Independent medical research

Non-dipping blood pressure profile

Ewing's sarcoma and primitive neuroectodermal tumours

Cushing's syndrome and bone mineral density
MISSION STATEMENT

The mission of the journal is to serve the need of the internist to practice up-to-date medicine and to keep track with important issues in health care. With this purpose we publish editorials, original articles, reviews, controversies, consensus reports, papers on specialty training and medical education, book reviews and correspondence.

EDITORIAL INFORMATION

Editor in chief
Anton F.H. Stalenhoef, Radboud University Nijmegen Medical Centre, Department of General Internal Medicine, Nijmegen, the Netherlands

Associate editors
Joost P.H. Drenth, Nijmegen, the Netherlands
Jack F.M. Wetzels, Nijmegen, the Netherlands
Theo Thien, Nijmegen, the Netherlands

Editorial board
J.V. Bonventre, Massachusetts, USA
H. Brunner, Nijmegen, the Netherlands
J.J. Cornelissen, Rotterdam, the Netherlands
S.A. Danner, Amsterdam, the Netherlands
J.T. van Dissel, Leiden, the Netherlands
J.P. Droz, Lyon, France
R.O.B. Gans, Groningen, the Netherlands
A.R.J. Girbes, Amsterdam, the Netherlands
D.E. Grobbee, Utrecht, the Netherlands
D.L. Kastner, Bethesda, USA
R.B.M. Landewé, Maastricht, the Netherlands
M.M. Levi, Amsterdam, the Netherlands
B. Lipsky, Seattle, USA

R.L.J.F. Loffeld, Zaandam, the Netherlands
Ph. Mackowiak, Baltimore, USA
J.W.M. van der Meer, Nijmegen, the Netherlands
G. Parati, Milan, Italy
A.J. Rabelink, Leiden, the Netherlands
D.J. Rader, Philadelphia, USA
J.A. Romijn, Leiden, the Netherlands
J.L.C.M. van Saase, Rotterdam, the Netherlands
P. Speelman, Amsterdam, the Netherlands
C.D.A. Stehouwer, Maastricht, the Netherlands
E. van der Wall, Utrecht, the Netherlands
R.G.J. Westendorp, Leiden, the Netherlands

Editorial office 'The Netherlands Journal of Medicine'
Geeralien Derksen-Willemsen
Radboud University Nijmegen Medical Centre
Department of General Internal Medicine 463
PO Box 9101, 6500 HB Nijmegen
The Netherlands
Tel.: +31 (0)24-361 04 59
Fax: +31 (0)24-354 17 34
E-mail: g.derksen@aig.umcn.nl
http://mc.manuscriptcentral.com/nethjmed

CITED IN

Biosis database; embase/excerpta medica; index medicus (medline) science citation index, science citation index expanded, isi alerting services, medical documentation services, current contents/clinical medicine, PubMed.
Contents

EDITORIAL

Independent medical research
J.W.M. van der Meer, A.M. de Gier, W.P.M. van Swaaij, M.B. Katan

124

REVIEW

Causes and consequences of a non-dipping blood pressure profile
A.M. Birkenhäuser, A.H. van den Meiracker

127

ORIGINAL ARTICLES

Ewing’s sarcoma and primitive neuroectodermal tumours in adults: single-centre experience in the Netherlands

132

Cushing’s syndrome and bone mineral density: lowest Z scores in young patients
A.W. van der Eerden, M. den Heijer, W.J. Oyen, A.R. Hermus

137

CASE REPORTS

Coma with ECG abnormalities: consider tricyclic antidepressant intoxication
J. Veris-van Dieren, L. Valk, L. van Geijlswijk, D. Tjan, A. van Zanten

142

Successful management of chronic myeloid leukaemia with leucapheresis during a twin pregnancy
R. Klaasen, P. de Jong, P.W. Wijermans

147

A woman with Cushing’s syndrome after use of an Indonesian herb: a case report
P.C. Oldenburg-Ligtenberg, M.M.L. van der Westerlaken

150

PHOTO QUIZ

Sine waves
A. Alkhatib S.E. McGuire

153

Answer to photo quiz

155

LETTER TO THE EDITOR

Extensive jejunal diverticulosis in a family, a matter of inheritance?
A.D. Koch, E.J. Schoon

154

MONTHLY NJM ONLINE HITLIST

For all articles published in January 2007

156
ABOUT THE COVER (NETh J Med 2007, Vol. 65, No. 2)

Unfortunately, the details about the cover print of Frans de Groot and Josée Wuyts were printed incorrectly.
The correct details are:
Title: ‘Herstel mijn huis’
Size: 92 x 63 cm
Price: €400,-

COVER (NETh J Med 2007, Vol. 65, No. 3)

The cover should be printed in full colour (see below).
The publisher and editorial board of the Netherlands Journal of Medicine regret that these omissions have occurred.

PHOTO QUIZ (NETh J Med 2007, Vol. 65, No. 3)

Regretably figure 2 in the photo quiz (Chalupa P, Holub M, Kašpar M. Abdominal pain in a veterinarian with cysts in the liver. Neth J Med 2007;65:119) has been printed incorrectly. Below you will find the correct figure 2.

Figure 2. CT scan demonstrates cystoid multiseptic lesion in the right liver lobe
Independent medical research?

J.W.M. van der Meer1,2,*, A.M. de Gier1,5, W.P.M. van Swaaij1,3,5, M.B. Katan1,4,5

1Royal Netherlands Academy of Arts and Sciences, Amsterdam, the Netherlands, 2Department of General Internal Medicine, Radboud University Nijmegen Medical Centre, Nijmegen, the Netherlands, 3Faculty of Science and Technology University of Twente, Enschede, the Netherlands, 4Institute for Health Sciences, VU University, Amsterdam, the Netherlands, *corresponding author: j.vandermeer@aig.umcn.nl

Concerns about undue influence of sponsors on research have already been heard for decades. Efforts to change this situation have had insufficient impact. Here we draw attention to an innovative proposal by the Royal Netherlands Academy of Sciences on how to prevent sponsor-induced bias.

Is the problem still urgent? Several studies suggest that it is. In a recent survey of the 289 most-cited clinical trials published between 1994 and 2003, Ioannidis’ group analysed the origin of authors (academic or nonacademic) and the source of finance of these trials. This study was done within the context of the International Campaign to Revitalise Academic Medicine (ICRAM). During the period of observation, the proportion of investigations financed by pharmaceutical industries increased significantly: no less than 65 of the 77 most-cited clinical trials were (co-)financed with money from industry. Obviously the increasing influence of industry is a reason for concern, especially when the boundaries of influence are unclear. Industrial involvement, particularly in drug trials, usually starts with the design of the study, the choice of the comparator drugs, and the selection of the clinical investigators. Often industry has a major involvement in the collection and control of the data, as well as in data analysis. Even the (ghost)writing of the article may be done by the sponsor.

It is of quite some concern, as Kjaergard and Als-Nielsen have pointed out, that authors of trials with competing interests, i.e., those funded by for-profit organisations, are significantly more positive towards the results of their investigation than those without. This observation fits in with the results of the systematic review by Lexchin et al., which demonstrates that studies sponsored by pharmaceutical companies are more likely to have outcomes favourable to these sponsors than investigations that received other funding. A recent survey of major clinical trials in the cardiovascular field showed similar results: trials funded by for-profit organisations were more likely to report positive findings than those supported by not-for-profit organisations. Similar bias was seen in nutrition studies supported by dairy and beverage companies.

But there are more reasons for concern: after publication, the study results may serve as promotional material and be selectively used to inform prescribers and potential consumers. In this process the investigators may be used as a vehicle. In addition, industrial influence may sneak into the process of development of professional protocols and guidelines. A recent, rather scary example is the Surviving Sepsis Campaign, a basically marvellous initiative aiming at standardising and improving the basic care for patients with sepsis. In this campaign, however, one particular industry, with a major interest, seems to have gained a pivotal position.

In a recently published perspective in the New England Journal of Medicine, this story has been described in detail. The attempts of industry to influence the development and – more seriously – the contents of practice guidelines are not new. It has been found that 87% of authors of guidelines have ties with industry and these are often not revealed.

Of concern are also the relationships between industry and members of institutional review boards (IRB). In a recent survey, Campbell et al. investigated the financial relationships between IRB members and industry and found that some 36% of these members had some kind of financial relationship with industry. Formal disclosure of relationships with industry is not required by 33% of IRB. Of the respondents, 15.6% reported that in their experience at least once a protocol had been presented in a biased way by an IRB member with industrial ties.

Also in daily practice, there is a strong influence of industry. In fact, it is a rather sad finding that
representatives of pharmaceutical industries often have a greater influence on prescribing habits than prevailing hospital protocols and objective appraisals in the literature. The persuasion of these pharmaceutical representatives is often reached with the help of ‘beads and mirrors’, rather than through solid information. These practices still persist despite initiatives to regulate the interaction between pharmaceutical industry and prescribing physicians. The Dutch Ministry of Health has issued a series of regulations in this respect, starting in 1999, and initially these measures met with quite some effect, but it is our impression that the effect has waned in recent years. It is interesting to note that industry is not the only party that tries to influence outcome and reporting of science. For instance, governmental bodies may assign investigations and selectively use the outcome of the research and may require that the results of the investigations are kept secret.14

If we return to the core of the problem, it is clear that the influence of industry on clinical research is too strong and difficult to disentangle. In 2001, the International Committee of Medical Journal Editors (ICMJE) took the important initiative to ask for a disclosure of conflicts of interests of all authors (and to publish those as part of the article that reports the investigation). A further definite step forward – aimed at preventing selective reporting – is the registration of clinical trials in a public repository at their inception.9 See also www.clinicaltrials.gov. In this repository, the role of the sponsor is also revealed. Still, there is a need for better regulation of the relationship between sponsor or client and researcher. In a recent advice to the Dutch Minister of Science and Education, The Royal Netherlands Academy of Arts and Sciences (KNAW) voiced its concern about the independence of the investigator, and proposes a code of conduct (figure 1) to be signed by the university or other research institute that performs the investigation.15 The declaration proposes that research institutes that wish to be certified as adherent to this code must maintain a list of all the research contracts concluded by them. Contracts would be open to inspection by the National Council on Research Integrity (LOWI) which resides within the Academy (figure 1, Clause 9). The Council could demand a copy of a specific contract, and could therefore perform random or directed inspections. If the text of a contract were found to violate the code, the Council could revoke the certification of the research institute. As yet, the Dutch government has not declared whether it will implement this code. We feel that acceptance and implementation of this code is an essential next step to create clarity in the relationship between research institutes and sponsors, and in the field of medicine it may help to control the unwanted influence of pharmaceutical industry.

**CONFLICTS OF INTEREST**

J.W.M. van der Meer received an unrestricted grant from Glaxo Smithkline for research of chronic fatigue syndrome. M.B. Katan is a member of the editorial board of PloS Medicine.

**REFERENCES**


Causes and consequences of a non-dipping blood pressure profile

A.M. Birkenhager*, A.H. van den Meiracker

Department of Internal Medicine, Vascular Pharmacology, Erasmus Medical Centre, 's Gravendijkwal 230, 3015 GD Rotterdam, the Netherlands, *corresponding author: tel.: +31 (0)10-463 21 96; fax: +31 (0)10-463 45 31; e-mail: a.birkenhager@erasmusmc.nl.

ABSTRACT

The development and clinical application of ambulatory blood pressure monitoring (ABPM) has brought several of the main features of the circadian blood pressure (BP) rhythm to light. ABPM has shown to be a very useful method in cardiovascular risk assessment and remains the only method of diagnosing a non-dipping blood pressure profile. A ‘non-dipping’ BP profile is currently regarded as a risk factor in its own right for cardiovascular (CV) events and target organ damage. Nevertheless, the reliability of ABPM in assessing dipping status is still being questioned. Furthermore, a clear-cut definition of ‘non-dipping’ has not been established so far. The pathophysiological mechanism(s) of a non-dipping profile might involve abnormalities in extracellular volume and/or vascular resistance regulation. In addition, differences in daytime and nighttime activity, sleep quality and body position during sleep are involved as well. A reduction in cardiovascular risk by a pharmacologically induced switch from a non-dipper to a dipper status might be expected, but remains to be proven.

KEYWORDS

ABPM, non-dipping, hypertension, circadian

INTRODUCTION

Since the development and clinical application of ambulatory blood pressure monitoring (ABPM), various studies have shown that assessing the circadian blood pressure (BP) profile is more predictive than office BP readings in estimating cardiovascular (CV) risk. A special advantage of ABPM as compared with all other forms of BP measurements is that information is obtainable about BP during the night. Compared with daytime values BP in most subjects is considerably lower during the night and the attenuation of this physiological nocturnal decline should be regarded as abnormal. Besides being of interest for research purposes, the clinical relevance of this abnormality relates to its close association with hypertensive target organ damage, its increased risk for future CV events and its association with clinical conditions such as certain secondary forms of hypertension, renal function impairment and disturbances of the autonomic nervous system. Of note, a non-dipping profile in some circumstances might be favourable. For instance in non-dipping patients with an already challenged cerebral perfusion, a medically induced nocturnal BP lowering may induce hypoperfusion in that area.

In this overview we first go into the definition and reproducibility of a non-dipping BP profile, then we describe possible mechanisms and finally we discuss its clinical relevance.

