Haematology Research Review™

Making Education Easy

Issue 28 - 2018

In this issue:

Abbreviations used in this issue

AAV = adeno-associated viral
CVT = cerebral vein thrombosis
DOAC = direct oral anticoagulant
DVT = deep vein thrombosis
FIX = factor IX
ICH = intracerebral haemorrhage
LMWH = low-molecular-weight heparin
OR = odds ratio
PE = pulmonary embolism
VTE = venous thromboembolism

Welcome to issue 28 of Haematology Research Review.

This issue begins with research from JAMA studying associations of prior warfarin, DOAC (direct oral anticoagulant) and no oral anticoagulant use with in-hospital mortality in patients with ICH. New research on clotting parameters has identified some that could help to predict DVT recurrence following discontinuation of anticoagulation. An interesting paper reporting promising results of a gene therapy with autologous CD34-positive cells transduced with the BB305 vector for severe β-thalassaemia is also included, as is one reporting a good safety profile with stable increases in FIX activity in haemophilia B after administration of a single infusion of AAV-5 (adeno-associated virus-5) vector combined with a liver-specific promoter driving expression of a codon-optimised wild-type human FIX gene.

As always, we invite you to send us your comments and feedback.

Kind regards,

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Association of intracerebral hemorrhage among patients taking non-vitamin K antagonist vs vitamin K antagonist oral anticoagulants with in-hospital mortality

Authors: Inohara T et al.

Summary: This US registry-based retrospective cohort study included 141,311 patients admitted to hospital with ICH, and assessed the association between oral anticoagulant use <7 days before hospital arrival and in-hospital mortality. Compared with patients without prior oral anticoagulant use, the risk of in-hospital mortality was higher among patients with prior use of warfarin (adjusted OR 1.62 [97.5% CI 1.53, 1.71]) and among those with prior use of DOACs (1.21 [1.11, 1.32]), but prior DOAC use, compared with prior warfarin use, was associated with a lower risk of in-hospital mortality (0.7 [0.69, 0.81]). The difference in mortality between DOAC- and warfarin-treated patients was numerically greater among patients with prior use of dual antiplatelet therapy (32.7% vs. 47.1%; adjusted OR 0.50 [97.5% CI 0.29, 0.86]) than among those taking these agents without prior antiplatelet therapy (26.4% vs. 31.7%; 0.77 [0.70, 0.85]).

Comment (LY): This large cohort of patients with ICH has some interesting take-home messages. Most of the patients were not receiving either anticoagulants or antiplatelet agents. DOAC recipients made up a small fraction of the cohort, mostly using Xa inhibitors. No specific reversal agents would have been widely available, other than in clinical trials, during the study period. These patients had a lower mortality than patients receiving warfarin, who were more numerous; no information is available about the management of the warfarinised patients, specifically use of prothrombin complex concentrates. Interestingly, three-quarters of the patients receiving warfarin had an international normalised ratio of <3. It will be interesting to track the impact, if any, of reversal agents on the outcome of DOAC-related haemorrhage.

Reference: JAMA 2018;319:463–73
Abstract

For more information, please go to http://www.medsafe.govt.nz

Effectiveness and safety of anticoagulants for the treatment of venous thromboembolism in patients with cancer

Authors: Streiff MB et al.

Summary: VTE recurrence and major bleeding rates were reported for patients with newly diagnosed cancer who received treatment for a first VTE with rivaroxaban (n=707), LMWH (n=660) or warfarin (n=1061). Compared with LMWH, rivaroxaban use was associated with a trend for a lower VTE recurrence rate at 6 months (13.2% vs. 17.1% [p=0.060]), which reached statistical significance at 12 months (16.5% vs. 22.2% [p=0.030]). Compared with warfarin, rivaroxaban was associated with significantly lower 6- and 12-month VTE recurrence rates (13.2% vs. 17.5% [p=0.014]) and 15.7% vs. 19.9% [p=0.017], respectively).

