure resulting from a fall in left ventricular filling and stroke volume. The inspiratory decrease in left ventricular filling can be attributed, at least in part, to a rise in transmural pressure elevating left ventricular afterload and a fall in compliance by right ventricular distention. These alterations are augmented by positive airway pressures when expiration is impaired, for instance during broncho-obstructive syndromes, and confounded by pre-existent alterations in the pulmonary vasculature that may result in pulmonary hypertension and right ventricular overload. Indeed, the pulsus paradoxus is the clinically most common example of heart lung interactions,[21] which can occur in patients with (severely exacerbated) broncho-obstructive pulmonary disease or with pericardial tamponade. In the latter, the sign may necessitate further diagnostics and treatment by pericardiocenthesis.[21]

The impaired expiration following broncho-obstruction in the course of asthma or chronic obstructive pulmonary disease may result in intrinsic positive end-expiratory pressure (PEEPi) with hyperinflation that may hamper venous return and thereby contribute to arterial hypotension and tachycardia in these patients.[22] This may be only partially counteracted by large negative swings in pleural pressure during inspiratory attempts increasing venous return. Treatment of underlying disease and broncho-obstruction to diminish right ventricular overloading remains of crucial importance and can be done by noninvasive ventilation with continuous positive airway pressure (CPAP or bilevel positive airway pressure) counteracting PEEPi, for instance.[22,27]

SLEEP APNOEA

Noninvasive ventilation is also commonly used in the treatment of sleep apnoea syndromes. Here, nocturnal delivery of CPAP by face mask augments lung volumes and improves gas exchange to combat or prevent hypoxaemia, thereby unloading the heart and increasing cardiac output and tissue oxygenation. Even on the long term, benefits of cardiovascular status have been described that may relate to less sympathetic overstimulation, among others.[23,24]

CARDIOGENIC PULMONARY OEDEMA

The circulatory effects of noninvasive ventilation (bilevel positive airway pressure and CPAP) in this condition or its nocturnal prevention are generally considered beneficial by unloading the heart, particularly when systolic function is compromised.[24-27] Unloading of respiratory muscle may decrease systemic oxygen requirements and unloading of the heart may reduce myocardial oxygen requirements and thus favourably effect coronary oxygen supply to demand ratios. By these cardiopulmonary mechanisms, CPAP decreases morbidity and perhaps mortality.[25,26] CPAP may also increase cardiac function and output, at high pulmonary capillary wedge pressures and alleviation of myocardial ischaemia. Hence, this noninvasive treatment is considered safe and can particularly be used in congestive heart failure-induced respiration disorders.[24]

MECHANICAL VENTILATION

Acute respiratory failure may necessitate intubation and mechanical ventilation. The changes in airway pressure during positive pressure ventilation are opposite to those in spontaneous ventilation and, in contrast to the latter, synchronous with lung volume changes. The reversed pulsus paradoxus refers to the inspiratory increase in left ventricular output when right ventricular output falls, and vice versa during expiration.[9] Obviously, pulmonary blood volume changes contribute to different behaviours of the right and left ventricle through the respiratory cycle. Moreover, left ventricular stroke volume variations (SVV) and thereby pressure variations, induced by mechanical ventilation and measured by noninvasive techniques, increase during a fall in biventricular preload and cardiac output,[28] thereby predicting a rise in cardiac output during fluid challenges; this is referred to as fluid responsiveness.[29] Hence, these variations may guide fluid treatment of critically ill and mechanically ventilated patients, in the absence of spontaneous respiratory efforts, constant ventilatory rates and volumes, and regular heart rates.[29] Finally, weaning from mechanical ventilation can be associated with recurrent pulmonary oedema, particularly in patients with poor left ventricular function.

Acute cor pulmonale evidenced by echocardiography can be treated by adjustment of ventilatory settings and amelioration of arterial hypotension by, for example, the use of vasopressors. Obviously, the modes of ventilatory support may modulate some of the effects described, but further discussion is beyond the scope of this review. It may suffice here to state that ventilatory support modes that alter pressure volume relations in the lungs may favourably affect haemodynamics when lowering mean airway pressure and preventing overdistention. Conversely, recruitment manoeuvres by applying temporary high airway pressures may have negative circulatory effects.[10,13]

Finally, a fall in cardiac output may partly offset a rise in arterial O_2 saturation by PEEP so that O_2 delivery to

the tissues may not fall even when cardiac output does. Therefore, authors have defined best PEEP as the level with highest O_2 delivery to the tissues.

FLUID THERAPY

Understanding heart-lung interactions is important since fluid loading to compensate for the fall in venous return with mechanical ventilation may only help increasing cardiac output when the right heart is fluid responsive by operating in the steep part of its function curve.[5] Right ventricular distention is generally believed to limit or preclude a rise in cardiac output upon fluid loading, particularly when right ventricular ischaemia ensues and the ventricle operates in the plateau of its function curve (*figure 1*). Fluid loading is thus usually counterproductive in right ventricular overloading and may worsen right ventricular failure. In contrast, fluid administration is the treatment of choice to enhance venous return and tissue oxygenation, if deemed necessary on clinical grounds, when cardiac dimensions have decreased, but this is often at the expense of a supranormal plasma volume and resultant tissue oedema.[5] Imaging the heart may thus be helpful in individual treatment decisions.

During broncho-obstructive syndromes, fluid loading may also be counterproductive in the case of concomitant pulmonary hypertension and cor pulmonale, when right ventricular overload precludes an increase in cardiac output with an attempted increase in preload. In contrast, exacerbations of chronic obstructive pulmonary disease may be accompanied by diastolic dysfunction or even overt left ventricular failure and administration of diuretics is often attempted to treat any concomitant pulmonary overhydration. This may in turn aggravate a fall in cardiac output and arterial hypotension. Hence, assessing volaemic status and heart function to guide treatment is important but hard at the bedside of patients with exacerbated chronic obstructive pulmonary disease and hypotension, and may therefore necessitate echocardiography.

CONCLUSIONS

The effect of airway pressures and volumes on cardiac loading and function is complex. The predominance of effects on cardiac output depends on the underlying function of the heart and pulmonary vasculature. Although the preload effect of increasing airway pressures often predominates in patients, a detrimental rise in right ventricular afterload is unpredictable and should be evaluated in case of severe haemodynamic compromise and absence of fluid responsiveness. This can be done by echocardiography and may have therapeutic implications. Heart-lung interactions may play a role in the manifestations and treatment of a variety of disorders.

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REVIEW

ANCA-associated vasculitides: advances in pathophysiology and treatment

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ABSTRACT

Substantial progress has been made over the last two decades in our understanding of the immunopathogenesis of antineutrophil cytoplasmic antibodies (ANCA) associated vasculitides. Compelling evidence from *in vitro* studies and experimental models in conjunction with clinical trials has confirmed that ANCA directly contribute to the evolution and progression of the disease process. Continuous development in our understanding of the mechanisms that drive the disease may ultimately allow us to tailor the multitude of novel therapies, which are rapidly becoming available, to the requirements of individual patients.
In this review we endeavour to provide a brief overview of the recent advances in ANCA-associated vasculitides and outline basic principles for diagnosis and treatment of these complex multisystem diseases.

KEYWORDS

ANCA, vasculitis, autoimmune disease, Wegeners granulomatosis, microscopic polyangiitis, Churg Strauss vasculitis

INTRODUCTION

Antineutrophil cytoplasmic antibodies (ANCA) associated vasculitides principally comprises three overlapping but distinct disease entities: Wegener's granulomatosis (WG), microscopic polyangiitis (MPA) and Churg-Strauss syndrome (CSS). ANCA-associated systemic vasculitis (AAV) has an incidence of 20 per million and a prevalence of 144 per million in the UK.[1] In vasculitis, the blood vessels are the primary site of autoimmune inflammation, and the pathological consequence is destruction of the vessel wall, seen histologically as fibrinoid necrosis. The disease may be limited to a single organ or vascular bed, but commonly affects multiple organ systems. AAV has substantial morbidity and mortality, often presenting with aggressive renal failure or pulmonary haemorrhage, and requiring therapy with toxic immunosuppression.[2] Left untreated, AAV has a universally poor prognosis with mortality approaching 100% within five years.[3] The introduction of treatment regimens based on cyclophosphamide and glucocorticoids has transformed AAV from a rapidly fatal disease, to one of chronic morbidity and reduced survival often preceded by end-stage renal disease.

ANCA were originally discovered by chance in the early 1980s by Davies *et al.* in serum samples from patients with necrotising glomerulonephritis.[4] They became serological hallmarks of the disease after Van der Woude showed that they mainly occur in WG.[5] They are directed against constituents of granules of neutrophils and lysosomes of monocytes. Indirect immunofluorescence detects two distinct patterns perinuclear (pANCA) and cytoplasmic (cANCA). Myeloperoxidase (MPO) is the primary antigenic target of pANCAs, and proteinase 3 (PR3) the major autoantigen associated with cANCAs, as determined by antigen-specific ELISA. Positive immunofluorescence in conjunction with a positive ELISA is highly specific for AAV, i.e. >95% of double-positive patients with suspected nephritis reveal vasculitis on renal biopsy. PR3-ANCAs are mainly detected in WG, whereas MPO-ANCAs are predominantly found in MPA and CSS. The availability of ANCA testing has profoundly reduced the diagnostic delay in these conditions, but at the same time increased the median age at diagnosis and the incidence of vasculitis. This suggests that there was a considerable underdiagnosis of vasculitis, particularly in the elderly, in the past.

The pathogenesis of AAV has not been fully elucidated, but a number of genetic and environmental factors have been implicated. There are striking geographic differences

in the incidence of the different AAVs (i.e. WG, the most common form of AAV in northern and central Europe, is virtually nonexistent in Japan and rare in China).[6] The most strongly linked environmental factors are silica exposure, infections particularly with *Staphylococcus aureus* and drugs such as propylthiouracil.[6]

DIAGNOSIS

Early diagnosis of AAV is crucial to prevent irreversible organ damage and to allow initiation of appropriate therapy. Heterogeneity in severity and organ distribution are the biggest stumbling blocks to making a speedy diagnosis. AAV should be on the list of differential diagnoses in any patient presenting with an inflammatory condition.

Patients will typically have fluctuating constitutional symptoms, which may vary in intensity and include malaise, tiredness, mild fevers and nonspecific aches and pains. The prodromal phase of AAV usually lasts months and may often be misinterpreted as viral infection, postviral syndrome or malignancy.

The kidney is the most commonly affected organ in AAV. Renal involvement is initially asymptomatic until renal failure occurs. The first sign of rapidly progressive glomerulonephritis is frequently the detection of blood and protein on urinary dipstick testing, which are always present in renal vasculitis. The presence of red cell casts in the urine sediment, combined with a rapidly rising serum creatinine, is strongly significant for a crescentic glomerulonephritis, as seen in AAV.[2,7]

Patients with AAV frequently present with symptoms suggestive of upper respiratory tract infections including sinusitis, deafness or hoarseness. More aggressive nasal inflammation, nasal septal defects, necrosis and collapse of the bridge, and severe nasal crusting or bleeding are more commonly associated with WG.[8] Lower respiratory tract involvement includes cough, exertional dyspnoea and haemoptysis. Chest X-rays may reveal the characteristic cavitating pulmonary lesions or raise suspicion of pulmonary haemorrhage. Maturity onset asthma in conjunction with a raised eosinophil count is a classic feature of CSS.

Skin lesions are common and can range from a painless purpuric rash to necrotic ulceration.

The involvement or isolated disease of the orbit is a feature of AAV, and usually presents with episcleritis and scleritis; this can lead to loss of vision, especially in cases of retinal vasculitis, retinal venous infarction or retro-orbital granulomas.

Gastrointestinal involvement in AAV is rare, but is more commonly seen in other vasculitic conditions such Henoch-Schoenlein purpura and polyarteriitis nodosa.

Mononeuritis multiplex of peripheral nerves with an axonal pattern is common in AAV, particularly CSS,[9] but direct involvement of the brain is rarely seen at presentation.

Cardiac disease is not common in AAV, but valvular infarction can occur and eosinophilic cardiomyopathy occurs in a significant number of patients with severe CSS.

The combination of prodromal symptoms, with one or many characteristic organ manifestations, in conjunction with a positive ANCA titre, leads to a probable diagnosis of AAV. Obtaining a confirmatory biopsy is highly recommended prior to committing patients to prolonged immunosuppression and will help to optimise treatment decisions. ANCA levels may be negative in early or limited disease, and occasional patients with biopsy-confirmed disease remain persistently ANCA negative.

TREATMENT

Early diagnosis must be associated with rapid therapy induction to prevent irreversible organ damage, such as renal scarring, visual impairment and peripheral nerve weakness. The therapeutic goal is to control disease activity and prevent relapse, while minimising risks of acute immunosuppression and late complications, such as cardiovascular disease, osteoporosis and malignancy.

The initial treatment phase is aimed at achieving remission and standard induction therapy with cyclophosphamide and glucocorticoids can induce remission in up to 90% of patients.[10] Cyclophosphamide can be administered as continuous or intravenous/oral therapy, with equal rates of remission induction. However, pulsed strategies have a lower cumulative exposure and fewer cases of leucopenia, but a higher relapse rate (*tables 1* and *2*).[11] In those with more severe disease, plasma exchange improves renal survival,[12] and is widely used in patients with pulmonary haemorrhage, but without clear evidence to support this particular indication. Methotrexate (MTX) can be substituted for cyclophosphamide for remission induction in less severe early systemic disease, with fewer side effects but a higher relapse rate.[13] One small randomised trial of renal vasculitis shows similar remission induction rates for mycophenolate mofetil (MMF) and cyclophosphamide,[14] and the efficacy of MMF is currently being studied in a large multicentre trial (MYCYC). Reducing treatment toxicity is a key emphasis of ongoing trials through substitution of cyclophosphamide with potentially less toxic agents, such as MMF, or by reducing cumulative steroid exposure. A recent study assessing outcome and adverse events of 524 newly diagnosed patients with AAV prospectively recruited to four European trials shows that overall one-year mortality was 11.1%. Importantly, 59% of those deaths were secondary to adverse events and only 14% to disease activity.[15] This clearly shows that in the first year the greatest risk to patients is therapy-associated adverse events.

Table 1. *European Vasculitis Study Group (EUVAS) Trials*

Trial	Number of Patients	Description	Outcome
CYCAZEREM	WG 95 MPA 60	Cyclophosphamide *vs* AZT for maintenance therapy	Replacement of cyclophosphamide with AZT does not increase relapse rate
NORAM	WG 94 MPA 6	MTX *vs* cyclophosphamide for induction of remission	MTX achieved comparable rates of remission but higher relapse rates
CYCLOPS	WG 61 MPA 99	Pulsed *vs* continuous oral cyclophosphamide for remission induction	Equal remission induction, higher relapse rates in pulsed regimens
MEPEX	WG 42 MPA 95	Plasma exchange *vs* high-dose methylprednisolone as adjunctive therapy in patients with severe renal impairment	Increased rate of renal recovery in plasma exchange group
RITUXVAS	Total 44	Rituximab *vs* cyclophosphamide for disease induction in renal AAV	Completed recruitment but results not yet published
MYCYC	72 recruited to date	MMF *vs* cyclophosphamide for induction of remission	Recruiting – target 140 patients
PEXIVAS	Target 500	Double cross over to examine steroid dose and plasma exchange for induction of remission	Protocol agreed, ethical approval, not yet recruiting

WG = Wegener's granulomatosis; MPA = microscopic polyangiitis; AZT = azathioprine; MTX = methotrexate; AAV = ANCA-associated vasculitides; MMF = mycophenolate mofetil.

