International Journal of Sports Science 2020, 10(4): 82-91
DOI: 10.5923/j.sports.20201004.03

The Effects of Cocoa Flavanols on Endothelial Function and Exercise Performance

James M. Heilman, Yunsuk Koh*

Department of Health, Human Performance, and Recreation, Baylor University, Waco, TX, USA

Abstract Cocoa flavanols are a commercially available nutritional supplement and may positively affect endothelial function through multiple physiological mechanisms. The purpose of this study was to evaluate the current status of cocoa flavanols in the literature, specifically in regard to their effects on endothelial function and exercise performance. A total of 27 qualifying studies were extensively examined for the use of cocoa flavanol supplementation and its effects on endothelial function. In addition, the role of cocoa flavanols in both aerobic and anaerobic exercise performance was evaluated. Although a positive effect of cocoa flavanols including the improvement in flow-mediated dilation and blood pressure has been well-documented, there is some variability in terms of the effects of certain doses on different populations. The existing literature offers both direct and theoretical evidence for positive effects on un-trained and moderately trained populations. However, more research is needed in order to better understand the effects of cocoa flavanols on aerobic and anaerobic exercise performance. Cocoa flavanols have the potential to be effective in a number of ways in both healthy and clinical populations. However, upper and lower limits for proper doses should be established for different populations.

Keywords Nutraceuticals, Sport performance, Vascular function

1. Introduction

Among the many commercially available supplements are cocoa flavanols, which are extracted from beans of the cacao tree. [1] Cocoa flavanols have previously been studied in the literature. [2–4] There are many proposed theories regarding their physiological effects on endothelial function that may tie into cardiovascular health and exercise performance. However, not all of the exact mechanisms and reasons for this are known. In addition, there is some variability in terms of the correct dose needed to elicit any of these responses. There are also inconsistencies regarding the proper intake duration to yield significant results. Overall, it is important to address these discrepancies in order to fully understand the current state of cocoa flavanols in the literature. A clear understanding is necessary to observe the effects that a moderate dose may have on endothelial function and exercise performance.

Many physiological mechanisms are thought to contribute to the positive effects of cocoa flavanols on endothelial function. These include decreases in known vasoconstrictors and nitric oxide (NO) scavengers, as well as an increase in the enzyme endothelial nitric oxide synthase (eNOS), which is essential for NO production. [2] Together, these changes lead to a robust increase in NO availability. [2] Cocoa flavanols appear to improve endothelial function through a combination of decreased blood pressure and enhanced flow-mediated dilation (FMD). [3,5–7]

It is also necessary to observe the effects that cocoa flavanol supplementation may have on exercise performance. There is little research on this topic, but there may be positive effects on both aerobic and anaerobic exercise performance. [8] The extent to which cocoa flavanols may aid performance still warrants more research. [8,9] It is important to determine whether or not this lesser-known nutritional supplement can impact various aspects of exercise performance, as well as the populations that may benefit from it. If cocoa flavanols are found to enhance endothelial function, they could be utilized in clinical settings to help lower the risk or severity of hypertension and cardiometabolic disease. [6] If cocoa flavanol supplementation is found to positively influence exercise performance, coaches and athletes could utilize this lesser-known nutritional supplement as a method of enhancing performance in both practice and competition.

* Corresponding author:
Yunsuk_Koh@baylor.edu (Yunsuk Koh)
Published online at http://journal.sapub.org/sports

2. Methods

Both *PubMed* and *Google Scholar* databases were utilized to search for appropriate studies published between 2003 and 2018. Using the following keywords including ‘cocoa flavanols,' 'endothelial function,' and 'exercise,' relevant studies were searched and evaluated. The article screening process was in accordance with the guidelines set forth by the preferred reporting items for systematic reviews and meta-analyses (PRISMA) statement. [10] Studies were evaluated based on a variety of inclusion and exclusion criteria. Accepted studies needed to utilize cocoa-based flavanols, as opposed to other known sources of dietary flavanols (fruits, tea, etc.). In addition, studies needed to include components of endothelial function and/or exercise performance. Finally, studies using both human and animal models were included. Studies were excluded if they did not include relevant information regarding cocoa flavanol supplementation with exercise or with chronic disease related to poor endothelial function. Finally, studies were excluded if supplementation methods were unclear.

3. Results

At the completion of the PRISMA screening process, 27 out of 59 qualifying studies were included for the review. As shown in Figure. 1, a total of 32 studies were excluded for not meeting the abovementioned criteria, such as not including cocoa flavanols (N=17), endothelial function (N=7), or cocoa-based sources of cocoa flavanols (N=8). [6]

Of the 27 studies included in the review, the majority (N=24) exhibited significant results demonstrating how cocoa flavanols enhance endothelial function and exercise performance. A smaller number of studies reported no change following cocoa flavanol supplementation (N=3).

