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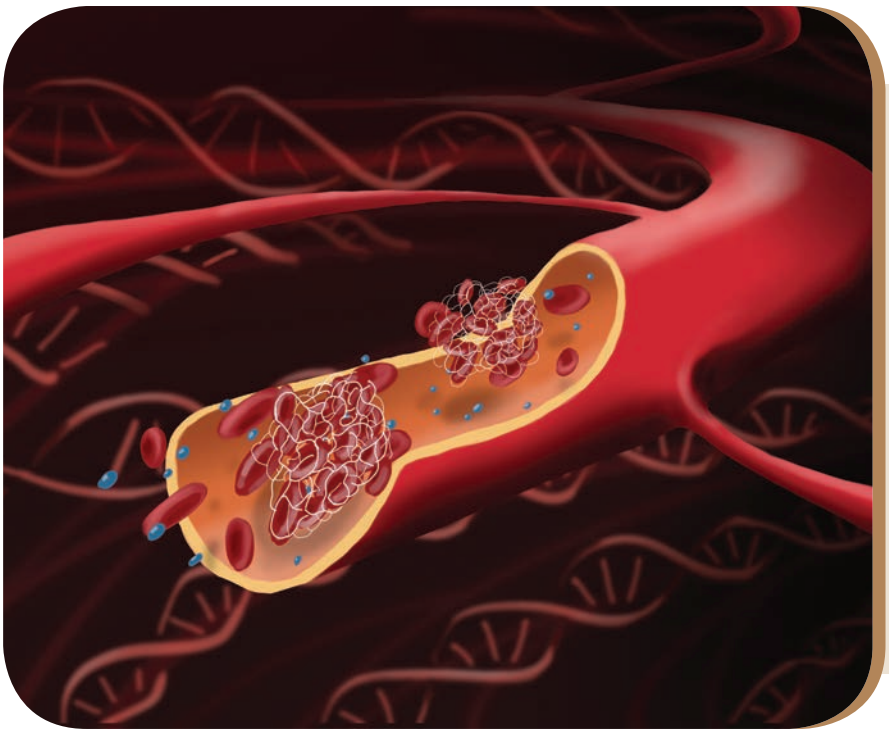

Direct oral anticoagulants (DOACs) are prescribed for conditions such as stroke, but they are associated with bleeding complications.

A study investigated the association between polymorphisms in fibrinogen genes and bleeding risk in patients who received DOACs in South Korea. The study found that 2 genotypes were related to bleeding risk for DOACs and developed a risk scoring system to predict it.

Illustration by Xinyu Li

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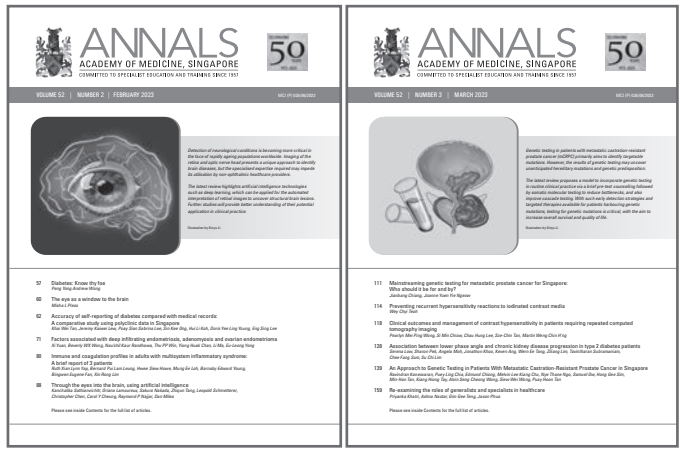

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EDITORIAL

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Incorporating assessment of fibrinogen gene polymorphisms and bleeding risk in patients treated with direct oral anticoagulants

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Direct oral anticoagulants (DOACs) have become entrenched as the dominant anticoagulant over the last decade for patients with venous thrombosis and atrial fibrillation.[1] Compared to warfarin, bleeding risk is similar or lower for patients on DOACs but clinically relevant bleeding is still a risk, especially for patients with impaired organ function.[2] Furthermore, current bleeding risk assessment tools, such as the Hypertension, Abnormal renal/liver function, Stroke, Bleeding history or predisposition, Labile international normalised ratio, Elderly, Drugs/alcohol concomitantly (HAS-BLED) score, were designed for patients on warfarin. As such, tools for the prediction of bleeding risk in patients taking DOACs are clearly wanting.

Various studies have been performed to determine if genetic polymorphisms could predict bleeding risk for patients on antithrombotic agents, including DOACs.[3] These existing works have been predominantly focused on the pharmacodynamic and pharmacokinetic aspects of drug metabolism and how these pharmacogenetic factors are associated with clinical outcomes.[4,5] In this issue of the *Annals*, Choi et al. adopted a novel genetic polymorphism approach to study bleeding in patients taking DOACs.[6] The group of Korean investigators examined polymorphisms in genes encoding fibrinogen and 2 targeted representative variants of prothrombin and FX genes. Fibrinogen, prothrombin and FX are crucial clotting factors in the generation of fibrin clots. It is theoretically plausible that polymorphisms in these genes, especially fibrinogen genes, could contribute to differences in bleeding risk in this patient population.

In this study, patients treated with DOACs demonstrated a significant association between genetic polymorphisms in fibrinogen genes and bleeding. The study included 468 patients on DOACs, of which 50 had bleeding episodes (14 of which were major). Single nucleotide polymorphisms (SNPs) in the prothrombin (*F2* rs5896) and fibrinogen gamma chain (*FGG* rs1800792) genes were significantly associated with bleeding risk. While s1800792 was not the strongest overall predictor in the model, it is the strongest predictor among all fibrinogen gene SNPs. Applied to a scoring system, a significant and almost linear association with bleeding risk was found. Allelic and genotypic frequencies as well as Hardy-Weinberg Equilibrium estimates were provided for each single nucleotide polymorphism to justify how the authors arrived at their conclusions.

The association between bleeding risk and the genetic polymorphisms in prothrombin and fibrinogen genes refers to all DOACs in the study. One may argue the clinical usefulness of detecting these non-modifiable factors. However, if these findings are validated, the presence of these SNPs can help identify patients with higher bleeding risk to physicians for early review and closer follow-up. It would be interesting to compare the performance of this novel approach to the more established risk assessment models.

Another key function of applying a bleeding risk assessment score is to highlight to the prescribers of anticoagulants any potential modifiable factors amenable to intervention. This study identified 2 modifiable factors, namely, anaemia and overdosing of DOACs, both of which were strongly associated with bleeding. Overdosing of DOACs as a factor was given the highest weightage in the proposed risk score model and is probably most amenable to intervention. Inappropriate dosing and overdosing of DOACs have been well-reported to be associated with adverse outcomes in patients taking DOACs.[7] Different types of DOACs are currently available and each DOAC has its own specific variations in dosing according to indications, creatinine function, age and concomitant drugs, to name a few. This has understandably created potential dosing problems and may put patients at risk of bleeding and/or thrombosis. Continuous education of prescribers as well as checks and prompts on the electronic prescribing system can mitigate the problem.

While it is logical that patients with coagulation factor deficiencies taking antithrombotic agents would be at higher bleeding risk, the mechanism of how carrying SNPs in fibrinogen and prothrombin genes contribute to

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heightened bleeding risk is less apparent. No functional assays or biological data have been reported on how the 2 SNPs detected in this study contribute to increased bleeding. Further mechanistic studies are needed to investigate the effects of these SNPs. Only Asian populations living in Korea were included in this study, thus reducing the generalisability of the results. As genetic variation and its effects could vary across race and ethnic groups, it should be applied with caution. Also, genetic polymorphism testing is not a routinely available test and there are many other confounders that are not considered in the risk scoring system.

Confirming the study findings in a broader population with other ethnic groups could lead to significant findings and methodologies that could be applied to stratify dosing for patients on DOACs. As we eagerly await more advancements in genetic polymorphism testing in the use of antithrombotic agents, and further confirmatory and validation of the findings in this study, let us not forget about getting the basics right—knowing DOACs and our patients well to minimise bleeding risk for our patients.

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EDITORIAL

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Promise and pitfalls of ChatGPT for patient education on coronary angiogram

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The past decade has seen extraordinary and rapid progress in the field of artificial intelligence (AI), which produces computer systems capable of performing tasks that typically require human intelligence. These advancements have yielded wide-ranging applications across various domains that are revolutionising industries and transforming the way humans live and work. Accordingly, potential applications of AI technology in the healthcare field are being investigated, such as deep learning for image analysis to detect lesions and provide diagnostic support as well as data mining and machine learning for analysing patient data to predict disease risks.[1,2]

Chat Generative Pre-trained Transformer (ChatGPT; OpenAI, San Francisco, USA) is an advanced conversational AI model that generates human-like responses based on a large dataset of text and can engage in interactive and contextually relevant conversations. This model has experienced remarkable growth since its public launch in November 2022, finding widespread use across online platforms and serving as a virtual assistant on devices and improving customer support. It has also become a valuable tool for language learners, offering conversational practice and feedback. Moreover, individuals are enjoying personal interactions with ChatGPT, seeking advice, brainstorming ideas, and enjoying its companionship. Given the widespread use of this technology, it is inevitable that patients will utilise it for gathering medical information. Accessing medical information via ChatGPT may make it easier for patients to self-inform and ask questions that they might find difficult to ask healthcare professionals directly. In addition, it has the potential to be valuable for healthcare providers, as it can help convey complex medical information in a more understandable manner. However, there is insufficient evidence thus far to confirm whether ChatGPT can provide accurate and useful information to patients.

In this issue of *Annals,* Koh et al. aimed to explore the quality of medical information provided and assess the opportunities and challenges of patient education using this natural-language AI model, focusing on the coronary angiogram procedure.[3] The authors employed a conversational approach, posing common questions about coronary angiography to ChatGPT and evaluating its responses across different domains. The strengths of ChatGPT's answers were evident. They were comprehensive, systematic and presented in plain language accessible to laypersons. Yet, the model still emphasised the importance of involving healthcare professionals in discussing individual circumstances and acknowledged its limitations in providing personalised recommendations.

Moreover, the study identified certain limitations in ChatGPT's responses. Factual inaccuracies were infrequent but present, including confusion between antiplatelet and anticoagulation drugs, inaccurate indications and risks of angiography, and incorrect contraindications. Some significant omissions were also noted, such as the exclusion of active acute coronary syndromes as an indication for angiography. Inaccurate assumptions were also observed such as the suggestion of routine sedation during the procedure. Additionally, the model appeared inflexible in expanding recommendations beyond the specific line of questioning, limiting its ability to consider non-cardiac causes of symptoms. Given that the medical information provided to patients can potentially influence their decision-making, it is crucial not to overlook the issues associated with ChatGPT.

Previous studies have also reported that ChatGPT may provide inaccurate medical information. A recent study examined the appropriateness of AI model responses to questions based on guideline-based cardiovascular prevention topics and found errors in 16% (21 out of 25 questions) of ChatGPT's responses.[4] Another study evaluating the accuracy of ChatGPT in answering clinical questions based on the Japanese society of hypertension guidelines revealed an overall accuracy rate of only 64.5% for ChatGPT's answers.[5]

The limitations of ChatGPT can be attributed to various factors. First, the inclusion of information from diverse internet sources introduces the possibility of

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presenting inaccurate information to users. Second, the need for probing and prompting for information that healthcare providers would typically provide during counselling highlights the limitations of the model in conveying comprehensive information. Such omissions may impact patients' decision-making processes. Third, the conversational nature and scoping of the topic might restrict the model's flexibility in addressing lateral aspects, especially when considering differential diagnoses. Furthermore, the model's reliance on data inputs with cut-off dates may result in outdated or incomplete information, and thus effort should be directed towards improving the accuracy of information provided by ChatGPT. This may involve incorporating real-time medical updates and ensuring access to reliable, up-to-date sources.

In addition to the challenges revealed by Koh's research, there are several issues to address before ChatGPT can be considered a useful tool for medical information-gathering for patients in the future. There is concern regarding the risk of data breaches and the leakage of personal information given by patients, and implementing robust security measures and data protection protocols would be crucial to minimise these risks and safeguard patient privacy. Additionally, ChatGPT's lack of human-like emotions and ethical judgement poses challenges when dealing with important medical decisions or providing information tailored to individual situations.

In conclusion, ChatGPT demonstrates potential as an adjunct tool for patients to acquire health information, and understanding the strengths and limitations of such natural-language AI platforms is essential for both patients and healthcare providers. Incorporating these models into healthcare systems alongside ongoing improvements holds promise for enhancing healthcare delivery. Nonetheless, it is vital to recognise that current AI models cannot replace the pivotal role of healthcare providers in delivering personalised care.

Conflict of interest

The authors have no conflicts of interest to disclose.

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ORIGINAL ARTICLE

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Association between genetic polymorphisms in fibrinogen genes and bleeding risk in patients treated with direct oral anticoagulants

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ABSTRACT

Introduction: This study aimed to investigate the association between polymorphisms in fibrinogen genes and bleeding risk in patients receiving direct oral anticoagulants (DOACs).

Method: Patients treated with DOACs from June 2018 to December 2021 were enrolled in the study. Genotyping was done for rs2070011, rs6050, and rs2070022 in fibrinogen alpha chain (*FGA*); rs1800788, rs4220, and rs4463047 in fibrinogen beta chain (*FGB*); and rs2066865 and rs1800792 in fibrinogen gamma chain (*FGG*), along with *F2* rs5896 and *F10* rs5960. Multivariable logistic regression analysis was performed to investigate the risk factors for bleeding and to develop a risk scoring system.

Results: A total of 468 patients were included in the analysis, 14 of whom experienced major bleeding and 36 experienced clinically relevant non-major bleeding. In the multivariable analysis, overdose, anaemia, *F2* rs5896, and *FGG* rs1800792 were found to be significantly associated with bleeding risk. Specifically, patients with the TT genotype of *F2* rs5896 and the CC genotype of *FGG* rs1800792 had 2.1 times (95% confidence interval [CI] 1.1–3.9) and 2.7 times (95% CI 1.2–5.9) higher bleeding risk than the C allele and T allele carriers, respectively. Based on the risk scoring system, patients with 0, 1, 2, 3, 4, and 5 points were predicted to have 5.2%, 10.8%, 22.4%, 32.3%, 42.3%, and 61.8% of bleeding risk, respectively.

Conclusion: To our knowledge, this is the first study to investigate the effects of polymorphisms in fibrinogen genes on DOAC response. After validation, these results will be useful for personalised DOAC therapy.

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Keywords: bleeding, direct oral anticoagulant, fibrinogen, thrombin, pharmacogenomics

INTRODUCTION

Direct oral anticoagulants (DOACs) are widely prescribed for the prevention and treatment of stroke, systemic embolism and venous thromboembolism.[1] Their mechanism of action involves direct binding to and inhibition of activated coagulation factors—factor Xa and thrombin—thereby preventing excessive blood clotting.[2] Overall, DOACs have favourable efficacy and safety profiles but are commonly associated with the complication of bleeding.[3] Hernandez et al. reported cumulative 1-year incidence rates of intracranial bleeding, gastrointestinal bleeding, and any bleeding

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CLINICAL IMPACT

What is New

- This study reports the effects of fibrinogen gene polymorphisms on bleeding risk in Asian patients taking direct oral anticoagulants.
- We propose a new bleeding risk scoring system that incorporates clinical (anaemia and overdose) and genetic risk factors (*F2* rs5896 and *FGG* rs1800792).

Clinical Implications

- An understanding of pharmacogenomics can prevent bleeding complications associated with direct oral anticoagulants.

event at 1%, 6–11%, and 19–26% in DOAC-treated patients, respectively.[4] Several recent works have suggested that some individuals with a genetic predisposition are more susceptible to DOAC-related haemorrhage.[5] For example, Paré et al. showed that the T allele of carboxylesterase 1 (*CES1*) rs2244613 is associated with an increased risk of any bleeding event in patients treated with dabigatran.[6] However, most studies were limited to pharmacokinetics-related genes.[5]

Fibrinogen is one of the important factors in the coagulation cascade.[7] After activating coagulation factor X to Xa and subsequently activating prothrombin to thrombin—both targets for DOACs—fibrinogen is cleaved into fibrin monomers, which form fibrin clots to aggregate platelets and promote coagulation.[8] As fibrinogen is involved in haemostasis, its abnormality leads to prothrombin time prolongation and increased bleeding risk.[9] Previous clinical studies revealed that plasma fibrinogen concentration is related to coagulation and haemorrhage.[10,11] Fibrinogen consists of 3 polypeptide chains—Aα, Bβ and γ—which are encoded by fibrinogen alpha chain (*FGA*), fibrinogen beta chain (*FGB*) and fibrinogen gamma chain (*FGG*) genes, respectively.[12] The fibrinogen gene family is clustered on chromosome 4 and spans ~50 kb, comprising *FGB*, *FGA* and *FGG*. As several single nucleotide polymorphisms (SNPs) of fibrinogen genes are reportedly related to fibrinogen levels,[13] we hypothesised that these genetic features may affect bleeding risk. Therefore, this study aims to investigate the association between polymorphisms in fibrinogen genes and bleeding risk in DOAC-treated patients.

METHOD

Study patients

This retrospective study was conducted using prospectively collected samples; the study cohort has been previously described in detail.[14,15] Briefly, patients treated with DOACs (direct thrombin inhibitors or factor Xa inhibitors) from June 2018 to December 2021 at Ewha Womans University Mokdong Hospital and Ewha Womans University Seoul Hospital were included in the study. For control groups, we only included patients who had at least a 3-month follow-up. Patients were excluded if they (1) were younger than 20 years old, (2) experienced infarction-related events, (3) had minor or unverified bleeding, and (4) withdrew consent. The primary outcome was any 1-year bleeding event, defined as a combined endpoint of major bleeding and clinically relevant non-major bleeding (CRNMB) by the International Society on Thrombosis and Haemostasis criteria;[16,17] therefore, we excluded patients who experienced bleeding episodes ≥1 year after DOAC treatment. The following demographic and clinical information were collected from electronic medical records: sex, age, body weight, creatinine clearance (CrCl), CHA_2DS_2-VASc score (Congestive heart failure, Hypertension, Age ≥75 years, Diabetes mellitus, Stroke, Vascular disease, Age 65–74 years, Sex category),[18] modified HAS-BLED (Hypertension, Abnormal renal/liver function, Stroke, Bleeding history or predisposition, Labile INR, Elderly, Drugs/alcohol concomitantly), excluding the liable INR,[19] type and dose of DOAC, social history (smoking and alcohol status), comorbidity and co-medication.

The study was performed in accordance with the Declaration of Helsinki principles and was approved by the Institutional Review Board (IRB) of Ewha Womans University Mokdong Hospital (IRB number: 2018-04-006) and Ewha Womans University Seoul Hospital (IRB number: 2019-05-038). All patients provided informed consent before participation.