Table 2. *Drugs commonly used to treat AAV*

Drug	Dose	Mechanism of action
Cyclophosphamide	Oral 2 mg/kg/day Pulse 15 mg/kg every 2 to 3 weeks Patients over 65 years of age not more than 100 mg/day (oral)	Alkylating agent – breakdown products covalently bind DNA leading to mutations and apoptosis
Azathioprine	Oral 2 mg/kg/day	Purine antimetabolite interferes with de novo purine synthesis and impairs cell proliferation
Methotrexate	Starting dose 10 mg/kg/once weekly Dose can be titrated up to maximum 25 mg/kg	Inhibition of dihydrofolate reductase thereby disrupting DNA synthesis and cell division
Prednisolone	Standard dose for remission induction 1 mg/kg/day Dose reduction during induction to ~12,5 mg/day at 3 months	Inhibits cytokine, chemokine synthesis and release. Reduces localisation of inflammatory cells to inflamed areas
Mycophenolate	Standard dose 1 or 1.5 g twice daily. Gradually increase dose to reduce gastrointestinal side effects	Inhibits de novo guanine synthesis. Lymphocytes lack salvage pathway resulting in impaired proliferation, antibody production and adhesion

The value of B-cell depletion by anti-CD20 antibodies has been tested in two randomised prospective controlled trials (RITUXVAS and RAVE, the latter was also double blind) *(table 1)*. Importantly, anti-CD20 was used with reduced doses of cyclophosphamide: only two pulses were given in RITUXVAS and none in RAVE. Although these studies have been completed, they are not yet published; however, preliminary reports suggest efficacy that is at least equivalent to cyclophosphamide. An advantage of rituximab is its lesser immunosuppressive properties.

Following remission, usually after three months, cyclophosphamide is replaced by azathioprine (AZT) or MTX and continued for at least 18 months to reduce relapse risk (*table 1*).[16] Relapse is more common in WG than in MPA, and this has been associated with nasal carriage of *Staphylococcus aureus*.[17] Patients who remain ANCA positive after induction therapy have a fourfold higher risk of relapse than those who become ANCA negative.[18] Generally speaking, patients who receive more potent induction therapy tend to have a lower rate of relapse when treatment is reduced or withdrawn.[11,13] AZT and MTX are associated with intolerance in a small number of cases, and fail to maintain remission in approximately 30% of patients. A significant proportion of patients relapsing on AZT can be rescued by switching to MTX, but MMF is increasingly used as a second-line agent in patients who relapse during maintenance therapy.

Careful monitoring of patients in remission is important as it contributes to early relapse detection and therapy adjustment, limits relapse severity and prevents further irreversible organ damage. Another important consideration of long-term follow-up is to carefully monitor disease and therapy-associated organ damage. Patients with AAV have a twofold increased risk of developing malignancies in comparison with the general population. The risk increase varies according to the affected organ and is highest for bladder cancer, nonmelanoma skin cancer and lymphoma.[19] Measures to reduce the risk of cardiovascular disease should be integral to the management of systemic vasculitis. A

recent study by Morgan *et al*. shows that patients with AAV have a greater risk of cardiovascular disease, in particular patients with pre-diagnosis cardiovascular disease and those with markedly impaired renal function.[20] Osteoporosis risk is markedly increased, especially in those patients who are exposed to a high cumulative steroid dose and prophylaxis is recommended to reduce fracture risk.[21]

A significant number of patients develop refractory disease.[22] Anti-T-cell therapies using antithymocyte globulin or anti-CD52 antibodies have been tested in small studies with some efficacy.[23,24] However, high relapse rates have occurred after withdrawal of therapy, as well as high rates of infectious complications. 15-Deoxyspergualine, a novel immunosuppressant whose mode of action remains unclear but is suggested to include suppression of NF-κB, has shown efficacy with respect to induction of remission in open-label studies of refractory patients with AAV but, as with anti-T-cell therapies, relapses occur rapidly after discontinuation of the drug.[25,26] Rituximab has been used for refractory disease and in a cross-sectional study of 64 patients, full remission was achieved in 75% and partial remission in 23%. Relapses occurred in 58% of patients who had achieved a full remission; 38 of these patients were retreated, with 84% achieving a further full remission.[27] Finally, the initial promise of anti-TNF therapies was not fulfilled in a randomised, controlled trial of etanercept compared with placebo, added to standard therapy for induction and maintenance of remission in WG.[28] Many patients had pre-existing disease and had received prior immunosuppressive therapies, including cyclophosphamide, which may have contributed to the enhanced number of solid organ tumours that occurred in the etanercept group.

LESSONS FROM ANIMAL MODELS/ ANCA PATHOGENICITY

A pathogenetic role of ANCA has been postulated since their close association with small-vessel vasculitis was discovered. Clinically, ANCA titres are frequently related to disease activity or relapse.[29] Therefore, a rising ANCA in a patient in clinical remission should raise suspicion of relapse and increase vigilance and follow-up frequency. *In vitro* experiments show that ANCA induce neutrophil activation by engagement of their target antigens MPO and PR3.[30] Adhesion studies under flow conditions, in which neutrophils are perfused through glass microslides coated with platelets or endothelial cells, show that ANCA play an important role in adhesion and migration. Activation of endothelial cells with low concentrations of TNFα followed by infusion of ANCA IgG resulted in stabilised adhesion and a tenfold increase in the number of transmigrating neutrophils.[31,32] Adhesion and migration require activation of neutrophil β2 integrins and involve the chemokine receptor CXCR2.[33]

A number of experimental models provide evidence that MPO-ANCA can induce crescentic glomerulonephritis, pulmonary capillaritis and systemic vasculitis. Immunisation of MPO-knockout mice with murine MPO induced MPO-ANCA, and when these were injected, immunodeficient or wild-type mice developed pauci-immune focal necrotising glomerulonephritis.[34] A similar approach did generate PR3-ANCA, but passive transfer of these did not induce vasculitis,[35] but significantly aggravated the local inflammatory response induced by subcutaneous TNF-α administration, thus providing evidence to support PR3-mediated tissue damage *in vivo*.

Little *et al*. have developed a rat model of focal necrotising crescentic glomerulonephritis and pulmonary capillaritis induced through immunisation with purified human MPO. This model has the advantage that it specified the amount of MPO required to induce disease, and more importantly, all animals developed renal and pulmonary damage with reduced variability in severity between animals.[36,37] In the same study, Little *et al*. explored the effects of MPO-ANCA on the induction of leucocyte-endothelium interactions using intravital microscopy of mesenteric venules.[36] Localised administration of the chemokine CXCL-1 (a rat homologue of interleukin-8) to the mesenterium of both immunised and naive rats, pretreated with purified IgG from sera of MPO-immunised rats, led to increased leucocyte adherence, transmigration and focal haemorrhage at chemokine application sites. This work confirms the direct pathogenic effect of MPO-ANCA, and suggests that ANCA pathogenicity is at least in part mediated through promotion of neutrophil adhesion to endothelium *in vivo*.

It has been assumed that complement activation is not involved in the pathogenesis of AAV because of the paucity of immunoglobulin and complement deposits in affected blood vessels and the absence of hypocomplementaemia. Recent evidence, however, points to an important role of complement activation in AAV; *in vitro* activation of human neutrophils by MPO-ANCA or PR3-ANCA leads to complement activation including activation of C3a.[38] *In vivo* complement depletion with cobra venom factor prevented the development of vasculitis following injection of MPO IgG or transfer of anti-MPO splenocytes. Furthermore, a common complement pathway inhibiting C5 antibody prevented or ameliorated MPO IgG-mediated glomerulonephritis when given before or after disease induction, respectively.[39] Studies using mice deficient in specific

complement pathways show that MPO IgG-mediated glomerulonephritis is dependent on the alternative complement pathway.[38] Immunofluorescence microscopy shows deposition of the complement component C3c in glomerular capillaries or mesangium in 33% of patients with AAV and this was associated with elevated proteinuria and more severe renal injury.[40] Overall, these studies support a crucial role for alternative pathway complement activation in AAV and suggest that complement inhibition may be a target for future therapies.

Recently, Kain *et al.*[41] were able to induce pauci-immune glomerulonephritis by immunising rats with rabbit immunoglobulin specific to human lysosomal-associated membrane protein-2 (LAMP-2), which cross-reacts with rat LAMP-2. All rats developed severe renal injury; 22% of glomeruli exhibited focal capillary necrosis after 24 hours, and 21% of glomeruli developed crescents within 48 hours. Anti-human LAMP-2 interestingly shares 100% homology and cross-reacts with the bacterial adhesin FimH, which also induces antibodies to human and rat LAMP-2, as well as causing pauci-immune glomerulonephritis when injected into rats, indicating that fimbriated Gram-negative bacteria are involved in the pathogenesis of AAV by triggering autoimmunity. In the study by Kain *et al.*, anti-LAMP-2 was detectable in almost all patients with focal necrotising glomerulonephritis, which represents a much higher prevalence than that of ANCA directed against PR3 or MPO. This raises the question of whether these antibodies are specific for AAV or more generally involved in vasculitic lesions.

SUMMARY

It has become increasingly clear over the last two decades that ANCA IgG is pathogenic in AAV. This is supported by evidence from *in vitro* and *in vivo* animal studies, as well as clinical observations from patients with AAV. At the same time, ANCA testing has contributed enormously to more rapid diagnosis of new cases, and early recognition of relapse in patients with AAV. Conventional treatments, such as cyclophosphamide and glucocorticoids, have remained the mainstay of therapy in generalised disease, but some progress has been made in minimising treatment dose. Novel therapies aimed at selected cell populations or blocking specific pathogenic pathways offer hope for more selectively treating this heterogeneous group of patients, while avoiding nonspecific immunosuppression and its adverse effects. However, this will not only require controlled trials to evaluate these novel therapies, but also a concerted effort to further enhance our understanding of the mechanisms behind ANCA pathogenesis.

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REVIEW

Bleeding in patients using new anticoagulants or antiplatelet agents: risk factors and management

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ABSTRACT

The most important adverse effect of antithrombotic treatment is the occurrence of bleeding. In case of serious or even life-threatening bleeding in a patient who uses anticoagulant agents or when patient on anticoagulants needs to undergo an urgent invasive procedure, anticoagulant treatment can be reversed by various specific strategies. Heparin and heparin derivatives can be counteracted by protamine sulphate, whereas the anticoagulant effect of vitamin K antagonists may be neutralised by administration of vitamin K or prothrombin complex concentrates. The antihaemostatic effect of aspirin and other antiplatelet strategies can be corrected by the administration of platelet concentrate and/or desmopressin, if needed. Recently, a new generation of anticoagulants with a greater specificity towards activated coagulation factors has been introduced and most of these agents are currently being evaluated in clinical studies, showing promising results. The new-generation anticoagulants include specific inhibitors of factor IIa or factor Xa (including pentasaccharides) and antiplatelet agents belonging to the class of thienopyridine derivatives. A limitation of the new class of anti-IIa and anti-Xa agents may be the lack of an appropriate strategy to reverse the effect if a bleeding event occurs, although in some cases the administration of recombinant factor VIIa may be an option.

KEYWORDS

Anticoagulants, haemorrhage, heparin, pentasaccharides, vitamin K antagonists, aspirin, clopidogrel, prasugrel, cangrelor

INTRODUCTION

Anticoagulant agents are usually for prevention and treatment of a wide range of cardiovascular diseases. The most frequently used anticoagulants are heparin or its derivatives, vitamin K antagonists (such as warfarin or coumadin) and antiplatelet agents, including aspirin and thienopyridine derivatives, such as clopidogrel or prasugrel. A myriad of clinical studies have demonstrated that these agents (alone or in combination) can prevent or treat acute or chronic thromboembolic complications, such as in patients with atrial fibrillation or prosthetic heart valves, after myocardial infarction, percutaneous coronary interventions, or ischaemic stroke, and in patients with venous thrombosis or pulmonary embolism.[1] The most important complication of treatment with anticoagulants is haemorrhage, which may be serious, may cause long-term debilitating disease, or may even be life-threatening.[2] In a very large series of 34,146 patients with acute ischaemic coronary syndromes, anticoagulant-associated bleeding was associated with a fivefold increased risk of death during the first 30 days and a 1.5-fold higher mortality between 30 days and six months.[3] Major bleeding was an independent predictor of mortality across all subgroups that were analysed. In some clinical situations the incidence of serious bleeding complications may annihilate or even overwhelm the efficacy of antithrombotic agents, as has been shown in the secondary prevention of patients with ischaemic stroke by vitamin K antagonists.[4] Nevertheless, in many situations clinical studies show a favourable balance between efficacy and safety in favour of anticoagulant treatment. However, if severe bleeding occurs or if a patient needs to undergo an urgent invasive procedure, such as emergency surgery, it may be required to reverse the anticoagulant effect of the various agents. Depending on the clinical situation, i.e. the severity of the

bleeding or the urgency and estimated risk of the invasive procedure, this reversal may take place in a few hours, but in some cases immediate reversal is necessary (*table 1*).[5] Generally, each (immediate) reversal of anticoagulant treatment also needs to take into consideration the indication for the antithrombotic agents. For example, the interruption of combined aspirin and clopidogrel treatment in a patient in whom an intracoronary stent has recently been inserted will markedly increase the risk of acute stent thrombosis with consequent downstream cardiac ischaemia or infarction. Likewise, in a patient with a prosthetic mitral valve and atrial fibrillation, interruption of vitamin K antagonists may increase the risk of valve thrombosis and cerebral or systemic embolism. Each of these specific clinical situations requires a careful and balanced assessment of the benefits and risks of reversing anticoagulants (and potential strategies to keep the period of reversal as short as possible). In this article, we will describe the various strategies to reverse the anticoagulant effect of the currently most widely used antithrombotic agents and the new generation of anticoagulants.

INCIDENCE AND RISK FACTORS FOR BLEEDING IN PATIENTS ON VKAS

Vitamin K antagonists (VKAs) (such as warfarin, coumadin, acenocoumarol or phenprocoumon) are often used for prevention and treatment of a wide range of cardiovascular diseases. The most important complication of treatment with these agents is haemorrhage, which may be life-threatening.[2] In well-controlled patients in clinical trials treatment with VKAs increases the risk of major bleeding by 0.5%/year and the risk of intracranial haemorrhage by about 0.2%/year.[6]

The most important risk factor for haemorrhage in users of VKAs is the intensity of the anticoagulant effect.[6] Studies indicate that with a target INR of >3.0 the incidence of major bleeding is twice as high as in studies with a target INR of 2.0 to 3.0.[7] In a meta-analysis of studies in patients with prosthetic heart valves, a lower INR target range resulted in a lower frequency of major bleeding and intracranial haemorrhage with a similar antithrombotic efficacy.[8] A retrospective analysis of outpatients using warfarin who presented with intracranial haemorrhage demonstrated that the risk of this complication doubled for each 1 unit increment of the INR.[9] Patient characteristics constitute another important determinant of the risk bleeding. Elderly patients have a twofold increased risk of bleeding[10] and the relative risk of intracranial haemorrhage (in particular at higher INRs) was 2.5 (95% CI 2.3 to 9.4) in patients >85 years compared with patients 70 to 74 years.[11] Recently, genetic factors have been identified that may affect the risk of bleeding. Common polymorphisms in the P450 CYP2C9 enzyme were found to be associated with slow metabolism of VKAs and (possibly) a higher risk of bleeding.[6,12] Other genetic factors that may influence the requirement of VKAs are variants in the vitamin K epoxide reductase complex subunit 1 gene (*VKORC1*).[13] Comorbidity, such as renal or hepatic insufficiency, may also significantly increase the risk of bleeding. A case-control study in 1986 patients on VKAs showed that this comorbidity increased

Table 1.