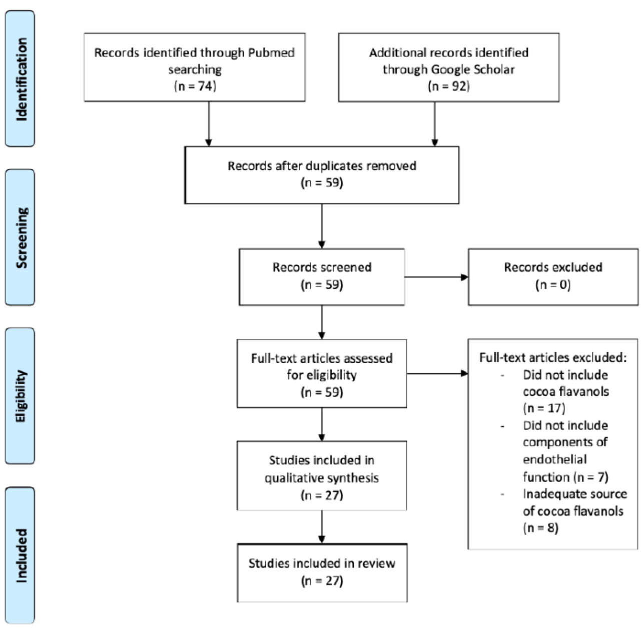

Figure 1. Screening process for eligible studies

4. Discussion

4.1. Cocoa Flavanols and Nitric Oxide

Cocoa flavanols may impact a variety of physiological mechanisms related to endothelial function through an increase in NO availability. [2] Specifically, cocoa flavanols are thought to increase eNOS activity and inhibit a variety of factors, such as superoxide anion, angiotensin-converting enzyme (ACE), and endothelin-1 (ET-1). [2,7,11,12] NO is a well-known vasodilator, causing blood vessel diameter to increase and allowing for a subsequent enhancement in blood flow. [2] Endothelial cells use an amino acid known as L-arginine to form NO. [13,14] eNOS, known as one of the limiting factors of NO availability, catalyzes this reaction. Indeed, there is a direct relationship between eNOS and NO availability. Once NO has been synthesized, it moves into the tunica media of the vasculature via diffusion. [14] NO then binds to the heme group of guanylyl cyclase, an important enzyme that catalyzes the conversion of guanosine triphosphate (GTP) to cyclic guanosine monophosphate (cGMP). [13,14] cGMP acts as a second messenger in the process of vasodilation, signaling vascular muscle tissue to stop contracting. [13] This is the pathway by which NO-mediated vasodilation occurs, and it is essential to understand this pathway when studying the effects of cocoa flavanols on endothelial function. The basic relationship between cocoa flavanols and vasodilation is depicted below in Figure. 2.

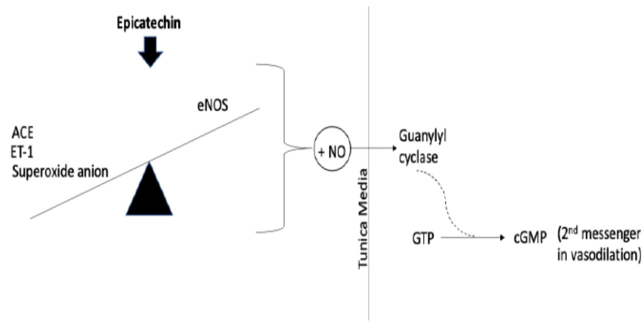

Figure 2. Effects of cocoa flavanol-derived epicatechin on NO availability and vasodilation. ACE = angiotensin-converting enzyme, cGMP = cyclic guanosine monophosphate, eNOS = endothelial nitric oxide synthase, ET-1 = endothelin-1, GTP = guanosine triphosphate, NO = nitric oxide

Cocoa flavanols contain two very important polyphenols that are thought to influence endothelial function: catechin and epicatechin. [2] Epicatechin is thought to play the greater role in endothelial function as it is the most plentiful polyphenol in cocoa powder and other cocoa products. [2,15,16] It is theorized that epicatechin causes a variety of physiological changes that lead to increased availability of NO. [16,17] One such change is that cocoa flavanols (especially epicatechin) cause an increase in eNOS activity. The exact mechanisms behind this enhancement are unclear. However, previous research has proposed that cocoa flavanols may inhibit arginase activity. [17] Arginase plays a role in the breakdown of L-arginine. As mentioned above, L-arginine is used to form NO with the help of eNOS. Therefore, upregulated L-arginine availability allows for eNOS to be much more active in converting L-arginine to NO. [4,17] Other studies have confirmed that cocoa flavanol supplementation does indeed lead to increased eNOS activity, resulting in greater NO availability. [4,13,17] Indeed, it is clear that cocoa flavanols have a significant effect on eNOS.

4.2. Cocoa Flavanols and Oxidants

Cocoa flavanols also appear to impact oxidants contained in endothelial cells, specifically superoxide anion. Superoxide anion's main function is to break down leftover NO within endothelial cells. [2,13] Cocoa-derived epicatechin has been shown to suppress cellular superoxide anion concentration, likely due to its ability to clear free radicals. [13] In addition, further research has speculated that cocoa flavanols may further limit superoxide anion concentration via inhibition of nicotinamide adenine dinucleotide phosphate (NADPH) oxidase, an enzyme involved with superoxide anion-mediated breakdown of NO. [2] Suppression of superoxide anion is yet another mechanism brought about by cocoa flavanols that appears to enhance endothelial function.