Genotyping

To select SNPs in 3 fibrinogen genes (*FGA*, *FGB* and *FGG*), genetic information was obtained from the dbSNP,[20] HaploReg 4.1[21] and PharmGKB databases.[22] Based on the functionality, minor allele frequency and linkage disequilibrium pattern, the following SNPs were selected: rs2070011, rs6050 and rs2070022 in *FGA*; rs1800788, rs4220 and rs4463047 in *FGB*; and rs2066865 and rs1800792 in *FGG*. As thrombin and factor Xa are the main targets of DOACs, rs5896

and rs5960, which are respectively the representative mutations of *F2* and *F10*,[23,24] were additionally included as potential confounding variables along with the selected SNPs.

Genomic DNA was extracted from blood or saliva samples collected in EDTA vacutainer tubes or Oragene OG-600 kits (DNA Genotek, Ottawa, ON, Canada), respectively. DNA was extracted following manufacturers' protocols from blood samples using the QIAamp DNA Blood Mini Kit (QIAGEN GmbH, Hilden, Germany) or saliva samples using the PrepIT-L2p kit (DNA Genotek, Ottawa, ON, Canada). Genotyping was performed using the TaqMan assay (Applied Biosystems, Foster City, CA, US).

Statistical analysis

Allele frequency was calculated from genotype frequency, and deviation from Hardy–Weinberg Equilibrium (HWE) was tested using chi-squared test. Chi-squared test and unpaired t-test were used to determine differences between case and control groups. Multivariable logistic regression was performed to determine independent predictors for bleeding risk. All variables with *P* values <0.05 in the univariate analysis were included along with sex and age, and backward elimination was performed for variable selection. The crude and adjusted odds ratio (OR) with the 95% confidence interval (CI) were estimated from univariate and multivariable analyses, respectively. The attributable risk was calculated as (adjusted OR−1)/adjusted OR. The Hosmer–Lemeshow goodness-of-fit was checked for model fitness.

A risk scoring system was developed according to the results of the multivariable logistic regression analysis. To assign the score for each risk factor, its OR was divided by the lowest value and rounded to the nearest integer. The area under the receiver operating characteristic (AUROC) curve was used to evaluate the performance of the risk scoring system.

All statistical analyses were performed using SPSS Statistics version 20 (IBM Corp, Armonk, NY, US). A *P* value of <0.05 was considered statistically significant.

RESULTS

A total of 576 patients treated with DOACs were enrolled. Among them, 108 patients were excluded for the following reasons: patients with <3 months of follow-up in the control group (n=5), patients with infarction-related events (n=25), patients with minor or unverified bleeding (n=23), patients with bleeding episodes >1 year after DOAC treatment (n=43), a patient who withdrew the consent (n=1), and a patient whose sample was not analysed (n=1). Therefore, 468 patients, including 14 patients who experienced major bleeding and 36 who experienced CRNMB, were included in the analysis.

Overall, 37.4% of patients were female (Table 1). The mean (standard deviation) age and weight were 69.2 (10.1) years old and 67.4 (11.7) kg, respectively. Approximately 90% of the patients had ≥2 CHA_2DS_2-VASc scores, whereas two-thirds of them had ≥2 modified HAS-BLED scores. Atrial fibrillation was the most prevalent comorbidity (98.5%), followed by hypertension (67.5%). Beta-blockers (71.2%) and statins (58.3%) were the most common comedications in the study population. Among clinical factors, the dose of DOAC and anaemia were significantly associated with bleeding risk. Despite not being statistically significant, patients with CrCl <30 (*P*=0.086) and a history of stroke/transient ischaemic attack/thromboembolism (*P*=0.076) tended to have a higher bleeding risk.

The distribution of allelic and genotypic frequencies is presented in Supplementary Table S1, and all SNPs are in HWE. The results of the genetic association analysis are shown in Table 2. The TT genotype of *F2* rs5896 was associated with an increased bleeding risk compared to the CC or CT genotypes. Among the SNPs in fibrinogen-related genes, *FGA* rs2070011 and *FGG* rs1800792 showed a significant association with bleeding risk; patients with variant homozygotes of *FGA* rs2070011 and *FGG* rs1800792 had a higher bleeding risk than patients with the wild-type allele.

Table 3 shows the result of the multivariable logistic regression analysis. Even after the adjustment of confounders, overdose, anaemia, *F2* rs5896, and *FGG* rs1800792 were still found to be significantly associated with bleeding risk. According to the results, a DOAC overdose increased bleeding risk by 6.2 times (95% CI 1.7–23.1), whereas anaemia increased the risk by 2.3 times (95% CI 1.2–4.3). In addition, patients with the TT genotype of *F2* rs5896 and the CC genotype of *FGG* rs1800792 had 2.1 times (95% CI 1.1–3.9) and 2.7 times (95% CI 1.2–5.9) higher bleeding risk, respectively, than the wild allele carriers. The attributable risks of *F2* rs5896 and *FGG* rs1800792 were calculated as 52.0% and 62.6%, respectively. Hosmer–Lemeshow test showed a good fit of the model (χ^2=3.161, *P*=0.367).

For the risk scoring system, overdose (3 points), anaemia (1 point), *F2* rs5896 (1 point), and *FGG* rs1800792 (1 point) were summed up. Theoretically, the risk scores may range from 0 to 6; however, no patient had 6 points. Based on the risk scoring system, patients with 0, 1, 2, 3, 4, and 5 points had approximately 4.6%,

Table 1. Baseline characteristics of the study population.

	Patients with bleeding (n=50)	Patients without bleeding (n=418)	*P* value
Sex			0.925
Male	31 (62.0)	262 (62.7)	
Female	19 (38.0)	156 (37.3)	
Age (years)			0.760
<65	16 (32.0)	125 (29.9)	
≥65	34 (68.0)	293 (70.1)	
Body weight (kg)			0.925
<60	12 (25.0)	98 (24.4)	
≥60	36 (75.0)	304 (75.6)	
Creatinine clearance (mL/min)			0.086
<30	5 (10.4)	18 (4.5)	
≥30	43 (89.6)	384 (95.5)	
CHA_2DS_2-VASc			0.147
<2	3 (6.0)	55 (13.2)	
≥2	47 (94.0)	363 (86.8)	
Modified HAS-BLED			0.870
<2	17 (34.0)	147 (35.2)	
≥2	33 (66.0)	271 (64.8)	
Type of DOAC			0.195
Direct thrombin inhibitors	3 (6.0)	51 (12.2)	
Factor Xa inhibitors	47 (94.0)	367 (87.8)	
Dose of DOAC			0.014
Underdose	18 (36.0)	133 (31.8)	
Standard dose[a]	28 (56.0)	278 (66.5)	
Overdose	4 (8.0)	7 (1.7)	
Alcohol	17 (37.0)	135 (35.4)	0.848
Smoking	7 (14.0)	57 (13.6)	0.944
Comorbidities			
Atrial fibrillation	49 (98.0)	398 (98.5)	0.788
Hypertension	34 (68.0)	282 (67.5)	0.939
Diabetes mellitus	14 (28.0)	119 (28.5)	0.945
Heart failure	5 (10.0)	78 (18.7)	0.130
Previous stroke /TIA/thromboembolism	28 (56.0)	179 (42.8)	0.076
Previous haemorrhage	4 (8.0)	18 (4.3)	0.277
Anaemia	22 (44.0)	107 (25.6)	0.006
Comedications			
Antiplatelet drugs	3 (6.0)	51 (12.2)	0.195
ACEI/ARBs	19 (38.0)	186 (44.5)	0.381
Beta-blockers	38 (76.0)	295 (70.6)	0.424
Calcium channel blockers	13 (26.0)	116 (27.8)	0.793
Diuretics	11 (22.0)	109 (26.1)	0.533
Statins	28 (56.0)	245 (58.6)	0.723

ACEIs: angiotensin converting enzyme inhibitors; ARBs: angiotensin II receptor blockers; DOACs: direct oral anticoagulants; NA: not available; TIA: transient ischaemic attack. [a]According to the FDA-approved labelling.

11.9%, 21.7%, 22.2%, 40.0%, and 100.0% of bleeding risk. Fig. 1 shows the predicted probability of bleeding risk versus the developed risk score; patients with 0, 1, 2, 3, 4, and 5 points were predicted to have 5.2%, 10.8%, 22.4%, 32.3%, 42.3%, and 61.8% of bleeding risk, respectively. The AUROC curve was 0.680 (95% CI 0.601–0.758).

DISCUSSION

In this study, we analysed the polymorphisms of fibrinogen-related genes and investigated whether these mutations, along with other possible confounding variables, can affect bleeding risk in patients treated with DOACs. The main finding of our study is that the DOAC-associated bleeding risk is related to *F2* rs5896 and *FGG* rs1800792, along with anaemia and overdose.

Among the clinical factors, anaemia was one of the predictors for bleeding risk. Anaemia reduces the red blood cell count in the blood and disrupts platelet adherence to endothelial cells, leading to bleeding due to impaired haemostasis.[25] Clinically, most bleeding risk assessment tools for patients taking anticoagulants, which were developed before DOACs were introduced, have included anaemia as a risk factor.[26] For example, patients received 1 point and 1.5 points for anaemia in the $HEMORR_2HAGES$ (Hepatic or renal disease, Ethanol abuse, Malignancy, Older age, Reduced platelet count or function, Re-bleeding, Hypertension, Anaemia, Genetic factors, Excessive fall risk and

Table 2. Association between genetic polymorphisms and bleeding risk in patients treated with direct oral anticoagulants.

Genetic polymorphisms	Molecular Consequence	Grouped genotypes	Patients with bleeding (n=50)	Patients without bleeding (n=418)	*P* value
F2 rs5896 (C>T)	Missense	CC, CT	26 (52.0)	279 (67.4)	0.030
		TT	24 (48.0)	135 (32.6)	
F10 rs5960 (C>T)	Synonymous	CC	6 (12.2)	97 (23.4)	0.076
		CT, TT	43 (87.8)	318 (76.6)	
FGA rs2070011 (T>C)	5'-UTR	TT, TC	35 (70.0)	341 (81.8)	0.047
		CC	15 (30.0)	76 (18.2)	
FGA rs6050 (T>C)	Missense	TT	15 (30.0)	88 (21.1)	0.151
		TC, CC	35 (70.0)	329 (78.9)	
FGA rs2070022 (G>A)	3'-UTR	GG	34 (68.0)	318 (76.6)	0.179
		GA, AA	16 (32.0)	97 (23.4)	
FGB rs1800788 (C>T)	Upstream	CC	8 (16.0)	50 (12.1)	0.428
		CT, TT	42 (84.0)	364 (87.9)	
FGB rs4220 (G>A)	Intronic	GG	34 (68.0)	307 (73.6)	0.397
		GA, AA	16 (32.0)	110 (26.4)	
FGB rs4463047 (T>C)	Downstream	TT	15 (31.3)	90 (22.1)	0.152
		TC, CC	33 (68.8)	318 (77.9)	
FGG rs2066865 (G>A)	Downstream	GG	16 (32.0)	87 (20.9)	0.074
		GA, AA	34 (68.0)	329 (79.1)	
FGG rs1800792 (T>C)	Upstream	TT, TC	40 (80.0)	381 (91.6)	0.019
		CC	10 (20.0)	35 (8.4)	

UTR: untranslated region.

Table 3. Multivariable logistic regression analysis for bleeding risk in patients treated with direct oral anticoagulants.

Predictors	Crude OR (95% CI)	Adjusted OR (95% CI)	Attributable risk (%)
Female	1.03 (0.56–1.88)		
Age ≥65 (years)	0.91 (0.48–1.70)		
Overdose	5.11 (1.44–18.10)	6.19 (1.67–23.05)**	83.86
Anaemia	2.28 (1.25–4.16)	2.29 (1.23–4.28)**	56.43
F2 rs5896 TT	1.91 (1.06–3.45)	2.09 (1.13–3.85)*	52.04
FGA rs2070011 CC	1.92 (1.000–3.70)		
FGG rs1800792 CC	2.72 (1.25–5.90)	2.67 (1.20–5.94)*	62.58

OR: odds ratio; CI: confidence interval. *P<0.05; **P<0.01. Multivariable analysis was performed with the factors with P<0.05 in the univariate analysis along with sex and age.

Stroke) and RIETE (Registro Informatizado Enfermedad TromboEmbolica) risk scoring systems, respectively, whereas ATRIA (AnTicoagulation and Risk factors In Atrial fibrillation) assigned 3 points for anaemia. Similar results have been observed for anaemia as a bleeding risk factor after DOAC treatment. According

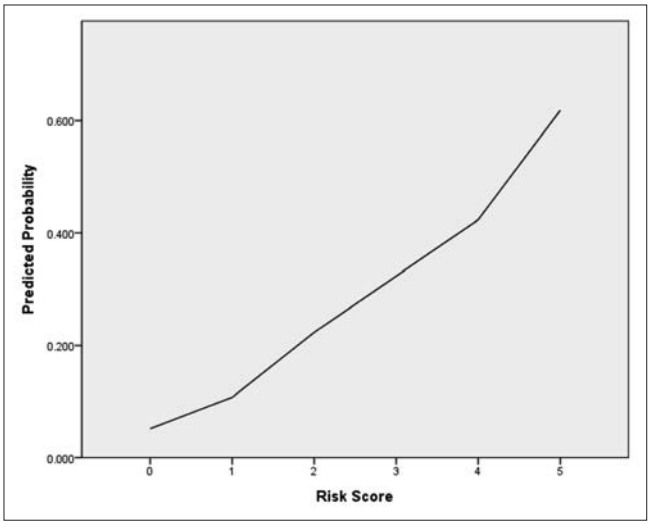

Fig. 1. The predicted probability of bleeding risk versus the developed risk score.

to the post hoc analysis of the RE-LY (Randomised Evaluation of Long-term anticoagulation therapY) trial, which evaluated dabigatran versus warfarin in patients with atrial fibrillation, anaemia was associated with a 2.1-fold higher risk of major bleeding.[27] The secondary analysis of ARISTOLE (Apixaban for Reduction In Stroke and Other ThromboembolLic Events in atrial fibrillation), a representative randomised controlled trial of apixaban versus warfarin,[28] also showed that anaemia increased the major bleeding risk by 1.92 times in patients taking apixaban. Thus, as anaemia is a modifiable risk factor for bleeding, it should be properly evaluated and managed in patients taking DOACs.

DOAC overdose is the most attributable risk factor for bleeding in our study. According to recent meta-analyses for the off-label overdosing of DOAC in patients with atrial fibrillation, patients who received a DOAC overdose were more likely to experience major bleeding than patients who received an on-label dose of DOAC.[29,30] Thus, tailored dosing of DOAC may be required for each patient for effective and safe treatment.

Among the genetic factors, rs1800792, located in the 5' flanking region of the *FGG*, was the strongest predictor for bleeding risk. This SNP was listed in the GTEx database as a significant expression quantitative trait locus for the *FGG* gene[31] and was related to fibrinogen levels.[32] Previous studies showed that fibrinogen gene haplotypes, including rs1800792, are associated with coagulation-related disorders, such as ischaemic stroke[33] and myocardial infarction.[34] Additional effects of rs1800792 have also been suggested; rs1800792 is associated with platelet counts[35] and interacts with interleukin levels.[36] As coagulation and fibrinolytic systems are complex and interconnected, many biomarkers can be involved, including coagulation factors, platelets, and inflammatory cytokines.[37] Further studies are needed to elucidate the detailed functional mechanisms of this SNP.

In addition to *FGG* rs1800792, the TT genotype of rs5896 in *F2* is associated with an increased bleeding risk. This SNP is a missense mutation of *F2*, which substitutes threonine (Thr) with methionine (Met) at position 165 in prothrombin.[20] Similar results were observed in the pharmacogenetic study of warfarin, another oral anticoagulant inhibiting vitamin K-related coagulation factors. According to D'Ambrosio,[38] variant allele carriers of rs5896 require a lower adjusted warfarin dose than those with wild-type homozygotes (2.9 mg vs 4.2 mg), which is related to the over-anticoagulation response in variant allele carriers. Shikata et al. also showed that a variant allele of rs5896 is related to increased warfarin sensitivity.[39] This effect can be explained by the conformational change of prothrombin. Thr 165 is located in the kringle 1 domain of prothrombin, which is extensively involved in protein–protein interactions with other clotting factors.[40] At position 165, the substitution of Thr, an amino acid with a polar side chain, with Met, an amino acid with a non-polar side chain, may disrupt hydrogen bond interactions and induce conformational changes of the three-dimensional structure of prothrombin. Although approximately 90% of the patients received Xa inhibitors, the *F2* SNP was identified as a genetic risk factor because the conversion of prothrombin to thrombin is required to complete the coagulation cascade in the presence of coagulation factor Xa.[7]

This study has several limitations. First, only Asian populations living in Korea were included, thereby reducing the generalisability of the results. As genetic variation and its effects can vary across race and ethnic groups, it should be applied with caution. Second, there were possible confounders not considered in this scoring system, which could affect the results. Lastly, the follow-up period was limited to 1 year to exclude the possibility of non-drug-related haemorrhage.

CONCLUSION

Despite these limitations, to our knowledge, this is the first study to investigate the genetic effects of polymorphisms in the fibrinogen genes on DOAC response. Although genotyping is not routinely performed at the initiation of DOAC, this study will be helpful for further personalise DOAC therapy. To generalise the results and apply them to clinical settings, the results should be validated in different populations.

Funding

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Conflict of interest

The authors have no conflict of interest to declare.

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ORIGINAL ARTICLE

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Effects of sex on clinical outcomes of hypertrophic cardiomyopathy in Singapore

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ABSTRACT

Introduction: Despite the growing recognition that sex can affect the presentation and outcomes in hypertrophic cardiomyopathy (HCM), this relationship is understudied in Asians. Therefore, we aimed to explore sex differences in Asian patients with HCM.

Method: A total of 295 consecutive patients diagnosed with HCM were recruited from a tertiary cardiology centre from 2010 to 2017 over a mean of 3.9±2.7 years. We evaluated the effects of sex on the outcomes of HCM in Asian patients.

Results: HCM patients were more commonly men (72%). Women were older and had more comorbidities, including hypertension and atrial fibrillation. On transthoracic echocardiography, the indexed left ventricular end-systolic and end-diastolic volumes were similar, but more women had more-than-moderate mitral regurgitation and had a smaller left ventricular outflow tract (LVOT). Women more commonly had findings of obstructive physiology with significant LVOT obstruction, defined as >30 mmHg at rest. The use of implantable cardioverter defibrillators was similar across sexes. On multivariable analysis, women were found to be more likely to develop progressive heart failure requiring admission (hazard ratio [HR] 2.10, 95% confidence interval [CI] 1.05–4.71, *P*=0.021) but had a lower rate of all-cause mortality (HR 0.36, 95% CI 0.19–0.70, *P*=0.003).

Conclusion: Women diagnosed with HCM were older, had more comorbidities and were more likely to develop heart failure while men had a higher risk of all-cause mortality.