DEFINITION

A ‘non-dipping BP profile’ is usually defined as a nocturnal BP fall of less than 10%. This definition requires further clarification. First of all, should one look at systolic BP (SBP) alone, or should diastolic BP (DBP) and/or mean arterial pressure (MAP) be taken into account as well? The effect of physical activity on SBP and DBP is unequal. With increased levels of activity there is an almost linear increase in SBP, whereas DBP tends to decrease. So, dipping classification may vary with the BP index taken. Most monitors used for ABPM measure BP oscillometrically. With this technique MAP, rather than SBP or DBP, is assessed most accurately. It might be
proposed, therefore, to use MAP as the BP index for classification of dipping status.

Secondly, arguments can be raised that in the definition, ‘nocturnal’ should be substituted with ‘sleeping’. The process of BP dipping is not likely to occur when a person does not sleep at night. Nightshift workers exemplify this. During the first 24-hour period of the nightshift, a dipping pattern switches to a non-dipping pattern. Gradually, the non-dipping pattern changes back to a dipping pattern during the following days. The BP dip in these subjects is then seen during their daytime sleeping period.\textsuperscript{16-21} To define the sleeping period, various (combinations of) methods are available. A simple method is to use diary card entries. Some prefer a short fixed time period to define nighttime, for instance from midnight to 6 am, thereby excluding to a large extent overlapping periods that patients may be either awake or have gone to bed.\textsuperscript{22} The morning BP surge will be excluded when this latter method is chosen. Since non-dippers have a rather modest morning BP surge as compared with dippers,\textsuperscript{23} this will only be of minor consequence for their nocturnal dipping percentage. Nevertheless, subjects classified as (borderline) dippers by the use of other methods might be classified as non-dippers with this method. A more recent method is the use of activity and posture monitoring, which is highly accurate, especially when combined with the diary card entry method.\textsuperscript{24} Finally, where does the 10% cut-off point come from, and why should a binary distribution be favoured over a continuous one? In our literature search we found no evidence of the 10% cut-off being more discriminative than other neighbouring cut-off points. Although arbitrary, the 10% cut-off is easy to use and seems to be quite practical so far.\textsuperscript{24}

**Reproducibility**

The clinical usefulness of a non-dipping BP pattern obviously depends on its reoccurrence from one occasion to another. Unfortunately, this is not always the case. For instance, a study by Manning et al. showed that only 54% of 79 untreated hypertensives and normotensives could be consistently classified as dippers, after performing three ABPMs within six months.\textsuperscript{25} In another study, in which two ABPMs were performed separated by more than one year in 170 hypertensives, no less than 40% had changed their dipping status. It should be remarked that although recordings were performed while patients were off antihypertensive treatment, between the two measurements subjects were treated with BP-lowering medication.\textsuperscript{26} A recent study showed more promising results in reproducibility.\textsuperscript{27} Sixty-five recently diagnosed untreated hypertensives underwent repeated ABPM. The nocturnal dipping pattern remained unchanged in 82% of the patients; 12% converted from a non-dipping to a dipping status after the repeated measurements. Also in studies in which physical activity during the day was observed more objectively, dipping status seemed to be more reproducible.\textsuperscript{28,29} A possible explanation is that subjects have a greater tendency to behave according to study protocol than when their activity is only controlled by a diary. They are also more likely to behave approximately the same during subsequent ABP measurements.

Change in body position from one night to another potentially affects reproducibility.\textsuperscript{30} When subjects are lying on their side rather than on their back, it can make a difference of about 12 to 14 mmHg if the cuff is attached to the upper arm or the forearm.\textsuperscript{31,32} Measurement of arm position during ABPM is possible with activity and posture monitoring systems. Using these systems correction of effects of changes in arm position during repeated recordings is possible. However, in a small study correcting for changes in body position during the night did not improve the reproducibility of dipping status.\textsuperscript{33}

In conclusion, reproducibility of dipping or non-dipping status is not perfect. Classification of dipping status and its reproducibility can be improved when measurements are done on like days, when daytime activity and duration of nighttime bed rest are objectively observed and when changes in the position of the cuffed arm during the night are taken into account.

**Proposed Mechanisms**

Inactivity and sleep are the two factors explaining the normally occurring nocturnal decline in BP. It might be argued, therefore, that daytime inactivity and poor sleep quality contribute to a decrease in this decline. For instance, it has been suggested that subjects with a more pronounced risk of CV events may be more likely to be more inactive during the day and therefore are also more prone to be diagnosed as non-dippers.\textsuperscript{34} Although daytime inactivity and poor sleep quality may explain the non-dipping phenomenon, contradictory arguments can be given as well. First, in studies comparing dippers and non-dippers daytime BP in both groups is usually similar.\textsuperscript{35} Second, non-dipping also occurs in patients with good sleep quality according to their diary input.\textsuperscript{36} Third, as summarised in table 1, non-dipping is related to a number of clinical conditions that usually have no influence on daytime activity and/or sleep quality.

Concerning the underlying haemodynamics, a normal dipping pattern is mainly due to a decrease in cardiac output (CO), whereas nighttime systemic vascular resistance (SVR) remains similar to daytime SVR or is even increased.\textsuperscript{37,38} The nocturnal decrease in CO is mainly
caused by a decrease in heart rate (HR), with stroke volume compared with daytime values remaining unchanged. A few studies have compared the day-night changes in CO and SVR in dippers and non-dippers. 19,33,36-41 The findings of these studies are not uniform. Thus a non-dipping profile might be caused by a diminished nocturnal decrease in CO, an exaggerated increase in SVR or a combination of these factors. An important reason for these discrepant findings is that the diurnal variation of CO and SVR, unlike BP, is strongly influenced by diurnal changes in posture and daily activity. 19

Looking at conditions associated with non-dipping may be helpful in explaining its underlying mechanism. Autonomic dysfunction is almost always associated with a non-dipping BP profile and sometimes even with nocturnal hypertension. 35,36 Due to impairment of the sympathetic nervous system (SNS), an excessive volume of blood will be pooled in the lower part of the body when assuming the upright position. In addition, the kidneys retain fluid retention during the day, which is related to a low renal perfusion pressure. When assuming the horizontal position this pooled blood is remobilised, causing an increase in stroke volume and CO and hence in BP, which cannot be counteracted by the baroreflex due to an impaired autonomic function. The observation that impaired renal function, hyperaldosteronism and hyperparathyroidism are frequently associated with non-dipping supports the role of excessive extracellular fluid volume in the pathogenesis of non-dipping. This is further substantiated by studies showing that in sodium-sensitive hypertensives a non-dipping BP can be converted to a dipping BP profile with a sodium-restricted diet or use of diuretics. 42,43 Although one study shows that the opposite can also occur. 44 In patients with the obstructive sleep apnoea syndrome and pheochromocytoma, a relatively high sympathetic tone or an increased concentration of circulating catecholamines are likely operative in the non-dipping BP pattern observed with these conditions. We suggest that in these conditions, both an inappropriate nocturnal increase in venous and arterial tone explain the non-dipping BP pattern.

RELEVANCE

The clinical relevance of establishing a non-dipping BP pattern lies in its proven association with more severe hypertensive target organ damage and its improved prediction of an increased CV risk, not only in hypertensive, but also in normotensive subjects. 45-49 Left ventricular hypertrophy, carotid intima-media thickening, microalbuminuria and cerebrovascular diseases are much more prevalent in non-dippers than in dippers. 1,4,50-52 Furthermore, it is well recognised that a non-dipping BP pattern is associated with renal function impairment. 11-14 Conversely, limited evidence indicates that such a pattern also accelerates the progression of renal dysfunction. 54-56

CONCLUSIONS

A non-dipping BP pattern has been well established as an entity with potentially important clinical implications. Although a nocturnal BP decline of less than 10% compared with daytime values is usually regarded as indicative for the diagnosis of non-dipping, it should be remarked that this threshold is arbitrary. In addition, it is not well settled which index of BP, i.e. SBP, DBP, MAP or some combination of these indexes, should be used. On theoretical grounds, we have argued to base the diagnosis on a less than 10% nocturnal decline of MAP. An unsolved problem is the imperfect reproducibility of a non-dipping status. Taking into account daytime and nighttime physical activity and subjective sleep quality and performing recordings on like days, reproducibility can almost certainly be improved. The mechanism underlying a non-dipping BP profile remains unknown.

Table 1. Associated conditions and other influences

<table>
<thead>
<tr>
<th>Endocrine conditions</th>
<th>Renal dysfunction</th>
<th>Disturbances of the autonomic nervous system</th>
<th>Miscellaneous</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aldosteronism 5,35</td>
<td>Chronic kidney damage 10,14</td>
<td>Pure autonomic failure 37,60</td>
<td>Salt-sensitive hypertension 46,48,50,64</td>
</tr>
<tr>
<td>Hypercortisolism 6,64</td>
<td>Renal transplantation 13,40</td>
<td>Diabetic neuropathy 65,67</td>
<td>Pre-eclamptic toxemia 58</td>
</tr>
<tr>
<td>Pheochromocytoma 59</td>
<td>Unilateral nephrectomy 55</td>
<td>Uraemic neuropathy 32</td>
<td>Malignant hypertension 70</td>
</tr>
<tr>
<td>Acromegaly 71</td>
<td></td>
<td>Familial amyloidotic polyneuropathy 72</td>
<td>Cardiac transplantation 71,74</td>
</tr>
<tr>
<td>Hyperthyroidism 75</td>
<td></td>
<td>Obstructive sleep apnoea syndrome 75</td>
<td>Ethnicity 77,77</td>
</tr>
<tr>
<td>Hyperparathyroidism 78</td>
<td></td>
<td></td>
<td>Disturbances in circadian plasma melatonin changes 79</td>
</tr>
</tbody>
</table>

* Use of immunosuppressive therapy may play a role as well. 33,34,35 ** People of African ancestry have a higher prevalence of non-dipping than Caucasians.
in many instances. Evidence is accumulating that volume-related factors are frequently involved. This explains the association of non-dipping with salt-sensitive forms of hypertension, renal function impairment and mineralocorticoid-induced forms of hypertension. How this volume dependency of BP translates into a non-dipping BP pattern requires further investigation.

Until the present day, there are still no specific therapeutic recommendations based on dipping status. As discussed, there is limited evidence that with certain antihypertensive agents, for instance diuretics, or changing the timing of drug administration, a non-dipping BP pattern can be reversed into a dipping pattern. Whether CV outcome improves by changing the dipping status pharmacologically remains to be proven.

ACKNOWLEDGMENTS

A.M.B. is supported by a fellowship of the Dutch Kidney Foundation (Grant application Co4.2087).

REFERENCES


43. Uzu T, Kirmura G. Diuretics shift circadian rhythm of blood pressure from non-dipper to dipper in essential hypertension. Circulation 1999;100:1655-6.
Ewing’s sarcoma and primitive neuroectodermal tumours in adults: single-centre experience in the Netherlands

C.H. Smorenburg¹, C.J. van Groeningen¹, O.W.M. Meijer², M. Visser³, E. Boven*¹

¹Departments of Medical Oncology, ²Radiotherapy, ³Pathology, VU University Medical Center, Amsterdam, the Netherlands, ⁴Present address: Department of Internal Medicine, Medical Centre Alkmaar, PO Box 501, 1800 AM Alkmaar, the Netherlands, ⁵corresponding author: tel.: +31 (0)20-444 43 36, fax: +31 (0)20-444 40 79.

ABSTRACT

Background: Ewing’s sarcoma and peripheral primitive neuroectodermal tumours (PNET) are rare tumours and closely related. They occur most often in children and adolescents. Few studies have been published on treatment outcome in adult patients.

Methods: We performed a retrospective analysis of patients aged >16 years who were primarily treated at our university hospital for Ewing’s sarcoma or PNET. In general, treatment consisted of long-term multiantiagent chemotherapy, interrupted by individualised local treatment consisting of surgery and/or radiotherapy. We reviewed clinical features and outcomes to present our experience with Ewing’s sarcoma and PNET in adults.

Results: From 1979 to 2002, 27 patients with Ewing’s sarcoma (20) or PNET (7) were treated. There were 22 men and 5 women, with a median age of 25 years (range 17-49). Ten patients presented with metastases predominantly in lungs (4) or bones (6). Combination therapy consisted of chemotherapy (27), surgery (16) and radiotherapy (16). After a median follow-up of ten years, 14 patients have died (toxicity = 2, progressive disease = 12) and 13 patients are alive and free of disease. Five-year overall survival was 58%. All four patients with bone metastases died, while all five patients presenting with lung metastases are disease-free. Five-year overall survival was 58%.

All four patients with bone metastases died, while all five patients presenting with lung metastases are disease-free. Conclusion: The five-year overall survival of 58% in this small series on adult patients is in line with paediatric study outcomes. Patients with lung metastases may even be cured by multimodality therapy. We therefore strongly advocate referral of patients with this rare disease to a specialised oncology centre.