	Time until restoration of haemostasis after cessation of therapeutic dose	Antidote	Remark
Heparin	3-4 hrs	Protamine sulphate 25-30 mg; immediate reversal	1 mg of protamin per 100 anti-Xa units given in the last 2-3 hrs
LMW heparin	12-24 hrs	(Partially) protamine sulphate 25-50 mg; immediate reversal	1 mg of protamine per 100 anti-Xa units given in the last 8 hrs
Pentasaccharides	Fondaparinux: 24-30 hrs Idraparinux: 5-15 days	Recombinant factor VIIa 90 µg/kg (?); immediate thrombin generation	Based on laboratory endpoints, no systematic experience in bleeding patients
Vitamin K antagonists	Acenocoumarol: 18-24 hr Warfarin: 60-80 hrs Phenprocoumon: 8-10 days	Vitamin K iv; reversal in 12-16 hrs Vitamin K orall;: reversal in 24 hrs PCCs: immediate reversal	Dose of vitamin K or PCCs depends on INR and bodyweight
Oral thrombin and factor Xa inhibitors	Dependent of compound, usually within 12 hrs	Recombinant factor Xa for Xa inhibitors, unsure for IIa inhibitors	Based on laboratory endpoints, no systematic experience in bleeding patients
Aspirin	5-10 days (time to produce unaffected platelets)	DDAVP (0.3-0.4 µg/kg) and/or platelet concentrate; reversal in 15-30 min	Cessation not always required, also dependent on clinical situation and indication
Clopidogrel Prasugrel	1-2 days	Platelet concentrate, possibly in combination with DDAVP (0.3-0.4 µg/kg); reversal in 15-30 min	Cessation not always desirable, also dependent on clinical situation and indication

LMW heparin = low-molecular-weight heparin; PCC = prothrombin complex concentrate; DDAVP = de-amino d-arginin vasopressin or desmopressin.

the risk of bleeding by about 2.5.[14] Another very important determinant of the risk of bleeding is the use of other medication, in particular agents affecting platelet function. Two meta-analyses, comprising six trials with a total of 3874 patients and ten trials with a total of 5938 patients, found a relative risk of major bleeding when VKAs were combined with aspirin of 2.4 (95% CI 1.2 to 4.8) and 2.5 (95% CI 1.7 to 3.7), respectively.[15,16] A population-based case-control study confirmed the high risk of upper gastrointestinal bleeding in patients using VKAs in combination with aspirin and/or clopidogrel.[17] Nonsteroidal anti-inflammatory agents (NSAIDs) are also associated with an enhanced risk of gastrointestinal bleeding. The combined use of VKAs and NSAIDs may result in an 11-fold higher risk of hospitalisation for gastrointestinal bleeding as compared with the general population.[18] This risk is not significantly lower when using selective inhibitors of COX-2.[19]

In case of major bleeding it may be required to reverse the anticoagulant effect of the various agents.[5] When interrupting the administration of VKAs important differences in the half-lives of the various agents (9 hours for acenocoumarol, 36-42 hours for warfarin, and 90 hours for phenprocoumon, respectively) need to be taken into account.[20] The most straightforward intervention to counteract the effect of VKAs is the administration of vitamin K.[21] There is quite some debate on the use of vitamin K in patients with a too high INR but no signs of bleeding. However, a recent randomised controlled trial did not find any difference in bleeding or other complications in nonbleeding patients with INR values of 4.5 to 10 who were treated with vitamin K or placebo.[22] In patients with clinically significant bleeding, administration of vitamin K is crucial to reverse the anticoagulant effect of VKAs. Vitamin K can be given orally and intravenously, whereas the parenteral route has the advantage of a more rapid onset of the treatment.[23] After the administration of intravenous vitamin K, the INR will start to drop within two hours and will be completely normalised within 12 to 16 hours,[24] whereas after oral administration it will take up to 24 hours to normalise the INR.[21] Intramuscular injections of vitamin K should be avoided in patients who are anticoagulated and subcutaneous administration of vitamin K results in a less predictable bioavailability.[23] A potential concern with the use of parenteral vitamin K is the occurrence of anaphylactic reactions, although the incidence of this complication is very low, in particular with the more modern micelle preparations.[25]

In case of very serious or even life-threatening bleeding, immediate correction of the INR is mandatory and can be achieved by the administration of vitamin K-dependent coagulation factors. Theoretically, these factors are present in fresh frozen plasma; however, the amount of plasma that is required to correct the INR is very large, carries the risk of fluid overload, and will probably take hours to administer.[26] Therefore, prothrombin complex concentrates (PCCs), containing all vitamin K-dependent coagulation factors, are more useful. Although PCCs can indeed be given using fixed dose schemes, it has been shown that individualised dosing regimens based on INR at presentation and body weight are more effective.[27] A prospective study in patients using VKA and presenting with bleeding also found that PCCs resulted in at least satisfactory and sustained haemostasis in 98%.[28] In recent years the safety of PCCs, in particular regarding the transmission of blood-borne infectious diseases, has markedly improved owing to several techniques, such as pasteurisation, nanofiltration, and addition of solvent detergent. The risk of disseminated intravascular coagulation (DIC) due to traces of activated coagulation factors in PCCs comes from older literature and modern PCCs do not seem to be associated with eliciting DIC.[27]

HEPARIN AND LOW-MOLECULAR-WEIGHT (LMW) HEPARIN

Herparin and heparin derivatives act by binding to antithrombin and thereby about 1000-fold potentiating the anticoagulant effect of this endogenous inhibitor towards thrombin and factor Xa (and some other coagulation factors). Heparin has a relatively short half-life of about 60 to 90 minutes and therefore the anticoagulant effect of therapeutic doses of heparin will be mostly eliminated at three to four hours after termination of continuous intravenous administration.[30] The anticoagulant effect of high-dose subcutaneous heparin, however, will take a longer time to abolish. If a more immediate neutralisation of heparin is required, intravenous protamine sulphate is the antidote of choice. Protamine, derived from fish sperm, binds to heparin to form a stable biologically inactive complex. Each mg of protamine will neutralise approximately 100 units of heparin. Hence, the protamine dose in a patient on a stable therapeutic heparin dose of 1000 to 1250 U/h should be about 25 to 30 mg (sufficient to block the amount of heparin given in the last two to three hours). The maximum dose of protamine is 50 mg. Since the half-life of protamine is only about ten minutes, the reversal of therapeutic dose subcutaneous heparin requires a repeated infusion of protamine sulphate (e.g. repeated after one hour). The effect of protamine can be monitored by measuring the activated partial thromboplastin time (aPTT), which should normalise after its administration.

The reversal of LMW heparin is more complex, as protamine sulphate will only neutralise the anti-factor IIa activity and has no or only partial effect on the smaller heparin fragments causing the anti-factor Xa activity of the compound.[31,32] The net effect of protamine reversal of LMW heparin is not completely clear. There are no clinical studies that have systematically studied this and small case

series and experimental animal studies show contradictory results.[32-34] As the aPTT is not useful as a monitoring assay when using LMW heparin, it can not be used for the monitoring of the neutralising effect of protamine either. Given the relatively long half-life of LMW heparin, the lack of an adequate strategy to reverse its anticoagulant action may sometimes cause a problem in clinical situations. A practical approach is to give 1 mg of protamine per 100 anti-factor Xa units of LMW heparin given in the last eight hours (whereas 1 mg of enoxaparin equals 100 anti-factor Xa units). If bleeding continues, a second dose of 0.5 mg per 100 anti-factor Xa units can be given.

The most important adverse effect of protamine is an allergic response, including haemodynamic and respiratory problems.[35] Most adverse reactions can be prevented or minimised by slowing the rate of administration of the drug or by pretreatment with steroids and antihistamines. Risk factors for an adverse reaction are sensitivity to fish (as may occur in traditional fishermen that are often exposed to fish proteins when cutting themselves), a history of vasectomy (which may demolish the blood-testis barrier with consequent formation of antisemen antibodies) and a history of receiving protamine sulphate containing insulin. Initial reports that the use of protamine sulphate could lead to an increased risk of rebound thrombosis, in particular ischaemic stroke[36,37] were not confirmed in a recent randomised controlled study.[38]

There are some other strategies to reverse (mostly unfractionated) heparin, such as platelet factor-4, heparanase, or extracorporeal heparin-removal devices, but none of these approaches have been properly evaluated and they are not currently approved for clinical use.[39-41]

PENTASACCHARIDES

Pentasaccharides are recently developed synthetic compounds that effectively bind and potentiate antithrombin to block factor Xa. Since they lack the additional glycosaminoglycan saccharide residues to bind to thrombin, they have an effect on factor Xa exclusively. The prototype pentasaccharide (and the only one approved for clinical use so far) is fondaparinux. Another pentasaccharide that is currently under study is idraparinux. The main difference between these two agents is the elimination half-life, which is 15 to 20 hours for fondaparinux and $5^1/_2$ days for idraparinux. This means that idraparinux can be administered once weekly, which renders the subcutaneous route of administration less cumbersome. Pentasaccharides were shown to be effective in the prophylaxis and treatment of venous thromboembolism and are currently evaluated in other types of thrombosis.[42] The (very) long half-life of pentasaccharides necessitates the availability of a suitable antidote if major bleeding complicates the treatment, which may especially occur in patients who are treated with therapeutic doses of this type of anticoagulation. So far, there is no antidote for the pentasaccharides that have been studied in controlled clinical studies.[43] The only agent that has been systematically evaluated to reverse the anticoagulant effect of pentasaccharides is recombinant factor VIIa (rVIIa). Two randomised placebo-controlled studies in healthy volunteers have tested the hypothesis that rVIIa may be useful as a suitable antidote for pentasaccharide anticoagulation.[44,45] In the first study, 16 subjects were treated with therapeutic doses of the pentasaccharide fondaparinux and after two hours (at the time of maximal anticoagulation) challenged with rVIIa or placebo. Injection of rVIIa (90 μg/kg) after fondaparinux normalised the prolonged aPTT and prothrombin time (PT) and reversed the decrease in prothrombin activation fragments 1+2 (F(1+2)), as observed with fondaparinux alone. Thrombin-generation time and endogenous thrombin potential, which were inhibited by fondaparinux, normalised up to six hours after rVIIa injection. In the second study 12 subjects received a single subcutaneous dose of 7.5 mg idraparinux (which is threefold higher than the currently recommended dose). The inhibition of thrombin generation by idraparinux, as reflected by an increased thrombin generation time (TGT) and decreased level of prothrombin fragment 1+2 (F_{1+2}), was partially reversed by injection of rVIIa three hours after idraparinux administration. The administration of rVIIa one week after treatment with idraparinux (when much lower, though still therapeutic, doses of the pentasaccharide were present) resulted in an nearly complete reversal of anticoagulation, reflected by normalisation of thrombin generation time and other markers of thrombin generation. As mentioned, there are no controlled trials in patients who present with pentasaccharide-induced bleeding but there is some anecdotal experience suggesting that rVIIa may indeed be able to stop bleeding in patients anticoagulated with fondaparinux.

NEW DIRECT FACTOR XA INHIBITORS

In recent years a large number of new antithrombotic agents have been developed and tested in clinical trials and many of these new agents will become available for clinical practice in the very near future.[46] The need for new anticoagulant agents is quite obvious. Firstly, the current agents are insufficiently effective. For example, 10 to 15% of patients undergoing major orthopaedic surgery develop venous thromboembolism, despite prophylaxis with low-molecular-weight (LMW) heparin.[47] Furthermore, the available anticoagulants are relatively unsafe, mostly due to the occurrence of bleeding as

discussed above. Lastly, current anticoagulant agents are often cumbersome with regards to their clinical use, requiring repeated laboratory control and frequent dose adjustments. Increasing knowledge on the function of the haemostatic system *in vivo* has resulted in a new generation of anticoagulant agents.

Some of these new classes of anticoagulants are directed at factor Xa. Prototypes of these agents are rivaroxaban and apixaban, which have shown promising results in initial experimental and clinical studies.[48,49] Rivaroxaban was evaluated in a series of trials in patients undergoing major orthopaedic surgery (RECORD studies), which showed a higher efficacy of the direct anti-Xa inhibitor compared with enoxaparin and similar bleeding rates.[50,51] Apixaban was also compared with enoxaparin in patients undergoing knee replacement surgery and was shown to be equally effective but had significantly less bleeding complications (2.9% in the apixaban group compared with 4.3% in the enoxaparin group).[52] In dose-ranging trials in patients with acute venous thromboembolism. rivaroxaban and apixaban were as effective as LMW heparin but rivaroxaban was associated with a lower incidence of bleeding complications (2.2 *vs* 8.8%).[53,54] Rivaroxaban was also studied in patients with acute coronary syndromes and showed a dose-dependent efficacy but also increased rates of major bleeding at higher doses.[55] Similarly, apixaban showed a similar pattern and exhibited 2.5-fold increased bleeding rates, in particular in patients using simultaneous antiplatelet agents.[56] Taken together, compared with LMW heparin, direct factor Xa inhibitors result in a lower bleeding risk at doses achieving equivalent efficacy and a similar bleeding risk at doses achieving higher efficacy. This means that for some clinical situations these drugs may represent an important improvement; however, the risk of (major) bleeding is still present.

Dependent on the severity of the clinical situation and in view of the half-life of the direct Xa inhibitors, cessation of medication may be sufficient to reverse the anticoagulant effect in case of bleeding. However, if immediate reversal of anticoagulation is required, there is no evidence of any antidotes against the anticoagulant effect of any of these orally available factor Xa inhibitors so far. Based on the experience with rVIIa in the reversal of the anticoagulant effect of fondaparinux, one can postulate that rVIIa may be an effective antidote for these agents; however, direct proof has not been demonstrated.

DIRECT THROMBIN INHIBITORS

Another important group of new anticoagulants is the class of direct thrombin inhibitors. Thrombin is the central enzyme in the coagulation process, not only mediating the conversion of fibrinogen to fibrin, but also being the most important physiological activator of platelets and various other coagulation factors. Inhibition of thrombin can be achieved by administration of heparin, but in view of the limited capability of the heparin-antithrombin complex to inhibit surface-bound thrombin, new antithrombin-independent anticoagulants have been developed.[57] The prototype of these thrombin inhibitors is hirudin, originally derived from the saliva from leeches (*hirudo medicinalis*) and nowadays produced by recombinant technology. Melagatran is a synthetic thrombin inhibitor, which has predictable pharmacokinetic properties and can thus be used in a fixed dose.[58] Moreover, the pro-drug ximelagatran is relatively quickly absorbed after oral ingestion and results in a sufficient systemic availability, rendering this agent suitable for long-term use as an oral anticoagulant. Despite clinical trials on prevention and treatment of venous thromboembolism and in patients with atrial fibrillation showing a promising efficacy of (xi) melagatran, the compound has been withdrawn by the manufacturer due to the occurrence of enhanced liver enzymes in 6 to 7% of patients. Recently, dabigatran, also a direct thrombin inhibitor with good and relatively stable bioavailability after oral ingestion, was introduced and licensed for prevention of venous thromboembolism after orthopaedic surgery. Indeed, clinical trials evaluating dabigatran against LMW heparin in patients undergoing major orthopaedic surgery show similar or slightly better efficacy of the direct thrombin inhibitor and similar bleeding rates.[59,60] The largest group of patients using long-term anticoagulants, however, are those with atrial fibrillation. In these patients dabigatran (150 mg twice daily) showed a significantly lower rate of thromboembolic complications compared with warfarin (relative risk 0.66; 95% CI 0.53 to 0.82) but also a slightly lower risk of major haemorrhage (3.11% per year in the dabigatran group *vs* 3.36% per year in the warfarin group).[61] Based on these findings and if confirmed by other ongoing major trials, it may be quite likely that in the future oral anticoagulant treatment with vitamin K antagonists is going to be replaced by treatment with directly acting anticoagulants, such as direct thrombin inhibitors. However, the risk of major bleeding is still relatively large and requires adequate management strategies.