4.3. Cocoa Flavanols and ACE

Cocoa flavanols have also been reported to inhibit angiotensin-converting enzyme (ACE), an enzyme that is crucial in the conversion of angiotensin I to angiotensin II. Angiotensin II is a potent vasoconstrictor, which causes smooth muscle to contract and its diameter to decrease. [2,12,18] There is evidence that cocoa flavanols may inhibit ACE activity; thus, there is less vasoconstriction in the blood vessel. [2,5,11,12,19] This may lead to improved endothelial function through an increase in flow-mediated dilation (FMD). [17] There is no definitive evidence of the exact mechanism behind this inhibition. However, Actis-Goretta and colleagues proposed that, while monomers of epicatechin and catechin can inhibit ACE activity, polymers contained in high amounts in flavanol-rich foods (specifically cocoa products) are the most effective. [12] The polymers include more hydroxyl groups than the monomers, meaning that the polymers of epicatechin and catechin can create more hydrogen bonds with ACE. This causes ACE inhibition and a subsequent decrease in vasoconstriction. [7] This potential mechanism is quite relevant, as it brings forth yet another pathway by which cocoa flavanols stimulate enhanced endothelial function.

4.4. Cocoa Flavanols and ET-1

The final proposed physiological mechanism is the inhibition of endothelin-1 (ET-1). ET-1 is a potent vasoconstrictor activated by the onset of tissue inflammation. [11] Located within the endothelium, ET-1 brings about vasoconstriction by causing an increase in superoxide anion, resulting in decreased NO availability within the endothelial cells. [11] There is evidence that cocoa flavanol supplementation (epicatechin in particular) can result in reduced expression of the ET-1 gene by inhibiting its transcription. [11] This in turn reduces the amount of ET-1 within endothelial cells, thereby decreasing the concentration of superoxide anion even further and adding to the strength of aforementioned mechanisms.

4.5. Clinical Significance of Previous Studies

In addition to the mechanistic physiological effects brought about by cocoa flavanols, it is important to observe how previous research has measured possible enhancements in endothelial function and the methodology used to produce significant results. Cocoa flavanols significantly impact both blood pressure and FMD%. [20,21] Both are measurements of endothelial function frequently used in clinical settings. [20] Even though cocoa flavanol supplementation has shown promising results, there are discrepancies within the literature regarding the proper dose and intake period necessary to elicit significant positive effects.

Blood pressure is commonly used in clinical settings as a measurement of endothelial function and the presence of hypertension. [19] Many previous studies have measured the potential effects of cocoa flavanol supplementation on resting blood pressure with interesting results. A previous study conducted by Fraga and colleagues observed that a dose of only 168 mg of cocoa flavanols for 14 days led to a significant decrease (5 mmHg) in diastolic blood pressure (DBP) and a significant decrease (5 mmHg) in mean blood pressure in 18-20 year old male soccer players. [7] This shows that cocoa flavanols can have a positive impact on blood pressure, even in normotensive young adults. In contrast, there is evidence that doses of 33 mg, 372 mg, and 712 mg, respectively, do not have any significant effect on blood pressure in older adults with mild hypertension. [5] The authors of the latter dose response study reported that a dose of only 1052 mg for 6 weeks significantly reduced blood pressure, causing 5.3 mmHg and 3.0 mmHg decreases in systolic blood pressure (SBP) and DBP, respectively. [5] Another similar study supplementing 213 mg of cocoa flavanols for 14 days found no significant change in blood pressure for moderately trained males. [8] However, a study involving healthy males and females aged 35-60 using a 450 mg dose measured after 14 days observed significant decreases in both SBP and DBP of 4.4 and 3.9 mmHg, respectively. [21] These are peculiar results, as one study found significant effects with a low dose for only 14 days, while the other only found significant results with a much higher dose for 6 weeks. Thus, more research must be done in order to develop a clearer dose-dependent relationship between cocoa flavanols and significant reductions in blood pressure. [8]

There is even further discrepancy within the literature between different meta-analyses on the subject. According to Ried and colleagues, a dose range of 49-3,675 mg of cocoa flavanols resulted in an average decrease of 2 mmHg in both SBP and DBP over a span of at least two weeks. [22] In addition, another meta-analysis reported that an even smaller dose range (5-174 mg) resulted in a significant reduction in SBP by an average of 4.5 mmHg and DBP by 2.5 mmHg. [23] There are certainly discrepancies between proper doses and intake durations for cocoa flavanols to have a meaningful effect on blood pressure. However, it appears that even lower doses may have an effect if administered for at least two weeks. [7,22]

There is also evidence that cocoa flavanol supplementation can positively affect FMD%, a popular method of measuring endothelial function of the brachial artery using ultrasound-imaging technology. [17,24,4] FMD involves measurement and analysis of brachial artery diameter before and after a short period of blood vessel occlusion using a rapidly deflating cuff on the forearm. [4] According to Davison, Coates, Buckley, and Howe, a high cocoa flavanol dose of 902 mg for 12 weeks enhanced FMD% by 2.4% in sedentary adults aged 18-65. [6] Other studies showed improvements in FMD% between 1.40 and 3.99% with dose ranges of 371 to 918 mg over durations of 1 day to 2 weeks. [3,4,17,25,26] The considerable variability between doses and duration intakes certainly does not result in any specific insights. However, similar to blood pressure, lower doses administered for at least 2 weeks can still result in significant effects in FMD. [17]

4.6. Cocoa Flavanols and Exercise

In addition to having physiological and clinical relevance, cocoa flavanols may have a positive impact on exercise performance as well through enhancements in endothelial function. [8] Cocoa flavanol supplementation has been used in studies measuring both aerobic and anaerobic exercise performance. Aspects of performance within these areas have been analyzed in the literature with conflicting results.