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Keywords: atrial fibrillation, cardiology, cardiomyopathy, heart failure, hypertrophic cardiomyopathy, sex

INTRODUCTION

Hypertrophic cardiomyopathy (HCM) is a common cause of inherited cardiomyopathy.[1] The phenotypic expression of HCM is highly diversified with varying extents of myocardial hypertrophy, which can affect different parts of the heart and result in varying extents of left ventricular outflow tract (LVOT) obstruction, diastolic dysfunction and arrhythmic potential. The large spectrum of phenotypic expression accounts for differences in clinical course and risk profile.[2-6]

Sex is known to be associated with differences in the presentation and outcomes in many cardiovascular conditions, including heart failure, valvular heart disease and coronary artery disease.[7-9] There is increasing recognition that sex could have a similar impact in HCM where studies have pointed out that women tend to show poorer outcomes than men.[10-15]

Most studies on this matter involve Western populations. Studies on Asian populations are less common. In this study, we aimed to evaluate sex-specific differences in

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CLINICAL IMPACT

What is New

- Hypertrophic cardiomyopathy (HCM) is more common among men in Singapore.
- Women with HCM have a higher risk of progression to heart failure while men have a higher risk of all-cause mortality.
- Women tend to present later usually because of atypical symptoms (such as lack of chest pain) compared with men and delayed recognition of new symptoms.

Clinical Implications

- Measures for early detection of HCM and disease progression for women and a holistic approach to reduce risk for all-cause mortality for men may improve outcomes of patients with HCM.

the presentations and outcomes of HCM in an Asian cohort.

METHOD

This study comprised 295 consecutive patients diagnosed with HCM who were recruited at National University Heart Centre, Singapore from 2010 to 2017. The subjects were retrospectively identified from a comprehensive echocardiography database. We devised a search strategy for all subjects with reported left ventricular hypertrophy or HCM on the indication or diagnosis of transthoracic echocardiograms performed from 2010 to 2017. Subsequently, chart reviews were conducted to identify patients who were diagnosed with HCM, which yielded the final cohort of 295 subjects. Outcome data were collected for patients up to 31 December 2021, giving a study period from the index echocardiogram till last follow-up. Patients recruited either did not have hypertension or were well controlled. Patients who did not have a clear diagnosis of HCM (e.g. equivocal between hypertensive heart disease or HCM) were excluded. Ethics approval was obtained from the National Healthcare Group Domain Specific Review Board (Reference number 2018/01329).

We collected baseline demographics, clinical data and echocardiographic parameters from the index echocardiogram. Outcome data were obtained by screening the Singapore electronic medical record. Echocardiographic data were interpreted and analysed according to the American Society of Echocardiography. The diagnosis of HCM was reached based on clinical and echocardiographic evaluation in accordance with consensus guidelines, where the presence of myocardial hypertrophy in the absence of local or systemic aetiologies with loading conditions would constitute HCM.[16] All echocardiograms were re-evaluated by 2 independent cardiologists to determine the HCM subtype, and uncertain cases were adjudicated by an independent experienced echocardiologist. The patients were followed for a mean of 3.9±2.7 years. Outcome data were collected until 31 December 2021 based on the review of the hospital medical records. Outcomes collected included first admission for heart failure as well as all-cause mortality. Mortality data in our hospital records are synchronised to the national death registry, such that we were able to capture all mortality events. However, we were unable to ascertain the cause of death if the death occurred outside our hospital.

Continuous variables were expressed as mean (±standard deviation), while categorical variables were expressed as number (proportion). Binary logistic regression was used to establish the association between sex differences and HCM outcomes. All-cause mortality and progression heart failure were assessed using Cox regression analysis. *P* values were 2-sided and deemed significant if <0.05. The statistical analysis was performed using SPSS Statistics version 27 (IBM Corp, Armonk, NY, US).

RESULTS

There was male predominance (n=211, 71.5%). Women were about 9 years older than men (66.6±18.4 years versus 55.6±15.1 years, *P*<0.001) at the time of diagnosis (Fig. 1), and formed most of those diagnosed above the age of 80.

In terms of comorbidities, women were more likely to have atrial fibrillation (25.0% vs 12.3%, *P*=0.008) and hypertension (56.0% vs 41.2%, *P*=0.023). The presence of other comorbidities, including hyperlipidaemia, diabetes mellitus, ischaemic heart disease, chronic kidney disease and peripheral vascular disease, was similar across the sexes. Men were more likely to be smokers than women (4.8% vs 26.1%, *P*<0.001) (Table 1).

On echocardiography, women were more likely to have significant LVOT gradient of >30 mmHg (28.6% vs 11.4%, *P*<0.001) as suggested by the echocardiographic finding of a smaller LVOT diameter (19.1 mm vs 21.5 mm, *P*<0.001). Women also had greater prevalence of more-than-moderate mitral regurgitation (11.9% vs 3.8%, *P*=0.013). While women had smaller left ventricular end-diastolic volumes (LVEDV), there was no significant difference after indexing for body

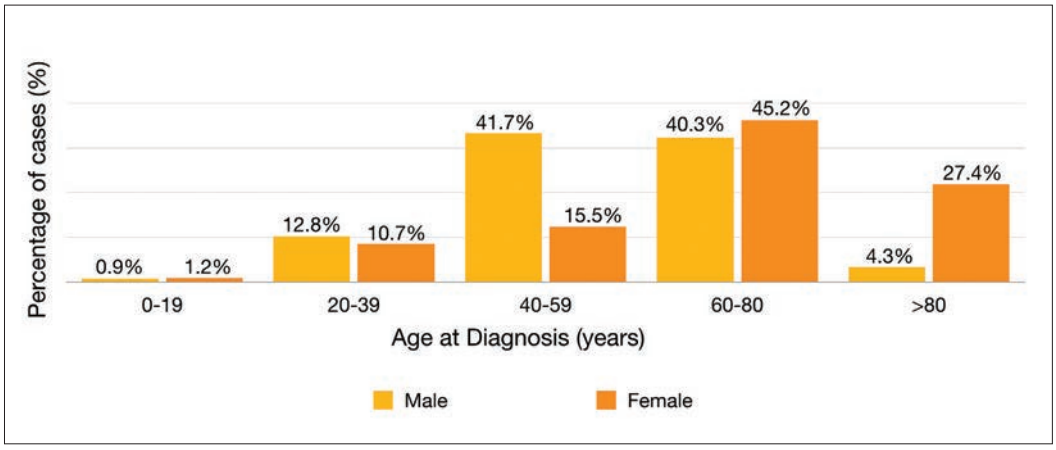

Fig. 1. Distribution of hypertrophic cardiomyopathy patients based on stratified age.

surface area. Women were more likely to have elevated cardiac filling pressures with a greater proportion with E/e' >14 (63.1% vs 38.9%, *P*<0.001) (Table 2). A total of 106 patients had a cardiac magnetic resonance imaging (cardiac MRI) done as part of their evaluation. Among these subjects, 85 had some extent of late gadolinium enhancement (LGE) detected on the cardiac MRI. However, we were unable to quantify the extent of LGE on these scans as quantification of LGE was not routinely performed in our centre during the study period.

At initial presentation, about half of the patients in both men and women's arms were asymptomatic. Most of these patients were screened because of an incidental abnormal electrocardiogram, and diagnosis was then confirmed by echocardiography and clinical review. For symptomatic patients, more women presented with dyspnoea (21.4% vs 9.5%, *P*=0.007) while more men had chest pain (17.9% vs 30.3%, *P*=0.031) (Table 3). There were 17 patients who already had implantable cardioverter defibrillators (ICDs) implanted prior to the index echocardiogram in this study.

In terms of long-term outcomes, women were more likely to progress to heart failure requiring admission (Fig. 2), but had a lower risk of all-cause mortality (Fig. 3). On multivariable Cox regression analysis, these associations persisted after correcting for age, hypertension, diabetes mellitus, left ventricular ejection function, atrial fibrillation and high-risk features such as a maximal wall thickness >30 mm and a significant LVOT gradient of >30 mmHg (hazard ratio [HR] 2.10, 95% confidence interval [CI] 1.05–4.71, *P*=0.021; and HR 0.36, 95% CI 0.19–0.70, *P*=0.003, respectively) (Table 3). The use of implantable cardioverter-defibrillators was similar across both sexes (*P*=0.848). Rates of arrhythmic events, namely, ventricular tachycardia/ventricular fibrillation (VT/VF) events either resuscitated or fatal, and appropriate ICD therapies were low and similar across both sexes.

We stratified the population by both sex and age for further analysis against a composite endpoint of stroke, progression to heart failure and all-cause mortality (Fig. 4). When stratified by age, there was no significant difference in outcomes in younger patients under the age of 50 years. However, women above 50 years experienced poorer overall outcomes (*P*=0.015) than their male counterparts.

DISCUSSION

Our study found that in an Asian cohort, there were more male patients with HCM, while women were older at diagnosis and more commonly displayed an obstructive physiology. Women also had a higher risk of developing progressive heart failure requiring admission. On the other hand, men had a worse prognosis in terms of all-cause mortality.

Our findings that there is male predominance among HCM patients while women tended to be older at the point of diagnosis are consistent with other studies globally.[11–14] However, it is unclear whether there is a significant genotypic or hormonal contribution to such findings. Otherwise, perhaps there are socioeconomic factors at play where a bias in screening or diagnostic strategies could have led to delayed or underdiagnosis in women. Some postulated there may be a genetic modifier that affects the degree and progression of cardiac hypertrophy, which could have contributed to the later onset of clinically manifest disease and thus later presentation in women. Our findings are consistent with literature where women are older and have higher rates

Table 1. Baseline characteristics and circumstances leading to diagnosis.

	Overall (n=295)	Men (n=211)	Women (n=84)	*P* value
Mean age at diagnosis (±SD), years	58.7 (±16.8)	55.6 (±15.1)	66.6 (±18.4)	**<0.001**
Ethnicity, no. (%)				
Chinese	192 (65.1)	134 (53.5)	58 (69.0	0.120
Malay	55 (18.6)	39 (18.5)	16 (19.0)	
Indian	36 (12.2)	30 (14.2)	6 (7.1)	
Others	121 (34.70)	78 (3.38)	4 (4.8)	
Body mass index, mean (±SD), kg/m^2	25.3 (±4.5)	25.6 (±4.3)	24.4 (±4.9)	**0.031**
Systolic blood pressure, mean (±SD), mmHg	127.2 (±20.8)	126.6 (±19.9)	128.5 (±22.9)	0.481
Smoking, no. (%)	59 (20)	55 (26.1)	4 (4.8)	**<0.001**
Past medical history				
Hypertension, no. (%)	134 (45.4)	87 (41.2)	47 (56.0)	**0.023**
Hyperlipidaemia, no. (%)	119 (40.3)	78 (37.0)	41 (48.8)	0.062
Diabetes mellitus, no. (%)	47 (15.9)	34 (16.1)	13 (15.5)	0.893
Ischaemic heart disease, no.(%)	82 (27.8)	58 (27.5)	24 (28.6)	0.851
Chronic kidney disease, no. (%)	3 (1.0)	2 (0.9)	1 (1.2)	0.852
Peripheral vascular disease, no. (%)	2 (0.7)	1 (0.5)	1 (1.2)	0.513
Atrial fibrillation, no. (%)	47 (15.9)	26 (12.3)	21 (25.0)	**0.008**
Reason for evaluation				
Asymptomatic, no. (%)	142 (48.1)	103 (48.8)	39 (46.4)	0.331
Symptomatic, no. (%)	153 (51.9)	108 (51.2)	45 (53.6)	
Clinical symptoms (if any), no. (%)				
Chest pain	79 (26.8)	64 (30.3)	15 (17.9)	**0.031**
Palpitations	42 (14.2)	27 (12.8)	15 (17.9)	0.264
Dyspnoea	38 (12.9)	20 (9.5)	18 (21.4)	**0.007**
Syncope	17 (5.8)	14 (6.6)	3 (3.6)	0.316
Family history, no. (%)				
Family history of HCM	18 (6.1)	13 (6.2)	5 (6.0)	0.946
Family history of SCD	3 (1.0)	1 (0.5)	2 (2.4)	0.185
Medication use, no. (%)				
Antiplatelet	90 (30.5)	67 (31.8)	23 (27.4)	0.462
Oral anticoagulation	58 (19.7)	33 (15.6)	25 (29.8)	**0.006**
Beta blocker	190 (64.4)	136 (64.5)	54 (64.3)	0.978
ACE inhibitor/ARB	82 (27.8)	56 (26.5)	26 (31.0)	0.460
CCB	58 (19.7)	34 (16.1)	24 (28.6)	**0.016**
Diuretics	49 (16.6)	28 (13.3)	21 (25.0)	**0.016**

ACE: angiotensin converting enzyme; ARB: aldosterone receptor blocker; CCB: calcium channel blocker; HCM: hypertrophic cardiomyopathy; SCD: sudden cardiac death; SD: standard deviation
P values in bold are statistically significant.

Table 2. Echocardiographic data.

	Overall (n=295)	Men (n=211)	Women (n=84)	*P* value
LVEF, mean (±SD)	64.8 (±11.6)	64.7 (±10.7)	64.8 (±13.7)	0.984
LVEF <50%, no. (%)	20 (6.8)	12 (5.7)	8 (9.4)	0.242
LVEDV, mean (±SD), mL	93.4 (±34.3)	96.4 (±34.8)	86.1 (±32.0)	**0.021**
LVEDVi, mean (±SD), mL/m^2	53.70 (±18.66)	55.22 (±19.66)	53.10 (±18.28)	0.378
LVESV, mean (±SD), mL	30.0 (±18.2)	30.5 (±19.1)	28.6 (±15.8)	0.404
LVESVi, mean (±SD), mL/m^2	17.20 (±9.95)	18.25 (±10.02)	16.79 (±9.92)	0.258
E/e', mean (±SD)	15.57 (±8.41)	14.51 (±7.56)	18.24 (±9.79)	**0.001**
E/e' >14, no. (%)	135 (45.8)	82 (38.9)	53 (63.1)	**<0.001**
LA diameter, mean (±SD), mm	43.2 (±9.8)	43.1 (±8.5)	43.4 (±12.5)	0.851
LVMI, mean (±SD), g/m^2	138.5 (±46.9)	139.3 (±47.0)	136.4 (±47.0)	0.637
Maximum wall thickness, mean (±SD), mm	20.4 (±4.6)	20.9 (±4.6)	19.4 (±4.4)	**0.010**
Maximum wall thickness >30 mm, no. (%)	8 (2.7)	7 (3.3)	1 (1.2)	0.010
LVOT gradient ≥30 mmHg, no. (%)	48 (16.3)	24 (11.4)	24 (28.6)	**<0.001**
More than moderate MR, no. (%)	18 (6.1)	8 (3.8)	10 (11.9)	**0.013**
Pattern of hypertrophy, no. (%)				
Localised basal hypertrophy	53 (20.0)	36 (17.1)	17 (20.2)	0.342
Reverse curvature septal hypertrophy, no (%)	44 (14.9)	17 (8.1)	9 (10.7)	
Apical	106 (35.9)	75 (35.5)	31 (36.9)	
Concentric	87 (29.5)	61 (28.9)	26 (31.0)	
Mid cavity	1 (0.3)	1 (0.5)	0 (0)	
Others	4 (1.4)	3 (1.4)	1 (1.2)	
Presence of LGE on CMR imaging	85 (28.8)	66 (31.3)	19 (22.6)	0.140

CMR: cardiovascular magnetic resonance; LA: left atrium; LGE: late gadolinium enhancement; LVEDV: left ventricular end-diastolic volume; LVEDVi: indexed left ventricular end-diastolic volume; LVEF: left ventricular ejection fraction; LVESV: left ventricular end systolic volume; LVESVi: indexed left ventricular systolic volume; LVMI: left ventricular mass index; LVOT: left ventricular outflow tract, MR: mitral regurgitation; SD: standard deviation
P values in bold are statistically significant.

Table 3. Eventual outcomes.

Variable	Overall (n=295)	Men (n=211)	Women (n=84)	*P* value
ICD implant, no. (%)	44 (14.9)	32 (15.2)	12 (14.3)	0.848
Resuscitated VT/VF or appropriate ICD therapy, no. (%)	5 (1.7)	5 (2.4)	0	**<0.001**
Stroke, no. (%)	36 (12.2)	20 (9.5)	16 (19.0)	**0.026**
Myocardial infarction, no. (%)	39 (13.2)	27 (12.8)	12 (14.3)	0.733
Heart failure, no. (%)	33 (11.2)	14 (6.6)	19 (22.6)	**<0.001**
Death (all-cause), no. (%)	65 (22.0)	44 (20.9)	21 (25)	0.439
Follow-up duration, mean (±SD), years	3.92 (±2.72)	3.91 (±2.73)	3.92 (±2.71)	0.982

ICD: implantable cardioverter defibrillator; VT/VF: ventricular tachycardia/ventricular fibrillation; SD: standard deviation
P values in bold are statistically significant.

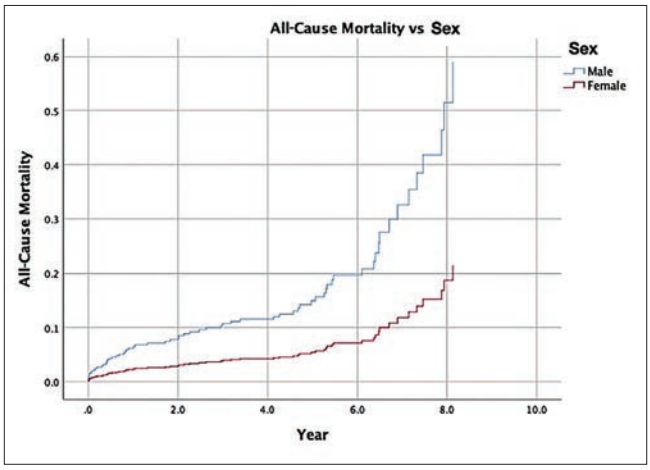

Fig. 2. Cox regression curves for all-cause mortality.
Hazard ratio 0.36, 95% confidence interval 0.19–0.70, *P*=0.003

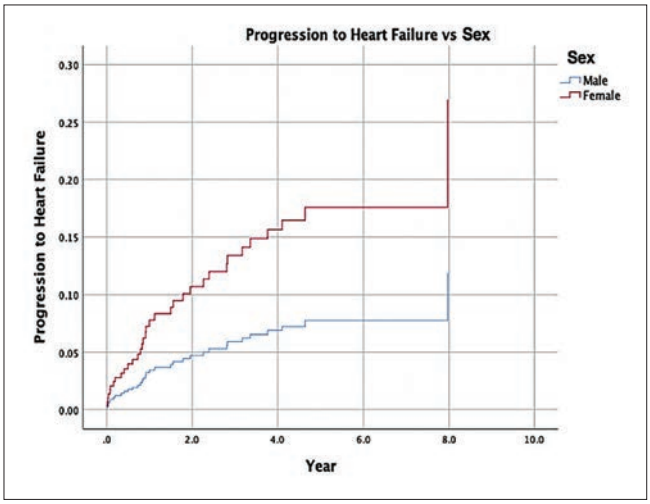

Fig. 3. Cox regression curves for heart failure.
Hazard ratio 2.10, 95% confidence interval 1.05–4.71, *P*=0.021

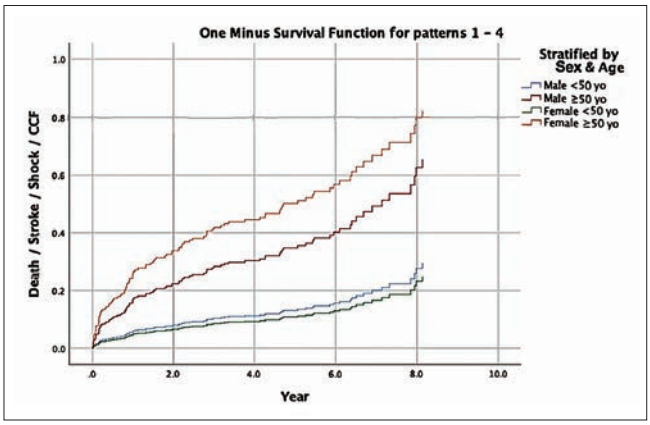

Fig. 4. Progression of hypertrophic cardiomyopathy patients to stroke, congestive heart failure and death as stratified by age and sex.
CCF: congestive cardiac failure; yo: years old

of obstructive physiology. Studies have alluded to the role of oestrogen in modulating myocardial hypertrophy in animal models while others have shown that there are differences in gene expression and therefore phenotypic expression between sexes.[17-20]

Beyond biochemical and physiologic differences between the sexes, there could also be psychosocial differences where women approach their own health and seek medical attention in a manner different from men. Studies have shown that women are often less aware of their risk of developing cardiovascular disease. Sometimes, their physicians are similarly biased and could less frequently explore such conditions with them.[21,22] It is well described that women tend to present later often because of atypical symptoms or delayed recognition and response to new symptoms. Taking ischaemic heart disease for example, women with chest pain have been shown to be more delayed in seeking medical attention, leading to delayed diagnosis and intervention.[23] The Variation in Recovery: Role of Gender on Outcomes of Young AMI Patients study showed that women with eventual ST-elevation myocardial infarction were likely to present without chest pain.[24] This is also seen in our cohort where more men presented with chest pain than women (30.3% vs 17.9%, *P*=0.031).

