Canola oil rich in oleic acid improves diastolic heart function in diet-induced obese rats

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Abstract Obesity is a leading cause of cardiovascular disease. It directly affects heart structure and function and contributes to heart failure. Diet is a major factor involved in the development of obesity along with genetic factors. We examined the effects of monounsaturated and polyunsaturated fatty acid-rich oils on cardiac structure and function in the diet-induced rodent model of obesity (DIO). Obese prone (OP) rats were fed a high-fat diet (HF; 55% of kcal) for 12 weeks; Sprague–Dawley rats fed commercial chow served as control. Echocardiography was performed to assess the cardiac structure and function in all rats at 12 weeks. OP rats fed the HF diet showed significant impairment in diastolic function compared to control rats. The HF diet containing high oleic canola oil significantly improved diastolic function of OP rats compared to the HF diet with lard. In conclusion, canola oil rich in oleic acid, when incorporated into an HF diet, prevents the development of diastolic dysfunction in DIO rats.

Keywords Diet-induced obesity • Edible oil • Diastolic dysfunction

Introduction

Obesity is one of the most widespread diseases in developed countries and in developing countries among young and adult populations [1, 2]. The prevalence of overweight and obesity has increased more than twofold since 1980; 2.1 billion adults were overweight in 2013, and 600 million were clinically obese [1–3]. Obesity is associated with physiological changes that cause or contribute to a wide variety of metabolic diseases, including type 2 diabetes, hypertension and cardiovascular disease (CVD) [4]. Because of its deleterious effects on various CVD risk factors and its adverse direct effects on cardiac structure and function, obesity is a major contributor to CVD [5]. In this scenario, obesity therapy should be designed to reduce CVD risk and mortality.

Several strategies are being used to combat obesity and its associated complications [6]. Lifestyle modifications based on exercise and calorie restriction appear to be the best solution; however, they are hard to maintain, and result in only a modest weight loss which is not sufficient for individuals who are morbidly obese. Bariatric surgery is the most successful strategy available, but it comes with risks and complications and remains an expensive option for most people. Pharmacological agents offer a non-invasive, simple, and accessible alternative. However, although reasonably effective, pharmacological agents are often accompanied by side-
effects that render them less valuable. Here arises the need for alternative strategies to prevent/manage obesity and its associated complications. One such avenue may be the use of more economical foods that are regularly available and possess health benefits. While commonly used edible oils have been widely studied for their beneficial effects for CVD management [7], their application to CVD as a consequence of obesity is largely unexplored. In this regard, it is imperative to identify the most useful edible oil that has superior properties in preventing obesity-associated CVD and CVD risk factors. However, no study has examined the comparative effects of long-term consumption of edible oils on heart structure and function in an animal model of diet-induced obesity (DIO). Given the indisputable role of diet and the polygenic nature of obesity in humans, the DIO model is considered the most appropriate for studying human obesity and obesity-related CVD [8]. In this study, we examined the effects of monounsaturated and polyunsaturated fatty acid-rich oils on cardiac structure and function in DIO rats. The oils included in the diet for this study were high oleic canola oil, the most widely used oil in industrial food production, conventional canola oil, a conventional canola oil-flax oil blend (3:1), high linoleic acid safflower oil, and soybean oil, which were compared with a lard-based control diet that is typically used for the DIO model.

**Materials and methods**

The experimental protocols were approved by the University of Manitoba Animal Care Committee and are in agreement with the Canadian Council on Animal Care and Use of Experimental Animals.

**Animal model and diet**

Six-week-old selectively bred obese prone (OP) and Sprague–Dawley (SD) rats were purchased from Charles River, St Constant, Quebec, Canada. After a 12- to 17-day acclimation period, one group of OP rats was fed a high-fat lard-based diet and another five groups of OP rats were fed high-fat diets (HF, energy from fat 55%, carbohydrate 30% and protein 15%) formulated with high oleic canola, canola, canola-flaxseed (3:1 blend), safflower, or soybean oil, respectively, for a period of 12 weeks. The diet formulations and fatty acid composition of the diets are shown in Tables 1 and 2, respectively. The reference control group SD rats were fed commercial chow (Prolab RMH 3000; energy from fat 14%) for 12 weeks. Metabolic parameters for the OP rats fed the HF diets with the various edible oils have been previously reported [9, 10].