The combination of cocoa flavanol supplementation and aerobic exercise performance has been given the most amount of attention in the literature. This likely due to the effects of cocoa flavanols on NO availability. [2,8] According to Patel, Brouner, and Spendiff, 14 days of 213 mg of cocoa flavanol supplementation resulted in a 6% increase in maximal oxygen uptake (VO_{2max}). [8] The authors also observed a decrease in oxygen consumption during moderate intensity exercise among moderately-trained male participants. [8] In contrast, another study reported that 900 mg of cocoa flavanol supplementation did not have a significant effect on a 30-minute cycling time trial in well-trained male participants with an average age of 30. [27] These are interesting results, as one study showed significant results with a smaller dose, while another found nothing using a much larger dose. [8,27] However, there is one notable difference between the two studies. Patel, Brouner, and Spendiff tested moderately trained individuals with an average VO_{2max} of 41 ml/kg/min, while Decroix and colleagues tested well-trained cyclists with an average VO_{2max} of 63 ml/kg/min. [8,27] Therefore, cocoa flavanol supplementation may only enhance aerobic performance in participants that are recreationally active or moderately trained.

Very little research has been performed on the effects of cocoa flavanol supplementation on anaerobic exercise performance. One relevant study reported that 213 mg taken for 14 days led to a significant (17%) increase in distance covered during an all-out time trial on a cycle ergometer. [8] Even though this is an interesting finding, further research needs to be performed using more recognized methods of testing anaerobic performance. Studies have, however, proposed ways of enhancing exercise performance through indirect mechanisms. [7,28,29] Exercise-related oxidative stress has been analyzed before and after cocoa flavanol supplementation with promising results. Doses of both 168 mg and approximately 2800 mg of cocoa flavanols had positive effects on oxidative stress caused by exercise. [7,28] In contrast, a study using downhill treadmill running as a stimulus for muscle damage found no significant enhancement in recovery from 350 mg of cocoa flavanols. [29] It is important to note, however, that the latter study utilized endurance-trained participants with an average VO_{2max} of 64.4 ml/kg/min. [29] These results point to training status as a potential limitation for the effects of cocoa flavanol supplementation. Overall, there are interesting findings that point to a possible indirect benefit of cocoa flavanols on exercise performance. There is a great need for more research on the subject.

There may be some interesting connections between endothelial function, clinical health, and exercise performance. In-depth studies on pathways related to NO availability have uncovered significant mechanistic effects that bring about an increase in vasodilation together with a decrease in vasoconstriction. [2,11,13,18,19,30] Nitric oxide availability is increased by cocoa flavanols through increased eNOS availability, as well as inhibition of superoxide anion, ACE, and ET-1. The comprehensive effects of these mechanisms allows for enhanced endothelial function. [2,11,13,18,19,30]

The physiological effects of cocoa flavanols can easily be measured in clinical settings using both blood pressure and FMD. Previous research on both methods has shown promising results for having implications on assessing clinical populations [17,19,23] This means that there is potential for cocoa flavanols to be used in conjunction with other treatments to help those who suffer from chronic diseases related to vascular dysfunction. Previous evidence shows that cocoa flavanols can benefit clinical measurements of endothelial function, such as blood pressure and FMD%. [3,4,7,17,25,26]

After a thorough review, it appears that doses between 168 and 918 mg per day for at least 14 days yield positive results in both blood pressure and FMD%. [3,4,13,17,25,26] This shows that even lower doses can have a significant impact on clinical measurements of endothelial function. It also presents definitive evidence that a daily intake of cocoa flavanols for 14 days can lead to significant beneficial effects. The clinical impact of this information remains somewhat unclear. However, there is potential for cocoa flavanols to attenuate risk factors for certain chronic diseases. [6,11,20,31] Further research is needed to determine how this may affect other populations. In addition, lower and upper dose guidelines need to be established for various populations.

Cocoa flavanols could certainly be supplemented in conjunction with exercise performance as well. Even though there is more conflicting evidence in this area, the mechanisms behind the supplement's functional use are quite clear. Aerobic performance stands to gain the most from cocoa flavanols. More research is needed in this area. However, after a review of the literature, it appears that the training status of individuals may play a role. This could mean that highly trained individuals are simply too fit to benefit from cocoa flavanols. Future research should compare and contrast different training statuses with an equal dose of cocoa flavanols. Even though anaerobic performance evidence is lacking, there are some potential tie-ins to recovery from oxidative stress that could lead to enhanced anaerobic exercise performance. [7,28] This may allow athletes to recover from high intensity activity much faster and allow them to subsequently increase their performance. Specific studies involving anaerobic exercise performance and markers of recovery would be quite helpful in this area. A summary of each article reviewed is included below in Table 1.

