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EDITORIAL

Evidence based: to be or not to be?

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Evidence-based medicine (EBM) is advocated as the reference standard not only for practising physicians, but also for healthcare managers, politicians and insurance companies. Perhaps this is all very well, but if we put EBM in the heart of medicine, wouldn't it be convenient if we were to have perfect agreement on its definition? Unfortunately, this is not the case.

Boldly spoken, there are two, quite different conceptions of what EBM really stands for. One is that it represents a way of practising medicine in which clinical decisions are based on epidemiological evidence as exclusively as possible. Any medical intervention, either diagnostic or therapeutic, that is not supported by epidemiological evidence should ideally be avoided, and not be reimbursed. If evidence for a particular disease is scarce, extrapolation of epidemiological evidence from neighbouring domains is, if anything, preferable to not considering epidemiological findings at all. Only if evidence is completely lacking, and life is clearly at risk, are 'non-evidence-based' decisions occasionally acceptable.

The alternative approach to EBM is that findings from epidemiological studies are never (!) directly translated to individual patient care without explicitly taking into consideration 1) knowledge from other domains (such as basic sciences and pathobiology), 2) clinical experience and 3) the patient's individual context. From this perspective on EBM, consideration of epidemiological evidence is just one of several elements of clinical decision making, and no hierarchy between these elements is defined a priori. To practice this type of EBM, one needs a comprehensive set of skills on top of thorough knowledge of epidemiological evidence: understanding basic sciences and pathobiological principles, allowing previous clinical experience to compete with 'hard facts', and having an open attitude for individual patient context.

Considering these various ways of looking at EBM, what is the 'official definition'? One of the founding fathers of EBM, David Sackett, has been very clear about this. He stated that EBM '*integrates the best external evidence with individual clinical expertise and patients' choice*'. With respect to the sources of the external evidence that is referred to, Sackett emphasises that it encompasses '*clinically relevant research, often from the basic sciences of medicine…*'.[1] Sackett indeed expressed grave concerns that EBM was misinterpreted by physicians and even abused by policy makers, by interpreting it in the restricted fashion, i.e. epidemiology as the only, if not certainly superior, justification for medical decision making. Other proponents of true EBM, such as Vandenbroucke, have been very supportive in conveying ideas on the appropriate use and implementation of EBM.[2]

Why is it important to understand EBM in its correct, broad context, and not accept the more restricted definition of 'clinical decision making predominantly based on epidemiological evidence'? The answer to this question is multifactorial.[3] Briefly, a few points should be made. The first is that epidemiological evidence is available for only a small fraction of our daily work. One study estimated that of the commonly applied therapeutic interventions, less than 15% are supported by solid epidemiological evidence (http://clinicalevidence.com). In accordance, the majority of guideline recommendations are not based on solid epidemiological evidence either,[4] though many believe this to be so. The second point to be made is that, even if epidemiological studies are available, they commonly do not include the type of patients we see in daily practice. The proportion of patients with a particular disease who meet inclusion criteria is usually far less than 50%,[5] and the reasons for exclusion are often clinically relevant.[6] Failing to acknowledge the importance of differences in patient profiles, or failing to identify relevant individual context, causes substantial harm.[7] The final reason not to rely too much on epidemiological evidence is that conclusions from epidemiological studies simply do not always reflect 'the truth'. Although data can themselves never lie, various sources of bias that precede data analysis compromise the reproducibility of epidemiological studies. Some even propose that 'the majority of epidemiological literature is false'.[8]

It is for such reasons that Sackett insisted that '*External clinical evidence can inform, but can never replace, individual clinical expertise, and it is this expertise that decides whether the external evidence applies to the individual patient at all and, if so, how it should be integrated into a clinical decision ... Good doctors use both individual clinical expertise and the best available external evidence, and neither alone is enough. Without clinical expertise, practice becomes tyrannised by epidemiologcal evidence, for even excellent external evidence may be inapplicable to or inappropriate for an individual patient.*' We should be supporting this paradigm. The resolute and restrictive 1:1 translation of epidemiological findings to individual patients is a crime towards our patients and ourselves. If you practise EBM in the narrow context, you will find that the evidence razor is as useful as a barber's shop on the steps of the guillotine.

Intuitively, proper use of the EBM definition may make sense, but everywhere around us, the danger of more restrictive definitions taking over is evident. This happens in care institutions, such as hospitals, as well as in political and financial institutions, where the narrow definition of EBM is abused to regulate care and its associated costs.

In this month's issue of the Netherlands Journal of Medicine, Ubbink and colleagues address knowledge of, implementation, and attitudes towards evidence-based practice (EBP).[9] They conclude that, overall, doctors and nurses embrace the EBP paradigm, but find it difficult to implement EBP for many reasons. Some of these reasons were practical (e.g. time constraints), others were more intellectual (doubt regarding methodology, etc). Their data are informative. The differences between doctors' and nurses' attitudes are particularly interesting, and the study identifies potential ways of improving epidemiological knowledge. The authors should be commended for their work.

The question does, however, arise as to what exactly the respondents were asked to reflect upon. In other words, was EBP clearly defined *before* doctors and nurses were requested to fill in their survey? In the original application of the McColl questionnaire they used, the questionnaire itself was accompanied by a separate letter containing a clear definition of EBM: '*conscientious, explicit and judicious use of current best evidence in making decisions about the care of individual patients. Its practice means integrating individual clinical expertise with the best available external clinical evidence from systematic research*'.[10] It is hard, if not impossible, to disagree with this approach to medicine. Its implementation refers to a 'mindset' rather than a 'nominal style of practice', and requires balanced skills, including knowledge of basic sciences, epidemiological evidence, a good memory for storing clinical experience in and, perhaps above all, ability to listen to patients and recognise relevant individual context. From the answers to the questions in Ubbink's paper, it seems that many respondents may have interpreted EBP in the more restrictive context. Thus, answers and differences between them may have been related to different interpretations of the exact definition of EBP. The impression that EBP was interpreted in its restrictive definition is further strengthened by how questions were literally phrased and how answers were interpreted. For example, asking 'which competences are considered essential to *change from* experience-based to evidence-based practice' might suggest that the two should be regarded as mutually exclusive, whereas they in fact are not. Also, the 'barriers to apply EBP' reported in table 4 are noteworthy. Yes, the literature indeed reports conflicting results, has methodological shortcomings, and often does not apply to the physician's situation. To me, these is not 'barriers to applying EBP', they are integral parts of evidence-based medicine, as they should be.

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REVIEW

Neuroimmune mechanisms in functional bowel disorders

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ABSTRACT

The enteric nervous system regulates diverse functions including gastrointestinal motility and nociception. The sensory neurons detect mechanical and chemical stimuli while motor neurons control peristalsis and secretion.
In addition to this extensive neuronal network, the gut also houses a highly specialised immune system which plays an important role in the induction and maintenance of tolerance to food and other luminal antigens and in the protection of the epithelial barrier against pathogenic invasion.
It is now increasingly recognised that the gastrointestinal immune system and the enteric nervous system closely interact. This review will focus on two common functional gastrointestinal disorders in which neuroimmune interaction is involved in the pathophysiology: i.e. postoperative ileus and irritable bowel syndrome. Postoperative ileus arises after almost every abdominal surgical procedure. Handling of the bowel results in local inflammation and activation of inhibitory neuronal pathways resulting in a generalised impairment of gastrointestinal motor function or ileus. On the other hand, postinfectious irritable bowel syndrome (PI-IBS) occurs in 10 to 30% of patients who suffer from infectious gastroenteritis. PI-IBS patients develop abnormal gastrointestinal sensitivity, motility and secretion which contribute to abdominal pain and discomfort, bloating and abnormal bowel function (diarrhoea and/or constipation). Biopsy studies revealed persistent low-grade inflammation and altered immunological function which may lead to abnormal pain perception and motor activity within the gastrointestinal tract.

KEYWORDS

Post operative ileus, post infectious irritable bowel syndrome, macrophage, mast cell

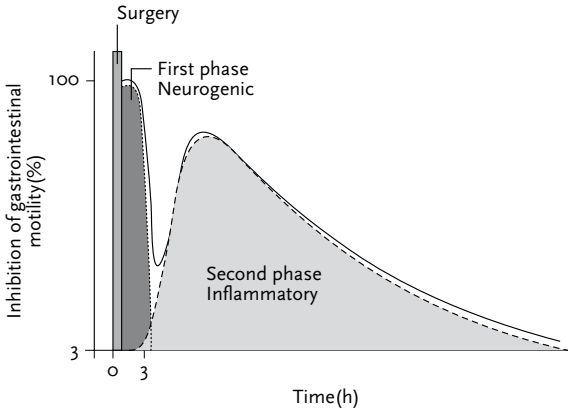
INTRODUCTION

It is generally accepted that there is bi-directional communication between the central nervous system (CNS) and the enteric nervous system (ENS). Indeed, the brain is continuously informed by afferent nerves detecting gut activity, whereas it is well established that psychological state or stress has a major influence on gut function. Interestingly, recent evidence shows that this bi-directional communication along the brain-gut axis is not confined to gut digestion and motility, but also involves immunological mechanisms, i.e. the immune system affects neuromuscular function whereas the nervous system has a major modulatory input on the immune system. In this review, we present evidence to support this bi-directional communication in two gastrointestinal disease models, postoperative ileus and postinfectious irritable bowel syndrome.

PATHOPHYSIOLOGY OF POSTOPERATIVE ILEUS

Postoperative ileus (POI) occurs after every abdominal surgical procedure. It presents clinically as the inability to tolerate food, the absence of bowel sounds, lack of flatus and defecation and abdominal distention. On average, this period lasts two to four days for conventional abdominal procedures.
By now, it is well established that POI is caused by transient inhibition of gastrointestinal motility which involves the entire gastrointestinal tract. Intestinal handling triggers two different phases, each with its own dynamics and underlying pathophysiological mechanism (*figure 1*). The first or early phase is neurally mediated and involves neural reflexes activated during and immediately following surgery. In a later phase, leucocytes enter the manipulated

Figure 1. *Schematic representation of the two phases involved in postoperative ileus*



The first neurogenic phase starts during abdominal surgery and ends soon after it. The second inflammatory phase starts approximately three to four hours after surgery, lasts much longer and is therefore clinically more relevant. (Figure adapted from Gut 2009; 58:1300-11).

intestinal segments, impairing the contractile properties of the inflamed intestine. This second phase starts three to four hours after surgery and is triggered by activation of resident macrophages in the muscularis externa. This later phase is responsible for the sustained and thus clinically more relevant inhibition of gastrointestinal motility.

THE LATE INFLAMMATORY PHASE OF POI

The second, long-lasting phase of POI is mainly due to inflammation of the intestinal muscularis.[1,2] Mainly from animal studies, it is now generally accepted that inflammation of the intestinal muscularis is the key mechanism impairing gastrointestinal motility resulting in postoperative ileus. Also in humans, we recently demonstrated that intestinal manipulation yields influx of neutrophils into the manipulated intestinal loops. Most evidence so far has identified mast cells, most probably peritoneal mast cells, and resident macrophages as the main players of the innate immune system involved in the inflammatory response to intestinal handling.

Resident peritoneal mast cells are a minor population of differentiated cells in the peritoneal cavity. Activation of these peritoneal mast cells by intestinal manipulation and the subsequent release of mediators such as histamine and mMCP-1 (murine mast cell protease 1) was demonstrated in rodent models and even in human.[3,4] Tryptase, one of the typical mediators released by mast cells, was significantly increased in the peritoneal lavage collected during abdominal surgery by gentle inspection of the intestine. In an animal model, mast cell stabilisers such as ketotifen and doxantrazole reduced the inflammatory response and improved gastric emptying 24 hours after abdominal surgery.[5,6] Moreover, transgenic mice that lack mast cells failed to develop an intestinal infiltrate following intestinal manipulation while reconstitution of these mast cells restored the capacity of mutant animals to recruit leucocytes to the intestine after surgery.[6] These data clearly support a key role for peritoneal mast cell activation in the development of POI but to date, the exact triggers activating these cells are still unclear.

Although tissue damage following intestinal handling will certainly contribute to the inflammatory cascade, mast cell activation is considered the most important step (*figure 2*). The mediators released by mast cells increase intestinal permeability, facilitating translocation of intraluminal bacteria and bacterial products (*figure 2*). In animal studies, introduction of fluorescent lipopolysaccharide or fluorescent microbeads into the intestines prior to surgery results in translocation of this fluorescent material through the mucosa into the intestinal wall. Once the beads enter the intestinal wall, they were transported to the lymph nodes via the lymphatic system or phagocytosed by the resident macrophages.[7] These data confirm that bacterial translocation occurs in response to surgery and may contribute to the activation of the immune system, in particular resident macrophages.

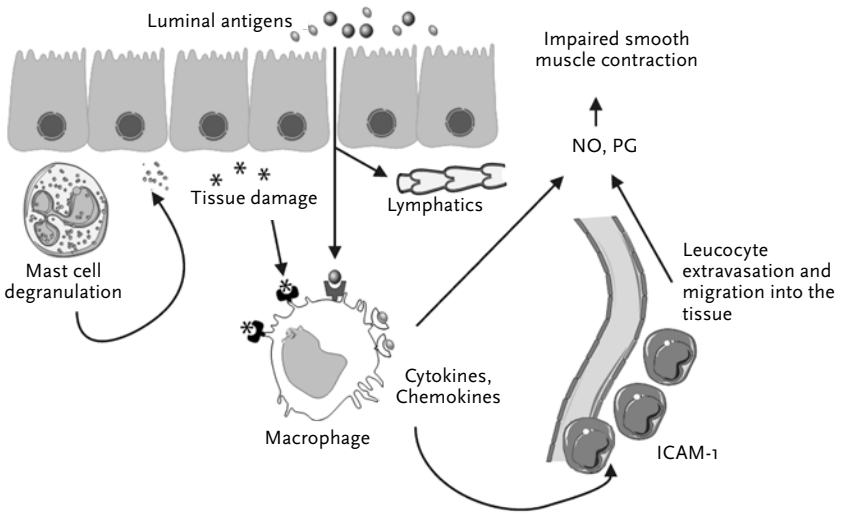
Residential macrophages are normally quiescent macrophages organised into a layer or 'network' at the level of the myenteric plexus and at the serosal side of the intestine. It has been suggested that these macrophages become activated by translocated bacterial antigens but also by the degradation products of extracellular matrix released during surgical manipulation (*figure 2*). Animal studies showed that pharmacological or genetic depletion of resident macrophages decreased the release of inflammatory mediators and diminished the recruitment of leucocytes in the muscularis.[8] In addition, macrophage-altered animals had near normal *in vitro* jejunal circular muscle function and gastrointestinal transit despite surgical manipulation, clearly illustrating the importance of these phagocytes in POI.

Stimulated macrophages secrete proinflammatory cytokines and chemokines leading to the upregulation of adhesion molecules (ICAM1) in the endothelium and the progressive influx of leucocytes (*figure 2*). Leucocytes and activated resident macrophages will synthesise enzymes such as iNOS and COX-2 which contribute to the impaired gastrointestinal motility, the main characteristic of POI.

NEUROIMMUNE INTERACTIONS IN POI

Proinflammatory cytokine production by innate immune cells and their release in the blood stream is critically associated with the pathology of inflammatory disorders

Figure 2. *Proposed immune mechanisms underlying impaired intestinal contractility following abdominal surgery*



Mast cells are activated by intestinal handling and release substances which increase mucosal permeability. This allows luminal bacteria or bacterial products such as lipopolysaccharides to enter the lymphatic system or to interact with Toll-like receptors on residential macrophages. The degrade products of the extracellular matrix that are released upon tissue damage by intestinal manipulation may also directly activate residential macrophages. The latter will release inflammatory cytokines and chemokines, resulting in upregulation of endothelial adhesion molecules including intercellular adhesion molecule-1 (ICAM-1). The latter attracts invasion of leucocytes into the intestinal muscularis externa. These leucocytes and the resident macrophages concomitantly produce large amounts of nitric oxide (NO) and cyclo-oxygenase 2 (Cox-2). Cox-2 stimulation results in production of prostaglandins (PG) which together with NO, impair the contractile activity of the smooth muscle cells.

including POI. Recently it became clear that the brain can downregulate this inflammatory response through a parasympathetic anti-inflammatory pathway.

