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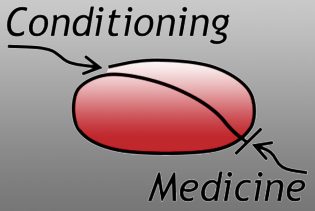

REVIEW ARTICLE | OPEN ACCESS

Is there a role for remote ischemic conditioning in preventing 5-fluorouracil-induced coronary vasospasm?

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Cardiac ischemia associated with chemotherapy has been linked to several anti-neoplastic agents and is multifactorial in etiology. Coronary artery vasospasm is one of the most commonly reported effects of cancer therapy that can lead to myocardial ischemia or infarction. The chemotherapy agent 5-fluorouracil (5-FU) or its oral pro-drug capecitabine can result in coronary vascular endothelial dysfunction causing coronary artery spasm, and possibly coronary thrombosis. These drugs have also been shown to be associated with myocardial infarction, malignant ventricular arrhythmias, heart failure, cardiogenic shock, and sudden death. The proposed mechanisms underlying cardiotoxicity induced by 5-FU are vascular endothelial damage followed by thrombus formation, ischemia secondary to coronary artery vasospasm, direct toxicity on myocardium, and thrombogenicity. There remains a pressing need to discover novel and effective therapies that can prevent or ameliorate 5-FU associated cardiotoxicity. To this point, promising overlap has been observed between proposed remote ischemic conditioning (RIC) cardioprotective mechanisms and 5FU-associated cardiotoxic cellular pathways. RIC, in which transient episodes of limb ischemia and reperfusion (induced by inflations and deflations of a pneumatic cuff placed on the upper arm or thigh), confer both cardioprotective and vasculoprotective effects, and may therefore prevent 5-FU coronary artery spasm/cardiotoxicity. In this review, we will be discussing the following potentially therapeutic aspects of RIC in ameliorating 5-FU associated cardiotoxicity: sequential phases of 5-FU cardiotoxicity as possible targets for dual windows of cardioprotection characteristic of RIC; protective effects of RIC on endothelial function and microvasculature in relation to 5-FU induced endothelial dysfunction/microvascular dysfunction; reduction in platelet activation by RIC in the context of 5-FU induced thrombogenicity; and the utility of improvement in mitochondrial function conferred by RIC in 5-FU induced cellular toxicity secondary to mitochondrial dysfunction.

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Introduction

Cardiovascular disease is the leading cause of morbidity and mortality worldwide (Benjamin et al., 2019). The World Health Organization (WHO) estimates that 17 million people die each year of cardiovascular disease, accounting for 30% of all deaths (Balukumar et al., 2016). Cancer is the second leading cause of death globally and is associated with 9 million deaths each year (Leal et al., 2016). In fact, cancer has now overtaken cardiovascular disease as the leading cause of mortality in high-income countries (Dagenais et al., 2019). According to the WHO, the incidence of cancer is expected to rise by about 70% over the next 20 years (WHO, 2018). Half of those diagnosed with cancer will survive for at least a decade, but this survival rate is expected to increase significantly in the future, leading to worsening burden of cancer-related complications experienced by the global population (Lucas et al., 2017,Cancer Research UK, 2019). Significant advances in cancer therapy have greatly reduced the mortality of cancer patients, with non-malignant comorbid conditions becoming important determinants of their quality of life and overall survival (Siegel et al., 2012). Among this heterogeneous group of comorbid conditions, cardiovascular diseases are a major contributor to overall morbidity and mortality in cancer survivors and patients with active cancer (Barac et al., 2015).

Heart disease and cancer share common risk factors in an ageing population and are further linked through cardiotoxic effects of contemporary cancer treatment (Moser et al., 2006; Weaver et al., 2013; Ghosh et al., 2017). Many cancer patients have subclinical cardiovascular disease, which can be worsened by the pro-inflammatory and hypercoagulable states associated with cancer (Blann 2011; Demers et al., 2012; Ghosh et al., 2018).

Cardiotoxicity secondary to 5-fluorouracil/capecitabine

Cardiac ischemia associated with chemotherapy has been linked to several anti-neoplastic agents and is multifactorial in etiology (Iliescu et al., 2016). Coronary artery vasospasm is one of the most commonly reported effects of cancer therapy that can lead to myocardial ischemia or infarction (Stewart and Pavlakis 2010; Nair and Steingart 2011). The chemotherapy agent 5-fluorouracil (5-FU) or its oral pro-drug capecitabine can result in coronary vascular endothelial dysfunction causing coronary artery spasm, and possibly coronary thrombosis, with a wide range of reported incidence between 1% and 68% (Pai 2000; Van Cutsem et al., 2002). These agents are used to treat solid cancers, including gastrointestinal, breast, head, neck, and pancreatic cancers (Polk et al., 2014). These drugs have also been shown to be associated with myocardial infarction or malignant ventricular arrhythmias (Kosmas et al., 2008). Capecitabine is converted to 5-FU in a three-step process involving several enzymes (Malet-Martino and Jolimaitre 2002). The last step is catalyzed by thymidine phosphorylase (Malet-Martino and Jolimaitre 2002). Many body tissues express thymidine phosphorylase, but this enzyme is expressed in higher concentrations in some carcinomas than in the surrounding normal tissues (Malet-Martino and Jolimaitre 2002). Based on this theory, the concentration of 5-FU at the tumor site should be increased compared to the concentration of 5-FU in healthy tissues, resulting in fewer side-effects involving healthy tissue (Malet-Martino and Jolimaitre 2002). The incidence of capecitabine-associated cardiac side-effects is 3-35%, gathered from the few studies examining capecitabine cardiotoxicity (Van Cutsem et al., 2002; Ng and Cunningham 2005; Jensen 2006; Kosmas et al., 2008; Koca et al., 2011). Case reports of cardiotoxicity after administration of capecitabine are similar to intravenous 5-FU treatment, with the predominant symptom being chest pain (Frickhofen et al., 2002; Cardinale and Colombo 2006; Coughlin et al., 2008). Other less frequent adverse effects are cardiac arrhythmias, myocardial infarction, heart failure, cardiogenic shock, and sudden death (Saif and Shah 2009; Kelly et al., 2013; Polk et al., 2013). Chest pain onset is often abrupt during infusion of 5-FU, but can also be delayed, presenting within the first 72 hours after 5-FU administration (Wacker et al., 2003; Saif and Shah 2009). Often, angina is accompanied by electrocardiogram (ECG) changes including ST-segment depression and prolonged repolarization abnormalities (Saif and Shah 2009).

Cardiac enzymes are infrequently elevated in patients experiencing angina following 5-FU (around 14% of cases) (Holubec et al., 2007; Saif and Shah 2009), and echocardiography has shown regional or global hypokinesis that usually returns to baseline within 48 hours of 5-FU cessation (Saif and Shah 2009). In these cases, significant coronary artery disease and acute plaque rupture is usually ruled out on coronary angiography, which leads to the consideration of coronary artery vasospasm (Cardinale and Colombo 2006; ,Lu et al., 2006). In a review of 377 patients with 5-FU-induced cardiotoxicity, cardiovascular risk factors such as smoking, diabetes, hypercholesterolemia, and family history of heart disease were found in 37% of the patients. Smoking was the most common risk factor among these groups of patients (Saif and Shah 2009). Previous or concomitant radiation therapy may play a role in 5-FU-induced cardiac toxicity as radiation can cause small-vessel thrombosis. 5-FU is a radio-sensitizer and may enhance radiation-induced thrombosis (Fajardo 1973; May et al., 1990). There is a higher incidence of angina with administration through continuous infusion compared to bolus infusion (Sudhoff et al., 2004; Saif and Shah 2009). It is unclear if this effect is dose-dependent, and although cessation of 5-FU results in resolution of angina, symptoms have been reported to last up to 12 hours (Tsavaris et al. 2002). Re-initiation of 5-FU has been associated with increased incidence of angina with serious complications including acute coronary syndrome, hypotension, cardiac failure, and even death (Sudhoff et al., 2004; Saif and Shah 2009).