Table 1. Summary of Studies Reviewed

Citation	Research Question	Sample Characteristics	CF Dose/ Duration	Results	Other Insights
Actis-Goretta, Ottaviani, & Fraga, 2006	Do flavanol-rich foods work to inhibit ACE activity?	Performed *in vitro* on rat lungs, kidneys, and testes	N/A	Flavanol-rich foods cause ACE inhibition because polymers of epicatechin and catechin have more hydroxyl groups available to create hydrogen bonds with ACE proteins.	- The relationship still needs to be studied *in vivo* - Researchers used wine, chocolate, and tea
Allgrove, Farrell, Gleeson, Williamson, & Cooper, 2011	Does dark chocolate have any effect on oxidative stress caused by exercise?	N = 20 healthy, active men, aged 18-26	- 40 g dark chocolate (approx. 1400 mg CF) 2x/day versus placebo - 2 weeks in duration	- CF's cause a significant decrease in oxidative stress from intense exercise ($p<.001$)	- 90 minutes of cycling at 60% VO_{2max} with 30-second sprints every 10 minutes
Aprotosoaie, Miron, Trifan, Luca, & Costache, 2016	What role do cocoa polyphenols play in aspects of cardiovascular health? Is there a relationship between antioxidant activity and cardiovascular health?	Review; humans, various populations	- 520-990 mg/day - At least 2 weeks or more to be effective	- SBP decreased 2.77-11.30 mmHg, depending on dose - DBP decreased 2.20-7.60 mmHg depending on dose	- There is a positive effect on hypertension - Anti-inflammatory effects - Enhanced vascular function - Antiatherogenic effects - 100-200 mg CF per serving could decrease iron absorption by approx. 70%
Balzer, et al., 2008	Does daily consumption of cocoa powder enhance vascular function in medicated, diabetic patients?	N = 10, aged 50-80, type 2 diabetes for at least 5 yrs., male and female	- Doses of 75, 371, and 963 mg - 30 days, 3x per day	- Caused significant (30%) increase in FMD ($p < .0001$) - Acute increases in FMD are dose-dependent	- FMD is a strong indicator of vascular function- Cocoa powder enhances vascular function in older adults with type 2 diabetes - Dry cocoa beverage mix provided by Mars, Inc.
Berry, Davison, Coates, Buckley, & Howe, 2010	Can consumption of cocoa flavanols enhance BP responsiveness to exercise in overweight/obese individuals?	N = 21, 55 yrs., BMI = 31.6, male and female	- 701 mg (high dose) - 22 mg (low dose) - Measured 2 hours post consumption with	- High dose significantly increased both FMD ($p < .001$) and decreased spike in BP from exercise in overweight/obese individuals	- Cocoa flavanols have acute effects on BP - May allow CVD patients to safely exercise at moderate intensity because less stress is put on the

			3- to 7-day washout (crossover design)		already-damaged heart
Brewster, Perazella, & Setaro, 2003	To explore the RAAS and its potentially negative impacts on cardiovascular health when overactive	Review	N/A	- Explanation of RAAS and how it affects cardiovascular and renal function	- Strong relationship between RAAS and sympathetic nervous system response - ACE blockers widely used to deter effects of overactive RAAS
Davison, et al., 2010	What is the minimum dose of cocoa-flavanol needed to induce BP benefits?	N = 52 male and female (post-menopausal women) with mild Hypertension	- 33, 372, 712, and 1052 mg - Measurements taken at 0, 3, and 6 weeks	- Reductions in 24-hr avg. BP at 1052 mg - SBP decreased 5.3 ± 5.1 mmHg ($p = .001$) DBP decreased 3.0 ± 3.2 mmHg ($p = .002$) - Other doses had no effect	- Not enough evidence to show dose-response relationship with CF & BP - 1052 mg is on lower end of dose-response relationship
Davison, Coates, Buckley, & Howe, 2008	Do cocoa flavanols and exercise have any effect on cardiometabolic function in obese individuals?	N = 49, male and female, aged 18-65, sedentary, BMI >25 kg/m^2	- 902 mg/day - 12 weeks	- High flavanol (902 mg) produced acute (2 hours post) enhanced FMD by 2.4% ($p < .01$) - Effects on FMD continued through 12 weeks of supplementation - Increased insulin sensitivity	- No change in body composition - High flavanol and moderate exercise can decrease CVD risk factors in obese individuals - Dairy-based cocoa mi
Desch, et al., 2010	Do cocoa flavanols decrease BP?	Meta-analysis; Healthy normotensive & stage 1 hypertensive adults (N = 297)	- Doses of 5-174 mg/day - 2-18 weeks in duration	- Doses of 5-174 mg/day - 2-18 weeks in duration	- Variations in doses & chronic usage side-effects need further study
Decroix, et al., 2017	Does acute cocoa flavanol consumption affect various exercise-related changes (antioxidant capacity, oxidative stress, NO production, inflammation, exercise performance & recovery	N = 12 well-trained male cyclists	- 900 mg - Measurements taken 90 minutes and 3 hours post-consumption	- No effect on TNF-a - Increase in total antioxidant levels during rest & exercise - No effect on inflammation, NO, exercise performance, or recovery	- Negative results most likely from participants' training status - Could see effect in un-trained, non-athletes, or clinical populations - Well-trained athletes may not benefit from CF's because they are already healthy
Decroix, Soares, Meeusen, Heyman, & Tonoli, 2018	Do cocoa flavanols have any effect on exercise performance and recovery?	Review; Humans, various populations	- 186-903 mg - Durations of 2-12 weeks	- CF can increase FMD - CF can decrease oxidative stress brought on by exercise - No effect on exercise performance - Reduction in BP	- More research is needed on this topic - More research needed on effect of CF on exercise-induced inflammation
Field & Newton, 2013	Do cocoa flavanols have any anti-hypertensive effects? If so, what mechanisms are behind this?	Review; Humans, various populations	- 520-990 mg - 2-18 weeks in duration	- Cocoa polyphenols appear to have anti-hypertensive effects - Could have significant positive impact on clinical populations	- Mechanisms include: eNOS upregulation and inhibition of superoxide anion, endothelin-1, and ACE - It is important to find adequate sources of