No established antidote is available in case of serious bleeding complicating the anticoagulant treatment for any of the direct thrombin inhibitors. Again, the half life of most of the agents is relatively short, hence with less serious bleeding interruption of treatment will be sufficient to reverse the anticoagulant effect. However, if immediate reversal is required, it is not clear which would be the best strategy. In a controlled clinical study in healthy subjects the melagatran-induced effects on aPTT, thrombin generation and platelet activation were not affected by the administration of rVIIa.[62] Based on

these results it seems that rVIIa is not effective in reversing direct thrombin inhibition. Since, however, rVIIa was able to correct the melagatran-induced prolongation of the prothrombin time and increased thrombin precursor protein concentrations, it might be that higher doses of rVIIa will have some effect in this situation, but this needs to be studied in future experiments.

ASPIRIN

Aspirin is effective in the secondary prevention of atherothrombotic disease, in particular coronary artery disease, cerebrovascular thromboembolism and peripheral arterial disease.[63] As a consequence, aspirin is one of the most widely used agents in the Western world. Aspirin increases the risk of bleeding, in particular gastrointestinal bleeding, and has been associated with a small but consistent increase in intracerebral haemorrhage. In addition, it has been shown that the use of aspirin is associated with increased perioperative blood loss in major procedures, although this does not necessarily translate into clinically relevant endpoints, such as the requirement for transfusion or reoperation.[64] Over the last years the approach to the patient who uses aspirin and who presents with bleeding or needs to undergo an invasive procedure has changed considerably. In fact, in current clinical practice bleeding can almost always be managed with local haemostatic procedures or conservative strategies without interrupting aspirin and also most invasive procedures do not require the cessation of aspirin when adequate attention is given to local haemostasis. In contrast, interruption of aspirin has been associated with an increased risk of thromboembolic complications, potentially due to a rebound hypercoagulability. Obviously, in special clinical circumstances, such as intracranial bleeding or the need to undergo a neurosurgical or ophthalmic procedure, the antihaemostatic effect of aspirin needs to be reversed immediately. The most rigorous measure to achieve that is the administration of platelet concentrate after cessation of aspirin. Another approach is the administration of de-amino d-arginin vasopressin (DDAVP, desmopressin). DDAVP is a vasopressin analogue that despite minor molecular differences has retained its antidiuretic properties but has much less vasoactive effects.[65] DDAVP induces release of the contents of the endothelial cell associated Weibel Palade bodies, including von Willebrand factor. Hence, the administration of DDAVP results in a marked increase in the plasma concentration of von Willebrand factor (and associated coagulation factor VIII) and (also by yet unexplained additional mechanisms) a remarkable augmentation of primary haemostasis as a consequence. DDAVP is effective in patients with mild haemophilia A or von Willebrand's disease and in patients with qualitative platelet defects, such as in uraemia or liver cirrhosis. DDAVP also seems capable of correcting the aspirin-induced platelet dysfunction, although large clinical studies employing relevant outcome parameters are missing.[66] The combined effect of platelet concentrate and subsequent administration of DDAVP has also been advocated to correct the aspirin effect on platelets. The standard dose of DDAVP is 0.3 to 0.4 μg/kg in 100 ml saline over 30 minutes and its effect is immediate.

THIENOPYRIDINE DERIVATIVES

Clopidogrel and prasugrel belong to the class of thienopyridine derivatives, which act by blocking the adenosine diphosphate (ADP) receptor on the platelet. Clinical studies have shown that clopidogrel is as good as aspirin in the secondary prevention of atherothrombotic events.[67] Importantly, the combination of aspirin and clopidogrel is vastly superior over aspirin alone in patients who have received intracoronary stents or in other patients with high-risk coronary artery disease. There is ample evidence that dual platelet inhibition of aspirin plus clopidogrel has a significantly higher efficacy than aspirin alone in patients with acute coronary syndromes who have undergone coronary interventions for at least a year (and possibly longer) after the event. However, the increased efficacy of the combined use of aspirin and clopidogrel is also associated with a significantly higher bleeding risk.[68] Prasugrel is another thienopyridine derivative that after rapid and almost complete absorption after oral ingestion irreversibly binds to the ADP receptor. Prasugrel has a stronger antiplatelet effect than clopidogrel because of more effective metabolism and less dependence on cytochrome P450 enzymes that may be subject to genetic polymorphisms.[69] Prasugrel was shown to be more effective than clopidogrel in preventing ischaemic events in patients with ST-elevation myocardial infarction undergoing primary percutaneous coronary interventions (with or without stent).[70] Rates of major bleeding were similar between clopidogrel and prasugrel; however, the rate of serious bleeding in patients requiring emergency coronary artery bypass grafting (CABG) was higher in the prasugrel group. In patients with acute coronary syndromes, prasugrel was also more effective than clopidogrel in preventing cardiovascular death, myocardial infarction and stroke; however, major bleeding rates were higher in the prasugrel group (2.4 *vs* 1.8%).[71] Of note, this disadvantage of prasugrel did not outweigh the efficacy benefit, and the net clinical benefit (defined as the efficacy gain minus the increased risk of major bleeding) was preserved in favour of prasugrel. Recently, a third thienopyridine derivative has been introduced: cangrelor. The advantage of this compound over the other members

of this group is the faster onset of action, which may be critical in acute coronary syndromes. However, two major clinical trials comparing cangrelor with clopidogrel in patients undergoing percutaneous coronary interventions did not show a higher efficacy of cangrelor but did demonstrate a significantly higher risk of bleeding.[72,73] Taken together, dual platelet inhibition, in particular with clopidogrel or even more outspoken with prasugrel, is highly effective in high-risk patients with coronary artery disease but the bleeding risk with dual platelet inhibition is something to take into account and strategies to reverse the antiplatelet effect may be warranted in case of serious bleeding.

The decision whether or not the interrupt or even reverse antithrombotic treatment with dual platelet inhibition in case of serious bleeding or the need to perform an invasive procedure will depend on the specific clinical situation but also on the indication for the antithrombotic treatment (see above). Especially in patients with recent implantation of an intracoronary stent (in the last 6 to 12 weeks), cardiologists will often not or only reluctantly agree to cessation of treatment.[74] In this period re-endothelialisation of the stent has not yet occurred and the patient is very vulnerable to acute thrombotic occlusion of the stent. In patients with drug-eluting stents this period may be even longer. If, however, the decision is made to stop and even reverse the treatment with aspirin and clopidogrel, administration of platelet concentrate is probably the best way to correct the haemostatic defect.[75] In addition, DDAVP was shown to correct the defect in platelet aggregation caused by clopidogrel, so this may be another option.[76]

CONCLUSION

Conventional anticoagulant treatment can be reversed by specific interventions when the clinical situation requires immediate correction of haemostasis. For the new generation of anticoagulants, no specific antidotes are available, although some interventions are promising but need further evaluation. Antiplatelet therapy with aspirin, alone or in combination with thienopyridine derivatives, such as clopidogrel and prasugrel, can be reversed but this is often not required and sometimes not desirable in view of the indication for this treatment.

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ORIGINAL ARTICLE

Survey into blood glucose control in critically ill adult patients in the Netherlands

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ABSTRACT

Background: To study current clinical practice in blood glucose (BG) control in adult intensive care units (ICUs) in the Netherlands.

Methods: We performed a national survey focusing on blood glucose targets, insulin administration, BG control guidelines, and opinions regarding BG control aiming for normoglycaemia (known as intensive insulin therapy, IIT).

Results: The completed questionnaire was returned by 88/113 (78%) of the participating centres. In 98% (86/88) of the ICUs some sort of BG control was being practised. Half of the ICUs (42/86, 48%) used tight BG targets as with IIT; 28/86 (33%) and 13/86 (15%) used more liberal targets of 4.4 to 7.0 mmol/l and 4.4 to 8.0 mmol/l, respectively. Eighty-two (93%) reported having a local guideline on BG control (or IIT). The BG threshold to start insulin was 7.0±1.3 mmol/l *vs* 7.8±1.3 mmol/l in ICUs that practised IIT *vs* ICUs that practised less tight BG control, respectively (p=0.005). In 28/86 (33%) measurement of the BG values was done according to a strict time schedule (i.e., BG values were measured on predefined time points). While respondents were fairly agreed on the benefits of IIT, opinions regarding ease of implementation and time needed to apply this strategy varied. In addition, severe hypoglycaemia was considered a serious side effect of IIT.

Conclusion: Approximately half of the ICUs in the Netherlands reported having implemented IIT. However, the full guideline as used in the original studies on IIT was hardly ever implemented. Concerns about severe hypoglycaemia, at least in part, hampers implementation of IIT.

KEYWORDS

Glucose control; intensive insulin therapy; guideline; survey

INTRODUCTION

The optimal blood glucose target and best way to control blood glucose are currently undecided for adult critically ill patients. Although intensive insulin therapy (IIT, blood glucose control aiming at blood glucose levels of 4.4 to 6.1 mmol/l) improved the mortality and morbidity of adult critically ill surgical and medical patients in two randomised controlled trials,[1,2] the benefit of IIT was questioned in three recent studies[3-5] and one meta-analysis.[6] Concerns about severe hypoglycaemic events associated with implementation of IIT have been another reason to advise against implementing IIT and to accept higher blood glucose targets,[7-10] thereby potentially losing any benefit that is associated with IIT.[11-13] Other factors potentially impeding implementation of IIT may include the belief that this strategy is time consuming and costly, and the lack of a common guideline for IIT.

Understanding of current blood glucose control practice patterns, as well as beliefs and concerns surrounding blood glucose control, in particular IIT, is essential for development of (inter)nationally accepted guidelines for blood glucose control in adult critically ill patients. We hypothesised that IIT is far from being implemented in critically ill patients in the Netherlands, and if implemented that it differs significantly from the guideline

as originally described.[1,2] This postal survey amongst adult intensive care units (ICUs) in the Netherlands explored this hypothesis.

MATERIALS AND METHODS

This survey was conducted with the approval of the institutional review board of the Academic Medical Centre, Amsterdam, the Netherlands, which waived the need for informed consent. Respondents were told that consent for participation in the survey was implied if they answered and returned the questionnaire.

Study population

A questionnaire was sent to ICU physicians and/or ICU nurses in the Netherlands. Neonatal and paediatric ICUs were excluded from the survey. The medical directors of all adult ICUs were contacted and asked to appoint one ICU physician and/or one registered ICU nurse engaged in blood glucose control to complete the questionnaire, after which these participants were contacted.

Definitions

In the survey 'blood glucose control (with insulin)' was defined as any strategy aiming at a certain blood glucose level or range (with insulin); IIT was defined as blood glucose control aiming at the tight blood glucose targets of 4.4 to 6.1 mmol/l.[1,2] Hypoglycaemia is defined as blood glucose level between 2.2 to 4.4 mmol/l, severe hypoglycaemia as ≤2.2 mmol/l.

The questionnaire

A first draft of the questionnaire was developed at an informal meeting with ICU physicians and ICU nurses from four Dutch hospitals. This first draft was sent for review to two ICU physicians, four ICU nurses and one expert in informatics. They independently reviewed the text and added comments and new questions. The second draft was sent to the same experts, who all approved the questionnaire.

Questionnaire items

The questionnaire started with questions regarding demographic data, i.e., type of organisation, size and volume of responding ICU. This was followed by questions on blood glucose control and IIT. Four different aspects of blood glucose control and IIT were surveyed: 1) the availability of a guideline for blood glucose control or IIT; 2) rules for insulin administration and blood glucose targets; 3) specific measures surrounding blood glucose control or IIT; 4) opinions and behaviour in relation to IIT. The questionnaire ended with itemised statements on IIT, to which the respondents were asked to respond on a visual analogue scale (VAS, ranging from 1 for complete disagreement, to 10 for complete agreement).

Questionnaire format and pretesting

A printed questionnaire was first tested in a small subset of three ICU physicians and three ICU nurses in the Academic Medical Centre, Amsterdam, the Netherlands, to ensure that each question and statement was clear. Unclear questions and statements were rephrased with the help of these caregivers. Finally, the questionnaire was printed in an A5-format booklet.

Administration of the questionnaire

We sent the questionnaire to the selected respondents for self-administration. To maximise the response rate, we enclosed a postage-paid return envelope, and after three weeks a postal reminder was sent, and reminder phone calls were made. After a two-month response-free period, the survey was considered to be complete.

Data management and statistical analysis

Descriptive statistics of dichotomous or ordinal variables are proportions. Continuous variables are expressed by the mean ± standard deviations (SD), medians and interquartile ranges (IQR), or the odds ratio and 95% confidence interval. We determined the significance of differences between variables with χ^2 analysis (for categorical variables) and independent t-test (for continuous variables). A p value of <0.05 was considered to be significant. Effect sizes of dichotomous data or data aggregated into dichotomous data were expressed as relative risks. Multiple factors considered to be modifiers for dichotomous dependent variables were analysed by multivariate logistic regression analysis. If suitable we expressed statistical uncertainty as 95% confidence limits. Analysis were performed using SPSS version 16.0 software (SPSS Inc., Chicago, IL, USA).

RESULTS

Response rate

Of 113 adult ICUs, 88 returned a completed questionnaire (response rate 78%). Questionnaires were returned for 68% (60/88) by an ICU physician and 32% (28/88) by an ICU nurse.

ICU characteristics

The characteristics of responding ICUs are given in *table 1*.

Availability of guidelines for blood glucose control or IIT

In 98% (86/88) of the responding ICUs some sort of blood glucose control was being practised. Approximately half of these ICUs (42/86, 48%) used the tight blood glucose

Table 1. *Characteristics of responding ICU*

	N = 88
Type of hospital*, N (%)	
• Academic centres	12 (14%)
• Non-academic training centres	47 (53%)
• Non-academic nontraining centres	27 (31%)
Type of ICU (organisation)**, N (%)	
• Closed-format	79 (90%)
• Open-format	8 (10%)
Type of ICU (specialities), N (%)	
• Mixed medical-surgical	81 (92%)
• Surgical	6 (7%)
• Neurosurgery	1 (1%)
Number of ICU beds available for mechanical ventilation, N (%)	
• >20 beds	13 (15%)
• 15-20 beds	5 (6%)
• 5-15 beds	48 (55%)
• <5 beds	21 (24%)
Number of admissions per year***, N (%)	
• >2000	8 (9%)
• 1500-2000	7 (8%)
• 1000-1500	18 (20%)
• 500-1000	39 (44%)
• <500	9 (10%)
Staffing, median (IQR)	
• Board-certified ICU physicians	3.7 (2-5) FTE
• Board-certified ICU nurses	34 (24-55) FTE
• ICU fellows (13 academic or training ICU)	6 (2-10)
• Number of patients a physician attended for during office hours	6 (4-8)
• Number of patients a physician attended for during the evening/weekend	9 (6-12)
• Number of nurses per bed per 24 hours	3 (2-3)

*** two missing values; ** one missing value, *** seven missing values.**

targets as in the original studies on IIT by Van den Berghe *et al.* (i.e., 4.4 to 6.1 mmol/l);[1,2] 28/86 (33%) and 13/86 (15%) of responding ICUs used more liberal targets of 4.4 to 7.0 mmol/l and 4.4 to 8.0 mmol/l, respectively. One ICU reported accepting blood glucose values up to 10 mmol/l. Three ICUs did not use a range, but aimed for a blood glucose level of 6.5 mmol/l.