While the causative relationship is unclear, endothelin-1 levels have been noted to be elevated in angina patients with 5-FU infusion (Sudhoff et al., 2004). Patients with known pre-existing history of coronary artery disease also have a higher incidence of angina, and are considered to have an increased risk of developing cardiac ischemia (Labianca et al., 1982; Giza et al., 2017).

In addition to high doses of 5-FU, prior mantle radiation, or simultaneous administration of another cardiotoxic chemotherapeutic agent are factors that can contribute to development of cardiac ischemia in patients treated with anti-metabolite drugs (de Forni et al., 1992,Anand 1994). In one large study, myocardial ischemia was reported in 4% of patients receiving high-dose, continuous infusion of 5-FU (Tsavaris et al. 2002). However, the failure of ergonovine and 5-FU to produce direct coronary artery vasospasm during cardiac catheterization has questioned the hypothesis of abnormal vasoreactivity being the predominant mechanism causing 5-FU associated myocardial ischemia (Freeman and Constanza 1988; Stewart et al., 2010). Age of the patient did not appear to influence the occurrence of cardiotoxicity (Labianca et al., 1982).

Need for novel therapies to prevent 5-FU associated cardiotoxicity

The proposed mechanisms underlying cardiotoxicity induced by 5-FU are vascular endothelial damage followed by thrombus formation, ischemia secondary to coronary artery vasospasm, direct toxicity on myocardium, and thrombogenicity. Patients developing ischemic events usually have recurrences if the drug is subsequently administered, so consideration must be given to

withholding future 5-FU therapy if a patient develops ischemic events while on the drug (Anand 1994; Akpek and Hartshorn 1999). Although treatment with vasodilators have been proposed as prophylaxis against coronary artery vasospasm, limited effectiveness of this prophylactic therapy has been observed (Patel et al., 1987). Pharmacogenomic studies and genetic profiling may help predict the occurrence and streamline the treatment of 5-FU-induced coronary artery vasospasm. Further research is required to explore the cardioprotective effect of agents such as coenzyme complex, GLP-1 analogues, and degradation inhibitors on 5-FU-induced coronary artery vasospasm.

Postulated mechanisms of 5-FU associated cardiotoxicity – an overview

Patients with coronary artery vasospasm may have ECG findings suggestive of coronary occlusion, including ST-segment elevation as well as biochemical evidence of myocardial injury with troponin elevation even in the absence of occlusive epicardial vessel disease on coronary angiography or computed tomography (CT) imaging of the coronary vessels. In fact, patients with 5-FU-associated cardiotoxicity have consistently been shown to lack significant coronary stenosis on coronary angiography (Shoemaker et al., 2004; Alter et al., 2006; Camaro et al., 2009; Atar et al., 2010; Tajik et al., 2010).

The underlying mechanism of 5-FU associated cardiotoxicity is not well established and is likely to be multifactorial (Polk et al., 2014). The mechanism to explain 5-FU cardiac effects that is best supported by preclinical and clinical data is coronary artery vasospasm (de Forni et al., 1992; Akhtar et al., 1993; Mossseri et al., 1993; Porta et al., 1998; Sudhoff et al., 2004,Alter et al., 2006,Floyd et al., 2005; Dalzell and Samuel 2009). Preclinical models provide *in vitro* evidence of concentration-dependent vasoconstriction by 5-FU on vascular smooth muscle cells (Mosseri et al., 1993). Clinical data include the documentation of coronary artery spasm angiographically following intravenous (IV) 5-FU, and some cases of successful prophylaxis against coronary artery vasospasm with calcium channel antagonists (Kleiman et al., 1987; Luwaert et al., 1991; Shoemaker et al., 2004; Sudhoff et al., 2004).

However, some characteristics of 5-FU cardiotoxicity are inconsistent with this hypothesis. Coronary artery vasospasm has not been consistently shown angiographically during symptomatic attacks, and reintroduction of 5-FU in patients with a previous adverse cardiac event has not resulted in coronary spasm as evidenced by coronary angiography (Burger and Mannino 1987; Mizuno et al., 1995). In some patients with suspected 5-FU-related cardiotoxicity, ergonovine provocation has failed to induce coronary artery vasospasm (Freeman and Constanza 1988). Echocardiography has demonstrated a reduced ejection fraction and global akinesia of the left ventricular myocardium during attacks, which did not correspond to the segmental distribution of the major coronary arteries (de Forni et al., 1992). Vasodilator drugs are also not consistently protective (Patel et al, 1987; Oleksowicz and Bruckner 1988; Eskilsson and Albertsson 1990; Cwikiel et al., 1996; Akpek and Hartshorn 1999; Saif and Shah 2009).

Therefore, other pathophysiologic mechanisms probably contribute, including myocarditis (Tsibiribi et al., 2006), a direct myocardial toxic effect secondary to the antimetabolite effects of the drug causing a cardiomyopathic picture (Patel et al., 1987; Jensen et al., 2010), or a thrombogenic effect due to endothelial injury (Sasson et al., 1994; Cwikiel et al., 1996; Kuropkat et al., 1999; Jensen et al., 2012). As 5-FU is rapidly cleared from the bloodstream after bolus administration with a half-life of 15 to 20 minutes, a direct effect of the drug seems unlikely to be the cause of cardiotoxicity. There is also a higher incidence of angina with administration through continuous infusion compared to bolus infusion (Sudhoff et al., 2004; Saif and Shah 2009).

Of note, the metabolite of 5-FU, alpha-fluoro-beta-alanine (FBAL), is further catabolized into fluoroacetate, which is known to be highly cardiotoxic (Arellano et al., 1998; Muneoka et al., 2005). The lack of reported cardiac toxicity from fluoropyrimidines administered with the dihydropyrimidine dehydrogenase (DPD) enzyme inhibitors eniluracil and