KEYWORDS

Adults, Ewing’s sarcoma, PNET

INTRODUCTION

The family of Ewing’s sarcoma forms a distinct entity within the group of malignant mesenchymal tumours which includes Ewing’s sarcoma of bone and soft tissue, peripheral primitive neuroectodermal tumour (PNET) and Askin tumour (PNET of the thoracic wall). The tumour was named after James Ewing, an American pathologist, who was the first to describe this disease in 1921. Morphologically, Ewing’s sarcoma and PNET are small round-cell tumours consisting of undifferentiated cells with uniform nuclei and scanty cytoplasm. The tumours are characterised by the expression of O13, a cell-surface antigen encoded by mica. Approximately 95% of patients with Ewing’s sarcoma have a characteristic t(11;22)(q24;q12) or t(21;22)(q22;q12) chromosomal translocation, which results in fusion of the EWS gene on chromosome 22 and the FLI 1 gene on chromosome 11 or the ERG gene on chromosome 21. This translocation results in a chimeric transcription factor containing a DNA-binding domain. In vitro, the EWS/ETS fusion protein blocks the differentiation of pluripotent marrow stromal cells, which suggests a critical function in tumorigenesis of Ewing’s sarcoma. The majority of patients with Ewing’s sarcoma and PNET are younger than 30 years of age, with a peak incidence at the age of 15 years. Ewing’s sarcoma is the second most common primary malignancy of bone, with an annual incidence in all ages of 0.6 per million in the United Kingdom. The overall male to female ratio is approximately 1.5:1, bone being the primary site in 60% of cases. Although Ewing’s sarcoma is a rare tumour, every physician should realise that adequate therapy of Ewing’s sarcoma requires a multidisciplinary approach right at the time of diagnosis. Treatment consists of multiantiagent chemotherapy for a prolonged period of time, even in apparently nonmetastatic disease, because of the high risk of early haematogenous metastases.
feasible, chemotherapy is interrupted by aggressive local therapy of the primary tumour consisting of surgery and/or radiotherapy. This multimodality treatment has resulted in a remarkable improvement in overall five-year survival of only 20% in the 1960s to approximately 50% nowadays. The optimal drug combination and duration of multiagent chemotherapy is being investigated by international study groups, such as the (European Intergroup) Cooperative Ewing’s Sarcoma Study (EI)CESS and the American Paediatric Oncology Group (POG). Of notice, the vast majority of patients included in these studies are children and adolescents. Although almost half of all patients are being treated by medical oncologists in the adult setting, only few studies have reported treatment outcomes for adult patients.

To investigate the outcome of multimodality treatment in adult patients in the VU University Medical Center, we performed a retrospective analysis of patients treated at our university hospital.

METHODS

Patients
Using the registration of diagnosis of the Departments of Medical Encoding and Pathology, we collected the names of patients aged >16 years who were primarily treated at our hospital for Ewing’s sarcoma or PNET between 1979 and 2002. The following characteristics were registered from the patient charts: age, gender, size and localisation of the primary tumour, presence and localisation of metastases. In addition, treatment modalities (cytotoxic agents, number of cycles of chemotherapy, surgery, radiotherapy), treatment outcome (response, progression, pathological response), time to progression, time to death or end of follow-up were recorded. This analysis used data obtained until January 2006.

Diagnosis of all cases was based on biopsy specimens, which were reviewed by an experienced pathologist at our centre. The diagnosis of PNET is based on markers of neuroepithelial differentiation and has been used internationally since 1988. The method of detection of characteristic chromosomal translocations using reverse transcriptase polymerase chain reaction (RT-PCR) became available in 1993, and was not routinely performed in this series.

Staging
Staging was based on physical examination, computed tomography (CT) scan or MRI scan of the primary tumour, CT scan of the chest and bone scan. In addition, a bone marrow aspiration was taken as part of the initial staging procedure. During treatment, the response of tumour lesions was evaluated every three months using similar radiographic techniques. Responses to chemotherapy were defined as progressive disease (any new lesion or increase in tumour size), stable disease (<50% decrease in tumour size), partial response (>50% tumour size reduction) and complete response (no viable tumour cells in resected pathology specimen).

Treatment
The standard regimen of chemotherapy has only slightly changed throughout time. Until 1992, chemotherapy consisted of 12 cycles of three weeks of vincristine, actinomycin D, cyclophosphamide and adriamycin (VACA) or (for patients with a high risk of recurrence) vincristine, actinomycin D, ifosfamide and adriamycin (VAIA), according to the schedule used in the CESS 86 study. After 1992, patients received 14 cycles of etoposide, vincristine, actinomycin D, ifosfamide and adriamycin (EVAIA), as given in the EICES 92 protocol for high-risk patients. According to this protocol, chemotherapy was repeated every three weeks (= 1 cycle), while adriamycin was alternated with actinomycin D. In case of insufficient bone marrow recovery (white blood cell count <2.0 x 10^9/l and/or platelets <80 x 10^9/l), the next cycle of chemotherapy was postponed and granulocyte-colony stimulating factor (G-CSF) was added to subsequent cycles. Chemotherapy was interrupted for local therapy of the primary tumour after four to six cycles. Local treatment was individualised and consisted of surgery, surgery followed by radiotherapy, or radiotherapy only. Preferably, a wide excision of the tumour was performed. Otherwise, a marginal resection was followed by radiotherapy or, in the case of (functional) irresectability, only radiotherapy was given. If histological examination of a radically resected tumour revealed more than 10% of vital tumour cells, radiotherapy was also administered postoperatively. Radiotherapy was given at a dose of 45 to 55 Gy in 25 to 30 fractions, depending on the individual indication. During radiotherapy, chemotherapy was continued in which actinomycin D and adriamycin were temporarily omitted. Adjuvant radiotherapy of lung metastases was scheduled after completing chemotherapy.

Statistics
Event-free and overall survival were estimated by the Kaplan-Meier method, using the computer programme SPSS version 9.0. Group comparisons were made using the log-rank test.

RESULTS

Patient characteristics
From 1979 to 2002, 27 patients with Ewing’s sarcoma (20) or PNET (7) were primarily treated in our hospital. Another seven patients with Ewing’s sarcoma or PNET were referred because of recurrent disease and were not included in this
series. Patient characteristics are depicted in table 1. Twenty-two men (81%) and five women (19%) were diagnosed with a median age of 25 years (range 17 to 49). The primary tumour originated in the bone in 14 patients (56%) (humerus = 3, pelvis = 3, femur = 2, tibia = 2, spine = 1, fibula = 1, metatarsal = 1, ethmoid = 1) and had an extraneous origin in 13 patients (chest wall = 5, soft tissue = 4, upper limb = 1, lower limb = 2, leptomeningeal = 1). The median tumour size was 8 cm (range 3.3 to 25 cm). At presentation, 10 out of 27 patients (37%) had metastatic disease, predominantly in the lungs (6) and bones (4). A bone marrow aspiration was performed in 14 patients, being normal in 12 patients. In two patients with tumour cells in bone marrow aspirate the diagnosis of bone metastases had already been made. RT-PCR analysis of characteristic chromosomal translocations was not routinely performed; analysis in three out of three patients revealed a t(11;22) translocation.

Treatment
An overview of treatment modalities is given in table 2. Chemotherapy was given to all patients, three of whom were treated with the VAIA regimen, five with the VACA regimen and 19 (70%) with the EVAIA regimen. The median number of cycles was nine (range 1 to 14). Regimen and 19 (70%) with the EVAIA regimen. The median number of cycles was nine (range 1 to 14).

\(table 2\)

<table>
<thead>
<tr>
<th>Variable</th>
<th>No. of patients (n=27)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>5</td>
</tr>
<tr>
<td>Male</td>
<td>22</td>
</tr>
<tr>
<td>Age</td>
<td></td>
</tr>
<tr>
<td>Median (years)</td>
<td>25</td>
</tr>
<tr>
<td>Range (years)</td>
<td>17-49</td>
</tr>
<tr>
<td>&gt;20 years</td>
<td>16</td>
</tr>
<tr>
<td>Site of primary tumour</td>
<td></td>
</tr>
<tr>
<td>Lower limb</td>
<td>8</td>
</tr>
<tr>
<td>Pelvis</td>
<td>3</td>
</tr>
<tr>
<td>Spine</td>
<td>1</td>
</tr>
<tr>
<td>Soft tissue</td>
<td>4</td>
</tr>
<tr>
<td>Chest wall</td>
<td>5</td>
</tr>
<tr>
<td>Upper limb</td>
<td>4</td>
</tr>
<tr>
<td>Head and neck</td>
<td>2</td>
</tr>
<tr>
<td>Size of primary tumour</td>
<td></td>
</tr>
<tr>
<td>&lt;5 cm</td>
<td>6</td>
</tr>
<tr>
<td>5-10 cm</td>
<td>8</td>
</tr>
<tr>
<td>&gt;10 cm</td>
<td>11</td>
</tr>
<tr>
<td>Unknown</td>
<td>2</td>
</tr>
<tr>
<td>Site of metastasis</td>
<td></td>
</tr>
<tr>
<td>Lung only</td>
<td>5</td>
</tr>
<tr>
<td>Lung + bone + bone marrow</td>
<td>1</td>
</tr>
<tr>
<td>Bone only</td>
<td>2</td>
</tr>
<tr>
<td>Bone + bone marrow</td>
<td>1</td>
</tr>
<tr>
<td>Liver + peritoneum</td>
<td>1</td>
</tr>
</tbody>
</table>

Response to first-line chemotherapy in 22 patients was partial or complete response (13), stable disease (3), progressive disease (4) and toxic death (2). Nine out of 13 patients with a response to chemotherapy are alive and disease-free. Three patients with stable disease and four patients with progressive disease finally died of the disease despite second-line chemotherapy in two of them. Two patients with bone metastases died of neutropenic sepsis after the first cycle. Of notice, bone marrow aspirates of both of them showed tumour cells. Four patients received chemotherapy (4 to 14 courses) after complete resection of the primary tumour, and are still alive. Second-line chemotherapy was used for progressive (2) or recurrent (2) disease in four patients and consisted of VAI, cisplatin/etoposide or carboplatin/etoposide. All of them died of progressive disease due to lack of treatment response.

In four patients, complete resection was the initial treatment due to an undefined diagnosis of the incisional biopsy. Chemotherapy was given postoperatively due to a final diagnosis of Ewing’s sarcoma or PNET. After a follow-up of 3.1 to 18.3 years, these four patients are alive and disease-free. Resection of the primary tumour was performed after three to five cycles of chemotherapy in 12 patients. Data on resection margins of one patient could not be retrieved. Resection margins were free of tumour in seven patients and irradical in four patients, respectively. Data on viability of tumour cells were lacking in five patients. All three patients whose tumour did not contain any vital cells at histological examination are alive. Three out of four patients with vital tumour cells died of progressive disease.

Radiotherapy was given on the site of the primary tumour in 15 patients, in nine of them after preceding surgery. One patient with leptomeningeal disease started with radiotherapy on the neuraxis, followed by chemotherapy. This patient died of progressive disease.

After completing chemotherapy, five patients received radiotherapy for lung metastases (whole lung irradiation = 3, partial lung irradiation = 2). At present, these five patients are disease-free and may be considered as being cured after a follow-up of 6.8 to 26.5 year.

Survival
After a median follow-up of 10.0 years (range 3.2 to 26.5 years), a total of 14 patients have died and 13 patients are alive and disease-free (table 2). Figure 1 depicts the survival probability according to Kaplan-Meier. In our series, five-year overall survival was 58%, with a median overall survival of 120 months (95% CI 107 to 227). The five-year overall survival was 52 and 60% for patients with nonmetastatic and metastatic disease, respectively (not significant). Of notice, all four patients with bone metastases died of either progressive disease (2) or neutropenic sepsis (2), while five out of six patients with lung metastases were cured (p<0.005, log rank). In this study, no other significant correlations were observed between patient characteristics and survival.
DISCUSSION

This is the first Dutch series of adult patients with Ewing’s sarcoma and PNET. The five-year overall survival of 58% in our patients with nonmetastatic disease is quite similar to the reported series on adult patients that range from 35 to 60% (Table 3).8-14 In our series, five-year overall survival was not worse in patients with metastatic disease as compared with nonmetastatic disease, which is likely due to the small number of patients with metastasis (n=10) in our series. Larger series report an overall survival of 20 to 30% for patients with metastatic disease.17 This modest outcome, however, still contrasts favourably with low five-year overall survival rates of the majority of solid malignant tumours with distant metastases.

Poor prognostic factors for Ewing’s sarcoma and PNET in adult patients are large tumours,9,10 primary tumour of the pelvis,8,10 metastases at presentation11,12 and advanced age.11 Similar prognostic factors were observed in two large studies including mainly paediatric patients.17,18 Advanced age as a poor prognostic factor may either be related to biological aspects of more aggressive disease or a lower dose intensity of chemotherapy to be delivered. Preliminary data of a large retrospective German analysis of 1426 patients reported both age >15 years at diagnosis and treatment outside paediatric oncology units as significantly poor prognostic factors.19 A large study in children has reported a five-year overall survival of 61 to 72%,18 which is higher than that observed in adult patients.8-14 In two other studies in adult patients, however, age >30 years did not appear to be related to a dismal outcome.10,13 Metastases at presentation are correlated with a poor prognosis.11,12 The localisation of metastases, however, is of importance. We observed a long-term disease-free survival in five out of six patients with lung metastases. In contrast,

Table 2. Primary treatment modalities and treatment outcome

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. of patients</th>
<th>No. of patients alive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chemotherapy alone</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>CT → Surgery → CT</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>CT → Radiotherapy → CT</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>CT → Surgery + Radiotherapy → CT</td>
<td>9</td>
<td>4</td>
</tr>
<tr>
<td>Surgery → Chemotherapy</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Radiotherapy → Chemotherapy</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>

CT = chemotherapy.