In terms of eventual outcomes, our study concurs with literature that women with HCM have a higher risk of progression to heart failure. Some have postulated that the development of heart failure in HCM progresses along 3 major pathways, namely, LV systolic dysfunction, LVOT obstruction, and the absence of obstruction with preserved systolic function.[25] First, in terms of systolic function, we found that there was no significant difference in systolic function across the sexes. With regard to the second mechanism, several studies have shown that despite similar degrees of hypertrophy, women were more likely to demonstrate obstructive physiology.[12,13,26,27] We made a similar observation in our experience where women were more likely to have significant obstructive disease with a significant LVOT gradient at rest. However, even after accounting for obstructive physiology in the multivariable analysis, women were still more likely to develop heart failure. This leads us to postulate that heart failure in women would have occurred mostly via the third pathway with diastolic dysfunction. This is supported by the finding where women were more likely to have elevated filling pressures with a higher E/e' and were also more likely to have had prior atrial fibrillation. All of these are risk factors for progressive diastolic dysfunction and would

be in keeping with other heart failure cohorts where females were at higher risk of developing heart failure with preserved ejection fraction (HFpEF).[28]

Another interesting finding is that the risk of progression to heart failure really starts to diverge at older ages. We noticed that when stratified by younger age, there was a large divergence in outcomes in women compared with men above the age of 50 years. One hypothesis is that the female hormonal profile represents a point of difference that leads to this great divergence in outcomes. Postmenopausal endocrine changes in women could also have an important role in the difference of presentations and outcomes. Studies have shown that oestrogen deficiency is associated with impaired ventricular relaxation, myocardial hypertrophy and fibrosis.[29,30] Oestrogen deficiency postmenopause could have contributed to the greater risk of progression to heart failure. Beyond HCM and even within HFpEF, women have also been shown to have greater age-dependent changes in diastolic ventricular function and arterial stiffness, and therefore have a greater risk of progression to heart failure. Nevertheless, further studies focusing on postmenopausal changes and outcomes in women are needed to support this hypothesis.

While women have a higher risk of progressing to heart failure, it was actually men who had a higher risk of all-cause mortality. Unfortunately, as this was a retrospective study, the specific cause of death was often unavailable and we were unable to adjudicate whether this higher rate of mortality was due to cardiovascular causes or related to HCM. This could be studied in future prospective studies.

Limitations

A limitation is that only all-cause mortality was reported, as the specific cause of death was often unavailable. This means that we were unable to adjudicate the rates of cardiovascular mortality, including the risk of sudden cardiac death in the study. Furthermore, genetic testing for HCM was uncommon in our institution and thus this information was not available in our cohort. Finally, as a retrospective single-centre study, our findings are subject to its inherent limitations and we can only establish relation, not causality.

CONCLUSION

Women diagnosed with HCM tended to be older, had more comorbidities and were at higher risk of developing heart failure, while men had a high risk of all-cause mortality.

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Conflict of interest

The authors report no conflict of interest for this study.

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ORIGINAL ARTICLE

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Clinical outcomes of hospitalised individuals with spin-induced exertional rhabdomyolysis

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ABSTRACT

Introduction: Exertional rhabdomyolysis (ER) is caused by myocyte breakdown after strenuous physical activity. In recent years, the incidence of spin-induced ER (SER) has been increasing. We describe the clinical characteristics, management and outcomes of patients admitted for SER.

Method: A review was conducted for all patients admitted to Singapore General Hospital for SER from 1 March 2021 to 31 March 2022. All patients with the admission diagnosis of "rhabdomyolysis", "raised creatine kinase (CK) level", or "elevated CK level" with a preceding history of spin-related physical exertion were included. Patients without a history of exertion, with a history of non-spin related exertion, or with a peak serum CK <1000 U/L were excluded.

Results: There were 93 patients in our final analysis; mean age was 28.6±5.6 years and 66 (71.0%) were female patients. Mean body mass index was 25.0±5.7 kg/m^2; 81 (87.1%) patients were first-time spin participants. All patients had muscle pain, 68 (73.1%) had dark urine, 16 (17.2%) muscle swelling and 14 (15.1%) muscle weakness. There were 80 (86.0%) patients with admission CK of >20,000 U/L. Mean admission creatinine was 59.6±15.6 μmol/L. Mean intravenous (IV) hydration received was 2201±496 mL/day, oral hydration 1217±634 mL/day and total hydration 3417±854 mL/day. There was 1 (1.1%) patient with acute kidney injury, which resolved the next day with IV hydration.

Conclusion: Inpatient management of SER includes laboratory investigations, analgesia and hydration. Risk of complications is low in SER patients. SER patients without risk factors for complications can be considered for hospital-at-home management with bed rest, aggressive hydration and early outpatient review.

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Keywords: emergency medicine, exertional rhabdomyolysis, home care, spinning, sports medicine

INTRODUCTION

Rhabdomyolysis is a clinical and biochemical syndrome caused by the breakdown of myocytes and release of intracellular components into the bloodstream.[1] A subset of rhabdomyolysis is exertional rhabdomyolysis (ER), which is caused by strenuous physical activity. Risk factors for ER include lack of physical endurance, increased duration and intensity of exercise, raised environmental temperature, male sex, genetically inherited metabolic diseases such as sickle cell trait or myopathies, concomitant infection, and consumption of medications or performance enhancing drugs (e.g. non-steroidal anti-inflammatory drugs [NSAIDs], statins, creatine and anabolic steroids).[2-4] ER most commonly presents as muscle pain, swelling, weakness and myoglobinuria.[5] There is no defined creatine kinase (CK) level diagnostic of ER, but a commonly used cut-off is CK >1,000 U/L or 5 times the upper limit of normal in the presence of symptoms.[6-9] Complications of ER include acute kidney injury (AKI), metabolic acidosis, electrolyte abnormalities, arrhythmias, compartment syndrome, disseminated intravascular coagulation and rarely, death.[6]

Spin is a form of high intensity interval training (HIIT) that has garnered widespread popularity worldwide due to its celebrity following, use of loud and fast-paced music, dim lighting with spotlights to encourage participants to exercise as hard as possible, for as long as possible. In the last decade, spin-induced exertional rhabdomyolysis (SER) emerged as a subset

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CLINICAL IMPACT

What is New

- First-time spin participants are more prone to developing spin-induced exertional rhabdomyolysis (SER).
- SER has a low risk of complications.

Clinical Implications

- Spin can lead to rhabdomyolysis.
- Low-risk patients with SER can be considered for hospital-at-home care.

of ER.[10-15] Young and healthy individuals without pre-existing conditions have very low risk of developing complications of ER.[16] The mainstays of treatment for ER are aggressive hydration, correcting electrolyte imbalances and preventing further muscular injury.[17-19] Traditionally, most patients are treated as inpatients with intravenous (IV) hydration and monitoring. This imposes a strain on limited hospital resources.[20]

There is a move in recent years towards delivering hospital care for patients at home for various conditions, including ER.[21,22] Patients receiving hospital-at-home care are reviewed regularly by doctors and other healthcare professionals, who can perform blood tests and administer IV therapy. Ko et al. reported 108 patients who participated in a hospital-at-home programme, among whom 5 had rhabdomyolysis.[22] However, as the intensity of monitoring and access to treatment are limited in a hospital-at-home setting, it is unclear if all SER patients are suitable for hospital-at-home care. Furthermore, apart from a guideline for military recruits,[23] guidance on safely managing ER patients as inpatients or outpatients is lacking.

In this study, we aimed to describe the clinical characteristics, inpatient management and clinical outcomes of patients admitted for SER. We also aim to identify SER patients who may be suitable for hospital-at-home care.

METHOD

Data source and study population

We performed a case note review for all patients admitted to the Department of Internal Medicine, Singapore General Hospital (DIM, SGH) between 1 March 2021 and 31 March 2022. SGH is the largest tertiary hospital in Singapore with a bed capacity of 1750. All patients with the admission diagnosis of "rhabdomyolysis", "raised CK level" or "elevated CK level" with a preceding history of spin-related physical exertion were included. Patients without a history of exertion, or with a history of non-spin related exertion, or with a peak serum CK value of less than 1000 U/L were excluded per CK cut-off for diagnosis of rhabdomyolysis.[6-9]

The following data were collected from patients' electronic medical records: baseline demographics, date and type of physical exertion, past medical history, clinical presentation, serum levels of CK, creatinine, electrolytes, alanine transaminase (ALT), aspartate transaminase (AST), hydration (both IV and oral) received, and analgesia prescribed. For patients who had multiple episodes of physical exertion prior to admission, the date of the first episode of physical exertion was taken as the date of exertion. Due to institutional laboratory limitations, CK values above 20,000 U/L were reported as >20,000 U/L. Other data collected include complications of rhabdomyolysis, length of stay (LOS) and readmissions within 30 days.

Our study was reviewed, and ethical approval was waived by the SingHealth Centralised Institutional Review Board (Reference Number: 2022/2486).

Institutional practice

In our institution, the Emergency Physician decides on the initial diagnostic tests, management, and whether admission is required. In general, all cases of rhabdomyolysis with or without AKI are admitted to the DIM. Treatment with IV fluids and analgesia is started in the Emergency Department (ED). Inpatient management varies as there are no treatment guidelines. Blood tests such as CK levels, creatinine, ALT, AST are performed every 1 to 2 days based on physician discretion. Volume of IV hydration and type of analgesia also varies depending on the managing physician, and is based on clinical findings such as hydration status and symptom improvement.

Statistical analysis

All statistical analysis was conducted using R version 4.1.3 (R Foundation, Vienna, Austria). Descriptive statistics were used for the demographic information and clinical characteristics. Results were presented either as a number and percentage for categorical variables or mean±standard deviation for continuous variables. Pearson's correlation was used to calculate the correlations between ALT/AST and creatinine/LOS. A *P* value of <0.05 was taken to be statistically significant.

RESULTS

There was a total of 101 patients admitted with exertional rhabdomyolysis from 1 March 2021 to 31 March 2022. There were 8 patients who had non-spin-induced exertion prior to presentation and were excluded from statistical analysis (2 induced by gym, 2 running, 1 CrossFit, 1 cycling/weights, 1 sit-ups/weights and 1 push-ups). There were 93 patients who had attended spin class prior to hospital presentation for ER and were included in the final analysis (Fig. 1).

Majority of patients (81, 87.1%) were first-time participants in a spin class. Mean age was 28.6±5.6 years and there were 66 (71.0%) female patients. Mean BMI was 25.0±5.7 kg/m^2. Eleven (11.8%) patients had chronic medical comorbidities (2 patients had hypertension and the other 9 patients had any of the following comorbidities: hyperlipidaemia/asthma/allergic rhinitis/bronchiectasis/epilepsy/mitral valve prolapse/polycystic ovarian syndrome/Sjögren's syndrome/obsessive compulsive disorder/obstructive sleep apnea/gastroesophageal reflux disease/chronic spontaneous urticarial/eczema). Only 1 patient had a history of statin use for hyperlipidaemia. There were no patients with genetically-inherited metabolic diseases, such as sickle cell trait or myopathies. With regard to symptoms, all patients had muscle pain, 68 (73.1%) had dark urine, 16 (17.2%) had muscle swelling and 14 (15.1%) had muscle weakness. Mean days of exertion to presentation to hospital was 3.5±1.2 days (Table 1).

Table 1. Baseline characteristics and clinical presentation of patients admitted for spin-induced exertional rhabdomyolysis.

	Spin-induced ER n=93
Sex, no. (%)	
Male	27 (29.0)
Female	66 (71.0)
Age, mean (SD), years	28.6 (5.6)
BMI, mean (SD), kg/m^2	25.0 (5.7)[a]
Risk factors for ER, no. (%)	
First episode spin	81 (87.1)
Statin use	1 (1.1)
Myopathy	0 (0)
Symptoms, no. (%)	
Muscle pain	93 (100)
Muscle swelling	16 (17.2)
Muscle weakness	14 (15.1)
Dark urine	68 (73.1)
Number of days from exertion to presentation, mean (SD)	3.5 (1.2)

BMI: body mass index; ER: exertional rhabdomyolysis; SD: standard deviation
[a] Data were available for 66 patients for BMI.

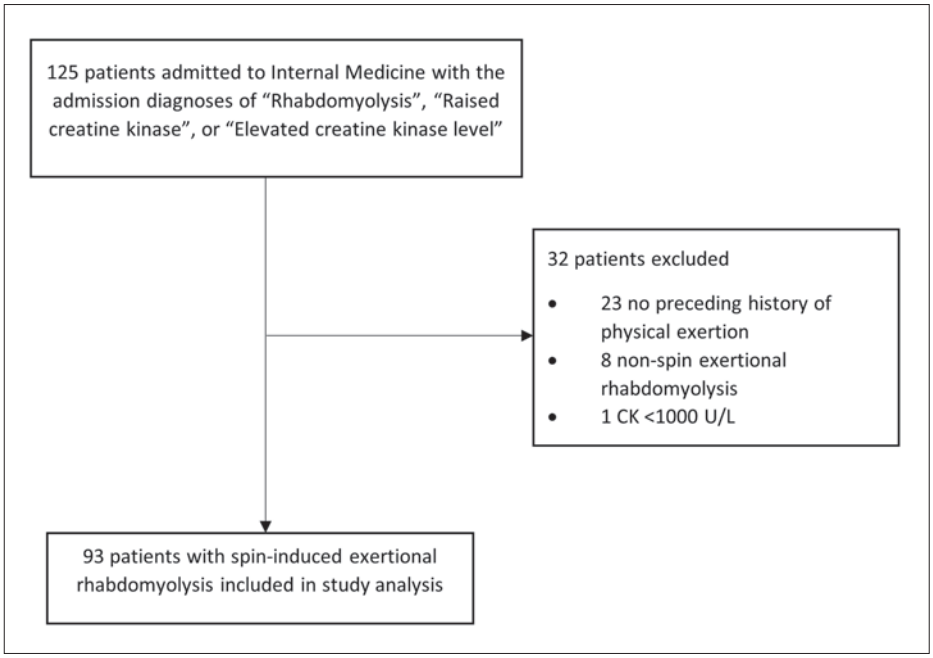

Fig. 1. Flowchart of study design.
CK: creatine kinase

There were 80 (86.0%) patients with an admission CK of >20,000 U/L. Patients with admission CK >20,000 U/L required a mean of 7.9±1.3 days after exertion for CK values to decrease to <20,000 U/L. For patients who had a quantifiable CK <20,000 U/L on admission, a mean of 2.3±1.1 days was needed to achieve a downward trend. There were 83 (89.2%) patients with peak CK >20,000 U/L. With regard to discharge CK, 17 (18.3%) patients discharged with CK >20,000 U/L, without a measurable downtrend of CK due to laboratory limitations (Table 2). Among them, 13 (76.5%) presented for follow-up appointments and all showed a downtrend in CK. Overall mean LOS was 5.5±1.8 days (Table 2). Mean LOS was longer for patients with admission CK >20k (5.74±1.71 vs 4.15±1.77 days, *P*=0.01). Patients who were discharged with CK >20,000 U/L had a shorter LOS (4.12±1.17 days) than those who were discharged with CK <20,000 U/L (5.83±1.77 days, *P*<0.01). There was no significant difference in the incidence of complications like AKI (*P*=1) and electrolyte abnormalities (*P*=0.40) in those were discharged with CK >20,000 U/L and <20,000 U/L. No patient was discharged with a documented uptrend in CK.

Mean initial creatinine was 59.6±15.6 μmol/L, mean peak creatinine was 60.5±15.8 μmol/L, and mean discharge creatinine was 51.8±14.1 μmol/L. Liver function test was done for 88 (94.6%) patients. Mean initial ALT and AST were 191.3±128.2 U/L and 806.6±608.5 U/L, respectively. Mean peak ALT and AST were 270.0±164.2 U/L and 1002.1±680.5 U/L, respectively. Mean ALT and AST on discharge were 219.5±138.2 U/L and 444.9±406.2 U/L, respectively (Table 2). Hepatitis and human immunodeficiency virus (HIV) screens were performed for 4 patients with raised liver enzymes and were negative. Abdominal ultrasound scans were performed for 2 patients, of which 1 scan was normal and the other showed hepatic steatosis. Neither admission ALT nor AST were significantly correlated to admission creatinine and LOS (admission ALT versus admission creatinine: *r*=-0.02, *P*=0.85, 95% confidence interval (CI) -0.23 – 0.19; admission ALT vs LOS: *r*=0.06, *P*=0.57, 95% CI -0.15 – 0.27; admission AST vs admission creatinine: *r*=0.00, *P*=1.00, 95% CI -0.21 – 0.21; admission AST vs LOS: *r*=0.13, *P*=0.23, 95% CI -0.08 – 0.33).

Mean IV hydration received was 2201±496 mL/day, mean oral hydration achieved by patients was 1217±634 mL/day, and mean total hydration was 3417±854 mL/day. The maximum volume of IV hydration received per day was 4250 mL. All patients received IV hydration up until at least the day before discharge.

Table 2. Laboratory investigations of patients admitted for spin-induced exertional rhabdomyolysis.