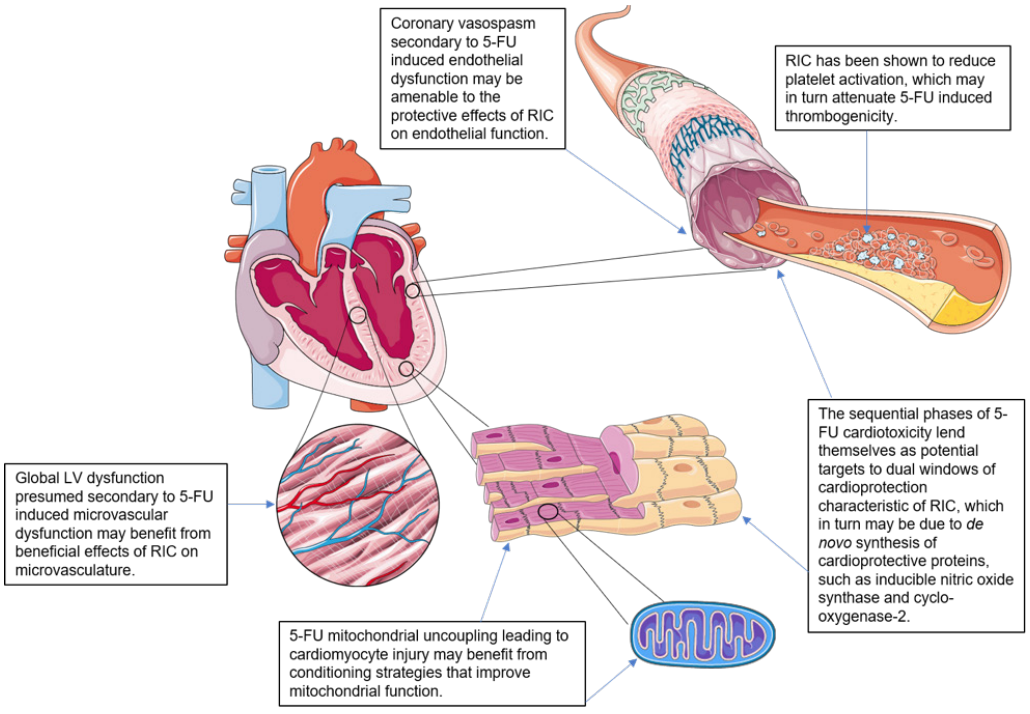

Figure 1. Potential cellular targets in 5-fluorouracil cardiotoxicity amenable to RIC cardioprotection.
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gimeracil lends further support to the theory that metabolic pathways leading to FBAL generation may be a significant pathophysiologic component of cardiotoxicity (Marsh et al., 2002; Guo et al, 2003; Yip et al., 2003).

5-FU administration can evoke a Takotsubo type of cardiomyopathy, a transient regional myocardial dysfunction that is precipitated by physical or emotional stress and thought to be related to exaggerated sympathetic stimulation (Stewart et al., 2010; Basselin et al., 2011; Dechant et al., 2012; Grunwald et al., 2012). The ECGs of patients with presumed Takotsubo cardiomyopathy often reveal ST-segment elevation, and cardiac enzymes are frequently mildly elevated, with a characteristic pattern of left ventricular dysfunction of non-segmental distribution. Finally, individual sensitivity to cardiotoxicity might result from inherited variations in the catabolic enzyme pathways responsible for the metabolism of 5-FU, leading to variable levels of cardiotoxic degradation products.

There remains a pressing need to discover novel and effective therapies that can prevent or ameliorate 5-FU associated cardiotoxicity. In this regard, remote ischemic conditioning (RIC), in which transient episodes of limb ischemia and reperfusion (induced by inflations and deflations of a pneumatic cuff placed on the upper arm or thigh), confer both cardioprotective and vasculoprotective effects, and may therefore prevent 5-FU coronary artery spasm/cardiotoxicity (see Figure 1) (Przyklenk et al., 1993; Chong et al., 2017; Chong et al., 2019).

Potential cellular targets in 5-fluorouracil cardiotoxicity amenable to RIC cardioprotection

1. Two phases of 5-FU cardiotoxicity amenable to two windows of RIC cardioprotection

While coronary vasoconstriction may be observed in patients during or immediately after 5-FU injection, clinical features of toxicity do not typically manifest until after the end of an infusion or even hours to days later (Grem 2000; Jensen et al., 2010, Basselin et al., 2011). 5-FU is also rapidly cleared from the bloodstream after bolus administration with a half-life of 15 to 20 minutes, therefore a direct effect of the drug seems unlikely to be the only cause of cardiotoxicity. There is also a higher incidence of angina with administration through continuous infusion compared to bolus infusion (Sudhoff et al., 2004; Saif and Shah 2009). The delayed phase of 5-FU cardiotoxicity could be explained in part by the generation of toxic breakdown products of 5-FU. FBAL (a metabolite of 5-FU) is further catabolized into highly-cardiotoxic fluoroacetate. Metabolic pathways leading to FBAL generation may be a significant pathophysiologic component of 5-FU cardiotoxicity (Marsh et al., 2002; Guo et al, 2003; Yip et al., 2003).

Gross evidence of myocarditis has been demonstrated in rabbits exposed to 5-FU (Becker et al., 1999) where left ventricular hypertrophy, myocardial necrosis, thickening of intramyocardial arterioles, and disseminated apoptosis in myocardial and endothelial cells have been demonstrated. The use of a high single dose of 5-FU in this study was intended to differentiate the acute toxic effects of 5-FU, which resulted in thrombogenesis and spasm due to endothelial lesions, from delayed cardiotoxicity after four injections at 7-day intervals, which lead to apoptosis of myocardial and endothelial cells without evidence of spasm. These results also support an alternative mechanism for 5-FU cardiotoxicity beyond vasospasm and ischemia.

The sequential phases of 5-FU cardiotoxicity lend themselves as potential targets to dual windows of cardioprotection characteristic of RIC. The protective effects of RIC on endothelial function do not display tachyphylaxis, suggesting that RIC may confer long-term cytoprotective effects against acute ischemia and reperfusion. It is currently thought that a single limb RIC stimulus confers 2 windows of protection, the first occurring immediately and lasting 2-3 hours, and the second window of preconditioning (SWOP), appearing 12-24 hours later and lasting 2-3 days (Hausenloy and Yellon 2010). RIC appears to extend the window of protection to 8 days. The SWOP may be due to *de novo* synthesis of cardioprotective proteins, such as inducible nitric oxide synthase and cyclo-oxygenase-2 (Hausenloy and Yellon 2010). This memory effect could be explained by epigenetic changes in the vasculature, which can extend the protective effect beyond conventional SWOP.

2. Endothelial and vascular smooth muscle dysfunction and the potential role of RIC

Immediately following the intravenous administration of 5-FU, coronary artery vasospasm has been directly visualized during coronary angiography (Heistad et al., 1984; Lopez et al., 1989; Luwaert et al.,1991), as has brachial artery vasoconstriction (38,85). Arterial vasospasm can be related to endothelial dysfunction (an endothelial-dependent mechanism) or primary vascular smooth muscle dysfunction (an endothelial-independent mechanism) (Sara et al., 2018). Endothelial dysfunction is the reaction of the vasculature to a range of insults and clinical circumstances (Reddy et al., 1994; Bonetti et al., 2003), and represents the initial stage of atherosclerosis. It is characterized by an abnormal vasodilatory response to increased flow/shear stress or endothelial-dependent vasodilating agents such as acetylcholine (Vita et al., 1990; Hasdai et al., 1996; Suwaidi et al., 2000; Schwartz et al., 2010). In normal physiology, acetylcholine induces vasodilation through the release of nitric oxide from endothelial cells, which induces vascular muscle cell relaxation, and in turn vessel dilatation through the cyclic-guanosine monophosphate (cGMP) pathway (Hasdai et al., 1997). Any damage to endothelial cells disrupts this process and upon acetylcholine administration, paradoxical vasoconstriction occurs instead (Sara et al., 2018). In the coronary arteries, endothelial function is assessed by invasive pharmacologic provocation during coronary angiography with excessive vasoconstriction representing endothelial dysfunction (Hasdai et al., 1998; Pyke and Tschakovsky 2005; Dalzell and Samuel 2009; Schwartz et al., 2010). Endothelial-independent vascular smooth muscle dysfunction leads to vasoconstriction in the presence of a functionally intact endothelium, and can also be assessed with invasive pharmacologic provocation using nitroglycerin (Hasdai et al., 1998).