Comment (LY): Since warfarin was shown to be associated with a higher rate of cancer-associated VTE recurrence compared with dalteparin (LMWH), an effective oral agent has been something resembling a holy grail to avoid the need for long-term daily injections in this population. This retrospective cohort study of around 2500 cancer-associated VTE patients used modelling to create equivalent cohorts of warfarin, LWMH and rivaroxaban to avoid the obvious confounding created by patient/physician selection of medication. As is typical in cancer-associated VTE, the rates of major bleeding and VTE recurrence were relatively high. However, the results confirm that rivaroxaban appears to be a reasonable choice of therapy in this difficult group. Randomised trials are confirming these real-world cohort results, and in this review, the first randomised trial of rivaroxaban in the cancer population is discussed. As this agent becomes available in NZ in August, we can expect use to increase rapidly.

Reference: Am J Hematol 2018;93:664–71
Abstract

Antiphospholipid antibodies induce thrombosis by PP2A activation via apoER2-Dab2-SHC1 complex formation in endothelium

Authors: Sacharidou A et al.

Summary: Experiments conducted by these researchers showed in endothelial cells that the apoER2 (apolipoprotein E receptor-2) cytoplasmic tail serves as a scaffold for antiphospholipid antibody-induced assembly and activation of the heterotrimeric PP2A (protein phosphatase 2A). The activating L309 methylation of the PP2A catalytic subunit by leucine methyl transferase-1 was shown to be promoted by disabled-2 recruitment to the apoER2 NPXY motif. There was concurrent recruitment of the PP2A scaffolding subunit to the proline-rich apoER2 C terminus, along with two distinct regulatory PP2A subunits that mediate inhibitory dephosphorylation of Akt and endothelial nitric oxide synthase, by SHC-1 (Src homology domain-containing transforming protein-1). In murine experiments, the coupling of these processes in the endothelium was shown to underlie antiphospholipid antibody-invoked thrombosis. The researchers commented that elucidation of the workings of the pathogenesis of antiphospholipid syndrome-related thrombosis has identified numerous potential new therapeutic targets.

Comment (LY): The clinical and laboratory phenotype of the antiphospholipid syndrome was first recognised in the 1980s; however, the mechanism by which antiphospholipid antibodies induced a prothrombotic phenotype remained unclear. These investigators have already shown that the interaction with B2GP-1 induces a reduction in endothelial nitric oxide but the specifics of this interaction were unclear. In this elegant work, more of the molecular links through the apoER2 receptor on the vascular endothelium are elucidated. The potential for targeting this mechanism is an exciting future possibility, other than the current standard of anticoagulation.

Reference: Blood 2018;131:2097–110
Abstract

myeloma new zealand

Myeloma NZ is a new foundation in NZ to provide a deeper level of support for those who affected by multiple myeloma. If patients or their loved one have been diagnosed with multiple myeloma, Myeloma NZ can help them learn about treatment options and point them to information and services to help them cope with the disease. www.multiplemyeloma.org.nz/

Independent commentary by Dr Laura Young, a haematologist specialising in thrombosis and haemostasis at Auckland Hospital as part of the Thrombosis Unit and Haemophilia Centre. She also has a part-time lecturing position in the Department of Molecular Medicine and Pathology at the University School of Medicine. **For full bio CLICK HERE.**

Independent commentary by Dr Paul Ockelford, a haematologist and Clinical Associate Professor at the University of Auckland School of Medicine and Director of both the Adult Haemophilia Centre and Thrombosis Unit at Auckland City Hospital. He serves on the Medical Advisory panel of the New Zealand Haemophilia Foundation and the National Haemophilia Management Group. **For full bio CLICK HERE.**

Antiphospholipid antibodies and recurrent thrombosis after a first unprovoked venous thromboembolism

Authors: Kearon C et al.

Summary: In this research, levels of anticardiolipin antibodies, anti-β2 glycoprotein-1 antibodies and lupus anticoagulant were measured on two occasions, ~6 months apart, in 307 participants with a first unprovoked VTE from a prospective cohort study. The association between antiphospholipid antibodies and recurrent thrombosis was explored in patients who stopped anticoagulant therapy in response to negative D-dimer results (n=290). Compared with patients without antiphospholipid antibodies, the likelihood of recurrent VTE was increased significantly in those who had the same antiphospholipid antibodies detected on two occasions (9% of patients; hazard ratio 2.7 [p=0.03]) or two or three different antiphospholipid antibody types on either the same or different occasions (3.8%; 4.5 [p=0.006]); the association for those with any APA on detected on ≥1 occasion did not reach significance (25.9%; 1.8 [0.9, 3.7]). No associations were evident between antiphospholipid antibody detection and D-dimer levels.