Table 3. Survival data in adult patients with Ewing’s sarcoma or PNET

<table>
<thead>
<tr>
<th>Author</th>
<th>Ref.</th>
<th>Chemotherapy</th>
<th>No. of patients</th>
<th>No. of patients with M1</th>
<th>5-year overall survival (%)</th>
<th>5-year overall survival in M0 (%)</th>
<th>5-year overall survival in M1 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sincovics</td>
<td>8</td>
<td>VACA</td>
<td>50</td>
<td>16</td>
<td>35</td>
<td>52</td>
<td>19</td>
</tr>
<tr>
<td>Verrill</td>
<td>9</td>
<td>IVAD</td>
<td>59</td>
<td>17</td>
<td>52</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Fizzazi</td>
<td>10</td>
<td>VACA, IVAD, VAD</td>
<td>182</td>
<td>53</td>
<td>54</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>Baldini</td>
<td>11</td>
<td>VAD or VACA</td>
<td>37</td>
<td>11</td>
<td>49</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Bacci</td>
<td>12</td>
<td>VACA, EVAIA</td>
<td>23</td>
<td>n.k.</td>
<td>53</td>
<td>n.k.</td>
<td>n.k.</td>
</tr>
<tr>
<td>Martin</td>
<td>13</td>
<td>Combination n.k.</td>
<td>59</td>
<td>9</td>
<td>60</td>
<td>33</td>
<td></td>
</tr>
<tr>
<td>Laurence</td>
<td>14</td>
<td>High dose + PSCT</td>
<td>46</td>
<td>10</td>
<td>63</td>
<td>71</td>
<td>34</td>
</tr>
<tr>
<td>This study</td>
<td></td>
<td>VACA, VAIA or EVAIA</td>
<td>27</td>
<td>10</td>
<td>58</td>
<td>52</td>
<td>60</td>
</tr>
</tbody>
</table>

M0 = no metastases; M1 = metastatic disease; PSCT = peripheral stem cell transplantation; n.k. = not known; n.s. = not significant.

VACA = vincristine, actinomycin D, cyclophosphamide, adriamycin; IVAD = ifosfamide, vincristine, adriamycin, actinomycin D; EVAIA = etoposide, vincristine, actinomycin D, ifosfamide, adriamycin.
all four patients with bone metastases, including a patient with simultaneous lung metastases, died of progressive disease. Likewise, paediatric studies have also reported a better prognosis for patients with lung metastases as compared with those with bone metastases. Although two out of five patients with lung metastases received successful partial irradiation, whole lung irradiation in such patients is being advised in EICESS protocols. According to EICESS protocols, two separate bone marrow biopsies should be performed as part of the initial staging procedure to define the intensity of treatment. In our series in which patients did not participate in a clinical trial, a bone marrow aspiration was only taken in half of the patients. Histopathological examination revealed bone marrow metastases in two patients, who also had bone metastases visualised on a bone scan. Both patients died of neutropenic sepsis. In addition to immunohistochemistry, Schleiermacher et al. used a PCR technique targeting EWS-specific transcripts and detected micrometastases in bone marrow in 18 out of 92 patients (20%) with Ewing’s sarcoma. This subgroup of patients indeed had an impaired prognosis and an increased risk of systemic relapse. Detection of micrometastases in bone marrow, however, lacks therapeutic consequences and PCR on bone marrow is therefore not routinely recommended.

None of the four patients with progressive or recurrent disease in our study responded to second-line chemotherapy. Progressive or recurrent disease still has a poor outcome, despite aggressive therapy including high-dose chemotherapy and peripheral stem cell transplantation (PSCT). Thus far, no standard chemotherapy can be recommended in this setting and identification of novel agents is warranted. Despite improvement in the treatment outcome of Ewing’s sarcoma and PNET during the past decades, almost half of all patients ultimately die of this disease. It is clear that for optimal efficacy of combined modality therapy with a curative intention, a multidisciplinary approach by experienced medical oncologists, surgeons and radiotherapists is essential. With regard to the low incidence of this tumour, we therefore strongly advocate referral of these patients to a specialised oncology centre and participation in international trials.

REFERENCES

Cushing’s syndrome and bone mineral density: lowest Z scores in young patients

A.W. van der Eerden¹, M. den Heijer¹²*, W.J Oyen³, A.R Hermus¹

Departments of ¹Endocrinology, ²Epidemiology and Biostatistics, ³Nuclear Medicine, Radboud University Nijmegen Medical Centre, Nijmegen, the Netherlands; *corresponding author: tel.: +31 (0)24-361 45 99, fax.: +31 (0)24-361 88 09, e-mail: m.denheijer@endo.umcn.nl.

ABSTRACT

Background: Patients with Cushing’s syndrome have a high prevalence of osteoporotic fractures. Little is known about factors determining bone mineral density (BMD) in these patients.

Objective: To evaluate which factors influence BMD at the time of diagnosis of Cushing’s syndrome.

Methods: In 77 consecutive patients with Cushing’s syndrome with a median age of 41.1 (interquartile range 31.1 to 52.2) years we measured BMD of the lumbar spine and the femoral neck at the time of diagnosis. From the medical records we obtained information on possible predictors of BMD. We compared BMD with a reference population by means of the Z score. Adjustment for other variables than age and sex was made with linear regression models.

Results: Patients with Cushing’s syndrome had a low Z score in both the lumbar spine (–1.07 SD (95% CI –1.43 to –0.71 SD)) and in the femoral neck (–0.81 SD (95% CI –1.06 to –0.55 SD)). 82% of patients had osteopenia at one or both sites (T score lower than –1 SD), including 31% with osteoporosis (T score –2.5 SD or lower). The main determinant of the Z score at both sites and for both sexes was age. Z score increased by about 0.4 SD per decade. 81% of patients <40 years had osteopenia at one or both sites, including 31% with osteoporosis. For patients ≥40 years these percentages were 83 and 32%, respectively.

Origin of Cushing’s syndrome, average 24-hour serum cortisol, duration of symptomatic glucocorticoid excess, sex, body mass index, menstrual status and duration of amenorrhoea did not influence the Z score.

Conclusions: At the time of diagnosis, BMD in patients with Cushing’s syndrome is low compared with sex- and age-matched controls. The prevalence of osteopenia and osteoporosis at the time of diagnosis of Cushing’s syndrome is independent of age. Treatment with bisphosphonates should be considered in patients of all ages with Cushing’s syndrome who have a decreased BMD.

KEYWORDS

Cushing, endogenous glucocorticoid excess, bone mineral density, Z score, age, osteoporosis

INTRODUCTION

The association between hypercorticism and loss of skeletal mass was first described by Harvey Cushing.¹ It is most likely that several factors act together in causing bone loss in Cushing’s syndrome. Glucocorticoids impair bone formation through direct effects on cells of osteoblastic lineage and they enhance bone resorption due to direct effects on osteoclasts.²,³ So, the coupling between bone resorption and bone formation, crucial to the normal process of bone remodelling, appears to be lost in active Cushing’s syndrome.⁴ Furthermore, there is a decrease in intestinal calcium absorption and in renal calcium reabsorption.⁵,⁶ Glucocorticoid-induced inhibition of gonadotropin and growth hormone secretion may also play a role, as well as glucocorticoid-induced decrease in muscular strength, leading to impaired physical activity.⁵,⁷ Bone loss due to treatment with pharmacological doses of glucocorticoids is more profound in trabecular than in cortical bone.⁵,⁸ This is supposedly related to the greater surface-to-volume ratio of trabecular bone compared with cortical bone. Since bone remodelling takes place at bone surfaces, trabecular bone responds more rapidly to either positive or negative changes in bone balance.⁹ In patients with endogenous hypercortisolism, bone loss in the lumbar spine, consisting mainly of trabecular bone, also seems to be more profound than bone loss in the femoral neck, consisting mainly of cortical bone.⁴,¹⁰⁻¹² So far, little is known about factors that determine bone mineral density (BMD) in patients with untreated Cushing’s syndrome. The aim of the present study was to evaluate which factors influence BMD at the time of diagnosis of Cushing’s syndrome.
PATIENTS AND METHODS

Between 1989 and 2003, all patients newly diagnosed with Cushing’s syndrome referred to the Department of Endocrinology of the Radboud University Nijmegen Medical Centre in Nijmegen, the Netherlands underwent measurement of BMD of both the lumbar spine (L1-L4, posterior-anterior) and the right femoral neck. Before treatment of hypercortisolism, BMD was measured by dual-energy X-ray absorptiometry (DEXA, Hologic Inc., Waltham MA, before 1998 QDR-1000, thereafter QDR-4500 Elite). The scanner was calibrated daily by means of phantom measurements. Vertebrä that showed artefacts due to fracture, spinal deformation and/or degenerative disease were excluded from the analysis. For each measured value of BMD (g/cm²) a T score and a Z score were calculated. A Z score of 0 SD means that the BMD is average for age and sex. From the medical records we also derived information on possible predictors of BMD.

To assess the effect of endogenous hypercortisolism on BMD we used the Z score, which implicitly means comparison with a reference group. To assess determinants of BMD within the group of Cushing patients we also used the Z score as measure of BMD, which means that comparisons are adjusted for age and sex. The comparisons were made by calculating mean differences in Z score between subgroups with corresponding 95% confidence intervals (95% CI). Adjustment for other variables than age and sex were made with linear regression models. To assess correlations between variables, we used Pearson correlation coefficients. To evaluate the risk for osteoporotic fractures we used the T score.

RESULTS

A total of 58 patients with untreated pituitary-dependent and 21 patients with untreated adrenal-dependent Cushing’s syndrome were seen during the study period. For unknown reasons, BMD measurements were not carried out in two patients with pituitary-dependent Cushing’s syndrome. Furthermore, we excluded patients with ectopic ACTH secretion from our analysis. Thus, BMD values of 56 patients with pituitary-dependent and 21 patients with adrenal-dependent Cushing’s syndrome (15 adrenal adenoma, 4 adrenal carcinoma, and 2 bilateral macronodular hyperplasia) were available. The group consisted of 19 men and 58 women, median age 41.1 (interquartile range 31.1 to 52.2) years. The average level of serum cortisol in blood samples taken over 24 hours at four hourly intervals was median 0.578 (interquartile range 0.498 to 0.668) μmol/l. Of the women, 21 were oligomenorrhoeic or eumenorrhoeic, whereas 31 women were postmenopausal or had been amenorrhoeic for at least six months secondary to the Cushing’s syndrome.

Two patients had undergone extirpation of the uterus, and four women were on oral contraceptive agents. Median body mass index was 27.7 (interquartile range 25.2 to 32.5) kg/m². The characteristics of patients of different ages, and patients with Cushing’s syndrome of different origins, did not differ with respect to the variables used in our analyses, except of course that older female patients were more often amenorrhoeic. None of the patients had previously taken drugs known to interfere with bone metabolism.

BMD at diagnosis of Cushing’s syndrome

Before treatment, patients with Cushing’s syndrome had a low mean BMD. The decrease in BMD seemed to be more severe in the lumbar spine than in the femoral neck, although the difference was not statistically significant. In the lumbar spine we found a mean BMD of 0.89 g/cm² (95% CI 0.85 to 0.93 g/cm²), corresponding to a mean Z score of –1.07 SD (95% CI –1.43 to –0.71 SD), whereas in the femoral neck a mean BMD of 0.76 g/cm² (95% CI 0.73 to 0.79 g/cm²) was found with a mean Z score of –0.81 SD (95% CI –1.06 to –0.55 SD). In 73% of the patients, the Z score at one or both sites was lower than –1 SD, including 16% who had a Z score of –2.5 SD or lower. A total of 82% had osteopenia at one or both sites, including 31% with osteoporosis according to World Health Organisation criteria. There was no significant difference in Z score between Cushing syndromes of different origins. For patients with pituitary-dependent Cushing’s syndrome we found mean Z scores in the lumbar spine of –1.08 SD (95% CI –1.52 to –0.61 SD) and in the femoral neck of –0.66 SD (95% CI –0.99 to –0.33 SD). However, for those with adrenal-dependent Cushing’s syndrome we found Z scores in the lumbar spine of –1.14 SD (95% CI –1.52 to –0.74 SD) and in the femoral neck of –1.14 SD (95% CI –1.55 to –0.74 SD).

Factors predictive of BMD

Table 1 shows the relative influence of various variables on the Z score at the time of diagnosis of Cushing’s syndrome. Variables related to Cushing’s syndrome, such as origin of Cushing’s syndrome, average 24-hour serum level of cortisol and duration of symptomatic glucocorticoid excess, did not exert an influence on Z scores. In females, BMD of the femoral neck was lower than in males. When correcting for the other variables analysed, femoral neck BMD in females was 0.13 g/cm² (95% CI 0.05 to 0.21) lower than in males. Interestingly, in males as well as in females, age correlated positively with Z score at both the lumbar spine and the femoral neck. Z scores increased by about 0.4 SD per decade (figure 1). Of female patients <40 years, 82% had a T score lower than –1 SD at one or both sites and 27% had a T score of –2.5 SD or lower. For female patients ≥40 years these percentages were 86 and 31%, respectively. Percentages for male patients were comparable. Of male patients under the age of 40 years, 79% had a T score lower.
than –1 SD at one or both sites including the 36% who had a T score of -2.5 SD or lower. For male patients ≥40 years these percentages were 60 and 40%, respectively.

**DISCUSSION**

The main finding of the present study is that in male as well as in female patients with Cushing’s syndrome, age correlated positively with Z scores, both in the lumbar spine and in the femoral neck. In Cushing patients of either sex the prevalence of osteopenia as well as of osteoporosis at either the lumbar spine or the femoral neck or at both sites was the same in patients <40 years compared with patients of ≥40 years. This explains why also young patients with Cushing’s syndrome have a considerable fracture risk.