RIC has been demonstrated to ameliorate endothelial dysfunction. The mechanisms responsible for these vascular effects are unclear but may relate to shear stress adaptations, augmentation of endothelium-dependent vasodilation and production of nitric oxide (Kimura et al., 2007), circulation of vasoactive mediators such as nitric oxide (Kimura et al., 2007), and systemic antioxidant and anti-inflammatory effects. Kharbanda et al (2002) reported that RIC in one arm attenuated the endothelial dysfunction (assessed by flow-mediated dilatation) induced by a sustained episode of limb ischemia and reperfusion in the contralateral arm. Luca et al (2013) demonstrated improved endothelial function following acute ischemia and reperfusion in healthy volunteers that received 7 days of daily RIC. In the study by Pryds et al (2017), RIC applied daily for 28 days in chronic ischemic heart failure patients was shown to improve global longitudinal strain (GLS) in patients with the highest NT-proBNP plasma levels and lowered systolic blood pressure.

The beneficial effects of RIC on both NT-proBNP and GLS may relate to less myocardial wall stress, caused by reduction in afterload (as evidenced by lowered systemic blood pressure), and this may due to the release of known vasodilatory mediators

of RIC such as adenosine and nitric oxide (Pryds et al., 2017). These beneficial vasodilatory effects could be conferred on patients receiving 5-FU through RIC administration peri- and during chemotherapy. Indeed, RIC activation of pathways producing vasodilatory mediators such as adenosine and nitric oxide can potentially bypass/override the deleterious effect of dysfunctional endothelium in vasoconstriction by delivering vasodilatory mediators direct to vessel smooth muscle lining, facilitating cGMP-mediated muscle relaxation, and vessel dilatation.

3. Dysfunctional coronary microvasculature with vasospasm and the potential role of RIC

Echocardiography has demonstrated global akinesia of the left ventricular myocardium in 5-FU associated cardiotoxicity not corresponding to segmental myocardial distribution of the major coronary arteries (de Forni et al., 1992). The discordance between echocardiographic and angiographic findings could undermine the epicardial arterial vasospasm theory in patients receiving 5-FU. However, microvascular vasospasm could be postulated to explain global, non-segmental akinesia. Endothelial-dependent and endothelial-independent dysfunction also affects the coronary microvasculature, often in the absence of affecting the epicardial vessels (Kinhult et al., 2001) where it leads to global versus segmental ischemia. Since the coronary microvasculature cannot be directly visualized, its function is assessed through measurements of coronary blood velocity and flow with intravascular Doppler techniques and also with pharmacologic provocation at coronary angiography (Hasdai et al., 1998).

Beneficial effects on vascular and endothelial function have been reported in the brachial artery and forearm microcirculation in healthy volunteers following daily RIC for 7 days, suggesting vascular effects of RIC which extend from conduit arteries to the skin microvasculature (Jones et al., 2014). It would be of clinical interest to explore if these beneficial effects can also be conferred onto coronary microvasculature, particularly in the case of diffuse myocardial ischemia induced by 5-FU administration. RIC-induced upregulation of vasodilatory mediators would be expected to enhance vascular smooth muscle relaxation and vessel dilatation at the microvascular level in 5-FU associated coronary arterial dysfunction. Interestingly, the vascular effects induced by RIC were shown to still be present 8 days after the end of the intervention, suggesting a 'memory' effect (Jones et al., 2014) that extends beyond the usual 2-3 days observed with the SWOP in terms of its cardioprotective effect (Marger et al., 1993).

4. 5-FU induced thrombogenicity and role of RIC induced fibrinolysis

Damaged endothelium exposes tissue factors, initiating platelet aggregation that is further propagated by the release of von Willebrand factor and fibrin aggregation, resulting in thrombi. 5-FU may lead to thrombotic occlusive disease, and studies of rabbit endothelium exposed to 5-FU have shown areas of platelet aggregation and fibrin formation (Yudkin et al., 1999; Jensen and Sorensen 2012). Regulating the initiation of thrombus formation is an additional aspect of endothelial function and studies have characterized abnormal endothelial function by identifying altered levels of endothelium-derived markers such as von Willebrand factor and fibronectin (Spasojevic et al., 2005; 2008), suggesting a role of endothelium-associated thrombogenicity in 5-FU cardiotoxicity.

Pryds et al (2017) have investigated the effect of RIC applied daily on platelet function in chronic ischemic heart failure patients. RIC was shown to have no effect on platelet aggregation in response to collagen or arachidonic acid in heart failure patients or platelet turnover, which differs from the effects of a single limb RIC stimulus which was reported to reduce platelet activation (Lanza et al., 2016; Pedersen et al., 2017). However, RIC did increase fibrinolysis in both heart failure and control patients, suggesting it may reduce the risk of thrombosis (Pryds et al., 2017). This effect may be of benefit in 5-FU associated thrombogenicity.

5. Direct myocardial cellular damage by 5-FU and the potential role of RIC on mitochondria

Direct cardiomyocyte and vascular cell damage could also contribute to 5-FU-induced cardiotoxicity. Animal studies have demonstrated dose-dependent pathological changes to cardiomyocytes (Dickson et al., 1999) and endothelial cells (Lamberti et al., 2012), which could be a representation of the initial insult and subsequent 'reaction to injury' that leads to endothelial dysfunction in response to 5-FU. These changes are thought to be caused by induction of apoptosis with an absence of necrosis as opposed to that seen with direct cytotoxicity (Matsubara et al., 1980), as is the mechanism in neoplastic cells. Other animal models have demonstrated specific biochemical changes in cardiomyocytes, including increased oxygen consumption, depletion of high-energy phosphate compounds, and citrate accumulation (Tamatsu et al., 1984; Millart et al., 1992; Durak et al., 2000) occurring independently of changes in blood and oxygen supply. This is thought to be secondary to reduced aerobic efficiency caused by 5-FU-related mitochondrial uncoupling (Tamatsu et al., 1984), which in turn leads to hypoxic cell injury.

Characteristics of mitochondrial dysfunction include changes in the mitochondrial membrane potential, a reduction in the adenosine triphosphate (ATP) level and the inhibition of mitochondrial oxygen consumption (Pieczenik and Neustadt 2007). Excessive formation of reactive oxygen species (ROS) contributes to mitochondrial dysfunction (Litvinova et al., 2015). In particular, superoxide anion generated by the mitochondria, namely by complexes I and III of the electron transport chain (ETC), is the precursor of most ROS and a mediator in oxidative chain reactions (Litvinova et al., 2015). Dismutation of superoxide produces hydrogen peroxide, which in turn may be partially reduced to hydroxyl radicals, causing more damage to various mitochondrial and cellular components (Turrens 2003). Free radical damage to mitochondria may lead to decreased affinity of mitochondrial proteins for substrates or coenzymes (Liu et al., 2003).

Previous studies have suggested an association of RIC with improved mitochondrial function (Kleinbongard et al., 2018). In mitochondria of isolated perfused rat hearts after RIC *in vivo*, there was preserved mitochondrial respiration after ischemia/reperfusion (Ferko et al., 2014). In mitochondria taken from atrial tissue of patients undergoing cardiac surgery with RIC, there was also preserved respiration when the atrial tissue was obtained after aortic cross-clamping, but not when obtained before aortic cross-clamping (Slagsvold et al., 2014a; 2014b). The different conditioning strategies including preconditioning, remote preconditioning, and postconditioning target mitochondria and can improve their function (Boenglet et al., 2018), with potential to ameliorate direct cellular toxicity secondary to mitochondrial dysfunction conferred by 5-FU.