	Spin-induced ER n=93
Creatine kinase (CK)	
Admission CK >20,000 U/L, no. (%)	80 (86.0)
Peak CK >20,000 U/L, no. (%)	83 (89.2)
Discharge CK >20,000 U/L, no. (%)	17 (18.3)
No. of days from exertion to CK <20,000 U/L, mean (SD)[a]	7.9 (1.3)
Creatinine, mean (SD), μmol/L	
Admission	59.6 (15.6)
Peak	60.5 (15.8)
Discharge	51.8 (14.1)
ALT, mean (SD), U/L	
Admission	191.3 (128.2)
Peak	270.0 (164.2)
Discharge	219.5 (138.2)
AST, mean (SD), U/L	
Admission	806.6 (608.5)
Peak	1002.1 (680.5)
Discharge	444.9 (406.2)
Length of stay, mean (SD), days	5.5 (1.8)

ALT: alanine transaminase, AST: aspartate transaminase; ER: exertional rhabdomyolysis; SD: standard deviation
[a] For patients with admission CK >20,000 U/L and discharge CK <20,000 U/L.

Types of IV fluids prescribed for hydration included sodium chloride solution, Plasma-Lyte-A (Baxter International), and Hartmann's solution. None of the patients were given sodium bicarbonate to alkalinise the urine. With regard to analgesia, 80 (86.0%) patients received paracetamol or paracetamol-orphenadrine, 11 (11.8%) patients received systemic NSAIDs, 31 (33.3%) patients received topical NSAIDs (ketoprofen patch) and 36 (38.7%) patients received weak opioids (codeine or tramadol) (Table 3).

With regard to complications, 1 (1.1%) patient had a disease-related complication and developed AKI. The patient with AKI was a 21-year-old man with an admission creatinine of 127 mmol/L and received IV hydration of 2400 mL within 24 hours of admission. Repeat creatinine the next day was normal at 100 mmol/L. One (1.1%) patient had a treatment-related complication and developed thrombophlebitis (Table 3).

Table 3. Treatment and complications of spin-induced exertional rhabdomyolysis patients.

	Spin-induced ER n=93
Hydration, mean (SD), mL/day	
Intravenous	2201 (496)
Oral	1217 (634)
Total	3417 (854)
Analgesia, no. (%)	
Paracetamol/paracetamol-orphenadrine	80 (86.0)
Topical NSAIDs	31 (33.3)
Systemic NSAIDs	11 (11.8)
Weak opioids	36 (38.7)
Complications, no. (%)	
Acute kidney injury	1 (1.1)
Electrolyte abnormalities[a]	32 (34.4)

ER: exertional rhabdomyolysis; NSAIDs: non-steroidal anti-inflammatory drugs; SD: standard deviation
[a] Metabolic acidosis (bicarbonate <19 mmol/L): 3 patients, metabolic alkalosis (bicarbonate >29 mmol/L): 15 patients, hypokalaemia: 4 patients, hyperkalaemia: 1 patient, hypomagnesaemia: 2 patients, hypermagnesaemia: 1 patient, hyperphosphataemia: 2 patients, metabolic alkalosis and hypokalaemia: 3 patients, metabolic alkalosis and hyperphosphataemia: 1 patient.

Electrolyte levels for calcium, magnesium and phosphate were tested for 41 (44.1%) patients. Electrolyte and metabolic abnormalities were detected: 3 (3.2%) patients with metabolic acidosis (bicarbonate <19 mmol/L), 15 (16.1%) with metabolic alkalosis (bicarbonate >29 mmol/L), 4 (4.3%) with hypokalaemia, 1 (1.1%) with hyperkalaemia, 2 (2.2%) with hypomagnesaemia, 1 (1.0%) with hypermagnesaemia, 2 (2.2%) with hyperphosphataemia, 3 (3.2%) with metabolic alkalosis and hypokalaemia, and 1 (1.1%) with metabolic alkalosis and hyperphosphataemia. A total of 7 (18.1%) patients received oral treatment and no patients required IV therapy to correct electrolyte abnormalities. There were no other complications, such as the need for renal replacement therapy, disseminated intravascular coagulopathy or compartment syndrome. There were no readmissions to our group of healthcare institutions within 30 days (Table 3).

DISCUSSION

The majority of patients in our study were young adult women. There were no patients with pre-existing myopathies or sickle cell disease, which reflects the low incidence of these conditions in Singapore. The majority of SER (81, 87.1%) developed in patients who attended their first spin class. Common presenting symptoms were muscle pain (93, 100%), dark urine (68, 73.1%), muscle swelling (16, 17.2%) and muscle weakness (14, 15.1%). There were 80 (86.0%) patients with an admission CK of >20,000 U/L and mean LOS was 5.5±1.8 days. Only 1 patient had mild AKI which resolved with IV hydration. There were few complications, no patients required dialysis, no inpatient referrals made to other specialties for reasons related to ER and no readmissions within 30 days. Increased awareness of SER in spin participants and instructors is important so that they can recognise symptoms and seek treatment early. First-time spin participants should attend a beginner class and start at a lower intensity to reduce the risk of SER.

Several studies have suggested CK >20,000 U/L as the cut-off for inpatient management.[23,24] There were 86.0% of patients in our study who had CK >20,000 U/L at presentation, and did not develop complications after adequate IV hydration. There were 89.2% of patients who had a mean peak CK >20,000 U/L. Quantifying the exact value of CK would allow more research to be done to adjust the thresholds for admission and increased risk of developing complications. In addition, when assessing the necessity for inpatient management, the number of days to presentation needs to be taken into consideration as CK levels may take up to 7 days to peak depending on the type of exercise.[25-27] The mean number of days required for CK to fall below 20,000 U/L was 7.9±1.3 days post-exertion; 17 (18.3%) patients were discharged with CK >20,000 U/L and all were given up a follow-up appointment within 1 month. Of the 13 patients who attended their follow-up appointment, all had a downward trend in CK. CK and myoglobin levels individually have been shown to have no correlation with AKI or mortality,[28,29] although higher admission CK levels are associated with a longer LOS (P<0.01).[30]

Hence, persistently high CK levels should not be a contraindication to patients' discharge as long as the patient has improved symptomatically and does not have any complications. Test for CK levels are often repeated daily and this practice should be reevaluated with consideration given to the time taken for levels to fall. However, CK levels should not be the main factor determining the patient's suitability for discharge. In this study, other criteria used for discharge included improvement of symptoms, ambulatory status and absence of complications. Other factors that may be considered include severity of symptoms, ability to tolerate oral hydration, the availability of a caregiver (if necessary) and the accessibility of medical care.

Among the 41 patients whose additional electrolytes such as serum calcium, magnesium and phosphate were checked, 6 patients were found to have mild electrolyte abnormalities and 2 with hypomagnesaemia were treated with oral therapy. For patients with liver function tests performed, there was no correlation with outcomes (either creatinine levels or LOS). Average peak ALT and AST were 270.0±164.2 U/L and 1002.1±680.5 U/L, respectively. Some patients were subjected to additional investigations in the absence of abdominal signs or symptoms, such as hepatitis/HIV screens (4 patients), and abdominal ultrasound scans (2 patients) which returned negative. Furthermore, the mean discharge ALT (219.5±138.2 U/L) was higher than admission ALT (191.3±128.2 U/L). The utility of performing tests for electrolytes levels (such as calcium, magnesium, phosphate) and liver function is unclear. Any abnormalities should be treated conservatively and further tests are only indicated if the clinical or biochemical picture suggests concomitant pathologies. Further investigation for metabolic myopathies should be considered in patients with recurrent ER, persistently high CK beyond 8 weeks, positive family history of rhabdomyolysis, exertion-induced muscle cramps or intolerance to exercise.[31]

Mean IV hydration received by patients was 2201±496 mL/day and mean oral hydration 1217±634 mL/day. All patients were kept on IV hydration at least until the day before discharge. Studies report fluid hydration for ER commencing at rates of 200–1000 mL/h, and subsequently reduced to a maintenance rate of 120–300 mL/h guided by a urine output target of 300 mL/h.[18,23,32] Type of fluid and addition of electrolytes or mannitol has not been shown to affect LOS or demonstrate clear utility, respectively.[18,23,32] While the SER patients in our study received a lower volume of hydration than suggested in literature, no patient developed AKI during admission, and the creatinine levels of the only patient with AKI normalised within 24 hours. More studies are needed to determine if SER patients at low risk of complications and who can tolerate large volumes of oral hydration are able to be managed with oral hydration alone.

The use of NSAIDs for pain control is not recommended in the management of ER due to the risk of AKI.[33] Despite this, 11 (11.8%) of patients were prescribed NSAIDS. More should be done to increase awareness among physicians regarding the preferred class of analgesia to avoid medication-related AKI. While the patients prescribed NSAIDS in our study did not develop AKI, several case reports have shown such patients developing AKI.[34-36]

Our patient who had AKI showed rapid resolution by the second day of admission after receiving IV hydration and did not require renal replacement therapy. Among SER patients, a systematic review found that 7.2% and 4.1% of patients developed AKI and compartment syndrome, respectively.[37] Patients with AKI were older, had elevated creatinine on arrival, presented later to the hospital or had a history of NSAID use.[34,38,39] The ER-related complication rate found in our study is much lower at 1.1%. The higher complication rate in previous studies may be contributed by publication bias, where the more severe cases of SER or SER with complications are more likely to be reported. Singapore studies have also found few cases of AKI in patients with SER, which is similar to our results.[10,14,40] Due to the increased incidence of SER, there have also been news articles and online educational materials to increase public awareness of SER in Singapore. Increased awareness of SER is important to allow for timely intervention and prevention of complications. Hence, patients with normal creatinine on admission, or mildly elevated creatinine that normalises, all remain potential candidates for hospital-at-home care.

Our institution is running a pilot hospital-at-home care programme as an alternative to in-hospital management. In this programme, selected patients are discharged from the inpatient ward but continue to receive hospital care at home. These patients would be reviewed daily by a doctor and nurse, either physically or via teleconsultation, depending on clinical condition. During physical visits, IV medications and IV hydration can be given, and multiple visits per day can be arranged. Blood tests can be done daily, and results are obtained within a few hours of sample collection. In the event that complications occur during hospital-at-home care, there are channels for patients to contact the healthcare team for assessment and transfer to hospital if required.

ER patients have been treated under hospital-at-home programmes and as outpatients in small case series and did not develop any complications.[22,24] The low complication rate in our study further supports that SER patients who require admission can be considered for hospital-at-home management. They should be able to ambulate, do not have severe AKI or have worsening AKI despite initial hydration, do not have severe electrolyte abnormalities, and have not developed complications related to ER. Patients with underlying medical comorbidities, such as congestive cardiac failure, liver or renal impairment, or syndrome of inappropriate secretion of anti-diuretic hormone, which limit their hydration should be excluded from hospital-at-home management and admitted for in-hospital management instead.

Our study has a few limitations. First this is a single-centre study and our study population is limited to patients who were admitted to the DIM service. It does not capture patients who were either discharged directly from the ED or admitted to another service. These numbers are likely to be small as the current practice in our institution is for rhabdomyolysis patients to be admitted to DIM. Second our institution's laboratory did not routinely quantify CK levels above 20,000 U/L unless specifically requested by the patient's managing physician. Hence, the exact CK values on admission and at peak for majority of our patients are unknown, and the mean peak CK values reported in our study are underestimations. In addition, charting of the volumes of IV and oral hydration commences after patients are admitted to an inpatient ward. Data on hydration received while patients are still in the ED waiting for an inpatient bed were not captured, leading to an underestimation of hydration volumes on day 1 of admission. Readmissions to healthcare institutions outside of our healthcare cluster were not captured in our data. Data collection coincided with the period of COVID-19 pandemic where indoor gyms were closed or operated at a lower capacity due to Singapore's prevailing social distancing measures, likely resulting in fewer people attending spin class than usual. Thus, the reported incidence of SER is likely to be an underestimate.

There are several potential areas for future research, such as the admission criteria for inpatient care of SER, relative CK value for which discharge is safe, and optimal amount of intravenous and/or oral hydration. More prospective studies are also required to determine patient suitability, and feasibility, safety and outcomes of the management of SER patients in the hospital-at-home setting.

CONCLUSION

Inpatient care of SER patients consists mainly of basic laboratory investigations, oral analgesia and hydration. As the risk of complications is low, majority of SER patients without risk factors for complications can be considered for hospital-at-home care. More can be done to increase public awareness of SER to reduce the incidence and healthcare burden of this preventable condition.

Disclosure

The authors report no conflict of interest or relationships with industry.

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REVIEW ARTICLE

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Molecular testing in non-small cell lung cancer: A consensus recommendation

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ABSTRACT

Introduction: Lung cancer remains an important cause of cancer-related mortality in Singapore, with a greater proportion of non-smokers diagnosed with non-small cell lung cancer (NSCLC) in the past 2 decades. The higher prevalence of targetable genomic alterations in lung cancer diagnosed in Singapore compared with countries in the West, as well as the expanding therapeutic landscape for NSCLC in the era of precision medicine, are both factors that underscore the importance of efficient and effective molecular profiling.

Method: This article provides consensus recommendations for biomarker testing for early-stage to advanced NSCLC. These recommendations are made from a multidisciplinary group of lung cancer experts in Singapore with the aim of improving patient care and long-term outcomes.

Results: The recommendations address the considerations in both the advanced and early-stage settings, and take into account challenges in the implementation of biomarker testing as well as the limitations of available data. Biomarker testing for both tumour tissue and liquid biopsy are discussed.

Conclusion: This consensus statement discusses the approaches and challenges of integrating molecular testing into clinical practice for patients with early- to late-stage NSCLC, and provides practical recommendations for biomarker testing for NSCLC patients in Singapore.

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Keywords: non-small cell lung cancer, biomarkers, adenocarcinoma, liquid biopsy, molecular testing

INTRODUCTION

Lung cancer is a leading cause of cancer mortality worldwide, with an estimated 2.21 million new cases and 1.80 million deaths in 2020.[1] In Singapore, lung cancer is the third most frequent cancer in men and women, and accounts for the highest and third highest number of cancer deaths among men and women, respectively.[2]

In the last 2 decades, therapies that target sensitising driver mutations such as epidermal growth factor receptor (*EGFR*) and anaplastic lymphoma kinase (*ALK*) tyrosine kinase inhibitors (TKIs) have transformed the treatment landscape for lung cancer, with the list of targetable driver mutations and therapies still expanding today. Immune checkpoint inhibitors (ICI) have also significantly improved survival outcomes in patients with non-small cell lung cancer (NSCLC) through invigorating the adaptive immune system to eliminate cancer cells.

With this backdrop, these guidelines aim to provide Singapore medical practitioners who diagnose and treat NSCLC patients with evidence-based, standardised pathways for molecular testing at diagnosis and progression. It is hoped that this would (1) streamline the workflow between cancer diagnosis, molecular testing, and treatment, and (2) enable practitioners to offer patients the most effective available treatment.

METHOD

The guideline development group included practitioners from Singapore's public and private sectors who are involved in the diagnosis and management of NSCLC

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CLINICAL IMPACT

What is New

- To our knowledge, this is the first consensus recommendation developed in Singapore for biomarker testing in early to advanced NSCLC.

Clinical Implications

- Patients with advanced, non-squamous NSCLC should have upfront testing done for at least *EGFR* mutation, *ALK* rearrangement and PD-L1.
- Plasma ctDNA testing should not be used in lieu of tissue diagnosis unless sampling is inadequate or unfeasible.
- Tumour biopsy is preferred in EGFR TKI resistance. Plasma ctDNA testing is an acceptable alternative.
- PD-L1 testing should be considered in stage I–III NSCLC, as well as *EGFR* testing in non-squamous histologies.

patients, namely, respiratory physicians, cardiothoracic surgeons, medical and radiation oncologists, and pathologists. A list of proposed statements was developed and each statement was deliberated upon at a consensus meeting organised by the Lung Cancer Consortium Singapore and held virtually on 11 February 2022. For each consensus recommendation statement, criteria for levels of evidence and grade of recommendation were adapted from European Society for Medical Oncology (ESMO) Standard Operating Procedures for Clinical Practice Guidelines[3] and collectively determined by the authors in the context of an Asian clinical landscape.

Tissue and liquid biopsy considerations

Tissue biopsy sample type and minimum material requirements

Methods of obtaining tumour material for microscopic evaluation and biomarker testing include transthoracic needle biopsy, bronchoscopy, thoracoscopy, mediastinoscopy and surgery. Minimally invasive sampling methods are favoured as they carry lower risks of complications; therefore, many tissue specimens are small samples that contain limited tumour material. Formalin-fixed, paraffin-embedded material resulting from routine laboratory processing is commonly used for diagnosis and for biomarker testing including immunohistochemistry (IHC), fluorescence in-situ hybridisation (FISH), and molecular (including next-generation sequencing, or NGS) panels.

Prior to molecular testing, tissue specimens are evaluated for tumour cellularity (TC), defined by tumour cells as a percentage of total number of nucleated cells, with minimum TC cut-offs depending on the assay. The College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology (CAP/IASLC/AMP) 2018 guidelines recommend use of assays that can detect molecular alterations in specimens with as little as 20% TC.[4] For programmed death-ligand 1 (PD-L1) IHC and FISH testing, minimum numbers of tumour cells are considered instead.[5-7] Other technical considerations, including fixative, duration of fixation, and age of the paraffin block or sections, also come into play.[4,8]

Plasma-based testing methodology and limitations

The role of liquid biopsy using plasma circulating tumour DNA (ctDNA) is an area of considerable interest, as it enables biomarker testing with less risk and a faster turnaround time compared to tissue biopsy. Limitations include false negative results due to low tumour cell content as a proportion of the total cellularity, absence of shedding, and the short half-life of ctDNA.[9]

Challenges and barriers to biomarker testing

Use of targeted therapies for NSCLC is contingent on the accurate identification of driver mutations through molecular testing. Engagement with key stakeholders in Singapore revealed various challenges when adopting NGS panel testing, including costs to patients, long turnaround times of 10 working days on average delaying commencement of therapy, lack of local guidelines, insufficient education of both patients and the clinical community, and lack of conclusive cost-benefit studies.[10] Cost was the most cited concern, as NGS panel testing ranges approximately SGD1500–5000 (USD1130–3760). Given the cap of SGD600 (USD450) per year that can be offset by the national medical savings scheme (i.e. MediSave) for cancer diagnostics, a significant out-of-pocket cost remains for most patients with newly diagnosed NSCLC. It is hoped that this guideline will pave the way for affordable reflex testing pathways that streamline turnaround times.

Treatment-naive, advanced NSCLC

Current and emerging biomarkers for NSCLC

NSCLC is characterised by molecularly defined subsets, several of which are therapeutically tractable. The identification of activating mutations in the tyrosine kinase domain of the *EGFR* gene with associated tumour

response to *EGFR*-directed targeted therapy[11,12] led to a paradigm shift in NSCLC treatment. Since then, the discovery of many other oncogenic alterations has followed, several with approved targeted therapies, including ROS proto-oncogene 1 (*ROS1*), Kirsten rat sarcoma viral oncogene homolog (*KRAS*) G12C, B-Raf proto-oncogene (*BRAF*) V600E, rearranged during transfection (*RET*), hepatocyte growth factor receptor (*MET*) exon 14 (*MET*ex14) skipping, Erb-B2 receptor tyrosine kinase 2 (*ERBB2*) and neurotrophic tyrosine receptor kinase (*NTRK*).