The first studies on the role of the central nervous system in regulating systemic inflammatory responses were performed by Tracey *et al*. They demonstrated that activation of afferent vagus nerve fibres by inflammatory endotoxins or cytokines stimulated a parasympathetic anti-inflammatory pathway. Acetylcholine, the principle vagal neurotransmitter, significantly attenuated the release of proinflammatory cytokines[9] while direct electrical stimulation of the peripheral vagus nerve during lethal endotoxaemia prevented the development of shock.[9] Since then, the anti-inflammatory effect of vagal nerve stimulation has been demonstrated in models of pancreatitis,[10] ischaemia[11] and colitis.[12] Also in a mouse model of POI, De Jonge *et al*. showed that the vagus nerve exerts an anti-inflammatory action *via* activation of the alpha7 subunit of the nicotinic receptor.[3]

THERAPY

Various reports on animal research confirmed the modulatory role of the cholinergic anti-inflammatory pathway on the intestinal immune system,[3,12] whereas vagotomy ameliorated inflammation. These reports open new perspectives in the development of new anti-inflammatory compounds. Agonists that mimic the effect of the vagal nerve can be developed to treat POI. We previously showed that pretreatment with a specific alpha7 nicotinic agonist indeed prevents inflammation and postoperative ileus in mice.[13] An alternative approach is to activate the endogenous vagal anti-inflammatory system. This can be achieved by feeding a high fat diet. Dietary fat induces the release of cholecystokinin which activates the vagal nerve indeed resulting in an anti-inflammatory effect[14] in animal models of sepsis and POI.[14] Maybe this clarifies the protective effect of fast-track surgery against POI.[15-17] Fast-track surgery promotes postoperative early ambulation and early oral hydration and nutrition.

Finally, as mast cells are playing an important role in the initiation of the inflammatory cascade triggered by intestinal handling, and mast cell stabilisation has proven efficient in our murine model, we designed a pilot study evaluating the effect of the mast cell stabiliser ketotifen on postoperative ileus in patients undergoing major abdominal surgery for gynaecological malignancy.[5] In this study, mast cell stabilisation restored gastric emptying and relieved abdominal cramping.[5]

In summary, intestinal inflammation due to handling of the intestines activates mast cells and resident macrophages which results in an influx of leucocytes and an inflammatory response leading to impaired motility and ileus. Minimising intestinal handling and fast track surgery shorten POI while mast cell stabilisation and drugs or interventions mimicking the effect of the vagal anti-inflammatory pathway may represent new approaches for the treatment of POI.

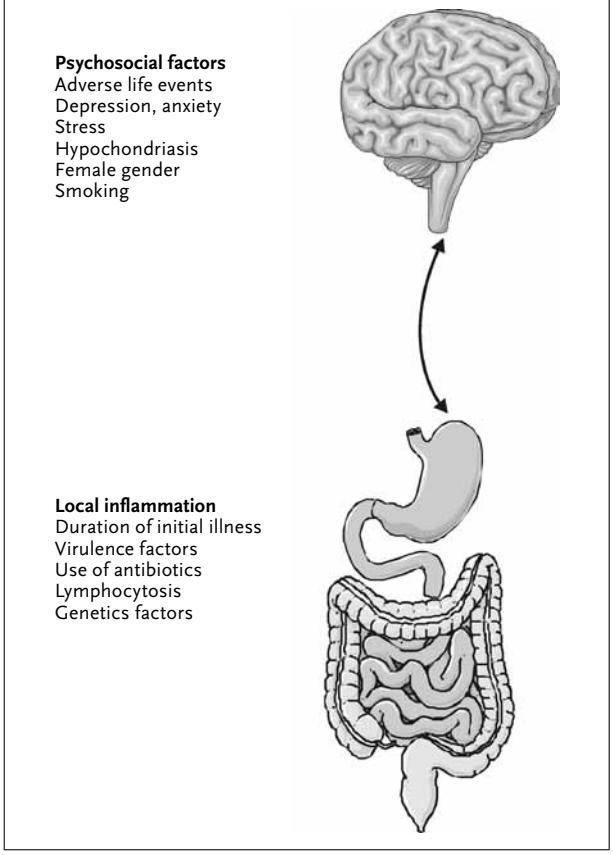
PATHOPHYSIOLOGY OF POSTINFECTIOUS IRRITABLE BOWEL SYNDROME

The irritable bowel syndrome or IBS is the most common gastrointestinal disorder affecting approximately 15% of individuals worldwide. IBS is characterised by chronic abdominal pain, discomfort, bloating, and alteration of bowel habits in the absence of any detectable organic cause. Increased abdominal pain perception or visceral hypersensitivity is considered an important pathophysiological mechanism explaining the clinical presentation of IBS. The diagnosis is based on IBS symptoms as described in Rome III criteria as no clinical tests nor imaging techniques can positively identify IBS.

In some individuals, IBS has an acute onset following an infectious gastroenteritis. This postinfectious syndrome has consequently been termed 'postinfectious IBS' (PI-IBS). Published studies have reported the incidence of PI-IBS after an infection ranges between 5% and 32%. Various bacterial pathogens including *Campylobacter, Shigella, Salmonella* and *Escherichia coli* have been implicated in the development of PI-IBS. Whether all organisms confer an equivalent risk remains unclear.

Potential risk factors for developing long-standing gastrointestinal symptoms after an acute infection include female gender and environmental factors (smoking, psychological distress) (*figure 3*). Especially a recent traumatic life event or a neurotic personality trait increase the susceptibility to develop PI-IBS. Genetic risk factors that underlie the susceptibility to develop postinfectious IBS include gene alterations in TLR9, a gene responsible for bacterial recognition, IL6, involved in inflammatory response and cadherin-1 (CDH1), involved in epithelial integrity.[18] In addition, factors related to the pathogen or the infection including the duration and severity increase the risk of developing postinfectious IBS (*figure 3*). Based on these risk factors it appears that the interaction between local inflammatory factors and psychosocial factors plays an important role in the development of PI-IBS, which fits well with the current theory of brain-gut dysfunction in patients with IBS. The exact mechanisms behind the development of long-standing gastrointestinal symptoms after an infectious event are not clear.

Figure 3. *Interaction between local inflammation and psychosocial factors in determining the risk of developing postinfectious IBS*



INTESTINAL BARRIER FUNCTION

Compromised epithelial barrier function has been associated with both IBS and PI-IBS.[19,20] Marshall *et al.* demonstrated a high incidence of postinfectious IBS with increased permeability following an acute bacterial gastroenteritis after a waterborne outbreak. This aberrant barrier function in IBS patients is due to decreased expression of tight junction proteins including zona occludens protein 1 (ZO-1).[21] It remains unclear, however, why these proteins are affected in IBS patients. Interestingly, increased permeability was evoked in mice when faecal supernatants from patients with diarrhoea-predominant IBS were applied to the colonic mucosa,[22] clearly indicating the presence of certain components that affect epithelial barrier function. It has been speculated that proteases such as tryptase (endogenous or deriving from commensal bacteria) may play an important role. Recent studies demonstrated the role of protease-activated receptors (PAR) in barrier dysfunction and in the generation of IBS symptoms. Activation of PAR1 and 2 on enteric neurons provokes the release of neuropeptides[23] which in turn activate their receptors present on endothelium and mast cells. Such PAR-induced micro-inflammation might participate in the generation of IBS symptoms as low levels of inflammation have been proposed to be involved in the pathogenesis of hypersensitivity. PAR2-induced permeability and rectal hypersensitivity could be inhibited by a tight junction blocker.[24] These studies demonstrate an important role for increased intestinal permeability in the development of PI-IBS hypersensitivity.

NEUROIMMUNE INTERACTIONS IN PI-IBS

Mast cell activation

As stated above, increased abdominal pain perception or visceral hypersensitivity is the hallmark of IBS. A variety of animal models of visceral hypersensitivity demonstrated immune activation with subsequent microscopic inflammation as the underlying mechanism of abnormal pain perception. Especially the role of mast cell activation in postinfectious IBS has been extensively studied. In PI-IBS animal models using *Trichinella spiralis*[25-27] or *Nippostrongylus brasiliensis*[28,29] infections, the development of visceral hypersensitivity has been associated with increased mast cell numbers and/or activation. Similarly, several studies indicated an increase in mast cell numbers in close proximity of nerve fibres in intestinal biopsies of IBS patients[30] demonstrating interactions between mast cells and nerves may be relevant for symptom generation. In addition, the supernatant of patient biopsies which contains more mast cell mediators[31] evoked an increased *in vivo* pain response to colonic distension in mice.[32] The importance of mast cell mediators was also addressed in a study by Buhner *et al*., who reported that the supernatants of colonic biopsy samples from IBS patients but not of those from healthy controls, activate human submucosal neurons.[33] Interestingly, this activation was not associated with IBS subtype (constipation/diarrhoea/alternating IBS), which indicates that it might be a general pathogenetic mechanism of IBS. In another study, peripheral blood mononuclear cell supernatants from patients with postinfectious IBS, but not of controls, activated mouse colonic pelvic or lumbar splanchnic nerves, which further suggests that the immune system is potentially involved in the generation of visceral hypersensitivity in IBS.[34] Taken together, mast cells release neurally active mediators (histamine, proteases, prostaglandins) which sensitise afferent neurons inducing increased visceral pain perception. On the other hand, mast cells also express a variety of neuropeptide receptors, demonstrating the significance of mast cells as end effector cells of the brain gut axis in the intestinal mucosa.

Adaptive immunity

It has been hypothesised that an adaptive immune response may underlie visceral hypersensitivity. Serial rectal biopsies taken from patients who developed IBS after a *Campylobacter jejuni* gastroenteritis showed a persistent inflammatory infiltrate, with an increase in enterochromaffin cells, mast cells and lymphocytes.[20] The recent finding that antibodies against nonspecific bacterial products, flagellin, are increased in IBS patients[35] together with the increase in IgG+ B cells in PI-IBS patients[36] underscores the importance of an adaptive immune response in PI-IBS. Therefore, it is hypothesised that PI-IBS patients develop a chronic low-grade immune response against commensal microbiota. Although these studies indeed indicate a role for immune activation in PI-IBS, there are some discrepancies in the literature that remain to be further studied. Two studies reported increased numbers of T lymphocytes[20,37] in PI-IBS whereas another study described normal T cell numbers in intestinal tissue.[38] Also the cytokine profile of mucosal T cells of (PI-) IBS patients has not been studied yet. More detailed studies of T and B cell activity and altered antibody production in IBS will most certainly result in a better understanding of IBS symptom generation.

THERAPY

Although the mechanisms by which initial inflammation triggers a state of visceral hypersensitivity remain unclear, it is thought to be related to modulation of visceral sensory neurons by mast cell mediators. More evidence of mast cell involvement in IBS is provided by drug studies such as a study by Klooker *et al*. In this study, treatment of IBS patients with the mast cell stabiliser ketotifen resulted in decreased visceral hypersensitivity and improved intestinal symptoms.[39] Another small study reported reduction in mast cell numbers and mediators following treatment with the anti-inflammatory drug mesalazine.[40] In mice, early corticosteroid therapy has been shown to attenuate postinfectious neuromuscular dysfunction.[41] However, a small randomised controlled trial with 29 PI-IBS patients given 30 mg prednisolone/day for three weeks was negative.[42] As stated above, disruption of intestinal barrier integrity plays an important role in PI-IBS and improving its function and integrity by glutamine supplementation may represent a new therapeutic approach.[43] Also therapeutic alteration of the GI microbiota by probiotic bacteria was shown to improve IBS symptoms and restore intestinal homeostasis.[44]

In summary, PI-IBS is triggered by an acute gastroenteritis and is characterised by increased mucosal permeability and a chronic low-grade inflammatory response in the mucosa. Mast cell activation plays a crucial role and their mediators sensitise nociceptive nerve fibres, thereby inducing increased abdominal pain perception. There is no standard treatment available so far but pilot studies indicate the beneficial role of mast cell stabilisers, anti-inflammatory drugs, glutamine supplementation and probiotics.

CONCLUSION

In conclusion, postoperative ileus and postinfectious IBS are the result of abnormal neuroimmune interactions. Inflammation in POI involves mast cell and subsequent

macrophage activation followed by an influx of leucocytes and impaired motor function. The vagal nerve exerts a cholinergic anti-inflammatory pathway and its activation may represent a new therapeutic approach in the treatment of POI. In contrast, inflammation in PI-IBS involves chronic loss of epithelial barrier integrity followed by mast cell activation and chronic low-grade inflammation. Release of proinflammatory mast cell mediators sensitise nociceptive neurons resulting in increased visceral pain sensitivity. Future research on the nature of the inflammatory response will provide insight and new tools to prevent and treat POI and PI-IBS.

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REVIEW

Orthostatic proteinuria: a harmless variant of protein loss?

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ABSTRACT

A 28-year-old young woman was referred to our department of Internal Medicine for analysis of unintentional weight loss. At initial analysis, a persistent proteinuria was found with no evident relation to her weight loss. Anamnestic as well as additional studies showed no evidence of a primary kidney disease. After this exclusion, orthostatic proteinuria was confirmed by simple urine analysis. Since the weight loss had not yet been explained, an analysis followed at the Department of Gastointestinal and Liver Diseases where inflammatory bowel disease (IBD) was found. Literature study shows that proteinuria may be associated with IBD. This concerns mainly selective tubular protein loss, without a distinctive change in protein loss with a change in position. Orthostatic proteinuria, therefore, remained the most likely diagnosis. In this case, the patient was advised to check both urine and kidney function annually.

KEYWORDS

Orthostatic proteinuria, kidney function, proteinuria, inflammatory bowel disease

INTRODUCTION

In recent years, there has been much attention for proteinuria, focusing mainly on its pathological significance in the context of renal diseases, progression of this disease, and for proteinuria as a marker of secondary injury of other causative diseases such as arteriosclerosis or diabetes. In addition, protein loss itself could cause damage to glomeruli and tubules which could deliver further deterioration of renal function. However, proteinuria is not always a harbinger of renal damage and loss of renal function. This case shows that isolated proteinuria without renal disease or renal damage may occur and may have an innocent origin.

CASE REPORT

A 28-year-old, previously healthy athletic young woman was referred to the Internal Medicine clinic in relation to unintentional weight loss. In a period of five months, she had lost about 12 kilos reducing her weight to 65 kilos (body mass index 18.6). She had had no fever. She denied gastrointestinal complaints and had a normal eating pattern and unchanged defecation. Also diuresis was normal. There were no joint or skin disorders, nor cardiopulmonary symptoms.

She did not smoke, alcohol consumption was limited and she took no medications or vitamins. Her family history was negative for kidney disease and diabetes.

On physical examination no abnormalities were found except the slender habit. Blood pressure was normal, 110/65 mmHg RR.

Laboratory showed no abnormalities; in particular kidney and liver function tests were normal. No inflammatory markers were found and thyroid functions were also normal. Determination of anti-TTG (tissue transglutaminase) was repeatedly negative. Serology revealed no evidence of parasitic infections; a triple faeces test was also negative. Stool examination was negative for elastase and lipase. A chest X-ray revealed no abnormalities; an ultrasound found normal kidneys, in size as well as the aspect of the parenchyma and cortex.

The urine screen tested repeatedly positive for protein, without erythrocyturia, leucocyturia or glucosuria. Quantitative research showed a 24-hour protein excretion of 0.30 g/24 hours (2.10 litre volume, creatinine excretion 13.9 mmol /24 hrs) and a micro-albumin excretion of 127 mg/24 hrs (9.1 micro-albumin/creatinine ratio mg/mmol).

In summary, neither anamnestic nor additional studies were indicative of IgA nephropathy, post-infectious or other primary or secondary renal disorders. Because we suspected orthostatic proteinuria, we asked the patient to collect a single morning urine sample immediately after rising in the morning and a second sample on the same day at normal work and effort patterns.

The morning urine was completely free of protein, the second sample of urine showed a marked proteinuria with a protein/creatinine ratio of 30.2 mg/mmol and 17.1 mg of micro-albumin/creatinine ratio/mmol. Because of this finding, we concluded the proteinuria to be orthostatic proteinuria. The patient refused to start ACE inhibition, partly because of the predominantly good prognosis of this illness and her low-normal blood pressure.

Because we could not explain the substantial weight loss over 12 kilos, she was referred for further analysis. At gastroduodenoscopy, a normal surface of gastric and duodenal mucosa was found and the biopsies were negative for coeliac disease. Colonoscopy, however, yielded an image of a mild sigmoiditis which, on microscopic examination, was suspicious for Crohn's disease. Literature studies found an association with inflammatory bowel diseases and slight to moderate proteinuria without loss of renal function. However, this could not explain the postural aspect of the proteinuria; therefore we did not doubt the existence of orthostatic proteinuria. Regarding this sigmoiditis, the patient remained for therapy on the Gastrointestinal and Liver Diseases ward.