Conclusion and future directions

There remains a pressing need to discover novel and effective therapies that can prevent or ameliorate 5-FU associated cardiotoxicity. The proposed mechanisms underlying cardiotoxicity induced by 5-FU are multifactorial and include vascular endothelial damage followed by thrombus formation, ischemia secondary to coronary artery vasospasm, direct

toxicity on myocardium and thrombogenicity (Chong and Ghosh 2019). There is promising overlap between proposed RIC cardioprotective mechanisms with 5-FU-associated cardiotoxic cellular pathways. Therefore, further studies are needed to investigate the therapeutic potential of RIC for preventing 5-FU coronary artery spasm/cardiotoxicity.

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Conflict of interests

None.

References

Akhtar SS, Salim KP, Bano ZA (1993) Symptomatic cardiotoxicity with high-dose 5-fluorouracil infusion: a prospective study. Oncology 50:441–4.

Akpek G, Hartshorn K (1999) Failure of oral nitrate and calcium channel blocker therapy to prevent 5-fluorouracil-related myocardial ischemia: a case report. Cancer Chemother Pharmacol. 43:157–61.

Alter P, Herzum M, Soufi M, Schaefer JR, Maisch B (2006) Cardiotoxicity of 5-fluorouracil. Cardiovasc Hematol Agents Med Chem. 4:1–6.

Anand AJ (1994) Fluorouracil cardiotoxicity. Ann Pharmacother. 28:374–8.

Arellano M, Malet-Martino M, Martino R, Gires P (1998) The anti-cancer drug 5-fluorouracil is metabolized by the isolated perfused rat liver and in rats into highly toxic fluoroacetate. Br J Cancer. 77:79–86.

Atar A, Korkmaz ME Ozin B (2010) Two cases of coronary vasospasm induced by 5-fluorouracil. Anadolu Kardiyol Derg. 10:461–2.

Balukumar P, Maung-U K, Jagadeesh G (2016) Prevalence and prevention of cardiovascular disease and diabetes mellitus. Pharmacol Res. 113:600–9.

Barac A, Murtagh G, Carver JR, Chen MH, Freeman AM, Herrmann J, Iliescu C, Ky B, Mayer EL, Owkuosa TM, Plana JC, Ryan TD, Rzeszut AK, Douglas PS (2015) Cardiovascular health of patients with cancer and cancer survivors: aroadmap to the next level. J Am Coll Cardiol. 65:2739–46.

Basselin C, Fontanges T, Descotes J, Chevalier P, Bui-Xuan B, Feinard G, Timour Q (2011) 5-Fluorouracil-induced Tako-Tsubo-like syndrome. Pharmacotherapy. 31:226.

Becker K, Erckenbrecht JF, Haussinger D, Frieling T (1999) Cardiotoxicity of the antiproliferative compound fluorouracil. Drugs. 57:475–84.

Benjamin EJ et al. Heart Disease and Stroke Statistics-2019 Update: A Report From the American Heart Association (2019). Circulation. 139(10):e56–528.

Blann AD, Dunmore S (2011) Arterial and venous thrombosis in cancer patients. Cardiol Res Pr. 2011:394740.

Boengler K, Lochnit G, Schulz R (2018) Mitochondria "THE" target of myocardial conditioning. Am J Physiol Circ Physiol [Internet]. Jul 13;315(5):H1215–31. Available from: https://doi.org/10.1152/ajpheart.00124.2018

Bonetti PO, Lerman LO, Lerman A (2003) Endothelial dysfunction: a marker of atherosclerotic risk. Arterioscler Thromb Vasc Biol. 23(2):168–75.

Burger AJ, Mannino S (1987) 5-Fluorouracil-induced coronary vasospasm. Am Heart J. 114:433–6.

Camaro C, Danse PW, Bosker HA (2009) Acute chest pain in a patient treated with capecitabine. Neth Heart J. 17:288–91.

Cancer Research UK [Internet]. Available from: www.cancerresearchuk.org

Cardinale D, Colombo, A Colombo N (2006) Acute coronary syndrome induced by oral capecitabine. Can J Cardiol. 22:251–3.

Chong J, Bulluck H, Yap EP, Ho AF, Boisvert WA, Hausenloy DJ (2018). Remote ischemic conditioning in ST-segment elevation myocardial infarction - an update. Cond Med. 1:13–22.

Chong JH, Ghosh AK (2019) Coronary Artery Vasospasm Induced by 5-fluorouracil: Proposed Mechanisms, Existing Management Options and Future Directions. Interv Cardiol (London, England) 14:89–94.

Chong JH, Bulluck H, Ho AF, Boisvert WA, Hausenloy DJ (2019) Chronic remote ischemic conditioning for cardiovascular protection. Cond Med. 2:164–9.

Coughlin S, Das S, Lee J, Cooper J (2008) Capecitabine induced vasospastic angina. Int J Cardiol. 130:e34--6.

Cwikiel M; Eskilsson J, Wieslander JB, Stjernquist U, Albertsson M (1996) The appearance of endothelium in small arteries after treatment with 5-fluorouracil. An electron microscopic study of late effects in rabbits. Scanning Microsc. 10:805.

Dagenais GR et al. (2019) Variations in common diseases, hospital admissions, and deaths in middle-aged adults in 21 countries from five continents (PURE): a prospective cohort study. Lancet (London, England). 2019 Sep 3.

Dalzell JR, Samuel LM (2009) The spectrum of 5-fluorouracil cardiotoxicity. Anticancer Drugs. 20:79–80.

Dechant C, Baur M, Böck R, Czejka M, Podczeck-Schweighofer A, Dittrich C,Christ G (2012) Acute Reversible Heart Failure Caused by Coronary Vasoconstriction due to Continuous 5-Fluorouracil Combination Chemotherapy. Case Rep Oncol 5:296–301.

de Forni M, Malet-Martino MC, Jaillais P, Shubinski RE, Bachaud , Lemaire L, Canal P, Chevreau C, Carrie D, Soulie P et al. (1992) Cardiotoxicity of high-dose continuous infusion fluorouracil: a prospective clinical study. J Clin Oncol Am Soc Clin Oncol. 10:1795–801.

Demers M, Krause DS, Schatzberg D MK, Voorhees JR, Fuchs TA Scadden DT, Wagner DD (2012) Cancers predispose neutrophils to release extracellular DNA traps that contribute to cancer-associated thrombosis. Proc Natl Acad Sci U S A. 109:13076–81.

Dickson EW, Reinhardt CP, Renzi FP, Becker RC PWA, SO H (1999) Ischemic preconditioning may be transferable via whole blood transfusion: preliminary evidence. J Thromb Thrombolysis. 8:123–9.

Durak I, Karaayvaz M, Kavutcu M, Cimen MY, Kacmaz M, Buyukkocak S, Ozturk HS (2000) Reduced antioxidant defense capacity in myocardial tissue from guinea pigs treated with 5-fluorouracil. J Toxicol Environ Health A. 59:585–9.