Comment (LY): Continuing the theme of antiphospholipid syndrome, this interesting analysis explored antiphospholipid antibody positivity in a study of the value of a point of care qualitative D-dimer assay for predicting recurrence risk. In the study, the minority who had a positive D-dimer remained on anticoagulants and the others discontinued. Antiphospholipid antibodies were measured at three timepoints and did not influence decision making, although an unknown number of patients may have been excluded prior to enrolment on the basis of positive antiphospholipid antibody tests. As noted in other cohorts, recurrence risks were significantly elevated in those who had antiphospholipid antibody tests positive on more than one occasion and/or more than one type of positive antiphospholipid antibody test. Interestingly however, quantitative D-dimer levels were not statistically significantly higher in the group with positive antiphospholipid antibodies as one might expect, given the presumed activation of thrombosis on the endothelium. The study largely confirms established dogma regarding the significance of both more than one assay and sustained positivity over time.

Reference: Blood 2018;131:2151–60
Abstract

NZMA New Zealand Medical Association

Time spent reading this publication has been approved for CME for Royal New Zealand College of General Practitioners (RNZCGP) General Practice Educational Programme Stage 2 (GPEP2) and the Maintenance of Professional Standards (MOPS) purposes, provided that a Learning Reflection Form is completed. Please **CLICK HERE** to download your CPD MOPS Learning Reflection Form. One form per review read would be required.

Time spent reading this publication has been approved for CNE by The College of Nurses Aotearoa (NZ) for RNs and NPs. For more information on how to claim CNE hours please **CLICK HERE**

Altered plasma clot properties increase the risk of recurrent deep vein thrombosis

Authors: Cieslik J et al.

Summary: The ability of abnormal clot properties to predict recurrent DVT was explored in 320 consecutive adults who had experienced a first DVT. Plasma clot properties were recorded following 3 months of anticoagulant treatment (mean duration 10 months) since the index event. The 12-month DVT recurrence rate was 6.6%, with one-quarter of patients experiencing recurrence over median follow-up of 44 months. Significant associations were seen between DVT recurrence and faster formation of denser fibrin networks and 4% higher maximum absorbance of plasma clots that displayed impaired fibrinolytic degradation (25% clot lysis time prolongation) and a 5% slower rate of increase in D-dimer levels during clot degradation. Independent predictors of DVT recurrence were proximal DVT alone, higher C-reactive protein level, D-dimer level, peak thrombin level, lower fibrin clot permeability, shorter lag phase, decreased D-dimer level during clot degradation and prolonged clot lysis time ($p<0.05$ for all). The risk of DVT recurrence was greatest in patients with reduced fibrin clot permeability and prolonged clot lysis time (OR 15.8 [95% CI 7.5, 33.5]).

Comment (LY): It is appealing and logical to think that properties of clot breakdown as well as formation would predict the likelihood of recurrent DVT. Inhibitors of fibrinolysis were included for a while in thrombophilia panels before being discarded. However previous efforts to correlate recurrence with thrombin generation did not show a correlation. In this work using a laborious technique to measure clot breakdown as well as other measures of thrombin generation, a correlation was demonstrated. None of these techniques are currently in regular clinical use however the idea is appealing and perhaps investigation of assessment of active fibrinolysis as a marker of thrombosis risk will continue.

Reference: Blood 2018;131:797–807
Abstract

Gene therapy in patients with transfusion-dependent β-thalassemia

Authors: Thompson AA et al.

Summary: These two phase 1–2 studies evaluated the safety and efficacy of gene therapy with lentiviral transfer of a marked β-globin in 22 patients aged 12–35 years with transfusion-dependent β-thalassaemia, from whom autologous CD34+ cells were obtained and transduced *ex vivo* with LentiGlobin BB305 vector encoding HbAT87Q (adult haemoglobin with a T87Q amino acid substitution). Following myeloablative busulfan conditioning, the cells were reinfused. At a median of 26 months postinfusion, all but one of the participants with a non-β0/β0 genotype (n=13) no longer required RBC transfusions; their HbAT87Q levels were 3.4–10.0 g/dL and their total haemoglobin levels were 8.2–13.7 g/dL. Biological markers of dyserythropoiesis were corrected in evaluated participants with near-normal haemoglobin levels. In participants with a β0/β0 genotype or two copies of the IVS1-110 mutation (n=9), the median annualised transfusion volume fell by 73% and three discontinued RBC transfusions. Treatment-related adverse events were consistent with autologous stem-cell transplantation, and clonal dominance related to vector integration was not seen.

