platelet counts of less than 20×10^9/L and bleeding problems. They have confirmed that, in the group studied, response is more rapid with IVIG than with methylprednisolone and more durable if initial treatment is followed by oral steroids. They also confirmed that neither approach affected the natural history of the disease. However, since the study group consisted of newly diagnosed patients, it is hard to tease out spontaneous remissions from responses to therapy. In addition, the extent of bleeding was variable and did not include those with the severest haemorrhage, the group for whom treatment is most needed.

Like other studies Goudeau and colleagues measured response in terms of increment in platelet counts. There is no clear evidence that, except for patients with life-threatening haemorrhage, differences in a day or two to reach haemostatically safe levels are clinically important. Nevertheless, in patients with severe or life-threatening bleeding, the immediate aim is as rapid an increase in platelet count as possible, and, overall, trials have shown that multimodal therapy—ie, generally with a combination of steroids in some form, and intravenous immunoglobulin—is appropriate. Until there are trials that clearly show the clinical benefit of one treatment over another, the choice will often depend on personal bias.

To demonstrate clinical benefit what is still required are trials in which assessment is based on clinical endpoints. The difficulty is that such studies will require large numbers of patients. Meanwhile, for all other patients without severe life-threatening bleeding, there should be an initial period of judicious observation, since treatment may often be unnecessary and may increase the risk of infection. Without treatment most such patients will have a normal or near-normal quality of life.[9] There is the prospect of specific immune-based therapies,[10,11] but these new treatments are also likely to be associated with an increased incidence of adverse effects, particularly infection. Hence treatment for AITP should always be restricted to those with clinical need.

Adrian C Newland

Department of Haematology, Barts and the London School of Medicine and Dentistry, London E1 1BB, UK
(e-mail: a.c.newland@mds.qmw.ac.uk)

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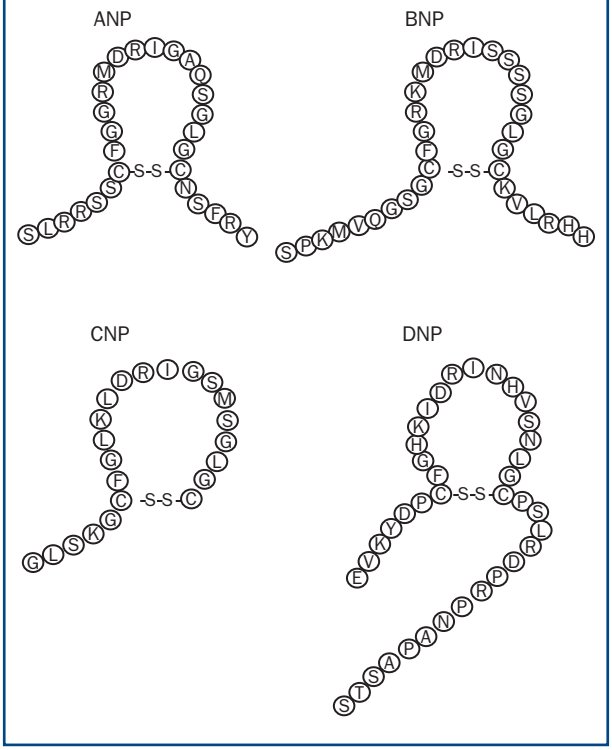
Dendroaspis natriuretic peptide: endogenous or dubious?

Dendroaspis natriuretic peptide (DNP), a 38-aminoacid peptide, was isolated from the venom of the Green Mamba (*Dendroaspis angusticeps*) by Schweitz and colleagues in 1992.[1] It has structural similarities to the two known human cardiac natriuretic peptides, in particular in sharing the characteristic 17-aminoacid disulphide ring (figure), with both and having 12 and 14 aminoacid residues in common with human atrial natriuretic peptide (ANP) and brain natriuretic peptide (BNP), respectively. DNP is as potent as rat ANP in relaxing pre-constricted rat aortic strips, and prior relaxation by ANP excludes further relaxant effects from DNP (and vice versa). In common with ANP and BNP, DNP induces formation of the second messenger cyclic guanosine monophosphate (cGMP) by cultured bovine aortic endothelial cells and has the same dose-response characteristics as rat ANP. Furthermore, relaxation of an isolated canine coronary-artery preparation by DNP was reduced by HS-142, an inhibitor of particulate guanylate cyclase.[2] Finally, DNP displaces radiolabelled rat ANP from specific binding sites on cultured rat aortic

Aminoacid sequence and structure of human ANP, BNP and CNP, and of dendroaspis natriuretic peptide



myocytes.[1] In summary, DNP has structural features, receptor characteristics, second-messenger activity, and some biological effects (vasorelaxation) in common with ANP and BNP. So, 9 years after its discovery, can DNP be said to be a member of the cardiac natriuretic peptide family in man?