Eskilsson J, Albertsson M (1990) Failure of preventing 5-fluorouracil cardiotoxicity by prophylactic treatment

with verapamil. Acta Oncol. 29:1001–3.
Fajardo LF, Stewart JR (1973) Pathogenesis of radiation-induced myocardial fibrosis. Lab Invest. 29:244–55.
Ferko M, Kancirova I, Jasova M, Carnicka S, Murarikova M, Waczulikova I, Sumbalova Z, Kucharska J, Ulicna O, Ravingerová T, Ziegelhöffer A (2014) Remote ischemic preconditioning of the heart: protective responses in functional and biophysical properties of cardiac mitochondria. Physiol Res. 63 Suppl 4:S469-78.
Floyd JD, Nguyen DT, Lobins RL, Bashir Q, Doll DC, Perry MC (2005) Cardiotoxicity of cancer therapy. J Clin Oncol. 23:7685–96.
Freeman NJ, Costanza ME (1988) 5-fluorouracil-associated cardiotoxicity. Cancer. 61:36–45.
Frickhofen N, Beck FJ, Jung B, Fuhr HG, Andrasch H, Sigmund M (2002) Capecitabine can induce acute coronary syndrome similar to 5-fluorouracil. Ann Oncol. 13:797–801.
Ghosh AK, Walker JM (2017) Cardio-Oncology - A new subspecialty with collaboration at its heart. Indian Heart J. 69(4):556–62.
Ghosh AK, Crake T, Manisty C, Westwood M (2018) Pericardial Disease in Cancer Patients. Curr Treat Options Cardiovasc Med. 20(7):60.
Giza DE, Boccalandro F, Lopez-Mattei J, Iliescu G, Karimzad K, Kim P, Iliesch C (2017) Ischemic Heart Disease: Special Considerations in Cardio-Oncology. Curr Treat Options Cardiovasc Med. 19:37.
Grem JL (2000) 5-Fluorouracil: forty-plus and still ticking. A review of its preclinical and clinical development. Invest New Drugs. 18:299–313.
Grunwald MR, Howie L, Diaz LA (2012) Takotsubo cardiomyopathy and Fluorouracil: case report and review of the literature. J Clin Oncol. 30:e11-4.
Guo X-D, Harold N, Saif MW, Schuler B, Szabo E, Hamilton JM, Manohan BP, Quinn MG, Cliatt J, Nguyen D, Grollman F, Thoman RR, McQuigan EA, Wilson R, Takimoto CH, Grem JL (2003) Pharmacokinetic and pharmacodynamic effects of oral eniluracil, fluorouracil and leucovorin given on a weekly schedule. Cancer Chemother Pharmacol. 52:79–85.
Hasdai D, Cannan CR, Mathew V, Holmes DRJ, Lerman A (1996) Evaluation of patients with minimally obstructive coronary artery disease and angina. Int J Cardiol. 53:203–8.
Hasdai D, Gibbons RJ, Holmes DRJ, Higano ST, Lerman A (1997) Coronary endothelial dysfunction in humans is associated with myocardial perfusion defects. Circulation. 96:3390–5.
Hasdai D, Holmes DRJ, Higano ST, Burnett JCJ, Lerman A (1998) Prevalence of coronary blood flow reserve abnormalities among patients with nonobstructive coronary artery disease and chest pain. Mayo Clin Proc. 73:1133–40.
Hausenloy DJ, Yellon DM (2010) The second window of preconditioning (SWOP) where are we now? Cardiovasc drugs Ther. 24:235–54.
Heistad DD, Armstrong ML, Marcus ML, Piegors DJ, Mark AL (1984) Augmented responses to vasoconstrictor stimuli in hypercholesterolemic and atherosclerotic monkeys. Circ Res. 54:711–8.
Holubec LJ, Topolcan O, Finek J, Salvet J, Svoboda T, Svobodova S, Mrazkova P, Ludvikova M (2007) Dynamic monitoring of cardio-specific markers and markers of thyroid gland function in cancer patients--a pilot study. Anticancer Res. 27:1883–6.
Iliescu CA, Grines CL, Herrmann J YE, Cilingiroglu M, Charitakis K, Hakeem A, Toutouzas KP, Leesar MA, Marmagkiolis K (2016) SCAI expert consensus statement: evaluation, management, and special considerations of cardio-oncology patients in the cardiac catheterization laboratory (endorsed by the cardiological society of India, and sociedad Latino Americana de Cardiologia interve. Catheter Cardiovasc Interv Off J Soc Card Angiogr Interv. 87:E202-23.
Jensen SA, Hasbak P, Mortensen J, Sorensen JB (2010) Fluorouracil induces myocardial ischemia with increases of plasma brain natriuretic peptide and lactic acid but without dysfunction of left ventricle. J Clin Oncol. 28:5280–6.
Jensen SA, Sorensen JB (2006) Risk factors and prevention of cardiotoxicity induced by 5-fluorouracil or capecitabine. Cancer Chemother Pharmacol. 58:487–93.
Jensen SA, Sorensen JB (2012) 5-fluorouracil-based therapy induces endovascular injury having potential significance to development of clinically overt cardiotoxicity. Cancer Chemother Pharmacol. 69:57–64.
Jones H, Hopkins N, Bailey TG, Green DJ, Cable NT, Thijssen DH (2014) Seven-day remote ischemic preconditioning improves local and systemic endothelial function and microcirculation in healthy humans. Am J Hypertens. 27:918–25.
Kelly C, Bhuva N, Harrison M, Budkley A, Saunders M (2013) Use of raltitrexed as an alternative to 5-fluorouracil and capecitabine in cancer patients with cardiac history. Eur J Cancer. 49:2303–10.
Kharbanda RK, Mortensen UM, White PA, Kristiansen SB, Schmidt MR, Hoschtitzky JA, Vogel M, Sorensen K, Redington AN, MacAllister R (2002) Transient limb ischemia induces remote ischemic preconditioning *in vivo*. Circulation. 106:2881–3.
Kimura M, Ueda K, Goto C, Jitsuiki D, Nishioka K, Umemura T, Noma K, Yoshizuma M, Chayama K, HIgashi Y(2007) Repetition of ischemic preconditioning augments endothelium-dependent vasodilation in humans: role of endothelium-derived nitric oxide and endothelial progenitor cells. Arterioscler Thromb Vasc Biol. 27:1403–10.
Kinhult S, Albertsson M, Eskilsson J, Cwikiel M (2001) Antithrombotic treatment in protection against thrombogenic effects of 5-fluorouracil on vascular endothelium: a scanning microscopy evaluation. Scanning. 23:1–8.
Kleiman NS, Lehane DE, Geyer CE, Pratt CM, Young JB (1987) Prinzmetal's angina during 5-fluorouracil chemotherapy. Am J Med. 82:566.
Kleinbongard P, Gedik N, Kirca M, Stoian L, Frey U, Zandi A, Thielmann M, Jakob H, Peters J, Kamler M, Heusch G (2018) Mitochondrial and Contractile Function of Human Right Atrial Tissue in Response to Remote Ischemic Conditioning. J Am Heart Assoc. 7:e009540.
Koca D, Salman T, Unek IT, Oztop I, Ellidokuz H, Eren M, Yilmaz U (2011) Clinical and electrocardiography changes in patients treated with capecitabine. Chemotherapy. 57:381–7.
Kosmas C, Kallistratos MS, Kopterides P, Syrios J, Skopelitis H, Mylonakis N, Karabelis A, Tsavaris N (2008) Cardiotoxicity of fluoropyrimidines in different schedules of administration: a prospective study. J Cancer Res Clin Oncol. 134:75–82.
Kuropkat C, Griem K, Clark J, Rodriguez ER, Hutchinson J, Taylor SG (1999) Severe cardiotoxicity during 5-fluorouracil chemotherapy: a case and literature report. Am J Clin Oncol. 22:466–70.
Labianca R, Beretta G, Clerici M, Fraschini P Luporini G (1982) Cardiac toxicity of 5-fluorouracil: a study on 1083