					cocoa flavanols that are low in sugar/fat
Fraga, et al., 2005	Does cocoa flavanol consumption have an effect on vascular health in younger adults?	N = 28 healthy male soccer players aged 18-20	- 168 mg/day - 14 days in duration	- Caused significant 5 mmHg decrease in DBP - 5 mmHg decrease in mean BP - 11% decrease in plasma cholesterol	- There is still evidence that conflicts with these results - Future research should focus on how cells metabolize flavanols
Fraga, et al., 2011	Do cocoa-flavanols assist in enhancing NO production, having an optimal effect on BP?	Review; Humans, various populations	- 30-1000 mg - 2-18 weeks in duration were adequate	- Cocoa flavanols cause a decrease in BP - Multiple mechanisms are at play -Epicatechin may up-regulate eNOS	- Kuna Indians of Panama have low incidence of Hypertension & CVD, but after moving to urban areas it increases - CF's help body maintain adequate levels of NO - CF's & flavanols in general may positively affect oxidative stress
Jumar & Schmieder, 2016	Are there any other antihypertensive effects to be gained from cocoa flavanols besides a decrease in BP?	Review; N = 1297, various populations	- Up to 994 mg/day - Measured 2 hours post-consumption - 2-18 weeks in duration is recommended	- CF's cause vasodilation because of a decrease in ROS, inhibition of ACE, & stimulation of eNOS - Average decrease in BP by 3.0 mmHg (for both SBP and DBP)	- Increased intake of flavanol-containing foods causes decrease in stroke and MI - Positive effects on hypertension are better in younger patients (<50 yrs)
Katz, Doughty, & Ali, 2011	What effects do cocoa & chocolate have on human health & certain diseases?	Review; Humans, various clinical populations	- 88-917 mg/day - At least 2-4 weeks in duration is necessary	- May improve insulin resistance - May improve BP - Un-processed cocoa is healthiest choice - Increased FMD between 1.45% and 3.99%	-Dark chocolate consumption is an adequate way to receive cocoa flavanols
Mogollon, et al., 2013	Does cocoa flavanol consumption have an effect on endothelial function and/or blood pressure on healthy, pregnant women?	N = 44 healthy, pregnant women	- 400 mg per day - 12 weeks in duration	- Significant increase in plasma epicatechin 180 min post-consumption - No effect on FMD, blood pressure, or MAP within or between groups	- Compliance checked via theobromine measurements - A larger sample size may produce more significant results in the future
Monahan, et al., 2011	Does acute cocoa ingestion increase endothelial function (measured by FMD)?	N = 23 healthy older adults (63 ± 2 yrs.)	- Doses of 69, 180, 465, and 1095 mg - Measurements taken both 1 hour and 2 hours post-consumption	- Dose-dependent relationship between cocoa ingestion & FMD -1 hour post: FMD increased by 1.0% with 465 mg; increased by 1.6% with 1095 mg - 2 hours post: FMD increased by 1.4% with 465 mg cocoa; increased by 2.5% with 1095 mg	- Flavanol intake may improve CV health by enhancing FMD - Epicatechin is likely the primary marker in cocoa - Doses smaller than 465 mg were not effective

Monahan, 2012	- Does cocoa/chocolate ingestion have any effect on FMD and cardiovascular health in humans?	Review; Humans, various populations	- 179-963 mg - Measured 2 hours post-consumption - Durations greater than 1 week	- High levels of cocoa flavanols (963 mg) can acutely increase FMD - Moderate intake (371 mg) also shows results (anything above 179 mg is effective) - Peak effects of epicatechin on FMD at 2 hrs after ingestion - Similar effects in chronic (>1 week) durations - Overall, cocoa/chocolate increases FMD in many different populations	-Pure epicatechin ingestion increases FMD - Even 5 g of cocoa can have an effect on FMD - Possible increase in NO availability - Effects on FMD are dose-dependent
Patel, Brouner, & Spendiff, 2015	Does dark chocolate have an effect on oxygen cost of moderate intensity cycling?	N = 9 males aged 20-22, moderately trained	- 213 mg per day - 14 days in duration	- Significantly increased VO_2max by 6% - Chronic supplementation produced increased GET (gas exchange threshold) and TT performance (+17% distance covered) - No statistical difference for BP	- Dark chocolate can act as "ergogenic aid" for shorter duration exercise at moderate intensity -20 min moderate intensity cycle - Distance time trial (2 min)
Peschek, Pritchett, Bergman, & Pritchett, 2014	Do cocoa flavanols have any effect on muscular recovery after exercise?	- N = 8 male endurance athletes aged 18-44 years - Average VO_{2max} of 64.4 ml/kg/min	-350 mg consumed post-exercise - Measurements take 24- and 48-hours post-exercise	There was no significant difference between CF's or placebo on creatine kinase or muscle soreness after a muscle damage protocol	- Downhill treadmill running to induce muscular inflammation - Advanced training status of participants may have played a role in no significant effects - Crossover design
Ried, Fakler, & Stocks, 2017	Do chocolate or coca products containing high flavanols affect blood pressure?	Meta-analysis; N = 1804 Adults with/without hypertension	- 49-3,675 mg per day - Durations of at least 2 weeks	- SBP significantly lowered in those with hypertension and pre-hypertension - Not lowered in normotensive - 2 mmHg average reduction in BP (short term)	- Younger participants respond better to CF's - More research needed on adverse effects of chronic cocoa consumption
Sansone, et al., 2015	What effect do cocoa flavanols have on endothelial function in those at risk for or suffering from cardiovascular disease?	N = 100 healthy males and females aged 35-60	- 450 mg twice per day - 4 weeks in duration	- CF group FMD increased 1.2% - CF group SBP significantly decreased by 4.4 mmHg - CF group DBP decreased by 3.9 mmHg	- Authors used Framingham Risk Score (FRS) - CF group showed significant decrease in FRS for 10-year risk of cardiac events - Even healthy adults may benefit from CF's
Schroeter, et al., 2006	What is the specific mechanism behind the beneficial effects of cocoa flavanols on cardiovascular health?	N = 16 overweight males 25-32 years	- 917 mg taken on two different days - Measurements taken 1, 2, 3, 4, and 6 hours post-consumption	- FMD increased significantly from hour 1 to hour 4 in CF group - No other significant effects on FMD	- Epicatechin is an important part of the mechanism for improved endothelial function from CF ingestion - Use of eNOS inhibitor showed that increases in FMD via epicatechin