A commercially available polyclonal antibody to dendroaspis peptide (Phoenix Pharmaceuticals, Mountain View, CA) has been used for radioimmunoassay measurements of DNP in plasma and tissue and for immunohistochemistry studies.[3–5] Schirger and colleagues[3] from the Mayo Clinic reported DNP-like immunoreactivity in human plasma, with concentrations in 19 cases of heart failure (mean 37·0 [SEM 15·0] pg/mL) exceeding ($p<0·05$) those in 19 normal controls (6·3 [1·0] pg/mL). These data, together with concentrations now reported by Emilio Fábrega and colleagues[6] in Spain in 20 healthy controls (median 106·5, range 67–246 pg/mL), 12 patients with cirrhosis but no ascites (136·5, 100–250 pg/mL), and 44 with cirrhosis plus ascites (249·0, 101–508 pg/mL), constitute the sum total (ie, 39 healthy people and 75 patients) of published material on plasma concentrations of DNP-like immunoreactivity in man. The threefold discrepancy in plasma immunoreactive DNP concentrations in healthy volunteers between the two groups calls for further investigation into the authenticity of the peptide(s) measured and/or interfering compounds.

The three published immunohistochemistry studies have confined attention to human and canine cardiac atria and canine ventricle.[3–5] DNP-like immunoreactive staining has consistently been observed within atrial myocyte cytoplasm in both peripheral and perinuclear locations. Normal (n=3) and failing human hearts (n=4) did not seem to differ.[3] Immunoreactive staining was more intense in atria from dogs with heart failure than in normal animals, with corresponding differences in tissue peptide concentrations on radioimmunoassay (0·4 [0·1] in normal dogs *vs* 2·9 [0·5] pg/mg of protein in those with experimental heart failure).[5] Differences were also found in ventricular tissue (0·5 [0·2] *vs* 2·1 [0·3] pg/mg protein).[5]

Using the Phoenix antibody, our laboratory has found plasma DNP-like immunoreactivity in normal human and heart failure plasma that is similar to data reported by Schirger and colleagues, and we have also observed positive immunostaining in ovine atria and ventricles (unpublished).

The bioactivity of dendroaspis peptide has been investigated in healthy dogs and in animals with pacing-induced heart failure.[4,5] DNP induced natriuresis and diuresis, decreased cardiac-filling pressure, lowered systemic arterial pressure, suppressed renin secretion, and increased plasma and urine cGMP.

These reports, although tantalising, do not establish DNP as a member of the mammalian cardiac natriuretic peptide family or even as an endogenous peptide in human beings. The presence of DNP-like immunoreactivity in plasma by radioimmunoassay and in cardiac tissue by immunohistochemistry does not constitute proof of endogenous authentic human DNP, since both methods are vulnerable to artifact and lack of specificity because of cross-reaction to unknown proteins. Several criteria (long since satisfied for ANP, BNP, and the C-type natriuretic peptide [CNP]) must be fulfilled. The peptide must be identified in human blood or tissue by high-performance chromatography linked to immunoassay, followed by purification and analysis to establish the human aminoacid sequence. The gene must be identified in the Green Mamba so that appropriate probes for the cloning of the human homologue can be generated. The response of endogenous plasma and tissue concentrations of the peptide to physiological and pathophysiological stimuli must be assessed by use of validated immunoassays based on antibodies raised to authenticated human peptide, with human DNP also acting as the assay standard since, as with BNP, there might be sizeable differences between species in DNP structure. The bioactivity of administered peptide must be tested in the species of origin. None of these tests has been passed. DNP has been isolated only from Green Mamba venom. There are no published reports of successful fractionation of plasma or tissue DNP-like immunoreactivity from human samples by high-performance liquid chromatography as a first step to isolation and sequencing of human peptide. The gene has not been cloned. These gaps in the chain of evidence are the more glaring when compared with the rapid progress achieved for ANP, BNP, and CNP in the 1980s and early 1990s.[7–11] An additional approach might be to go back to *Dendroaspis angusticeps* to elucidate the site of DNP synthesis (is the heart involved?), and identify the gene encoding the peptide or its biological activity. It might be, for example, that DNP is a primitive, ancestral cardiac natriuretic peptide, an evolutionary precursor to ANP and BNP, or even snake BNP itself since the structure of BNP varies considerably between species. As of now, it is unclear whether DNP, or a closely similar peptide, is an endogenous entity in man.

**A Mark Richards, John G Lainchbury, M Gary Nicholls, A Vicky Cameron, Timothy G Yandle*

Christchurch Cardioendocrine Research Group, Christchurch School of Medicine and Health Sciences, PO Box 4345, Christchurch, New Zealand
(e-mail: mark.richards@cdhb.govt.nz)

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