We found that in patients with newly diagnosed endogenous Cushing’s syndrome, the mean Z score is approximately 1 SD below control values, consistent with previous reports. The decrease in BMD seems to be more severe in the lumbar spine than in the femoral neck; however, as in previous reports the difference was not statistically significant. Our data do not show a significant difference in BMD between patients with adrenal-dependent and pituitary-dependent Cushing’s syndrome, neither in the group as a whole, nor in male or female patients separately. This confirms the findings in a very recent study by Tauchmanová et al. In contrast, Ohmori et al. did find a lower BMD in females with adrenal-dependent Cushing’s syndrome than in females with pituitary-dependent Cushing’s syndrome, both in the lumbar spine and in the femoral neck. In their study body mass index was significantly higher in patients with a pituitary origin of the disease than in those with an adrenal origin, which may explain their findings. Minetto et al. also found that lumbar spine BMD, but not femoral neck BMD, was lower in patients with adrenal-dependent than in patients with pituitary-dependent Cushing’s syndrome.

In our study severity of Cushing’s syndrome, expressed as average 24-hour cortisol levels in serum, and duration of symptomatic glucocorticoid excess, did not influence Z scores. These findings are in line with observations in patients treated with pharmacological doses of glucocorticoids. In patients treated with glucocorticoids the daily dose does not correlate with BMD, and bone loss occurs particularly during the first months of treatment. At diagnosis of Cushing’s syndrome, the duration of glucocorticoid excess is usually more than a year, so it is not surprising that we did not observe a correlation between duration of symptomatic glucocorticoid excess and BMD. In our group of patients neither body mass index nor menstrual status showed a relation with Z scores. This is in line with observations published in a very recent report, but is in contrast with the situation in healthy subjects, where body mass index and menstrual status strongly influence BMD. The calculation of Z scores for BMD as measured by the two types of DEXA scanners used in our study was based on the same reference population, i.e. NHANES reference

### Table 1. Influence of variables on Z score (SD) of the lumbar spine (L1-L4) and the femoral neck at the time of diagnosis of Cushing’s syndrome

<table>
<thead>
<tr>
<th>Lumbar spine</th>
<th>Linear regression coefficient</th>
<th>Linear regression coefficient</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Origin of Cushing’s syndrome (adrenal versus pituitary)</strong></td>
<td>-0.08 (-0.35 to 0.70)</td>
<td>0.04 (-0.03 to 0.07)</td>
</tr>
<tr>
<td>Average 24h serum cortisol (µmol/l)</td>
<td>-1.61 (-3.89 to 0.67)</td>
<td>0.04 (-0.02 to 0.06)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>0.04 (0.02 to 0.06)</td>
<td>0.04 (-0.02 to 0.06)</td>
</tr>
<tr>
<td>Sex (male vs. female)</td>
<td>-0.69 (-1.47 to 0.09)</td>
<td>-0.47 (-1.03 to 0.06)</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>0.17 (0.05 to 0.88)</td>
<td>0.17 (0.05 to 0.88)</td>
</tr>
<tr>
<td>Secondary amenorrhoea vs oligo-/eumenorrhoea</td>
<td>0.04 (-0.02 to 0.11)</td>
<td>0.04 (-0.02 to 0.11)</td>
</tr>
<tr>
<td>Length of period of secondary amenorrhoea before DEXA measurement (years)</td>
<td>0.04 (0.01 to 0.06)</td>
<td>0.04 (0.01 to 0.06)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Femoral neck</th>
<th>Linear regression coefficient</th>
<th>Linear regression coefficient</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Origin of Cushing’s syndrome (adrenal vs. pituitary)</strong></td>
<td>-0.47 (-1.01 to 0.09)</td>
<td>0.04 (-0.02 to 0.06)</td>
</tr>
<tr>
<td>Average 24h serum cortisol (µmol/l)</td>
<td>0.35 (1.20 to 1.99)</td>
<td>0.04 (0.01 to 0.05)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>0.04 (0.01 to 0.05)</td>
<td>0.04 (0.01 to 0.05)</td>
</tr>
<tr>
<td>Sex (male vs. female)</td>
<td>0.47 (-0.12 to 1.06)</td>
<td>0.47 (-0.12 to 1.06)</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>0.00 (-0.05 to 0.04)</td>
<td>0.00 (-0.05 to 0.04)</td>
</tr>
<tr>
<td>Secondary amenorrhoea vs oligo-/eumenorrhoea'</td>
<td>0.32 (-0.26 to 0.91)</td>
<td>0.32 (-0.26 to 0.91)</td>
</tr>
<tr>
<td>Length of period of secondary amenorrhoea before DEXA measurement (years)</td>
<td>0.01 (-0.04 to 0.06)</td>
<td>0.01 (-0.04 to 0.06)</td>
</tr>
</tbody>
</table>

*Adjusted for age, sex, body mass index, origin of Cushing’s syndrome, and average 24-hour serum cortisol; **Average of serum cortisol in blood samples taken at 0.00 h, 4.00 h, 8.00 h, 12.00 h, 16.00 h, and 20.00 h; *Secondary amenorrhoea is defined as postmenopausal or amenorrhoeic for at least 6 months secondary to Cushing’s syndrome.

population. Besides presenting Z-scores, for the purpose of clinical interpretation we pooled BMD values obtained by two types of DEXA scanners. This pooling is justified by the results of a study in which Litaker et al. compared repeatedly measured values of BMD within 219 individuals, as obtained by Hologic QDR-1000/W and 4500W DXA scanners. The difference between the mean values as measured by the two types of machines was only 0.02 g/cm$^2$ in white females and 0.05 g/cm$^2$ in white males. The differences found by Litaker et al. clearly pose a problem for the interpretation of longitudinal follow-up studies, measuring the gradual effect of the course of disease or cure within individuals. For cross-sectional studies, in contrast, describing a point estimate for the large effect of a situation such as the presence of Cushing’s syndrome, the disagreement between the machines is negligible. Indeed, the difference between the machines is far smaller than the difference we found between white, mostly female, patients with Cushing’s syndrome and the NHANES reference population (i.e. a decrease of 1.07 SD = 0.11 g/cm$^2$ in lumbar spine and a decrease of 0.81 SD = 0.08 g/cm$^2$ in femoral neck).$^{13}$ The low R$^2$ values we found using multivariate regression (0.145 for lumbar spine and 0.315 for femoral neck) indicate that there is a lot of variation in BMD that cannot be explained by the variables we used for our analyses. Therefore, one or more as yet undetermined (e.g. genetic) factors exert a strong influence on BMD in Cushing patients.

Osteoporosis is a well-known side effect of treatment with glucocorticoids. For patients in whom prednisone treatment is started with an intended treatment duration of at least three months at a dose of ≥7.5 mg per day, it is advised to add bisphosphonates.$^{23}$ Although results in one nonrandomised study on alendronate therapy in patients treated for Cushing’s disease are promising,$^{12}$ at present there is no consensus that patients with endogenous Cushing’s syndrome should start on bisphosphonates as soon as hypercortisolism has been demonstrated. One of the reasons for not giving bisphosphonates is the fact that BMD after cure of Cushing’s syndrome recovers spontaneously.$^{4,11,13}$ However, normalisation of BMD in Cushing’s syndrome may last approximately ten years and it is not certain whether complete recovery occurs in every patient. At the time of diagnosis, 82% of our patients had a T score lower than −1 SD, and 31% had a T score of −2.5 SD or lower, consistent with a previous report.$^{17}$

![Figure 1: Influence of age on Z score of the lumbar spine (L1-L4; A and C) and the femoral neck (B and D) at the time of diagnosis of Cushing’s syndrome in both sexes](image-url)
Other authors found even higher prevalences. As a T score lower than –1 SD is associated with a 1.5- to 2-fold increase of the risk of fractures and a T score of –2.5 SD or lower is associated with a 3.5- to 4.5-fold increase of fracture risk in eucorticoid postmenopausal persons. Cushing patients have a considerable fracture risk. Moreover, at comparable levels of BMD, patients treated with glucocorticoids have a higher risk of fracture than nonusers, probably reflecting glucocorticoid-induced deterioration of bone quality. Indeed, the prevalence of osteoporotic fractures in patients with endogenous Cushing’s syndrome (30 to 76%) is higher than expected on the basis of BMD alone, particularly at the vertebral level.

In summary, 82% of patients with untreated Cushing’s syndrome had a T score lower than –1 SD, including the 31% of patients who had a T score of –2.5 SD or lower. Surprisingly, we found that the prevalence of osteopenia and osteoporosis at the time of diagnosis of Cushing’s syndrome was independent of age. As recovery of bone loss after treatment of Cushing’s syndrome may last many years, treatment with bisphosphonates should be considered in patients with Cushing’s syndrome who have a decreased BMD, independent of age. A randomised trial with sufficient power and duration of follow-up is needed to definitively prove that treatment with bisphosphonates accelerates the recovery of bone loss after successful treatment of Cushing’s syndrome and that treatment with bisphosphonates prevents fractures in patients with this disorder.

The data in this article were presented as a poster at Annual Meeting of the Endocrine Society 2006, in Boston, USA.

REFERENCES

Coma with ECG abnormalities: consider tricyclic antidepressant intoxication

J. Veris-van Dieren', L. Valk', I. van Geijlswijk², D. Tjan¹, A. van Zanten*¹

Departments of ¹Intensive Care, ²Clinical Pharmacology, Gelderse Vallei Hospital, PO Box 9025, 6710 HN Ede, the Netherlands, *corresponding author: tel.: +31 (0)318-434 115, fax: +31 (0)318-434 116, e-mail: zantena@zgv.nl.

A B S T R A C T

Two comatose patients with a psychiatric history were admitted to our hospital. On admission both presented with major cardiovascular instability and needed urgent intensive care treatment. Although initially not suspected, the coma was caused by tricyclic antidepressant intoxication (TCA). Serious neurological, anticholinergic and cardiovascular effects may occur in TCA intoxication. In any comatose patient with ECG changes and a psychiatric history, TCA intoxication should be strongly suspected.

K E Y W O R D S

Tricyclic antidepressant poisoning, intoxication, coma, ECG abnormalities, cardiogenic shock

I N T R O D U C T I O N

In the differential diagnosis of comatose patients with a history of depression and/or suicide attempts intoxications should be considered. Tricyclic antidepressants (TCA), alone or in combination with other drugs, are often used in intentional drug overdosing. Serious neurological (coma, convulsions), anticholinergic and cardiovascular (hypotension and ventricular arrhythmias) effects may occur, and therefore patients are frequently admitted to intensive care units (ICUs).¹,² However, at initial presentation the aetiology of coma may not be immediately clear, and many other causes can be considered. The following two cases illustrate both the severity of toxic effects and the complexity in diagnosing coma due to TCA poisoning in the clinical setting. ECG abnormalities may provide clues for the final diagnosis.

C A S E 1

A 66-year-old female patient was transported to the emergency department in the early morning. The last two days she had been lethargic, and presented influenza-like symptoms. Overnight she became comatose. Her medical history revealed a manic depression, currently treated by a psychiatrist, auto-intoxication, lithium-induced hypothyroidism, gestational hypertension, and cerebral concussion/contusion. No medications were missing. She had been on nortriptyline (for two months), valproic acid (for five years), propranolol, codeine phosphate and calcium.

On arrival to the emergency department, the patient was comatose. Her coma score was E1M1V1. Pupils were moderately dilated (4 mm) and slow reactions to light were noticed. The patient was hyperventilating. Blood pressure was immeasurable. She had cold, white acra. Rectal temperature was 40.5 °C.

Arterial blood gas analysis showed a pH of 7.34 (7.35 to 7.45), pCO₂ 4.9 (4.5 to 6.0 kPa), pO₂ 8.3 (9.5 to 13 kPa), bicarbonate of 19.2 (22 to 26 mmol/l), base excess –5.5 (–2 to +2), and SO₂ 90% (92 to 99%). Other laboratory results were CRP 11 (0 to 5 mg/l), Hb 8.9 (7.5 to 10.0 mmol/l) and leucocytes 12.5 (4 to 11/µl). The renal tests showed a urea of 20.5 (3.0 to 7.0 mmol/l), creatinine 366 (50 to 90 mmol/l) and were known to have been in the normal range for the last two years. The sodium was 135 (135 to 145 mmol/l) and potassium 5.9 (3.5 to 4.7 mmol/l), with ionised calcium 1.08 (1.15 to 1.29 mmol/l). From the liver tests only the aspartate aminotransferase of 447 (0 to 45 U/l) and alanine aminotransferase of 323 (0 to 45 U/l) were abnormal. Creatine kinase was 6476 (0 to 170 U/l) and troponin-I 0.34 (0 to 0.48 µg/l). Arterial lactate was 2.2 (0.5 to 1.7 mmol/l). Blood glucose level was 20.9 (4.0 to 10.0 mmol/l). Ethanol level was <0.1 g/00 (below detection).
CT scanning of the brain showed no bleeding or cerebral infarction. Cerebrospinal fluid was clear (leucocytes $4 \times 10^3/\text{nl}$, erythrocytes $302 \times 10^3/\text{nl}$, glucose $9.5 \text{ mmol/l}$ and total protein $0.53 \text{ g/l}$).

A 12-lead ECG revealed a ventricular escape rhythm of 40 beats/min and widened QRS complexes, 0.24 sec, with right bundle branch block (RBBB) and left anterior fascicular block (LAFB) configurations.