There is consensus across international guidelines such as the ESMO[13] and National Comprehensive Cancer Network (NCCN)[14] that molecular subtyping is imperative for therapeutic decision-making and should be performed prior to treatment initiation, where possible. Prevalence of oncogenic driver, access to both testing platforms and suitable treatments are factors for consideration. Both ESMO and NCCN recommend systematic testing of *EGFR*, *ALK*, *ROS1*, *BRAF* and *NTRK* in advanced NSCLC as these have approved first-line targeted therapies. PD-L1 expression with tumour proportion score (TPS) should also be evaluated for advanced NSCLC, as the level of PD-L1 expression is an important factor that influences the choice of front-line treatment in NSCLC without driver mutations.[15,16]

Additional molecular testing for *KRAS*, *MET*ex14 skipping mutation, *ERBB2* and *RET* are recommended by the NCCN and IASLC given recent approvals. Emerging biomarkers such as *MET* amplifications and *NRG1* rearrangements are also of increasing interest with novel therapies in development, though these are not routinely performed in clinical practice. The role of tumour mutational burden (TMB) in predicting response to immune checkpoint inhibition is also unclear, with inconsistent results in terms of survival[17,18] and thus is not currently incorporated into routine practice.

Multiplex versus single testing strategies

Given the expanding list of therapeutically relevant biomarkers and approved targeted therapies, molecular profiling at diagnosis is paramount. While single biomarker assays such as polymerase chain reaction, Sanger sequencing, IHC and FISH have traditionally been implemented due to accessibility and cost, multiplexed NGS assays are now increasingly utilised in practice to evaluate multiple targetable alterations in small tissue biopsy specimens with excellent sensitivity and specificity.[19,20] Regardless, the optimal approach in an Asian population with a high prevalence of *EGFR*-mutated NSCLC remains unclear from a cost-effectiveness perspective, with multiplex and sequential testing both showing merit in different healthcare systems.[21-24] Adding to the complexity is the lack of established gold standard methods of detection for emerging biomarkers, the adoption of panel-based NGS assays as companion diagnostics for many recently approved targeted drugs, as well as the practical issues of accessibility to testing platform and cost of therapy.

In an Asian population like Singapore's, where there is a high prevalence of targetable alterations, upfront molecular testing for at least *EGFR* mutation, *ALK* rearrangement and PD-L1 should be done on tissue biopsy or cytology material for advanced stage non-squamous NSCLC; and results should be available prior to treatment initiation. Where there is access to relevant targeted therapies, multiplex testing including *ROS1*, *KRAS* G12C, *BRAF* V600E, *MET*ex14 skipping, *RET*, *ERBB2* and *NTRK* should also be considered where available.

Practical considerations in molecular testing

Turnaround time is an important consideration in the implementation of molecular testing as it could lead to delays in initiation of optimal therapy. While molecular testing should ideally be conducted at the same institution where histological diagnosis is made to optimise turnaround time, feasibility of on-site testing is influenced by adequate caseload and assay complexity, and thus may not be available at every centre. The Asian Expert Consensus Recommendations on Biomarker Testing in Metastatic and Nonmetastatic NSCLC recommend a turnaround time of 2 weeks or less from time of sample receipt to result reporting.[25]

Another strategy to optimise turnaround time is reflex testing, where molecular testing is initiated by pathologists upon histological diagnosis of non-squamous NSCLC. This has been shown to optimise tissue utilisation and improve profiling rates[26] over on-demand testing, which is ordered by the treating physician after review of histology results. Importantly, reflex testing is contingent on pathologist access to relevant clinical information as overall costs could increase if molecular profiling is ordered inappropriately, underscoring the need for good communication in a multidisciplinary team and adequate patient counselling at point of biopsy when feasible.

Role of plasma-based tests in treatment-naive, advanced NSCLC

Tumour tissue genotyping and plasma ctDNA analysis are both valuable tools in the diagnostic work up of advanced NSCLC. There has been increasing uptake of the latter given the growing list targetable molecular

alterations and the inherent issues of tissue biopsies including yield and attrition.[27,28] Notably, studies have demonstrated ctDNA testing to generally have very high specificity, but significantly compromised sensitivity, with approximately 30% false negative rate depending on platform used.[14] The decision to adopt either testing strategy, be it concurrently or sequentially, should be individualised to the clinical context, methodologies available, and expected results.[29] In particular, the use of ctDNA testing may be considered in specific clinical scenarios, most notably if the patient is medically unsuitable for invasive tissue sampling, or if there is insufficient material for molecular testing following the pathologic confirmation of a NSCLC diagnosis.

Recommendations for molecular testing in treatment-naive, advanced NSCLC (Fig. 1)

- All patients with advanced, non-squamous NSCLC should have upfront testing done for at least *EGFR* mutation, *ALK* rearrangement and PD-L1 done on tissue biopsy or cytology material. This should also be performed for non-smokers with advanced squamous NSCLC [I, A].
- Upfront or sequential multiplex testing including *ROS1*, *KRAS* G12C, *BRAF* V600E, *MET*ex14 skipping, *RET*, *ERBB2* and *NTRK* should also be considered where available, especially if there is access to relevant targeted therapies [II, A].
- Molecular profiling results should be available prior to treatment initiation where possible [V, A].
- Sequential single gene testing strategies are not preferred [II, A].
- Reflex molecular testing is preferred on histological diagnosis of advanced non-squamous NSCLC [V, C].
- Plasma ctDNA testing should not be used in lieu of a histological tissue diagnosis of NSCLC [V, C]. Plasma ctDNA can be considered for the genotyping of newly diagnosed advanced NSCLC where tissue sampling is not feasible or insufficient for molecular analysis [V, C].
- Tumour mutational burden (TMB) does not have an established role in routine clinical practice [II, C].

Advanced EGFR-mutated NSCLC with disease progression on tyrosine kinase inhibitor

EGFR tyrosine kinase inhibitors (TKIs) are the standard f irst line treatment for metastatic NSCLC harbouring sensitising *EGFR* mutations—most commonly exon 19 deletion and exon 21 L858R mutations. However, patients invariably develop resistance to *EGFR* TKIs, through secondary mutations in *EGFR*, phenotypic transformation, or the activation of alternative pathways.[30]

After the first- or second-generation TKI treatments, 50% of patients develop *EGFR* exon 20 T790M mutations, while other resistance mechanisms include

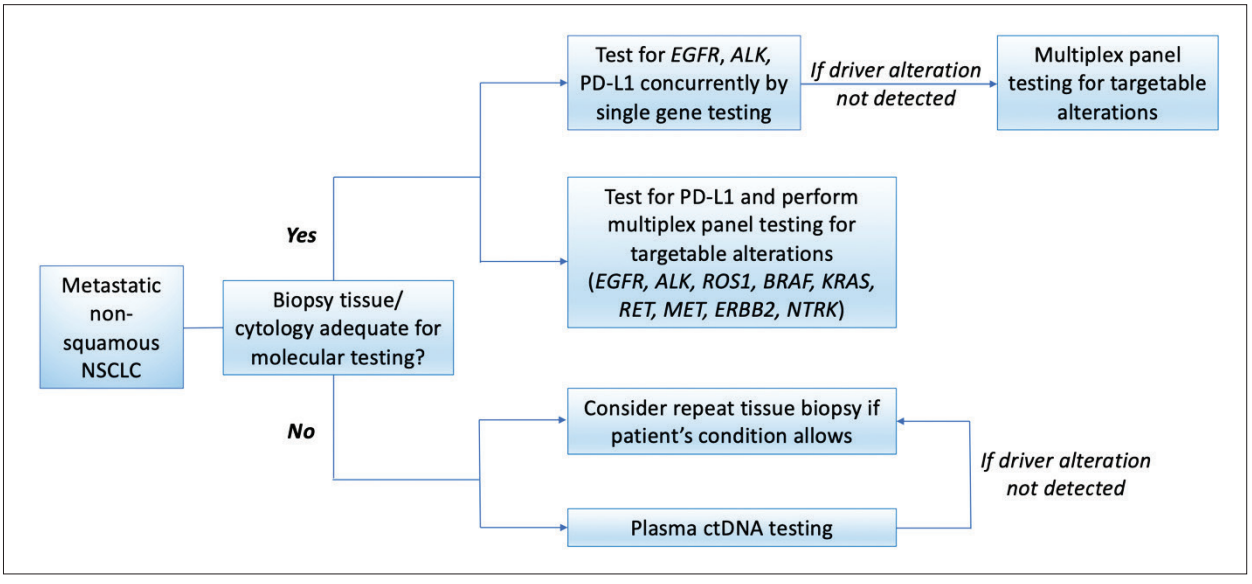

Fig. 1. Biomarker testing recommendations for metastatic non-squamous non-small cell lung cancer (NSCLC).
ALK: anaplastic lymphoma kinase; BRAF: B-Raf proto-oncogene; ctDNA: circulating tumour DNA; EGFR: epidermal growth factor receptor; ERBB2: Erb-B2 receptor tyrosine kinase 2; KRAS: Kirsten rat sarcoma viral oncogene homolog; MET: hepatocyte growth factor receptor; NTRK: neurotrophic tyrosine receptor kinase; PD-L1: programmed death-ligand 1; RET: rearranged during transfection; ROS1: ROS proto-oncogene 1

HER2 amplification (10–15%), small cell transformation (10%), *MET* amplification (5%), *KRAS* or *BRAF* mutations (approximately 1% each).[31] The mechanism of resistance has treatment implications, particularly in that patients harbouring T790M mutations benefit from treatment from third-generation *EGFR* TKI osimertinib, based on the results of the AURA3 study.[32] Recent years have seen the approval of multiple targeted agents against off-target resistance mutations, with many others in early phases of development.

On progression on first- or second-generation *EGFR* TKI, T790M testing should be done as a minimum, in view of the high proportion of T790M resistance mutations and the availability of effective agents. With regard to modality of T790M testing, tissue biopsy remains the gold standard, allowing concurrent evaluation for histologic transformation to small cell. However, this is not always possible due to risks or feasibility of obtaining tissue. ctDNA testing for plasma *EGFR* analysis of T790M is a reasonable alternative, with a positive predictive value of 0.85 and a negative predictive value of 0.60 compared to tissue testing.[33] Studies have shown that patients who are negative for tissue T790M but positive for plasma ctDNA T790M were confirmed to be T790M positive by NGS and had clinical benefit from osimertinib.[34] Based on this, in patients who are positive for plasma T790M, it would not be necessary to pursue a tissue biopsy as they may be treated with osimertinib; in patients who are negative for plasma T790M, we would recommend further tissue biopsy to confirm T790M status and rule out false negatives.

NGS can also be considered, either sequentially f ollowing a negative T790M result, or as an alternative to single gene testing. Use of upfront NGS allows optimal tissue utilisation, and the opportunity to pick up alternative driver mutations, but is limited by cost and turnaround time.

The profile of resistance mechanisms to third-generation EGFR TKI osimertinib differs from first- and second-generation EGFR TKIs, and also depends on whether osimertinib is given in the first or second line. Notably, T790M mutations are absent in cases of treatment failure on osimertinib.[35] Again, up to 15% demonstrate transformation, and there are various targetable mutations that occur. While these targetable mutations may have available therapies, they tend to happen at a relatively low frequency of <5%. Tissue biopsy for histopathology and NGS can be considered to identify subsequent treatment options. In addition to histological transformation, *MET* amplification and alternate drivers (e.g. *RET* re-arrangement), have been described. However, in the majority of cases, mechanisms of resistance remain unknown and this remains an unmet medical need subject to ongoing research efforts.

Recommendations for molecular testing in advanced, EGFR TKI-resistant NSCLC

- Tumour biopsy is preferred for detection of *EGFR* T790M in the resistance setting. Plasma ctDNA testing is an acceptable alternative approach; however a negative result should be followed by tissue testing for *EGFR* T790M, as well as alternative mechanisms of resistance when feasible [I, A].
- In patients who develop disease progression on third-generation EGFR TKI, tissue biopsy for histopathology and multiplex testing can be considered [III, C].

Unresectable stage III NSCLC

Stage III NSCLC is an intrinsically heterogeneous disease, with distinct molecular and phenotypic sub-types. In unresectable disease, radiotherapy (RT) is the curative local therapy and treatment intensification with concurrent platinum-based doublet chemotherapy is the standard of care.[36] The introduction of immune checkpoint inhibitors (ICI) to concurrent chemoradio-therapy (CRT) represented a paradigm shift in the management of stage III disease. In the phase III PACIFIC trial, the addition of 1 year of consolidation durvalumab therapy resulted in a significant improvement in overall survival, which was sustained at long-term follow-up, with an estimated 5-year overall survival rate of 42.9% for durvalumab versus 33.4% for placebo.[37]

In spite of these improved outcomes, there are concerns regarding the applicability of these results to re al-world clinical practice. In a patient population commonly associated with high incidence of comorbidi-ties, multi-modality CRT with ICI is difficult to tolerate. In a Dutch multi-centre study of 855 eligible patients, only 52% received curative-intent, multi-modality therapy and only 57% patients who underwent CRT received ICI. Main predictors for not receiving ICI included age ³70, diabetes and ³grade 3 dysphagia post-CRT.[38] This highlights the need for biomarkers to identify patients at risk of relapse, who are most responsive to ICI and will benefit most from treatment intensification.

PD-L1 status is a potential biomarker being considered in the unresectable stage III setting. In a post-hoc exploratory analysis of the PACIFIC study performed at the request of the European Medicines Agency

(EMA), patients with PD-L1 status ³25% benefited more from durvalumab (HR 0.53, 95% CI 0.33 0.85) compared to patients with PD-L1: 1 to 24% (HR 0.69, 95% CI 0.43–1.10) and <1% (HR 1.05, 95% CI 0.69–1.62).[39] As such, durvalumab was restricted for patients with PD-L1 ³1%, a decision that was disputed by experts in the field, citing issues with the unplanned nature of the analysis, assessment of PD-L1 status on pre-CRT samples and only available in 63% of patients, as well as the over-performance of the control arm in the PD-L1 <1% sub-group compared to the intention-to-treat placebo arm.[40] Subsequent trial designs are now incorporating PD-L1 status as a stratification factor and this will shed more light on its utility as a biomarker.[41] While awaiting these results, PD-L1 testing in patients with adequate tissue obtained at diagnosis may be considered to facilitate counselling with patients for consolidation durvalumab.

In the PACIFIC study, *EGFR*-mutated patients represented only 6% of all patients. Post-hoc exploratory analysis of this subgroup demonstrated similar survival outcomes (HR for PFS and OS 0.91 and 1.02, respectively) and wide 95% confidence intervals owing to the small numbers.[42] *ALK* rearrangements were also not accounted for in this study. Retrospective analyses of oncogene-addicted stage III unresectable NSCLC have also similarly demonstrated limited activity of durvalumab consolidation.[43,44] Moreover, a lack of efficacy had also been demonstrated with ICI in patients with *EGFR* mutations and *ALK* rearrangements in the advanced setting, generating uncertainty regarding its clinical benefit post-CRT.[45,46] Given that there is insufficient data to exclude these patients from receiving adjuvant durvalumab, molecular testing for *EGFR* mutations and *ALK* rearrangement may be considered at diagnosis for facilitate clinical decision making.

Molecularly directed approaches are being explored in unresectable stage III NSCLC. The randomised phase III LAURA study is assessing the efficacy of maintenance third-generation EGFR TKI osimertinib post-CRT and is expected to read out in 2023.[47] Given the superior disease-free survival seen with osimertinib in early-stage completely resected *EGFR* mutated disease,[48] LAURA with a similar design and primary endpoint is expected to have a positive outcome as well. This could potentially lend further support to testing for sensitising *EGFR* mutations Furthermore, given the high relapse rates following curative treatment for stage III disease, challenges with performing molecular testing on archival tissue, high prevalence of targetable genetic alterations in our local context as well as cost effectiveness of performing panel-based testing, there is merit in conside-ring reflex testing for PD-L1 expression, *EGFR* mutation and *ALK* rearrangements for non-squamous histologies as well as for other targetable genetic alterations with approved therapies in the advanced setting.[21,49]

Recommendations for molecular testing in unresectable stage III NSCLC (Fig. 2)

- Molecular testing for *EGFR* mutation may be considered at diagnosis on tissue specimen for unresectable stage III non-squamous NSCLC [II, C].
- PD-L1 testing on tissue specimen at diagnosis for unresectable stage III NSCLC may be considered [II, C].
- Multiplex testing to include all targetable genetic alterations with approved therapies (such as *ALK*, *ROS1*, *KRAS* G12C, *BRAF* V600E, *MET*ex14 skipping, *RET*, *ERBB2*, *NTRK*) in advanced NSCLC is beneficial and may be considered [V, C].
- Reflex testing at diagnosis for PD-L1 expression and *EGFR* mutation for non-squamous NSCLC may be considered [V, C].

Resected stage I–III NSCLC

EGFR mutation testing in early-stage NSCLC

Approximately 20% of lung cancer in Singapore is diagnosed in the early stages (stages I–II),[50] where treatment comprises surgical resection and mediastinal node dissection. Stage III lung cancer accounts for approximately 14% of lung cancer in Singapore, where surgical resection may be performed in patients with single station N2 disease as part of multimodality management.[50] A significant proportion of patients with stage I–III NSCLC eventually recur despite curative intent surgery,[51] and thus adjuvant strategies continue to be evaluated with the aim of improving outcomes. The following studies highlight the role of *EGFR* biomarker testing in the selection of suitable patients for adjuvant *EGFR* TKI.

The phase III randomised ADAURA trial[48] enrolled a total of 682 patients with *EGFR* mutation-positive (Ex19del or L858R), completely resected stage IB–IIIA NSCLC to receive 3 years of osimertinib versus placebo following recovering from surgery and standard adjuvant chemotherapy, if given. HR for the primary outcome measure of disease-free survival (DFS) in patients with stage II and IIIA disease was 0.23 (95% CI 0.18, 0.30; 242/470 events; 51% maturity),[52] and the 3-year DFS rate was 84% with osimertinib versus 34% with placebo. Additionally, fewer patients in the osimertinib arm experienced central nervous system (CNS) recurrences,

with a CNS DFS HR of 0.24 (95% CI 0.14, 0.42; 63/470 events).[52]

The CTONG 1104 trial[53]—a randomised phase III trial in patients with *EGFR* mutation-positive completely resected stage II–IIIA (N1-N2) NSCLC—compared adjuvant gefitinib versus standard doublet chemotherapy with DFS as the primary endpoint. Although DFS benefit was lost at 5 years (DFS at 3 and 5 years were 40.3% and 23.4% for gefitinib, versus 33.2% and 23.7% for chemotherapy), the overall survival of 75.5m in completely resected N1-N2 NSCLC was the best in this group of patients.[54]

The ongoing Neo-ADAURA trial[55] is a phase III randomised study assessing neoadjuvant osimertinib as monotherapy or in combination with chemotherapy in patients with resectable stage II–IIIB *EGFR*-mutated NSCLC, versus standard of care chemotherapy alone, followed by surgery and adjuvant treatment as per investigator's choice, including osimertinib for up to 3 years. Patients are selected for *EGFR* mutation status and it is expected that this trial will further support *EGFR* biomarker testing in early non-squamous NSCLC.