Comment (PO): β-thalassaemia is a common genetic disorder in Africa, Asia and the Middle East. Approximately 75% have severe disease, are transfusion dependent and need iron chelation to reduce iron overload. Treatment-related complications are high. Allogeneic bone marrow transplant is the only curative treatment, but limited by lack of suitable donors. Successful gene therapy for β0/β0 was first reported in 2010. This report uses a lentiviral vector derived from that patient. Autologous CD34+ patient cells (haematopoietic stem cells) were transduced *ex vivo* and returned to the patient after single-agent busulfan conditioning. At a median 2 years, 12 of 13 less severe (non-β0/β0) patients became transfusion-independent and transfusions were decreased by 73% in the β0/β0 patients. Haemoglobin levels became normal in several individuals with no vector-related adverse effects. Further development of curative haematopoietic stem-cell gene therapy for this monogenic disorder can be expected. The major challenge is how the therapy will be available to the largest population of patients living in third-world countries.

Reference: N Engl J Med 2018;378:1479–93
Abstract

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Gene therapy with adeno-associated virus vector 5–human factor IX in adults with hemophilia B

Authors: Miesbach W et al.

Summary: Adults with haemophilia B with a severe-bleeding phenotype received 5×10^{12} (n=5) or 2×10^{13} (n=5) genome copies of AMT-060 (AAV-5 vector combined with a liver-specific promoter that drives expression of a codon-optimised wild-type human FIX gene) per kilogram. For the respective low- and high-dose groups, mean endogenous FIX activity increased to 4.4 and 6.9 IU/dL, annualised FIX use fell by 81% and 73%, and mean annualised spontaneous bleeding rates decreased by 53% and 70%. Both groups also had stable FIX activity, and eight of the nine participants receiving FIX at enrolment stopped prophylaxis. Limited, asymptomatic, transient alanine aminotransferase level elevations, with no decreases in FIX activity or capsid-specific T-cell response, affected one low-dose recipient and two high-dose recipients; all were treated with prednisolone.

Comment (PO): Haemophilia is another single-gene disorder with sustained endogenous factor expression after gene therapy. Modest rises in the clotting factor are sufficient to reduce spontaneous bleeding. Recent reports differ based on the AAV vector used, the vector dose and the gene inserted (wild type or mutant for FIX). AAV8 transduced with wild-type FIX led to durable FIX activity (~5%) in ten patients for >4 years. Unfortunately there was a capsid-specific immune T-cell response in two-thirds of those receiving the highest vector dose, with significant loss of transduced liver cells and FIX levels. AAV5 has also been used for factor VIII gene transfer. Advantages are a lower potential for immune rejection and a lower population prevalence of neutralising antibodies (3.2% vs. 19% for AAV8). This interim analysis reports favourable activity levels, reduced bleed frequency and less product dependence. A mild alanine aminotransferase level rise in three patients responded to steroids. There was no capsid-specific immune rejection.

Reference: Blood 2018;131:1022–31
Abstract

Comparison of an oral factor Xa inhibitor with low molecular weight heparin in patients with cancer with venous thromboembolism

Authors: Young AM et al.

Summary: Patients with active cancer (58% with metastases) and symptomatic PE, incidental PE or symptomatic lower-extremity proximal DVT were randomised to receive dalteparin 200 IU/kg daily for 1 month then 150 IU/kg daily for months 2–6 (n=203) or rivaroxaban 15mg twice daily for 3 weeks, then 20mg once daily for a total of 6 months (n=203) in the SELECT-D trial. Compared with dalteparin, rivaroxaban was associated with a significantly lower 6-month cumulative VTE recurrence rate (4% vs. 11%; hazard ratio 0.43 [95% CI 0.19, 0.99]) and a similar 6-month cumulative major bleeding rate (6% vs. 4%; 1.83 [0.68, 4.96]), but a higher clinically relevant nonmajor bleeding rate (13% vs. 4%; 3.76 [1.63, 8.69]).