Six out of 86 ICUs (7%) reported that they did not have a (written or electronic) guideline on blood glucose control or IIT (two academic, three non-academic teaching and one non-academic nonteaching ICUs; all closed-format ICUs); 56 (65%) reported having both a physician- and a nurse-based guideline on blood glucose control or IIT; 7% (6/86) and 21% (18/86) said that the guideline on blood glucose control or IIT was either physician-based or nurse-based, respectively. Availability of a guideline was not statistically different between the different ICU types (p=0.97). Within ICUs that practised IIT the availability of a guideline was similar compared with ICUs that practised less tight blood glucose control (p=0.31).

Rules for insulin administration

The mentioned blood glucose threshold to start insulin was 7.4±1.3 mmol/l (range 6 to 12 mmol/l). ICUs that practised IIT said that they started insulin at a lower blood glucose value than ICUs that practised less tight blood glucose control (7.0±1.3 mmol/l *vs* 7.8±1.3 mmol/l, p=0.005).

The majority of respondents, 73% (64/86), said that they applied blood glucose control in all patients, irrespective of the referring speciality. This was not different for centres using IIT and less tight blood glucose control; there were no differences between medical and surgical ICUs either. In 24% (21/88) of the ICUs blood glucose control was not initiated in patients who were expected to stay on the ICU <3 days. Diabetes mellitus or no need for mechanical ventilation were only seldom a reason for not applying blood glucose control (2/80 and 10/77, respectively).

ICU physicians and ICU nurses were allowed to initiate insulin in similar frequencies (51% (45/86) and 64% (56/86), respectively), with no differences between ICUs that applied IIT and ICUs that aimed for less tight blood glucose levels (p=0.89). The same applied for insulin dose adjustments; dosing adjustments were made in 63% (55/86) and 75% (66/86) by ICU physicians and ICU nurses, respectively. ICU physicians were reported to have the exclusive legal responsibility for insulin dosing in 31/86 (35%) ICUs, while this responsibility was exclusively with ICU nurses in 28/86 (11%) and with both in 28/86 (32%); in 13/86 (15%) of the responding ICUs this was not mentioned. There were no differences between ICUs that practised IIT and ICUs that aimed for less tight blood glucose levels (p=0.65).

In most ICUs (66%, 49/88) adjustments in insulin dosing were made as a consequence of blood glucose values and according to a flow chart. In other ICUs, the insulin dose was adjusted with the help of specially developed software (6%, 6/88), or a calculation formula (14%, 12/88).

Specific measures surrounding blood glucose control or IIT

In the majority of ICUs timing of blood glucose measurement was unclear and/or highly variable. Measurement of the blood glucose values was only done according to a strict time schedule (i.e., blood glucose values were measured at predefined time points, usually in addition to the possibility to measure them in between) in approximately one third of ICUs (34%, 28/86). Of the ICU physicians, 86% (76/88) individually determined the time of the next blood glucose measurement, while for the ICU nurses this was 80% (71/88).

There were no differences between ICUs that practised IIT and ICUs that aimed for less tight blood glucose targets (p=0.65).

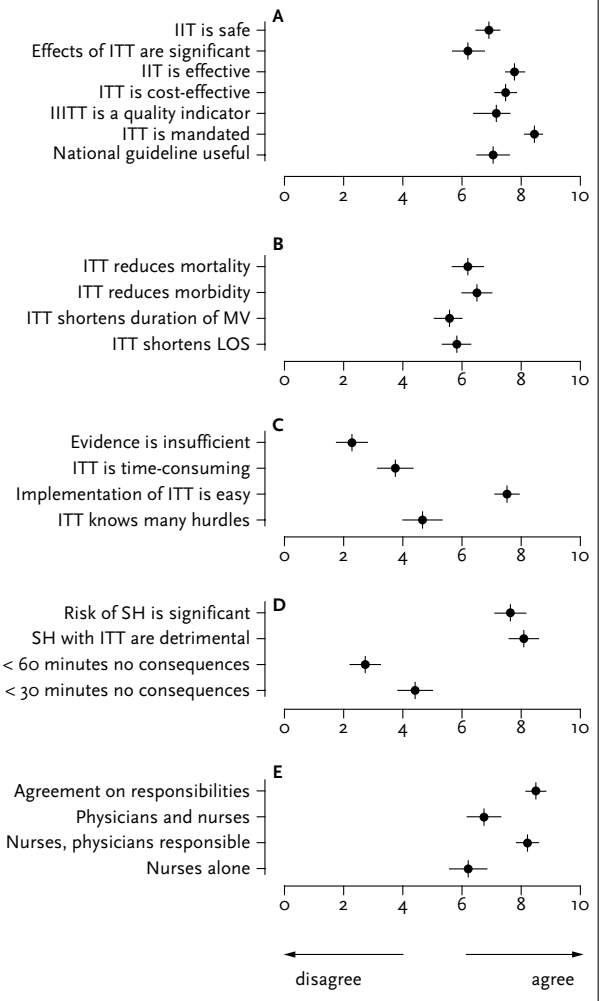
Guidelines provided adjustments for insulin dosing when patients received parenteral or enteral nutrition in 46% (40/86) of participating ICUs; corticosteroid therapy

(18/86; 21%) or the presence of diabetes mellitus was mentioned in 17/86 (19%) of the responses. Of the local guidelines, 53% (47/88) indicated that (tight) blood glucose targets should no longer be aimed for if patients were on oral feeding. Blood glucose control was to be discontinued if the patient was transferred to a step-down facility in 78% (69/86) of cases. Surprisingly, blood glucose control was also to be stopped if patients were in the predefined blood glucose ranges for two days or longer (38%, 33/86). Severe hypoglycaemia was seldom reported as a reason to stop blood glucose control (3%, 3/86).

Opinions and behaviour in relation to IIT

Figure 1 shows responses to the itemised statements regarding IIT. Most respondents agreed on the statements that IIT is safe and (cost)-effective. In agreement with this, most respondents thought of IIT as a mandatory ICU strategy and application of IIT as an indicator of ICU quality. While respondents were fairly agreed on the benefits of IIT, opinions regarding ease of implementation and time needed to apply IIT varied. In addition, severe hypoglycaemia was considered a serious side effect of IIT. Finally, there was good agreement on responsibilities regarding starting and dosing insulin. In contrast to what was practised in the original studies on IIT, the responsibility of ICU nurses varied widely across ICUs in the Netherlands.

Figure 1. *Responses to itemised statements on IIT (A: general statements; B: effects of IIT; C: implementation aspects; D: risks of hypoglycaemia; E: statements on responsibilities surrounding IIT). Respondents were asked to respond on a visual analogue scale (VAS), ranging from 1 for complete disagreement, to 10 for complete agreement. Data are means + 95% confidence intervals. IIT = intensive insulin therapy; LOS = length of stay; MV = mechanical ventilation; SH = severe hypoglycaemia; 60 (or 30) minutes no consequences: severe hypoglycaemia <60 (or 30) minutes is without clinical consequences.*



DISCUSSION

In this survey into blood glucose control and IIT we showed fair but incomplete implementation of IIT in ICUs in the Netherlands. Indeed, while approximately half of the ICUs mentioned that they used IIT and a fast majority stated that they had a guideline on glucose control, insulin was started at much higher blood glucose values than in the original studies by Van den Berghe *et al.*[1,2] Also, in contrast to the studies from Leuven, insulin was started and its dose adjusted by both ICU physicians and ICU nurses, rather than by ICU nurses alone. This survey demonstrates the need for a clear (inter)nationally accepted guideline on blood glucose control.

Implementation of a guideline is a complex process, involving numerous successive steps.[14,15] One of the first steps is an environmental scan, such as (regional and/or local) surveys into current practice, behaviour and potential hurdles (including concerns and (mis)beliefs) for adopting a certain guideline. Surveys also allow insight into translational errors of complex guidelines. We demonstrated several differences between guidelines on blood glucose control and the guideline as used by Van den Berghe *et al.*[1,2] The guideline in Leuven combined a simple set of rules on blood glucose control, next to targeting at tight blood glucose levels of 4.4 to 6.1 mmol/l: a) start of insulin is advised even with low blood glucose levels, even when within targets (thus at fairly low blood glucose levels); b) initiation and dose adjustment of insulin is done solely by ICU nurses (and never by ICU physicians, who are in fact banned from blood glucose control in Leuven); c) blood glucose measurements are to be performed at predefined time points (i.e., every four hours, but measurements can be done in between if needed; the decision is left to the discretion of the attending ICU nurse), d) although

severe hypoglycaemia is clearly stated as a potentially dangerous side effect, (mild) hypoglycaemia is not a reason for stopping insulin and infusing glucose, but a reason to be more careful and take more frequent blood glucose measurements to adjust the dose of insulin when the risk of severe hypoglycaemia increases or persists. The present survey clearly shows that these aspects are not translated into the currently used guidelines on blood glucose control. Although it is difficult, if not impossible, to determine whether all the above-mentioned aspects add to the success of IIT in the two original studies on IIT,[1,2] it is our belief that if we want to implement true IIT we should implement what was practised in these two positive studies. The results from this survey, therefore, have been used in an implementation project aiming for complete implementation of IIT in four hospitals in the Netherlands, which we will report on after completion of the project.[16]

The benefit of IIT in adult critically ill patients has been questioned recently.[3,4,6] Indeed, three recent randomised controlled trials did not confirm the beneficial effects of IIT.[3-5] It must be mentioned, however, that in all three trials blood glucose control in the intervention group was less tight than in the original studies in Leuven.[1,2] Also, compared with the control groups of the two original studies there was improved blood glucose control in the control groups, further decreasing the contrast between the study arms of these three negative studies. In addition, the first two confirmation studies may have been (severely) underpowered, one study due to the fact that it was stopped prematurely because the safety board considered the higher incidence of severe hypoglycaemia to be significant and dangerous.[3] A meta-analysis of randomised controlled trials by Wiener *et al.* showed that hospital mortality did not differ between blood glucose control and usual care overall; also, mortality was not influenced when stratified by blood glucose targets or ICU settings.[6] However, in our view Wiener *et al.* incorrectly meta-analysed the results from all the studies, including those in which IIT was said to be practised but actually not achieved.[3,4] The most recent meta-analysis on IIT by Griesdale *et al.*[17] showed that particularly surgical patients may benefit from IIT by lower mortality (RR=0.63; CI=0.44 to 0.91), confirming the original findings by Van den Berghe. The difference with the meta-analysis of Wiener *et al.* is predominantly explained by the inclusion of a recent Chinese study performed in a surgical ICU.[18] One recent paediatric study adds to the evidence on the benefit of IIT in critically ill subjects.[19] This randomised controlled trial showed IIT to improve short-term outcome of patients in a paediatric ICU. Of note, in this study exactly the same guideline, though with different (age-adjusted, lower) blood glucose targets, was used as in the two former studies from Leuven.[1,2]

It seems that blood glucose control is not a completely nurse-driven strategy in many ICUs, in contrast to what is practised in Leuven. Indeed, starting insulin as well as making dose adjustments were reported to be done by both ICU physicians and nurses. This may be a misconception: in particular the continuous presence of ICU nurses at the bedside may prevent deterioration of glucose control. For instance, changes in feeding, the most important cause of severe hypoglycaemia with IIT,[20] are recognised earlier by ICU nurses allowing them to adjust the insulin dose more swiftly. Similarly, giving full control of insulin dosing to those carers, who are constantly present (i.e., ICU nurses), allows shorter durations of both hyperglycaemia and hypoglycaemia.

Our survey suggests concern about severe hypoglycaemia is one reason to accept higher blood glucose values, which is a frequently mentioned barrier to implementation of IIT.[21,22] It seems contradictory that the respondents indicated IIT in itself to be safe, but when specifically asked for their opinion pertaining to severe hypoglycaemia, their replies indicated concerns about a higher occurrence and potential safety issues. This is an interesting contradiction, but might be explained by the fact that the respondents did not directly link the occurrence of severe hypoglycaemia to IIT per se. Indeed, opinion leaders sturdily point to the high incidence of (severe) hypoglycaemia as (one) reason not to aim for normoglycaemia.[7-10] Moreover, several large trials have even been stopped due to a high incidence of (severe) hypoglycaemia although predefined endpoints had not yet been reached.[3,23] Consequently, implementation of IIT is far from complete, and frequently local guidelines still accept higher blood glucose levels than those accepted in the original studies on IIT.[1,2] Severe and prolonged hypoglycaemia can indeed cause complications and mortality.[24,25] Although hypoglycaemia occurs more often in patients who are most severely ill and have a long stay on the ICU, this association does not suffice to conclude that severe hypoglycaemia actually causes death. Solid evidence for a causal relationship between short-lasting IIT-induced severe hypoglycaemia in the ICU setting and risk of death is lacking. A retrospective nested case-control study that was carefully matched for type and severity of illness as well as duration of ICU stay and thus for exposure time to insulin infusions, however, suggested no causal relationship between severe hypoglycaemia and mortality.[26] Moreover, experimental data showed that glucose reperfusion, rather than hypoglycaemia itself, is the cause of neuronal damage.[27]

Results from our survey are different from results from three surveys in Canada,[28] the United Kingdom[21] and Australia/New Zealand.[22] First, we found that insulin is started at lower blood glucose levels. Indeed, McMullin *et al.* reported thresholds for hyperglycaemia to be remarkably high: the median threshold was 10 mmol/l (IQR 9 to 11 mmol/l), with ICU nurses acting on 0.5 mmol/l higher blood glucose levels.[28] Of interest, in

the survey by McMullin *et al*. blood glucose control was judged not to be important for surgical patients, the targeted patients in the first study on IIT in ICU patients by Van den Berghe *et al*. Our survey showed that ICUs practise blood glucose control or IIT in all patient groups, irrespective of the referring speciality. Second, the level of IIT implementation is higher than in the UK and Australia/New Zealand. Mackenzie *et al*.[21] reported that only 25% of ICUs aimed for blood glucose levels similar to those used in the studies by Van den Berghe *et al*. Mitchell *et al*. also found that only a few ICUs have adopted blood glucose control.[22] The majority of the ICU nurses in the UK (82%) reported having concerns regarding severe hypoglycaemia in the patients receiving blood glucose control, although a clear reasoning for these feelings was lacking.[21] In the survey in Australia/New Zealand, reasons for not implementing IIT were also concerns about the risk of severe hypoglycaemia, but also doubts about the external validation of the original study by Van den Berghe *et al*.[22] Our results are, at least in part, in line with a recent survey by Hishberg *et al*. on stated blood glucose control practice in North American ICUs.[29] In this survey, 83% of adult ICU physicians preferred a target blood glucose level between 4.4 and 6.1 mmol/l, which is even higher than in our survey. In the North American survey many ICU clinicians (60%) mentioned hypoglycaemia to be more dangerous than hyperglycaemia, which seems in line with opinions in our survey.