					are dependent on this enzyme
Tsukamoto, et al., 2018	Do cocoa flavanols enhance exercise-induced improvements in executive function and memory function?	- N = 10 healthy, active males aged 22.6 ± 0.3 - Peak VO2 = 44.5 ml/kg/min	- 563 mg per day - Exercise testing 70 minutes post-consumption	Cocoa flavanol consumption improves executive function after moderate aerobic exercise, but not memory function	Improvements in cerebral blood flow and endothelial function in general may be behind this
Vlachojannis, Erne, Zimmermann, & Chrubasik-Hausmann, 2016	Which doses of cocoa flavanols have an effect on cardiovascular aspects of health?	Review; Human, various populations	- 400-900 mg - Durations of 2 weeks or more	- Cocoa with 100 mg epicatechin can increase FMD by 1.1%(dose-related) - 400-900 mg CF may increase FMD - BP decreased after 2 weeks consumption for all studies	- 100 – 500 g chocolate (for CF's) and 50 – 200 g chocolate (for epicatechin) - Chocolate products should provide amounts of epicatechin & CF's on labels
Westphal & Luley, 2011	Does consumption of flavanols mend endothelial dysfunction caused by a high fat meal?	N = 18 healthy students (25 ± 2.5 yrs.)	- High dose of 918 mg or low dose of 14 mg - Measurements taken over span of 6 hours post-consumption	- High levels of cocoa flavanols decrease endothelial dysfunction from a high fat meal - FMD was significantly better after CF intake ($p < .001$) - Most likely from increased eNOS	- Epicatechin is most likely responsible for increases in FMD - Interaction between epicatechin and eNOS is important

Abbreviations: ACE = angiotensin converting enzyme, BP = blood pressure, CF = cocoa flavanols, CVD = cardiovascular disease, DBP = diastolic blood pressure, eNOS = endothelial nitric oxide synthase, FMD = flow-mediated dilation, MI = myocardial infarction, NO = nitric oxide, RAAS = renin-angiotensin aldosterone system, ROS = reactive oxygen species, SBP = systolic blood pressure, TT = time trial

5. Conclusions

The present review explored the current status of cocoa flavanols in the literature, specifically relating to endothelial function and exercise performance. Their positive effects on endothelial function are well documented, and cocoa flavanols could be utilized in clinical settings in the ongoing battle against vascular dysfunction. In addition, exercise performance may stand to benefit as well. Cocoa flavanols hold a large amount of un-tapped potential in various areas of research. Further work is certainly necessary to fully understand their benefits. Overall, it is important to realize that cocoa flavanols may be beneficial to human health, and that their effects on endothelial function can be applied in many directions to benefit different populations. With future research in more specific directions, cocoa flavanols could likely be seen as a more effective and useful nutritional supplements.

REFERENCES

[1] Tsukamoto H, Suga T, Ishibashi A, et al. Flavanol-rich cocoa consumption enhances exercise-induced executive function improvements in humans. *Nutrition*. 2018; 46: 90-96.

[2] Aprotosoaie CA, Miron A, Trifan A, Luca SV, Costache I-I. The Cardiovascular Effects of Cocoa Polyphenols—An Overview. *Dis* . 2016; 4(4).

[3] Katz DL, Doughty K, Ali A. Cocoa and chocolate in human health and disease. *Antioxid Redox Signal*. 2011; 15: 2779+-2779+.

[4] Monahan KD, Feehan RP, Kunselman AR, Preston AG, Miller DL, Lott MEJ. Dose-dependent increases in flow-mediated dilation following acute cocoa ingestion in healthy older adults. *J Appl Physiol*. 2011; 111(6): 1568-1574.

[5] Davison K, Berry NM, Misan G, Coates AM, Buckley JD, Howe PRC. Dose-related effects of flavanol-rich cocoa on blood pressure. *J Hum Hypertens*. 2010;24:568+-568+.

[6] Davison K, Coates AM, Buckley JD, Howe PRC. Effect of cocoa flavanols and exercise on cardiometabolic risk factors in overweight and obese subjects. *Int J Obes (Lond)*. 2008; 32(8): 1289-1296.

[7] Fraga CG, Actis-Goretta L, Ottaviani JI, et al. Regular consumption of a flavanol-rich chocolate can improve oxidant stress in young soccer players. *Clin Dev Immunol*. 2005; 12(1): 11-17.