DISCUSSION

Isolated asymptomatic proteinuria is frequently encountered in daily practice.[1] It is easy to detect, and an estimated quantification by using the dipstick is easy to make, based on coloration of the strip. When finding proteinuria, one should always consider further analysis because proteinuria may be a symptom of a primary renal disease or a complication of another disease such as diabetes, hypertension or systemic disease

In recent years, random screening for proteinuria has increased, partly because of the 'kidney screening' introduced by the Kidney Foundation in the Netherlands. This campaign has now been discontinued because its goal was achieved: attention to and highlighting of early symptoms and the silent course of renal disease and thus to emphasise the importance of screening for proteinuria in risk groups, such as diabetics or patients with hypertension.[2] Early detection of proteinuria could lead to earlier diagnosis and treatment of kidney diseases as well as slowing down the progression of complications of diseases such as diabetes or hypertension.[1,2]

Nevertheless, the existence of isolated asymptomatic proteinuria should not always raise suspicion of a primary renal disease or complication of other diseases. It is therefore essential to make a distinction between innocent causes of transient proteinuria and pathological causes which need treatment in short notice.[1]

ORTHOSTATIC PROTEINURIA

Orthostatic proteinuria was first reported in the Lancet by Pavy in 1885, who described a proteinuria with a cyclic character which consisted of protein-free urine in the morning and at night, but proteinuria at daytime. In 1887, Striling found a relation with position and called it postural proteinuria. Soon, it became known that this cyclic proteinuria was present in 15 to 30% of children and was related to physical disturbances such as headache, dizziness, paleness and collapse. Causal explanations were numerous: anatomical changes, metabolic disorders, glomerular disorders, some kind of infectious or septic kidney disruption, cardiovascular disorder or just a weak constitution were all thought to be possible causes. Nevertheless, it was mostly believed to be a mechanical disorder caused by a hyperlordosis in the lumbar spine, which caused an extension of the renal vein while standing that disappeared while lying down, thus causing hydronephrosis and proteinuria with a variability in protein loss between day and night. As the muscles of the lumbar spines strengthen while growing up and the hyperlordosis disappears, this would explain why this disorder particularly exists in children.

When only orthostatic proteinuria is present, it was believed to be a benign disorder with benign causes, which would probably disappear over time. Urine was checked once in a while for the amount of protein loss. However, when urine samples also showed significant haematuria or cylinders, a renal disorder was suspected.

The first thesis about orthostatic proteinuria in our country dates from 1918 and was written by P.H. Kramer. He tested urine samples from soldiers for protein and found orthostatic proteinuria in 8% of healthy individuals, especially after hard work and effort or long periods of standing. This thesis again emphasised the mechanical anatomical explanation, but also found a relation with a weak cardiovascular system, both of which were supposed to cause congestion of the kidneys.[3]

Nowadays, orthostatic proteinuria can still be defined as isolated proteinuria that occurs in the upright position and disappears in a supine position. This distinguishes orthostatic proteinuria from other benign causes of proteinuria, such as some types of proteinuria during pregnancy, hyperthermia, but also exercise-induced, cold-induced, and orthostatic proteinuria.[2,4] There is a

fixed, reproducible form and a transient form, but the latter could possibly be regarded as a different kind of proteinuria.[2,5] Orthostatic proteinuria is the most common cause of protein loss in children (60%) and adolescents (75%), but its prevalence decreases during ageing and is rare in adults over 30 years.[2,6] The severity of proteinuria can not be used diagnostically, nor for prognostic purposes: proteinuria could even be found in the nephrotic range.[5] While this form of proteinuria has long been known and has been described in many textbooks, maybe the pathophysiology is still not complete or perhaps misunderstood. Several mechanisms may be responsible for the development of proteinuria, including changes in glomerular permeability or inadequate tubular dysfunction, but permanent renal damage is not necessary for proteinuria to occur.[2]

HAEMODYNAMIC MECHANISM

A number of interesting hypotheses attribute proteinuria to altered renal haemodynamics and associated changes in glomerular filtration. Generally, the degree of protein loss in a standing position is greater than in the supine position, even in normal physiology and a healthy kidney.[5-7] Up to 20% of healthy volunteers would lose more protein in a standing than in lying position, while total proteinuria loss does not exceed 150 mg/day.[6] This can be explained by the increase of angiotensin II and noradrenaline in the standing position, which causes renal efferent vasoconstriction and arteriolar resistance, increases the glomerular filtration pressure and glomerular filtration rate and thus causes an increase of proteinuria.[5-7] It could therefore be assumed that orthostatic proteinuria is an 'exaggerated' response to angiotensin and is thus a variant of a normal response.[5,6]
Moreover, it is shown that this increase in protein loss is a selective proteinuria, which supports this hypothesis.[6] In many other forms of proteinuria, such as in pathological glomerulonephritis, also an increase in non-selective protein loss is seen in an upright position. The local glomerular haemodynamics may also change in the standing position, again caused by angiotensin II. By increasing the glomerular filtration pressure and filtration fraction in local efferent vasoconstriction, the intrinsic size selectivity of the basement membrane changes, increasing filtration of large proteins.[9] Damage to the glomerular basement membrane by continued elevated pressures and increased proteinuria could be a logical result.[9] Other studies suggest that non-haemodynamic effects of angiotensin II, which acts as a local endogenous hormone, cause increased production of free oxygen-radicals, upregulation of cytokines and leukotrienes, profibrotic growth factors and, eventually, an increased production of extracellular metalloproteins,[7,9] resulting in proteinuria.

NUTCRACKER PHENOMENON: OBSTRUCTION MECHANISM

Especially in paediatric literature, much attention is paid to the so-called nutcracker phenomenon as an explanation for orthostatic proteinuria. This phenomenon was first described in 1972. It is thought to be caused by a transient partial obstruction of the left renal vein because of its anatomical location between the abdominal aorta and the superior mesenteric artery.[9,10-12] Although rare, the nutcracker phenomenon causes a variety of symptoms of (left-sided) microscopic and macroscopic haematuria, ureter and parapelvic varices and unexplained flank pain. Also an association was found with chronic fatigue in children.[12,13] This obstruction, which occurs especially in the standing position, also leads to stimulation of angiotensin II by the decreased renal blood flow.[10] Proteinuria may occur or increase in the same way as described above.[5,7,9]
The nutcracker obstruction can be visualised with Doppler ultrasound and MRI, but the gold standard is renal angiography or retrograde renography.[10-13] However, using imaging techniques to show obstruction does not give reliable answers to the haemodynamic significance of this obstruction, so the nutcracker syndrome should solely be a clinical diagnosis.[12] A surgical approach may be chosen if the nutcracker syndrome causes severe symptoms such as massive haematuria causing refractory anaemia or persistent flank pain. For asymptomatic proteinuria, however, surgical intervention is not indicated. ACE inhibition may be considered, with significant reduction or even disappearance of the protein loss.[8,9] However, after cessation of treatment, proteinuria usually reappears. Because of the presumed benign course, it remains unclear whether medical therapy or conservative management should be chosen.[8,9]

IMMUNOLOGICAL MECHANISM

Still unclear and maybe even controversial remains the significance of subtle but pathological changes found on renal biopsy. These changes are seen in the glomerular basement membrane, best shown by immunofluorescence. There seems to be a possible relationship to complement activation (especially C3 and C4 activation, found in basement membrane in orthostatic proteinuria).[5,6] To increase the knowledge base in this area, further study and research is required, in order that clearer statements can be delivered.

DIAGNOSTICS

The diagnostics of orthostatic proteinuria are easy to determine in different ways. The most reliable, but less

practical method consists of a 24-hour urine collection, which is separated into a 16-hour collection during the day and an 8-hour collection during the night. The supine position should be taken two hours before finishing the 16-hour day collection, to avoid contamination of the following 8-hour collection of urine.
An easier alternative is calculating a micro-albumin/creatinine ratio in two different urine samples: one first morning urine sample and a sample during the day. The normal value is <0.5 mg/mmol. When the second sample is both dipstick positive for protein and shows an increased micro-albumin/creatinine ratio, orthostatic proteinuria can be strongly suspected.[7,9,14]

COURSE AND PROGNOSIS

Although it is generally accepted that proteinuria itself could be harmful to the kidney, deterioration of renal function is uncommon and progression to end-stage renal disease has not been described; proteinuria usually decreases and disappears over the years.[1,2,5-7,14,15] To our knowledge, the period during which renal function and proteinuria should be monitored is not explicitly stated. Also, the frequency at which urine samples and kidney function should be checked has not been specified, but annual monitoring seems to be sufficient and reasonable. When renal function deteriorates and with a persistently increasing proteinuria, one should consider other kidney diseases and refer the patient to a nephrologist.[1,2,14,15]

RELATIONSHIP BETWEEN PROTEINURIA AND IBD?

As described earlier, proteinuria is frequently noted as a secondary phenomenon in disorders other than renal diseases, and can also occur secondary to inflammatory bowel diseases (IBD).[16-18] Proteinuria in inflammatory bowel disease is variable in nature, but seems partly correlated with histopathological staging of the disease and disease activity.[16-18] Some authors even suggest that the degree of proteinuria can be used as a marker for the degree of disease activity.[16,17] This relationship especially seems to exist for the loss of tubular proteins (e.g. microglobulin).[17] The suggestion that proteinuria could be caused by treatment of inflammatory bowel disease can be ignored in our case, since at the time of the analysis and diagnosis, the patient was not taking any medication.[13,16] To our knowledge, only accidental relationships but no causal ones between orthostatic proteinuria and inflammatory bowel disease have been described in literature.

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ORIGINAL ARTICLE

Long-term follow-up of organ-specific antibodies and related organ dysfunction in type 1 diabetes mellitus

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ABSTRACT

Objective: Diabetes mellitus type 1 (DM1) is associated with other autoimmune disorders. To our knowledge, there are no longitudinal data considering the long-term clinical relevance of organ-specific antibodies (OS-Ab) in DM1 patients. We performed a long-term retrospective longitudinal study in order to investigate the presence and diagnostic accuracy (positive predictive value: PPV and negative predictive value: NPV) of OS-Ab in DM1 patients.

Research design and methods: In a retrospective longitudinal study, the presence of OS-Ab and related organ function were analysed in 396 DM1 patients (184 F/212 M, age 44±13 years, age at onset of DM1 21±13 years), with a median follow-up time of 23±10 years.

Results: OS-Ab frequencies at baseline were: antibodies against thyroglobulin (Tg-Ab) 4.3%, antibodies against thyroid peroxidase (TPO-Ab) 8.1%, Tg- and/or TPO-Ab 10.4%, antibodies against parietal cells (PCA) 5.8% and antibodies against adrenal cortex (ACA) 0.5%. The occurrence of (sub)clinical hypothyroidism was higher in patients with Tg-Ab (47%) or TPO-Ab (42%) than in those without these antibodies (6.2 and 5.1%, respectively, p<0.001). PPV and NPV for Tg-Ab were 0.60 and 0.88, respectively, for TPO-Ab 0.54 and 0.91. Also in patients with PCA, organ dysfunction occurred more often (61%) than in patients without PCA (9.7%, p<0.001). PPV for PCA was 0.61 and NPV 0.90. NPV and PPV for ACA could not be calculated because of the low prevalence.

Conclusion: Long-term follow-up of 396 DM1 patients shows that the presence of thyroid antibodies and/or parietal cell antibodies is clearly associated with dysfunction of the corresponding organ.

KEYWORDS

Type 1 diabetes mellitus, autoimmune antibodies, organ-specific antibodies, autoimmune thyroiditis, autoimmune gastritis; Addison's disease

INTRODUCTION

Type 1 diabetes mellitus (DM1) is associated with other immune-mediated disorders,[1,2] such as autoimmune thyroiditis,[3-6] Addison's disease[7] and pernicious anaemia.[8,9] In the past years, extensive research has been performed to predict the occurrence of autoimmune diseases by the presence of organ-specific antibodies (OS-Ab), as recently reviewed.[10]

Thyroid antibodies (Th-Ab) are directed against thyroglobulin (Tg-Ab) or against thyroid peroxidase (TPO-Ab). TPO-Ab prevalences in DM1 populations vary between 5.5 and 46.2% and in control populations between 0 and 27.0%.[5,9,11-32] Tg-Ab prevalences in DM1 populations vary between 2.1 and 40% and in control populations between 0 and 20%.[5,12-15,18-20,22,25,29,32,33] The prognostic significance of Th-Ab has been studied in several longitudinal non-diabetic populations.[34-36] The risk of developing overt hypothyroidism per year in TPO-Ab positive individuals is higher than in TPO-Ab negative individuals (4.3 and 2.6%, respectively).[34,37] Parietal cell antibodies (PCA) are directed against the parietal cells in the stomach,[38,39] chronically targeting H^+/K^+ ATPase, which can lead to atrophic gastritis, hypochlorhydria or achlorhydria, and a decline in intrinsic factor production, causing hypergastrinaemia, vitamin B12 malabsorption and ultimately pernicious anaemia.[39,39] Hypochlorhydria may also impair iron absorption which can lead to iron deficiency anaemia.[40-45] The PCA prevalences in DM1 populations

range from 3 to 34% and in control populations from 0 to 13%.[9,11,13,16,18-20,22,24,25,27,32,46-50] To our knowledge, no prospective studies have been published concerning PCA in DM1.

Adrenocortical autoimmune disease, also called primary adrenal insufficiency or Addison's disease, is the result of humoral and cell-mediated inflammation of the adrenal cortex.[51] Adrenal cortex antibodies (ACA) are directed against 21-hydroxylase, a microsomal cytochrome P450 enzyme that converts 17-α-hydroxyprogesterone and progesterone into 11-deoxycortisol and 11-deoxycorticosterone.[52] The ACA prevalences in DM1 populations range from 0 to 4% and in control populations from 0 to 0.7%.[16,24,25,32,46-48,53-58] To date, only one longitudinal study has been performed that studied ACA: Betterle *et al*. performed a longitudinal analysis of 15 DM1 patients with organ-specific autoimmune disease who were positive for ACA: 40% developed Addison's disease during a mean observation period of 3.2 years.[59]

CLINICAL PROBLEM AND RESEARCH QUESTION

Early detection of antibodies and latent organ-specific dysfunction is important to alert physicians to take appropriate action in order to prevent full-blown disease.[60] Although from a clinical point of view it is of utmost importance to be able to determine the prognostic significance of OS-Ab, most studies so far have had a cross-sectional design. Obviously, longitudinal studies are needed to fill this gap in knowledge. Therefore, we performed a retrospective longitudinal study in order to investigate the prevalence and clinical relevance of thyroid antibodies, parietal cell antibodies and adrenocortical antibodies, and the prevalence of corresponding organ dysfunction during more than 20 years follow-up of 396 patients with diabetes mellitus type 1.

RESEARCH DESIGN AND METHODS

Research design

A total of 396 consecutive patients with DM1 from the Diabetes Outpatient Department of the Leiden University Medical Center were included in this retrospective longitudinal study between 1981 and 1998. We assessed the presence of OS-Ab and / or autoimmune thyroid disease, Addison's disease, or macrocytic, normocytic or microcytic anaemia during more than 20 years of follow-up.

ANTIBODY DETECTION METHODS

PCA and ACA were measured by indirect immunofluorescence using tissue slides of Scimedx (Denville, NJ, USA). Thyroid antibodies (TPO-AB and Tg-Ab) were measured by radioimmunoassay (DiaSorin, Saluggia, Italy). The Tg-Ab assay range is from 5 to 6500 kU/l, reference value <100 kU/l; the TPO-Ab reference range is <60 kU/l. Both assays had coefficients of variation of <10%. Monkey tissue was used to detect Th-Ab and ACA, whereas rat tissue was used to detect PCA. Both tissue slides were manufactured by SciMedex. Patients who were weakly positive or doubtfully positive for antibodies were not taken into account; only positive, strongly positive and negative patients were considered.

ENDOCRINE ASSESSMENTS

Serum thyroid-stimulating hormone (TSH) and FT4 were measured by time resolved fluoroimmunoassay and serum T4 and T3 by in-house radioimmunoassay methods. Reference values for T3 were 1.1 to 3.1 nmol/l, for T4 70 to 160 nmol/l, for free T4 10 to 24 pmol/l and for TSH 0.3 to 4.8 mU/l. Overt clinical hypothyroidism was defined as elevated TSH levels and T3, T4, or free T4 levels under the lower limit of normal. Subclinical hypothyroidism was defined as an elevated TSH level with normal T3, T4, or free T4 levels. Overt clinical hyperthyroidism was defined as both suppressed TSH levels and T3, T4, or free T4 levels above the upper limit of normal. Subclinical hyperthyroidism was defined as a suppressed TSH level with normal T3, T4, or free T4 levels.