She was endotracheally intubated and mechanical ventilation was started. Chest X-ray showed a pneumonic infiltrate, possibly due to aspiration of gastric contents. The patient was admitted to the ICU. A pulmonary artery catheter was inserted and a pacemaker lead was introduced for ventricular pacing at 80 beats/min. High-dose inotropes (epinephrine, dobutamine and dopamine) and vasoconstrictors (norepinephrine) were commenced. The patient gradually improved over 36 hours, and was eventually transferred to an internal medicine department. She denied taking overdoses. Nortriptyline levels on the day of admission appeared to be $909 \mu\text{g/l}$ (normal 75 to 250 $\mu\text{g/l}$), a toxic level. The valproic acid level was 15 mg/l (subtherapeutic, normal 50 to 120 mg/l). The renal tests normalised within a few days. The transient renal dysfunction was probably due to anticholinergic effects of nortriptyline and acute tubular necrosis caused by hypoperfusion of the kidneys during the period of hypotension and bradycardia. After six days patient was readmitted to the ICU with massive gastrointestinal blood loss and shock. On endoscopy, a visible vessel in the pylorus was coagulated. Severe anaemia (Hb 2.8 mmol/l) and acute stress caused an acute anterolateral myocardial infarction. The ECG normalised except for abnormal lateral repolarisation due to the myocardial infarction. Paresis of the right arm resulting from compression of the plexus brachialis during her comatose period at home persisted. After two weeks, the patient was discharged from the hospital, and she was further treated by a psychiatrist and a cardiologist in the outpatient setting.

**CASE 2**

A 44-year-old woman had been comatose for a few hours and presented to the emergency department. She had a history of hypertension and autointoxication. She had recently been admitted to a psychiatric hospital and was on weekend leave. During this time she became unconscious with involuntary movements. She had not been ill before. It was unknown if she had taken any medication. She was on olanzapine, oxazepam, paroxetine, temazepam, atenolol, irbesartan and hydrochlorothiazide.

She presented comatose, snoring but moving all her limbs. Her EMV score was E1M4V1. Pupil reactions were normal. She demonstrated no lateralisation and reflexes were symmetrical. No meningeal signs were noted. Blood pressure was 100/50 mmHg, pulse 114 beats/min and temperature 37.5 °C. Clinical examination of the thorax and abdomen were normal. An ECG showed a first-degree AV block and RBBB (figure 2), a PQ interval of 0.24 seconds, with a QRS width of 0.14 seconds. Arterial blood gases were normal. Other laboratory results were glucose 7.8 (4.0 to 10.0 mmol/l) and normal liver and kidney functions.
She was admitted to the ICU for observation. Intoxication with benzodiazepines was assumed because her medication list revealed these medications. However, there was no response after the administration of anexate.

In the evening she developed tonic-clonic seizures for which clonazepam 6 mg/24 hours was started. Serum lithium <0.1 (0.4 to 1.2 mmol/l) and ethanol <0.1 were below detection limits. On the second day she still did not wake up. CT brain, cerebrospinal fluid and EEG were normal. The patient developed respiratory failure after i.v. midazolam (used for lumbar puncture) in combination with a pneumonia. She was intubated, ventilated and needed inotropic support. After four days, the patient was extubated and was transferred to the internal medical ward. Nortriptyline levels were 1149 μg/l (therapeutic range 75 to 250, toxic) and the paroxetine level was 167 μg/l (therapeutic range 10 to 75, maximal 250, this level can be attributed to a daily dose of 40 mg, not being a trough level), both taken during the day of admission. Five days later the nortriptyline level was to 352 μg/l. So, she had unexpectedly taken nortriptyline, which had been prescribed earlier, way beyond its therapeutic level.

**Discussion**

Of all the antidepressants, TCAs are the most toxic drugs often used for suicide attempts. Diagnosis of TCA intoxication can be very difficult. The clinical signs, however severe, are nonspecific. The mechanism of toxicity of TCAs is probably related to four pharmacological actions: anticholinergic, vascular α1-antiadrenergic action, adrenergic reuptake inhibition at nerve terminals and fast sodium channel blockade, a quinidine-like effect in the heart. Serious anticholinergic effects include decreased bowel movements resulting in ileus, hyperthermia causing rhabdomyolysis, urinary retention, renal failure, and confusion.

Cardiovascular and neurological effects are most common (table 1). By inhibiting sodium channels, TCAs can delay the propagation of the depolarisation and repolarisation in the myocardium. This can lead to prolongation of the PR, QRS and QT intervals. AV blocks and bundle branch blocks can be seen. Severe ventricular tachycardia and sinus tachycardia may occur. Our two patients both presented with RBBB and one of them also had LAFB and first-degree atrioventricular block, which fully recovered. Hypotension may be the result of a combination of diminished myocardial contractility (inhibition of fast sodium channels), vascular dilatation (blockade of α1-adrenergic neurotransmitters) and norepinephrine (NA) reuptake inhibition (can lead to NA depletion). Case one had a cardiogenic shock caused by the TCA intoxication.

<table>
<thead>
<tr>
<th>Table 1. ECG changes in TCA overdosing</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Primary dysrhythmia</strong></td>
</tr>
<tr>
<td>Sinus tachycardia</td>
</tr>
<tr>
<td>Supraventricular arrhythmias</td>
</tr>
<tr>
<td>Ventricular tachycardia</td>
</tr>
<tr>
<td>Ventricular ectopy</td>
</tr>
<tr>
<td>Asystole</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>
Severe neurological symptoms include drowsiness, ataxia, hypertonia and hyperreflexia with extensor plantar responses. Agitation and delirium are known presentations. Respiratory depression may occur with the coma. Convulsions occur in more than 5% of patients and often in combination with QRS prolongation. In the second case, anexate was administered because we initially suspected benzodiazepine intoxication to be more likely than TCA intoxication. However, when TCA intoxication is suspected, especially in combination with benzodiazepine intoxication, anexate should not be used. If anexate is used in this situation, there is an increased risk that treatment of convulsions is more difficult since TCA and benzodiazepines can act as competitive antagonists. Another possible symptom, although rare, is lung injury. Pulmonary oedema and ARDS-like symptoms can occur, in combination with nonspecific changes on the chest X-ray.\textsuperscript{12,13} The manifestations of acute toxicity can vary greatly between patients. It is hard to predict which patients will have more serious symptoms, such as seizures or arrhythmias. Individual variation in absorption, protein binding and metabolism might give a variation in plasma TCA levels, making plasma levels an unreliable predictor of toxicity.\textsuperscript{4,14} Toxicity has been suggested to be more likely if plasma concentrations are >100 \( \mu \text{g/l} \), but has also been seen at lower levels.\textsuperscript{4} TCAs have a long half-life, due to the high protein binding and large volume of distribution. These drugs are rapidly absorbed in the gastrointestinal tract. After metabolism by hepatic enzymes the metabolites are conjugated and excreted by the kidneys.\textsuperscript{14} Hypoalbuminaemia and acidemia increases the amount of free drug.\textsuperscript{9} Plasma TCA levels are difficult to determine, and are not readily available. The plasma level of TCA (no distinction can be made between the different kinds of TCA) can only be measured within 30 minutes with the fluorescence polarisation immunoassay.\textsuperscript{4} This technique was not available in our hospital. The value of levels of TCA metabolites in red blood cells is under investigation and seems to have a better correlation with QRS duration.\textsuperscript{15} TCA are extensively metabolised by several cytochrome P450 enzymes. Therefore, drug levels can be influenced by comedication with hepatic clearance. Valproic acid is a CYP 2C9 inhibitor; although the main metabolic pathway of nortriptyline is CYP2D6, the interaction between valproic acid and nortriptyline has been previously described and hesitantly attributed to CYP2C9 or CYP2C19.\textsuperscript{16,17} The intoxication is likely due to the fact that the patient had been on valproic acid for five years at low drug levels and nortriptyline was added recently. The patient denied having taken an overdose. This drug interaction may have reduced nortriptyline elimination. The second patient was on paroxetine, which is known to be a potent inhibitor of CYP 2D6. Since she had a high level of paroxetine, it is most likely that this interaction took place, especially since the long half-life of nortriptyline after the initial (due to treatment with activated charcoal) sharp decline of levels (1148 day 0, 437 day 3, 416 day 4, 352 day 5), had a terminal half-life of about 100 hours. An ECG is an essential diagnostic tool in assessing the clinical severity of overdose, because impaired conduction can progress into arrhythmias with cardiovascular collapse. However, it can never be used as the sole method of risk determination.\textsuperscript{18,19} The accuracy of the ECG is influenced by the time of drug ingestion. Directly after intake the ECG is usually still normal, with abnormalities developing after several hours. Repeat ECGs are necessary when TCA intoxication is suspected. A QRS >100 to 120 msec, QTc >500 ms and R/S ratio in aVR >0.7 (R wave >0.3mm), with right-axis deviation, increase the risk of arrhythmias.\textsuperscript{18,19,20} Heart rate analysis and variation can be useful, but is still controversial.\textsuperscript{18} When the QRS is >100 msec, there is a greater risk of seizures.\textsuperscript{18,6,10,14} Both our patients had a QRS >100 msec, but only one (patient two) demonstrated seizures. It might be that the R/S ratio in aVR may be a better predictor of complications. In some studies the R/S ratio in aVR was a better predictor than the QRS interval for association with arrhythmias or seizures.\textsuperscript{8,12} Niemann and coworkers suggested that the right bundle may be more susceptible to TCA-induced conduction block, which could explain why both patients had RBBB.\textsuperscript{23} The time of resolution of ECG abnormalities varies widely, possibly related to the severity of the toxicity.\textsuperscript{20} Specific treatment to prevent arrhythmias could be justified for patients with QRS >100 msec or R/S ratio >0.7. Patients with hypoxia, acidosis, seizures or other significant noncardiac complications should receive specific treatment. The moment a TCA overdose is suspected, rapid gastrointestinal decontamination should be initiated to prevent further absorption of TCA. Since TCA slows down gastrointestinal movement, activated charcoal is still useable several hours after ingestion. In case of nortriptyline intoxication associated with severe acidosis, sodium bicarbonate administration may be considered to diminish direct cardiac toxicity.\textsuperscript{24} However, it is likely that not the correction of the acidosis, but the administration of sodium causes this beneficial effect. Experimental data suggest that hypertonic saline is even more effective than sodium bicarbonate.\textsuperscript{25} Although administration of sodium bicarbonate may have immediate effects on cardiac toxicity in critical situations it can decrease renal excretion. In fact acidification of urine promotes elimination.\textsuperscript{26} Bicarbonate administration and hyperventilation have been associated with (near) fatal
alkalosis if not properly monitored. Therefore we advise that sodium bicarbonate is used with caution in TCA intoxication and suggest considering hypertonic saline as an alternative.

CONCLUSIONS

We have presented two comatose patients with a TCA intoxication. On admission to the ICU, a TCA intoxication was not immediately diagnosed or even considered. ECG clues could have led to earlier diagnosis and specific toxicological analysis. So, in any comatose patient with ECG changes and a psychiatric history, TCA intoxication should be strongly suspected.

REFERENCES

SUCCESSFUL MANAGEMENT OF CHRONIC MYELOID LEUKAEMIA WITH LEUCAPHERESIS DURING A TWIN PREGNANCY

R. KLAASEN1*, P. DE JONG2, P.W. WIJERMANS1

DEPARTMENTS OF 1HAEMATOLOGY, 2OBSTETRICS AND GYNAECOLOGY, HAGA HOSPITAL, LEYWEG 275, 2545 CH THE HAGUE, THE NETHERLANDS, *CORRESPONDING AUTHOR: TEL.: +31 (0) 70-359 25 56, FAX: +31 (0) 70-359 22 09, E-MAIL: ruthklaasen@casema.nl.

ABSTRACT

We present a 36-year-old woman in whom chronic myeloid leukaemia (CML) was diagnosed during a twin pregnancy. Because of hyperleucocytosis, we started leucapheresis also with the goal of gaining time for discussing treatment options. As this strategy was so effective and our patient was reluctant to take medication, we decided to continue this treatment.

KEYWORDS

CML, pregnancy, leucapheresis

INTRODUCTION

Fortunately, chronic myeloid leukaemia (CML) rarely occurs in pregnant women. Pregnancy does not appear to affect the natural course of CML. Various treatment modalities have been used in the management of this disease during pregnancy. This case report describes the successful use of leucapheresis in a patient with CML during a twin pregnancy. In the discussion we describe possible treatment strategies and explain why we choose this treatment in our patient.

CASE REPORT

A 36-year-old gravida 3 para 2 was referred to our clinic because a raised white blood cell count was noted after an amniotic fluid puncture. At that moment she was 16 weeks pregnant of dizygote twins. She complained of headache, fatigue and dyspnoea on effort which she had attributed to her pregnancy. Physical examination revealed splenomegaly and a uterus in conformity with a 16-week twin pregnancy.

Her haematological profile showed a white blood cell count of 140 x 10^9/l, haemoglobin of 7.1 mmol/l and platelets of 262 x 10^9/l. The differential count was 54% neutrophils, 10% bands, 4% metamyelocytes, 8% myelocytes, 8% promyelocytes, 7% myeloblasts, 1% eosinophils and 0% basophils, 7% lymphocytes and 1% monocytes. A bone marrow aspiration revealed 5% blasts and hypercellularity. Multiple small one nucleus megakaryocytes were seen. Conventional cytogenetic studies showed the presence of the Philadelphia chromosome. Reverse transcriptase polymerase chain reaction confirmed the presence of a head to tail fusion of the breakpoint cluster region (BCR) on chromosome 22 with the Abelson murine leukaemia virus gene (ABL) located on chromosome 9 as well in blood as in bone marrow obtained by aspiration.