patients. Tumori. 68:505–10.
Lamberti M, Porto S, Marra M, Zappavigna S, Grimaldi A, Feola D, Pesce D, Naviglio S, Spina A, Sannolo N, Caraglia M (2012) 5-Fluorouracil induces apoptosis in rat cardiocytes through intracellular oxidative stress. J Exp Clin Cancer Res. 31:60.
Lanza GA, Stazi A, Villano A, Torrini F, Milo M, Laurito M, Flego D, Aurigemma C, Liuzzo G, Crea F (2016) Effect of Remote Ischemic Preconditioning on Platelet Activation Induced by Coronary Procedures. Am J Cardiol. 117:359–65.
Leal YA, Fernadez-Garrote LM, Mohar-Betancourt A, Meneses-García A (2016) The importance of registries in cancer control. Salud Publica Mex. 58:309–16.
Litvinova L, Atochin DN, Fattakhov N, Vasilenko M, Zatolokin P, Kirienkova E (2015) Nitric oxide and mitochondria in metabolic syndrome [Internet]. Vol. 6, Frontiers in Physiology . 2015. p. 20. Available from: https://www.frontiersin.org/article/10.3389/fphys.2015.00020
Liu C-S, Tsai C-S, Kuo C-L, Chen H-W, Lii C-K, Ma YS, Wei YW (2003) Oxidative stress-related alteration of the copy number of mitochondrial DNA in human leukocytes. Free Radic Res. 37:1307–17.
Lopez JA, Armstrong ML, Piegors DJ, Heistad DD (1989) Effect of early and advanced atherosclerosis on vascular responses to serotonin, thromboxane A2, and ADP. Circulation. 79:698–705.
Lu JI, Carhart RL, Graziano SL Gajra A (2006) Acute coronary syndrome secondary to fluorouracil infusion. J Clin Oncol Off J Am Soc Clin Oncol. 24:2959–60.
Luca MC, Liuni A, McLaughlin K, Gori T, Parker JD (2013) Daily ischemic preconditioning provides sustained protection from ischemia-reperfusion induced endothelial dysfunction: a human study. J Am Heart Assoc. 2(1):e000075.
Lucas G, Marcu A, Piano M, Grosvenor W, Mold F, Maguire R, Ream E (2017) Cancer survivors' experience with telehealth: a systematic review and thematic synthesis. J Med Internet Res. 19:e11.
Luwaert RJ, Descamps O, Majois F, Chaudron JM Beaudiun M (1991) Coronary artery spasm induced by 5-fluorouracil. Eur Hear J. 12:468–70.
Malet-Martino M, Jolimaitre P, Martino R (2002) The prodrugs of 5-fluorouracil Curr Med Chem Anticancer Agents 2:267–310.
Marber MS, Latchman DS, Walker JM, Yellon DM (1993) Cardiac stress protein elevation 24 hours after brief ischemia or heat stress is associated with resistance to myocardial infarction. Circulation. 88:1264–72.
Marsh JC, Catalano P, Huang J, Graham DL, Cornfeld MJ, O'Dwyer PJ, Benson AB (2002) Eastern Cooperative Oncology Group Phase II Trial (E4296) of Oral 5-Fluorouracil and Eniluracil as a 28-Day Regimen in Metastatic Colorectal Cancer. Clin Colorectal Cancer [Internet]. 2:43–50. Available from: https://doi.org/10.3816/CCC.2002.n.010
Matsubara I, Kamiya J, Imai S (1980) Cardiotoxic effects of 5-fluorouracil in the guinea pig. Jpn J Pharmacol. 30:871–9.
May D, Wandl U, Becher R, Niederle N, Schmidt CG (1990) Kardinale nebenwirkungen von 5-fluorouraci. Dtsch Med Wochenschr. 115:618–21.
Millart H, Brabant L, Lorenzato M, Lamiable D, Albert O, Choisy H (1992) The effects of 5-fluorouracil on contractility and oxygen uptake of the isolated perfused rat heart. Anticancer Res. 12:571–6.
Mizuno Y, Hokamura Y, Kimura T, Kimura Y, Kaikita K, Yasue H (1995) A case of 5-fluorouracil cardiotoxicity simulating acute myocardial infarction. Jpn Circ J. 59:303–7.
Moser EC, Noordijk EM, van Leeuwen FE le CS, Baars JW, Thomas J, Carde P, Meerwaldt JH, van Glabbeke M, Kluin-Nelemans HC (2006) Long-term risk of cardiovascular disease after treatment for aggressive non-Hodgkin lymphoma. Blood. 107:2912–9.
Mosseri M, Fingert HJ, Varticovski L, Chokshi S, Isner JM (1993) *In vitro* evidence that myocardial ischemia resulting from 5-fluorouracil chemotherapy is due to protein kinase C-mediated vasoconstriction of vascular smooth muscle. Cancer Res. 53:3028–33.
Muneoka K, Shirai Y, Yokoyama N, Wakai T, Hatakeyama K (2005) 5-Fluorouracil cardiotoxicity induced by alpha-fluoro-beta-alanine. Int J Clin Oncol. 10:441–3.
Naib T, Steingart RM Chen CL (2011) Sorafenib-associated multivessel coronary artery vasospasm. Herz. 36:348–51.
Ng M, Cunningham D Norman AR (2005) The frequency and pattern of cardiotoxicity observed with capecitabine used in conjunction with oxaliplatin in patients treated for advanced colorectal cancer (CRC). Eur J Cancer. 41:1542–1546.
Oleksowicz L, Bruckner HW (1988) Prophylaxis of 5-fluorouracil-induced coronary vasopasm with calcuim channel blockers. Am J Med. 85:750.
Pai VB Nahata M (2000) Cardiotoxicity of chemotherapeutic agents: incidence, treatment and prevention. Drug Saf. 22:263–302.
Patel B, Kloner RA, Ensley J, Al-Sarraf M, Kish J Wynne J (1987) 5-fluorouracil cardiotoxicity: left ventricular dysfunction and effect of coronary vasodilators. Am J Med Sci. 294:238–43.
Pedersen CM, Cruden NL, Schmidt MR, Lau C, Botker HE, Kharbanda RK, Newby DE (2011) Remote ischemic preconditioning prevents systemic platelet activation associated with ischemia-reperfusion injury in humans. Vol. 9, Journal of thrombosis and haemostasis : JTH. England; p. 404–7.
Pieczenik SR, Neustadt J (2007) Mitochondrial dysfunction and molecular pathways of disease. Exp Mol Pathol. 83:84–92.
Polk A, Vistisen K, Vaage-Nilsen M, Nielsen DL (2014) A systematic review of the pathophysiology of 5-fluorouracil-induced cardiotoxicity. BMC Pharmacol Toxicol. 15:1–11.
Polk A, Vaage-Nilsen M, Vistisen K, Nielsen DL (2013) Cardiotoxicity in cancer patients treated with 5-fluorouracil or capecitabine: a systematic review of incidence, manifestations and predisposing factors. Cancer Treat Rev. 39:974–84.
Porta C, Moroni M, Ferrari S, Nastasi G (1998) Endothelin-1 and 5-fluorouracil-induced cardiotoxicity. Neoplasma. 45(2):81–2.
Pryds K, Nielsen RR, Jorsal A, Hansen MS, Ringgaard S, Refsgaard J, Kim WY, Petersen AK, Bøtker HE, Schmidt MR (2017) Effect of long-term remote ischemic conditioning in patients with chronic ischemic heart failure. Basic Res Cardiol. 112:67.
Pryds K, Kristiansen J, Neergaard-Petersen S, Nielsen RR, Schmidt MR, Refsgaard J, Kristensen SD1, Bøtker HE, Hvas AM, Grove EL (2017) Effect of long-term remote ischaemic conditioning on platelet function and fibrinolysis in patients with chronic ischaemic heart failure. Thromb Res. 153:40–6.
Przyklenk K, Bauer B, Ovize M, Kloner RA Whittaker P (1993) Regional ischemic 'preconditioning'' protects remote virgin myocardium from subsequent sustained coronary occlusion. Circulation. 87:893–9.
Pyke KE, Tschakovsky ME (2005) The relationship between

shear stress and flow-mediated dilatation: implications for the assessment of endothelial function. J Physiol. 568(Pt 2):357–69.