Between 1978 and 1986, cortisol was measured by in-house radioimmunoassay with an interassay coefficient of variation of 10% and with a detection limit of 50 nmol/l. Between 1986 and 1994, a fluorescence energy-transfer immunoassay Syva-Advance (Syva Company, Palo Alto, CA) was used, with an interassay variation coefficient of 3.6 to 6.1% and a detection limit of 50 nmol/l. From 1994, cortisol was measured by fluorescence-polarisation assay on a TDx (Abbott Laboratories, Abbott Park, IL). The interassay variation coefficient is 5 to 6% above 500 nmol/l and amounts to 12% under 200 nmol/l. The detection limit is 20 nmol/l. The methods correlated well with each other, and therefore no correction factors were introduced for follow-up of patients. Reference values for morning cortisol were 0.20 to 0.60 µmol/l.

Adrenocorticotropic hormone (ACTH) has been measured since 1986 using an immunoradiometric assay (Nichols Institute Diagnostics, San Juan Capistrano, CA) with a detection limit of 3 ng/l. The intra- and interassay average variations ranged from 2.8 to 7.5% across the sample range observed. If Addison's disease had to be excluded because of the presence of ACA antibodies or because of a clinical suspicion, an ACTH stimulation test with 250 µg synacthen was used. The test was interpreted as normal when the cortisol level exceeded 0.55 umol/l at 60 minutes after stimulation; hypocortisolism was diagnosed when the cortisol level failed to reach this value.

The haemoglobin (Hb) levels and mean corpuscular volume (MCV) were determined with an automated analysis system (Coulter Counter; Coulter Electronics, Hialeah, Florida). Reference values for Hb were 7.5 to 10 mmol/l for women and 8.5 to 10 mmol/l for men; the reference value for MCV was 80 to 100 fl for both sexes.

Serum levels of vitamin B12 were determined using the Dual Count Solid Phase No-Boil Assay (Diagnostic Products Corp., Los Angeles, California). Vitamin B12 deficiency was defined as serum vitamin B12 levels lower than 150 pmol/l.

Parietal cell dysfunction was diagnosed when atrophic gastritis, macrocytic anaemia or pernicious anaemia was present. Macrocytosis was defined as a high MCV without anaemia. Pernicious anaemia was defined as anaemia with a high MCV in the presence of atrophic gastritis. Microcytic and normocytic anaemias were also taken into account, since achlorhydria can cause iron deficiency and subsequent microcytic anaemia, which can result in normocytic anaemia when combined with macrocytic anaemia.

STATISTICAL ANALYSIS

Patients' data were analysed using SPSS 16.0 (ANOVA and Chi Square). Prevalence of organ dysfunction was compared between antibody positive and negative DM1 patients. Positive predictive value (PPV) was calculated as the number of patients with organ dysfunction divided by the total number of patients with OS-Ab for whom organ function was tested. Negative predictive value (NPV) was calculated as the number of patients with organ dysfunction divided by the total number of patients without OS-Ab for whom organ function was tested.

RESULTS

The age at the time of the study of the 396 patients (184 females and 212 males) was 44±13 years, age at onset of DM1 was 21±13 years. Median time from referral to final assessment was 21 (8 to 70) years and was comparable for antibody positive and negative patients (*table 1*).

Table 1. *Prevalence of organ-specific antibodies and corresponding organ dysfunction in 396 DM1 patients*

Antibodies	**Tg-Ab**		**Hyperthyroidism**		**Tg- and/or TPO-Ab**			**PCA**			**ACA**	
	-	+	-	+	-	+		-	+		-	+
N (total)	333 (84.1%)#	17 (4.3%)	308 (77.7)#	32 (8.1%)	295 (74.5)#	41 (10.4%)		362 (91.4)#	23 (5.8%)		392 (98.9)#	2 (0.5%)
% F	42%	71%**	42%	78%**	41%	76%**		45%	70%*		46%	100%
Age (baseline)	43.4 ±12.9	45.8 ±10.7	43.2 ±12.9	45.3 ±10.5	43.1 ±13.0	45.4 ±11.1		43.6 ±12.5	43.4 ±17.7		43.6 ±12.8	59.0 ±17.0
DM duration (baseline)	22.4 ±10.0	22.6 ±10.0	22.5 ±10.0	21.7 ±11.1	22.4 ±10.1	22.5 ±11.2		22.7 ±10.2	21.9 ±12.2		22.7 ±10.4	27.5 ±26.2
Organ dysfunction (total)	11.7%	60.0%	9.4%	53.4%	9.1%	52.9%		9.7%	60.9%			
Subclinical hypothyroidism	0.8%	13.3%	0.9%	11.5%	0.9%	14.7%	Macrocytosis	1.4%	4.3%	Hypocortisolism	2.4%	0
Clinical hypothyroidism	5.5%	33.3%	4.3%	30.8%	3.6%	29.4%	Macrocytic anaemia	0.3%	4.3%	Hypercortisolism	4.9%	0
Hyperthyroidism	3.1%	0	1.7%	3.8%	1.8%	2.9%	Pernicious anaemia	0.3%	8.7%			
Graves	2.3%	13.3%	2.6%	7.7%	2.7%	5.8%	Normocytic anaemia	5.1%	26%			
							Microcytic anaemia	2.6%	17.4%			
Diagnostic accuracy	NPV 0.88	PPV 0.60	NPV 0.91	PPV 0.53	NPV 0.91	PPV 0.53		NPV 0.90	PPV 0.61			
AB+ vs AB -	p<0.001		p<0.001		p<0.001			p<0.001			NS	

Data are presented as mean ± SD unless stated otherwise * p<0.05 ** p<0.01, # total patient numbers do not add up to 396 since weakly positive patients were left out of the analysis; Tg-Ab = antibodies against thyroglobulin; TPO-Ab = antibodies against thyroid peroxidise; PCA = antibodies against parietal cells; ACA = antibodies against adrenal cortex; F = female; DM = diabetes mellitus; hyperthyroidism = hyperthyroidism without thyroid stimulating antibodies; Graves = Graves' disease; PA = pernicious anaemia; Addison = Addison's disease; PPV = positive predictive value; NPV = negative predictive value; AB^+ vs AB^- = level of significance for the difference in organ dysfunction frequency between AB-positive and AB-negative patients.

ANTIBODY PREVALENCES AND ORGAN DYSFUNCTION

Altogether, 396 patients were tested for Th-Ab, PCA and ACA. All patients had islet cell antibodies (ICA), since this was obligatory for the diagnosis of DM1. Of the patients tested for Tg-Ab, 4.3% were positive. 60.0% of the Tg-Ab-positive patients tested had organ dysfunction (PPV 0.60, NPV 0.88). In patients positive for Tg-Ab, the occurrence of organ dysfunction was significantly higher than in patients negative for those antibodies (60.0 *vs* 11.7%, p<0.001).

Of the patients tested for TPO-Ab, 8.1% were positive; 53.4% of the TPO-Ab positive patients tested had organ dysfunction (PPV for hypothyroidism was 0.53, NPV 0.91). This was significantly higher than in patients negative for TPO-Ab (53.4 *vs* 9.4%, p<0.001).

Of the patients, 10.4% were positive for either TPO-Ab, Tg-Ab, or both. Of these patients, 52.9% had organ dysfunction at testing (PPV 0.53, NPV 0.91), which was significantly higher than in patients negative for these antibodies (52.9 *vs* 9.1%, p<0.001).

Of the patients tested for PCA, 5.8% were positive; 60.8% had organ dysfunction (PPV 0.61, NPV 0.90). In patients positive for PCA, the occurrence of organ dysfunction was significantly higher than in patients negative for those antibodies (60.9 *vs* 9.7%, p<0.001).

Of the patients tested for ACA, two were ACA positive. None of them had signs of adrenal dysfunction.

Fifteen patients had multiple antibodies: nine had Th-Ab (either TPO-Ab, Tg-Ab, or both) and PCA, two had Th-Ab and ACA and four had Th-Ab (and, like all included patients, ICA). However, none of these patients had the combination of different types of organ dysfunction leading to the clinical diagnosis of one of the polyglandular syndromes.

Table 1 shows antibody prevalences and organ dysfunction in all patients tested. *Figures 1 and 2* show the occurrence of different types of organ dysfunction in patients positive for thyroid or parietal cell antibodies, compared with patients negative for those antibodies.

There was a female predominance for Tg-Ab and TPO-Ab (p<0.001), and for PCA a tendency towards female predominance (p=0.06). The two ACA positive patients were female.

DISCUSSION

As shown in our recent review,[10] most studies performed in the past to investigate the relevance of organ-specific antibodies in DM1 used a cross-sectional design; no longitudinal studies have been performed to date. In order to investigate the predictive value of these OS-Ab in

Figure 1. *Difference in prevalence of thyroid dysfunction in patients negative (Th-AB-) and positive (Th-AB+) for thyroid antibodies (p<0.001)*

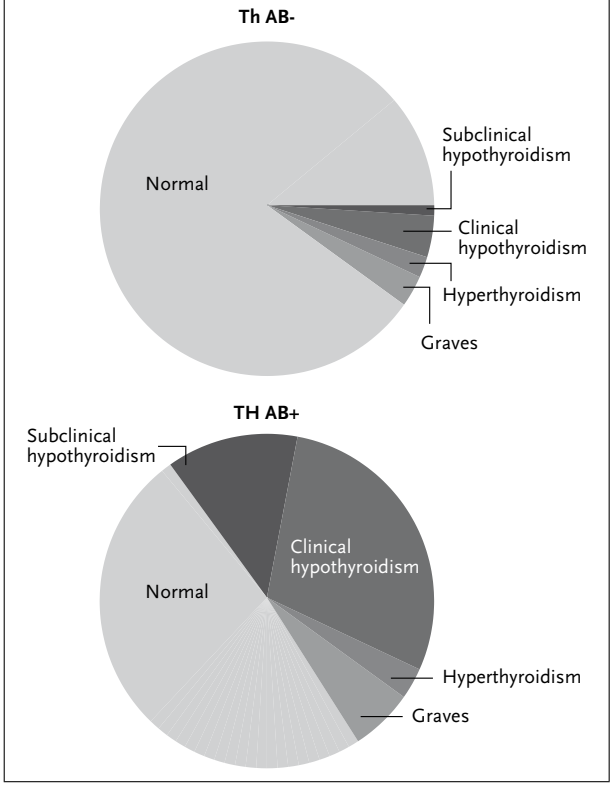
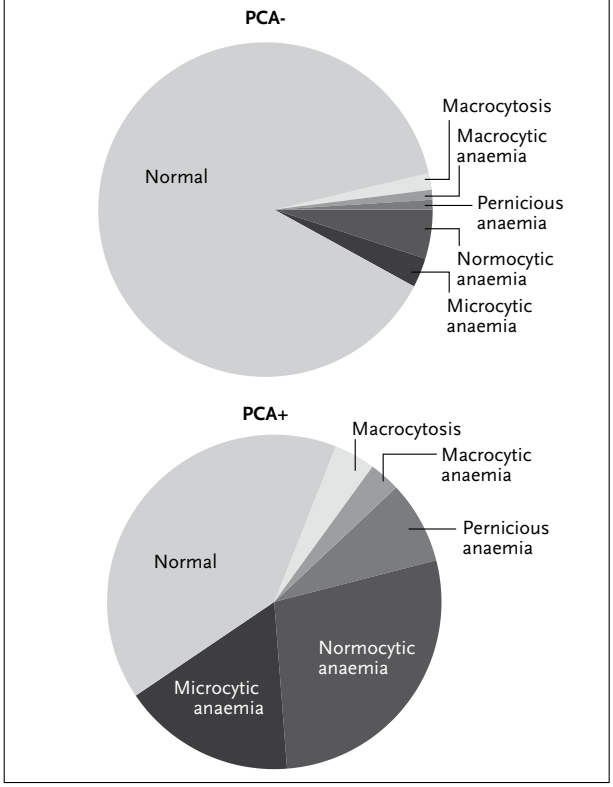


Figure 2. *Difference in prevalence of macro-, normo- and microcytic anaemia in patients negative (PCA-) and positive (PCA+) for parietal cell antibodies (p<0.001)*



DM1 patients, we performed a retrospective longitudinal study on the prevalence and clinical relevance of thyroid antibodies, parietal cell antibodies and adrenocortical antibodies. We report on the presence of OS-Ab and on the development of corresponding organ dysfunction during more than 20 years follow-up of 396 patients with DM1, median follow-up being comparable for antibody positive and negative patients.

As expected, the frequency of (subclinical) hypothyroidism, macrocytic haematological profile and different types of anaemia was significantly higher in DM1 patients with than in DM1 patients without thyroid and gastric antibodies.

Among our population of DM1 patients, the organ most frequently affected by OS-Ab was the thyroid gland. Of all the patients, 10.4% tested were positive for thyroid antibodies, which was within the range of prevalence found by other authors.[5,9,11-31] TPO-Ab were more frequent than Tg-Ab, which is also in accordance with the literature. The prevalence of hypothyroidism was significantly higher among Th-Ab positive patients than among Th-Ab negative patients and this was true for both Tg-Ab and TPO-Ab.

The PCA prevalence in our DM1 population was 5.8%, which was within the range of prevalence found by other authors.[11,13,20,22,24,25,27,46-50] Of 23 PCA-positive patients, 9% had a macrocytic blood picture, 9% pernicious anaemia and 43% had normocytic or microcytic anaemia, which was significantly higher than in PCA-negative patients.

In accordance with previous studies, we found a low prevalence of ACA (0.5%) in our DM1 population. Only two patients were ACA positive, both without signs of adrenal dysfunction. The low prevalence of ACA in our population makes it impossible to determine the predictive value of these antibodies, but high positive predictive values have been reported in the literature.[10,16,46,55,58,59,61]

In summary, this study is the first to investigate the long-term clinical relevance of organ-specific antibodies in DM1 patients in a longitudinal manner. The presence of thyroid and parietal cell antibodies is associated with an increased risk of developing (sub)clinical hypothyroidism and different types of anaemia.

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CASE REPORT

An illustrated case of doxorubicin-induced radiation recall dermatitis and a review of the literature

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ABSTRACT

Radiation recall dermatitis (RRD) is a rare cutaneous reaction occurring within a previously irradiated field, precipitated by certain drugs. A case of RRD most likely induced by doxorubicin is presented and illustrated together with a review of the literature.

KEYWORDS

Recall dermatitis, radiotherapy, doxorubicin

WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT?

Radiation recall dermatitis (RRD) is defined as the appearance of skin reactions in previously irradiated skin after the administration of certain response-inducing drugs. The incidence of RRD is poorly documented but generally this condition is regarded as rare. The first published case, by D'Angio and colleagues in 1962,[1] was triggered by dactinomycin. Some medications have been documented to be more commonly associated with RRD, such as doxorubicin, gemcitabine, and docetaxel. Here we present a case of RRD most likely induced by doxorubicin.

CASE REPORT

Patient and tumour characteristics and radiotherapy regimen

A 44-year-old female patient was treated for breast cancer. She was not taking any medication or alcohol; she smoked 10 to 15 cigarettes daily. Her medical history revealed episodes of eczema for many years for which she had frequently used topical corticosteroids. Slight atrophy of the skin as a result of this was predominantly seen on her hands, but not on her breast. The tumour was a 2.2 cm large grade III infiltrating ductal carcinoma, the receptors for oestrogen and progesterone were both positive, HER2/neu was negative. Axillary dissection showed ten nodes, six of these were invaded by metastases. The axillary apex node was negative. An ultrasound of the liver, a chest X-ray and a bone scintigraphy were normal. Six weeks after breast-conserving surgery, she was irradiated to the left breast, axilla and internal mammary chain, with a boost to the lumpectomy area by a simultaneous integrated boost technique. An elective dose of 50.4 Gy in 28 once-daily fractions of 1.81 Gy was delivered by 6 MV photons without bolus material to the skin by intensity modulated radiotherapy (IMRT) treatment planning. The boost area received 28 fractions of 0.49 Gy by 8 MeV electrons with 0.5 cm bolus material (cumulative dose boost area: 64.4 Gy). She was checked weekly during irradiation and a grade I erythema developed in the 5th week, only to the skin of the axilla but not to the breast.

Exactly 14 days after completion of radiotherapy she started her adjuvant chemotherapy. At the start of chemotherapy, the slight erythema had completely resolved. For personal reasons she decided to photographically document her skin condition prospectively.