Because of the high white blood cell count, the dyspnoea and unknown influence of a high blood cell count on the blood supply of the placenta we started treatment immediately. To gain time to discuss the different treatment options we started leucapheresis. Our patient was very reluctant to take medication, especially chemotherapy, and leucapheresis was well tolerated and very effective, so we decided to continue this therapy. We discussed the risks of only symptomatic treatment with the patient and her partner. Termination of pregnancy was considered but has never been a real option because our patient was already 16 weeks pregnant. All the symptoms disappeared when the leucocyte count dropped below 100 x 10^9/l. Elo-haes, normally used for a better separation of cells, was not used because the effect on unborn babies is not known. Sodium citrate was used as anticoagulation. After every pheresis fluid replacement was carried out with fresh frozen plasma and physiological saline. Overall, 14
Leucapheresis procedures were needed (figure 1). During the 30th week of gestation the leucocyte count rose to over 100 x 10⁹/l. Simultaneously, the symptoms of our patient reappeared. Again, these symptoms disappeared after restarting leucapheresis. To prevent haemodynamic problems the volume of the pheresis was reduced each time towards the end of the pregnancy and so the frequency of pheresis was increased. At a gestational age of 36 weeks and 5 days a caesarean section was carried out because of breech presentation of both babies. Two girls with a birth weight of 2690 and 2640 g were born. They had normal apgar scores and a normal acid-base equilibrium. One of the babies has a club foot which was contributed to mechanical problems in utero. After delivery we initially planned to start treatment with imatinib and cytarabine according the HOVON protocol. Our patient refused this proposal and was treated with only imatinib. One year later a three log reduction in the BCR-ABL clone was noticed. Both children are developing normally.

**DISCUSSION**

When our patient presented at 16 weeks' gestation, she had significant hyperleucocytosis. Although the leucocyte count was not high enough to meet the criteria for leucostasis, we were concerned because of potential microvascular complications, especially placental blood flow. To gain time for discussing treatment options we started leucapheresis.

There are several treatment options for CML. For a few years now, imatinib has been the treatment of choice. Little is known about possible teratogenic effects in humans. Imatinib is teratogenic in rats, but not in rabbits, and impaired spermatogenesis occurred in rats, dogs and monkeys. Hensley and Ford described three patients in clinical trials who became pregnant and proceeded to term. Two patients delivered normal infants. One baby had a hypospadias. Anecdotal reports may suffer from reporting bias. The chemotherapeutic agents hydroxyurea and cytarabine have both been used in pregnant women with different haematological malignancies. Theoretically, these drugs may be hazardous for foetal development. However, there are several reports that using chemotherapy from the second trimester does not result in malformations. On the other hand, a few studies report fine congenital abnormalities of the joints when using cytarabine during pregnancy.

Alpha-interferon appeared to be a safer alternative as it has no known effects on DNA and, being a large molecule, is assumed not to cross the placenta barrier to any great extent. Very low levels of IFN-α in newborns compared with maternal levels confirmed this hypothesis. Alpha-interferon has not been associated with teratogenicity in either animal studies or in humans receiving the drug during pregnancy. One newborn of a mother treated with α-interferon during pregnancy was born with severe thrombocytopenia probably due to α-interferon. Therefore, α-interferon with or without low-dose cytarabine was one of the treatment options and hydroxyurea was a good alternative. Although leucapheresis has the advantage that exposure to potentially teratogenic agents can be avoided, the overall time to progression is not prolonged. Especially for our patient with a high Sokal index of 1.4, this could be a disadvantage. Haemodynamic instability is one of the complications of leucapheresis and it is imaginable that in pregnant patients this risk is even greater. In PubMed we found eight case reports of pregnant women treated with leucapheresis for different haematological malignancies and none of them mention problems of haemodynamic instability. All these mothers delivered healthy babies.
Because our patient was very reluctant to take medication during her pregnancy and leucapheresis was very effective, we decided to continue this strategy. Two healthy babies were born and the mother showed a three log reduction in the BCR-ABL clone one year following imatinib treatment, which has a very good prognosis. This is the second report of a successful outcome of leucapheresis during a twin pregnancy with CML.

REFERENCES


Klaasen, et al. Successful management of CML with leucapheresis during a twin pregnancy.

APRIL 2007, VOL. 65, NO. 4

149
A 69-year-old woman developed Cushing’s syndrome after long-term use of Sinatren®, an Indonesian over-the-counter drug, which was pharmacologically analysed three times before the correct content was discovered. After discontinuation she developed adrenal insufficiency, for which she needed substitution of steroids. Physicians should be aware of the presence of corticosteroids in over-the-counter products, that are not mentioned on the instruction leaflet.

**KEYWORDS**

Cushing’s syndrome, herbal drugs

**INTRODUCTION**

In the literature several reports have been published about the systemic effects of corticosteroid therapy applied by intra-articular injections, enemas, topical therapy, and inhalation. We present a case of a woman who developed a clear Cushing’s syndrome after long-term use of an Indonesian mixture, Sinatren®.

**CASE REPORT**

A 69-year-old woman developed Cushing’s syndrome after long-term use of Sinatren®, an Indonesian over-the-counter drug, which was pharmacologically analysed three times before the correct content was discovered. After discontinuation she developed adrenal insufficiency, for which she needed substitution of steroids. Physicians should be aware of the presence of corticosteroids in over-the-counter products, that are not mentioned on the instruction leaflet.

**KEYWORDS**

Cushing’s syndrome, herbal drugs

**INTRODUCTION**

In the literature several reports have been published about the systemic effects of corticosteroid therapy applied by intra-articular injections, enemas, topical therapy, and inhalation. We present a case of a woman who developed a clear Cushing’s syndrome after long-term use of an Indonesian mixture, Sinatren®.

**CASE REPORT**

A 68-year-old woman was referred to our hospital because of progressive fatigue and dizziness for a few years. Her medical history revealed eczema of the face and extremities, hay fever, nonspecific chest pain for which she had a cardiac evaluation in 1988, and cerebral concussion after a collapse of unknown origin in 1999. On admission, she was on budesonide nasal spray, 200 mg per day, fexofenadine 120 mg once daily and diltiazem 180 mg once daily. Previously, she had used a steroid-containing ointment as a local therapy for her eczema, but this had been discontinued for more than six months. Her complaints consisted mainly of tiredness and she had developed full, reddish cheeks and neck region. She denied an increase in weight, but had noticed spontaneous bruising on her extremities and her skin had become thinner in the past few years. She had no chest pain or dyspnoea, but she did report emotional lability and easy agitation.

On physical examination clear signs of Cushing’s syndrome were present. There was a centripetal obesity, especially in the face (so-called ‘moon face’) with plethora over the cheeks, in the neck, trunk and abdomen, with the extremities relatively spared and wasted. Enlarged fat pads filling the supraclavicular fossae were noted. The blood pressure was 115/70 mmHg, weight 57 kg and height 165 cm. There were no ecchymoses present, nor oedema.

Laboratory results were as follows (normal values in parentheses): haemoglobin 8.0 mmol/l (7.4 to 9.9), white cell count 7.6 x 10⁹/l (3.5 to 11.0) with 67% neutrophils (50 to 70%) and 23% lymphocytes (25 to 40%), glucose 5.5 mmol/l (3.5 to 6), creatinine 59 µmol/l (-105), sodium 139 mmol/l (136 to 146), potassium 3.1 mmol/l (3.5 to 4.5), thyroid-stimulating hormone 0.5 µIU/ml (0.1 to 5), Ca²⁺ 1.28 mmol/l (1.13 to 1.30), alkaline phosphatase 78 IU/l (-100), lactate dehydrogenase 89 IU/l (-110), albumin 38 g/l (37-58), cortisol <0.02 µmol/l (0.25 to 0.76), ACTH 5 ng/l (10 to 20).

Budesonide nasal spray was discontinued. She was only taking fexofenadine and diltiazem as before, but no other medications or products. Cortisol levels were detectable within two weeks: 0.39 µmol/l (9.00 am), 0.28 µmol/l (04.00 pm); high-dose synacthen test (250 µg ACTH): t=0 cortisol 0.51 µmol/l, t=30 min cortisol 0.77 µmol/l.
At that time she felt better and showed no signs of adrenal insufficiency.

After three months she reported reappearance of her symptoms. She was no longer using nasal sprays. However, she mentioned that she had started using Sinatren®, an Indonesian product which was claimed to contain only herbal constituents (figures 1 and 2). She purchased it at the local market in Indonesia for treatment of her eczema. She had used it on and off for years, but she did not know exactly what the product contained. Because we had previously discussed the possibility that the symptoms and biochemical results could be due to exogenous corticosteroids, she had approached her pharmacist for analysis of these Sinatren® sachets. UV spectrophotometrical and thin layer chromatographic analyses repeatedly reported acetaminophen being the only pharmacological constituent. Being satisfied with these results, she again started taking Sinatren® on a daily basis. At that time, the cortisol levels were again not detectable (cortisol <0.02 μmol/l), the result of ACTH being 3 ng/l. It was then assumed that the Sinatren® sachets had to be the source of the steroids, even though there was no direct evidence at that time. Sinatren® was stopped immediately. The sachets were therefore reanalysed in our own pharmacy.

After two weeks she developed signs of a relative adrenal insufficiency with flu-like muscle pain, nausea with a weight loss of 5 kg, and stiff hands. She also complained of acute low back pain. Her blood pressure was 95/70 mmHg. There were no signs of oedema. She was admitted to the hospital for clinical testing and analysis of her low back pain. Cortisol and ACTH values were 0.33 μmol/l and 14 ng/l respectively (9.00 am). Other laboratory values were as follows: creatinine 52 μmol/l, sodium 134, and potassium 3.6. Repeatedly measured cortisol values between 8.00 and 9.00 am were low (cortisol 0.22 and 0.29 μmol/l). In combination with the clinical presentation we assumed that she had signs of a relative adrenal insufficiency and therefore a synacthen test was not performed. Substitution therapy with hydrocortisone was started (10-5 mg). An X-ray of the lumbar spine was taken, revealing loss of height of several corpora (Th11, Th12, L1, and L2) and secondary thoracolumbar kyphosis. Bone densitometry showed low values of the bone mineral density for the lumbar region (T score –3.8, Z score –1.7) and both colla (T score –3.3, Z score –1.6) indicating osteoporosis. After a few days of bed rest and medication for the low back pain, she was able to resume daily activities again. She was also being treated with bisphosphonates and calcium supplements. At that time, the results of the Sinatren® analysis by our hospital pharmacy department became known. High-performance liquid chromatography (HPLC) revealed the presence of dexamethasone and acetaminophen (figure 3).
DISCUSSION

This case illustrates how difficult it can be to prove the presence of corticosteroids in an innocent appearing drug. In this case the patient was not aware of the content of an Indonesian mixture, Sinatren, presented and sold as a herbal drug, which led to Cushing’s syndrome with suppression of the hypothalamic-pituitary axis. It took a long time to prove the presence of corticosteroids in this product. Three analyses were needed before the evidence was found. The first examination was not conclusive. The Research Institute of Dutch Pharmacists (WINAP) performed the second examination. They analysed two different sachets of Sinatren (LNA registration numbers: C1037, yellow sachet; C1038, brown sachet). After UV spectrophotometrical and thin layer chromatographic examinations they excluded the presence of steroids in the sachets. The brown sachets contained acetylsalicylic acid in a concentration of 200 mg/g.

Only after the third analysis with high-performance liquid chromatography (HPLC) in our own hospital was it shown that dexamethasone is present in Sinatren*: the yellow sachet contained approximately 0.6 mg dexamethasone per gram powder and 1 mg acetylsalicylic acid per gram powder, the brown sachet contained approximately 3.1 mg ibuprofen per gram powder and approximately 37 mg acetylsalicylic acid per gram powder. This again underlines the importance of the choice of a suitable method for analysing pharmacologically active products. Only HPLC proved to have a detection limit low enough to recognise the presence of a pharmacologically significant amount of dexamethasone.

Both UV analysis and thin layer chromatography had detection limits far above the pharmacologically relevant concentration and could not measure the lower quantity of dexamethasone present in this case. Interestingly, ‘sinatren’ is the Indonesian word for express train (phonetically it sounds in Dutch like ‘sneltrein’) suggesting a rapid action.

Some reports have been published about adrenal insufficiency after treatment with intra-articular steroid injection,2,4 enemas,2,4 and topical steroid therapy1 and after inhaled corticosteroids.5,7 However, literature about adrenal insufficiency after ingestion of herbal or alternative mixtures is very scarce. Only three reports have passed on this matter, one of them in the Chinese language.8-10 An excellent review on herbal remedies has been published11 and is very much worth reading.

This case underscores the importance of taking a medical history carefully, with special interest to the use of regular and alternative (herbal) drugs. Physicians should be aware of potential, previously described or yet unknown, hazards related to these products. Even when products appear to be completely harmless, a thorough analysis with a method capable of producing clinically relevant results is warranted if no other source of exogenous steroids can be detected.

REFERENCES


APRIL 2007, VOL 65, NO. 4

152
Sine waves

A.A. Alkhatib, S.E. McGuire

Department of Internal Medicine, the University of Texas Health Science Center, 6431 Fannin, Ste 1.134, Houston, TX 77030, *corresponding authors: tel.: +1 832-659-7837, fax: +1 713-500-6530, e-mail: amer.a.alkhatib@uth.tmc.edu; tel.: +1 713-256-3881, fax: +1 713-500-6530, e-mail: seanmcguire@sbcglobal.net

A 42-year-old African-American man, with AIDS (CD4 count 374), Hepatitis B virus infection and end-stage renal disease on regular dialysis three times per week, presented to the emergency centre with profound weakness for the past 24 hours. Upon review, the patient admitted to having missed his last two dialysis appointments. An ECG was obtained and the result is presented in figure 1. The patient was treated with intravenous calcium chloride, insulin, dextrose and bicarbonate. His serum potassium level was 8.9 mEq/l. The patient was admitted to the Medical ICU and urgently dialysed. His serum potassium corrected to 4.3. A postdialysis ECG is shown in figure 2.