Several limitations to our survey should be mentioned. Most important, the response from participating ICUs could be either by ICU physicians and/or nurses. However, responses from different care providers in centres from which we received both a response from an ICU physician and an ICU nurse were not different. Second, although the response rate is very high (78%), institutions that did not respond to the survey could potentially be less likely to be convinced of the benefits of IIT and/or less likely to practise IIT. This may have influenced the results, but we consider this unlikely because both academic and non-academic hospitals were broadly represented in the survey. Third, one questionnaire per participating centre may not be an adequate way of interpreting the standard of care pertaining to IIT in the participating centres. Nevertheless, we think that in general the responses were a reflection of department policies, because in an accompanying letter, the medical directors were specifically asked to reply in such a manner. Fourth, a survey only asks for current policy, and does not test whether a certain strategy is truly (and correctly) applied. For instance, one report on IIT in Finland showed that while it was implied that IIT was performed, the median blood glucose level of 6.2 mmol/l with 53% of blood glucose measurements above target suggested that implementation of IIT was rather 'loose'.[30] Finally, it can be questioned whether our findings are relevant to other countries.

In conclusion, many ICUs have adopted some form of blood glucose control, in half of the ICUs even with the tight blood glucose targets as used in the original studies on IIT. However, not all aspects of the original guideline, as used in Leuven, are fully appreciated. One reason for not implementing IIT seems concerns about severe hypoglycaemia, although it is questionable whether this fear is rational.

ACKNOWLEDGEMENTS

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CASE REPORT

Severe non-type-1 *Legionella pneumophila* infection without pneumonia

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ABSTRACT

We present a patient with myalgia and ongoing fever without respiratory symptoms caused by a *Legionella pneumophilia* infection. We conclude that in patients with fever of unknown origin legionellosis should be considered, even in the absence of pulmonary symptoms. When considering legionellosis, diagnostic tests should include the urinary antigen test.

KEYWORDS

Extrapulmonary legionellosis, fever of unknown origin, *Legionella pneumophila* non-type-1, *Legionella* urinary antigen test

INTRODUCTION

Legionella is considered a relatively common cause of pneumonia. The clinical picture ranges from an influenza-like syndrome known as Pontiac fever, to a severe pneumonia requiring mechanical ventilation.[1] In legionnellosis, extrapulmonary manifestation in the absence of pneumonia is not common. To our knowledge, there are no reports in the literature of *Legionella* as the explanation for fever of unknown origin. We present a patient referred because of myalgia and ongoing fever without respiratory symptoms. Since the *Legionella* urinary antigen test can be falsely negative in 10% of infections, serotyping should be performed and repeated.

CASE REPORT

A 56-year old man was admitted for evaluation of fever, myalgia, fatigue and 10 kg weight loss in four weeks. Initially he had experienced headaches and transient coughing. Fever began ten days before admission. In the week preceding admission his general practitioner had prescribed amoxicillin/clavulanate and clarithromycin. His medical history was unremarkable. The patient had travelled as an IT professional to Egypt and Jordan 14 weeks before referral. On physical examination the only remarkable finding was a rectal temperature of 38.5° Celsius. Laboratory results showed an elevated erythrocyte sedimentation rate, mild leucocytosis, elevated C-reactive protein and abnormal liver function tests (*table 1*). Both chest X-ray and computed tomography (CT) scan were normal and did not reveal pulmonary infiltrates. Abdominal ultrasound and CT scan showed neither lymphomas nor a liver abscess. The differential diagnosis was infection, lymphoreticular disease and autoimmune disorders. Extensive diagnostic tests did not reveal a cause: tests were performed to detect viral hepatitis, cytomegalovirus, Epstein-Barr virus, respiratory syncytial virus, parainfluenza, human immunodeficiency virus, dengue, rickettsioses, *Mycoplasma pneumoniae*, *Chlamydia psittaci*, *Coxiella burnetii*, *Brucella*, malaria, lues, leishmaniasis, schistosomiasis and leptospirosis. Biopsy of the temporal artery showed no signs of vasculitis. Bone marrow aspiration revealed normal trilinear haematopoiesis, whereas cultures remained negative, as did staining and culture for mycobacteria. Serology for *Legionella*, a specimen taken one day after admission, revealed no positive ELISA titres. The urinary *Legionella* antigen test was not performed on admission. Despite empirical treatment with intravenous amoxicillin/clavulanate and nonsteroid-anti-inflammatory drugs, his fever and myalgia persisted. Two weeks after admission, seroconversion for *Legionella* IgM from 6 E/ml to 254 E/ml was demonstrated. Serotyping revealed *L. pneumophila* non-type-1 infection (possibly type 5 with an elevated titre of 1:256, *table 2*). Intravenous ciprofloxacin and rifampicin were administered (400 mg and 600 mg twice

Table 1. *Laboratory values*

	Unit	Admission	Reference range
Erythrocyte sedimentation rate	mm/u	99	0-15
Haemoglobin	mmol/l	8.9	8.5-11.0
Leucocytes	10.9/l	12.3	4.3-10.0
Thrombocytes	10.9/l	676	150-400
Bilirubin, total	µmol/l	20	0-17
Aspartate	U/l	118	0-37
Alanine	U/l	187	0-41
Lactate dehydrogenase	U/l	423	0-450
Alkaline phosphatase	U/l	731	25-120
Gamma-glutamyltransferase	U/l	244	0-50
Creatine kinase	U/l	<20	0-200
C-reactive protein	mg/l	152	0-10

Table 2. *Legionella pneumophila subroup titres*

	Serum titre
L. pneumophila 1	Negative
L. pneumophila 2	1:128
L. pneumophila 3	1:64
L. pneumophila 4	1:64
L. pneumophila 5	1:256
L. pneumophila 6	1:64
L. pneumophila polyvalent	Negative

daily respectively). The patient recovered well. On inquiry, he had installed a steam shower at home eight weeks before admission. Afterwards the cultures from this shower for *Legionella* spp. remained negative.

DISCUSSION

In adults with legionellosis, systemic manifestations besides pneumonia are often reported. The absence of pulmonary symptoms though is rare. Therefore in our patient, an important clue to the clinical diagnosis was missing: his chest X-ray was normal. In a prospective study, Tan *et al*. describe chest X-ray findings in 43 patients with a diagnosis of community-acquired pneumonia due to legionellosis.[2] In 40 of the 43 patients, admission radiographs were compatible with pneumonia, and in three patients admission radiographs were normal. Atypical presentation of legionellosis is described in a Japanese study from 2002.[3] Clinical features and CT-scan findings are described in an outbreak of *Legionella* pneumonia in eight patients. All patients were febrile; however respiratory symptoms were observed in only four. Chest CT scan, though, showed abnormalities in all eight patients (ground glass opacity, consolidation, pleural effusion). Although rare, extrapulmonary symptoms may be the presenting features of legionellosis: endocarditis, arthritis and liver function disturbances have all been described in the absence of pneumonia.[4,5] Lowry *et al*. reported 22 patients with extrapulmonary legionellosis, five (23%) of whom died.[6] Transient nonproductive coughing was one of patient's initial complaints (the other was headache). Protracted upper respiratory tract infection can be the explanation of his symptoms. Lieberman *et al*. found the same pathogens in upper and lower respiratory tract infection (RTI).[7] This serological study challenges the distinction between upper and lower RTI. Patients with upper RTI are usually not admitted and general practitioners do not perform serology for self-limiting diseases such as upper RTI: the true prevalence of *L. pneumophila* upper RTI is not known. The patient did not recover until combination therapy for *Legionella* was prescribed. Dutch Working Party on Antibiotic Policy (SWAB) guidelines advise treatment with macrolides or fluorochinolones for proven *Legionella* infection.[8] The general practitioner had already prescribed clarithromycin. After seroconversion the patient was treated with ciprofloxacillin, but fever did not resolve completely. De Vries *et al*. suggest combination therapy, for example rifampicin, in case of failing monotherapy.[9] We added rifampicin, which has a good sensitivity for *Legionella*. Both the patient's travelling and his installing a steam shower are well-known causes of legionellosis. Since the incubation period of *Legionella* is usually less than three weeks, his visit to the subtropics 14 weeks before admission seems a less likely cause given the time frame. *Legionella* is an intracellular Gram-negative bacterium, of which *Legionella (L.) pneumophila* type 1 is responsible for approximately 90% of *Legionella* infections in the Netherlands.[10] The urinary antigen test only detects type 1. Therefore, when legionellosis is suspected, serology should be performed and repeated in case of a negative urinary antigen test. We found no evidence in literature for a predilection for extrapulmonary sites for non-type 1 infections.

CONCLUSION

Legionella pneumophila type 1 is responsible for the majority of cases of legionellosis in the Netherlands. Pulmonary symptoms are key findings in this diagnosis. We report a case of severe non-type-1 *Legionella pneumophila* infection manifesting itself as ongoing fever, but without pneumonia. We conclude that in patients with fever of unknown origin legionellosis should be considered, even without pulmonary symptoms. Furthermore, when considering legionellosis, diagnostic tests should include the urinary antigen test. In approximately 10% of legionellosis though, this test is false-negative, due to one of the more seldom reported non-type-1 subgroups. Serology should be performed and repeated.

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CASE REPORT

Vinblastine, rituximab and HAART, treatment of an HIV-positive patient with multicentric Castleman's disease

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ABSTRACT

An HIV-positive man from Somalia presented with severe malaise, weight loss, relapsing fever, lymphadenopathy and splenomegaly. An FDG-PET-scan-guided lymph node biopsy revealed the characteristic histological features of the plasma cell variant of Castleman's disease. A high HHV-8 viral load was detected in the serum (7980 copies/ml). Treatment with HAART, rituximab and vinblastine resulted in a full and rapid recovery and lowered HHV-8 viral load to undetectable levels.

KEYWORDS

Castleman's disease, HAART, HHV-8, rituximab, vinblastine

INTRODUCTION

A well-known benign lymphoproliferative disease (LPD) is infectious mononucleosis, induced by the Epstein-Barr virus (EBV) of the γ-herpes family. Another γ-herpes virus is human herpes virus 8 (HHV-8), one of the causes of a much rarer LPD, known as multicentric Castleman's disease (MCD). Both viruses have also been linked to lymphoproliferative disease and lymphoma. Especially immunocompromised hosts such as post-transplantation and HIV-positive patients are susceptible to uncontrolled infection with these viruses and their associated diseases;[1,2] however, MCD also occurs in HIV-negative patients.

Here we describe the case of an HIV-positive man from Somalia with MCD who responded well to treatment with HAART, rituximab and vinblastine. In addition we will give a brief review of the literature. The unicentric hyaline vascular, the unicentric plasma cell and the not otherwise specified variant occur in a different population and are rarely or not related to HHV-8. This case report concerns only the HHV-8-associated multicentric plasma cell and plasmablastic variants.

CASE

A 49-year-old man from Somalia had been HIV-positive since 2004. Because he had always been asymptomatic, and his CD4 count had never dropped below 400 cells/mm^3, antiretroviral therapy had not been initiated.

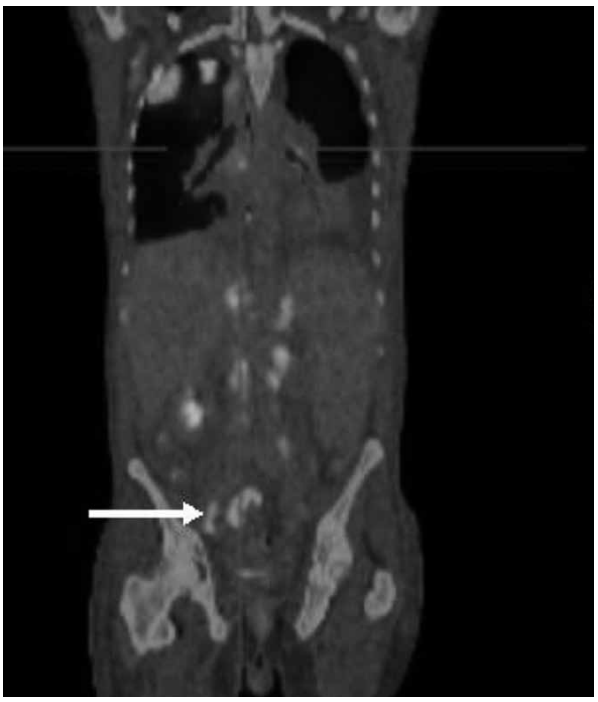
The patient presented with a solitary enlarged lymph node in the neck and marked hepatosplenomegaly. Laboratory examination revealed a normocytic anaemia (haemoglobin 3.7 mmol/l) and a declining CD4 count (290/mm^3).

The differential diagnosis included (opportunistic) infections such as tuberculosis but also malignant lymphoma. A computed tomography (CT) scan of the neck, chest and abdomen revealed pleural effusion, extensive lymphadenopathy and hepatosplenomegaly, but no pulmonary infiltrates or tumours. The Mantoux test proved negative and a biopsy of an enlarged lymph node in the neck displayed no signs of lymphoma, tuberculosis or other opportunistic infection. A bone marrow aspirate revealed an increased number of polyclonal plasma cells, but no signs of other pathology.

Within five weeks the patient's condition deteriorated with a non-productive cough, relapsing fever and severe weight loss (10 kg). The laboratory results revealed a Coombs-positive normocytic anaemia (haemoglobin 4.9 mmol/l), without clinical haemolysis. Also, an elevated C-reactive protein was detected (186 mg/ml). Lactate dehydrogenase was not increased with 204 U/l.

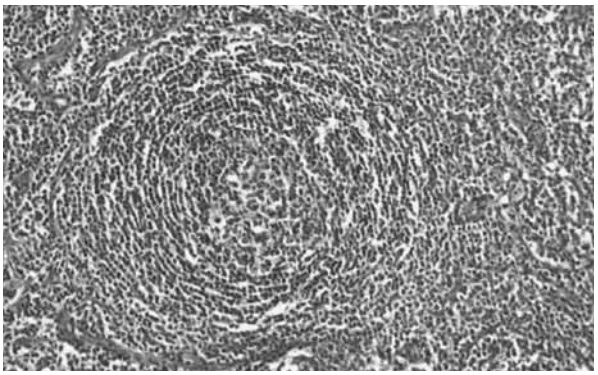
Cytology of the pleural effusion revealed unspecific reactive cells. A second biopsy of an axillary lymph node displayed follicular hyperplasia only. An FDG-PET-scan revealed FDG-PET positive lesions, among which the right axillary lymph nodes (possibly an effect of the second biopsy), and lymph nodes in the right inguinal region (*figure 1A*).
The FDG-PET positive inguinal lymph node was extracted. It showed the characteristic features of the plasma cell variant of Castleman's disease, with hyperplastic follicles surrounded by a concentric mantle of plasma cells (*figure 1B*). The serum load of HHV-8 was 7980 copies/ml. Serum IL-6 was 28 pg/ml (in other HIV-positive MCD patients, IL-6 levels above 4500 pg/ml have been reported).[3]

Figure 1A. *Pet scan*



The PET-scan revealed several hot-spots. The arrow demarks the location of the lymph node that displayed the characteristic features of Castleman's disease.

Figure 1B. *A lymphoid follicle surrounded by a concentric mantle of plasma cells, as seen in the plasma cell variant of Castleman's disease*



Treatment with antiretroviral therapy was started together with a regime containing both vinblastine (6 mg/m^2 three times, once every other week) and rituximab (375 mg/m^2, four times weekly). Within two weeks, after the start of therapy, the patient started to recover; he regained 10 kg of weight and his temperature normalised. Four months after treatment, the HHV-8 viral load was undetectable (<150 copies/ml).