Chemotherapy

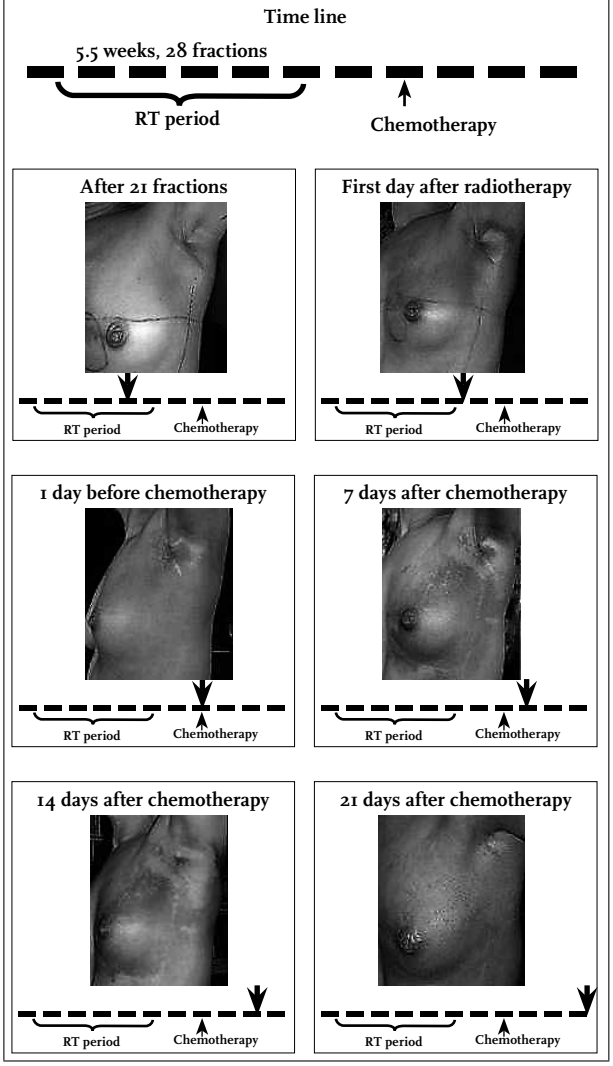
As adjuvant chemotherapy she received TAC in a three-weekly schedule; taxotere, doxorubicin and cyclophosphamide. A dose of 120 mg docetaxel (Taxotere®), 800 mg cyclophosphamide (Endoxan®) and 80 mg doxorubicin was administered.

Epicrisis

Two days after the administration of the first TAC course she started to experience a general malaise consisting of feeling cold and shaking. A severe pain developed in the irradiated area. On day 4 after administration of TAC chemotherapy, the irradiated skin started to show an erythema with a purplish aspect. A patchy superficial epidermiolysis developed in the affected skin designated as grade III.[2] The pain subsided after five days, the skin reaction started to recover after 14 days.

Since the indication of adjuvant chemotherapy was considered to be crucial,[3] (adjuvant online), she was encouraged to continue without doxorubicin. On day 22 the second chemotherapy course was administered consisting of TC. The docetaxel dose remained 120 mg, but the cyclophosphamide was escalated to 975 mg. Neither the pain nor the skin reaction reoccurred. Subsequent courses could be administered without reappearance of the recall phenomena. The appearance of the skin over time is depicted in *figure 1*.

Figure 1. *The evolution over time of the skin reaction with respect to the overall treatment time of radiotherapy and the first adjuvant chemotherapy*



DISCUSSION AND REVIEW OF THE LITERATURE

RRD (radiation recall dermatitis) involves the appearance of skin reactions in previously irradiated skin in response to the administration of several drugs, as described in *table 1*. Although the number of published cases is low, there is probably an underscoring of its true incidence. Furthermore, patients need to have had radiotherapy first and therefore this condition is only described in an oncological setting. The severity of the skin reaction is described by the Common Toxicity Criteria version 4.0.[2]

Table 1. *Literature review on drugs probably involved in radiation recall dermatitis*

Drug	References
Doxorubicin and pegylated liposomal doxorubicin	9, 15-18
Docetaxel	6, 19-27
Gemcitabine	20, 28-36
Paclitaxel	37, 38
Methotrexate	39, 40
Tamoxifen	41-44
Dactinomycin	1
Vinblastine	45
Carboplatin	30, 37
Dacarbazine	46
Cetuximab	47
Cyclophosphamide	48, 49
Capecitabine	50-52
Others	Bevacizumab[53], Trastuzumab[54], Pemetrexed[55-57], Gatifloxacin[58], Levofloxacin[59], Lanreotide[60], Gefitinib[61], Arsenic trioxide[62], Interferon alfa-2b[63], Oxaliplatin[64,65], Idarubicin[10], Simvastatin[66], Bleomycin[67]

TEMPORAL RELATIONSHIP BETWEEN USE OF DRUGS AND END OF RADIOTHERAPY

Camidge and Price[4] have further defined the clinical entity and have made a clear separation between radiosensitisation and RDD. They suggest designating any reaction occurring within seven days after administration of drugs as sensitisation and not as RRD. In our case the interval was 14 days and can, by this Camidge definition, therefore be called RDD. Although this interval of two weeks can be considered relatively short, much longer intervals have been described. To the best of our knowledge, the longest reported interval for RDD is seven years by Mayer *et al.*[5] Burdon *et al.* have reported a 15-year interval for adriamycin-induced stomatitis.[6]

PATHOGENESIS

Although the precise mechanism of action for RRD is not known, several mechanisms have been proposed including changes in vascularisation, DNA repair, radiation-impaired epithelial function of stem cells, increased stem cell sensitivity, and increased sensitivity to drugs.[4,7] None of these hypotheses have been proven. Furthermore, the recall phenomena are not only seen in the skin, but also on mucosa and other internal organs.[8-11]

RADIOTHERAPY CHARACTERISTICS

There is no relationship between the occurrence of RDD and the applied radiation dose. Therapeutic schedules well below 20 Gy have been described to elicit RDD as well.[4]
It is a fact that the incidence of radiation-induced skin reactions in the case of breast-conserving therapy predominantly depends on breast volume, beam energy and the use of IMRT.[12] In the patient we have presented the breast was relatively small and she was treated by 6 MV photon IMRT. Based on these characteristics the anticipated chance of moist desquamation was low. Indeed during and in the first two weeks after radiotherapy skin reactions were very mild and desquamation did not occur. The severe adverse effects on the skin were only seen directly after the start of adjuvant chemotherapy. This makes the radiotherapy itself a very unlikely cause of her malaise.

DRUGS ASSOCIATED WITH RRD

Besides chemotherapeutic agents, many more drugs (such as antibiotics, monoclonal antibodies, biological response modifiers) are also able to elicit RRD, but few are administered in close temporal relation to radiotherapy.[13] Several drugs are enumerated in *table 1*.

THERAPEUTIC INTERVENTIONS

There are no proven interventions to relieve symptoms or to enhance recuperation. Once the RDD has occurred almost all reports advise to discontinue the triggering drug. However, a rechallenge does not always result in the occurrence the skin reactions.[4]
Care must be taken when studying the literature on this subject. There are many studies on how to reduce acute radiation skin reactions with creams, amifostine, N-acetylcysteine, etc.[14] But there are no studies on how to manage the skin that was intact after completion of radiotherapy and subsequently developed RDD. Caloglu *et al*. have produced an algorithm on how to manage the recall phenomena.[7] In case of 'severe' reactions they suggest prescribing systemic or topical steroids, nonsteroidal anti-inflammatory agents, and antihistamines. However, the available scoring system[2] subdivides RDD into the usual Grades I to V (there is no 'severe' in the CTC version 4.0) and there is no evidence for these agents.
In conclusion, this case report adds to the already existing knowledge that RRD is a rare though possibly underreported event. To the best of our knowledge, this is the first published case with a prospectively collected series of photographs on the evolution of skin reactions both during the radiotherapy course and the subsequent RDD occurrence and resolution.

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CASE REPORT

Herpes simplex virus oesophagitis in a pregnant woman

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ABSTRACT

Herpes simplex virus (HSV) oesophagitis is well described in immunocompromised patients. In immunocompetent individuals HSV oesophagitis is rare. We present a case of HSV oesophagitis in a pregnant woman. A possible explanation for HSV oesophagitis during pregnancy is the decreased cellular immunity, leading to an increased frequency and severity of viral infections. Antiviral therapy is advocated in pregnancy.

KEYWORDS

Herpes simplex virus, oesophagitis, pregnancy

What was known on this topic?
Herpes simplex virus oesophagitis is a well-known phenomenon in immunocompromised hosts. In immunocompetent individuals this condition is rare.

What does this add?
We present a case of herpes simplex virus oesophagitis in a healthy, pregnant woman, which has never been described before. A possible explanation is the decreased cellular immunity during pregnancy, leading to an increased frequency and severity of viral infections.

INTRODUCTION

Herpes simplex virus (HSV) oesophagitis is a well-known phenomenon in immunocompromised hosts. In immunocompetent individuals this condition is rare. HSV oesophagitis during pregnancy has never been described before in the English literature. We present a case of a pregnant woman with HSV oesophagitis, together with an overview of the characteristics of HSV oesophagitis, within the scope of pregnancy.

CASE REPORT

A 25-year-old woman, gravida 2 para 0, was admitted to our hospital at 22 5/7 weeks gestation with a four-week history of epigastric pain. One week before hospitalisation she noted fever, together with aggravation of the epigastric pain radiating to the back. Intake of food was hampered by odynophagia and vomiting. The medical history revealed nephrolithiasis. Antacids and laxative suppositories were prescribed for the symptoms, next to antibiotics because of a supposed cystitis. Advanced ultrasonography at 20 weeks had shown no signs of congenital abnormalities.
Physical examination revealed a tachycardia of 104 beats/min and a body temperature of 39.2 °C. There was epigastric tenderness on palpation. Blood testing showed elevated inflammatory markers: leucocytes 13.4 /nl and C-reactive protein 101 mg/l. There were no signs of preeclampsia or HELLP syndrome. Ultrasonography of the upper abdomen showed no abnormalities. Oesophagogastroduodenoscopy revealed mucosal erythema and multiple fibrin exudates of the entire oesophagus, which became partially confluent in appearance (*figure 1*). Biopsies were taken for histopathological analysis and culture. Histopathology showed multinucleated giant cells and Cowdry's type A inclusion bodies (*figure 2*). Immunohistochemical stains were positive for HSV (*figure 3*), fungal stains were negative. HSV type 1 was cultured from the oesophageal tissue.

Figure 1. *Oesophagogastroduodenoscopy showing HSV oesophagitis. There is extensive ulceration throughout the oesophagus, volcano-like in appearance, with raised edges*

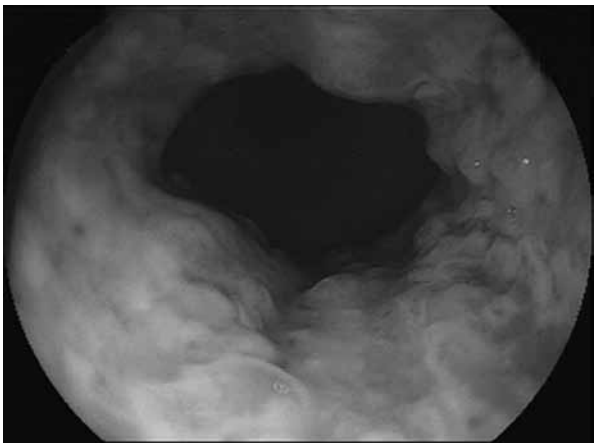


Figure 2. *Histopathology of HSV oesophagitis. There are multinucleated giant cells showing characteristic nuclear inclusion bodies*

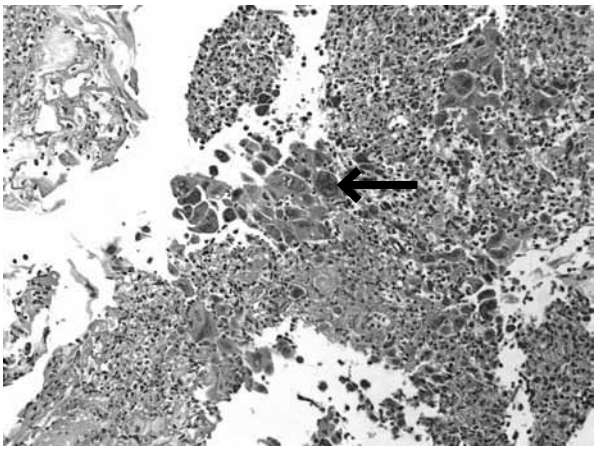
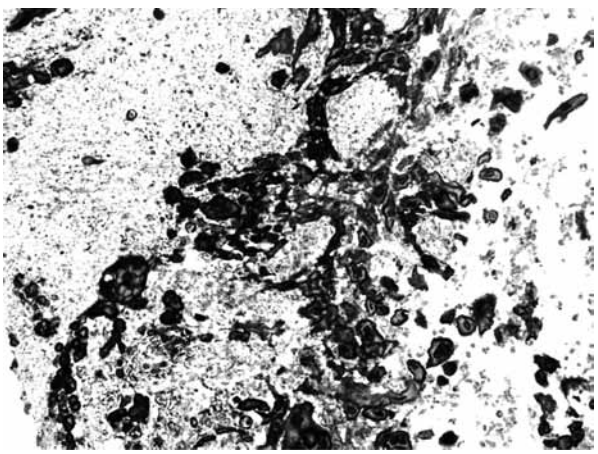


Figure 3. *Positive immunohistochemical stains for HSV in the oesophagus*



She was treated with acyclovir intravenously for seven days (15 mg/kg in four doses). Within two days she was able to drink and eat a soft diet. Complete relief of symptoms was achieved in five days. She was discharged from the hospital on the 13th day. Serology for HSV showed seroconversion, indicating primary infection. The patient had no history of HSV infection and no genital lesions were seen during gynaecological inspection. Retrospectively, her partner had had possible orolabial HSV lesions several weeks before, indicating a possible source of infection. Human immunodeficiency virus serology was negative.

At 41 2/7 weeks gestation she delivered a healthy daughter of 3280 gram with Apgar scores of 9 and 10 at one and five minutes, respectively. No congenital abnormalities were noticed. At follow-up three months postpartum she had normal levels of serum immunoglobulins. T-lymphocyte quantitation revealed high-normal values: CD4 lymphocyte count 2.15 x 10^9/l (0.56-1.49 x 10^9/l), CD8 lymphocyte count 1.17 x 10^9/l (0.26-0.99 x 10^9/l). The CD4/CD8 lymphocyte ratio was 1.84 (normal).

DISCUSSION

HSV type 1 (HSV-1) and 2 (HSV-2) belong to the human herpes viruses.[1] Orolabial herpes infections are usually caused by HSV-1.[2] As a consequence HSV oesophagitis is also predominantly caused by HSV-1 while HSV-2 is mostly responsible for genital herpes infections.[2,3]

The incidence of HSV oesophagitis is 1.8% in autopsy patients.[4] Most of these patients were immunocompromised. In immunocompetent individuals HSV oesophagitis is rare. HSV oesophagitis may represent primary disease or reactivation of a latent infection. The characteristic patient is a young adult male with the acute onset of the triad odynophagia, retrosternal chest pain and fever.[3] Prior exposure to a family member with possible HSV lesions has been reported in about 20% of the cases.[3,5]

Oesophagogastroduodenoscopy is the diagnostic procedure of choice. HSV oesophagitis has typical endoscopic findings.[5] In the early stage vesicles are seen, which then deepen and confluence to form discrete, circumscribed ulcers with raised edges. These lesions may have a punched-out or volcano-like appearance. Cobblestoning can be seen when the lesions coalesce. Exudates are present in a substantial number of patients. Biopsies from the ulcer edges should be obtained for histopathology and viral culture to make the definitive diagnosis. The histological characteristics of HSV oesophagitis are ground-glass appearance in the nuclei, Cowdry type A nuclear inclusion bodies and multinuclear cells.[6]

During pregnancy significant changes occur in the maternal immune system. The alterations are initiated at the placental site by the hormones of pregnancy. Sridama *et al.* demonstrated a decrease of CD4 lymphocytes throughout pregnancy which is responsible for the maternal immunodeficiency.[7] This mechanism may be important for the protection of the foetus. However, due to the decreased levels of CD4 lymphocytes, pregnant women have a higher risk of viral infections and severity of symptoms.[1,7] After pregnancy, the CD4 lymphocyte levels normalise within five months.

In case of a primary HSV infection in pregnancy the risk of intrauterine transmission is less than 5%.[1] After intrauterine transmission there is an increased risk of disseminated infection of the foetus which may lead to early embryonic or foetal damage, intrauterine growth restriction, preterm birth, major developmental or congenital anomalies, or even miscarriage or stillbirth.[1]

Treatment of HSV oesophagitis with acyclovir, a nucleoside analogue, is well established in immunocompromised patients.[8] In immunocompetent individuals HSV oesophagitis is generally a self-limiting disease. However, to prevent complications such as bleeding or perforation and to hasten recovery, it is advisable to initiate antiviral therapy in an early stage of disease also in immunocompetent patients, particularly in those with severe odynophagia.[8,9]

The essence of treatment of HSV infections in pregnancy should be prevention of vertical transmission of HSV to the foetus or neonate. Acyclovir crosses the placenta and is excreted by the foetal kidney, but there is no accumulation in the foetus.[10,11] Stone *et al.* have studied the effects of systemic acyclovir administration during pregnancy.[12] The risk of birth defects of live births exposed to systemic acyclovir during first trimester is 3.2% and for exposures during any trimester is 2.6%. As the expected rate of congenital abnormalities in the general population is 3.2%, no difference is seen in the acyclovir versus the general population and acyclovir is therefore classified as a category B drug.[1] Pregnant women are relatively immunocompromised and therefore antiviral therapy is indicated in case of HSV oesophagitis.