[8] Patel RK, Brouner J, Spendiff O. Dark chocolate supplementation reduces the oxygen cost of moderate intensity cycling. *J Int Soc Sports Nutr*. 2015; 12(1): 47.

[9] Decroix L, Soares DD, Meeusen R, Heyman E, Tonoli C. Cocoa Flavanol Supplementation and Exercise: A Systematic Review. *Sport Med*. 2018; 48(4): 867-892.

[10] Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration. *J Clin Epidemiol*. 2009; 62(10): e1-e34.

[11] Field D, Newton G. The anti-hypertensive actions of cocoa polyphenols - a review. *Curr Top Nutraceuticals Res*. 2013; 11(4): 113-128.

[12] Actis-Goretta L, Ottaviani JI, Fraga CG. Inhibition of Angiotensin Converting Enzyme Activity by Flavanol-Rich Foods. *J Agric Food Chem*. 2006; 54(1): 229-234.

[13] Fraga CG, Litterio MC, Prince PD, Calabró V, Piotrkowski B, Galleano M. Cocoa flavanols: effects on vascular nitric oxide and blood pressure. *J Clin Biochem Nutr*. 2011; 48(1): 63-67.

[14] Maiorana A, O'Driscoll G, Taylor R, Green D. Exercise and the Nitric Oxide Vasodilator System. *Sport Med*. 2003; 33(14): 1013-1035.

[15] Mogollon JA, Bujold E, Lemieux S, et al. Blood pressure and endothelial function in healthy, pregnant women after acute and daily consumption of flavanol-rich chocolate: a pilot, randomized controlled trial. *Nutr J*. 2013; 12(1): 41.

[16] Schroeter H, Heiss C, Balzer J, et al. (-)-Epicatechin mediates beneficial effects of flavanol-rich cocoa on vascular function in humans. *Proc Natl Acad Sci U S A*. 2006; 103(4): 1024-1029.

[17] Monahan KD. Effect of cocoa/chocolate ingestion on brachial artery flow-mediated dilation and its relevance to cardiovascular health and disease in humans. *Arch Biochem Biophys*. 2012; 527(2): 90-94.

[18] Brewster UC, Perazella MA, Setaro JF. The Renin-Angiotensin-Aldosterone System: Cardiorenal Effects and Implications for Renal and Cardiovascular Disease States. *Am J Med Sci*. 2003; 326(1): 15-24.

[19] Jumar A, Schmieder RE. Cocoa Flavanol Cardiovascular Effects Beyond Blood Pressure Reduction. *J Clin Hypertens*. 2016; 18(4): 352-358.

[20] Balzer J, Rassaf T, Heiss C, et al. Sustained benefits in vascular function through flavanol-containing cocoa in medicated diabetic patients a double-masked, randomized, controlled trial. *J Am Coll Cardiol*. 2008; 51(22): 2141-2149.

[21] Sansone R, Rodriguez-Mateos A, Heuel J, et al. Cocoa flavanol intake improves endothelial function and Framingham Risk Score in healthy men and women: a randomised, controlled, double-masked trial: the Flaviola Health Study. *Br J Nutr*. 2015; 114(8): 1246-1255.

[22] Ried K, Fakler P, Stocks NP. The effect of cocoa on blood pressure. *Cochrane Database Syst Rev*. 2017; 4: 1-88.

[23] Desch S, Schmidt J, Kobler D, et al. Effect of Cocoa Products on Blood Pressure: Systematic Review and Meta-Analysis. *Am J Hypertens*. 2010; 23(1): 97-103.

[24] Harris RA, Nishiyama SK, Wray DW, Richardson RS. Ultrasound assessment of flow-mediated dilation: a tutorial. *Hypertension*. 2010;55(5):1075-1085.

[25] Vlachojannis J, Erne P, Zimmermann B, Chrubasik - Hausmann S. The Impact of Cocoa Flavanols on Cardiovascular Health. *Phyther Res*. 2016; 30(10): 1641-1657.

[26] Westphal S, Luley C. Flavanol-rich cocoa ameliorates lipemia-induced endothelial dysfunction. *Heart Vessels*. 2011; 26(5): 511-515.

[27] Decroix L, Tonoli C, Soares DD, et al. Acute cocoa Flavanols intake has minimal effects on exercise-induced oxidative stress and nitric oxide production in healthy cyclists: a randomized controlled trial. *J Int Soc Sports Nutr*. 2017; 14(1): 28.

[28] Allgrove J, Farrell E, Gleeson M, Williamson G, Cooper K. Regular dark chocolate consumption's reduction of oxidative stress and increase of free-fatty-acid mobilization in response to prolonged cycling. *Int J Sport Nutr Exerc Metab*. 2011; 21(2): 113-123.

[29] Peschek K, Pritchett R, Bergman E, Pritchett K. The Effects of Acute Post Exercise Consumption of Two Cocoa-Based Beverages with Varying Flavanol Content on Indices of Muscle Recovery Following Downhill Treadmill Running. *Nutr*. 2014; 6(1).

[30] Jiménez R, Duarte J, Perez-Vizcaino F. Epicatechin: Endothelial Function and Blood Pressure. *J Agric Food Chem*. 2012; 60(36): 8823-8830.

[31] Berry NM, Davison K, Coates AM, Buckley JD, Howe PRC. Impact of cocoa flavanol consumption on blood pressure responsiveness to exercise. *Br J Nutr*. 2010; 103(10): 1480-1484.