| Table 1 Diet formulations |
|---------------------------|
| g/kg diet | L | HC | C | CF | SB | SF |
|---------------------------|
| Cornstarch | 209 | 209 | 209 | 209 | 209 | 209 |
| Maltodextrin | 69.4 | 69.4 | 69.4 | 69.4 | 69.4 | 69.4 |
| Sucrose | 100 | 100 | 100 | 100 | 100 | 100 |
| Cellulose | 63.8 | 63.8 | 63.8 | 63.8 | 63.8 | 63.8 |
| Casein | 186.2 | 186.2 | 186.2 | 186.2 | 186.2 | 186.2 |
| High oleic canola oil | 0 | 308.3 | 0 | 0 | 0 | 0 |
| Canola oil | 0 | 0 | 308.3 | 231.2 | 0 | 0 |
| Flaxseed oil | 0 | 0 | 0 | 77.1 | 0 | 0 |
| Safflower oil | 0 | 0 | 0 | 0 | 0 | 308.3 |
| Soybean oil | 28.5 | 0 | 0 | 0 | 308.3 | 0 |
| Lard | 279.8 | 0 | 0 | 0 | 0 | 0 |
| AIN-93G-MX* | 44.6 | 44.6 | 44.6 | 44.6 | 44.6 | 44.6 |
| AIN-93-VX* | 12.7 | 12.7 | 12.7 | 12.7 | 12.7 | 12.7 |
| t-Cystine | 3 | 3 | 3 | 3 | 3 | 3 |
| Choline bitartrate | 3.2 | 3.2 | 3.2 | 3.2 | 3.2 | 3.2 |
| t-Butylhydroquinone | 0.037 | 0.037 | 0.037 | 0.037 | 0.037 | 0.037 |

*L* lard, *HC* high-oleic canola, *C* conventional canola, *CF* conventional canola/flax, *SB* soybean, *SF* safflower

| Table 2 Fatty acid composition of diets |
|---------------------------|
| Fatty acid* | L | HC | C | C/F | SB | SF |
|---------------------------|
| Total SFA | 49 | 7 | 7 | 8 | 15 | 10 |
| PA (C16:0) | 24 | 4 | 4 | 4 | 10 | 6 |
| SA (C18:0) | 21 | 2 | 2 | 2 | 4 | 3 |
| Total MUFA | 42 | 78 | 66 | 54 | 21 | 17 |
| OA (C18:1) | 39 | 76 | 64 | 53 | 20 | 16 |
| Total PUFA | 9 | 16 | 27 | 38 | 63 | 73 |
| LA (C18 2n6) | 8 | 14 | 19 | 18 | 54 | 73 |
| ALA (C18 3n3) | 1 | 2 | 8 | 20 | 9 | 0.2 |
| LA/ALA | 8 | 7 | 2 | 1 | 6 | 365 |
| Total n–6 | 8 | 14 | 19 | 19 | 54 | 73 |
| Total n–3 | 1 | 2 | 8 | 20 | 9 | 0.4 |
| n–6/n–3 | 7 | 7 | 2 | 1 | 6 | 183 |

*ALA* α-linolenic acid, *LA* linoleic acid, *OA* oleic acid, *PA* palmitic acid, *SA* stearic acid, *L* lard, *HC* high oleic canola, *C* conventional canola, *CF* conventional canola/flax, *SB* soybean, *SF* safflower

| Assessment of cardiac structure and function in vivo by echocardiography |
|---------------------------|
| Cardiac structure and function were assessed at week 12 by transthoracic echocardiography according to our standard |
procedure with a Sonos 5500 ultrasound system (Agilent Technologies, Andover, MA, USA) equipped with a 12-MHz (s12) transducer [11, 12]. All echocardiography measurements were carried out on anesthetized animals. Structural parameters [left ventricular posterior wall thickness (LVPW) and interventricular septum (IVS) thickness, left ventricular internal dimension (LVID)], and systolic function [left ventricular ejection fraction (EF)] were analyzed by 2D-guided M-Mode echocardiography, while diastolic function [isovolumic relaxation time (IVRT)] was measured by pulse wave Doppler echocardiography.

Measurement of oxidative stress marker

Oxidative stress was determined by measuring the concentration of lipid peroxidation marker, malondialdehyde (MDA), in heart tissues from all groups. The quantification was done using a lipid peroxidation (MDA) assay kit (Abcam, Cambridge, MA, USA) by following the manufacturer’s instructions [12]. A portion of LV tissue was homogenized in lysis buffer and the supernatant was collected after centrifugation and used for protein estimation and assay. The MDA concentration was calculated as amount of MDA present in the sample/protein concentration of the sample, and values are reported in nmol/mg protein.

Measurement of inflammation marker

The level of inflammation marker, tumor necrosis factor-α (TNF-α) was quantified using a TNF-α ELISA kit (Thermo Fisher Scientific, MA, USA) by following the manufacturer’s instructions [11]. The heart tissue sample was prepared as mentioned in the MDA assay method. The TNF-α concentration was calculated as amount of TNF-α present in the sample/protein concentration of the sample, and values are reported in pg/mg protein.

Statistics

Data were analyzed by one-way ANOVA and Duncan’s multiple range test was used for post hoc means testing using Statistical Analysis Software (SAS; version 9.1.3, SAS Institute Inc., Cary, NC). Data is reported as mean values with standard error of the mean (SEM). Differences were considered statistically significant at \( p < 0.05 \).