In conclusion, HSV oesophagitis is a rare occurrence in pregnancy. It should be considered in patients with odynophagia, retrosternal chest pain and fever. Oesophagogastroduodenoscopy with biopsy for histopathology and viral culture should be performed to confirm the diagnosis. Antiviral therapy is advocated in pregnancy.

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CASE REPORT

Elevated plasma creatinine due to creatine ethyl ester use

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ABSTRACT

Creatine is a nutritional supplement widely used in sport, physical fitness training and bodybuilding. It is claimed to enhance performance. We describe a case in which serum creatinine is elevated due to the use of creatine ethyl esther. One week after withdrawal, the plasma creatinine had normalised.

There are two types of creatine products available: creatine ethyl esther (CEE) and creatine monohydrate (CM). Plasma creatinine is not elevated in all creatine-using subjects. CEE, but not CM, is converted into creatinine in the gastrointestinal tract. As a result the use of CEE may be associated with elevated plasma creatinine levels. Since plasma creatinine is a widely used marker for renal function, the use of CEE may lead to a false assumption of renal failure.

KEYWORDS

Creatine, creatinine, bodybuilding

INTRODUCTION

The use of nutritional supplements continues to increase in sports and physical fitness, with creatine as one of the most widely used substances. Among visitors to Dutch fitness centres, 3.2% had used creatine supplementation in the past year.[1] Creatine is claimed to increase muscle strength and is most popular among bodybuilders. It is considered a harmless product and is not regarded as a performance enhancing drug by the *World Anti Doping Agency*. We describe a patient who had a substantial elevation of his plasma creatinine level, which normalised after withdrawal of the creatine-containing supplement he was using. Furthermore we explain why serum creatinine is not elevated in all subjects using creatine.

CASE REPORT

A 38-year-old man visited our outpatient clinic because he was worried about his fertility after years of anabolic steroids usage. One year before oligoasthenoteratospermia was diagnosed. His medical history was otherwise unremarkable. He had not been using anabolic steroids over the past two years. In the seven years before, he had been taking various types of anabolic steroids, stacked (eq. combining different anabolic steroids) mostly in cycles lasting on average three months with occasional use in between cycles. At first presentation he had only been using protein shakes and creatine powder for a week (creatine ethyl esther). He exercised four times a week for one and a half hour, mostly strength training. He did not mention any mental or physical complaints.

Physical examination was unremarkable except for a very muscular appearance. Routine blood tests showed an elevated creatinine (247 μmol/l; reference range 64 to 104 μmol/l), urea (9 μmol/l; reference range 3 to 7.5 μmol/l) and creatine kinase (227 μmol/l; reference range 0 to 170 μmol/l) levels. The results of other blood tests (including a complete blood count, liver enzymes and electrolytes) were within normal limits. The urine screening test was negative for haemoglobin, leucocytes or protein. Ultrasound of the kidneys showed normal sized kidneys without signs of hydronephrosis or kidney stones.

We advised the patient to stop the creatine and protein shakes and repeated blood tests six days later. The plasma creatinine levels had normalised to 76 μmol/l and the urea to 5.5 μmol/l. We concluded that the elevated plasma creatinine level was due to oral supplementation with creatine.

In addition we measured serum creatine levels. Creatine was mildly elevated while using creatine (61 μmol/l; reference range 6 to 50 μmol/l); after withdrawal the level normalised to 38 μmol/l.

DISCUSSION

In the presented case, the plasma creatinine level was elevated due to oral supplementation with creatine ethyl esther. Creatine is a supplement widely used by athletes, mostly amateur bodybuilders. It is not on the World Anti Doping Agency's list of banned substances and can be easily obtained via internet, in fitness centres and in health or sport shops. Creatine is not considered a pharmaceutical drug and therefore, its use is not always mentioned by a patient, nor is it asked for by the doctor.

Creatinine is a widely used measure to estimate renal function. It is a chemical waste molecule that is generated from creatine phosphate metabolism by skeletal muscle.

Plasma creatinine concentration is equal to its production rate divided by the metabolic clearance rate. The plasma creatinine concentration is a well established marker of renal function because its production rate is fairly stable and it is exclusively eliminated by renal excretion.[2] The tubular secretion of creatinine is limited and there is little to no tubular reabsorption. Clinicians intuitively associate elevated creatinine levels with impaired renal function. In most cases, this is correct and further tests to explore the cause of renal dysfunction are justified. However, our case illustrates that, in rare cases, plasma creatinine levels may be elevated in individuals with normal renal function due to an elevated 'production' of creatinine.

Creatine (α-methyl guandino-acetic acid), which was first identified in 1832, is a nitrogenic composite which is manufactured in the liver, and to lesser extent in the kidneys and pancreas, from three amino acids: methionine, arginine and glycine.[3] Creatine is also obtained through the diet, mainly by consuming fish and red meat. Of the body creatine pool 95% is located in skeletal muscle, the remaining 5% is located in liver, brain, kidneys, and testes.[4]

Creatine can be converted to phosphocreatine by binding inorganic phosphate through the reversible reaction of creatine kinase. Adenosine diphosphate (ADP) and phosphocreatine provide adenosine triphosphate (ATP) and creatine (*figure 1*). Energy is provided to the body from the hydrolysis of ATP into ADP. Creatine supplementation is thought to increase skeletal muscle's ability to resynthesise ATP from ADP and in theory this could explain the presumed increase in muscle strength by supplementation of creatine.[5-7] Studies aimed to determine the effect of creatine supplementation on muscular strength or performance show conflicting results.[8-11]

It has been described before that circulating levels of creatinine may be increased in users of creatine supplements.[12-14] There are two types of creatine supplements widely available: creatine monohydrate (CM) and creatine ethyl esther (CEE). CEE is converted to CM by modifying an acid moiety through ester bond attachment. This can be achieved by solvating CM in dry ethanol in an acidic atmosphere. CEE is claimed to have a better solubility in lipids leading to higher absorption rates.[15] However, in contrast to CM, CEE can be converted to creatinine in the gastrointestinal tract. This is supported by the results of a study published by Spillane *et al.* who measured serum creatine and creatinine levels in healthy individuals at various time points after oral ingestion of CEE, CM or placebo. Serum creatine increased 1.5 fold six days after ingestion of CM, but only marginally after ingestion of CEE. Creatinine levels, on the other hand, approximately tripled after ingestion of CEE and only marginally increased after ingestion of CM.[11] The mean increase in serum creatinine levels in the group that used CEE in this study corresponded to the increase seen in our patient.

In conclusion, elevated serum creatinine does not always indicate impaired renal function. The use of CEE, but not CM, may lead to a harmless elevation of plasma creatinine levels and may provoke unnecessary concern. We advise to pay particular attention to the use of supplements in patients, and to explicitly ask for the use of creatine in professional or amateur athletes. The use of CEE should be stopped six days prior to blood sampling.

Figure 1. *The reversible conversion of creatine into phosphocreatine by binding inorganic phosphate with creatine kinase as catalyst. The reverse reaction provides ATP from ADP*

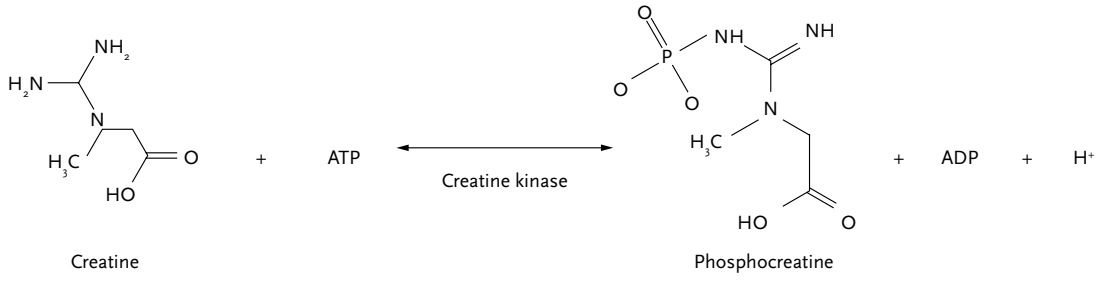

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PHOTO QUIZ

Blistering of the hand in a breast cancer patient

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CASE REPORT

A 48-year-old patient presented with redness and tenderness of the dorsal side of her left hand, which had started two to three days after administration of the third cycle of vinorelbine, which she was receiving as palliative treatment for breast cancer. A week afterwards blisters developed (*figures 1 and 2*).

She was diagnosed with breast cancer in 2005, and in 2007 she developed multiple metastases in ovary, bone and liver. Previously, she had been treated with three lines of hormonal therapy and three lines of chemotherapy. Her medical history was otherwise unremarkable. She had never experienced skin problems before. During administration of vinorelbine no pain or swelling of the hand was noticed.

Figure 1.

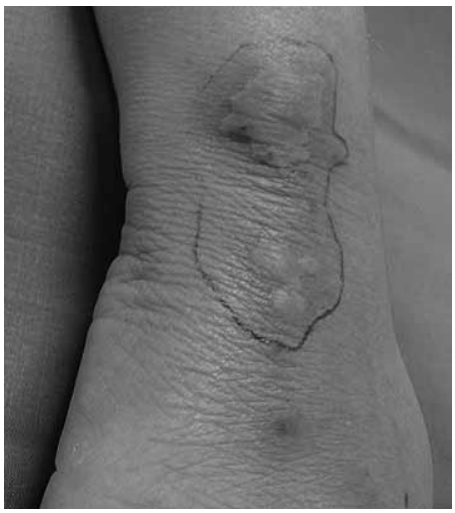
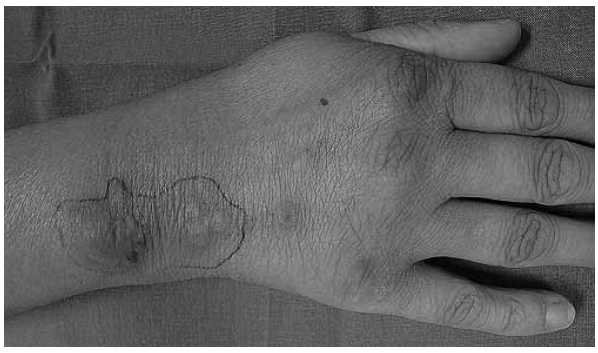


Figure 2.



WHAT IS YOUR DIAGNOSIS?

See page 85 for the answer to this photo quiz.

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A crackling handshake

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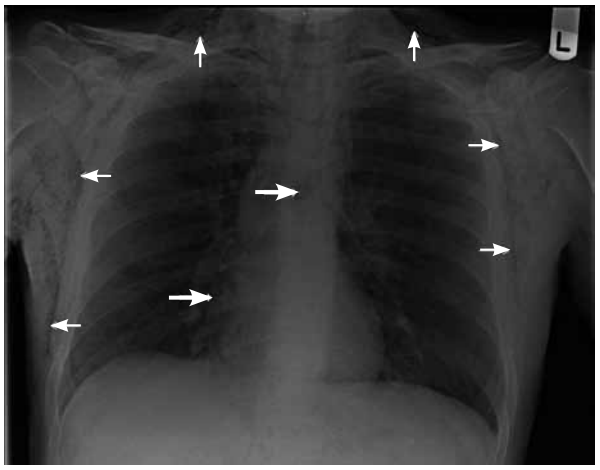
CASE REPORT

A 27-year-old man was admitted with excessive vomiting two days after completing his final regimen of adjuvant chemotherapy (bleomycin, etoposide, cisplatinum) for testicular carcinoma (pT1N2M0, stage IIB). He had been vomiting for several weeks despite antiemetics. A remarkable crepitus was noticed upon shaking the patient's hand. At further physical examination there was crackling of the skin of the entire right arm, chest and neck. The patient did not have any fever. Examination of the heart and lungs was unremarkable. Besides a slight leucocytopenia and thrombocytopenia, laboratory investigation revealed no further abnormalities. *Figure 1* shows the chest X-ray at presentation.

Figure 1. *Chest X-ray: subcutaneous emphysema of the lateral chest wall (small arrows) with signs of pneumomediastinum (large arrows). Also visible are slight paracardial pulmonary interstitial changes*



WHAT IS YOUR DIAGNOSIS?

See page 86 for the answer to this photo quiz

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PHOTO QUIZ

Slipped capital femoral epiphysis as manifestation of a rare endocrinological disease

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CASE REPORT

Patient A presented at the age of 15 to the emergency department with acute hip complaints after a fall. At the age of six years, nodules were removed from her tongue. Examination revealed a flexion, abduction and exorotation contracture of the left hip with a decreased range of motion. Radiology confirmed a slipped capital femoral epiphysis (SCFE) of the left hip (*figure 1*). The SCFE was surgically treated with cannulated screw fixation. Peroperatively the blood pressure remained stable.

A few months later the patient was referred to an ENT specialist because of a nodule in the neck. Fine needle aspiration of the lymph node revealed medullary thyroid carcinoma. The patient had thickened lips, nodules on the tongue and a marfanoid appearance (*figure 2*).

Patient B visited an orthopaedic surgeon at the age of 16 because of pain of the left hip on exertion for the last six months. On examination left hip flexion was diminished; however, abduction and adduction were within the normal range. Radiology of the left hip showed that the capital femoral epiphysis had slipped in a dorsomedial direction. Surgical treatment with a cannulated screw fixation was performed successfully.

At the age of 17, the patient returned with an SCFE of the contralateral hip. Radiology confirmed a slip of the right epiphysis of the femoral head. No complications occurred during screw fixation.

Preoperatively, a nodule in the thyroid had been noticed for which the patient was referred to a paediatrician. Thin needle biopsy showed no malignancy. Because of progressive growth of this tumour in the following years, which resulted in cosmetic complaints, the patient underwent a right hemi-thyroidectomy. Pathological examination surprisingly identified medullary thyroid carcinoma.

WHAT IS YOUR DIAGNOSIS?

See page 94 for the answer to this photo quiz.

Figure 1. *A typical example of an acute slipped capital femoral epiphysis*

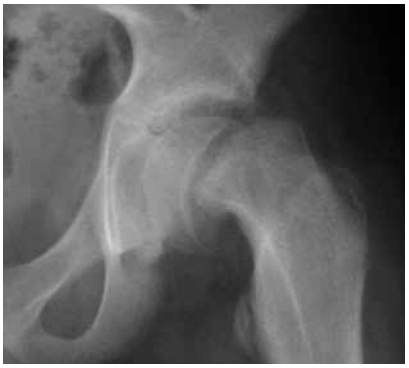
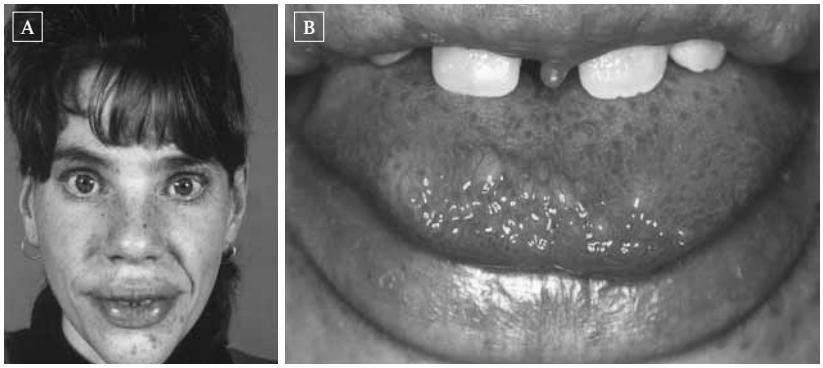


Figure 2. *a) A patient with neurofibromas of the tongue and eyelids, thick lips and marfanoid phenotype. b) Close-up of neurofibromas of the tongue (permission granted)*



ANSWER TO PHOTO QUIZ (PAGE 82)

BLISTERING OF THE HAND IN A BREAST CANCER PATIENT

DIAGNOSIS

Despite the absence of pain or swelling during infusion of vinorelbine, extravasation of vinorelbine is the most likely cause of the skin lesions. Extravasation trauma can be caused by hypertonic solutions, vasoconstrictive solutions and irritating solutions such as chemotherapeutic agents. The reported incidence of extravasation in intravenously administrated chemotherapy ranges from 0.01 and 6%.[1,2] Vinca alkaloids, such as vinorelbine, are classified as vesicant. Vesicants are chemical compounds that have the ability to cause chemical burns with severe skin damage and blistering. In some cases extravasation leads to skin necroses, with histopathology showing separation of dermis from necrotic epidermis.[3]

When discovered early, extravasations can be treated with dilution by subcutaneous flushing with saline. This is preferably done within the first eight hours, but an attempt can still be made within the first 24 hours. In addition, hot packs can help to prevent damage in vinca alkaloids, non-DNA binding vesicants, while cold packs have shown to increase damage in animal studies. In contrast, cold packs can prevent further damage in case of extravasation of DNA-binding vesicants, such as anthracyclines and taxanes.[2]

In our case the patient did not complain of swelling or pain during infusion. Her complaints started after two days and blistering appeared after nine days. Usually, the most severe extravasation damage is noted days to weeks after extravasation has taken place. Because there were no symptoms during intravenous administration and the amount of affected skin was limited, probably just a small amount of vinorelbine had extravasated.