Comment (PO): This trial adds to the increasing evidence to support the use of the new target-specific anti-Xa anticoagulants for thrombosis complicating malignancy. Edoxaban (not available in NZ) and now rivaroxaban (due for Pharmac listing on August 1st) are effective as alternatives to LMWH. Both agents were compared with dalteparin. Rivaroxaban showed an approximate halving of VTE recurrence, compared with the LMWH comparator, but there was no difference in overall survival or symptomatic or fatal PE (insufficient study power). Major bleed rates are similar (6% rivaroxaban vs. 4% LMWH) and broadly in line with the edoxaban study. Nonmajor but clinically relevant bleeding was three-fold higher with rivaroxaban. This was predominantly GI tract and urological. No CNS bleeds were reported. The optimal duration of VTE treatment in cancer remains unresolved but usually while risk factors persist. These results are likely to be practice changing for those currently treated with extended enoxaparin.

Reference: J Clin Oncol; Published online May 10, 2018
Abstract

Targeting anticoagulant protein S to improve hemostasis in hemophilia

Authors: Prince R et al.

Summary: These researchers investigated targeting of protein S to promote haemostasis in haemophilia via rebalancing coagulation. In haemophilic mice, targeting the protein S gene resulted in protection against bleeding, especially intra-articular. The mice were observed to have increases in thrombin generation, resistance to activated protein C and TFPI (tissue factor pathway inhibitor), and an improved fibrin network. In plasma of patients with haemophilia, blocking protein S was found to normalise thrombin generation *in vitro*. Protein S and TFPI-α were detected in the joints of the haemophilic mice, and they were strongly expressed in the joints of patients with haemophilia A patients, when compared with patients with haemophilia B, during on-demand therapy. In contrast, expression of protein S and TFPI was decreased in patients with haemophilia A receiving prophylaxis, comparable to patients with osteoarthritis.

Comment (PO): Disruptive therapies to rebalance the haemostatic pathway by targeting inhibitory coagulation proteins are already in clinical trial. Inhibitor targets include antithrombin, using small silencing RNA molecules, and antibodies against TFPI. Protein S, encoded by the *PROS1* gene, is another potential target. As a cofactor for activated protein C-mediated inactivation of Va/VIIIa, and inhibition of Xa by TFPI, it is a key regulator of thrombin generation. In an elegant series of murine experiments, and with observations on testing clinical samples from haemophilia patients, proof of concept that protein S and TFPI contribute to haemarthrosis and haemophilia joint damage is provided. They are produced within joints by fibroblast-like synoviocytes. In the presence of protein S, haemarthrosis increases synovial TFPI expression. Targeting protein S either with a specific antibody or an inhibitory sRNA protects against haemarthrosis. This offers the potential for another therapeutic target in haemophilia, but the merits of this approach await future studies.

Reference: Blood 2018;131:1360–71
Abstract

The effect of recanalization on long-term neurological outcome after cerebral venous thrombosis

Authors: Rezoagli E et al.

Summary: This retrospective observational study investigated recanalisation rates following CVT (cerebral vein thrombosis) and its prognostic role in long-term neurological outcomes in a cohort of 508 patients who had experienced an acute first episode of CVT and had ≥1 imaging test during follow-up. There was no differential representation of complete or partial recanalisation in patients who underwent scans at different periods of time (from a 28-day to 3-month period, up to a 1- to 3-year-period). Among patients with available modified Rankin scale scores at the time of follow-up imaging (n=483), 92.8% had a good score (0–1). The likelihood of a favourable modified Rankin scale score at follow-up was increased with CVT recanalisation (OR 2.56 [95% CI 1.59, 4.13]), but decreased with cancer (0.29 [0.09, 0.88]) and a personal history of VTE (0.36 [0.14, 0.92]).

Comment (PO): The estimated incidence of CVT is ~1.5 per 100,000. The mortality approximates 4% in the acute phase and up to 10% overall. About 12% have a poor neurological outcome. How important is thrombus recanalisation? This large cohort study indicates that recanalisation exceeds 80% within 3 months. Median treatment duration was 6 months. Recanalisation was independently associated with complete neurological recovery, which approached 95% by 3 years after the event. Higher recanalisation rates are seen in those with single site and pregnancy-related episodes, but anticoagulant duration is not predictive of recanalisation. Young women (aged <39 years) with hormone-associated events have a good outcome, as shown in the ISCVT study (Stroke 2009), but those with cancer and a personal history of VTE do less well.

Reference: J Thromb Haemost 2018;16:718–24
Abstract