Reddy KG, Nair RN, Sheehan HM, Hodgson JM (1994) Evidence that selective endothelial dysfunction may occur in the absence of angiographic or ultrasound atherosclerosis in patients with risk factors for atherosclerosis. J Am Coll Cardiol. 23:833–43.

Saif MW, Shah MM Shah AR (2009) Fluoropyrimidine-associated cardiotoxicity: revisited. Expert Opin Drug Saf. 8:191–202.

Salepci T, Seker M, Uyarel H, Gumus M, Bilici A, Ustaalioglu BB OA, Sonmez B, Orcun A, Ozates M, Irmak R Yaylaci M (2010) 5-Fluorouracil induces arterial vasoconstrictions but does not increase angiotensin II levels. Med Oncol. 27:416–20.

Sara JD, Kaur J, Khodadadi R, Rehman M, Lobo R, Chakrabarti S, Herrmann J, Lerman A, Grothey A (2018) 5-fluorouracil and cardiotoxicity: a review. Ther Adv Med Oncol [Intern et].10:1758835918780140–1758835918780140. Available from: https://www.ncbi.nlm.nih.gov/pubmed/29977352

Sasson Z, Morgan CD, Wang B, Thomas G, MacKenzie B, Platts ME (1994) 5-Fluorouracil related toxic myocarditis: case reports and pathological confirmation. Can J Cardiol.10(8):861–4.

Schwartz BG, Economides C, Mayeda GS, Burstein S Kloner RA (2010) The endothelial cell in health and disease: its function, dysfunction, measurement and therapy. Int J Impot Res. 22:77–90.

Siegel R, DeSantis C, Virgo K, Stein K MA, Smith T, Cooper D, Gansler T, Lerro C, Fedewa S, Lin C, Leach C, Cannady RS, Cho H, Scoppa S, Hachey M, Kirch R, Jemal A, Ward E (2012) Cancer treatment and survivorship statistics. CA Cancer J Clin. 2012;62(4):220–41.

Slagsvold KH, Moreira JBN, Rognmo O, Hoydal M, Bye A, Wisloff U, Wahba A (2014) Remote ischemic preconditioning preserves mitochondrial function and activates pro-survival protein kinase Akt in the left ventricle during cardiac surgery: a randomized trial. Int J Cardiol. 177:409–17.

Slagsvold KH, Rognmo O, Hoydal M, Wisloff U, Wahba A (2014) Remote ischemic preconditioning preserves mitochondrial function and influences myocardial microRNA expression in atrial myocardium during coronary bypass surgery. Circ Res. 114:851–9.

Spasojevic I, Maksimovic V, Zakrzewska J, Bacic G (2005) Effects of 5-fluorouracil on erythrocytes in relation to its cardiotoxicity: membrane structure and functioning. J Chem Inf Model. 45:1680–5.

Spasojević I, Jelić S, Zakrzewska J, Bacić G (2008) Decreased oxygen transfer capacity of erythrocytes as a cause of 5-fluorouracil related ischemia. Molecules [Internet]. 14:53–67. Available from: https://www.ncbi.nlm.nih.gov/pubmed/19127237

Stewart T, Pavlakis N, Ward M (2010) Cardiotoxicity with 5-fluorouracil and capecitabine: more than just vasospastic angina. Intern Med J. 40:303–7.

Shoemaker LK, Arora U, Rocha, Lima CM (2004) 5-fluorouracil-induced coronary vasospasm. Cancer Control. 11:46–9.

Sudhoff T, Enderle MD, Pahlke M, Petz C, Teschendorf C Graeven U, Schmiegel W (2004) 5-Fluorouracil induces arterial vasocontractions. Ann Oncol Off J Eur Soc Med Oncol. 15:661–4.

Suwaidi JA, Hamasaki S, Higano ST, Nishimura RA, Holmes DRJ, Lerman A (2000) Long-term follow-up of patients with mild coronary artery disease and endothelial dysfunction. Circulation. 101:948–54.

Tajik R, Saadat H, Taherkhani M, Movahed MR (2010) Angina induced by 5-fluorouracil infusion in a patient with normal coronaries. Am Hear Hosp J. 8:E111--E112.

Tamatsu H, Nakasawa M, Imai S, Watari H (1984) 31P-Topical Nuclear Magnetic Resonance (31P-TMR) Studies of Cardiotoxic Effects of 5-Fluorouracil (5-FU) and 5'-Deoxy-5-Fluorouridine (5'-DFUR). Jpn J Pharmacol. 34:375-9.

Tsavaris N, Kosmas C, Vadiaka M, Efremidis M ZA, et al. Beldecos D, Sakelariou D, Koufos C, Stamatelos G (2002) Cardiotoxicity following different doses and schedules of 5-fluorouracil administration for malignancy – a survey of 427 patients. Med Sci Monit Int Med J Exp Clin Res. 8(:P151-7.

Tsibiribi P, Bui-Xuan C, Bui-Xuan B, Lombard-Bohas C, Duperret S BM, Tabib A, Maujean G, Descotes J, Timour Q (2006) Cardiac lesions induced by 5-fluorouracil in the rabbit. Hum Exp Toxicol. 25:305–9.

Turrens JF (2003) Mitochondrial formation of reactive oxygen species. J Physiol. 552(Pt 2):335–44.

Van Cutsem E, Hoff PM, Blum JL, Abt M, Osterwalder B (2002) Incidence of cardiotoxicity with the oral fluoropyrimidine capecitabine is typical of that reported with 5-fluorouracil. Annals of oncology: Off J Eur Soc Med Oncol. 13:484–5.

Vita JA, Treasure CB, Nabel EG, McLenachan JM, Fish RD, Yeung AC, Vekshtein VI, Selwyn AP, Ganz P (1990) Coronary vasomotor response to acetylcholine relates to risk factors for coronary artery disease. Circulation. 81:491–7.

Wacker A, Lersch C, Scherpinski U, Reindl L Seyfarth M (2003) High incidence of angina pectoris in patients treated with 5-fluorouracil. A planned surveillance study with 102 patients. Oncology. 65:108–12.

Weaver KE, Foraker RE, Alfano CM, Rowland JH A, NK, Bellizzi KM, Hamilton AS, Oakley-Girvan I, Keel G, Aziz NM (2013) Cardiovascular risk factors among long-term survivors of breast, prostate, colorectal, and gynecologic cancers: a gap in survivorship care? J cancer Surviv Res Pract. 7:253–61.

WHO. (2018, September 12).Cancer. Available from: https://www.who.int/en/news-room/fact-sheets/detail/cancer

Yip D, Karapetis C, Strickland AH, Steer C, Holford C, Knight S, Harper P (2003) A dose-escalating study of oral eniluracil/5-fluorouracil plus oxaliplatin in patients with advanced gastrointestinal malignancies. Ann Oncol [Internet]. 14:864–6. Available from: https://doi.org/10.1093/annonc/mdg254

Yudkin JS, Stehouwer CD, Emeis JJ, Coppack SW (1999) C-reactive protein in healthy subjects: associations with obesity, insulin resistance, and endothelial dysfunction: a potential role for cytokines originating from adipose tissue? Arterioscler Thromb Vasc Biol. 19:972–8.