EPIDEMIOLOGY

While the exact epidemiology remains unknown, HHV-8-associated MCD is rare. Despite its relation to immunodeficiency, several studies demonstrate a low CD4 count to be only a minor risk factor.[4,5]

CLINICAL PRESENTATION

Patients generally present with waxing and waning systemic symptoms, such as malaise, fever, weight loss, lymphadenopathy, hepatosplenomegaly and skin rash. HHV-8 is related to Kaposi sarcoma, which frequently occur in MCD patients. Rarely MCD patients present with pemphigus. Cytopenias, raised lactate dehydrogenase and C-reactive protein, as well as increased liver enzymes and hypergamma-globulinaemia are generally found.[6,7] Often, autoimmune anaemia is observed. Bone marrow examination may reveal significant plasmocytosis. Seldom, patients present with a haemophagocytic syndrome.

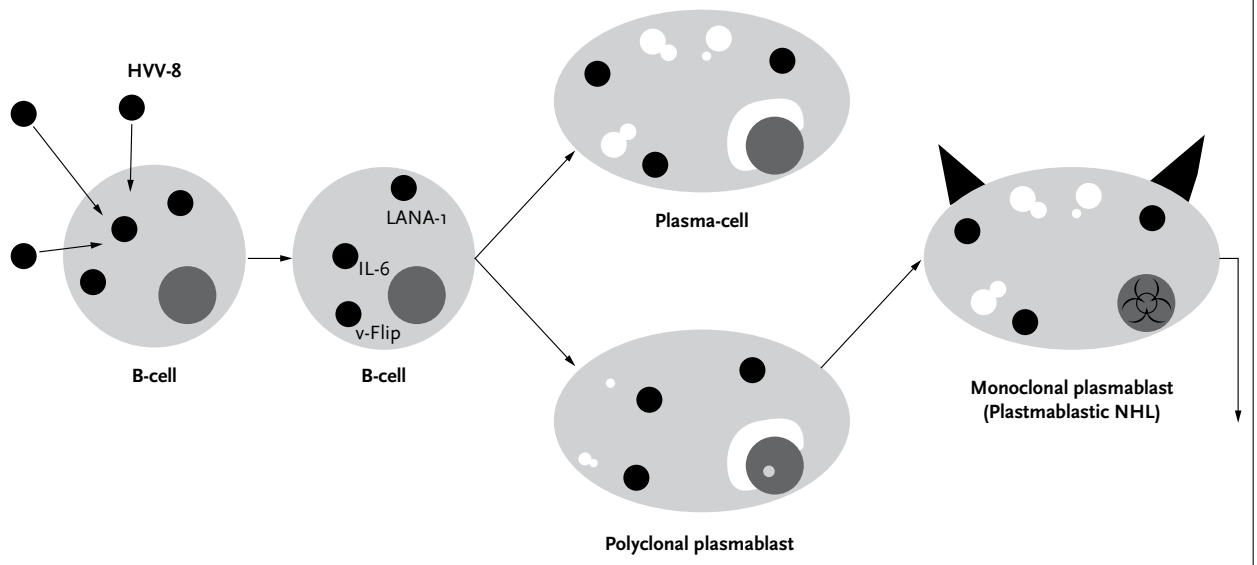
HUMAN HERPES VIRUS 8

The aetiology of plasma cell and plasmablastic MCD involves HHV-8.[8] The virus resides in B-cells, endothelial and epithelial cells, and produces a protein strongly analogous to interleukin 6 (IL-6). When a high dose of IL-6 is infused in mice, plasmocytosis, an expansion of B-cells and organomegaly develop.[9] HHV-8 produces other proteins: v-FLIP, which has antiapoptotic properties and LANA-1, which makes B-cells skip vital steps in the differentiation towards plasma cells.[10,11]

PROGNOSIS

Even though the B-cell proliferation is initially polyclonal, the clinical course of MCD is severe with a high mortality. Most patients eventually die of a SIRS-like syndrome with multi-organ failure.[6,7,12] Chronic proliferation and deviant differentiation of B-cells eventually induce monoclonal plasmablastic non-Hodgkin lymphoma (*figure 2*).

Figure 2. *HHV-8 infects B-cells. Pirating the cell's replication mechanism, it produces several proteins among which IL-6, v-Flip and LANA-1. IL-6 stimulates B-cell proliferation and differentiation into plasma cells. V-Flip and LANA-1 may induce aberrant deviation into plasmablasts, which might eventually lead to the development of a non-Hodgkin lymphoma*



Non-Hodgkin lymphoma has been reported to occur in an estimated 20% of HHV-8-associated MCD patients.[13]

TREATMENT

Evidence is based on small series and case reports; therefore, no standard therapy is defined. Few clinical results on HHV-8-targeting antiviral therapy have been published, but ganciclovir and cidofovir seem to inhibit HHV-8 replication both *in vitro* and *in vivo*.[14,15] Also splenectomy may improve clinical condition and blood count transiently, and could be considered an adjunct to systemic treatment.[11] Recently a case report described complete and lasting remission in an HIV-negative MCD patient after splenectomy without any systemic therapy.[16] Mortality rates among HIV-positive MCD patients were higher in the pre-HAART era than in the HAART era.[5] In a small series of immunodeficient HIV-positive MCD patients, initiation of HAART therapy initially worsened symptoms, possibly related to an immune reconstitution syndrome.[17]

Due to the aggressive nature of MCD and the high risk of subsequent non-Hodgkin lymphoma, chemotherapy is indicated. Moreover, vinblastine and etoposide are known to effectively suppress symptoms.[12,18] When such treatment is stopped, however, MCD generally relapses rapidly. More-toxic CHOP chemotherapy may be effective as well, also in the long run.[19] It must be noted that such intensive chemotherapy is often poorly tolerated by these very ill patients.

Another approach is the use of rituximab, a monoclonal antibody, directed against the CD20 protein on B-cells and plasmablasts. By inducing antibody-dependent cell-mediated cytotoxicity, complement-dependent cytotoxicity and apoptosis, rituximab effectively eliminates almost the entire B-cell population.[20]

The two largest studies (n=21 and n=24) on rituximab in the treatment of HIV-positive, HHV-8-associated MCD patients showed promising results in lowering HHV-8 viral load,[21] with disease-free survival rates of 71% after one year in the first study[22] and 95% after two years in the second study.[21] A small number of patients died of progressive MCD, an effect possibly attributable to sudden increases in HHV-8 viral load shortly after administration. Also mild exacerbations of Kaposi sarcoma were reported in both trials during treatment. These results have not been compared in a clinical trial with mild or intensive chemotherapy; however, the results in pretreated patients seem to be better than the results of chemotherapy alone.

The mechanism underlying the beneficial effect of rituximab possibly relates to a reservoir function of B-cells and plasmablasts, essential for the survival and proliferation of HHV-8. What contradicts this is that not all plasmablasts express CD20 and that HHV-8 is also known to reside in epithelial and endothelial cells.

DISCUSSION

Bone marrow or lymph nodes will not always display signs characteristic of MCD. In this case, an FDG-PET scan helped out and ultimately revealed a lymph node of diagnostic value.

When comparing the different treatment modalities available to HHV-8-associated MCD in HIV patients, anti(retro)viral therapy, splenectomy, chemotherapy and/or immunotherapy may be used. The case for chemotherapy is clear, however, intensive chemotherapy is often too toxic for the patient's generally poor clinical condition. Moreover, relatively mild chemotherapy alone induces acceptable response rates and seems to control symptoms. The results of immunotherapy with rituximab are impressive: monotherapy induces long-lasting response after relapse on chemotherapy. The combination of rituximab with mild chemotherapy seems to be effective as a first-line therapy. Because this combination is a relatively nontoxic treatment with favourable results, it seems to be a reasonable first choice.

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PHOTO QUIZ

Abnormal chest X-ray in a patient with mononeuritis multiplex

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CASE REPORT

A 74-year-old man with a medical history of hypertension, polymyalgia rheumatica (PMR) and steroid-induced diabetes mellitus, visited our outpatient clinic in 2000 (at the age of 65) with mononeuritis multiplex of the legs. Laboratory analysis showed elevated inflammation parameters (erythrocyte sedimentation rate (ESR) 118 mm after one hour, C-reactive protein 99 mg/l) and normocytic anaemia (haemoglobin 6.4 mmol/l). Chest X-ray was normal besides an elongated aorta. Later chest X-rays, from 2004 onwards, were abnormal (*figures 1* and *2*).

WHAT IS YOUR DIAGNOSIS?

See page 92 for the answer to this photo quiz.

Figure 1. *Chest X-ray of the patient in 2004*

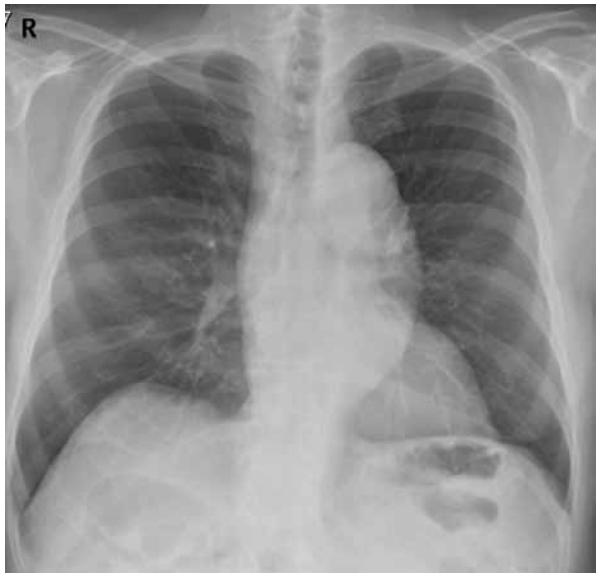
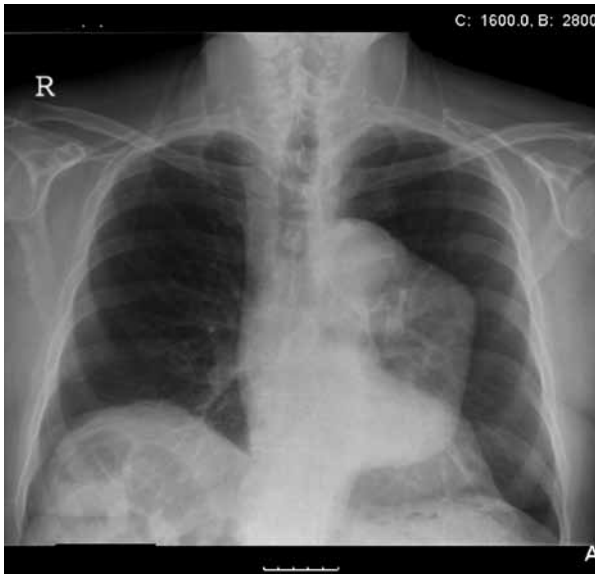


Figure 2. *Chest X-ray in 2008*



ANSWER TO PHOTO QUIZ (PAGE 91)

ABNORMAL CHEST X-RAY IN A PATIENT WITH MONONEURITIS MULTIPLEX

DIAGNOSIS

Biopsy of the sural nerve showed vasculitis and axonal degeneration. Biopsy of the temporal artery showed disruption of the lamina elastica, indicating arteritis. The diagnosis giant cell arteritis (GCA) was made and treatment with high-dose prednisolone was started. In 2004, computed tomography scan of the abdomen revealed an aneurysm of the thoracic aorta, maximum diameter 41 mm. The aneurysm was followed up yearly and treated by endovascular surgery in 2008 (maximum diameter 56 mm).

GCA typically occurs in patients older than 50 years, who present with new headache and increased ESR. Other symptoms of vasculitis may occur, as in this case mononeuritis multiplex of the legs. GCA can occur as a separate disease, or in a spectrum with PMR. Temporal artery biopsy is the gold standard for diagnosis. It is important to keep in mind that the disease affects large- and medium-sized arteries, especially the proximal aorta and its branches. Aneurysms of the thoracic aorta can develop years after onset of the disease as a late complication, and are 17 times more common in patients with giant-cell arteritis compared with the normal population.[1] The optimal frequency and imaging modality for monitoring and follow-up of aortic aneurysms in GCA have yet to be determined. Bongartz and Matteson suggest yearly abdominal ultrasound, chest X-ray and transthoracic echo. However, for 'high-risk patients', they suggest CT or MRI angiography at the time of diagnosis and after one year, followed by yearly abdominal ultrasound, chest X-ray and transthoracic echo.[2] The authors have defined high-risk patients as those with aortic insufficiency murmur, or PMR with ESR >100 mm/1h, or at least two of the following: hypertension, hyperlipidaemia, PMR, coronary artery disease.

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PHOTO QUIZ

Acute abdomen after deceleration trauma

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CASE REPORT

An 86-year-old female presented at the emergency room with abdominal pain, nausea and vomiting. She had been without symptoms until she fell on her buttocks one hour before. The pain gradually increased with time. She had no fever, diarrhoea or blood loss.

Her medical history revealed a hysterectomy and several fall accidents. Because of constipation she used macrogol if necessary.

On examination she was an ill-looking lady in obvious pain. Pulse rate, temperature and blood pressure were within the normal range. There was no audible peristalsis over her abdomen, all regions were extremely painful with guarding and rebound tenderness. Abdominal pain precluded rectal digital examination and optimal examination of the lower extremities.

Laboratory results showed normal values for C-reactive protein (CRP), erythrocyte sedimentation rate (ESR) and leucocytes. Serum amylase was slightly elevated (183 U/l, upper normal limit (UNL) is 50 U/l), lactate was 2.4 mmol/l (UNL 2.2 mmol/l) and glucose 11.2 mmol/l (UNL is 6.4). The urine sample revealed no abnormalities.

Plain radiography showed no abnormalities of the chest and abdomen. No fractures were evident on skeletal radiography of lumbar spine and pelvis. With a working diagnosis of acute abdomen after a deceleration trauma computed tomography of the abdomen was performed (*figures 1 and 2*).

Figure 1. *CT scan of the abdomen*

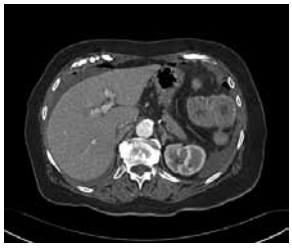
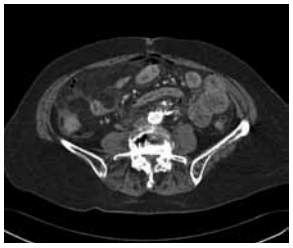


Figure 2. *CT scan of the abdomen*



WHAT IS YOUR DIAGNOSIS?

See page 94 for the answer to this photo quiz.

ANSWER TO PHOTO QUIZ (PAGE 93)

ACUTE ABDOMEN AFTER DECELERATION TRAUMA

On computed tomography accumulation of fluid and extraluminal air were seen. In the right lower quadrant obliteration of mesenteric fat was recognised. Because of our patients older age and history of constipation a perforated diverticulum was considered as a possible diagnosis. On laparotomy extensive adhesions were found in the lower abdomen, as well as a small incisional hernia after the previous hysterectomy with perforation of the ileum at the edge of the hernia. Segmental resection of the perforated part of the small bowel was performed followed by anastomosis. Postoperatively, antibiotic treatment was initiated. Unfortunately our patient died 11 days after laparotomy due to postoperative complications.