As differential diagnosis, non-chemical skin burns and paraneoplastic syndromes such as neutrophilic dermatosis of the dorsal hands (variation of Sweet's syndrome) should be considered. However, our patient had no history of skin burns. Neutrophilic dermatosis usually presents with pustules rather than blisters and is associated with haematological malignancies rather than breast cancer.[4]

The patient was referred to the plastic surgeon for conservative treatment. The wound was treated as a second-degree burn. The wound was dressed with impregnated gauzes, Cuticerin, 7.5 x 7.5 cm and a compress. Her hand recovered well in three weeks.

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ANSWER TO PHOTO QUIZ (PAGE 83)

A CRACKLING HANDSHAKE

DIAGNOSIS

The air in the mediastinum was initially thought to originate from a rupture of the oesophagus due to ongoing vomiting. Upon gastroscopy oesophagitis grade C was seen but no visible oesophageal rupture. Another explanation for air in the mediastinum can be a rupture of the bronchus, but this was thought to be unlikely. The patient was treated with meropenem and fluconazole as mediastinitis could not be ruled out. He recovered from the subcutaneous emphysema and was discharged from hospital. A few months later the patient complained of progressive dyspnoea. He was admitted to the intensive care unit because of renal and respiratory failure and required mechanical ventilation. Progressive pulmonary interstitial abnormalities with pleural effusion were seen on the CT scan of the chest. Under the probable diagnosis of bleomycin-induced pneumonitis (BIP) with superimposed pulmonary infection, he was treated with high-dose corticosteroids and broad-spectrum antibiotics. Therapy was unsuccessful and the patient died a few days later. Section was not performed. The possibility of bleomycin-induced lung injury as the cause of the subcutaneous emphysema and pneumomediastinum had not been considered at the initial presentation.

The pulmonary toxicity of bleomycin has been recognised for many years and has various forms of presentation. The first fatal case of subcutaneous emphysema and pneumomediastinum as the initial presentation of BIP has recently been described.[1] Bleomycin therapy can cause damage to tissues missing the enzyme bleomycin hydrolase, including the skin and lung.[2] It is thought that free radicals and cytokines cause endothelial damage leading to the pulmonary changes. The prevalence is estimated at 2 to 40% in patients receiving bleomycin. Mortality is approximately 2%. Pneumomediastinum without pneumothorax is extremely rare.[3] Risk factors for developing BIP are poor renal function, age over 40, stage IV disease and cumulative bleomycin doses of more than 300 mg.[4] BIP can develop during therapy, but has also been reported up to six months after discontinuation of bleomycin therapy. Symptoms and findings are non-specific, including dyspnoea and dry cough. Although hard evidence is lacking, numerous case reports suggest that treatment consisting of high-dose prednisolone is favourable.

Symptoms of extensive subcutaneous emphysema in a patient treated with bleomycin can be an early warning for the development of bleomycin toxicity. Awareness of the sequelae is indispensible.

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SPECIAL ARTICLE

Implementation of evidence-based practice: outside the box, throughout the hospital

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ABSTRACT

Background: Evidence-based practice (EBP) is a generally accepted means to improve healthcare quality. However, not all healthcare professionals and managers apply EBP in daily practice. We investigated EBP attitudes, knowledge and the perceived barriers and facilitators to practising EBP, to define tailor-made interventions for improving evidence-based behaviour.

Methods: In this cross-sectional survey, doctors and nurses from five major specialities of a university hospital were invited to complete the McColl and Barriers questionnaires.

Results: Response rates were 70% (305/435) for doctors and 74% (396/537) for nurses. They were welcoming towards EBP, but considered time constraints, knowledge gaps and poor availability of evidence as major barriers to implement EBP. They also mentioned contradicting results (75%) and flawed methodology (69%), while nurses frequently mentioned unawareness of (75%), or difficulty in reading and interpreting research papers (70%). Regarding EBP knowledge, 6/8 common EBP terms could be explained by 54% of doctors but by only 15% of nurses. Facilitating factors among doctors concerned the availability and accessibility of high-level evidence and communication of evidence during various clinical meetings and handovers for clinical decision making. Among nurses, promoting factors involved more teaching and instances to incorporate EBP in clinical practice. Both groups desired more managerial support in terms of motivation and opportunities.

Conclusions: Doctors and nurses have embraced the EBP paradigm as an important means to improve quality of clinical patient care, but its application is still cumbersome. This paper offers a tailored programme for implementation and sustainment of EBP, corroborated by professional and managerial role-models.

KEYWORDS

Evidence-based practice, implementation, patient care management, quality assurance, quality of healthcare, questionnaires

INTRODUCTION

Societal and patients' demands for professional and resource accountability have fostered the introduction of evidence-based practice (EBP) in healthcare and education.[1,2] Hospital executive boards and insurance companies stress the use of EBP to prevent practices that are unsafe or lack empirical support, to reduce unacceptable individual variance, and ultimately to increase efficiency and quality in healthcare.[3]

However, reality proves that healthcare professionals have been unresponsive to embrace EBP in daily practice. Implementation by doctors might be hampered by a perceived lack of time, knowledge or resources,[4,5] while in the nursing realm the body of knowledge is still burgeoning. A joint venture of role-modelling teachers, doctors, nurses and managers is desirable yet missing to really make EBP work and to enhance the quality of care for patients.[6,7]

Randomised trials or systematic reviews may be scarce and available evidence may merely stem from bias-prone study designs, or be lacking altogether.[8] Nevertheless, it is important to be aware of this level of evidence behind the interventions we offer our patients,[9] as it guides the strength of our recommendations and can help clinical decision making.[10] Hence, the question emerges as to how to overcome possible limitations of, and reluctance to implement EBP.

Improvements in evidence-based behaviour can only be realised if awareness of, and a positive attitude towards,

EBP are secured first.[11] Moreover, promoting change in clinical practice is more likely to be successful if a change strategy is based on the specific barriers and facilitators perceived by the professionals involved.[12]
Therefore, the aim of this study was to determine the attitude towards and awareness of the EBP principle among doctors, nurses and managers within a university hospital and the barriers experienced in practising EBP, in order to define a tailor-made intervention programme to structurally facilitate and sustain evidence-based behaviour.

METHODS

This survey was conducted at the five largest departments (Internal Medicine, Surgery, Obstetrics & Gynaecology, Paediatrics and Neurology) of the Academic Medical Center, a 1000-bed university hospital in Amsterdam, the Netherlands. Approval for the survey was obtained from the medical and nursing managers of each of these departments. Ethical approval was deemed unnecessary.
To assess the attitudes towards and knowledge and barriers of the EBP principle, we combined two questionnaires, i.e. the Barriers scale and the McColl questionnaire.[5,13] The Barriers scale addresses the perceptions of barriers to the utilisation of research findings in clinical practice. This five-point scale of 29 items has been validated in various settings worldwide to assess EBP implementation barriers.[14,15] The McColl questionnaire addresses attitude (on a 10 cm visual analogue scale), awareness and actual use of EBP, and has also been applied widely.[16-18] Both questionnaires were translated into Dutch by means of forward-backward translation,[19] and distributed as paper or electronic versions. To assess EBP knowledge among doctors and nurses a list of common EBP terms relevant to their clinical practices was provided. We added two non-existing dummy terms to these lists to gauge any socially desirable answering.[20] For doctors, these were 'Fixed event rate' and 'Random benefit ratio', and for nurses 'Dosage chance' and 'Absolute treatment increase'. All clinical specialists, trainees and nurses, including those with managerial tasks, of the five departments were invited to complete the questionnaires. Respondents' general characteristics, including their age, gender, level of education, working experience, previous EBP training and literature search facilities were also recorded.

DATA ANALYSIS

The answers to the 29 possible barriers were dichotomised, i.e. items scored as 'barrier' or 'large barrier' were counted as barriers. Means and standard deviations (SD) or medians and inter-quartile ranges (IQR) were calculated, depending on the distribution of the parameter. Differences were expressed as mean differences with 95% confidence intervals (CI). To compare the means of the attitude scores towards EBP between different subgroups, the Student's t-test was used. Differences between median values were analysed using the Mann-Whitney U test. Statistical analysis was performed using IBM-SPSS version 18.0 (SPSS Inc., Chicago, Illinois, USA).

RESULTS

A total of 305 doctors and 396 nurses responded (response rates of 70 and 74%, respectively). Characteristics of the respondents are summarised in *table 1*.

McColl questionnaire

About two-thirds of the doctors and nearly half of the nurses stated to have had some training in literature searching (*table 1*). About a quarter of the doctors and less than 10% of the nurses had attended a formal EBP course in the past. These data did not differ substantially among the five departments. Doctors regularly searched for evidence in the literature, which contrasted sharply with the nurses. Doctors had easy access to PubMed, both at home and at work. In contrast, nurses did not always realise they could access PubMed at home, despite having internet facilities.
When asked which competences they considered essential to change from experience-based to evidence-based practice, the majority of doctors stated a combination of searching and critical appraisal skills, finding and applying evidence-based summaries, and using evidence-based guidelines. The same question was repeated for perceived future needs, showing a small shift towards the exclusive use of evidence-based guidelines. For now and for the future, the majority of the nurses preferred to rely entirely on evidence-based guidelines and protocols.
The EBP principle enjoyed a welcoming attitude (*table 2*). This was more so among doctors (72 on a scale of 100) than among nurses (55; mean difference 16.5, 95% CI 13.8 to 19.2). Neurologists and internal medicine nurses had the most positive attitudes (scores of 77 and 71, respectively). The same was true for the perceived EBP attitude of their colleagues.
Both doctors and nurses considered research findings to be very useful in daily practice and they very much agreed with the statement that EBP improves patient care (*table 2*). Surgeons tended to find practising EBP rather demanding (score of 56), but not all were convinced about the purported limitations of EBP that it would be time-consuming for busy professionals or that evidence would be frequently lacking. For instance, this was not so among the neurologists (score of 35), who also felt that

Table 1. *Respondents' characteristics, EBM proficiency and literature search facilities*

	DOCTORS (n=305)						NURSES (n=396)					
	Internal medicine	**Surgery**	**Paediatrics**	**Neurology**	**Obs&Gyn**	**Total**	**Internal medicine**	**Surgery**	**Paediatrics**	**Neurology**	**Obs&Gyn**	**Total**
N respondents/total (%)	98/142 (69%)	29/43 (67%)	80/128 (63%)	36/55 (65%)	62/67 (93%)	305/435 (70%)	31/54 (57%)	84/122 (67%)	192/255 (75%)	46/53 (87%)	43/53 (81%)	396/537 (74%)
Males	45%	62%	42%	74%	29%	46%	27%	23%	9%	24%	12%	15%
Mean age (SD[1])	38 (9)	38 (8)	41 (9)	36 (11)	38 (11)	39 (10)	33 (11)	37 (13)	40 (10)	40 (13)	41 (12)	39 (12)
Senior staff[2]	42%	52%	73%	37%	48%	52%	29%	18%	19%	24%	19%	20%
Median working experience in years (IQR[3])	7 (4-16)	9 (4-17)	10 (6-17)	5 (2-10)	8 (4-19)	8 (4-17)	6 (2-13)	13 (4-27)	19 (8-28)	17 (5-28)	16 (7-30)	16 (6-27)
Previous literature search training	56%	69%	69%	75%	76%	67%	55%	41%	45%	52%	47%	45%
Previous critical appraisal training	62%	62%	73%	67%	53%	64%	45%	33%	41%	39%	40%	39%
Previous EBM training	10%	28%	29%	19%	36%	23%	4%	5%	9%	11%	12%	8%
Median number of searches last year (IQR[3])	100 (50-300)	30 (20-125)	100 (50-200)	200 (100-425)	100 (50-300)	100 (50-300)	4 (0-15)	0 (0-2)	1 (0-4)	1 (0-10)	0 (0-3)	1 (0-4)
Current way[4] of using evidence	57% all	62% all	69% all	81% all	58% all	64% all	46% guidelines	68% guidelines	66% guidelines	60% guidelines	68% guidelines	64% guidelines
Future way[4] of using evidence	58% all	72% all	67% all	83% all	66% all	66% all	39% all; 22% guidelines	26% guidelines; 22% all; 18% skills + guidelines	23% all; 24% search + guidelines; 23% guidelines	24% all; 20% skills; 20% guidelines	24% guidelines 21% all; 17% skills	23% all; 23% guidelines
Most suitable way[4] of using evidence for own specialism	26% all; 25% guidelines; 23% EB summaries	32% all; 32% guidelines	37% all; 28% guidelines	51% all; 14% skills; 11% guidelines	40% all; 39% guidelines	36% all; 28% guidelines	29% guidelines; 25% all	40% guidelines; 28% all	42% guidelines; 25% skills + guidelines	46% guidelines; 22% skills + guidelines	44% guidelines 21% all	40% guidelines; 20% EB summaries
PubMed access at home	97%	86%	95%	100%	90%	94%	46%	22%	38%	56%	35%	38%
PubMed access at work	100%	100%	100%	100%	100%	100%	96%	84%	87%	93%	82%	88%
Internet at home	100%	97%	98%	100%	100%	99%	96%	89%	97%	98%	91%	95%
Internet at work	100%	100%	100%	100%	100%	100%	86%	100%	78%	91%	100%	79%

[1]SD = standard deviation; [2]I.e., medical specialists and senior or head nurses; [3]IQR = inter-quartile range; [4](Combinations of) a. searching and critical appraisal skills; b. use of evidence-based summaries; c. using evidence-based guidelines or protocols.

Table 2. *Current attitudes towards EBP; scores can range from 0 to 100*

	Doctors mean (SD)	Nurses mean (SD)	Difference (95% CI)	P-value (Student t-test)
Your current attitude towards EBP Least positive (0) ↔ Extremely positive (100)	71.5 (15.7)	55.0 (21.6)	16.5 (13.8 to 19.2)	<0.001
Attitude of your colleagues towards EBP Least positive ↔ Extremely positive	73.3 (13.5)	48.1 (19.2)	25.2 (22.8 to 27.6)	<0.001
How useful are research findings in daily practice? Useless ↔ Extremely useful	70.0 (15.4)	62.0 (18.4)	8.0 (5.5 to 10.6)	<0.001
What percentage of your clinical practice is evidence-based? 0% ↔ 100%	50.2 (18.1)	43.8 (20.6)	6.4 (3.5 to 9.3)	<0.001
Practising EBP improves patient care Completely disagree ↔ Fully agree	79.0 (13.8)	74.3 (17.6)	4.7 (2.4 to 7.1)	<0.001
EBP is of limited value in clinical practice, because a scientific basis is lacking Completely disagree ↔ Fully agree	41.6 (23.7)	48.6 (20.7)	-7.0 (-10.4 to -3.6)	<0.001
Implementing EBP, however worthwhile as an ideal, places another demand on already overloaded doctors/nurses Completely disagree ↔ Fully agree	44.3 (24.9)	55.2 (23.2)	-10.9 (-14.5 to -7.2)	<0.001

scientific evidence was broadly available in their speciality (score of 28) (data from each separate speciality not shown in tables).

Doctors estimated that only half of their clinical practice was evidence-based, which was even lower (44%) according to the nurses (*table 2*). The respondents of the obstetrics/gynaecology department estimated their practice was most evidence-based (doctors 58%, nurses 53%), while the paediatrics department regarded their practice as least evidence-based (doctors 39%, nurses 42%). This might be related to the barriers paediatricians noted that available evidence cannot easily be extrapolated to children and that clinical trials in children are scarce.