Results

Heart mass (assessed by heart to tibia length ratio, Fig. 1) was significantly higher in the SD reference group compared to all other groups at 12 weeks. Cardiac wall thickness (assessed by LVPWd, and IVSd, Fig. 2a, b) was comparable between groups at 12 weeks. In contrast, LVIDd was significantly higher in the SD reference group when compared to other groups except the canola/flax oil group (Fig. 2c). EF was significantly lower in the SD reference group compared to other groups at 12 weeks (Fig. 3a), which is consistent with an earlier study reported by us [11]. IVRT was significantly higher in OP rats fed the HF diet containing the various oil sources compared to the SD reference group (Fig. 3b). Our data showed that a high oleic canola oil HF diet prevented the increase in IVRT in OP rats compared to OP rats fed other oils. In order to understand the potential mechanism of HF diet induced cardiac dysfunction that we observed in our study, we examined the levels of oxidative stress and inflammatory markers as oxidative stress and inflammation are known to play a major role in the genesis of cardiac abnormalities. At 12 weeks, the level of MDA (a widely used marker of oxidative stress) was not significantly different between HF fed rats and SD rats fed with a standard diet (commercial chow) or HF+ dietary oil fed rats (Fig. 4a). In contrast, the level of TNF-α (inflammatory marker) was significantly higher in HF fed rats in comparison to SD rats fed with standard diet for 12 weeks (Fig. 4b). In addition, the canola oil and soybean treated groups had significantly lower levels of TNF-α when compared to HF fed rats, whereas the high oleic canola treatment group showed a trend \( (p = 0.059) \) towards lowering the inflammation marker TNF-α in the heart tissue of HF fed rats.

Discussion

The results of this study show that HF feeding for 12 weeks did not result in cardiac hypertrophy in OP rats as we did not observe a significant increase in heart to tibia length.
ratio and wall thickness, which are the indicators of cardiac concentric hypertrophy. Our results also showed that there was no significant dilation of heart (increased LVID which is a marker of cardiac eccentric hypertrophy) due to HF feeding for 12 weeks. Cardiac functional assessment suggested that HF feeding for 12 weeks did not lead to a reduction in ability of the heart to contract and eject blood (reduction in EF). Overall, our data suggests that 12 week HF diet feeding did not result in significant adverse cardiac remodeling and systolic dysfunction. However, OP rats fed the HF diet had significant impairment in the ability of the heart to relax during the diastolic filling phase (as evident by increased IVRT), indicating that diet-induced obesity resulted in diastolic dysfunction as characterized by incomplete filling of the ventricles. We thus demonstrate for the first time that incorporation of high oleic canola oil into a HF diet prevents development of diastolic dysfucntion in DIO rats. Interestingly, other edible oils such as soybean oil and high linoleic safflower oil were not effective in improving diastolic dysfunction. Furthermore, it must be noted that conventional canola oil by itself or in combination with flax oil was not effective in preventing the development of abnormalities in diastolic function in DIO rats. Based on dietary fatty acid composition, these results suggest that the high oleic content in canola oil is cardioprotective, and prevents the development of obesity-induced cardiac complications. It is also possible that the other minor constituents in high oleic canola oil may have contributed in part to the beneficial effects. Future studies are needed to identify the involvement of other factors.

There are limited published studies in which the experimental treatment has used high oleic oil, and
specifically high oleic canola oil, as a strategy to prevent obesity-related (diet induced) cardiac complications. Although high oleic oils such as those from canola and sunflower may have beneficial effects on cardiovascular risk factors such as lowering LDL-cholesterol in hypercholesterolemic individuals [13] or in healthy subjects [14], there is no literature to indicate direct beneficial effects of high oleic oils on heart structure and function in vivo, with the exception of our study. There are reports showing direct anti-apoptotic effects of oleic acid observed in vitro, i.e., in adult cardiomyocytes exposed to TNF-α [15] and palmitic acid [16]. We previously reported that high oleic canola treatment increased the level of the anti-inflammatory cytokine adiponectin in epididymal adipose tissue of HF fed rats [10]. In the current study, there was a trend \( p = 0.059 \) for high oleic canola treatment to lower the inflammation marker TNF-α in heart tissue of HF fed rats, suggesting that this may have contributed to the observed high oleic canola mediated cardioprotection. It must also be noted that the safflower oil and soybean oil used in our study had high linoleic acid content, not high oleic acid (see Table 2). This informs us that it is worthwhile exploring the cardiac health benefits of foods having high oleic acid content. The safflower and soybean industries also produce high oleic acid oils, and further studies are needed to test whether high oleic acid containing oils from different plant sources will be equally effective for preventing CVD risk factors and cardiac impairment.

In conclusion, our discovery has important implications for the edible oil industry because high oleic oils are being promoted for heart health, and the positive results obtained in our study indicates further research is needed on their effects on cardiac structure and function in humans in the context of CVD prevention and management.

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Compliance with ethical standards

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Conflict of interest The authors report no conflict of interest.

Ethical approval The experimental protocols were approved by the University of Manitoba Animal Care Committee and are in agreement with the Canadian Council on Animal Care and Use of Experimental Animals.

Informed consent This article does not contain any studies with human participants performed by any of the authors.

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