PDL1 testing in early-stage NSCLC

The phase III Impower010 trial[56] assessed the role of adjuvant atezolizumab versus best supportive care (BSC) in 882 completely resected stage IB–IIIA NSCLC patients after surgery and cisplatin-based chemotherapy. A significant improvement in DFS was demonstrated for the atezolizumab arm (median DFS 42 versus 35 months; HR 0.79, 95% CI 0.64–0.96). Notably, the greatest magnitude of DFS benefit was observed in stage II–IIIA patients with PD-L1-expressing tumour cells ³50% (median DFS not evaluable versus 35.7 months; HR 0.43, 95% CI 0.27–0.68), leading to local regulatory approval for this subgroup.

The KEYNOTE 091 study[57] randomised 1177 participants with stage IB–IIIA NSCLC after complete surgical resection with or without adjuvant chemotherapy to receive either pembrolizumab or placebo for 1 year. An improvement in the primary endpoint DFS with pembrolizumab in all-comers regardless of PD-L1 expression was demonstrated (median 53.6 vs 42.0 mo; HR 0.76; 95% CI 0.63–0.91; *P*=0.0014), but there was a non-significant trend towards improvement in those with PD-L1 ≥50%. Overall survival results are immature and regulatory approvals are still pending.

In the neoadjuvant setting, the CheckMate 816 trial[58] randomised patients with stage IB–IIIA resectable NSCLC to received platinum-based chemotherapy with or without nivolumab, followed by resection. The median event-free survival was 31.6 months (95% CI, 30.2 to not reached) with nivolumab plus chemotherapy and 20.8 months (95% CI, 14.0–26.7) with chemotherapy alone (HR for disease progression, disease recurrence, or death, 0.63; 97.38% CI, 0.43–0.91; *P*=0.005). This benefit was demonstrated across disease stages, histologies, tumour mutational burden and PD-L1 expression levels, but notably a greater event-free survival benefit was observed in patients with positive PD-L1 expression.

Adjuvant and neoadjuvant immunotherapy strategies continue to be investigated in the setting of resected NSCLC, and these studies highlight the utility and limitations of PD-L1 as a biomarker in the selection of treatment approaches. Nonetheless, given recent regulatory approvals, PD-L1 expression testing should be performed to facilitate clinical decision making in stage IB–IIIA NSCLC.

Testing for other biomarkers

Other biomarker such as *ALK*, *ROS1*, *KRAS* G12C, *BRAF* V600E, *MET*ex14 skipping, *RET*, *ERBB2* and *NRTK* are targetable mutations that are often tested for in advanced non-squamous NSCLC. Where available and feasible, extended testing for biomarkers, e.g. ALK, should be considered as emerging adjuvant or neoadjuvant studies exclude these subpopulations of oncogene-driven cancers. The ongoing ALINA trial[59] compares alectinib versus chemotherapy as adjuvant treatment for patients with resected stage IB–IIIA *ALK*+ NSCLC. There is no data yet available from ALINA, or any studies to guide the use of adjuvant targeted therapies for the other mutations in the early-stage setting.

Recommendations for molecular testing in resected stage I–III NSCLC (Fig. 2)

PD-L1 expression testing on tissue specimen should be performed for stage IB–IIIA NSCLC [I, A], as well as at least *EGFR* mutation testing for non-squamous histologies [I, A].

CONCLUSION

Comprehensive molecular testing is of increasing importance given the myriad of approved targeted therapies, as well as the numerous ongoing clinical trials that evaluate new therapies for specific genomic alterations in NSCLC. Importantly, biomarker testing strategies must evolve in parallel with emerging data both in the advanced and early-stage settings. This consensus aims to provide a framework for molecular testing in early and advanced NSCLC, tailored to the

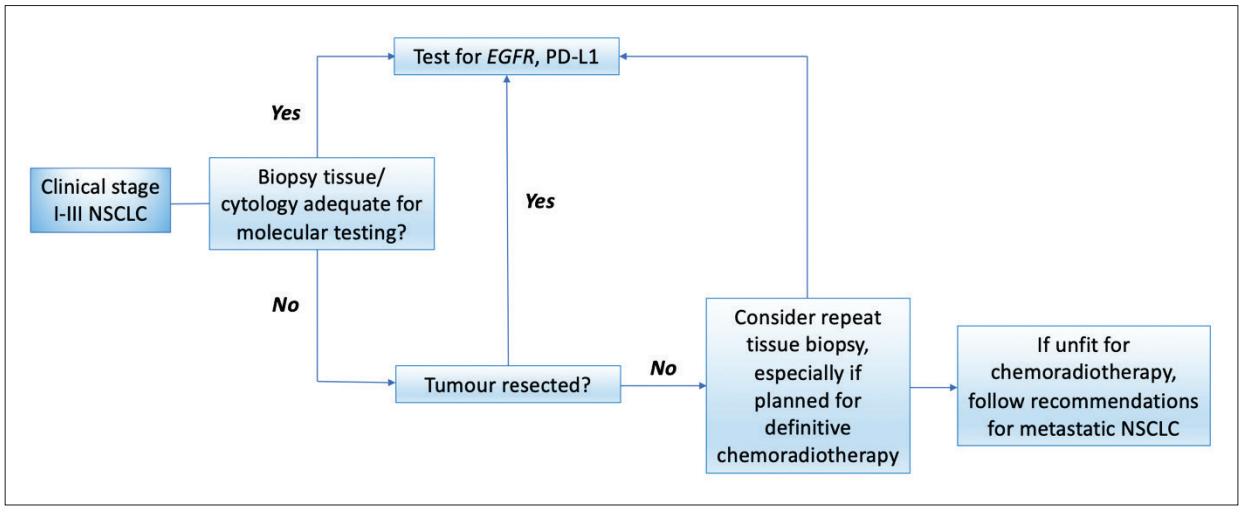

Fig. 2. Biomarker testing recommendations for clinical stage I–III non-small cell lung cancer (NSCLC).
EGFR: epidermal growth factor receptor; PD-L1, programmed death-ligand 1

Singapore context where the presence of genomic alterations in NSCLC is high, with the aim of timely diagnosis and better patient outcomes.

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COMMENTARY

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Leveraging ChatGPT to aid patient education on coronary angiogram

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ABSTRACT

Natural-language artificial intelligence (AI) is a promising technological advancement poised to revolutionise the delivery of healthcare. We aim to explore the quality of ChatGPT in providing medical information regarding a common cardiology procedure—the coronary angiogram—and evaluating the potential opportunities and challenges of patient education through this natural-language AI model in the broader context. In a conversational manner, we asked ChatGPT common questions about undergoing a coronary angiogram according to the areas of: description of procedure, indications, contraindications, complications, alternatives, and follow-up. The strengths of the answers given by ChatGPT were that they were generally presented in a comprehensive and systematic fashion, covering most of the major information fields that are required. However, there were certain deficiencies in its responses. These include occasional factual inaccuracies, significant omissions, inaccurate assumptions, and lack of flexibility in recommendations beyond the line of questioning, resulting in the answers being focused solely on the topic. We would expect an increasing number of patients who may choose to seek information about their health through these platforms given their accessibility and perceived reliability. Consequently, it is prudent for healthcare professionals to be cognisant of both the strengths and deficiencies of such models. While these models appear to be good adjuncts for patients to obtain information, they cannot replace the role of a healthcare provider in delivering personalised health advice and management.

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Keywords: artificial intelligence, cardiology, coronary artery disease, medical education, public health, quality of life

Natural-language artificial intelligence (AI) is a promising technological advancement poised to revolutionise the delivery of healthcare.[1] Traditionally, inclusion of technology in the augmentation of healthcare communication comprised the use of chatbots, which is limited by a predetermined set of queries and matched answers.[2] However, natural-language AI models prompt a paradigm shift, given that they can interpret colloquial inputs to provide new texts based on the large datasets that it was trained. Chat Generative Pre-trained Transformer (ChatGPT) is an example of an open natural-language AI conversational platform that has recently been developed. While there are many other natural-language models, ChatGPT's free, intuitive, and user-friendly interface has attracted the most significant group of users and analysis of its output. It has not only revolutionised the non-medical fraternity with wide-ranging abilities, including creative writing, essay writing, prompt writing, code writing, and answering questions,[3] but has also made inroads in the medical field.[4] Given its accessibility and reported medical proficiency,[5] we aim to explore the quality of ChatGPT in providing medical information regarding a common cardiology procedure—the coronary angiogram—and evaluating the potential opportunities and challenges of patient education through this natural-language AI model in the broader context.

In a conversational manner, we asked ChatGPT (https://openai.com/blog/chatgpt) common questions about undergoing a coronary angiogram according to the areas of: description of procedure, indications, contraindications, complications, alternatives, and follow-up.

The types of questions asked and the evaluation of the outputs' strengths and deficiencies are outlined in Table 1, with the exact replies made available in the Supplementary Appendix.

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The strengths of the answers given by ChatGPT were that they were generally presented in a comprehensive and systematic fashion, covering most of the major information fields that are required. The language used was easy to understand by the layperson, and medical terminology unfamiliar to persons without clinical experience were avoided. Most responses also appropriately concluded that it was important to involve the healthcare professional in discussing the specific circumstances of the individual, and acknowledged its own deficiencies of providing personalised recommendations.

However, there were certain deficiencies in its responses. First, while infrequent, there were some factual inaccuracies. These included inadequate differentiation between antiplatelet and anticoagulation among blood thinners and the resultant decision to continue or discontinue it prior to the procedure; certain inaccurate indications for angiography (e.g. family history, prior stroke, monitoring); some inaccurate risks of angiography (e.g. blood clot—rather than calcifications—dislodgement by catheter); and incorrect contraindications (e.g. severe heart failure). Second, there were some significant omissions. For example, it excluded the important indication of active acute coronary syndromes, for which coronary evaluation via angiography would be recommended. Third, there were some inaccurate assumptions made. For example,

Table 1. Evaluation of outputs by ChatGPT on patient education on coronary angiogram.

Questions	Strengths	Deficiencies
1. Procedural description		
Can I find out more about coronary angiogram?	Offered a generally factually accurate explanation of how a coronary angiogram is performed.	Some details lacking, including the expected duration.
How long is the procedure? Will I be awake or asleep for the procedure? Is it painful?	Provided generally reasonable and accurate answers.	The role of sedation may vary and should be acknowledged. In most instances, the procedure will be carried out under local anaesthesia and sedation only if needed. The need for general anaesthesia is extremely rare.
What do I need to do beforehand to prepare?	Provided both medical and logistics advice on what to do and prepare prior to the procedure.	–
Should I stop or take blood thinners for my angiogram?	Reasonable attempt at providing general advice and mentioned that the physician needs to individualise advice.	Could have better differentiated the differences between antiplatelet and anticoagulation. Continuation of antiplatelet is usually routine and the decision to continue anticoagulation may vary depending on the individual, access site and indication for anticoagulation. Did not include common antiplatelets that are used nowadays, like ticagrelor and prasugrel, especially in the setting of acute coronary syndromes.
2. Indications		
What are some reasons why I need to go for an angiogram? If I have chest pain when climbing up the stairs should I go for an angiogram? I get increasingly breathless, is an angiogram useful? What are the benefits of going for an angiogram?	Balanced answer towards the role of angiogram in the presence of symptoms stating that the symptoms may be indicative of heart disease but will need further consultation with a physician that may entail other diagnostic testing. Decision for angiography will vary depending on the situation.	Provided several incorrect indications that would not necessitate an angiogram—these include prior stroke, family history and "monitoring of progress of heart disease". Excluded the important indication of active acute coronary syndromes usually requiring angiograms. Should include simple mention on potential non-cardiac causes of chest pain or breathlessness.
If I have a heart attack, should I go for an angiogram?	Provided a firm recommendation that in such instances, an angiogram should be performed urgently or emergently, especially if symptoms are ongoing.	–

Table 1. Evaluation of outputs by ChatGPT on patient education on coronary angiogram. (Cont'd)

Questions	Strengths	Deficiencies
3. Contraindications		
What are some reasons I should not go for an angiogram?	Provided a comprehensive list of both absolute and relative contraindications.	The contraindication of "severe heart failure" should be better explained, as patients generally may undergo coronary angiography to work up low ejection fraction. This is probably more accurately stated as active heart failure, and that steps to first optimise the patient prior to undergoing coronary angiography in the elective setting.
4. Complications		
What are some complications of an angiogram? What are the harms of going for an angiogram?	Provided comprehensive list of both common and rarer complications.	Some inaccurate portions, including the risk of dislodgement of blood clots by the catheter potentially causing heart attack or stroke, although this is usually caused by calcifications.
5: Alternatives		
Are there alternatives to an angiogram?	Provided reasonable lists of alternatives.	–
Would you recommend I go for it?	Good explanation of its limitation that as an AI language model, it cannot provide medical advice or make recommendations for individual cases.	–
6: Follow-up		
What do I need to do after an angiogram?	Provided reasonable guidelines on what to do.	–
Do I need a follow up appointment? Are there medications to take after an angiogram?	Provided a reasonable list of recommended follow up plans, including a list of common medications to take.	Some inaccurate parts, including the angiogram being able to show that a patient has high cholesterol.

it suggested that sedation is usually given but this may vary. Lastly, the model also appeared inflexible in recommendations beyond the line of questioning, resulting in the answers only focused on the topic. For example, the model was not able to consider non-cardiac causes of common clinical presentations of chest pain and breathlessness in the context of asking about symptoms and need for coronary angiography.

The shortcomings of a natural-language AI platform like ChatGPT may be due to several issues.

First, given that the models are limited by the data inputs, latest developments may not be fully captured by datasets that the software was trained on. Further, given that information had been derived from all sources of internet, there may be inaccurate information that are presented to the reader.

Second, the model required further probing and prompting for information that would otherwise have normally been given by healthcare providers as part of the counselling process. Such omissions in the

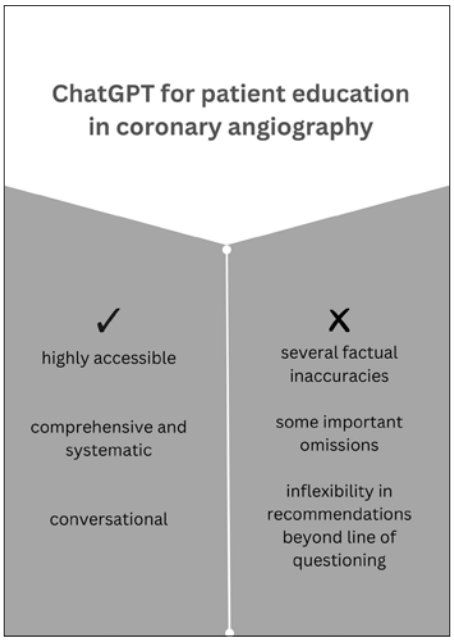

Fig. 1. Overview of strengths and deficiencies of ChatGPT patient education on coronary angiogram.

initial explanation may result in potentially important information not being given to patients or caregivers that may subsequently impact their decision-making process.

Third, the inability for the model to be flexible in recommendations beyond the line of questioning may be due to the conversational nature and scoping of the topic. This may be acceptable if patients only require more knowledge about the particular topic, but it can be counter-productive when it involves looking laterally, especially for differential diagnoses.

Overall, the performance of ChatGPT was thought-provoking. We would expect an increasing number of patients who may choose to seek information about their health through these platforms given its accessibility and perceived reliability. Consequently, it is prudent for healthcare professionals to be cognisant of both the strengths and deficiencies of such models. The ability to harness and incorporate these models into our healthcare systems may transform and improve healthcare delivery bringing potential benefits to both patients and physicians alike, particularly with constant improvements of this model; the release of improved iterations, including the paid version of ChatGPT Plus, which operates on increasingly more datapoints and parameters, may further improve the accuracy and performance of their outputs.[6] Nevertheless, while these models appear to be good adjuncts for patients to obtain information, they cannot replace the role of a healthcare provider in delivering personalised health advice and management.

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Systemic methotrexate in the management of ectopic pregnancy and pregnancy of unknown location

Dear Editor,

Ectopic pregnancy (EP) occurs in 2% of all spontaneous conceptions. It can be a life-threatening condition and is the most common cause of mortality during the first trimester of pregnancy, contributing to 7% of all pregnancy-related deaths.[1,2] The risk factors for EP include tubal damage following surgery or infection, smoking, and in vitro fertilisation. The fallopian tube is the most common location of ectopic implantation, accounting for more than 90% of cases.

EP can be managed surgically, medically, and occasionally by expectant management. Medical management of EP has grown in popularity, and several observational studies have reported success rates with a single dose of systemic methotrexate (MTX) in the range of 65–95%, with 3–27% of women requiring a second dose.[1]

Pregnancy of unknown location (PUL) is defined as the condition when there is no evidence of an intra-uterine pregnancy, extra-uterine pregnancy or retained products of conception on a transvaginal ultrasound scan despite a positive pregnancy test. A single dose of MTX 50 mg/m^2 has been used successfully in women who present with symptomatic persistent PUL, leading to a subsequent resolution of serum β-human chorionic gonadotropin (hCG) levels.

We conducted a review of the medical management of EP and PUL with MTX at KK Women's and Children's Hospital, Singapore. Also examined are predictive factors for its success rate, efficacy of MTX, the need for a second dose of MTX, and the need for surgery.

This single-centre study included women with a diagnosis of EP or PUL from January to December 2019. Per institutional guidelines, the inclusion criteria for medical management of EP were haemodynamic stability, non-acute abdomen on clinical examination, β-hCG levels <5000 IU/L, tubal or adnexal mass <2 cm, and absence of fetal cardiac activity.

These women were followed up with serum β-hCG levels on day 5, day 12 and weekly thereafter until serum β-hCG levels fall below 25 IU/L. Treatment success was defined as the resolution of the serum β-hCG level to <25 IU/L. Treatment failure was defined as the need for surgical intervention. The need for a second MTX injection was not considered a treatment failure.

A total of 135 patients were included in the final analysis. Among them, 126 patients received 1 dose of MTX, while 9 patients required a second dose. The overall success rate of medical management with MTX for EP was 73.3% after 1 dose and 77% after including patients who received 2 doses of MTX (Fig. 1). The success rate of medical management for PUL was 100% (n=11) and that for fallopian tube EP was 76% (n=91/120). The overall success rate for non-tubal EPs was 50% (n=4). A total of 31 patients (23%) in our study had failure of treatment for their EPs with MTX. Among them, 27 patients had 1 dose of MTX, while 4 patients received 2 doses of MTX.