Specialists estimated a slightly but significantly lower percentage of their practice to be evidence-based (47.8%) than their trainees did (52.9%); mean difference 5.1%, 95% CI 1.0 to 9.3%. However, their attitude to EBP was not different from the trainees. Furthermore, EBP attitude among the oldest quartile of specialists (51 to 65 years) was not significantly different from the youngest quartile (aged below 37). There were also no meaningful differences in attitude between male and female doctors. Senior nurses showed a more positive attitude towards EBP than non-senior registered nurses (scores of 67 *vs* 52, respectively; mean difference 14.9, 95% CI 10.3 to 19.5), and were more convinced that EBP improves patient care (79 *vs* 73, respectively; mean difference 6.0, 95% CI 2.2 to 9.8). Doctors and nurses with a managerial role (i.e. heads of department, nursing managers) did not give conspicuous responses.

Figure 1 shows the proportion of common EBP terms the doctors and nurses said they understood. Half of the doctors had (some) understanding about all of the eight terms provided. The two dummy terms were least known, but still 39 and 49% of the doctors, respectively, claimed

Figure 1. *Doctors' (top panel) and nurses' (bottom panel) knowledge of common EBP terms; terms with an asterisk are meaningless dummy terms*

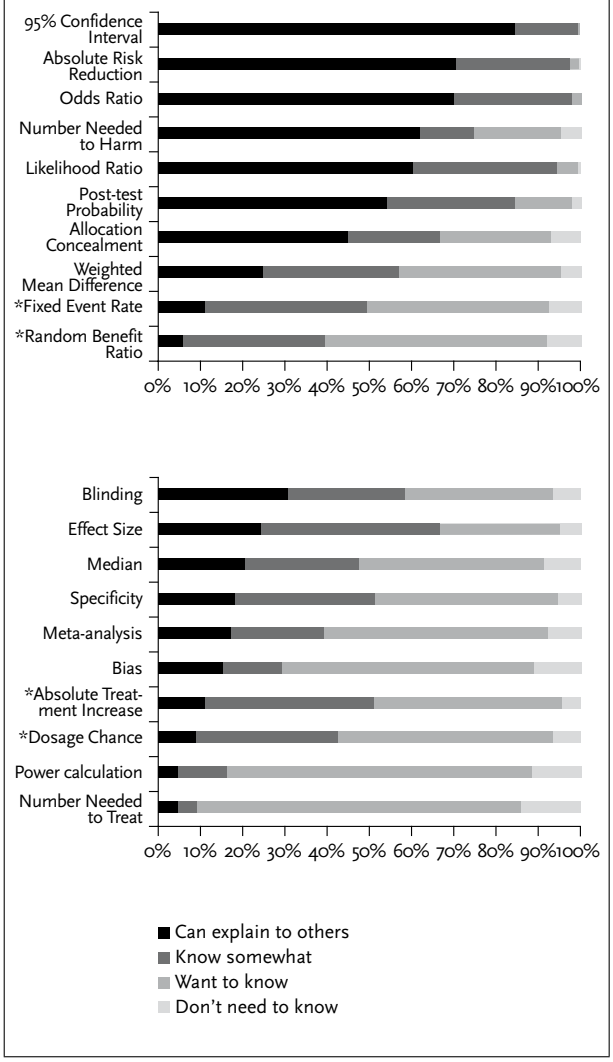

Table 3. *Doctors' and nurses' awareness of common sources of evidence*

Source	Unknown	Familiar	Read	Used
		DOCTORS		
ACP (American College of Physicians) Journal Club[1]	70.0%	17.7%	6.8%	5.5%
CBO (Dutch Institute for Healthcare Improvement)[2]	19.3%	16.2%	18.6%	45.9%
Cochrane library[3]	0.0%	6.3%	22.0%	71.7%
Evidence-based medicine[1]	37.0%	38.0%	12.5%	12.5%
National Guideline Clearinghouse[2]	60.7%	18.6%	6.8%	13.9%
TRIP database[3]	70.6%	15.0%	7.2%	7.2%
		NURSES		
CBO (Dutch Institute for Healthcare Improvement)[2]	62.9%	19.5%	8.7%	8.9%
Cochrane library[3]	49.1%	28.9%	11.5%	10.5%
Evidence-based nursing[1]	36.9%	41.5%	18.1%	3.6%
CINAHL (Cumulative Index to Nursing & Allied Health Literature)[3]	70.6%	14.8%	9.0%	5.6%
LEVV (Netherlands Centre of Excellence in Nursing)[2]	56.8%	21.7%	15.0%	6.5%
Verpleegkunde (Dutch-Flemish Scientific Nursing Journal)[3]	23.2%	36.7%	34.9%	5.2%

[1]Source offering pre-appraised evidence; [2]Source offering (evidence-based) guidelines; [3]Source offering evidence from various study designs and aggregation levels.

to have some knowledge about their meaning. Half of the nurses had (some) understanding of three out of the eight terms. Real and dummy terms were known equally well among nurses (suggesting socially desirable answering to this part of the questionnaire), while 'power calculation' and 'number needed to treat' were virtually unknown.

The respondents' familiarity with sources of evidence are summarised in *table 3*. Databases with systematic literature reviews (Cochrane) and national guidelines (CBO) were most widely used for clinical decision making (by 72 and 46% of the doctors, respectively). This was also true for nurses, but to a much lesser extent (10.5 and 8.9%, respectively). Sources offering pre-appraised evidence (e.g. ACP Journal Club, Evidence-based Medicine) were not (yet) used habitually.

Table 4. *Top-five barriers to applying EBP as stated by doctors and nurses*

DOCTORS	
The literature reports conflicting results	75.3%
The research has methodological shortcomings	69.4%
The doctor has insufficient time to read research	66.3%
The doctor is unaware of the research	61.9%
The doctor feels the results are not applicable to his/her situation	58.4%
NURSES	
The nurse has insufficient time to read research	76.5%
The nurse is unaware of the research	75.4%
The research is not reported clearly and readably	70.2%
Statistical analyses are unintelligible	69.9%
Insufficient time to implement new ideas at the workplace	69.5%

BARRIERS SCALE

The top-five barriers as perceived among doctors and nurses are shown in *table 4*. For over 75% of the doctors, inconsistent literature results were the biggest hurdle. Time constraints to read and unawareness of literature results were considered to be the major impediments by more than 75% of the nurses, but also by many doctors. Nurses in particular had difficulties with reading papers in a foreign (English) language.

The major facilitating factors as reported by doctors and nurses (*table 5*) could be summarised as: constant involvement by colleagues, staff and management in learning and applying EBP in daily clinical practice, structural promotion and facilitation of EBP activities by the management, and clear and easily accessible protocols and guidelines.

Table 5. *Major facilitating factors to apply EBP as stated by doctors and nurses*

- Dedicated time to learn and practise EBP
- Management support
- Promotion and integration of EBP among all disciplines involved in patient care
- Communication of (new) evidence at various meetings, rounds or handovers
- Easily accessible sources of evidence-based guidelines and protocols
- Role-modelling EBP experts and managers
- Availability of pre-appraised or aggregate evidence
- Promotion of EBP by spreading successes of evidence-based interventions
- More well-designed and well-performed, clinically relevant research

DISCUSSION

The majority of healthcare professionals and staff in the larger clinical specialities within our university hospital appear to be quite EBP-minded. They appreciate that research findings are useful for daily clinical practice and consider the EBP paradigm an important tool to improve the quality of patient care. However, important barriers are still obstructing the implementation of EBP in daily clinical practice.

These findings, in particular barriers such as time constraints, knowledge gap and poor availability of evidence, occur consistently among the various medical specialists and nurses alike and have also been signalled in many other specific settings and specialities throughout Europe.[20-24] Moreover, observed barriers appear to be consistent over time and geographical region.[15] These observations have prompted various single-focus teaching initiatives, e.g. teach-the-teacher modules.[25] However, available evidence is not convincing as to whether stand-alone teaching modules actually improve EBP skills, attitudes or behaviour.[26]

Effective implementation strategies should take a broader approach and involve not only medical and nursing schools and residency educational programmes, but also management policy and health systems.[27,28] Such implementation strategy should be a multifocal, comprehensive programme for all the professionals involved ('professional in the lead') and tailored to their desires and perceived barriers.[29] After all, excellent evidence-based patient care cannot be attained without the interaction of the different managerial, research, and healthcare professionals.

Table 6. *Structural incorporation of EBP at various levels*

National	• Governmental enforcement of EBP in healthcare and educational institutions
	• Professional societies' quality assurance and guidelines policy
Board of hospital directors	• Strategic aims
	• Five-year planning
	• Workplace visits and internal audits
	• Stimulation funds
	• Annual invitation of visiting professors on EBP related topics
Management	• Staff planning and recruitment of EBP-minded leadership and role-modelling personnel
	• Yearly performance interviews including EBP activities
	• Budgetary allowances for EBP education and EBP experts on every ward
	• Professional atmosphere that embraces EBP
Education	• Structural part of medical and nursing curricula
	• Structural postgraduate courses and e-learning modules
	• Collaboration and interaction between teachers and clinicians
Services	• Medical library facilities
	• Content management system allowing access to guidelines, protocols and condensed recommendations
	• Generally accessible database for critically appraised topics (CATs)
	• Help service for searching databases
Local workplace	• Journal clubs, grand rounds, handovers, regular (research) meetings
	• Dedicated time and personnel for EBP activities
	• Easy access to computers and databases
	• Research on yet unproven interventions

IMPLICATIONS

Based on the results from our and other groups, and considering the various challenges and opportunities for EBP implementation, we have summarised our suggestions for structural incorporation of EBP at various hierarchical levels in *table 6*. An EBP implementation programme should firstly be promoted and facilitated by the management, and epitomised by role models among the various specialities and professionals. Furthermore, EBP activities should be part of quality indicators, departmental audits, and certification. Second, it should include teaching modules for undergraduate students and (preferably integrated) postgraduate courses for nurses and doctors.[30] However, not every healthcare professional needs to be trained up to an expert level at which (s) he can find, appraise, implement as well as generate evidence.[31,32] Rather, every department should at least have some EBP experts, doctors as well as nurses, to ignite and sustain the EBP approach, while every professional should have a critical attitude towards their clinical practice. Third, the programme should enable a local easy-to-use and easy-to-access database with updated evidence-based guidelines and protocols,[33] because awareness and use of internet sources of evidence is still imperfect.[34] Finally, it should make the most of opportunities during regular clinical meetings, such as handovers, grand rounds and journal clubs, to present and discuss available evidence. These discussions could help overcome the possible conflicting opinions about existing evidence and may help reach an agreement about the policy of choice. This requires an open culture in which feedback, communication prowess and respectful arguing are basic attitudes.

The overall welcoming attitude towards EBP as found in our survey offers an excellent opportunity to improve the apparently deficient EBP knowledge, skills and facilities. Both our survey and the presently available evidence

have given input for more outside-the-box thinking and a wide-ranging, structured approach to improve and sustain the implementation of the EBP paradigm throughout and even beyond the hospital. Because it is clear that many other institutions face the same challenges, the proposed structural implementation programme is likely to be useful for wider implementation.

LIMITATIONS

The outcomes of our survey may show a flattering picture. First, the non-responders may have been less EBP-prone. On the other hand, the responders did relate many barriers to EBP implementation, indicating they had not swallowed EBP whole. Second, the survey was based on self-reported knowledge rather than actual EBP knowledge and behaviour, while the dummy terms revealed an inflated level of knowledge. Hence, the factual EBP level is probably lower. Third, at the time of this survey our institution was not virginal in terms of EBP education. Since the 1990s, our doctors and nurses have been ushered into the EBP principle. More than half of the doctors and about 40% of the nurses now stated to have had some training in critical appraisal. It is therefore likely that other, non-teaching hospitals will be much less familiar with EBP. This underlines the need for further improvement initiatives.

Finally, our finding that doctors outperform nurses in EBP proficiency may be due to the fact that nurses are lagging behind in EBP education and only a few of them have been educated at a master degree level. This explains to some extent why nurses have difficulties with reading scientific (mainly English) literature. Fortunately, the number of available undergraduate and postgraduate EBP modules is now growing on a national and international scale.[34] In our hospital the EBP knowledge level is likely to have improved in the mean time, after the educational efforts during recent years. Hence, some of the items in our ongoing EBP implementation programme that have been employed in our institution gradually seem to be bearing fruit. Thus, we are confident the proposed multifaceted approach will be even more helpful for a successful implementation and assurance of EBP activities in daily clinical practice. Future verification measurements are needed to confirm adherence to EBP behaviour and its effect on patient-relevant outcomes.[36]

CONCLUSION

In our quest to clinical excellence of patient care, the adoption of the EBP paradigm through a tailor-made structural programme in collaboration with all stakeholders appears to be pivotal to make a substantial contribution to this goal.

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ANSWER TO PHOTO QUIZ (PAGE 84)

SLIPPED CAPITAL FEMORAL EPIPHYSIS AS MANIFESTATION OF A RARE ENDOCRINOLOGICAL DISEASE

DIAGNOSIS

Slipped capital femoral epiphysis (SCFE) can be a manifestation of the multiple endocrine neoplasia syndrome type 2 (MEN 2).[1] MEN 2 syndrome is subdivided into MEN 2a and MEN 2b and both have medullary thyroid carcinoma as the most common feature. However, hyperparathyroidism is characteristic for MEN 2a whereas patients with MEN 2b can be recognised by neurofibromas of the tongue and marfanoid habitus.[2]

The diagnosis of acute SCFE is easier than that of chronic SCFE.[3] Both disorders present with pain in the hip or with referred pain in the knee. Patients with acute SCFE typically have a contracture by flexion, abduction and exorotation. However, in patients with chronic SCFE the only presenting symptom can be a mild limp.[3]

SCFE can be difficult to diagnose on anteroposterior radiographs.[3] For chronic SCFE a lateral radiograph according to Lauenstein (hips in 90° flexion and maximal abduction) is advised.[4] SCFE often occurs bilaterally, therefore bilateral imaging at presentation and also during follow-up is indicated.[4] Treatment of acute and chronic SCFE is surgical.[4] Awareness of the association between MEN 2 and SCFE could help to identify patients earlier. This is crucial in order to prevent metastatic medullary thyroid carcinoma. Pheochromocytoma can also be part of MEN 2 and could cause severe hypertensive crisis or arrhythmias perioperatively. Complications of SCFE are avascular necrosis, chondrolysis and coxarthrosis if the diagnosis is missed.

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LETTER TO THE EDITOR

Anakinra for the inflammatory complications of chronic granulomatous disease

Dear Editor,

With great interest, we read Seger's review on chronic granulomatous disease (CGD).[1] It is a very up-to-date and clear review. Seger refers to our observation that there is a strongly upregulated production of interleukin-1β in patients with CGD, demonstrating that a deficiency of NADPH-dependent reactive oxygen species leads to increased inflammasome activation.[2] We have taken this observation further in a 39-year-old patient with CGD (p91-phox mutation), who was suffering from perirectal granulomas that were refractory against corticosteroid therapy. We treated the patient with recombinant interleukin-1 receptor antagonist (subcutaneous anakinra, 100 mg daily) for three months with a good response.

In his review, Seger states that anti-TNF drugs may be used in such cases, but only for a short period, because of the infectious hazards. As anakinra is much safer in this respect,[3,4] we would propose to try anakinra first in CGD patients with granulomas. Since anakinra has fewer side effects (pain and inflammation at the injection site due to the preservative) than corticosteroids one might even ask the question whether anakinra should be preferred over the latter drugs.

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LETTER TO THE EDITOR

Hereditary persistence of alpha-fetoprotein

Dear Editor,

The article by Houwert *et al*. about Hereditary persistence of alpha-fetoprotein (AFP)[1] correctly underlines that increased serum AFP concentrations do not necessarily reflect malignant or hepatic disease. Unfortunately, their list of conditions (*table 4*) associated with increased serum AFP is incomplete.

Ataxia with oculumotor apraxia type 2 (AOA2), ataxia telangiectasia (AT) and variant AT are three conditions belonging to the autosomal recessive ataxias that are characterised by increased serum AFP levels.[2] For (variant) AT, AFP may be in de range of 200 to 500 ng/ml.[3] All three conditions are caused by a defective DNA repair mechanism, which is most seriously affected in AT. Particularly in AT and variant AT, the diagnosis should not be overlooked. First, because various complications may develop including diabetes mellitus, immunodeficiciency and severe lung restriction. Second because maligancy often develops in the 2nd or 3rd decade. Third, because the increased radiosensitivity precludes the use X-ray diagnostics, radiotherapy and alkylating agents.

We feel the need to emphasise the presence of high serum AFP levels in AT, because the diagnosis can easily be missed for several years, particularly in patients with variant AT who may have only subtle neurological problems until adulthood. In these cases, high AFP levels may be the key to the correct diagnosis and appropriate patient management. Ignorance of the diagnosis of (variant) AT will put a patient at risk after mechanical ventilation,[4] when X-ray diagnostics are perfomed, or – in case of proven malignancy – when DNA-damaging therapy is given.

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