WHAT IS YOUR DIAGNOSIS?

See page 155 for the answer to this photo quiz.

Figure 1. Wide-complex tachycardia with sine waves

Figure 2. Post-dialysis ECG
Extensive jejunal diverticulosis in a family, a matter of inheritance?

ABSTRACT

Diverticulosis of the jejunum is a rare finding (0.06 to 1.3%). Possible complications are bacterial overgrowth, malabsorption, bleeding, mechanical obstruction, volvulus and perforation. At present only one case report on familial jejunal diverticulosis has been published. We describe three patients with jejunal diverticulosis within one family, which might suggest inheritance.

CASE REPORT

The family consists of a mother and four children; no information is available about the father (figure 1).

Patient A: the mother died at the age of 83 due to infection and rapid dehydration. She had documented colonic cancer for which she was operated at the age of 50. She suffered from celiac sprue and chronic anaemia. Autopsy revealed extensive jejunal diverticulosis.

A son had a myocardial infarction at the age of 50 and colonic cancer was diagnosed at the age of 60. He shows no symptoms related to jejunal diverticulosis. Another son died of a myocardial infarction, no other data are available.

Patient B: a son is currently being treated intermittently for bacterial overgrowth due to jejunal diverticulosis diagnosed with enteroclysis.

Patient C: a daughter came to our hospital with complaints of cramp-like abdominal pain, nausea, vomiting, flatulence and weight loss. She had lost over 25 kg in two years time, and her BMI had dropped to 18 kg/m². Gastroduodenoscopy showed no abnormalities, biopsies excluded coeliac sprue. H₂-breathing tests were positive for lactose and glucose, suggesting bacterial overgrowth. Enteroclysis demonstrated jejunal diverticula (figure 2). She was treated with doxycycline for bacterial overgrowth due to jejunal diverticulosis. This alleviated most of her symptoms and she gained 13 kg in weight. She kept having episodes of abdominal pain and vomiting. She underwent a diagnostic laparoscopy with resection of a diverticulum, which was probably the cause of these transient symptoms related to ileus. Laparoscopy revealed jejunal diverticula involving about half of the small bowel.

Figure 1. Pedigree chart compatible with an autosomal dominant trait

Figure 2. Enteroclysis of patient C demonstrating jejunal diverticulosis
One of the diverticula was stapled and histology showed pneumatosis intestinalis probably due to her severe form of bacterial overgrowth or as a result of her motility disorder. She was treated over a long period of time with a lactose-free diet, cobalamin injections every three months and doxycycline. A surgical treatment is not feasible as it would involve a small bowel resection extending over 50% of the total small bowel length.

As was calculated in the article by Sternberg et al., simple calculation shows a chance of $1.28 \times 10^{-5}$ that three people in a family of five have jejunal diverticulosis coincidentally:

$\frac{3! \cdot (1-p)^2 \cdot (4!/2! \cdot 2!)}{p}$; $p=0.013$ (the highest prevalence mentioned)

Although it is statistically only partially correct to use prevalence as a $P$ value, it is a very strong indication that coincidence is highly unlikely.

**CONCLUSION**

We present a family with jejunal diverticulosis in the absence of apparent connective tissue disease. The inheritance pattern is compatible with an autosomal dominant inheritance trait, which clearly infers a genetic cause. As such it is the second family to be described.

A.D. Koch, E.J. Schoon
Catharina Hospital, Department of Gastroenterology, Michelangelolaan 2, 5623 EJ Eindhoven, the Netherlands

**REFERENCES**


**ANSWER TO PHOTO QUIZ (ON PAGE 153)**

**SINE WAVES**

ECG findings with hyperkalaemia are well described in the literature. When the potassium level is between 5.5 and 6.5 mEq/l, large-amplitude T waves may be seen. PR prolongation, P-wave flattening or disappearance, QRS-complex widening, and conduction blocks with escape beats are typical findings, which may be associated with a potassium level of 6.5 to 8.0 mEq/l. Sine-wave appearance is an ominous ECG finding which precedes ventricular fibrillation. Usually sine waves correlate with a potassium level >8.0 mEq/l. It should be emphasised that hyperkalaemia may present with minimal ECG changes.

**REFERENCE**

Parnassus

Bart Elfrink

Bart Elfrink (1954) lives and works in Nijmegen. This woodcut, specially made for this last cover provided by Galerie Unita, is based on a series of pictures and drawings of Parnassus mountain. Bart Elfrink took these pictures during one of his many trips to Greece. Every year he travels to Greece to work and teach in his house and studio in the Greek province of Epirus. Besides portraits, the landscape, especially the Greek landscape, has been his main inspiration for his work as painter for many years. Sometimes, he also creates graphic work like this woodcut.

Bart Elfrink exposes his work in the Netherlands and abroad. As a member of a renowned family of painters, his work was exhibited in Museum Valkhof in Nijmegen in 2006.

More prints by Bart Elfrink can be seen at www.bartelfrink.exto.nl

An original print is available at a price of € 200 and can be ordered from Galerie Unita, Rijksstraatweg 109, 6573 CK Beek-Ubbergen, the Netherlands or by mail: galerie-unita@planet.nl or www.galerie-unita.com.

© 2007 Van Zuiden Communications B.V. All rights reserved.
The table lists online hits for all articles that were published in the January issue of the Netherlands Journal of Medicine. This is based on analysis of our user log file on 8 March 2007 (www.njmonline.nl)

<table>
<thead>
<tr>
<th>Article</th>
<th>Online hits</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>EDITORIALS</strong></td>
<td></td>
</tr>
<tr>
<td>Announcements from the Editorial Board of the Netherlands Journal of Medicine</td>
<td>139</td>
</tr>
<tr>
<td>The ups and downs of sirolimus in kidney transplantation, and the importance of reporting negative findings</td>
<td>136</td>
</tr>
<tr>
<td><strong>REVIEWS</strong></td>
<td></td>
</tr>
<tr>
<td>Evidence-based guideline on management of colorectal liver metastases in the Netherlands</td>
<td>166</td>
</tr>
<tr>
<td>Clinical indicators: development and applications</td>
<td>454</td>
</tr>
<tr>
<td><strong>ORIGINAL ARTICLES</strong></td>
<td></td>
</tr>
<tr>
<td>Inferior results with basic immunosuppression with sirolimus in kidney transplantation</td>
<td>128</td>
</tr>
<tr>
<td>Binge drinking causes endothelial dysfunction, which is not prevented by wine polyphenols: a small trial in healthy volunteers</td>
<td>137</td>
</tr>
<tr>
<td><strong>CASE REPORTS</strong></td>
<td></td>
</tr>
<tr>
<td>Cisplatin-induced hyperglycaemic hyperosmolar coma</td>
<td>125</td>
</tr>
<tr>
<td>Therapeutic hypothermia after prolonged cardiopulmonary resuscitation for pulseless electrical activity</td>
<td>141</td>
</tr>
<tr>
<td><strong>PHOTO QUIZ</strong></td>
<td></td>
</tr>
<tr>
<td>An unusual cause of hypertrichosis</td>
<td>159</td>
</tr>
<tr>
<td><strong>LETTER TO THE EDITOR</strong></td>
<td></td>
</tr>
<tr>
<td>Mesothelioma: a case report</td>
<td>123</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>1708</td>
</tr>
</tbody>
</table>
**Aims and scope**
The Netherlands Journal of Medicine publishes papers in all relevant fields of internal medicine. In addition to reports of original clinical and experimental studies, reviews on topics of interest or importance, case reports, book reviews and letters to the editor are welcomed.

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

**Language**
The language of the Journal is English. English idiom and spelling is used in accordance with the Oxford dictionary. Thus: Centre and not Center, Tumour and not Tumor, Haematology and not Hematology.

**Submission**
All submissions to the Netherlands Journal of Medicine should be submitted online through Manuscript Central at http://mc.manuscriptcentral.com/nethjmed. Authors should create an account and follow the instructions. If you are unable to submit through Manuscript Central contact the editorial office at g.derksen@aig.umcn.nl, tel.: +31 (0)24-361 04 59 or fax: +31 (0) 24-354 17 34.

**Preparation of manuscripts**
Type all pages with double spacing and wide margins on one side of the paper. To facilitate the reviewing process, number the lines in the margin and the pages. Subheadings should not exceed 55 characters, including spaces. Abbreviations: Measurements should be abbreviated according to SI units. All other abbreviations or acronyms should be defined on the first appearance in the text. Use a capital letter for generic names of substances and materials. A Covering letter should accompany the manuscript, identifying the corresponding person (with the address, telephone number, fax number and e-mail address). Conflicts of interest, commercial affiliations, consultations, stock or equity interests should be specified. In the letter one to three sentences should be dedicated to what this study adds. The letter should make it clear that the final manuscript has been seen and approved by all authors. All authors should sign the letter. The letter should either be submitted through http://mc.manuscriptcentral.com/nethjmed or faxed to the editorial office (+31 (0)24-354 17 34).

Divide the manuscript into the following sections: Title page, Abstract, Keywords, Introduction, Materials and methods, Results, Discussion, Acknowledgements, References, Tables and Figures with Legends.

The Title page should include authors’ names, degrees, academic addresses, correspondence address, including telephone number, fax number, e-mail address and grant support. Also the contribution of each author should be specified.

The title should be informative and not exceed 90 characters, including spaces. Avoid use of extraneous words such as ‘study’, ‘investigation’ as well as priority claims (new, novel, first). Give a running title of less than 50 characters. If data from the manuscript have been presented at a meeting, list the name, date and location of the meeting and reference and previously published abstracts in the bibliography. Give a word count (including references, excluding tables and legends) at the bottom of this page.

The Abstract, not exceeding 250 words, should be written in a structured manner and with particular care. In original articles, the Abstract should consist of the following paragraphs: Background, Methods, Results and Conclusion. They should briefly describe the problem being addressed in the study, how the study was performed and which measurements were carried out, the most relevant results, and what the authors conclude from the results.

**Keywords:** Include three to five keywords.

The Introduction should be brief and set out the purposes for which the study has been performed.

The Materials and methods should be sufficiently detailed so that readers and reviewers can understand precisely what has been done without studying the references directly. The description may be abbreviated when well-accepted techniques are used.

The Results should be presented precisely, without discussion.

The Discussion should directly relate to the study being reported. Do not include a general review of the topic, but discuss the pertinent literature.

**Acknowledgement:** All funding sources should be credited here. Also a statement of conflicts of interest should be mentioned.
References should be numbered consecutively as they appear in the text (after the punctuation and in square brackets). Type the reference list with double spacing on a separate page. References should be in the language they are published in, conform the ‘Vancouver’ style for biomedical journals (N Engl J Med 1991;324:424-8).

Journal abbreviations should conform to the style used in the Cumulated Index Medicus. Examples:


Please note that all authors should be listed when six or less; when seven or more, list only the first three and add et al. Do not include references to personal communications, unpublished data or manuscripts either ‘in preparation’ or ‘submitted for publication’. If essential, such material may be incorporated into the appropriate place in the text. Recheck references in the text against the reference list after your manuscript has been revised.

The use of bibliographic software programmes that are designed to generate reference lists such as Reference Manager© or Endnote© is highly encouraged. Authors can use the predefined output ‘Vancouver’ style from these programmes.

Tables should be typed with double spacing each on a separate page, numbered consecutively with Arabic numerals, and should contain only horizontal lines. Provide a short descriptive heading above each table with footnotes and/or explanation underneath.

Figures must be suitable for high-quality reproduction (>300 DPI). Submit line drawings made in Word or other computer programmes but not in a PowerPoint file. Colour figures are occasionally possible and will be charged to the authors. Legends for figures should be typed, with double spacing, on a separate page.

Case reports
Case reports containing concise reports on original work will be considered for publication. Case reports which are relevant for understanding the pathophysiology or clinical presentation of disease may also be accepted under this heading. Selection of case reports will be based on criteria as outlined in a special report by the editors (Drenth et al. The case for case reports in the Netherlands Journal of Medicine. Neth J Med 2006;64(7):262-4). We advise potential authors to take notice of the instructions in this report. Articles published in this section should be no longer than 1000 words, and supplied with a summary of about 60 words, preferably no more than two figures and/or tables, and no more than 15 references.

Mini reviews
Mini reviews are concise notes that bring the reader up to date with the recent developments in the field under discussion. The review article should mention any previous important reviews in the field and contain a comprehensive discussion starting with the general background of the field. It should then go on to discuss the salient features of recent developments. The authors should avoid presenting material which has already been published in a previous review. The manuscript should be divided as follows: title page, abstract and main text. The text may be subdivided further according to the areas to be discussed. The text should not exceed 2500 words.

Letters to the editor (correspondence)
Letters to the editor will be considered by the editorial board. Letters should be no more than 400 words. Please use SI units for measurements and provide the references conform the Vancouver style (N Engl J Med 1991;324:424-8). No more than one figure is allowed. For letters referring to articles previously published in the Journal, the referred article should be quoted in the list of references.

Photo quiz
A photo quiz should not exceed 500 words and include no more than two figures and four references conform the Vancouver style. Abbreviations of measurements should be quoted in SI units.

Book reviews
The editorial board will consider articles reviewing books.

Reviewing process
After external and editorial review of the manuscript the authors will be informed about acceptance, rejection or revision. We require revision as stated in our letter.

Proofs
Proofs will be sent to the authors to be carefully checked for printer’s errors. Changes or additions to the edited manuscript cannot be allowed at this stage. Corrected proofs should be returned to the editorial office within two days of receipt.

Offprints
These are not available. The first author receives a sample copy of the Journal with the published article.