Traumatic bowel perforation can be caused by penetrating trauma, including accidental perforation during laparoscopy or endoscopy. Blunt abdominal trauma, occasionally related to seat belts in motor vehicle accidents, can lead to small bowel perforation as well. In patients with blunt abdominal trauma bowel injury occurs in 1%, leading to perforation in approximately 40% of these patients. The jejunum and ileum are involved most frequently.[1] In our patient perforation was related to a deceleration trauma. Probably her older age and impaired intestinal mobility due to herniation and adhesions made her vulnerable to small bowel perforation. Deceleration trauma has previously been described as a possible cause of small bowel perforation.[2]

Traumatic small bowel perforation is an uncommon injury and diagnosis can be laborious.[3] Diagnostic delay has a marked effect on mortality.[4] A multi-modal approach is mandatory for an accurate diagnosis: the reconstruction of the mechanism of injury, serial physical examination, laboratory data (especially white blood cells and amylase), diagnostic abdominal lavage and appropriately selected imaging modalities. Despite improvements in diagnostic workup traumatic bowel perforation still has a mortality of 10.6%.[4]

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PHOTO QUIZ

Blindness, confusion and seizures in a cancer patient

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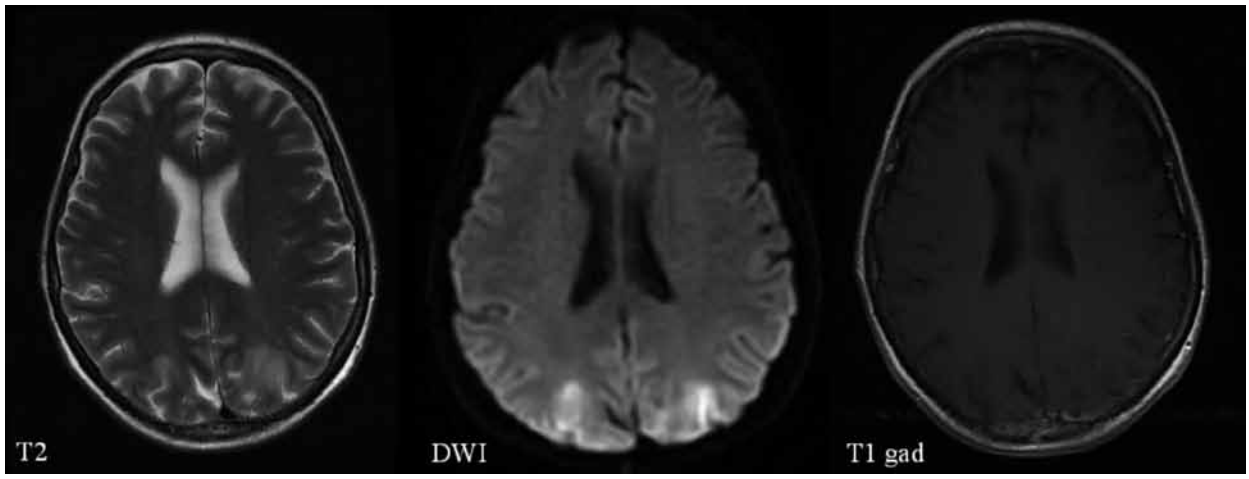
CASE REPORT

A 58-year-old woman with metastatic adenocarcinoma of unknown primary developed headache, confusion and bilateral cortical blindness of acute onset. This was followed shortly by two episodes of generalised seizures. She last received chemotherapeutic treatment with epirubicin, oxaliplatin and capecitabine two months previously. She had no other significant past medical history and was not on any medications. Blood pressure was within normal limits and her laboratory tests were unremarkable. A magnetic resonance imaging of her brain was performed (*figure 1*).

WHAT IS YOUR DIAGNOSIS ?

See page 96 for the answer to this photo quiz.

Figure 1. *Magnetic resonance imaging*



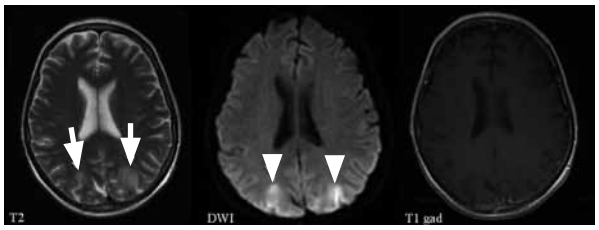
DIAGNOSIS

From the clinical presentation, the differential diagnosis includes cerebral metastases, malignant leptomeningeal involvement and posterior reversible encephalopathy syndrome (PRES). The magnetic resonance imaging showed high signal intensity on T2-weighted images (T2) affecting the cortex and subcortical white matter of both occipital lobes symmetrically (*figure 2,* white arrows). Corresponding hyperintensities were seen on diffusion-weighted images (DWI) suspicious for cerebral ischaemia (*figure 2,* white arrowheads). There was no enhancement on gadolinum-enhanced T1-weighted images (T1 gad) to suggest metastatic disease. The clinical and imaging findings were highly suggestive of PRES with possible cerebral ischaemia. She was treated with supportive care with anticonvulsants and her symptoms completely resolved in 12 days.

PRES can occur in patients with eclampsia, uncontrolled hypertension with resultant encephalopathy, renal impairment and those receiving immunosuppressive treatments such as ciclosporin and tacrolimus. Previous exposure to chemotherapeutic agents has also been implicated.[1] The pathophysiology of PRES is not well understood but is believed to be due to a paucity of sympathetic innervation within the posterior circulation, making it more prone to interruption of autoregulation with disruption of the blood brain barrier. This results in vasogenic oedema with hydrostatic leakage and interstitial fluid accumulation in the cortex and subcortical white matter. The clinical presentation of PRES is similar to that experienced by our patient.

PRES is commonly diagnosed on magnetic resonance imaging of the brain with cortical/subcortical vasogenic oedema affecting mainly the occipital and parietal lobes in a bilateral symmetrical fashion. This manifests as areas of high signal intensity on T2-weighted images. There is no restricted diffusion (no high signal intensity) on DWI with vasogenic oedema. DWI is thus helpful in distinguishing PRES from other conditions such as cerebral ischaemia and infarction, which result in cytotoxic oedema that manifests as restricted diffusion (high signal intensity).[2] Occasionally, as in our patient, concurrent ischaemia may complicate PRES with resultant cytotoxic oedema and hence restricted diffusion on DWI.[3] Despite this, our patient experienced complete resolution of symptoms. This reversibility together with the clinical presentation and imaging findings of lesions in a bilateral symmetrical posterior cerebral distribution is typical of PRES. Clinical recovery usually occurs in most patients within days.[4] Early recognition is essential as delayed diagnosis can result in complications such as infarction. Repeat imaging is recommended to document resolution of abnormalities.

Figure 2. *Magnetic resonance imaging*



There were areas of high signal intensity on the T2-weighted images affecting the cortex and subcortical white matter of both occipital lobes symmetrically (white arrows). Corresponding hyperintensities were seen on diffusion-weighted images suspicious for cerebral ischaemia (white arrowheads). There was no enhancement on gadolinum-enhanced T1-weighted images to suggest metastatic disease.

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PHOTO QUIZ

Oedema and Crohn's disease

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CASE REPORT

A 43-year-old man was admitted to the hospital for analysis and treatment of severe oedema. He had suffered from Crohn's disease since 1988, which had responded well to prednisolone in the past. However, he did not tolerate treatment with azathioprine and treatment with infliximab was unsuccessful. His Crohn's disease was not well controlled, due in part to the patient's poor follow-up, non-compliance and intolerance for medication, resulting in (peri)anal fistulas and frequent exacerbations. In 1993, the terminal ileum had to be resected.

Blood tests at admission showed a creatinine of 78 µmol/l (50-95 µmol/l), urea of 2.3 mmol/l (2.5-6.4 mmol/l), sodium of 147 mmol/l (135-145 mmol/l), potassium of 3.1 mmol/l (3.2-4.7 mmol/l) and albumin of 15 g/l (32-48 g/l). There was proteinuria of 5.28 g/24 h; in 1989 the urine dipstick was negative for protein. Liver tests were abnormal, with ASAT 216 U/l (11-35 U/l), ALAT 266 U/l (15-35 U/l), alkaline phosphatase 372 U/l (40-120 U/l), γGT 709 U/l (8-35 U/l) and bilirubin 13 µmol/l (5-19 µmol/l). The urinary sediment was acellular.

Our patient was admitted with nephrotic syndrome. Treatment with diuretics and ACE inhibition was started. In regard to the abnormal liver tests, we suspected primary sclerosing cholangitis (PSC). Both liver biopsy (*figure 1*) and kidney biopsy (*figure 2*) were performed.

Figure 1. *PAS staining of liver biopsy*

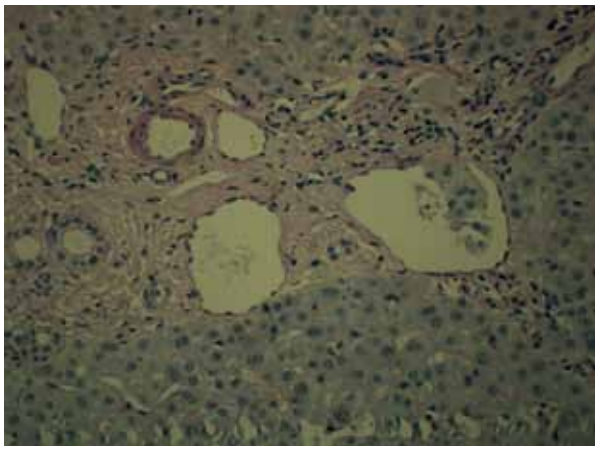
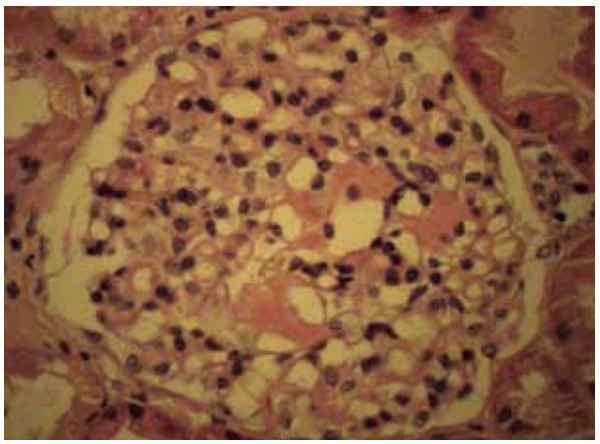


Figure 2. *PAS staining of kidney biopsy*



WHAT IS YOUR DIAGNOSIS?

See page 98 for the answer to this photo quiz.

ANSWER TO PHOTO QUIZ (PAGE 97)

OEDEMA AND CROHN'S DISEASE

DIAGNOSIS

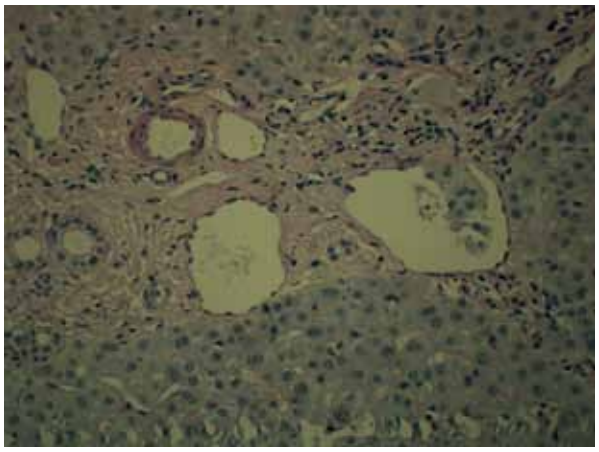
Liver biopsy showed no PSC but depositions of amyloid (*figure 1*). Kidney biopsy also showed depositions of amyloid (*figure 2*). CT imaging and colonoscopy showed no active Crohn's disease, but leucocyte scintigraphy showed activity in the anastomotic area of the ileocaecum. He was treated with prednisolone and 6-mercaptopurine. The oedema diminished, proteinuria declined to 4 g/24 h and the albumin increased to 20 g/l.

Chronic inflammatory disease can be complicated by systemic amyloidosis. Proteins accumulate in tissues as nonsoluble fibrils, resulting in progressive organ failure and eventually death.[1] In this reactive type of amyloidosis, the serum amyloid A protein (SAA) plays an important role. It is synthesised by hepatocytes under regulation of proinflammatory cytokines.[2] The fibrils are derived from these acute phase proteins via a process of cleavage, misfolding and aggregation.[1] The mean plasma concentration of SAA in healthy persons is 3 mg/l, while the concentration during an acute phase response can increase to more than 2000 mg/l.[3] Overproduction of SAA increases the risk of development of AA amyloidosis; it is unclear why this affects just a small group of patients with chronic inflammatory disease.[4,5] The kidney is the organ most affected.[6] Deposition of amyloid results in proteinuria and progressive loss of renal function. Other sites of deposition can be the digestive tract, the liver, the autonomic nervous system and the heart.[7] Treatment consists of removing the stimulus, in this case preventing the persistence of the acute phase response by effectively treating the inflammatory bowel disease.[8] No treatment has a direct effect on the development of SAA,[3] although a recent study showed treatment with eprodisate to be promising.[7]

The Crohn's disease of our patient was difficult to treat, partly due to erratic care and multiple intolerances. This resulted in ongoing inflammation. The end result is manifest AA amyloidosis, presenting as nephrotic syndrome. The SAA plasma concentration was not determined prior to the treatment with prednisolone and 6-mercaptopurine. It was normal during treatment, an indication of improved control of the Crohn's disease.

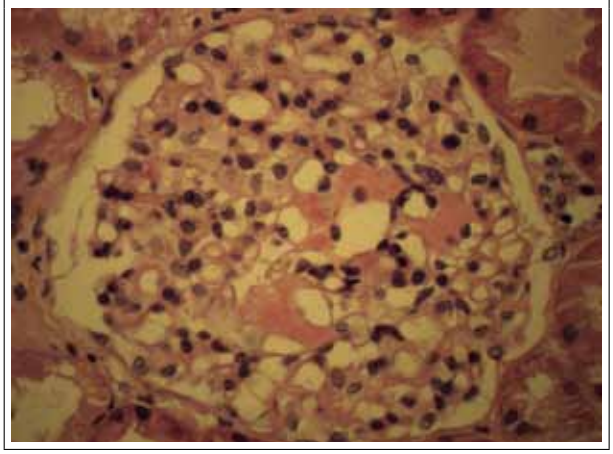
In case of nephrotic syndrome in patients with inflammatory bowel disease, amyloidosis as a rare complication should be considered.

Figure 1. *PAS staining of portal tract in liver biopsy*



The arterial wall shows amorphous depositions which stain positive in amyloid stains (Congo red, SAB, Thioflavine, immunoperoxidase for AA amyloid). There is no intrasinusoidal amyloid deposition.

Figure 2. *PAS staining of kidney biopsy, showing mesangial depositions of amyloid*



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INFORMATION FOR AUTHORS

Aims and scope

The *Netherlands Journal of Medicine* publishes papers in all relevant fields of internal medicine. In addition to reports of original clinical and experimental studies, reviews on topics of interest or importance, case reports, book reviews and letters to the editor are welcomed.

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Manuscripts submitted to the Journal should report original research not previously published or being considered for publication elsewhere. Submission of a manuscript to this Journal gives the publisher the right to publish the paper if it is accepted. Manuscripts may be edited to improve clarity and expression.

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2. Kaplan NM. Clinical Hypertension. 7th ed. Baltimore: Williams & Wilkins; 1998.
3. Powell LW, Isselbacher KJ. Hemochromatosis. In: Braunwald E, Fauci AS, Kasper DL, et al., editors. Harrison's Principles of Internal Medicine. 15th edition. New York: McGraw-Hill; 2001. p. 2257-61.

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