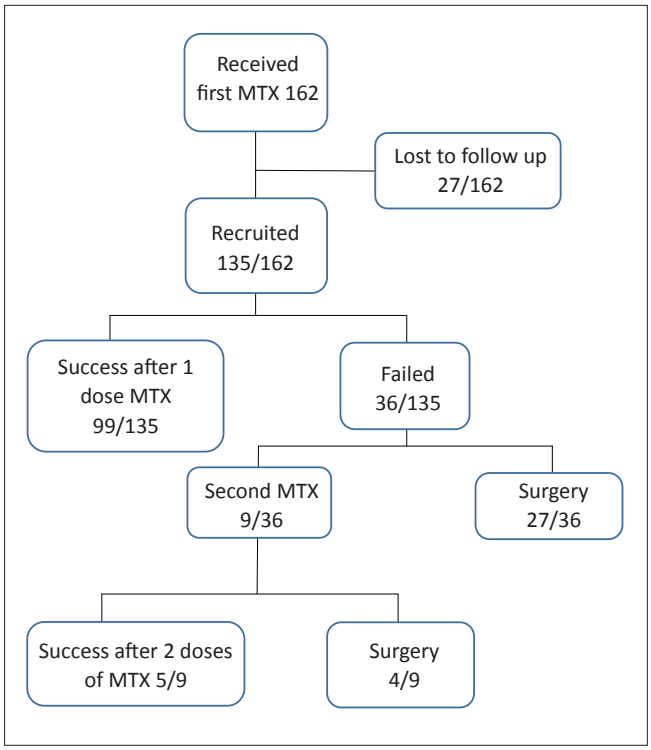

Fig. 1. Flowchart of systemic methotrexate (MTX) and success rate.

The success rate of MTX treatment was related to the initial β-hCG levels. When the initial β-hCG level was less than 1500 IU/L, the success rate was 88% (n=86/98) (P<0.0001); when the initial β-hCG level was between 1500–3000 IU/L, the success rate was 67% (n=14/21). When the initial β-hCG level ranged 3000–4000 IU/L, the success rate was 50% (n=3/6); when the initial

β-hCG level was between 4000–5000 IU/L, the success rate was 14% (n=1/7). Women should be informed of the reduced success rate with a high initial serum β-hCG level when discussing treatment options, and caution should be exercised in this group of women.

In our series, 23% (31/135) required surgery. Of these, majority (n=29) were tubal EPs and 2 were non-tubal EPs (1 scar EP and 1 interstitial EP). In this group, 17 patients presented as an emergency with either abdominal pain or vaginal bleeding necessitating an emergency surgery. The rest (n=14) underwent surgery for persistently high β hCG levels.

In our study population, the overall MTX treatment success rate was 88% when the initial β-hCG level was <1500 IU/L. The multivariate analysis that looked into various parameters in our study showed that only β-hCG level <1500 IU/L on D1 was a statistically significant predictive factor of MTX treatment success (P<0.0001). Other studies conducted by Potter, Kirk and Dudley et al. also reported that treatment success rates are higher with lower β-hCG levels, and the success rates were 81–98% if the serum β-hCG levels were <1000 IU/L, compared with 38% if β-hCG levels were >5000 IU/L.[3-5]

There was no reported success of MTX when the initial β-hCG level was over 5000 IU/L in our study. This is in keeping with a previous systematic review of 503 patients that showed a statistically significant increase in failure rates when initial β-hCG levels were >5000 IU/L compared with those who had initial levels of <5000 IU/L (odds ratio 5.5, 95% confidence interval 3.0–9.8).[6] Therefore, women with an initial serum β-hCG level of >5000 IU/L should be offered surgery as the first line management option; this is in keeping with the 2019 NICE guidelines.[7]

In conclusion, our review showed that the overall success rate for the medical management of EP and PUL with MTX was 77% with no serious treatment related morbidity. Therefore, a systemic MTX treatment for EP and PUL in a carefully selected group of women is a safe alternative to surgery. Our study emphasises that the initial serum β-hCG level <1500 IU/L is the single most important prognostic indicator of treatment success. Women desirous of the medical management of their EP or PUL with initial β-hCG level >3000 IU/L should be thoroughly counselled. They must also be informed of the risks, and estimated duration and number of follow-ups. Surgery should be offered as the first line with β-hCG levels >5000 IU/L.

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Improving neonatal counselling service for premature births

Dear Editor,

Despite the substantial advancement of neonatal care leading to increased survival of infants of periviable gestation, as young as 22 weeks,[1] the anticipated birth of an extremely low gestational age infant remains challenging for both the parents and physician, with regard to decision-making in initiating resuscitation post-delivery. Ideally, the eventual decision should be well informed, ethically sound and mutually agreed upon by the medical team, and alongside parental wishes pre-delivery.[2,3]

In our hospital, parents facing periviable pregnancies at risk of preterm deliveries (between 23+0 and 24+6 weeks of gestation, without fetal malformation) are counselled by a neonatologist from the High-Risk Consult Team. We conducted a single-centre, cross-sectional study from 1 January 2021 to 1 January 2022, utilising post-neonatal counselling surveys to improve our neonatal counselling service while evaluating the understanding among Asian parents. This study received exemption from the Institutional Review Board.

The counselling sessions, each lasting 30–45 minutes, mostly took place at bedside, in a single to 5-bedded room. Parents were counselled together, occasionally with a family member or friend, on survival rates based on current and subsequent gestations (depending on fetal weight), short-term morbidities (i.e. intraventricular haemorrhage, retinopathy of prematurity, chronic lung disease, necrotising enterocolitis and sepsis), and long-term outcomes (i.e. developmental impairment, cerebral palsy, and hearing and visual impairment). Options of full resuscitation or "comfort care" at birth were discussed. All information was conveyed with simple verbal-only illustrations of the expected outcomes in percentages or ratios. A self-administered questionnaire in English was distributed to parents within 72 hours after the counselling and was collected within 24 hours after distribution.

The questionnaire sought parents' opinions mainly on the environment in which the counselling took place and the information conveyed by the counsellor (Table 1). Parents were asked to answer each question based on a 5-point Likert scale (1 for strongly disagree and 5 for strongly agree). The answers to the questionnaire were assumed as a consensus between the parents unless the mother was counselled alone. The questionnaire ended with a text box for any suggestions for a better future counselling session.

We enrolled 20 randomly selected married couples who underwent counselling through purposive sampling. Nineteen mothers were carrying singleton fetus and 1 mother had twins. Enforced COVID-19-related hospital visitation restrictions resulted in half of the mothers being counselled alone. Forty percent of mothers found it helpful to have additional support (family member or friend) during the counselling. Most mothers felt that the sessions were conducted in an appropriate place with sufficient amount of time and were ready to discuss about their baby's future during the counselling. Ninety percent of mothers understood their baby's potential outcomes and liked that they were given information on outcomes of prematurity. Most parents (85%) preferred prematurity outcomes to be given in percentages, compared with 50% who favoured ratios and 40% who requested for pictorial guides.

Ninety percent of parents had adequate question-asking opportunities, and 95% of them agreed that our counselling has helped answer questions about their baby. Eighty-five percent of parents decided on the resuscitation plans prior to counselling, and 90% of parents were more confident about their decision post-counselling, with more than half them feeling less anxious post-counselling. Sixty percent of the parents requested for further meetings with their counsellors.

In Asian culture, there are often prenatal birth observances perceived to protect the expectant mother and her unborn child. Expectant mothers are to avoid looking at "unsightly images" and refrain from uttering certain words. Hence, a traditional verbal-only neonatal counselling has been the method of choice in most Asian populations over the last few decades, similar to our study finding. Furthermore, we observed a poor response with regard to visitation to our Neonatal Intensive Care Unit post-counselling, possibly due to fear of what they may potentially witness.

Most parents were satisfied and felt that our neonatal counselling session increased their confidence while reducing their anxiety levels in decision-making for their unborn child. This finding echoed observations in earlier studies, despite the occasional discordance between physician and parents during the decision-making process.[4] Requests were made from parents to

Table 1. Results of questionnaire from 20 enrolled parents.

1. Settings and environment, no. (%)	**Disagree**	**Neutral**	**Agree**
The meeting place was appropriate	2 (10)	1 (5)	17 (85)
Ready to discuss baby's future	1 (5)	5 (25)	14 (70)
Duration of meeting was sufficient	1 (5)	3 (15)	16 (80)
Preferred additional family member/friend during counselling	0 (0)	12(60)	8 (40)
2. Information-giving and counsellor approach, no. (%)	**Disagree**	**Neutral**	**Agree**
Understood information provided about baby's outcome	0 (0)	2 (10)	18 (90)
Liked that they were given information about outcomes of prematurity	1 (5)	1 (5)	18 (90)
Preferred that they were given information in the form of:			
Percentages	1 (5)	2 (10)	17 (85)
Ratio	1 (5)	9 (45)	10 (50)
Pictorial guide	5 (25)	7 (35)	8 (40)
Had sufficient opportunity to ask questions about baby	0 (0)	2 (10)	18 (90)
Made decision regarding resuscitation prior to counselling	1 (5)	2 (10)	17 (85)
Felt more confident about decision regarding resuscitation after counselling session	0 (0)	2 (10)	18 (90)
Felt counselling helped answer all questions about baby	0 (0)	1 (5)	19 (95)
Preferred further meetings with counsellor	1 (5)	7 (35)	12 (60)
3. Others, no. (%)	**Disagree**	**Neutral**	**Agree**
Preferred a visit/orientation to the Neonatal Intensive Care Unit	2 (10)	10 (50)	8 (40)
Felt counselling session helped them feel less anxious about baby	2 (10)	7 (35)	11 (55)

highlight positive outcomes like survivors of premature births, expressing the need for "hope" during this difficult period, and for physicians to raise other matters such as "pain and suffering" as well as the financial burden apart from disability and deaths.[5] In our survey, one parent mentioned that "it would be nice to include some successful cases as examples so that parents know that there may be some hope since all babies are very precious." This was evident in studies examining parental perception regarding neonatal counselling.[5-7]

Nevertheless, there is still a knowledge gap within the community regarding babies being born extremely premature. Information given during neonatal counselling may have been misunderstood due to limited health literacy,[8] leading to increased requests from parents to supplement our counselling with written information or pictorial guides. Parents believe that these tools will further help them decide between active resuscitation versus "comfort care", and augment their understanding on short- and long-term morbidities.[4,5,7] One mother commented, "a pictorial guide on the risks involved at each premature stage would be extremely helpful as it would better allow parents to digest the information overload." In recent years, there are burgeoning publications in the Western population on supplementing neonatal counselling, either with written material or visual aid to bridge this knowledge gap. Earlier randomised-control studies found that women who were counselled with visual or decisional aids, compared with those who were not, were able to retain the complexity of information regarding potential disabilities of premature babies and anticipated longer duration of hospitalisation, without impacting maternal decisional conflict.[8-10] So far, to our knowledge, there are no similar studies found in the Asian population.

Our study limitations include small sample size and potential for bias, as most survey forms were completed by expectant mothers alone. Some of the mothers were in labour, thus influencing their decision-making. However, the survey was done fairly soon after the counselling to reduce recall bias. Future implemented post-delivery

survey could gauge if the counselling had prepared them adequately for the arrival of their premature baby.

In conclusion, our traditional verbal-only counselling at the threshold of viability was sufficient in helping parents make decisions regarding their pregnancies. Supplementing future counselling sessions with written materials or pictorial guides could further assist decision-making, improve knowledge gap, and reduce discordance between physicians and parents.

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Script concordance test to assess diagnostic and management reasoning in acute medicine

Dear Editor,

Clinical reasoning, an essential skill for patient care, can be difficult to assess. We created and validated a script concordance test (SCT) to assess clinical reasoning in acute medicine. This tool was used to provide feedback and targeted remediation for Postgraduate-Year-1 (PGY1) doctors, guide teaching and learning, and facilitate programme evaluation.

The SCT is a validated method for assessing the interpretation of medical data, hypothesis evaluation, and clinical judgement under conditions of uncertainty.[1] An SCT presents brief clinical scenarios and asks examinees to evaluate diagnostic or management options while considering new information (Supplementary Fig. S1). Questions have a degree of uncertainty with no single correct answer. Responses are compared with a scoring key derived from a reference panel of experts.[2]

Clinical reasoning research has largely focused on diagnostic reasoning. With increasing recognition that management reasoning is a distinct skill[3] that is less well understood, calls have been made for more studies in the area.[4] We thus studied differences between diagnostic and management reasoning across internal medicine (IM) doctors of varying seniority. We obtained Institutional Review Board approval (SingHealth CIRB 2020/2591).

We developed an Acute Internal Medicine SCT (AIM-SCT) blueprinted for common acute medical conditions. From 50 clinical scenarios, item analysis was performed to discard overly concordant (i.e. poor discriminant value) or discordant (i.e. measurement error) questions. Final question selection ensured representation across all medical specialties, in accordance with the blueprint. Questions were classified as "diagnostic reasoning" or "management reasoning" by three of our authors independently; there was 100% agreement. The final AIM-SCT consisted of 73 items (26 scenarios), with 37 items (13 scenarios) testing diagnostic reasoning and 36 items (13 scenarios) testing management reasoning.

We used published processes[5] to derive the scoring key. The reference panel (20 senior residents, equivalent to specialty registrars or fellows) was selected based on the following criteria: (1) currency in on-site senior IM call duties within the past four months, (2) currency in intensive care medicine within the past four months, (3) completed formal IM training, and (4) relevant postgraduate qualifications (e.g. American Board of Internal Medicine and Membership of the Royal College of Physicians). All major medical specialties were represented.

PGY1s, medical officers (postgraduate years 2–5) and senior residents completed the SCT asynchronously using a web response form with a 75-minute time limit. Responses were scored against the scoring key and expressed as a percentage of the total score.

Statistical analysis was conducted with SPSS Statistics version 28.0. (IBM Corp, Armonk, NY, US). Test reliability was assessed using Cronbach's alpha. SCT scores between seniority levels were compared using one-way analysis of variance (ANOVA), followed by pairwise comparisons with Bonferroni correction. Two independent sample t-tests were conducted to compare scores between PGY1s in their first posting (i.e. first 4 months of PGY1 training) and PGY1s in their second or third postings (i.e. latter 8 months of PGY1 training). Diagnostic and management reasoning scores were compared within each seniority level using Wilcoxon signed-rank test. Correlations between diagnostic and management reasoning scores were explored through Pearson correlation. Significance was set at $P<0.05$.

Thirty-three PGY1s, 22 medical officers and 20 senior residents completed the SCT. All participants completed the test asynchronously using a web response form within a 75-minute time limit. Test reliability was high (Cronbach's alpha=0.85); all items showed item-total correlations of >0.05. Total SCT scores increased with seniority; mean percentage scores (standard deviation) increased from 60.3 (8.9) in PGY1s to 70.4 (7.5) in medical officers, and 78.2 (4.5) in senior residents (all pairwise $P<0.01$; Table 1). First-posting PGY1s (those in their first 4 months of PGY1 training, n=10) scored significantly lower than second and third-posting PGY1s (those in their latter 8 months of PGY1 training, n=23), with mean percentage (standard deviation) scores: 54.3 (8.7) versus 62.9 (7.8), $P=0.015$.

PGY1s scored significantly higher in diagnostic reasoning 62.0 (8.9) than in management reasoning 58.6 (10.4), mean score difference 3.34, 95% confidence interval (CI) 0.707–5.99, $P=0.024$. No significant differences between diagnostic and management reasoning scores were found for medical officers and senior residents (Table 1). Overall, combining all

Table 1. Comparison of total, diagnostic and management reasoning scores.

	PGY1 (n=33)	Medical Officer (MO) (n=22)	Senior Resident (SR) (n=20)	*P* value[a]	Pairwise *P* value[b]
Total SCT scores					
Mean (SD) Median (Q1, Q3)	60.3 (8.9) 62.8 (50.9, 68.0)	70.4 (7.5) 71.4 (65.6, 75.6)	78.2 (4.5) 77.8 (75.2, 82.3)	<0.001	PGY1 vs MO: <0.001 PGY1 vs SR: <0.001 MO vs SR: 0.004
Diagnostic versus management reasoning scores					
Diagnostic score Mean (SD) Median (Q1, Q3)	62.0 (8.9) 63.4 (53.3, 70.0)	70.9 (8.6) 71.0 (63.6, 77.4)	78.6 (7.3) 76.4 (72.7, 85.9)	<0.001	PGY1 vs MO <0.001 PGY1 vs SR <0.001 MO vs SR 0.013
Management scores					
Mean (SD) Median (Q1, Q3)	58.6 (10.4) 57.0 (48.6, 67.6)	70.0 (8.5) 72.4 (63.5, 77.5)	77.7 (6.3) 77.7 (72.4, 83.6)	<0.001	PGY1 vs MO <0.001 PGY1 vs SR <0.001 MO vs SR 0.018
Score difference (diagnostic minus management)					
Mean (SD) (95% CI of Mean) Median (Q1, Q3)	3.34 (1.30) (0.71, 5.99) 4.74 (-3.17, 4.74)	1.00 (1.80) (-2.74, 4.75) 2.59 (-5.19, 2.59)	0.87 (2.32) (-4.00, 5.73) 0.56 (-7.91, 10.1)	N/A	N/A
***P* value for score difference**[c]	0.024	0.322	0.681		

PGY1: Postgraduate-Year-1; SCT: script concordance test; SD: standard deviation
[a] Comparing the 3 groups using 1-way ANOVA.
[b] Group pairwise comparisons using 2 independent sample t-test.
[c] Using the Wilcoxon signed-rank test.

seniority levels, there was a moderate correlation between diagnostic and management reasoning scores (r=0.688, P<0.001).

Our AIM-SCT demonstrates construct validity with good discrimination between seniority levels among IM doctors. It is a feasible tool to assess clinical reasoning using a resource-lean online examination without direct examiner involvement. To the best of our knowledge, this is the first SCT developed in acute IM.

Our SCT is the first, as far as we know, to explore differences in diagnostic and management reasoning performance.[4] We postulate several explanations for our finding that PGY1s, but not more senior clinicians, performed better in diagnostic than management reasoning. First, management reasoning may be more complex than diagnostic reasoning, requiring the weighing of testing and treatment thresholds, consideration of the value of care, and management of uncertainty,[6] and thus may take more time and experience to develop. Our finding may support the hypothesis of a separate management script.[6,7] Second, management reasoning requires shared decision-making and follow-up[4]—skills that may not develop until later in postgraduate training as junior doctors learn at the workplace via situated learning.[8] Third, undergraduate medical curricula may prioritise diagnostic over management reasoning. Singapore's national outcomes framework for medical graduates includes a list of conditions for which both diagnostic and management competencies are required, and a separate list of conditions where graduates are only expected to diagnose but not manage.[9]

Our study has several limitations. First, all reference panel members come from a single academic medical centre. A prior study found local experience to be associated with improved SCT performance,[10] suggesting that clinical reasoning may be context-specific and not immediately transferrable to other settings. Second, SCTs do not test features of management reasoning such as the dynamic interplay between people, systems and priorities, as well as communication and shared decision-making,[4,6] which are perhaps better assessed with workplace-based assessments.

In conclusion, our AIM-SCT is a reliable, valid and feasible tool to assess clinical reasoning in acute medicine. Plans are underway to incorporate our SCT into the formative assessment of PGY1s to provide

individualised feedback and plan targeted interventions. Finally, our finding that management reasoning may develop differently from diagnostic reasoning requires empirical validation, and a better understanding of how management reasoning develops will be valuable in its teaching and assessment.

Disclosure

No author has any affiliation or financial involvement with any commercial organisation with a direct financial interest in this manuscript.

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